Data Evaluation Report on Field Exposure of *Xenopus laevis* to Atrazine and Other Triazines in South Africa: Exposure Characterization and Assessment of Laryngeal and Gonadal Responses

EPA MRID Number 458677-01

Data Requirement:

EPA DP Barcode D288775

EPA MRID 458677-01
EPA Guideline 70-1(Special Study)

Test material: **Purity:** not reported

Common name Atrazine
chemical name: IUPAC
CAS name 6-chloro-N-ethyl-N'-(1-methylethyl)-1,3,5-triazine-2,4-diamine
CAS No. 1912-24-9
synonyms
EPA PC Code: 80803

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CITATION: Smith, E., L DuPreez and K. Solomon. 2003. Field exposure of *Xenopus laevis* to atrazine and other triazines in South Africa: exposure characterization and assessment of laryngeal and gonadal responses. The Institute of Environmental & Human Health, Texas Tech University, Lubbock, Texas 79490 (USA) and School of Environmental Sciences and Development, Potchefstroom University for CHE, Private Bag X6001, Potchefstroom 2520 (South Africa). Sponsor: Syngenta Crop Protection, Inc., Laboratory Study ID ECORISK Number SA-01B

EXECUTIVE SUMMARY:

This study is the second phase of a project to assess the possible effects of exposure to atrazine and other triazines on African clawed frogs (*Xenopus laevis*) in their South African native range under field conditions. The objective of this phase of the study was to characterize the exposure of frog populations to atrazine and related triazines in surface waters of both reference (no corn/no atrazine-triazine use) and experimental (corn growing/atrazine-triazine use) ponds from November 2001 through early June 2002. During the second two months of the study, rainfall was roughly double the 10-year average and resulted in all of the earthen ponds overflowing their banks for as long as two months. Based on EFED's analysis of raw residue data, maximum atrazine residues in experimental ponds over the study period ranged from 1.46 to 11.6 ug/L while reference ponds ranged from 0.41 to 1.62 ug/L. However, the atrazine degradate diaminochlorotriazine (DACT) maximum residues ranged from 4.6 to 8.2 ug/L at experimental sites and form 6.8 to 7.4 ug/L at reference sites. Two other atrazine degradates, i.e., desethylated atrazine (DEA)and desisopropyl atrazine (DIA) showed roughly similar maximum residue levels in both experimental and reference sites. Maximum residues of terbuthylazine ranged from 1.8 to 5.3 ug/Lat experimental sites and from 2.4 to 2.8 ug/L at reference sites. According to the report, triazine residues in the reference ponds were likely due to wind effect; the authors speculate that the high rain events during the sampling period likely reduced atrazine levels and that "frogs living in these dams [ponds] were undoubtedly exposed to much higher atrazine and other triazine levels than had been recorded during the present study." Given that atrazine and/or its degradates were present in reference ponds at levels at times equivalent to some experimental pond sites and the authors concede that atrazine exposure prior to the winter floods was likely higher at all sites, it is unclear how the study can differentiate atrazine effects on frogs at reference and experimental sites. The high variability in exposure could potentially confound any attempt to document significant differences in effects.

The objective of this study was to examine the effects of atrazine on *X. laevis* in its native habitat (South Africa). Initially the study was intended to test whether morphological and biochemical differences existed between clawed frogs in atrazine-exposed (experimental) versus non-exposed (reference) ponds. The criteria for differentiating reference and experimental sites included production of corn and use of atrazine in the vicinity, plus the presence of *X. laevis* in a pond. Based on an initial survey of the sampling area, five experimental (atrazine exposure) and three reference (no atrazine exposure) sites were selected (458677-09). However, subsequent sampling during later phases of the study revealed that the reference sites all contained measurable residues of atrazine, its degradates, and terbutyhylazine (triazine herbicide not registered for use in the USA) that were, in some cases, higher than sites considered representative of atrazine exposure

I. MATERIALS AND METHODS

GUIDELINE FOLLOWED: Nonguideline Study
COMPLIANCE: Not conducted under full GLP; however, most practices as defined by 40 CFR Part 160, August 19, 1989 were established for this study, including but not limited to:

- Written, authorized protocol
- Written, authorized Standard Operating Procedures for all key procedures.
- Organization and Personnel were sufficient in terms of number, education, training and experience.
- Facilities were of suitable size and construction
- Equipment used was of appropriate design and adequate capacity.
- Test material identity, strength, purity and composition were characterized.
- Independent QA Inspections were conducted.
- Final report was written
- Raw data, documentation, records, protocols, and final report were archived.

