Final Report to the Government of Japan

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on the

USDA MATURITY STUDY:

Determining the Relationship between Chronological and Physiological Age in the U.S. Fed-Beef Population

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RESEARCH TEAM:
Mr. Barry Carpenter, Deputy Administrator, AMS, Livestock and Seed Program (AMS/LS)
Mr. Martin O'Connor, Chief, Standardization Branch, AMS/LS
Dr. Michael Feil, Chief, Statistics Branch, AMS
Dr. Justin Ransom, Marketing Specialist, AMS/LS
Dr. Jimmy Wise, Marketing Specialist, AMS/LS
Dr. Lawrence Yates, Marketing Specialist, AMS/LS
Dr. Mohammed Koohmarie, Meat Animal Research Center, Agricultural Research Service
Dr. Jim Hrubovcak, Economist, Office of the Chief Economist
Dr. Keith Belk, Professor, Colorado State University
Dr. John Scanga, Assistant Professor, Colorado State University
Dr. Gary Smith, Distinguished University Professor, Colorado State University

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PREFACE: As stated in the ANNEX, of the "Terms of Reference: Physiological Maturity of Beef Cattle Carcasses", the purpose of this study was to determine an appropriate end-point physiological maturity score that will assure exclusion of steers and heifers with a chronological age of 21 months and older from a certification program for export to Japan. The following report utilizes a deterministic approach to data analysis as agreed upon by the Government of Japan (GOJ) and the United States Government (USG).

INTRODUCTION

On October 23, 2004, the USG and the GOJ decided that USG would implement an interim program for beef and beef products that would result in restoration of beef trade. That program, known as a Beef Export Verification (BEV) Program, requires that U.S. exporting companies must comply with specified requirements which the U.S. Department of Agriculture's (USDA) Agricultural Marketing Service (AMS) will certify. One of the specified requirements is that only beef and beef products from animals 20 months of age or younger may be sold to Japan. During discussions with GOJ, the USG described the production system for the U.S. fed beef population and the necessity for cattle to grow efficiently—and therefore be harvested at youthful ages—in order to maximize profitability. Also, the USG explained that documentation proving chronological age of fed cattle currently is available for only an estimated 5% of the U.S. fed beef population; therefore, other technical options needed to be reviewed in order to scientifically

prove the age of the cattle. Following this discussion, it was decided to conduct a 45-day study to evaluate the relationship between physiological and chronological age of beef cattle. The short time frame of this study resulted in several challenges, but the greatest challenge was finding older animals of exact known ages to be used in the study.

After release of the joint press statement that addressed criteria for restoring trade in beef and beef products, the Agricultural Marketing (AMS) Service, Livestock and Seed (LS) Program conducted a study in which steers and heifers of known ages were identified and evaluated as they were harvested and chilled. The United States Standards for Grades of Carcass Beef (effective January 31, 1997) were used as the criteria for assessing physiological maturity. The purpose of this study was to establish an overall physiological maturity score (e.g., A^{20}, A^{30}, A^{40}, A^{50}, et cetera) that would effectively allow sortation of carcasses of steers and heifers that are younger than 21 months of age *versus* those that are 21 months of age or older for purposes of qualifying products for export to Japan via the mutually agreeable BEV.

The study was conducted, data was analyzed, and a final report was generated for presentation to the GOJ on January 19, 2005 in Tokyo.

USDA, AMS, MEAT GRADING AND CERTIFICATION

Each year, approximately 160 USDA/AMS graders evaluate the physiological maturity and other grade factors of approximately 27 million carcasses. Of the steers and heifers graded, it is estimated that approximately 90% are 20 months of age and younger, and only outliers are older than 24 months of age. The official standards for grades of steer and heifer beef were revised in 1965 to place added emphasis on physiological skeletal maturity (ossification) in grading carcasses. As cattle advance in chronological age, physiological maturity causes the amount of

collagen cross-linkage in muscle to increase, resulting in tough meat; therefore, cattle with advanced physiological skeletal maturity also have advanced physiological muscle maturity, and thus should be excluded from the premium grades of USDA Prime, Choice, Select, and Standard. Since physiological maturity was added to the grade standards, physiological maturity has been used to classify maturity and to assist graders in the determination of the quality (i.e., expected palatability of the cooked lean product) of beef carcasses. At the time of grading, 36 - 48 hours after slaughter, USDA/AMS graders evaluate each carcass in order to determine the quality grade. USDA/AMS graders evaluate both physiological maturity and other factors to assist them in determining the final USDA Quality Grade. This system allows graders to identify and segregate beef carcasses according to quality differences within the U.S. beef population for purposes of establishing value, which ultimately then is used in the in the marketing system to send economic signals upstream and downstream in the marketing chain; resulting in higher quality beef in a value-driven marketing system. Use of pictures depicting critical evaluation decision points are used by USDA/AMS graders to standardize and assure accuracy and precision of carcass evaluation and quality grade assignment.

Since 1985, USDA/AMS has routinely conducted grading audit reviews to maintain accuracy of grade placement across the industry. In the Meat Grading and Certification (MGC) Branch, extensive training is conducted during the first two years of employment and stringent qualification requirements are established to insure accuracy of grade placement by graders-in-training, journeyman graders (graders with at least two years of experience), and expert graders (supervisors). In the current grading system, there is one supervisor for every nine graders, which demonstrates the level of hands-on commitment the AMS/LS/MGC Branch possess, in

order to assure an accurate evaluation and application of the official USDA Quality and Yield Grades (Appendix A).

Reviews (internal and by an independent third party) are conducted to characterize the current carcass population and to evaluate performance of on-line graders. These reviews are conducted randomly at each major processing facility where graders are stationed. In addition, data from these reviews provide an accurate description of the approximately 475 million cattle slaughtered since 1985. Since these intensive reviews began, the accuracy of all factors affecting accurate placement (assignment) of carcass grades on more than 30,000 carcasses have been evaluated for both USDA Quality and Yield Grades. Statistical evaluation of grader performance indicates that national grading accuracy is 94 % with respect to correct grade assignment. In a recent national correlation on overall maturity, evaluators agreed with the expert panel 98% of the time at A^{50} and 99% of the time at A^{60} (Figure 1). This determination was made within a 20 unit (degree) increment from official maturity scores assigned to carcasses by a panel of experts for each carcass. The average grader difference between the official committee and the individual grader was 3 maturity units (degrees) lower (per carcass) than the official (Figure 2) over a range from A^{50} to C^{00}. When taking into account the range in maturity scores under consideration with this review, the supervisory scores for overall maturity tended to overestimate overall maturity at A^{50}, essentially equaled zero at A^{60} and slightly underestimated maturity at A^{70} (Figure 2). A similar trend was noticed in another national correlation on overall maturity one year earlier. Once a beef export verification program is in place, grader training and accreditation programs will be conducted to assure accuracy of graders in determining compliance with the established overall maturity threshold.

The skeletal maturity classification system segregates cattle into 5 different maturity groups; A (youngest), B, C, D, and E (oldest). Carcass maturity is determined by evaluating the size, shape, and ossification of the bones and cartilages along the split vertebral column of the carcass. Special attention is paid to the split chine bones, as the greatest difference in A maturity carcasses begin to occur along the split chine surface. In split chine bones, visually-evident changes in ossification (i.e., the degree to which cartilage has converted to bone) occur at an earlier stage of maturity in the posterior portion of the vertebral column (sacral vertebrae) and at progressively later stages of maturity in the lumbar, thoracic, and other anterior vertebrae. Changes in ossification occur in the cartilaginous tips of *spinous processes* (chine bones) located on the apex (dorsal extremity)of split thoracic vertebrae; these changes are especially useful in evaluating physiological maturity and are referred to frequently in the grade standards. The size and shape of the rib bones also are important considerations in evaluating differences in maturity.

