UNITED STATES ENVIRONMENTAL PROTECTION AGENCY
WASHINGTON, D.C. 20460

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OCT 21 1991

OFFICE OF
PESTICIDES AND TOXIC
SUBSTANCES

**MEMORANDUM**

**SUBJECT:** Requested review of a fourteen day repeated dose oral toxicity/hormone study, and measurements of various hormones in rat serum for evidence of possible hormonal mediated carcinogenesis.

Tox.Chem No.: 063 and 740
MRID No.: 414793-01 and 415109-01
HED Project No.: 0-1484
Submission No.: 266495

To: Jude Andreasen, PM Team#76
Registration Division
Insecticide-Rodenticide Branch (H7505)

From: John C. Redden, Toxicologist 9/18/91
Section 3
Toxicology Branch 1
Health Effects Division (H7509C)

Thru: Henry Spencer, Ph.D. 10/2/91
Acting, Section Head Section 3
Toxicology Branch 1
Health Effects Division (H7509C) 10/15/91

**ACTION:**

The registrant has submitted MRID No. 414793-01, "Simazine Technical Measurement of various Hormones in Rat Serum", and MRID No. 415109-01,"14-Day Repeated Dose Oral Toxicity/Hormone Study in Female Albino Rats with Atrazine and Diaminochlorotriazine", for evidence of possible hormonal mediated carcinogenesis. These studies were submitted in response for hormone data on chlorotriazines for atrazine and simazine special data call-ins.

**CONCLUSIONS:**

MRID No. 414793-01 reports the methods and results of measurements of various hormones in rat serum from CIBA-GEIGY Study No. 852004, "Simazine Technical: 104-Week Oral Toxicity/ Carcinogenicity Study in Rats". The dose groups for this study were 0, 10, 100, and 1000 ppm of Simazine. Serum was collected from selected males and females who survived 104 weeks of treatment, and

were examined endocrinologically to "assess any possible correlation between altered hormone levels and tumor development in Sprague-Dawley Rats".

Male rats for all dose groups showed no positive or negative trends nor any significant differences in mean when compared to the control group. Female rats for all dose groups showed negative trends for estradiol ($E_2$), triiodothyronine ($T_3$) and progesterone, when compared with the control mean values. Positive trends occurred for all dose groups when compared with the control mean values for prolactin and growth hormone. Follicle stimulating hormone(FSH) and estradiol levels were increased at 100 and 1000 ppm. Growth hormone (GH) levels were increased at 10 and 1000 ppm.

Lifetime administration of Simazine Technical may alter serum hormone levels in Sprague-Dawley rats. However, variation in the data from such small numbers of animals make it difficult to have great confidence in the results. Clarification of trend values for female FSH is required. Core classification is supplementary as this type study is not addressed in the EPA guidelines. The study is not upgradeable.

In MRID No. 415109-01,"14-Day Repeated Dose Oral Toxicity/Hormone Study in Female Albino Rats with Atrazine and Diaminochlorotriazine", Atrazine or diaminochlorotriazine (DACT) were administered orally to female Sprague-Dawley rats at doses of 0, 100, 200, and 400 mg/kg/day. The 400 mg/kg dose was reduced to 300 mg/kg/day on day 4 because of a death in each of the high dose groups.

All treated groups demonstrated reduced body weight/body weight gain. Animals receiving Atrazine doses of ≥200 mg/kg and DACT doses ≥100 mg/kg had a thin and hunched appearance, rough coat and the presence of few or no feces. Absolute spleen weight was decreased in the 300 mg/kg DACT-treated group. Relative spleen weights were increased in the 100 and 200 mg/kg DACT-treated groups.

Estrogen levels were lower at doses ≥ 200 mg/kg of atrazine. Luteinizing hormone(LH) was reduced in the ≥ 300 mg/kg of atrazine. Estrogen and progesterone levels were depressed at doses of ≥ 200 mg/kg DACT. LH and prolactin levels were decreased in the 100 and 200 mg/kg DACT.

