Data Evaluation Report on Methods Development for the Study of Mechanism of Action of Atrazine in Adult and Metamorphosing *Xenopus laevis* and Rana clamitans: Aromatase Induction

EPA MRID Number 458677-08

Data Requirement:

EPA DP Barcode D288775

EPA MRID 458677-08
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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EPA PC Code 080803

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CITATION: Villeneuve, D. L., K. Coady, M. Hecker, M. B. Murphy, P. D. Jones and J. P. Giesy. 2003. Methods development for the study of mechanism of action of atrazine in adult and metamorphosing *Xenopus laevis* and *Rana clamitans*: aromatase induction. Aquatic Toxicology Laboratory, Michigan State University, National Food Safety and Toxicology Center, E. Lansing, MI. Sponsor: Syngenta Crop Protection, Inc., Laboratory Study ID ECORISK Number MSU-01.

EXECUTIVE SUMMARY:

In three separate studies, two involving adult male African clawed frogs (*Xenopus laevis*) and one with adult female *Xenopus*, frogs were exposed to either atrazine at 25 µg/L or to freshwater under static renewal conditions with 50% exposure solution changes every 72 hours. In the first study, 15 males were exposed for 26 days (single tank per treatment); in the second study 15 males were exposed for 43 days (single tank per treatment); and in the third study 13 females (6 in one replicate and 7 in the second replicate) were exposed for 47 days. Overall mortality was 3%, 7% and 19% in the 26, 43 and 47-day exposures, respectively; mortality was primarily associated with disease (fungal/bacterial) and was positively correlated (r=0.77) with the number of exposure days. Homogenates from a single testes and from brain were used to measure aromatase activity (CYP19 gene expression) in males. Aromatase activity in the testes was at or near the level of detection (LOD = 0.025 fmol/h/mg protein). Mean brain aromatase activity from atrazine-treated males (8.4 ± 4.2 fmol/h/mg protein) was not statistically different (p = 0.678) from controls (7.1 ± 4.2 fmol/h/mg protein) after 26 days of exposure. Following 43 days of exposure, mean brain aromatase activity in atrazine-treated males (5.8 ± 3.4 fmol/h/mg protein) was not statistically different (p = 0.199) from controls (10.4 ± 7.1 fmol/h/mg protein); however, in the second study, atrazine contaminated the controls at levels as high 0.25 µg/L. In the third exposure using female frogs, ovarian aromatase activity in atrazine-treated females averaged 4.5 ± 1.7 fmol/h/mg protein and did not differ statistically (p=0.447) from controls (5.4 ± 2.1 fmol/h/mg protein). Mean aromatase activity of brain homogenates from atrazine-treated females was 7.3 ± 5.0 fmol/h/mg protein did not differ significantly (p= 0.582) from control females (8.9 ± 4.2 fmol/h/mg protein).

With a single tank per treatment in the first two studies, it is not possible to document tank effects. Hhowever, the authors note that there was considerable variability in aromatase activity between frogs receiving the same treatment with coefficients of variability at or exceeding 100%. The third exposure where treatments were replicated confirmed that tank effects were high enough to potentially confound the ability of the study design to detect treatment effects. Additionally, with only one atrazine concentration tested, the ability of the study to discriminate potential "low-dose" effects is limited. Atrazine contamination of controls in the second study further confounded the test's ability to discriminate subtle effects.

The study recommends that future testing use replication, higher sample sizes and a broader range of atrazine levels to test for potentially subtle effects. Although not discussed in the study, water quality may have also compromised the study given the correlation of mortality with the number of days the frogs were confined and the references to the diseased state of the animals that had succumbed. Apparently, *Xenopus* are susceptible to bacterial septicemia (red-leg disease) if poor water quality persists.

Although it is indicated in the introduction that plasma sex steroids were also analyzed, these data and associated methods were not presented in this report. The only mention of sex steroid measurements was an indication that blood samples were collected by cardiac puncture.

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.
- Independent A Inspection was conducted of raw data.
- Final Report was written
- Raw data, documentation, records, protocols, and final report was archieved.

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* obtained from Xenopus Express® (Plant City, FL)

B. STUDY DESIGN:

Objective:

1. To test hypothesis that waterborne exposure to 25 µg/L atrazine could up-regulate aromatase activity in sexually mature *X. laevis*; male and female *X. laevis* were tested.

2. Test hypothesis that exposure to atrazine causes decreases in plasma testosterone and increases in estradiol (consistent with an increase in aromatase activity).

1. Experimental Conditions

a) Range-finding Study:

b) Definitive Study

Table 1 . Experimental Parameters

Parameter	Details
Acclimation: period: Conditions: (same as test or not) Feeding: Health: (any mortality observed)	 several weeks freshwater (treated well water) not reported mortality observed during study; frog with leision
Duration of the test	Exposure 1: 26-day Exposure 2: 43-day Exposure 3: 47-day
Test condition static/flow- through Type of dilution system for flow-through method. Renewal rate for static renewal	 static renewal NA 50% test solution change every 72 hours
Aeration, if any	not reported

Parameter	Details
Test vessel Material: (glass/stainless steel) Size: Fill volume:	 fiberglass 600 L 120 L
Source of dilution water Quality:	Treated well water (MSU-University Research Containment Facility;
Water parameters: Hardness pH Dissolved oxygen Total Organic carbon Particulate Matter Ammonia Nitrite Metals Pesticides Chlorine Temperature {Salinity for marine or estuarine species} Intervals of water quality measurement	 no water quality data provided at all No mention of whether water quality measurements were taken at all
Number of replicates/groups: negative control: water treated ones: atrazine at 25 µg/L	Exposure 1: single tank for each treatment (25 µg atrazine/L plus control) Exposure 2: single tank for each treatment (25 µg atrazine/L plus control) Exposure 3: 2 replicates per treatment

