

## **A Rapid and Easy Method to Introduce *Daphnia magna* into Test Vessels**

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The method normally used to introduce *D. magna* into test vessels is to pipette the organisms with a drop of water using a smooth glass tube. Unfortunately this method has several disadvantages.

- a) Counting of *Daphnia* in a small drop of water is rather strenuous and leads relatively often to mistakes.
- b) The method is very time consuming.
- c) Due to the drop of water it is practically impossible to retain the test concentration.

To overcome these problems we have developed a new method. We use Fluorocarbon Monofilament fabrics 2x1.3 cm in size, with a mesh-opening of 70  $\mu\text{m}$ , and a thread diameter of 80  $\mu\text{m}$ . This piece of fabric is laid on washed and dried chromatography paper. A drop of water normally containing more than the required number of animals is pipetted onto the fabric. Because of the underlaid chromatography paper the water disappears immediately. The surplus of *Daphnia* is removed by use of a stainless steel spatula and the fabric is dipped into the test solution using stainless steel forceps. The *Daphnia* swim away and the fabric without test animals is removed and discarded.

This method has proved to have the following advantages:

- a) Counting of *Daphnia* on the fabric is easy and almost no mistakes occur.
- b) We found this method three times quicker than the "pipette method".
- c) The test concentration is not changed.

The use of fluorocarbon and stainless steel guarantees that no leachable substances can enter the test solution and that there is no loss of test material by adsorption. To ensure that there is no damage to *Daphnia* with this method we carried out the following experiment.

According to the German standard method (1) we made a series of 40 controls with the "fabric method", resulting in a total of 200 test animals. We recorded the number of dead animals after 24, 48, 72 and 96 h and compared these results with the data derived from another series of 40 controls using the "pipette method". The results are shown in Table 1.

TABLE 1

	Percent of dead Daphnia after			
	24 h	48 h	72 h	96 h
Pipette method	0.5	0.5	2.5	30
Fabric method	0.5	1.0	1.5	23.5

Obviously these results did not show a worse survival rate with the new method. Furthermore the behaviour of the test animals showed no abnormality and no damage could be detected by microscopical control. The EC 50 of numerous tests with  $K_2Cr_2O_7$  did not show any significant differences between the two methods. Because of these results we think that our method is superior to the method used until now.

#### R E F E R E N C E S

- 1) DEUTSCHES INSTITUT FÜR NORMUNG: Bestimmung der biologischen Wirkung von Wasserinhaltsstoffen auf Kleinkrebse. DIN 38412 Part 11 (Draft) (1980).