Hormonal changes appear to result from 14 days of treatment by gavage at ≥ 200 mg/kg Atrazine and ≥ 100 mg/kg DACT. DACT appears more toxic than the parent, Atrazine. The results suggest that subacute administration of atrazine and diaminochlorotriazine alters serum hormone levels in female Sprague-Dawley rats. This may also occur at lower doses in chronic studies. The Toxicology Branch acknowledges the data, but awaits the chronic studies before evaluating evidence of possible hormonal mediated carcinogenesis. Core classification is supplementary as this type study is not addressed in the guidelines. The study is not upgradeable.

Reviewed by: John C. Redden, Toxicologist 9/18/91
Section III, Tox. Branch I
Secondary reviewer: Henry Spencer, Ph.D. 10/2/91
Section III, Tox. Branch I

DATA EVALUATION REPORT

GUIDELINES § Not Applicable

STUDY TYPE: 14-Day Repeated Dose Oral Toxicity/Hormone

TOX. CHEM NO: 063

MRID NO.: 415109-01

TEST MATERIAL: Atrazine and diaminochlorotriazine

SYNONYMS: Not applicable

STUDY NUMBER: HLA No. 483-268

SPONSOR: Ciba-Geigy Corporation, P.O. Box 18300, Greensburo, NC 27419

TESTING FACILITY: Hazleton Laboratories America, Inc., 9200 Leesburg Pike Vienna, Virginia 22180

TITLE OF REPORT: 14-Day Repeated Dose Oral Toxicity/Hormone Study in Female Albino Rats with Atrazine and Diaminochlorotriazine

AUTHOR(S): Sandra L. Morseth, Ph.D.

REPORT ISSUED: March 6, 1990

CONCLUSION:

Atrazine or diaminochlorotriazine (DACT) were administered orally to female Sprague-Dawley rats at doses of 0, 100, 200, and 400 mg/kg/day. The 400 mg/kg dose was reduced to 300 mg/kg/day on day 4 for both atrazine and DACT, because one animal in the Group 5 (400 mg/kg/day Atrazine), and 1 animal in the Group 8 (400 mg/kg/day DACT) died. Once the dose was changed to 300 mg/kg/day 1 an additional animal died in Group 5, and 7 animals died in Group 8. One animal in the 200 mg/kg/day DACT dose group also died.

All treated groups demonstrated reduced body weight/body weight gain. Animals at Atrazine doses of ≥200 mg/kg and DACT doses ≥100 mg/kg had a thin and hunched appearance, rough coat and the presence of few or no feces. Absolute spleen weight was

decreased in the 300 mg/kg DACT-treated group and relative spleen weights were significantly increased in the 100 and 200 mg/kg DACT-treated groups.

The positive control group had significantly higher levels of prolactin and somewhat elevated levels of luteinizing hormone(LH), when compared with the vehicle control group. Estrogen levels were significantly lower at doses ≥ 200 mg/kg of atrazine. LH was significantly reduced in the ≥ 300 mg/kg of atrazine. Estrogen and progesterone levels were depressed at doses of ≥ 200 mg/kg DACT. While LH and prolactin levels were decreased in the 100 and 200 mg/kg DACT. Hormonal changes appear to result from 14 days of treatment by gavage at ≥ 200 mg/kg Atrazine and ≥ 100 mg/kg DACT. DACT appears more toxic than the parent, Atrazine. The results suggest that subacute administration of atrazine and diaminochlorotriazine alters serum hormone levels in female Sprague-Dawley rats. This may also occur at lower doses in chronic studies. The Toxicology Branch acknowledges the data, but awaits the chronic studies before evaluating evidence of possible hormonal mediated carcinogenesis.

Core Classification: Core supplementary as this study type is not addressed in the guidelines. The study is not upgradeable.

A. MATERIALS:

1. Test compound: Atrazine, a white powder, Lot No. FL850612, 97.0% pure. Diaminochlorotriazine, a white powder, Lot No. FL871776, 97.4%.