Parameter	Details
Number of organisms per replicate /groups:	Exposure 1: 15 adult frogs/treatment Exposure 2: 15 adult frogs/treatment Exposure 3: 6 adults in Rep 1 and 7 adults in Rep 2
Biomass loading rate	Exposure 1 and 2: 15 adults/120 L = 0.13 frogs/L Exposure 3: 6 to 7 adults/120 L = 0.05 - 0.06 frogs/L
Test concentrations: nominal: measured:	 25 µg/L Exposure 1: 30.6 ± 2.1 µg/L Exposure 2: 34 ± 8 µg/L Exposure 3: 32 ± 16 µg/L
Solvent (type, percentage, if used)	none
Lighting	not reported
Feeding	not reported
Recovery of chemical Level of Quantitation Level of Detection	ELISA (Envirogard Triazine®; Strategic Diagnostics Newark, DE) LOD 0.025 µg/L (Envirogard); 0.05 µg/L (Beacon)
Positive control {if used, indicate the chemical and concentrations}	none
Other parameters, if any	NA

2. Observations:

Table 2: Observations

Criteria	Details
Parameters measured including the sublethal effects/toxicity symptoms	Catalytic activity of aromatase (CYP19) measured in gonad and brain homogenates. Blood analyzed for steroid hormone levels
Observation intervals	daily
Were raw data included?	Yes
Other observations, if any	

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 appeared to differ in any substantive way, the difference was reported in the text below.]

Although blood was collected for steroid hormone analysis, these data were not reported in this study. Catalytic activity of aromatase (CYP19) was measured using the tritiated water release assay based on method of Lephart and Simpson (1991); protein concentration was determined using fluorescamine assay based on method of Kennedy and Jones (1994). Aromatase level was expressed as fmol/h/mg protein.

Atrazine concentrations in exposure tanks are based on samples collected following solution renewal after 72-hrs static exposure Only 50% of the exposure solutions were changed at renewal though.

Exposure 1 (26 days): one atrazine-treated frog died apparently from disease (open lesions on its legs). Single testes were taken for analysis; none of the testes homogenates had measurable aromatase activity; mean aromatase activity to the testicular homogenates was not statistically different among treatments (p=0.586). Positive control (*Xenopus* ovarian tissue).

Immature *Xenopus* ovarian exhibited roughly similar aromatase levels as 2 mg/mL bovine serum albumin; whereas *X. laevis* adult ovary contained roughly 26 fmol/h/mg protein. Mean aromatase activity from atrazine-treated males (8.4 ± 4.2 fmol/h/mg protein) was not statistically different (p = 0.678) from controls (7.1 ± 4.2 fmol/h/mg protein).

Atrazine concentrations averaged 30.6 ± 2.1 µg/L, while the control tank contained less than 0.025 µg/L (LOD for ELISA).

Exposure 2 (43 days): one animal from each treatment died of unknown causes. Animals were reported to appear healthy. Testicular homogenates had low to non-detectable levels of aromatase and again, there was no statistical difference (p = 0.764) between treatments. Positive control *X. laevis* ovarian homogenates yielded measurable aromatase activity, and activity was inhibited by addition of 7.5 µL of 559 µM 4-androstein-4-ol-3, 17-dione (a specific inhibitor of aromatase).

Atrzine-treated mean brain aromatase activity (5.8 ± 3.4 fmol/h/mg protein) was not statistically different (p = 0.199) from controls (10.4 ± 7.1 fmol/h/mg protein). A negative control (*Rana pipiens* ovarian homogenate) did not yield a significant response in the aromatase assay. There were significant differences among mean aromatase activity in replicate tanks (tank effects) within each treatment group.

Atrazine concentrations over the 43-day exposure period averaged 34 ± 8 µg/L but ranged as high as 50 µg/L. Atrazine in control tanks ranged up to 0.26 µg/L but average 0.07 ± 0.09 µg/L.

Exposure 3 (47 days): two atrazine-treated and three control females died; overall mortality was 19%. Fungal or bacterial infections were considered the probable cause of the mortalities although the surviving animals are reported as appearing healthy.

Ovarian aromatase activity in atrazine-treated females averaged 4.5 ± 1.7 fmol/h/mg protein and did not differ statistically (p=0.447) from controls (5.4 ± 2.1 fmol/h/mg protein). The report notes that overall ovarian aromatase activity was approximately 33 times greater than that observed for testicular homogenates from Exposure II.

The mean aromatase activity of brain homogenates from atrazine-treated females was 7.3 ± 5.0 fmol/h/mg; protein did not differ significantly (p= 0.582) from control females (8.9 ± 4.2 fmol/h/mg protein) .

Atrazine concentrations in each of the atrazine groups averaged 32 ± 16 µg/L and 28 ± 15 µg/L; however concentrations ranged as high as 70 µg/L. Atrazine concentrations in control tanks were generally less than the limit of detection and averaged 0.018 ± 0.02 µg/L and 0.016 ± 0.019 µg/L in the two replicates.

The report concludes that there were no statistically significant differences in mean brain homogenate aromatase activity among treatment groups in all three exposures (p= 0.75). For ovarian homogenates from exposure 3, there were no statistically significant differences between atrazine and control females. As a whole "the results of the three exposures do no support the hypothesis that waterborne exposure to approximately 25 µg/L atrazine causes changes in brain or gonadal aromatase activity in adult *X. laevis* relative to that in *X. laevis* exposed to laboratory water."

C. REPORTED STATISTICS:

D. VERIFICATION OF STATISTICAL RESULTS: Basic analyses in attached SAS© [Statistical Analysis System, Release 8.01, Cary, North Carolina] output)

E. STUDY DEFICIENCIES: Water quality data were not provided; the purity of atrazine was not provided; and atrazine was detected in the negative control in Exposure 2. In Exposures 1 and 3, mortality was attributed to disease. The feeding rate and lighting cycle were not discussed in the study.

F. REVIEWER'S COMMENTS:

It is unclear why a negative control (*Rana pipiens* ovarian homogenate) did not yield a significant response in the aromatase assay. In other words, why didn't aromatase from a different species convert testosterone to estrogen since the substrates are presumably the same?

