DATA EVALUATION RECORD
AQUATIC INVERTEBRATE LIFE CYCLE TEST
§ 72-4(b)

1. **CHEMICAL:** Methomyl PC Code No.: 090301

2. **TEST MATERIAL:** Methomyl technical Purity: 98.6%

3. **CITATION:**

Authors: Ward, T.J., J.P. Magazu and R.L. Boeri

Title: Methomyl Technical: Flow-Through Chronic Toxicity to the Mysid, *Americamysis bahia* (Formerly Known as *Mysidopsis bahia*).

Study Completion Date: October 1, 1999

Laboratory: T.R. Wilbury Laboratories, Inc.
40 Doaks Lane
Marblehead, Massachusetts 01945

Sponsor: E.I. DuPont de Nemours and Company
Wilmington, Delaware 19898

Laboratory Report ID: DuPont-1157

MRID No.: 45013203

DP Barcode: D264036

4. **REVIEWED BY:** Gregory S. Hess, Staff Scientist, Dynamac Corporation

Signature: **Date:** 7/26/01

APPROVED BY: Kathleen Ferguson, Ph.D., Senior Staff Scientist, Dynamac Corporation

Signature: **Date:** 7/26/01

5. **APPROVED BY:** Todd Phillips, Ph.D., Biologist, OPP/EFED/ERBIV

Signature: **Date:** 5/13/04

6. STUDY PARAMETERS:

Scientific Name of Test Organisms: *Americamysis bahia*

Age of Test Organism: <24 Hours old

Definitive Test Duration 28 days

Study Method: Flow-through

Type of Concentrations: Mean-measured

7. CONCLUSIONS:

In this 28-day life cycle toxicity test, *Americamysis bahia* neonates were exposed under flow-through conditions to Methomyl technical (98.6% purity) at mean measured concentrations of 0 (negative control), 0.0145, 0.0291, 0.0591, 0.121 and 0.253 mg a.i./L. Measured values were 85% to 101% of nominal concentrations. Prior to sexual maturity and pairing, there were 60 mysids/treatment level: 15 mysids/compartment, 2 compartments/chamber, and 2 replicate chambers/concentration. Following pairing, there were 16 to 20 mysid pairs/treatment: one pair/compartment, 6 to 10 compartments/chamber, and 2 chambers/concentration.

First-generation mysids were observed for mortality and signs of abnormal behavior once daily throughout the study. On day 7, three mysids exposed to 0.121 mg/L were swimming erratically; however, since this effect occurred in only one replicate and on only one day, it was not believed to be significant. Second-generation mysids were counted and discarded once daily during the reproduction period. Endpoints included survival of first-generation mysids (female and combined sexes), number of young produced per female, number of young/number of reproductive days, and wet and dry weight (sexes separated) and length (sexes separated) of surviving first-generation mysids by 28 days.

Prior to pairing on Day 15, the study authors reported the 7 day EC50 for immobility (mortality) to be 0.153 mg/L with 95% confidence limits of 0.121 to 0.253 mg/L. The 14 day EC50 for immobility was 0.147 mg a.i./L with 95% confidence limits of 0.121 to 0.253 mg a.i./L. The 21 day and 28 day EC50 values for immobility were 0.141 and 0.131 mg a.i./L with 95% confidence limits of 0.121 to 0.253 and 0.0591 to 0.253 mg a.i./L, respectively.

Mean lengths ranged from 7.1 to 7.5 mm, mean wet weights ranged from 3.2 to 3.4 mg, and mean dry weights ranged from 0.62-0.70 mg for all treatment groups, including the controls.

The most sensitive measure of toxicity determined by statistical analysis of survival, growth,

and reproduction data was the number of young per surviving female. **The NOEC was 0.0291, the LOEC was 0.0591, and the MATC was 0.0415 mg a.i./L**.

This study is classified as Supplemental because of the deviations from the guideline requirements for an aquatic invertebrate life-cycle toxicity test using *Americamysis bahia* (72-4(b)).