In the very youngest A maturity carcasses (A^{00}), cartilage on the ends of the chine bones show no ossification, cartilage is evident on all of the vertebrae of the spinal column, and the sacral vertebrae show distinct separation. In addition, split vertebrae usually are soft and porous and very red in color. In such carcasses, rib bones are relatively round and have only a slight tendency toward flatness. However, the specifications for skeletal ossification in the oldest of A maturity carcasses consists of carcasses that have slightly red and slightly soft chine bones plus evidence of ossification in cartilage on the ends of the thoracic vertebrae. In addition, sacral vertebrae will be completely fused (i.e., no differentiation among individual vertebra), cartilage on the ends of lumbar vertebrae will be nearly completely ossified, and rib bones will become slightly wide and slightly flat.

For carcasses to be considered by an evaluator as A^{40}, they must have: (1) some evidence of cartilage in all vertebrae, (2) distinct separation of the sacral vertebrae and caps that show considerable evidence of cartilage, (3) caps on the lumbar vertebrae that tend to be partially ossified, (4) no ossification of the thoracic vertebrae, (5) split vertebrae surfaces that tend to be soft, porous and red, (6) ribs that have some tendency toward flatness, and (7) lean texture that is very fine, and lean that is light red in color.

On the other hand, for carcasses to be evaluated as A^{50}, they must have: (1) some evidence of cartilage in all vertebrae, (2) separation of the sacral vertebrae caps show evidence of cartilage, (3) caps on the lumbar vertebrae that tend to be nearly moderately ossified, (4) no ossification of the thoracic vertebrae, (5) split vertebrae surfaces that tend to be moderately soft, porous and moderately red, (6) ribs that have some tendency toward flatness and narrow, and (7) lean texture that is very fine and lean that is moderately light red in color (Images 1 and 2; Table 1).

U.S. BEEF INDUSTRY

There are 796,436 beef herds and 91,989 dairy herds in the U.S. which currently account for a national inventory consisting of 32.9 million beef cows, 9.0 million dairy cows, and 29 million feeder calves. About 90% of beef cow herds produce less than 100 head, but account for only 50% of total U.S. cattle inventory. An estimated 2,100 feedlots are responsible for generating 87% of the cattle harvested, and only 1,781 companies have the capacity to feed more than 1,000 cattle at any given time—with total one-time capacity for about 14 million head.

Currently, about 60 of the 706 total beef packing plants harvest about 17.2 million young (12 to 18 months of age) steers and 11.1 million young heifers each year. The largest four packing firms account for about 81% and approximately 29 plants operated by the largest five firms

account for approximately 88% to 90%, of the total number of cattle harvested each year. The typical large packing plant harvests in excess of 5,000 cattle each day. Approximately 6.7 million culled beef and dairy cows, bulls, and stags are harvested at non-fed-beef processing facilities each year. Thus, about 35 million cattle are harvested each year in the U.S., of which only a small fraction (about 19%) are culled animals, and of which an even smaller number are of a chronological age to have contracted the Bovine Spongiform Encephalopathy (BSE) infectious agent and show clinical signs of the disease. Based on current international scientific knowledge, such a population should pose an extremely low risk—even if BSE is present in the population—of transmitting a food safety threat.

The U.S. beef industry is structured like an hour-glass; as described above, cattle from many hundreds of thousands of producers flow to a much smaller number of feedlots, and then to even fewer numbers of packing plants. Following harvest, numbers of further-processors, distributors, traders, retailers, foodservice establishments and, finally, consumers are vastly larger. Thus, product flow through the industry is such that constriction and consolidation of product occurs most at the packing level, and although control of food safety is exerted across all sectors of the industry, controls to prevent transmission of infectious agents that are not susceptible to cooking temperatures are most efficient and effective when exerted stringently in packing plants.

Mainstream beef production systems in the U.S. differ substantially from beef production systems employed by producers in Japan, resulting in generally younger cattle at harvest in the U.S. Typical fed-cattle production flow pathways are provided in Appendix C. Generally, cattle are weaned from their mothers when they are less than 8 months of age, pastured or fed for an additional 4 to 11 months, and fed a high-concentrate diet in a finishing feedlot for 100 to 150 days.

Rarely are mainstream fed cattle harvested when chronologically 21 months of age and older. Available data suggest that the mean age of fed cattle at harvest is about 16 to 17 months of age, and 97% are harvested before 20 months of age.

Management pressures (health and nutritional) in the U.S. beef production system, for economic reasons, result in more efficient growth performance and earlier weaning of calves as time proceeds. Therefore, there is no reason to believe that cattle will not continue to be younger at harvest in the future; it certainly is unlikely that any incentives exist to promote production of older cattle at harvest in the future.

U.S. ANIMAL HEALTH POLICIES

Establishment of BSE mitigation procedures in the United States began in 1989 following scientific recognition of the disease as an infectious agent among cattle.

The first "fire wall" erected against transmission of BSE in the U.S. cattle population, implemented in 1989, prevented importation of animal feeds, animals, and some animal products from countries with confirmed cases of BSE.

In 1990, USDA-Animal Plant Health Inspection Service (APHIS) initiated a surveillance testing program which, for 13 years, yielded no positive cases of the disease. The surveillance testing program evaluated 20,526 cattle in 2003 (47 times the Organization International Epizooties (OIE)'s recommended surveillance level).

Following discovery of the single Canadian-born case of BSE in the U.S., a targeted surveillance program was implemented on June 1, 2004 that will result in the testing of 268,000 high risk (> 30 MOA, non-ambulatory, exhibiting neurological disorders) cattle upon completion, plus an additional 20,000 low risk cattle. This new surveillance program is designed to provide a 99% level of confidence that the disease will be diagnosed if it occurs at a rate of 1 positive BSE animal in 10 million cattle. To date (January 10, 2005), of 178,336 total animals tested, no additional BSE-infected cattle have been detected in the U.S.

In 1997, the U.S. Food and Drug Administration, which is responsible for regulating the rendering industry and animal feed manufacturers, implemented a ban on feeding mammalian-derived feedstuffs to ruminant animals. Because epidemiological evidence from Europe indicates that BSE is primarily spread to cattle via consumption of feedstuffs that are contaminated with the infectious agent, monitoring and compliance enforcement by FDA-CVM of the regulated feed ban has generated substantial historical documentation that more than 99.4% of feed manufacturers comply with the regulation (the 0.6% non-compliant manufacturers were cited for minor infractions not posing a threat to animal health). The U.S. mammalian-to-ruminant feed ban is largely responsible for the fact that no U.S. born cattle have ever been diagnosed with BSE.

In addition to the regulatory requirements of FDA, and in order to establish compliance documentation insuring that no beef is produced from cattle that may have received meat and bone meal, many beef packers require producer signed affidavits prior to purchase, affirming that no mammalian derived proteins are fed to purchased cattle. Because, legally, providing misleading information on a signed affidavit in the U.S. violates labeling laws and requirements, it is

considered by the courts to be a felony. This provides strong incentive to cattle feeders to insure that cattle receive no prohibited feedstuffs.

