
Part II

Environmental Protection Agency

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40 CFR Part 79

Fuels and Fuel Additives Registration Regulations; Final Rule
ENVIRONMENTAL PROTECTION AGENCY

40 CFR Part 79

[FRL-4892-7]
RIN 2060-AC10

Fuels and Fuel Additives Registration Regulations

AGENCY: Environmental Protection Agency (EPA).

ACTION: Final rule.

SUMMARY: This final rule establishes new requirements for the registration of designated fuels and fuel additives (F/FAs) as authorized by sections 211(b)(2) and 211(e) of the Clean Air Act (CAA).

The registration requirements are organized within a three-tier structure. Tier 1 requires F/FA manufacturers to perform a literature search on the health and welfare effects of F/FA emissions, characterize the emissions, and provide qualitative exposure information. Tier 2 requires biological testing for the examination of subchronic systemic and organ toxicity, as well as the assessment of specific health effects endpoints. When necessary, Tier 3, which includes follow-up studies or other additional tests, may be required. The rule permits adequate existing test data to be submitted in lieu of conducting new duplicative tests. It also includes special provisions for small businesses and certain types of products, and a grouping system which permits manufacturers of similar F/FA products to share the costs of compliance.

DATES: This regulation is effective May 27, 1994. The incorporation by reference of certain publications listed in the regulations is approved by the Director of the Federal Register as of June 27, 1994.

The information collection requirements contained in 40 CFR 79.51, 79.52, and 79.57 through 79.68 have not been approved by the Office of Management and Budget (OMB) and are not effective until OMB has approved them. EPA will publish a document in the Federal Register announcing OMB approval of the information collection requirements.

ADDRESSES: The record for this rulemaking is contained in Docket No. A-90-07. The docket is located at the Air Docket, Room M-1500, 401 M Street SW., Washington, DC 20460; phone (202) 260-7548 or 7549; fax (202) 260-4000. The docket is open for public inspection from 8 a.m. until 4 p.m., Monday through Friday. As provided in 40 CFR part 2, a reasonable fee may be charged by EPA for photocopying services. Electronic copies of major F/FA rulemaking documents can be obtained through the Office of Air Quality Planning and Standards (OAQPS) Technology Transfer Network Bulletin Board System (TTNBBS). Details on how to access TTNBBS are included in Section XIV of this preamble.

FOR FURTHER INFORMATION CONTACT: Regarding F/FA registration procedures: James Caldwell (phone 202-233-9303) or Joseph Fernandes (phone 202-233-9016). USEPA, OMS Field Operations and Support Division, Mail Code 6406J, 401 M Street SW., Washington, DC 20460. Regarding technical requirements: Kent Helmer (phone 313-741-7825). USEPA, OMS Regulation Development and Support Division, Mail Code RDSD-12, 2565 Plymouth Road, Ann Arbor, MI 48105.

SUPPLEMENTARY INFORMATION:

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I. Introduction

Over 2,300 fuels and 4,800 fuel additives were registered by EPA as of March 1994 and, to some degree, each of them produces emissions which may contribute to potentially harmful air pollution. The primary purpose of today's rule is to establish registration requirements which will provide information for identifying and evaluating the potential adverse effects of designated F/FA emissions and for guiding the direction of related regulatory actions in the future as specified in section 211 of the CAA.

Previous actions have implemented CAA sections 211(a) and 211(b)(1), which govern the general registration of F/FAs, as well as CAA section 211(f). Today's rule amends 40 CFR part 79 by adding regulatory provisions requiring the testing of F/FAs as a requirement for registration, as stipulated in section 211(b)(2) and section 211(e) of the CAA.

In addressing these additional statutory provisions, this rule focuses on the identification and evaluation of potential adverse health effects associated with F/FA evaporative and combustion emissions. The required health effects evaluation is organized in a tiered structure, and includes emission characterization, literature search, and biological testing requirements. Although this testing framework focuses on the evaluation of health effects, F/FA manufacturers are also required to perform data searches to obtain information on the potential welfare effects of F/FA emissions. In addition, EPA will continue to use existing procedures under CAA section 211(f) for the evaluation of potential effects of F/FAs on ECS performance.

The ultimate use of the registration information to be submitted in compliance with this rule is to guide EPA in potential future regulatory actions under CAA section 211(c). Section 211(c) provides authority for the possible control or prohibition of any fuel or fuel additive whose emission products cause or contribute to air pollution which may reasonably be anticipated to endanger the public health or

welfare. Evidence of adverse effects of F/FA emissions on ECS performance, obtained under CAA section 211(f) or from other sources, could also be used by EPA to support such regulatory decisions.

II. Background

A. Legal Authority and Statutory History

The legal authority for the F/FA registration program is provided by section 211 of the CAA. Section 211(a), 42 U.S.C. section 7545, authorizes EPA to designate any fuel or fuel additive and prohibits manufacturers of designated fuels or additives from selling such products unless they have been registered by EPA in accordance with CAA section 211(b). In 1975, EPA issued regulations (40 CFR part 79) implementing basic registration requirements, as stipulated by CAA section 211(b)(1), that included: commercial identifying information, range of concentration, purpose-in-use, and chemical composition.

Section 211(b)(2) of the CAA also gives EPA discretionary authority to establish additional registration requirements. According to this section, EPA ``may also require the manufacturer of any fuel or fuel additive to conduct tests to determine potential public health effects of such fuel or fuel additive (including, but not limited to, carcinogenic, teratogenic, or mutagenic effects)," and to furnish other ``reasonable and necessary" information to identify F/FA emissions and determine their effects on vehicular emission control performance and on the public health and welfare. The statute further stipulates that testing for health effects is to be conducted according to procedures and protocols established by the Administrator, and that test results will not be considered confidential. Once the manufacturer has completed registration requirements and has given assurances that the Agency will be notified of future changes in that information, CAA section 211(b)(3) directs the Administrator to register the fuel or fuel additive.

EPA did not exercise its discretionary authority to require testing of F/FAs under CAA section 211(b)(2) as part of the general registration regulations issued in 1975. However, in the CAA Amendments of 1977 (PL 95-95, August 7, 1977), Congress added section 211(e), which made implementation of section 211(b)(2) mandatory and contained additional provisions requiring the implementation of the regulations within one year of enactment of the CAA Amendments. In an effort to fulfill this requirement, EPA published an Advanced Notice of Proposed Rulemaking (ANPRM) in 1978 (see 43 FR 38607, August 29, 1978; Docket ORD-78-01). However, the rulemaking process did not go forward during the next ten years and the rule was not finalized. Nevertheless, this action remained on EPA's regulatory agenda and a development plan for

the rulemaking was created in 1988.

In 1989, a citizens group brought a lawsuit [Thomas v. Browner, C.A. No. 89-6269 (D. Oreg. 1989)] challenging EPA's failure to promulgate F/FA testing regulations within the one-year deadline stipulated in CAA section 211(e). EPA entered into a Consent Decree in settlement of this lawsuit which, together with subsequent modifications, established the rulemaking schedule. Accordingly, a new ANPRM was published on August 7, 1990 (55 FR 32218) and a Notice of Proposed Rulemaking (NPRM) was published on April 15, 1992 (57 FR 13168). Public hearings as well as periods for written commentary followed both of these publications. On February 24, 1994, EPA published a Notice of Reopening of Comment Period (59 FR 8886) requesting public comment on several compliance-related and technical issues that needed clarification and/or reconsideration. Today's action culminates the rulemaking process by promulgating F/FA registration requirements under CAA sections 211(b)(2) and 211(e).

B. Public Participation

In the months following the publication of the ANPRM (55 FR 32218) and the associated public hearing (on September 26, 1990), EPA explored the feasibility and appropriateness of applying regulatory negotiation procedures (under the provisions of section 583 of the Negotiated Rulemaking Act of 1990) to the development of this rule. Interviews and meetings were held with representatives of a variety of affected industry groups and environmental organizations, to assess their interest and willingness to participate in potential negotiations. This process indicated that there was insufficient support for regulatory negotiation among a number of key parties. A traditional rulemaking procedure was then followed to develop this rule.

Following publication of the proposed rule, EPA held a public hearing on May 28, 1992, and accepted comments until June 30, 1992. Public response on the NPRM included five oral presentations at the hearing and the subsequent submission of 42 written comments. EPA also received 13 written comments on the issues discussed in the Notice of Reopening of Comment Period (referred to as the ``Reopening Notice" in later portions of this document). A transcript of both public hearings and copies of all written comments are available in public Docket No. A-90-07.

A discussion of comments received since the NPRM and EPA's responses are included in the ``Summary and Analysis of Comments for the Fuels and Fuel Additives Registration Regulations," which is available in the public docket referenced above. All public commentary was carefully considered in developing this final rule. Major areas of comment are described in the relevant sections of this preamble.

C. Additional Information on the Effective Date

The effective date of this rule is May 27, 1994. EPA notes that the general requirement (under 5 U.S.C. 553(d), the Administrative Procedure Act (APA)), that publication or service of a substantive rule be made not less than 30 days before its effective date, does not apply here. Under 5 U.S.C. 559, the APA states that a subsequent statute does not supersede or modify the APA except to the extent that it does so expressly. CAA section 307(d)(1)(E) specifically applies to the promulgation or revision of any regulation pertaining to any fuel or fuel additive under CAA section 211. CAA section 307(d)(1) further provides that ``[t]he provisions of sections 533 through 557 and section 706 of title VI shall not, except as expressly provided in this subsection, apply to actions to which this subsection applies." Nowhere does subsection 307(d) expressly provide that section 553(d) of title 5 applies.

Further, CAA section 211(e)(2) expressly provides that the time period for providing the ``requisite information" under section 211(e)(2) is based on the ``date of promulgation" of the rule. Therefore, the requirements under CAA section 211(e)(2) are effective on May 27, 1994. Additionally, even if section 553(d) were to apply to the portion of the rule promulgated under the authority of section 211(b) of the CAA, there is good cause under section 553(d)(3) of the APA to provide less than 30 days notice following publication in order to simplify implementation of the rule by establishing one effective date for the rule's requirements. As discussed in the following sections, this final rule provides a six year time period for completing Tier 2 testing, commencing on May 27, 1994. EPA believes this to be sufficient for the regulated industry to comply with the rule and that, given this compliance schedule, a reasonable amount of notice is provided for this type of information-gathering regulation. Finally, EPA has taken steps to provide notice of this final action to the regulated industry upon signature of the rule. For these reasons, EPA believes that establishing the effective date as May 27, 1994 is reasonable.

III. Overview of Program Requirements

A. Overall Scope and Approach

The requirements of this rule apply to all types of F/FAs which have been designated to be registered by EPA (see Section III.A.3). Based on the provisions of CAA section 211(e), the requirements must be satisfied both by manufacturers of F/FA products registered at the time of promulgation as well as manufacturers of F/FA products seeking

registration after promulgation. Considering the large number of F/FA products to be evaluated and the potential burden of the program on the regulated industry, this final rule maintains the grouping system and the tiered approach proposed in the NPRM.

The grouping system allows manufacturers of similar products to share the costs of testing. Rather than mandating comprehensive testing as a routine registration requirement for every registered fuel and fuel additive, the grouping system permits the testing of one product as a representative of all relatively similar products (see Section IV of this preamble). In addition, the testing program is designed to address testing needs on a tiered basis, with allowance for more rigorous, resource-intensive requirements contained in each successive tier. A detailed description of the scope and requirements of each individual tier is provided in Sections VII, VIII, and IX of this preamble, and procedures for generating the emissions to be tested are explained in Section VI. Additional special provisions to reduce the burden on the regulated industry are discussed in Section X.

EPA believes that the program required by this final rule is consistent with the CAA and reflects a reasonable and cost-conscious approach to a very complex regulatory area. The following sections present a general overview of the main provisions of the rule.

1. Tiered Approach

As depicted in Figure 1, the registration program's requirements are organized within a three-tier structure. In part, each tier is intended to function as a screen for determining the need for more rigorous requirements in subsequent tiers. Consistent with CAA section 211(e)(3), which authorizes EPA to avoid duplication of effort, the tiered approach permits F/FA manufacturers to use test results and other information which may already be available about their products.

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a. Content of Tiers. Except as may be modified by any applicable special provisions, the requirements of Tiers 1 and 2 are mandatory for all fuels and fuel additives. These requirements may be satisfied by manufacturers either on an individual basis or by way of a group submission consistent with the provisions of the grouping system.

Under Tier 1, F/FA manufacturers are required to perform a literature search on the health and welfare effects of F/FA emissions, characterize the emissions, and provide a qualitative exposure analysis based on total annual production volume and market distribution data

(see Section VII for details on Tier 1 requirements). The modeling analyses proposed in the NPRM have been eliminated from Tier 1 as explained in Section III.C of this preamble. Tier 2 (see Section VIII) includes biological testing for specific health effect endpoints, as well as general systemic and organ toxicity. The Tier 2 biological testing requires the exposure of laboratory animals to the whole emissions of fuels or additive/base fuel mixtures.\1\ To the extent that previously conducted studies are available which are at least comparable to the specified guidelines for the chemical and/or biological tests required in Tiers 1 and 2, such existing data may be submitted in lieu of performing and reporting on new duplicative tests (see Section VII.A.2).

\1\An additive must be mixed with the base fuel of its associated fuel family prior to generating emissions for testing (see Sections IV.A and IV.B.1).

The results of Tiers 1 and 2 are to be reported to EPA according to the report formats described in Section XII. EPA will evaluate these results to determine if additional testing or analysis may be indicated under the provisions of Tier 3. For the purpose of peer review during this evaluation process, EPA may furnish the submitted data to, and consult with, other organizations, such as the Health Effects Institute. Tier 3 tests will be determined on a case-by-case basis at EPA's discretion, as discussed in Section IX. The specific objectives and scope of Tier 3 tests will vary depending on the concerns identified in the earlier tiers or any other information available to EPA.

b. Timing of Requirements: Registered F/FAs. EPA proposed to require that Tier 1 and Tier 2 data be submitted within three years for registered F/FAs. In their comments on the proposal the regulated industry suggested that it would not be possible to complete Tier 2 testing within three years. As detailed in the Summary and Analysis of Comments, these commenters stated that the number of F/FAs to be tested, the time needed for development of detailed test protocols, and the lack of available test facilities were inconsistent with the three-year time frame for completion of Tier 1 and Tier 2 for all F/FAs (or groups). None of the commenters, however, suggested an alternative time frame within which the testing of all F/FAs (or groups) could be completed.

EPA has thoroughly considered these comments and, accordingly, has taken a number of measures to streamline the program. As discussed later in this preamble, such measures include the deletion from the

final rule of some requirements proposed in the NPRM (e.g., quantitative modeling requirements), modification of the Tier 2 testing scenario for greater efficiency, simplification of some of the grouping rules, and the addition of special provisions which will result in a smaller number of F/FA products requiring testing.

Nevertheless, EPA recognizes that the number of laboratory facilities currently available to conduct the required emission-based toxicological tests is very limited. EPA expects that the promulgation of this rule will create a demand for testing laboratories which will encourage the reactivation, modification, and/or expansion of existing laboratories, as well as the development of new facilities, to accommodate the requirements of the F/FA registration program. However, there is likely to be a lag between the demand for and the availability of laboratory capacity. Thus, while EPA believes that some groups could complete the testing required by the rule in three years, it is likely that not all of the F/FAs to be tested could complete the requirements in the three-year time frame.

Considering these factors, the final rule allows a six-year period for the conduct of Tier 2 testing for registered F/FAs. This longer period will provide the necessary start-up time for laboratories with previous experience in conducting studies reasonably similar to those required in Tier 2 to adapt or build the necessary facilities, organize the key technical personnel, and conduct verification procedures. In this regard, it is not necessary for each laboratory providing services for this program to employ experts in each of the toxicology specialty fields covered by Tier 2. EPA recognizes that some of the required expertise may be in short supply, and envisions that laboratories may subcontract with subspecialists as needed for evaluation of test results. Furthermore, biological laboratories which currently offer inhalation toxicology testing services, but are not equipped to generate and deliver engine emissions for such testing, should be able to obtain the additional equipment and engineering expertise they will need in a relatively short period of time. Addition of emission generation capabilities by such laboratories will be facilitated by the fact that the final rule allows the use of relatively inexpensive and possibly portable engine dynamometers for generating the required emissions for toxicological testing related both to light-duty and heavy-duty engine applications.

EPA estimates that the necessary toxicology laboratory capacity will begin to come on line within 8-15 months of the effective date of this rule, with expansion of capacity continuing for an additional 12-24 months thereafter. The initial period should coincide with the start-up time needed by the regulated industry prior to beginning the required Tier 2 testing. Manufacturers' start-up activities will include review and understanding of the requirements, formation and

functional organization of groups, acquisition of required test fuel supplies, and contracting for data gathering and testing services. EPA estimates that these activities can generally be accomplished in 6-12 months.

With six years provided for full Tier 2 completion, sufficient time should then be available for completion of all Tier 2 testing. However, this assumes that the regulated industry will not purposely delay the onset of testing. If all F/FA groups wait until the fourth and fifth years to begin the Tier 2 testing, it will again become likely that some will not be able to complete the requirements on time. In such instances, the responsible manufacturers will have failed to comply with the requirements of this rule and will be subject to enforcement action and/or loss of registration.

Thus, in the case of registered F/FA products, this final rule requires the submission within three years from the effective date of all applicable Tier 1 requirements plus either: (1) Submittal of all Tier 2 requirements or (2) evidence of a contractual obligation with a qualified laboratory to conduct the required Tier 2 tests.\2\ If, within the first three years, a contract for Tier 2 is submitted rather than the Tier 2 data itself, then the final Tier 2 report is due to EPA no later than six years of the effective date of this rule. Both Tiers 1 and 2 are mandatory. Failure to submit Tier 2 data for a registered F/FA within six years of the effective date of this rule will subject the manufacturer to enforcement action and/or revocation of the registration. In the case of F/FAs for which Tier 2 testing is not required because of special provisions, all applicable requirements are due to EPA within the initial three-year period after promulgation of this rule.

\2\To be qualified, a laboratory must be able to perform inhalation toxicology tests in compliance with the Good Laboratory Practice requirement in this rule, including monitoring by an onsite Quality Assurance Unit. It must also be able to properly and safely store, transport, and use F/FAs. The study director must be a professional scientist with a doctoral degree in toxicology or equivalent. Other individuals engaged in the conduct of the studies shall have the education, training, and/or experience to enable proper performance of the assigned functions. The laboratory's animal handling facility must be registered and in good standing with the U.S. Department of Agriculture. Accreditation with a recognized independent organization which sets laboratory animal handling standards [e.g., the American Association for Accreditation of Laboratory Animal Care (AAALAC)] is required.

Existing F/FA registrations are also conditional on satisfaction of any Tier 3 requirements which might be prescribed by the Agency pursuant to CAA section 211(b). When Tier 3 testing is prescribed for a registered F/FA product, the existing registration will be extended for that time which EPA specifies as necessary for completion of the additional requirements. Maintenance of registration will depend upon satisfactory compliance with these requirements.

EPA is promulgating Tier 2 testing requirements under the authority of both sections 211(b) and 211(e) of the CAA. The requirements for Tier 2 testing are all within EPA's discretion under section 211(b). Section 211(b) gives the Administrator broad authority ``for the purpose of registration of fuels and fuel additives" to require manufacturers ``to conduct tests to determine potential public health effects of such fuel or fuel additive." EPA interprets ``for the purpose of registration" to encompass both gaining and maintaining registration for F/FAs. This interpretation is supported by section 211(e), which requires implementation of section 211(b) authority with respect to both registered and new F/FAs and mandates that EPA require testing of F/FAs. The legislative history supports such a view. The 1977 House Report,\3\ upon which section 211(e) was based, states:

\3\H. Rept. No. 294, 95th Cong., 1st Sess. 308, reprinted in 1977, U.S. CODE CONG. & ADMIN. NEWS 1077, 1387.

Section 220 of the Committee bill is intended to express the Committee's disapproval of EPA's past handling of its authority in this area and of its proposed future plans * * * Instead, an aggressive, preventative approach to the gathering of necessary information is mandated * * *

The bill mandates the Administrator to promulgate regulations within one year after enactment. * * * These regulations must require testing by the manufacturer of the fuel or fuel additive, except insofar as paragraph (3) otherwise permits.

All of these requirements are mandatory.

Tier 2 involves testing ``to determine potential public health effects" of F/FAs and, therefore, is within the type of testing that section 211(b) allows EPA to require. The timing requirements of Tier 2 are also within EPA's discretion under section 211(b), for section 211(b) leaves submission deadlines to EPA's discretion.

The timing for Tier 2 submissions is also governed, at least in part, by section 211(e)(2). That section requires that, for F/FAs registered when the rule is promulgated, ``requisite information" be submitted within three years of the promulgation date. ``Requisite

information" is not defined in the statute. EPA proposed to interpret ``requisite information" as the data required by Tiers 1 and 2. In part this was based on EPA's understanding that Congress intended that the testing rule promulgated under section 211(e) would require manufacturers to conduct testing--not merely conduct a literature search and compile studies that had already been conducted. As a practical matter, however, EPA now believes that the Tier 2 tests cannot be completed for all F/FAs within three years. EPA believes this makes its proposed definition of ``requisite information" unreasonable and requires a different interpretation of ``requisite information."

EPA considered redefining ``requisite information" to mean studies that could be completed for all F/FAs within three years. Given the time frame, laboratory availability, and the number of groups to be tested, EPA was not sure that any meaningful health effects testing could be accomplished for all groups within three years. Certainly, such testing could not include testing of combustion and evaporation products for all groups. As discussed elsewhere, EPA believes that the testing of combustion and evaporative emissions included in Tier 2 is part of the basic testing necessary to evaluate potential health effects, because people are exposed to both combustion and evaporative emissions. Therefore, EPA would require this testing under section 211(b) regardless of whether the testing is required by section 211(e)(2). If EPA had chosen to interpret ``requisite information" to be testing that could be done in three years, EPA would have, in any case, required such testing in addition to the Tier 2 testing required by this rule. This would likely have delayed submission of the information that EPA believes to be necessary (i.e., the combustion emissions testing and evaporative emissions testing).

In today's rule, EPA interprets ``requisite information" as either data required by Tiers 1 and 2 or data required by Tier 1 and a commitment to conduct Tier 2 testing. EPA believes that this meets the congressional mandate to require emissions speciation testing and a demonstration that manufacturers are making progress in their testing by requiring submission of evidence of a contract with a qualified laboratory to conduct the Tier 2 studies. In addition, this interpretation imposes testing costs on manufacturers only for types of studies that EPA believes are necessary and useful, and it requires manufacturers to finish testing in a time frame that EPA believes is possible to meet.

Even if ``requisite information" were interpreted to mean only data required by Tier 1, EPA would still impose Tier 2 testing and timing requirements as contained in today's rule. In that case, section 211(e) would require Tier 1 data to be submitted within three years of this rule's promulgation, and section 211(b) would provide authority for EPA to impose the Tier 2 testing requirements in the time frame set

forth in this rule.

c. Timing of Requirements: Registrable and New F/FAs. Consistent with section 211(e), for F/FAs not yet registered, all test requirements must be satisfied prior to registration, including any Tier 3 requirements which EPA judges to be necessary. However, as discussed in the Reopening Notice, this final rule makes a distinction between ``registrable'' and truly ``new'' F/FA products. Registrable F/FAs are products\4\ that are not registered as of the effective date of this rule but that meet the program's criteria for grouping with a currently registered fuel or bulk additive\5\ in the same fuel family.\6\ Conversely, a F/FA product not registered as of the effective date of this rule is designated as ``new'' if it does not meet the program's criteria for grouping with a currently registered fuel or bulk additive in the same fuel family. In the above definitions, the term ``currently'' refers to the date on which EPA receives the basic registration data for the F/FA product in question.\7\

\4\Registration is product-specific. Thus, if a particular fuel or additive product has not been registered by its manufacturer, then that manufacturer does not have the right to introduce, market, and/or sell this product, even if a compositionally similar or identical product has been registered by another manufacturer.

\5\A ``bulk additive,'' sometimes called a ``general use'' additive, is defined as a product added to fuel at the refinery as part of the original blending stream or after the fuel is transported from the refinery, but before the fuel is purchased for introduction into the fuel tank of a motor vehicle. In contrast, an ``aftermarket additive,'' sometimes called a ``consumer additive,'' is an additive product which is added by the end-user directly to fuel in a motor vehicle or engine to modify the performance or other characteristics of the fuel, the engine, or its emissions.

\6\``Fuel family'' refers to the primary categorization of F/FAs in the grouping system of this registration program, as described in Section IV. A fuel family is defined as a set of F/FAs which share basic chemical and physical formulation characteristics and can be used in the same engine or vehicle. In the definition of ``registrable'', the restriction ``in the same fuel family'' means that the similarity of an applicant F/FA product to a bulk additive currently registered for use in another fuel family will not suffice to make the applicant F/FA product registrable. This restriction is consistent with the general principles of the grouping system, which permits grouping of F/FAs only within the defined fuel families.

\7\Revision of an existing registration (e.g., addition or

deletion of a currently-registered bulk additive to an existing fuel registration) does not constitute a new registration (assuming any added bulk additive is registered for use in fuels of the same type). However, test requirements may change if the revision causes the fuel product to change from one group to another.

For registrable products, similar testing and compliance requirements apply as those pertaining to currently registered F/FAs in the same group. Upon the manufacturer's submittal of the basic registration data and other pre-Tier 1 application requirements for a registrable product, registration will be granted by EPA. Once registered, these products will be legally able to enter the market. The manufacturer will have the same period of time after the effective date of this rule for the completion of Tiers 1 and 2 as the applicable group of existing F/FAs, and can satisfy these requirements either by joining the existing group or by testing individually. On the other hand, manufacturers of new F/FA products (i.e., F/FAs not registered as of the effective date of this rule and not fitting the registrable criteria) are required to submit all testing requirements prior to registration, including Tier 3 when prescribed by the Agency. Thus, if EPA identifies a need for additional testing at the Tier 3 level for a new F/FA product, registration will not occur until satisfactory completion of all such requirements.

As discussed in the NPRM and in the Reopening Notice, EPA interprets CAA section 211(b) in conjunction with CAA section 211(c), which gives EPA authority to control or prohibit the manufacture, introduction into commerce, offering for sale, or sale of any fuel or fuel additive if the Administrator finds that the emission products of such fuel or fuel additive ``cause[s], or contribute[s], to air pollution which may reasonably be anticipated to endanger the public health or welfare." In light of this responsibility, EPA believes that it should exercise particular caution in registering new F/FA products and that it should have the necessary information to evaluate fully the potential public health consequences of such new F/FAs prior to allowing their introduction into the market. Thus, before granting registration to manufacturers of new F/FAs, under the authority of CAA section 211(b), this final rule requires that they comply with all testing requirements.

Figure 2 summarizes the decision process for determining whether an unregistered F/FA product (i.e., a F/FA product not registered as of the effective date of this rule) is ``registrable" and thus handled much like a currently registered F/FA product, or whether an unregistered F/FA product is ``new" and must complete all testing requirements before registration is granted.

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An unregistered F/FA product which meets the criteria for grouping only with a currently registered aftermarket additive (and not with a currently registered fuel and/or bulk additive) is not designated as registrable. This does not necessarily preclude an unregistered aftermarket additive from being registrable (since aftermarket additives can group with fuels and bulk additives), nor does it affect the registration status of currently registered aftermarket additives.

For example, an unregistered detergent additive (either bulk or aftermarket) intended for use in gasoline and meeting the criteria for grouping with baseline gasoline fuels and bulk additives will be registrable.\8\ On the other hand, an unregistered chromium-containing additive (either bulk or aftermarket) intended for use in gasoline will be considered ``new'' rather than ``registrable,'' because there are no currently registered chromium-containing fuels or bulk additives in the gasoline fuel family with which the applicant additive could be grouped. Even if a chromium-containing product had previously been registered as an aftermarket additive for gasoline [e.g., as a ``grandfathered'' product registered prior to the ban of such aftermarket additives under CAA section 211(f)(1)(B)]\9\ or as a bulk additive for use in another fuel family (e.g., diesel fuel), the applicant additive would still be considered ``new''.

\8\The grouping criteria for the baseline gasoline category are described in Section IV.B.2.a.

\9\Until the 1990 CAA Amendments went into effect, the statutory language of section 211(f) was interpreted as applying only to unleaded gasoline fuels and related bulk additives. Thus, prior to November 15, 1990 (the effective date of the CAA Amendments), aftermarket additives intended for use in unleaded gasoline and containing elements in addition to carbon, hydrogen, oxygen, nitrogen, and/or sulfur were allowed to be registered. Under the 1990 CAA Amendments, all types of motor vehicle F/FAs were placed under section 211(f) jurisdiction. [However, section 211(f) provisions do not apply until certification requirements are promulgated for the specific motor vehicle fuel or fuel additive.] All aftermarket additives that were not ``substantially similar'' and were introduced on or after November 15, 1990 were banned. However, this ban did not apply retroactively. Thus, ``non-

substantially similar" gasoline aftermarket additives which had been registered prior to November 15, 1990 have been allowed to retain their registrations. These are so-called ``grandfathered" aftermarket additives.

As discussed in the Reopening Notice, EPA believes that the distinctions between registrable and new F/FAs, both in terms of their definitions and their respective compliance requirements, reflect reasonable regard for the public health and welfare without undue interference in the F/FA marketplace. Because registrable F/FAs are defined such that they must be reasonably similar in composition and usage to current F/FAs, their entry into the market will generally not be expected to increase the health or welfare risks potentially related to current F/FA emission exposures, assuming the overall rate of usage does not increase substantially.

Today's rule implements EPA's policy that F/FAs that may pose new or different health risks to the public should not be allowed on the market until EPA has determined that adequate health testing has been conducted. Because it would cause significant hardships to pull all currently used products off the market until they were tested, products that are already registered may continue to be sold. If these principles were strictly applied (i.e., EPA refused to register any specific product that does not currently have a registration), there could be significant stagnation in the marketplace--a new company that wanted to sell the same unleaded gasoline that everyone else is selling would be prevented from getting a registration until it had tested its gasoline product. Thus, today's rule allows new registrants to sell products that are similar to registered products in terms of: (1) Expected health effects; and (2) usage (and, therefore, type or extent of exposure) currently allowed by law.

If an unregistered product can group with a registered product, EPA has determined that the products should have similar health effects. To ensure that usage (and, therefore, type or extent of exposure) is similar to a registered product, under today's rule, a manufacturer cannot rely on registration of an aftermarket fuel additive or on a F/FA in a different fuel family. Considering a fuel or bulk additive to be registrable based on an aftermarket additive registration could significantly increase the public exposure to that F/FA. Generally, aftermarket additives are relatively limited in distribution and usage and, therefore, in exposure. Thus, in seeking a registration for a new product, a manufacturer cannot rely on the registration of aftermarket additives or any F/FA product in a fuel family other than the one for which the registration is sought.

This is consistent with Congress' intent in CAA section

211(f)(1)(B) to preclude introduction into commerce of new aftermarket additives which do not fit the ``substantially similar'' criteria.\10\<SUP>, \11\ Looking only to F/FAs in the same fuel family to determine registrability is also intended to prevent potential increases in exposure to untested products. Expanding the use of an additive from one fuel family to another (e.g., from diesel fuel to gasoline) would significantly increase the overall size of the potential market for the product and thus the potential exposure to its emissions.

\10\See memorandum from James W. Caldwell to Mary T. Smith regarding ``Review of Notifications Submitted Pursuant to 40 CFR 79 for Compliance with the `Substantially Similar' Rule for Unleaded Gasoline,'' available in Docket A-90-07, Item No. IV-B-07.

\11\The grouping criteria in this final rule (see Section IV) allow aftermarket additives to join the baseline group if they contain no elements in addition to carbon, hydrogen, oxygen, nitrogen, and/or sulfur, even if they may sometimes be used by consumers in an amount greater than the gasoline ``substantially similar'' restriction of 2500 ppm.

EPA interprets section 211(e) to support the distinction between registered or registrable F/FAs and new F/FAs. EPA believes that the reference in CAA section 211(e)(2) (A) and (B) to a ``fuel or fuel additive which is registered'' or ``which is not registered'' is ambiguous as to whether it refers to the F/FA product generally or to a particular product-specific registration. Given this ambiguity, EPA believes that it is reasonable to interpret the phrase ``fuel or fuel additive which is registered'' to refer to the F/FAs generally. A contrary interpretation would result in EPA allowing numerous types of unleaded gasoline to be sold under existing registrations, while at the same time denying a registration (until completion of necessary testing) to a new company that wanted to sell the same type of unleaded gasoline that many others now sell. As discussed above, for ``registrable'' products, such an interference in the market would not likely result in any public health benefit. Therefore, EPA believes it is reasonable to interpret the phrase ``registered'' to include both F/FAs that are either ``registered'' or ``registrable.'' ``Registrable'' F/FAs are sufficiently similar in composition and use to existing F/FAs that one would not expect them to have dissimilar health effects; and therefore, it is reasonable to interpret the phrase ``registered fuel or fuel additive'' to include not only those F/FAs that are identical, but also those that can group with existing F/FAs.

Alternatively, EPA believes that the above approach is consistent with section 211 because EPA interprets section 211(e)(3) (A) and (B) in conjunction with section 211(e)(3)(C), which gives EPA authority to exempt any F/FA product from duplicative testing. Thus, even if one interprets the phrase ``fuel or fuel additive which [is/is not] registered'' to mean either that an identical F/FA product must already have a registration, or to refer to a product-specific registration, EPA believes it is reasonable to interpret section 211(e)(3)(C) to allow F/FAs that are similar in composition and usage to those already on the market to group with those similar F/FAs and complete the testing with the other F/FAs in their group. At the same time, EPA believes that for F/FAs that differ significantly in composition or usage from currently registered F/FAs, such testing would not be duplicative of testing of registered F/FAs; and therefore, EPA is authorized under section 211(e) to require this information prior to registration. Under either theory, EPA's authority to obtain information prior to registration is not limited to Tier 1 and Tier 2 data, because section 211(b)(2)(B) gives EPA authority to require for registration any information necessary to assess the effects of emissions on public health or welfare. Therefore, EPA interprets section 211 (e) and (b) to give it the authority to require any necessary health or welfare effects information for F/FAs that are significantly different in composition or usage from currently registered products.

d. Changes to 40 CFR Part 79. This final rule includes revisions to the current 40 CFR part 79 registration regulations which are necessary to properly implement the new testing requirements in Subpart F. These consist of various conforming changes in registration procedures, requirements, and terminology.

The submission by which a manufacturer requests registration of a fuel or fuel additive product is now called an ``application'' rather than a ``notification,'' in order to better reflect the additional submissions and requirements upon which registration is now contingent, and to avoid confusion with the various notifications concerning testing requirements which will be transmitted to applicants and registrants by EPA. Conforming changes have been made in procedures for notifying an applicant that a submission does not comply with registration requirements and for granting registration.

As discussed in Section IV of this preamble, in order to determine in which of the F/FA group(s) established under the criteria set forth in this rule (see Sec. 79.56) a particular fuel additive should be enrolled, and to administer applicable testing requirements separately for each such F/FA group, it is essential that fuel additives be deemed to be registered only for those specific types of fuel for which they will be sold and used. The Agency interprets and is already

administering the existing part 79 in this manner. However, in the conforming changes included in this rule, part 79 has been clarified to confirm that registration of fuel additives is fuel specific, and that EPA considers sale or distribution of a fuel additive product for use in a fuel for which it is not registered to be unlawful.

In addition, a manufacturer seeking to register a fuel product or a fuel additive product for use in vehicles manufactured after 1974 must demonstrate that it is ``substantially similar'' to fuels or additives utilized in the certification of vehicles for 1975 or subsequent model years, or that the manufacturer has obtained a waiver under CAA section 211(f)(4). This expressly codifies the manner in which EPA has administered the registration program since it adopted criteria implementing the statutory prohibition in CAA section 211(f)(1) on introduction into commerce of fuels and fuel additives which are not ``substantially similar.''

2. Program Focus On Emissions

CAA section 211 gives the Agency discretion to determine the focus of the F/FA testing program under CAA section 211(b). EPA is exercising its discretion by focusing this rule on the testing of emissions, because the main purpose of the testing program is to provide EPA with information that can be used in regulatory decision-making under CAA section 211(c).

Section 211(c) of the CAA gives EPA the authority to regulate F/FAs based on the impact of their emissions on public health or welfare. Specifically, it allows the Administrator to control or prohibit the manufacture, introduction into commerce, or sale of any fuel or fuel additive whose emission product(s) cause or contribute to harmful air pollution. The legislative history of the provision also supports a focus on emissions, since House and Senate Reports on the CAA Amendments of 1970 link the information to be obtained under CAA section 211(b) to EPA's authority to regulate emissions under CAA section 211(c).\12\ Thus in the NPRM, EPA proposed to focus this rule's requirements on the potential emissions-based effects of F/FAs rather than on the effects of the raw (i.e., uncombusted) F/FA product. Public comment received after publication of the NPRM generally supported the proposed emissions-based focus of the rule. Accordingly, EPA has retained this focus in today's action. The health effects testing requirements of this final rule specifically address the effects of inhalation exposure to F/FA combustion and evaporative emissions. The required testing focuses on the evaluation of health effects of the whole emissions of the fuel or additive/base fuel mixture of interest and not on the toxicity of the individual emission products.

\12\H. Rep. No. 1146, 91st Cong. 2nd Sess. (1980) at 13,

reprinted in Environment and Natural Resources Division of the Library of Congress, 93rd Cong., 2nd Sess.; A Legislative History of the Clean Air Act Amendments of 1970 (Comm. Print 1974) (``Leg. Hist." at 433-434).

For the purposes of this rule, combustion emissions are the primary exhaust products of the combustion of a fuel or additive/base fuel mixture in a motor vehicle engine and do not include secondary atmospheric transformation products. EPA recognizes that secondary air pollutants are a factor in the characterization of overall risks associated with F/FA emissions. However, it is not feasible to include this type of laboratory testing as a standard requirement at this time. When required in specific instances, transformation products will be addressed under Tier 3, as described in Section III.C.

As proposed, evaporative emission testing is to be performed for F/FAs meeting specific volatility criteria. While some commenters asked EPA to eliminate the evaporative emission testing from the program, today's rule maintains this requirement. EPA's decision is supported by the legislative history, which expresses the concerns of Congress about the public health impacts of emissions from both combustion and evaporative sources. Public exposure to evaporative emissions is still significant and, for many F/FAs, the toxicity of evaporative emissions as a whole mixture has not been characterized. Thus, this rule includes requirements for the characterization and biological testing of evaporative emissions in certain circumstances.

While combustion emissions are inevitable products of the engine combustion process, the significance of evaporative emissions depends on the type of F/FA product. As proposed in the NPRM, this final rule specifies criteria for determining the need for evaporative emission testing. For fuels that are supplied to motor vehicle engines by way of sealed containment and delivery systems, evaporative emissions testing is less important, since human and environmental exposure should be extremely low or nonexistent. Thus, evaporative emissions testing under this final rule would not apply to methane (compressed natural gas or liquified natural gas) or propane (liquified petroleum gas) formulations.

For liquid F/FAs, the significance of vaporization varies widely, depending largely on the volatility of the fuel or additive/base fuel mixture. Thus, this final rule uses the Reid Vapor Pressure (RVP) of a fuel or additive/base fuel mixture to determine its applicability for evaporative emissions testing. An RVP of 2.0 pounds per square inch (psi) is designated as the threshold for determining the need for evaporative emission testing for fuels. That is, fuels with RVP of 2.0 psi or greater are subject to the evaporative emissions testing

requirements, while those with RVP less than 2.0 psi are excused from the evaporative emission testing requirements under Tier 1 and Tier 2.

With respect to additives, the NPRM proposed to require evaporative emission testing if the RVP of the additive/base fuel mixture was increased by 0.1 psi or more in comparison with the RVP of the base fuel alone. However, methods for measurement of vapor pressure have a reproducibility of about 0.3 psi.\13\ To account for this limitation of measurement accuracy, especially when dealing with low pressure measurements, today's rule uses a 0.4 psi criterion (i.e., 0.1 <plus-minus>0.3) for additives. Accordingly, this final rule requires the evaporative emission testing of additives when the RVP of the associated fuel in the additive/base fuel mixture is increased by 0.4 psi and the resulting RVP of the additive/base fuel mixture is 2.0 psi or more.\14\ For example, an additive that causes an increase of 0.6 psi when mixed with a fuel with a vapor pressure of 1.0 psi (i.e., the resulting RVP of the additive/base fuel mixture is 1.6), need not be tested for evaporative emissions. On the other hand, an additive that causes an increase of 1.1 psi when mixed with a fuel with RVP of 1.0 psi is required to undergo evaporative emission testing because the resulting RVP of the additive/base fuel mixture is 2.1 psi.

\13\See ``Standard Test Method for Vapor Pressure of Petroleum Products (Mini Method)," ASTM D 5191-91.

\14\The requirement to test the evaporative emissions of a qualifying additive product do not apply if the manufacturer intends to satisfy the test requirements of the additive as part of a group, of which another member product or a base fuel serves as the group representative, and the manufacturer does not specifically test the additive apart from the group. See Section IV for a discussion on grouping provisions.

The above defined thresholds are used by EPA in determining the applicability of evaporative emission testing for the purposes of Tier 1 and Tier 2. However, EPA retains the authority to require evaporative emission testing under Tier 3 for fuels or additive/base fuel mixtures with low vapor pressure, e.g., RVP less than 2.0, if there is a health or welfare concern associated with the evaporative emissions of the fuel or additive/base fuel mixture in question. For example, if a highly toxic substance is present in a fuel or additive/base fuel mixture, EPA could require evaporative emission testing under Tier 3, even if the RVP of the F/FA product in question is below 2.0 psi. These special cases will be handled on a case-by-case basis under Tier 3.

3. Program Applicability

The requirements of this rule apply to manufacturers (including importers) of designated fuels or fuel additives and to any F/FA manufacturer [see Sec. 79.2(d) and (f) as amended in this rule] seeking registration under CAA section 211(a) and 211(b). A fuel is defined to be any material which is capable of releasing energy or power by combustion or other chemical or physical reaction [see 40 CFR Sec. 79.2(c)]. A fuel additive is defined as any substance that is intentionally added to a fuel (including any added to a motor vehicle's fuel system) and that is not intentionally removed prior to sale or use (see 40 CFR Sec. 79.2(e), as amended in this rule), including both bulk and aftermarket additives.

At the present time, the designation of F/FAs encompasses both leaded and unleaded gasoline F/FAs and diesel F/FAs produced and commercially distributed for use in motor vehicles. F/FAs intended only for off-road use (e.g., farm and construction equipment, aircraft, boats, railroad engines) are not currently designated to be registered, and thus are not subject to the requirements of today's rule. However, if off-road F/FAs become designated in the future (according to provisions under CAA section 213), this rule will be modified, as needed, to cover them as well.

While the designated F/FAs include leaded gasoline formulations, CAA section 211(n) provides that ``after December 31, 1995 it shall be unlawful for any person to sell, offer for sale, supply, offer for supply, dispense, transport, or introduce into commerce, for use as fuel in any motor vehicle (as defined in section 7554(a) of this title) any gasoline which contains lead or lead additives." Because of the upcoming ban of leaded F/FAs, compliance with the requirements in today's rule will be superfluous for manufacturers of leaded products. Thus, the leaded fuel family has been deleted from this final rule.

While alternative fuels and their additives are currently on the market, they are not yet designated and thus not yet required to be registered. However, because they are currently used and EPA contemplates their future designation, this final rule includes provisions for their registration and testing. The alternative fuels for which provisions are included are: methanol, ethanol, compressed natural gas (CNG), liquified natural gas (LNG), and liquified petroleum gas (LPG). EPA is currently developing the proposal for the designation of these alternative fuels and their additives. In that proposal, EPA currently intends to propose to set an effective date for the final designation rule far enough in the future so that these F/FAs could complete whatever pre-registration testing would be required prior to the time they would be required to be registered. EPA currently believes the delay of the effective date of the designation rule would be justified by the need to minimize disruptions in an existing market for alternative F/FAs.

This rule contains a number of special provisions which reduce or modify the program's requirements for certain manufacturers or certain classes of products. Such special provisions apply to small businesses (as defined in this rule) and manufacturers of experimental F/FAs, relabeled products, and aerosols. These provisions are described in Section X of this preamble.

B. Health Evaluation Requirements

The testing program established in this rule focuses on the identification and evaluation of potential adverse health effects associated with inhalation exposure to F/FA emissions. The Tier 2 testing program of this rule addresses, in addition to the areas of inquiry mandated by the statute (carcinogenicity, teratogenicity, and mutagenicity), specific assessments designed to detect potential pulmonary, neurotoxic, and general reproductive effects of F/FA emissions.

In the NPRM, short-term (42-day) tests were proposed under Tier 2 to address each of the health effect endpoints described above. However, in response to public commentary and EPA's own analysis, the Tier 2 testing requirements have been modified in this final rule to enhance the efficiency and feasibility of the program. Today's rule uses a comprehensive 90-day subchronic inhalation protocol and ancillary tests to examine general systemic and organ toxicity (including pulmonary toxicity), as well as the specific areas of concern described above.

These evaluations require the exposure of laboratory animals to the whole emissions of F/FAs. Tier 2 tests are to be conducted for both combustion and (when applicable) evaporative emissions. The subchronic inhalation protocol allows the examination of specific endpoints within the 90-day testing framework. For example, pulmonary and neurotoxic effects are examined in conjunction with the subchronic study standard histopathological requirements. The neurotoxicity assessment also includes a biochemical assay to measure the level of glial fibrillary acidic protein (GFAP). Coordinated with the 90-day study is a battery of three assays used in the evaluation of carcinogenicity and mutagenicity: the in vitro Salmonella assay, the in vivo micronucleus assay, and the in vivo sister chromatid exchange assay. A fertility assessment that looks at both reproductive and teratogenic effects is also coordinated with the general toxicity study. The assessment for reproductive effects involves the mating of exposed animals, the measurement of reproductive cycles, and the histopathology of male and female reproductive organs. The teratogenic assessment requires the exposure of pregnant females to F/FA emissions and the subsequent examination of the uterus and its contents just prior to the normal

time of parturition.

In addition to the evaluation of the health effects described above, EPA retains the authority under Tier 3 to require additional testing on a case-by-case basis on those endpoints evaluated under Tier 2 and/or on other endpoints of concern. Further discussion about the specific requirements of Tiers 1, 2, and 3 is provided in Sections VII-IX.

C. Welfare Evaluation Requirements

CAA section 211(b)(2)(B) states that the Administrator may require manufacturers to furnish ``reasonable and necessary'' information for determining ``the extent to which F/FA emissions affect the public health or welfare''. The term ``welfare effects'' encompasses a variety of complex and interrelated factors. In terms of motor vehicle F/FA emissions, welfare effects could include the impact of air pollution on the public health and the environment, including a broad range of effects on aquatic and terrestrial ecosystems, cultivated crops and other vegetation, natural and man-made materials, wildlife, and stratospheric ozone. Air pollution effects on the public welfare also include important environmental concerns such as noxious odors or visibility impairment, which may detract from human well-being.

Except for stipulating that welfare effects should be addressed, the statute gives EPA broad discretion about how to address welfare effects. EPA recognizes that, at the present time, scientific experience and laboratory screening methods for the evaluation of welfare effects are more limited than in the area of health effects. Thus, today's rule limits the routinely required welfare evaluation to requirements that are coincident or concurrent with the evaluation of health effects. These include the literature search, emission characterization, and exposure analysis requirements of Tier 1. While at this time EPA is not requiring biological testing for welfare effects, the Administrator retains the authority to require additional evaluation and/or testing of welfare effects at the Tier 3 level, when the outcome of lower tiers demonstrates both significant environmental toxicity and exposure potential. EPA will determine the need for Tier 3 welfare effects testing on a case-by-case basis.

In the NPRM, EPA proposed to require modeling analyses for atmospheric reactivity, environmental fate/partitioning, and exposure as part of the welfare evaluation. EPA requested comments on the feasibility of requiring such modeling analyses as a routine requirement for registration. Commenters urged EPA to limit the modeling requirements due to the lack of standardized methods in this area of study. Recognizing the limitations of modeling methods and the availability of existing data for some of the areas of study of

interest in this rule, this final rule does not require modeling analyses as part of Tier 1.

EPA recognizes that other EPA programs are actively researching and controlling mobile and stationary source contributions to major air pollution problems such as tropospheric/stratospheric ozone, global warming, and acid rain. Furthermore, modeled ozone reactivity data are already available for most conventional and alternative fuels. EPA believes that if additional modeling is deemed necessary, this could be performed by manufacturers under Tier 3. Also, EPA may conduct simple modeling, using the emission data submitted by registrants under Tier 1, if needed for regulatory decisions.

Regarding environmental fate and exposure modeling, EPA recognizes that these types of analyses will be extremely difficult due to the complex nature of F/FA emissions. Because both environmental partitioning models and exposure models address single compounds rather than mixtures, it would be unduly burdensome and unreasonable to require all registrants to perform these analyses on each individual emission constituent. Requiring the modeling of each individual emission product would also result in duplication of information and, therefore, would be inconsistent with the original intent of the statute.

In addition, the environmental models are applicable only to a limited number of emission products for which appropriate physical/chemical data are available in order to perform the analysis. In terms of available exposure models, these usually rely on carbon monoxide monitoring data and related emission rates to estimate potential exposures. This means that the applicability of available exposure models is somewhat limited to the analysis of compounds whose chemical/physical behavior is similar to carbon monoxide.

Based on the above factors, EPA believes that quantitative evaluations of potential exposures and environmental fate/partitioning of F/FA emissions will be better addressed at the Tier 3 level on a case-by-case basis, where they can be focused on specific compounds of potential environmental concern.

D. Requirements for Emission Control System Testing

CAA section 211(b)(2) requires F/FA manufacturers to provide information to determine their products' effects on ECS performance. The NPRM stated that EPA intended to continue addressing ECS performance through the existing waiver application program under CAA section 211(f). The waiver program prevents the introduction into commerce of F/FAs which would significantly degrade the performance of emission control equipment. Under CAA section 211(f), F/FA formulations which do not meet specific chemical and physical criteria considered to

be ``substantially similar'' to EPA certification fuel (see interpretive rule in 56 FR 5352), cannot be introduced into commerce unless a waiver is issued by EPA. The waiver process then requires the applicant to demonstrate, through testing if necessary, that ``such fuel or fuel additive or a specified concentration thereof, and the emission products of such fuel or fuel additive or a specified concentration thereof, will not cause or contribute to a failure of any emission control device or system (over the useful life of any vehicle in which such device or system is used) to achieve compliance by the vehicle with the emission standards to which it has been certified.'' For products already registered that do not meet ``substantially similar'' criteria, i.e., grandfathered products that were registered prior to the implementation of the waiver application program, EPA proposed to establish a mechanism that would permit the public to submit petitions to EPA requesting ECS testing for a particular fuel or fuel additive of concern.

Today's rule reflects EPA's judgment that the mechanisms already established under CAA section 211(f) are adequate for the ECS testing of F/FAs. EPA's previous experience with the waiver application process has demonstrated the practical value of the ``substantially similar'' concept for determining whether a F/FA product needs to be tested for its effects on emission control equipment. EPA is not aware of instances in which products meeting ``substantially similar'' criteria were later discovered to have adverse effects on vehicular emission control performance. The implementation of another ECS testing program under section 211(b) would be duplicative and, therefore, inconsistent with Congress' intent. Thus, as proposed, today's rule refers to the waiver application process under CAA section 211(f)\15\ for the ECS testing of ``substantially similar'' F/FAs and F/FAs required to obtain a waiver under CAA section 211(f)(4). Products which conform to applicable ``substantially similar'' criteria are not required to undergo ECS testing before they can be registered. On the other hand, new F/FAs which do not meet ``substantially similar'' criteria are subject to the standard 211(f) application process prior to registration.

\15\An example of a waiver decision can be found in 53 FR 33846.

EPA recognizes that there are grandfathered F/FA products (see Section III.A above) which fall outside the regulatory domain of CAA section 211(f). These grandfathered products include gasoline aftermarket additives introduced prior to the 1990 CAA Amendments. Therefore, statutory authority for the ECS evaluation and regulatory

control of grandfathered products exists under CAA sections 211 (b) and (c) rather than section 211(f). EPA judges that requiring ECS evaluation of all grandfathered products, without evidence of ECS problems, would be unreasonable and unnecessarily burdensome on the industry. Instead, today's rule provides a petition mechanism for the ECS evaluation of grandfathered products. Under this mechanism, EPA could require ECS testing of grandfathered products, similar to the testing which a waiver applicant would generally conduct, if so petitioned by outside parties or if other information available to the Agency indicates that such evaluation is appropriate. Such information might be obtained as a result of the emission characterization requirements included in this final rule. In addition, vehicle manufacturers or other outside parties are allowed to submit petitions to EPA requesting the testing of grandfathered products based on evidence of potential harm to vehicular ECS. If EPA judges that ECS testing is warranted after reviewing the petition arguments, emission characterization results and/or other available information, the authority provided by CAA sections 211(b) and 211(c) to require specific grandfathered products to test for ECS effects.

IV. Grouping System

A. Objectives and Rationale

CAA section 211(e) provides a number of mechanisms by which EPA may reduce the costs and burdens of compliance with the registration requirements set forth in CAA section 211(b). In particular, CAA section 211(e)(3)(B) permits the Administrator to ``provide for cost-sharing with respect to the testing of any fuel or fuel additive which is manufactured or processed by two or more persons, or otherwise provide for shared responsibility'' so that the program requirements can be met without duplication of effort. In accordance with this provision, today's rule maintains the grouping system proposed in the NPRM, which permits manufacturers of similar F/FAs, on a voluntary basis, to pool their resources and efforts to satisfy the registration requirements. The groups defined by the specifications in this final rule are the only groups permitted for satisfying the requirements of the registration program.

As proposed in the NPRM, the grouping system allows similar fuels and additives to be grouped together, rather than creating separate fuel groups and additive groups. This convention recognizes that, to meet the requirements of this final rule, an additive must be mixed with its associated base fuel\16\ prior to generating the emissions for testing. To the extent that the resulting additive/base fuel mixture is similar to existing fuel formulations, the tests conducted on the

emissions of the additive/base fuel mixture will be duplicative of tests conducted on the related fuels. To avoid potential duplication, this final rule maintains the proposed approach, in which closely-related fuels and additives are grouped together. Accordingly, the manufacturers of fuels and the related additives can fulfill their individual registration responsibilities through jointly-supported testing rather than through duplicative independent efforts. By grouping similar fuels and additives together, the grouping scheme also avoids the need to define each generic product or product component as either a ``fuel'' or an ``additive.'' This would otherwise present a problem when a given substance (or mixture) can serve as either a fuel or an additive (e.g., ethanol).

\16\Base fuel specifications for each fuel family are described in Section V.

In the NPRM, EPA developed criteria for sorting individual F/FAs into groups of related formulations based on similarities in the chemical/physical properties of the ``raw'' fuel or additive/base fuel mixture. EPA has maintained this approach in the final rule. EPA expects F/FAs within each group to have similar emission characteristics and thus essentially the same general effects on the public health and welfare. Therefore, chemical or toxicologic information associated with individual members of a given group can reasonably be generalized to all F/FAs in the group. EPA will consider tests performed on a selected representative of a group to apply to all members of the group for purposes of compliance with registration requirements, for deciding whether to require additional testing under Tier 3, or for taking regulatory action under CAA section 211(c).

While each manufacturer of a fuel or fuel additive will still be held individually accountable for compliance with the registration program, the grouping system provides an opportunity for meeting the program requirements in a more cost-effective manner. Participation in the F/FA grouping system is strictly voluntary, and any manufacturer may choose to fulfill the requirements on an individual basis. Those who choose to take advantage of the grouping opportunity will be able to share their planning efforts, research capabilities, and financial resources to satisfy the information-gathering and testing requirements of the F/FA registration program. To satisfy the chemical and biological testing requirements, the required tests will be done on the selected representative for the respective group, rather than being repeated for each of the F/FAs in the group. The results of the tests on the group representative will then be submitted jointly for all

members of the group, with applicable costs to be shared by the respective manufacturers (based on their cost-sharing agreements, as discussed in Section IV.C). Manufacturers who question whether the results obtained for their group's representative are valid for their own products may conduct confirmatory tests on their products on an independent basis and at their own cost. However, until such independent test results are made available to EPA, the original results submitted on behalf of the group will be considered valid for all member products, and could be applied by EPA to support regulatory decisions under CAA section 211(c) or requirements for further testing under CAA section 211(b).

The F/FA grouping system is expected to provide a number of benefits to the F/FA manufacturers who are responsible for registration while increasing the efficiency and functionality of the registration program itself. First, the grouping system will reduce the overall costs of the registration program by avoiding the generation and submission of essentially redundant information by individual manufacturers with similar products. In addition, by reducing the number of individual formulations that will be subject to testing, the grouping system is expected to ease the pressure and demands on limited laboratory capacity.

B. Grouping Approach and Criteria

The basic conceptual framework for the grouping system is illustrated in Figure 3. First, each fuel or additive is sorted into one of six broad ``fuel families.'' F/FAs in each fuel family are then subdivided into three ``F/FA categories.'' The categories are further subdivided into ``F/FA groups''--the ``working'' units of the grouping system. It is among the members of the F/FA groups that cooperative evaluation and testing efforts can be pursued using designated group representatives. This grouping system is very similar to the approach that was proposed in the NPRM, with the exception that the original proposal has been simplified in today's rule by eliminating the separate concept of ``formulation class.'' EPA judged that the ``formulation class'' concept could be confusing, and was not necessary for the structure or implementation of the grouping system. The key parameters and relationships within this grouping framework are further explained in the following sections. A summary of the grouping system is provided in Table F94-7 (see Sec. 79.56) of the accompanying regulatory text for this rule.

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Generic rules for categorization and grouping are used to determine specific F/FA groups based on the raw composition of the particular products under consideration. The first step entails the selection of the applicable fuel family and category for the product according to the criteria discussed below. Rules specific to the categories then define the proper F/FA group. After the group is formed and arrangements are made for cooperative testing efforts, applicable criteria will be applied to select a representative of the group to be used in group-sponsored testing. In determining the category and group to which a fuel or fuel additive belongs, impurities present in trace amounts can be ignored unless otherwise restricted in the definition of the particular fuel family. Impurities in fuels or fuel additives are substances which are present through contamination, or remain naturally, after processing is completed.

1. Fuel Families

This final rule defines six fuel families, as follows: (1) Gasoline (containing more than 50 percent gasoline by volume), (2) diesel (containing more than 50 percent diesel by volume; includes both diesel #1 and diesel #2 formulations), (3) methanol (containing at least 50 percent methanol by volume), (4) ethanol (containing at least 50 percent ethanol by volume), (5) methane (includes compressed natural gas and liquified natural gas containing at least 50 mole percent methane), and (6) propane (i.e., liquid petroleum gas containing at least 50 percent propane by volume). A manufacturer seeking to register a formulation which does not fit the criteria for inclusion in any of the above fuel families shall contact EPA for guidance in classifying and testing such formulation.

In the NPRM, EPA proposed to establish two gasoline fuel families: unleaded and leaded. As explained previously, EPA is not including a leaded fuel family in this final rule due to the upcoming ban of leaded F/FAs under CAA section 211(n). Thus, the unleaded fuel family has been renamed to become simply the ``gasoline'' fuel family in this final rule.

Fuel families consist of a constellation of F/FA products sharing basic characteristics in regard to their chemical/physical properties and engine/vehicle applicability. For ease of reference, the gasoline and diesel families are regarded as the ``conventional'' fuel families, while the remaining four are referred to as the ``alternative'' fuel families. If a manufacturer develops a F/FA product that does not meet the definition of any of the fuel families included in this rule, EPA will define additional fuel families to include such a product.

Each fuel family includes not only the fuels referenced in the name of the family, but also bulk and aftermarket additives which are

intended for use in such fuels. Additives which are registered for use in more than one type of fuel are assigned to each applicable fuel family. For example, an additive product that is registered as both a gasoline additive and a diesel additive belongs to both the gasoline fuel family and the diesel fuel family. Furthermore, the manufacturer of such additive product will be required to test the additive in each applicable fuel family. The multiple testing responsibility can be satisfied by the manufacturer individually or by participating in the applicable groups for each fuel family. For instance, if an additive product meets the baseline criteria for both gasoline and diesel, then the product will be assigned to two groups, i.e., the gasoline baseline group and the diesel baseline group. The manufacturer of such additive product will then be able to share the costs of testing with other manufacturers of baseline gasoline F/FAs and baseline diesel F/FAs.

Today's rule specifies the chemical and physical characteristics of ``base fuel'' formulations for each defined fuel family. These are generic formulations (rather than any particular commercial fuel) with average or normative characteristics for a given fuel family. Once an additive has been assigned to the applicable fuel family or families, determination of the proper category and group (for each applicable fuel family) for the additive is based on the properties of the mixture that results when the additive is mixed in the base fuel of the applicable family at the maximum concentration recommended for use by the additive manufacturer. Moreover, it is this mixture that is used for the generation and testing of additive emissions.\17\ Tests conducted on the emissions of the base fuel then serve as one control (the other being a clean-air exposure group) against which tests on the emissions of the additive/base fuel mixture are to be compared. Further discussion on the definition and use of base fuels is presented in Section V.

\17\Special provsions related to the testing of additives are discussed in Section VI.F.

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2. F/FA Categories

Fuel families are subdivided into three F/FA categories: ``baseline,'' ``non-baseline,'' and ``atypical.'' The baseline category consists of fuels and associated fuel additives which resemble the respective base fuel for a particular fuel family in terms of elemental composition and which conform with certain quantitative limits for particular constituents. It is important to understand that a baseline category is not limited to base fuels; the baseline category and group criteria defined below for each fuel family are considerably less

restrictive than the respective base fuel definitions (specified in Section V). Non- baseline F/FAs contain no chemical elements other than those allowed in the baseline category, but they exceed the allowable baseline limit for certain constituents for the respective fuel family. The atypical category consists, in general, of F/FAs that contain chemical elements in addition to those allowed in the baseline category. (In a few circumstances, the atypical category also includes F/FAs that exceed specified baseline limits for certain constituents, as discussed below.) As mentioned above, the category determination for fuel additives is based on the properties of the mixture which results when the additive is mixed in the appropriate base fuel at the maximum concentration recommended for use by the additive manufacturer. If the fuel or additive/base fuel mixture contains both non-baseline and atypical constituents, the formulation is characterized as atypical. Thus, atypical constituents take precedence over non-baseline constituents.

In establishing the F/FA categories (and the groups within them), EPA has sought to avoid overly narrow definitions which would result in unnecessary and duplicative testing by manufacturers, as well as overly broad definitions which would cause potentially important toxicologic differences between F/FAs to be obscured. A basic guideline EPA has used to find the proper balance between these two unsatisfactory scenarios is to ask whether the emissions of a single group representative (i.e., test substance) can reasonably be expected to reflect the chemical and toxicologic properties of the emissions of the F/FAs proposed to be classified together. In applying this guideline, EPA has kept in mind both the homogenizing effect of combustion processes, as well as the need in some cases to examine the effects of evaporative emissions, which generally retain the characteristics of the raw F/FA to a greater degree.

These considerations have led EPA to depend on the chemical elements in F/FA formulations as a primary criterion for categorization and grouping. Another key criterion is the presence of significant amounts (by volume, weight, or potential potency) of F/FA constituents that are likely to change the characteristics of the combustion or evaporative emissions in substantive ways.

Accordingly, the baseline category in each fuel family is generally comprised of F/FAs containing no elements in addition to carbon, hydrogen, oxygen, nitrogen, and/or sulfur.\18\ These elements are the fundamental chemical building blocks of all of the fuel families defined in this rule. Omitting any of these elements from the list of permissible baseline elements would eliminate all or most F/FAs from the baseline categories. On the other hand, allowing additional elements in the baseline definition would introduce substances not characteristic of most F/FA products in the fuel family. If a group

representative (test substance) did not contain the additional element, it could not reasonably be assumed to reflect the presence and activity of F/FAs that do contain the element. Conversely, if the group representative did contain the additional element, then the results of the testing would be influenced by the presence and activity of this element, and would therefore not be valid for the large majority of the baseline F/FAs. Thus, allowing F/FAs with additional elements to be included in the baseline categories (and groups) would violate the principles of the grouping system and the objectives of the testing program.

\18\The exceptions to this generalization (discussed in later sections) are small amounts of chlorine permitted in baseline methanol F/FAs, and small amounts of chlorine and copper permitted in baseline ethanol F/FAs. Also, trace contamination with elements other than carbon, hydrogen, oxygen, nitrogen, and sulfur do not cause F/FAs to be eliminated from baseline categories or groups.

F/FA formulations containing elements in addition to those allowed in the baseline category of a fuel family are classified in the atypical category for that family. As described further below, such F/FAs are then subdivided into groups based primarily on which atypical element(s) they contain. Moreover, the rules for choosing representatives of these atypical groups emphasize the atypical constituent(s). This approach assures separate testing of F/FA products with unique compositional characteristics that can reasonably be expected to appear in the emissions and may thus have distinct emissions-based toxicologic effects. EPA believes that this approach best effectuates CAA section 211(e) by avoiding duplicative testing of similar F/FA products while requiring ``each'' F/FA to be tested.

Between the baseline category and the atypical category in each fuel family is the non-baseline category. Broad generalizations about the non-baseline F/FA categories are somewhat more difficult to make, since they are distinguished from the respective baseline categories by various attributes other than elemental composition. In the case of gasoline and diesel F/FAs, the distinction is based primarily on the presence of significant concentrations of oxygenating compounds. As discussed further below, the presence of such compounds may have a large impact on F/FA emission profiles. Classification of the oxygenated F/FAs into separate categories from the baseline F/FAs (and further subdivision into separate groups) is necessary to assure testing of representatives that can reasonably reflect the differences in these emission mixtures and, possibly, their health effects. Similar

principles apply to the non-baseline categories in the alternative fuel families. In the case of alcohol fuels, non-baseline F/FAs are those which have a substantial non-alcohol and non-gasoline component in the formulation. Non-baseline propane and methane formulations are those containing significant amounts of substances other than propane and methane, respectively. In all of these cases, the non-baseline definitions serve to ensure that F/FAs with properties that are likely to result in significantly different emission profiles, with possibly different toxicologic effects, are not subsumed in the baseline category.

The following sections describe the criteria which determine F/FA categories for each fuel family. It should be noted that the criteria that define baseline F/FA products for each fuel family are not meant to be fuel specifications. The baseline criteria used for grouping purposes in this final rule consider the potential health implications of the composition of the fuel or additive/base fuel mixture and might differ from previously established commercial fuel specifications, such as those established by the American Society for Testing and Materials (ASTM), the California Air Resources Board (CARB), or federal ``substantially similar'' criteria.

a. Gasoline. EPA discussed in the NPRM two options (Option A and Option B) for distinguishing the baseline and non-baseline categories for the gasoline fuel family (see 57 FR 13187-13188). Today's action uses Option A for the classification of gasoline products. A discussion supporting this decision is included in the ``Summary and Analysis of Comments for the Fuels and Fuel Additives Registration Regulations'' (available in public docket A-90-07). A synopsis of EPA's analysis follows.

The major differences between the two alternative grouping options considered by EPA in the NPRM were: (1) The cutoff point for oxygen content to distinguish between baseline and non-baseline products, and (2) the approach for handling F/FAs that have received a waiver under CAA section 211(f). Under Option A, the baseline category was to be limited to F/FAs having less than 1.5 weight percent oxygen by weight. Because the cut-off point of 1.5 percent is consistent with the minimum oxygen requirement for reformulated gasolines, reformulated gasolines would not be considered baseline under Option A. Instead, gasolines with 1.5 percent or more oxygen were to be sorted into different non-baseline groups depending on the oxygenated compounds they contained. In contrast, Option B used an oxygen cutoff point of 2.7 weight percent, based on current ``substantially similar'' criteria (see 56 FR 5352). F/FAs which exceeded this limit but had been granted a waiver for the excess oxygen were also to be designated as baseline.