Results Synopsis:

Survival

NOEC: 0.121 mg a.i./L
LOEC: 0.253 mg a.i./L
MATC: 0.175 mg a.i./L
LC_{50}/EC_{50} (95% C.I.): 0.131 (0.0591-0.253) mg a.i./L

Reproduction (number of young per surviving female)

NOEC: 0.0291 mg a.i./L
LOEC: 0.0591 mg a.i./L
MATC: 0.0415 mg a.i./L
LC_{50}/EC_{50} (95% C.I.): 0.059 (0.023-0.16) mg a.i./L

Wet Weight

NOEC: 0.121 mg a.i./L
LOEC: 0.253 mg a.i./L
MATC: 0.175 mg a.i./L
LC_{50}/EC_{50} (95% C.I.): >0.253 mg a.i./L

Dry Weight

NOEC: 0.121 mg a.i./L
LOEC: 0.253 mg a.i./L
MATC: 0.175 mg a.i./L
LC_{50}/EC_{50} (95% C.I.): >0.253 mg a.i./L

Length

NOEC: 0.121 mg a.i./L
LOEC: 0.253 mg a.i./L
MATC: 0.175 mg a.i./L
LC_{50}/EC_{50} (95% C.I.): >0.253 mg a.i./L

8. ADEQUACY OF THE STUDY:

A. Classification: Supplemental

B. Rationale: This study is classified as Supplemental because of the deviations from the guideline requirements for an aquatic invertebrate life-cycle toxicity test using *Americamysis bahia* (72-4(b)). Specifically, second-generation mysids should have been maintained for at least 4 days to monitor survival, development, and behavior.

C. Repairability: This study can be upgraded to core if additional data is submitted that addresses the deviations listed in the next section.

9. GUIDELINE DEVIATIONS:

1) Survival of first-generation mysids should have been provided for individual sexes. The reviewer was unable to derive these data for males from any of the raw data tables provided.

2) Second-generation mysids should have been maintained for at least 4 days to monitor survival, development, and behavior.

3) On day 27, dead males in paired cages were not replaced with extra males.

4) Temperature was not continuously recorded throughout the entire definitive toxicity test; it was not recorded for all of day 6 and a portion of day 7.

5) Mysids were not fed on the last day of the test.

6) Analytical retention times were slightly outside the range specified in the protocol.

7) The analytical LOQ was less than twice as low as the lowest tested concentration (LOQ = 0.010 ppm).

8) The number of mysid pairs ranged from 16 to 20 pairs/treatment instead of the recommended 20 pairs/treatment.

10. SUBMISSION PURPOSE: Reregistration

11. MATERIALS AND METHODS:

A. Test Organisms/Acclimation

Guideline Criteria	Reported Information
Species An estuarine shrimp species, preferably *Americamysis bahia*	*Americamysis bahia*
Source/Supplier	In-house cultures which were started with mysids from a commercial supplier (Aquatic BioSystems, Inc., Fort Collins, Colorado).
Age at Beginning of Test <24 hours old	<24 hours old
Parental Acclimation Parental stock must be maintained separately from the brood culture in dilution water and under test conditions. Mysids should be in good health.	During acclimation, parent mysids were maintained under test conditions. Parental mysids were not treated for disease and were free of apparent disease, injuries, and abnormalities at the time neonates to be used in the test were collected.
Parental Acclimation Period At least 14 days	14 Days
Brood Stock Test started with mysids from: - one brood stock, or - brood stock which has not obtained sexual maturity or had been maintained for >14 days in a laboratory with same food, water, temperature, and salinity used in the test.	During the 21 day period before test start, the temperature range was 23.9 to 24.3°C, the pH was 7.6 to 8.0, the salinity was 14 to 16 ppt, and dissolved oxygen concentration was always at least 7.5 mg/L. Mysids were fed live brine shrimp, *Artemia salina*.