In an effort to be proactive in keeping BSE out of the U.S., USDA commissioned Harvard University to conduct a BSE risk analysis in 2001. Results suggested that the risk of a BSE "outbreak" in the U.S. was highly unlikely. Following announcement by the Canadian government on May 26, 2003, that a Canadian-born cow had been confirmed BSE-positive, USDA commissioned a second Harvard Risk Analysis, adding to the model the fact that BSE had been diagnosed in a cow in North America. This report, once again, indicated that risk of BSE spreading in the U.S. cow herd, even if contaminated ruminant feeds or infected animals entered the U.S., was highly remote and that the U.S., in a worst-case scenario, should eradicate BSE within 20 years. Following the December 23, 2003 discovery in Washington State of a single case of BSE in a Canadian-born cow, along with the subsequent implementation of additional "firewalls" to prevent transmission of the disease, the risk of BSE spreading in the U.S. has once again been assessed to be low by Harvard.

Following discovery of the U.S. BSE case in December 2003, the Secretary of Agriculture commissioned an International Expert Subcommittee review of U.S. policies regarding BSE. The Expert Subcommittee made several positive comments regarding U.S. actions, (a) acknowledging the U.S. science-based approach to policy formulation, (b) commending the thorough epidemiological investigation and concurring with conclusions of the investigation, (c) stating that tracing and recall of the rendered meat and bone meal that may have been contaminated was appropriate and effective, (d) confirming actions to prohibit air injection/retraction stunning

methods, (e) commending prohibition of non-ambulatory cattle from entering the human food supply, (f) recommending adoption of rapid BSE screening tests to increase surveillance efforts, (g) commending efforts to accelerate implementation of a national U.S. animal identification and traceability program, and (h) recognizing U.S. containment and proper destruction of Specific Risk Materials.

The assessment by the International Expert Subcommittee was in general agreement with the Harvard Risk Analysis in that (a) the U.S. may find more cases of BSE, but not on a widespread basis, (b) BSE will not amplify in the U.S. and the country is on a path to elimination of the disease, (c) additional specific measures could hasten elimination (most of these subsequently were implemented by USDA), and (d) some differences in specific assumptions or interpretations of scientific knowledge between the Harvard Risk Analysis and the International Expert Subcommittee require further understanding and scientific investigation.

U.S. FOOD SAFETY POLICIES

Additional public health policies were implemented by the U.S. on January 12, 2004 to further reduce risk of transmitting BSE to humans through dissemination of contaminated, or potentially-infectious, beef items in the human food chain.

The first of these preventative regulations enacted by USDA- Food Safety Inspection Service (FSIS), excluded non-ambulatory (downer) cattle from the human food supply. Any animal not able to walk at the time of antemortem inspection is condemned and not allowed to enter the food supply.

Secondly, FSIS mandated that all parts of a carcass derived from an animal being included in the BSE surveillance program test must be retained from the food supply until results of the test are returned. In practice, animals identified for testing in the surveillance program are not allowed into processing plants, but rather are diverted to inedible rendering facilities at the time of testing and are not included in the human food supply, regardless of the BSE surveillance test results.

The third mitigation measure banned use of air-injection stunning practices and equipment as these devices can relocate central nervous system tissue into the circulatory system causing distribution within the carcass. This regulation was included in the January 12, 2004 notice for purposes of officially banning the practice and to insure compliance among countries which export beef products to the U.S.; however, the U.S. beef industry had recognized potential for such contamination and voluntarily eliminated these devices and practices during the late 1990s.

The final piece of the January 12, 2004 regulation required removal and control of disposition of Specified Risk Materials (SRMs) from beef animals. USDA-FSIS stipulated that spinal cord, vertebral column, brain, skull, eyes, trigeminal ganglia, and dorsal root ganglia must be removed and properly disposed of (e.g., inedible rendering) from cattle that are 30 months of age or older; and tonsils and distal ileum (via removal of the entire small intestine) must be discarded from all cattle regardless of age. Chronological age of cattle is determined via dentition or known date-of-birth (by individual animal or by lot of cattle) so that carcasses and carcass parts generated from animals 30 months of age or older are identified and segregated throughout production. All SRMs are removed from the human food chain, prevented from being processed in advanced meat recovery systems, and are disposed of through inedible rendering—most packing plants accomplish appropriate disposition of SRMs by placing them under

"lock-and-key." Plants also were required to develop and implement Standard Operating Procedures preventing cross-contamination of carcasses and carcass parts with SRM tissues.

In practice, most SRM tissues are discarded by packers irrespective of cattle age. The only exception to this is the vertebral column; when bone-in rib or loin subprimals from cattle younger than 30 months of age are purchased by packing plant customers, the vertebral column—although "chined" with a saw to remove the vertebral foreman—remains intact with the resulting subprimal cut.

MATERIALS

The federally inspected beef slaughter population in nine packing plants was surveyed during a 4-week period (November 2004). Information was collected from packing companies that slaughtered source verified cattle that were of known age and agreed to allow data collection in their plants. Source verification data were obtained for a total of 4,493 cattle from which sample carcasses were derived during the course of normal packing plant operations. Production information concerning cattle/carcasses included in this study was, at a minimum, date of birth (exact or in a birth date interval). Additional live animal information included: (1) gender (steers vs. heifers); (2) breed (predominately British, predominately Continental European, *Bos indicus* influenced, and Holstein); (3) management (backgrounded vs. stocker cattle); and (4), growth promotion regimen (implanted vs. non-implanted). The physiological maturity (bone and lean maturity) of each carcass was assigned by USDA Meat Grading and Certification staff that had expert knowledge of the interpretation and application of the United States Standards for Grades

of Carcass Beef (effective January 31, 1997). All evaluations were performed in accordance with this standard.

The chronological age of each animal (from which sample carcasses were derived) was measured in days. Animal age was calculated by subtracting the slaughter date from the earliest possible date of birth, and then dividing the resulting difference by 30 (a month is defined as being comprised of 30 days). The birth date interval was calculated using the difference between the earliest and latest possible dates of birth. When cattle born within a birth date interval were used in this study, all of the cattle in the birth date interval were assigned the oldest possible birth date available in the interval; thus, the youngest animals in the group could have been up to two months younger than their estimated birth date. The birth date intervals to be used consisted of a time-frame of 62 days or less; resulting in a sample population of 3,338 total carcasses that met the protocol for the study. Subsequent analysis techniques only were applied to these resulting data (n = 3,338).

The chronological age of cattle from which sample carcasses were derived formed the basis for stratifying the sample population into the fixed effect classifications of (a) those carcasses from cattle that were less 21 months of age, and (b) those carcasses from cattle that were equal to, or greater than, 21 months of age. Data were further divided into two groups; those from carcasses of cattle for which exact birth dates were known, and those from carcasses of cattle for which only a birth date interval (62-day window) was available.

