

**REPORT FOR ACUTE TOXICITY TESTING OF
APPLIED POLYMER SYSTEMS, INC.
SILT STOP PRODUCTS**

NORCROSS, GEORGIA

TEST PERIOD: OCTOBER 3-18, 2000

Prepared for:

APPLIED POLYMER SYSTEMS, INC.

Norcross, Georgia

October 2000

**REPORT FOR ACUTE TOXICITY TESTING OF
APPLIED POLYMER SYSTEMS, INC.
SILT STOP PRODUCTS**

NORCROSS, GEORGIA

TEST PERIOD: OCTOBER 3-18, 2000

Prepared for:

**Applied Polymer Systems, Inc.
Norcross, Georgia**

Prepared by:

**BioTox Laboratory
Law Engineering and Environmental Services, Inc.
Kennesaw, Georgia**

October 2000

October 30, 2000

Mr. Steven R. Iwinski
Applied Polymer Systems, Inc.
4015 Holcomb Bridge Rd.
Suite 350-931
Norcross, GA 30092

Subject: **Applied Polymer Systems, Inc. Acute Toxicity Testing, October 3-18, 2000**

Dear Mr. Iwinski:

Law Engineering and Environmental Services, Inc. (LAW), BioTox Laboratory has completed 48-hr. acute static toxicity testing on seven polymer samples of Silt Stop Products submitted by you on September 29, 2000 and October 12, 2000. Toxicity testing was initiated by LAW on October 3 and 16, 2000 to determine the LC₅₀ (the concentration of the sample at which 50% of the test organism, *Daphnia magna*, die). Samples were prepared according to your instruction and diluted with laboratory formulated water to the following test concentrations: 26.25 ppm, 52.5 ppm, 105 ppm, 210 ppm, and 420 ppm. Test results indicated no acute mortality to *Daphnia magna* in the 702AA, 702B, 702C, 705, 730B, or the 740 polymer samples. Since these samples did not exhibit 50% or greater mortality, an LC₅₀ was not calculable. However, samples 703D (with flocculant), 703D (without flocculant), and 770 exhibited mortality rates below 50%. The LC₅₀ was calculated as 383 ppm for 703D (with flocculant), 105 ppm for 703D (without flocculant), and 98 ppm for sample 770.

Results are summarized in the accompanying report. Detailed testing documentation, including chemical and physical measurements (Appendix A), test organism documentation (Appendix B), and polymer sample test data/reference toxicant data (Appendix C).

LAW appreciates the opportunity to provide these testing services to you and Applied Polymer Systems, and we look forward to serving your future needs. If there are any questions, please do not hesitate to contact Mr. Andrew S. Peiken at (770) 421-7027.

Sincerely,

LAW ENGINEERING AND ENVIRONMENTAL SERVICES, INC.

Andrew S. Peiken
BioTox Laboratory Manager

Margaret E. Tanner
Environmental Engineer

Attachment: Data Report

TEST SUMMARY

I. Client

Client: Applied Polymer Systems, Inc.
4015 Holcomb Bridge Rd.
Suite 350-931
Norcross, GA 30092

Product Tested: Silt Stop Products: 702AA, 702B, 702C, 703D (with flocculant),
703D (without flocculant), 705, 730B, 740, 770

Laboratory: BioTox Laboratory
Law Engineering and Environmental Services, Inc.
3200 Town Point Drive NW, Suite 100
Kennesaw, Georgia 30144

II. Laboratory Accreditation

Laboratory: BioTox Laboratory
Law Engineering and Environmental Services, Inc.
3200 Town Point Drive N.W., Suite 100
Kennesaw, Georgia 30144

Accreditor: State of Florida, Dept. of Health and Rehabilitative Services
Office of Laboratory Services, Environmental Water

Accreditation ID: Lab ID. E87477, Certificate No. 477-128

Category: Bioassay

Effective: July 1, 2000 through June 30, 2001

III. Tests Conducted

Test: Water Flea, *Daphnia magna*
48-hr. Acute Static Toxicity Test
EPA/600/4-90/027F, Fourth Edition, Sept. 1991

Test Dates: October 3-5, 2000, October 4-6, 2000, and October 16-18, 2000

Source, Age of Organisms: In-house cultures, ID No. MAGM04092700 / <24-hr old
and MAGM042100600 / <24 hr old