After careful evaluation, EPA has selected Option A for the grouping of gasolines in this final rule. In arriving at this decision,

EPA considered the testing and potential health effects implications of both grouping options. The main purpose of the grouping system is to sort F/FAs based on the similarities of their emission components. After analyzing existing emission characterization data, EPA concluded that Option B did not provide an adequate approach for the testing of gasoline F/FAs because it does not distinguish between formulations that may have significantly different emission characteristics. EPA's evaluation showed that the emissions from oxygenated gasolines are not the same as for non-oxygenated gasolines and that, furthermore, the emission profiles differ according to the particular oxygenated compound present in a fuel formulation. Differences in emission species will affect the toxicological characteristics of the fuel or additive/base fuel mixture. Option B was found inappropriate because it would have allowed the grouping of gasolines containing significant amounts of different oxygenated compounds into one single group. These different compounds may have distinguishable toxic effects. Therefore, EPA selected Option A for the grouping of gasolines in this final rule in order to adequately examine the potential health effects of the different oxygenated compounds. EPA believes this best effectuates the goal of CAA Section(e) to require testing of each fuel or fuel additive.

In this final rule, gasoline formulations are defined as those containing more than 50 percent gasoline by volume. Based on current ``substantially similar" criteria (see interpretative rule at 56 FR 5352), the sulfur content for all gasoline formulations in the gasoline fuel family is limited to 0.1 percent by weight.

F/FAs in the baseline gasoline category must contain no elements in addition to carbon, hydrogen, nitrogen, oxygen, and/or sulfur. Gasoline baseline formulations must possess, at the time of manufacture, all the physical and chemical properties of an unleaded gasoline and applicable volatility class standards as specified in the latest version of ASTM standard for Automotive Spark-Ignition Engine Fuel, D 4814. As discussed above, the oxygen content of baseline gasolines must be less than 1.5 percent by weight. The baseline gasoline category includes all gasoline fuels and additives (evaluated as additive/base fuel mixtures) meeting the above criteria.

The non-baseline gasoline category is comprised of F/FAs which conform to the baseline specifications in terms of elemental composition, but exceed the specified baseline oxygen limit. Thus, this category includes gasoline formulations with no elements in addition to carbon, hydrogen, oxygen, nitrogen, and/or sulfur, which have been blended with oxygenates (i.e., alcohol, ether, ester, furan, and any other compound used to increase the oxygen content of the gasoline formulation), such that the total oxygen content of the gasoline-oxygenate blend is at least 1.5 weight percent. Included in the non-

baseline gasoline category are reformulated gasolines and oxygenated gasolines with at least 1.5 percent oxygen (by weight), including a number of formulations which have previously been granted CAA section 211(f) waivers on oxygen content.

The atypical category in the gasoline fuel family includes F/FAs which contain elements in addition to carbon, hydrogen, oxygen, nitrogen, and/or sulfur. (Trace contamination by other elements does not cause a F/FA to be classified as atypical, however.)

The baseline gasoline group is restricted to formulations that are derived from conventional petroleum sources. Thus, gasolines derived from synthetic crude oils are excluded from the baseline category. Synthetic crude oils can be prepared from coal, shale and tar sands, heavy oil deposits, and other non-conventional petroleum sources. Compared to petroleum, these synthetic crude oils must be extensively upgraded before they can be refined into useful products. Because of the nature of their sources, these synthetic products are likely to contain a variety of unknown contaminants with unknown health effects. With little specific data currently available on their composition, EPA believes that separate non-baseline classifications are most appropriate for grouping these products. Gasoline formulations derived from a particular synthetic crude oil source (e.g., coal) will be permitted to group together. The representative of each such group will be the first such product to seek registration.

b. Diesel. Diesel formulations are defined as those containing more than 50 percent diesel by volume. The sulfur content for all diesel formulations in the diesel fuel family is limited to 0.05 percent by weight, based on current EPA limits (55 FR 34120).

The diesel fuel family includes both diesel #1 and diesel #2 formulations.

As originally proposed, the diesel baseline category includes diesel formulations containing no elements in addition to carbon, hydrogen, oxygen, nitrogen, and/or sulfur. Baseline diesel formulations must also possess, at the time of manufacture, all the physical and chemical properties of a diesel fuel as specified in the latest version of ASTM standard D 975. Oxygen content of baseline diesel formulations must be less than 1.0 percent by weight. The baseline diesel category includes all diesel fuels that meet the above criteria.

The diesel baseline definition is consistent with existing information in EPA's F/FA registration data base, which indicates that most commercial diesel fuels, including their bulk additives, consist of carbon, hydrogen, oxygen, nitrogen, and/or sulfur. While some currently registered diesel fuels contain additives with additional elements, the objectives of the testing program are inconsistent with broadening the diesel baseline definition to include other elements with potentially different health effects from those of basic diesel

formulations. A broader baseline definition would mean that the atypical diesel F/FAs would not be separately examined. Limiting baseline diesel F/FAs to those containing no elements in addition to carbon, hydrogen, oxygen, nitrogen, and/or sulfur ensures the conduct of separate health effects evaluations for the emissions of diesel formulations containing atypical elements.

Similar to baseline gasoline, the baseline diesel category excludes fuels derived from synthetic crude oil sources. Thus, such formulations are included in the non-baseline category of the diesel family. The non-baseline diesel category also includes diesel formulations with 1.0 percent or more oxygen by weight. Examples of non-baseline diesel formulations are alcohol blends and biodiesel formulations.

c. Methanol. F/FAs in the methanol fuel family are defined as those containing at least 50 percent methanol by volume. The baseline methanol category is comprised of methanol and methanol-gasoline F/FAs that: (1) Contain at least 50 percent methanol by volume, (2) contain no more than 4 percent by volume of substances other than methanol and gasoline, and (3) contain no elements in addition to carbon, hydrogen, oxygen, nitrogen, sulfur, and/or chlorine. The sulfur content of baseline methanol formulations is limited to 0.004 percent by weight. Chlorine (as chloride) is limited to no more than 0.0001 percent by weight. Chlorine is allowed in methanol baseline formulations because it is a common contaminant remaining from methanol production.

The baseline methanol category includes all methanol fuels meeting the above criteria and is divided into two groups: M100 group and M85 group. The M100 group includes methanol-gasoline formulations containing at least 96 percent methanol by volume, while the M85 group consists of methanol formulations containing 50-95 percent methanol by volume.

F/FAs within the baseline M100 group are required to contain odorants and bitterants. These formulations should have a distinctive and noxious taste, for purposes of preventing purposeful or inadvertent human consumption. The elemental composition of the odorant and bitterant is limited to carbon, hydrogen, oxygen, nitrogen, sulfur, and chlorine. Baseline methanol formulations in the M85 group must comply with the elemental composition specified above for all baseline methanol F/FAs, but need not have added odorants and bitterants.

The non-baseline methanol category is comprised of methanol formulations (i.e., containing at least 50 percent methanol by volume) that meet the baseline limits on elemental composition, but contain more than 4 percent by volume of substances other than methanol and gasoline. a

Atypical methanol F/FAs contain elements in addition to those allowed in the baseline methanol category or exceed the specified limits for sulfur or chlorine.

d. Ethanol. Ethanol formulations in the ethanol fuel family are defined as those containing at least 50 percent ethanol by volume. The final rule defines a single group (represented by E85) for the baseline category of ethanol F/FAs. Although in the NPRM, EPA proposed two baseline groups for the ethanol fuel family (i.e., E100 and E85), EPA expressed its intention to establish a single group for baseline ethanol formulations in the Reopening Notice. As discussed in the Reopening Notice, the rationale behind this decision is that fuel ethanol is required to contain at least five percent denaturant, which means that, in actuality, E100 formulations contain only 95 percent ethanol (i.e., E95). Furthermore, gasoline is normally used as the denaturant for ethanol fuels. EPA judged that there was little incremental value in requiring tests of E95 in addition to E85. Thus, the final rule creates a single baseline ethanol group represented by E85. However, EPA retains the authority to require testing on other members of any F/FA group under Tier 3 (see Section IX.A).

The baseline ethanol category is comprised of ethanol and ethanol-gasoline F/FAs that: (1) Contain at least 50 percent ethanol by volume, (2) contain no more than 5 percent by volume of substances other than ethanol and gasoline, and (3) contain no elements in addition to carbon, hydrogen, oxygen, nitrogen, sulfur, chlorine, and/or copper. The sulfur content of ethanol baseline formulations is limited to 0.004 percent sulfur by mass. Chlorine (as chloride) and copper are allowed in the baseline ethanol formulations at a maximum level of 0.0004 percent by mass for chloride and 0.07 mg/l for copper. Chlorine and copper are permitted in the baseline ethanol formulations because they are common contaminants remaining from ethanol production. The baseline ethanol category includes all ethanol fuels meeting the above criteria.

The non-baseline ethanol category is comprised of ethanol formulations (i.e., containing at least 50 percent ethanol by volume) that meet the baseline limits on elemental composition, but contain more than 5 percent by volume of substances other than ethanol and gasoline.

Atypical ethanol F/FAs contain elements in addition to those specified in the baseline ethanol category or exceed the specified limits for sulfur, chlorine, or copper.

e. Methane. Methane F/FAs are defined as those containing at least 50 mole percent methane, including both compressed natural gas (CNG) and liquified natural gas (LNG). Baseline methane formulations must contain no elements in addition to carbon, hydrogen, oxygen, nitrogen, and/or sulfur, and must contain no more than 20 mole percent of non-methane hydrocarbons. Sulfur content for baseline methane formulations (including additives) is limited to 16 parts per million (ppm) by volume. Methane formulations must contain added odorants with an elemental composition that satisfies the baseline methane definition.

The baseline methane category includes all methane fuels (and associated additives) meeting the above criteria.

Non-baseline methane formulations are those that exceed the limit of 20 mole percent non-methane hydrocarbons. Atypical methane formulations include products containing elements in addition to carbon, hydrogen, nitrogen, oxygen, and/or sulfur, or exceed the baseline sulfur limit of 16 ppm by volume.

f. Propane. Propane formulations are defined as those containing at least 50 percent propane by volume. The baseline propane category includes LPG formulations containing no elements in addition to carbon, hydrogen, oxygen, nitrogen, and/or sulfur. Baseline LPG products are restricted to a maximum of 20 percent by volume for non-propane hydrocarbons. Sulfur content (including additives) is restricted to 123 ppm by weight. LPG formulations must have a distinctive odor. The elemental composition of odorants added to LPG formulations is limited to carbon, hydrogen, oxygen, nitrogen, and sulfur. The baseline propane category includes all propane fuels (and associated additives) meeting the above criteria.

Non-baseline propane formulations are those that exceed the specified limit for non-propane hydrocarbons. Atypical propane formulations include LPG products that contain elements in addition to carbon, hydrogen, nitrogen, oxygen, and/or sulfur, or exceed the baseline sulfur limit of 123 ppm by weight.

3. F/FA Groups

The F/FA groups are subdivisions of the F/FA categories and represent the final level of product classification within the grouping system. The groups are the actual operating units of the grouping system. The objective underlying the group definitions is to sort F/FAs together when it is reasonable to assume that their emission products will be essentially the same on a qualitative basis.

A summary table of the F/FA grouping system is included in the regulations (see Table F94-7 in Sec. 79.56). In this table, the fuel families serve as column headings and the categories define the rows. The resulting combination of fuel families and categories (i.e., the boxes in the table) contain the F/FA groups. Within each category, one or more groups are defined according to the presence of differing constituents in the raw fuel or additive/base fuel mixture. The number of groups in a particular F/FA category depends on the variability among the products in that category. For example, the atypical category for each fuel family potentially consists of many groups that are defined according to the atypical element(s) or constituent(s) specified for the particular family. Within each group, one formulation is chosen to represent all of the member products in compliance with the registration requirements. Related costs may be shared by participating F/FA manufacturers within each group.

Groups within the Baseline Categories. The baseline category for each defined fuel family contains a single F/FA group, with the exception of the baseline methanol category. As discussed above, the baseline methanol category includes two groups: the M100 group and the M85 group. The representative to be used in required emission characterization and health effects tests for each baseline group is the designated base fuel for the respective fuel family (see Section V). For example, all gasoline formulations meeting the gasoline baseline criteria are sorted into one group, to be represented in testing by the designated gasoline base fuel. The same holds true for diesel, ethanol, methane, and propane fuel families. In the case of methanol, baseline formulations are divided into two groups and testing is performed on two representatives, one for each of the designated baseline groups, i.e., M100 base fuel and M85 base fuel.

Groups within the Non-Baseline Categories. Non-baseline categories are defined for each fuel family. F/FAs in non-baseline groups include products that comply with the baseline elemental composition restrictions for the respective fuel family, but do not meet quantitative limits on certain baseline components (e.g., oxygen content). Non-baseline groups are defined according to the constituent(s) that differentiate the fuel or additive/base fuel mixture from the baseline products in the respective fuel family. The representative for each non-baseline group is the member of the group with the highest concentration of the non-baseline constituent.

(a) Gasoline. Gasoline formulations which comply with the baseline elemental composition criteria, except that they have a total oxygen content of 1.5 weight percent or more, are designated as non-baseline. These products are grouped according to the specific oxygenate compound (e.g., any specific alcohol, ether, or methanol/co-solvent combination) used to increase the oxygen content of the gasoline formulation. Thus, separate non-baseline groups are defined for ethanol, methyl tertiary butyl ether (MTBE), ethyl tertiary butyl ether (ETBE), tertiary amyl methyl ether (TAME), diisopropyl ether (DIPE), di-methyl ether (DME), tertiary amyl ethyl ether (TAEE), etc.

In the NPRM, EPA had proposed to define additional non-baseline groups for fuels containing combinations of oxygenate compounds (or for which the registration contained multiple oxygenate additives), with separate groups defined for each combination recorded in a registration. However, EPA decided not to require the testing of oxygenate combinations in this final rule during the routine Tier 1 and Tier 2 testing program. EPA believes that the testing of fuels with individual oxygenates will satisfy the main objectives of the program by providing basic information about the potential health effects of particular oxygenated compounds in gasolines. Requiring routine testing of every recorded combination was judged unreasonable, as it resulted

in a number of groups that did not reflect actual formulations in use. If there is concern about the toxicity of specific mixtures of oxygenated compounds, EPA may require additional testing under Tier 3 on a case-by-case basis.

An exception to this treatment of oxygenate combinations occurs in the case of non-baseline formulations containing methanol. Existing ``substantially similar'' criteria currently limit the use of methanol as an oxygenate in gasoline to 0.3 percent by volume (i.e., 0.1 percent by weight), unless the formulation contains appropriate alcohol co-solvents. Thus, methanol-containing gasoline formulations with 1.5 weight percent oxygen or more must also contain a co-solvent. Accordingly, in the grouping system, each methanol and co-solvent combination used in gasoline formulations defines a different non-baseline group [e.g, methanol and isopropyl alcohol (IPA), methanol and tertiary butyl alcohol (TBA), methanol and butanol, etc.]. Those oxygenate compounds used as co-solvents for methanol need to be identified as such in a fuel's registration. If an oxygenate is not identified as a methanol co-solvent, even if it appears in a fuel registration that also includes methanol, then EPA will assume that it defines a gasoline/oxygenate group separate from the methanol/gasoline mixture.

Within each non-baseline gasoline group, a formulation consisting of the base gasoline fuel blended with the highest weight percent of the oxygenate or methanol/co-solvent combination registered for any member F/FA product will serve as the group representative that will be tested to comply with the program's requirements. The selection of the group representative is to be based on the highest actual concentration-in-use or the highest recommended concentration-in-use, whichever is the greater, for the particular oxygenate or oxygenate/co-solvent blend.

EPA recognizes that current fuel registration procedures allow manufacturers to include in the original registration a list of all the potential additives that might be used in the marketed fuel, along with the applicable range of concentration-in-use for each alternative. Under these circumstances, this final rule makes the non-baseline producer responsible for the testing of each oxygenate listed in the registration. For example, if a gasoline fuel registration lists methanol/co-solvent, ethanol, MTBE, and ETBE, then the manufacturer is responsible for separately testing each of four gasoline/oxygenate blends: gasoline-methanol/co-solvent, gasoline-ethanol, gasoline-MTBE, and gasoline-ETBE. The multiple testing responsibility can be satisfied by the manufacturer individually or by participating in four applicable groups. In each group, a formulation consisting of the base gasoline fuel blended with the highest concentration of the oxygenate listed for any member fuel or additive/base fuel mixture would serve as the group

representative to be tested to comply with the program's requirements.

The existing fuel registration procedures also allow manufacturers to report a range of concentration-in-use for each bulk additive listed as a potential component of the registered fuel. Thus, it is possible for the same registration to include formulations under both baseline and non-baseline definitions. If so, the manufacturer is responsible for testing formulations in both categories covered by the indicated range listed in the registration. In other words, if the reported range of concentration-in-use of an added oxygenate could include gasoline formulations with less than 1.5 weight percent oxygen as well as formulations with 1.5 weight percent oxygen or more, then the manufacturer is responsible for testing formulations in both baseline and non-baseline categories. For example, suppose a gasoline registration includes two potential oxygenates with respective concentration-in-use (shown here in terms of the resulting oxygen content in the formulation), as follows: ethanol (0 to 3.5 percent oxygen by weight) and ETBE (0 to 2.7 percent oxygen by weight). Because the indicated ranges include both baseline and non-baseline formulations, the manufacturer would be responsible for the testing of three formulations: baseline gasoline, a non-baseline gasoline-ethanol blend, and a non-baseline gasoline-ETBE blend. If the manufacturer chooses to participate in grouping arrangements, then he/she would be sharing the cost of the testing for the representative of each of these three groups.

(b) Diesel. Non-baseline diesel formulations contain at least 1.0 percent oxygen by weight. Non-baseline formulations include alcohol blends, ether blends, biodiesels (e.g., diesel-soy methyl ester blend), and other formulations containing oxygenating compounds. Separate non-baseline groups are defined for each added alcohol or ether (e.g., methanol, ethanol, DME, etc.) and for other oxygenating compounds by class (e.g., peroxides, nitroso compounds, nitro compounds, alkyl nitrites, alkyl nitrates, animal-source alkyl esters, vegetable-source alkyl esters, furans, etc.).

Diesel fuel manufacturers are responsible for the testing of each added alcohol, ether, or oxygenate class included in their fuel registration. For example, if the registration includes added methanol and soy methyl ester, the manufacturer will be responsible for testing two non-baseline formulations: (1) A diesel-methanol blend and (2) a diesel formulation containing a vegetable-source alkyl ester. In order to satisfy the testing requirements, the manufacturer may perform the tests individually or take advantage of the grouping provisions to share the testing costs with other manufacturers of similar products. In the above example, the manufacturer will be able to group with other manufacturers of diesel formulations containing methanol and with other manufacturers of formulations containing other vegetable-source alkyl

esters (e.g., rape methyl ester).

For each diesel non-baseline group defined by the presence of an alcohol, ether, or class of oxygenating compound, the representative to be used in testing will be a formulation consisting of the diesel base fuel blended with the highest actual or recommended concentration-in-use of the particular alcohol, ether, or class of oxygenating compound, as recorded for any member of the group. For example, if manufacturers form a group of non-baseline diesel formulations containing vegetable-source alkyl esters, the group representative will be a diesel formulation containing the highest volume percent of any of the vegetable-source alkyl esters represented in the group. The alkyl ester is to be added to the base diesel fuel for conducting the required emission characterization and toxicity tests.

EPA recognizes that current registration procedures allow manufacturers to include in the original diesel fuel registration a list of all the potential oxygenating compounds that might be used in the marketed fuel, along with the applicable range of concentration-in-use for each alternative. As with gasoline formulations, this final rule requires the diesel fuel producer to test each alcohol, ether, or class of oxygenate listed in the registration. Also, if a registration lists a range of oxygen content that defines both baseline and non-baseline formulations, then the manufacturer is required to test both a baseline formulation and a non-baseline formulation.

(c) Methanol. Non-baseline methanol formulations conform with the baseline limits in terms of elemental composition, but contain more than 4 percent by volume of substances other than methanol and gasoline. Individual groups are defined for each non-methanol, non-gasoline component, and for each unique combination of such components. The representative of each non-baseline methanol group will be the group member with the highest concentration (i.e., percent by volume) of the non-methanol, non-gasoline component(s).

(d) Ethanol. Non-baseline ethanol formulations conform with the baseline limits in terms of elemental composition, but contain more than 5 percent by volume of substances other than ethanol and gasoline. Individual groups are defined for each non-ethanol, non-gasoline component, and for each unique combination of such components. The representative of each non-baseline ethanol group will be the group member with the highest concentration (i.e., percent by volume) of the non-ethanol, non-gasoline component(s).

(e) Methane. There is only one non-baseline methane group. This group contains all methane formulations conforming with the baseline criteria except that they exceed the allowable limit for non-methane hydrocarbons (i.e., 20 mole percent). The representative for the non-baseline methane group will be the member formulation containing the highest concentration of non-methane hydrocarbons.

(f) Propane. Non-baseline propane formulations are those which conform with the baseline criteria except that they exceed the allowable limit for non-propane hydrocarbons (i.e., 20 percent by volume). All non-baseline propane formulations are sorted into a single group. The representative for the non-baseline propane group will be the member formulation containing the highest concentration of non-propane hydrocarbons.

Groups within the Atypical Categories. Atypical groups within each fuel family are defined according to the distinctive atypical constituent(s). Separate groups are established for any single atypical constituent and any unique combination of atypical constituent(s) which occurs among the products in each category. For example, if a gasoline fuel contains sodium, and no other atypical element, then this atypical fuel will group with other gasoline fuels or additive/base fuel mixtures containing sodium as their only atypical constituent. However, if a gasoline fuel contains sodium and potassium, then this fuel will define a separate group for formulations containing both sodium and potassium. As explained previously, EPA believes that this approach is reasonable because different atypical elements may have distinct toxicological effects. Thus, while similarly composed F/FAs may group together, EPA believes that testing distinct F/FAs separately best effectuates CAA Section 211(e), which states that ``each'' F/FA shall be tested.

Groups are further subdivided according to the presence of polymers containing atypical element(s) in their molecular structure. F/FAs containing polymers are considered atypical for a respective fuel family only if the F/FA product as a whole contains one or more atypical elements. If the polymer contains an atypical element as part of its molecular structure, then the atypical polymer defines a separate atypical group. For example, the presence of polyethylene in a gasoline product does not in itself make that product atypical because polyethylene contains no elements in addition to carbon, hydrogen, oxygen, nitrogen, and sulfur. On the other hand, if the gasoline product contains chlorinated polyethylene, then the product is considered atypical because of the chlorine content (chlorine is an atypical element for the gasoline fuel family). Such product could group with other atypical gasoline products containing chlorinated polymers. However, if the atypical gasoline product contains polyethylene and chlorine as two different components of the formulation, the product will group with other atypical gasoline products containing chlorine in non-polymer constituents.

For groups defined by a single atypical constituent, the representative to be used in satisfying the group's testing requirements will be the member fuel or additive/base fuel mixture with the highest actual or recommended concentration-in-use of the atypical

constituent. Within a group of such products containing a unique combination of two or more atypical elements, the representative shall be the product which has the highest total concentration of atypical elements. In the case that two or more products within such a group contain the same and highest concentration of atypical constituents, the process specified for selecting the representative gives precedence to the highest total concentration of the atypical constituents in the following priority order: (1) Total concentration of metals, (2) total concentration of halogens, (3) total concentration of other atypical elements (including sulfur, if applicable), (4) total concentration of polymers containing atypical elements, (5) total concentration of oxygen.

As discussed previously, current fuel registration procedures allow manufacturers to include in their registration a list of potential bulk additives to be used in the fuel. As a result, registrations could include several additives containing one or more atypical constituents with the same purpose-in-use, but which are not intended by the fuel manufacturer to be used at the same time. If several additives for the same purpose-in-use are listed in a single registration, and if these additives contain different atypical elements, the manufacturer is responsible for testing each individual atypical additive separately. This means that each unique atypical additive listed in a registration for the same purpose-in-use will define a different testing group. On the other hand, if a fuel registration includes additives with different functions and different atypical elements, and if these additives are normally blended together in the same formulation, then the manufacturer is allowed to test them together (or to participate in an applicable group). For example, if a diesel fuel registration lists two atypical biocide additives, one containing boron and the other containing chlorine, the fuel manufacturer would then be responsible for testing two formulations (one diesel formulation containing boron and one diesel formulation containing chlorine). However, if the registration includes a boron-containing biocide and a chlorine-containing detergent, then the manufacturer may test the two additives together.

C. Implementation of Grouping System and Cost-Sharing Provisions

The grouping system included in this final rule allows manufacturers of similar F/FAs, on a voluntary basis, to pool their resources and efforts to satisfy the registration requirements. The primary objectives of the grouping system and cost-sharing provisions are to reduce the overall costs of the registration program and maximize the efficiency of the program by avoiding duplication of effort. The grouping and cost-sharing provisions included in today's

rule are supported by CAA section 211(e)(3)(B), which permits manufacturers of similar F/FAs to share the testing costs of the program so that requirements can be met without duplication. Although this rule allows manufacturers to comply with the program's requirements by participation in a group, each manufacturer continues to be individually subject to this rule and responsible for testing under this rule.

The practical implementation of the grouping system involves two major tasks: (1) The organization and administration of group functions, and (2) the development of equitable arrangements for cost-sharing. Backed by its experience with respect to the TSCA testing program, EPA judges that the F/FA industry, under the aegis of its various trade associations or other third parties, is capable of accomplishing these tasks with little or no Agency assistance and interference. EPA's experience with cost-shared testing under TSCA regulations (40 CFR part 791) indicates that manufacturers prefer to work out their own cost-sharing arrangements, and EPA anticipates that F/FA registration applicants will likewise prefer to work out their own cost-sharing agreements. Public comments from the regulated industry support this assumption. Thus, EPA intends for manufacturers to work out cost-sharing agreements by themselves. However, if F/FA manufacturers cannot work out cost reimbursement, this rule allows F/FA manufacturers to use procedures similar to existing TSCA procedures [see Sec. 79.56(c) of this rule] for resolution of disputes.

In addition to establishing cost-sharing mechanisms, F/FA manufacturers will also need to develop agreements concerning the division of responsibilities among group members for meeting the specific requirements of the registration program. EPA expects the participation of industry trade associations in the formation of groups and management of these activities. These associations should be able to establish ``third-party'' mechanisms whereby individual manufacturers can enroll their products in appropriate groups while minimizing the extent to which confidential data must be revealed. Each manufacturer needs to determine whether the grouping and cost-sharing advantages outweigh the possible competitive risks involved.

In general, F/FA manufacturers should be able to determine the appropriate groups for their products without EPA involvement, according to the grouping criteria specified in this final rule, and to enroll their products into those groups. However, EPA recognizes that some Agency involvement might be needed in some special cases. When appropriate, based on EPA's discretion, the Agency will provide limited guidance for those manufacturers needing assistance with the application of the grouping criteria to their specific products.

Manufacturers of F/FAs registered prior to the effective date of this rule are required to notify EPA within six months after the

effective date of this rule if they intend to comply with the rule as part of a group and, if so, to identify the person or entity which is organizing the testing (see Section XII.A). In this case, groups of producers would organize prospectively to complete the same program requirements for their similar products and cost-sharing arrangements could be reached in advance of testing.

Manufacturers of F/FAs not registered prior to the effective date of this rule are expected to conduct the required testing individually, unless they certify to EPA that they intend to rely on data to be submitted (and/or previously submitted) by an existing group or individual manufacturer of a similar registered product. The certification needs to include assurances that the original submitter has been notified (see Section XII.A for notification requirements) and that the manufacturer intends to comply with reimbursement as provided in this rule.

Under the reimbursement provisions in this rule, there will be a fifteen years ``reimbursement period'' for the original submitter (individual or group) to obtain reimbursement from those manufacturers that rely on previously submitted data. This period has been lengthened from the originally proposed five years in response to public comments.

V. Base Fuel Specifications and Formulation Requirements

In this final rule, EPA is establishing chemical and physical specifications to represent base fuel formulations for each defined fuel family. EPA has adopted the method proposed in the reformulated gasoline rulemaking (56 FR 31176), which uses sales-weighted averages of fuel survey data to determine national average chemical and physical parameters, to establish base fuel specifications for gasoline and diesel. Because comparable survey data are not available for alternative fuels, the base fuels for the alternative fuel families are based on CARB definitions and limited survey information.

The generic base fuel formulations will function as archetypes of the F/FAs in each fuel family and will serve as the test substance or group representatives for the baseline group(s) for the respective fuel family. The use of consistently formulated base fuels will facilitate the comparison of the emission and health effect test results from the many fuel and fuel additive products within each fuel family. The base fuels will also serve as the fuel substrates into which additives undergoing evaluation will be mixed prior to emission generation and testing. Tests conducted on the emissions of the base fuel will then serve as controls against which tests on the emissions of the additive/base fuel mixture will be compared.

In addition to defining chemical and physical parameters for each base fuel, EPA is also specifying the allowable additive(s) to be

included in the base fuel. EPA recognizes that commercial fuels typically contain additives to control fuel quality and enhance fuel performance, as well as to help in fuel production and distribution. Ideally, in order to better isolate the health effects associated with a particular additive or fuel, the base fuel would not contain additives unless they were the actual test subjects. However, several bulk additive types are common to most of the fuels within a given fuel family, and these should arguably be included as part of the base fuel. As a practical matter, it would be difficult in some instances to find a fuel that did not contain certain additive types used by refiners to facilitate production or distribution. EPA is thus requiring that base fuels contain a limited complement of the additives which are essential for the fuel's production or distribution and/or for the successful operation of the test vehicle/engine throughout the mileage accumulation and emission generation periods required under this rule. Since additives may have a substantial effect on emissions, for purposes of standardization it is important to specify the additive types which are to be contained in the base fuels. However, the selection of the specific product within each specified additive functional category is left to the formulator of the base fuel and/or the manufacturer responsible for the testing. Unless otherwise restricted, the presence of trace contaminants does not preclude the use of a fuel or fuel additive as a component of a base fuel.

Additive requirements for each defined base fuel are discussed in the following sections. Additives used as base fuel components are to be added at the minimum treatment rate needed for effective performance. In contrast, additives to be tested must be mixed in the base fuel at the maximum in-use concentration recommended by their manufacturers.\19\ When a fuel additive is tested, any additive normally contained in the base fuel which serves the same function as the test subject additive must be removed from the base fuel formulation. For example, if a corrosion inhibitor is to be tested, this test additive would replace the corrosion inhibitor normally included as a component in the base fuel. This substitution requirement may preclude the use of certain multi-functional additives as base fuel components (in the case where the subject additive serves one of the functions of the multi-functional additive), since it would not be possible to replace a portion of a multi-functional additive with the test subject additive.

\19\Special provisions related to the testing of additives are discussed in Section VI.F.

Note: The specifications in the following sections describe the

base fuel(s) for each fuel family, which serve the test fuel functions discussed above. These base fuel specifications are not the same as the criteria which permit F/FAs to join the baseline group within a fuel family. The baseline group criteria are provided in the preceding section of this preamble.

A. Gasoline

For the gasoline base fuel, EPA is requiring the use of the reformulated gasoline summer baseline fuel as specified in CAA Section 211(k)(10)(B)(i). This unleaded gasoline fuel, which is free of oxygenates, was determined from fuel survey data and will be used to represent all grades of conventional gasoline. This base fuel has the same specifications as the industry average gasoline used in many recent fuel emission studies, including the Auto/Oil Program\20\ and EPA's reformulated gasoline testing program. Selecting this formulation as the base gasoline fuel allows the comparison of emission characterization results from the F/FA testing program with a larger body of current emission data. The blending tolerances for the gasoline base fuel are consistent with certain blending tolerances specified in the RFG rule (59 FR 7716).

\20\Auto/Oil Air Quality Improvement Research Program, Technical Bulletin #1, December 1990; available in Docket A-90-07, Item No. IV-A-08.

The gasoline base fuel must contain the following additives: deposit control, corrosion inhibitor, demulsifier, anti-oxidant, and metal deactivator. In addition to the above required additives, the final rule allows manufacturers to use anti-static additives in the gasoline base fuel, if needed. Anti-static additives are not required in gasoline base fuel because this type of additives is not considered essential for the fuel's production, distribution, or the vehicle operation. Thus, anti-static additives should be used only as a safety measure on a case-by-case basis, as needed (e.g., when static problems present a risk of explosion). The required and permissible gasoline base fuel additives may contain no elements in addition to carbon, hydrogen, oxygen, nitrogen, and/or sulfur.

In the Reopening Notice, EPA proposed to preclude the use of sulfur-containing additives in the gasoline base fuel. However, in response to a number of comments from the regulated industry, this final rule permits up to 15 ppm sulfur to be included in the additives. The total sulfur content in the base fuel, including any sulfur

contributed by the additive components, must equal 339 ppm (within a tolerance of <plus-minus> 25 ppm). A summary of the gasoline base fuel specifications and its additive components is provided in the accompanying regulations [see Table F94-1 in Sec. 79.55(b)].

B. Diesel

Reflecting its predominant usage, #2 diesel is selected in this final rule as the base fuel for diesel. The specifications for the diesel base fuel were determined by calculating an industry average diesel fuel from 1990 industry and government diesel fuel survey data. The sources of data and methods of calculations are contained in the docket for this rulemaking.\21\ The blending tolerances for the diesel base fuel have been set to be comparable to those used in the gasoline base fuel. An exception to this general methodology is the base fuel specification for sulfur level. The required sulfur level (0.05 weight percent) reflects current on-road diesel fuel sulfur limits (55 FR 34120).

\21\See memorandum from James Greaves to Docket A-90-07 (Item No. IV-B-01) regarding ``Revised Base Diesel Fuel Determination Procedures for the Fuels and Fuel Additives Rulemaking."

The additives required as diesel base fuel components are: corrosion inhibitor, demulsifier, anti-oxidant, and metal deactivator. In addition to the above required additives, the final rule allows the use of anti-static and flow improver additives in the diesel base fuel, as needed. As with gasoline, anti-static additives are not required because they should only be used in the case of static accumulation problems. Similarly, flow improvers may be used on a need basis to improve cold weather handling.

As in the gasoline base fuel, the diesel base fuel additives may contain sulfur, as well as carbon, hydrogen, oxygen, and nitrogen. The total sulfur content in the diesel base fuel formulation, including any sulfur contributed by the additives, may not exceed 0.05 percent by weight. A summary of the diesel base fuel specifications and allowed additive components is provided in the regulatory text [see Table F94-2 in Sec. 79.55(c)].

C. Alternative Fuels

EPA has used CARB definitions and other available information to establish base fuel specifications for each alternative fuel family

(see Tables F94-3--F94-6 in Sec. 79.55). However, due to rapidly developing technology, the fuel additive package requirements for these fuels are not as well established as for gasoline and diesel. In fact, there is only limited information available on the additive requirements for the successful long-term operation of each alternative fuel/vehicle combination. Hence, it is the responsibility of the F/FA manufacturers who are required to test such base fuels (in consultation with EPA), to comply with the additive requirements of the manufacturer of the particular vehicle/engine used for the testing of alternative F/FAs. If the manufacturer of an alternatively-fueled vehicle or engine specifies that additives (beyond those specified in the regulations), are essential for operation, then the F/FA manufacturer should submit a request to EPA to use those additional additives as components of the base fuel at the minimal effective level. EPA will publish a document in the Federal Register whenever approving such a request to modify a base fuel.

1. Methanol

The methanol fuel family contains two fuel groups, one for M100 fuels and one for M85 fuels. Each of these methanol groups has its own base fuel. These base fuels may only contain the elements carbon, hydrogen, oxygen, nitrogen, sulfur, and chlorine. The chlorine (as chloride) is permitted as a contaminant remaining from methanol production, and is limited to no more than 0.0001 percent by mass. The sulfur content may not exceed 0.002 percent by mass in the base M100 fuel and may not exceed 0.004 percent by mass in the base M85 fuel.

The M100 base fuel must consist of 100 percent chemical grade methanol by volume. The M85 base fuel is to contain 85 percent chemical grade methanol by volume, blended with 15 volume percent base gasoline fuel (meeting the gasoline base fuel specifications outlined in Section V.A., above). Specifications for the methanol base fuels are listed in Table F94-3 in Sec. 79.55(d) of the regulations.

Some gasoline detergents have been shown to cause intake system deposits when used in M85 applications. Likewise, lubricating oils containing calcium have been shown to cause injector tip deposits in M100 applications. Therefore, EPA recommends that F/FA manufacturers determine the methanol compatibility of lubricating oils as well as fuel additives used in the gasoline portion of the M85 base fuel.

2. Ethanol

The ethanol fuel family contains one group, represented by E85 base fuel. The E85 base fuel is to contain 85 percent chemical grade ethanol by volume, blended with 15 volume percent base gasoline. The ethanol base fuel may only contain the elements carbon, hydrogen, oxygen, nitrogen, sulfur, chlorine, and copper. The chlorine (as chloride) is permitted as a contaminant remaining from ethanol production, and is limited to no more than 0.0004 percent by mass. The sulfur content may

not exceed 0.004 percent by mass. Copper, also a contaminant from ethanol production, is limited to 0.07 mg/L.

Additives used in the gasoline component of E85 base fuel must be ethanol-compatible. The base fuel specifications for E85 are summarized in Table F94-4 in Sec. 79.55(e) of the regulatory text.

3. Methane

The methane fuel family is represented by a natural gas base fuel whose specifications are within the proposed ranges for natural gas certification fuel (as proposed in 57 FR 52912). This base fuel may only contain the elements carbon, hydrogen, oxygen, nitrogen, and sulfur, with the sulfur limited to 16 parts per million (by volume). The methane base fuel must contain added odorant for leak detection purposes, used at a level such that at ambient conditions the fuel has a distinctive odor potent enough for its presence to be detected down to a concentration in air of not over \1/5\ (one-fifth) of the lower limit of flammability.

In the Reopening Notice, EPA proposed that any sulfur in the methane base fuel be limited to that contained in the odorant additive. In response to public comment, this restriction has been removed; however, the total sulfur in the methane base fuel formulation, including that contributed by any additives, may not exceed 16 parts per million. The methane base fuel specifications are listed in Table F94-5 in Sec. 79.55(f) of the accompanying regulations.

4. Propane

The propane fuel family is represented by a commercial LPG base fuel. The propane base fuel may only contain the elements carbon, hydrogen, oxygen, nitrogen, and sulfur, with the sulfur limited to 123 ppm (by weight). The propane base fuel must contain added odorant, for leak detection purposes, at a level such that at ambient conditions the fuel has a distinctive odor potent enough for its presence to be detected down to a concentration in air of not over \1/5\ (one-fifth) of the lower limit of flammability. As in the case of the methane base fuel, the final rule does not require the sulfur in the formulation to be contained only in the odorant additive. Rather, the sulfur limitation applies to the fuel/additive mixture in combination. The propane base fuel specifications are listed in Table F94-6 in Sec. 79.55(g) of the regulatory text.

VI. Emission Generation

A. General Approach

As part of the registration requirements, F/FA manufacturers are required to conduct a detailed characterization of the combustion and evaporative emissions of their products, as well as biological tests in

which animals are exposed to these emissions. The next sections describe the methods specified in the rule for generating the emissions to be used in these chemical and biological tests.

As proposed in the NPRM, combustion emissions are to be generated using applicable portions of the FTP.\22\ To control some of the inherent variability of FTP emissions generated under transient engine operation,\23\ this final rule requires the use of a mixing chamber or other apparatus (see Section VI.B.2). This is one of the approaches discussed for consideration in the Reopening Notice. EPA is permitting the use of either the engine dynamometer or the chassis dynamometer for emission generation during biological testing using FTP or FTP-equivalent cycles. For the reasons discussed in the Reopening Notice, EPA has decided to require the use of non-catalyzed emissions (i.e., untreated exhaust emissions)\24\ for biological testing in order to assure that the test animals are exposed to the full range of emission species potentially resulting from the combustion of F/FAs. A brief summary of the rationale behind this decision is included below.

\22\Federal Test Procedure (FTP) are the standard exhaust and evaporative emissions test procedures described in 40 CFR part 86 and used by EPA to certify new vehicles.

\23\Transient engine operation is achieved by varying the engine speed and/or engine load, which typically results in an emission stream varying in quantity and composition over time.

\24\Exhaust emission not subject to an aftertreatment device such as a functional catalyst or particulate trap.

It is important to keep in mind that the purpose of this program is not to test the effectiveness of emission control devices or to directly evaluate the emission performance of various vehicles and engines. Rather, it is to examine the potential toxicologic effects of the emissions produced by F/FAs in use. With modern emission control technology in place, most of the ambient air pollutant species attributable to automobile exhaust come from two sources: Malfunctioning vehicles (``high emitters'') and normal vehicles during their cold start period, when their engines run rich and their catalytic converters have not yet reached effective operating temperatures. The variety of emissions from these two important sources are not well represented by hot, catalyzed exhaust generated from well-maintained, modern vehicles. Emissions during the cold-start include hundreds of organic chemical species which are generated before the catalytic converter reaches its effective temperature. Once the catalytic converter is warmed-up, its efficiency increases to the point

where only a dozen or so simple compounds remain in readily measurable amounts in the catalyzed exhaust. Thus, the use of catalyzed exhaust in the biological testing program would exclude from the tests relevant emission species that could potentially be harmful to human health or the environment. In fact, laboratory animals would be exposed to only very few of the organic emission species associated with the combustion of the fuel or additive of interest. In contrast, the ambient air normally contains the full range of combustion emissions, since cold-start emissions are continuously reintroduced and some ``high emitters'' are always in operation. Since humans experience long-term exposure to these emissions, EPA believes it is important that they be included in the test exposure atmosphere. EPA's analysis\25\ of non-catalyzed emission data demonstrates that emissions that receive no aftertreatment represent a comprehensive aggregate of characteristic combustion products at enriched concentrations, including the species which may otherwise be emitted only during the cold start or by high-emitting vehicles. In order to simulate emissions that include the full range of potential species produced in the combustion of F/FAs, EPA is requiring the use of non-catalyzed emissions for biological testing in this program.

\25\See memorandum from Stephen Mayotte to Docket A-90-07 (Item No. IV-B-02) regarding ``Engine-out versus Tailpipe Emissions in Light-duty Vehicles.''

With the exception of exhaust after-treatment devices, this final rule requires that all normally required emission control equipment be present and fully operational on all test vehicles and heavy-duty engines used in the generation of non-catalyzed emissions. In order to maintain the appropriate operation of the exhaust system while obtaining non-catalyzed emissions, EPA requires the use of non-functional aftertreatment devices (e.g., a blank catalyst with no catalytic wash coat) in order to simulate the back pressure, residence time, and mixing characteristics usually provided by normally functioning aftertreatment devices. Special emission generation allowances for the testing of specific additives which are introduced for use in conjunction with certain aftertreatment devices are discussed in Section VI.F.

B. Combustion Emission Generation

1. For Emission Characterization

Manufacturers are required under Tier 1 to characterize the

combustion emissions of their F/FAs. Depending on the fuel family in question, vapor-phase, semi-volatile, and particulate emissions may be required to be characterized.\26\ As discussed in Section VII.B., the emission characterization requirements include the measurement of hydrocarbons, carbon monoxide, oxides of nitrogen, particulates, aldehydes, ketones, alcohols, ethers, polycyclic aromatic compounds, and atypical products, as applicable.

\26\Examples of general sampling procedures for vehicle emissions are discussed in Schuetzle, D., ``Sampling of Vehicle Emissions for Chemical Analysis and Biological Testing," Environmental Health Perspectives, Volume 47, pp. 65-80, 1983.

Both untreated (non-catalyzed) and treated (tailpipe)\27\ emissions generated using FTP conditions are to be characterized. Characterization of the tailpipe emissions will allow comparison of emissions from the test F/FA product with results from other studies. Characterization of the non-catalyzed emissions will be used to identify the emissions to which animals will be exposed in the biological tests.

\27\Tailpipe emissions are emissions downstream from all normally present emission aftertreatment devices, i.e., catalytic converters and/or particulate traps.

Applicable FTP procedures to be used in generating emissions are specified in 40 CFR part 86. The Urban Dynamometer Driving Schedule (UDDS)\28\ and the Engine Dynamometer Driving Schedule (EDS)\29\ cycles of the FTP shall be used in the emission generation for light-duty vehicles and heavy-duty vehicles, respectively. The motoring portion of the heavy-duty test cycle can be eliminated, at the manufacturer's option, for the generation of emissions. This will allow the use of relatively inexpensive dynamometer equipment without compromising the value of the test.

\28\UDDS is a 1372 second transient speed driving sequence used by EPA to simulate typical urban driving. The UDDS for light-duty vehicles is described in 40 CFR part 86, Appendix I(a).

\29\EDS is the transient engine speed versus torque time sequence commonly used in heavy-duty engine evaluation. The EDS for

heavy-duty diesel engines is described in 40 CFR part 86, Appendix I(f)(2).

As discussed earlier, this final rule allows the use of a vehicle or engine for emission generation using FTP procedures. In the case of F/FAs normally used in light-duty vehicle applications, if an engine is to be used, the appropriate speed versus torque trace for the UDDS must be determined in a vehicle on a chassis dynamometer prior to emission generation. The engine used for emission generation in this testing program must then be operated under specific speed and torque conditions that simulate the UDDS.

In light-duty vehicle testing, vapor phase emission samples are to be collected for each segment of the FTP cycle (i.e., Bag 1, Bag 2, and Bag 3). In addition, a semi-volatile sample and a particulate sample are to be collected during the driving cycle for light-duty vehicles. The heavy-duty testing procedure includes two tests: a cold-start test and a hot-start test. All three emission phases (i.e., vapor, semi-volatile, and particulate) are to be collected for each heavy-duty test. Some modifications to the standard FTP may be required for collection of semi-volatile and particulate emissions, which are required for emission characterization and in-vitro biological testing (see next section). Special procedures may also be necessary in order to characterize emissions from F/FAs containing atypical elements. Good engineering and analytical chemistry practices should be followed while modifying the applicable test cycle for the collection of fractions not specified in 40 CFR part 86. Such modifications must be described in detail in the discussion of emission generation procedures to be included in the report provided to EPA, as discussed in Section XII.B.

Vapor-phase emissions are to be collected and stored in Tedlar bags for subsequent chemical analysis. These emissions can be stored for only a limited period of time before chemical changes may occur. The critical time period is a function of the composition of the emissions, storage temperature and pressure, type of storage container, exposure to ultraviolet light, and the amount of deterioration that is considered acceptable. The maximum allowable storage times for emissions which are to be subjected to chemical analysis will vary depending on the speciation protocol, and are identified in relevant parts of the regulatory text.

The particulate fraction may be collected on a single filter instead of on multiple filters as prescribed in the FTP. Although the filter collection procedures outlined in the CFR were designed for heavy-duty emission testing, these methods are applicable and can be used in light-duty applications as well. Similarly, semi-volatile phase emissions are to be collected on one apparatus for the entire driving

cycle. Semi-volatile emissions are collected immediately downstream from the particulate collection filters using porous polymer beds or other equipment designed for their capture.\30\ After collection, the soluble organic fractions of the particulate and semi-volatile emissions are to be separately extracted using appropriate laboratory procedures.\32\ Because the extracted materials are much more stable than gaseous combustion emissions, they can be stored up to six months if protected from ultraviolet light and maintained at or below -20 deg.C. Particulate phase emissions can be stored either on the collection filter or after extraction. Semi-volatile phase emissions must be extracted immediately after collection. The duration of the collection process which will be needed to obtain sufficient quantities of the test substance will vary depending on the emission characteristics of the engine and fuel or additive/base fuel mixture, and on the requirements of the biological test protocol. If an insufficient amount of particulate or semi-volatile material is obtained during a single driving cycle, the FTP may be repeated as required and the extracted organic fractions combined.

\30\An example procedure using a porous polymer resin as a trapping medium is described in Stump, F. et al., ``Trapping Gaseous Hydrocarbons for Mutagenic Testing," SAE Technical Paper Series No. 820776, 1982; Available in Docket A-90-07 (Item No. II-J-14).

\31\Examples of particulate and semi-volatile emission collection and analysis methods are described in 40 CFR Sec. 86.1301-1344 and in Coordinating Research Council Report No. 551 (entitled ``Chemical Methods For The Measurement Of Unregulated Diesel Emissions--Carbonyls/Aldehydes, Particulate Characterization, Sulfates, PAH/NO<INF>2PAH," August 1987; available in Docket A-90-07, Item No. II-J-15).

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2. For Biological Testing

Non-catalyzed emissions are to be generated for conducting biological tests, following the same procedures described above for emission characterization.

In vitro biological testing (i.e., the Salmonella assay) is to be conducted on extracts of the particulate and semi-volatile emission phases separately. Particulate and semi-volatile emissions are to be collected in a manner identical to the procedure used for particulate and semi-volatile emission characterization.

The in vivo biological testing requires the generation of whole untreated emissions for a minimum of six hours per day, five days per week, for 13 weeks. To generate these emissions, light-duty vehicles

(or engines) with non-functional after treatment devices (e.g., blank catalyst with no catalytic wash coat) are to be operated under FTP or FTP-equivalent engine conditions. The continuous generation of emissions throughout the required exposure period requires light-duty vehicles/engines to be driven through repeated UDDS cycles and heavy-duty engines to be operated over repeated EDS cycles. If desired, registrants may automate their emission generation system.

As discussed in the Reopening Notice, EPA was concerned about the inherent variability of FTP-generated emissions. To accommodate the FTP transient cycle within the biological testing program, this rule requires the use of an apparatus to provide a more stable exposure environment for biological testing. For this purpose, EPA recommends the development and use of a large dilution/mixing/integration chamber located between the constant volume sampling (CVS) system and the final dilution apparatus, just prior to the exposure chamber containing the test animals. The mixing chamber will allow the necessary adjustment of the exhaust concentrations and integration of the large concentration swings typical of FTP exhaust, prior to exposing the test animals. This chamber must meet certain performance specifications based on the average concentration of total hydrocarbons in the exhaust. That is, the average concentration of total hydrocarbons leaving the mixing chamber must be within ten percent of the average concentration of total hydrocarbons entering the chamber. Much of the CVS system concentration variability is associated with the rapidly changing dilution ratios that result from rapidly changing exhaust flow rates. EPA recognizes that vehicle exhaust sampling devices, such as mini-diluters,\32\ are being developed to maintain constant dilution ratios during transient testing. These systems will eliminate much of the concentration variability of classical CVS exhaust. As discussed in the Reopening Notice, these systems are currently under development and their use at this time is limited. However, today's rule will allow their use if they can meet the performance specifications defined above as well as other requirements of the testing program.

\32\A discussion on mini-diluter technology can be found in: American Industry/Government Emissions Research (AIGER) Cooperative Research and Development Agreement, ``Specifications for Advanced Emissions Test Instrumentation," AIGER PD-94-1, Revision 5.0, February 1994; available in Docket A-90-07, Item No. IV-A-09.

The combustion emissions generated for animal testing are to be diluted prior to delivery to the test animals. The CVS system, commonly used to condition exhaust for sampling and analysis, provides for

controlled ambient air dilution of the combustion emissions. However, water condensation can be a problem during CVS system sample conditioning, depending upon vehicle fuel consumption and fuel economy, dilution air humidity, and exhaust/diluent ratio.\33\ The use of pre-dried dilution air will lower the sample humidity, thus permitting lower dilution ratios and higher concentration of hydrocarbons to be achieved without condensation of water vapor. The minimum dilution ratio will vary with fuel composition. For example, a minimum dilution ratio of about 1:5 raw exhaust (dewpoint about 125 deg.F) with dry, clean filtered air is expected for gasoline fuels to reduce the water concentration to a dewpoint of about 68 deg.F. The minimum dilution ratio (maximum exhaust flow rate) occurs at about 200 seconds into the UDDS transient driving cycle. The dilution ratio is expected to be greater for methanol, ethanol, and natural gas fuels than for gasoline fuels because the exhaust water concentrations are greater with these alternative fuels. Heated transfer ducts or tubing can be used to avoid water condensation in much of the system, but the dilution/mixing/integration chamber will generally be at or near laboratory temperature (about 70 deg.F), and CVS dilution will have to be adequate to assure that the cumulative integrated chamber dew point remains below laboratory temperature at all times.

\33\An example procedure on how to deal with water vapor condensation problems is found in Black and Snow, ``Constant Volume Sampling System Water Condensation," SAE 940970, 1994. This paper describes a ``spreadsheet" procedure for detailed, second by second, determination of diluted exhaust dew point and the necessary CVS system flow rates to avoid water vapor condensation.

After initial dilution to preserve the character of the emissions, the exhaust stream may be further diluted to achieve the desired biological exposure concentrations. In testing the emissions of a particular fuel or additive/base fuel mixture, a manufacturer shall determine an optimum range of dilutions with which to characterize the health effects of the test substance. The range of dilutions shall include, at a minimum, an overtly or highly toxic concentration, a minimally toxic or non-toxic concentration, and a concentration of emissions having an intermediate level of toxicity. The selected concentrations must allow the determination of a concentration-response relationship (see Section VIII.A.3). EPA recommends that manufacturers review available literature for information on the design of inhalation studies.\34\

\34\An example reference is Phalen, R. F., ``Inhalation Studies: Foundations and Techniques," CRC Press, Inc., Boca Raton, Florida, 1984.

One important factor to consider in determining the exposure concentrations or dilutions is the effect of carbon monoxide (CO) concentration in test animals. The CO concentration in the emissions is expected to be a limiting factor in establishing the appropriate dilutions for the testing of F/FAs. Anoxia, among other negative health effects from this combustion product, may mask the more subtle health effects of F/FA emissions. EPA recommends that manufacturers review available literature on previous toxicity studies for information on appropriate CO concentrations that have been used in the exposure of laboratory animals to automobile emissions.\35\\36\\37\

\35\Stara et al., ``Long-Term Effects of Air Pollutants in Canine Species," EPA/600/8-80/014, 1980.

\36\Brightwell, J. et al., ``Neoplastic and Functional Changes in Rodents after Chronic Inhalation of Engine Exhaust Emissions," In: Ishinishi, N. et al., (eds), Carcinogenic and Mutagenic Effects of Diesel Engine Exhaust, Elsevier Science Publishers, Amsterdam, pp. 471-485, 1986; available in Docket A-90-07, Item No. IV-A-17.

\37\Pepelko, W. E. et al., ``Effect of 90 Days Exposure to Catalytically Treated Automobile Exhaust in Rats," Environmental Research, Volume 19, pp. 91-101, 1979.

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3. Verification Testing

A number of mechanisms can cause emissions to be captured in the dilution and sampling system before they can be characterized or used for animal exposures. Verification testing is required to determine the ratio (``recovery factor") of emissions that exit the sampling system to those that enter the system. This ratio must be high in order for subsequent emission testing to be meaningful.

EPA requires testing to verify the exposure atmosphere and to monitor the performance of the dilution/sampling system and mixing chamber, ensuring the repeatability of test results. Verification testing of the dilution/sampling system must be accomplished by injecting a known sample at the inlet and measuring the amount that exits the sample probe. For example, an injected hydrocarbon sample could be detected with a gas chromatograph and flame ionization detector to estimate the recovery factor. Similar verification procedures apply to the verification testing of the mixing chamber.

Verification procedures for the dilution/sampling system and mixing chamber are included in Sec. 79.57(e)(2)(v) of the accompanying regulations. Additional requirements include the monitoring of conditions (e.g., air flow, CO levels, etc.) in the inhalation exposure chamber and verification of test animal exposure levels (see Sec. 79.61).

C. Evaporative Emission Generation

Section III.A.2 discusses the RVP criteria which determine the applicability of evaporative emission testing to specific fuels and additive/base fuel mixtures. Evaporative emissions from in-use vehicles include diurnal, hot soak, resting and running loss emissions, and refueling emissions. However, to simplify the generation and collection procedures and to supply evaporative emissions of sufficient concentration for biological exposure testing, today's rule requires that evaporative emissions be generated using an evaporative emission generator (EEG). Emissions to be used both for characterization tests and biological exposure tests are to be generated in this way. The EEG is a fuel tank or vessel to which heat is applied to cause a portion of the fuel or additive/base fuel mixture to evaporate at a desired rate. Manufacturers will have flexibility in the design of the EEG used to test their particular F/FA. The size and/or number of EEG units to be used for evaporative emission testing will depend on the rate of emissions needed for the inhalation study. The vapor pressure of the F/FA product may influence the required tank size, as well. Emission rate modifications shall not be adjusted by temperature control, since emission composition is sensitive to temperature changes.

In general, the composition of evaporative emissions from vehicles does not resemble fully-evaporated whole samples of raw fuels or fuel additives. This phenomenon is due to differences in the vapor pressure of the fuel or fuel additive components and the effects of evaporative emission control equipment. To simulate this phenomenon with the EEG, procedures are to be followed to ensure that the evaporated fraction contain a reasonable representation of potential evaporated emission compounds. The EEG will be run at 13<plus-minus>05 deg.F and will be equipped with a drain. The fuel will be drained and replenished periodically in order to maintain a constant composition and prevent the build-up of heavier compounds in the non-evaporated portion. The concentration of emissions of the evaporated fuel or additive/base fuel mixture in the vapor space of the EEG during the time emissions are being withdrawn for testing shall not vary more than ten percent from the equilibrium concentration in the vapor space of emissions generated from fresh fuel or additive/base fuel mixture in the evaporative chamber.

EPA recognizes that other methods may also be suitable for generating F/FA evaporative emission mixtures for the testing purposes of this program. One possible alternative method was suggested in a comment received by EPA in response to the Reopening Notice.\38\ Based on the distillation properties of the test formulation, the suggested method would involve the distillation, condensation, and storage of the light-end components of the test fuel mixture, with revaporization of this whole fraction to generate test atmospheres. Other alternatives may also be valid. To accommodate these potential alternatives, the final rule contains a provision (see Sec. 79.57(f)(5) of the accompanying regulations) which permits manufacturers to request approval for methods other than the EEG for generating evaporative emission test atmospheres. To be granted, such requests must include supporting information which demonstrates (among other requirements) that the proposed procedures will generate emissions reasonably similar to in-use evaporative emission mixtures and that the generated emissions will be sufficiently concentrated to be useful in the context of toxicology tests. Approved procedures will be placed in the public docket.

\38\Comments of the American Petroleum Institute on U.S. EPA's Fuels and Fuel Additives Registration Regulations, March 28, 1994 (Item IV-D-49 in Docket A-90-07).

For applicable F/FAs, evaporative emission characterization requirements include the measurement of total volatile organic compounds with speciation of the hydrocarbon compounds, alcohols, ethers, and atypical compounds. Characterization requirements are discussed in Section VII.B.

For biological testing, evaporative emissions will be diluted and routed to the animal chambers in a manner similar to the method used for combustion emissions health effects testing, as described in the previous section, except that a mixing chamber is not required. The rate of emission generation shall be high enough to supply the biological exposure chamber with sufficient emissions to allow for a minimum of fifteen air changes per hour.

The concentration of total hydrocarbons in the evaporative emission stream routed to the biological exposure chambers is to be diluted to three separate concentrations to establish a range of responses similar to combustion emission testing (see previous section). Evaporative emissions are not constrained by CO, NO<INF>X, or CO<INF>2 levels, and hence can be used at higher concentrations than combustion emissions. Verification testing is required for evaporative emissions in a manner

analogous to the verification testing performed for combustion emissions.

D. Vehicle Selection

EPA is requiring that new vehicles or engines be used for the combustion emission generation and testing of F/FAs to avoid the carry-over effects from previously used fuels. All F/FAs must be tested in vehicles or engines (corresponding to chassis or engine dynamometer testing, respectively) that have been operated exclusively on the fuel or additive/base fuel mixture to be tested.

EPA is also requiring that vehicles and engines used for the testing of F/FAs be unaltered from original equipment manufacturer (OEM) specifications (with the exception of modifications in aftertreatment devices as described in Section VI.A). Rebuilds and alteration kits will only be allowed upon EPA's approval, when a F/FA manufacturer demonstrates to EPA that OEM equipment suitable for their F/FA product's testing is unavailable.

As proposed in the NPRM (57 FR 13192-13193), vehicle and engine selection must follow the criteria outlined in Sec. 79.57(a) of the accompanying regulations. The selection method is described in detail in a memorandum entitled, ``Vehicle Selection Procedures for the Proposed Fuels and Fuel Additives Rulemaking" (see Item No. II-B-6 in the public docket of this rule). As proposed, the final rule does not differentiate between light-duty vehicles and light-duty trucks. Thus, vehicles/engines are separated into two classes: light-duty and heavy-duty. The vehicle or engine selected must be a new vehicle or engine of the model year in which testing begins. However, vehicle selection criteria are to be based on technology characteristics of the previous model year. Any one of the top five selling models (based on sales figures from the year prior to testing) with the appropriate technology in a fuel group may be chosen. Each test vehicle or engine must be equipped with all of the normally required and functioning emission control equipment, with the exception of aftertreatment devices, when applicable (see Section VI.A).

Considering the practical constraints of the rule, EPA is requiring that only one vehicle or engine model be used to generate emissions for these tiers. Although EPA recognizes that emission composition is somewhat dependent on vehicle models and may even vary in replicate tests of the same vehicle/fuel combination, the use of untreated exhaust in the testing program will greatly reduce the significance of these potential sources of variability. The purpose of the testing program is to determine potential health effects of F/FA emissions and not to establish in-use fleet average emission

levels for different types of vehicles. However, EPA reserves the right to require the testing of F/FAs in additional vehicles or engines, under Tier 3, if there is concern for technology-based differences in toxicological effects. Furthermore, EPA could require the use of catalyzed exhaust to perform tests under Tier 3.

Although EPA is routinely requiring only one vehicle or engine for the testing of F/FAs, EPA foresees that at least one backup vehicle/engine of the identical model may be needed to replace vehicles/engines that wear out or malfunction during the course of testing. The probability of needing a replacement vehicle or engine increases in the case of testing F/FAs containing atypical elements that require additional mileage accumulation (see next section). The decision concerning the timing of vehicle and engine replacements is the responsibility of the F/FA manufacturer seeking registration. EPA recommends that backup vehicles/engines (if present) accumulate mileage along with the primary test vehicle, so as to minimize testing interruptions if the backup vehicle/engine is needed. Manufacturers may, at their own discretion, alternate between backup vehicles (or engines) during testing to further decrease the probability of problems or interruptions. Similarly conditioned vehicles/engines (i.e., primary and backup vehicle/engine) would be expected to generate comparable emissions. Emissions from backup vehicles/engines must have their emissions characterized prior to use in the biological studies. Wide discrepancies between the emissions of primary and backup vehicle/engine emissions may be cause to void a test.

During emission generation, vehicles and engines must be maintained in good condition by following the OEM recommendations for service schedule and parts replacement. If unscheduled maintenance becomes necessary, the vehicle or engine must be repaired to OEM specifications, using OEM or OEM-approved parts. In addition, the manufacturer is required to measure the basic emissions (as described in Section VII.B.2.a) after the unscheduled maintenance and before resuming testing, to demonstrate that the post-maintenance emissions are within 20 percent of pre-maintenance emission levels. If the basic emissions cannot be brought within 20 percent of their previous levels, then the manufacturer must restart testing using a new vehicle or engine. Provisions in the regulations allow for a limited amount of emission generation disruption without voiding the biological test.

E. Mileage Accumulation

New vehicles (or engines) to be used in emission generation for the testing of F/FAs are required to undergo a break-in period in which the vehicle (or engine) is run exclusively on the fuel or additive/base fuel mixture to be tested. The mileage accumulation requirements of

this final rule follow the approaches discussed in the Reopening Notice. These requirements serve the purpose of stabilizing the emissions from the new vehicle or engine.

Vehicles to be used in the evaluation of baseline and non-baseline F/FAs are required to accumulate at least 4,000 miles prior to emission testing. For engines operated on an engine dynamometer, the minimum break-in requirement is 125 hours of operation for testing baseline and non-baseline F/FAs. The 4,000 mile/125 hour mileage accumulation requirements are consistent with the emission stabilization procedures used in EPA's new vehicle certification program.\39\ An intact aftertreatment device must be used when accumulating mileage in the evaluation of baseline and non-baseline F/FAs. Mileage can be accumulated in a number of ways, i.e., on a test track, on a dynamometer, on the street, or as part of a manufacturer's fleet. No specific driving cycle is required, but it must include a reasonable amount of transient operation.

\39\40 CFR 86.094-26. Mileage and service accumulation; emission requirements.

For atypical F/FAs, the minimum mileage accumulation required prior to testing is also 4,000 miles for test vehicles or 125 hours for test engines. After completion of the 4,000 mile/125 hour minimum mileage accumulation, an attempt should be made to identify and measure the atypical element(s) in the emissions. Mileage accumulation must continue until either: (1) 50 percent or more of the input mass of each atypical element is measured in the emissions (i.e., vapor, semi-volatile, and particulate combined), or (2) a maximum mileage accumulation equivalent to 40 percent of the average useful life of the applicable vehicle/engine (e.g., 40,000 miles for light-duty vehicles, 116,000 miles for heavy-duty vehicles, or engine equivalent) has been reached. When either of these conditions has been met, emission generation can begin for purposes of emission characterization or biological testing. Because the presence of atypical species in specific emission fractions will be dependent on the nature of the particular atypical element, EPA recommends examination of all emission fractions (i.e., vapor, semi-volatile, and particulate). The determination of the appropriate intervals for conducting emissions measurements is left to the manufacturer's discretion.

Manufacturers of atypical F/FAs may choose to accumulate the required mileage using a vehicle/engine equipped with either an intact aftertreatment device or with a non-functional aftertreatment device (e.g., a blank catalyst without its catalytic wash coat).\40\ However,

the sampling and analysis of emissions for detecting the atypical element(s) of interest, prior to emission characterization or biological testing, must be done with a non-functional aftertreatment device. A brief period of warm-up driving (i.e., 10 miles or equivalent time) needs to precede the sampling for the detection of atypical element(s).

\40\If the manufacturer chooses to accumulate mileage without a functional aftertreatment device, and if the manufacturer wishes to do this outside of a laboratory/test track setting, then a memorandum of exemption for product testing must be obtained by applying to the Director of the Field Operations and Support Division [see Sec. 79.51(e)(6)(iv) of this rule].

During the mileage accumulation period for the testing of any fuel or fuel additive, vehicles and engines used for emission generation must be maintained in good condition by following the recommended maintenance practices in the appropriate vehicle or engine owner's manual. Maintenance requirements were described in the previous section.

F. Special Requirements for Additives

This section describes provisions for the testing of specific types of additives that might require modification in emission generation requirements. These include additives used in conjunction with aftertreatment devices, additives used infrequently, and diesel additives produced exclusively for use in diesel #1 fuels.

As explained in the above sections, EPA is requiring that emissions used for Tier 1 and Tier 2 testing be generated from vehicles/engines with non-functional aftertreatment devices. In response to comments received on the Reopening Notice, however, EPA is including in this final rule a special allowance for specific types of additives that are designed to work in conjunction with aftertreatment devices. In the case of fuel additives specifically intended to enhance the effectiveness of exhaust aftertreatment devices, the related aftertreatment device may be used on the emission generation vehicle/engine during all mileage accumulation and testing.