B. Test System

Guideline Criteria	Reported Information
Source of Dilution Water May be natural (sterilized and filtered) or a commercial mixture; water must be free of pollutants.	Natural seawater collected from the Atlantic Ocean at T.R. Wilbury Lab in Marblehead, Mass. and adjusted to a salinity of 15-17 ppt with deionized water. The water was carbon filtered, passed through a 20 µm particle filtered, UV sterilized and aerated continuously prior to use. A 5 µm filter and UV sterilizer were placed on the dilution water feed line just prior to the diluter. Periodic contaminant analysis was provided.
Does water support test animals without observable signs of stress?	Yes.
Water Temperature 27°C for mysids - At test termination, mean-measured temperature for each chamber should be within 1°C of selected test temperature. - Must be within 3°C of the mean of the time-weighted averages. - Must not differ by >2°C between chambers during the same interval.	Mean: 25.1 ± 0.5°C Range: 23.1-26.6°C
Salinity 15-30 ‰ - The difference between highest and lowest measured salinities should be less than 5 ‰.	Range: 13-16 ‰
pH 7.6 and 8.2	Range: 7.7-8.1
Dissolved Oxygen 60-100% saturation	Range: 7.3-8.3 mg O_2/L; 100% saturation in 15 ppt SW at 25°C is 7.6 mg O_2/L; 60% saturation in 15 ppt SW at 25°C is 4.6 mg O_2/L (p. 43 of the definitive study)

Guideline Criteria	Reported Information
Photoperiod 16-hr light/8-hr dark (14-hr light/10-hr dark also acceptable)	16-hr light/8-hr dark, including a 15 minute transition period between light and dark.
Test Chambers 1. Material: All glass, No. 316 stainless steel, or perflorocarbon plastic 2. Size: Typically 30 x 45 x 15 cm (20.25 L) 3. Fill depth: 10 cm 4. Were chambers identical and covered during the test?	1. Glass 2. 20-L aquaria (20 x 40 x 25 cm) 3. Up to 8 L fill volume; fill depth not reported 4. All chambers were identical; chambers were loosely covered.
Test Compartments (within chambers) - 250-mL glass beakers with side cutouts covered with nylon mesh or stainless steel screen, or - 90- or 140-mm id glass Petri dish bottoms with collars made of 200-250 µm mesh screen	- Prior to pairing (on Day 15), mysids were maintained in two retention chambers consisting of two petri dishes (approx. 10 cm diameter with approx. 13 cm high Nitex® screen (typically 350um mesh) collar attached with silicone adhesive). - Following pairing, the reproductive compartments were 6-cm diameter x 13 cm high glass Petri dishes covered with nylon mesh.
Type of Dilution System Intermittent flow proportional diluters or continuous flow serial diluters should be used.	Intermittent low proportional diluter

Guideline Criteria	Reported Information
Toxicant Mixing 1. Mixing chamber is recommended but not required; aeration should not be used for mixing. 2. If a mixing chamber was not employed, was it demonstrated that the test solution was completely mixed before introduction into the test system? 3. Was flow splitting accuracy within 10%?	1. Mixing chambers not employed. Aeration not reported. 2. Not reported 3. Not reported
Flow Rate 1. 5-10 volume additions per 24 hours. 2. Did the flow rate maintain the toxicant level and the DO at ≥60% of saturation? 3. Were the meter systems calibrated before study and checked twice daily during test period?	1. 9 volume additions every 24 hours 2. Yes 3. Yes. The diluter was calibrated before and after the test, and observed for normal operation twice daily.
Solvents - Acceptable solvents include triethylene glycol, ethanol, acetone, and methanol. - Solvent should not exceed 0.1 mL/L in a flow-through system.	None
Aeration Dilution water should be vigorously aerated, but the test tanks should not be aerated.	Dilution water was aerated but test tanks were not.