In Exposure 2, atrazine in control tanks ranged up to 0.26 µg/L, but averaged 0.07 ± 0.09 µg/L. While the report states that this suggests "atrazine concentrations in control water was, on average, at least 485 times lower than that in atrazine-treated water," the levels detected were well-above the detection limit of the ELISA assay (LOD = 0.025). Atrazine in controls was at 0.1 µg/L on March 18, 0.25 µg/L on March 21, and 0.24 µg/L on March 30. Thus, exposure to atrazine in the controls was not an isolated event and may have confounded the ability of the study to detect treatment-related effects.

The level of total mortality was positively correlated (r=0.77) with the length of exposure and in most cases was associated with either bacterial and/or fungal disease.

In the second exposure, there were significant differences among mean aromatase activity in replicate tanks (tank effects) within each treatment group.

Although the report concluded that "the results of the three exposures do no support the hypothesis that waterborne exposure to approximately 25 µg/L atrazine causes changes in brain or gonadal aromatase activity in adult *X. laevis* relative to that in *X. laevis* exposed to laboratory water," the authors acknowledged that the magnitude of variability in gonadal and brain aromatase activity among individual frogs from the same treatment limited the power of the experiments to detect subtle differences even if they occurred. Large sample sizes would be required to test for subtle effects. Additionally, with only one tank per treatment in Exposures 1 and 2, the ability to account for tank effects is very limited and as the authors noted could potentially yield a false-negative or false positive response.

Furthermore, the studies only examined a single atrazine exposure concentration, i.e., 25 µg/L. The authors note that one hypothesis that has been raised is that high concentrations of atrazine may up-regulate aromatase to the point where the estrogen produced causes negative feedback on the hypothalmic-pituitary-axis resulting in down-regulation of aromatase expression. They conclude that future testing should use a wider range of concentrations.

Although it is indicated in the introduction that plasma sex steroids were also analyzed, these data and associated methods were not presented in this report. The only mention of sex steroid measurements was an indication that blood samples were collected by cardiac puncture.

G. CONCLUSIONS:

Aromatase activity in testes was too low to reliably quantify. While atrazine treatment of adult male *X. laevis* did not significantly affect brain aromatase activity relative to controls, the ability of the current studies to detect treatment effects appears limited. Since mortality over the three studies was positively correlated with the length of the study and because disease appeared to be an issue in two out of three studies, water quality may have been a problem; however, water quality data were not reported for the studies. Additionally, the second study (Exposure 2) was compromised by the discovery of atrazine in the control exposure. With a single tank per treatment (no replicates) and a single atrazine treatment (25 µg/L) per study, variability within treatments made it difficult to clearly differentiate treatment effects and to test the hypothesis.

H. REFERENCES:

Kennedy, S. W. and S. P. Jones. 1994. Simultaneous measurement of cytochrome P4501A catalyitc activity and total protein-concentration with fluorescence plate reader. Analytical Biochemistry 222(1): 217 - 223.

Laphart, E. D. and E. R. Simpson. 1991. Assay of aromatase activity. Methods in Enzymology 206: 477 - 483.

NONPARAMETRIC COMPARISON OF GONADAL AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED 264

Obs	SAMPLE	_TYPE_	_FREQ_	MEAN	STD	CV
1	A1	0	2	11.1919	5.11863	45.735
2	A10	0	2	2.1999	0.02387	1.085
3	A11	0	2	1.8184	0.09016	4.958
4	A12	0	2	0.6189	0.14642	23.658
5	A13	0	2	18.8785	0.89090	4.719
6	A14	0	2	-0.3838	0.34889	-90.914
7	A15	0	2	-1.7780	0.36618	-20.595
8	A2	0	2	19.5097	0.08537	0.438
9	A3	0	2	1.0145	0.35979	35.465
10	A4	0	2	13.9604	0.21978	1.574
11	A7	0	2	17.5941	0.01244	0.071
12	A8	0	2	6.4782	1.36797	21.116
13	A9	0	2	17.6115	0.57149	3.245
14	C1	0	2	-0.8158	0.25213	-30.907
15	C10	0	2	12.0858	0.74018	6.124
16	C11	0	2	14.7327	0.07916	0.537
17	C12	0	2	3.1996	0.31515	9.850
18	C13	0	2	7.2257	0.47387	6.558
19	C14	0	2	0.5245	0.04568	8.709
20	C15	0	2	12.5170	0.11766	0.940
21	C2	0	2	-0.0897	0.25792	-287.405
22	C3	0	2	14.6997	5.50738	37.466
23	C4	0	2	1.7527	0.36438	20.790
24	C5	0	2	1.2051	1.46082	121.218
25	C6	0	2	13.9926	3.28426	23.471
26	C8	0	2	18.9569	0.13915	0.734
27	C9	0	2	-0.2912	0.07412	-25.456

MEAN BRAIN AROMATASE ACTIVITY IN CONTROL AND ATRAZINE-TREATED FROGS (EXP 1) 265

Obs	GROUP	_TYPE_	_FREQ_	MEAN	STD	CV
1	ATRAZIN	0	13	8.36265	8.27945	99.0051
2	CONTROL	0	14	7.12111	7.06613	99.2279

ANOVA FOR BRAIN AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED FROGS (EXP 1) 266

The ANOVA Procedure

Class Level Information

Class	Levels	Values
GROUP	2	ATRAZIN CONTROL

Number of observations 27

Dependent Variable: MEAN

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	1	10.390314	10.390314	0.18	0.6780
Error	25	1471.684807	58.867392		
Corrected Total	26	1482.075121			

R-Square	Coeff Var	Root MSE	MEAN Mean
0.007011	99.39913	7.672509	7.718889

Source	DF	Anova SS	Mean Square	F Value	Pr > F
GROUP	1	10.39031393	10.39031393	0.18	0.6780

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

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
GROUP	1	1928.1	1928.1	1.54	0.2255
Error	25	31216.8	1248.7		

Bartlett's Test for Homogeneity of MEAN Variance

Source	DF	Chi-Square	Pr > ChiSq
GROUP	1	0.3013	0.5831

Dunnett's t Tests for MEAN

NOTE: This test controls the Type I experimentwise error for comparisons of all treatments against a control.