2. Test animals: Species: rat, Strain: Sprague-Dawley Crl:CD BR, Age: 12 weeks, Weight: 219.0 to 300.6 grams, Source: Charles River Laboratories, Inc., Raleigh, North Carolina.

B: STUDY DESIGN:

1. Animal Assignment: Animals were acclimated to laboratory conditions for 21 days prior to treatment. Animals were smeared daily to determine which animals were normally cycling. 120 hormonally cycling animals, about 12 weeks of age were assigned to the study. Distilled water and Purina Certified Rodent Chow 5002 was available ad libitum. Temperature and Relative Humidity were recorded daily and ranged from 70-78 F and 28-60% respectively. A 14-hour light/10-hour dark cycle was maintained throughout the study. Animals were assigned to the dose groups by a computer-generated weight randomization process. The dose groups were as follows:

| Group No. | Dose mg/kg/day | No. Animals | Test Materials |
|---|---|---|---|
| 1 (Control) | 0 | 15 | vehicle |
| 2 (Positive control) | 0 | 15 | vehicle and metoclopramide |
| 3 (Low) | 100 | 15 | atrazine |
| 4 (Mid) | 200 | 15 | atrazine |
| 5 (high) | 400* | 15 | atrazine |
| 6 (Low) | 100 | 15 | DACT |
| 7 (Mid) | 200 | 15 | DACT |
| 8 (High) | 400* | 15 | DACT |

* dose changed to 300 mg/kg after day 4.

Animals were identified by ear tags and house in stainless steel wire cages.

2. Compound Preparation and Administration: Hazleton worked under the assumption that the test materials were 100 % pure (atrazine 97% and DACT 97.4% in actuality). Dosing suspensions were prepared by mixing the test materials with a mixture of corn starch and distilled water. Fresh dosing suspensions were prepared weekly. Dosing suspensions were administered by oral gavage at a constant volume of 10 ml per kg/bwt. The oral route was chosen because this is the likely route of human exposure. The positive control was prepared daily by diluting metoclopramide with 0.9% saline. The metoclopramide was administered by intraperitoneal injection, about 20 minutes before sacrifice at a dose volume of 1 ml per kg/bwt.

3. Analytical Chemistry: Stability under refrigeration was checked on the 100 and 400 mg/kg/day dose of both test materials on days 0, 3, and 7. Homogenicity was tested by taking samples from the top, middle, and bottom of the above mentioned dose solutions before the commencement of dosing.

4. Statistics: Body weight and changes from the control group were compared statistically to the treated groups. Organ weights were examined by one-way analysis of variance techniques. If the data was heterogenous, analysis was then performed on rank-transformed data. Hormone means excluded values detected to be outliers at $p \leq 0.110$ by Dixon's test. Group comparisons were done at the 5% two-tailed probability level.

5. Quality Assurance: A quality assurance statement was not available from the contract laboratory. However, a GLP statement was present from the sponsor stating that the study was "conducted in accordance with good and acceptable scientific practices". The statement was signed and dated 3/22/90.

C. METHODS AND RESULTS:

1. Observations: All animals were observed twice daily for mortality and moribundity. Animals were vaginally smeared daily to determine the stage of estrous.

Results: Animals receiving Atrazine doses of ≥200 mg/kg and DACT doses ≥100 mg/kg had a thin and hunched appearance, rough coat and the presence of few or no feces. Other observations seen in all groups, except Group 2, were rhinorrhea, low body temperature, urine stains, alopecia, and other excretions. One animal in group 7 on the third day of dosing had a red fluid in her cage pan. Eleven animals were found dead during the study; 2 in Group 5, 1 in Group 7, and 8 in Group 8.

2. Body Weights: Animals were weighed at the initiation of treatment, weekly, and at sacrifice.

Results: When compared to the control mean values body weight for Groups 5, 7, and 8 were significantly lower at week 1. Similarly, Groups 4, 5, 7, and 8 for week 2 were significantly lower when compared to the control body weight mean values. Mean body weight gains for Groups 3-8 were significantly lower when compared to the control values for the weeks 0-1 and weeks 0-2.

