Updates to OECD Guidance Document 23: Aqueous-Phase Aquatic Toxicity Testing of Difficult Test Chemicals

Gilly Stoddart,[i] Christopher Faßbender,[i] Marlies Halder,[ii] Edward Salinas,[iii] and Wesley Hunter[iv]

[i]PETA International Science Consortium Ltd., Society Building, 8 All Saints Street, London N1 9RL, United Kingdom
[ii]European Commission Joint Research Centre, Via Enrico Fermi 2749, I - 21027 Ispra VA, Italy
[iii]BASF SE, Experimental Toxicology and Ecology, 67056 Ludwigshafen, Germany
[iv]US Food and Drug Administration, Center for Veterinary Medicine, 7500 Standish Place, Rockville, MD 20855, USA

Abstract

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The Organisation for Economic Co-operation and Development (OECD) Guidance Document on Aqueous-Phase Aquatic Toxicity Testing of Difficult Test Chemicals (GD 23) provides critical guidance that supplements OECD Test Guidelines (TGs) for studies conducted for regulatory purposes. OECD GDs undergo periodic review and updates to incorporate scientific progress, changing regulatory needs, and animal-welfare considerations. GD 23 was first published in 2000 and was recently updated to provide state-of-the-art approaches for aquatic toxicity tests involving difficult-to-test chemicals. This poster provides an overview of the updated GD 23, including expansion of the guidance on testing of poorly water-soluble test chemicals and substances of unknown or variable composition, complex reaction products, and biological materials (UVCBs). As part of the updates, particular attention was paid to updating exposure methods that do not employ a solvent in order to eliminate solvent effects and reduce the number of animals used in aquatic toxicity tests (i.e. via eliminating the need for a solvent control). The updated GD 23 is a useful tool to aid regulators, industry, and contract research organisations in conducting (or reviewing) valid and reliable aquatic toxicity studies while minimising both the number of animals used and the need to repeat studies.

The views, conclusions, and recommendations expressed in this poster are those of the authors and do not necessarily represent the policies or positions of the PETA International Science Consortium Ltd., the International Council on Animal Protection in OECD Programmes (ICAPO), the European Commission, the US Food and Drug Administration (FDA), or the OECD.

Introduction

- GD 23 provides guidance on maintaining consistent exposure to the dissolved test chemical throughout the test while minimising conditions that may lead to experimental artefacts (e.g. physical effects).
- The aim of this project was to update GD 23 with new techniques. Particular attention was paid to updating methods available for testing poorly water-soluble test chemicals while avoiding the use of solvents, thereby eliminating the need for a solvent control group and thus reducing the number of animals used.
- In the EU, nearly 180,000 fish were used for toxicological and other safety evaluations in 2011.[1] In the interest of animal welfare and efficient use of resources, it is important to avoid the unnecessary use of animals whenever possible.
- Revisions to GD 23 were initiated in 2014 by the European Commission and ICAPO. The US FDA Center for Veterinary Medicine joined the effort in 2016 to ensure GD 23 was fully updated.
- The updated GD 23 was approved by the OECD WNT[2] in April 2018 with the title Guidance Document on Aqueous-Phase Aquatic Toxicity Testing of Difficult Test Chemicals.[3]
- Content of GD 23:

Preparing stock and test solution, and selecting exposure systems, for chemicals that are:	
• Poorly/sparingly water soluble	• Coloured
• Volatile	• Hydrophobic
• Easily degraded	• Ionisable
• Highly adsorptive	• Multi-component substances
• Complex forming	• Alloys

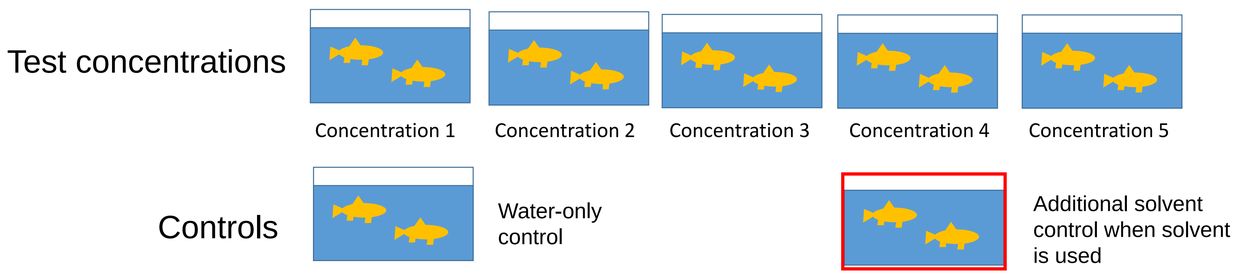
Additional topics:	
• Preliminary assessment of stability	• Testing at low chemical concentrations
• Exposure system considerations	• Sampling schedules
• Testing at water saturation	• Calculation and expression of test results

- This poster describes some key updates to GD 23.