C. Test Design

Guideline Criteria	Reported Information
Duration of the Test Approximately 28 days. Was the test terminated within 7 days of the median time of first brood release in the controls?	28 Days No; The day of first brood release in the control group was Day 16 and the study was not terminated within 7 days of this date.
Nominal Concentrations Negative control, a solvent control (when applicable), and at least five treatment levels, one of which must adversely affect a life stage and one must not affect any life stage. The dilution factor should not be >50%.	Negative control and 0.017, 0.033, 0.063, 0.130 and 0.250 mg a.i./L
Distribution Number of mysids before pairing: Minimum of 15 mysids per compartment, 2 compartments per chamber, 2 chambers per concentration for a total of 60/treatment level. Number of mysids after pairing: ≥20 randomly selected pairs/treatment (excess males should be held in separate compartment in same treatment to replace paired males).	Total = 60 mysids/treatment level: 15/mysids per compartment, two compartments per chamber, and two replicate chambers per concentration. Actual number ranged from 6 - 10 pairs/treatment. One pair per compartment, 6 - 10 compartments per chamber, and 2 replicate chambers per concentration.
Pairing Should be conducted when most of the mysids are sexually mature, usually 10-14 days after test initiation. All pairing should occur on the same day.	All pairing was conducted on Day 15.
Test organisms randomly or impartially assigned to test vessels?	Yes
Were treatments randomly assigned to individual test chamber locations?	Yes

Guideline Criteria	Reported Information
Feeding Mysids should be fed live brine shrimp nauplii at least once daily. 150 live brine shrimp nauplii per mysid per day or 75 twice a day is recommended.	Juvenile mysids were fed live brine shrimp (*Artemia* sp.) nauplii three times a day at a rate of approximately 150 brine shrimp/mysid/day except during the final 17 hours before the determination of wet and dry weight.
Counts Live adult mysids should be counted at initiation, at pairing, and daily after pairing. Live young must be counted and removed daily. Missing or impinged animals should be recorded.	Live adult mysids were counted once daily throughout the test. After pairing, live young were counted and removed once daily until test termination. Missing or impinged animals were recorded.
Controls Negative control and carrier control (when applicable) are required.	Negative control was included.
Water Parameter Measurements 1. Temperature should be monitored daily in one chamber and at least three times in all chambers. 2. Salinity should be measured daily in at least one test vessel. 3. pH should be measured at the beginning, the end, and at least weekly during the test in the control vessels and highest test level. 4. Dissolved oxygen must be measured at each concentration at least once a week.	Temperature was measured daily in each replicate test chamber. Also measured continuously in one negative control replicate except on day 6 and a portion of day 7 when the recorder failed. Salinity, pH and DO were measured in each replicate test chamber daily.

Guideline Criteria	Reported Information
Chemical Analysis Toxicant concentration must be measured in one chamber at each toxicant level every week.	Toxicant concentration was measured on Days 0, 7, 14, 21, and 28. Analysis was performed using HPLC in conjunction with a diode array detector. The LOQ was 0.010 mg a.i./L, the LOD was 0.0050 mg/L.

Comments: 2nd generation mysids were not maintained for at least 4 days to monitor survival, development, and behavior.

12. REPORTED RESULTS

A. General Results

Guideline Criteria	Reported Information
Quality assurance and GLP compliance statements were included in the report?	Yes
Chemical Analysis For all test groups, a) the measured concentration of the test material should not be <50% of the time-weighted average measured concentration for >10% of the duration of the test, and b) the measured concentration should not be >30% of the time-weighted average measured concentration for >5% of the duration of the test.	- All criteria met.

Guideline Criteria	Reported Information
Controls - Survival of the paired first-generation controls must be ≥70%. - ≥75% of the paired first-generation female controls produced young, or - The average number of young produced by the first-generation female controls was ≥3.	- Yes, survival was 83%. - Not reported - Average number of young produced was 4.6.
Data Endpoints Must Include 1. Survival of first-generation mysids, gender specified 2. Number of live young produced per female 3. Dry weight and length of each first generation mysid alive at the end of the test, gender specified **Data Endpoints Should Also Include** 4. Incidence of morphological findings. 5. Survival, development, and behavior of second-generation mysids for at least 4 days.	Data Endpoints Included 1. Survival of first-generation mysids noted, but not gender specified 2. Yes 3. Yes 4. Yes, morphological findings (abnormal development) were evaluated, but none were observed in this study. 5. Not addressed in study Results and Discussion.
Raw data must include 1. Survival of first-generation mysids, gender specified 2. Number of live young produced per female 3. Terminal weight and length measurements, individual and gender specified	1. Survival of first-generation mysids was noted, but not gender specified. 2. Provided 3. Provided

Toxicity Observations: No sublethal effects other than growth and reproductive effects were noted during the test except for three mysids exposed to 0.121 mg a.i./L that were swimming erratically on day 7. The authors assumed that since this effect occurred only in the replicate on this one day, it was not believed to be significant.