3. Sacrifice and Gross Pathology: After two weeks of treatment 2-3 animals in diestrous in each group were sacrificed by decapitation, without anesthesia. Sacrifices were done at least two hours after the gavage dose. Blood was collected at this time for hormone analyses.

Results: One group 4 animal had a moderate amount of red fluid in the thoracic cavity and a thickened pericardial sac containing a small amount of pale green material. Group 7 animal B78175 had a moderately enlarged spleen. Another, Group 7 animal B78168 had a small amount of fluid in the thoracic cavity, a thickened pericardial sac and a gelatinous thymus.

4. Organ Weights: After trimming the uterus, ovaries, spleen, thymus, pituitary, and mammary tissue were weighed. In addition the uterus, pituitary, and mammary tissue were frozen in liquid nitrogen for possible future study.

Results: Group 4-8 uterus mean weight values compared to the control values for final body weight were significantly lower. Group 3-8 thymus and Group 8 spleen mean weight values were significantly lower when compared to control mean values. Thymus mean organ weight compared to mean organ weight for the control group were significantly lower

for Groups 4-8. The spleen weights were significantly greater than controls for Groups 6 and 7.

5. Hormone Analyses: Clots were removed from the blood before the blood was centrifuged. Serum was then collected and frozen at -20 C. The serum was analyzed for prolactin, luteinizing hormone(LH), follicle stimulating hormone(FSH), progesterone, and estrogen.

Results: The positive control group (Table 1) had significantly higher levels of prolactin and somewhat elevated levels of LH, when compared to those of the vehicle control group. Estrogen levels were significantly lower at doses ≥ 200 mg/kg of atrazine and LH was significantly reduced in the ≥ 300 mg/kg of atrazine. Estrogen and progesterone levels were depressed at doses of ≥ 200 mg/kg DACT. LH and prolactin levels were decreased in the 100 and 200 mg/kg DACT treatments respectively.

D. STUDY AUTHOR'S CONCLUSIONS:

Atrazine or diaminochlorotriazine (DACT) were administered orally to female Sprague-Dawley rats at doses of 0, 100, 200, and 400 mg/kg/day. The 400 mg/kg dose was reduced to 300 mg/kg/day on day 4 for both atrazine and DACT, because one animal in the Group 5 (400 mg/kg/day Atrazine), and 1 animal in the Group 8 (400 mg/kg/day DACT) died. Once the dose was changed to 300 mg/kg/day 1 additional animal died in Group 5, and 7 additional animals died in Group 8. One other animal in the 200 mg/kg/day DACT dose group also died. All treated groups demonstrated reduced body weight/body weight gain. Animals at Atrazine doses of ≥200 mg/kg and DACT doses ≥100 mg/kg had a thin and hunched appearance, rough coat and the presence of few or no feces. Mean absolute spleen weights were decreased in the 300 mg/kg DACT-treated group and relative spleen weights were significantly increased in the 100 and 200 mg/kg DACT-treated groups. The positive control group had significantly higher levels of prolactin and somewhat elevated levels of LH, when compared with the vehicle control group. Estrogen levels were significantly lower at doses ≥ 200 mg/kg of atrazine and LH was significantly reduced in the ≥ 300 mg/kg of atrazine. Estrogen and progesterone levels were depressed at doses of ≥ 200 mg/kg DACT, and LH and prolactin levels were decreased in the 100 and 200 mg/kg DACT. The results suggest that subacute administration of atrazine and diaminochlorotriazine alter serum hormone levels on female Sprague-Dawley rats. This may also occur at lower doses in chronic studies.

E. REVIEWER'S DISCUSSION AND INTERPRETATION OF RESULTS:

The Toxicology Branch acknowledges the data, and notes that DACT appears more toxic than the parent, Atrazine. Hormonal changes appear to result from 14 days of treatment by gavage at ≥ 200 mg/kg Atrazine and ≥ 100 mg/kg DACT. This reviewer agrees with the conclusions noted by the study author. Core Classification is Core supplementary as this study type is not addressed in the guidelines. The study is not upgradeable.