Regarding infrequently used additives, EPA recognizes that some aftermarket additives are intended by the manufacturer (as stated in the additive's instructions for use) to be added to the fuel tank only at infrequent intervals. During mileage accumulation, these types of additives may be applied according to the manufacturer's

specifications. However, during emission generation and testing, EPA requires that each tankful of fuel used contain the fuel additive at its maximum recommended level. In the case of bulk additives used intermittently for the direct purpose of conditioning or treating a fuel during storage or transport, or for treating or maintaining the storage, pipeline, and/or other component of the fuel distribution system (and not the vehicle/engine for which the fuel is ultimately intended), EPA also requires that the additive be added to the base fuel at the maximum concentration recommended by the additive manufacturer for treatment of the fuel or distribution system component. If the manufacturer of infrequently used aftermarket or bulk additives (as described above) is concerned that the test vehicle/ engine may be adversely affected and/or the emissions may be subject to artifacts due to overuse of these types of additives, then that manufacturer may submit a request for a modification in test procedure requirements. Any such request must include test data (e.g., emission characterization data) to support the claim that procedural modification is needed, as well as a suggested substitute procedure.

In order to simplify diesel additive testing and allow comparability between the test results of all diesel additives evaluated in this program, EPA requires that all diesel additives (including those produced exclusively for use in #1 diesel fuels) be tested on the #2 diesel base fuel (specified in Section V). If a manufacturer is concerned that the emissions generated using a blend of their #1 diesel fuel additive with the #2 diesel base fuel may be subject to artifacts due to this blending, then that manufacturer may submit a request for a modification in test procedures. Any such request must include supporting data (e.g., emission characterization data) and suggested test modifications.

VII. Tier 1 Requirements

The scope of Tier 1 encompasses: (1) a literature search for available information on the composition and effects of F/FA emissions on public health and welfare, (2) a chemical analysis to characterize the emissions of fuels or additive/base fuel mixtures, and (3) a qualitative discussion of potential exposures using information on total production volume and market distribution patterns of the particular fuel(s) or additive/base fuel mixture(s).

A. Literature Search

1. Scope

The registration program requires F/FA manufacturers to conduct a comprehensive data search that will

include all relevant existing information concerning previous emission characterization and health effects and welfare studies. The data search must address the chemical composition and potential adverse effects of whole combustion emissions, relevant combustion emission fractions (e.g., particulate phase), and whole evaporative emissions, as applicable. The literature search must also address each of the individual combustion and evaporative (where different) emission products identified by the required emission speciation procedures, with the exception of carbon monoxide, carbon dioxide, nitrogen oxides, benzene, 1,3-butadiene, acetaldehyde, and formaldehyde. Special literature search requirements for non-baseline and atypical F/FAs are described in Section VII.A.3.

Information considered applicable to a given fuel or additive includes data obtained from the testing of emissions from the fuel or additive in question or from other similar products. For this purpose, ``similar'' products are those which meet the criteria for enrollment in the same F/FA group as the subject fuel or additive, pursuant to the grouping system criteria discussed in Section IV. F/FA manufacturers who choose to participate in the grouping system may pool information about all member products for purposes of their joint submission and may also make use of available data on other products which are not enrolled in the group but share the designated formulation characteristics of group members. Similarly, a manufacturer who chooses not to participate in the grouping system could include any test results which may be available for products which could theoretically be assigned to the same group as the manufacturer's own product.

The survey on health effects studies is not restricted to the particular endpoints and experimental protocols included in Tier 2. Studies using other scientifically acceptable methods or protocols addressing all health effects of F/FA emissions must also be included in the Tier 1 report. Most often, data will be available from experiments conducted with laboratory animals, but other applicable studies must also be considered. Evidence for potential toxicity or lack of toxicity in exposed humans may be available from epidemiological studies, clinical studies, occupational exposures, or case reports. In general, referenced experiments must be concerned with the health effects of inhalation exposure to F/FA emissions (combustion and evaporative). However, data collected from relevant studies using other routes of exposure must also be included. Available results from in vitro tests, comparative metabolism studies, and structure-activity analyses are also considered relevant and must be included in the summary report for health effects of F/FA emissions.

The data search must include available literature on welfare effects, including, but not limited to, the exposure and response of plants and animals to whole emissions and individual components of

emissions, the potential for bioaccumulation, and the concentration and persistence of emission products in the air, soil, and water. Available results of exposure modeling analyses, environmental and atmospheric fate modeling studies, field studies, monitoring studies, accident evaluations, or environmental simulation experiments must be included to characterize potential exposures and the environmental impact of F/FA emissions. Specific ecological studies addressing the potential environmental effects of F/FA emissions on vegetation, livestock, wildlife, aquatic species, and soil organisms must be included. In addition, the data search must address the welfare effects of F/FA emissions concerning their contribution to odor and visibility nuisances.

Both public and in-house available sources must be included in the literature survey. Information on the health and environmental effects of
F/FAs is to be compiled from peer- reviewed scientific journals and other literature as well as internal industry studies, government-sponsored reports, proceedings of scientific meetings, and other documented sources. In general, EPA will place greater confidence in studies that have been subject to peer review. A search of appropriate commercially available chemical, toxicological, and environmental data bases must be conducted to obtain information from published sources. An example list of commercially available data bases that may be used to obtain information on potential health and environmental effects, as well as environmental fate data, is available in the public docket of this rule.\41\

\41\See memorandum from Ines del C. Figueroa to Docket A-90-07 (Item No. IV-B-03) regarding ``List of Data Bases."

In the NPRM, EPA proposed that literature searches cover at least fifteen years. However, in response to public comments, EPA has increased this time period to cover at least thirty years prior to the date of submission, so that important information from earlier testing will not be omitted. In addition, literature searches must be current as of six months prior to the beginning of testing. The thirty years are not meant to be an absolute limit for data collection. EPA encourages F/FA manufacturers to do a comprehensive search that will include all relevant available information, regardless of the age of the data.

The information to be submitted to EPA as a result of the data search includes the following items: (1) Brief text summary of the general findings and conclusions, including references, (2) a printed

copy of the outputs from the data base searches, including reference list and associated abstracts, (3) complete documentation in scientific journal format of unpublished in-house or other privately-conducted studies, and (4) tables summarizing the protocols and results of all cited studies, organized by health or environmental endpoint and type of emissions (e.g., whole combustion emission, individual emission product). In addition, the person(s) or contractor(s) conducting the literature search and summary must be identified. Further discussion on the reporting requirements of this final rule is included in Section XII.

2. Adequate Existing Information

The primary purpose of the literature search is to provide EPA with a comprehensive survey of the available data on health and welfare effects of F/FAs. A secondary function of the literature search is to enable F/FA manufacturers to document the extent to which the emission characterization in Tier 1 and/or the evaluation of health effects included in Tier 2 have already been addressed by previous adequate testing and/or analysis. If adequate testing/analysis exists, F/FA manufacturers may submit such previous data in compliance with the requirements of the registration program. For example, if previous emission characterization studies addressing the speciation requirements of this program are available in the literature, then F/FA manufacturers may submit those studies in lieu of new characterization tests. Similarly, F/FA manufacturers could use the literature search to determine the availability of adequate biological tests in compliance with Tier 2 requirements.

To satisfy the testing requirements of Tier 1 and/or 2 with previously conducted studies, reports of such previous tests must be sufficiently detailed to allow EPA to judge the adequacy of protocols, techniques, experimental design, statistical analyses, and data interpretation. Documentation must be sufficient to determine if the previously conducted studies were performed in a manner consistent with generally accepted scientific principles, good laboratory practices, and the specific testing guidelines in question. The age of the data will be considered but will not be the ultimate determining factor in deciding if an existing study is adequate. Although changes in technological approaches and methodology might preclude the use of some older studies, EPA recognizes that older literature can be useful for the purposes of this program. Thus, the quality of the study will be the deciding factor in determining the adequacy of existing studies, not the age per se. Additional criteria to be used in determining the adequacy of existing data/studies in relation to Tier 2 compliance are provided in Section VIII.C.

3. Special Requirements for Non-Baseline and Atypical F/FAs

EPA recognizes that many of the individual chemical species that will be present in the emissions of non-baseline and atypical F/FAs will also be present in the emissions of baseline products in the same fuel family. Non-baseline formulations, as defined in this rule, contain the same elements as baseline formulations. Thus, on a qualitative basis, the emission products from non-baseline F/FAs are expected to overlap with those of baseline F/FAs in the same fuel family. For atypical F/FAs, the main differentiating characteristic is the presence of atypical element(s) which are not included in the baseline category for a particular fuel family. The composition of the emissions for atypical products, therefore, is expected to consist mainly of those species present in the emissions of baseline F/FAs (for the same fuel family), with the addition of compounds which host the specific atypical element(s) of interest.

In addition to requiring literature data on the potential health and welfare effects of the whole combustion and evaporative (where different) emissions of the particular F/FA product, Tier 1 also specifies that a literature search be conducted on each of the emission products of the tested fuel or additive/base fuel mixture. Because of the substantial overlap in the emission species of F/FAs in different categories within the same fuel family, however, this requirement could result in significant duplication of effort and waste of resources. To avoid this outcome, as authorized under CAA section 211(e)(3)(C), this final rule allows manufacturers of non-baseline and atypical F/FAs to limit the literature search done for individual emission species to only those compounds which are different from the compounds typically present in the emissions of baseline F/FAs for the same fuel family.

In order to take advantage of this reduction in requirements, manufacturers of non-baseline or atypical F/FAs must compare the emission characterization results of their products with emission characterization data for baseline F/FAs. Such data may be available from private sources, in-house testing, or from publicly available literature or data bases. For example, emission characterization data for baseline gasoline are expected to be available in published literature from studies sponsored by the Auto/Oil Program.\42\ The data base ``SPECIATE'' might also be useful in identifying baseline emissions species for gasoline.\43\ Other applicable literature on gasoline and diesel emissions can be obtained in the NRC Report on ``Feasibility of Assessment of Health Risks from Vapor-Phase Organic Chemicals in Gasoline and Diesel Exhaust."\44\ Emission characterization data for alternative fuels is available in a variety of CARB reports.<SUP>45,46

\42\An example reference is ``The Auto/Oil Air Quality

Improvement Research Program SP-920," (published by SAE, Inc., February 1992). Similar information may be obtained from other Auto/Oil publications.

\43\``SPECIATE--VOC/PM Speciation Data Base Management System," Version 1.5, EPA-454/C-93-013, October 1992. This data base can be obtained electronically from the CHIEF Bulletin Board System (modem phone no. 919-541-5742). For information on this data base, call 919-541-5285 (INFO CHIEF).

\44\Published by National Academic Press, Washington, DC, 1983 (see Appendix A of Report).

\45\``Definition of Low-Emission Motor Vehicle in Compliance with the Mandates of Health and Safety Code Section 39037.05," CARB, May 19, 1989.

\46\``Proposed Reactivity Adjustment Factors for Transitional Low-Emission Vehicles," Technical Support Document, CARB, September 27, 1991.

B. Characterization of Emissions

1. Scope

The chemical analysis requirements of Tier 1 satisfy the provision in CAA section 211(b)(2)(B) requiring information ``to determine the emissions resulting from the use of the fuel or additive contained in such fuel." The characterization of emissions in Tier 1 will provide a useful inventory of potentially harmful F/FA emission products for further study and evaluation in support of the F/FA testing program, risk assessments, and future regulatory actions.

F/FA manufacturers are responsible for the generation, collection, and sampling of the combustion and, if applicable, the evaporative emissions of their F/FAs, and for the conduct of tests to determine the identity and concentration of individual emission products. In general, the required procedures are directed toward the detection and measurement of selected chemical classes and compounds. The analyses include: (1) the measurement of basic emissions (i.e., total hydrocarbons, carbon monoxide, oxides of nitrogen, and particulates), (2) the speciation of volatile hydrocarbon compounds, aldehydes, ketones, alcohols, ethers, and polycyclic aromatic compounds, and (3) the speciation of atypical emission products (when atypical elements are known to be present in the raw fuel or additive formulation). Speciation requirements are summarized in Table 1.

Table 1.--Emission Characterization/Measurement Requirements

	Speciated Emissions

Emission Type	Basic Emissions<SUP>a	Polycyclic Hydrocarbons	Ketones and Aldehydes	Alcohols and Ethers<SUP>b	Aromatic Compounds<SUP>c	Atypicals<SUP>d
Combustion emissions:						
Vapor phase.....	X	X	X	X	.....	X
Semivolatile phase.....	.....	.....	.....	.....	X	X
Particulate phase.....	X	.....	.....	.....	X	X
Evaporative emissions:<SUP>e						
Evaporative emission generator<SUP>f.....	X<SUP>g	X	.<SUP>.....<SUP>...		X	.<SUP>.....<SUP>... X

<SUP>aBasic emissions=total hydrocarbons, carbon monoxide, oxides of nitrogen, and particulates (see Section VII.B.2.a below).

<SUP>bRequired if alcohols or ethers exist in the uncombusted fuel or additive/base fuel mixture.

<SUP>cIncludes specific polycyclic aromatic hydrocarbons (PAHs), nitrated polycyclic aromatic hydrocarbons (NPAHs), and poly-chlorinated dibenzodioxins/dibenzofurans (PCDD/PCDFs). PAH and NPAH speciation is not required for F/FAs in the methane (CNG, LNG) and propane (LPG) families, or for F/FAs in the atypical categories of other fuel families. Chlorine-containing atypical F/FAs are subject to the dioxins/furans speciation requirements.

<SUP>dManufacturers of atypical products must examine all emission fractions for the measurement and identification of potential atypical species.

<SUP>eOnly applicable to F/FAs required to measure evaporative emissions.

<SUP>fEvaporative emissions are to be generated using an evaporative emission generator as described in Section VI.C.

<SUP>gThe only basic emission required to be measured for evaporative emissions is total hydrocarbons.

2. Speciation Procedures

Section VI describes the required procedures for the generation of both combustion and evaporative emissions. Characterization of combustion emissions must be done both for non-catalyzed emissions and

for tailpipe emissions. As discussed in Section III.A.2, the evaporative emissions of some F/FAs are also required to undergo emission characterization analysis. To provide an indication of the variability, the emissions must be generated and characterized three times on three different days. Collection and speciation of background samples is required.

The CAA authorizes EPA to require information to characterize F/FA emissions, while giving EPA discretion to specify the particular protocols to be used for this purpose. The following sections identify the general emission product categories of interest and discuss currently available protocols which are suitable for their analyses. EPA recognizes that scientific methods can be expected to advance in the future. Thus, the use of the protocols referenced in this final rule is not mandated. Rather, EPA will hold F/FA manufacturers accountable for state-of-the-art methods and good analytical chemistry and laboratory practices, such as those described in the article ``Principles of Environmental Analysis."\47\

\47\Keith et al., ACS Committee on Environmental Improvement, ``Principles of Environmental Analysis," The Journal of Analytical Chemistry, Volume 55, pp. 2210-2218, 1983; available in Docket A-90-07, Item No. II-J-12.

Today's rule does not discourage the use of any validated method to perform the characterization of emissions, or the submittal of existing speciation results obtained from validated methods, as long as the data address the speciation requirements of the F/FA registration program. EPA acknowledges the state-of-the-art methods of the Auto/Oil Air Quality Improvement Research Program (Auto/Oil Program) for the characterization of emissions. In fact, the speciation requirements included in this rule for fuels composed primarily of hydrocarbon compounds of twelve carbons (C12) or less (e.g., gasoline) are based on such methodology.<SUP>48, 49, 50 Where applicable, EPA will accept results from the Auto/Oil Program as adequate data in lieu of new testing. However, the Auto/Oil Program might not address all the emission characterization requirements of today's rule, so additional procedures (e.g., for the analysis of polycyclic aromatic compounds) might be needed. EPA recognizes that characterization data have already been submitted to EPA in relation to the Auto/Oil Program. F/FA manufacturers need not resubmit this information, but are required to reference these data (e.g., report number, applicable page numbers, etc.) on the Tier 1 report so EPA can verify the adequacy of the information being used in compliance with the F/FA registration program

for the particular
F/FA product or group representative. Although resubmission of the raw emission data is not required, manufacturers are still responsible for providing a summary discussion of the emission characterization results in the Tier 1 report as outlined in Section XII.B.

\48\Jensen, T. E. et al., ``Advanced Emission Speciation Methodologies for the Auto/Oil Air Quality Improvement Program--I. Hydrocarbons and Ethers," SAE 920320 In: Auto Oil Air Quality Improvement Research Program, SP-920, February 1992.

\49\Swarin, S. J. et al., ``Advanced Speciation Methodologies for the Auto/Oil Air Quality Improvement Research Program--II. Aldehydes, Ketones, and Alcohols," SAE 920321, In: Auto Oil Air Quality Improvement Research Program, SP-920, February 1992.

\50\Siegl, W. O. et al., ``Improved Emission Speciation Methodology for Phase II of the Auto/Oil Air Quality Improvement Research Program--Hydrocarbons and Oxygenates," SAE 930142, 1993.

For the characterization of diesel
F/FAs, EPA recognizes the procedures under the Air Pollution Research Advisory Council (APRAC) program. The work done by APRAC provides speciation guidelines for unregulated diesel emissions and addresses diesel combustion compounds of concern to EPA. As with the Auto/Oil Program studies, existing applicable APRAC speciation studies will be considered adequate data in lieu of new testing. However, today's rule requires manufacturers of diesel F/FAs to perform speciation procedures for hydrocarbons which might not be included in the APRAC program. These are discussed in the following sections.

a. Characterization of Basic Emissions. EPA proposed to require the characterization of ``regulated emissions" for fuel/vehicle types for which certification procedures existed at the time of the publication of the NPRM (i.e., gasoline, diesel, and methanol). Today, EPA is terming this requirement ``basic emissions," instead of ``regulated emissions," because certification requirements are not established yet for all the F/FAs included in this rule. To be consistent and avoid confusion, the term ``basic emissions" is used for all F/FA families included in this rule. Based on the current regulated emissions and taking into consideration the objectives of this program, EPA selected four basic emissions for measurement, as follows: Total hydrocarbons, carbon monoxide, oxides of nitrogen, and particulates.

The four basic emissions are to be measured in combustion emissions as a routine requirement for all F/FA families, as shown in Table 1. Only total hydrocarbons are required to be measured in evaporative

emissions. Manufacturers are referred to the vehicle certification procedures in 40 CFR part 86 for general guidance on the measurement of the basic emissions of interest to this rule.

b. Characterization of Hydrocarbons. As shown in Table 1, this rule requires the speciation of hydrocarbons for the vapor phase of combustion emissions and for evaporative emissions generated using an evaporative emission generator. The speciation is to be performed using methods that identify and determine the concentration of all hydrocarbon compounds containing twelve or fewer carbon atoms. The Auto/Oil Program procedures referenced above provide an acceptable speciation method for hydrocarbons.

c. Characterization of Aldehydes and Ketones. Speciation of aldehydes and ketones containing a maximum of eight carbon atoms is required only for the vapor phase of combustion emissions. A test procedure for formaldehyde measurement is included in 40 CFR part 86 for formaldehyde. F/FA manufacturers are also referred to the Auto/Oil Program procedures referenced above for the analysis of aldehydes and ketones. Additional applicable procedures are available in ASTM D 5197-91, ``Standard Test Method for Determination of Formaldehyde and Other Carbonyl Compounds in Air (Active Sampler Methodology)."

d. Characterization of Alcohols and Ethers. Alcohol and ether compounds containing six or fewer carbon atoms are to be characterized for both evaporative and combustion emissions, whenever the fuel or additive/base fuel mixture under evaluation contains alcohols or ethers. If a F/FA formulation contains an alcohol or ether with more than six carbon atoms, then this manufacturer is required to measure their presence in the emissions, as well as alcohols or ethers with fewer number of carbon atoms. For example, if an ether containing seven carbon atoms (e.g., isopropyl tertiary butyl ether) is part of a fuel formulation being tested in this program, then its manufacturer must characterize ethers with seven or fewer carbon atoms.

In addition to the Auto/Oil Program procedures referenced above, a test procedure for the characterization of alcohols and ethers is described in 40 CFR part 80, Appendix F, entitled ``Test Method for Determination of C1-C4 Alcohols and MTBE in Gasoline by Gas Chromatography". This procedure can be used for the identification of ethers in addition to MTBE, but will require appropriate modifications for application to gas phase samples.

e. Characterization of Polycyclic Aromatic Compounds. In the NPRM, EPA proposed a broad requirement for the identification and measurement of polycyclic aromatic compounds. In the final rule, this requirement is narrowed to a limited number of specified compounds which are of significant concern in terms of their potential non-carcinogenic and/or carcinogenic effects. Included are specified polycyclic aromatic hydrocarbon (PAH) and nitrated polycyclic aromatic hydrocarbon (NPAH)

compounds as well as individual compounds and classes of polychlorinated dibenzodioxins/dibenzofurans (PCDD/PCDFs).

In addition to specifying particular polycyclic aromatic compounds for analysis, the final rule reduces the families and/or categories of F/FAs which are subject to these requirements. PAH and NPAH speciation need not be done for F/FAs in the methane and propane fuel families, nor for F/FAs in the atypical categories of other fuel families. Furthermore, speciation of dioxins/furans is required only for F/FAs which contain chlorine as an atypical element. This is consistent with the requirement applicable to atypical F/FAs in general, that all emission species containing the relevant atypical elements be identified and measured (see section f, below).

While EPA believes that characterization of dioxins/furans is also important in the case of baseline and non-baseline F/FAs, the NPRM did not propose to require this procedure on a wider basis. EPA has thus refrained from including mandatory requirements for speciation of dioxins/furans in the case of baseline and non-baseline F/FAs. Instead, for manufacturers of F/FAs other than chlorine-containing atypical F/FAs, dioxin/furan characterization is included in the final rule only on a voluntary basis. EPA strongly encourages manufacturers of baseline and non-baseline F/FAs to collect the necessary emission samples and conduct these voluntary procedures at the same time that mandatory emission characterization requirements are being fulfilled. The recent attention and concern about the potential health effects of dioxins/furans, combined with the current dearth of information on the specific sources and generation of these compounds, increases the likelihood that these procedures will be prescribed under EPA's discretionary Tier 3 authority if the necessary data are not otherwise submitted on a voluntary basis. If so, the incremental costs are likely to be considerably higher than if the procedures were conducted in conjunction with the standard Tier 1 emission characterization tasks.

As was proposed, the final rule requires the measurement and speciation of polycyclic aromatic compounds in both the semi-volatile phase and particulate phase of combustion emissions. While, in the past, these compounds have been analyzed primarily in the particulate phase, the quantity of these compounds in the semi-volatile phase at the temperatures encountered in dilute exhaust may also be important.

Particulate and semi-volatile phase emissions are to be collected using methods described in Section VI.B.1. The soluble organic fraction (SOF) is to be extracted from the filter and polymer bed separately. The extracts of the two phases are to be tested separately for PAHs and NPAHs, but may be combined before testing for dioxins/furans. Examples of protocols suitable for characterizing polycyclic aromatic compounds are available in the literature.<SUP>51, 52, 53, 54, 55, 56

\51\Coordinating Research Council, ``Chemical Methods for the Measurement of Unregulated Diesel Emissions," CRC Report No. 551, 1987; available in Docket A-90-07, Item No. II-J-15.

\52\Tejada, S.B., ``Fluorescence Detection and Identification of Nitro Derivatives of Polynuclear Aromatic Hydrocarbons by On-Column Catalytic Reduction to Aromatic Amines," Analytical Chemistry, Volume 58, Number 8, pp. 1827-1834, July 1986.

\53\Tejada, S.B. et al., ``Analysis of Nitroaromatics in Diesel and Gasoline Car Emissions," SAE Paper No. 820775, 1982.

\54\Schuetzle D., ``Analysis of Nitrated Polycyclic Aromatic Hydrocarbons in Diesel Particulates," Analytical Chemistry, Volume 54, pp. 265-271, 1982.

\55\John J. H. et al., ``A review of diesel particulate control technology and emissions effects--1992 Horning Memorial Award Lecture," SAE Technical Paper Series No. 940233, 1994.

\56\A protocol for identification and measurement of poly-chlorinated dibenzodioxins and dibensofurans is provided in 40 CFR part 60, Appendix A, Method 23.

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f. Characterization of Emissions with Atypical Elements. F/FAs containing chemical elements other than those included in the baseline formulations for the respective fuel family are classified as atypical formulations (see Section IV.B.2). In addition to the emission characterization requirements described above, producers of atypical F/FAs are required to identify and measure the emission products containing the associated atypical element(s). For example, if a gasoline additive product contains chlorine, then this manufacturer must identify and measure all emission compounds that contain chlorine. Due to the nature of atypical products, special procedures for the generation of emissions are required (see Section VI.E).

The presence of atypical species in specific emission fractions will be dependent on the nature of the particular atypical element/compound of concern. In view of this, EPA recommends that manufacturers of atypical products examine all emission fractions (i.e., vapor, semi-volatile, and particulate) for the measurement and identification of potential atypical species. Because of the variety of potential elements and reaction products involved, all of the necessary chemical/analytical procedures cannot be specified in this final rule. The selection of the particular method(s) for measuring atypical elements or compounds is left to the manufacturer. However, the procedures used must be state-of-the-art and based on sound analytical chemistry principles applicable to the atypical element or compound of concern.

3. Quality Assurance

While today's rule requires emissions to be generated and characterized three times as a way to evaluate the repeatability of the test results, additional quality assurance procedures are needed to control variability during the characterization of emissions. Laboratories conducting emission characterization/speciation analyses are required to perform verification testing to examine the repeatability and accuracy of test procedures. For this purpose, a prepared mixture of chemical compounds, as appropriate for each particular procedure, should be subjected to the speciation protocols. The use of analytical standards and controls for calibration of instruments is also required to assure precision and accuracy of results.

EPA reserves the right in this final rule to audit testing facilities involved in the generation and characterization of emissions, as well as the health effects testing of F/FAs. Such audits will be organized and administered by EPA at its own expense. The audit procedures could include a requirement that facilities submit a completed questionnaire in which equipment and procedural information is described. EPA might make recommendations based on the submitted information and/or might follow up with a visit to observe the performance of the protocols. The audit could also include EPA distribution of ``blind'' samples for analysis at participating laboratories (at their expense). The audit will not have the purpose of certifying that the laboratory is ``EPA approved''. Rather, it will have the purpose of determining the weaknesses of laboratories and the acceptability of the laboratory's current performance.

C. Exposure Analysis

In the NPRM, EPA proposed to require modeling or other analytic methods to evaluate potential exposures, expected atmospheric reactivity, and environmental partitioning of emission products. However, as discussed in Section III.C., this final rule does not require modeling analyses to be performed as routine requirements under Tier 1. Instead, quantitative modeling efforts will be required on a case-by-case basis as needed under Tier 3.

Nevertheless, EPA believes that exposure data are still critical for the assessment of the potential risks associated with the emissions of F/FAs in question. For this purpose, today's rule requires manufacturers to provide a qualitative discussion of potential population exposures based on the production and use of the particular fuel or additive (or group of F/FAs) in question. This qualitative analysis must consider the actual and/or projected total annual production volumes and the market distribution patterns (e.g., percent

of sales by state or region) of the particular product or group of products. Group submissions must assess the cumulative exposure resulting from all members of the group. A quantitative analysis is encouraged when appropriate data are available, including any existing modeling data, to support the exposure analysis. As discussed earlier, EPA retains the authority to require from manufacturers more exhaustive exposure analysis for particular products of concern under Tier 3 (including modeling), based on the EPA evaluation of Tier 1 and Tier 2 results or other available information.

VIII. Tier 2 Requirements

In the NPRM, EPA proposed short-term (42-day) tests under Tier 2 for the evaluation of six health effects endpoints: carcinogenicity, mutagenicity, teratogenicity, reproductive toxicity, neurotoxicity, and pulmonary toxicity. EPA examined the proposed Tier 2 program and found that similar requirements among the various proposed tests (in regard to animal subjects, exposure scenarios, and general technical principles) provided the opportunity to combine several endpoint tests within the same exposure protocol. In view of this, EPA has modified the Tier 2 testing program to allow for concurrent test performance in a more cost-effective manner.

The revised Tier 2 testing program enhances efficiency and feasibility, while providing better health effects information. In fact, the design of the Tier 2 testing program makes best use of animals (minimum number of animals used), laboratory capacity, and financial resources. The basic Tier 2 testing framework of this final rule consists of a 90-day subchronic inhalation study to examine general systemic and organ toxicity (including pulmonary effects), with the addition of ancillary tests that allow the assessment of several specific health effect endpoints (carcinogenicity, mutagenicity, teratogenicity, reproductive toxicity, and neurotoxicity) within the same exposure schedule. A fertility assessment is coordinated with the 90-day study to examine reproductive and teratogenic effects.

Brief descriptions of test guidelines for the evaluation of each health effect endpoint are provided in the sections below. Most of these testing guidelines are modified versions of guidelines previously published under TSCA (40 CFR part 798, revised as of July 1, 1992) and/or the test guidelines which accompanied the NPRM. Detailed protocols for the Tier 2 testing program are included in Sec. 79.62-Sec. 79.68 of the accompanying regulations. Figure 4 shows a diagram of the suggested timing and organization of the Tier 2 studies within the general 90-day subchronic exposure schedule.

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A. General Methodology

1. Exposure Duration

As mentioned previously, EPA originally proposed a program that included six separate tests for the evaluation of the endpoints of concern. Because of cost considerations, a minimum six-week (42-day) exposure period was proposed in the NPRM, instead of the traditional 90-day test. EPA requested comments on the adequacy of the proposed exposure period and exposure regimen, and on the possibility of extending these tests to 90 days for comparability to historical data. Comments were also requested on the possible use of an alternative approach for the testing program, the Screening Information Data Set (SIDS) protocol developed for use by the Organization for Economic Cooperation and Development (OECD). The standard SIDS protocol is designed as a single-study screen (45-day) for repeat dose, reproductive, and developmental effects.

Public comments from industry supported the 90-day test over either the proposed 42-day test or the SIDS protocol. Commenters expressed concern regarding both of these protocols because relatively less scientific experience and historical comparison data are available for these shorter test scenarios. Upon reconsideration, EPA agrees that the 90-day protocol is more suitable and more cost effective than either of the alternatives proposed in the NPRM for purposes of the F/FA registration program. EPA recognizes that the shorter tests might miss some adverse health effects that might be identified with the 90-day exposure. Apart from the study duration, the SIDS protocol includes somewhat less information for each endpoint, in comparison with the selected 90-day subchronic inhalation study, with relatively small savings in time and cost. For example, the SIDS protocol includes a fertility screen (i.e., one-generation reproductive study), but does not include an examination of the fetus for teratogenic effects.

Thus, today's final rule requires F/FA manufacturers to use the more standard 90-day subchronic inhalation protocol for the evaluation of health effects, with an exposure regimen of at least six hours per day, five days per week. EPA judges the exposure regimen of five days per week for the 90-day protocol to be the minimum acceptable exposure period for the purposes of the Tier 2 evaluation. Also, the 90-day protocol provides a broad and efficient testing approach that allows the evaluation of several endpoints at the end of the same exposure period using the same exposed test animals. As a result, the modified

Tier 2 program provides a reduction in the number of animals needed to perform the evaluation of the endpoints of concern and savings on emission generation and testing costs.

2. Animal Model and Laboratory Practices

In general, the Tier 2 testing program requires the exposure of live laboratory animals to whole F/FA emissions. Rodent species are required and rats are specifically recommended. Animal facilities must be operated in compliance with the ``Guide for the Care and Use of Laboratory Animals" (U.S. DHHS Publication (NIH) 86-23, 1985). To ensure the quality and integrity of test results, the performance of all studies will be required to conform with good laboratory practice (GLP) standards. GLP standards specific to this rule are included in Sec. 79.60. As proposed in the NPRM, the GLP standards are based on those published in 40 CFR part 792 (revised as of July 1, 1992) for conducting tests under TSCA, with modifications to accommodate the specific goals of this rule. The GLP standards address facility, equipment, organization, quality assurance, and personnel requirements, as well as specifications for proper care of laboratory animals, handling of test substances, instrumentation issues, conduct of studies, record keeping, and reporting of results.

3. Exposure Route and Concentrations

With the exception of the Salmonella assay, the Tier 2 testing program is based on the inhalation exposure of laboratory animals to diluted whole emissions. Such studies require an exposure system designed to ensure the controlled generation, dilution, and delivery of F/FA emissions to the laboratory animals for prolonged periods. Section VI describes the methodology for the generation of F/FA combustion and evaporative emissions and the procedures to deliver the emissions to the test animals. Requirements for hardware, maintenance, and the use of emission generation and inhalation systems are included in Sec. 79.57 and Sec. 79.61 of this rule.

Before testing the emissions of a particular fuel or additive/base fuel mixture, a manufacturer must determine an appropriate range of exposure concentrations to be used in the characterization of potential health effects. The objective is to select exposure concentrations to determine a reasonable concentration-response curve that may predict the potential health risks associated with a particular exposure. Concentrations should be spaced to produce test groups with a range of toxic effects. In order to accomplish this, EPA requires that at least three concentration levels be used to construct the concentration-response curve. These levels should correspond to, at a minimum: (1) an overtly or highly toxic concentration, (2) a concentration having an intermediate level of toxicity, and (3) a minimally toxic or non-toxic level. The highest concentration should result in toxic effects but not produce a level of fatalities which would prevent a meaningful

interpretation of the resulting data. The lowest concentration should produce minimal or no observable toxic effects. If more than one intermediate concentration level is used, the concentrations should be spaced to produce a gradation of toxic effects. Due to the inherent toxicity of most vehicle emissions, it might be impossible to precisely select an exposure level which results in no observable evidence of toxicity, or a no- observed-adverse-effect-level (NOAEL). Thus, EPA is not requiring manufacturers to specifically achieve a NOAEL. Instead, EPA recommends that manufacturers use available scientific approaches (e.g., range-finding test and extrapolation of data results) to design the study with reasonable concentration spacing so as to improve the probability of achieving a NOAEL. In recognition of the possibility that the highest achievable exposure concentration (considering the limiting CO concentration) may also be non-toxic, provisions are made for ``limit tests,'' where appropriate. If a test at the highest achievable concentration produces no observable toxic effect(s), then a full study using three concentration levels might not be necessary (see regulatory text for specific endpoint tests).

B. Subchronic Inhalation Study and Endpoint Tests

As described earlier, the Tier 2 health effects testing program includes a 90-day subchronic inhalation study and ancillary assays/ tests for the examination of specific health effects endpoints. Specific assays or analyses for carcinogenicity, mutagenicity, teratogenicity, reproductive toxicity, and neurotoxicity may be conducted at the end of the 90-day study, coordinated with the 90-day exposure, or conducted separately.

In the NPRM, EPA proposed a separate acute pulmonary test (i.e., lung lavage assay) for the evaluation of pulmonary toxicity. However, the Tier 2 testing framework of this final rule does not require a separate test for the assessment of pulmonary effects because the 90-day subchronic study includes gross pathology and histopathology of the lungs and respiratory tract. Pulmonary effects in this final rule will be examined as part of the standard 90-day inhalation study. Positive results at the end of the exposure period for pulmonary toxicity will be indicated by abnormal gross or histopathological findings relative to appropriate control animals.

1. Subchronic Inhalation Study

The subchronic inhalation study is designed to determine a concentration-response relationship for potential toxic effects in rodents, resulting from exposure to vehicle/engine emissions over a period of 90 days. This test will provide valuable information on general systemic and target organ toxicity, including pulmonary effects. This information is considered an essential component for the

assessment of potential health hazards resulting from the exposure to F/FA emissions. The exposure period of the subchronic study (i.e., 90 days) covers approximately one tenth of the life span for the recommended test animal species (i.e., rats). Although life-shortening or tumors are not likely to be observed within the 90 days of exposure, the subchronic study should be able to identify a wide variety of adverse effects.

The specific guidelines for the subchronic inhalation study are included in Sec. 79.62 of this final rule. The subchronic study requires that animals be observed and weighed during the exposure period. Ophthalmological examination, blood chemistry analysis, organ examination, and histopathology are basic requirements of the 90-day protocol. Hematology and clinical biochemistry determinations are required to be carried out after 30 days of exposure and just prior to termination. Hematology analyses include: hematocrit, hemoglobin concentration, erythrocyte count, total and differential leukocyte count, and a measure of clotting potential (e.g., clotting time, prothrombin time, thromboplastin time, or platelet count). Clinical biochemical testing includes assessment of electrolyte balance, carbohydrate metabolism, and liver and kidney function. Other specific biochemical tests are described in the regulatory text of the final rule.

At the end of the exposure period, tissues and/or organs from a subgroup of the test animals are specially preserved according to the requirements of the neurological, pulmonary, and reproductive organ examinations. Tissues/organs from the main test population are preserved using standard techniques for the general toxicity evaluation. Test animals will be subjected to a full gross necropsy which includes examination of the external surface of the body, all orifices, and the cranial, thoracic, and abdominal cavities and their contents. All major organs must be weighed. Gross pathology must be performed on the following target organs and tissues: liver, kidneys, lungs, adrenals, brain, and gonads. Histopathology must be performed on all gross lesions and specific organs/tissues, as follows: respiratory tract (i.e., lungs, nasopharyngeal tissues, trachea), brain, heart, sternum with bone marrow, salivary glands, liver, spleen, kidneys, adrenals, pancreas, reproductive organs (i.e., uterus, cervix, ovaries, testes, epididymides), aorta, gall bladder, esophagus, stomach, intestinal tract, urinary bladder, representative lymph node, and peripheral nerve/tissue. Other organs and tissues must be preserved in a suitable medium for possible future histopathological examination, as described in the regulatory text of the final rule.

As described earlier, the 90-day subchronic inhalation study will serve as a basic framework for the Tier 2 testing program. The following sections provide brief descriptions of the ancillary assays

and other additional test and/or measures performed under Tier 2.

2. Carcinogenicity and Mutagenicity Assays

For the evaluation of carcinogenicity and mutagenicity, Tier 2 includes a battery of three genotoxic assays: Salmonella, micronucleus (MN) and sister chromatid exchange (SCE). These assays are specific for mutagenic/carcinogenic outcomes at a cellular level, but the tests may not be indicative of non-mutagenic or initiation/promotion cancer mechanisms.

In general, the Tier 2 genotoxic assays are considered cost-effective indicators of mutagenicity and, by implication, predictors of suspect carcinogens. The rationale for using these tests for the assessment of potential mutagenic and carcinogenic effects is based on the general assumption that cancer is a multi-stage process involving a variety of events that can include genotoxic steps. The general consensus among scientists is that many of the cancers may be attributable to adverse genetic changes. Because genotoxic steps are generally implicated early in the process of cancer development, their detection has assumed the status of presumptive carcinogen identification. EPA recognizes that this working assumption has some limitations because there is always a possibility of having false positive (mutagenic noncarcinogens) or false negatives (nonmutagenic carcinogens) when evaluating the carcinogenic potential of the test substance. However, these limitations do not preclude the usefulness and the effectiveness of the genotoxic assays (i.e., Salmonella, MN, and SCE) as cost-effective predictors of potential mutagenic and carcinogenic effects in the context of the Tier 2 testing program.

A description of the Tier 2 assays required for the evaluation of carcinogenicity and mutagenicity is provided in the following sections. The two in vivo assays (MN and SCE) can be conducted concurrently with the 90-day subchronic inhalation study (i.e., same animal population). The Salmonella assay is run separately, because it does not require the exposure of live animals to emissions.

a. Salmonella Assay. The Salmonella assay is an in vitro test for mutagenicity and, by implication, for carcinogenicity. The assay makes use of five mutant strains of the bacterium Salmonella typhimurium which cannot grow in a medium deficient in histidine due to an inherited inability to produce this amino acid. Exposure to mutagenic or carcinogenic substances can elicit reverse mutations, such that the bacteria regain their ability to grow in a histidine-deficient medium. In this test, bacteria will be exposed to the semi-volatile and particulate extracts of combustion emissions (see Section VI.B.2 for information on sampling of combustion emissions for testing). Test procedures for this assay have been described in previous literature.\57\ After exposure, the cells will be plated on histidine-deficient media (both with and without metabolic activation) and

incubated for a designated period of time. The number of emissions-induced mutant colonies (revertants) growing on the plates will then be compared to the number of spontaneous revertants in control cultures. The testing guidelines for the Salmonella assay are included in Sec. 79.68 of this final rule.

\57\Huisingh, J.L., et al, ``Mutagenic and Carcinogenic Potency of Extracts of Diesel and Related Environmental Emissions: Study Design, Sample Generation, Collection, and Preparation," In: Health Effects of Diesel Engine Emissions, Vol. II, W.E. Pepelko, et al. (Eds.), US EPA, Cincinnati, EPA-600/9-80-057b, pp. 788-800, 1980; available in Docket A-90-07, Item No. II-J-13.

A positive result for the Salmonella assay occurs when there is a statistically significant concentration-related increase in the number of revertants or a reproducible and statistically significant positive response for at least one of the test concentrations.

b. In Vivo Micronucleus (MN) Assay. Micronuclei are sub-cellular structures containing chromosomes and chromosome fragments not incorporated into the main nucleus during cell division. While micronuclei do form under natural conditions, exposure to potentially mutagenic or carcinogenic agents can cause an increase in micronucleated cells. In this assay, live rodents will be exposed by inhalation to the emissions of the particular fuel or additive/base fuel mixture (this assay is applicable to the evaluation of both combustion and evaporative emissions). Subsequently, erythrocytes in the bone marrow will be sampled, stained, and viewed under a light microscope. The number of erythrocytes containing micronuclei will then be counted and compared with erythrocytes from untreated animals. The use of erythrocytes in this procedure facilitates the visualization of micronuclei, since their primary nucleus is normally extruded during cell development. The testing guidelines for the MN assay are contained in Sec. 79.62 and Sec. 79.64 of this final rule.

A positive result for the MN assay is determined by a statistically significant concentration-related increase in the number of micronucleated erythrocytes or a reproducible and statistically significant positive response for at least one of the test concentrations.

c. In Vivo Sister Chromatid Exchange (SCE). SCEs are believed to be caused by chromosome strand breakage resulting in exchanges of genetic material between the halves of a chromosome ``pair" (i.e., the chromatids). While some SCEs occur normally, an increase in the frequency of such exchanges may be indicative of carcinogenic activity.

In this assay, animals which have undergone inhalation exposure to the emissions will be sacrificed (this assay is applicable to the evaluation of both combustion and evaporative emissions). Peripheral blood lymphocytes will then be isolated and cultured. The cells will be treated with a DNA base analog (bromodeoxyuridine, BrdU) and with a spindle inhibitor such as colchicine. After appropriate staining for labeled DNA, SCEs will be scored from cells arrested in the second mitotic division and the results compared with appropriate controls. Details on the SCE testing procedures are included in Sec. 79.62 and Sec. 79.65 of this final rule.

A positive result for the SCE assay is determined by a statistically significant concentration-related increase in the number of SCE or a reproducible and statistically significant positive response for at least one of the test concentrations.

3. Fertility Screen for Reproductive and Teratologic Effects

The fertility screen involves mating of test animals previously exposed (by inhalation) to F/FA emissions to examine the effects of such exposure on conception. The females will continue their exposures throughout pregnancy and will be assessed for teratologic effects on their offspring. This test is applicable to the evaluation of both combustion and evaporative emissions. The fertility assessment is coordinated with the 90-day subchronic inhalation study and replaces the separate reproductive and teratology studies originally proposed in the NPRM.

The reproductive assessment includes vaginal cell smears to track effects on the estrous cycle. Commenters discouraged the use of frequent vaginal smearing. In response to the comments, vaginal cyclicity determinations will be performed on the test subjects for two weeks prior to the start of the exposure period (for culling acyclic females), and will resume after seven weeks of exposure (for four weeks or until the female is confirmed pregnant). The assessment for reproductive effects also includes a mating trial and the weighing and histopathological examination of male and female reproductive organs (i.e., uterus, ovaries, testes, epididymis, and seminal vesicles), all of which can be performed within the context of the 90-day subchronic inhalation study.

As compared with appropriate control animals, positive results for reproductive effects include: changes in the length or stages of the estrous cycle as indicated by the vaginal cytology data, changes in reproductive organ weights, and pathological changes found during gross or microscopic examination of male or female reproductive organs. Changes in fecundity, fertility or litter size (number of normal fetuses) will also be regarded as positive results.

To assess potential teratogenic effects, each dam will be sacrificed on the day prior to normal parturition and its uterus

examined for embryonic or fetal deaths. Viable fetuses will be counted and then examined for skeletal and soft tissue anomalies. These results will be evaluated relative to the number of spontaneous embryonic or fetal deaths and abnormalities in unexposed controls. The required analysis covers specific fetal effects outcomes, i.e., in utero death, growth alteration, and structural abnormalities.

Although the NPRM proposed the use of two different species for the evaluation of teratogenic effects, today's action requires the use of only one species to satisfy the Tier 2 requirements. This modification will reduce the number of animals needed for Tier 2 tests, as requested by some of the commenters, while maintaining an adequate approach for the screening of teratology effects.

The above described combined protocol will detect fertility problems and teratogenic effects. EPA realizes that the protocol will not detect adverse effects on reproductive development that might appear in the offspring as they grow and mature (since pups are examined just before birth). However, EPA judges that the combined protocol provides a reasonable screen for both reproductive and teratogenic effects. The testing guidelines for the fertility assessment are included in Sec. 79.62 and Sec. 79.63 of this final rule.

4. Neurotoxicity Screen

EPA proposed in the NPRM to require the Functional Observational Neurotoxicity Battery (FONB) for the evaluation of neurotoxic effects. In addition to the FONB, EPA asked for comment on the possible inclusion of a biochemical assay to measure the level of glial fibrillary acidic protein (GFAP). Comments were received on both proposed tests.

The regulated industry objected to the use of the FONB on the basis of their belief that it would be likely to give artifactual test results. Commenters also expressed concern about the use of the GFAP assay. They considered it to be a sensitive marker for neurotoxicity, but discouraged its use because they perceived that adequate historical data are not available for this test. While not necessarily agreeing with either of these comments, EPA has reconsidered its original proposal regarding the specific content of the Tier 2 neurotoxicity assessment.

EPA considers the FONB to be a well-validated standard test and recognizes its extensive use in the past. In the context of this F/FA emissions testing program, however, EPA believes that, rather than requiring the FONB on a standard basis within the Tier 2 testing regimen, the FONB is more appropriately reserved for use when a neurotoxicity concern has been identified and additional testing is needed to clarify the nature and/or significance of the potential adverse effects (e.g., within Tier 3).

In regard to the GFAP assay, EPA judges that concerns about the amount of historical data are not sufficient justification to prevent its use in the F/FA program, given its high specificity and potential applicability as a screening test. This final rule thus includes the GFAP assay as an element in the Tier 2 assessment of potential neurotoxic effects.

The GFAP assay is a biochemical assay that measures the level of a major intermediate filament protein of astrocytes (cells of the supporting structure of the nervous system) from brain tissues. An increase in the GFAP level is highly specific for detecting the existence and location of chemical-induced injury to the central nervous system (CNS) associated with astrocytic hypertrophy. However, due to its specificity, the assay does not provide information on other potential sites for neurotoxic effects. To provide a more comprehensive evaluation, today's rule supplements the GFAP assay with an expanded neurohistopathology examination as part of the 90-day subchronic inhalation study. The neurohistopathology involves the examination of several brain sections, including the cerebrum, cerebellum, medulla, cervical bulb of the spinal cord, and peripheral nerves (e.g., tibial or sciatic nerve). Testing guidelines for the neurotoxicity evaluation are included in Sec. 79.62, Sec. 79.66 (neuropathology assessment), and Sec. 79.67 (GFAP) of this final rule. Positive results at the end of the exposure period for neurotoxicity will be indicated by an increase in the GFAP level and/or abnormal gross or histopathological findings relative to appropriate control animals. The neurotoxicity screen is applicable to the evaluation of both combustion and evaporative emissions.

C. Adequate Endpoint Information in Lieu of Tier 2 Tests

One of the functions of the data search requirement under Tier 1 (as discussed in Section VII.A.2) is to enable F/FA manufacturers to examine the available literature and determine if adequate data exists (for both combustion and evaporative emissions, as applicable) that would satisfy the Tier 2 testing requirements, so that duplication of effort can be avoided. In addition to existing test data from protocols similar to those specified in the Tier 2 testing program, EPA will consider results from other test protocols to be adequate in lieu of new testing, as long as the alternative methods provide comparable information. Table 2 provides criteria for determining what constitutes adequate existing data in lieu of the specified Tier 2 tests, and includes an example list of comparable tests for each Tier 2 endpoint. EPA recognizes that changes and scientific advances in toxicology testing may result in the development of additional techniques and methods that could be applicable to the Tier 2 testing requirements of

this program in the future. In deciding if a specific protocol is acceptable in lieu of a Tier 2 test, manufacturers must also address other specific criteria for Tier 2 requirements, as explained below.

Table 2.--Criteria for Determining Adequacy of Existing Data in Lieu of Tier 2 (T2) Tests

Endpoint	Minimum requirements for T2 testing in the absence of adequate existing data	existing data to be considered adequate substitutes for T2 tests	Examples of other existing studies which may be acceptable
Carcinogenicity.....	Salmonella assay, in vivo Micronucleus assay, and in vivo Sister Chromatid Exchange assay.	Salmonella assay plus two other assays (at least one of which shall be in vivo).	Alternative to all assays: Lifetime cancer study. In vivo assays: Sister Chromatid Exchange (SCE), Chromosomal Aberrations (CA), Micronucleus (MN), Unscheduled DNA Synthesis (UDS). In vitro or microbial assays: E. coli Reverse Mutation, DNA Repair, Yeast Mutation, Yeast Mitotic Recombination, Mouse Lymphoma, CHO/ V79 Mutation, UDS, CA, SCE, Cell Transformation.
Mutagenicity.....	Salmonella assay and in vivo Micronucleus assay.	Salmonella assay plus one in vivo assay.	MN, Dominant Lethal Assay, CA, Heritable Translocation Assay, Specific Locus Assay.
Teratogenicity.....	T2 fertility/teratology assessment with 90-day exposure.	FDA/Phase II (gd6-15) Study.	If fetal effects analysis is included: Two-generation study, Reproductive Assessment by Continuous Breeding (RACB), One-generation study.
Adult reproductive effects.	T2 fertility/teratology assessment with 90-day exposure.	T2 fertility/teratology assessment with 90-day exposure.	Two-generation study, Reproductive Assessment by Continuous Breeding (RACB), One-generation study.
Neurotoxicity.....	GFAP assay and neuro-	GFAP assay and neuro-	Detailed

characterization of			
	histopathology with 90-day exposure.	histopathology with 90-day exposure.	neurotoxicity using behavior, neurophysiological, and/or neurochemical assessments (e.g., EPA Neurotoxicity Assessment).
Pulmonary effects....	T2 respiratory tract pathology after 90-day exposure.	T2 respiratory tract pathology after 90-day exposure.	Chronic toxicity study, with or without lifetime cancer study; subchronic toxicity study.

In general, for existing information to fulfill the Tier 2 testing requirements, it must include the in vivo inhalation exposure to whole motor vehicle emissions (combustion or evaporative, as applicable), except for the few in vitro studies acceptable for carcinogenicity/ mutagenicity assessment. In order to be acceptable, previous toxicity studies must include exposure to non-catalyzed emissions, as required in this final rule. EPA is requiring the use of an evaporative emission generator for the evaporative emission testing. However, EPA will accept previous inhalation exposure studies of whole evaporative emissions in which the emissions were generated using standard evaporative emission procedures under FTP conditions. Raw product tests, using F/FAs in the uncombusted state, are not considered adequate replacements for Tier 2 combustion emission testing. In addition, studies using whole aerosolized preparations or tests on individual emission products of the fuel or additive cannot be used as substitutes for whole emission testing for either combustion or evaporative emissions. The reason for this requirement is that, as explained in Section III.A.2 of this preamble, this rule focuses on the effects of whole emissions rather than raw F/FAs or individual emission products. Tests performed on the emissions of F/FAs which are classified in the same group as the subject fuel or additive are considered relevant, but tests on products not conforming to the grouping criteria of the subject fuel or additive do not apply.

Other important parameters to consider in determining if existing studies are adequate include: the type and number of test subjects, the number and adequacy of dosages, the methodology and duration of exposure, and the technical methods used for monitoring the progress of the test and for analyzing the results. Generally, 90-days is the minimum acceptable length of exposure. However, an existing study having a shorter exposure period might be considered adequate if the test results are positive, i.e., adverse effects are observed. Previously-conducted studies using mammals other than rodents may be

acceptable in lieu of rodent testing, if the existing studies meet all other applicable criteria for adequacy.

For carcinogenicity and mutagenicity, the Salmonella assay is always required because of its broad sensitivity and specificity for detecting chemical exposures having mutagenic and/or carcinogenic potential. Manufacturers should note (see Table 2) that when a registrant needs to conduct new Tier 2 testing, two in vivo assays (MN and SCE), in addition to the Salmonella assay, are required to satisfy the carcinogenicity endpoint. However, in lieu of new testing, a manufacturer may rely on one existing in vivo assay for carcinogenicity, in addition to the Salmonella assay and another in vitro assay. In other words, two in vivo tests for carcinogenicity are required if new testing is performed, but only one in vivo study is required if the manufacturer relies on existing carcinogenicity information. The reason for this is that the incremental costs of performing the two specified in vivo assays (MN and SCE) within the 90-day inhalation protocol (as required in this final rule) is low in comparison with the amount of useful data obtained. This approach is cost-effective because the same animals used in the inhalation study can also be used for the in vivo carcinogenicity assays. In fact, fewer resources will be spent to do both the MN and SCE assays within a single group of inhalation-exposed animals than to conduct just one of these tests along with a separate in vitro carcinogenicity assay (in addition to the Salmonella assay).

D. Alternative Tier 2 Provision

The Tier 2 tests described above pertain to all designated F/FAs, unless mitigated by special provisions or comparable data from adequately performed and documented previous studies. In general, EPA considers this standard testing program to be necessary for the health effects evaluation of F/FA emissions, even if further evaluation may be required under the provisions of Tier 3. However, this final rule also adopts the special provision discussed in the Reopening Notice, under which EPA retains the authority to modify the standard Tier 2 test requirements in certain instances.

The exercise of this authority will be done wholly at EPA's initiative and discretion. The alternative Tier 2 provision is intended to provide a degree of flexibility to EPA when available information indicates that, in a specific case, another testing regimen is preferable to the standard set of Tier 2 tests. There are three scenarios under which EPA generally anticipates the possible use of this provision.

First, for a particular fuel or fuel additive (or group), information may be available (independent of the requirements of this

rule) which may cause EPA to be concerned about potential health effects related to an endpoint not specifically addressed in Tier 2. In such an instance, the alternative Tier 2 provision allows EPA to require additional studies targeted to the identified area of concern, even though these studies are not normally included in Tier 2. While the standard structure of this rule also allows EPA to prescribe the additional tests under Tier 3, the alternative Tier 2 provision enables EPA to prescribe and receive the desired data earlier in the process. This flexibility is particularly important given that this final rule allows up to six years for Tier 2 submittal. When the additional testing can be coordinated with the standard Tier 2 testing program, the alternative Tier 2 provision will also save costs relative to conducting the additional tests at a separate point in time.

Second, independent of the information to be submitted under this rule for a particular F/FA product (or group), EPA may identify a potentially significant public health risk related to a Tier 2 endpoint, such that EPA knows that more definitive testing will be required for this endpoint than is ordinarily required under Tier 2. Again, EPA could require such testing under Tier 3 after the evaluation of Tier 2. However, the alternative Tier 2 provision can facilitate earlier and potentially more efficient acquisition of the required data. If appropriate to the case at hand, EPA would substitute the more definitive endpoint test for the standard Tier 2 test (with appropriate deadline adjustment). In such a case, EPA's authority to waive the requirement to provide the respective Tier 2 test derives from its authority in section 211(e) to provide exemptions from testing when such testing would be duplicative. In this example, because the substituted test would address the endpoint more rigorously than the standard Tier 2 assessment, it would be duplicative to require both evaluations.

Third, EPA may identify concerns about the effects of F/FA emissions involving different engine and/or emission control technologies than those ordinarily required for generating the emissions tested in Tier 2. For example, biological testing using catalyzed instead of non-catalyzed emissions might be required if emission species of concern are present in the catalyzed exhaust of a fuel or additive that are not represented in the untreated exhaust.\58\ In this case, EPA could prescribe a Tier 2 program using catalyzed instead of non-catalyzed emissions under the alternative Tier 2 provision. Otherwise, the manufacturer of this product would likely be required to conduct a second series of biological tests with catalyzed emissions, under Tier 3.

\58\As described in Section VI.A, the use of non-catalyzed

emissions are normally required in the Tier 2 toxicology tests. Under Tier 1, however both catalyzed and non-catalyzed emissions are required to be characterized.

In summary, the alternative Tier 2 provision will give EPA the flexibility, when indicated, to prescribe additional tests to be performed along with the standard Tier 2 program, to substitute different tests, and/or to modify the underlying vehicle/engine specifications for Tier 2. When EPA exercises its authority under this special provision, it will allow an appropriate time for completion of the prescribed alternative tests. EPA may also use the alternative Tier 2 authority to waive certain Tier 2 endpoint evaluations (generally on occasions when additional and/or more rigorous tests are being required for other Tier 2 endpoints). However, Tier 2 endpoint tests will not be waived in the absence of adequate information or requirements for more rigorous testing of the endpoint(s).

EPA intends to exercise this special authority only in exceptional cases. When EPA decides to use the alternative Tier 2 provision, EPA will notify the responsible manufacturer (or group) by certified mail letter of the specific modifications in lieu of the standard Tier 2 program, along with a schedule for compliance and submittal of test results. The manufacturer (or group of manufacturers) will have 60 days to comment on the prescribed alternative Tier 2 testing program and timing requirements. If the responsible manufacturer does not provide any comments, EPA will assume that the manufacturer has consented in full with the prescribed testing regimen. EPA will publish a notice in the Federal Register to inform the public of its intent to require alternative testing for a particular F/FA manufacturer and that a copy of the letter to the manufacturer is available in the public record of this rule for review and comment. Additional correspondence between EPA and the responsible manufacturer regarding alternative testing requirements will also be placed in the public record. After receipt and review of all comments received (or, if no comments are received), EPA will publish a notice of final action on the proposed alternative Tier 2 requirements in the Federal Register.

In the Reopening Notice, EPA proposed to notify manufacturers of proposed alternative Tier 2 requirements within 18 months of promulgation of the final rule (for registered F/FAs) or within 18 months of EPA's receipt of intent to register (for currently unregistered F/FAs). Comments received from the industry indicated that this notification period was too long relative to the proposed three year deadline for submittal of Tier 2 results. However, EPA believes the proposed notification period is reasonable, given that this final rule allows manufacturers up to six years for submittal of Tier 2

results. Furthermore, EPA believes that restricting the time period for exercising the alternative Tier 2 authority is unnecessary and disadvantageous to the public interest as well as the regulated industry, since this provision establishes a mechanism to provide needed data on a timely basis and to eliminate unnecessary screening studies when substitute tests will be required. Therefore, while EPA will endeavor to notify manufacturers of proposed alternative Tier 2 requirements within 18 months, this final rule permits EPA to notify manufacturers of proposed alternative Tier 2 requirements at any time prior to EPA's receipt of Tier 2 data. If a manufacturer receiving such notification has already begun the standard Tier 2 toxicology testing, then EPA will refrain from requiring the testing; however, in such cases, the manufacturer is required to submit the results of the standard Tier 2 tests within one year of the date when testing began. In other cases, EPA will consider the potential costs, burdens, and timing factors in making its final decisions on alternative Tier 2 requirements.

IX. Tier 3 Requirements

A. Scope

On the basis of the submitted Tier 1 and/or Tier 2 data, or any other available information, EPA will determine whether further testing and/or analysis for the subject fuel or fuel additive is needed under the provisions of Tier 3. Given the variety of evaluations included in Tiers 1 and 2 and the wide range of possible interrelated outcomes which could be obtained, EPA proposed to use its discretion in determining the need for Tier 3 testing on a case-by-case basis. Decisions on the need for follow-up testing within Tier 3 would depend on expert scientific judgment as to the availability of adequate data to enable a health risk evaluation and the need for more definitive information for developing regulatory decisions.

EPA requested comments on the proposed discretionary nature of Tier 3 determinations and on a possible alternative approach involving the establishment of ``automatic triggers'' for Tier 3 decisions, i.e., specific outcomes of Tiers 1 and 2 which would make Tier 3 testing mandatory. Responses included both support and opposition to the proposed Tier 3 discretionary approach. EPA evaluated all comments and determined that decisions on Tier 3 requirements should remain at the discretion of EPA. In order to accomplish the goals of the program, it is essential for EPA to be able to examine the Tier 1 and Tier 2 data prior to prescribing additional tests. The need for and content of Tier 3 testing will most often be dependent on the results of the earlier tiers. In these cases, decisions on Tier 3 can only be reasonably

specified after EPA's review of the applicant's initial submittal. The purpose of Tier 3 is not to fill all data gaps, but to establish a program that provides the Administrator with the necessary and reasonable information to make regulatory decisions. Based on this rationale, EPA judges that it is neither practical nor desirable to specify criteria which will automatically force F/FA manufacturers to perform additional testing under Tier 3. Thus, EPA will use its discretion to determine Tier 3 requirements on a case-by-case basis, allowing EPA to target specific regulatory needs. Although EPA can use the Tier 3 data to support regulatory actions, referral to Tier 3 level is not mandatory before beginning actions under CAA section 211(c).

The need for Tier 3 testing and/or analysis will depend, in part, on whether Tier 1 and/or Tier 2 data provide sufficient toxicity and exposure information to determine the potential health risks associated with a particular fuel or fuel additive. The endpoints to be addressed and the nature of the studies to be performed under Tier 3 are to be determined on a case-by-case basis. Tier 3 studies will most often be required to further explicate the results of the tests/analyses performed under Tiers 1 and 2 or to address other areas of concern highlighted by the literature search. If additional toxicity testing is required under Tier 3, the test might entail whole emissions (as in Tier 2), or the testing of one or more individual emission species identified to be of particular concern. EPA could also use other information (available outside this program) to require testing under Tier 3.

While the specific objectives and scope of Tier 3 testing will vary depending on the concerns identified in the earlier tiers or any other information available to EPA, examples of possible areas for further testing are: chronic/lifetime studies, chemical disposition/metabolism studies, exposure studies, dosimetry analyses, additional emission characterization/speciation, additional modeling analysis, environmental toxicity tests, testing using different emission generation procedures or emission control systems, or any other additional evaluation approach EPA deems necessary to assess the health and/or welfare effects of a particular fuel or fuel additive. The previous examples and the discussion below in no way limit the scope of Tier 3 or EPA's authority to require further testing under this program.

Today's rule specifically includes under Tier 3 any health effects testing to be performed on aerosol additives (see Section X.C for details on the special provision for aerosol products). Due to the special nature and use of this type of product, EPA judges that any testing needs will require non-standard test procedures that can be better addressed under Tier 3. EPA will review the composition information and literature data on the specific aerosol product and

will determine if health effects testing is needed on a case-by-case basis. For example, if available literature, submitted under Tier 1, indicates that a component of an aerosol product is highly toxic, and the product is widely produced, then Tier 3 testing on the aerosol product would likely be indicated.

Modeling of potential exposures, atmospheric reactivity, and/or environmental fate/partitioning may also be required under Tier 3, as explained in Section III.C. For example, if a fuel or additive is widely distributed and Tier 2 data indicate serious concern for health effects, EPA might require more extensive exposure analysis and/or modeling under Tier 3 to better define potential risks.

Although the grouping mechanism included in this rule allows F/FA manufacturers to submit Tier 1 and Tier 2 data on the representative of a designated group for purposes of registration, EPA retains the authority under Tier 3 to require testing on any member product of a group. Thus, when follow-up testing is required under Tier 3 authority, the specified test(s) could be required to be conducted on the selected representative or on any other member of an existing group. Testing on additional F/FA products could be required if EPA identifies a concern for any member of the group other than the group representative. For example, the testing representative for the M85 group (consisting of methanol formulations containing 50-95 percent methanol) is an M85 base fuel containing 85 percent methanol by volume. In the future, other methanol fuels could enter the market and be used extensively, triggering a concern for exposure and potential health risks. In this case, EPA could require Tier 3 testing for another methanol formulation within the M85 group.

Another possible Tier 3 effort could involve the testing of combinations of oxygenates. Although groups for non-baseline gasolines are defined on the basis of individual oxygenates, EPA could request additional testing to address mixtures of oxygenates under Tier 3 authority. For example, if a registered non-baseline gasoline formulation containing a mixture of oxygenates is widely produced and used, resulting in high exposures and potential health risks, EPA could require testing of this formulation under Tier 3. Tier 3 tests for this formulation could involve standard Tier 1 or Tier 2 tests, testing for other endpoints not addressed under Tier 2, or testing for other areas of concern (e.g., exposure analysis, environmental testing, etc.).

EPA also retains the authority to require additional testing using different vehicle/engine technologies and/or emission generation specifications than those prescribed for Tier 1 and Tier 2 analyses. To reduce the costs of the program, today's action requires the use of a single vehicle model for emission generation. However, if EPA determines that emissions from other applicable vehicle/engine technologies might differ significantly for a given fuel or additive/

base fuel mixture, then EPA may require additional emission characterization and/or toxicological testing under Tier 3 using different vehicle/engine technology. Also, although standard Tier 2 tests are to be conducted using non-catalyzed emissions (as discussed in Section VI.A), EPA could require the use of catalyzed emissions for testing under Tier 3.

When a determination has been made that Tier 3 testing is required, EPA will inform the responsible manufacturer by certified mail of the purpose and nature of the testing to be performed along with a schedule for compliance and submittal of the Tier 3 report to EPA. EPA will also publish a notice in the Federal Register, notifying the public that the letter to the manufacturer is available in the public record for review and comment. The affected manufacturer's comments and EPA's response to these comments will be placed in the public record, as well. After receipt and review of all comments received (or, if no comments are received), EPA will publish a notice of final action on the proposed Tier 3 requirements in the Federal Register.

EPA proposed in the NPRM to provide the responsible manufacturer or group a 30-day comment period to respond to EPA's requirements under Tier 3. In their commentary, the regulated industry asked EPA to extend this comment period to 60 days to allow for appropriate time for review, analysis, and preparation of a written response to EPA regarding the designated protocol(s) to be used for Tier 3 tests. EPA judged that this request was reasonable, thus today's rule gives F/FA manufacturers 60 days to comment on the EPA-prescribed Tier 3 requirements.

The responsible manufacturer is expected to submit detailed protocols for review and approval by EPA prior to beginning Tier 3 testing. Tier 3 tests must comply with the pre-approved specifications given by EPA. If manufacturers experience unforeseen difficulties while conducting the prescribed Tier 3 tests approved by EPA (e.g., excess mortality observed half-way through a chronic bioassay), they will be allowed to request a modification of the requirements. This mechanism would apply to unusual circumstances that are outside the control of the manufacturer. If testing problems are identified, EPA must be notified as soon as possible so that requirements can be modified.

B. Criteria for Referral to Tier 3

This section presents some of the guidelines and considerations which EPA will use in determining the necessity for additional testing under a discretionary Tier 3 testing approach. Consistent with the discretionary decision-making process for Tier 3, this discussion is not intended to provide an exhaustive, limiting, or definitive listing of relevant criteria.

The decision to require manufacturers to submit additional testing on the health, environmental, or welfare effects of F/FA emissions will take into account the cumulative information provided by Tiers 1 and 2, including previous scientific data, emissions characterization data, biological test results, and any ancillary information which may be available to EPA. Thus, decisions to require Tier 3 level testing will be made only after all the requirements of Tiers 1 and 2 have been adequately satisfied (with the exception of special cases as discussed in Section VIII.D). Adherence to this principle will prevent unnecessarily costly or poorly targeted decisions based on piecemeal, out-of-context information, and will promote more precise identification and evaluation of data gaps, and more cost-efficient coordination of potential test requirements.