Alpha	0.05
Error Degrees of Freedom	25
Error Mean Square	58.86739
Critical Value of Dunnett's t	2.05959

Comparisons significant at the 0.05 level are indicated by ***.

GROUP Comparison	Difference Between Means	Simultaneous 95% Confidence Limits	
CONTROL - ATRAZIN	-1.242	-7.328	4.845

NONPARAMETRIC COMPARISON OF BRAIN AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED FR 270

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable MEAN
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
ATRAZIN	13	190.0	182.0	20.607442	14.615385
CONTROL	14	188.0	196.0	20.607442	13.428571

Wilcoxon Two-Sample Test

Statistic	190.0000
Normal Approximation	
Z	0.3639
One-Sided Pr > Z	0.3579
Two-Sided Pr > |Z|	0.7159
t Approximation	
One-Sided Pr > Z	0.3594
Two-Sided Pr > |Z|	0.7188

Z includes a continuity correction of 0.5.

Kruskal-Wallis Test

Chi-Square	0.1507
DF	1
Pr > Chi-Square	0.6979

Median Scores (Number of Points Above Median) for Variable MEAN
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
ATRAZIN	13	6.0	6.259259	1.321968	0.461538
CONTROL	14	7.0	6.740741	1.321968	0.500000

Median Two-Sample Test

Statistic	6.0000
Z	-0.1961
One-Sided Pr < Z	0.4223
Two-Sided Pr > |Z|	0.8445

Median One-Way Analysis

Chi-Square	0.0385
DF	1
Pr > Chi-Square	0.8445

MEAN GONADAL AROMATASE ACTIVITY IN CONTROL AND ATRAZINE-TREATED FROGS (EXP 1) 273

Obs	SAMPLE	_TYPE_	_FREQ_	MEAN	STD	CV
1	A1	0	2	0.40032	1.40426	350.79
2	A10	0	2	-0.06537	0.04967	-75.99
3	A11	0	2	0.00952	0.22021	2314.17
4	A12	0	2	-0.33007	0.17387	-52.68
5	A13	0	2	-0.04765	0.14865	-311.96
6	A14	0	2	-0.41950	0.00438	-1.04
7	A15	0	2	-0.14911	0.14151	-94.90
8	A2	0	2	-0.52470	0.90598	-172.67
9	A3	0	2	-0.39147	0.30402	-77.66
10	A4	0	2	-0.47445	0.00000	0.00
11	A5	0	2	0.11161	0.20003	179.21
12	A7	0	2	-0.21175	0.02073	-9.79
13	A8	0	2	-0.34509	0.02312	-6.70
14	A9	0	2	-0.08582	0.10868	-126.65
15	C1	0	2	-0.59029	0.29282	-49.61
16	C10	0	2	-0.84140	0.11866	-14.10
17	C11	0	2	-0.01878	0.11230	-597.83
18	C12	0	2	-0.19693	0.05968	-30.30
19	C13	0	2	-0.05506	0.02920	-53.03
20	C14	0	2	-0.03542	0.36070	-1018.23
21	C15	0	2	0.40039	0.08395	20.97
22	C2	0	2	-0.46908	0.05875	-12.52
23	C3	0	2	-0.27553	0.13632	-49.48
24	C4	0	2	-0.38774	0.48604	-125.35
25	C5	0	2	-0.03013	0.00000	0.00
26	C6	0	2	-0.27841	0.00000	0.00
27	C8	0	2	-0.33768	0.12295	-36.41
28	C9	0	2	-0.21742	0.07923	-36.44

MEAN GONADAL AROMATASE ACTIVITY IN CONTROL AND ATRAZINE-TREATED FROGS (EXP 1) 273

Obs	GROUP	_TYPE_	_FREQ_	MEAN	STD	CV
1	ATRAZIN	0	14	-0.18025	0.25669	-142.408
2	CONTROL	0	14	-0.23811	0.29774	-125.044

ANOVA FOR GONADAL AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED FROGS (EXP 1) 274

The ANOVA Procedure

Class Level Information

Class	Levels	Values
GROUP	2	ATRAZIN CONTROL

Number of observations 28

Dependent Variable: MEAN

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	1	0.02342954	0.02342954	0.30	0.5866
Error	26	2.00900245	0.07726932		
Corrected Total	27	2.03243199			

R-Square	Coeff Var	Root MSE	MEAN Mean
0.011528	-132.8879	0.277974	-0.209179

Source	DF	Anova SS	Mean Square	F Value	Pr > F
GROUP	1	0.02342954	0.02342954	0.30	0.5866

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

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
GROUP	1	0.00313	0.00313	0.25	0.6229
Error	26	0.3280	0.0126		

Bartlett's Test for Homogeneity of MEAN Variance

Source	DF	Chi-Square	Pr > ChiSq
GROUP	1	0.2744	0.6004

Dunnett's t Tests for MEAN

NOTE: This test controls the Type I experimentwise error for comparisons of all treatments against a control.

Alpha	0.05
Error Degrees of Freedom	26
Error Mean Square	0.077269
Critical Value of Dunnett's t	2.05558
Minimum Significant Difference	0.216

Comparisons significant at the 0.05 level are indicated by ***.

GROUP Comparison	Difference Between Means	Simultaneous 95% Confidence Limits	
CONTROL - ATRAZIN	-0.05785	-0.27382	0.15811

NONPARAMETRIC COMPARISON OF GONADAL AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED 278

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable MEAN
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
ATRAZIN	14	208.0	203.0	21.763884	14.857143
CONTROL	14	198.0	203.0	21.763884	14.142857

Wilcoxon Two-Sample Test

Statistic	208.0000
Normal Approximation	
Z	0.2068
One-Sided Pr > Z	0.4181
Two-Sided Pr > |Z|	0.8362
t Approximation	
One-Sided Pr > Z	0.4189
Two-Sided Pr > |Z|	0.8377

Z includes a continuity correction of 0.5.

