

# Alternative to animal experiments: Fish cell test internationally certified

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For the first time ever, a toxicity test with cultured gill cell lines from fish has been ISO-certified. The test is used to determine the acute toxicity of water samples and chemicals to fish. This is an important milestone because there is a lack of recognised alternatives to experiments with live fish.

In 2017, more than 7,500 ecotoxicological tests were carried out on fish in Switzerland alone with the aim of protecting humans, animals and the environment. For many years, Eawag has been researching alternatives in order to reduce or even replace fish experiments. One of these alternatives involves experiments with a gill cell line of rainbow trout (RTgill W1 cell line), which can be used to reliably determine the acute toxicity of water samples and many chemicals to fish.

Under the leadership of Prof Kristin Schirmer, the Department of Environmental Toxicology has continuously refined the method over the last few years. In an international round-robin study, six laboratories from industry and academia took part and determined the robustness, transferability and comparability of the method with the RTgill-W1 cell line on the basis of six selected test chemicals. The results show that all laboratories were able to provide reproducibly comparable results using this procedure. This methodology has recently also been extended to comply with ISO standards, which serve as standard references for researchers and manufacturers in tests.

### Kristin, what does this ISO certification mean?

Our test can now be used by anyone who wants to test the fish acute toxicity of water samples or certain chemicals – from sewage plant operators to environmental agencies or chemical companies.

T +41 58 765 55 11 F +41 58 765 50 28 info@eawag.ch www.eawag.ch



The test saves time, is cost-effective and, of course, does not require any laboratory animals whatsoever.

### What hurdles have to be overcome in order to have a test ISO-certified?

The certification process takes a very, very long time. The first experiments with the fish cell line we used date back to my doctoral thesis twenty years ago. Back then we published a scientific article which showed for the first time that the fish acute toxicity of waste water samples can be detected with gill cells of rainbow trout. But it wasn't until a few years later, in 2007, that we received funding from the European Chemical Industry Council (CEFIC) to further advance the method and make it available for chemicals testing. In 2013 we published a study that showed that our method achieved the same toxicity levels for over 30 chemicals as experiments with live animals. A Norwegian colleague was so enthusiastic about the results that he proposed certification to the ISO Commission. But we then had to prove that our method also worked in other laboratories, which is why we set up a round-robin study organised by my colleague Melanie Fischer. In February of this year, ISO agreed to certify our method following the successful round-robin study and several ballots.

## Are manufacturers of chemicals now allowed to do without animal testing and rely solely on this test?

I'm afraid not. The regulatory guidelines of the OECD continue to apply to the manufacture of chemicals. These require experiments with live fish which are exposed to the chemical in order to determine how many fish survive after four days at what concentrations. However, the test with RTgill-W1 cells can now be used, for example, in product development in order to decide in advance whether an animal experiment is worth carrying out. Naturally, we very much hope that our test will also be recognised by the authorities in the future as an alternative to animal testing. We have therefore also submitted the test to the OECD and are now in the process of completing all the necessary forms. Thanks to the ISO certification, the chances are good that our alternative will soon be established.

### **Original articles**

Repeatability and reproducibility of the RTgill-W1 cell line assay for predicting fish acute toxicity academic.oup.com/toxsci/advance-article/doi/10.1093/toxsci/kfz057/5368498

ISO 21115:2019 Water quality - Determination of acute toxicity of water samples and chemicals to a fish gill cell line (RTgill-W1) www.iso.org/standard/69933.html

Predicting Fish Acute Toxicity Using a Fish Gill Cell Line-Based Toxicity Assay https://pubs.acs.org/doi/abs/10.1021/es303505z

### Links

The Eawag spinoff Aquatox solutions offers the ISO-certified method. Gill cells instead of fish for toxicity tests

### Contact





Kristin Schirmer Group leader and deputy head of department Tel. +41 58 765 5266 kristin.schirmer@eawag.ch

https://www.eawag.ch/en/info/portal/news/news-archive/archive-detail/alternative-to-animal-experiments-fish-cell-test-internationally-certified