RESULTS AND DISCUSSION

The overall sample population consisted of 4,493 carcasses. Of these, 43.9% were steers,

50.7% were heifers, and 5.4% were of mixed gender. Examination by breed showed that 35.5% were British, 0.5% were Continental, 7.7% were Holstein, 56% were a British-Continental cross and 0.4% were *Bos indicus*. When the distribution of the different production factors was examined, no information was available for 20.5% of the carcasses, 45.2% were from cattle that were backgrounded, 16.7% of the cattle were grazed on grass/wheat, and 7.7% were calf fed. Stratification of the sample population by growth promotion regime indicated that 81.8% were treated with growth-promoting implants, while the remaining 18.3% did not receive implants. Percentages of carcasses with birth date intervals of 0 to 60 days, 61 to 90 days, and greater than 90 days was 37.6, 52.8, and 9.8%, respectively (Tables, 2-6, respectively).

Data obtained from the reduced sample population (n = 3,338) were analyzed independent of the total sample and ranked by chronological age group and physiological maturity; these data included information from carcasses for which exact birth dates were known, and from carcasses for which only a birth date interval (62-day duration) was available. In the smaller sub-sample population of cattle from known birth date intervals, 43.5% were steers and 50.6% were heifers, and the mixed (both sexes in a group) cattle were 5.9%. The distribution of breeds remained similar to the larger population, with 43.8, 0.6, 10.4, 44.7, and 0.5% of the sub-sample were of British, Continental, Holstein, British X Continental and *Bos Indicus* origin, respectively. In addition, the production information in the sub-sample population (n=3,338) (Tables 7-9, respectively) resembled that of the larger population (N=4,493).

Rank ordering of the sub-sample by maturity score indicated that an overall maturity score of A^{40} would serve as an appropriate threshold for insuring that all beef and beef product exported to Japan would be obtained from cattle 20 months of age or younger. While this approach was

excessively conservative considering other U.S. firewalls that are in place to prevent transmission of BSE, and little to no prevalence of BSE in the national herd, it does provide the greatest level of assurance that no products originating from cattle greater than 21 months of age will be allowed into Japan. This was demonstrated by the 250 head (Tables 10 and 11) of cattle 21 months of age and older that all had maturity scores greater than A^{40}. In addition, 92.3% (3,082 out of 3,338) of the population of cattle in this data set had an overall maturity score greater than A^{40}, thus effectively eliminating a large majority of the entire available population. Furthermore, data from the present 3,338-head study indicated that no carcasses assessed as A^{40} in physiological maturity resulted from cattle older than 17 months of age (Figure 3).

Nonparametric analysis indicates that there is a statistically significant difference between the ages of carcasses from cattle having an overall maturity score of A^{40} and A^{50} ($P < 0.05$). In Figure 4, it is clear that there is a difference in the age distribution of the carcasses with these two maturity scores.

Additional figures (Figures 5-9) provide frequency distributions for carcasses evaluated within each overall maturity classification (e.g., A^{30}, A^{40}, A^{50}, et cetera). The figures of chronological ages by physiological maturity score suggest an overall maturity score of A^{40} be utilized to achieve certainty that those cattle 21 months of age and older will be effectively segregated. A maturity score of A^{40} provides the assurance that no beef from cattle equal to or greater than 21 months of age would be exported to Japan.

RISK ASSESSMENT

To date, in the United States, 178,336 head of cattle have been tested in the enhanced surveillance program that began in June 2004. Of the cattle tested, most (if not all) were older than

24 months of age; therefore, since no animals have tested positive for BSE, it will be difficult to predict the potential number of infected cattle in the young age group. However, European data can be used for this purpose. Use of European data though will overestimate the potential infected cattle in this age group since those data were generated from countries in which BSE has been detected through less intensive surveillance testing (than the USA; 47 times the recommended level by the OIE). Also, European data represent a more extreme value for the potential presence of the disease agent in this young age group they did not differentiate the age of the cattle below 35 months. Thus, we will consider those cattle below this age as younger cattle.

In Europe, there was a total of one case that was identified as BSE-positive below the age of 35 months during the last 10 years. In addition, there was a total of 12 million cattle that were tested during 2002, the most extreme surrogate for other years (European Union Statistics). Thus, the rate of BSE-positive cattle in this young age group is 0.6 per million in contrast to 309.5 (per 1,000,000) for those older than 35 months of age. **In Europe, the likelihood of a cow to be tested positive and being infected is 500 times higher if it is older than 35 months of age, as compared to those younger than 35 months of age**. Given this information, we can extrapolate the information to form the basis for determining the risk of BSE in the young population of cattle in the U.S.:

1. We can estimate the potential number of infected cattle at a young age in the U.S. if the disease is present using as follows:

    a. A total of 34,907,000 head are slaughtered in the U.S. of which 28,255,000 are youthful steers and heifers. Let us assume that 50% of the steers and heifers in the U.S. fed beef population are younger than 30 MOA (14,127,500). This is the most

liberal approach, as the data indicate that more than 90% of the steers and heifers are under 30 MOA.

b. Application of the above rate of infection/detection of 0.6 per million therefore translates to a total of 9.0 (8.47 animals) potentially infected cattle in the system if the disease is present in the U.S. at the level of 1 in a million of cattle population. **Even if we consider the level of ID_{50} that has been estimated by Japanese scientists, the potential level of distribution of the agent in the food chain is negligible.**

c. A total of 178,336 cattle were tested for BSE in the U.S. in June through January 10, 2005, and 10 million cattle were tested in EU member-countries in 2002. Thus, we would expect approximately 0.0178 animals in the U.S. young age group to be positive for BSE if the disease is present.

To summarize, there is an extremely low level of potential for cattle to be infected in the U.S., particularly in the youthful fed-beef population, even when the two extreme options (b or c above) are considered.

CONCLUSION

In total, the 4,493 samples collected in this study accurately replicated the current dynamics of the U.S. fed-beef population. After 26% of the data population was discarded due to the wide birth date intervals, the resulting sample population maintained it's resemblance to the initial sample population, although the relative percentage of cattle 21 months of age and older increased.

Results demonstrated that, by using a deterministic analysis of data, an overall maturity

score of A^{40} would serve as an appropriate threshold for insuring that 100% of beef exported to Japan would be obtained from cattle 20 months of age or younger. While this approach was excessively conservative considering other firewalls that are in place to prevent the transmission of BSE in the U.S, and little to no prevalence of the disease in the national herd, it does provide the greatest level of assurance that no products originating from cattle 21 months of age and older will be allowed to be exported to Japan.

The most liberal method that could be used to mitigate the risk of exporting beef and beef products from any cattle 21 months of age and older would be to use the most conservative data analysis, which suggested that A^{40} would serve as the proper threshold to identify carcasses that originate from cattle that are 20 months of age and younger.

Image 1. Photographic demonstration of the lumbar vertebrae of a carcass with A^{40} Overall Maturity.

Image 2. Photographic demonstration of the lumbar vertebrae of a carcass with A^{50} Overall Maturity.

Table 1. The description of maturity characteristics within A maturity.

	A^{00}	A^{40}	A^{50}	A^{100}
All Vertebrae	Some evidence of cartilage in all vertebrae	Some evidence of cartilage in all vertebrae	Some evidence of cartilage in all vertebrae	
Sacral Vertebrae	Show distinct separation	Show distinct separation, caps show considerable evidence of cartilage	Show separation, caps show evidence of cartilage	Completely fused
Lumbar Vertebrae	No ossification	Caps tend to be partially ossified	Caps tend to be nearly moderately ossified	Nearly completely ossified
Thoracic Vertebrae	No ossification	No ossification	No ossification	Some evidence of ossification
Split Vertebrae Surfaces	Soft, porous and very red	Tend to be soft, porous and red	Tend to be moderately soft, porous and moderately red	Slightly red and slightly soft
Ribs	Only slight tendency toward flatness	Tendency toward flatness	Some tendency toward flatness and narrow	Slightly wide and slightly flat
Lean Texture and Color	Very fine, light grayish red	Very fine, light red	Very fine, moderately light red	Fine, moderately light red

Table 2. Stratification of total sample population (N=4,493) by sex.