A. MATERIALS:

1. Test Material Atrazine

Description: Not reported

Lot No./Batch No. : Not reported

Purity: Not reported
Stability of Compound Under Test Conditions: Not reported

Storage conditions of test chemicals: Not reported

2. Test organism:

Species: African clawed frog (*Xenopus laevis*)
.
Age at test initiation: Adults
Weight at study initiation: (mean and range) not reported
Length at study initiation: (mean and range) not reported

Source: Adult *X. laevis* were field collected in two areas (3 non-corn growing areas and 5 corn growing areas) in the vicinity of Potchefstrooom, South Africa, using traps baited with liver and meat scraps.

B. STUDY DESIGN:

Objective: 1. To characterize the exposure of populations of *Xenopus laevis* larvae, metamorphs and adults to atrazine and related triazines in surface water in reference (no corn production or atrazine/triazine use) and exposed habitats in the proximity of corn production and atrazine/triazine use in the Potchestroom region of South Africa.

1. Experimental Conditions

A total of 8 sampling sites in two adjacent regions (5 in Viljoenskroon corn growing region = E; 3 in non-corn growing Potchefstroom region = C) in South Africa. Experimental site selection based on proximity of corn, previous and/or planned use of atrazine and terbuthylazine, and presence of *Xenopus*. Reference sites based on absence of corn production, absence of triazine amd terbuthylazine in the water, and presence of *Xenopus*.

Biweekly water/sediment samples collected from early November 2001 to early June 2002. Within 5 hours of collection water samples kept at 4°C and transported to testing lab within 24 hours of collection. Water temperature, conductivity, dissolved oxygen and pH were recorded at reference points on each sampling site.

Pesticide and metabolite determinations in water conducted at Department of Microbiology of the School of Environmental Sciences and Development at the Potchefstroom University. Quality control conducted at CSIR (Pretoria, South Africa) and SGS (Midrand, South Africa).

Climatological conditions characterized in terms of air temperature and rainfall.

Agricultural practices in each of the study site catchment areas characterized as to crop, stage of crop development, *e.g*., just planted, in flower *etc*., and pesticides applied..

Compounds of interest included atrazine, its metabolites desethylated atrazine (DEA), desisopropyl atrazine (DIA), diaminochlorotriazine (DACT), and terbuthylazine plus simazine and acetochlor.

Non-corn growing sites had secchi disc readings ranging from 6.5 to 32 cm; pH ranged from 5.1 to 8.8; some of the control ponds were subject to drying (semi-permanent).

Corn-growing sites had pond surface areas ranging from 2,400 m^2 to 68,000 m^2; pH ranged from 7.2 - 10.8 and secchi disc readings ranging from 6.5 to 207 cm.

II. RESULTS and DISCUSSION: [All results discussed in this section and the next are those reported by the study authors. Although supplemental data are typically used in a qualitative manner only, EFED verified spreadsheet data and ran basic statistical analyses on the major study parameters. See attached appendix. If results differed in any substantive way, the difference was reported in the text below.]

Rainfall during November and December of sampling period were more than double (~150 to 200 mm) the long term average of approximately 100 mm precipitation. Air temperature over the study period were relatively consistent with 10-year minimums and maximums. High rainfall in November and December resulted in the majority of the area being planted in corn late in the season. Although corn is typically planted in November, the high rains delayed planting until the first week of January. Because of the heavy rains, all ponds overflowed their embankments until early January.

The following insecticides were used in the study's catchment areas: terbufos (79.2 to 460 g/ha), cypermethrin (6.6 g/100 m row), monocrotophos (1.5 L/ha). Herbicides included acetochlor, atrazine, terbuthylazine, S-metolaclhor, simazine, cyanazine and dimthenomid.

No herbicides were detected in any of the sediment samples analyzed. The level of detection for water samples ranged from 0.5 to 1.0 µg/L. The report records values of <0.6 µg/L as 0.25 µg/L.

Table 1 presents maximum residues of atrazine and its degradates DACT, DEA and DIA; while maximum atrazine residues were roughly an order of magnitude higher at experimental sites, reference sites had detectable residues of atrazine. Maximum residues of DACT, DEA and DIA were similar in both reference and experimental sites. Both DEA and DACT had peak residues in reference ponds that were generally higher than all of the experimental pond sites except E8. The highest residues of DACT in reference ponds occurred in early December, early to mid-February and mid- to late May; a relatively similar pattern occurred in experimental ponds. Additionally, terbuthylazine in reference pond C6 tended to remained relatively constant at around 1 µg/L until March when it spiked to over 2 µg/L; only experimental ponds E1, E6 and E8 had monthly terbuthylazine residues higher than reference pond C6. While the high rain events in November and December may have explained potential triazine contamination of reference sites, the spikes in February and May suggest that triazine contamination of reference sites was recurrent and may not have been associated with unusual weather. According to the report though, triazine residues in the reference ponds were likely due to wind effect and the authors speculate that the high rain events during the sampling period likely reduced atrazine levels and that "frogs living in these dams were undoubtedly exposed to much higher atrazine and other triazine levels than had been recorded during the present study.