Ultimately, EPA must be able to decide whether or not the use of a fuel or fuel additive is likely to create unacceptable health or welfare risks. If a risk decision is made possible by the information from Tiers 1 and 2, then Tier 3 will not be required. However, if such a risk decision cannot be made on the basis of the Tiers 1 and 2 data, then Tier 3 testing will be mandated. Therefore, to make a determination on the need for Tier 3 testing, EPA scientists will evaluate the extent to which the results of Tiers 1 and 2 are adequate for such decisions, guided by the basic principles of risk assessment.

A risk assessment requires the merging of a health effects assessment (including hazard identification and concentration-response relationship) and an exposure assessment. Such an assessment can range from a qualitative to a highly quantitative analysis, depending upon the extent of the available data. EPA recognizes that a quantitative assessment might not be possible at the end of Tier 2.\59\ However, Tiers 1 and 2 might indicate that little hazard is present and that exposures may be quite low and limited geographically. In such a case, there may be no reason to pursue further testing at the Tier 3 level to improve risk assessment information. On the other hand, Tiers 1 and 2 might suggest that a hazard is likely and that exposures could be significant because of the production volume and ubiquitous use of a product, but the data may still be inadequate for a quantitative risk assessment. In this case, Tier 3 testing could be indicated to provide the needed information.

\59\However, if adequate information exists, EPA does not rule out the possibility of conducting such risk assessment.

In general, the principles and critical data elements of the risk assessment process will provide a useful guide for identifying whether

meaningful information gaps remain and for determining the specific objectives of potential Tier 3 testing. However, EPA does not intend to conduct a formal risk assessment as part of its decision on whether to promote a fuel or fuel additive (or group) to Tier 3. Rather, EPA will evaluate the quality and certainty of the toxicity and concentration-response data and consider qualitatively whether such data weighs in favor of or against further testing. A formal risk assessment will be more likely to be developed at such a time that there is a need for action to control or prohibit a product under the regulatory authority of CAA section 211(c).

The following sections discuss key factors which EPA will consider in identifying the need for and content of Tier 3 testing.

1. Statistical Issues

As previously mentioned, scientific judgment will be exercised in determining whether Tier 3 testing is indicated. An important factor in such judgments will be the interpretation of and significance ascribed to ``negative'' results obtained in Tiers 1 and 2. To address this issue, EPA will consider statistical information such as the probability of Type I and Type II errors.

A Type I error occurs when a false positive conclusion is made, while a Type II error is a false negative conclusion. The acceptability of a specific Type II error is related to the acceptability of false negatives in the particular study being performed. For example, from a toxicological perspective, screening assays often have a relatively high probability of producing false negative (Type II) outcomes, since some major aspects of organ or tissue toxicity are not being examined. Thus, an acceptable Type II error for screening assays will typically be high. However, the level of Type II error considered acceptable should be tempered by the goal of the study. A higher false negative conclusion (e.g., Type II error of 0.2) will generally be acceptable if it refers to an effect of minimal severity at a high-exposure test level relative to ambient concentration and if few people are likely to be exposed. The converse will also hold true.

Scientifically sound statistical analyses are a crucial part of any reliable study and will provide key information for EPA to make judgments on whether or not Tier 3 testing is needed. While it is not feasible to list all possible scenarios and results for each Tier 2 endpoint, the above discussion describes how some of the statistical factors will be incorporated into EPA's decisions.

2. Exposure Assessment

The Tier 1 requirements will provide EPA with information on the composition of F/FA emissions and potential exposures to F/FA products. This information includes: (1) Types and emission rates of speciated emission components, (2) possible literature search findings on ambient, occupational, or epidemiological exposures, (3) literature

information on the potential fate and environmental effects, and (4) a qualitative exposure analysis (submitted by the manufacturer or group) based on the production and use of the F/FA product(s). As mentioned above, this information will be considered qualitatively by EPA in determining the extent of potential exposures and whether additional testing or analysis is needed to assess risks quantitatively. As discussed earlier, EPA has the authority to require exposure modeling or analysis under Tier 3.

Significant public health concerns might sometimes be revealed by the submitted information on product composition, total annual production volume and market distribution data, and emissions data. This might be the case, for example, if there was a significant annual release of emission compounds with known toxicities, or if the anticipated exposures approached or exceeded current estimates of apparently safe levels of known toxicants. In the case of high-volume fuels and their associated bulk additives, EPA will generally assume that human and environmental exposures will be of sufficient level and extent that significant observed adverse effects could indicate a need for follow-up in Tier 3. This exposure assumption reflects the high production and consumption of these products, either at the present time or as anticipated in the future. Thus, decisions to refer these products to Tier 3 will be based on the degree to which additional testing is needed to clarify the results and potential health effect and environmental implications identified in the previous tiers. On the other hand, it cannot be assumed that fuel additives used in relatively low concentrations or produced in relatively low volumes will automatically be excused from Tier 3. For these products, test results indicative of severe health effects and/or high exposure levels (e.g., during consumer use) might be cause for escalation to Tier 3.

3. Health Assessment

General criteria for evaluating the potential public health effects associated with fuel and additive emissions will include: (1) The number of positive and negative outcomes related to each endpoint, (2) the identification of a concentration-effect relationship, (3) the statistical sensitivity and significance of such studies, (4) the severity of the observed effects (e.g., whether the effects will likely lead to incapacitating or irreversible conditions), and (5) the consistency and clarity of apparent mechanisms, target organs, and outcomes. Additional parameters which will influence the decision on whether to require Tier 3 will include: (1) The nature and amount of known toxic agents in the emissions stream and (2) the observation of lesions which specifically implicate inhalation as an important exposure route for inducing adverse health effects.

These criteria will be evaluated in conjunction with the Tier 1 and Tier 2 results to determine whether or not higher level testing is

needed. In this decision, both the biological and statistical significance of the Tiers 1 and 2 results will be taken into account. Generally, escalation to Tier 3 may be judged necessary when remaining uncertainties about the significance of observed outcomes and/or potential exposures interfere with EPA's ability to make reasonable estimates of potential health risks. On the other hand, if no statistically significant effects are obtained at any exposure level in a scientifically sound Tier 2 study (or existing test submitted in lieu of Tier 2 testing and not contradicted by other published reports of equal or greater reliability), and if other major sources of concern do not arise (e.g., toxic effects of structurally related compounds), then Tier 3 testing is not likely to be required for the endpoint in question.

This discussion assumes that relevant, high-quality statistical analyses have been done to permit the negative test results to be properly evaluated and interpreted. The statistical analyses recommended for Tier 2, including determination of Type I and II error (as discussed in Section IX.B.1), should enable reasonable conclusions to be drawn as to the significance of negative findings. Factors to be taken into account include the toxicological nature of the findings and the exposure levels used in the test. For example, if the statistical analyses are applied to a ``severe'' endpoint (e.g., major fetal abnormalities, major lung pathology, etc.) and the exposure in question is moderate, then a relatively low Type II error level may be appropriate. In contrast, if a high concentration limit test causes a relatively minor effect (e.g., a small change in estrous cyclicity), a higher Type II error may be allowed, effectively increasing the chance of false negative conclusions.

4. Evaluation of Tier 2 Results

The specific outcomes which will be considered positive and negative results for each Tier 2 test were mentioned briefly in the previous descriptions of Tier 2 requirements (see Section VIII.B) and are defined and interpreted more precisely in the regulatory text of this final rule. For example, three primary assays (i.e., Salmonella, MN, and SCE) are included in Tier 2 for the screening of carcinogenicity and mutagenicity. As compared with appropriate controls, a statistically significant concentration-related positive response in any one of these assays could be cause for concern, as will be positive outcomes for at least one concentration in two or more of these tests. Such outcomes will be indicative of mutagenic and/or carcinogenic risk. Positive results will also indicate that the emissions could initiate some of the mechanisms involved in carcinogenesis. However, these results will generally not in themselves be sufficient to determine whether the emissions were in fact carcinogenic because the development of cancer is a multi-step process.

Depending on the internal and historical consistency of the results and their relationship to projected exposures, further testing might be required to determine the significance of the mutagenic and/or carcinogenic activity/risks in human populations exposed by inhalation. In contrast, if no statistically significant results are obtained in the three assays and no conflicting results are found in the literature or in any other Tier 2 tests, then Tier 3 follow-up of potential carcinogenic/mutagenic effects is not likely to be required.

To take another example, determination of the need to investigate further reproductive or teratogenic risks will take into account the outcome of the results of the estrous cyclicity measurements, the histopathological evaluation of reproductive organs, the outcome of the fertility screen, and the outcome of the teratogenicity evaluation. If negative results are obtained in the above evaluations (according to statistically sound principles), and if these results are not refuted by the existing literature, then additional testing is not likely to be required at the Tier 3 level for reproductive or teratogenic effects. Positive results for the teratogenicity study will include a decrease in neonatal viability relative to that in control studies, a significant change in the proportion of viable male versus female fetuses or offspring, the presence of soft tissue or skeletal abnormalities, and an increased rate of embryonic or fetal resorption. Other positive outcomes related to reproductive effects, such as decreased fertility, decreased litter size, abnormal changes in vaginal cytology or reproductive organ histopathology, will be indicative of hazards to the adult reproductive systems. The need for additional evaluation under Tier 3 will depend on the specificity, severity, and consistency of results, the presence or absence of a concentration-effect relationship, and the significance of these outcomes in view of projected exposures. The greater the remaining uncertainty regarding the risk of reproductive or teratogenic effects after analysis of such factors, the higher will be the likelihood that Tier 3 will be required.

Similarly, consistent negative results (according to statistically sound principles) obtained in other Tier 2 tests, in the absence of significant related concerns raised in the literature, will (in all likelihood) make Tier 3 unnecessary. If adverse effects are found at Tier 2 and/or reported in the literature, EPA will determine if Tier 3 follow-up is required by attempting to evaluate the nature, severity, and significance of the findings in light of the likely exposures. If EPA determines that Tier 3 testing is required to resolve the remaining uncertainties, the Tier 3 requirements will reflect both positive and negative results. For example, if the results of Tier 2 were positive for pulmonary effects but negative for neurotoxicity (according to criteria discussed earlier), and if these results were consistent with

the literature, only pulmonary toxicity would be a likely candidate for Tier 3 follow-up testing.

C. Potential Tier 3 Tests

To be most cost-effective, Tier 3 testing will be designed to address specific data gaps regarding health effects endpoints of concern or health and environment-related issues requiring further analysis. For instance, Tier 3 requirements could potentially include further emission characterization procedures, perhaps involving additional vehicles and/or more rigorous pre-conditioning methods, to identify and quantify harmful emission products with greater precision. Higher-order modeling calculations or exposure field studies could be required to resolve uncertainties in the Tier 1 emissions exposure information. Health or welfare effects testing requirements will be aimed at providing sufficient information to make sound conclusions about the degree of health or welfare risk. If more than one endpoint is of concern, EPA will attempt to reduce testing costs by permitting combined protocols insofar as possible.

Tier 3 tests for specific endpoints could require the determination of a NOAEL. Depending on the endpoint under evaluation, consideration will be given to including a mid-duration examination in the case of chronic inhalation tests. A mid-duration evaluation will be useful for affirming the adequacy of exposure levels and, in some cases, might enable interim risk conclusions to be drawn which will avoid the need for further examination. Inhalation studies will generally make use of rodent species, but higher order mammals could be required.

While Tier 3 testing requirements will be targeted to critical areas of concern, EPA will also exercise its judgment to avoid the false economy of establishing overly narrow requirements. Just as requirements for too many assays would be wasteful of resources, requirements for too few assays might result in inconclusive findings, creating needs for still further testing at greater total expense than would have been necessary at the start. Similarly, EPA will consider the value of including secondary evaluations as useful and low-cost adjuncts to tests already required. For example, if the histopathology of a specified target organ was the primary examination required at the conclusion of an inhalation exposure, other organs could be weighed and saved in storage for a limited time period, at low incremental expense. If indicated, these other organs would then be available for subsequent examination, avoiding the possible need to repeat the chronic inhalation procedures to assess the effects on other organs.

Because the specific health testing requirements which will be imposed in Tier 3 will be tailored to individual circumstances, precise test guidelines cannot be provided in advance. However, some examples

of testing scenarios which might be required under Tier 3 are cited in Sec. 79.54 in the accompanying regulatory text. Where possible, existing standard guidelines for these tests are referenced. It should be recognized, however, that such guidelines might need to be revised to accommodate emission inhalation requirements and/or to evaluate certain structures or functions which the current guidelines do not adequately address. Study parameters which might require modification include exposure routes and concentrations, species selection, number of animal subjects, examination procedures and frequencies, and analytic requirements. Furthermore, interim advances in the underlying science and testing technology may provide superior approaches which could be available for use by the time Tier 3 requirements are implemented.

X. Special Provisions

The following sections describe special provisions included in the F/FA registration program to avoid duplication of effort, to alleviate the financial impact on small businesses, and to ease the burdens of the program on the regulated industry in general.

A. Experimental F/FAs

EPA requested comments on the possibility of providing a temporary program exemption or deferment for experimental F/FAs. Eligibility for this special provision was to be limited to unregistered products (i.e., F/FAs which are not registered as of the effective date of this final rule) or registered products that had not been placed into wholesale or retail commerce prior to promulgation of this rule.

The regulated industry provided comments questioning the need for a special provision for experimental F/FAs under this rulemaking because a provision for experimental products already exists under existing registration rules. EPA agrees with the comments received and today's rule relies on existing regulations under 40 CFR 79.4(a)(3) and 79.4(b)(2) for the exemption of experimental F/FAs. 40 CFR section 79.4(a)(3) exempts fuels used for research, development or testing, and 40 CFR section 79.4(b)(2) similarly exempts fuel additives. Based on these existing provisions, any designated F/FA product sold to automobile, engine, or component manufacturers for research, development or test purposes, or sold to automobile manufacturers for factory fill, and not in any case offered for commercial sale to the public, is exempted from the registration requirements of today's rule.

B. Relabeled Products

A company's product is registered as ``relabeled'' if it is simply a repackaged and rebranded version of a formulation which is already registered by another manufacturer and is procured from that manufacturer for sale or use. Requiring companies which sell relabeled products to conduct the testing program in today's rulemaking would clearly duplicate the efforts of the original manufacturer. Thus, under the authority of CAA section 211(e)(3)(C), which provides that the Administrator may ``exempt any person from such regulations with respect to a particular fuel or fuel additive upon a finding that any additional testing of such fuel or fuel additive would be duplicative of adequate existing testing," today's rule includes a special provision exempting relabeled products from the evaluation and testing requirements. For relabeled products, only basic registration information will be required, as described below in Section XII.A.

About half of the 4,800 fuel additives registered (as of March 1994) are relabeled products. Manufacturers of these relabeled products will therefore not be required to comply with the health and welfare effects assessment provisions of the F/FA registration program.

C. Aerosols

Several commenters requested an exemption for aftermarket aerosol additives because of the nature of their mode of application and their low frequency of use. EPA examined available data on aerosol products and concluded that the required testing procedures of this rule are not well suited to this type of product. The evaluation of currently registered additives indicates that aerosols include carburetor cleaners and engine starters that are sprayed into the air intake valve of the engine and are used only intermittently and, even then, for only a very brief period of time (i.e., a few seconds). Because of their intermittent use and method of use, it is unlikely that their recommended application would affect the overall characteristics of vehicle emissions. Direct exposure to the aerosol product itself is likely to be a more important source of potential hazards than is exposure to its combustion or evaporative emission products. Thus, the evaluation of potential health and welfare effects of aerosols would require a different testing regimen tailored to the specific nature of these products.

Today's rule therefore establishes a special provision for aerosols. Manufacturers of aftermarket aerosol additives are required to provide only the basic registration data required for all F/FA manufacturers (see Section XII.A), plus a literature survey of existing information on their products and a discussion on the potential exposures. Thus, the standard emission characterization and Tier 2 tests are not required for aerosol products. The literature search (as

described in Section VII.A) must include existing data on potential health and welfare effects on the uncombusted aerosol product as a whole and on the individual components of the product. The analysis of potential exposures should be based on the total annual production volume data and the market distribution of the product, as explained in Section VII.C. EPA will review the submitted information and will determine if there is a need to require testing on a case-by-case basis. Thus, if testing of an aerosol is prescribed by EPA, the testing is to be performed under Tier 3. Using this approach, EPA will be able to tailor specific tests (as needed) addressing the particular problems related to aerosol product exposure.

D. Small Business Provisions

In the NPRM, EPA requested comments on the possibility of establishing special provisions for small businesses. The regulated industry expressed concerns regarding the costs of the program and the potential impact of the F/FA regulations on the financial status of small companies. EPA understands that small businesses might be particularly affected by the F/FA regulations due to their smaller resource base, generally lower rate of representation in trade organizations and, consequently, their potentially limited opportunity to participate in grouping and cost-sharing arrangements. In view of these circumstances, EPA is including in today's rule special provisions to alleviate the economic impact of this rule on small manufacturers of F/FAs, taking into consideration the comments received.

EPA examined the distribution of currently registered F/FA companies across various sales ranges and identified a sales level which will define a ``small business'' for the purposes of this rule. A comparison between registered fuel manufacturers and registered additive manufacturers indicates that the distribution across sales ranges is similar for both industries. Each industry appears to be made up of many small companies and relatively few larger companies. Approximately a fifth of registered fuel manufacturers and a third of registered additive manufacturers have sales above $100 million. For both industries, most of the companies with sales under $100 million tend to cluster under $50 million. Thus, within each of the industries, companies tend to cluster above $100 million and below $50 million in sales. Furthermore, companies with sales below $50 million also tend to have sales below $10 million, especially within the additive industry.

Based on the analysis of F/FA manufacturers' sales data, EPA identified $50 million as a reasonable sales level for differentiating between small and large companies within each industry.\60\ As a result, this final rule defines a small business as any motor vehicle

fuel or fuel additive manufacturer with total annual sales of less than $50 million.\61\ The small business provisions established for this rule are defined below.

\60\In cases where subsidiary, divisional, or other complex business arrangements exist, the business entity to which this sales level pertains is the parent company with ultimate ownership. The ``ultimate'' parent is defined as the uppermost headquarters or topmost company encompassing all related parents, subsidiaries, divisions, branches, or other operating units. This definition follows that used by the Small Business Administration. It also helps to ensure that companies will not subdivide merely to become eligible for the small business provisions of this program.

\61\``Total annual sales'' means the average of the manufacturer's sales revenue in each of the previous three years (i.e., the three years prior to the submittal of the supplemental notification form required under this rule).

Special provisions for small F/FA companies were developed based on assumptions regarding the degree to which manufacturers will have the opportunity to group with other manufacturers to share costs, and the degree to which information on similar products can be expected to be submitted by larger companies. A review of EPA's registration data base shows that nearly every registered F/FA manufacturer produces at least one baseline or non-baseline product. Hence, even if generous special provisions are provided for small manufacturers of these products, EPA is assured that test data on such products will still be submitted by other, larger companies which do not qualify for the special provisions. Therefore, in regard to registration of products in the baseline and non-baseline categories, this final rule requires small businesses (i.e., companies with annual sales less than $50 million) to submit only the basic registration data for their baseline and/or non-baseline products. Such companies are not required to meet the Tier 1 and Tier 2 requirements in order to register their baseline and non-baseline F/FAs. Since the larger companies will still be submitting the data for these products, this provision does not compromise EPA's ability to get the necessary information to evaluate the potential health and welfare effects of baseline and non-baseline products.

The above assumption, however, is not valid in the case of atypical F/FAs. Analysis of current registrations shows that there are fewer atypical products than baseline products and fewer large companies that produce atypical F/FAs. Therefore, if the same small business

provisions described above for baseline/non-baseline F/FAs were also applied to atypical F/FAs, then there would be no data submitted for many atypical F/FAs. EPA would then be unable to evaluate the health and welfare effects for these atypical products. Nevertheless, EPA realizes that the testing program will be particularly burdensome for very small manufacturers of atypical F/FAs. In part, this is due to the added compliance requirements for atypical products (e.g., more rigorous mileage accumulation requirements prior to emission generation (see Section VI.E) and more extensive emission characterization requirements (see Section VII.B). An even greater impact results from the fact that grouping opportunities for these manufacturers might be quite limited. Not only do atypical products tend to be unique, but also, the smallest manufacturers rarely are members of the dominant trade associations which are most likely to take the lead in organizing and administering F/FA group functions. Thus, high program costs could fall on manufacturers with low financial resources and perhaps few opportunities to share the costs.

Based on the above reasons, this final rule includes additional special provisions applicable to the atypical F/FAs of small businesses. However, these provisions apply to fewer manufacturers and are less liberal than those established for baseline and non- baseline products. Specifically, for manufacturers of atypical products with less than $10 million in annual sales (rather than the $50 million sales level applicable to manufacturers of baseline/non-baseline F/FAs), the minimal requirements of the program include only basic registration and Tier 1 data. These requirements fall between those applicable to small manufacturers of baseline/non-baseline F/FAs and the general requirements of the program for larger manufacturers.

In summary, manufacturers qualifying for small business special provisions for their baseline and non-baseline products (having less than $50 million annual sales) are excused from both Tier 1 and Tier 2 requirements, while small manufacturers qualifying for special provisions for their atypical products (having less than $10 million annual sales) are excused only from Tier 2 requirements for these products. Since small business provisions are based on both the annual sales and the product category, the possibility exists for a manufacturer to have some products excused from program requirements while having to comply with testing requirements for others. For example, an additive manufacturer having both baseline and atypical products and annual sales of $30 million can use the special small business provision for the baseline additives, but not for the atypical products.

Pursuant to CAA section 211(b), F/FAs which are excused from any program requirements under these special provisions may still be subject to testing under Tier 3 at EPA's discretion (on a case-by-case

basis). The Tier 3 testing might include (but would not be necessarily limited to) information which would otherwise have been required under the provisions of Tier 1 and/or Tier 2.

XI. Timing and Compliance Requirements

The timing and compliance requirements for the F/FA registration program are dependent on the type of product and the registration status of the product. As discussed in Section III.A of this preamble, both Tier 1 and Tier 2 are mandatory requirements for all F/FAs (or groups), except as may be modified by any applicable special provisions. Special provisions affecting the content and/or timing of these requirements are discussed in Sections VII.A.3, VIII.D, and X of this preamble. For F/FAs registered as of the effective date of this rule, Tier 1 data and evidence of a suitable contractual arrangement for satisfactory completion of Tier 2 requirements must be submitted to EPA within three years of the effective date. The results of Tier 2 must be submitted to EPA no later than six years from the effective date. The schedule for completion of any Tier 3 requirements which EPA may prescribe will be determined based on the nature of the particular requirements. The general reporting format for submittal of all of these requirements is described in Section XII.

For registrable F/FAs, i.e., F/FAs not registered as of the effective date of this rule but meeting the criteria for grouping with a currently registered fuel or bulk additive in the same fuel family, the content and timing of requirements is essentially the same as for the currently registered F/FAs. Thus, manufacturers of these products will be granted registration and be permitted to market registrable F/FAs upon EPA's receipt of basic registration data for such products. In contrast, for manufacturers of new F/FAs (i.e., F/FAs that are not currently registered and do not meet the registrable criteria), all testing requirements must be completed prior to registration and introduction into commerce, including Tier 3 when prescribed by EPA.

After receipt of Tier 1 and/or Tier 2 data, EPA will determine whether the submitted information is in compliance with the specified guidelines and whether further testing of a particular fuel or fuel additive is required under the provisions of Tier 3. For registered F/FAs, EPA intends to determine the adequacy of the submitted data within two years after receipt. However, if EPA is unable to inform the registered manufacturer of the adequacy of the Tier 1 and/or Tier 2 data within two years after submittal, EPA retains the authority to require that satisfactory data be submitted if, upon subsequent review, EPA finds that the original submittal was inadequate for compliance. In such a case, EPA will not hold the manufacturer liable for penalties for violating this rule from the time period between the date on which

the data were due and the date on which EPA informs the responsible manufacturer of a violation. Regarding new F/FAs, EPA will send a notification of compliance within six months after submission of Tier 1 and Tier 2 data. If the manufacturer of the new F/FA product does not receive a notification of compliance with Tier 1 and Tier 2 within this time frame, then the manufacturer should assume that the Tier 1 and Tier 2 requirements have been satisfactorily met. EPA's determination of the need for Tier 3 testing for new F/FAs will occur within six months after EPA notifies the manufacturer of satisfactory compliance with Tier 1 and Tier 2 requirements or within twelve months of the Tier 1 and Tier 2 submittal, whichever occurs first.

If Tier 3 testing is deemed necessary, EPA will notify the responsible manufacturer (or group) by certified letter of the specific Tier 3 requirement(s) along with a schedule for compliance and a deadline for submittal of the Tier 3 report to EPA (see Section IX.A). This final rule gives the responsible manufacturer (or group) 60 days to comment on the prescribed Tier 3 requirements, compliance schedule, and submission deadline. In the event that EPA receives no comment within the given period, the manufacturer will be assumed to have consented in full to the prescribed Tier 3 requirements. Compliance with Tier 3 requirements is not optional.

Registered (and registrable) F/FAs required to undergo Tier 3 testing will retain their registration for that time determined to be necessary for the completion of Tier 3 tests. This registration will be contingent on the satisfactory compliance with the Tier 3 requirements according to a timetable determined by EPA to be appropriate to those requirements. When Tier 3 is prescribed for new F/FAs (i.e., those not meeting the registrable criteria), EPA may withhold registration until completion of all testing requirements. For new F/FAs, EPA will determine whether the Tier 3 requirements have been met within one year of receiving the Tier 3 submittal. If Tier 3 requirements are satisfied, then EPA will send a notification to the manufacturer granting registration to the new F/FA product. Registration of new F/FAs will not occur until that time when EPA determines that all Tier 3 requirements have been satisfactorily met.

As described above, EPA's review times for data on new F/FAs are shorter than those for registered F/FAs. The reason for this discrepancy is that manufacturers of new F/FAs are barred from marketing such products until EPA approves their compliance with all testing requirements and grants them registration. On the other hand, manufacturers of registered products can maintain their registration, and thus their ability to sell their products, while EPA is reviewing their submitted data. Thus, to ensure that undue hindrance is not created for manufacturers of new products wanting to enter the marketplace, EPA has abbreviated the review times for new F/FA

products.

Notwithstanding the granting of a registration (or continued registration for registered F/FAs), if EPA determines that a fuel or fuel additive causes or contributes to air pollution that may reasonably be anticipated to endanger the public health or welfare, then EPA could invoke available regulatory authority under CAA section 211(c). Referral to Tier 3 is not required for EPA to begin a regulatory action under 211(c).

If additional testing is needed to make up for deficiencies in information content or testing technique/procedures related to Tier 1, Tier 2, and/or Tier 3, then the original compliance deadlines will still be in force. Manufacturers of existing products who fail to submit data in the prescribed time frames or who submit data from tests that do not comply with the specified guidelines will be in violation of this rule and will be subject to the penalties specified in CAA section 211(d). According to CAA section 211(d), persons who fail to submit any information or conduct any tests required by the Administrator under CAA section 211(b) shall be liable to the United States for a civil penalty of not more than $25,000 for every day of such violation plus the amount of economic benefit or savings resulting from the violation. Each day after the due date for submission of data will constitute a separate day of violation. Civil penalties will be assessed in accordance with CAA sections 205(b) and (c), which permit EPA to proceed either in court or in an administrative action. If a group of manufacturers commits to performing joint testing, each manufacturer would separately be in violation of the rule. However, the Administrator would retain the authority to remit or mitigate any penalty under CAA section 211(d).

In addition to the above penalties, the district courts of the United States have jurisdiction to compel the furnishing of information and/or the conduct of tests required under CAA section 211(b). This means that, in addition to the financial penalties, persons failing to submit data or comply with the specified guidelines would still need to submit the data originally required. Furthermore, if EPA determines that the data requirements of the rule were not met, EPA could revoke the registration of the fuel(s) or additive(s) in question.

Because EPA recognizes that unusual circumstances, outside the control of the manufacturer, may occasionally interfere with the manufacturer's ability to comply with the provisions of the rule, today's rule contains a mechanism to allow manufacturers to request modification of the requirements under some specific circumstances. This special mechanism allows persons who experience unforeseen problems or accidents in conducting the EPA-prescribed tests to request modification of the requirements in order to avoid being in violation of the rule. This mechanism would apply to unusual mechanical problems

or other unavoidable problems that could arise during the performance of the required tests. The modification requests must be submitted as soon as the manufacturer is aware of the difficulty, but not later than thirty days following the event precipitating the request. Additional details on this special mechanism for modification of requirements is included in the regulatory text of this rule.

XII. Reporting Requirements

The materials to be submitted to EPA include the basic registration data, a summary report with Tier 1 and Tier 2 results, and associated appendices. If the results of Tiers 1 and 2 are submitted at the same time, then the summary report must include both Tier 1 and Tier 2 information and associated appendices, as described below. If Tier 1 and Tier 2 results are submitted separately, then two individual reports must be provided to EPA, i.e., Tier 1 report and Tier 2 report. In such a case, each individual report must include the summary information applicable to the respective tier (including a cover page, executive summary, test substance information, a summary of tier results, conclusions, and associated appendices). If the Tier 2 report for registered F/FAs is not submitted within three years after the effective date of the final rule, then evidence of a suitable arrangement for completion of Tier 2 (e.g., a copy of a signed contract with a qualified laboratory to conduct the required Tier 2 tests) must be submitted to EPA prior to that date. F/FA manufacturers who must conduct additional testing under Tier 3 are required to submit a Tier 3 report when the designated Tier 3 testing is complete. The nature of the information to be included in the basic registration data, reports, and associated appendices is described below.

A. Basic Registration Data

The basic information already required for F/FA registration includes product and manufacturer identification, concentration and purpose-in-use, and specific compositional data. Today's rule adds the following items to the basic registration data requirements: total annual production volume data, marketing distribution data, notification about group participation, and notification on the use of special provisions (i.e., relabeled products, aerosols, and small business, as discussed in Section X). Manufacturers of F/FAs registered as of the effective date of this rule must submit the additional basic registration data items to EPA within six months of that date. Other manufacturers are strongly encouraged to submit the basic registration data prior to starting the evaluation tiers (i.e., Tier 1, Tier 2, and/or Tier 3).

The production volume information is to be reported in units of gallons per year for F/FA products that are generally sold in liquid form and kilograms per year for F/FA products that are generally sold in solid form. For F/FAs already in production, the submitted figure must reflect the most recent annual period as well as the volume projected to be produced in the third subsequent year. For products not yet in production, the best estimate of expected total production volume during the third year of production must be provided. Market distribution data for each product must also be provided. For fuels and bulk additives, the distribution data must be reported as the percent of total annual sales volume marketed in each Petroleum Administration for Defense District (PADD), as defined in Sec. 79.59(b)(3) of this rule. For aftermarket additives, the distribution data must be reported as the percent of total annual sales volume marketed in each state. For products not yet in production, the manufacturer must report projected distribution data by PADD or state, as applicable.

Manufacturers of F/FAs registered as of the effective date of this rule who intend to comply with registration requirements as part of a group must identify the person or entity which is organizing the testing for the applicable group. Similarly, if an applicant is relying on another manufacturer's (or group's) previous registration materials in compliance with the testing requirements for an unregistered product, then the other manufacturer or group must be identified. In addition, the manufacturer of the unregistered product must provide evidence that the original submitter has been notified and that reimbursement will occur.

The basic registration data must be submitted (or resubmitted) individually for each product being registered, using EPA forms which are in effect at the time of the submittal. This requirement pertains to all F/FA products registered as of the effective date of this rule, including relabeled products, as well as those for which first-time registration is sought after promulgation of this rule. If the basic registration data previously submitted for a currently registered fuel or additive is accurate and complete, then a statement asserting that this is so will suffice in lieu of the submittal of duplicate information. A finding by EPA that this information is not, in fact, accurate and complete as claimed will result in the report being considered inadequate.

A fuel manufacturer may at any time modify an existing fuel registration by submitting a request to EPA to add or delete a bulk additive to the existing registration information for such fuel product, provided that any additional additive must be registered by EPA for use in the specific fuel family to which the fuel product belongs. The addition or deletion of a bulk additive to a fuel registration does not necessarily cause the fuel to be considered

``new". However, if the change affects the grouping of such registered fuel, it may affect the testing responsibilities of the fuel manufacturer.

B. Summary Report

This report will provide a summary of the evaluation procedures, results, and conclusions, pertaining to Tier 1 and/or Tier 2 requirements.\62\ References used to support Tier 1 and/or Tier 2 conclusions must be cited in the report. A cover page must be included, identifying the test substance, the manufacturer's name and address, a designated contact person and phone number, and grouping information (if applicable). The grouping information must identify the group name or grouping criteria, all products and manufacturers to which the report pertains, and the name and address of the responsible organization or entity reporting for the group. The body of the summary report must be divided into the following sections.

\62\These reporting requirements may pertain to separate submittals for Tier 1 and Tier 2 or a single submittal for both tiers, depending on the relative timing of these compliance activities.

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1. Executive Summary

This section must include a brief description of the general results and conclusions for the tier(s) included in the report (i.e., Tier 1 and/or Tier 2), emphasizing information and test data which provide evidence for potential adverse health and/or welfare effects.

2. Test Substance Information

This section must include a detailed test substance description, including (as applicable) base fuel parameter values or test fuel composition (if other than base fuel), and test additive composition. The base fuel description must include the types and concentrations of base fuel additive components and values for each of the parameters specified in the base fuel definition for the applicable fuel family. Similar parameter values must be identified for test fuels other than base fuels.

3. Tier 1 Summary

This section is intended to provide an overview of Tier 1 analyses. Detailed procedural descriptions, tables, and other outputs are to be included in the appendices.

a. Literature Search. The search methods must be described, including the identity of data bases and time periods accessed. Any in-

house and/or other unpublished studies included in the literature search must also be described briefly. The results and conclusions of the literature search with respect to potential health and welfare effects of the subject fuel or fuel additive must be summarized. If test documentation provided by the literature search is used to satisfy some or all of the other program requirements, the relevant studies must be discussed and their adequacy to fulfill the specific purposes of the associated program requirements must be justified. Finally, the person(s) or contractors conducting the search are to be identified.

b. Emission Generation and Characterization. This section of the summary report must identify the vehicle selected and describe the procedures followed in vehicle/engine preparation and maintenance and in the generation, storage, and processing of emissions for testing. For group submissions, the report must include a complete description of the group representative used in the generation of emissions. A description of the analytic methods used to characterize the F/FA emission products must also be provided. Problems encountered in generating and/or characterizing the emissions must be discussed, including attempts to resolve the problems and their potential effects on testing outcomes. The laboratories performing these procedures must be identified.

c. Exposure Analysis. This section must include a qualitative discussion on the potential exposures to the general, area-specific, and/or special at-risk population groups based on the production and use of the particular fuel or additive in question. For group submissions, the analysis must consider potential exposures due to all members of the group. When available, EPA recommends the use of existing modeling data to support the exposure analysis.

4. Tier 2 Summary

For each study, the objectives, principles, and general procedures must be outlined and the findings and conclusions summarized. Discussion must be included regarding problems encountered during the performance of the tests and the methods used to resolve them. This discussion must include the impact which such problems may have had on the study outcomes.

5. Conclusions

Further testing needs must be identified or else a discussion must be provided explaining why the results of Tiers 1 and/or Tier 2 should not trigger Tier 3 testing requirements.

C. Appendices

Detailed information in support of the general discussions contained in the summary report are to be submitted as appendices to the report. In regard to the literature search, the appendices must

contain (1) summary tables of existing studies regarding health and environmental effects, including such information as the type of study, species/strain used, exposure concentration(s), duration of study, endpoints evaluated, results (incidence and statistical significance), and references, (2) a complete copy of reference lists and associated abstracts obtained from data base searches (in printed form or on 3\1/2\ inch (IBM compatible) computer diskettes), (3) complete documentation of in-house studies and other unpublished information sources, and (4) complete documentation (e.g., copies of journal articles) of previous studies which are being cited in satisfaction of Tier 1 and/or Tier 2 test requirements. Appendices to the emission characterization section must contain detailed protocols, copies of all relevant laboratory reports, a list of all speciated emission products and their emission rates, and documentation and results of calibration/verification procedures. For the section that discusses potential exposures, an appendix must be provided for detailed background information on the production volume and market distribution data used in the exposure analysis. If exposure models are used, background calculations and/or model data must also be included in an appendix.

An appendix is also required for each of the tests conducted in compliance with Tier 2 requirements. These appendices must contain the full detailed study protocol, complete laboratory report, statistical analysis of the findings, and scientific conclusions. These materials must conform to the reporting requirements of the individual study guidelines as well as the general standards for recordkeeping and reporting specified in the GLP standards of this final rule (see Sec. 79.60). A final appendix must be provided, containing laboratory certifications and associated personnel credentials.

D. Tier 3 Report

Reports for additional tests required under the Tier 3 provisions must include a cover page with identifying information as described above for the Tier 1 and 2 summary report(s). The report must begin with a discussion of the concerns arising under the previous tiers which led to the Tier 3 requirements, the specific objectives of the additional studies, and a summary of pertinent results and conclusions. References used in support of Tier 3 conclusions must be cited in the report. The Tier 3 summary discussion must be supported with appendices containing the kinds of documentation discussed above with respect to Tier 2. The laboratory conducting the required tests must be identified, and relevant certifications and personnel credentials provided.

E. Confidential Business Information

CAA section 211(b)(2)(B) states that the results of tests ``conducted in conformity with test procedures and protocols established by the Administrator,'' pursuant to CAA section 211(b)(2)(A), shall not be considered confidential. Thus, health and welfare information supplied to EPA in compliance with Tier 1, Tier 2, and Tier 3 testing requirements will be made available to the public.

Manufacturers (or groups) claiming business confidentiality on any information submitted under the F/FA testing program must make a claim of confidentiality in writing at the time of submittal of the reporting requirements. To assert a business confidentiality claim the submitter must clearly mark the confidential information and must submit a separate document setting forth the claim and listing each location at which the confidential information appears in the submitted data. If any person subsequently requests access to the test data submitted under the F/FA testing program (other than health and welfare effects information) and such information is subject to a claim of business confidentiality, the request and any subsequent disclosure will be governed by the provisions of 40 CFR part 2.

XIII. Administrative Requirements

A. Administrative Designation and Regulatory Analysis

Under Executive Order 12866 (58 FR 51735), EPA must determine whether the regulatory action is ``significant'' and therefore subject to the Office of Management and Budget (OMB) review and the requirements of the Executive Order. The Order defines ``economically significant regulatory action'' as one that is likely to result in a rule that may: (1) Have an annual effect on the economy of $100 million or more or adversely affect in a material way the economy, a sector of the economy, productivity, competition, jobs, the environment, public health or safety, or State, local, or tribal governments or communities; (2) create a serious inconsistency or otherwise interfere with an action taken or planned by another agency; (3) materially alter the budgetary impact of entitlements, grants, user fees, or loan programs or the rights and obligations of recipients thereof; or (4) raise novel legal or policy issues arising out of legal mandates, the President's priorities, or the principles set forth in the Executive Order.

A regulatory support document which presents EPA's analysis of the cost impacts of this final rule is available for review in the public docket (A-90-07). EPA estimates that the costs to industry for submittal of the requisite data for Tiers 1 and 2 would total approximately $66 million, assumed to be incurred over the first three-year period after promulgation of this final rule. Thus, the average

annual cost during this period would be about $22 million. In the subsequent three years, Tier 3 requirements might cost an additional $1 million annually, per product or group. If ten products or groups were required to conduct Tier 3 testing in the three-year period following the initial compliance period, the cost would be $10 million per year. These projected overall costs are far less than the $100 million annual cost criterion which is a major determinant in defining an ``economically significant regulatory action.'' In addition, this final rule is not expected to adversely affect in a material way the economy, a sector of the economy, productivity, competition, jobs, the environment, public health or safety, or State, local, or tribal governments or communities.

Although not ``economically significant'' based on the above criteria, this final rule is still considered a ``significant regulatory action'' pursuant to the terms of Executive Order 12866 and was thus submitted to OMB for review. Any written comments from OMB and any EPA response to OMB's comments are available in the public docket for this rule.

B. Regulatory Flexibility Act

Under section 605 of the Regulatory Flexibility Act, 5 U.S.C. 601 et seq., the Administrator is required to assess the economic impact of regulatory actions on small businesses. Accordingly, a Regulatory Flexibility Analysis (RFA) has been prepared for this rule and is presented as part of the Regulatory Support Document (available in the public docket). The RFA compares the financial impacts of this rule on small F/FA manufacturers to the impacts on large F/FA manufacturers. The analysis explains the small business definition specifically developed in this rule to provide special provisions for small F/FA manufacturers (see Section X.D) and assesses the effectiveness of these provisions.

This final rule defines a small business as any motor vehicle fuel or fuel additive manufacturer with total annual sales of less than $50 million. A comparison of companies classified as small under this definition to those classified as small by the Small Business Administration (SBA) reveals that the F/FA program definition classifies a larger number of F/FA manufacturers as small, and more closely groups companies with similar financial characteristics. Based on this analysis, EPA determined that the small business definition established in this rule is reasonable and applicable to the F/FA industry.

Impacts of this rule on F/FA manufacturers were determined by projecting the effects of the estimated compliance costs on each company's return on assets (ROA). In general, a reduction in ROA (after

compliance costs) to less than 2.5 percent is indicative of financial distress. A ROA less than -4 percent indicates that a company is in severe financial distress, and a ROA less than -30 percent generally indicates closure. According to these ROA thresholds, results of the RFA show that approximately 68 unique F/FA manufacturers (43 additive manufacturers, 23 fuel manufacturers, and 2 that produce both fuels and additives) could potentially be pushed into some level of financial distress. All of these companies are classified as small (i.e., have sales less than $50 million). The majority of these companies would fall into the first level of financial distress; however, less than one-fifth of them (12 companies) would potentially be in danger of closure. In relation to the total population of F/FA manufacturers, the companies potentially falling into some level of financial distress account to about six percent, with about 1.2 percent potentially in danger of closure.

As previously described, this final rule includes two types of special provisions for small businesses. With respect to baseline and non-baseline F/FAs, all small manufacturers (i.e., annual sales under $50 million) are excused from all Tier 1 and Tier 2 testing requirements. A segment of these small manufacturers, i.e., those having sales under $10 million, are also excused from Tier 2 testing responsibility for their atypical F/FAs. Nevertheless, all twelve of the manufacturers who are projected to be in danger of closure as a result of this rule are very small companies with one or more atypical F/FAs. The RFA analysis shows that the special provision for small manufacturers with atypical products lowers overall compliance costs for 60 such manufacturers by roughly $20 million (as compared with costs that would otherwise occur in the absence of this special provision). Without this provision, an additional 15 manufacturers of atypical products would potentially be pushed into closure.

It should be noted that the RFA has assumed no changes in prices, sales, product mix, or financial strategies. In many cases, a portion of regulatory costs can be actually passed on to consumers or back to suppliers. Manufacturers also have the option of reformulating a product to a ``baseline'' standard which has much lower compliance costs, or even of dropping products with the greatest cost and smallest profit potential. A more detailed discussion on circumstances which could mitigate compliance cost impacts is provided in the RFA.

C. Recordkeeping Requirements

The information collection requirements in this rule have been submitted for approval to OMB under the Paperwork Reduction Act, 44 U.S.C. 3501 et seq. An Information Collection Request document has been prepared by EPA (ICR #1696.01) and a copy may be obtained from Sandy

Farmer, Information Policy Branch, EPA, 401 M Street SW., Washington, DC 20460 or by calling 202-260-2740. These requirements are not effective until OMB approves them and a technical amendment to that effect is published in the Federal Register.

This collection of information has an estimated annual reporting and recordkeeping burden averaging 43 hours per response. These estimates include time for reviewing instructions, searching existing data sources, gathering and maintaining the data needed, and completing and reviewing the collection of information.

Send comments regarding the burden estimate or any other aspect of this collection of information, including suggestions for reducing this burden to Chief, Information Policy Branch; EPA; 401 M Street, SW., (Mail Code 2136); Washington, DC 20460; and to the Office of Information and Regulatory Affairs, Office of Management and Budget, Washington, DC 20503, marked ``Attention: Desk Officer for EPA."

XIV. Electronic Availability of Rulemaking Documents

Electronic copies of the preamble, the Regulatory Support Document and Regulatory Flexibility Analysis, the Summary and Analysis of Comments, and the regulations for the Fuels and Fuel Additives Registration rulemaking are available on the Office of Air Quality Planning and Standards (OAQPS) Technology Transfer Network Bulletin Board System (TTNBBS). Instructions for accessing TTNBBS and downloading F/FA files are described below.

TTNBBS can be accessed using a dial-in telephone line (919-541-5742) and a 1200, 2400, or 9600 bps modem (equipment up to 14.4 Kbps can be accommodated). The parity of the modem should be set to N or none, the data bits to 8, and the stop bits to 1. When first signing on to the bulletin board, the user will be required to answer some basic informational questions to register into the system. After registering, proceed through the following options from a series of menus:

(M) OMS
(K) Rulemaking and Reporting
(3) Fuels
(4) Fuels/Fuel Additives

A list of ``.ZIP" files will be displayed, all of which relate to the Fuels and Fuel Additives Registration rulemaking. The four documents listed will be in the form of ``.ZIP" files and are identified by the following titles:

``FFA--PRE.ZIP" (Preamble)
``FFA--RSD.ZIP" (Regulatory Support Document and Regulatory

Flexibility Analysis)
``FFA--COM.ZIP" (Summary and Analysis of Comments)
``FFA--REG.ZIP" (Regulations)

File information can be obtained from the ``READ.ME" file. Choose from the following options when prompted:

<D>ownload, <P>rotocol, <E>xamine, <N>ew, <L>ist, <H>elp or <ENTER> to exit.

To download a file, e.g., <D> filename.ZIP, the user needs to choose a file transfer protocol appropriate for the user's computer from the options listed on the terminal. The user's computer is then ready to receive the file by invoking the user's resident file transfer software. Programs and instructions for de-archiving compressed files can be found under <S>ystems Utilities from the top menu, under <A>rchivers/de-archivers.

TTNBBS is available 24 hours a day, 7 days a week except Monday morning from 8-12 EST, when the system is down for maintenance and backup. For help in accessing the system, call the systems operator at 919-541-5384 in Research Triangle Park, North Carolina, during normal business hours EST.

List of Subjects in 40 CFR Part 79

Environmental protection, Fuel additives, Fuels, Gasoline, Incorporation by reference, Motor vehicle pollution, Penalties, Reporting and recordkeeping requirements.

Dated: May 27, 1994.
Carol M. Browner,
Administrator.

Part 79 of title 40 of the Code of Federal Regulations is amended as follows:

1. The authority citation for part 79 is revised to read as follows:

Authority: 42 U.S.C. 7414, 7524, 7545 and 7601.

2. Section 79.2 is amended by revising paragraphs (d), (e), and (f) to read as follows:

Sec. 79.2 Definitions.

* * * * *

(d) Fuel manufacturer means any person who, for sale or introduction into commerce, produces, manufactures, or imports a fuel or causes or directs the alteration of the chemical composition of, or the mixture of chemical compounds in, a bulk fuel by adding to it an additive.

(e) Additive means any substance that is intentionally added to a fuel named in the designation (including any added to a motor vehicle's fuel system) and that is not intentionally removed prior to sale or use.

(f) Additive manufacturer means any person who produces, manufactures, or imports an additive for use as an additive and/or sells or imports for sale such additive under the person's own name.

* * * * *

3. Section 79.3 is revised to read as follows:

Sec. 79.3 Availability of information.

The availability to the public of information provided to, or otherwise obtained by, the Administrator under this part shall be governed by part 2 of this chapter except as expressly noted in subpart F of this part.

4. Section 79.4 is amended by revising paragraph (b)(1) to read as follows:

Sec. 79.4 Requirement of registration.

* * * * *

(b) Additives. (1) No manufacturer of any fuel additive designated under this part shall, after the date by which the additive must be registered under this part, sell, offer for sale, or introduce into commerce such additive for use in any type of fuel designated under this part unless the Administrator has registered that additive for use in that type of fuel.

* * * * *

5. Section 79.6 is revised to read as follows:

Sec. 79.6 Requirement for testing.

Provisions regarding testing that is required for registration of a designated fuel or fuel additive are contained in subpart F of this part.

6. Section 79.10 is revised to read as follows:

Sec. 79.10 Application for registration by fuel manufacturer.

Any manufacturer of a designated fuel who wishes to register that fuel shall submit an application for registration including all of the information set forth in Sec. 79.11. If the manufacturer produces more than one grade or brand of a designated fuel, a manufacturer may include more than one grade or brand in a single application, provided that the application includes all information required for registration of each such grade or brand by this part. Each application shall be signed by the fuel manufacturer and shall be submitted on such forms as the Administrator will supply on request.

7. Section 79.11 is amended by revising the introductory text of the section, removing the period in paragraph (h) and adding a semicolon and paragraphs (i) and (j) to read as follows:

Sec. 79.11 Information and assurances to be provided by the fuel manufacturer.

Each application for registration submitted by the manufacturer of a designated fuel shall include the following:

* * * * *

(i) The manufacturer of any fuel which will be sold, offered for sale, or introduced into commerce for use in motor vehicles manufactured after model year 1974 shall demonstrate that the fuel is substantially similar to any fuel utilized in the certification of any 1975 or subsequent model year vehicle or engine, or that the manufacturer has obtained a waiver under 42 U.S.C. 7545(f)(4); and

(j) The manufacturer shall submit, or shall reference prior submissions, including all of the test data and other information required prior to registration of the fuel by the provisions of subpart F of this part.

8. Section 79.12 is revised to read as follows:

Sec. 79.12 Determination of noncompliance.

If the Administrator determines that an applicant for registration of a designated fuel has failed to submit all of the information required by Sec. 79.11, or determines within the applicable period provided for Agency review that the applicant has not satisfactorily completed any testing which is required prior to registration of the

fuel by any provision of subpart F of this part, he shall return the application to the manufacturer, along with an explanation of all deficiencies in the required information.

9. Section 79.13 is amended by revising paragraph (a) to read as follows:

Sec. 79.13 Registration.

(a) If the Administrator determines that a manufacturer has submitted an application for registration of a designated fuel which includes all of the information and assurances required by Sec. 79.11 and has satisfactorily completed all of the testing required by subpart F of this part, the Administrator shall promptly register the fuel and notify the fuel manufacturer of such registration.

* * * * *

10. Section 79.20 is revised to read as follows:

Sec. 79.20 Application for registration by additive manufacturer.

Any manufacturer of a designated fuel additive who wishes to register that additive shall submit an application for registration including all of the information set forth in Sec. 79.21. Each application shall be signed by the fuel additive manufacturer and shall be submitted on such forms as the Administrator will supply on request.

11. Section 79.21 is amended by revising the introductory text of the section and paragraph (d) and adding paragraphs (h) and (i) to read as follows:

Sec. 79.21 Information and assurances to be provided by the additive manufacturer.

Each application for registration submitted by the manufacturer of a designated fuel additive shall include the following:

* * * * *

(d) The specific types of fuels designated under Sec. 79.32 for which the fuel additive will be sold, offered for sale, or introduced into commerce, and the fuel additive manufacturer's recommended range of concentration and purpose-in-use for each such type of fuel.

* * * * *

(h) The manufacturer of any fuel additive which will be sold, offered for sale, or introduced into commerce for use in any type of fuel intended for use in motor vehicles manufactured after model year

1974 shall demonstrate that the fuel additive, when used at the recommended range of concentration, is substantially similar to any fuel additive included in a fuel utilized in the certification of any 1975 or subsequent model year vehicle or engine, or that the manufacturer has obtained a waiver under 42 U.S.C. 7545(f)(4).

(i) The manufacturer shall submit, or shall reference prior submissions, including all of the test data and other information required prior to registration of the fuel additive by the provisions of subpart F of this part.

12. Section 79.22 is revised to read as follows:

Sec. 79.22 Determination of noncompliance.

If the Administrator determines that an applicant for registration of a designated fuel additive has failed to submit all of the information required by Sec. 79.21, or determines within the applicable period provided for Agency review that the applicant has not satisfactorily completed any testing which is required prior to registration of the fuel additive by any provision of subpart F of this part, he shall return the application to the manufacturer, along with an explanation of all deficiencies in the required information.

13. Section 79.23 is amended by removing paragraph (b), by redesignating paragraph (c) as paragraph (b), and by revising paragraph (a) to read as follows:

Sec. 79.23 Registration.

(a) If the Administrator determines that a manufacturer has submitted an application for registration of a designated fuel additive which includes all of the information and assurances required by Sec. 79.21 and has satisfactorily completed all of the testing required by subpart F of this part, the Administrator shall promptly register the fuel additive and notify the fuel manufacturer of such registration.

* * * * *

14. Section 79.31 is amended by revising paragraph (b) to read as follows:

Sec. 79.31 Additives.

* * * * *

(b) All designated additives must be registered by July 7, 1976.

* * * * *

15. A new subpart F, consisting of Secs. 79.50-79.68, is added to part 79 to read as follows:

Subpart F--Testing Requirements for Registration

Subpart F--Testing Requirements for Registration

Sec. 79.50 Definitions.

The definitions listed in this section apply only to subpart F of this part.

Additive/base fuel mixture means the mixture resulting when a fuel additive is added in specified proportion to the base fuel of the fuel family to which the additive belongs.

Aerosol additive means a chemical mixture in aerosol form generally used as a motor vehicle engine starting aid or carburetor cleaner and not recommended to be placed in the fuel tank.

Aftermarket fuel additive means a product which is added by the end-user directly to fuel in a motor vehicle or engine to modify the performance or other characteristics of the fuel, the engine, or its

emissions.

Atypical element means any chemical element found in a fuel or additive product which is not allowed in the baseline category of the associated fuel family, and an ``atypical fuel or fuel additive'' is a product which contains such an atypical element.

Base fuel means a generic fuel formulated from a set of specifications to be representative of a particular fuel family.

Basic emissions means the total hydrocarbons, carbon monoxide, oxides of nitrogen, and particulates occurring in motor vehicle or engine emissions.

Bulk fuel additive means a product which is added to fuel at the refinery as part of the original blending stream or after the fuel is transported from the refinery but before the fuel is purchased for introduction into the fuel tank of a motor vehicle.

Emission characterization means the determination of the chemical composition of emissions.

Emission generation means the operation of a vehicle or engine or the vaporization of a fuel or additive/fuel mixture under controlled conditions for the purpose of creating emissions to be used for testing purposes.

Emission sampling means the removal of a fraction of collected emissions for testing purposes.

Emission speciation means the analysis of vehicle or engine emissions to determine the individual chemical compounds which comprise those emissions.

Engine Dynamometer Schedule (EDS) means the transient engine speed versus torque time sequence commonly used in heavy-duty engine evaluation. The EDS for heavy-duty diesel engines is specified in 40 CFR part 86, appendix I(f)(2).

Evaporative Emission Generator (EEG) means a fuel tank or vessel to which heat is applied to cause a portion of the fuel to evaporate at a desired rate.

Evaporative emissions means chemical compounds emitted into the atmosphere by vaporization of contents of a fuel or additive/fuel mixture.

Evaporative fuel means a fuel which has a Reid Vapor Pressure (RVP, pursuant to 40 CFR part 80, appendix ``E'') of 2.0 pounds per square inch or greater and is not supplied to motor vehicle engines by way of sealed containment and delivery systems.

Evaporative fuel additive means a fuel additive which, when mixed with its specified base fuel, causes an increase in the RVP of the base fuel by 0.4 psi or more relative to the RVP of the base fuel alone and results in an additive/base fuel mixture whose RVP is 2.0 psi, or greater. Excluded from this definition are fuel additives used with fuels which are supplied to motor vehicle engines by way of sealed

containment and delivery systems.

Federal Test Procedure (FTP) means the body of exhaust and evaporative emissions test procedures described in 40 CFR 86 for the certification of new motor vehicles to Federal motor vehicle emissions standards.

Fuel family means a set of fuels and fuel additives which share basic chemical and physical formulation characteristics and can be used in the same engine or vehicle.

Manufacturer means a person who is a fuel manufacturer or additive manufacturer as defined in Sec. 79.2 (d) and (f).

Nitrated polycyclic aromatic hydrocarbons (NPAH) means the class of compounds whose molecular structure includes two or more aromatic rings and contains one or more nitrogen substitutions.

Non-catalyzed emissions means exhaust emissions not subject to an effective aftertreatment device such as a functional catalyst or particulate trap.

Oxygenate compound means an oxygen-containing, ashless organic compound, such as an alcohol or ether, which may be used as a fuel or fuel additive.

Polycyclic aromatic hydrocarbons (PAH) means the class of hydrocarbon compounds whose molecular structure includes two or more aromatic rings.

Relabeled additive means a fuel additive which is registered by its original manufacturer with EPA and is also registered and sold, unchanged in composition, under a different label and/or by a different entity.

Semi-volatile organic compounds means that fraction of gaseous combustion emissions which consists of compounds with greater than twelve carbon atoms and can be trapped in sorbent polymer resins.

Urban Dynamometer Driving Schedule (UDDS) means the 1372 second transient speed driving sequence used by EPA to simulate typical urban driving. The UDDS for light-duty vehicles is described in 40 CFR part 86, appendix I(a).

Vapor phase means the gaseous fraction of combustion emissions.

Vehicle classes/subclasses means the divisions of vehicle groups within a vehicle type, including light-duty vehicles, light-duty trucks, and heavy-duty vehicles as specified in 40 CFR part 86.

Vehicle type means the divisions of motor vehicles according to combustion cycle and intended fuel class, including, but not necessarily limited to, Otto cycle gasoline-fueled vehicles, Otto cycle methanol-fueled vehicles, diesel cycle diesel-fueled vehicles, and diesel cycle methanol-fueled vehicles.

Whole emissions means all components of unfiltered combustion emissions or evaporative emissions.

Sec. 79.51 General requirements and provisions.

(a) Overview of requirements. (1) All manufacturers of fuels and fuel additives that are designated for registration under this part are required to comply with the requirements of subpart F of this part either on an individual basis or as a participant in a group of manufacturers of the same or similar fuels and fuel additives, as defined in Sec. 79.56. If manufacturers elect to comply by participation in a group, each manufacturer continues to be individually subject to the requirements of subpart F of this part, and responsible for testing under this subpart. Each manufacturer, subject to the provisions for group applications in Sec. 79.51(b) and the special provisions in Sec. 79.58, shall submit all Tier 1 and Tier 2 information required by Secs. 79.52, 79.53 and 79.59 for each fuel or additive, except that the Tier 1 emission characterization requirements in Sec. 79.52(b) and/or the Tier 2 testing requirements in Sec. 79.53 may be satisfied by adequate existing information pursuant to the Tier 1 literature search requirements in Sec. 79.52(d). The adequacy of existing information to serve in compliance with specific Tier 1 and/or Tier 2 requirements shall be determined according to the criteria and procedures specified in Secs. 79.52(b) and 79.53 (c) and (d). In all cases, EPA reserves the right to require, based upon the information contained in the application or any other information available to the Agency, that manufacturers conduct additional testing of any fuel or additive (or fuel/additive group) if EPA determines that there is inadequate information upon which to base regulatory decisions for such product(s). In any case where EPA determines that the requirements of Tiers 1 and 2 have been satisfied but that further testing is required, the provisions of Tier 3 (Sec. 79.54) shall apply.

(2) Laboratory facilities shall perform testing in compliance with Good Laboratory Practice (GLP) requirements as those requirements apply to inhalation toxicology studies. All studies shall be monitored by the facilities' Quality Assurance units (as specified in Sec. 79.60).

(b) Group Applications. Subject to the provisions of Sec. 79.56 (a) through (c), EPA will consider any testing requirements of this subpart to have been met for any fuel or fuel additive when a fuel or fuel additive which meets the criteria for inclusion in the same group as the subject fuel or fuel additive has met that testing requirement, provided that all fuels and additives must be individually registered as described in Sec. 79.59(b). For purposes of this subpart, a determination of which group contains a particular fuel or additive will be made pursuant to the provisions of Sec. 79.56 (d) and (e). Nothing in this subsection (b) shall be deemed to require a manufacturer to rely on another manufacturer's testing.

(c) Application Procedures and Dates. Each application submitted in

compliance with this subpart shall be signed by the manufacturer of the designated fuel or additive, or by the manufacturer's agent, and shall be submitted to the address and in the format prescribed in Sec. 79.59. A manufacturer who chooses to comply as part of a group pursuant to Sec. 79.56 shall be covered by the group's joint application. Subject to any modifications pursuant to the special provisions in Secs. 79.51(f) or 79.58, the schedule for compliance with the requirements of this subpart is as follows:

(1) Fuels and fuel additives with existing registrations. (i) The manufacturer of a fuel or fuel additive product which, pursuant to subpart B or C of this part, is registered as of May 27, 1994 must submit the additional basic registration data specified in Sec. 79.59(b) before November 28, 1994.

(ii) For these products, the manufacturer must also satisfy the requirements and time schedules in either of the following paragraphs (c)(1)(ii) (A) or (B) of this section:

(A) Within May 27, 1997, all applicable Tier 1 and Tier 2 requirements must be submitted to EPA, pursuant to Secs. 79.52, 79.53, and 79.59; or

(B) Within May 27, 1997, all applicable Tier 1 requirements (pursuant to Secs. 79.52 and 79.59), plus evidence of a contract with a qualified laboratory (or other suitable arrangement) for completion of all applicable Tier 2 requirements, must be submitted to EPA. For this purpose, a qualified laboratory is one which can demonstrate the capabilities and credentials specified in Sec. 79.53(c)(1). In addition, within May 26, 2000, all applicable Tier 2 requirements (pursuant to Secs. 79.53 and 79.59) must be submitted to EPA.

(iii) In the case of such fuels and fuel additives which, pursuant to applicable special provisions in Sec. 79.58, are not subject to Tier 2 requirements, all other requirements (except Tier 3) must be submitted to EPA before May 27, 1997.

(iv) In the event that Tier 3 testing is also required (under Sec. 79.54), EPA shall determine an appropriate timeline for completion of the additional requirements and shall communicate this schedule to the manufacturer according to the provisions of Sec. 79.54(b).

(v) The manufacturer may at any time modify an existing fuel registration by submitting a request to EPA to add or delete a bulk additive to the existing registration information for such fuel product, provided that any additional additive must be registered by EPA for use in the specific fuel family to which the fuel product belongs. However, the addition or deletion of a bulk additive to a fuel registration may effect the grouping of such registered fuel under the criteria of Sec. 79.56, and thus may effect the testing responsibilities of the fuel manufacturer under this subpart.

(2) Registrable fuels and fuel additives. (i) A fuel product which

is not registered pursuant to subpart B of this part as of May 27, 1994 shall be considered registrable if, under the criteria established by Sec. 79.56, the fuel can be enrolled in the same fuel/additive group with one or more currently registered fuels. A fuel additive product which is not registered for a specific type of fuel pursuant to subpart C of this part as of May 27, 1994 shall be considered registrable for that type of fuel if, under the criteria established by Sec. 79.56, the fuel/additive mixture resulting from use of the additive product in the specific type of fuel can be enrolled in the same fuel/additive group with one or more currently registered fuels or bulk fuel additives. For the purpose of this determination, currently registered fuels and bulk additives are those with existing registrations as of the date on which EPA receives the basic registration data (pursuant to Sec. 79.59(b)) for the product in question.

(ii) A manufacturer seeking to register under subpart B of this part a fuel product which is deemed registrable under this section, or to register under subpart C of this part a fuel additive product for a specific type of fuel for which it is deemed registrable under this section, shall submit the basic registration data (pursuant to Sec. 79.59(b)) for that product as part of the application for registration. If the Administrator determines that the product is registrable under this section, then the Administrator shall promptly register the product, provided that the applicant has satisfied all of the other requirements for registration under subpart B or subpart C of this part, and contingent upon satisfactory submission of required information under paragraph (c)(2)(iii) of this section.

(iii) Registration of a registrable fuel or additive shall be subject to the same requirements and compliance schedule as specified in paragraph (c)(1) of this section for existing fuels and fuel additives. Accordingly, manufacturers of registrable fuels or additives may be granted and may retain registration for such products only if any applicable and due Tier 1, 2, and 3 requirements have also been satisfied by either the manufacturer of the product or the fuel/additive group to which the product belongs.

(3) New fuels and fuel additives. A fuel product shall be considered new if it is not registered pursuant to subpart B of this part as of May 27, 1994 and if, under the criteria established by Sec. 79.56, it cannot be enrolled in the same fuel/additive group with one or more currently registered fuels. A fuel additive product shall be considered new with respect to a specific type of fuel if it is not expressly registered for that type of fuel pursuant to subpart C of this part as of May 27, 1994 and if, under the criteria established by Sec. 79.56, the fuel/additive mixture resulting from use of the additive product in the specific type of fuel cannot be enrolled in the same fuel/additive group with one or more currently registered fuels or

bulk fuel additives. For the purpose of this determination, currently registered fuels and bulk additives are those with existing registrations as of the date on which EPA receives the basic registration data (pursuant to Sec. 79.59(b)) for the product in question. For such new product, the manufacturer must satisfactorily complete all applicable Tier 1 and Tier 2 requirements, followed by any Tier 3 testing which the Administrator may require, before registration will be granted.

(d) Notifications. Upon receipt of a manufacturer's (or group's) submittal in compliance with the requirements of this subpart, EPA will notify such manufacturer (or group) that the application has been received and what, if any, information, testing, or retesting is necessary to bring the application into compliance with the requirements of this subpart. EPA intends to provide such notification of receipt in a timely manner for each such application.

(1) Registered fuel and fuel additive notification. (i) The manufacturer of a registered fuel or fuel additive product who is notified that the submittal for such product contains adequate information pursuant to the Tier 1 and Tier 2 testing and reporting requirements (Secs. 79.52, 79.53, and 79.59 (a) through (c)) may continue to sell, offer for sale, or introduce into commerce the registered product as permitted by the existing registration for the product under Sec. 79.4.

(ii) If the manufacturer of a registered fuel or fuel additive product is notified that testing or retesting is necessary to bring the Tier 1 and/or Tier 2 submittal into compliance, the continued sale or importation of the product shall be conditional upon satisfactorily completing the requirements within the time frame specified in paragraph (c)(1) of this section.

(iii) EPA intends to notify the manufacturer of the adequacy of the submitted data within two years of EPA's receipt of such data. However, EPA retains the right to require that adequate data be submitted to EPA if, upon subsequent review, EPA finds that the original Tier 1 and/or Tier 2 submittal is not consistent with the requirements of this subpart. If EPA does not notify the manufacturer of the adequacy of the Tier 1 and/or Tier 2 data within two years, EPA will not hold the manufacturer liable for penalties for violating this rule for the period beginning when the data was due until the time EPA notifies the manufacturer of the violation.

(iv) If the manufacturer of a registered fuel or fuel additive product is notified (pursuant to Sec. 79.54(b)) that Tier 3 testing is required for its product, then the manufacturer may continue to sell, offer for sale, introduce into commerce the registered product as permitted by the existing registration for the product under Sec. 79.4. However, if the manufacturer fails to complete the specified Tier 3

requirements within the specified time, the registration of the product will be subject to cancellation under Sec. 79.51(f)(6).

(v) EPA retains the right to require additional Tier 3 testing pursuant to the procedures in Sec. 79.54.

(2) New fuel and fuel additive notification. (i) Within six months following its receipt of the Tier 1 and Tier 2 submittal for a new product (as defined in paragraph (c)(3) of this section), EPA shall notify the manufacturer of the adequacy of such submittal in compliance with the requirements of Secs. 79.52, 79.53, and 79.59 (a) through (c).