B. Statistical Results:

Statistical Method: Data that were statistically analyzed included: (1) the number of first generation mysids surviving 28 days of exposure, (2) the number of young per surviving female after 28 days of exposure (defined as the sum of the total number of young each day divided by the number of surviving females that day), (3) total length of surviving first generation mysids at the conclusion of the test, (4) blotted wet weight of surviving first generation mysids at the conclusion of the test, and (5) dry weight of surviving first generation mysids at the conclusion of the test.

No sublethal effects (other than effects on growth and reproduction) were observed at any concentration at test termination; hence, statistical analysis of sublethal effect data was not warranted.

Data were analyzed by standard statistical techniques (ASTM, 1990) using TOXSTAT version 3.5 (Gulley et al., 1996) and Stephan (1983). A Shapiro-Wilk's test was used to determine if data were normally distributed and Bartlett's test was used to determine if variances were homogeneous. A one-way analysis of variance (ANOVA) and Dunnett's test were then used to compare treatment and control means for reproduction and dry weight and William's test (non-parametric) was used for survival, length and wet weight data. Survival data were arc sine [square root (Y)] transformed prior to analysis. All calculations were performed using mean measured concentrations of the active ingredient. The EC50 values for immobility and 95% confidence limits were calculated for days 7, 14, 21 and 28 by the binomial method. The NOEC is the highest tested concentration at which no measured biological parameter is statistically different ($\propto = 0.05$) from the controls. The LOEC is the lowest tested concentration at which any measured biological parameter is statistically different ($\propto = 0.05$) from the controls. The MATC is the geometric mean of the NOEC and the LOEC.

Most sensitive endpoint: Reproduction (number of young per surviving female).

Results Synopsis

Endpoint	Method	NOEL	LOEL	LC_{50}/EC_{50} (95% C.I.)[a]	MATC
Survival	William's	0.121	0.253	0.131 (0.0591-0.253)	0.175
Reproduction	ANOVA/Dunnett's	0.0291	0.0591	NA	0.041
Wet weight	William's	0.121	0.253	NA	0.175
Dry weight	ANOVA/Dunnett's	0.121	0.253	NA	0.175
Length	William's	0.121	0.253	NA	0.175

[a] The EC50 value reported by the study authors was based upon day 28 immobility, which was assumed to be synonymous with survival by the reviewer and, thus, equivalent to an LC50 value.

13. VERIFICATION OF STATISTICAL RESULTS:

Statistical Method: Survival data were subjected to visual analysis followed by tests to confirm adverse treatment effects and analyzed for normality and homogeneity of variance prior to statistical comparisons with Dunnett's test for multiple comparisons via TOXSTAT software. Reproduction, length, wet and dry weight data were subjected to visual analysis followed by William's tests via TOXSTAT software to assess treatment effects after confirming normality and homogeneity of variance. EC50 values were estimated for reproduction, wet and dry weight and length with the probit method via NUTHATCH software. The LC50 based upon mortality data was estimated via the probit method using TOXANAL software.

Endpoint	Method	NOEL	LOEL	LC_{50}/EC_{50} (95% C.I.)	MATC
Survival	ANOVA/Dunnet's test for multiple comparisons	0.121	0.253	0.106 (0 to ∞)*	0.175
Reproduction	William's	0.0291	0.0591	0.059 (0.023-0.16)	0.0415
Wet weight	William's	0.121	0.253	> 0.253	0.175
Dry weight	William's	0.121	0.253	> 0.253	0.175
Length	William's	0.121	0.253	>0.253	0.175

*The reviewer determined the LC50 value using the probit method because the number of organisms studied was too large for the binomial method.