Kruskal-Wallis Test

Chi-Square	0.0528
DF	1
Pr > Chi-Square	0.8183

Median Scores (Number of Points Above Median) for Variable MEAN
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
ATRAZIN	14	8.0	7.0	1.347151	0.571429
CONTROL	14	6.0	7.0	1.347151	0.428571

Median Two-Sample Test

Statistic	8.0000
Z	0.7423
One-Sided Pr > Z	0.2290
Two-Sided Pr > |Z|	0.4579

Median One-Way Analysis

Chi-Square	0.5510
DF	1
Pr > Chi-Square	0.4579

AVERAGE ATRAZINE CONCENTRATIONS IN CONTROL EXPOSURE TANKS BY DATE 280

Obs	MONTH	DAY	_TYPE_	_FREQ_	MEAN
1	3	3	0	2	0.03820
2	3	6	0	2	0.06553
3	3	9	0	2	0.01873
4	3	12	0	2	0.00515
5	3	15	0	2	0.03088
6	3	18	0	2	0.11453
7	3	21	0	2	0.26317
8	3	24	0	2	0.01652
9	3	30	0	2	0.22902
10	4	5	0	2	0.00473
11	4	8	0	2	0.00725

AVERAGE ATRAZINE CONCENTRATION IN CONTROL EXPOSURE TANK ACROSS ENTIRE STUDY (EXP 1) 281

Obs	_TYPE_	_FREQ_	MEAN	STD	CV
1	0	11	0.072156	0.092187	127.761

AVERAGE ATRAZINE CONCENTRATIONS IN 25 UG/L EXPOSURE 1 TANKS BY DATE 282

Obs	MONTH	DAY	_TYPE_	_FREQ_	MEAN
1	3	3	0	2	22.5051
2	3	6	0	2	32.4627
3	3	9	0	2	25.4194
4	3	12	0	2	25.8225
5	3	15	0	2	38.2852
6	3	18	0	2	34.8589
7	3	21	0	2	44.3471
8	3	24	0	2	49.5251
9	3	30	0	2	35.0879
10	4	5	0	2	33.9842
11	4	8	0	2	32.9775

AVERAGE ATRAZINE CONCENTRATION IN 25 UG/L EXPOSURE TANK ACROSS ENTIRE STUDY 283

Obs	_TYPE_	_FREQ_	MEAN	STD	CV
1	0	11	34.1160	8.02663	23.5275

MEAN BRAIN AROMATASE ACTIVITY FOLLOWING EXPOSURE 2 284

Obs	_TYPE_	_FREQ_	MEAN	STD	CV
1	0	28	8.07322	9.41514	116.622

ANOVA FOR BRAIN AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED FROGS (EXPOSURE 2 285

The ANOVA Procedure

Class Level Information

Class	Levels	Values
GROUP	2	EXP REF

Number of observations 28

Dependent Variable: FMOL

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	1	150.032040	150.032040	1.74	0.1988
Error	26	2243.380177	86.283853		
Corrected Total	27	2393.412216			

R-Square	Coeff Var	Root MSE	FMOL Mean
0.062685	115.0583	9.288910	8.073222

Source	DF	Anova SS	Mean Square	F Value	Pr > F
GROUP	1	150.0320396	150.0320396	1.74	0.1988

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

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
GROUP	1	70229.1	70229.1	3.77	0.0632
Error	26	484906	18650.2		

Bartlett's Test for Homogeneity of FMOL Variance

Source	DF	Chi-Square	Pr > ChiSq
GROUP	1	6.2027	0.0128

Dunnett's t Tests for FMOL

NOTE: This test controls the Type I experimentwise error for comparisons of all treatments against a control.

Alpha	0.05
Error Degrees of Freedom	26
Error Mean Square	86.28385
Critical Value of Dunnett's t	2.05558
Minimum Significant Difference	7.2169

Comparisons significant at the 0.05 level are indicated by ***.

GROUP Comparison	Difference Between Means	Simultaneous 95% Confidence Limits	
REF - EXP	4.630	-2.587	11.846

NONPARAMETRIC COMPARISON OF BRAIN AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED FR 289

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable FMOL
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
EXP	14	186.0	203.0	21.763884	13.285714
REF	14	220.0	203.0	21.763884	15.714286

Wilcoxon Two-Sample Test

Statistic	186.0000
Normal Approximation	
Z	-0.7581
One-Sided Pr < Z	0.2242
Two-Sided Pr > |Z|	0.4484
t Approximation	
One-Sided Pr < Z	0.2275
Two-Sided Pr > |Z|	0.4549

Z includes a continuity correction of 0.5.

Kruskal-Wallis Test

Chi-Square	0.6101
DF	1
Pr > Chi-Square	0.4347

Median Scores (Number of Points Above Median) for Variable FMOL
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
EXP	14	6.0	7.0	1.347151	0.428571
REF	14	8.0	7.0	1.347151	0.571429

Median Two-Sample Test

Statistic	6.0000
Z	-0.7423
One-Sided Pr < Z	0.2290
Two-Sided Pr > |Z|	0.4579

Median One-Way Analysis

Chi-Square	0.5510
DF	1
Pr > Chi-Square	0.4579

MEAN ADULT MALE GONADAL AROMATASE ACTIVITY IN ATRAZINE-TREATED AND CONTROL FROGS (EXP 2) 291

Obs	GROUP	_TYPE_	_FREQ_	MEAN	STD	CV
1	ATRAZIN	0	14	0.13818	0.09607	69.525
2	CONTROL	0	14	0.15601	0.19786	126.827

ANOVA FOR ADULT MALE GONADAL AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED FROGS (292

The ANOVA Procedure

Class Level Information

Class	Levels	Values
GROUP	2	ATRAZIN CONTROL

Number of observations 28

Dependent Variable: FMOL

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	1	0.00222571	0.00222571	0.09	0.7640
Error	26	0.62891899	0.02418919		
Corrected Total	27	0.63114470			

R-Square	Coeff Var	Root MSE	FMOL Mean
0.003526	105.7346	0.155529	0.147094

Source	DF	Anova SS	Mean Square	F Value	Pr > F
GROUP	1	0.00222571	0.00222571	0.09	0.7640

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

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
GROUP	1	0.00540	0.00540	1.08	0.3074
Error	26	0.1296	0.00499		

Bartlett's Test for Homogeneity of FMOL Variance

Source	DF	Chi-Square	Pr > ChiSq
GROUP	1	6.0347	0.0140

Dunnett's t Tests for FMOL

NOTE: This test controls the Type I experimentwise error for comparisons of all treatments against a control.