(A) If EPA notifies the manufacturer that testing, retesting, or additional information is necessary to bring the Tier 1 and Tier 2 submittal into compliance, the manufacturer shall remedy all inadequacies and provide Tier 3 data, if required, before EPA shall consider the requirements for registration to have been met for the product in question.

(B) If EPA does not notify the manufacturer of the adequacy of the Tier 1 and Tier 2 submittal within six months following the submittal, the manufacturer shall be deemed to have satisfactorily completed Tiers 1 and 2.

(ii) Within six months of the date on which EPA notifies the manufacturer of satisfactory completion of Tiers 1 and 2 for a new product, or within one year of the submittal of the Tier 1 and Tier 2 data (whichever is earlier), EPA shall determine whether additional testing is currently needed under the provisions of Tier 3 and, pursuant to Sec. 79.54(b), shall notify the manufacturer of its determination.

(A) If the manufacturer of a new fuel or fuel additive product is notified that Tier 3 testing is required for such product, then EPA shall have the authority to withhold registration until the specified Tier 3 requirements have been satisfactorily completed. EPA shall determine whether the Tier 3 requirements have been met, and shall notify the manufacturer of this determination, within one year of receiving the manufacturer's Tier 3 submittal.

(B) If EPA does not notify the manufacturer of potential Tier 3 requirements within the prescribed timeframe, then additional testing at the Tier 3 level is deemed currently unnecessary and the manufacturer shall be considered to have complied with all current registration requirements for the new fuel or additive product.

(iii) Upon completion of all current Tier 1, Tier 2, and Tier 3 requirements, and submission of an application for registration which includes all of the information and assurances required by Sec. 79.11 or Sec. 79.21, the registration of the new fuel or additive shall be granted, and the registrant may then sell, offer for sale, or introduce into commerce the registered product as permitted by Sec. 79.4.

(iv) Once the new product becomes registered, EPA reserves the

right to require additional Tier 3 testing pursuant to the procedures specified in Sec. 79.54.

(e) Inspection of a testing facility. (1) A testing facility, emissions analysis or health and/or welfare effects, shall permit an authorized employee or duly designated representative of EPA, at reasonable times and in a reasonable manner, to inspect the facility and to inspect (and in the case of records also to copy) all records and specimens required to be maintained regarding studies to which this rule applies. The records inspection and copying requirements shall not apply to quality assurance unit records of findings and problems, or to actions recommended and taken, except the EPA may seek production of these records in litigation or informal hearings.

(2) EPA will not consider reliable for purposes of showing that a test substance does or does not present a risk of injury to health or the environment any data developed by a testing facility or sponsor that refuses to permit inspection in accordance with this section. The determination that a study will not be considered reliable does not, however, relieve the sponsor of a required test of any obligation under any applicable statute or regulation to submit the results of the study to EPA.

(3) Effects of non-compliance. Pursuant to sections 114, 208, and 211(d) of the CAA, it shall be a violation of this section and a violation of 40 CFR part 79, subpart F to deny entry to an authorized employee or duly designated representative of EPA for the purpose of auditing a testing facility or test data.

(f) Penalties and Injunctive Relief. (1) Any person who violates these regulations shall be subject to a civil penalty of up to $25,000 for each and every day of the continuance of the violation and the economic benefit or savings resulting from the violation. Action to collect such civil penalties shall be commenced in accordance with paragraph (b) of section 205 of the Clean Air Act or assessed in accordance with paragraph (c) of section 205 of the Clean Air Act, 42 U.S.C. 7524 (b) and (c).

(2) Under section 205(b) of the CAA, the Administrator may commence a civil action for violation of this subpart in the district court of the United States for the district in which the violation is alleged to have occurred or in which the defendant resides or has a principal place of business.

(3) Under section 205(c) of the CAA, the Administrator may assess a civil penalty of $25,000 for each and every day of the continuance of the violation and the economic benefit or savings resulting from the violation, except that the maximum penalty assessment shall not exceed $200,000, unless the Administrator and the Attorney General jointly determine that a matter involving a larger penalty amount is appropriate for administrative penalty assessment. Any such

determination by the Administrator and the Attorney General shall not be subject to judicial review.

(4) The Administrator may, upon application by the person against whom any such penalty has been assessed, remit or mitigate, with or without conditions, any such penalty.

(5) The district courts of the United States shall have jurisdiction to compel the furnishing of information and the conduct of tests required by the Administrator under these regulations and to award other appropriate relief. Actions to compel such actions shall be brought by and in the name of the United States. In any such action, subpoenas for witnesses who are required to attend a district court in any district may run into any other district.

(6) Cancellation.

(i) The Administrator of EPA may issue a notice of intent to cancel a fuel or fuel additive registration if the Administrator determines that the registrant has failed to submit in a timely manner any data required to maintain registration under this part or under section 211(b) or 211(e) of the Clean Air Act.

(ii) Upon issuance of a notice of intent to cancel, EPA will forward a copy of the notice to the registrant by certified mail, return receipt requested, at the address of record given in the registration, along with an explanation of the reasons for the proposed cancellation.

(iii) The registrant will be afforded 60 days from the date of receipt of the notice of intent to cancel to submit written comments concerning the notice, and to demonstrate or achieve compliance with the specific data requirements which provide the basis for the proposed cancellation. If the registrant does not respond in writing within 60 days from the date of receipt of the notice of intent to cancel, the cancellation of the registration shall become final by operation of law and the Administrator shall notify the registrant of such cancellation. If the registrant responds in writing within 60 days from the date of receipt of the notice of intent to cancel, the Administrator shall review and consider all comments submitted by the registrant before taking final action concerning the proposed cancellation. The registrants' communications should be sent to the following address: Director, Field Operations and Support Division, 6406J--Fuel/Additives Registration, U.S. Environmental Protection Agency, 401 M Street SW., Washington, DC 20460.

(iv) As part of a written response to a notice of intent to cancel, a registrant may request an informal hearing concerning the notice. Any such request shall state with specificity the information the registrant wishes to present at such a hearing. If an informal hearing is requested, EPA shall schedule such a hearing within 60 days from the date of receipt of the request. If an informal hearing is held, the

subject matter of the hearing shall be confined solely to whether or not the registrant has complied with the specific data requirements which provide the basis for the proposed cancellation. If an informal hearing is held, the designated presiding officer may be any EPA employee, the hearing procedures shall be informal, and the hearing shall not be subject to or governed by 40 CFR part 22 or by 5 U.S.C. 554, 556, or 557. A verbatim transcript of each informal hearing shall be kept and the Administrator shall consider all relevant evidence and arguments presented at the hearing in making a final decision concerning a proposed cancellation.

(v) If a registrant who has received a notice of intent to cancel submits a timely written response, and the Administrator decides after reviewing the response and the transcript of any informal hearing to cancel the registration, the Administrator shall issue a final cancellation order, forward a copy of the cancellation order to the registrant by certified mail, and promptly publish the cancellation order in the Federal Register. Any cancellation order issued after receipt of a timely written response by the registrant shall become legally effective five days after it is published in the Federal Register.

(g) Modification of Regulation. (1) In special circumstances, a manufacturer subject to the registration requirements of this rule may petition the Administrator to modify the mandatory testing requirements in the test standard for any test required by this rule by application to Director, Field Operations and Support Division, at the address in paragraph (f)(6)(iii) of this section.

(i) Such request shall be made as soon as the test sponsor is aware that the modification is necessary, but in no event shall the request be made after 30 days following the event which precipitated the request.

(ii) Upon such request, the Administrator may, in circumstances which are outside the control of the manufacturer(s) or his/their agent and which could not have been reasonably foreseen or avoided, modify the mandatory testing requirements in the rule if such requirements are infeasible.

(iii) If the Administrator determines that such modifications would not significantly alter the scope of the test, EPA will not ask for public comment before approving the modification. The Administrator will notify the test sponsor by certified mail of the response to the request. EPA will place copies of each application and EPA response in the public docket. EPA will publish a notice in the Federal Register annually describing such changes which have occurred during the previous year. Until such Federal Register notice is published, any modification approved by EPA shall apply only to the person or group who requested the modification; EPA shall state the applicability of

each modification in such notice.

(iv) Where, in EPA's judgment, the requested modification of a test standard would significantly change the scope of the test, EPA will publish a notice in the Federal Register requesting comment on the request and proposed modification. However, EPA may approve a requested modification of a test standard without first seeking public comment if necessary to preserve the validity of an ongoing test undertaken in good faith.

(2) [Reserved]

(h) Special Requirements for Additives. An additive which is a direct test subject, either because it is the chosen representative of a group or because it is not a member of a group, is subject to the following rules:

(1) All required emission characterization and health effects testing procedures shall be performed on the mixture which results when the additive is combined with the base fuel for the appropriate fuel family (as specified in Sec. 79.55) at the maximum concentration recommended by the additive manufacturer pursuant to Sec. 79.21(d). This combination shall be known as the additive/base fuel mixture.

(i) The appropriate fuel family to be utilized for the additive/ base fuel mixture is the fuel family which contains the specific type(s) of fuel for which the additive is presently registered or for which the manufacturer of the additive is seeking registration.

(ii) Fuels and additives belonging to more than one fuel family.

(A) If a fuel or additive product is registered in two or more fuel families as of May 27, 1994, then the manufacturer of that product is responsible for testing (or participating in group testing of) each formulation in compliance with the requirements of this subpart for each fuel family in which the manufacturer wishes to maintain a product registration for its fuel or additive.

(B) If a fuel or additive manufacturer is seeking to register such product in two or more fuel families, then the product shall be considered, for testing and registration purposes, to be a member of each fuel family in which the manufacturer is seeking registration. The manufacturer is responsible for testing (or participating in group testing of) each formulation in compliance with the requirements of this subpart for each fuel family in which the manufacturer wishes to obtain a product registration for its fuel or additive.

(iii) In the case of the methanol fuel family, which contains two base fuels (M100 and M85 base fuels, pursuant to Sec. 79.55(d)), the applicable base fuel is the one which represents the fuel/additive group (specified in Sec. 79.56(e)(4)(i)(C)) containing fuels of which the most gallons are sold annually.

(iv) Aftermarket additives which are intended by the manufacturer to be added to the fuel tank only at infrequent intervals shall be

applied according to the manufacturer's specifications during mileage accumulation, pursuant to Sec. 79.57(c). However, during emission generation and testing, each tankful of fuel used must contain the fuel additive at its maximum recommended level. If the additive manufacturer believes that this maximum treatment rate will cause adverse effects to the test engine and/or that the engine's emissions may be subject to artifacts due to overuse of the additive, then the manufacturer may submit a request to EPA for modification of this requirement and related test procedures. Such request must include objective evidence that the modification(s) are needed, along with data demonstrating the maximum concentration of the additive which may actually reach the fuel tanks of vehicles in use.

(v) Additives produced exclusively for use in #1 diesel fuel shall be tested in the diesel base fuel specified in Sec. 79.55(c), even though that base fuel is formulated with #2 diesel fuel. If a manufacturer is concerned that emissions generated from this combination of fuel and additive are subject to artifacts due to this blending, then that manufacturer may submit a request for a modification in test procedure requirements to the EPA. Any such request must include supporting test results and suggested test modifications.

(vi) Bulk additives which are used intermittently for the direct purpose of conditioning or treating a fuel during storage or transport, or for treating or maintaining the storage, pipeline, and/or other components of the fuel distribution system itself and not the vehicle/ engine for which the fuel is ultimately intended, shall, for purposes of this program, be added to the base fuel at the maximum concentration recommended by the additive manufacturer for treatment of the fuel or distribution system component. However, if the additive manufacturer believes that this treatment rate will cause adverse effects to the test engine and/or that the engine's emissions may be subject to artifacts due to overuse of the additive, then the manufacturer may submit a request to EPA for modification of this requirement and related test procedures. Such request must include objective evidence that the modification(s) are needed, along with data demonstrating the maximum concentration of the additive which may actually reach the fuel tanks of vehicles in use.

(2) EPA shall use emissions speciation and health effects data generated in the analysis of the applicable base fuel as control data for comparison with data generated for the additive/base fuel mixture.

(i) The base fuel control data may be:

(A) Generated internally as an experimental control in conjunction with testing done in compliance with registration requirements for a specific additive; or

(B) Generated externally in the course of testing different

additive(s) belonging to the same fuel family, or in the testing of a base fuel serving as representative of the baseline group for the respective fuel family pursuant to Sec. 79.56(e)(4)(i).

(ii) Control data generated using test equipment (including vehicle model and/or engine, or Evaporative Emissions Generator specifications, as appropriate) and protocols identical or nearly identical to those used in emissions and health effects testing of the subject additive/base fuel mixture would be most relevant for comparison purposes.

(iii) If an additive manufacturer chooses the same vehicle/engine to independently test the base fuel as an experimental control prior to testing the additive/base fuel mixture, then the test vehicle/engine shall undergo two mileage accumulation periods, pursuant to Sec. 79.57(c). The initial mileage accumulation period shall be performed using the base fuel alone. After base fuel testing, and prior to testing of the additive/base fuel mixture, a second mileage accumulation period shall be performed using the additive/base fuel mixture. The procedures outlined in this paragraph shall not preclude a manufacturer from testing a base fuel and the manufacturer's additive/base fuel mixture separately in identical, or nearly identical, vehicles/engines.

(i) Multiple Test Potential for Non-Baseline Products. (1) When the composition information reported in the registration application or basic registration data for a gasoline or diesel product meets criteria for classification as a non-baseline product (pursuant to Sec. 79.56(e)(3)(i)(B) or Sec. 79.56(e)(3)(ii)(B)), then the manufacturer is responsible for testing (or participating in group testing) of a separate formulation for each reported oxygenating compound, specified class of oxygenating compounds, or other substance which defines a separate non-baseline fuel/additive group pursuant to Sec. 79.56(e)(4)(ii)(A) or (B). For each such substance, testing shall be performed on a mixture of the relevant substance in the appropriate base fuel, formulated according to the specifications for the corresponding group representatives in Sec. 79.56(e)(4)(ii).

(2) When the composition information reported in the registration application or basic registration data for a non- baseline gasoline product contains a range of total oxygenate concentration-in-use which encompasses gasoline formulations with less than 1.5 weight percent oxygen as well as gasoline formulations with 1.5 weight percent oxygen or more, then the manufacturer is required to test (or participate in applicable group testing of) a baseline gasoline formulation as well as one or more non-baseline gasoline formulations as described in paragraph (h)(1) of this section.

(3) When the composition information reported in the registration application or basic registration data for a non- baseline diesel product contains a range of total oxygenate concentration-in-use which

encompasses diesel formulations with less than 1.0 weight percent oxygen as well as diesel formulations with 1.0 weight percent oxygen or more, then the manufacturer is required to test (or participate in applicable group testing) of a baseline diesel formulation as well as one or more non-baseline diesel formulations as described in paragraph (h)(1) of this section.

(j) Multiple Test Potential for Atypical Fuel Formulations. When the composition information reported in the registration application or basic registration data for a fuel product includes more than one atypical bulk additive product (pursuant to Sec. 79.56(e)(2)(iii)), and when these additives belong to different fuel/additive groups (pursuant to Sec. 79.56(e)(4)(iii)), then:

(1) When such disparate additive products are for the same purpose-in-use and are not ordinarily used in the fuel simultaneously, the fuel manufacturer shall be responsible for testing (or participating in the group testing of) a separate formulation for each such additive product. Testing related to each additive product shall be performed on a mixture of the additive in the applicable base fuel, as described in paragraph (g)(1) of this section, or by participation in the costs of testing the designated representative of the fuel/additive group to which each separate atypical additive product belongs.

(2) When the disparate additive products are not for the same purpose-in-use, the fuel manufacturer shall nevertheless be responsible for testing a separate formulation for each such additive product, as described in paragraph (g)(1) of this section, if these additives are not ordinarily blended together in the same commercial formulation of the fuel.

(3) When the disparate additive products are ordinarily blended together in the same commercial formulation of the fuel, then the fuel manufacturer shall be responsible for the testing of a single test formulation containing all such simultaneously used atypical additive products. Alternatively, this responsibility can be satisfied by enrolling such fuel product in a group which includes other fuel or additive products with the same total combination of atypical elements as that occurring in the fuel product in question. If the basic registration data for the subject fuel includes any alternative additives which contain atypical elements not represented in the test formulation, then the fuel manufacturer is also responsible for testing a separate formulation for each such additional disparate additive product.

(k) Emission Control System Testing. If any information submitted in accordance with this subpart or any other information available to EPA shows that a fuel or fuel additive may have a deleterious effect on the performance of any emission control system or device currently in use or which has been developed to a point where in a reasonable time

it would be in general use were such effect avoided, EPA may, in its judgment, require testing to determine whether such effects in fact exist. Such testing will be required in accordance with such protocols and schedules as the Administrator shall reasonably require and shall be paid for by the fuel or fuel additive manufacturer.

Sec. 79.52 Tier 1.

(a) General Specifications. Tier 1 requires manufacturers of designated fuels or fuel additives (or groups of manufacturers pursuant to Sec. 79.56) to supply to the Administrator: the identity and concentration of certain emission products of such fuels or additives; an analysis of potential emissions exposures; and any available information regarding the health and welfare effects of the whole and speciated emissions. In addition to any information required under Sec. 79.59 and in conformance with the reporting requirements thereof, manufacturers shall provide, pursuant to the timing provisions of Sec. 79.51(c), the following information.

(b) Emissions Characterization. Manufacturers must provide a characterization of the emission products which are generated by evaporation (if required pursuant to Sec. 79.58(b)) and by combustion of the fuel or additive/base fuel mixture in a motor vehicle. For this purpose, manufacturers may perform the characterization procedures described in this section or may rely on existing emission characterization data. To be considered adequate in lieu of performing new emission characterization procedures, the data must be the result of tests using the product in question or using a fuel or additive/base fuel mixture meeting the same grouping criteria as the product in question. In addition, the emissions must be generated in a manner reasonably similar to those described in Sec. 79.57, and the characterization procedures must be adequately performed and documented and must give results reasonably comparable to those which would be obtained by performing the procedures described herein. Reports of previous tests must be sufficiently detailed to allow EPA to judge the adequacy of protocols, techniques, and conclusions. After the manufacturer's submittal of such data, if EPA finds that the manufacturer has relied upon inadequate test data, then the manufacturer will not be considered to be in compliance until the corresponding tests have been conducted and the results submitted to EPA.

(1) General Provisions.

(i) The emissions to be characterized shall be generated, collected, and stored according to the processes described in Sec. 79.57. Characterization of combustion and evaporative emissions

shall be performed separately on each emission sample collected during the applicable emission generation procedure.

(ii) As provided in Sec. 79.57(d), if the emission generation vehicle/engine is ordinarily equipped with an emission aftertreatment device, then all requirements in this section for the characterization of combustion emissions must be completed both with and without the aftertreatment device in a functional state. The emissions shall be generated three times (on three different days) without a functional aftertreatment device and, if applicable, three times (on three different days) with a functional aftertreatment device, and each such time shall be analyzed according to the remaining provisions in this paragraph (b) of this section.

(iii) Measurement of background emissions. It is required that ambient/dilution air be analyzed for levels of background chemical species present at the time of emission sampling (for both combustion and evaporative emissions) and that background chemical species profiles be reported with emissions speciation data. Background chemical species measurement/analysis during the FTP is specified in Secs. 86.109-94(c)(5) and 86.135-94 of this chapter.

(iv) Concentrations of emission products shall be reported in units of grams (g) per mile and in units of weight percent of measured total hydrocarbons.

(v) Laboratory practice must be of high quality and must be consistent with state-of-the-art methods as presented in current environmental and analytical chemistry literature. Examples of analytical procedures which may be used in conducting the emission characterization/speciation requirements of this section can be found among the references in paragraph (b)(5) of this section.

(2) Characterization of the combustion emissions shall include, for products in all fuel families (except when expressly noted in this section):

(i) Determination of the concentration of the basic emissions as follows: total hydrocarbons, carbon monoxide, oxides of nitrogen, and particulates. Manufacturers are referred to the vehicle certification procedures in 40 CFR part 86, subparts B and D (Secs. 86.101 through 86.145 and Secs. 86.301 through 86.348) for guidance on the measurement of the basic emissions of interest to this subpart.

(ii) Characterization of the vapor phase of combustion emissions, as follows:

(A) Determination of the identity and concentration of individual species of hydrocarbon compounds containing 12 or fewer carbon atoms. Such characterization shall begin within 30 minutes after emission collection is completed.

(B) Determination of the identity and concentration of individual species of aldehyde and ketone compounds containing eight or fewer

carbon atoms. Characterization of these emissions captured in cartridges shall be performed within two weeks if the cartridge is stored at room temperature, and one month if the cartridge is stored at 0 deg.C or less. If the emissions are sampled using the impinger method, the sample must be stored in a capped sample vial at 0 deg.C or less and characterized within one week.

(C) Determination of the identity and concentration of individual species of alcohol and ether compounds containing six or fewer carbon atoms, for those fuels and additive/base fuel mixtures which contain alcohol and/or ether compounds containing from one to six carbon atoms in the uncombusted state. For fuel and additive formulations containing alcohols or ethers with more than six carbon atoms in the uncombusted state, alcohol and ether species with that higher number of carbon atoms or less must be identified and measured in the emissions. Such characterization shall begin within four hours after emission collection is completed.

(iii) Characterization of the semi-volatile and particulate phases of combustion emissions to identify and measure polycyclic aromatic compounds, as follows:

(A) Analysis for polycyclic aromatic compounds shall not be conducted at or soon after the start of a recommended engine lubricant change interval.

(B) Analysis for polycyclic aromatic hydrocarbons (PAHs) and nitrated polycyclic aromatic hydrocarbons (NPAHs), specified in paragraph (b)(2)(iii)(D) of this section, need not be done for any fuels and additives in the methane or propane fuel families, nor for fuels and additives in the atypical categories of any other fuel families, pursuant to the definitions of such families and categories in Sec. 79.56.

(C) Analysis for poly-chlorinated dibenzodioxins and dibenzofurans (PCDD/PCDFs), specified in paragraph (b)(2)(iii)(E) of this section, is required only for fuels and additives which contain chlorine as an atypical element, pursuant to paragraph (b)(2)(iv) of this section, which requires all individual emission products containing atypical elements to be determined for atypical fuels and additives. However, manufacturers of baseline and nonbaseline fuels and fuel additives in all fuel families, except those in the methane and propane fuel families, are strongly encouraged to conduct these analyses on a voluntary basis.

(D) The analytical method used to measure species of PAHs and NPAHs should be capable of detecting at least 1 ppm (equivalent to 0.001 microgram (<greek-m>g) of compound per milligram of organic extract) of these compounds in the extractable organic matter. The concentration of each individual PAH or NPAH compound identified shall be reported in units of microgram per mile. Each compound which is present at 0.001

<greek-m>g per mile or more must be identified, measured, and reported. The following individual species shall be measured:

(1) PAHs:

(i) Benzo(a)anthracene;

(ii) Benzo[b]fluoranthene;

(iii) Benzo[k]fluoranthene;

(iv) Benzo(a)pyrene;

(v) Chrysene;

(vi) Dibenzo[a,h]anthracene; and

(vii) Indeno[1,2,3-c,d]pyrene.

(2) NPAHs:

(i) 7-Nitrobenzo[a]anthracene;

(ii) 6-Nitrobenzo[a]pyrene;

(iii) 6-Nitrochrysene;

(iv) 2-Nitrofluorene; and

(v) 1-Nitropyrene.

(E) The analytical method used to measure species and classes of PCDD/PCDFs should be capable of detecting at least 1 part per trillion (ppt) (equivalent to 0.001 picogram (pg) of compound per milligram of organic extract) of these compounds in the extractable organic matter. The concentration of each individual PCDD/PCDF compound identified shall be reported in units of picograms (pg) per mile. Each compound which is present at 0.5 pg per mile or more must be identified, measured, and reported.

(1) With respect to measurement of PCDD/PCDFs only, the liquid extracts from the particulate and semi-volatile emissions fractions may be combined into one sample for analysis.

(2) The manufacturer is referred to 40 CFR part 60, appendix A, Method 23 for a protocol which may be used to identify and measure any potential PCDD/PCDFs which might be present in exhaust emissions from a fuel or additive/base fuel mixture.

(3) The following individual compounds and classes of compounds of PCDD/PCDFs shall be identified and measured:

(i) Individual tetra-chloro-substituted dibenzodioxins (tetra-CDDs);

(ii) Individual tetra-chloro-substituted dibenzofurans (tetra-CDFs);

(iii) Penta-CDDs and penta-CDFs, as one class;

(iv) Hexa-CDDs and hexa-CDFs, as one class;

(v) Hepta-CDDs and hepta-CDFs as one class; and

(vi) Octo-CDDs and octo-CDFs as one class.

(iv) With respect to all phases (vapor, semi-volatile, and particulate) of combustion emissions generated from those fuels and additive/base fuel mixtures classified in the atypical categories (pursuant to Sec. 79.56), the identity and concentration of individual

emission products containing such atypical elements shall also be determined.

(3) For evaporative fuels and evaporative fuel additives, characterization of the evaporative emissions shall include:

(i) Determination of the concentration of total hydrocarbons for the applicable vehicle type and class in 40 CFR part 86, subpart B (Secs. 86.101 through 86.145).

(ii) Determination of the identity and concentration of individual species of hydrocarbon compounds containing 12 or fewer carbon atoms. Such characterization shall begin within 30 minutes after emission collection is completed.

(iii) In the case of those fuels and additive/base fuel mixtures which contain alcohol and/or ether compounds in the uncombusted state, determination of the identity and concentration of individual species of alcohol and ether compounds containing six or fewer carbon atoms. For fuel and additive formulations containing alcohols or ethers with more than six carbon atoms in the uncombusted state, alcohol and ether species with that higher number of carbon atoms or less must be identified and measured in the emissions. Such characterization shall begin within four hours after emission collection is completed.

(iv) In the case of those fuels and additive/base fuel mixtures which contain atypical elements, determination of the identity and concentration of individual emission products containing such atypical elements.

(4) Laboratory quality control. (i) At a minimum, laboratories performing the procedures specified in this section shall conduct calibration testing of their emissions characterization equipment before each new fuel/additive product test start-up. Known samples representative of the compounds potentially to be found in emissions from the product to be characterized shall be used to calibrate such equipment.

(ii) Laboratories performing the procedures specified in this section shall agree to permit quality control inspections by EPA, and for this purpose shall admit any EPA Enforcement Officer, upon proper presentation of credentials, to any facility where vehicles are conditioned or where emissions are generated, collected, stored, sampled, or characterized in meeting the requirements of this section. Such laboratory audits may include EPA distribution of ``blind'' samples for analysis by participating laboratories.

(5) References. For additional background information on the emission characterization procedures outlined in this paragraph, the following references may be consulted:

(i) ``Advanced Emission Speciation Methodologies for the Auto/Oil Air Quality Improvement Program--I. Hydrocarbons and Ethers," Auto Oil Air Quality Improvement Research Program, SP-920, 920320, SAE, February

1992.

(ii) ``Advanced Speciation Methodologies for the Auto/Oil Air Quality Improvement Research Program--II. Aldehydes, Ketones, and Alcohols," Auto Oil Air Quality Improvement Research Program, SP-920, 920321, SAE, February 1992.

(iii) ASTM D 5197-91, ``Standard Test Method for Determination of Formaldehyde and Other Carbonyl Compounds in Air (Active Sampler Methodology)."

(iv) Johnson J. H., Bagley, S. T., Gratz, L. D., and Leddy, D. G., ``A Review of Diesel Particulate Control Technology and Emissions Effects--1992 Horning Memorial Award Lecture," SAE Technical Paper Series, SAE 940233, 1994.

(v) Keith et al., ACS Committee on Environmental Improvement, ``Principles of Environmental Analysis," The Journal of Analytical Chemistry, Volume 55, pp. 2210-2218, 1983.

(vi) Perez, J.M., Jabs, R.E., Leddy, D.G., eds. ``Chemical Methods for the Measurement of Unregulated Diesel Emissions (CRC-APRAC Project No. CAPI-1-64), Coordinating Research Council, CRC Report No. 551, August, 1987.

(vii) Schuetzle, D., ``Analysis of Nitrated Polycyclic Aromatic Hydrocarbons in Diesel Particulates," Analytical Chemistry, Volume 54, pp. 265-271, 1982.

(viii) Siegl, W.O., et al., ``Improved Emissions Speciation Methodology for Phase II of the Auto/Oil Air Quality Improvement Research Program--Hydrocarbons and Oxygenates", SAE Technical Paper Series, SAE 930142, 1993.

(ix) Tejada, S. B. et al., ``Analysis of Nitroaromatics in Diesel and Gasoline Car Emissions," SAE Paper No. 820775, 1982.

(x) Tejada, S. B. et al., ``Fluorescence Detection and Identification of Nitro Derivatives of Polynuclear Aromatic Hydrocarbons by On-Column Catalytic Reduction to Aromatic Amines," Analytical Chemistry, Volume 58, pp. 1827-1834, July 1986.

(xi) ``Test Method for Determination of C1-C4 Alcohols and MTBE in Gasoline by Gas Chromatography," 40 CFR part 80, appendix F.

(c) Exposure Analysis. Using annual and projected production volume, marketing, and distribution data submitted as part of the basic registration data, specified in Sec. 79.59(b), manufacturers shall provide a qualitative discussion of the potential public health exposure(s) of the general population and any special at-risk populations to the emission products of their fuel or additive product(s). The analysis accompanying a group submission shall address the characteristics of the cumulative exposure resulting from the use of all fuel or additive products in the group. Modeling and other quantitative approaches to the analysis are encouraged when the appropriate data is available.

(d) Literature Search. (1) Manufacturers of fuels and fuel additives shall conduct a literature search and compilation of information on the potential toxicologic, environmental, and other public welfare effects of the emissions of such fuels and additives. The literature search shall include all available relevant information from in-house, industry, government, and public sources pertaining to the emissions of the subject fuel or fuel additive or the emissions of similar fuels or additives, with such similarity determined according to the provisions of Sec. 79.56.

(2) The literature search shall address the potential adverse effects of whole combustion emissions, evaporative emissions, relevant emission fractions, and individual emission products of the subject fuel or fuel additive except as specified in the following paragraph. The individual emission products to be included are those identified pursuant to the emission characterization procedures specified in paragraph (b) of this section, other than carbon monoxide, carbon dioxide, nitrogen oxides, benzene, 1,3-butadiene, acetaldehyde, and formaldehyde.

(3) In the case of the individual emission products of non-baseline or atypical fuels and additives (pursuant to Sec. 79.56(e)(2)), the literature data need not be submitted for those emission products which are the same as the combustion emission products of the respective base fuel for the product's fuel family (pursuant to Sec. 79.55). For this purpose, data on the base fuel emission products for the product's fuel family:

(i) May be found in the literature of previously-conducted, adequate emission speciation studies for the base fuel, or for a fuel or additive/fuel mixture capable of grouping with the base fuel (see, for example, the references in paragraph (b)(5) of this section).

(ii) May be compiled while gathering internal control data during emissions characterization studies on the manufacturer's non-baseline or atypical product; or

(iii) May be obtained from various manufacturers in the course of their testing different additive(s) belonging to the same fuel family, or in the testing of a base fuel serving as representative of the baseline group for the respective fuel family.

(e) Data bases. The literature search must include the results of searching appropriate commercially available chemical, toxicologic, and environmental databases. The databases shall be searched using, at a minimum, CAS numbers (when applicable), chemical names, and common synonyms.

(f) Search period. The literature search shall cover a time period beginning at least thirty years prior to the date of submission of the reports specified in Secs. 79.59(b) through (c) and ending no earlier than six months prior to the date on which testing is commenced or

reports are submitted in compliance with this subpart.

(g) References. Information on base fuel emission inventories may be found in references in paragraphs (b)(5)(i) through (xi) of this section, as well as in the following:

(1) Auto/Oil Air Quality Improvement Research Program, Technical Bulletin #1, December 1990.

(2) Keith et al., ACS Committee on Environmental Improvement, ``Principles of Environmental Analysis,'' The Journal of Analytical Chemistry, Volume 55, pp. 2210-2218, 1983.

(3) ``The Composition of Gasoline Engine Hydrocarbon Emissions--An Evaluation of Catalyst and Fuel Effects''--SAE 902074 and ``Speciated Hydrocarbon Emissions from Aromatic, Olefin, and Paraffinic Model Fuels''--SAE 930373.

Sec. 79.53 Tier 2.

(a) Generally. Subject to the provisions of Sec. 79.53(b) through (d), the combustion emissions of each fuel or fuel additive subject to testing under this subpart must be tested in accordance with each of the testing guidelines in Secs. 79.60 through 79.68, except that fuels and additives in the methane and propane fuel families (pursuant to Sec. 79.56(e)(1)(v) and (vi)) need not undergo the Salmonella mutagenicity assay in Sec. 79.68). Similarly, subject to the provisions of Sec. 79.53(b) through (d), the evaporative emissions of each designated evaporative fuel and each designated evaporative fuel additive subject to testing under this subpart must be tested according to each of the testing guidelines in Secs. 79.60 through 79.67 (excluding Sec. 79.68, Salmonella typhimurium Reverse Mutation Assay).

(b) Manufacturer Determination. Manufacturers shall determine whether the information gathered pursuant to the literature search in Sec. 79.52(d) contains the results of adequately performed and adequately documented previous testing which provides information reasonably comparable to that supplied by the health tests described in Secs. 79.62 through 79.68 regarding the carcinogenicity, mutagenicity, neurotoxicity, teratogenicity, reproductive/fertility measures, and general toxicity effects of the emissions of the fuel or additive. When manufacturers make an affirmative determination, they need submit only the information gathered pursuant to Sec. 79.52(d) for such tests. EPA maintains final authority in judging whether the information is an adequate substitution in lieu of conducting the associated tests. EPA's determination of the adequacy of existing information shall be guided by the considerations described in paragraph (d) of this section. If EPA finds that the manufacturer has relied upon inadequate test data, then the manufacturer will not be considered to be in compliance until

the corresponding tests have been conducted and the results submitted to EPA.

(c) Testing. (1) All testing required pursuant to this section must be done in accordance with the procedures, equipment, and facility requirements described in Secs. 79.57, 79.60, and 79.61 regarding emissions generation, good laboratory practices, and inhalation exposure testing, respectively, as well as any other requirements described in this subpart. The laboratory conducting the animal studies shall be registered and in good standing with the United States Department of Agriculture and regularly inspected by United States Department of Agriculture veterinarians. In addition, the facility must be accredited by a generally recognized independent organization which sets laboratory animal care standards. Use of inadequate test protocols or substandard laboratory techniques in performing any testing required by this subpart may result in cancellation of all affected registrations.

(2) Carcinogenic or mutagenic effects in animals from emissions exposures shall be determined pursuant to Sec. 79.64 In vivo Micronucleus Assay, Sec. 79.65 In vivo Sister Chromatid Exchange Assay, and Sec. 79.68 Salmonella typhimurium Reverse Mutation Assay. Teratogenic effects and reproductive toxicity shall be examined pursuant to Sec. 79.63 Fertility Assessment/Teratology. General toxicity and pulmonary effects shall be determined pursuant to Sec. 79.62 Subchronic Toxicity Study with Specific Health Effect Assessments. Neurotoxic effects shall be determined pursuant to Sec. 79.66 Neuropathology Assessment and Sec. 79.67 Glial Fibrillary Acidic Protein Assay.

(d) EPA Determination. (1) After submission of all information and testing, EPA in its judgment shall determine whether previously conducted tests relied upon in the registration submission are adequately performed and documented and provide information reasonably comparable to that which would be provided by the tests described herein. Manufacturers' submissions shall be sufficiently detailed to allow EPA to judge the adequacy of protocols, techniques, experimental design, statistical analyses, and conclusions. Studies shall be performed using generally accepted scientific principles, good laboratory techniques, and the testing guidelines specified in these regulations.

(2) EPA shall give appropriate weight when making this determination to the following factors:

(i) The age of the data;

(ii) The adequacy of documentation of procedures, findings, and conclusions;

(iii) The extent to which the testing conforms to generally accepted scientific principles and practices;

(iv) The type and number of test subjects;

(v) The number and adequacy of exposure concentrations, i.e., emission dilutions;

(vi) The degree to which the tested emissions were generated by procedures and under conditions reasonably comparable to those set forth in Sec. 79.57; and

(vii) The degree to which the test procedures conform to the testing guidelines set forth in Secs. 79.60 through 79.68 and/or furnish information comparable to that provided by such testing.

(3) The test animals shall be rodents, preferably a strain of rat, and testing shall include all of the endpoints covered in Secs. 79.62 through 79.68. All studies shall be properly executed, with appropriate documentation, and in accord with the individual health testing guidelines (Secs. 79.60 through 79.68) of this part, e.g., 90-day, 6-hour per day exposure, minimum.

(4) In general, the data in a manufacturer's registration submittal shall be adequate if the duration of a test's exposure period is at least as long, in days and hours, as the inhalation exposure specified in the related health test guideline(s). Data from tests with shorter exposure durations than those specified in the guidelines may be acceptable if the test results are positive (i.e., exhibit adverse effects) and/or include a demonstrable concentration-response relationship.

(5) Data in support of a manufacturer's registration submittal shall directly address the effects of inhalation exposure to the whole evaporative and exhaust emissions of the respective fuel or additive or to the whole evaporative and exhaust emissions of other fuels or additives which satisfy the criteria in Sec. 79.56 for classification into the same group as the subject fuel or fuel additive. Data obtained in the testing of a raw liquid fuel or additive/base fuel mixture or a raw, aerosolized fuel or additive/base fuel mixture shall not be adequate to support a manufacturer's registration submittal. Data from testing of evaporative emissions cannot substitute for test data on combustion emissions. Data from testing of combustion emissions cannot substitute for test data on evaporative emissions.

Sec. 79.54 Tier 3.

(a) General Criteria for Requiring Tier 3 Testing. (1) Tier 3 testing shall be required of a manufacturer or group of manufacturers at EPA's discretion when remaining uncertainties as to the significance of observed health effects, welfare effects, and/or emissions exposures from a fuel or fuel/additive mixture interfere with EPA's ability to make reasonable estimates of the potential risks posed by emissions

from the fuel or additive products. Tier 3 testing may be conducted either on an individual basis or a group basis. If performed on a group basis, EPA may require either the same representative to be used in Tier 3 testing as was used in Tier 2 testing or may select a different member or members of the group to represent the group in the Tier 3 tests.

(2) In addition to the criteria specific to particular tests as summarized and detailed in the testing guidelines (Secs. 79.62 through 79.68), EPA may consider a number of factors (including, but not limited to):

(i) The number of positive and negative outcomes related to each endpoint;

(ii) The identification of concentration-effect relationships;

(iii) The statistical sensitivity and significance of such studies;

(iv) The severity of the observed effects (e.g., whether the effects would be likely to lead to incapacitating or irreversible conditions);

(v) The type and number of species included in the reported tests;

(vi) The consistency and clarity of apparent mechanisms, target organs, and outcomes;

(vii) The presence or absence of effective health test control data for base-fuel-only versus additive/base fuel mixture comparisons;

(viii) The nature and amount of known toxic agents in the emissions stream; and

(ix) The observation of lesions which specifically implicate inhalation as an important exposure route.

(3) Consideration of exposure. EPA retains discretion to consider, in addition to available toxicity data, any Tier 1 data on potential exposures to emissions from a particular fuel or fuel additive (or group of fuels and/or fuel additives) in determining whether to require Tier 3 testing. EPA may consider, but is not limited to, the following factors:

(i) Types and emission rates of speciated emission components;

(ii) Types and emission rates of combinations of compounds or elements of concern;

(iii) Historical and/or projected production volumes and market distributions; and

(iv) Estimated population and/or environmental exposures obtained through extrapolation, modeling, or literature search findings on ambient, occupational, or epidemiological exposures.

(b) Notice. (1) EPA will determine whether Tier 3 testing is necessary upon receipt of a manufacturer's (or group's) submittal as prescribed under Sec. 79.51(d). If EPA determines on the basis of the Tier 1 and 2 data submission and any other available information that further testing is necessary, EPA will require the responsible

manufacturer(s) to conduct testing as described elsewhere in this section. EPA will notify the manufacturer (or group) by certified letter of the purpose and nature of any proposed testing and of the proposed deadline for completing the testing. A copy of the letter will be placed in the public record. EPA will provide the manufacturer a 60-day comment period after the manufacturer's receipt of such notice. EPA may extend the comment period if it appears from the nature of the issues raised that further discussion is warranted. In the event that no comment is received by EPA from the manufacturer (or group) within the comment period, the manufacturer (or group) shall be deemed to have consented to the adoption by EPA of the proposed Tier 3 requirements.

(2) EPA will issue a notice in the Federal Register of its intent to require testing under Tier 3 for a particular fuel or additive manufacturer and that a copy of the letter to the manufacturer outlining the Tier 3 testing for that manufacturer is available in the public record for review and comment. The public shall have a minimum of thirty (30) days after the publication of this notice to comment on the proposed Tier 3 testing.

(3) EPA will include in the public record a copy of any timely comments concerning the proposed Tier 3 testing requirements received from the affected manufacturer or group or from the public, and the responses of EPA to such comments. After reviewing all such comments received, EPA will adopt final Tier 3 requirements by sending a certified letter describing such final requirements to the manufacturer or group. EPA will also issue a notice in the Federal Register announcing that it has adopted such final Tier 3 requirements and that a copy of the letter adopting the requirements has been included in the public record.

(4) Prior to beginning any required Tier 3 testing, the manufacturer shall submit detailed test protocols to EPA for approval. Once EPA has determined the Tier 3 testing requirements and approves the test protocols, any modification to the requirements shall be governed by Sec. 79.51(f).

(c) Carcinogenicity and Mutagenicity Testing. (1) A potential need for Tier 3 carcinogenicity and/or mutagenicity testing may be indicated if the results of the In vivo Micronucleus Assay, required under Sec. 79.64, the In vivo Sister Chromatid Exchange Assay, required under Sec. 79.65, the Salmonella mutagenicity assay required under Sec. 79.68, or relevant pathologic findings under Sec. 79.62 demonstrate a statistically significant dose-related positive response as compared with appropriate controls. Alternatively, Tier 3 carcinogenicity testing and/or mutagenicity testing may be required if there are positive outcomes for at least one concentration in two or more of the tests required under Secs. 79.64, 79.65, and 79.68.

(2) The testing for carcinogenicity required under this paragraph

may, at EPA's discretion, be conducted in accordance with 40 CFR 798.3300 or 798.3320, or their equivalents (see suggested references following each health effects testing guideline). The testing for mutagenicity required under this paragraph may likewise be conducted in accordance with 40 CFR 798.5195, 798.5500, 798.5955, 798.7100, and/or other suitable equivalent testing (see suggested references following each health effects testing guideline). EPA may supplement or modify guidelines as required to ensure that the prescribed testing addresses the identified areas of concern.

(d) Reproductive and Teratological Effects Testing. (1) A potential need for Tier 3 testing may be indicated if the results of the Fertility Assessment/Teratology study required under Sec. 79.63 or relevant findings under Sec. 79.62 demonstrate, in comparison with appropriate controls, a statistically significant dose-related positive response in one or more of the possible test outcomes. Similarly, Tier 3 testing may be indicated if statistically significant positive results are confined to either sex, or to the fetus as opposed to the pregnant adult.

(2) The testing for reproductive and teratological effects required under this paragraph may, at EPA's discretion, be conducted in accordance with 40 CFR 798.4700 and/or by performance of a reproductive assay by continuous breeding. These guidelines may be modified or supplemented by EPA as required to ensure that the prescribed testing addresses the identified areas of concern.

(e) Neurotoxicity Testing. (1) A potential need for Tier 3 neurotoxicity testing may be indicated if either the results of the Neuropathology Assessment required under Sec. 79.67 shows concentration-related effects in exposed animals or the Glial Fibrillary Acidic Protein Assay required under Sec. 79.66 demonstrates a statistically significant concentration-related positive response as compared with appropriate controls. Similarly, Tier 3 neurotoxicity testing may be indicated if relevant results under Sec. 79.62 demonstrate a statistically significant positive response in comparison to appropriate controls.

(2) The testing for neurotoxicity required under this paragraph may, at EPA's discretion, be conducted in accordance with 40 CFR 798.3260 and 40 CFR part 798 subpart G. These guidelines may be modified or supplemented by EPA as required to ensure that the prescribed testing addresses the identified areas of concern.

(f) General and Pulmonary Toxicity Testing. (1) A potential need for Tier 3 general and/or pulmonary toxicity testing may be indicated if, in comparison with appropriate controls, the results of the Subchronic Toxicity Study, pursuant to Sec. 79.62, demonstrate abnormal gross analysis or histopathological findings (especially as relates to lung pathology from whole-body preserved test animals) or persistence

or delayed occurrence of toxic effects beyond the exposure period.

(2) A potential need for Tier 3 testing with respect to other organ systems or endpoints not addressed by specific Tier 2 tests, e.g., hepatic, renal, or endocrine toxicity, may be demonstrated by findings in the Tier 2 Subchronic Toxicity Study (pursuant to Sec. 79.62) or by findings in the Tier 1 literature search of adverse functional, physiologic, metabolic, or histopathologic effects of fuel or additive emissions to such other organ systems or any other information available to EPA. In addition, findings in the Tier 1 emission characterization of significant levels of a known toxicant to such other organ systems and endpoints may also indicate a need for relevant health effects testing. The testing required under this paragraph may include tests conducted in accordance with 40 CFR 798.3260 or 798.3320. These guidelines may be modified or supplemented by EPA as necessary to ensure that the prescribed testing addresses the identified areas of concern.

(3) The testing for general/pulmonary toxicity required under this paragraph may, at EPA's discretion, be conducted in accordance with 40 CFR 798.2450 or 798.3260. These guidelines may be modified or supplemented by EPA as necessary to ensure that the prescribed testing addresses the identified areas of concern. Pulmonary function measurements, host defense assays, immunotoxicity tests, cell morphology/morphometry, and/or enzyme assays of lung lavage cells and fluids may be specifically required.

(g) Other Tier 3 Testing. (1) A manufacturer or group may be required to use up-to-date modeling, sampling, monitoring, and/or analytic approaches at the Tier 3 level to provide:

(i) Estimates of exposures to the emission products of a fuel or fuel additive or group of products;

(ii) The expected atmospheric transformation products of such emissions; and

(iii) The environmental partitioning of such emissions to the air, soil, water, and biota.

(2) Additional emission characterization may be required if uncertainty over the identity of chemical species or rate of their emission interferes with reasonable judgments as to the presence and/or concentration of potentially toxic substances in the emissions of a fuel or fuel additive. The required tests may include characterization of additional classes of emissions, the characterization of emissions generated by additional vehicles/engines of various technology mixes (e.g., catalyzed versus non-catalyzed emissions), and/or other more precise analytic procedures for identification or quantification of emissions compounds. Additional emissions testing may also be required to evaluate concerns which may arise regarding the potential effects of a fuel or fuel additive on the performance of emission control

equipment.

(3) A manufacturer or group may be required to conduct biological and/or exposure studies at the Tier 3 level to evaluate directly the potential public welfare or environmental effects of the emissions of a fuel or additive, if significant concerns about such effects arise as a result of EPA's review of the literature search or emission characterization findings in Tier 1 or the results of the toxicological tests in Tier 2.

(4) With regard to group submittals, Tier 3 studies on a fuel or additive product(s) other than the originally specified group representative may be required if specific differences in the product's composition indicate that its emissions may have different toxicologic properties from those of the original group representative.

(5) Additional emission characterization and/or toxicologic tests may be required to evaluate the impact of different vehicle, engine, or emission control technologies on the observed composition or health or welfare effects of the emissions of a fuel or additive.

(6) Toxicological tests on individual emission products may be required.

(7) Upon review of information submitted for an aerosol product under Sec. 79.58(e), emissions characterization, exposure, and/or toxicologic testing at a Tier 3 level may be required.

(8) A manufacturer which qualifies for and has elected to use the special provisions for the products of small businesses (pursuant to Sec. 79.58(d)) may be required to conduct emission characterization, exposure, and /or toxicologic studies at the Tier 3 level for such products, as specified in Sec. 79.58(d)(4).

(9) The examples of potential Tier 3 tests described in this section do not in any way limit EPA's broad discretion and authority under Tier 3.

Sec. 79.55 Base fuel specifications.

(a) General Characteristics. (1) The base fuel(s) in each fuel family shall serve as the group representative(s) for the baseline group(s) in each fuel family pursuant to Sec. 79.56. Also, as specified in Sec. 79.51(h)(1), for fuel additives undergoing testing, the designated base fuel for the respective fuel family shall serve as the substrate in which the additive shall be mixed prior to the generation of emissions.

(2) Base fuels shall contain a limited complement of the additives which are essential for the fuel's production or distribution and/or for the successful operation of the test vehicle/engine throughout the mileage accumulation and emission generation periods. Such additives

shall be used at the minimum effective concentration-in-use for the base fuel in question.

(3) Unless otherwise restricted, the presence of trace contaminants does not preclude the use of a fuel or fuel additive as a component of a base fuel formulation.

(4) When an additive is the test subject, any additive normally contained in the base fuel which serves the same function as the subject additive shall be removed from the base fuel formulation. For example, if a corrosion inhibitor were the subject of testing and if this additive were to be tested in a base fuel which normally contained a corrosion inhibitor, this test additive would replace the corrosion inhibitor normally included as a component of the base fuel.

(5) Additive components of the methanol, ethanol, methane, and propane base fuels in addition to any such additives included below shall be limited to those recommended by the manufacturers of the vehicles and/or engines used in testing such fuels. For this purpose, EPA will review requests from manufacturers (or their agents) to modify the additive specifications for the alternative fuels and, if necessary, EPA shall change these specifications based on consistency of those changes with the associated vehicle manufacturer's recommendations for the operation of the vehicle. EPA shall publish notice of any such changes to a base fuel and/or its base additive package specifications in the Federal Register.

(b) Gasoline Base Fuel. (1) The gasoline base fuel is patterned after the reformulated gasoline summer baseline fuel as specified in CAA section 211(k)(10)(B)(i). The specifications and blending tolerances for the gasoline base fuel are listed in Table F94-1. The additive types which shall be required and/or permissible in the gasoline base fuel are listed in Table 1 as well.

Table F94-1.--Gasoline Base Fuel Properties

API Gravity	57.4<plus-minus>0.3
Sulfur, ppm	339<plus-minus>25
Benzene, vol%	1.53<plus-minus>0.3
RVP, psi	8.7<plus-minus>0.3
Octane, (R+M)/2	87.3<plus-minus>0.5
Distillation Parameters:	
10%, deg.F	128<plus-minus>5
50%, deg.F	218<plus-minus>5
90%, deg.F	330<plus-minus>5
Aromatics, vol%	32.0<plus-minus>2.7
Olefins, vol%	9.2<plus-minus>2.5

Saturates, vol%..... 58.8<plus-minus>2.0
Additive Types:
Required..... Deposit Control
Corrosion Inhibitor
Demulsifier
Anti-oxidant
Metal Deactivator
Permissible..... Anti-static

(2) The additive components of the gasoline base fuel shall contain compounds comprised of no elements other than carbon, hydrogen, oxygen, nitrogen, and sulfur. Additives shall be used at the minimum concentration needed to perform effectively in the gasoline base fuel. In no case shall their concentration in the base fuel exceed the maximum concentration recommended by the additive manufacturer. The increment of sulfur contributed to the formulation by any additive shall not exceed 15 parts per million sulfur by weight and shall not cause the gasoline base fuel to exceed the sulfur specifications in Table F94-1 of this section.

(c) Diesel Base Fuel. (1) The diesel base fuel shall be a #2 diesel fuel having the properties and blending tolerances shown in Table F94-2 of this section. The additive types which shall be permissible in diesel base fuel are presented in Table F94-2 as well.

Table F94-2.--Diesel Base Fuel Properties

API Gravity..... 33<plus-minus>1
Sulfur, wt%..... 0.05<plus-minus>0.0025
Cetane Number..... 45.2<plus-minus>2
Cetane Index..... 45.7<plus-minus>2
Distillation Parameters:
10%, deg.F..... 433<plus-minus>5
50%, deg.F..... 516<plus-minus>5
90%, deg.F..... 606<plus-minus>5
Aromatics, vol%..... 38.4<plus-minus>2.7
Olefins, vol%..... 1.5<plus-minus>0.4
Saturates, vol%..... 60.1<plus-minus>2.0
Additive Types:
Required..... Corrosion Inhibitor
Demulsifier
Anti-oxidant

	Metal Deactivator
Permitted.....	Anti-static
	Flow Improver
Not Permitted.....	Deposit Control

(2) The additive components of the diesel base fuel shall contain compounds comprised of no elements other than carbon, hydrogen, oxygen, nitrogen, and sulfur. Additives shall be used at the minimum concentration needed to perform effectively in the diesel base fuel. In no case shall their concentration in the base fuel exceed the maximum concentration recommended by the additive manufacturer. The increment of sulfur contributed to the base fuel by additives shall not cause the diesel base fuel to exceed the sulfur specifications in Table F94-2 of this section.

(d) Methanol Base Fuels. (1) The methanol base fuels shall contain no elements other than carbon, hydrogen, oxygen, nitrogen, sulfur, and chlorine.

(2) The M100 base fuel shall consist of 100 percent by volume chemical grade methanol.

(3) The M85 base fuel is to contain 85 percent by volume chemical grade methanol, blended with 15 percent by volume gasoline base fuel meeting the gasoline base fuel specifications outlined in paragraph (b)(1) of this section. Manufacturers shall ensure the methanol compatibility of lubricating oils as well as fuel additives used in the gasoline portion of the M85 base fuel.

(4) The methanol base fuels shall meet the specifications listed in Table F94-3.

Table F94-3.--Methanol Base Fuel Properties

M100:	
Chemical Grade MeOH, vol%.....	100
Chlorine (as chlorides), wt%, max.....	0.0001
Water, wt%, max.....	0.5
Sulfur, wt%, max.....	0.002
M85	
Chemical Grade MeOH, vol%,.....	85
Gasoline Base Fuel, vol%.....	15
Chlorine (as chlorides), wt%, max.....	0.0001
Water, wt%, max.....	0.5
Sulfur, wt%, max.....	0.004

(e) Ethanol Base Fuel. (1) The ethanol base fuel, E85, shall

contain no elements other than carbon, hydrogen, oxygen, nitrogen, sulfur, chlorine, and copper.

(2) The ethanol base fuel shall contain 85 percent by volume chemical grade ethanol, blended with 15 percent by volume gasoline base fuel that meets the specifications listed in paragraph (b)(1) of this section. Additives used in the gasoline component of E85 shall be ethanol-compatible.

(3) The ethanol base fuel shall meet the specifications listed in Table F94-4.

Table F94-4.--Ethanol Base Fuel Properties

E85:

Chemical Grade EtOH, vol%, min.	85
Gasoline Base Fuel, vol%	15
Chlorine (as chloride), wt%, max.	0.0004
Copper, mg/L, max.	0.07
Water, wt%, max.	0.5
Sulfur, wt%, max.	0.004

(f) Methane Base Fuel. (1) The methane base fuel is a gaseous motor vehicle fuel marketed commercially as compressed natural gas (CNG), whose primary constituent is methane.

(2) The methane base fuel shall contain no elements other than carbon, hydrogen, oxygen, nitrogen, and sulfur. The fuel shall contain an odorant additive for leak detection purposes. The added odorant shall be used at a level such that, at ambient conditions, the fuel must have a distinctive odor potent enough for its presence to be detected down to a concentration in air of not over \1/5\ (one-fifth) of the lower limit of flammability. After addition of the odorant, the methane base fuel shall contain no more than 16 ppm sulfur by volume.

(3) The methane base fuel shall meet the specifications listed in Table F94-5.

Table F94-5.--Methane Base Fuel Specifications

Methane, mole%, min.	89.0
Ethane, mole%, max.	4.5
Propane and higher HC, mole%, max.	2.3
C6 and higher HC, mole%, max.	0.2

Oxygen, mole%, max..... 0.6
Sulfur (including odorant additive) ppmv, max..... 16
Inert gases:
Sum of CO<INF>2 and N<INF>2, mole%, max..... 4.0

(g) Propane Base Fuel. (1) The propane base fuel is a gaseous motor vehicle fuel, marketed commercially as liquified petroleum gas (LPG), whose primary constituent is propane.

(2) The propane base fuel may contain no elements other than carbon, hydrogen, oxygen, nitrogen, and sulfur. The fuel shall contain an odorant additive for leak detection purposes. The added odorant shall be used at a level such that at ambient conditions the fuel must have a distinctive odor potent enough for its presence to be detected down to a concentration in air of not over \1/5\ (one-fifth) of the lower limit of flammability. After addition of the odorant, the propane base fuel shall contain no more than 120 ppm sulfur by weight.

(3) The propane base fuel shall meet the specifications listed in Table F94-6.

Table F94--6.--Propane Base Fuel Specifications

Vapor pressure at 100-F, psig, max..... 208
Evaporative temperature, 95%, deg.F, max..... -37
Propane, vol%, min..... 92.5
Propylene, vol%, max..... 5.0
Butane and heavier, vol%, max..... 2.5
Residue-evaporation of 100mL, max, mL..... 0.05
Sulfur (including odorant additive) ppmw, max..... 123

Sec. 79.56 Fuel and fuel additive grouping system.

(a) Manufacturers of fuels and fuel additives are allowed to satisfy the testing requirements in Secs. 79.52, 79.53, and 79.54 and the associated reporting requirements in Sec. 79.59 on an individual or group basis, provided that such products meet the criteria in this section for enrollment in the same fuel/additive group. However, each manufacturer of a fuel or fuel additive must individually comply with the notification requirements of Sec. 79.59(b). Further, if a manufacturer elects to comply by participation in a group, each manufacturer continues to be individually subject to the information requirements of this subpart.

(1) The use of the grouping provision to comply with Tier 1 and Tier 2 testing requirements is voluntary. No manufacturer is prohibited from testing and submitting its own data for its own product registration, despite its qualification for membership in a particular group.

(2) The only groups permitted are those established in this section.

(b) Each manufacturer who chooses to enroll a fuel or fuel additive in a group of similar fuels and fuel additives as designated in this section may satisfy the registration requirements through a group submission of jointly-sponsored testing and analysis conducted on a product which is representative of all products in that group, provided that the group representative is chosen according to the specifications in this section.

(1) The health effects information submitted by a group shall be considered applicable to all fuels and fuel additives in the group. A fuel or fuel additive manufacturer who has chosen to participate in a group may subsequently choose to perform testing of such fuel or fuel additive on an individual basis; however, until such independent registration information has been received and reviewed by EPA, the information initially submitted by the group on behalf of the manufacturer's fuel or fuel additive shall be considered applicable and valid for that fuel or fuel additive. It could therefore be used to support requirements for further testing under the provisions of Tier 3 or to support regulatory decisions affecting that fuel or fuel additive.

(2) Manufacturers are responsible for determining the appropriate groups for their products according to the criteria in this section and for enrolling their products into those groups under industry-sponsored or other independent brokering arrangements.

(3) Manufacturers who enroll a fuel or fuel additive into a group shall share the applicable costs according to appropriate arrangements established by the group. The organization and administration of group functions and the development of cost-sharing arrangements are the responsibility of the participating manufacturers. If manufacturers are unable to agree on fair and equitable cost sharing arrangements and if such dispute is referred by one or more manufacturers to EPA for resolution, then the provisions in Sec. 79.56(c) (1) and (2) shall apply.

(c) In complying with the registration requirements for a given fuel or fuel additive, notwithstanding the enrollment of such fuel or additive in a group, a manufacturer may make use of available information for any product which conforms to the same grouping criteria as the given product. If, for this purpose, a manufacturer wishes to rely upon the information previously submitted by another

manufacturer (or group of manufacturers) for registration of a similar product (or group of products), then the previous submitter is entitled to reimbursement by the manufacturer for an appropriate portion of the applicable costs incurred to obtain and report such information. Such entitlement shall remain in effect for a period of fifteen years following the date on which the original information was submitted. Pursuant to Sec. 79.59(b)(4)(ii), the manufacturer who relies on previously-submitted registration data shall certify to EPA that the original submitter has been notified and that appropriate reimbursement arrangements have been made.

(1) When private efforts have failed to resolve a dispute about a fair amount or method of cost-sharing or reimbursement for testing costs incurred under this subpart, then any party involved in that dispute may initiate a hearing by filing two signed copies of a request for a hearing with a regional office of the American Arbitration Association and mailing a copy of the request to EPA. A copy must also be sent to each person from whom the filing party seeks reimbursement or who seeks reimbursement from that party. The information and fees to be included in the request for hearing are specified in 40 CFR 791.20(b) and (c).

(2) Additional procedures and requirements governing the hearing process are those specified in 40 CFR 791.22 through 791.50, 791.60, 791.85, and 791.105, excluding 40 CFR 791.39(a)(3) and 791.48(d).

(d) Basis for Classification. (1) Rather than segregating fuels and fuel additives into separate groups, the grouping system applies the same grouping criteria and creates a single set of groups applicable both to fuels and fuel additives.

(2) Fuels shall be classified pursuant to Sec. 79.56(e) into categories and groups of similar fuels and fuel additives according to the components and characteristics of such fuels in their uncombusted state. The classification of a fuel product must take into account the components of all bulk fuel additives which are listed in the registration application or basic registration data submitted for the fuel product.

(3) Fuel additives shall be classified pursuant to Sec. 79.56(e) into categories and groups of similar fuels and fuel additives according to the components and characteristics of the respective uncombusted additive/base fuel mixture pursuant to Sec. 79.51(h)(1).

(4) In determining the category and group to which a fuel or fuel additive belongs, impurities present in trace amounts shall be ignored unless otherwise noted. Impurities are those substances which are present through contamination or which remain in the fuel or additive naturally after processing is completed.

(5) Reference Standards. (i) American Society for Testing and Materials (ASTM) standard D 4814-93a, ``Standard Specification for

Automotive Spark-Ignition Engine Fuel", used to define the general characteristics of gasoline fuels (paragraph (e)(3)(i)(A)(3) of this section) and ASTM standard D 975-93, ``Standard Specification for Diesel Fuel Oils", used to define the general characteristics of diesel fuels (paragraph (e)(3)(ii)(A)(3) of this section) have been incorporated by reference.

(ii) This incorporation by reference was approved by the Director of the Federal Register in accordance with 5 U.S.C. 552(a) and 1 CFR part 51. Copies may be obtained from the American Society for Testing and Materials (ASTM), 1916 Race Street, Philadelphia, PA 19103. Copies may be inspected at U.S. EPA, OAR, 401 M Street SW., Washington, DC, 20460 or at the Office of the Federal Register, 800 North Capitol Street NW., suite 700, Washington, DC.

(e) Grouping Criteria. The grouping system is represented by a matrix of three fuel/additive categories within six specified fuel families (see Table F94-7, Grouping System for Fuels and Fuel Additives). Each category may include one or more groups. Within each group, a representative may be designated based on the criteria in this section and joint registration information may be developed and submitted for member fuels and fuel additives.

Table F94-7.--Grouping System for Fuels and Fuel Additives

	Conventional Fuel Families				Alternative Fuel Families
Category					Methane (CNG, LNG)
	Gasoline (A)	Diesel (B)	Methanol(C)	Ethanol(D) (E)	Propane (LPG) (F)
Baseline.....	One group represented by gasoline base fuel.	One group represented by diesel base fuel.	Two groups: (1) M100 methanol-gasoline formulations with at least 96% ethanol)	One group (includes both CNG and LNG), represented by CNG base fuel.	One group (includes ethanol-gasoline formulations with at least 50% base fuel.	One group represented by LPG base fuel.

			methanol) represented by M100 base fuel.	represented by E85 base fuel (2) M85 (includes methanol-gasoline formulations with 50-95% methanol) represented by M85 base fuel.		
Non-baseline.....	One group for each gasoline-oxygenate blend or each gasoline-methanol/ co-solvent blend; one group for each synthetic crude-derived fuel.	One group for each oxygen-contributing compound or class of compounds; one group for each synthetic crude-derived fuel.	One group for each individual non-methane gasoline component exceeding the specified limit for one group for each non-methane synthetic crude-derived fuel.	One group for each individual non-methane gasoline component exceeding the specified limit for each unique combination of such components.	One group to include propane formulations and one group for each unique combination of such components.	One group to include butane and higher hydrocarbons.
Atypical.....	One group for each atypical element/ characteristic, or unique combination of atypical	One group for each atypical element/ characteristic, or unique combination of atypical	One group for each atypical element/ characteristic, or unique combination of atypical	One group for each atypical element/ characteristic, or unique combination of atypical	One group for each atypical element/ characteristic, or unique combination of atypical	One group for each atypical element/ characteristic, or unique combination of atypical

	elements/characteri stics.	elements/characteri stics.	elements/characteri stics.	elements/characteri stics.	elements/characteri stics.	elements/characteri stics.

(1) Fuel Families. Each of the following six fuel families (Table F94-7, columns A-F) includes fuels of the type referenced in the name of the family as well as bulk and aftermarket additives which are intended for use in those fuels. When applied to fuel additives, the criteria in these descriptions refer to the associated additive/base fuel mixture, pursuant to Sec. 79.51(h)(1). One or more base fuel formulations are specified for each fuel family pursuant to Sec. 79.55.

(i) The Gasoline Family includes fuels composed of more than 50 percent gasoline by volume and their associated fuel additives. The base fuel for this family is specified in Sec. 79.55(b).

(ii) The Diesel Family includes fuels composed of more than 50 percent diesel fuel by volume and their associated fuel additives. The Diesel fuel family includes both Diesel #1 and Diesel #2 formulations. The base fuel for this family is specified in Sec. 79.55(c).

(iii) The Methanol Family includes fuels composed of at least 50 percent methanol by volume and their associated fuel additives. The M100 and M85 base fuels are specified in Sec. 79.55(d).

(iv) The Ethanol Family includes fuels composed of at least 50 percent ethanol by volume and their associated fuel additives. The base fuel for this family is E85 as specified in Sec. 79.55(e).

(v) The Methane Family includes compressed natural gas (CNG) and liquefied natural gas (LNG) fuels containing at least 50 mole percent methane and their associated fuel additives. The base fuel for the family is a CNG formulation specified in Sec. 79.55(f).

(vi) The Propane Family includes propane fuels containing at least 50 percent propane by volume and their associated fuel additives. The base fuel for this family is a liquefied petroleum gas (LPG) as specified in Sec. 79.55(g).

(vii) A manufacturer seeking registration for formulation(s) which do not fit the criteria for inclusion in any of the fuel families described in this section shall contact EPA at the address in Sec. 79.59(a)(1) for further guidance in classifying and testing such formulation(s).

(2) Fuel/Additive Categories. Fuel/additive categories (Table F94-7, rows 1-3) are subdivisions of fuel families which represent the degree to which fuels and fuel additives in the family resemble the base fuel(s) designated for the family. Three general category types are defined in this section. When applied to fuel additives, the

criteria in these descriptions refer to the associated additive/base fuel mixture, pursuant to Sec. 79.51(h)(1).

(i) Baseline categories consist of fuels and fuel additives which contain no elements other than those permitted in the base fuel for the respective fuel family and conform to specified limitations on the amounts of certain components or characteristics applicable to that fuel family.

(ii) Non-Baseline Categories consist of fuels and fuel additives which contain no elements other than those permitted in the base fuel for the respective fuel family, but which exceed one or more of the limitations for certain specified components or characteristics applicable to baseline formulations in that fuel family.

(iii) Atypical Categories consist of fuels and fuel additives which contain elements or classes of compounds other than those permitted in the base fuel for the respective fuel family or which otherwise do not meet the criteria for either baseline or non-baseline formulations in that fuel family. A fuel or fuel additive product having both non-baseline and atypical characteristics pursuant to Sec. 79.56(e)(3), shall be considered to be an atypical product.