Gender	Frequency	%	Cumulative Frequency	Cumulative %
Steer	1970	43.9	1970	43.9
Heifer	2282	50.7	4252	94.6
Mixed	241	5.4	4493	100

Table 3. Stratification of total sample population (N=4,493) by breed.

Breed	Frequency	%	Cumulative Frequency	Cumulative %
British	1595	35.5	1595	35.5
Continental	20	0.5	1615	35.9
Holstein	347	7.7	1962	43.7
BritishXConti	2515	56.0	4477	99.6
Bos Indicus	16	0.4	4493	100

Table 4. Stratification of total sample population (N=4,493) by production system.

Production System	Frequency	%	Cumulative Frequency	Cumulative %
NSI	914	20.5	914	20.5
Backgrounded	2011	45.2	2925	65.7
Grass/Wheat	748	16.7	3673	81.8
Non-Implant	473	10.5	4146	92.3
Calf Fed	347	7.7	4493	100

Table 5. Stratification of total sample population (N=4,493) by growth promotion regime (GPR).

GPR	Frequency	%	Cumulative Frequency	Cumulative %
Implant	3673	81.8	3673	81.3
Non-Implant	820	18.3	4493	100

Table 6 Stratification of total sample population (N=4,493) by birth interval.

Birth Interval (days)	Frequency	%	Cumulative Frequency	Cumulative %
Exact	168	3.7	168	3.7
1-30	375	8.4	543	12.1
31-60	1147	26.1	1690	37.6
61-90	2367	52.8	4057	90.3
91-120	305	6.9	4362	97.1
121-150	103	2.3	4465	99.4
151 or greater	28	0.6	4493	100

Table 7. Stratification of sub-sample (n=3,338)* population by sex.

Gender	Frequency	%	Cumulative Frequency	Cumulative %
Steer	1452	43.5	1452	43.5
Heifer	1688	50.6	3140	94.1
Mixed	198	5.9	3338	100

Table 8. Stratification of sub-sample (n=3,338)* population by breed.

Breed	Frequency	%	Cumulative Frequency	Cumulative %
British	1462	43.8	1462	43.8
Continental	20	0.6	1482	44.4
Holstein	347	10.4	1829	54.8
BritishXConti	1493	44.7	3322	99.5
Bos Indicus	16	0.5	3338	100

Table 9. Stratification of sub-sample (n=3,338)* population by production system.

Production System	Frequency	%	Cumulative Frequency	Cumulative %
NSI	636	19.1	636	19.1
Backgrounded	1378	41.3	2014	60.4
Grass/Wheat	542	16.2	2556	76.6
Non-Implant	435	13.0	2991	89.6
Calf Fed	347	10.4	3338	100

Table 10. Distribution of cattle older than and younger than 21 months of age by overall maturity classification.

Maturity Score	20 MOA and younger (n)	Frequency %	21 MOA and older (n)	Frequency %
A^{20}	3	100.0	0	0.0
A^{30}	57	100.0	0	0.0
A^{40}	196	100.0	0	0.0
A^{50}	382	95.3	19	4.7
A^{60}	1072	94.0	69	6.0
A^{70}	894	90.9	89	9.1
A^{80}	349	89.0	43	11.0
A^{90}	70	78.7	19	21.3
B^{00} or older	65	78.7	11	14.5

Table 11. Contingency table characterizing the distribution of age among overall maturity scores.

Age in Months

Overall Maturity Score

	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	Total
A^{20}			1	1	1																3
A^{30}			3	1	47	6															57
A^{40}		2	19	12	92	69	2														196
A^{50}	1	7	31	28	42	135	100	10	18	10	19										401
A^{60}		1	58	174	155	79	164	105	297	39	69										1141
A^{70}		1	30	56	105	6	83	125	441	47	89										983
A^{80}				2	8		11	56	218	54	37	1	1					2	1	1	392
A^{90}			1	3	12		3	1	36	14	17						1	1			89
B^{00}				3	1	1		2	13	4	4					2		1	1		32
B^{10}				4	3			1	9												17
B^{20}				4					7		2										13
B^{30}				2	1				1									1			5
B^{40}				1																	1
B^{50}				1	1																2
B^{60}									1												1
C^{00}				2	1				2												5
Total	1	11	143	294	469	296	363	300	1048	168	237	1	1			2	1	5	2	1	3338

Figure 1. Evaluator Accuracy as a Function of the Overall Maturity Score
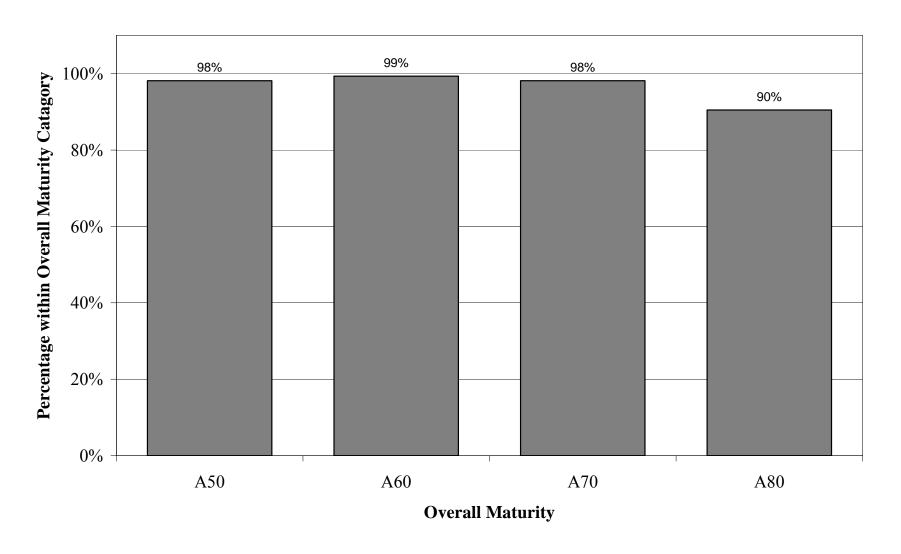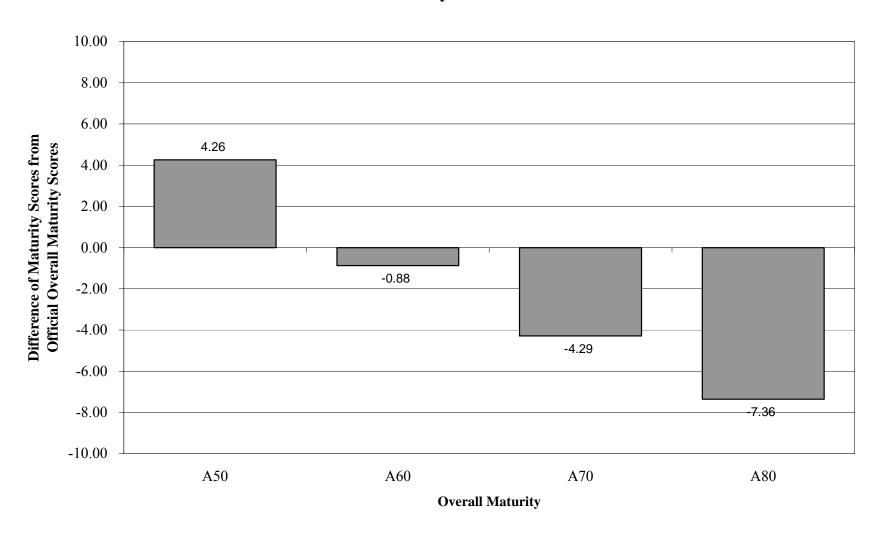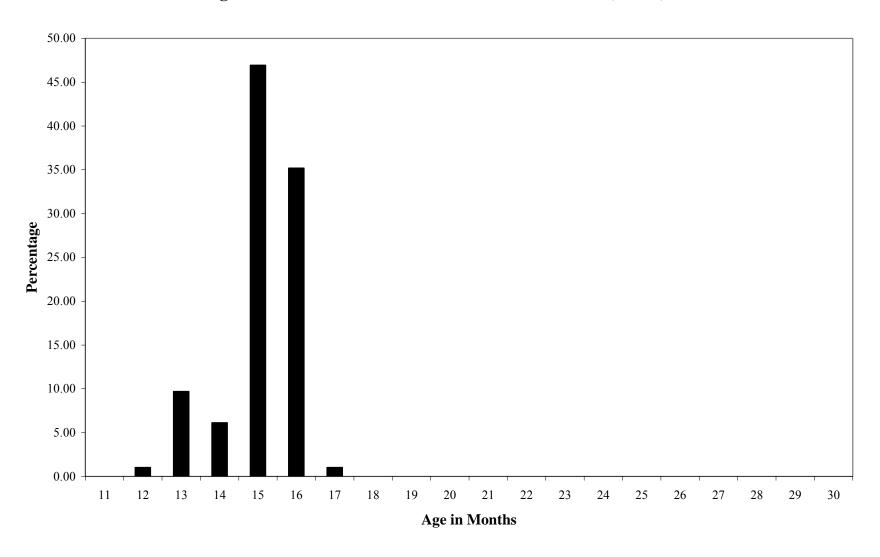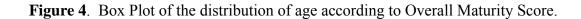
Figure 2. Average Difference in Maturity Scores as a Function of Official Overall Maturity Score

