VISCO, INC.
SEAL-ONCE

48-Hour Acute Toxicity Test Report

Mysidopsis bahia

Menidia beryllina

July 2003

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48-HOUR LC50 PRODUCT REPORT

Client.....Visco, Inc.	Project No.: DYN-3
Sample.....SEAL-ONCE	Test Date: .....July 2003

INTRODUCTION

A product identified as SEAL-ONCE (wood preservative) was delivered to Huther and Associates from Visco, Inc. on May 19, 2003. Two acute LC50 toxicity tests were requested: a static acute 48-hour definitive toxicity test using Mysidopsis Bahia (opossum shrimp) and a static acute 48-hour definitive toxicity test using Menidia beryllina (silverside minnow). Test procedures followed recommended methods contained in "*Methods for Measuring the Acute Toxicity of Effluents and Receiving Waters to Freshwater and Marine Organisms, Fourth Edition*", EPA/600/4-90/027F, August 1993.

SAMPLE PREPARATION

The product was prepared at a ratio of two parts water: one part product following the manufacturer's recommendation. The product went readily into solution. Three test concentrations were prepared by adding 0.037 mL of the two parts water: one part product (stock) to 1,000 mL laboratory water (0.0123 ml/L after dilution factor), 0.370 mL of stock to 1,000 mL laboratory water (0.123 ml/L after dilution factor), and 3.70 mL of stock to 1,000 mL laboratory water (1.230 ml/L after dilution factor). Laboratory water and control water were distilled, deionized water reconstituted with sea salts to a salinity of 26 ppt.

TEST DESIGN

Mysidopsis bahia

The 48-hour static, non-renewal, definitive *Mysidopsis bahia* test was conducted in 250 mL beakers containing 200 mL of test solution. The test was initiated July 8, 2003. Ten *M. bahia* juveniles were added to each of four replicate beakers per concentration. Juveniles originated from laboratory cultures and were five days old at test initiation. Juveniles were fed laboratory cultured *Artemia nauplii* during test exposure. The test was conducted for 48-hours during which survival was recorded daily.

A control of four replicate beakers containing ten *M. bahia* juveniles each in laboratory saltwater was conducted concurrently with the test. The test was terminated on July 10, 2003. Survival data was statistically analyzed using the Trimmed Spearman-Karber point estimate test to determine the lethal concentration to fifty percent (50%) of the test population (LC50) and associated 95% confidence limits.

RESULTS

Mysidopsis bahia ONCE:

The following estimated LC50 value was determined for SEAL-

48-Hour Definitive Test				
Conc. (ml/L)	# Exposed	# Alive	# Dead	% Survival
Control	40	39	1	97.5
0.0123	40	33	7	82.5
0.1230	40	19	21	52.5
1.2300	40	0	40	0.0

Percent Spearman-Karber Trim:	15.38%
Estimated LC50 (ml/L):	**0.10**
95% Upper C.L. (ml/L):	0.17
95% Lower C.L. (ml/L):	0.06

TEST DESIGN

The 48-hour static, non-renewal, definitive *Menidia beryllina* test was conducted in 250 mL beakers containing 200 mL of test solution. Ten *M. beryllina* larvae were added to each of four replicate beakers per concentration. Larvae originated from laboratory cultures and were ten days old at test initiation. Larvae were fed laboratory cultured *Artemia nauplii* during test exposure. The test was conducted for 48-hours during which survival was recorded daily.

A control of four replicate beakers containing ten M. beryllina larvae each in laboratory saltwater was conducted concurrently with the test. The test was terminated on July 10, 2003. Survival data was statistically analyzed using the Trimmed Spearman-Karber point estimate test to determine the lethal concentration to fifty percent (50%) of the test population and the associated ninety-five percent (95%) confidence limits.

RESULTS

The following estimated LC5O value was determined for SEAL-ONCE:

48-Hour Definitive Test				
Conc. (ml/L)	# Exposed	# Alive	# Dead	% Survival
Control	40	39	1	97.5
0.0123	40	40	0	100.0
0.1230	40	23	17	57.5
1.2300	40	0	40	0.0

Percent Spearman-Karber Trim:	0.00%
Estimated LC50 (ml/L):	**0.15**
95% Upper C.L. (ml/L):	0.21

95% Lower C.L. (ml/L):	0.06

DISCUSSION

Mysidopsis bahia was determined to be the more sensitive species to SEAL-ONCE, with an estimated 48-hour LC50 concentration of 0.10 ml/L. The estimated 48-hour LC50 concentration to Menidia beryllina was 0.15 ml/L.