Alpha	0.05
Error Degrees of Freedom	26
Error Mean Square	0.024189
Critical Value of Dunnett's t	2.05558
Minimum Significant Difference	0.1208

Comparisons significant at the 0.05 level are indicated by ***.

GROUP Comparison	Difference Between Means	Simultaneous 95% Confidence Limits	
CONTROL - ATRAZIN	0.01783	-0.10300	0.13867

NONPARAMETRIC COMPARISON OF ADULT MALE GONADAL AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZI 296

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable FMOL
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
ATRAZIN	14	224.0	203.0	21.763884	16.0
CONTROL	14	182.0	203.0	21.763884	13.0

Wilcoxon Two-Sample Test

Statistic	224.0000
Normal Approximation	
Z	0.9419
One-Sided Pr > Z	0.1731
Two-Sided Pr > |Z|	0.3462
t Approximation	
One-Sided Pr > Z	0.1773
Two-Sided Pr > |Z|	0.3546

Z includes a continuity correction of 0.5.

Kruskal-Wallis Test

Chi-Square	0.9310
DF	1
Pr > Chi-Square	0.3346

Median Scores (Number of Points Above Median) for Variable FMOL
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
ATRAZIN	14	9.0	7.0	1.347151	0.642857
CONTROL	14	5.0	7.0	1.347151	0.357143

Median Two-Sample Test

Statistic	9.0000
Z	1.4846
One-Sided Pr > Z	0.0688
Two-Sided Pr > |Z|	0.1376

Median One-Way Analysis

Chi-Square	2.2041
DF	1
Pr > Chi-Square	0.1376

MEAN ATRAZINE LEVEL BY SAMPLING DATE FOR CONTROL (C) AND ATRAZINE (A) TREATED TANKS 298

Obs	MONTH	DAY	SAMPLE	GROUP	_TYPE_	_FREQ_	MEAN	STD	CV
1	4	17	A1	ATRAZINE	0	2	21.3668	0.25186	1.1788
2	4	17	A2	ATRAZINE	0	2	6.8501	0.20184	2.9465
3	4	17	C1	CONTROL	0	2	0.0072	0.00365	50.6899
4	4	17	C2	CONTROL	0	2	0.0041	0.00070	17.0096
5	4	20	A1	ATRAZINE	0	2	22.4385	2.50752	11.1751
6	4	20	A2	ATRAZINE	0	2	4.5276	1.69332	37.3999
7	4	20	C1	CONTROL	0	2	0.0070	0.00132	18.7495
8	4	20	C2	CONTROL	0	2	0.0254	0.00179	7.0668
9	4	23	A1	ATRAZINE	0	2	18.2464	3.42110	18.7495
10	4	23	A2	ATRAZINE	0	2	32.0647	4.89450	15.2644
11	4	23	C1	CONTROL	0	2	0.0769	0.00724	9.4163
12	4	23	C2	CONTROL	0	2	0.0771	0.00045	0.5894
13	5	2	A1	ATRAZINE	0	2	25.6397	2.86526	11.1751
14	5	2	A2	ATRAZINE	0	2	17.2724	6.55444	37.9475
15	5	2	C1	CONTROL	0	2	0.0234	0.00963	41.2067
16	5	2	C2	CONTROL	0	2	0.0186	0.00865	46.5324
17	5	5	A1	ATRAZINE	0	2	17.7872	4.56176	25.6463
18	5	5	A2	ATRAZINE	0	2	17.4941	5.47574	31.3004
19	5	5	C1	CONTROL	0	2	0.0057	0.00027	4.7134
20	5	5	C2	CONTROL	0	2	0.0044	0.00008	1.7681
21	5	8	A1	ATRAZINE	0	2	21.1917	0.49956	2.3573
22	5	8	C1	CONTROL	0	2	0.0068	0.00052	7.6546
23	5	8	C2	CONTROL	0	2	0.0048	0.00057	11.7606
24	5	11	C1	CONTROL	0	2	0.0636	0.01413	22.2114
25	5	11	C2	CONTROL	0	2	0.0121	0.00445	36.8512
26	5	14	C1	CONTROL	0	2	0.0202	0.00107	5.3020
27	5	14	C2	CONTROL	0	2	0.0140	0.00530	37.9475
28	5	17	C1	CONTROL	0	2	0.0064	0.00300	46.5324
29	5	17	C2	CONTROL	0	2	0.0072	0.00025	3.5356
30	5	20	C1	CONTROL	0	2	0.0058	0.00219	37.9475
31	5	20	C2	CONTROL	0	2	0.0040	0.00073	18.1701
32	5	23	C1	CONTROL	0	2	0.0110	0.00672	61.1231
33	5	23	C2	CONTROL	0	2	0.0203	0.00892	43.8868
34	5	26	C1	CONTROL	0	2	0.0094	0.00094	10.0029
35	5	26	C2	CONTROL	0	2	0.0177	0.00239	13.5146
36	5	29	C1	CONTROL	0	2	0.0055	0.00197	35.7501
37	5	29	C2	CONTROL	0	2	0.0055	0.00065	11.7606
38	6	1	C1	CONTROL	0	2	0.0064	0.00101	15.8467
39	6	1	C2	CONTROL	0	2	0.0047	0.00087	18.7494