Test Concentrations: 26.25, 52.5, 105, 210, and 420 ppm

Dilution/Control Water: Blend of Diluted Mineral Water (20% Perrier/80% Milli-Q) and
Moderately hard synthetic freshwater (reagent grade chemicals
and Milli-Q)

Deviation From Test Protocol:

Upon preparing polymer sample 703D for testing, a thread-like material or flocculant was observed in the water column of the sample. The toxicity test performed October 3-6, 2000 indicated acute toxicity to the water flea. It was noted that the water fleas were physically entrapped in the material, possibly causing the observed acute toxicity. The test was re-run on October 16, 2000 with the thread-like material removed from the sample prior to testing.

IV. Results

Survival Data for *Daphnia magna* Exposed for 48-hr to Applied Polymer Systems, Inc. Silt Stop Products, October 3-6, 2000 and October 16-18, 2000.

Sample (ppm)	Survival at 48-hours (%)	LC ₅₀ (ppm)
702AA		
Laboratory Control ^a	100	>420
26.25	100	
52.5	100	
105	100	
210	100	
420	90	
702B		
Laboratory Control ^a	100	>420
26.25	100	
52.5	100	
105	95	
210	95	
420	94.4	
702C		
Laboratory Control ^a	100	>420
26.25	95	
52.5	100	
105	100	
210	100	
420	94.7	
702D (Silt Stop 100)		
Laboratory Control ^a	100	383
26.25	100	
52.5	100	
105	85	
210	50 ^b	
420	65 ^b	
702D (Silt Stop 400)		
Laboratory Control ^a	100	105
26.25	100	
52.5	100	
105	50 ^b	
210	30 ^b	
420	15 ^b	

Survival Data for *Daphnia magna* Exposed for 48-hr to Applied Polymer Systems, Inc. Silt Stop Products, October 3-6, 2000 and October 16-18, 2000 (continued).

702A		
Laboratory Control ^a	100	>100
26.25	100	
52.5	100	
105	95	
210	100	
420	90	
702B		
Laboratory Control ^a	100	>100
26.25	95	
52.5	100	
105	100	
210	100	
420	100	
702C		
Laboratory Control ^a	100	>100
26.25	100	
52.5	100	
105	100	
210	100	
420	100	
703D		
Laboratory Control ^a	100	98
26.25	100	
52.5	65 ^b	
105	50 ^b	
210	25 ^b	
420	0 ^b	

Calculated by: ASP 10/23/00
Checked by: RFP 10/23/00

^a Laboratory Control – Moderately hard synthetic freshwater.

^b Significantly different from the laboratory control using statistical comparisons.

V. Summary

48-hour acute toxicity testing was conducted on eight polymer samples of Silt Stop Products submitted by you on September 29 and October 12, 2000. Toxicity testing was initiated on October 3 and 16, 2000 to determine the LC₅₀ (the concentration of the sample at which 50% of the test organism, *Daphnia magna*, die). Samples were prepared according to your instruction and diluted with laboratory formulated water to the following test concentrations: 26.25 ppm, 52.5 ppm, 105 ppm, 210 ppm, and 420 ppm. Test results indicated no acute mortality to *Daphnia magna* in the 702AA, 702B, 702C, 705, 730B, or the 740 polymer samples. Since these samples did not exhibit 50% or greater mortality, an LC₅₀ was not calculable. However, samples 703D (with flocculant), 703D (without flocculant), and 770 exhibited mortality rates below as 50%. The LC₅₀ was calculated as 383 ppm for 703D (with flocculant), 105 ppm for 703D (without flocculant), and 98 ppm for sample 770.

