

## A TECHNIQUE FOR TESTING CHEMICALS AS MOLLUSCICIDES

F. S. Barbosa, J. M. Barbosa e T. Figueiredo

In the ordinary methods of testing molluscicides in the laboratory each group of snails, usually 10, is placed in a glass or plastic container into which a certain concentration of the chemical has been introduced. The number of containers corresponds to the number of concentrations required in each test.

Replications of the tests are usually necessary in order to increase the number of snails in each experiment. In doing that, additional containers are required.

A common and important error in mollusciciding laboratory experiments results from the tendency of the snails to crawl out of the toxic solutions into which they have been immersed. As far as the experience of the authors of the current paper is concerned there is no perfect device completely able to prevent the escape of the snails from the molluscicide.

In the technique described in the present paper the error mentioned above is eliminated and the number of containers can be greatly reduced.

### *Description of the technique*

A large glass container is used for each chemical concentration. The volume of

the container will depend on the number of snails to be tested in each chemical concentration. Keeping the proportion of 100 ml of the molluscicide solution per snails, a container holding five liters will receive 50 snails.

After filling the containers, (one for each concentration of the molluscicide to be tested), the snails can be introduced. Groups of 10 snails are placed inside bags of nylon net (mesh 1.5 mm diameter) and then plunged into the solutions. For larger snails the groups within each bag can be smaller. The number of bags to be placed in each container will depend on the volume of the liquid. In the experiments made in this laboratory five bags were used (totaling 50 snails) for each concentration of the chemical. Each bag is tied with a thread which bears an identification tag on its free end (Figs. 1 and 2).

The kind of net utilized is light enough to permit the bags to float in the solution. The snails are seen inside the bag coming up to the surface to breathe (fig. 3).

After the exposure period, the bags are taken out of the molluscicide, opened, and the snails are washed and transferred to fresh water. Bags are used only once.

School of Medicine, University of Pernambuco and Centro de Pesquisas Aggeu Magalhães, Recife, Brazil.

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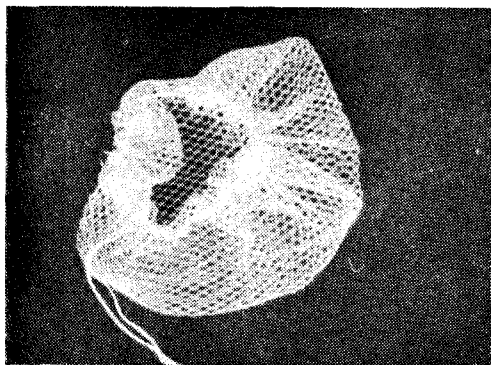


Figure 1 Nylon sacs used in the experiments.

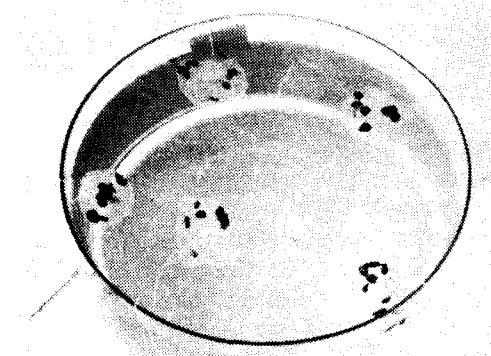


Figure 2 Nylon sacs immersed in the molluscicide solution.



Figure 3 Closer picture showing the snails inside floating sacs.

Snail controls are treated in the same way, i.e. placed inside similar bags and immersed into water in the tanks.

The survival of the snail controls has been determined in the laboratory. The snails inside the bags survive the same length of time as those kept outside the bag and immersed into the same water.

#### *Utilization and possibilities of the technique*

The technique described in the present paper, besides the advantage of excluding the most common error in the ordinary mollusciciding laboratory methods, is simple, rapid and economical. The number of containers is smaller and the employment of manpower reduced.

The technique is specially indicated for comparative purposes when confronting different species, strains, or stages of the snails with the same molluscicide. The technique is also very useful for the determination of time concentration curves. For that purpose the snails can be immersed into one solution prepared at each concentration of the chemical. Under such conditions the bags can be taken from the molluscicide upon the termination of the exposure time for each group of snails.

#### *Experimental evaluation of the technique*

The consistency of the technique and its reliability were tested by using the well-known molluscicide Bayluscide. Variation was studied in concentrations of 0.040 and 0.064 ppm which usually kill 40 to 60 per cent of the snails. The experiment was performed to study the variability of the technique in comparison with two other techniques.

Three groups of 10 snails, *Biomphalaria glabrata*, were tested. Snails of group 1 were used according to the classical technique recommended by WHO (2) and without any device to avoid the snails from escaping from the solutions. The same technique was employed for group 2 with just one difference. This group was closely watched during 24 hours to prevent the snails from crawling out of solutions. Every snail trying to get out of the molluscicide was gently pushed back into the solution. This method, although imprat-

TABLE 1. MOLLUSCIDING ACTIVITY OF BAYLUSCIDE AGAINST THREE EXPERIMENTAL GROUPS OF *B. GLABRATA*

Experimental groups	Chemical	concentration	Coefficient of variation at	
	LC <sub>50</sub>	Confidence interval 95%	0.064 ppm	0.040 ppm
1	0.074	0.051-0.197	73.4	210.0
2	0.052	0.038-0.070	25.4	106.6
3	0.040	0.031-0.052	24.7	66.8

ical for ordinary use, was employed to assure closer contact of the snails with molluscicide. Finally the third group of snails was tested in nylon sacs according to the technique described in this paper.

Table 1 shows the results of the tests with three experimental groups of snails for which the LC<sub>50</sub> was determined according to Litchfield & Wilcoxon (1) method.

Comparison of the three above treatments showed that the differences observed between group one to either group two or group three, were statistically significant at 5% probability.

Although the observed difference was

not significant when groups 2 and 3 were compared, in the latter group the LC<sub>50</sub> value was lower, the confidence interval narrower and the coefficient of variation lower, particularly at the concentration of 0.040 ppm.

This paper clearly demonstrates that when an efficient device is used to avoid the escaping of the snails from the molluscicide, results are much more consistent and variation is much less pronounced. In this case an important cause of additional variation other than that intrinsic variation observed in biological measurements is prevented.

#### REFERENCES

1. Litchfield, J. T. & Wilcoxon, F. (1949) A simplified method of evaluating dose effect experiments, *J. Pharmacol. exp. Ther.*, 95: 99
2. World Health Organization (1961) *Wld Hlth Org techn. Rep. Ser.*, 214

(Continuação da pág. 94)

Art. 6.º — O parecer da Comissão Julgadora deverá ser emitido até o início do Congresso da Sociedade Brasileira de Medicina Tropical nos anos pares, quando o prêmio será solenemente entregue ao vencedor. A primeira entrega de prêmio será em 1970.

Art. 7.º — Os casos omissos neste re-

gulamento serão resolvidos pelo Diretor do Instituto Brasileiro de Tropicologia Médica em conformidade com o Presidente da Sociedade Brasileira de Medicina Tropical e se fôr o caso com a Firma Patrocinadora e ao candidato não caberá nenhum recurso ao parecer da Comissão Julgadora ou de seu Presidente.