(3) This section defines the specific categories applicable to each fuel family. When applied to fuel additives, the criteria in these descriptions refer to the associated additive/base fuel mixture, pursuant to Sec. 79.51(h)(1).

(i) Gasoline Categories. (A) The Baseline Gasoline category contains gasoline fuels and associated additives which satisfy all of the following criteria:

(1) Contain no elements other than carbon, hydrogen, oxygen, nitrogen, and/or sulfur.

(2) Contain less than 1.5 percent oxygen by weight.

(3) Sulfur concentration is limited to 1000 ppm per the specifications cited in the following paragraph.

(4) Possess the physical and chemical characteristics of unleaded gasoline as specified by ASTM standard D 4814-93a (incorporated by reference, pursuant to paragraph (d)(5) of this section), in at least one Seasonal and Geographical Volatility Class.

(5) Derived from conventional petroleum sources only.

(B) The Non-Baseline Gasoline category is comprised of gasoline fuels and associated additives which conform to the specifications in paragraph (e)(3)(i)(A) of this section for the Baseline Gasoline category except that they contain 1.5 percent or more oxygen by weight and/or may be derived from synthetic crudes, such as those prepared from coal, shale and tar sands, heavy oil deposits, and other non-conventional petroleum sources.

(C) The Atypical Gasoline category is comprised of gasoline fuels and associated additives which contain one or more elements other than

carbon, hydrogen, oxygen, nitrogen, and sulfur.

(ii) Diesel Categories. (A) The Baseline Diesel category is comprised of diesel fuels and associated additives which satisfy all of the following criteria:

(1) Contain no elements other than carbon, hydrogen, oxygen, nitrogen, and/or sulfur. Pursuant to 40 CFR 80.29, highway diesel sold after October 1, 1993 shall contain 0.05 percent or less sulfur by weight;

(2) Contain less than 1.0 percent oxygen by weight;

(3) Diesel formulations containing more than 0.05 percent sulfur by weight are precluded by 40 CFR 80.29;

(4) Possess the characteristics of diesel fuel as specified by ASTM standard D 975-93 (incorporated by reference, pursuant to paragraph (d)(5) of this section); and

(5) Derived from conventional petroleum sources only.

(B) The Non-Baseline Diesel category is comprised of diesel fuels and associated additives which conform to the specifications in paragraph (e)(3)(ii)(A) of this section for the Baseline Diesel category except that they contain 1.0 percent or more oxygen by weight and/or may be derived from synthetic crudes, such as those prepared from coal, shale and tar sands, heavy oil deposits, and other non-conventional petroleum sources.

(C) The Atypical Diesel category is comprised of diesel fuels and associated additives which contain one or more elements other than carbon, hydrogen, oxygen, nitrogen, and sulfur.

(iii) Methanol Categories. (A) The Baseline Methanol category is comprised of methanol fuels and associated additives which contain at least 50 percent methanol by volume, no more than 4.0 percent by volume of substances other than methanol and gasoline, and no elements other than carbon, hydrogen, oxygen, nitrogen, sulfur, and/or chlorine. Baseline methanol shall contain no more than 0.004 percent by weight of sulfur or 0.0001 percent by weight of chlorine.

(B) The Non-Baseline Methanol category is comprised of fuel blends which contain at least 50 percent methanol by volume, more than 4.0 percent by volume of a substance(s) other than methanol and gasoline, and meet the baseline limitations on elemental composition in paragraph (e)(3)(iii)(A) of this section.

(C) The Atypical Methanol category consists of methanol fuels and associated additives which do not meet the criteria for either the Baseline or the Non-Baseline Methanol category.

(iv) Ethanol Categories. (A) The Baseline Ethanol category is comprised of ethanol fuels and associated additives which contain at least 50 percent ethanol by volume, no more than five (5) percent by volume of substances other than ethanol and gasoline, and no elements other than carbon, hydrogen, oxygen, nitrogen, sulfur, chlorine, and

copper. Baseline ethanol formulations shall contain no more than 0.004 percent by weight of sulfur, 0.0004 percent by weight of chlorine, and/or 0.07 mg/L of copper.

(B) The Non-Baseline Ethanol category is comprised of fuel blends which contain at least 50 percent ethanol by volume, more than five (5) percent by volume of a substance(s) other than ethanol and gasoline, and meet the baseline limitations on elemental composition in paragraph (e)(3)(iv)(A) of this section.

(C) The Atypical Ethanol category consists of ethanol fuels and associated additives which do not meet the criteria for either the Baseline or the Non-Baseline Ethanol categories.

(v) Methane Categories. (A) The Baseline Methane category is comprised of methane fuels and associated additives (including at least an odorant additive) which contain no elements other than carbon, hydrogen, oxygen, nitrogen, and/or sulfur, and contain no more than 20 mole percent non-methane hydrocarbons. Baseline methane formulations shall not contain more than 16 ppm by volume of sulfur, including any sulfur which may be contributed by the odorant additive.

(B) The Non-Baseline Methane category consists of methane fuels and associated additives which conform to the specifications in paragraph (e)(3)(v)(A) of this section for the Baseline Methane category except that they exceed 20 mole percent non-methane hydrocarbons.

(C) The Atypical Methane category consists of methane fuels and associated additives which contain one or more elements other than carbon, hydrogen, oxygen, nitrogen, and/or sulfur, or exceed 16 ppm by volume of sulfur.

(vi) Propane Categories. (A) The Baseline Propane category is comprised of propane fuels and associated additives (including at least an odorant additive) which contain no elements other than carbon, hydrogen, oxygen, nitrogen, and/or sulfur, and contain no more than 20 percent by volume non-propane hydrocarbons. Baseline Propane formulations shall not contain more than 123 ppm by weight of sulfur, including any sulfur which may be contributed by the odorant additive.

(B) The Non-Baseline Propane category consists of propane fuels and associated additives which conform to the specifications in paragraph (e)(3)(vi)(A) of this section for the Baseline Propane category, except that they exceed the 20 percent by volume limit for butane and higher hydrocarbons.

(C) The Atypical Propane category consists of propane fuels and associated additives which contain elements other than carbon, hydrogen, oxygen, nitrogen, and/or sulfur, or exceed 123 ppm by weight of sulfur.

(4) Fuel/Additive Groups. Fuel/additive groups are subdivisions of the fuel/additive categories. One or more group(s) are defined within each category in each fuel family according to the presence of

differing characteristics in the fuel or additive/base fuel mixture. For each group, one formulation (either a base fuel or a member fuel or additive product) is chosen to represent all the member products in the group in any tests required under this subpart. The section which follows describes the fuel/additive groups.

(i) Baseline Groups. (A) The Baseline Gasoline category comprises a single group. The gasoline base fuel specified in Sec. 79.55(b) shall serve as the representative of this group.

(B) The Baseline Diesel category comprises a single group. The diesel base fuel specified in Sec. 79.55(c) shall serve as the representative of this group.

(C) The Baseline Methanol category includes two groups: M100 and M85. The M100 group consists of methanol-gasoline formulations containing at least 96 percent methanol by volume. These formulations must contain odorants and bitterants (limited in elemental composition to carbon, hydrogen, oxygen, nitrogen, sulfur, and chlorine) for prevention of purposeful or inadvertent consumption. The M100 base fuel specified in Sec. 79.55(d) shall serve as the representative for this group. The M85 group consists of methanol-gasoline formulations containing at least 50 percent by volume but less than 96 percent by volume methanol. The M85 base fuel specified in Sec. 79.55(d) shall serve as the representative of this group.

(D) The Baseline Ethanol category comprises a single group. The E85 base fuel specified in Sec. 79.55(e) shall serve as the representative of this group.

(E) The Baseline Methane category comprises a single group. The CNG base fuel specified in Sec. 79.55(f) shall serve as the representative of this group.

(F) The Baseline Propane category comprises a single group. The LPG base fuel specified in Sec. 79.55(g) shall serve as the representative of this group.

(ii) Non-Baseline Groups-- (A) Non-Baseline Gasoline. The Non-Baseline gasoline fuels and associated additives shall sort into groups according to the following criteria:

(1) For gasoline fuel and additive products which contain 1.5 percent oxygen by weight or more, a separate non-baseline gasoline group shall be defined by each oxygenate compound or methanol/co-solvent blend listed as a component in the registration application or basic registration data of any such fuel or additive.

(i) Examples of oxygenates occurring in non-baseline gasoline formulations include ethanol, methyl tertiary butyl ether (MTBE), ethyl tertiary butyl ether (ETBE), tertiary amyl methyl ether (TAME), diisopropyl ether (DIPE), dimethyl ether (DME), tertiary amyl ethyl ether (TAEE), and any other compound(s) which increase the oxygen content of the gasoline formulation. A separate non-baseline gasoline

group is defined for each such oxygenating compound.

(ii) Each unique methanol and co-solvent combination (whether one, two, or more additional oxygenate compounds) used in a non-baseline fuel shall also define a separate group. An oxygenate compound used as a co-solvent for methanol in a non-baseline gasoline formulation must be identified as such in its registration. If the oxygenate is not identified as a methanol co-solvent, then the compound shall be regarded by EPA as defining a separate non-baseline gasoline group. Examples of methanol/co-solvent combinations occurring in non-baseline gasoline formulations include methanol/isopropyl alcohol, methanol/ butanol, and methanol with alcohols up to C8/octanol (Octamix).

(iii) For each such group, the representative to be used in testing shall be a formulation consisting of the gasoline base fuel blended with the relevant oxygenate compound (or methanol/co-solvent combination) in an amount equivalent to the highest actual or recommended concentration-in-use of the oxygenate (or methanol/co-solvent combination) recorded in the basic registration data of any member fuel or additive product. In the event that two or more products in the same group contain the same and highest amount of the oxygenate or methanol/co-solvent blend, then the representative shall be chosen at random for such candidate products.

(2) An oxygenate compound or methanol/co-solvent combination to be blended with the gasoline base fuel for testing purposes shall be chemical-grade quality, at a minimum, and shall not contain a significant amount of other contaminating oxygenate compounds.

(3) Separate non-baseline gasoline groups shall also be defined for gasoline formulations derived from each particular non-conventional petroleum source or process.

(i) Such groups may include, but are not limited to, the following: coal-derived gasoline formulations; chemically-synthesized gasoline formulations (including those using recycled chemical/petrochemical products); tar sand-derived gasoline formulations; shale-derived gasoline formulations; and other types of soil-recovered products used in formulating gasolines.

(ii) In any such group, the first product to be registered or to apply for EPA registration shall be the representative of that group. If two or more such products are registered or apply for first registration simultaneously, then the representative shall be chosen by a random method from among such candidate products.

(4) Pursuant to Sec. 79.51(i), non-baseline gasoline products may belong to more than one fuel/additive group.

(B) Non-Baseline Diesel. The Non-Baseline diesel fuels and associated additives shall sort into groups according to the following criteria:

(1) For diesel fuel and additive products which contain 1.0 percent

oxygen by weight or more, a separate non-baseline diesel group shall be defined by each individual alcohol or ether listed as a component in the registration application or basic registration data of any such fuel or additive. For each such group, the representative to be used in testing shall be a formulation consisting of the diesel base fuel blended with the relevant alcohol or ether in an amount equivalent to the highest actual or recommended concentration-in-use of the alcohol or ether recorded in the basic registration data of any member fuel or additive product.

(2) A separate non-baseline diesel group is also defined for each of the following classes of oxygenating compounds: mixed nitroso-compounds; mixed nitro- compounds; mixed alkyl nitrates; mixed alkyl nitrites; peroxides; furans; mixed alkyl esters of plant origin; and mixed alkyl esters of animal origin. For each such group, the representative to be used in testing shall be formulated as follows:

(i) From the class of compounds which defines the group, a particular oxygenate compound shall be chosen from among all such compounds recorded in the registration application or basic registration data of any fuel or additive in the group.

(ii) The selected compound shall be the one recorded in any member product's registration application with the highest actual or recommended maximum concentration-in-use. This compound, when mixed into the diesel base fuel at the indicated maximum concentration, shall serve as the group representative.

(iii) In the event that two or more oxygenate compounds in the relevant class have the highest recorded concentration-in-use, then the oxygenate compound to be used in the group representative shall be chosen at random from the qualifying candidate compounds.

(3) A separate non-baseline diesel group shall also be defined for each diesel fuel derived from a particular synthetic petroleum source or process.

(i) Such groups include, but shall not be limited to, the following: coal-derived diesel formulations; chemically-synthesized diesel formulations (including those using recycled chemical/ petrochemical products); tar sand-derived diesel formulations; shale-derived diesel formulations; and other types of soil-recovered products used in formulating diesel fuel(s).

(ii) In any such group, the first product to be registered or to apply for EPA registration shall be the representative of that group. If two or more products are registered or apply for first registration simultaneously, then the representative shall be chosen by a random method from among such candidate products.

(4) Pursuant to Sec. 79.51(i), non-baseline diesel products may belong to more than one fuel/additive group.

(C) Non-Baseline Methanol. The Non-Baseline methanol formulations

are sorted into groups based on the non-methanol, non-gasoline component(s) of the blended fuel. Each such component occurring separately and each unique combination of such components shall define a separate group.

(1) The representative of each such non-baseline methanol group shall be the group member with the highest percent by volume of non-methanol, non-gasoline component(s).

(2) In case two or more such members have the same and highest concentration of non-methanol, non-gasoline component(s), the representative of the group shall be chosen at random from among such equivalent member products.

(D) Non-Baseline Ethanol. The Non-Baseline ethanol formulations are sorted into groups based on the non-ethanol, non-gasoline component(s) of the blended fuel. Each such component occurring separately and each unique combination of such components shall define a separate group.

(1) The representative of each such non-baseline ethanol group shall be the group member with the highest percent by volume of non-ethanol, non-gasoline component(s).

(2) In case two or more such members have the same and highest concentration of non-ethanol, non-gasoline component(s), the representative of the group shall be chosen at random from among such equivalent member products.

(E) Non-Baseline Methane. The Non-Baseline methane category consists of one group. The group representative shall be the member fuel or fuel/additive formulation containing the highest concentration-in-use of non-methane hydrocarbons. If two or more member products have the same and the highest concentration-in-use, then the representative shall be chosen at random from such products.

(F) Non-Baseline Propane. The Non-Baseline propane category consists of one group. The group representative shall be the member fuel or fuel/additive formulation containing the highest concentration-in-use of butane and higher hydrocarbons. If two or more products have the same and the highest concentration-in-use, then the representative shall be chosen at random from such products.

(iii) Atypical groups.

(A) As defined for each individual fuel family in Sec. 79.56(e)(3), fuels and additives meeting any one of the following criteria are considered atypical.

(1) Gasoline Atypical fuels and additives contain one or more elements in addition to carbon, hydrogen, oxygen, nitrogen, and sulfur.

(2) Diesel Atypical fuels and additives contain one or more element in addition to carbon, hydrogen, oxygen, nitrogen, and sulfur.

(3) Methanol Atypical fuels and additives contain:

(i) one or more element in addition to carbon, hydrogen, oxygen, nitrogen, sulfur, and chlorine, and/or

(ii) sulfur in excess of 0.004 percent by weight, and/or

(iii) chlorine in excess of 0.0001 percent by weight.

(4) Ethanol Atypical fuels and additives contain:

(i) one or more element in addition to carbon, hydrogen, oxygen, nitrogen, sulfur, chlorine, and copper, and/or

(ii) sulfur in excess of 0.004 percent by weight, and/or

(iii) contain chlorine (as chloride) in excess of 0.0004 percent by weight, and/or

(iv) contain copper in excess of 0.07 mg/L.

(5) Methane Atypical fuels and additives contain:

(i) one or more element in addition to carbon, hydrogen, oxygen, nitrogen, and sulfur, and/or

(ii) sulfur in excess of 16 ppm by volume.

(6) Propane Atypical fuels and additives contain:

(i) one or more element in addition to carbon, hydrogen, oxygen, nitrogen, and sulfur, and/or

(ii) sulfur in excess of 123 ppm by weight.

(B) General rules for sorting these atypical fuels and additives into separate groups are as follows:

(1) Pursuant to Sec. 79.51(j), a given atypical product may belong to more than one atypical group.

(2) Fuels and additives in different fuel families may not be grouped together, even if they contain the same atypical element(s) or other atypical characteristic(s).

(3) A fuel or additive containing one or more atypical elements attached to a polymer compound must be sorted into a separate group from atypical fuels or fuel additives containing the same atypical element(s) in non-polymer form. However, the occurrence of a polymer compound which does not contain an atypical element does not affect the grouping of a fuel or additive.

(C) Specific rules for sorting each family's atypical fuels and additives into separate groups, and for choosing each such group's representative for testing, are as follows:

(1) A separate group is created for each atypical element (or other atypical characteristic) occurring separately, i.e., in the absence of any other atypical element or characteristic, in one or more fuels and/or additives within a given fuel family.

(i) Consistent with the basic grouping guidelines provided in Sec. 79.56(d), a fuel product which is classified as atypical because its basic registration data or application lists a bulk additive containing an atypical characteristic, may be grouped with that additive and/or with other fuels and additives containing the same atypical characteristic.

(ii) Within a group of products containing only one atypical element or characteristic, the fuel or additive/base fuel mixture with

the highest concentration-in-use or recommended concentration-in-use of the atypical element or characteristic shall be the designated representative of that group. In the event that two or more fuels or additive/base fuel mixtures within the group contain the same and highest concentration of the single atypical element or characteristic, then the group representative shall be selected by a random method from among such candidate products.

(2) A separate group is also created for each unique combination of atypical elements (and/or other specified atypical characteristics) occurring together in one or more fuels and/or additives within a given fuel family.

(i) Consistent with the basic grouping guidelines provided in Sec. 79.56(d), a fuel which is classified as atypical because its basic registration data lists one bulk additive containing two or more atypical characteristics, may be grouped with that additive and/or with other fuels and/or additives containing the same combination of atypical characteristics. Grouping of fuels containing more than one atypical additive shall be guided by provisions of Sec. 79.51(j).

(ii) Within a group of such products containing a unique combination of two or more atypical elements or characteristics, the designated representative shall be the product within the group which contains the highest total concentration of the atypical elements or characteristics.

(iii) In the event that two or more products within a given atypical group contain the same and highest concentration of the same atypical elements or characteristics then, among such candidate products, the designated representative shall be the product which, first, has the highest total concentration of metals, followed in order by highest total concentration of halogens, highest total concentration of other atypical elements (including sulfur concentration, as applicable), highest total concentration of polymers containing atypical elements, and, lastly, highest total concentration of oxygen.

(iv) If two or more products have the same and highest concentration of the variable identified in the preceding paragraph, then, among such products, the one with the greatest concentration of the next highest variable on the list shall be the group representative.

(v) This decision-making process shall continue until a single product is determined to be the representative. If two or more products remain tied at the end of this process, then the representative shall be chosen by a random method from among such remaining products.

Sec. 79.57. Emission generation.

This section specifies the equipment and procedures that must be used in generating the emissions which are to be subjected to the characterization procedures and/or the biological tests specified in Secs. 79.52(b) and 79.53 of these regulations. When applicable, they may also be required in conjunction with testing under Secs. 79.54 and 79.58(c). Additional requirements concerning emission generation, delivery, dilution, quality control, and safety practices are outlined in Sec. 79.61.

(a) Vehicle and engine selection criteria. (1) All vehicles and engines used to generate emissions for testing a fuel or additive/fuel mixture must be new (i.e., never before titled) and placed into the program with less than 500 miles on the odometer or 12 hours on the engine chronometer. The vehicles and engines shall be unaltered from the specifications of the original equipment manufacturer.

(2) The vehicle/engine type, vehicle/engine class, and vehicle/engine subclass designated to generate emissions for a given fuel or additive shall be the same type, class, and subclass which, over the previous three years, has consumed the most gallons of fuel in the fuel family applicable to the given fuel or additive. No distinction shall be made between light-duty vehicles and light-duty trucks for purposes of this classification.

(3) Within this vehicle/engine type, class, and subclass, the specific vehicles and engines acceptable for emission generation are those that represent the most common fuel metering system and the most common of the most important emission control system devices or characteristics with respect to emission reduction performance for the model year in which testing begins. These vehicles will be determined through a survey of the previous model year's vehicle/engine sales within the given subclass. These characteristics shall include, but need not be limited to, aftertreatment device(s), fuel aspiration, air injection, exhaust gas recirculation, and feedback type.

(4) Within the applicable subclass, the five highest selling vehicle/engine models that contain the most common such equipment and characteristics shall be determined. Any of these five models of the current model year (at the time testing begins) may be selected for emission generation.

(i) If one or more of the five models is not available for the current model year, the choice of model for emission generation shall be limited to those remaining among the five.

(ii) If fewer than five models of the given vehicle/engine type are available for the current model year, all such models shall be eligible.

(5) When the fuel or fuel additive undergoing testing is not commonly used or intended to be used in the vehicle/engine types prescribed by this selection procedure, or when rebuilding or

alteration is required to obtain a suitable vehicle/engine for emission generation, the manufacturer may submit a request to EPA for a modification in test procedure requirements. Any such request must include objective test results which support the claim that a more appropriate vehicle/engine type is needed as well as a suggested substitute vehicle/engine type. The vehicle/engine selection in this case shall be approved by EPA prior to the start of testing.

(6) Once a particular model has been chosen on which to test a fuel or additive product, all mileage accumulation and generation of emissions for characterization and biological testing of such product shall be conducted on that same model.

(i) If the initial test vehicle/engine fails or must be replaced for any reason, emission generation shall continue with a second vehicle/engine which is identical to, or resembles to the greatest extent possible, the initial test vehicle/engine. If more than one replacement vehicle/engine is necessary, all such vehicles/engines shall be identical, or resemble to the greatest extent possible, the initial test vehicle/engine.

(ii) Manufacturers are encouraged to obtain, at the start of a test program, more than one emission generation vehicle/engine of the identical model, to ensure the availability of back-up emission generator(s). All backup vehicles/engines must be conditioned and must have their emissions fully characterized, as done for the initial test vehicle/engine, prior to their use as emission generators for biological testing. Alternating between such vehicles/engines regularly during the course of testing is permissible and advisable, particularly to allow regular maintenance on such vehicles/engines during prolonged health effects testing.

(b) Vehicle/engine operation and maintenance. (1) For the purpose of generating combustion emissions from a fuel or additive/base fuel mixture for which the relevant class is light duty, either a light-duty vehicle shall be operated on a chassis dynamometer or a light-duty engine shall be operated on an engine dynamometer. When the relevant class is heavy duty, the emissions shall be generated on a heavy-duty engine operated on an engine dynamometer. In both cases, the vehicle or engine model shall be selected as described in paragraph (a) of this section and shall have all applicable fuel and emission control systems intact.

(2) Except as provided in Sec. 79.51(h)(2)(iii), the fuel or additive/base fuel mixture being tested shall be used at all times during operation of the test vehicle or engine. No other fuels or additives shall be used in the test vehicle or engine once mileage accumulation has begun until emission generation for emission characterization and biological testing purposes is completed.

(3) Scheduled and unscheduled vehicle/engine maintenance.

(i) During emission generation, vehicles and engines must be maintained in good condition by following the recommendations of the original equipment manufacturer (OEM) for scheduled service and parts replacement, with repairs performed only as necessary. Modifications, adjustments, and maintenance procedures contrary to procedures found in 40 CFR part 86 for the maintenance of test vehicles/engines or performed solely for the purpose of emissions improvement are not allowed.

(ii) If unscheduled maintenance becomes necessary, the vehicle or engine must be repaired to OEM specifications, using OEM or OEM-approved parts. In addition, the tester is required to measure the basic emissions pursuant to Sec. 79.52(b)(2)(i) after the unscheduled maintenance and before resuming testing to ensure that the post-maintenance emissions shall be within 20 percent of pre-maintenance emissions levels. If the basic emissions cannot be brought within 20 percent of their previous levels, then the manufacturer shall restart the emissions characterization and health testing of its products combustion emissions using a new vehicle/engine.

(c) Mileage accumulation. (1) A vehicle/engine break-in period is required prior to generating emissions for characterization and/or biological testing under this subpart. The required mileage accumulation may be accomplished on a test track, on the street, on a dynamometer, or using any other conventionally accepted method.

(2) Vehicles to be used in the evaluation of baseline and non-baseline fuels and fuel additives shall accumulate 4,000 miles prior to emission testing. Engines to be used in the evaluation of baseline and non-baseline fuels and fuel additives shall accumulate 125 hours of operation on an engine dynamometer prior to emission testing.

(3) When the test formulation is classified as an atypical fuel or fuel additive formulation (pursuant to definitions in Sec. 79.56(e)(4)(iii)), the following additional mileage accumulation requirements apply:

(i) The test vehicle/engine must be operated for a minimum of 4,000 vehicle miles or 125 hours of engine operation.

(ii) Thereafter, at intervals determined by the tester, all emission fractions (i.e., vapor, semi-volatile, and particulate) shall be sampled and analyzed for the presence and amount of the atypical element(s) and/or other atypical constituents. Pursuant to paragraph (d) of this section, the sampled emissions must be generated in the absence of an intact aftertreatment device. Immediately before the samples are taken, a brief warmup period (at least ten miles or the engine equivalent) is required.

(iii) Mileage accumulation shall continue until either 50 percent or more of the mass of each atypical element (or other atypical constituent) entering the engine can be measured in the exhaust

emissions (all fractions combined), or the vehicle/engine has accumulated mileage (or hours) equivalent to 40 percent of the average useful life of the applicable vehicle/engine class (pursuant to regulations in 40 CFR part 86). For example, the maximum mileage required for light-duty vehicles is 40 percent of 100,000 miles (i.e., 40,000 miles), while the maximum time of operation for heavy-duty engines is the equivalent of 40 percent of 290,000 miles (i.e., the equivalent in engine hours of 116,000 miles).

(iv) When either condition in paragraph (c)(3)(iii) of this section has been reached, additional emission characterization and biological testing of the emissions may begin.

(d) Use of exhaust aftertreatment devices. (1) If the selected test vehicle/engine, as certified by EPA, does not come equipped with an emissions aftertreatment device (such as a catalyst or particulate trap), such device shall not be used in the context of this program.

(2) Except as provided in paragraph (d)(3) of this section for certain specialized additives, the following provisions apply when the test vehicle/engine, as certified by EPA, comes equipped with an emissions aftertreatment device.

(i) For mileage accumulation:

(A) When the test formulation does not contain any atypical elements (pursuant to definitions in Sec. 79.56(e)(4)(iii)), an intact aftertreatment device must be used during mileage accumulation.

(B) When the test formulation does contain atypical elements, then the manufacturer may choose to accumulate the required mileage using a vehicle/engine equipped with either an intact aftertreatment device or with a non-functional aftertreatment device (e.g., a blank catalyst without its catalytic wash coat). In either case, sampling and analysis of emissions for measurement of the mass of the atypical element(s) (as described in Sec. 79.57(c)(3)) must be done on emissions generated with a non-functional (blank) aftertreatment device.

(1) If the manufacturer chooses to accumulate mileage without a functional aftertreatment device, and if the manufacturer wishes to do this outside of a laboratory/test track setting, then a memorandum of exemption for product testing must be obtained by applying to the Director of the Field Operations and Support Division (see Sec. 79.59(a)(1)).

(2) [reserved]

(ii) For Tier 1 (Sec. 79.52), the total set of requirements for the characterization of combustion emissions (Sec. 79.52(b)) must be completed two times, once using emissions generated with the aftertreatment device intact and a second time with the aftertreatment device rendered nonfunctional or replaced with a non-functional aftertreatment device as described in paragraph (d)(2)(i)(B) of this section.

(iii) For Tier 2 (Sec. 79.53), the standard requirements for biological testing of combustion emissions shall be conducted using emissions generated with a non-functioning aftertreatment device as described in paragraph (d)(2)(i)(B) of this section.

(iv) For alternative Tier 2 requirements (Sec. 79.58(c)) or Tier 3 requirements (Sec. 79.54) which may be prescribed by EPA, the use of functional or nonfunctional aftertreatment devices shall be specified by EPA as part of the test guidelines.

(v) In the case where an intact aftertreatment device is not in place, all other manufacturer-specified combustion characteristics (e.g., back pressure, residence time, and mixing characteristics) of the altered vehicle/engine shall be retained to the greatest extent possible.

(3) Notwithstanding paragraphs (d)(1) and (d)(2) of this section, when the subject of testing is a fuel additive specifically intended to enhance the effectiveness of exhaust aftertreatment devices, the related aftertreatment device may be used on the emission generation vehicle/engine during all mileage accumulation and testing.

(e) Generation of combustion emissions--

(1) Generating combustion emissions for emission characterization. (i) Combustion emissions shall be generated according to the exhaust emission portion of the Federal Test Procedure (FTP) for the certification of new motor vehicles, found in 40 CFR part 86, subpart B for light-duty vehicles/engines, and subparts D, M and N for heavy-duty vehicles/engines. The Urban Dynamometer Driving Schedule (UDDS), pursuant to 40 CFR part 86, appendix I(a), shall apply to light-duty vehicles/engines and the Engine Dynamometer Driving Schedule (EDS), pursuant to 40 CFR part 86, appendix I(f)(2), shall apply to heavy-duty vehicles/engines. The motoring portion of the heavy-duty test cycle may be eliminated, at the manufacturer's option, for the generation of emissions.

(A) For light-duty engines operated on an engine dynamometer, the tester shall determine the speed-torque equivalencies (``trace'') for its test engine from valid FTP testing performed on a chassis dynamometer, using a test vehicle with an engine identical to that being tested. The test engine must then be operated under these speed and torque specifications to simulate the FTP cycle.

(B) Special procedures not included in the FTP may be necessary in order to characterize emissions from fuels and fuel additives containing atypical elements or to collect some types of emissions (e.g., particulate emissions from light-duty vehicles/engines, semi-volatile emissions from both light-duty and heavy-duty vehicles/engines). Such alterations to the FTP are acceptable.

(ii) Pursuant to Sec. 79.52(b)(1)(i) and Sec. 79.57(d)(2)(ii), emission generation and characterization must be repeated three times

when the selected vehicle/engine is normally operated without an emissions aftertreatment device and six times when the selected vehicle/engine is normally operated with an emissions aftertreatment device. In the latter case, the emission generation and characterization process shall be repeated three times with the intact aftertreatment device in place and three times with a non-functioning (blank) aftertreatment device in place.

(iii) From both light-duty and heavy-duty vehicles/engines, samples of vapor phase, semi-volatile phase, and particulate phase emissions shall be collected, except that semi-volatile phase, and particulate emissions need not be sampled for fuels and additives in the methane and propane families (pursuant to Sec. 79.56(e)(1)(v) and (vi)). The number and type of samples to be collected and separately analyzed during one emission generation/characterization process are as follows:

(A) In the case of combustion emissions generated from light-duty vehicles/engines, the samples consist of three bags of vapor emissions (one from each segment of the light-duty exhaust emission cycle) plus one sample of particulate-phase emissions and one sample of semi-volatile-phase emissions (collected over all segments of the exhaust emission cycle). If the mass of particulate emissions or semi-volatile emissions obtained during one driving cycle is not sufficient for characterization, then the driving cycle may be performed again and the extracted fractions combined prior to chemical analysis. Particulate-phase emissions shall not be combined with semi-volatile-phase emissions.

(B) In the case of combustion emissions generated from heavy-duty engines, the samples consist of one sample of each emission phase (vapor, particulate, and semi-volatile) collected over the entire cold-start cycle and a second sample of each such phase collected over the entire hot-start cycle (see 40 CFR 86.334 through 86.342).

(iv) Emission collection and storage. (A) Vapor phase emissions shall be collected and stored in Tedlar bags for subsequent chemical analysis. Storage conditions are specified in Sec. 79.52(b)(2).

(B) Particulate phase emissions shall be collected on a particulate filter (or more than one, if required) using methods described in 40 CFR 86.1301 through 86.1344. These methods, ordinarily applied only to heavy-duty emissions, are to be adapted and used for collection of particulates from light-duty vehicles/engines, as well. The particulate matter may be stored on the filter in a sealed container, or the soluble organic fraction may be extracted and stored in a separate sealed container. Both the particulate and the extract shall be shielded from ultraviolet light and stored at -20 deg.C or less. Particulate emissions shall be tested no later than six months from the date they were generated.

(C) Semi-volatile emissions shall be collected immediately

downstream from the particulate collection filters using porous polymer resin beds, or their equivalent, designed for their capture. The soluble organic fraction of semi-volatile emissions shall be extracted immediately and tested within six months of being generated. The extract shall be stored in a sealed container which is shielded from ultraviolet light and stored at -20 deg.C or less.

(D) Particulate and semi-volatile phase emission collection, handling and extraction methods shall not alter the composition of the collected material, to the extent possible.

(v) Additional requirements for combustion emission sampling, storage, and characterization are specified in Sec. 79.52(b).

(2) Generating whole combustion emissions for biological testing. (i) Biological tests requiring whole combustion emissions shall be conducted using emissions generated from the test vehicle or engine operated in general accordance with the FTP procedures cited in this section. The emissions shall be generated continuously throughout the animal exposure periods, diluted by an amount appropriate for the test being performed as specified in Sec. 79.61(d)(3), passed through a mixing chamber, and routed to the biological test chamber.

(ii) Light-duty test vehicles/engines shall be operated over the Urban Dynamometer Driving Schedule (or equivalent engine dynamometer trace, per paragraph (e)(1)(i)(A) of this section) and heavy-duty test engines shall be operated over the Engine Dynamometer Schedule (see 40 CFR part 86, appendix I).

(A) The tolerances of the driving cycle shall be two times those of the Federal Test Procedure and must be met 95 percent of the time.

(B) The driving cycle shall be repeated as many times as required for the biological test session.

(C) Light-duty dynamometers shall be calibrated prior to the start of a biological test (40 CFR 86.118-78), verified weekly (40 CFR 86.118-78), and recalibrated as required. Heavy-duty dynamometers shall be calibrated and checked prior to the start of a biological test (40 CFR 86.1318-84), recalibrated every two weeks (40 CFR 86.1318-84(a)) and checked as stated in 40 CFR 86.1318-84(b) and (c).

(D) The fuel reservoir for the test vehicle/engine shall be large enough to operate the test vehicle/engine throughout the daily biological exposure period, avoiding the need for refueling during testing.

(iii) An apparatus to integrate the large concentration swings typical of transient-cycle exhaust is to be used between the FTP-Constant Volume Sampler (CVS) source of emissions and the exposure chamber containing the animal test cage(s). The purpose of such apparatus is to decrease the variability of the biological exposure atmosphere.

(A) A large mixing chamber is suggested for this purpose. The

mixing chamber would be charged from the CVS at a constant rate determined by the exposure chamber purge rate. Flow to the exposure chamber would begin at the conclusion of the initial transient cycle with the associated mixing chamber charge.

(B) A potential alternative apparatus is a mini-diluter (see, for example, AIGER/CRADA, February, 1994 in Sec. 79.57(g)).

(C) The mixing chamber (or any alternative emission moderation apparatus) must function such that the average concentration of total hydrocarbons leaving the apparatus shall be within 10 percent of the average concentration of hydrocarbons entering the chamber.

(iv) Emission dilution. (A) Dilution air can be pre-dried to lower the relative humidity, thus permitting a lower dilution rate and a higher concentration of hydrocarbons to be achieved without condensation of water vapor.

(B) With gasoline fuels, a minimum dilution ratio of about 1:5 raw exhaust (dewpoint about 125 deg.F) with dry, clean filtered air is required to reduce the water concentration to a dewpoint of about 68 deg.F. The minimum dilution ratio (maximum exhaust flow rate) occurs at about 200 seconds into the UDDS transient driving cycle. Larger minimum dilution ratios are required if the dilution air includes water vapor. However, the minimum dilution ratio will vary with fuel composition. Fuels which generate greater engine exhaust water concentrations (e.g., alcohol and natural gas fuels) will require greater initial dilutions. Heated transfer ducts or tubing can be used to avoid water condensation in much of the system, but the mixing chamber described in paragraph (e)(2)(iii) of this section will generally be at or near laboratory temperature, and CVS dilution will have to be adequate to assure that the cumulative dew point in the chamber remains below laboratory temperature at all times (further guidance on this topic may be found in Black and Snow, 1994 in Sec. 79.57(g)).

(C) After the initial exhaust dilution to preserve the character of the exhaust, the exhaust stream can be further diluted in the mixing chamber (and/or after leaving the chamber) to achieve the desired biological exposure concentrations.

(v) Verification procedures. (A) The entire system used to dilute and transport whole combustion emissions (i.e., from exhaust pipe to outlet in the biological testing chamber) shall be verified before any animal exposures begin, and verified at least weekly during testing. (See procedures at 40 CFR 86.119-90 for light-duty vehicles and Sec. 86.1319-90 for heavy-duty engines.) Verification testing shall be accomplished by introducing a known sample at the end of the vehicle/ engine exhaust pipe into the dilution system and measuring the amount exiting the system. For example, an injected hydrocarbon sample could be detected with a gas chromatograph (GC) and flame ionization detector (FID) to determine the recovery factor.

(B) Verification of the integrity of the mixing chamber (or alternative apparatus) shall be determined before animal exposures begin and at least weekly thereafter. Composite values for weight percent total hydrocarbons shall be determined for the test vehicle/engine's dilute exhaust stream entering and exiting the mixing chamber apparatus. These values must be within 10 percent of each other.

(vi) Emission exposure quality control. (A) The tester shall incorporate the additional quality assurance and safety procedures outlined in Sec. 79.61(d) to control variability of emissions during the generation of exposure emissions during health effect testing.

(B) These procedures include requirements that the mean exposure concentration in the inhalation test chamber shall be within 10 percent of the target concentration (established in the developmental phase of testing) on 90 percent or more of exposure days and that daily monitoring of CO, CO<INF>2, NO<INF>X, SO<INF>X, and total hydrocarbons in the exposure chamber shall be required. Analysis of the particle size distribution shall also be performed to establish the stability and consistency of particle size distribution in the test exposure.

(C) The testing facility shall allow an audit of its premises, the qualifications, e.g., curriculum vitae, of its staff assigned to testing, and the specimens and records of the testing for registration purposes (as specified in Sec. 79.60).

(vii) In order to allow for unforeseen problems with the emission generation or dilution equipment, emission generation may be interrupted for up to four hours on a maximum of two occasions in any four-week period of testing. The amount of time for which emission generation was interrupted shall subsequently be added after the equipment problem is corrected. If the equipment problem causes more than four consecutive hours of emission generation to be interrupted, or if more than two such occasions occurs in any four-week period during testing, the interrupted tests shall be void. Testers shall be aware of concerns for backup vehicles/engines cited in paragraph (a)(7)(ii) of this section.

(3) Generating particulate and semi-volatile emissions for biological testing. (i) Salmonella mutagenicity testing, pursuant to Sec. 79.68, shall be conducted on extracts of the particulate and semi-volatile emission phases separately. These emissions shall be generated by operating the test vehicle/engine over the appropriate FTP driving cycle (see paragraph (e)(2)(ii) of this section) and collected and analyzed according to methods described in 40 CFR 86.1301 through 1344 (further information on this subject may be found in Perez, et al. CRC Report No. 551, 1987 listed in Sec. 79.57(g)).

(A) Particulate emissions shall be collected on particulate filters and extracted from the collection equipment for use in biological tests. The particulate emissions from all segments of the FTP or from

multiple FTP cycles may be collected on one or more filters, as necessary. The time spent collecting sufficient quantities of the test substances in emissions samples will vary, depending on the emission characteristics of the engine and fuel or additive/base fuel mixture and on the requirements of the biological test protocol.

(B) Semi-volatile emissions shall be collected immediately downstream from the particulate collection filters using porous polymer resin beds, or their equivalent, designed for their capture. Semi-volatile phase emissions shall be collected on one apparatus. The time spent collecting sufficient quantities of the test substances in emissions samples will vary, depending on the emission characteristics of the engine and fuel or additive/base fuel mixture and on the requirements of the biological test protocol.

(ii) The extraction method shall be determined by the specifications of the biological test for which the emissions are used.

(iii) Particulate and semi-volatile emission storage requirements are as specified in Sec. 79.57(e)(1)(iv).

(iv) Particulate and semi-volatile phase emission collection, handling and extraction methods shall not alter the composition of the collected material, to the extent possible.

(v) Particulate emissions shall not be combined with semi-volatile phase emissions.

(f) Generation of evaporative emissions for characterization and biological testing. (1) Except as provided in paragraph (f)(5) of this section, an evaporative emissions generator shall be used to volatilize samples of a fuel or additive/base fuel mixture for evaporative emissions characterization and biological testing. Emissions shall be collected and sampled using equipment and methods appropriate for use with the compounds being characterized and the requirements of the emission characterization analysis. In the case of potentially explosive test substance concentrations, care must be taken to avoid generating explosive atmospheres. The tester is referred to Sec. 79.61(d)(8) for considerations involving explosivity.

(2) Evaporative Emissions Generator (EEG) Description. An EEG is a fuel tank or vessel to which heat is applied causing a portion of the fuel to evaporate at a desired rate. The manufacturer has flexibility in designing an EEG for testing a particular fuel or fuel additive. The sample used to generate emissions in the EEG shall be renewed at least daily.

(i) The evaporation chamber shall be made from materials compatible with the fuels and additives being tested and shall be equipped with a drain.

(ii) The chamber shall be filled to 40 <plus-minus>5 percent of its interior volume with the fuel or additive/base fuel mixture being tested, with the remainder of the volume containing air.

(iii) The concentration of the evaporated fuel or additive/base fuel mixture in the vapor space of the evaporation chamber during the time emissions are being withdrawn for testing shall not vary by more than 10 percent from the equilibrium concentration in the vapor space of emissions generated from the fresh fuel or additive/base fuel mixture in the chamber.

(A) During the course of a day's emission generation period, the level of fuel in the EEG shall be maintained to within 7 percent of its height at the start of the daily exposure period.

(B) The fuel used in the EEG shall be drained at the end of each daily exposure. The EEG shall be refilled with a fresh supply of the test formulation before the start of each daily exposure.

(C) The vapor space of the evaporation chamber shall be well mixed throughout the time emissions are being withdrawn for testing.

(iv) The size of the evaporation chamber shall be determined by the rate at which evaporative emissions shall be needed in the test animal exposure chambers and the rate at which the fuel or the additive/base fuel mixture evaporates. The rate of evaporative emissions may be adjusted by altering the size of the EEG or by using one or more additional EEG(s). Emission rate modifications shall not be adjusted by temperature control or pressure control.

(v) The temperature of the fuel or additive/base fuel mixture in the evaporation chamber shall be 130 deg.F<plus-minus>5 deg.F. The vapors shall maintain this temperature up to the point in the system where the vapors are diluted.

(vi) The pressure in the vapor space of the evaporation chamber and the dilution and sampling apparatus shall stay within 10 percent of ambient atmospheric pressure.

(vii) There shall be no controls or equipment on the evaporation chamber system that change the concentration or composition of the vapors generated for testing.

(viii) Manufacturers shall perform verification testing of evaporative emissions in a manner analogous to the verification testing performed for combustion emissions.

(3) For biological testing, vapor shall be withdrawn from the EEG at a constant rate, diluted with air as required for the particular study, and conducted immediately to the biological testing chamber(s) in a manner similar to the method used in Sec. 79.57(e), excluding the mixing chamber therein. The rate of emission generation shall be high enough to supply the biological exposure chamber with sufficient emissions to allow for a minimum of fifteen air changes per exposure chamber per hour. Interruption of evaporative emissions exposures during biological testing for more than four consecutive hours, or on more than two separate occasions within a four-week period for less than four consecutive hours, shall cause the affected test(s) to be

void.

(4) For characterization of evaporative emissions, samples of equilibrated emissions to the vapor space of the EEG shall be withdrawn into Tedlar bags, then stored and analyzed as specified in Sec. 79.52(b).

(5) A manufacturer (or group of manufacturers) may submit to EPA a request for approval of an alternative method of generating evaporative emissions for use in emission characterization and biological tests required under this subpart.

(i) To be approved by EPA, the request must fully explain the rationale for the proposed method as well as the technical procedures, quality control, and safety precautions to be used, and must demonstrate that the proposed method will meet the following criteria:

(A) The emission mixture generated by the proposed procedures must be reasonably similar to the equilibrium composition of the vapor which occurs in the vehicle fuel tank head space when the subject fuel or additive/base fuel mixture is in use and near-maximum in-use temperatures are encountered.

(B) The emissions mixture generated by the proposed method must be sufficiently concentrated to provide adequate exposure levels in the context of the required toxicologic tests.

(C) The proposed method must include procedures to ensure that the emissions delivered to the biologic exposure chambers will provide a reasonably constant exposure atmosphere over time.

(ii) If EPA approves the request, EPA will place in the public record a copy of the request, together with all supporting procedural descriptions and justifications, and will notify the public of its availability by publishing a notice in the Federal Register.

(g) References. For additional background information on the emission generation procedures outlined in this paragraph (g), the following references may be consulted. Additional references can be found in Sec. 79.61(f).

(1) AIGER/CRADA (American Industry/Government Emissions Research Cooperative Research and Development Agreement, ``Specifications for Advanced Emissions Test Instrumentation" AIGER PD-94-1, Revision 5.0, February, 1994

(2) Black, F. and R. Snow, ``Constant Volume Sampling System Water Condensation" SAE #940970 in ``Testing and Instrumentation" SP-1039, Society of Automotive Engineers, Feb. 28-Mar. 3, 1994.

(3) Perez, J.M., Jass, R.E., Leddy, D.G., eds. ``Chemical Methods for the Measurement of Unregulated Diesel Emissions (CRC-APRAC Project No. CAPI-1-64), Coordinating Research Council, CRC Report No. 551, August, 1987.

(4) Phalen, R.F., ``Inhalation Studies: Foundations and Techniques", CRC Press, Inc., Boca Raton, Florida, 1984.

Sec. 79.58 Special provisions.

(a) Relabeled Additives. Sellers of relabeled additives (pursuant to Sec. 79.50) are not required to comply with the provisions of Secs. 79.52, 79.53 or 79.59, except that such sellers are required to comply with Sec. 79.59(b).

(b) Low Vapor Pressure Fuels and Additives. Fuels which are not designated as ``evaporative fuels'' and fuel additives which are not designated as ``evaporative fuel additives'' pursuant to the definitions in Sec. 79.50 need not undergo the emission characterization or health effects testing specified in Secs. 79.52 and 79.53 for evaporative emissions. At EPA's discretion, the evaporative emissions of such fuels and additives may be required to undergo Tier 3 testing, pursuant to Sec. 79.54.

(c) Alternative Tier 2 Provisions. At EPA's discretion, EPA may modify the standard Tier 2 health effects testing requirements for a fuel or fuel additive (or group). Such modification may encompass substitution, addition, or deletion of Tier 2 studies or study specifications, and/or changes in underlying engine or equipment requirements, except that a Tier 2 endpoint will not be deleted in the absence of existing information deemed adequate by EPA or alternative testing requirements for such endpoint. If warranted by the particular requirements, EPA will allow additional time for completion of the alternative Tier 2 testing program.

(1) When EPA intends to require testing in lieu of or in addition to standard Tier 2 health testing, EPA will notify the responsible manufacturer (or group) by certified letter of the specific tests which EPA is proposing to require in lieu of or in addition to Tier 2, and the proposed schedule for completion and submission of such tests. A copy of the letter will be placed in the public record. EPA intends to send the notification prior to November 27, 1995, or in the case of new fuels and additives (as defined in Sec. 79.51(c)(3)), within 18 months of EPA's receipt of an intent to register such product. However, EPA's notification to the manufacturer (or group) may occur at any time up to EPA's receipt of Tier 2 data for the product(s) in question. EPA will provide the manufacturer with 60 days from the date of receipt of the notice to comment on the tests which EPA is proposing to require and on the proposed schedule. If the manufacturer believes that undue costs or hardships will occur as a result of EPA's delay in providing notification of alternative Tier 2 requirements, then the manufacturer's comments should describe and include evidence of such hardship. In particular, if the standard Tier 2 toxicology testing for the fuel or additive in question has already begun at the time the manufacturer receives EPA's notification of proposed alternative Tier 2

requirements, then EPA shall refrain from requiring alternative Tier 2 tests provided that EPA receives the standard Tier 2 data and report (pursuant to Sec. 79.59(c)) within one year of the date on which the toxicology testing began.

(2) EPA will issue a notice in the Federal Register announcing its intent to require special testing in lieu of or in addition to the standard Tier 2 testing for a particular fuel or additive manufacturer or group, and that a copy of the letter to the manufacturer or group describing the proposed alternative Tier 2 testing for that manufacturer or group is available in the public record for review and comment. The public shall have a minimum of 30 days after the publication of this notice to comment on the proposed alternative Tier 2 testing.

(3) EPA will include in the public record a copy of any timely comments concerning the proposed alternative Tier 2 testing requirements received from the affected manufacturer or group or from the public, and the responses of EPA to such comments. After reviewing all such comments received, EPA may adopt final alternative Tier 2 requirements by sending a certified letter describing such final requirements to the manufacturer or group. In that event, EPA will also issue a notice in the Federal Register announcing that it has adopted final alternative Tier 2 requirements and that a copy of the letter adopting the requirements has been included in the public record.

(4) After EPA's receipt of a manufacturer's (or group's) submittals, EPA will notify the responsible manufacturer (or group) regarding the adequacy of the submittal and potential Tier 3 testing requirements according to the same relative time intervals and by the same procedures as specified in Sec. 79.51 (c) and (d) for routine Tier 1 and Tier 2 submittals.

(d) Small Business Provisions. (1) For purposes of these provisions, when subsidiary, divisional, or other complex business arrangements exist, manufacturer is defined as the business entity with ultimate ownership of all related parents, subsidiaries, divisions, branches, or other operating units. Total annual sales means the average of the manufacturer's total sales revenue in each of the three years prior to such manufacturer's submittal to EPA of the basic registration information pursuant to Sec. 79.59 (b)(2) through (b)(5).

(2) Provisions Applicable to Baseline and Non-baseline Products. A manufacturer with total annual sales less than $50 million is not required to meet the requirements of Tier 1 and Tier 2 (specified in Secs. 79.52 and 79.53) with regard to such manufacturer's fuel and/or additive products which meet the criteria for inclusion in a Baseline or Non-baseline group pursuant to Sec. 79.56. Upon such manufacturer's satisfactory completion and submittal to EPA of basic registration data specified in Sec. 79.59(b), the manufacturer may request and EPA shall

issue a registration for such product, subject to Sec. 79.51(c) and paragraphs (d)(4) and (d)(5) of this section.

(3) Provisions Applicable to Atypical Products. A manufacturer with total annual sales less than $10 million is not required to meet the requirements of Tier 2 (specified in Sec. 79.53) in regard to such manufacturer's fuel and/or additive products which meet the criteria for inclusion in an Atypical group pursuant to Sec. 79.56. Upon such manufacturer's satisfactory completion and submittal to EPA of basic registration data specified in Sec. 79.59(b) and Tier 1 information specified in Sec. 79.52 for an Atypical fuel or additive, the manufacturer may request and EPA shall issue a registration for such product, subject to Sec. 79.51(c) and paragraphs (d)(4) and (d)(5) of this section. Compliance with Tier 1 requirements under this paragraph may be accomplished by the individual manufacturer or as a part of a group pursuant to Sec. 79.56.

(4) Any registration granted by EPA under the provisions of this section are conditional upon satisfactory completion of any Tier 3 requirements which EPA may subsequently impose pursuant to Sec. 79.54. In such circumstances, the Tier 3 requirements might include (but would not necessarily be limited to) information which would otherwise have been required under the provisions of Tier 1 and/or Tier 2.

(5) The provisions in paragraphs (d)(2) and (d)(3) of this section are voluntary on the part of qualifying small manufacturers. Such manufacturers may choose to fulfill the standard requirements for their fuels and additives, individually or as a part of a group, rather than satisfying only the requirements specified in paragraphs (d)(2) and/or (d)(3) of this section. If a qualifying small manufacturer elects these special provisions rather than the standard requirements for a product, then EPA will generally assume that any additional information submitted by other manufacturers, for fuels and additives meeting the same grouping criteria (under Sec. 79.56) as that of the small manufacturer's product, is pertinent to further testing and/or regulatory decisions that may affect the small manufacturer's product.

(e) Aftermarket Aerosol Additives. (1) To obtain registration for an aftermarket aerosol fuel additive, the manufacturer shall provide existing information in the form of a literature search, a discussion of the potential exposure(s) to such product, and the basic registration data specified in Sec. 79.59(b).

(2) The literature search shall include existing data on potential health and welfare effects due to exposure to the aerosol product itself and its raw (uncombusted) components. The analysis for potential exposures shall be based on the actual or anticipated production volume and market distribution of the particular aerosol product, and its estimated frequency of use. Other Tier 1 and Tier 2 requirements are not routinely required for aerosol products. EPA will review the

submitted information and, at EPA's discretion, may require from the manufacturer further information and/or testing under Tier 3 on a case-by-case basis.

Sec. 79.59 Reporting requirements.

(a) Timing. (1) The manufacturer of each designated fuel or fuel additive shall submit to EPA the basic registration data detailed in paragraph (b) of this section. Forms for submitting this data may be obtained from EPA at the following address: Director, Field Operations and Support Division, 6406J--Fuel/Additives Registration, U.S. Environmental Protection Agency, 401 M Street, S.W., Washington, DC 20460.

(i) For existing products (pursuant to Sec. 79.51(c)(1)), manufacturers shall submit the basic registration data as specified in Sec. 79.59(b) to EPA by November 28, 1994.

(ii) For registrable products (pursuant to Sec. 79.51(c)(2)), manufacturers shall submit the basic registration data as specified in Sec. 79.59(b) to apply for registration for such product.

(iii) For new products (pursuant to Sec. 79.51(c)(3)), manufacturers are strongly encouraged to notify EPA of an intent to obtain product registration by submitting the basic registration data as specified in Sec. 79.59(b) prior to starting Tiers 1 and 2.

(2) The information specified in paragraph (c) of this section shall be submitted to the address in paragraph (a)(1) of this section at the conclusion of activities performed in compliance with Tiers 1 and 2 under the provisions of Secs. 79.52 and 79.53, according to the time constraints specified in Sec. 79.51 (c) through (d).

(3) The information specified in paragraph (d) of this section shall be submitted to EPA at the address in paragraph (a)(1) of this section at the conclusion of activities performed in compliance with Tier 3 under the provisions of Sec. 79.54.

(b) Basic Registration Data. Each manufacturer of a designated fuel or fuel additive shall submit the following data in regard to such fuel or fuel additive:

(1) The information specified in Sec. 79.11 or Sec. 79.21. If such information has already been submitted to EPA in compliance with subpart B or C of this part, and if such previous information is accurate and up-to-date, the manufacturer need not resubmit this information.

(2) Annual production volume of the fuel or fuel additive product, in units of gallons per year if most commonly sold in liquid form or kilograms per year if most commonly sold in solid form. For fuels and fuel additives already in production, the most recent annual production

volume and the volume projected to be produced in the third subsequent year shall be provided. For products not yet in production, the best estimate of expected annual volume during the third year of production shall be provided.

(3) Market distribution of the product. For fuels and bulk additives, this information shall be presented as the percent of total annual sales volume marketed in each Petroleum Administration for Defense District (PADD). The states comprising each PADD are listed in the following section. For aftermarket additives, the distribution data shall be presented as the percent of total annual sales volume marketed in each state. For a product not yet in production, the manufacturer shall present the distribution (by PADD or state, as applicable) projected to occur during the third year of production.

(i) The following states and jurisdictions are included in PADD I:

Connecticut
Delaware
District of Columbia
Florida
Georgia
Maine
Maryland
Massachusetts
New Hampshire
New Jersey
New York
North Carolina
Pennsylvania
Rhode Island
South Carolina
Vermont
Virginia
West Virginia

(ii) The following states are included in PADD II:

Illinois
Indiana
Iowa
Kansas
Kentucky
Michigan
Minnesota
Missouri
Nebraska

North Dakota
Ohio
Oklahoma
South Dakota
Tennessee
Wisconsin

(iii) The following states are included in PADD III:

Alabama
Arkansas
Louisiana
Mississippi
New Mexico
Texas

(iv) The following states are included in PADD IV:

Colorado
Idaho
Montana
Utah
Wyoming

(v) The following states are included in PADD V:

Alaska
Arizona
California
Hawaii
Nevada
Oregon
Washington

(4) Any applicable information pursuant to the grouping provisions in Sec. 79.56, as follows:

(i) If the manufacturer has enrolled or intends to enroll the product in a fuel/additive group, the relevant group and the person(s) or entity expected to submit information on behalf of the group must be identified.

(ii) If the manufacturer intends to rely on registration information previously submitted by another manufacturer (or group) for registration of other product(s) in the same fuel/additive group, then the original submitter and its product (or product group) shall be identified. In such cases, the manufacturer shall provide evidence that

the original submitter has been notified of the use of its registration data and that the manufacturer has complied or intends to comply with the proportional reimbursement required under Sec. 79.56(c) of this rule.

(5) Any applicable information pursuant to the special provisions in Sec. 79.58, as follows:

(i) If the manufacturer claims applicability of the special provisions for relabeled additives, pursuant to Sec. 79.58(a), then the manufacturer and brand name of the original product shall be given.

(ii) If the manufacturer claims applicability of any small business provisions pursuant to Sec. 79.58(d), the average of the manufacturer's total annual sales revenue for the previous three years shall be given.

(iii) If the manufacturer claims applicability of the special provisions for aerosol products, pursuant to Sec. 79.58(e), then the purpose and recommended frequency of use shall be given.

(c) Tier 1 and Tier 2 Reports. If the results of Tiers 1 and 2 are reported to EPA at the same time, then the report shall include the following documents in paragraphs (c)(1) through (7) of this section. If Tier 1 and Tier 2 results are submitted to EPA separately, then the separate Tier 1 report shall include only documents in paragraphs (c)(1) through (4), (c)(6), and associated appendices in paragraphs (c)(7) of this section, and the separate Tier 2 report shall include only documents in paragraphs (c)(1) through (3), (c)(5), (c)(6), and associated appendices in paragrpah (c)(7) of this section. In addition, pursuant to the requirements in Sec. 79.51(c)(1)(ii)(B), if the Tier 2 report for registered fuels and fuel additives is not submitted prior to May 27, 1997, then evidence of a suitable arrangement for completion of Tier 2 (e.g., a copy of a signed contract with a qualified laboratory for applicable Tier 2 services) must be submitted to EPA prior to that date.

(1) Cover page. (i) Identification of test substance,

(ii) Name and address of the manufacturer of the test substance,

(iii) Name and phone number of a designated contact person,

(iv) Group information, if applicable, including:

(A) Group name or grouping criteria,

(B) Name and address of responsible organization or entity reporting for the group,

(C) Product trade name and manufacturer of each member fuel and additive to which the report pertains.

(2) Executive Summary. Text overview of the significant results and conclusions obtained as a result of completing the requirements of Tier 1 and/or Tier 2, including references if used to support such results and conclusions.

(3) Test Substance Information. Test substance description, including, as applicable,

(i) Base fuel parameter values (including types and concentrations of base fuel additives) or test fuel composition (if a fuel other than the base fuel is used in testing). These values must be provided for each of the fuel parameters specified in Sec. 79.55 for the applicable fuel family.

(ii) Test additive composition and concentration

(4) Summary of Tier 1 (i) Literature Search. Pursuant to Sec. 79.52(d), the literature search shall include a text summary of the methods and results of the literature search, including the following:

(A) Identification of person(s) performing the literature search,

(B) Description of data sources accessed, search strategy used, search period, and terms included in literature search,

(C) Documentation of all unpublished in-house and other privately-conducted studies,

(D) Tables summarizing the protocols and results of all cited studies,

(E) Summary of significant results and conclusions with respect to the effects of the emissions of the subject fuel or fuel additive on the public health and welfare, including references if used to support such results and conclusions.

(F) Statement of the extent to which the literature search has produced adequate information comparable to that which would otherwise be obtained through the performance of applicable emission characterization requirements under Sec. 79.52(b) and/or health effects testing requirements under Sec. 79.53, including justifications and specific references.

(ii) Emission Characterization. Pursuant to Sec. 79.52(b), the emission characterization shall include:

(A) Name, address, and telephone number of the laboratory performing the characterization,

(B) Name and description of analytic methods used for characterization.

(iii) Exposure Analysis. Pursuant to Sec. 79.52(c), the exposure analysis shall include:

(A) A qualitative discussion of the potential exposure(s) of the general and any special at-risk populations to the emission products, based on annual and projected production volume, and market distribution data. For group submittals, this discussion shall address the characteristics of the cumulative exposure from the potential use of all fuel or additive products in the group.

(B) Identification of person(s) preparing the analysis.

(5) Summary of Tier 2. For each health effects test performed pursuant to the provisions of Sec. 79.53, the Tier 2 summary shall contain the following information:

(i) Name, address, and telephone number of the testing facility,

(ii) Summary of procedures (including quality assurance, quality control and compliance with Good Laboratory Practice Standards as specified in Sec. 79.60), findings, and conclusions, including references if used to support such results and conclusions,

(iii) Description of any problems and their resolution.

(6) Conclusions. The conclusions shall identify the need for further testing, if that need exists, or justify that current testing and/or available information is adequate for the tier(s) included in the report.

(7) Appendices. The appendices shall contain detailed documentation related to the summary information described in this section, including, at a minimum, the following five appendices:

(i) Literature search appendices shall contain:

(A) Copies of literature source outputs, including reference lists and associated abstracts from database searches, printed or on 3\1/2\ inch IBM-compatible computer diskettes;

(B) Summary tables organized by health or welfare endpoint and type of emission (e.g., combustion, evaporation, individual emission product), presenting in tabular form the following information at a minimum: number and species of test subjects, exposure concentrations/ duration, positive (i.e., abnormal) findings including numbers of test subjects involved, and bibliographic references;

(C) Complete documentation and/or reprints of articles for any previous study relied upon for satisfying emission characterization and/or Tier 2 test requirements; and

(D) Full reports for unpublished/in-house studies.

(ii) Emissions characterization appendices shall contain:

(A) Complete laboratory reports, including documentation of calibration and verification procedures;

(B) Documentation of the emissions generation procedures used; and

(C) Lists of speciated emission products and their emission rates reported in units of grams/mile.

(iii) Exposure analysis appendices may be submitted to report any detailed documentation of data used in the analyses and/or calculations determining potential exposures to population(s). If modeling data are used, these should be included in an appendix.

(iv) Tier 2 appendices shall contain, for each test performed:

(A) Complete protocol used;

(B) Documentation of emission generation procedures; and

(C) Complete laboratory report in compliance with the reporting standards in Sec. 79.60, including detailed test results and conclusions, and descriptions of any problems encountered and their resolution.

(v) Laboratory certification/accreditation information, personnel

credentials, and statements of compliance with the Good Laboratory Practices Standards specified in Sec. 79.60 and the requirements in Sec. 79.53(c)(1).

(d) Tier 3 Report. Subject to applicability as specified in Sec. 79.54, each manufacturer of a designated fuel or fuel additive, or each group of such manufacturers pursuant to the provisions of Sec. 79.56, shall submit the following information with respect to each Tier 3 test conducted for such fuels or fuel additives:

(1) The test objectives, including a summary of the reason(s) why such additional testing, beyond Tiers 1 and 2, was required;

(2) Name, address, and telephone number of each testing facility;

(3) Summary of test procedures, results and conclusions;

(4) Complete documentation of test protocols and emission generation procedures, complete laboratory reports in compliance with the reporting standards of Sec. 79.60, detailed test results and conclusions, including references if used to support such results and conclusions, and descriptions of any problems encountered and their resolution; and

(5) Laboratory certification information, personnel credentials, and statements of compliance with the Good Laboratory Practices Standards specified in Sec. 79.60.

(e) Availability of Information. (1) All health and safety test data and other information concerning health and welfare effects which is submitted by any manufacturer or group pursuant to Secs. 79.52(c), 79.53, or 79.54, shall be considered to be public information and shall be made available to the public by EPA upon request. A reasonable fee may be charged by EPA for copying such materials. Any manufacturer or group who claims that any information concerning the composition of a fuel or fuel additive product, or any other information, submitted under this subpart is confidential business information must state this claim in writing at the time of the submittal.

(2) To assert a business confidentiality claim concerning any information submitted under this subpart, the submitter must:

(i) Clearly mark the information as confidential at each location it appears in the submission; and

(ii) Submit with the information claimed as confidential a separate document setting forth the claim and listing each location at which the information appears in the submission.

(3) If any person subsequently requests access to information submitted under this subpart (other than health and safety test data and other information concerning health and welfare effects), and such information is subject to a claim of business confidentiality, the request and any subsequent disclosure shall be governed by the provisions of 40 CFR part 2.

Sec. 79.60 Good laboratory practices (GLP) standards for inhalation exposure health effects testing.

(a) General Provisions--(1) Scope. (i) This section prescribes good laboratory practices (GLPs) for conducting inhalation exposure studies relating to motor vehicle emissions health effects testing under this part. These directions are intended to ensure the quality and integrity of health effects data submitted pursuant to registration regulations issued under sections 211(b) or 211(e) of the Clean Air Act (CAA) (42 U.S.C. 7545).

(ii) This section applies to any study described by paragraph (a)(1)(i) of this section which any person conducts, initiates, or supports on or after May 27, 1994.

(iii) It is EPA's policy that all health effects data developed under sections 211(b) and (e) of CAA be in accordance with provisions of this section. If data are not developed in accordance with the provisions of this section, EPA may consider such data insufficient to evaluate the health effects of a motor vehicle's fuel or fuel additive emissions, unless the submitter provides additional information demonstrating that the data are reliable and adequate and EPA determines that the data are sufficient.

(2) Definitions. As used in this section, the following terms shall have the meanings specified:

Batch means a specific quantity or lot of a test fuel, additive/base fuel mixture, or reference substance that has been characterized according to Sec. 79.60(f)(1)(i).

CAA means the Clean Air Act.

Carrier means any material which is combined with engine/motor vehicle emissions or a reference substance for administration to a test system. ``Carrier'' includes, but is not limited to, clean, filtered air, water, feed, and nutrient media.

Control atmosphere means clean, filtered air which is administered to the test system in the course of a study for the purpose of establishing a basis for comparison with the test atmosphere for chemical or biological measurements.

Experimental start date means the first date the test atmosphere is applied to the test system.

Experimental termination date means the last date on which data are collected directly from the study.

Person includes an individual, partnership, corporation, association, scientific or academic establishment, government agency, or organizational unit thereof, and any other legal entity.

Quality assurance unit means any person or organizational element, except the study director, designated by testing facility management to perform the duties relating to quality assurance of the studies.

Raw data means any laboratory worksheets, records, memoranda, notes, or exact copies thereof, that are the result of original observations and activities of a study and are necessary for the reconstruction and evaluation of the report of that study. In the event that exact transcripts of raw data have been prepared (e.g., tapes which have been transcribed verbatim, dated, and verified accurate by signature), the exact copy or exact transcript may be substituted for the original source as raw data. ``Raw data" may include photographs, videotape, microfilm or microfiche copies, computer printouts, magnetic media, including dictated observations, and recorded data from automated instruments.

Reference substance means any chemical substance or mixture, analytical standard, or material other than engine/motor vehicle emissions and/or its carrier, that is administered to or used in analyzing the test system in the course of a study. A ``reference substance" is used to establish a basis for comparison with the test atmosphere for known chemical or biological measurements, i.e., positive or negative control substance.

Specimen means any material derived from a test system for examination or analysis.

Sponsor means person who initiates and supports, by provision of financial or other resources, a study or a person who submits a study to EPA in response to the CAA Section 211(b) or 211(e) Fuels and Fuel Additives Registration Rule or a testing facility, if it both initiates and actually conducts the study.