The most sensitive endpoint measured was reproduction (number of young per female):

NOEC: 0.0291 mg a.i./L
LOEC: 0.0591 mg a.i./L

EC50: 0.059 mg a.i./L 95% C.I.: 0.023 to 0.16 mg a.i./L
MATC: 0.0415 mg a.i./L

14. REVIEWER'S COMMENTS:

This study is classified as Supplemental because of the deviations listed in the previous section. However, this study does provide useful information regarding the chronic toxicity to mysids exposed to technical methomyl.

15. REFERENCES:

ASTM. 1990. Standard Guide for Conducting Life-Cycle Toxicity Tests with Saltwater Mysids. Designation E 12191-90. Approved March 30, 1990.

Gulley, D.D., A.M. Boelter and H.L. Bergman. 1996. TOXSTAT Version 3.5. Fish Physiology and Toxicology Laboratory, University of Wyoming, Laramie, Wyoming.

Japan MAFF. 1984. Good Laboratory Practice Standard. 59 NohSan No. 3850.

OECD. 1992. OECD Principles of Good Laboratory Practice. OECD Series on Principles of Good Laboratory Practice and Compliance Monitoring. Number 1, Environmental Monograph No. 45 OCDE/GD(92)32. Paris.

Stephan, C.E. 1983. Computer Program for the Calculation of LC_{50} Values. Personal Communication.

Stuck, H.C., H.M. Perry and R.W. Heard. 1979. Annotated Key to the Mysidacea of the North Central Gulf of Mexico. Gulf Research Reports, Vol. 6, No. 3, 225-238.

U.S. EPA. 1985. Standard Evaluation Procedure. Fish Early Life Stage. Hazard Evaluation Division, Office of Pesticide Programs. Washington, D.C.

U.S. EPA 1988. Pesticide Assessment Guidelines. Subdivision E, Hazard Evaluation: Wildlife and Aquatic Organisms. 72-4. Fish Early Life Stage and Aquatic Invertebrate Life-Cycle Studies. Ecological Effect Branch, Hazard Evaluation Division, Office of Pesticide Programs, Washington, D.C. Draft, March 1988.

U.S. EPA. 1993a. 40 CFR Part 160. Federal Insecticide, Fungicide, and Rodenticide Act (FIFRA); Good Laboratory Practice Standards; Final Rule.

U.S. EPA. 1993b. 40 CFR Part 792. Toxic Substances Control Act (TSCA); Good Laboratory Practice Standards. Final Rule.

U.S. EPA. 1998. Reregistration Eligibility Decision (RED). Methomyl EPA-738-F-019. 248 p.

16. OUTPUT OF REVIEWER'S STATISTICAL VERIFICATION:

Survival:

Title: 13203
File: 13203 . Transform: ARC SINE(SQUARE ROOT(Y))

ANOVA Table

SOURCE	DF	SS	MS	F
Between	5	1.7023	0.3405	25.0372
Within (Error)	6	0.0816	0.0136	
Total	11	1.7839		

(p-value = 0.0006)

Critical F = 8.7459 (alpha = 0.01, df = 5,6)
= 4.3874 (alpha = 0.05, df = 5,6)

Since F > Critical F REJECT Ho: All equal (alpha = 0.05)

Title: 13203
File: 13203 . Transform: ARC SINE(SQUARE ROOT(Y))

Dunnett's Test - TABLE 1 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	TRANSFORMED MEAN	MEAN CALCULATED IN ORIGINAL UNITS	TRANS T STAT	SIG 0.05
1	0	1.1458	0.8300		
2	0.0145	1.2490	0.9000	-0.8853	
3	0.0291	0.9814	0.6850	1.4100	
4	0.0591	0.9916	0.7000	1.3222	
5	0.121	0.8698	0.5800	2.3670	
6	0.253	0.0914	0.0000	9.0419	*

Dunnett critical value = 2.8300 (1 Tailed, alpha = 0.05, df = 5,6)

Title: 13203
File: 13203 . Transform: ARC SINE(SQUARE ROOT(Y))

Dunnett's Test - TABLE 2 OF 2 Ho:Control<Treatment

GROUP	IDENTIFICATION	NUM OF REPS	MIN SIG DIFF (IN ORIG. UNITS)	% OF CONTROL	DIFFERENCE FROM CONTROL
1	0	2			
2	0.0145	2	0.2996	36.1	-0.0700
3	0.0291	2	0.2996	36.1	0.1450