TABLE 1: 14-Day Repeated Dose Oral Toxicity/Hormone Study in Female Albino Rats with Atrazine and Diaminochlorotriazine Mean Hormone Analyses (extracted from the Final Report).

| | Prolactin ng/ml | LH pg/ml | FSH ng/ml | Progesterone ng/ml | Estrogen pg/ml |
|---|---|---|---|---|---|
| Group 1 (Control) | | | | | |
| Mean | 7.0 | 579.3 | 74.3 | 9.72 | 12.2 |
| S.D. | 6.2 | 380.0 | 28.5 | 7.8 | 9.3 |
| N | 14 | 15 | 15 | 13 | 14 |
| Group 2 (Positive Control) | | | | | |
| Mean | 559.3 | 349.9 | 65.0 | 10.79 | 14.69 |
| S.D. | 232.3 | 152.4 | 22.3 | 3.7 | 12.1 |
| N | 14 | 14 | 13 | 12 | 15 |
| Group 3 (Atrazine 100 mg/kg/day) | | | | | |
| Mean | 3.4 | 396.2 | 71.8 | 9.4 | 21.7 |
| S.D. | 1.4 | 191.0 | 31.9 | 7.3 | 15.6 |
| N | 15 | 14 | 15 | 14 | 15 |
| Group 4 (Atrazine 200 mg/kg/day) | | | | | |
| Mean | 3.7 | 652.2 | 170.5 | 14.3 | 3.3 |
| S.D. | 1.2 | 457.5 | 182.7 | 8.1 | 1.7 |
| N | 12 | 15 | 15 | 14 | 12 |
| Group 5 (Atrazine 400 first 4 days 300 mg/kg/day thereafter) | | | | | |
| Mean | 3.7 | 222.7 | 61.7 | 8.8 | 6.6 |
| S.D. | 1.7 | 101.5 | 27.0 | 6.1 | 3.5 |
| N | 12 | 11 | 12 | 11 | 13 |
| Group 6 (Diaminochlorotriazine 100 mg/kg/day) | | | | | |
| Mean | 2.8 | 285.1 | 79.1 | 16.9 | 11.4 |
| S.D. | 0.6 | 101.1 | 40.8 | 14.3 | 9.9 |
| N | 13 | 13 | 15 | 15 | 14 |
| Group 7 (Diaminochlorotriazine 200 mg/kg/day) | | | | | |
| Mean | 3.1 | 235.4 | 83.6 | 2.9 | 3.9 |
| S.D. | 0.8 | 156.0 | 24.1 | 1.3 | 1.6 |
| N | 11 | 13 | 12 | 10 | 11 |
| Group 8 (DACT 400 first 4 days 300 mg/kg/day thereafter) | | | | | |
| Mean | 7.6 | 228.6 | 98.5 | 2.2 | 3.7 |
| S.D. | 6.6 | 110.0 | 26.2 | 0.7 | 2.0 |
| N | 8 | 8 | 8 | 5 | 8 |

Means exclude values detected to be outliers at $p \leq 0.10$ by Dixon's test.

Reviewed by: John C. Redden, Toxicologist 9/18/91
Section III, Tox. Branch I
Secondary reviewer: Henry Spencer, Ph.D. 10/2/91
Section III, Tox. Branch I

DATA EVALUATION REPORT

GUIDELINES § Not Applicable

STUDY TYPE: Measurement of Various Hormones in Rat Serum

TOX. CHEM NO: 740

MRID NO.: 414793-01

TEST MATERIAL: Simazine Technical

SYNONYMS: Not available

STUDY NUMBER: 300-938

SPONSOR: Ciba-Geigy Corporation P.O. Box 18300, Greensboro, NC 27419

TESTING FACILITY: Hazleton Laboratories America, Inc., 9200 Leesburg Turnpike, Vienna, Virginia 22182

TITLE OF REPORT: Simazine Technical: Measurement of Various Hormones in Rat Serum