Reference ponds contained soft water (total hardness range 18 - 25 mg/L as CaCO3) while experimental ponds all contained moderately hard water (total hardness range: 182 - 200 mg/L). Lead residues ranged form 0.08 to 0.09 mg/L in reference ponds while it was nondetectable in experimental sites. Reference pond also seemed to be an outlier in terms of the amount of siltation; while all other ponds (reference and experimental) had silica levels ranging from 4.3 - 8.7 mg/L, reference pond C6 contained 49.2 mg/L. Additionally, pond C6 had the highest chromium (100.5 mg/Kg) and titanium (0.6 mg/L) residues

Table 1. Maximum residues of atrazine, diaminochlorotriazine (DACT), desethylated atrazine (DEA), desisopropyl atrazine (DIA), terbuthylazine, simazine and acetochlor detected in surface water collected from reference sites (no corn grown) and experimental sites (corn grown) from November 2001 to March 2002.

Residue	Reference Sites (C1, C3, and C6) µg/L	Experimental Sites (E1, E3, E4, E6, and E8) µg/L
Atrazine	0.41 - 1.62	1.46 - 11.6
DACT	6.83 - 7.38	4.59 - 8.16
DEA	0.38 - 2.21	0.57 - 1.9
DIA	0.45 - 1.34	0.69 - 0.93
Terbuthylazine	2.39 - 2.79	1.82 - 5.30
Simizine	0.25 - 0.25	0.25 - 3.10
Acetochlor	0.25 - 0.25	0.25 - 1.0

E. STUDY DEFICIENCIES:

Atrazine, its degradates and terbuthylazine are present in reference sites. Study failed to provide data on other pesticides.

F. REVIEWER'S COMMENTS:

This study is the second phase of a project to assess the possible effects of exposure to atrazine and other triazines on African clawed frogs (*Xenopus laevis*) in their South African native range under field conditions. The objective of this phase of the study was to characterize the exposure of frog populations to atrazine and related triazines in surface waters of both reference (no corn/no atrazine-triazine use) and experimental (corn growing/atrazine-triazine use) ponds from November 2001 through early June 2002. During the second two months of the study, rainfall was roughly double the 10-year average and resulted in all of the earthen ponds overflowing their banks for as long as two months. Based on EFED's analysis of raw residue data, maximum atrazine residues in experimental ponds over the study period ranged from 1.46 to 11.6 ug/L while reference ponds ranged from 0.41 to 1.62 ug/L. However, the atrazine degradate diaminochlorotriazine (DACT) maximum residues ranged from 4.6 to 8.2 ug/L at experimental sites and form 6.8 to 7.4 ug/L at reference sites. Two other atrazine degradates, i.e., desethylated atrazine (DEA)and desisopropyl atrazine (DIA) showed roughly similar maximum residue levels in both experimental and reference sites. Maximum residues of terbuthylazine ranged from 1.8 to 5.3 ug/Lat experimental sites and from 2.4 to 2.8 ug/L at reference sites. According to the report, triazine residues in the reference ponds were likely due to wind effect; the authors speculate that the high rain events during the sampling period likely reduced atrazine levels and that "frogs living in these dams [ponds] were undoubtedly exposed to much higher atrazine and other triazine levels than had been recorded during the present study." Given that atrazine and/or its degradates were present in reference ponds at levels at times equivalent to some experimental pond sites and the authors concede that atrazine exposure prior to the winter floods was likely higher at all sites, it is unclear how the study can differentiate atrazine effects on frogs at reference and experimental sites. The high variability in exposure could potentially confound any attempt to document significant differences in effects.

G. CONCLUSIONS:

This study is the second phase of a project to assess the possible effects of exposure to atrazine and other triazines on African clawed frogs (*Xenopus laevis*) in their South African native range under field conditions. The objective of this phase of the study was to characterize the exposure of frog populations to atrazine and related triazines in surface waters of both reference (no corn/no atrazine-triazine use) and experimental (corn growing/atrazine-triazine use) ponds from November 2001 through early June 2002. During the second two months of the study, rainfall was roughly double the 10-year average and resulted in all of the earthen ponds overflowing their banks for as long as two months. Based on EFED's analysis of raw residue data, maximum atrazine residues in experimental ponds over the study period ranged from 1.46 to 11.6 ug/L while reference ponds ranged from 0.41 to 1.62 ug/L. However, the atrazine degradate diaminochlorotriazine (DACT) maximum residues ranged from 4.6 to 8.2 ug/L at experimental sites and form 6.8 to 7.4 ug/L at reference sites. Two other atrazine degradates, i.e., desethylated atrazine (DEA)and desisopropyl atrazine (DIA) showed roughly similar maximum residue levels in both experimental and reference sites. Maximum residues of terbuthylazine ranged from 1.8 to 5.3 ug/Lat experimental sites and from 2.4 to 2.8 ug/L at reference sites. According to the report, triazine residues in the reference ponds were likely due to wind effect; the authors speculate that the high rain events during the sampling period likely reduced atrazine levels and that "frogs living in these dams [ponds] were undoubtedly exposed to much higher atrazine and other triazine levels than had been recorded during the present study." Given that atrazine and/or its degradates were present in reference ponds at levels at times equivalent to some experimental pond sites and the authors concede that atrazine exposure prior to the winter floods was likely higher at all sites, it is unclear how the study can differentiate atrazine effects on frogs at reference and experimental sites. The high variability in exposure could potentially confound any attempt to document significant differences in effects.