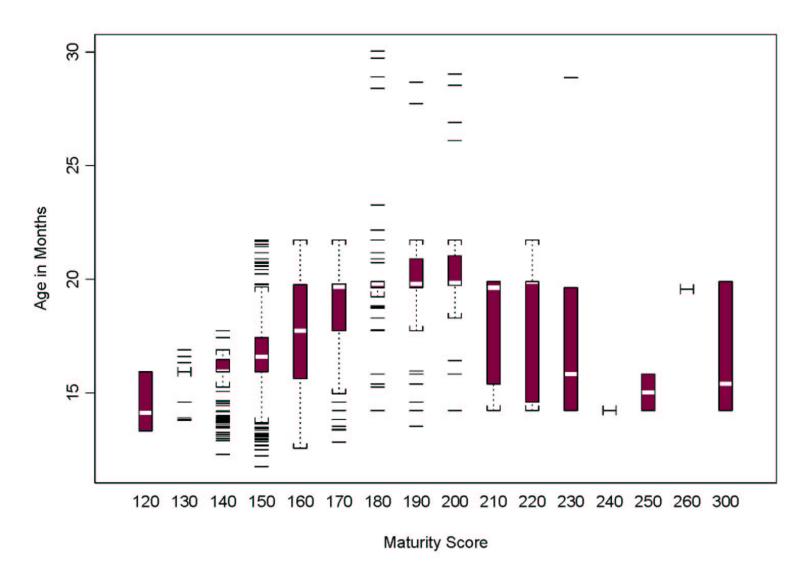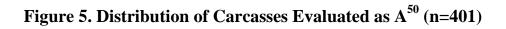**Figure 3. Distribution of Carcasses Evaluated as A^{40} (n=196)**

Figure 4. Box Plot of the distribution of age according to Overall Maturity Score.

Figure 5. Distribution of Carcasses Evaluated as A^{50} (n=401)

Figure 6. Distribution of Carcasses Evaluated as A^{60} (n=1141)

Figure 7. Distribution of Carcasses Evaluated as A^{70} (n=983)

Figure 8. Distribution of Carcasses Evaluated as A^{80} (n=392)