VI. Quality Assurance

Daphnia magna, NaCl Reference Toxicant, 007, 10/10/00

Survival: 48-hr LC₅₀ = 5278 mg/L NaCl (PASS)

48-hr LC₅₀ Range of Acceptability = 2640 to 7480 mg/L NaCl

GLOSSARY AND ABBREVIATIONS

Acute	Involving a stimulus severe enough to rapidly induce a response; in toxicity tests, a response observed in 96 hours or less typically is considered acute.
Chronic	Involving a stimulus that lingers or continues for a relatively long period of time, often one-tenth of the life span or more. A chronic effect can be lethality, growth, reduced reproduction, etc.
Chronic Value	A numeric value representing the geometric mean of the numeric values of concentrations analyzed as the NOEC (No Observed Effect Concentration) and the LOEC (Lowest Observed Effect Concentration) by chronic toxicity testing. The chronic value is an estimate of the toxicant concentration that will be the actual no effect concentration based on the chronic effect tested.
Critical Value	Minimum numeric value for a toxicity test endpoint (i.e., survival, growth, or reproduction) below which a given test result will be statistically significantly different from the control value.
DMW	Diluted Mineral Water
EC	Effective concentration, a point estimate of the toxicant concentration that would cause an adverse response such as death, immobilization, or serious incapacitation.
Ft-c	Foot candles - a measure of <u>light</u> intensity
Graphical Method	Log concentration versus percent mortality method. Toxicity test data are plotted on 2-cycle semi-log graph paper. The logarithmic axis (y axis) is used for percent effluent concentration, and the linear axis (x axis) is used for percent mortality. The graph provides a reasonably accurate estimate of the LC ₅₀ , but does not provide a confidence interval.
IC	Inhibition Concentration, a point estimate of the toxicant concentration that would cause a given percent reduction in a biological measurement such as fecundity or growth.
LC	Lethal Concentration, identical to EC when the observed response is death.
LC ₅₀	The toxicant concentration that is lethal to 50 percent of exposed organisms at a specific time of observation.
LCL	Lower 95% Confidence Limit
LOEC	Lowest-Observed-Effect-Concentration, the lowest concentration of toxicant to which organisms are exposed that causes adverse effects.
LWC	Lab Water Control, moderately hard synthetic freshwater prepared from MILLIPORE MILLI-Q ^R water and reagent grade chemicals.

NOEC	No-Observed-Effect-Concentration, the highest concentration of toxicant to which organisms are exposed that causes no observable adverse effects.
Probit Analysis	Probit Analysis consists of a group of statistical methods used to analyze data from concentration-response experiments, and provides an estimate of the LC ₅₀ and the precision of this estimate. In Probit Analysis, the percentages of affected organisms are converted to Probits (probability units), and the effluent concentrations are converted to logarithms. The relationship between the Probits and the logarithmic values of the concentrations is approximately linear. A Probit regression line drawn through the data points is used to estimate the LC ₅₀ and its precision estimate. To use Probit Analysis, at least two partial mortalities must be obtained in the toxicity test.
RWC	Receiving Water Control
UCL	Upper 95% Confidence Limit
μE/m ² /s	Micro-ergs per square meter per second - a measure of <u>light</u> intensity

REFERENCES

- Gulley, D.D., and WEST, Inc. 1996. TOXSTAT version 3.5. Fish Physiology and Toxicology Laboratory. Department of Zoology and Physiology. University of Wyoming. Laramie, Wyoming.
- Peltier, W., and C.I. Weber. eds. 1991. Methods for Measuring the Acute Toxicity of Effluents to Freshwater and Marine Organisms. Fourth Edition. Environmental Monitoring and Support Laboratory, U.S. Environmental Protection Agency, Cincinnati, OH. EPA 600/4-90/027F.
- U.S. Environmental Protection Agency. 1979. Methods for Chemical Analysis of Water And Wastes. Environmental Monitoring Laboratory, U.S. Environmental Protection Agency, Cincinnati, OH. EPA 600/4-79-020.
- U.S. Environmental Protection Agency. 1999. Errata for Effluent and Receiving Water Toxicity Testing Manuals: Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms; Short-Term Methods for Estimating the Chronic Toxicity of Effluent and Receiving Waters to Freshwater Organisms; and Short-Term Methods for Estimating the Chronic Toxicity of Effluents and Receiving Waters to Marine and Estuarine Organisms. U.S. Environmental Protection Agency, Office of Research and Development, Duluth, MN. EPA/600/R-98/182.