ANOVA FOR CHEMICAL RESIDUES ACROSS SAMPLING AREAS 51

Obs	SITE	RESIDUE	TREAT	_TYPE_	_FREQ_	MEAN	STD	MIN	MAX
1	E1	ATRA	CORN GROW	0	24	3.07583	1.82021	0.05	6.78
2	E1	DACT	CORN GROW	0	24	0.94542	1.64876	0.05	5.20
3	E1	DEA	CORN GROW	0	24	0.68417	0.46684	0.05	1.66
4	E1	DIA	CORN GROW	0	24	0.39042	0.24017	0.05	0.93
5	E1	TERB	CORN GROW	0	24	2.84875	1.46945	0.05	5.30
6	E3	ATRA	CORN GROW	0	24	0.79667	0.32979	0.14	1.46
7	E3	DACT	CORN GROW	0	24	1.07333	1.81710	0.05	5.96
8	E3	DEA	CORN GROW	0	24	0.30375	0.16691	0.05	0.64
9	E3	DIA	CORN GROW	0	24	0.25375	0.19147	0.05	0.81
10	E3	TERB	CORN GROW	0	24	0.87667	0.79778	0.05	3.31
11	E4	ATRA	CORN GROW	0	24	0.56333	0.43697	0.15	1.96
12	E4	DACT	CORN GROW	0	24	1.04875	1.51311	0.05	4.59
13	E4	DEA	CORN GROW	0	24	0.19333	0.11698	0.05	0.57
14	E4	DIA	CORN GROW	0	24	0.23333	0.18647	0.05	0.69
15	E4	TERB	CORN GROW	0	24	0.56250	0.60623	0.05	1.82
16	E6	ATRA	CORN GROW	0	24	2.19500	1.39259	0.05	4.46
17	E6	DACT	CORN GROW	0	24	1.46625	2.01606	0.05	6.40
18	E6	DEA	CORN GROW	0	24	0.51542	0.46881	0.05	1.90
19	E6	DIA	CORN GROW	0	24	0.27875	0.22601	0.05	0.86
20	E6	TERB	CORN GROW	0	24	1.77917	1.17789	0.05	4.43
21	E8	ATRA	CORN GROW	0	24	3.33875	2.58250	0.05	11.60
22	E8	DACT	CORN GROW	0	24	1.40375	2.34236	0.05	8.16
23	E8	DEA	CORN GROW	0	24	0.45875	0.37055	0.05	1.25
24	E8	DIA	CORN GROW	0	24	0.40208	0.23805	0.05	0.88
25	E8	TERB	CORN GROW	0	24	1.61542	1.12659	0.05	3.48
26	R1	ATRA	REFERENCE	0	24	0.24542	0.10496	0.05	0.41
27	R1	DACT	REFERENCE	0	24	1.01792	1.67744	0.05	6.88
28	R1	DEA	REFERENCE	0	24	0.12958	0.12743	0.05	0.60
29	R1	DIA	REFERENCE	0	24	0.19958	0.11845	0.05	0.47
30	R1	TERB	REFERENCE	0	24	0.32333	0.64632	0.05	2.11
31	R3	ATRA	REFERENCE	0	24	0.18042	0.15502	0.05	0.57
32	R3	DACT	REFERENCE	0	24	1.39167	1.90289	0.05	7.38
33	R3	DEA	REFERENCE	0	24	0.26333	0.56395	0.05	2.21
34	R3	DIA	REFERENCE	0	24	0.24000	0.27233	0.05	1.34
35	R3	TERB	REFERENCE	0	24	0.55708	0.88616	0.05	2.79
36	R6	ATRA	REFERENCE	0	24	0.24083	0.16981	0.05	0.69
37	R6	DACT	REFERENCE	0	24	1.17708	1.69010	0.05	6.82
38	R6	DEA	REFERENCE	0	24	0.10000	0.08038	0.05	0.38
39	R6	DIA	REFERENCE	0	24	0.16542	0.09385	0.05	0.45
40	R6	TERB	REFERENCE	0	24	1.08042	0.50886	0.48	2.67