Figure 9. Distribution of Carcasses Evaluated as A^{90} (n=89)
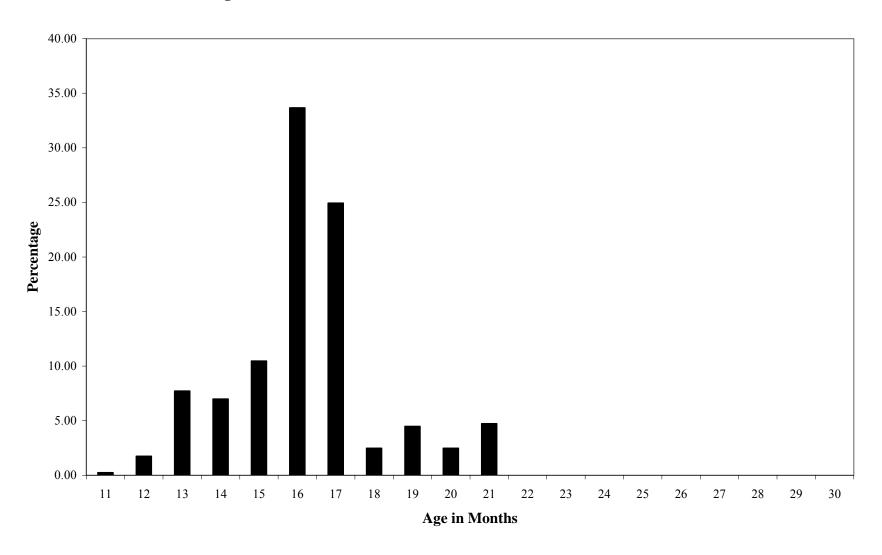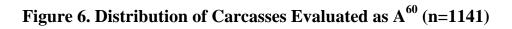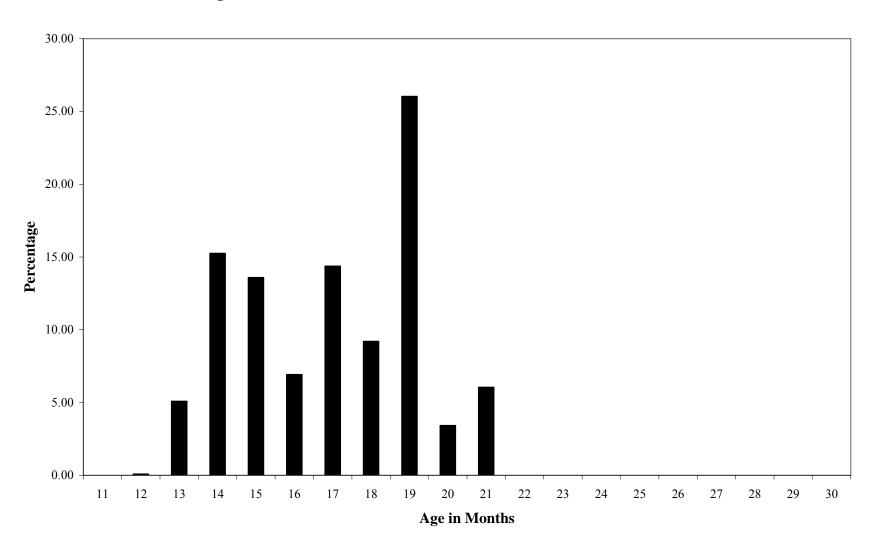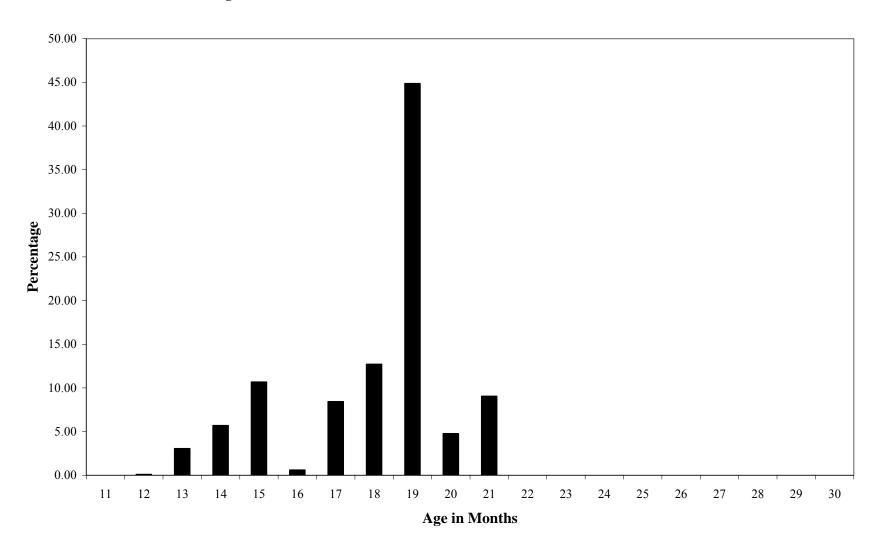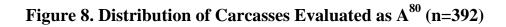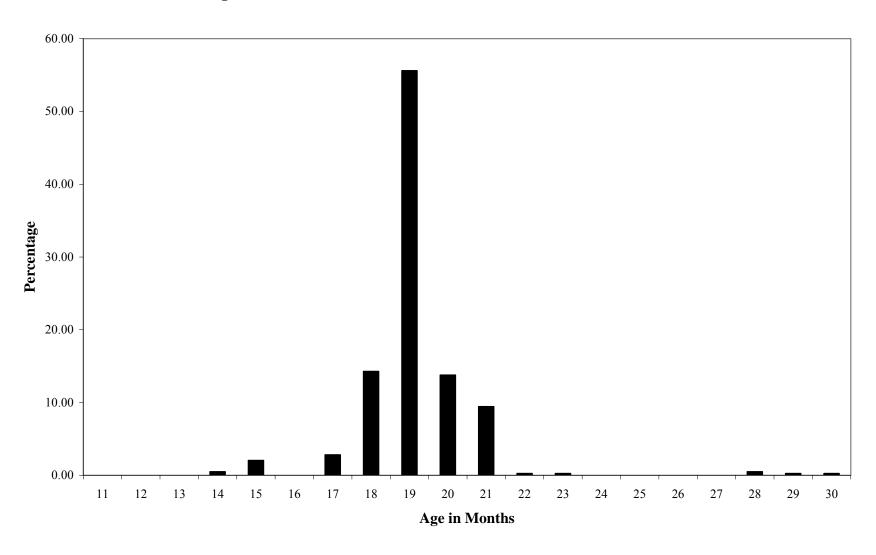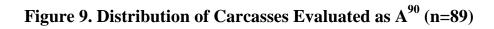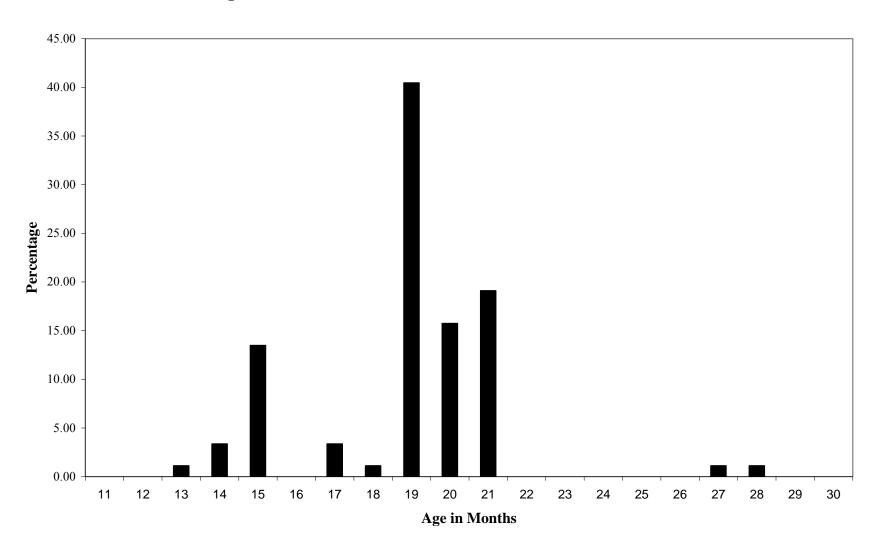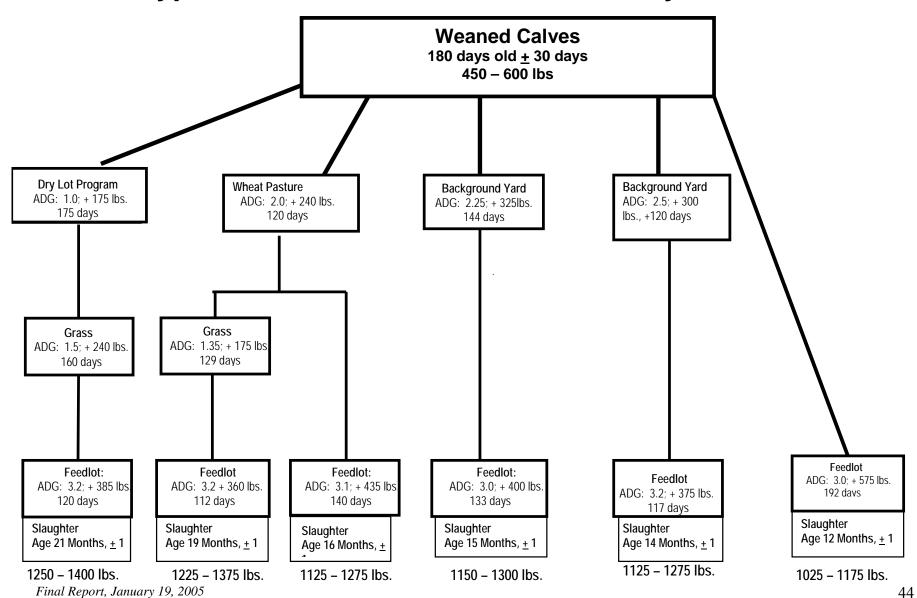
Appendix C
Typical U.S. Beef Cattle Production Systems

Weaned Calves
180 days old ± 30 days
450 – 600 lbs

Dry Lot Program
ADG: 1.0; + 175 lbs.
175 days

Wheat Pasture
ADG: 2.0; + 240 lbs.
120 days

Background Yard
ADG: 2.25; + 325lbs.
144 days

Background Yard
ADG: 2.5; + 300 lbs., +120 days

Grass
ADG: 1.5; + 240 lbs.
160 days

Grass
ADG: 1.35; + 175 lbs.
129 days

Feedlot:
ADG: 3.2; + 385 lbs.
120 days

Slaughter
Age 21 Months, ± 1

1250 – 1400 lbs.