Summary of Effluent Toxicity Test Conditions for the Water Flea (*Daphnia magna*) Acute Survival Test (EPA 600/4-90/027F)

1. Test type:	48-hr Static Acute
2. Temperature (°C):	20.0-21.0°C
3. Light quality:	Ambient laboratory illumination
4. Light intensity:	10-20 uE/m ² /s (50-100 ft-c) (ambient laboratory levels)
5. Photoperiod:	16-hr light, 8-hr dark
6. Test chamber size:	30 mL
7. Test solution volume:	25 mL/replicate
8. Age of test organisms:	<24-hr
9. No. neonates per test chamber:	5
10. No. replicate chambers per concentration:	4
11. No. neonates per concentration:	20
12. Feeding regime:	Fed 0.1 each of <i>Selenastrum</i> /YCT prior to test start and at 48-hr
13. Aeration:	None
14. Dilution water:	Blend of Diluted Mineral Water (20% Perrier/80% Milli-Q) and Moderately hard synthetic freshwater (reagent grade chemicals and Milli-Q)
15. Effluent concentrations (ppm):	26.25, 52.5, 105, 210, and 420
16. Dilution factor:	0.5
17. Test duration	48-hr
18. Effect measured:	Mortality - no movement on gentle prodding
19. Test acceptability:	90% or greater survival in controls
20. Sample handling:	Whole polymer samples held at room temperature.
21. Sample volume required:	1 L
22. Chemical parameters:	pH, DO, total residual chlorine, total alkalinity, total hardness, conductivity, temperature

Initial Chemical Characterization of Applied Polymer Systems, Inc. Silt Stop Products and Control Water Used in Acute Toxicity Tests, October 3-5, 2000, October 4-6, 2000, and October 16-18, 2000.

Parameter	420 ppm Polymer Sample									
	702AA	702B	702C	703D with flocculant	703D without flocculant	705	730 B	740	770	Control ^a
Dissolved Oxygen (mg/L)	7.40	7.50	7.30	7.40	7.50	7.40	7.40	7.20	7.30	7.70
pH	7.89	7.84	7.94	7.12	7.53	7.87	7.95	7.86	10.35	7.82
Total Alkalinity as CaCO ₃ (mg/L)	59.5	54.5	57.0	22.0	17.0	54.5	59.5	102	52.0	59.5
Total Hardness as CaCO ₃ (mg/L)	135.5	119.5	79.5	119.5	115.5	79.5	93.5	77.5	59.5	75.5
Conductivity @ 25°C (µmhos/cm)	351	321	241	335	322	243	253	260	276	221
Total Ammonia ^b (mg/L)	0.56 ^c	1.5 ^c	0.36 ^c	0.41 ^c	0.39 ^c	0.66 ^c	0.63 ^c	0.49 ^c	<0.01	<0.01

Determined according to EPA 600/4-79-020, 1979.

Entered by: ASP 10/23/00

Checked by: RFP 10/23/00

^a Control used in the water flea test was a blend of Diluted Mineral Water (20% Perrier/80% Milli-Q) and Moderately hard synthetic freshwater (reagent grade chemicals and Milli-Q)

^b Determined by Hach Colorimetric Test Kit (DPD Method in conjunction with spectrophotometer)

^c Caution should be used when interpreting ammonia data as there was interference noted (formation of a white flocculant) in the samples upon addition of the Nessler reagent. When ammonia is present, a sample will exhibit an orange color change upon addition of test reagents. In the Silt Stop polymer samples, there was no orange coloration, indicating that ammonia is not present and the observed measurement of ammonia by the spectrophotometer is artifactual and caused by the interference of the flocculant.

APPENDICES

APPENDIX A

Chemical and Physical Data

APPENDIX B

Test Organism Documents

LAW Engineering and Environmental Services, Inc.
BioTox Laboratory

Daphnia magna In-house Culture Source Document

Brood Board ID No.	Brood Board Date	Date/Time Cups Marked	Age of Neonates at Test Start	Water Type/Temp. (°C)	Food Type
MAGM040092700 and MAGM042100600	9/27/00 and 10/06/00	10/3/00 (0900), 10/4/00 (0900), and 10/16/00 (0900)	<24 hrs.	DMW/LWC (20.0)	YCT & <i>Selenastrum</i>

APPENDIX C

Test Data Sheets

**Water Flea
Polymer Test**

**Water Flea
Reference Toxicant Data Sheets**