MEAN ATRAZINE CONCENTRATION ACROSS SAMPLING DATES FOR EACH OF THE TWO CONTROL (C) AND ATRAZI 299

Obs	SAMPLE	GROUP	_TYPE_	_FREQ_	MEAN	STD	CV
1	A1	ATRAZINE	0	6	21.1117	2.8844	13.662
2	A2	ATRAZINE	0	5	15.6418	10.9158	69.786
3	C1	CONTROL	0	14	0.0182	0.0229	125.436
4	C2	CONTROL	0	14	0.0157	0.0191	121.373

MEAN ATRAZINE CONCENTRATION IN CONTROL AND ATRAZINE-TREATED TANKS OVER ENTIRE STUDY (EXP 3 300

Obs	GROUP	_TYPE_	_FREQ_	MEAN	MIN	MAX	STD	CV
1	ATRAZINE	0	11	18.6254	4.52761	32.0647	7.74480	41.582
2	CONTROL	0	28	0.0170	0.00403	0.0771	0.02070	121.980

MEAN BRAIN AROMATASE ACTIVITY BY SAMPLE AND NUMBER IN CONTROL (C) AND ATRAZINE-TREATETED (A) 301

Obs	SAMPLE	NUMBER	GROUP	_TYPE_	_FREQ_	MEAN	STD	CV
1	A1	1	ATRAZINE	0	2	20.9131	0.52971	2.5329
2	A1	2	ATRAZINE	0	2	1.9509	0.02533	1.2982
3	A1	3	ATRAZINE	0	2	1.3884	0.00843	0.6070
4	A1	4	ATRAZINE	0	2	3.8467	0.18493	4.8075
5	A1	5	ATRAZINE	0	2	5.6692	0.02706	0.4774
6	A1	6	ATRAZINE	0	2	0.9863	0.02995	3.0367
7	A2	1	ATRAZINE	0	2	7.5578	0.14937	1.9764
8	A2	2	ATRAZINE	0	2	8.7285	1.14801	13.1524
9	A2	3	ATRAZINE	0	2	7.6883	0.05418	0.7047
10	A2	4	ATRAZINE	0	2	-0.0514	0.00540	-10.5056
11	A2	5	ATRAZINE	0	2	21.1885	0.02985	0.1409
12	C1	1	CONTROL	0	2	11.8105	0.01860	0.1575
13	C1	2	CONTROL	0	2	2.1175	0.20755	9.8015
14	C1	3	CONTROL	0	2	6.2637	0.10634	1.6977
15	C1	4	CONTROL	0	2	21.4478	0.23323	1.0874
16	C1	5	CONTROL	0	2	4.1599	0.02262	0.5438
17	C2	1	CONTROL	0	2	15.0054	2.98537	19.8953
18	C2	2	CONTROL	0	2	1.0907	0.04346	3.9843
19	C2	3	CONTROL	0	2	7.0063	0.10293	1.4692
20	C2	4	CONTROL	0	2	8.3976	0.10458	1.2454
21	C2	5	CONTROL	0	2	12.2114	0.15241	1.2481

MEAN BRAIN AROMATASE ACTIVITY BY SAMPLE IN CONTROL AND ATRAZINE-TREATED FROGS (EXP 3) 302

Obs	SAMPLE	_TYPE_	_FREQ_	MEAN	STD	CV
1	A1	0	6	5.79244	7.61168	131.407
2	A2	0	5	9.02234	7.65441	84.838
3	C1	0	5	9.15987	7.76212	84.741
4	C2	0	5	8.74230	5.31368	60.781

MEAN BRAIN AROMATASE ACTIVITY IN CONTROL AND ATRAZINE-TREATED FROGS (EXP 3) 303

Obs	GROUP	_TYPE_	_FREQ_	MEAN	STD	CV
1	ATRAZINE	0	11	7.26057	7.43303	102.375
2	CONTROL	0	10	8.95109	6.27499	70.103

ANOVA FOR BRAIN AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED FROGS (EXP 3) 304

The ANOVA Procedure

Class Level Information

Class	Levels	Values
GROUP	2	ATRAZINE CONTROL

Number of observations 21

Dependent Variable: MEAN

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	1	14.9695821	14.9695821	0.31	0.5820
Error	19	906.8789562	47.7304714		
Corrected Total	20	921.8485383			

R-Square	Coeff Var	Root MSE	MEAN Mean
0.016239	85.65689	6.908724	8.065579

Source	DF	Anova SS	Mean Square	F Value	Pr > F
GROUP	1	14.96958206	14.96958206	0.31	0.5820

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

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
GROUP	1	1145.7	1145.7	0.31	0.5865
Error	19	71107.0	3742.5		

Bartlett's Test for Homogeneity of MEAN Variance

Source	DF	Chi-Square	Pr > ChiSq
GROUP	1	0.2554	0.6133

Dunnett's t Tests for MEAN

NOTE: This test controls the Type I experimentwise error for comparisons of all treatments against a control.

Alpha	0.05
Error Degrees of Freedom	19
Error Mean Square	47.73047
Critical Value of Dunnett's t	2.09309

Comparisons significant at the 0.05 level are indicated by ***.

GROUP Comparison	Difference Between Means	Simultaneous 95% Confidence Limits	
CONTROL - ATRAZINE	1.691	-4.628	8.009

NONPARAMETRIC COMPARISON OF BRAIN AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED FR 308

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable MEAN
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
ATRAZINE	11	107.0	121.0	14.200939	9.727273
CONTROL	10	124.0	110.0	14.200939	12.400000

Wilcoxon Two-Sample Test

Statistic	124.0000
Normal Approximation	
Z	0.9506
One-Sided Pr > Z	0.1709
Two-Sided Pr > |Z|	0.3418
t Approximation	
One-Sided Pr > Z	0.1766
Two-Sided Pr > |Z|	0.3531

Z includes a continuity correction of 0.5.