Feedlot
ADG: 3.2 + 360 lbs.
112 days

Slaughter
Age 19 Months, ± 1

1225 – 1375 lbs.

Feedlot:
ADG: 3.1; + 435 lbs.
140 days

Slaughter
Age 16 Months, ± 1

1125 – 1275 lbs.

Feedlot:
ADG: 3.0; + 400 lbs.
133 days

Slaughter
Age 15 Months, ± 1

1150 – 1300 lbs.

Feedlot
ADG: 3.2; + 375 lbs.
117 days

Slaughter
Age 14 Months, ± 1

1125 – 1275 lbs.

Feedlot
ADG: 3.0; + 575 lbs.
192 days

Slaughter
Age 12 Months, ± 1

1025 – 1175 lbs.

Appendix D

Consist of Sample Population with a Chronological Age Greater Than 20 Months and a Physiological Maturity Score of A^{50} and A^{60}

A total of 88 cattle out of the 3,338 head sample population were over 21 months of age (MOA) and were assessed as A^{50} or A^{60} overall physiological maturity.

Of the A^{50} cattle, 19 were >21 MOA and, of these:

- 2 were carcasses of cattle in which the exact date of birth (DOB) was known.
- 14 were in a 58/59 day range in DOB.
- 3 were in a 28 day range in DOB.

Of the A^{60} cattle, 69 were >21 MOA

- 4 were exact date of birth.
- 48 were in a 58/59 day range in DOB
- 17 were in a 28 day range in DOB

If you further categorize these data, of the A^{50} cattle >21 MOA using a 30 day month to determine MOA (N=19):

- 11 fall below 21 MOA when using a 30.4 day month to calculate MOA.
- 16 fall below 21 MOA when using a 31 day month to calculate MOA.
- 3 of the 19 A^{50} cattle would still be >21 MOA using a 31 day month (all 21.03 MOA).

Further categorization of the A^{60} cattle (N=88):

- 34 of these fall below 21 MOA when using a 30.4 day month to calculate MOA.
- 51 of these fall below the 21 MOA when using a 31 day month to calculate MOA.
- 18 of the 88 A^{60} cattle would still be >21 MOA using a 31 day month (again, all at an age of 21.03 MOA).

Appendix E

ESTIMATING THE PROBABILITY OF OBSERVING A CARCASS EVALUATED AS A^{40} OR LESS GIVEN THAT THE CARCASS WAS 21 MONTHS IN AGE

Data in this study indicated that no carcasses were evaluated as equal to or less than A^{40} in physiological maturity resulted from cattle older than 17 months of age. In terms of statistical analysis, however, the possibility of detecting a carcass 21 months of age or older in age and evaluated as A^{40} is not zero in any sampling environment. The purpose of the following exposition is to hypothetically compute probabilities for such occurrences.

Vertical Analysis

Considering that older chronological age essentially presents a higher maturity score, and that sample selections were made based on age but not on maturity grade, statistical analysis of this sample data set should be performed by taking age as the regressor ("vertical analysis"). Therefore, the result of the estimation will be the probability of observing at least one carcass from a bovine carcass evaluated as A^{40} (or less) physiological maturity in the cattle of 21 months of age (It is appropriate that cattle at 22 months of age and older should be excluded, since the sampling target was focused on those at 21 months of age threshold and younger).

Sub-samples

The probability of observing at least one carcass from a bovine animal evaluated as A^{40} (or less) that would also be 21 months of age was estimated using two different sub-samples of the total experimental population. In fact, no carcasses in either subpopulation actually were classified as equal to, or less than, A^{40} physiological maturity. The first sub-sample included n = 237 (sub-sample 1) carcasses that were evaluated as A^{50} physiological maturity or higher in the initial data set and that were also from live animals only 21 months of chronological age. The second sub-sample included n = 1,748 (sub-sample 2) carcasses that had a chronological age of 18 to 21 months and that also were evaluated as A^{50} physiological maturity or higher (i.e., it included the first sub-sample of n = 237 carcasses). These sub-sample populations were selected to exclude carcasses of cattle that actually were 22 months of chronological age or older, and the latter

sub-sample (i.e., sub-sample 2) population also reflected carcasses from those cattle of chronological ages described in the public presentation of the final report as the "buffer zone" (cattle less than 21 months of age but whose carcasses exceeded A^{40} physiological maturity; added to these were data from sub-sample 1). In other words, sub-sample 2 reflected an observation that there were not any carcasses evaluated as A^{40} or lower in the original experimental sample population that were older than 17 months of chronological age.

Analysis

Conclusions included in our initial experimental results (as presented in the final report and in the January 19, 2005 public meeting of experts) were actually derived by focusing on data from carcasses of cattle classified as equal to or less than A^{40} physiological maturity—among those carcasses, none were derived from cattle that were over 17 months of chronological age. Here, we intended to discern the probability of detecting a carcass that was over 21 months of chronological age using a sub-sample of the original experimental population that reflected (a) those that were exactly 21 months of age, and (b) those that were exactly 21 months of age plus those that were between 18 and 20 months of age (inclusively).

We chose to utilize a level of Type I statistical error of $\alpha = 0.01$ in these "vertical" non-parametric analyses for standard significance testing of hypotheses. Probabilities were computed as follows:

$P \leq 1 - \alpha^{1/n} [(1 - P)^n \geq \alpha \leftrightarrow P \leq 1 - \alpha^{1/n}]$. At $\alpha = 0.01$, the probability for sub-sample 1 (n = 237) that an animal would be A^{40} or less in physiological maturity was $P = 0.0192$, while the increased number of observations and the greater statistical power provided by sub-sample 2 (n = 1,748) yielded a probability that an animal would be A^{40} or less in physiological maturity of $P = 0.00263$

Conclusion

Clearly these results ($P = 0.0192$ and $P = 0.00263$) suggested that using a sub-sample of carcasses from cattle that are over 17 months of age results in a hypothetically low probability that carcasses in the parent population would generate carcasses that were equal to or less than A^{40} in physiological maturity, and when that sample size is magnified by over 7-fold, the same

probability also is reduced by a factor of greater than 7-fold. Such a risk is clearly further reduced—exponentially—given additional "firewalls" to BSE transmission and food contamination implemented by the U.S. government. This conclusion coincided nicely with conclusions from our initial analyses (as presented in the final report and in the January 19, 2005 public meeting of experts) which focused on data from carcasses of cattle classified as equal to or less than A^{40} physiological maturity—among those carcasses, none were derived from cattle that were over 17 months of chronological age. Based on initial experimental results, combined with these vertical non-parametric analyses, A^{40} is a threshold for insuring that no beef would be exported to Japan that is derived from cattle that are over 20 months of age.