Poorly/Sparingly Water-Soluble Test Chemicals

- Poorly/sparingly water-soluble test chemicals present unique challenges.
- Solvents are sometimes used to facilitate dissolution but can affect study results.
- Solvent use increases the number of animals used by up to 25 per cent per test, depending on the TG, because of the inclusion of both a solvent and negative control.

Example of aquatic toxicity test design:

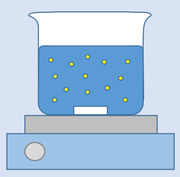
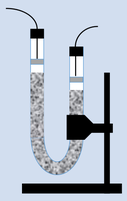
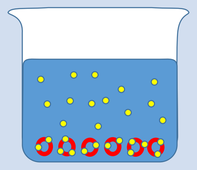
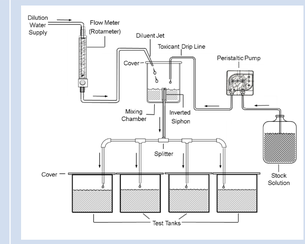


Number of Fish Used in Selected OECD TGs

OECD TG	# fish per control	# test concentrations	# fish per test concentration replicate	# of replicates	Total # fish per test if solvent used	# fish saved if no solvent used (%)
TG 203: Fish, Acute Toxicity	7	5	7	1	49	7 (14)
TG 215: Fish, Juvenile Growth	16	5	16	1	112	16 (14)
TG 212: Fish, Short-Term Toxicity Test on Embryo and Sac-Fry Stages	30	5	10	3	210	30 (14)
TG 210: Fish, Early-Life Stage Toxicity	20	5	20	4	560	80 (14)
TG 305: Bioaccumulation in Fish: Aqueous and Dietary Exposure (e.g. aquatic exposure)	36	2	4	9 time points	144	36 (25)

- Substantial revisions were introduced to, *inter alia*, reduce occasions when solvents are used.

Techniques Described in GD 23 That May Not Require Solvent in the Exposure System

Direct addition	Generator systems	Passive dosing	Flow-through exposure systems
 Stirring, shaking, blending, homogenisation, sonication, pH adjustment	 Liquid/liquid saturator  Saturator column	 Loaded polymer used as partitioning donor to maintain freely dissolved concentrations	 Maintain stable water quality and test concentrations (Diagram courtesy of EAG)
• Guidance on methods for dissolution of the test chemical is provided. • Undissolved test chemical must be removed prior to test initiation.	• Saturator column preparation techniques, temporal limitations of use, and analytics are described. • Test medium must not be recirculated through columns after being used in exposure.	• Guidance on loading principles and techniques is provided. • References to publications describing different geometries of dosing polymers are provided. • Limitations of the method are described.	• Recommendations are provided for the number of tank volume turnovers per 24 hours, flow-rate calibration, stock solution preparation, and the equilibrium pre-test period.

Updated Guidance for Multi-Component Substances

- Mixtures/preparations and UVCBs
- Individual components may have different physicochemical properties, such as:
  - Water solubility
  - Octanol-water partition constants
  - Melting points
- Different physicochemical properties create unique challenges when conducting aquatic toxicity tests and interpreting the results
- GD 23 provides more guidance on determining the physicochemical properties of UVCBs and conducting aquatic toxicity tests using water-accommodated fractions (WAFs).
- Considerations and challenges discussed include:
  - Identifying key or major components in the UVCB itself and in the WAF
  - Differing physicochemical properties for the UVCB components
  - Toxicity test methods of fully and partially soluble UVCBs, including generation of a WAF or water-soluble fraction
  - Reporting of toxicity test results

Conclusions

- The updated GD 23 represents a collaborative effort of numerous experts from industry, OECD member countries, non-government organisations, and academia.
- The updated GD 23 will help government agencies, industry, and contract research organisations conduct valid and reliable aquatic toxicity studies on difficult-to-test chemicals while minimising the number of animals used and the need to repeat studies.

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Note

[1]European Commission. 2013. Seventh Report on the Statistics on the Number of Animals Used for Experimental and Other Scientific Purposes in the Member States of the European Union. Available at: https://eur-lex.europa.eu/legal-content/EN/TXT/PDF/?uri=CELEX:52013DC0859&from=EN. Accessed 3 August 2018.
[2]WNT = Working Group of the National Coordinators for the Test Guidelines Programme
[3]https://www.oecd-ilibrary.org/environment/oecd-series-on-testing-and-assessment_20777876?page=3