Kruskal-Wallis Test

Chi-Square	0.9719
DF	1
Pr > Chi-Square	0.3242

Median Scores (Number of Points Above Median) for Variable MEAN
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
ATRAZINE	11	5.0	5.238095	1.171274	0.454545
CONTROL	10	5.0	4.761905	1.171274	0.500000

Median Two-Sample Test

Statistic	5.0000
Z	0.2033
One-Sided Pr > Z	0.4195
Two-Sided Pr > |Z|	0.8389

Median One-Way Analysis

Chi-Square	0.0413
DF	1
Pr > Chi-Square	0.8389

MEAN GONADAL AROMATASE ACTIVITY BY SAMPLE AND NUMBER IN CONTROL (C) AND ATRAZINE-TREATETED (310

Obs	SAMPLE	NUMBER	GROUP	_TYPE_	_FREQ_	MEAN	STD	CV
1	A1	1	ATRAZINE	0	2	4.3309	0.13618	3.1445
2	A1	2	ATRAZINE	0	2	3.8744	0.02522	0.6510
3	A1	3	ATRAZINE	0	2	7.3398	0.02316	0.3155
4	A1	4	ATRAZINE	0	2	5.1150	0.06204	1.2129
5	A1	5	ATRAZINE	0	2	8.3364	0.01106	0.1327
6	A1	6	ATRAZINE	0	2	7.3282	0.02467	0.3366
7	A2	1	ATRAZINE	0	2	1.7495	0.02643	1.5105
8	A2	2	ATRAZINE	0	2	4.9783	0.12473	2.5055
9	A2	3	ATRAZINE	0	2	3.3030	0.07831	2.3710
10	A2	4	ATRAZINE	0	2	0.2169	0.02263	10.4327
11	A2	5	ATRAZINE	0	2	2.8070	0.34990	12.4655
12	C1	1	CONTROL	0	2	3.5497	0.02948	0.8304
13	C1	2	CONTROL	0	2	1.6790	0.04290	2.5551
14	C1	3	CONTROL	0	2	4.5738	0.50058	10.9444
15	C1	4	CONTROL	0	2	3.9795	0.02188	0.5497
16	C1	5	CONTROL	0	2	3.1596	0.03938	1.2463
17	C2	1	CONTROL	0	2	10.6332	0.14104	1.3264
18	C2	2	CONTROL	0	2	9.6644	0.09341	0.9665
19	C2	3	CONTROL	0	2	3.3400	1.00685	30.1450
20	C2	4	CONTROL	0	2	6.0585	0.00074	0.0122
21	C2	5	CONTROL	0	2	7.5248	0.31761	4.2209

MEAN GONADAL AROMATASE ACTIVITY BY SAMPLE IN CONTROL AND ATRAZINE-TREATED FROGS (EXP 3) 311

Obs	SAMPLE	_TYPE_	_FREQ_	MEAN	STD	CV
1	A1	0	6	6.05412	1.84866	30.5355
2	A2	0	5	2.61093	1.77449	67.9639
3	C1	0	5	3.38835	1.09028	32.1774
4	C2	0	5	7.44418	2.90987	39.0892

MEAN GONADAL AROMATASE ACTIVITY IN CONTROL AND ATRAZINE-TREATED FROGS (EXP 3) 312

Obs	GROUP	_TYPE_	_FREQ_	MEAN	STD	CV
1	ATRAZINE	0	11	4.48904	2.49031	55.4753
2	CONTROL	0	10	5.41627	2.97674	54.9592

ANOVA FOR GONADAL AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED FROGS (EXP 3) 313

The ANOVA Procedure

Class Level Information

Class	Levels	Values
GROUP	2	ATRAZINE CONTROL

Number of observations 21

Dependent Variable: MEAN

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
Model	1	4.5034675	4.5034675	0.60	0.4468
Error	19	141.7650045	7.4613160		
Corrected Total	20	146.2684720			

R-Square	Coeff Var	Root MSE	MEAN Mean
0.030789	55.40006	2.731541	4.930574

Source	DF	Anova SS	Mean Square	F Value	Pr > F
GROUP	1	4.50346755	4.50346755	0.60	0.4468

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

Source	DF	Sum of Squares	Mean Square	F Value	Pr > F
GROUP	1	28.6086	28.6086	0.49	0.4920
Error	19	1107.1	58.2672		

Bartlett's Test for Homogeneity of MEAN Variance

Source	DF	Chi-Square	Pr > ChiSq
GROUP	1	0.2867	0.5923

Dunnett's t Tests for MEAN

NOTE: This test controls the Type I experimentwise error for comparisons of all treatments against a control.

Alpha	0.05
Error Degrees of Freedom	19
Error Mean Square	7.461316
Critical Value of Dunnett's t	2.09309

Comparisons significant at the 0.05 level are indicated by ***.

GROUP Comparison	Difference Between Means	Simultaneous 95% Confidence Limits	
CONTROL - ATRAZINE	0.9272	-1.5709	3.4253

NONPARAMETRIC COMPARISON OF GONADAL AROMATASE ACTIVITY BETWEEN CONTROL AND ATRAZINE-TREATED 317

The NPAR1WAY Procedure

Wilcoxon Scores (Rank Sums) for Variable MEAN
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
ATRAZINE	11	113.0	121.0	14.200939	10.272727
CONTROL	10	118.0	110.0	14.200939	11.800000

Wilcoxon Two-Sample Test

Statistic	118.0000
Normal Approximation	
Z	0.5281
One-Sided Pr > Z	0.2987
Two-Sided Pr > |Z|	0.5974
t Approximation	
One-Sided Pr > Z	0.3016
Two-Sided Pr > |Z|	0.6032

Z includes a continuity correction of 0.5.

Kruskal-Wallis Test

Chi-Square	0.3174
DF	1
Pr > Chi-Square	0.5732

Median Scores (Number of Points Above Median) for Variable MEAN
Classified by Variable GROUP

GROUP	N	Sum of Scores	Expected Under H0	Std Dev Under H0	Mean Score
ATRAZINE	11	5.0	5.238095	1.171274	0.454545
CONTROL	10	5.0	4.761905	1.171274	0.500000

Median Two-Sample Test

Statistic	5.0000
Z	0.2033
One-Sided Pr > Z	0.4195
Two-Sided Pr > |Z|	0.8389

Median One-Way Analysis

Chi-Square	0.0413
DF	1
Pr > Chi-Square	0.8389