4	0.0591	2	0.2996	36.1	0.1300
5	0.121	2	0.2996	36.1	0.2500
6	0.253	2	0.2996	36.1	0.8300

Reproduction:
45013203
File: 3203rd Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	control	2	4.600	4.600	4.775
2	0.0145	2	4.950	4.950	4.775
3	0.0291	2	3.350	3.350	3.350
4	0.0591	2	1.650	1.650	1.800
5	0.121	2	1.950	1.950	1.800
6	0.253	2	0.000	0.000	0.000

45013203
File: 3203rd Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
control	4.775				
0.0145	4.775	0.190		1.94	k= 1, v= 6
0.0291	**3.350**	**1.358**		**2.06**	**k= 2, v= 6**
0.0591	1.800	3.043	*	2.10	k= 3, v= 6
0.121	1.800	3.043	*	2.12	k= 4, v= 6
0.253	0.000	4.999	*	2.13	k= 5, v= 6

s = 0.920
Note: df used for table values are approximate when v > 20.

Length:
45013203
File: 3203ld Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	control	2	7.450	7.450	7.450
2	0.0145	2	7.450	7.450	7.450
3	0.0291	2	7.350	7.350	7.350

4	0.0591	2	7.300	7.300	7.300
5	0.121	2	7.150	7.150	7.150
6	0.253	2	0.000	0.000	0.000

45013203
File: 32031d Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
control	7.450				
0.0145	7.450	0.000		1.94	k= 1, v= 6
0.0291	7.350	0.522		2.06	k= 2, v= 6
0.0591	7.300	0.783		2.10	k= 3, v= 6
0.121	**7.150**	**1.567**		**2.12**	**k= 4, v= 6**
0.253	0.000	38.905	*	2.13	k= 5, v= 6

s = 0.191
Note: df used for table values are approximate when v > 20.

Wet weight:
45013203
File: 3203wd Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	control	2	3.250	3.250	3.350
2	0.0145	2	3.400	3.400	3.350
3	0.0291	2	3.400	3.400	3.350
4	0.0591	2	3.300	3.300	3.300
5	0.121	2	3.300	3.300	3.300
6	0.253	2	0.000	0.000	0.000

45013203
File: 3203wd Transform: NO TRANSFORMATION

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
control	3.350				
0.0145	3.350	0.385		1.94	k= 1, v= 6
0.0291	3.350	0.385		2.06	k= 2, v= 6

0.0591	3.300	0.192		2.10	k= 3, v= 6
0.121	**3.300**	**0.192**		**2.12**	**k= 4, v= 6**
0.253	0.000	12.509	*	2.13	k= 5, v= 6

s = 0.260
Note: df used for table values are approximate when v > 20.

Dry weight:
45013203
File: 3203dd Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 1 OF 2

GROUP	IDENTIFICATION	N	ORIGINAL MEAN	TRANSFORMED MEAN	ISOTONIZED MEAN
1	control	2	0.635	0.635	0.675
2	0.0145	2	0.695	0.695	0.675
3	0.0291	2	0.695	0.695	0.675
4	0.0591	2	0.660	0.660	0.660
5	0.121	2	0.635	0.635	0.635
6	0.253	2	0.000	0.000	0.000

45013203
File: 3203dd Transform: NO TRANSFORM

WILLIAMS TEST (Isotonic regression model) TABLE 2 OF 2

IDENTIFICATION	ISOTONIZED MEAN	CALC. WILLIAMS	SIG P=.05	TABLE WILLIAMS	DEGREES OF FREEDOM
control	0.675				
0.0145	0.675	1.141		1.94	k= 1, v= 6
0.0291	0.675	1.141		2.06	k= 2, v= 6
0.0591	0.660	0.713		2.10	k= 3, v= 6
0.121	0.635	0.000		2.12	k= 4, v= 6
0.253	0.000	18.106	*	2.13	k= 5, v= 6

s = 0.035
Note: df used for table values are approximate when v > 20.