AVERAGE CHEMICAL RESIDUES IN CORN-GROWING AND REFERENCE SITES IN SOUTH AFRICA 52

Obs	TREAT	RESIDUE	_TYPE_	_FREQ_	MEAN	STD	MIN	MAX
1	CORN GROW	ATRA	0	5	1.99392	1.27471	0.56333	3.33875
2	CORN GROW	DACT	0	5	1.18750	0.23203	0.94542	1.46625
3	CORN GROW	DEA	0	5	0.43108	0.19018	0.19333	0.68417
4	CORN GROW	DIA	0	5	0.31167	0.07898	0.23333	0.40208
5	CORN GROW	TERB	0	5	1.53650	0.89039	0.56250	2.84875
6	REFERENCE	ATRA	0	3	0.22222	0.03628	0.18042	0.24542
7	REFERENCE	DACT	0	3	1.19556	0.18756	1.01792	1.39167
8	REFERENCE	DEA	0	3	0.16431	0.08703	0.10000	0.26333
9	REFERENCE	DIA	0	3	0.20167	0.03734	0.16542	0.24000
10	REFERENCE	TERB	0	3	0.65361	0.38766	0.32333	1.08042

NONPARAMETRIC COMPARISON OF CHEMICAL RESIDUES ACROSS SAMPLING SITES 53

---------- RESIDUE=ATRA ----------

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable UG
Classified by Variable SITE

SITE	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
E1	24	3463.00	2316.0	254.513858	144.291667
E3	24	2665.00	2316.0	254.513858	111.041667
E4	24	2085.00	2316.0	254.513858	86.875000
E6	24	3345.00	2316.0	254.513858	139.375000
E8	24	3504.00	2316.0	254.513858	146.000000
R1	24	1313.50	2316.0	254.513858	54.729167
R3	24	934.00	2316.0	254.513858	38.916667
R6	24	1218.50	2316.0	254.513858	50.770833

Average scores were used for ties.

Kruskal-Wallis Test

Chi-Square	109.1485
DF	7
Pr > Chi-Square	<.0001

Median Scores (Number of Points Above Median) for Variable UG
Classified by Variable SITE

SITE	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
E1	24	21.0	12.0	2.297278	0.875000
E3	24	19.0	12.0	2.297278	0.791667
E4	24	9.0	12.0	2.297278	0.375000
E6	24	22.0	12.0	2.297278	0.916667
E8	24	21.0	12.0	2.297278	0.875000
R1	24	0.0	12.0	2.297278	0.000000
R3	24	1.0	12.0	2.297278	0.041667
R6	24	3.0	12.0	2.297278	0.125000

Average scores were used for ties.

Median One-Way Analysis

Chi-Square	110.4219
DF	7
Pr > Chi-Square	<.0001

NONPARAMETRIC COMPARISON OF CHEMICAL RESIDUES ACROSS SAMPLING SITES 55

---------- RESIDUE=DACT ----------

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable UG
Classified by Variable SITE

SITE	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
E1	24	2130.00	2316.0	243.756869	88.75000
E3	24	2220.00	2316.0	243.756869	92.50000
E4	24	2202.00	2316.0	243.756869	91.75000
E6	24	2449.50	2316.0	243.756869	102.06250
E8	24	2430.00	2316.0	243.756869	101.25000
R1	24	2238.00	2316.0	243.756869	93.25000
R3	24	2473.50	2316.0	243.756869	103.06250
R6	24	2385.00	2316.0	243.756869	99.37500

Average scores were used for ties.

Kruskal-Wallis Test

Chi-Square	1.8154
DF	7
Pr > Chi-Square	0.9693

Median Scores (Number of Points Above Median) for Variable UG
Classified by Variable SITE

SITE	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
E1	24	11.0	12.0	2.297278	0.458333
E3	24	11.0	12.0	2.297278	0.458333
E4	24	12.0	12.0	2.297278	0.500000
E6	24	12.0	12.0	2.297278	0.500000
E8	24	14.0	12.0	2.297278	0.583333
R1	24	11.0	12.0	2.297278	0.458333
R3	24	13.0	12.0	2.297278	0.541667
R6	24	12.0	12.0	2.297278	0.500000

Average scores were used for ties.