AUTHOR(S): R.L. Tacey

REPORT ISSUED: March 7, 1990

CONCLUSION:

MRID No. 414793-01 reports the methods and results of measurements of various hormones in rat serum from CIBA-GEIGY Study No. 852004, "Simazine Technical: 104-Week Oral Toxicity/Carcinogenicity Study in Rats". The dose groups for this study were 0, 10, 100, and 1000 ppm of Simazine. Serum was collected from selected males and females who survived 104 weeks of treatment, and were examined endocrinologically to "assess any possible correlation between altered hormone levels and tumor development in Sprague-Dawley Rats". Males were assayed for adrenocorticotropic hormone (ACTH), luteinizing hormone (LH), thyroid stimulating hormone (TSH), dihydrotestosterone (DHT), triiodothyronine ($T_3$), thyroxine ($T_4$), and Testosterone. Females were assayed for Estradiol($E_2$), Prolactin, follicle stimulating hormone(FSH), progestererone, LH, growth hormone(GH), TSH, $T_3$, $T_4$, and ACTH. Male rats for all dose groups showed no significant

positive or negative trends nor any significant differences in mean when compared to the control group. Female rats for all dose groups showed significant negative trends for estradiol ($E_2$), triiodothyronine ($T_3$) and progesterone, and positive trends occurred for all dose groups when compared with the mean for prolactin and growth hormone. FSH and estradiol levels were significantly increased at 100 and 1000 ppm, and growth hormone (GH) levels were increased at 10 and 1000 ppm. Clarification of trend values for female FSH is required. Variation in the data from such small numbers of animals make it difficult to have great confidence in the results. The Toxicology Branch acknowledges the data as preliminary in nature, and awaits the completed additional data before trying to draw conclusions on the relevant role of various hormones in rat serum with respect to possible hormonally mediated carcinogenesis.

Core Classification: Core classification is supplementary as this type study is not addressed in EPA guidelines.

A. MATERIALS, STUDY DESIGN, METHODS, AND RESULTS:

1. These samples were obtained from a previously submitted study MRID No. 40614405.

2. Statistics: Dixon's test was used to detect any outliers before statistical analysis followed by: Terpstra-Jonckheere test for monotone trend, simple linear regression analysis of both untransformed and ranked data, ANOVA of both untransformed and ranked data, and Dunnett's test for control versus treatment mean comparisons.

3. Hormone Analyses: The primary method of analysis was radioimmunoassay. Males were assayed for adrenocorticotropic hormone (ACTH), luteinizing hormone (LH), thyroid stimulating hormone (TSH), dihydrotestosterone (DHT), triiodothyronine ($T_3$), thyroxine ($T_4$), and Testosterone. Females were assayed for Estradiol($E_2$), Prolactin(prl), follicle stimulating hormone(FSH), progestererone(prog), luteinizing hormone (LH), growth hormone(GH), TSH, $T_3$, $T_4$, and ACTH. ACTH was given highest priority for males, but in females ACTH was last because of the limited sample volumes obta'..ed from females.

Results: Male rats for all dose groups showed no significant positive or negative trends nor any significant differences in mean when compared to the control group. Female rats(Table 1) for all dose groups showed significant negative trends for estradiol ($E_2$), triiodothyronine ($T_3$) and progesterone(prog), and positive trends occurred for all

dose groups when compared with the mean for prolactin and growth hormone(GH). FSH and estradiol levels were significantly decreased at 100 and 1000 ppm, and growth hormone (GH) levels were increased at 10 and 1000 ppm. FSH was also reported to have a negative trend. However, page 53 reports a positive trend, while the table on page 17 reports a negative trend.