Study means any experiment, at one or more test sites, in which a test system is exposed to a test atmosphere under laboratory conditions to determine or help predict the health effects of that exposure in humans, other living organisms, or media.

Study completion date means the date the final report is signed by the study director.

Study director means the individual responsible for the overall conduct of a study.

Study initiation date means the date the protocol is signed by the study director.

Test substance means a vapor and/or aerosol mixture composed of engine/motor vehicle emissions and clean, filtered air which is administered directly, or indirectly, by the inhalation route to a test system in a study which develops data to meet the registration requirements of CAA section 211(b) or (e).

Test system means any animal, microorganism, chemical or physical matrix, to which the test, control, or reference substance is administered or added for study. This definition also includes appropriate groups or components of the system not treated with the test, control, or reference substance.

Testing facility means a person who actually conducts a study, i.e., actually uses the test substance in a test system. ``Testing facility'' encompasses only those operational units that are being or have been used to conduct studies.

TSCA means the Toxic Substances Control Act (15 U.S.C. 2601 et seq.).

(3) Applicability to studies performed under grants and contracts. When a sponsor or other person utilizes the services of a consulting laboratory, contractor, or grantee to perform all or a part of a study to which this section applies, it shall notify the consulting laboratory, contractor, or grantee that the service is, or is part of, a study that must be conducted in compliance with the provisions of this section.

(4) Statement of compliance or non-compliance. Any person who submits to EPA a test in compliance with registration regulations issued under CAA section 211(b) or section 211(e) shall include in the submission a true and correct statement, signed by the sponsor and the study director, of one of the following types:

(i) A statement that the study was conducted in accordance with this section; or

(ii) A statement describing in detail all differences between the practices used in the study and those required by this section; or

(iii) A statement that the person was not a sponsor of the study, did not conduct the study, and does not know whether the study was conducted in accordance with this section.

(5) Inspection of a testing facility. (i) A testing facility shall permit an authorized employee or duly designated representative of EPA, at reasonable times and in a reasonable manner, to inspect the facility and to inspect (and in the case of records also to copy) all records and specimens required to be maintained regarding studies to which this section applies. The records inspection and copying requirements shall not apply to quality assurance unit records of findings and problems, or to actions recommended and taken, except the EPA may seek production of these records in litigation or formal adjudicatory hearings.

(ii) EPA will not consider reliable for purposes of showing that a test substance does or does not present a risk of injury to health or the environment any data developed by a testing facility or sponsor that refuses to permit inspection in accordance with this section. The determination that a study will not be considered reliable does not, however, relieve the sponsor of a required test of any obligation under any applicable statute or regulation to submit the results of the study to EPA.

(6) Effects of non-compliance. (i) Pursuant to sections 114, 208, and 211(d) of the CAA, it shall be a violation of this section and a violation of this rule (40 CFR part 79, subpart F) if:

(A) The test is not being or was not conducted in accordance with any requirement of this part; or

(B) Data or information submitted to EPA under part 79, including the statement required by Sec. 79.60(a)(4), include information or data that are false or misleading, contain significant omissions, or otherwise do not fulfill the requirements of this part; or

(C) Entry in accordance with Sec. 79.60(a)(5) for the purpose of auditing test data is denied.

(ii) EPA, at its discretion, may not consider reliable for purposes of showing that a chemical substance or mixture does not present a risk of injury to health any study which was not conducted in accordance with this part. EPA, at its discretion, may rely upon such studies for purposes of showing adverse effects. The determination that a study will not be considered reliable does not, however, relieve the sponsor of a required test of the obligation under any applicable statute or regulation to submit the results of the study to EPA.

(iii) If data submitted in compliance with registration regulations issued under CAA section 211(b) or section 211(e) are not developed in accordance with this section, EPA may determine that the sponsor has not fulfilled its obligations under 40 CFR part 79 and may require the sponsor to develop data in accordance with the requirements of this section in order to satisfy such obligations.

(b) Organization and Personnel. (1) Personnel. (i) Each individual engaged in the conduct of or responsible for the supervision of a study shall have education, training, and experience, or combination thereof, to enable that individual to perform the assigned functions.

(ii) Each testing facility shall maintain a current summary of training and experience and job description for each individual engaged in or supervising the conduct of a study.

(iii) There shall be a sufficient number of personnel for the timely and proper conduct of the study according to the protocol.

(iv) Personnel shall take necessary personal sanitation and health precautions designed to avoid contamination of test fuel and additive/base fuel mixtures, test and reference substances, and test systems.

(v) Personnel engaged in a study shall wear clothing appropriate for the duties they perform. Such clothing shall be changed as often as necessary to prevent microbiological, radiological, or chemical contamination of test systems and test, control, and reference substances.

(vi) Any individual found at any time to have an illness that may adversely affect the quality and integrity of the study shall be excluded from direct contact with test systems, fuel and fuel/additive mixtures, test and reference substances and any other operation or function that may adversely affect the study until the condition is corrected. All personnel shall be instructed to report to their

immediate supervisors any health or medical conditions that may reasonably be considered to have an adverse effect on a study.

(2) Testing facility management. For each study, testing facility management shall:

(i) Designate a study director as described in Sec. 79.60(b)(3) before the study is initiated.

(ii) Replace the study director promptly if it becomes necessary to do so during the conduct of a study.

(iii) Assure that there is a quality assurance unit as described in Sec. 79.60(b)(4).

(iv) Assure that test fuels and fuel/additive mixtures and test and reference substances have been identified as to content, strength, purity, stability, and uniformity, as applicable.

(v) Assure that personnel, resources, facilities, equipment, materials and methodologies are available as scheduled.

(vi) Assure that personnel clearly understand the functions they are to perform.

(vii) Assure that any deviations from these regulations reported by the quality assurance unit are communicated to the study director and corrective actions are taken and documented.

(3) Study director. For each study, a scientist or other professional person with a doctorate degree or equivalent in toxicology or other appropriate discipline shall be identified as the study director. The study director has overall responsibility for the technical conduct of the study, as well as for the interpretation, analysis, documentation, and reporting of results, and represents the single point of study control. The study director shall assure that:

(i) The protocol, including any changes, is approved as provided by Sec. 79.60(g)(1)(i) and is followed;

(ii) All experimental data, including observations of unanticipated responses of the test system are accurately recorded and verified;

(iii) Unforeseen circumstances that may affect the quality and integrity of the study are noted when they occur, and corrective action is taken and documented;

(iv) Test systems are as specified in the protocol;

(v) All applicable good laboratory practice regulations are followed; and

(vi) All raw data, documentation, protocols, specimens, and final reports are archived properly during or at the close of the study.

(4) Quality assurance unit. A testing facility shall have a quality assurance unit which shall be responsible for monitoring each study to assure management that the facilities, equipment, personnel, methods, practices, records, and controls are in conformance with the regulations in this section. For any given study, the quality assurance unit shall be entirely separate from and independent of the personnel

engaged in the direction and conduct of that study. The quality assurance unit shall conduct inspections and maintain records appropriate to the study.

(i) Quality assurance unit duties. (A) Maintain a copy of a master schedule sheet of all studies conducted at the testing facility indexed by test substance and containing the test system, nature of study, date study was initiated, current status of each study, identity of the sponsor, and name of the study director.

(B) Maintain copies of all protocols pertaining to all studies for which the unit is responsible.

(C) Inspect each study at intervals adequate to ensure the integrity of the study and maintain written and properly signed records of each periodic inspection showing the date of the inspection, the study inspected, the phase or segment of the study inspected, the person performing the inspection, findings and problems, action recommended and taken to resolve existing problems, and any scheduled date for re-inspection. Any problems which are likely to affect study integrity found during the course of an inspection shall be brought to the attention of the study director and management immediately.

(D) Periodically submit to management and the study director written status reports on each study, noting any problems and the corrective actions taken.

(E) Determine that no deviations from approved protocols or standard operating procedures were made without proper authorization and documentation.

(F) Review the final study report to assure that such report accurately describes the methods and standard operating procedures, and that the reported results accurately reflect the raw data of the study.

(G) Prepare and sign a statement to be included with the final study report which shall specify the dates inspections were made and findings reported to management and to the study director.

(ii) The responsibilities and procedures applicable to the quality assurance unit, the records maintained by the quality assurance unit, and the method of indexing such records shall be in writing and shall be maintained. These items including inspection dates, the study inspected, the phase or segment of the study inspected, and the name of the individual performing the inspection shall be made available for inspection to authorized employees or duly designated representatives of EPA.

(iii) An authorized employee or a duly designated representative of EPA shall have access to the written procedures established for the inspection and may request test facility management to certify that inspections are being implemented, performed, documented, and followed up in accordance with this paragraph.

(c) Facilities--(1) General. Each testing facility shall be of

suitable size and construction to facilitate the proper conduct of studies. Testing facilities which are not completely located within an indoor controlled environment shall be of suitable location/proximity to facilitate the proper conduct of studies. Testing facilities shall be designed so that there is a degree of separation that will prevent any function or activity from having an adverse effect on the study.

(2) Test system care facilities. (i) A testing facility shall have a sufficient number of animal rooms or other test system areas, as needed, to ensure proper separation of species or test systems, quarantine or isolation of animals or other test systems, and routine or specialized housing of animals or other test systems.

(ii) A testing facility shall have a number of animal rooms or other test system areas separate from those described in paragraph (a) of this section to ensure isolation of studies being done with test systems or test, control, and reference substances known to be biohazardous, including volatile atmospheres and aerosols, radioactive materials, and infectious agents. The animal handling facility must operate under the supervision of a veterinarian.

(iii) Separate areas shall be provided, as appropriate, for the diagnosis, treatment, and control of laboratory test system diseases. These areas shall provide effective isolation for the housing of test systems either known or suspected of being diseased, or of being carriers of disease, from other test systems.

(iv) Facilities shall have proper provisions for collection and disposal of contaminated air, water, or other spent materials. When animals are housed, facilities shall exist for the collection and disposal of all animal waste and refuse or for safe sanitary storage of waste before removal from the testing facility. Disposal facilities shall be so provided and operated as to minimize vermin infestation, odors, disease hazards, and environmental contamination.

(v) Facilities shall have provisions to regulate environmental conditions (e.g., temperature, humidity, day length, etc.) as specified in the protocol.

(3) Test system supply/operation areas. (i) There shall be storage areas, as needed, for feed, bedding, supplies, and equipment. Storage areas for feed and bedding shall be separated from areas where the test systems are located and shall be protected against infestation or contamination. Perishable supplies shall be preserved by appropriate means.

(ii) Separate laboratory space and other space shall be provided, as needed, for the performance of the routine and specialized procedures required by studies.

(4) Facilities for handling test fuels and fuel/additive mixtures and reference substances. (i) As necessary to prevent contamination or mixups, there shall be separate areas for:

(A) Receipt and storage of the test fuels and fuel/additive mixtures and reference substances;

(B) Mixing of the test fuels, fuel/additive mixtures, and reference substances with a carrier, i.e., liquid hydrocarbon; and

(C) Storage of the test fuels, fuel/additive mixtures, and reference substance/carrier mixtures.

(ii) Storage areas for test fuels and fuel/additive mixtures and reference substances and for reference mixtures shall be separate from areas housing the test systems and shall be adequate to preserve the identity, strength, purity, and stability of the substances and mixtures.

(5) Specimen and data storage facilities. Space shall be secured for archives for the storage and retrieval of all raw data and specimens from completed studies.

(d) Equipment--(1) Equipment design. Equipment used in the generation, measurement, or assessment of data and equipment used for facility environmental control shall be of appropriate design and adequate capacity to function according to the protocol and shall be suitably located for operation, inspection, cleaning, and maintenance.

(2) Maintenance and calibration of equipment. (i) Equipment shall be adequately inspected, cleaned, and maintained. Equipment used for the generation, measurement, or assessment of data shall be adequately tested, calibrated, and/or standardized.

(ii) The written standard operating procedures required under Sec. 79.60(e)(1)(ii)(K) shall set forth in sufficient detail the methods, materials, and schedules to be used in the routine inspection, cleaning, maintenance, testing, calibration, and/or standardization of equipment, and shall specify, when appropriate, remedial action to be taken in the event of failure or malfunction of equipment. The written standard operating procedures shall designate the person responsible for the performance of each operation.

(iii) Written records shall be maintained of all inspection, maintenance, testing, calibrating, and/or standardizing operations. These records, containing the date of the operation, shall describe whether the maintenance operations were routine and followed the written standard operating procedures. Written records shall be kept of non-routine repairs performed on equipment as a result of failure and malfunction. Such records shall document the nature of the defect, how and when the defect was discovered, and any remedial action taken in response to the defect.

(e) Testing Facilities Operation--(1) Standard operating procedures. (i) A testing facility shall have standard operating procedures in writing, setting forth study methods that management is satisfied are adequate to insure the quality and integrity of the data generated in the course of a study. All deviations in a study from

standard operating procedures shall be authorized by the study director and shall be documented in the raw data. Significant changes in established standard operating procedures shall be properly authorized in writing by management.

(ii) Standard operating procedures shall be established for, but not limited to, the following:

(A) Test system room preparation;

(B) Test system care;

(C) Receipt, identification, storage, handling, mixing, and method of sampling of test fuels and fuel/additive mixtures and reference substances;

(D) Test system observations;

(E) Laboratory or other tests;

(F) Handling of test animals found moribund or dead during study;

(G) Necropsy or postmortem examination of test animals;

(H) Collection and identification of specimens;

(I) Histopathology

(J) Data handling, storage and retrieval.

(K) Maintenance and calibration of equipment.

(L) Transfer, proper placement, and identification of test systems.

(iii) Each laboratory or other study area shall have immediately available manuals and standard operating procedures relative to the laboratory procedures being performed. Published literature may be used as a supplement to standard operating procedures.

(iv) A historical file of standard operating procedures, and all revisions thereof, including the dates of such revisions, shall be maintained.

(2) Reagents and solutions. All reagents and solutions in the laboratory areas shall be labeled to indicate identity, titer or concentration, storage requirements, and expiration date. Deteriorated or outdated reagents and solutions shall not be used.

(3) Animal and other test system care. (i) There shall be standard operating procedures for the housing, feeding, handling, and care of animals and other test systems.

(ii) All newly received test systems from outside sources shall be isolated and their health status or appropriateness for the study shall be evaluated. This evaluation shall be in accordance with acceptable veterinary medical practice or scientific methods.

(iii) At the initiation of a study, test systems shall be free of any disease or condition that might interfere with the purpose or conduct of the study. If during the course of the study, the test systems contract such a disease or condition, the diseased test systems shall be isolated, if necessary. These test systems may be treated for disease or signs of disease provided that such treatment does not interfere with the study. The diagnosis, authorization of treatment,

description of treatment, and each date of treatment shall be documented and shall be retained.

(iv) When laboratory procedures require test animals to be manipulated and observed over an extended period of time or when studies require test animals to be removed from and returned to their housing units for any reason (e.g., cage cleaning, treatment, etc.), these test systems shall receive appropriate identification (e.g., tattoo, color code, etc.). Test system identification shall conform with current laboratory animal handling practice. All information needed to specifically identify each test system within the test system-housing unit shall appear on the outside of that unit. Suckling animals are excluded from the requirement of individual identification unless otherwise specified in the protocol.

(v) Except as specified in paragraph (e)(3)(v)(A) of this section, test animals of different species shall be housed in separate rooms when necessary. Test animals of the same species, but used in different studies, shall not ordinarily be housed in the same room when inadvertent exposure to the test or reference substances or test system mixup could affect the outcome of either study. If such mixed housing is necessary, adequate differentiation by space and identification shall be made.

(A) Test systems that may be used in multispecies tests need not be housed in separate rooms, provided that they are adequately segregated to avoid mixup and cross-contamination.

(B) [reserved]

(vi) Cages, racks, pens, enclosures, and other holding, rearing, and breeding areas, and accessory equipment, shall be cleaned and sanitized at appropriate intervals.

(vii) Feed and water used for the test animals shall be analyzed periodically to ensure that contaminants known to be capable of interfering with the study and reasonably expected to be present in such feed or water are not present at greater than trace levels. Documentation of such analyses shall be maintained as raw data.

(viii) Bedding used in animal cages or pens shall not interfere with the purpose or conduct of the study and shall be changed as often as necessary to keep the animals dry and clean.

(ix) If any pest control materials are used, the use shall be documented. Cleaning and pest control materials that interfere with the study shall not be used.

(x) All test systems shall be acclimatized to the environmental conditions of the test, prior to their use in a study.

(f) Test fuels, additive/base fuel mixtures, and reference substances--(1) Test fuel, fuel/additive mixture, and reference substance identity (i) The product brand name/service mark, strength, purity, content, or other characteristics which appropriately define

the test fuel, fuel/additive mixture, or reference substance shall be reported for each batch and shall be documented before its use in a study. Methods of synthesis, fabrication, or derivation, as appropriate, of the test fuel, fuel/additive mixture, or reference substance shall be documented by the sponsor or the testing facility, and such location of documentation shall be specified.

(ii) The stability of test fuel, fuel/additive mixture, and reference substances under storage conditions at the test site shall be known for all studies.

(2) Test fuel, additive/base fuel mixture, and reference substance handling. Procedures shall be established for a system for the handling of the test fuel, fuel/additive mixture, and reference substance(s) to ensure that:

(i) There is proper storage.

(ii) Distribution is made in a manner designed to preclude the possibility of contamination, deterioration, or damage.

(iii) Proper identification is maintained throughout the distribution process.

(iv) The receipt and distribution of each batch is documented. Such documentation shall include the date and quantity of each batch distributed or returned.

(3) Mixtures of test emissions or reference solutions with carriers.

(i) For test emissions or each reference substance mixed with a carrier, tests by appropriate analytical methods shall be conducted:

(A) To determine the uniformity of the test substance and to determine, periodically, the concentration of the test emissions or reference substance in the mixture;

(B) When relevant to the conduct of the experiment, to determine the solubility of each reference substance in the carrier mixture before the experimental start date; and

(C) To determine the stability of test emissions or a reference solution in the test substance before the experimental start date or concomitantly according to written standard operating procedures, which provide for periodic analysis of each batch.

(ii) Where any of the components of the reference substance/carrier mixture has an expiration date, that date shall be clearly shown on the container. If more than one component has an expiration date, the earliest date shall be shown.

(iii) If a chemical or physical agent is used to facilitate the mixing of a test substance with a carrier, assurance shall be provided that the agent does not interfere with the integrity of the test.

(g) Protocol for and conduct of a study--(1) Protocol. (i) Each study shall have a written protocol that clearly indicates the objectives and all methods for the conduct of the study. The protocol

shall contain but shall not be limited to the following information:

(A) A descriptive title and statement of the purpose of the study.

(B) Identification of the test fuel, fuel/additive mixture, and reference substance by name, chemical abstracts service (CAS) number or code number, as applicable.

(C) The name and address of the sponsor and the name and address of the testing facility at which the study is being conducted.

(D) The proposed experimental start and termination dates.

(E) Justification for selection of the test system, as necessary.

(F) Where applicable, the number, body weight, sex, source of supply, species, strain, substrain, and age of the test system.

(G) The procedure for identification of the test system.

(H) A description of the experimental design, including methods for the control of bias.

(I) Where applicable, a description and/or identification of the diet used in the study. The description shall include specifications for acceptable levels of contaminants that are reasonably expected to be present in the dietary materials and are known to be capable of interfering with the purpose or conduct of the study if present at levels greater than established by the specifications.

(J) Each concentration level, expressed in milligrams per cubic meter of air or other appropriate units, of the test or reference substance to be administered and the frequency of administration.

(K) The type and frequency of tests, analyses, and measurements to be made.

(L) The records to be maintained.

(M) The date of approval of the protocol by the sponsor and the dated signature of the study director.

(N) A statement of the proposed statistical method.

(ii) All changes in or revisions of an approved protocol and the reasons therefor shall be documented, signed by the study director, dated, and maintained with the protocol.

(2) Conduct of a study. (i) The study shall be conducted in accordance with the protocol.

(ii) The test systems shall be monitored in conformity with the protocol.

(iii) Specimens shall be identified by test system, study, nature, and date of collection. This information shall be located on the specimen container or shall accompany the specimen in a manner that precludes error in the recording and storage of data.

(iv) In animal studies where histopathology is required, records of gross findings for a specimen from postmortem observations shall be available to a pathologist when examining that specimen histopathologically.

(v) All data generated during the conduct of a study, except those

that are generated by automated data collection systems, shall be recorded directly, promptly, and legibly in ink. All data entries shall be dated on the day of entry and signed or initialed by the person entering the data. Any change in entries shall be made so as not to obscure the original entry, shall indicate the reason for such change, and shall be dated and signed or identified at the time of the change. In automated data collection systems, the individual responsible for direct data input shall be identified at the time of data input. Any change in automated data entries shall be made so as not to obscure the original entry, shall indicate the reason for change, shall be dated, and the responsible individual shall be identified.

(h) Records and Reports--(1) Reporting of study results. (i) A final report shall be prepared for each study and shall include, but not necessarily be limited to, the following:

(A) Name and address of the facility performing the study and the dates on which the study was initiated and was completed, terminated, or discontinued.

(B) Objectives and procedures stated in the approved protocol, including any changes in the original protocol.

(C) Statistical methods employed for analyzing the data.

(D) The test fuel, additive/base fuel mixture, and test and reference substances identified by name, chemical abstracts service (CAS) number or code number, strength, purity, content, or other appropriate characteristics.

(E) Stability, and when relevant to the conduct of the study, the solubility of the test emissions and reference substances under the conditions of administration.

(F) A description of the methods used.

(G) A description of the test system used. Where applicable, the final report shall include the number of animals or other test organisms used, sex, body weight range, source of supply, species, strain and substrain, age, and procedure used for identification.

(H) A description of the concentration regimen as daily exposure period, i.e., number of hours, and exposure duration, i.e., number of days.

(I) A description of all circumstances that may have affected the quality or integrity of the data.

(J) The name of the study director, the names of other scientists or professionals and the names of all supervisory personnel, involved in the study.

(K) A description of the transformations, calculations, or operations performed on the data, a summary and analysis of the data, and a statement of the conclusions drawn from the analysis.

(L) The signed and dated reports of each of the individual scientists or other professionals involved in the study, including each

person who, at the request or direction of the testing facility or sponsor, conducted an analysis or evaluation of data or specimens from the study after data generation was completed.

(M) The locations where all specimens, raw data, and the final report are to be kept or stored.

(N) The statement, prepared and signed by the quality assurance unit, as described in Sec. 79.60(b)(4)(i)(G).

(ii) The final report shall be signed and dated by the study director.

(iii) Corrections or additions to a final report shall be in the form of an amendment by the study director. The amendment shall clearly identify that part of the final report that is being added to or corrected and the reasons for the correction or addition, and shall be signed and dated by the person responsible. Modification of a final report to comply with the submission requirements of EPA does not constitute a correction, addition, or amendment to a final report.

(iv) A copy of the final report and of any amendment to it shall be maintained by the sponsor and the test facility.

(2) Storage and retrieval of records and data. (i) All raw data, documentation, records, protocols, specimens, and final reports generated as a result of a study shall be retained. Specimens obtained from mutagenicity tests, wet specimens of blood, urine, feces, and biological fluids, do not need to be retained after quality assurance verification. Correspondence and other documents relating to interpretation and evaluation of data, other than those documents contained in the final report, also shall be retained.

(ii) All raw data, documentation, protocols, specimens, and interim and final reports shall be archived for orderly storage and expedient retrieval. Conditions of storage shall minimize deterioration of the documents or specimens in accordance with the requirements for the time period of their retention and the nature of the documents of specimens. A testing facility may contract with commercial archives to provide a repository for all material to be retained. Raw data and specimens may be retained elsewhere provided that the archives have specific reference to those other locations.

(iii) An individual shall be identified as responsible for the archiving of records.

(iv) Access to archived material shall require authorization and documentation.

(v) Archived material shall be indexed to permit expedient retrieval.

(3) Retention of records. (i) Record retention requirements set forth in this section do not supersede the record retention requirements of any other regulations in this subchapter.

(ii) Except as provided in paragraph (h)(3)(iii) of this section,

documentation records, raw data, and specimens pertaining to a study and required to be retained by this part shall be archived for a period of at least ten years following the completion of the study.

(iii) Wet specimens, samples of test fuel, additive/base fuel mixtures, or reference substances, and specially prepared material which are relatively fragile and differ markedly in stability and quality during storage, shall be retained only as long as the quality of the preparation affords evaluation. Specimens obtained from mutagenicity tests, wet specimens of blood, urine, feces, biological fluids, do not need to be retained after quality assurance verification. In no case shall retention be required for a longer period than that set forth in paragraph (h)(3)(ii) of this section.

(iv) The master schedule sheet, copies of protocols, and records of quality assurance inspections, as required by Sec. 79.60(b)(4)(iii) shall be maintained by the quality assurance unit as an easily accessible system of records for the period of time specified in paragraph (h)(3)(ii) of this section.

(v) Summaries of training and experience and job descriptions required to be maintained by Sec. 79.60(b)(1)(ii) may be retained along with all other testing facility employment records for the length of time specified in paragraph (h)(3)(ii) of this section.

(vi) Records and reports of the maintenance and calibration and inspection of equipment, as required by Sec. 79.60(d)(2) (ii) and (iii), shall be retained for the length of time specified in paragraph (h)(3)(ii) of this section.

(vii) If a facility conducting testing or an archive contracting facility goes out of business, all raw data, documentation, and other material specified in this section shall be transferred to the sponsor of the study for archival.

(viii) Records required by this section may be retained either as original records or as true copies such as photocopies, microfilm, microfiche, or other accurate reproductions of the original records.

Sec. 79.61. Vehicle emissions inhalation exposure guideline.

(a) Purpose. This guideline provides additional information on methodologies required to conduct health effects tests involving inhalation exposures to vehicle combustion emissions from fuels or fuel/additive mixtures. Where this guideline and the other health effects testing guidelines in 40 CFR 79.62 through 79.68 specify differing values for the same test parameter, the specifications in the individual health test guideline shall prevail for that health effect endpoint.

(b) Definitions. For the purposes of this section the following

definitions apply.

Acute inhalation study means a short-term toxicity test characterized by a single exposure by inhalation over a short period of time (at least 4 hours and less than 24 hours), followed by at least 14 days of observation.

Aerodynamic diameter means the diameter of a sphere of unit density that has the same settling velocity as the particle of the test substance. It is used to compare particles of different sizes, densities and shapes, and to predict where in the respiratory tract such particles may be deposited. It applies to the size of aerosol particles.

Chronic inhalation study means a prolonged and repeated exposure by inhalation for the life span of the test animal; technically, two years in the rat.

Concentration means an exposure level. Exposure is expressed as weight or volume of test aerosol/substance per volume of air, usually mg/m<SUP>3 or as parts per million (ppm) over a given time period. Micrograms per cubic meter (<greek-m>g/m<SUP>3) or parts per billion may be appropriate, as well.

Cumulative toxicity means the adverse effects of repeated exposures occurring as a result of prolonged action or increased concentration of the administered test substance or its metabolites in the susceptible tissues.

Inhalable diameter means that aerodynamic diameter of a particle which is considered to be inhalable for the organism. It is used to refer to particles which are capable of being inhaled and may be deposited anywhere within the respiratory tract from the trachea to the alveoli.

Mass median aerodynamic diameter (MMAD) means the calculated aerodynamic diameter, which divides the particles of an aerosol in half based on the mass of the particles. Fifty percent of the particles in mass will be larger than the median diameter, and fifty percent will be smaller than the median diameter. MMAD describes the particle distribution of any aerosol based on the weight and size of the particles. MMAD and the geometric standard deviation describe the particle-size distribution.

Material safety data sheet (MSDS) means documentation or information on the physical, chemical, and hazardous characteristics of a given chemical, usually provided by the product's manufacturer.

Reynolds number means a dimensionless number that is proportional to the ratio of inertial forces to frictional forces acting on a fluid. It quantitatively provides a measure of whether flow is laminar or turbulent. A fluid traveling through a pipe is fully developed into a laminar flow for a Reynolds number less than 2000, and fully developed into a turbulent flow for a Reynolds number greater than 4000.

Subacute inhalation toxicity means the adverse effects occurring as a result of the repeated daily exposure of experimental animals to a chemical by inhalation for part (less than 10 percent) of a lifespan; generally, less than 90 days.

Subchronic inhalation study means a repeated exposure by inhalation for part (approximately 10 percent) of a life span of the exposed test animal.

Toxic effect means an adverse change in the structure or function of an experimental animal as a result of exposure to a chemical substance.

(c) Principles and design criteria of inhalation exposure systems. Proper conduct of inhalation toxicity studies of the emissions of fuels and additive/fuel mixtures requires that the exposure system be designed to ensure the controlled generation of the exposure atmosphere, the adequate dilution of the test emissions, delivery of the diluted exposure atmosphere to the test animals, and use of appropriate exposure chamber systems selected to meet criteria for a given exposure study.

(1) Emissions generation. Emissions shall be generated according to the specifications in 40 CFR 79.57.

(2) Dilution and delivery systems.

(i) The delivery system is the means used to transport the emissions from the generation system to the exposure system. The dilution system is generally a component of the delivery system.

(ii) Dilution provides control of the emissions concentration delivered to the exposure system, serving the function of diluting the associated combustion gases, such as carbon monoxide, carbon dioxide, nitrogen oxides, sulfur dioxide and other noxious gases and vapors, to levels that will ensure that there are no significant or measurable responses in the test animals as a result of exposure to the combustion gases. The formation of particle species is strongly dependent on the dilution rate, as well.

(iii) The engine exhaust system shall connect to the first-stage-dilution section at 90 deg. to the axis of the dilution section. This is then connected to a right angle elbow on the center line of the dilution section. Engine emissions are injected through the elbow so that exhaust flow is concurrent to dilution flow.

(iv) Materials. In designing the dilution and delivery systems, the use of plastic, e.g., PVC and similar materials, copper, brass, and aluminum pipe and tubing shall be avoided if there exists a possibility of chemical reaction occurring between emissions and tubing. Stainless steel pipe and tubing is recommended as the best choice for most emission dilution and delivery applications, although glass and teflon may be appropriate, as well.

(v) Flow requirements. (A) Conduit for dilute raw emissions shall

be of such dimensions as to provide residence times for the emissions on the order of less than one second to several seconds before the emissions are further diluted and introduced to the test chambers. With the high flow rates in the dilute raw emissions conduit, it will be necessary to sample various portions of the dilute emissions for delivering differing concentrations to the test chambers. The unused portions of the emissions stream are normally exhausted to the atmosphere outside of the exposure facility.

(B) Dimensions of the dilute raw exhaust conduit shall be such that, at a minimum, the flow Reynolds number is 70,000 or greater (see Mokler, et al., 1984 in paragraph (f)(13) of this section). This will maintain highly turbulent flow conditions so that there is more complete mixing of the exhaust emissions.

(C) Wall losses. The delivery system shall be designed to minimize wall losses. This can be done by sizing the tubing or pipe to maintain laminar flow of the diluted emissions to the exposure chamber. A flow Reynolds number of 1000-3000 will ensure minimal wall losses. Also, the length of and number and degree of bends in the delivery lines to the exposure chamber system shall be minimized.

(D) Whole-body exposure vs. nose-only exposure delivery systems. Flow rates through whole-body chamber systems are of the order of 100 liters per minute to 500 liters per minute. Nose-only systems are on the order of less than 50 liters per minute. To maintain laminar flow conditions, the principles described in paragraph (c)(2)(v)(C) of this section apply to both systems.

(vi) Dilution requirements. (A) To maintain the water vapor, and dissolved organic compounds, in the raw exhaust emissions stream, a manufacturer/tester will initially dilute one part emissions with a minimum of five parts clean, filtered air (see Hinners, et al., 1979 in paragraph (f)(11) of this section). Depending on the water vapor content of a particular fuel/additive mixture's combustion emissions and the humidity of the dilution air, initial exhaust dilutions as high as 1:15 or 1:20 may be necessary to maintain the general character of the exhaust as it cools, e.g., M100. At this point, it is expected that the exhaust stream would be further diluted to more appropriate levels for rodent health effects testing.

(B) A maximum concentration (minimum dilution) of the raw exhaust going into the test animal cages is anticipated to lie in the range between 1:5 and 1:50 exhaust emissions to clean, filtered air. The minimum concentration (maximum dilution) of raw exhaust for health effects testing is anticipated to be in range between 1:100 and 1:150. Individual manufacturers will treat these ranges as approximations only and will determine the optimum range of emission concentrations to elicit effects in Tier 2 health testing for their particular fuel/fuel additive mixture.

(3) Exposure chamber systems--(i) Referenced Guidelines. (A) The U.S. Department of Health and Human Services ``Guide for the Care and Use of Laboratory Animals" (Guide), 1985 cited in paragraph (c)(3)(ii)(A)(4), and in paragraphs (d)(2)(i), (d)(2)(ii), (d)(2)(iii), (d)(4)(ii), and (d)(4)(iii) of this section, has been incorporated by reference.

(B) This incorporation by reference was approved by the Director of the Federal Register in accordance with 5 U.S.C. 552(a) and 1 CFR part 51. Copies may be purchased from the Superintendent of Documents, U.S. Government Printing Office, Washington, DC 20402. Copies may be inspected at U.S. EPA, OAR, 401 M Street SW., Washington, DC, 20460 or at the Office of the Federal Register, 800 North Capitol Street NW., suite 700, Washington, DC.

(ii) Exposure chambers. There are two basic types of dynamic inhalation exposure chambers, whole-body chambers and nose-/head-only exposure chambers (see Cheng and Moss, 1989 in paragraph (f)(8) of this section).

(A) Whole-body chambers. (1) The flow rate through a chamber shall be maintained at 15 air changes per hour.

(2) The chambers are usually maintained at a slightly negative pressure (0.5 to 1.5 inch of water) to prevent leakage of test substance into the exposure room.

(3) The exposure chamber shall be designed in such a way as to provide uniform distribution of exposure concentrations in all compartments (see Cheng et al., 1989 in paragraph (f)(7) of this section).

(4) Animals are housed in separate compartments inside the chamber, where the whole surface area of an animal is exposed to the test material. The spaces required for different animal species shall follow the Guide. In general, the volume of animal bodies occupy less than 5 percent of the chamber volume.

(B) Head/nose-only exposure chambers. (1) In head/nose-only exposure chambers, only the head (oronasal) portion of the animal is exposed to the test material.

(2) The chamber volume and flow rates are much less than in the whole-body exposure chambers because the subjects are usually restrained in a tube holder where the animal's breathing can be easily monitored. The head/nose-only exposure chamber is suitable for short-term exposures or when use of a small amount of test material is required.

(iii) Since whole-body exposure appears to be the least stressful mode of exposure, it is the preferred method. In general, head/nose only exposure, which is sometimes used to avoid concurrent exposure by the dermal or oral routes, i.e., grooming, is not recommended because of the stress accompanying the restraining of the animals. However,

there may be specific instances where it may be more appropriate than whole-body exposure. The tester shall provide justification for its selection.

(d) Inhalation exposure procedures--(1) Animal selection. (i) The rat is the preferred species for vehicle emission inhalation health effects testing. Commonly used laboratory strains shall be used. Any rodent species may be used, but the tester shall provide justification for the choice of that species.

(ii) Young adult animals, approximately ten weeks of age for the rat, shall be used. At the commencement of the study, the weight variation of animals used shall not exceed <plus-minus>20 percent of the mean weight for each sex. Animals shall be randomly assigned to treatment and control groups according to their weight.

(iii) An equal number of male and female rodents shall be used at each concentration level. Situations may arise where use of a single sex may be appropriate. Females, in general, shall be nulliparous and nonpregnant.

(iv) The number of animals used at each concentration level and in the control group(s) depends on the type of study, number of biological end points used in the toxicity evaluation, the pre-determined sensitivity of detection and power of significance of the study, and the animal species. For an acute study, at least five animals of each sex shall be used in each test group. For both the subacute and subchronic studies, at least 10 rodents of each sex shall be used in each test group. For a chronic study, at least 20 male and 20 female rodents shall be used in each test group.

(A) If interim sacrifices are planned, the number of animals shall be increased by the number of animals scheduled to be sacrificed during the course of the study.

(B) For a chronic study, the number of animals at the termination of the study must be adequate for a meaningful and valid statistical evaluation of chronic effects.

(v) A concurrent control group is required. This group shall be exposed to clean, filtered air under conditions identical to those used for the group exposed to the test atmosphere.

(vi) The same species/strain shall be used to make comparisons between fuel-only and fuel/additive mixture studies. If another species/strain is used, the tester shall provide justification for its selection.

(2) Animal handling and care. (i) A key element in the conduct of inhalation exposure studies is the proper handling and care of the test animal population. Therefore, the exposure conditions must conform strictly with the conditions for housing and animal care and use set forth in the Guide.

(ii) In whole-body exposure chambers, animals shall be housed in

individual caging. The minimum cage size per animal will be in accordance with instructions set forth in the Guide.

(iii) Chambers shall be cleaned and maintained in accordance with recommendations and schedules set forth in the Guide.

(A) Observations shall be made daily with appropriate actions taken to minimize loss of animals to the study (e.g., necropsy or refrigeration of animals found dead and isolation or sacrifice of weak or moribund animals). Exposure systems using head/nose-only exposure chambers require no special daily chamber maintenance. Chambers shall be inspected to ensure that they are clean, and that there are no obstructions in the chamber which would restrict air flow to the animals. Whole-body exposure chambers will be inspected on a minimum of twice daily, once before exposures and once after exposures.

(B) Signs of toxicity shall be recorded as they are observed, including the time of onset, degree, and duration.

(C) Cage-side observations shall include, but are not limited to: changes in skin, fur, eye and mucous membranes, respiratory, autonomic, and central nervous systems, somatomotor activity, and behavioral patterns. Particular attention shall be directed to observation of tremors, convulsions, salivation, diarrhea, lethargy, sleep, and coma.

(iv) Food and water will be withheld from animals for head/nose-only exposure systems. For whole-body-exposure systems, water only may be provided. When the exposure generation system is not operating, food will be available ad libitum. During operation of the generation system, food will be withheld to avoid possible contamination by emissions.

(v) At the end of the study period, all survivors in the main study population shall be sacrificed. Moribund animals shall be removed and sacrificed when observed.

(3) Concentration levels and selection. (i) In acute and subacute toxicity tests, at least three exposure concentrations and a control group shall be used and spaced appropriately to produce test groups with a range of toxic effects and mortality rates. The data shall be sufficient to produce a concentration-response curve and permit an acceptable estimation of the median lethal concentration.

(ii) In subchronic and chronic toxicity tests, testers shall use at least three different concentration levels, with a control exposure group, to determine a concentration-response relationship. Concentrations shall be spaced appropriately to produce test groups with a range of toxic effects. The concentration-response data may also be sufficient to determine a NOAEL, unless the result of a limit test precludes such findings. The criteria for selecting concentration levels has been published (40 CFR 798.2450 and 798.3260).

(A) The highest concentration shall result in toxic effects but not produce an incidence of fatalities which would prevent a meaningful

evaluation of the study.

(B) The lowest concentration shall not produce toxic effects which are directly attributable to the test exposure. Where there is a useful estimation of human exposure, the lowest concentration shall exceed this.

(C) The intermediate concentration level(s) shall produce minimal observable toxic effects. If more than one intermediate concentration level is used, the concentrations shall be spaced to produce a gradation of toxic effects.

(D) In the low, intermediate, and control exposure groups, the incidence of fatalities shall be low to absent, so as not to preclude a meaningful evaluation of the results.

(4) Exposure chamber environmental conditions. The following environmental conditions in the exposure chamber are critical to the maintenance of the test animals: flow; temperature; relative humidity; lighting; and noise.

(i) Filtered and conditioned air shall be used during exposure, to dilute the exhaust emissions, and during non- exposure periods to maintain environmental conditions that are free of trace gases, dusts, and microorganisms on the test animals. Twelve to fifteen air changes per hour will be provided at all times to whole-body-exposure chambers. The minimum air flow rate for head/nose-only exposure chambers will be a function of the number of animals and the average minute volume of the animals:

Q<INF>minimum(L/min)=2 x number of animals x average minute volume

(see Cheng and Moss, 1989 in paragraph (f)(8) of this section).

(ii) Recommended ranges of temperature for various species are given in the Guide. The recommended temperature ranges will be used for establishing temperature conditions of whole-body- exposure chambers. For rodents in whole-body-exposure chambers, the recommended temperature is 22 deg.C +/- 2 deg.C and for rabbits, it is 20 deg.C +/- 3 deg.C. Temperature ranges have not been established for head/ nose-only tubes; however, recommended maximum temperature limits have been established at the Inhalation Toxicology Research Institute (see Barr, 1988 in paragraph (f)(1) of this section). Maximum temperature for rats and mice in head/nose-only tubes is 23 deg.C.

(iii) Relative humidity. The relative humidity in the chamber air is important for heat balance and shall be maintained between 40 percent and 60 percent, but in certain instances, this may not be practicable. Testers shall follow Guide recommends for a 30 percent to 70 percent relative humidity range for rodents in exposure chambers.

(iv) Lighting. Light intensity of 30 foot candles at 3 ft. from the floor of the exposure facility is recommended (see Rao, 1986 in

paragraph (f)(16) of this section).

(5) Exposure Conditions. Study animals shall be exposed to the test atmosphere on a repeated basis for at least 6 hours per day on a 7-day per week basis for the exposure period. However, based primarily on practical considerations, exposure on a 5-day-per-week basis for a minimum of 6 hours per day is the minimum acceptable exposure period.

(6) Exposure atmosphere. (i) The exposure atmosphere shall be held as constant as is practicable and must be monitored continuously or intermittently, depending on the method of analysis, to ensure that exposure levels are at the target values or within stated limits during the exposure period. Sampling methodology will be determined based on the type of generation system and the type of exposure chamber system specified for the exposure study.

(A) Integrated samples of test atmosphere aerosol shall be taken daily during the exposure period from a single representative sample port in the chamber near the breathing zone of the animals. Gas samples shall be taken daily to determine concentrations (ppm) of the major vapor components of the test atmosphere including CO, CO<INF>2, NO<INF>X, SO<INF>2, and total hydrocarbons.

(B) To ensure that animals in different locations of the chamber receive a similar exposure atmosphere, distribution of an aerosol or vapor concentration in exposure chambers can be determined without animals during the developmental phase of the study, or it can be determined with animals early in the study. For head/nose-only exposure chambers, it may not be possible to monitor the chamber distribution during the exposure, because the exposure port contains the animal.

(C) During the development of the emissions generation system, particle size analysis shall be performed to establish the stability of an aerosol concentration with respect to particle size. Over the course of the exposure, analysis shall be conducted as often as is necessary to determine the consistency of particle size distribution.

(D) Chamber rise and fall times. The rise time required for the exposure concentration to reach 90 percent of the stable concentration after the generator is turned on, and the fall time when the chamber concentration decreases to 10 percent of the stable concentration after the generation system is stopped shall be determined in the developmental phase of the study. Time-integrated samples collected for calculating exposure concentrations shall be taken after the rise time. The daily exposure time is exclusive of the rise or the fall time.

(ii) Instrumentation used for a given study will be determined based on the type of generation system and the type of exposure chamber system specified for the exposure study.

(A) For exhaust studies, combustion gases shall be sampled by collecting exposure air in bags and then analyzing the collected air sample to determine major components of the combustion gas using gas

analyzers. Exposure chambers can also be connected to gas analyzers directly by using sampling lines and switching valves. Samples can be taken more frequently using the latter method. Aerosol instruments, such as photometers, or time-integrated gravimetric determination may be used to determine the stability of any aerosol concentration in the chamber.

(B) For evaporative emission studies, concentration of fuel vapors can usually be determined by using a gas chromatograph (GC) and/or infrared (IR) spectrometry. Grab samples for intermittent sampling can be taken from the chamber by using bubble samplers with the appropriate solvent to collect the vapors, or by collecting a small volume of air in a syringe. Intermediate or continuous monitoring of the chamber concentration is also possible by connecting the chamber with a GC or IR detector.

(7) Monitoring chamber environmental conditions may be performed by a computer system or by exposure system operating personnel.

(i) The flow-metering device used for the exposure chambers must be a continuous monitoring device, and actual flow measurements must be recorded at least every 30 minutes. Accuracy must be <plus-minus>5 percent of full scale range. Measurement of air flow through the exposure chamber may be accomplished using any device that has sufficient range to accurately measure the air flow for the given chamber. Types of flow metering devices include rotameters, orifice meters, venturi meters, critical orifices, and turbinemeters (see Benedict, 1984 in paragraph (f)(4) and Spitzer, 1984 in paragraph (f)(17) of this section).

(ii) Pressure. Pressure measurement may be accomplished using manometers, electronic pressure transducers, magnehelics, or similar devices (see Gillum, 1982 in paragraph (f)(10) of this section). Accuracy of the pressure device must be <plus-minus>5 percent of full scale range. Pressure measurements must be continuous and recorded at least every 30 minutes.

(iii) Temperature. The temperature of exposure chambers must be monitored continuously and recorded at least every 30 minutes. Temperature may be measured using thermometers, RTD's, thermocouples, thermistors, or other devices (see Benedict, 1984 in paragraph (f)(4) of this section). It is necessary to incorporate an alarm system into the temperature monitoring system. The exposure operators must be notified by the alarm system when the chamber temperature exceeds 26.7 deg.C (80 deg.F). The exposure must be discontinued and emergency procedures enacted to immediately reduce temperatures or remove test animals from high temperature environment when chamber temperatures exceed 29 deg.C. Accuracy of the temperature monitoring device will be <plus-minus>1 deg.C for the temperature range of 20-30 deg.C.

(iv) Relative humidity. The relative humidity of exposure chambers

must be monitored continuously and recorded at least every 30 minutes. Relative humidity may be measured using various devices (see Chaddock, 1985 in paragraph (f)(6) of this section).

(v) Lighting shall be measured quarterly, or once at the beginning, middle, and end of the study for shorter studies.

(vi) Noise level in the exposure chamber(s) shall be measured quarterly, or once at the beginning, middle, and end of the study for shorter studies.

(vii) Oxygen content is critical, especially in nose-only chamber systems, and shall be greater than or equal to 19 percent in the test cages. An oxygen sensor shall be located at a single position in the test chamber and a lower alarm limit of 18 percent shall be used to activate an alarm system.

(8) Safety procedures and requirements. In the case of potentially explosive test substance concentrations, care shall be taken to avoid generating explosive atmospheres.

(i) It is mandatory that the upper explosive limit (UEL) and lower explosive limit (LEL) for the fuel and/or fuel additive(s) that are being tested be determined. These limits can be found in the material safety data sheets (MSDS) for each substance and in various reference texts. The air concentration of the fuel or additive-base fuel mixture in the generation system, dilution/delivery system, and the exposure chamber system shall be calculated to ensure that explosive limits are not present.

(ii) Storage, handling, and use of fuels or fuel/additive mixtures shall follow guidelines given in 29 CFR 1910.106.

(iii) Monitoring for carbon monoxide (CO) levels is mandatory for combustion systems. CO shall be continuously monitored in the immediate area of the engine/vehicle system and in the exposure chamber(s).

(iv) Air samples shall be taken quarterly in the immediate area of the vapor generation system and the exposure chamber system, or once at the beginning, middle, and end of the study for shorter studies. These samples shall be analyzed by methods described in paragraph (d)(6)(ii)(B) of this section.

(v) With the presence of fuels and/or fuel additives, all electrical and electronic equipment must be grounded. Also, the dilution/delivery system and chamber exposure system must be grounded. Guidelines for grounding are given in 29 CFR 1910.304.

(9) Quality control and quality assurance procedures--(i) Standard operating procedures (SOPs). SOPs for exposure operations, sampling instruments, animal handling, and analytical methods shall be written during the developmental phase of the study.

(ii) Technicians/operators shall be trained in exposure operation, maintenance, and documentation, as appropriate, and their training shall be documented.

(iii) Flow meters, sampling instruments, and balances used in the inhalation experiments shall be calibrated with standards during the developmental phase to determine their sensitivity, detection limits, and linearity. During the exposure period, instruments shall be checked for calibration and documented to ensure that each instrument still functions properly.

(iv) The mean exposure concentration shall be within 10 percent of the target concentration on 90 percent or more of exposure days. The coefficient of variation shall be within 25 percent of target on 90 percent or more of exposure days. For example, a manufacturer might determine a mean exposure concentration of its product's exposure emissions by identifying ``marker'' compound(s) typical of the emissions of the fuel or fuel/additive mixture under study as a surrogate for the total of individual compounds in those exposure emissions. The manufacturer would note any concentration changes in the level of the ``marker'' compound(s) in the sample's daily emissions for biological testing.

(v) The spatial variation of the chamber concentration shall be 10 percent, or less. If a higher spatial variation is observed during the developmental phase, then air mixing in the chamber shall be increased. In any case, animals shall be rotated among the various cages in the exposure chamber(s) to insure each animal's uniform exposure during the study.

(e) Data and reporting. Data shall be summarized in tabular form, showing for each group the number of animals at the start of the test, the number of animals showing lesions, the types of lesions, and the percentage of animals displaying each type of lesion.

(1) Treatment of results. All observed results, quantitative and incidental, shall be evaluated by an appropriate statistical method. Any generally accepted statistical method may be used; the statistical methods shall be selected during the design of the study.

(2) Evaluation of results. The findings of an inhalation toxicity study should be evaluated in conjunction with the findings of preceding studies and considered in terms of the observed toxic effects and the necropsy and histopathological findings. The evaluation will include the relationship between the concentration of the test atmosphere and the duration of exposure, and the severity of abnormalities, gross lesions, identified target organs, body weight changes, effects on mortality and any other general or specific toxic effects.

(3) Test conditions. (i) The exposure apparatus shall be described, including:

(A) The vehicle/engine design and type, the dynamometer, the cooling system, if any, the computer control system, and the dilution system for exhaust emission generation;

(B) The evaporative emissions generator model, type, or design and

its dilution system; and

(C) Other test conditions, such as the source and quality of mixing air, fuel or fuel/additive mixture used, treatment of exhaust air, design of exposure chamber and the method of housing animals in a test chamber shall be described.

(ii) The equipment for measuring temperature, humidity, particulate aerosol concentrations and size distribution, gas analyzers, fuel vapor concentrations, chamber distribution, and rise and fall time shall be described.

(iii) Daily exposure results. The daily record shall document the date, the start and stop times of the exposure, number of samples taken during the day, daily concentrations determined, calibration of instruments, and problems encountered during the exposure. The daily exposure data shall be signed by the exposure operator and reviewed and signed by the exposure supervisor responsible for the study.

(4) Exposure data shall be tabulated and presented with mean values and a measure of variability (e.g., standard deviation), and shall include:

(i) Airflow rates through the inhalation equipment;

(ii) Temperature and humidity of air;

(iii) Chamber concentrations in the chamber breathing zone;

(iv) Concentration of combustion exhaust gases in the chamber breathing zone;

(v) Particle size distribution (e.g., mass median aerodynamic diameter and geometric standard deviation from the mean);

(vi) Rise and fall time;

(vii) Chamber concentrations during the non-exposure period; and

(viii) Distribution of test substance in the chamber.

(5) Animal data. Tabulation of toxic response data by species, strain, sex and exposure level for:

(i) Number of animals exposed;

(ii) Number of animals showing signs of toxicity; and

(iii) Number of animals dying.

(f) References. For additional background information on this exposure guideline, the following references should be consulted.

(1) Barr, E.B. (1988) Operational Limits for Temperature and Percent Oxygen During HM Nose-Only Exposures--Emergency Procedures [interoffice memorandum]. Albuquerque, NM: Lovelace Inhalation Toxicology Research Institute; May 13.

(2) Barr, E.B.; Cheng, Y.S.; Mauderly, J.L. (1990) Determination of Oxygen Depletion in a Nose-Only Exposure Chamber. Presented at: 1990 American Association for Aerosol Research; June; Philadelphia, PA: American Association for Aerosol Research; abstract no. P2e1.

(3) Barrow, C.S. (1989) Generation and Characterization of Gases

and Vapors. In: McClellan, R.O., Henderson, R.F. ed. Concepts in Inhalation Toxicology. New York, NY: Hemisphere Publishing Corp., 63-84.

(4) Benedict, R.P. (1984) Fundamentals of Temperature, Pressure, and Flow Measurements. 3rd ed. New York, NY: John Wiley and Sons.

(5) Cannon, W.C.; Blanton, E.F.; McDonald, K.E. The Flow-Past Chamber. (1983) An Improved Nose-Only Exposure System for Rodents. Am. Ind. Hyg. Assoc. J. 44: 923-928.

(6) Chaddock, J.B. ed. (1985) Moisture and humidity. Measurement and Control in Science and Industry: Proceedings of the 1985 International Symposium on Moisture and Humidity; April 1985; Washington, D.C. Research Triangle Park, NC: Instrument Society of America.

(7) Cheng, Y.S.; Barr, E.B.; Carpenter, R.L.; Benson, J.M.; Hobbs, C.H. (1989) Improvement of Aerosol Distribution in Whole-Body Inhalation Exposure Chambers. Inhal. Toxicol. 1: 153-166.

(8) Cheng,Y.S.; Moss, O.R. (1989) Inhalation Exposure Systems. In: McClellan, R.O.; Henderson, R.F. ed. Concepts in Inhalation Toxicology. New York, NY: Hemisphere Publishing Corp., 19-62.

(9) Cheng, Y.S.; Yeh, H.C.; Mauderly, J.L.; Mokler, B.V. (1984) Characterization of Diesel Exhaust in a Chronic Inhalation Study. Am. Ind. Hyg. Assoc. J. 45: 547-555.

(10) Gillum, D.R. (1982) Industrial Pressure Measurement. Research Triangle Park, NC: Instrument Society of America.

(11) Hinners, R.G.; Burkart, J.K.; Malanchuk, M. (1979) Animal Exposure Facility for Diesel Exhaust Studies.

(12) Kittelson, D.B.; Dolan, D.F. (1979) Diesel exhaust aerosols. In Willeke, K. ed. Generation of Aerosols and Facilities for Exposure Experiments. Ann Arbor, MI: Ann Arbor Science Publishers Inc., 337-360.

(13) Mokler, B.V.; Archibeque, F.A.; Beethe, R.L.; Kelly, C.P.J.; Lopez, J.A.; Mauderly, J.L.; Stafford, D.L. (1984) Diesel Exhaust Exposure System for Animal Studies. Fundamental and Applied Toxicology 4: 270-277.

(14) Moore, W.; et al. (1978) Preliminary finding on the Deposition and Retention of Automotive Diesel Particulate in Rat Lungs. Proc. of Annual Meeting of the Air Pollution Control Assn, 3, paper 78-33.7.

(15) Raabe, O.G., Bennick, J.E., Light, M.E., Hobbs, C.H., Thomas, R.L., Tillery, M.I. (1973) An Improved Apparatus for Acute Inhalation Exposure of Rodents to Radioactive Aerosols. Toxicol & Applied Pharmaco.; 1973; 26: 264-273.

(16) Rao, G.N. (1986) Significance of Environmental Factors on the Test System. In: Hoover, B.K.; Baldwin, J.K.; Uelner, A.F.; Whitmire, C.E.; Davies, C.L.; Bristol, D.W. ed. Managing conduct and

data quality of toxicology studies. Raleigh, NC: Princeton Scientific Publishing Co., Inc.: 173-185.
(17) Spitzer, D.W. (1984) Industrial Flow Measurement. Research Triangle Park, NC: Instrument Society of America.
(18) 40 CFR part 798, Health effects testing guidelines.
(19) 29 CFR part 1910, Occupational safety and health standards for general industry.
(20) Federal Register, 42 FR 26748, May 25, 1977.

Sec. 79.62 Subchronic toxicity study with specific health effect assessments.

(a) Purpose--(1) General toxicity. This subchronic inhalation study is designed to determine a concentration-response relationship for potential toxic effects in rats resulting from continuous or repeated inhalation exposure to vehicle/engine emissions over a period of 90 days. A subgroup of perfusion-fixed animals is required, in addition to the main study population, for more exacting organ and tissue histology. This test will provide screening information on target organ toxicities and on concentration levels useful for running chronic studies and establishing exposure criteria. Initial information on effective concentrations/exposures of the test atmosphere may be determined from the literature of previous studies or through concentration range-finding trials prior to starting this study. This health effects screening test is not capable of directly determining those effects which have a long latency period for development (e.g., carcinogenicity and life-shortening), though it may permit the detremination of a no-observed-adverse-effect level, or NOAEL.
(2) Specific health effects assessments (HEAs). These supplemental studies are designed to determine the potential for reproductive/ teratologic, carcinogenic, mutagenic, and neurotoxic health effect outcomes from vehicle/engine emission exposures. They are done in combination with the subchronic toxicity study and paragraph (c) of this section or may be done separately as outlined by the appropriate test guideline.
(i) Fertility assessment/teratology. The fertility assessment is an in vivo study designed to provide information on potential health hazards to the fetus arising from the mother's repeated exposure to vehicle/engine emissions before and during her pregnancy. By including a mating of test animals, the study provides preliminary data on the effects of repeated vehicle/engine emissions exposure on gonadal function, conception, and fertility. The fertility assessment/ teratology guideline is found in Sec. 79.63.
(ii) Micronucleus (MN) Assay. The MN assay is an in vivo

cytogenetic test which gives information on potential carcinogenic and/or mutagenic effects of exposure to vehicle/engine emissions. The MN assay detects damage to the chromosomes or mitotic apparatus of cells in the tissues of a test subject exposed repeatedly to vehicle/engine emissions. The assay is based on an increase in the frequency of micronucleated erythrocytes found in bone marrow from treated animals compared to that of control animals. The guideline for the MN assay is found in Sec. 79.64.

(iii) Sister Chromatid Exchange (SCE) Assay. The SCE assay is an in vivo analysis which gives information on potential mutagenic and/or carcinogenic effects of exposure to vehicle/engine emissions. The assay detects the ability of a chemical to enhance the exchange of DNA between two sister chromatids of a duplicating chromosome. This assay uses peripheral blood lymphocytes isolated from an exposed rodent test species and grown to confluence in cell culture. The guideline for the SCE assay is found in Sec. 79.65.

(iv) Neurotoxicity (NTX) measures. NTX measures include (A) histopathology of specified central and peripheral nervous system tissues taken from emission-exposed rodents, and (B) an assay of brain tissue levels of glial fibrillary acidic protein (GFAP), a major filament protein of astrocytes, from emission-exposed rodents. The guidelines for the neurohistopathology and GFAP studies are found in Sec. 79.66 and Sec. 79.67, respectively.

(b) Definitions. For the purposes of this section, the following definitions apply:

No-observed-adverse-effect-level (NOAEL) means the maximum concentration used in a test which produces no observed adverse effects. A NOAEL is expressed in terms of weight or volume of test substance given daily per unit volume of air (<greek-m>g/L or ppm).

Subchronic inhalation toxicity means the adverse effects occurring as a result of the continuous or repeated daily exposure of experimental animals to a chemical by inhalation for part (approximately 10 percent) of a life span.

(c) Principle of the test method. As long as none of the requirements of any study are violated by the combination, one or more HEAs may be combined with the general toxicity study through concurrent exposures of their study populations and/or by sharing the analysis of the same animal subjects. Requirements duplicated in combined studies need not be repeated. Guidelines for combining HEAs with the general toxicity study are as follows.

(1) Fertility assessment. (i) The number of study animals in the test population is increased when the fertility assessment is run concurrently with the 90-day toxicity study. A minimum of 40 females per test group shall undergo vaginal lavage daily for two weeks before the start of the exposure period. The resulting wet smears are examined

to cull those animals which are acyclic. Twenty-five females shall be randomly assigned to a for-breeding group with the balance of females assigned to a group for histopathologic examination.

(ii) All test groups are exposed over a period of 90 days to various concentrations of the test atmosphere for a minimum of six hours per day. After seven weeks of exposures, analysis of vaginal cell smears shall resume on a daily basis for the 25 for-breeding females and shall continue for a period of four weeks or until each female in the group is confirmed pregnant. Following the ninth week of exposures, each for-breeding female is housed overnight with a single study male. Matings shall continue for as long as two weeks, or until pregnancy is confirmed (pregnancy day 0). Pregnant females are only exposed through day 15 of their pregnancy while daily exposures continue throughout the course of the study for non-pregnant females and study males.

(iii) On pregnancy day 20, pregnant females are sacrificed and their uteri are examined. Pregnancy status and fetal effects are recorded as described in Sec. 79.63. At the end of the exposure period, all males and non-pregnant females are sacrificed and necropsied. Testes and epididymal tissue samples are taken from five perfusion-fixed test subjects and histopathological examinations are carried out on the remainder of the non-pregnant females and study males.

(2) Carcinogenicity/mutagenicity (C/M) assessment. When combined with the subchronic toxicity study, the main study population is used to perform both the in vivo MN and SCE assays. Because of the constant turnover of the cells to be analyzed in these assays, a separate study population may be used for this assessment. A study population needs only to be exposed a minimum of four weeks. At exposure's end, ten animals per exposure and control groups are anaesthetized and heart punctures are performed on all members. After separating blood components, individual lymphocyte cell cultures are set up for SCE analysis. One femur from each study subject is also removed and the marrow extracted. The marrow is smeared onto a glass slide, and stained for analysis of micronuclei in erythrocytes.

(3) Neurotoxicity (NTX) measures. (i) When combined with this subchronic toxicity study, test animals designated for whole-body perfusion fixation/lung histology and exposed as part of the main animal population are used to perform the neurohistology portion of these measures. After the last exposure period, a minimum of ten animals from each exposure group shall be preserved in situ with fixative. Sections of brain, spinal cord, and proximal sciatic or tibial nerve are then cut, processed further in formalin, and mounted for viewing under a light microscope. Fibers from the sciatic or tibial nerve sample are teased apart for further analysis under the microscope.

(ii) GFAP assay. After the last exposure period, a minimum of ten

rodents from each exposure group shall be sacrificed, and their brains excised and divided into regions. The tissue samples are then applied to filter paper, washed with anti-GFAP antibody, and visualized with a radio-labelled Protein A. The filters are quantified for degree of immunoreactivity between the antibody and GFAP in the tissue samples. A non-radioactive ELISA format is also referenced in the GFAP guideline cited in paragraph (a)(2)(iv) of this section. Note: Because the GFAP assay requires fresh, i.e., non-preserved, brain tissue, the number of test animals may need to be increased to provide an adequate number of test subjects to complete the histopathology requirements of both the GFAP and the general toxicity portion of the 90-day inhalation study.

(iii) The start of the exposure period for the NTX measures study population may be staggered from that of the main study group to more evenly distribute the analytical work required in both study populations. The exposures would remain the same in all other respects.

(d) Test procedures--(1) Animal selection--(i) Species and sex. The rat is the recommended species. If another rodent species is used, the tester shall provide justification for its selection. Both sexes shall be used in any assessment unless it is demonstrated that one sex is refractory to the effects of exposure.

(ii) Age and number. Rats shall be at least ten weeks of age at the beginning of the study exposure. The number of animals necessary for individual health effect outcomes is as follows:

(A) Thirty rodents per concentration level/group, fifteen of each sex, shall be used to satisfy the reporting requirements of the 90-day toxicity study. Ten animals per concentration level/group shall be designated for whole body perfusion with fixative (by gravity) for lung studies, and neurohistology and testes studies, as appropriate.

(B) Forty rodents, 25 females and ten males shall be added for each test concentration or control group when combining a 90-day toxicity study with a fertility assessment.

(C) The tester shall provide a group of 10 animals (five animals per sex per experimental/control groups) in addition to the main test population when performing the GFAP neurotoxicity HEA.

(2) Recovery group. The manufacturer shall include an group of 20 animals (10 animals per sex) in the test population, exposing them to the highest concentration level for the entire length of the study's exposure period. This group shall then be observed for reversibility, persistence, or delayed occurrence of toxic effects during a post-exposure period of not less than 28 days.

(3) Inhalation exposure. (i) All data developed within this study shall be in accordance with good laboratory practice provisions under Sec. 79.60.

(ii) The general conduct of this study shall be in accordance with the vehicle emissions inhalation exposure guideline in Sec. 79.61.

(4) Observation of animals. (i) All toxicological (e.g., weight loss) and neurological signs (e.g., motor disturbance) shall be recorded frequently enough to observe any abnormality, and not less than weekly for all study animals. Animals shall be weighed weekly.

(ii) The following is a minimal list of measures that shall be noted:

(A) Body weight;

(B) Subject's reactivity to general stimuli such as removal from the cage or handling;

(C) Description, incidence, and severity of any convulsions, tremors, or abnormal motor movements in the home cage;

(D) Descriptions and incidence of posture and gait abnormalities observed in the home cage;

(E) Description and incidence of any unusual or abnormal behaviors, excessive or repetitive actions (stereotypies), emaciation, dehydration, hypotonia or hypertonia, altered fur appearance, red or crusty deposits around the eyes, nose, or mouth, and any other observations that may facilitate interpretation of the data.

(iii) Any animal which dies during the test is necropsied as soon as possible after discovery.

(5) Clinical examinations. (i) The following examinations shall be performed on the twenty animals designated as the 90-day study population, exclusive of pregnant dams and those study animals targeted for perfusion by gravity:

(A) The following hematology determinations shall be carried out at least two times during the test period (after 30 days of exposure and just prior to terminal sacrifice at the end of the exposure period): hematocrit, hemoglobin concentration, erythrocyte count, total and differential leukocyte count, and a measure of clotting potential such as prothrombin time, thromboplastin time, or platelet count.

(B) Clinical biochemistry determinations on blood shall be carried out at least two times during the test period, after 30 days of exposure and just prior to terminal sacrifice at the end of the exposure period, on all groups of animals including concurrent controls. Clinical biochemical testing shall include assessment of electrolyte balance, carbohydrate metabolism, and liver and kidney function. The selection of specific tests will be influenced by observations on the mode of action of the substance. In the absence of more specific tests, the following determinations may be made: calcium, phosphorus, chloride, sodium, potassium, fasting glucose (with period of fasting appropriate to the species), serum alanine aminotransferase, serum aspartate aminotransferase, sorbitol dehydrogenase, gamma glutamyl transpeptidase, urea nitrogen, albumen, blood creatinine, methemoglobin, bile acids, total bilirubin, and total serum protein measurements. Additional clinical biochemistry shall be employed, where

necessary, to extend the investigation of observed effects, e.g., analyses of lipids, hormones, acid/base balance, and cholinesterase activity.

(ii) The following examinations shall initially be performed on the high concentration and control groups only:

(A) Ophthalmological examination, using an ophthalmoscope or equivalent suitable equipment, shall be made prior to exposure to the test substance and at the termination of the study. If changes in the eyes are detected, all animals shall be examined.

(B) Urinalysis is not required on a routine basis, but shall be done when there is an indication based on expected and/or observed toxicity.

(iii) Preservation by whole-body perfusion of fixative into the anaesthetized animal for lung histology of ten animals from the 90-day study population for each experimental and control group.

(6) Gross pathology. With the exception of the whole body perfusion-fixed test animals cited in paragraph (d)(1)(ii)(A) of this section, all rodents shall be subjected to a full gross necropsy which includes examination of the external surface of the body, all orifices and the cranial, thoracic, and abdominal cavities and their contents. Gross pathology shall be performed on the following organs and tissues:

(i) The liver, kidneys, lungs, adrenals, brain, and gonads, including uterus, ovaries, testes, epididymides, seminal vesicles (with coagulating glands), and prostate, constitute the group of target organs for histology and shall be weighed as soon as possible after dissection to avoid drying. In addition, for other than rodent test species, the thyroid with parathyroids, when present, shall also be weighed as soon as possible after dissection to avoid drying.

