COVANCE
THE DEVELOPMENT SERVICES COMPANY

AMENDMENT TO FINAL REPORT

Study Number:	2901/001	
Amendment Issue Date:	July 2009	
Amendment Number:	1	
Study Title:	PFH Ammonium Salt: Fish, Early Life Stage Toxicity Test to *Oncorhynchus mykiss* (Rainbow Trout)	
Authorised By:		
		Date 15 July 09
Quality Assurance Audit:		
		Date 15 July 09
Distribution:	Sponsor	
This amendment to final report contains three pages including this one.		
Documentation Section(s) amended and explanation for the change		
Page 1: Title page amended to read 'Amended Final Report 1' and issue date July 2009. Page 11: Date received of the test substance corrected from 4 November 2007 to 4 December 2007 The headers have changed to read 'Amended Final Report 1'		

Amended Final Report 1

Study Title	PFH Ammonium Salt: Fish, Early Life Stage Toxicity Test to *Oncorhynchus mykiss* (Rainbow Trout)
Study Guideline	OPPTS Biological Effects Test Guidelines 850.1400 Fish Early-Life Stage Toxicity Test
Authors	
Sponsor	Planning Dept. Chemical Division Daikin Industries, LTD. Umeda Center Bldg., 2-4-12, Nakazaki-Nishi Kita-Ku, Osaka, 530-8323 Japan
Study Monitor	
Test Facility	Covance Laboratories Ltd Otley Road, Harrogate North Yorkshire HG3 1PY UNITED KINGDOM
Study Number	2901/001
Covance Report Number	2901/001-D2149
Report issued	July 2009
Page Number	1 of 64

All procedures to be carried out on live animals as part of this study will be subject to the provisions of United Kingdom National Law, in particular the Animals (Scientific Procedures) Act 1986, to ensure that minimum suffering and distress is caused to animals on study.

MATERIALS AND METHODS

Protocol Adherence
The study was conducted in accordance with the agreed definitive protocol and three protocol amendments. Minor deviations that do not affect the integrity of the study are presented in Appendix 4.

Test Substance

Test substance name	PFH Ammonium Salt
Sponsor batch number	Lot 7001
Description	Clear liquid
Purity	50% solution
Storage	Refrigerated (1-10°C) in the dark
Expiry date	3 June 2009
Date received	4 December 2007

Test substance details were supplied by the Sponsor.

Test Organism
The Rainbow trout (*Oncorhynchus mykiss*) eggs and milt used in this study were supplied by a recognised external supplier. Supplier details are maintained in the study raw data.

At the start of the test, approximately 40 fertilised eggs were added randomly to each test vessel, divided equally between two incubation chambers per vessel. The fertilised eggs were less than 24 hours old on addition to the test system.

Egg addition was achieved by careful addition of the required number of eggs directly to the incubation chambers as soon as possible after receipt to avoid handling stress. The test started after the final batch of eggs had been added to the last incubation chamber.

Preparation of Test Media
The dilution water used for conducting the definitive test was dechlorinated mains water that had been passed through particulate and activated charcoal filters and treated with ozone (O_3) to remove residual algal cells and fungal spores for improved water quality. The typical constituents of dilution water are presented in Appendix 1.

The definitive test was conducted at nominal concentrations of 0.095, 0.304, 0.972, 3.11 and 9.96 mg/L PFH active moiety (equivalent to 0.2, 0.641, 2.05, 6.56 and 21.0 mg/L in terms of PFH ammonium salt with a purity of 50% and conversion factor of 1.054).

Concentrated aqueous stocks were prepared by direct addition of *ca* 48, 154, 492, 1574 and 5040 mg and made to a final volume of 15 L with treated mains water at each treatment level.