Median One-Way Analysis

Chi-Square	1.3264
DF	7
Pr > Chi-Square	0.9877

NONPARAMETRIC COMPARISON OF CHEMICAL RESIDUES ACROSS SAMPLING SITES 57

---------- RESIDUE=DEA ----------

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable UG
Classified by Variable SITE

SITE	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
E1	24	3596.50	2316.0	251.896139	149.854167
E3	24	2741.00	2316.0	251.896139	114.208333
E4	24	2145.00	2316.0	251.896139	89.375000
E6	24	2974.00	2316.0	251.896139	123.916667
E8	24	2979.00	2316.0	251.896139	124.125000
R1	24	1435.50	2316.0	251.896139	59.812500
R3	24	1449.00	2316.0	251.896139	60.375000
R6	24	1208.00	2316.0	251.896139	50.333333

Average scores were used for ties.

Kruskal-Wallis Test

Chi-Square	75.5240
DF	7
Pr > Chi-Square	<.0001

Median Scores (Number of Points Above Median) for Variable UG
Classified by Variable SITE

SITE	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
E1	24	23.000000	12.0	2.277249	0.958333
E3	24	17.166667	12.0	2.277249	0.715278
E4	24	11.166667	12.0	2.277249	0.465278
E6	24	17.166667	12.0	2.277249	0.715278
E8	24	17.166667	12.0	2.277249	0.715278
R1	24	4.333333	12.0	2.277249	0.180556
R3	24	3.000000	12.0	2.277249	0.125000
R6	24	3.000000	12.0	2.277249	0.125000

Average scores were used for ties.

Median One-Way Analysis

Chi-Square	71.2969
DF	7
Pr > Chi-Square	<.0001

NONPARAMETRIC COMPARISON OF CHEMICAL RESIDUES ACROSS SAMPLING SITES 59

---------- RESIDUE=DIA ----------

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable UG
Classified by Variable SITE

SITE	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
E1	24	3075.50	2316.0	254.029783	128.145833
E3	24	2274.50	2316.0	254.029783	94.770833
E4	24	2097.50	2316.0	254.029783	87.395833
E6	24	2326.50	2316.0	254.029783	96.937500
E8	24	3114.50	2316.0	254.029783	129.770833
R1	24	1983.00	2316.0	254.029783	82.625000
R3	24	1967.00	2316.0	254.029783	81.958333
R6	24	1689.50	2316.0	254.029783	70.395833

Average scores were used for ties.

Kruskal-Wallis Test

Chi-Square	25.6164
DF	7
Pr > Chi-Square	0.0006

The NPAR1WAY Procedure

Median Scores (Number of Points Above Median) for Variable UG
Classified by Variable SITE

SITE	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
E1	24	18.00	12.0	2.261098	0.750000
E3	24	11.50	12.0	2.261098	0.479167
E4	24	10.50	12.0	2.261098	0.437500
E6	24	11.50	12.0	2.261098	0.479167
E8	24	18.00	12.0	2.261098	0.750000
R1	24	10.50	12.0	2.261098	0.437500
R3	24	9.50	12.0	2.261098	0.395833
R6	24	6.50	12.0	2.261098	0.270833

Average scores were used for ties.

Median One-Way Analysis

Chi-Square	19.4252
DF	7
Pr > Chi-Square	0.0070

NONPARAMETRIC COMPARISON OF CHEMICAL RESIDUES ACROSS SAMPLING SITES 61

---------- RESIDUE=TERB ----------

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable UG
Classified by Variable SITE

SITE	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
E1	24	3715.50	2316.0	252.107852	154.812500
E3	24	2158.50	2316.0	252.107852	89.937500
E4	24	1664.00	2316.0	252.107852	69.333333
E6	24	3049.50	2316.0	252.107852	127.062500
E8	24	2890.50	2316.0	252.107852	120.437500
R1	24	1112.00	2316.0	252.107852	46.333333
R3	24	1395.50	2316.0	252.107852	58.145833
R6	24	2542.50	2316.0	252.107852	105.937500

Average scores were used for ties.

Kruskal-Wallis Test

Chi-Square	77.4362
DF	7
Pr > Chi-Square	<.0001

Median Scores (Number of Points Above Median) for Variable UG
Classified by Variable SITE

SITE	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
E1	24	21.0	12.0	2.297278	0.875000
E3	24	11.0	12.0	2.297278	0.458333
E4	24	7.0	12.0	2.297278	0.291667
E6	24	18.0	12.0	2.297278	0.750000
E8	24	16.0	12.0	2.297278	0.666667
R1	24	3.0	12.0	2.297278	0.125000
R3	24	6.0	12.0	2.297278	0.250000
R6	24	14.0	12.0	2.297278	0.583333

Average scores were used for ties.