D. STUDY AUTHOR'S CONCLUSIONS:

Serum was collected from selected males and females who survived 104 weeks of treatment, and were examined endocrinologically to "assess any possible correlation between altered hormone levels and tumor development in Sprague-Dawley Rats". Male rats for all dose groups showed no significant positive or negative trends nor any significant differences in mean when compared to the control group. Female rats for all dose groups showed significant negative trends for FSH, estradiol ($E_2$), triiodothyronine ($T_3$) and progesterone, and positive trends occurred for all dose groups when compared with the mean for prolactin and growth hormone. FSH and estradiol levels were significantly increased at 100 and 1000 ppm, and growth hormone (GH) levels were increased at 10 and 1000 ppm. Lifetime administration of Simazine Technical alters serum hormone levels in Sprague-Dawley rats.

E. REVIEWER'S DISCUSSION AND INTERPRETATION OF RESULTS:

The Toxicology Branch acknowledges the data as preliminary in nature, and awaits the completed additional data before trying to draw conclusions on the relevant role of various hormones in rat serum with respect to possible hormonally mediated carcinogenesis. Variation in the data from such small numbers of animals make it difficult to have great confidence in the results. FSH was reported to have both a negative trend (page 17) and a positive trend (page 53). Obviously, one of the pages is in error, and requires correction. Lifetime administration of Simazine Technical may cause alterations in serum hormone levels, as indicated above, in Sprague-Dawley rats. Core classification is supplementary since this type study is not addressed in EPA guidelines.

TABLE 1: Simazine Technical: 104-Week Oral Toxicity/Carcinogenicity Study Group Means of Hormone Assays Females (Values Detected as Outliers Excluded).

| | $E_2$ (pg/ml) | Prl (ng/ml) | FSH (ng/ml) | Prog (ng/ml) | LH (pg/ml) | GH (ng/ml) | TSH (ng/ml) | $T_4$ (µg/dl) | $T_3$ (µg/dl) |
|---|---|---|---|---|---|---|---|---|---|
| Gp.1(0 ppm) | | | | | | | | | |
| Mean | 11.5 | 28.8 | 160.3 | 38.9 | 140.8 | 11.2 | 1.3 | 2.1 | 58.8 |
| S.D. | 6.41 | 18.01 | 24.27 | 26.18 | 36.20 | 1.63 | 0.55 | 1.07 | 12.22 |
| N | 6 | 4 | 4 | 5 | 4 | 4 | 3 | 5 | 4 |
| Gp.2(10 ppm) | | | | | | | | | |
| Mean | 7.7 | 58.8 | 133.0 | 17.1 | 263.6 | 18.4 | 1.2 | 1.8 | 49.0 |
| S.D. | 4.19 | 50.95 | 15.19 | 9.17 | 95.05 | 4.78 | 0.46 | 0.70 | 14.71 |
| N | 4 | 4 | 4 | 4 | 5 | 4 | 5 | 5 | 4 |
| Gp.3(100 ppm) | | | | | | | | | |
| Mean | 5.2 | 92.2 | 123.0 | 6.9 | 232.8 | 15.3 | 1.9 | 1.8 | 40.5 |
| S.D. | 2.40 | 28.06 | 17.72 | 4.19 | 108.81 | 0.99 | 1.13 | 0.31 | 0.99 |
| N | 5 | 3 | 4 | 4 | 5 | 3 | 2 | 5 | 2 |
| Gp.4(1000 ppm) | | | | | | | | | |
| Mean | 1.9 | 203.6 | 94.5 | 11.0 | 182.9 | 37.4 | 2.0 | 1.2 | 34.4 |
| S.D. | 0.87 | 147.42 | 41.85 | 9.31 | 88.98 | 16.29 | 0.74 | 0.65 | 22.46 |
| N | 3 | 6 | 6 | 6 | 5 | 5 | 4 | 6 | 5 |
| Trend | -** | +** | -** | -* | NS | +** | NS | NS | -* |

NS=Not Significant at $p \geq 0.05$
*=Significant at $p \leq 0.05$
**=Significant at $p \leq 0.01$
Note page 53 of Final Report lists FSH trend as +**
Dixon's Test used to detect and eliminate outliers($p \leq 0.10$) before statistical analysis.
Statistical analyses: Terpstra-Jonckheere test;simple lirear regression; ANOVA);Dunnett's test.