OECD/OCDE

The Threshold Approach for Acute Fish Toxicity Testing

GENERAL CONSIDERATIONS

1. In the interest of sound science and animal welfare, it is important to avoid the unnecessary use of animals whenever possible. All information on a substance relevant to its potential acute aquatic toxicity should be evaluated prior to considering testing in fish. Sufficent information may already exist to classify a test substance as to its acute aquatic toxicity, e.g. it is well known that fish is not always the most sensitive species (1, 2). Therefore, information from validated (Q)SARs (3), *Daphnia* (TG 202; 4) and algae (TG 201; 5) should be utilized in a weight-of-evidence analysis. Significant information might be gained from experimental data from standard, non-standard studies or from non-standard species, data generated with QSARs, read-across, etc. If there is compelling evidence, using these methods, to suggest that the fish is likely to be at least a factor of about 10 less sensitive than invertebrates or algae, further acute fish toxicity testing may not be needed. If these data indicate that the fish is not the most sensitive species, an acute fish test might not be needed.

2. It is recommended that a weight-of-the-evidence analysis be used to evaluate existing information pertaining to acute fish toxicity of substances and to determine whether additional studies, other than an acute fish study, should be performed to help characterise such potential. Where an acute fish toxicity study is needed and for substances which have no testing history, it is recommended that the threshold approach be utilised to develop the relevant experimental data. It has the potential to significantly reduce the number of fish needed, especially if the test substance is likely to be more toxic to daphnia and algae than to fish.

3. The threshold approach represents both best practice and an ethical benchmark for *in vivo* testing for acute fish toxicity. The threshold approach described thereafter is based on the threshold/step-down approach initally described for pharmaceuticals (2) and further developed for chemical substances at the Euroepan Commission's Joint Research Centre (7, 8) taking into consideration existing OECD guidelines. It was subsequently affirmed and incorporated into the "Guidance on information requirements and chemcial safety assessment" for REACH (9). Several publications confirm the reduction potential of the threshold approach (10, 11).

DESCRIPTION OF THE THRESHOLD APPROACH

4. When acute fish toxicity data need to be generated the threshold approach should be applied. It includes the performance of tests according to OECD guidelines:

   a. TG 201 – Freshwater Alga and Cyanobacteria, Growth Inhibition Test
   b. TG 202 – Daphnia sp. Acute Immobilisation Test
   c. TG 203 – Fish, Acute Toxicity Test (Limit test)
   d. TG 203 – Fish, Acute Toxicity Test.

   The following step-wise procedure should be utilized (Figure):

5. Derivation of the threshold concentration (Step 1): The lowest EC50 value of invertebrate (e.g. daphnia) or algae is set as threshold concentration (TC). If these data are not available they need to be determined according to TG201 and TG202.

6. Assessment of acute fish toxicity (limit test) at the TC (Step 2): An acute fish test is performed according to the limit test described in TG 203 (paragraph 20) at the TC. If the TC is >100 mg/l, the test substance concentration should be 100mg/l in the limit test. The absence of mortality indicates that the fish is not the most sensitive species and that, with at least 90% of confidence, the LC50 is greater then the threshold concentration. If mortality occurs, a full study according to TG 203 should be conducted.

LITERATURE

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(3) OECD QSAR toolbox

(4) OECD (2004). Guideline 2002. Daphnia sp. Acute Immobilisation Test

(5) OECD (2006). Guideline 201. Freshwater Alga and Cyanobacteria, Growth Inhibition Test

(6) OECD (1992). Guideline 203. Fish, Acute Toxicity Test

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(9) ECHA (2008). Guidance on information requirements and chemical safety assessment. Chapter R.7B – Endpoint specific guidance (p. 41 ff, Chapter 7.8)

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Figure

The Threshold Approach for Acute Fish Toxicity Testing

	Activity	**Finding**	**Conclusion**
1	Derivation of threshold concentration (TC) - use existing EC50 values from invertebrates (e.g. daphnids) and algae OR - generate EC50 values according to OECD TG 201 and OECD TG 202	EC50 daphnids EC50 algae	lowest EC50 = TC
2	Assessment of acute fish toxicity (limit test) at the TC - Limit test according to OECD TG 203 at TC	no mortality mortality	LC50 >TC full OECD TG 203