Median One-Way Analysis

Chi-Square	46.4236
DF	7
Pr > Chi-Square	<.0001

ANOVA FOR CHEMICAL RESIDUES ACROSS SAMPLING AREAS 63

---------- RESIDUE=ATRA ----------

The ANOVA Procedure

Class Level Information

Class	Levels	Values
SITE	8	E1 E3 E4 E6 E8 R1 R3 R6

Number of observations 192

Dependent Variable: UG

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	297.3024703	42.4717815	27.66	<.0001
Error	184	282.5627875	1.5356673		
Corrected Total	191	579.8652578			

R-Square	Coeff Var	Root MSE	UG Mean
0.512710	93.20732	1.239220	1.329531

Source	DF	Anova SS	Mean Square	F Value	Pr > F
SITE	7	297.3024703	42.4717815	27.66	<.0001

Levene's Test for Homogeneity of UG Variance
ANOVA of Squared Deviations from Group Means

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
SITE	7	890.5	127.2	4.85	<.0001
Error	184	4830.0	26.2501		

Bartlett's Test for Homogeneity of UG Variance

Source	DF	Chi-Square	Pr > ChiSq
SITE	7	347.0	<.0001

Bonferroni (Dunn) t Tests for UG

NOTE: This test controls the Type I experimentwise error rate, but it generally has a higher Type II error rate than REGWQ.

Alpha	0.05
Error Degrees of Freedom	184
Error Mean Square	1.535667
Critical Value of t	3.17001
Minimum Significant Difference	1.134

Means with the same letter are not significantly different.

Bon Grouping	Mean	N	SITE
A	3.3388	24	E8
A			
B A	3.0758	24	E1
B			
B	2.1950	24	E6
C	0.7967	24	E3
C			
C	0.5633	24	E4
C			
C	0.2454	24	R1
C			
C	0.2408	24	R6
C			
C	0.1804	24	R3

ANOVA FOR CHEMICAL RESIDUES ACROSS SAMPLING AREAS 67

---------- RESIDUE=DACT ----------

The ANOVA Procedure

Class Level Information

Class	Levels	Values
SITE	8	E1 E3 E4 E6 E8 R1 R3 R6

Number of observations 192

Dependent Variable: UG

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	6.8600062	0.9800009	0.29	0.9577
Error	184	624.5005417	3.3940247		
Corrected Total	191	631.3605479			

R-Square	Coeff Var	Root MSE	UG Mean
0.010865	154.7464	1.842288	1.190521

Source	DF	Anova SS	Mean Square	F Value	Pr > F
SITE	7	6.86000625	0.98000089	0.29	0.9577

Levene's Test for Homogeneity of UG Variance
ANOVA of Squared Deviations from Group Means

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
SITE	7	158.5	22.6424	0.45	0.8688
Error	184	9242.4	50.2302		

Bartlett's Test for Homogeneity of UG Variance

Source	DF	Chi-Square	Pr > ChiSq
SITE	7	6.2865	0.5067

ANOVA FOR CHEMICAL RESIDUES ACROSS SAMPLING AREAS 70

---------- RESIDUE=DACT ----------

The ANOVA Procedure

Bonferroni (Dunn) t Tests for UG

NOTE: This test controls the Type I experimentwise error rate, but it generally has a higher Type II error rate than REGWQ.

Alpha	0.05
Error Degrees of Freedom	184
Error Mean Square	3.394025
Critical Value of t	3.17001
Minimum Significant Difference	1.6859

Means with the same letter are not significantly different.

Bon Grouping	Mean	N	SITE
A	1.4663	24	E6
A			
A	1.4038	24	E8
A			
A	1.3917	24	R3
A			
A	1.1771	24	R6
A			
A	1.0733	24	E3
A			
A	1.0488	24	E4
A			
A	1.0179	24	R1
A			
A	0.9454	24	E1

ANOVA FOR CHEMICAL RESIDUES ACROSS SAMPLING AREAS 71

---------- RESIDUE=DEA ----------

The ANOVA Procedure

Class Level Information

Class	Levels	Values
SITE	8	E1 E3 E4 E6 E8 R1 R3 R6

Number of observations 192

Dependent Variable: UG

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	7.03822500	1.00546071	8.40	<.0001
Error	184	22.01816667	0.11966395		
Corrected Total	191	29.05639167			

R-Square	Coeff Var	Root MSE	UG Mean
0.242226	104.4958	0.345925	0.331042

Source	DF	Anova SS	Mean Square	F Value	Pr > F
SITE	7	7.03822500	1.00546071	8.40	<.0001

Levene's Test for Homogeneity of UG Variance
ANOVA of Squared Deviations from Group Means

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
SITE	7	2.2597	0.3228	2.39	0.0233
Error	184	24.8825	0.1352		

Bartlett's Test for Homogeneity of UG Variance

Source	DF	Chi-Square	Pr > ChiSq
SITE	7	140.8	<.0001

ANOVA FOR CHEMICAL RESIDUES ACROSS SAMPLING AREAS 74

---------- RESIDUE=DEA ----------

The ANOVA Procedure

Bonferroni (Dunn) t Tests for UG

NOTE: This test controls the Type I experimentwise error rate, but it generally has a higher Type II error rate than REGWQ.