(ii) The following organs and tissues, or representative samples thereof, shall be preserved in a suitable medium for possible future histopathological examination: All gross lesions; lungs--which shall be removed intact, weighed, and treated with a suitable fixative to ensure that lung structure is maintained (perfusion with the fixative is considered to be an effective procedure); nasopharyngeal tissues; brain--including sections of medulla/pons, cerebellar cortex, and cerebral cortex; pituitary; thyroid/parathyroid; thymus; trachea; heart; sternum with bone marrow; salivary glands; liver; spleen; kidneys; adrenals; pancreas; reproductive organs: uterus; cervix; ovaries; vagina; testes; epididymides; prostate; and, if present, seminal vesicles; aorta; (skin); gall bladder (if present); esophagus; stomach; duodenum; jejunum; ileum; cecum; colon; rectum; urinary bladder; representative lymph node; (mammary gland); (thigh musculature); peripheral nerve/tissue; (eyes); (femur--including articular surface); (spinal cord at three levels--cervical, midthoracic, and lumbar); and (zymbal and exorbital lachrymal glands).

(7) Histopathology. Histopathology shall be performed on the following organs and tissues from all rodents:

(i) All gross lesions.

(ii) Respiratory tract and other organs and tissues, listed in paragraph (d)(6)(ii) of this section (except organs/tissues in parentheses), of all animals in the control and high dose groups.

(iii) The tissues mentioned in parentheses, listed in paragraph (d)(6)(ii) of this section, if indicated by signs of toxicity or target organ involvement.

(iv) Lungs of animals in the low and intermediate dose groups shall also be subjected to histopathological examination, primarily for evidence of infection since this provides a convenient assessment of the state of health of the animals.

(v) Lungs and trachea of the whole-body perfusion-fixed test animals cited in paragraph (d)(1)(ii)(A) of this section are examined for inhaled particle distribution.

(e) Interpretation of results. All observed results, quantitative and incidental, shall be evaluated by an appropriate statistical method. The specific methods, including consideration of statistical power, shall be selected during the design of the study.

(f) Test report. In addition to the reporting requirements as specified under Secs. 79.60 and 79.61(e), the following individual animal data information shall be reported:

(1) Date of death during the study or whether animals survived to termination.

(2) Date of observation of each abnormal sign and its subsequent course.

(3) Individual body weight data, and group average body weight data vs. time.

(4) Feed consumption data, when collected.

(5) Hematological tests employed and all results.

(6) Clinical biochemistry tests employed and all results.

(7) Necropsy findings.

(8) Type of stain/fixative and procedures used in preparing tissue samples.

(9) Detailed description of all histopathological findings.

(10) Statistical treatment of the study results, where appropriate.

(g) References. For additional background information on this test guideline, the following references should be consulted.

(1) 40 CFR 798.2450, Inhalation toxicity.

(2) 40 CFR 798.2675, Oral Toxicity with Satellite Reproduction and Fertility Study.

(3) General Statement of Work for the Conduct of Toxicity and Carcinogenicity Studies in Laboratory Animals (revised April, 1987/ modifications through January, 1990) appendix G, National Toxicology

Program--U.S. Dept. of Health and Human Services (Public Health Service), P.O. Box 12233, Research Triangle Park, NC 27709.

Sec. 79.63 Fertility assessment/teratology.

(a) Purpose. Fertility assessment/teratology is an in vivo study designed to provide information on potential health hazards to the fetus arising from the mother's repeated inhalation exposure to vehicle/engine emissions before and during her pregnancy. By including a mating of test animals, the study provides preliminary data on the effects of repeated vehicle/engine emissions exposure on gonadal function, conception, and fertility. Since this is a one-generation test that ends with examination of full-term fetuses, but not of live pups, it is not capable of determining effects on reproductive development which would only be detected in viable offspring of treated parents.

(b) Definitions. For the purposes of this section, the following definitions apply:

Developmental toxicity means the ability of an agent to induce in utero death, structural or functional abnormalities, or growth retardation after contact with the pregnant animal.

Estrous cycle means the periodic recurrence of the biological phases of the female reproductive system which prepare the animal for conception and the development of offspring. The phases of the estrous cycle for a particular animal can be characterized by the general condition of the cells present in the vagina and the presence or absence of various cell types.

Vaginal cytology evaluation means the use of wet vaginal cell smears to determine the phase of a test animal's estrous cycle and the potential for adverse exposure effects on the regularity of the animal's cycle. In the rat, common cell types found in the smears correlate well with the various stages of the estrous cycle and to changes occurring in the reproductive tract.

(c) Principle of the test method. (1) For a two week period before exposures start, daily vaginal cell smears are examined from a surplus of female test animals to identify and cull those females which are acyclic. After culling, testers shall randomly assign at each exposure concentration (including unexposed) a minimum of twenty-five females for breeding and fifteen non-bred females for later histologic evaluation. Test animals shall be exposed by inhalation to graduated concentrations of the test atmosphere for a minimum of six hours per day over the next 13 weeks. Males and females in both test and control groups are mated after nine weeks of exposure. Exposures for pregnant females continue through gestation day 15, while exposures for males

and all non- pregnant females shall continue for the full exposure period.

(2) Beginning two weeks before the start of the mating period, daily vaginal smears resume for all to-be-bred females to characterize their estrous cycles. This will continue for four weeks or until a rat's pregnancy is confirmed, i.e., day 0, by the presence of sperm in the cell smear. On pregnancy day 20, shortly before the expected date of delivery, each pregnant female is sacrificed, her uterus removed, and the contents examined for embryonic or fetal deaths, and live fetuses. At the end of the exposure period, males and all non-pregnant females shall be weighed, and various organs and tissues, as appropriate, shall be removed and weighed, fixed with stain, and sectioned for viewing under a light microscope.

(3) This assay may be done separately or in combination with the subchronic toxicity study, pursuant to the provisions in Sec. 79.62.

(d) Limit test. If a test at one dose level of the highest concentration that can be achieved while maintaining a particle size distribution with a mass median aerodynamic diameter (MMAD) of 4 micrometers (<greek-m>m) or less, using the procedures described in section 79.60 of this part produces no observable toxic effects and if toxicity would not be expected based upon data of structurally related compounds, then a full study using three dose levels might not be necessary. Expected human exposure though may indicate the need for a higher dose level.

(e) Test procedures--(1) Animal selection--(i) Species and strain. The rat is the preferred species. Strains with low fecundity shall not be used and the candidate species shall be characterized for its sensitivity to developmental toxins. If another rodent species is used, the tester shall provide justification for its selection.

(ii) Animals shall be a minimum of 10 weeks old at the start of the exposure period.

(iii) Number and sex. Each test and control group shall have a minimum of 25 males and 40 females. In order to ensure that sufficient pups are produced to permit meaningful evaluation of the potential developmental toxicity of the test substance, twenty pregnant test animals are required for each exposure and control level.

(2) Observation period. The observation period shall be 13 weeks, at a minimum.

(3) Concentration levels and concentration selection. (i) To select the appropriate concentration levels, a pilot or trial study may be advisable. Since pregnant animals have an increased minute ventilation as compared to non-pregnant animals, it is recommended that the trial study be conducted in pregnant animals. Similarly, since presumably the minute ventilation will vary with progression of pregnancy, the animals should be exposed during the same period of gestation as in the main

study. It is not always necessary, though, to carry out a trial study in pregnant animals. Comparisons between the results of a trial study in non-pregnant animals, and the main study in pregnant animals will demonstrate whether or not the test substance is more toxic in pregnant animals. In the trial study, the concentration producing embryonic or fetal lethalities or maternal toxicity should be determined.

(ii) The highest concentration level shall induce some overt maternal toxicity such as reduced body weight or body weight gain, but not more than 10 percent maternal deaths.

(iii) The lowest concentration level shall not produce any grossly observable evidence of either maternal or developmental toxicity.

(4) Inhalation exposure. (i) All data developed within this study shall be in accordance with good laboratory practice provisions under Sec. 79.60.

(ii) The general conduct of this study shall be in accordance with the vehicle emissions inhalation exposure guideline in Sec. 79.61.

(f) Test performance--(1) Study conduct. Directions specific to this study are:

(i) The duration of exposure shall be at least six hours daily, allowing appropriate additional time for chamber equilibrium.

(ii) Where an exposure chamber is used, its design shall minimize crowding of the test animals. This is best accomplished by individual caging.

(iii) Pregnant animals shall not be subjected to beyond the minimum amount of stress. Since whole-body exposure appears to be the least stressful mode of exposure, it is the preferred method. In general oronasal or head-only exposure, which is sometimes used to avoid concurrent exposure by the dermal or oral routes, is not recommended because of the associated stress accompanying the restraining of the animals. However, there may be specific instances where it may be more appropriate than whole-body exposure. The tester shall provide justification/reasoning for its selection.

(iv) Measurements shall be made at least every other day of food consumption for all animals in the study. Males and females shall be weighed on the first day of exposure and 2-3 times per week thereafter, except for pregnant dams.

(v) The test animal housing, mating, and exposure chambers shall be operated on a twenty-four hour lighting schedule, with twelve hours of light and twelve hours of darkness. Test animal exposure shall only occur during the light portion of the cycle.

(vi) Signs of toxicity shall be recorded as they are observed including the time of onset, degree, and duration.

(vii) Females showing signs of abortion or premature delivery shall be sacrificed and subjected to a thorough macroscopic examination.

(viii) Animals that die or are euthanized because of morbidity will

be necropsied promptly.

(2) Vaginal cytology. (i) For a two week period before the mating period starts, each female in the to-be-bred population shall undergo a daily saline vaginal lavage. Two wet cell smears from this lavage shall be examined daily for each subject to determine a baseline pattern of estrus. Testers shall avoid excessive handling and roughness in obtaining the vaginal cell samples, as this may induce a condition of pseudo-pregnancy in the test animals.

(ii) This will continue for four weeks or until day 0 of a rat's pregnancy is confirmed by the presence of sperm in the cell smear.

(3) Mating and fertility assessment. (i) Beginning nine weeks after the start of exposure, each exposed and control group female (exclusive of the histology group females) shall be paired during non-exposure hours with a male from the same exposure concentration group. Matings shall continue for a period of two weeks, or until all mated females are determined to be pregnant. Mating pairs shall be clearly identified.

(ii) Each morning, including weekends, cages shall be examined for the presence of a sperm plug. When found, this shall mark gestation day 0 and pregnancy shall be confirmed by the presence of sperm in the day's wet vaginal cell smears.

(iii) Two weeks after mating is begun, or as females are determined to be pregnant, bred animals are returned to pre-mating housing. Daily exposures continues through gestation day 15 for all pregnant females or through the balance of the exposure period for non-pregnant females and all males.

(iv) Those pairs which fail to mate shall be evaluated in the course of the study to determine the cause of the apparent infertility. This may involve such procedures as additional opportunities to mate with a proven fertile partner, histological examination of the reproductive organs, and, in males, examination of the spermatogenic cycles. The stage of estrus for each non-pregnant female in the breeding group will be determined at the end of the exposure period.

(4) All animals in the histology group shall be subject to histopathologic examination at the end of the study's exposure period.

(g) Treatment of results. (1) All observed results, quantitative and incidental, shall be evaluated by an appropriate statistical method. The specific methods, including consideration of statistical power, shall be selected during the design of the study.

(2) Data and reporting. In addition to the reporting requirements specified under Secs. 79.60 and 79.61, the final test report must include the following information:

(i) Gross necropsy. (A) All animals shall be subjected to a full necropsy which includes examination of the external surface of the body, all orifices, and the cranial, thoracic, and abdominal cavities

and their contents. Special attention shall be directed to the organs of the reproductive system.

(B) The liver, kidneys, adrenals, pituitary, uterus, vagina, ovaries, testes, epididymides and seminal vesicles (with coagulating glands), and prostate shall be weighed wet, as soon as possible after dissection, to avoid drying.

(i) At the time of sacrifice on gestation day 20 or at death during the study, each dam shall be examined macroscopically for any structural abnormalities or pathological changes which may have influenced the pregnancy.

(ii) The contents of the uterus shall be examined for embryonic or fetal deaths and the number of viable fetuses. Gravid uterine weights need not be obtained from dead animals where decomposition has occurred. The degree of resorption shall be described in order to help estimate the relative time of death.

(iii) The number of corpora lutea shall be determined in each pregnant dam.

(iv) Each fetus shall be weighed, all weights recorded, and mean fetal weights determined.

(v) Each fetus shall be examined externally and the sex determined.

(vi) One-half of the rat fetuses in each litter shall be examined for skeletal anomalies, and the remaining half shall be examined for soft tissue anomalies, using appropriate methods.

(ii) Histopathology. (A) Histopathology on vagina, uterus, ovaries, testes, epididymides, seminal vesicles, and prostate as appropriate for all males and histology group females in the control and high concentration groups and for all animals that died or were euthanized during the study. If abnormalities or equivocal results are seen in any of these organs/tissues, the same organ/tissue from test animals in lower concentration groups shall be examined.

Note: Testes, seminal vesicles, epididymides, and ovaries, at a minimum, shall be examined in perfusion-fixed (pressure or gravity method) test subjects, when available.

(B) All gross lesions in all study animals shall be examined.

(C) As noted under mating procedures, reproductive organs of animals suspected of infertility shall be subject to microscopic examination.

(D) The following organs and tissues, or representative samples thereof, shall be preserved in a suitable medium for future histopathological examination: all gross lesions; vagina; uterus; ovaries; testes; epididymides; seminal vesicles; prostate; liver; and kidneys/adrenals.

(3) Evaluation of results. (i) The findings of a developmental

toxicity study shall be evaluated in terms of the observed effects and the exposure levels producing effects. It is necessary to consider the historical developmental toxicity data on the species/strain tested.

(ii) There are several criteria for determining a positive result for reproductive/teratologic effects; a statistically significant dose-related decrease in the weight of the testes for treated subjects over control subjects, a decrease in neonatal viability, a significant change in the presence of soft tissue or skeletal abnormalities, or an increased rate of embryonic or fetal resorption or death. Other criteria, e.g., lengthening of the estrous cycle or the time spent in any one stage of estrus, changes in the proportion of viable male vs female fetuses or offspring, the number and type of cells in vaginal smears, or pathologic changes found during gross or microscopic examination of male or female reproductive organs may be based upon detection of a reproducible and statistically significant positive response for that evaluation parameter. A positive result indicates that, under the test conditions, the test substance does induce reproductive organ or fetal toxicity in the test species.

(iii) A test substance which does not produce either a statistically significant dose-related change in the reproductive organs or cycle or a statistically significant and reproducible positive response at any one of the test points may not induce reproductive organ toxicity in this test species, but further investigation , e.g., to establish absorption and bioavailability of the test substance, should be considered.

(h) Test report. In addition to the reporting requirements as specified under 40 CFR 79.60 and the vehicle emissions inhalation toxicity guideline as published in 40 CFR 79.61, the following specific information shall be reported:

(1) Individual animal data. (i) Time of death during the study or whether animals survived to termination.

(ii) Date of onset and duration of each abnormal sign and its subsequent course.

(iii) Feed and body weight data.

(iv) Necropsy findings.

(v) Male test subjects.

(A) Testicle weight, and body weight: testicle weight ratio.

(B) Detailed description of all histopathological findings, especially for the testes and the epididymides.

(vi) Female test subjects.

(A) Uterine weight data.

(B) Beginning and ending collection dates for vaginal cell smears.

(C) Estrous cycle length compared within and between groups including mean cycle length for groups.

(D) Percentage of time spent in each stage of cycle.

(E) Stage of estrus at time of mating/sacrifice and proportion of females in estrus between concentration groups.

(F) Detailed description of all histopathological findings, especially for uterine/ovary samples.

(vii) Pregnancy and litter data. Toxic response data by exposure level, including but not limited to, indices of fertility and time-to-mating, including the number of days until mating and the number of full or partial estrous cycles until mating.

(A) Number of pregnant animals,

(B) Number and percentage of live fetuses, resorptions.

(viii) Fetal data. (A) Numbers of each sex.

(B) Number of fetuses with any soft tissue or skeletal abnormalities.

(2) Type of stain/fixative and procedures used in preparing tissue samples.

(3) Statistical treatment of the study results.

(i) References. For additional background information on this test guideline, the following references should be consulted.

(1) 40 CFR 798.2675, Oral Toxicity with Satellite Reproduction and Fertility Study.

(2) 40 CFR 798.4350, Inhalation Developmental Toxicity Study.

(3) Chapin, R.E. and J.J. Heindel (1993) Methods in Toxicology, Vol. 3, Parts A and B: Reproductive Toxicology, Academic Press, Orlando, FL.

(4) Gray, L.E., et al. (1989) ``A Dose-Response Analysis of Methoxychlor-Induced Alterations of Reproductive Development and Function in the Rat" Fund. App. Tox. 12, 92-108.

(5) Leblond, C.P. and Y. Clermont (1952) ``Definition of the Stages of the Cycle of the Seminiferous Epithelium of the Rat." Ann. N. Y. Acad. Sci. 55:548-73.

(6) Morrissey, R.E., et al. (1988) ``Evaluation of Rodent Sperm, Vaginal Cytology, and Reproductive Organ Weight Data from National Toxicology Program 13-week Studies." Fundam. Appl. Toxicol. 11:343-358.

(7) Russell, L.D., Ettlin, R.A., Sinhattikim, A.P., and Clegg, E.D (1990) Histological and Histopathological Evaluation of the Testes, Cache River Press, Clearwater, FL.

Sec. 79.64 In vivo micronucleus assay.

(a) Purpose. The micronucleus assay is an in vivo cytogenetic test which uses erythrocytes in the bone marrow of rodents to detect chemical damage to the chromosomes or mitotic apparatus of mammalian

cells. As the erythroblast develops into an erythrocyte (red blood cell), its main nucleus is extruded and may leave a micronucleus in the cell body; a few micronuclei form under normal conditions in blood elements. This assay is based on an increase in the frequency of micronucleated erythrocytes found in bone marrow from treated animals compared to that of control animals. The visualization of micronuclei is facilitated in these cells because they lack a main nucleus.

(b) Definitions. For the purposes of this section the following definitions apply:

Micronuclei mean small particles consisting of acentric fragments of chromosomes or entire chromosomes, which lag behind at anaphase of cell division. After telophase, these fragments may not be included in the nuclei of daughter cells and form single or multiple micronuclei in the cytoplasm.

Polychromatic erythrocyte (PCE) means an immature red blood cell that, because it contains RNA, can be differentiated by appropriate staining techniques from a normochromatic erythrocyte (NCE), which lacks RNA. In one to two days, a PCE matures into a NCE.

(c) Test method--(1) Principle of the test method. (i) Groups of rodents are exposed by the inhalation route for a minimum of 6 hours/day over a period of not less than 28 days to three or more concentrations of a test substance in air. Groups of animals are sacrificed at the end of the exposure period and femoral bone marrow is extracted. The bone marrow is then smeared onto glass slides, stained, and PCEs are scored for micronuclei. Researchers may need to run a trial at the highest tolerated concentration of the test atmosphere to optimize the sample collection time for micronucleated cells.

(ii) This assay may be done separately or in combination with the subchronic toxicity study, pursuant to the provisions in Sec. 79.62.

(2) Species and strain. (i) The rat is the recommended test animal. Other rodent species may be used in this assay, but use of that species will be justified by the tester.

(ii) If a strain of mouse is used in this assay, the tester shall sample peripheral blood from an appropriate site on the test animal, e.g., the tail vein, as a source of normochromatic erythrocytes. Results shall be reported as outlined later in this guideline with ``normochromatic'' interchanged for ``polychromatic'', where specified.

(3) Animal number and sex. At least five female and five male animals per experimental/sample and control group shall be used. The use of a single sex or a smaller number of animals shall be justified.

(4) Positive control group. A single concentration of a compound known to produce micronuclei in vivo is adequate as a positive control if it shows a significant response at any one time point; additional concentration levels may be used. To select an appropriate concentration level, a pilot or trial study may be advisable.

Initially, one concentration of the test substance may be used, the maximum tolerated dose or that producing some indication of toxicity, e.g., a drop in the ratio of polychromatic to normochromatic erythrocytes. Intraperitoneal injection of 1,2-dimethyl-benz-anthracene or benzene are examples of positive control exposures. A concentration of 50-80 percent of an LD50 may be a suitable guide.

(d) Test performance--(1) Inhalation exposure. (i) All data developed within this study shall be in accordance with good laboratory practice provisions under Sec. 79.60.

(ii) The general conduct of this study shall be in accordance with the vehicle emissions inhalation exposure guideline in Sec. 79.61.

(2) Preparation of slides and sampling times. Within twenty-four hours of the last exposure, test animals will be sacrificed. One femur from each test animal will be removed and placed in fetal bovine serum. The bone marrow is removed, cells processed, and two bone marrow smears are made for each animal on glass microscope slides. The slides are stained with acridine- orange (AO) or another appropriate stain (Giemsa + Wright's, etc.) and examined under a microscope.

(3) Analysis. Slides shall be coded for study before microscopic analysis. At least 1,000 first-division erythrocytes per animal shall be scored for the incidence of micronuclei. Sexes will be analyzed separately.

(e) Data and report--(1) Treatment of results. In addition to the reporting requirements specified under Secs. 79.60 and 79.61, the final test report must include the criteria for scoring micronuclei. Individual data shall be presented in a tabular form including both positive and negative controls and experimental groups. The number of polychromatic erythrocytes scored, the number of micronucleated erythrocytes, the percentage of micronucleated cells, and, where applicable, the percentage of micronucleated erythrocytes shall be listed separately for each experimental and control animal. Absolute numbers shall be included if percentages are reported.

(2) Interpretation of data. (i) There are several criteria for determining a positive response, one of which is a statistically significant dose-related increase in the number of micronucleated polychromatic erythrocytes. Another criterion may be based upon detection of a reproducible and statistically significant positive response for at least one of the test substance concentrations.

(ii) A test substance which does not produce either a statistically significant dose-related increase in the number of micronucleated polychromatic erythrocytes or a statistically significant and reproducible positive response at any one of the test points is considered nonmutagenic in this system.

(3) Test evaluation. (i) Positive results in the micronucleus test provide information on the ability of a chemical to induce micronuclei

in erythrocytes of the test species under the conditions of the test. This damage may have been the result of chromosomal damage or damage to the mitotic apparatus.

(ii) Negative results indicate that under the test conditions the test substance does not produce micronuclei in the bone marrow of the test species.

(f) Test report. In addition to the reporting recommendations as specified under Sec. 79.60, the following specific information shall be reported:

(1) Test atmosphere concentration(s) used and rationale for concentration selection.

(2) Rationale for and description of treatment and sampling schedules, toxicity data, negative and positive controls.

(3) Historical control data (negative and positive), if available.

(4) Details of the protocol used for slide preparation.

(5) Criteria for identifying micronucleated erythrocytes.

(6) Micronucleus analysis by animal and by group for each concentration (sexes analyzed separately).

(i) Ratio of polychromatic to normochromatic erythrocytes.

(ii) Number of polychromatic erythrocytes with micronuclei.

(iii) Number of polychromatic erythrocytes scored.

(7) Statistical methodology chosen for test analysis.

(g) References. For additional background information on this test guideline, the following references should be consulted.

(1) 40 CFR 798.5395, In Vivo, Mammalian Bone Marrow Cytogenetics Tests: Micronucleus Assay.

(2) Cihak, R. ``Evaluation of Benzidine by the Micronucleus Test." Mutation Research, 67: 383-384 (1979).

(3) Evans, H.J. ``Cytological Methods for Detecting Chemical Mutagens." Chemical Mutagens: Principles and Methods for Their Detection, Vol. 4. Ed. A. Hollaender (New York and London: Plenum Press, 1976) pp. 1-29.

(4) Heddle, J.A., et al. ``The Induction of Micronuclei as a Measure of Genotoxicity. A Report of the U.S. Environmental Protection Agency Gene-Tox Program." Mutation Research, 123:61-118 (1983).

(5) Preston, J.R. et al. ``Mammalian In Vivo and In Vitro Cytogenetics Assays: Report of the Gene-Tox Program." Mutation Research, 87:143-188 (1981).

(6) Schmid, W. ``The micronucleus test for cytogenetic analysis", Chemical Mutagens, Principles and Methods for their Detection. Vol. 4 Hollaender A, (Ed. A ed. (New York and London: Plenum Press, (1976) pp. 31-53.

(7) Tice, R.E., and Al Pellom ``User's guide: Micronucleus assay

data management and analysis system", NTIS Order no. PB-90-212-598AS.

Sec. 79.65 In vivo sister chromatid exchange assay.

(a) Purpose. The in vivo sister chromatid exchange (SCE) assay detects the ability of a chemical to enhance the exchange of DNA between two sister chromatids of a duplicating chromosome. The most commonly used assays employ mammalian bone marrow cells or peripheral blood lymphocytes, often from rodent species.

(b) Definitions. For the purposes of this section, the following definitions apply:

C-metaphase means a state of arrested cell growth typically seen after treatment with a spindle inhibitor, i.e., colchicine.

Sister chromatid exchange means a reciprocal interchange of the two chromatid arms within a single chromosome. This exchange is visualized during the metaphase portion of the cell cycle and presumably requires the enzymatic incision, translocation and ligation of at least two DNA helices.

(c) Test method--(1) Principle of the test method. (i) Groups of rodents are exposed by the inhalation route for a minimum of 6 hours/day over a period of not less than 28 days to three or more concentrations of a test substance in air. Groups of animals are sacrificed at the end of the exposure period and blood lymphocyte cell cultures are prepared from study animals. Cell growth is suspended after a time and cells are harvested, fixed and stained before scoring for SCEs. Researchers may need to run a trial at the highest tolerated concentration of the test atmosphere to optimize the sample collection time for second division metaphase cells.

(ii) This assay may be done separately or in combination with the subchronic toxicity study, pursuant to the provisions in Sec. 79.62.

(2) Description. (i) The method described here employs peripheral blood lymphocytes (PBL) of laboratory rodents exposed to the test atmosphere.

(ii) Within twenty-four hours of the last exposure, test animal lymphocytes are obtained by heart puncture and duplicate cell cultures are started for each animal. Cultures are grown in bromo-deoxyuridine (BrdU), and then a spindle inhibitor (e.g., colchicine) is added to arrest cell growth. Cells are harvested, fixed, and stained and their chromosomes are scored for SCEs.

(3) Species and strain. The rat is the recommended test animal. Other rodent species may be used in this assay, but use of that species will be justified by the tester.

(4) Animal number and sex. At least five female and five male

animals per experimental and control group shall be used. The use of a single sex or different number of animals shall be justified.

(5) Positive control group. A single concentration of a compound known to produce SCEs in vivo is adequate as a positive control if it shows a significant response at any one time point; additional concentration levels may be used. To select an appropriate concentration level, a pilot or trial study may be advisable. Initially, one concentration of the test substance may be used, the maximum tolerated dose or that producing some indication of toxicity as evidenced by animal morbidity (including death) or target cell toxicity. Intraperitoneal injection of 1,2-dimethyl-benz-anthracene or benzene are examples of positive control exposures. A concentration of 50-80 percent of an LD50 would also be a suitable guide.

(6) Inhalation exposure. (i) All data developed within this study shall be in accordance with good laboratory practice provisions under Sec. 79.60.

(ii) The general conduct of this study shall be in accordance with the vehicle emissions inhalation exposure guideline in Sec. 79.61.

(d) Test performance--(1) Treatment. At the conclusion of the exposure period, all test animals are anaesthetized and heart punctures are performed. Lymphocytes are isolated over a Ficoll gradient and replicate cell cultures are started for each animal. After some 21 hours, the cells are treated with BrdU and returned to incubation. The following day, a spindle inhibitor (e.g., colchicine) is added to arrest cell growth in c-metaphase. Cells are harvested 4 hours later and second-division metaphase cells are washed and fixed in methanol:acetic acid, stained, and chromosome preparations are scored for SCEs.

(2) Staining method. Staining of slides to reveal SCEs can be performed according to any of several protocols. However, the fluorescence plus Giemsa method is recommended.

(3) Number of cells scored. (i) A minimum of 25 well-stained, second-division metaphase cells shall be scored for each animal for each cell type.

(ii) At least 100 consecutive metaphase cells shall be scored for the number of first, second, and third division metaphases for each animal for each cell type.

(iii) At least 1000 consecutive PBL's shall be scored for the number of metaphase cells present.

(iv) The number of cells to be analyzed per animal shall be based upon the number of animals used, the negative control frequency, the pre-determined sensitivity and the power chosen for the test. Slides shall be coded before microscopic analysis.

(e) Data and report--(1) Treatment of results. In addition to the reporting requirements specified under Secs. 79.60 and 61, data shall

be presented in tabular form, providing scores for both the number of SCE for each metaphase. Differences among animals within each group shall be considered before making comparisons between treated and control groups.

(2) Statistical evaluation. Data shall be evaluated by appropriate statistical methods.

(3) Interpretation of results. (i) There are several criteria for determining a positive result, one of which is a statistically significant dose-related increase in the number of SCE. Another criterion may be based upon detection of a reproducible and statistically significant positive response for at least one of the test concentrations.

(ii) A test substance which does not produce either a statistically significant dose-related increase in the number of SCE or a statistically significant and reproducible positive response at any one of the test concentrations is considered not to induce rearrangements of DNA segments in this system.

(iii) Both biological and statistical significance shall be considered together in the evaluation.

(4) Test evaluation. (i) A positive result in the in vivo SCE assay for either, or both, the lung or lymphocyte cultures indicates that under the test conditions the test substance induces reciprocal interchanges of DNA in duplicating chromosomes from lung or lymphocyte cells of the test species.

(ii) Negative results indicate that under the test conditions the test substance does not induce reciprocal interchanges in lung or lymphocyte cells of the test species.

(5) Test report. In addition to the reporting recommendations as specified under Secs. 79.60 and 79.61, the following specific information shall be reported:

(i) Test concentrations used, rationale for concentration selection, negative and positive controls;

(ii) Toxic response data by concentration;

(iii) Schedule of administration of test atmosphere, BrdU, and spindle inhibitor;

(iv) Time of harvest after administration of BrdU;

(v) Identity of spindle inhibitor, its concentration and timing of treatment;

(vi) Details of the protocol used for cell culture and slide preparation;

(vii) Criteria for scoring SCE;

(viii) Replicative index, i.e., [percent 1st division+(2 x percent 2nd division) + (3 x percent 3rd division) metaphases]/100; and

(ix) Mitotic activity, i.e., # of metaphases/1000 cells.

(f) References. For additional background information on this test

guideline, the following references should be consulted.

(1) 40 CFR 798.5915, In vivo Sister Chromatid Exchange Assay.

(2) Kato, H. ``Spontaneous Sister Chromatid Exchanges Detected by a BudR-Labeling Method." Nature, 251:70-72 (1974).

(4) Kligerman, A. D., et al. ``Sister Chromatid Exchange Analysis in Lung and Peripheral Blood Lymphocytes of Mice Exposed to Methyl Isocyanate by Inhalation." Environmental Mutagenesis 9:29-36 (1987).

(5) Kligerman, A.D., et al., ``Cytogenetic Studies of Rodents Exposed to Styrene by Inhalation", IARC Monographs no. 127 ``Butadiene and Styrene: Assesment of Health Hazards" (Sorsa, et al., eds), pp 217-224, 1993.

(6) Kligerman, A., et al., ``Cytogenetic Studies of Mice Exposed to Styrene by Inhalation.", Mutation Research, 280:35-43, 1992.

(7) Wolff, S., and P. Perry. ``Differential Giemsa Staining of Sister Chromatids and the Study of Sister Chromatid Exchanges Without Autoradiography." Chromosoma 48: 341-53 (1974).

Sec. 79.66 Neuropathology assessment.

(a) Purpose. (1) The histopathological and biochemical techniques in this guideline are designed to develop data in animals on morphologic changes in the nervous system associated with repeated inhalation exposures to motor vehicle emissions. These tests are not intended to provide a detailed evaluation of neurotoxicity. Neuropathological evaluation should be complemented by other neurotoxicity studies, e.g. behavioral and neurophysiological studies and/or general toxicity testing, to more completely assess the neurotoxic potential of an exposure.

(2) [Reserved]

(b) Definition. Neurotoxicity (NTX) or a neurotoxic effect is an adverse change in the structure or function of the nervous system following exposure to a chemical substance.

(c) Principle of the test method. (1) Laboratory rodents are exposed to one of several concentration levels of a test atmosphere for at least six hours daily over a period of 90 days. At the end of the exposure period, the animals are anaesthetized, perfused in situ with fixative, and tissues in the nervous system are examined grossly and prepared for microscopic examination. Starting with the highest dosage level, tissues are examined under the light microscope for morphologic changes, until a no-observed-adverse-effect level is determined. In cases where light microscopy has revealed neuropathology, the NOAEL may be confirmed by electron microscopy.

(2) The tests described herein may be combined with any other toxicity study, as long as none of the requirements of either are violated by the combination. Specifically, this assay may be combined with a subchronic toxicity study, pursuant to provisions in Sec. 79.62.

(d) Limit test. If a test at one dose level of the highest concentration that can be achieved while maintaining a particle size distribution with a mass median aerodynamic diameter (MMAD) of 4 micrometers (<greek-m>m) or less, using the procedures described in paragraph (a) of this section, produces no observable toxic effects and if toxicity would not be expected based upon data of structurally related compounds, then a full study using three dose levels might not be necessary. Expected human exposure though may indicate the need for a higher dose level.

(e) Test procedures--(1) Animal selection--(i) Species and strain. Testing shall be performed in the species being used in other NTX tests. A standard strain of laboratory rat is recommended. The choice of species shall take into consideration such factors as the comparative metabolism of the chemical and species sensitivity to the toxic effects of the test substance, as evidenced by the results of other studies, the potential for combined studies, and the availability of other toxicity data for the species.

(ii) Age. Animals shall be at least ten weeks of age at the start of exposure.

(iii) Sex. Both sexes shall be used unless it is demonstrated that one sex is refractory to the effects of exposure.

(2) Number of Animals. A minimum of ten animals per group shall be used. The tissues from each animal shall be examined separately.

(3) Control Groups. (i) A concurrent control group, exposed to clean, filtered air only, is required.

(ii) The laboratory performing the testing shall provide positive control data, e.g., results from repeated acrylamide exposure, as evidence of the ability of their histology procedures to detect neurotoxic endpoints. Positive control data shall be collected at the time of the test study unless the laboratory can demonstrate the adequacy of historical data for the planned study.

(iii) A satellite group of 10 female and 10 male test subjects shall be treated with the highest concentration level for the duration of the exposure and observed thereafter for reversibility, persistence, or delayed occurrence of toxic effects during a post-treatment period of not less than 28 days.

(4) Inhalation exposure. (i) All data developed within this study shall be in accordance with good laboratory practice provisions under Sec. 79.60.

(ii) The general conduct of this study shall be in accordance with the vehicle emissions inhalation exposure guideline in Sec. 79.61.

(5) Study conduct--(i) Observation of animals. All toxicological (e.g., weight loss) and neurological signs (e.g., motor disturbance) shall be recorded frequently enough to observe any abnormality, and not less than weekly.

(ii) The following is a minimal list of measures that shall be noted:

(A) Body weight;

(B) Subject's reactivity to general stimuli such as removal from the cage or handling;

(C) Description, incidence, and severity of any convulsions, tremors, or abnormal motor movements in the home cage;

(D) Descriptions and incidence of posture and gait abnormalities observed in the home cage; and

(E) Description and incidence of any unusual or abnormal behaviors, excessive or repetitive actions (stereotypies), emaciation, dehydration, hypotonia or hypertonia, altered fur appearance, red or crusty deposits around the eyes, nose, or mouth, and any other observations that may facilitate interpretation of the data.

(iii) Sacrifice of animals--(A) General. The goal of the techniques outlined for sacrifice of animals and preparation of tissues is preservation of tissue morphology to simulate the living state of the cell.

(B) Perfusion technique. Animals shall be perfused in situ by a generally recognized technique. For fixation suitable for light or electronic microscopy, saline solution followed by buffered 2.5 percent glutaraldehyde or buffered 4.0 percent paraformaldehyde, is recommended. While some minor modifications or variations in procedures are used in different laboratories, a detailed and standard procedure for vascular perfusion may be found in the text by Zeman and Innes (1963), Hayat (1970), and Spencer and Schaumburg (1980) under paragraph (g) of this section. A more sophisticated technique is described by Palay and Chan-Palay (1974) under paragraph (g) of this section.

(C) Removal of brain and cord. After perfusion, the bony structure (cranium and vertebral column) shall be exposed. Animals shall then be stored in fixative-filled bags at 4 deg.C for 8-12 hours. The cranium and vertebral column shall be removed carefully by trained technicians without physical damage of the brain and cord. Detailed dissection procedures may be found in the text by Palay and Chan-Palay (1974) under paragraph (g) of this section. After removal, simple measurement of the size (length and width) and weight of the whole brain (cerebrum, cerebellum, pons-medulla) shall be made. Any abnormal coloration or discoloration of the brain and cord shall also be noted and recorded.

(D) Sampling. Cross-sections of the following areas shall be examined: The forebrain, the center of the cerebrum, the midbrain, the cerebellum, and the medulla oblongata; the spinal cord at the cervical

swelling (C<INF>3-C<INF>6), and proximal sciatic nerve (mid-thigh and sciatic notch) or tibial nerve (at knee). Other sites and tissue elements (e.g., gastrocnemius muscle) shall be examined if deemed necessary. Any observable gross changes shall be recorded.

(iv) Specimen storage. Tissue samples from both the central and peripheral nervous system shall be further immersion fixed and stored in appropriate fixative (e.g., 10 percent buffered formalin for light microscopy; 2.5 percent buffered gluteraldehyde or 4.0 percent buffered paraformaldehyde for electron microscopy) for future examination. The volume of fixative versus the volume of tissues in a specimen jar shall be no less than 25:1. All stored tissues shall be washed with buffer for at least 2 hours prior to further tissue processing.

(v) Histopathology examination--(A) Fixation. Tissue specimens stored in 10 percent buffered formalin may be used for this purpose. All tissues must be immersion fixed in fixative for at least 48 hours prior to further tissue processing.

(B) Dehydration. All tissue specimens shall be washed for at least 1 hour with water or buffer, prior to dehydration. (A longer washing time is needed if the specimens have been stored in fixative for a prolonged period of time.) Dehydration can be performed with increasing concentration of graded ethanols up to absolute alcohol.

(C) Clearing and embedding. After dehydration, tissue specimens shall be cleared with xylene and embedded in paraffin or paraplast. Multiple tissue specimens (e.g. brain, cord, ganglia) may be embedded together in one single block for sectioning. All tissue blocks shall be labelled showing at least the experiment number, animal number, and specimens embedded.

(D) Sectioning. Tissue sections, 5 to 6 microns in thickness, shall be prepared from the tissue blocks and mounted on standard glass slides. It is recommended that several additional sections be made from each block at this time for possible future needs for special stainings. All tissue blocks and slides shall be filed and stored in properly labeled files or boxes.

(E) Histopathological techniques. The following general testing sequence is proposed for gathering histopathological data:

(1) General staining. A general staining procedure shall be performed on all tissue specimens in the highest treatment group. Hematoxylin and eosin (H&E) shall be used for this purpose. The staining shall be differentiated properly to achieve bluish nuclei with pinkish background.

(2) Peripheral nerve teasing. Peripheral nerve fiber teasing shall be used. Detailed staining methodology is available in standard histotechnological manuals such as AFIP (1968), Ralis et al. (1973), and Chang (1979) under paragraph (g) of this section. The nerve fiber teasing technique is discussed in Spencer and Schaumberg (1980) under

paragraph (g) of this section. A section of normal tissue shall be included in each staining to assure that adequate staining has occurred. Any changes shall be noted and representative photographs shall be taken. If a lesion(s) is observed, the special techniques shall be repeated in the next lower treatment group until no further lesion is detectable.

(F) Examination. All stained microscopic slides shall be examined with a standard research microscope. Examples of cellular alterations (e.g., neuronal vacuolation, degeneration, and necrosis) and tissue changes (e.g., gliosis, leukocytic infiltration, and cystic formation) shall be recorded and photographed.

(f) Data collection, reporting, and evaluation. In addition to information meeting the requirements stated under 40 CFR 79.60 and 79.61, the following specific information shall be reported:

(1) Description of test system and test methods. (i) A description of the general design of the experiment shall be provided. This shall include a short justification explaining any decisions where professional judgment is involved such as fixation technique and choice of stains; and

(ii) Positive control data from the laboratory performing the test that demonstrate the sensitivity of the procedures being used. Historical data may be used if all essential aspects of the experimental protocol are the same.

(2) Results. All observations shall be recorded and arranged by test groups. This data may be presented in the following recommended format:

(i) Description of signs and lesions for each animal. For each animal, data must be submitted showing its identification (animal number, treatment, dose, duration), neurologic signs, location(s) nature of, frequency, and severity of lesion(s). A commonly-used scale such as 1+, 2+, 3+, and 4+ for degree of severity ranging from very slight to extensive may be used. Any diagnoses derived from neurologic signs and lesions including naturally occurring diseases or conditions, shall also be recorded;

(ii) Counts and incidence of lesions, by test group. Data shall be tabulated to show:

(A) The number of animals used in each group, the number of animals displaying specific neurologic signs, and the number of animals in which any lesion was found; and

(B) The number of animals affected by each different type of lesion, the average grade of each type of lesion, and the frequency of each different type and/or location of lesion.

(iii) Evaluation of data. (A) An evaluation of the data based on gross necropsy findings and microscopic pathology observations shall be made and supplied. The evaluation shall include the relationship, if

any, between the animal's exposure to the test atmosphere and the frequency and severity of any lesions observed; and

(B) The evaluation of dose-response, if existent, for various groups shall be given, and a description of statistical method must be presented. The evaluation of neuropathology data shall include, where applicable, an assessment in conjunction with any other neurotoxicity studies, electrophysiological, behavioral, or neurochemical, which may be relevant to this study.

(g) References. For additional background information on this test guideline, the following references should be consulted.

(1) 40 CFR 798.6400, Neuropathology.
(2) AFIP Manual of Histologic Staining Methods. (New York: McGraw-Hill (1968).
(3) Chang, L.W. A Color Atlas and Manual for Applied Histochemistry. (Springfield, IL: Charles C. Thomas, 1979).
(4) Dunnick, J.K., et.al. Thirteen-week Toxicity Study of N-Hexane in B6C3F1 Mice After Inhalation Exposure (1989) Toxicology, 57, 163-172.
(5) Hayat, M.A. ``Vol. 1. Biological applications," Principles and techniques of electron microscopy. (New York: Van Nostrand Reinhold, 1970).
(6) Palay S.L., Chan-Palay, V. Cerebellar Cortex: Cytology and Organization. (New York: Springer-Verlag, 1974).
(7) Ralis, H.M., Beesley, R.A., Ralis, Z.A. Techniques in Neurohistology. (London: Butterworths, 1973).
(8) Sette, W. ``Pesticide Assessment Guidelines, Subdivision F, Neurotoxicity Test Guidelines." Report No. 540/09-91-123 U.S. Environmental Protection Agency 1991 (NTIS #PB91-154617).
(9) Spencer, P.S., Schaumburg, H.H. (eds). Experimental and Clinical Neurotoxicology. (Baltimore: Williams and Wilkins, 1980).
(10) Zeman, W., Innes, J.R.M. Craigie's Neuroanatomy of the Rat. (New York: Academic, 1963).

Sec. 79.67 Glial fibrillary acidic protein assay.

(a) Purpose. Chemical-induced injury of the nervous system, i.e., the brain, is associated with astrocytic hypertrophy at the site of damage (see O'Callaghan, 1988 in paragraph (e)(3) in this section). Assays of glial fibrillary acidic protein (GFAP), the major intermediate filament protein of astrocytes, can be used to document this response. To date, a diverse variety of chemical insults known to be injurious to the central nervous system have been shown to increase GFAP. Moreover, increases in GFAP can be seen at concentrations below

those necessary to produce cytopathology as determined by routine Nissl stains (standard neuropathology). Thus it appears that assays of GFAP represent a sensitive approach for documenting the existence and location of chemical-induced injury of the central nervous system. Additional functional, histopathological, and biochemical tests are necessary to assess completely the neurotoxic potential of any chemical. This biochemical test is intended to be used in conjunction with neurohistopathological studies.

(b) Principle of the test method. (1) This guideline describes the conduct of a radioimmunoassay for measurement of the amount of GFAP in the brain of vehicle emission-exposed and unexposed control animals. It is based on modifications (O'Callaghan & Miller 1985 in paragraph (e)(5), O'Callaghan 1987 in paragraph (e)(1) of this section) of the dot-immunobinding procedure described by Jahn et al. (1984) in paragraph (e)(2) of this section. Briefly, brain tissue samples from study animals are assayed for total protein, diluted in dot-immunobinding buffer, and applied to nitrocellulose sheets. The spotted sheets are then fixed, blocked, washed and incubated in anti-GFAP antibody and [I<SUP>125] Protein A. Bound protein A is then quantified by gamma spectrometry. In lieu of purified protein standards, standard curves are constructed from dilution of a single control sample. By comparing the immunoreactivity of individual samples (both control and exposed groups) with that of the sample used to generate the standard curve, the relative immunoreactivity of each sample is obtained. The immunoreactivity of the control groups is normalized to 100 percent and all data are expressed as a percentage of control. A variation on this radioimmunoassay procedure has been proposed (O'Callaghan 1991 in paragraph (e)(4) of this section) which uses a ``sandwich'' of GFAP, anti-GFAP, and a chromophore in a microtiter plate format enzyme-link immunosorbent assay (ELISA). The use of this variation shall be justified.

(2) This assay may be done separately or in combination with the subchronic toxicity study, pursuant to the provisions of Sec. 79.62.

(c) Test procedure--(1) Animal selection--(i) Species and strain. Test shall be performed on the species being used in concurrent testing for neurotoxic or other health effect endpoints. This will generally be a species of laboratory rat. The use of other rodent or non-rodent species shall be justified.

(ii) Age. Based on other concurrent testing, young adult rats shall be used. Study rodents shall not be older than ten weeks at the start of exposures.

(iii) Number of animals. A minimum of ten animals per group shall be used. The tissues from each animal shall be examined separately.

(iv) Sex. Both sexes shall be used unless it is demonstrated that one sex is refractory to the effects.

(2) Materials. The materials necessary to perform this study are [I<SUP>125] Protein A (2-10 <greek-m>Ci/<greek-m>g), Anti-sera to GFAP, nitrocellulose paper (0.1 or 0.2 <greek-m>m pore size), sample application template (optional; e.g., ``Minifold II", Schleicher & Schuell, Keene, NH), plastic sheet incubation trays.

(3) Study conduct. (i) All data developed within this study shall be in accordance with good laboratory practice provisions under Sec. 79.60.

(ii) Tissue Preparation. Animals are euthanized 24 hours after the last exposure and the brain is excised from the skull. On a cold dissecting platform, the following six regions are dissected freehand: cerebellum; cerebral cortex; hippocampus; striatum; thalamus/ hypothalamus; and the rest of the brain. Each region is then weighed and homogenized in 10 volumes of hot (70-90 deg.C) 1 percent (w/v) sodium dodecyl sulfate (SDS). Homogenization is best achieved through sonic disruption. A motor driven pestle inserted into a tissue grinding vessel is a suitable alternative. The homogenized samples can then be stored frozen at -70 deg.C for at least 4 years without loss of GFAP content.

(iii) Total Protein Assay. Aliquots of the tissue samples are assayed for total protein using the method of Smith et al. (1985) in paragraph (e)(7) of this section. This assay may be purchased in kit form (e.g., Pierce Chemical Company, Rockford, IL).

(iv) Sample Preparation. Dilute tissue samples in sample buffer (120 mM KCl, 20 mM NaCl, 2 mM MgCl<INF>2), 5 mM Hepes, pH 7.4, 0.7 percent Triton X-100) to a final concentration of 0.25 mg total protein per ml (5 <greek-m>g/20 <greek-m>l).

(v) Preparation of Standard Curve. Dilute a single control sample in sample buffer to give at least five standards, between 1 and 10 <greek-m>g total protein per 20 <greek-m>l. The suggested values of total protein per 20 <greek-m>l sample buffer are 1.25, 2.50, 3.25, 5.0, 6.25, 7.5, 8.75, and 10.0 <greek-m>g.

(vi) Preparation of Nitrocellulose Sheets. Nitrocellulose sheets of 0.1 or 0.2 micron pore size are rinsed by immersion in distilled water for 5 minutes and then air dried.

(vii) Sample Application. Samples can be spotted onto the nitrocellulose sheets free-hand or with the aid of a template. For free-hand application, draw a grid of squares approximately 2 centimeters by 2 centimeters (cm) on the nitrocellulose sheets using a soft pencil. Spot 5-10 <greek-m>l portions to the center of each square for a total sample volume of 20 <greek-m>l. For template aided sample application a washerless microliter capacity sample application manifold is used. Position the nitrocellulose sheet in the sample application device as recommended by the manufacturer and spot a 20 <greek-m>l sample in one application. Do not wet the nitrocellulose or

any support elements prior to sample application. Do not apply vacuum during or after sample application. After spotting samples (using either method), let the sheets air dry. The sheets can be stored at room temperature for several days after sample application.

(viii) Standard Incubation Conditions. These conditions have been described by Jahn et al. (1984) in paragraph (e)(2) of this section. All steps are carried out at room temperature on a flat shaking platform (one complete excursion every 2-3 seconds). For best results, do not use rocking or orbital shakers. Perform the following steps in enough solution to cover the nitrocellulose sheets to a depth of 1 cm.

(A) Incubate 20 minutes in fixer (25 percent (v/v) isopropanol, 10 percent (v/v) acetic acid).

(B) Discard fixer, wash several times in deionized water to eliminate the fixer, and then incubate for 5 minutes in Tris-buffered saline (TBS): 200 mM NaCL, 60 mM Tris-HCl to pH 7.4.

(C) Discard TBS and incubate 1 hour in blocking solution (0.5 percent gelatin (w/v)) in TBS.

(D) Discard blocking solution and incubate for 2 hours in antibody solution (anti-GFAP antiserum diluted to the desired dilution in blocking solution containing 0.1 percent Triton X-100). Serum anti-bovine GFAP, which cross reacts with GFAP from rodents and humans, can be obtained commercially (e.g., Dako Corp.) and used at a dilution of 1:500.

(E) Discard antibody solution, and wash in 4 changes of TBS for 5 minutes each time. Then wash in TBS for 10 minutes.

(F) Discard TBS and incubate in blocking solution for 30 minutes.

(G) Discard blocking solution and incubate for 1 hour in Protein A solution ([I\125\]-labeled Protein A diluted in blocking solution containing 0.1 percent Triton X-100, sufficient to produce 2000 counts per minute (cpm) per 10 <greek-m>l of Protein A solution).

(H) Remove Protein A solution (it may be reused once). Wash in 0.1 percent Triton X-100 in TBS (TBSTX) for 5 minutes, 4 times. Then wash in TBSTX for 2-3 hours for 4 additional times. An overnight wash in a larger volume can be used to replace the last 4 washes.

(I) Hang sheets to air-dry. Cut out squares or spots and count radioactivity in a gamma counter.

(ix) Expression of data. Compare radioactivity counts for samples obtained from control and treated animals with counts obtained from the standard curve. By comparing the immunoreactivity (counts) of each sample with that of the standard curve, the relative amount of GFAP in each sample can be determined and expressed as a percent of control.

(d) Data Reporting and Evaluation--(1) Test Report. In addition to information meeting the requirements stated under 40 CFR 79.60, the following specific information shall be reported:

(i) Body weight and brain region weights at time of sacrifice for

each subject tested;

(ii) Indication of whether each subject survived to sacrifice or time of death;

(iii) Data from control animals and blank samples; and

(iv) Statistical evaluation of results;

(2) Evaluation of Results. (i) Results shall be evaluated in terms of the extent of change in the amount of GFAP as a function of treatment and dose. GFAP assays (of any brain region) from a minimum of 6 samples typically will result in a standard error of the mean of +/- 5 percent. In this case, a chemically-induced increase in GFAP of 115 percent of control is likely to be statistically significant.

(ii) The results of this assay shall be compared to and evaluated with any relevant behavioral and histopathological data.

(e) References. For additional background information on this test guideline the following references should be consulted.

(1) Brock, T.O and O'Callaghan, J.P. 1987. Quantitative changes in the synaptic vesicle proteins, synapsin I and p38 and the astrocyte specific protein, glial fibrillary acidic protein, are associated with chemical-induced injury to the rat central nervous system, J. Neurosci. 7:931-942.

(2) Jahn, R., Schiebler, W. Greengard, P. 1984. A quantitative dot-immunobinding assay for protein using nitrocellulose membrane filters. Proc. Natl. Acad. Sci. U.S.A. 81:1684-1687.

(3) O'Callaghan, J.P. 1988. Neurotypic and gliotypic protein as biochemical markers of neurotoxicity. Neurotoxicol. Teratol. 10:445-452.

(4) O'Callaghan, J.P. 1991. Quantification of glial fibrillary acidic protein: comparison of slot-immunobinding assays with a novel sandwich ELISA. Neurotoxicol. Teratol. 13:275-281.

(5) O'Callaghan, J.P. and Miller, D.B. 1985. Cerebellar hypoplasia in the Gunn rat is associated with quantitative changes in neurotypic and gliotypic proteins. J. Pharmacol. Exp. Ther. 234:522-532.

(6) Sette, W.F. ``Pesticide Assessment Guidelines, Subdivision `F', Hazard Evaluation: Human and Domestic Animals, Addendum 10, Neurotoxicity, Series 81, 82, and 83" US-EPA, Office of Pesticide Programs, EPA-540/09-91-123, March 1991.

(7) Smith, P.K., Krohn, R.I., Hermanson, G.T., Mallia, A.K., Gartner, F.H., Provenzano, M.D., Fujimoto, E.K., Goeke, N.M., Olson, B.J., Klenk, D.C. 1985. Measurement of protein using bicinchoninic acid. Annal. Biochem. 150:76-85.

Sec. 79.68 Salmonella typhimurium reverse mutation assay.

(a) Purpose. The Salmonella typhimurium histidine (his) reversion system is a microbial assay which measures his<SUP>-<r-arrow> his<SUP>+ reversion induced by chemicals which cause base changes or frameshift mutations in the genome of the microorganism Salmonella typhimurium.

(b) Definitions. For the purposes of this section, the following definitions apply:

Base pair mutagen means an agent which causes a base change in DNA. In a reversion assay, this change may occur at the site of the original mutation or at a second site in the chromosome.

Frameshift mutagen is an agent which causes the addition or deletion of single or multiple base pairs in the DNA molecule.

Salmonella typhimurium reverse mutation assay detects mutation in a gene of a histidine-requiring strain to produce a histidine independent strain of this organism.

(c) Reference substances. These may include, but need not be limited to, sodium azide, 2-nitrofluorene, 9-aminoacridine, 2-aminoanthracene, congo red, benzopurpurin 4B, trypan blue or direct blue 1.

(d) Test method.--(1) Principle. Motor vehicle combustion emissions from fuel or additive/base fuel mixtures are, first, filtered to trap particulate matter and, then, passed through a sorbent resin to trap semi-volatile gases. Bacteria are separately exposed to the extract from both the filtered particulates and the resin-trapped organics. Assays are conducted using both test mixtures with and without a metabolic activation system and exposed cells are plated onto minimal medium. After a suitable period of incubation, revertant colonies are counted in test cultures and compared to the number of spontaneous revertants in unexposed control cultures.

(2) Description. Several methods for performing the test have been described. The procedures described here are for the direct plate incorporation method and the azo-reduction method. Among those used are:

(i) Direct plate incorporation method;

(ii) Preincubation method;

(iii) Azo-reduction method;

(iv) Microsuspension method; and

(v) Spiral assay.

(3) Strain selection--(i)Designation. Five tester strains shall be used in the assay. At the present time, TA1535, TA1537, TA98, and TA100 are designated as tester strains. The fifth strain will be chosen from the pool of Salmonella strains commonly used to determine the degree to which nitrated organic compounds, i.e., nitroarenes, contribute to the overall mutagenic activity of a test substance. TA98/1,8-DNP<INF>6 or

other suitable Rosenkranz nitro-reductase resistant strains will be considered acceptable. The choice of the particular strain is left to the discretion of the researcher. However, the researcher shall justify the use of the selected bacterial tester strains.

(ii) Preparation and storage of bacterial tester strains. Recognized methods of stock culture preparation and storage shall be used. The requirement of histidine for growth shall be demonstrated for each strain. Other phenotypic characteristics shall be checked using such methods as crystal violet sensitivity and resistance to ampicillin. Spontaneous reversion frequency shall be in the range expected as reported in the literature and as established in the laboratory by historical control values.

(iii) Bacterial growth. Fresh cultures of bacteria shall be grown up to the late exponential or early stationary phase of growth (approximately 108-109 cells per ml).

(4) Exogenous metabolic activation. Bacteria shall be exposed to the test substance both in the presence and absence of an appropriate exogenous metabolic activation system. For the direct plate incorporation method, the most commonly used system is a cofactor-supplemented postmitochondrial fraction prepared from the livers of rodents treated with enzyme-inducing agents, such as Aroclor 1254. For the azo-reduction method, a cofactor- supplemented postmitochondrial fraction (S-9) prepared from the livers of untreated hamsters is preferred. For this method, the cofactor supplement shall contain flavin mononucleotide, exogenous glucose 6-phosphate dehydrogenase, NADH and excess of glucose-6-phosphate.

(5) Control groups--(i) Concurrent controls. Concurrent positive and negative (untreated) controls shall be included in each experiment. Positive controls shall ensure both strain responsiveness and efficacy of the metabolic activation system.

(ii) Strain specific positive controls shall be included in the assay. Examples of strain specific positive controls are as follows:

(A) Strain TA1535, TA100: sodium azide;
(B) TA98: 2-nitrofluorene (without activation), 2-anthramine (with activation);
(C) TA1537: 9-aminoacridine; and
(D) TA98/1,8-DNP<INF>6: benzo(a)pyrene (with activation).

The papers by Claxton et al., 1991 and 1992 in paragraph (g) in this section will provide helpful information for the selection of positive controls.

(iii) Positive controls to ensure the efficacy of the activation system. The positive control reference substances for tests including a metabolic activation system shall be selected on the basis of the type of activation system used in the test. 2-Aminoanthracene is an example

of a positive control compound in plate-incorporation tests using postmitochondrial fractions from the livers of rodents treated with enzyme-inducing agents such as Aroclor-1254. Congo red is an example of a positive control compound in the azo-reduction method. Other positive control reference substances may be used.

(iv) Class-specific positive controls. The azo-reduction method shall include positive controls from the same class of compounds as the test agent wherever possible.

(6) Sampling the test atmosphere.--(i) Extracts of test emissions are collected on Teflon<Register>-coated glass fiber filters using an exhaust dilution setup. The particulates are extracted with dichloromethane (DCM) using Soxhlet extraction techniques. Extracts in DCM can be stored at dry ice temperatures until use.

(ii) Gaseous hydrocarbons passing through the filter are trapped by a porous, polymer resin, like XAD-2/styrene-divinylbenzene, or an equivalent product. Methylene chloride is used to extract the resin and the sample is evaporated to dryness before storage or use.

(iii) Samples taken from this material are then used to expose the cells in this assay. Final concentration of extracts in solvent/vehicle, or after solvent exchange, shall not interfere with cell viability or growth rate. The paper by Stump (1982) in paragraph (g) of this section is useful for preparing extracts of particulate and semi-volatile organic compounds from diesel and gasoline exhaust stream.

(iv) Exposure concentrations. (A) The test should initially be performed over a broad range of concentrations. Among the criteria to be taken into consideration for determining the upper limits of test substance concentration are cytotoxicity and solubility. Cytotoxicity of the test chemical may be altered in the presence of metabolic activation systems. Toxicity may be evidenced by a reduction in the number of spontaneous revertants, a clearing of the background lawn or by the degree of survival of treated cultures. Relatively insoluble samples shall be tested up to the limits of solubility. The upper test chemical concentration shall be determined on a case by case basis.

(B) Generally, a maximum of 5 mg/plate for pure substances is considered acceptable. At least 5 different concentrations of test substance shall be used with adequate intervals between test points.

(C) When appropriate, a single positive response shall be confirmed by testing over a narrow range of concentrations.

(e) Test performance. All data developed within this study shall be in accordance with good laboratory practice provisions under Sec. 79.60.

(1) Direct plate incorporation method. When testing with metabolic activation, test solution, bacteria, and 0.5 ml of activation mixture containing an adequate amount of postmitochondrial fraction shall be added to the liquid overlay agar and mixed. This mixture is poured over

the surface of a selective agar plate. Overlay agar shall be allowed to solidify before incubation. At the end of the incubation period, revertant colonies per plate shall be counted. When testing without metabolic activation, the test sample and 0.1 ml of a fresh bacterial culture shall be added to 2.0 ml of overlay agar.

(2) Azo-reduction method. When testing with metabolic activation, 0.5 ml of activation mixture containing 150 <greek-m>l of postmitochondrial fraction and 0.1 ml of bacterial culture shall be added to a test tube kept on ice. 0.1 ml of test solution shall be added, and the tubes shall be incubated with shaking at 30 deg.C for 30 minutes. At the end of the incubation period, 2.0 ml of agar shall be added to each tube, the contents mixed and poured over the surface of a selective agar plate. Overlay agar shall be allowed to solidify before incubation. At the end of the incubation period, revertant colonies per plate shall be counted. For tests without metabolic activation, 0.5 ml of buffer shall be used in place of the 0.5 ml of activation mixture. All other procedures shall be the same as those used for the test with metabolic activation.

(3) Other methods/modifications may also be appropriate.

(4) Media. An appropriate selective medium with an adequate overlay agar shall be used.

(5) Incubation conditions. All plates within a given experiment shall be incubated for the same time period. This incubation period shall be for 48-72 hours at 37 deg.C.

(6) Number of cultures. All plating shall be done at least in triplicate.

(f) Data and report--(1) Treatment of results. Data shall be presented as number of revertant colonies per plate, revertants per kilogram (or liter) of fuel, and as revertants per kilometer (or mile) for each replicate and dose. These same measures shall be recorded on both the negative and positive control plates. The mean number of revertant colonies per plate, revertants per kilogram (or liter) of fuel, and revertants per kilometer (or mile), as well as individual plate counts and standard deviations shall be presented for the test substance, positive control, and negative control plates.

(2) Statistical evaluation. Data shall be evaluated by appropriate statistical methods. Those methods shall include, at a minimum, means and standard deviations of the reversion data.

(3) Interpretation of results. (i) There are several criteria for determining a positive result, one of which is a statistically significant dose-related increase in the number of revertants. Another criterion may be based upon detection of a reproducible and statistically significant positive response for at least one of the test substance concentrations.

(ii) A test substance which does not produce either a statistically

significant dose-related increase in the number of revertants or a statistically significant and reproducible positive response at any one of the test points is considered nonmutagenic in this system.

(iii) Both biological and statistical significance shall be considered together in the evaluation.

(4) Test evaluation. (i) Positive results from the Salmonella typhimurium reverse mutation assay indicate that, under the test conditions, the test substance induces point mutations by base changes or frameshifts in the genome of this organism.

(ii) Negative results indicate that under the test conditions the test substance is not mutagenic in Salmonella typhimurium.

(5) Test report. In addition to the reporting recommendations as specified under 40 CFR 79.60, the following specific information shall be reported:

(i) Sampling method(s) used and manner in which cells are exposed to sample solution;

(ii) Bacterial strains used;

(iii) Metabolic activation system used (source, amount and cofactor); details of preparation of postmitochondrial fraction;

(vi) Concentration levels and rationale for selection of concentration range;

(v) Description of positive and negative controls, and concentrations used, if appropriate;

(vi) Individual plate counts, mean number of revertant colonies per plate, number of revertants per mile (or kilometer), and standard deviation; and

(vii) Dose-response relationship, if applicable.

(g) References. For additional background information on this test guideline, the following references should be consulted.

(1) 40 CFR 798.5265, The Salmonella typhimurium reverse mutation assay.

(2) Ames, B.N., McCann, J., Yamasaki, E. ``Methods for detecting carcinogens and mutagens with the Salmonella/mammalian microsome mutagenicity test," Mutation Research 31:347-364 (1975).

(3) Huisingh, J.L., et al.,``Mutagenic and Carcinogenic Potency of Extracts of Diesel and Related Environmental Emissions: Study Design, Sample Generation, Collection, and Preparation". In: Health Effects of Diesel Engine Emissions, Vol. II, W.E. Pepelko, R., M., Danner and N. A. Clarke (Eds.), US EPA, Cincinnati, EPA-600/9-80-057b, pp. 788-800 (1980).

(5) Claxton, L.D., Allen, J., Auletta, A., Mortelmans, K., Nestmann, E., Zeiger, E. ``Guide for the Salmonella typhimurium/mammalian microsome tests for bacterial mutagenicity" Mutation Research 189(2):83-91 (1987).

(6) Claxton, L., Houk, V.S., Allison, J.C., Creason, J., ``Evaluating the relationship of metabolic activation system concentrations and chemical dose concentrations for the Salmonella Spiral and Plate Assays" Mutation Research 253:127-136 (1991).
(7) Claxton, L., Houk, V.S., Monteith, L.G., Myers, L.E., Hughes, T.J., ``Assessing the use of known mutagens to calibrate the Salmonella typhimurium mutagenicity assay: I. Without exogenous activation." Mutation Research 253:137-147 (1991).
(8) Claxton, L., Houk, V.S., Warner, J.R., Myers, L.E., Hughes, T.J., ``Assessing the use of known mutagens to calibrate the Salmonella typhimurium mutagenicity assay: II. With exogenous activation." Mutation Research 253:149-159 (1991).
(9) Claxton, L., Creason, J., Lares, B., Augurell, E., Bagley, S., Bryant, D.W., Courtois, Y.A., Douglas, G., Clare, C.B., Goto, S., Quillardet, P., Jagannath, D.R., Mohn, G., Neilsen, P.A., Ohnishi, Y., Ong, T., Pederson, T.C., Shimizu, H., Nylund, L., Tokiwa, H., Vink, I.G.R., Wang, Y., Warshawsky, D., ``Results of the IPCS Collaborative Study on Complex Mixtures" Mutation Research 276:23-32 (1992).
(10) Claxton, L., Douglas, G., Krewski, D., Lewtas, J., Matsushita, H., Rosenkranz, H., ``Overview, conclusions, and recommendations of the IPCS Collaborative Study on Complex Mixtures" Mutation Research 276:61-80 (1992).
(11) Houk, V.S., Schalkowsky, S., and Claxton, L.D., ``Development and Validation of the Spiral Salmonella Assay: An Automated Approach to Bacterial Mutagenicity Testing" Mutation Research 223:49-64 (1989).
(12) Jones, E., Richold, M., May, J.H., and Saje, A. ``The Assessment of the Mutagenic Potential of Vehicle Engine Exhaust in the Ames Salmonella Assay Using a Direct Exposure Method" Mutation Research 97:35-40 (1985).
(13) Maron, D., and Ames, B. N., Revised methods for the Salmonella mutagenicity test, Mutation Research, 113:173-212 (1983).
(14) Prival, M.J., and Mitchell, V.D. ``Analysis of a method for testing azo dyes for mutagenic activity in Salmonella typhimurium in the presence of flavin mononucleotide and hamster liver S-9," Mutation Research 97:103-116 (1982).
(15) Rosenkranz, H.S., et.al. ``Nitropyrenes: Isolation, identification, and reduction of mutagenic impurities in carbon black and toners" Science 209:1039-43 (1980).
(16) Stump, F., Snow, R., et.al., ``Trapping gaseous hydrocarbons for mutagenic testing" SAE Technical Paper Series, No. 820776 (1982).
(17) Vogel, H.J., Bonner, D.M. ``Acetylornithinase of E. coli: partial purification and some properties," Journal of Biological

Chemistry. 218:97-106 (1956).

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