Alpha	0.05
Error Degrees of Freedom	184
Error Mean Square	0.119664
Critical Value of t	3.17001
Minimum Significant Difference	0.3166

Means with the same letter are not significantly different.

Bon Grouping			Mean	N	SITE
	A		0.68417	24	E1
	A				
B	A		0.51542	24	E6
B	A				
B	A	C	0.45875	24	E8
B		C			
B	D	C	0.30375	24	E3
B	D	C			
B	D	C	0.26333	24	R3
	D	C			
	D	C	0.19333	24	E4
	D				
	D		0.12958	24	R1
	D				
	D		0.10000	24	R6

ANOVA FOR CHEMICAL RESIDUES ACROSS SAMPLING AREAS 75

---------- RESIDUE=DIA ----------

The ANOVA Procedure

Class Level Information

Class	Levels	Values
SITE	8	E1 E3 E4 E6 E8 R1 R3 R6

Number of observations 192

Dependent Variable: UG

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	1.21022500	0.17288929	4.14	0.0003
Error	184	7.67894167	0.04173338		
Corrected Total	191	8.88916667			

R-Square	Coeff Var	Root MSE	UG Mean
0.136146	75.54545	0.204287	0.270417

Source	DF	Anova SS	Mean Square	F Value	Pr > F
SITE	7	1.21022500	0.17288929	4.14	0.0003

Levene's Test for Homogeneity of UG Variance
ANOVA of Squared Deviations from Group Means

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
SITE	7	0.0781	0.0112	1.07	0.3813
Error	184	1.9095	0.0104		

Bartlett's Test for Homogeneity of UG Variance

Source	DF	Chi-Square	Pr > ChiSq
SITE	7	35.1037	<.0001

ANOVA FOR CHEMICAL RESIDUES ACROSS SAMPLING AREAS 78

---------- RESIDUE=DIA ----------

The ANOVA Procedure

Bonferroni (Dunn) t Tests for UG

NOTE: This test controls the Type I experimentwise error rate, but it generally has a higher Type II error rate than REGWQ.

Alpha	0.05
Error Degrees of Freedom	184
Error Mean Square	0.041733
Critical Value of t	3.17001
Minimum Significant Difference	0.1869

Means with the same letter are not significantly different.

Bon Grouping	Mean	N	SITE
A	0.40208	24	E8
A			
A	0.39042	24	E1
A			
B A	0.27875	24	E6
B A			
B A	0.25375	24	E3
B A			
B A	0.24000	24	R3
B A			
B A	0.23333	24	E4
B			
B	0.19958	24	R1
B			
B	0.16542	24	R6

ANOVA FOR CHEMICAL RESIDUES ACROSS SAMPLING AREAS 79

---------- RESIDUE=TERB ----------

The ANOVA Procedure

Class Level Information

Class	Levels	Values
SITE	8	E1 E3 E4 E6 E8 R1 R3 R6

Number of observations 192

Dependent Variable: UG

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	7	118.3988167	16.9141167	18.58	<.0001
Error	184	167.4817500	0.9102269		
Corrected Total	191	285.8805667			

R-Square	Coeff Var	Root MSE	UG Mean
0.414155	79.14758	0.954058	1.205417

Source	DF	Anova SS	Mean Square	F Value	Pr > F
SITE	7	118.3988167	16.9141167	18.58	<.0001

Levene's Test for Homogeneity of UG Variance
ANOVA of Squared Deviations from Group Means

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
SITE	7	65.4317	9.3474	5.16	<.0001
Error	184	333.2	1.8111		

Bartlett's Test for Homogeneity of UG Variance

Source	DF	Chi-Square	Pr > ChiSq
SITE	7	41.4268	<.0001

ANOVA FOR CHEMICAL RESIDUES ACROSS SAMPLING AREAS 82

---------- RESIDUE=TERB ----------

The ANOVA Procedure

Bonferroni (Dunn) t Tests for UG

NOTE: This test controls the Type I experimentwise error rate, but it generally has a higher Type II error rate than REGWQ.

Alpha	0.05
Error Degrees of Freedom	184
Error Mean Square	0.910227
Critical Value of t	3.17001
Minimum Significant Difference	0.8731

Means with the same letter are not significantly different.

Bon Grouping		Mean	N	SITE
A		2.8488	24	E1
B		1.7792	24	E6
B				
C B		1.6154	24	E8
C B				
C B	D	1.0804	24	R6
C	D			
C	D	0.8767	24	E3
	D			
	D	0.5625	24	E4
	D			
	D	0.5571	24	R3
	D			
	D	0.3233	24	R1