

Field Trials to Evaluate the Effectiveness of the Molluscicide *N*-Tritylmorpholine in Irrigation Systems

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In field trials with the molluscicide N-tritylmorpholine (Frescon, WL 8008), a prolonged low-dosage technique has been developed for use in irrigation systems. In Tanzania a dose of 0.025 ppm applied for 30 days to the headworks of a 5000-acre (2025-ha) irrigation system gave effective control of Biomphalaria pfeifferi, the snail host of Schistosoma mansoni, for a period of 3-4 months. This was comparable to the effectiveness of other molluscicides and other dosage regimes but the technique had the advantages of simplicity, lower cost and lack of toxicity towards fish or other aquatic fauna. Similar trials have been carried out in Egypt and Southern Rhodesia. Theoretical and practical considerations involved in the choice of dosage regimes are discussed and a general formula is suggested for choosing dosage regimes in irrigation systems.

INTRODUCTION

Bilharziasis is a major public health problem of increasing importance in most irrigated areas in Africa. The intermediate-host snails of the carriers of this disease thrive in the relatively warm, slow-moving water of most irrigation systems in the tropics. Native and immigrant communities tend to congregate near the irrigation systems to meet their need for domestic water supplies. With few exceptions the inhabitants of such areas cannot, or will not, avoid contact with irrigation water, even if provided with an alternative supply. Man has created these ideal conditions for transmission of bilharziasis; he is truly the vector of this disease and in this capacity he has become extraordinarily efficient.

There is considerable actual and projected expansion of irrigated areas in Africa; the Aswan High Dam in Egypt, the Awash Valley scheme in Ethiopia, the Tana River scheme in Kenya, the Hippo Valley/Triangle area in Southern Rhodesia and the Limpopo River scheme in Mozambique are just a few examples. Although such measures as health education, environmental sanitation and improved design of irrigation waterways may prove invaluable in the

long run, they can do little to alleviate the immediate problem of bilharziasis in developing countries.

Several workers (e.g., McMullen & Harry, 1958; Macdonald, 1965) have pointed out the inadequacies of attacking the problem in only one of the parasite's hosts. In man, the use of drugs in mass-treatment campaigns is unlikely to achieve a worthwhile reduction in transmission, because this measure alone can offer little or no hope of protection against the risk of reinfection. In the snail, control through the use of molluscicides has most often been advocated in recent years and this measure alone has been successful in reducing the incidence of bilharziasis (Clarke, Shiff & Blair, 1961; McMullen et al., 1962). No dramatic reduction in incidence can be expected by this measure alone, because it is essentially a prophylactic one, but the number of infections can be expected to decline gradually over a period of years.

An integrated approach, involving both prophylactic molluscicides and therapeutic drugs, has obvious merit but has been criticized on the grounds of lack of effectiveness and excessive cost. These criticisms may be overcome by advances in technology, as has already happened in the field of malariaology (Macdonald, 1957). The development of DDT for mosquito control, in conjunction with the effective use of prophylactic and therapeutic

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agents, led to the World Health Organization's worldwide malaria eradication programme, an ambitious concept that has already paid handsome dividends. The problems of bilharziasis control are more complex and as yet little understood compared with those of malaria control, but the development of a new and better technology in control work may well provide the stimulus for a more vigorous attack.

In recent years there have been notable advances in the development of molluscicides, and the discovery of the new therapeutic compound Ambilhar (1-(5-nitro-2-thiazolyl)-2-imidazolidinone) may represent a parallel development. Outstanding among the molluscicides are the synthetic materials Bayluscide (the ethanolamine salt of 5,2'-dichloro-4'-nitrosalicylanilide), which has been widely tested, and a new compound, Frescon (*N*-tritylmorpholine). Shiff (1966) has already reported promising results with the latter compound (under the name WL 8008) in night-storage dams in Southern Rhodesia, and trials in irrigation systems are reported here.

The simplest method of treating an irrigation system with molluscicide is to apply a low concentration over a long period at the intake, relying on water carriage for distribution of the chemical and thus avoiding the time and trouble of treating canals individually. Sharaf El Din & El Nagar (1955) have reported on the use of copper sulfate in this way as a chemical barrier in the Gezira area of the Sudan to prevent repopulation by snails after "blanket" treatment of canal systems with 30 ppm of copper sulfate. Teesdale, Hadman & Nguriathi (1961), working in the Mwea/Tebera rice-irrigation scheme in Kenya, showed that a concentration of 0.25 ppm of copper as copper sulfate was ineffective against *Biomphalaria pfeifferi* after a distance of 3000 yd (2740 m) downstream of the application point. Earlier experiments by Chancellor, Coombs & Foster (1958) against aquatic weeds had shown that copper is selectively absorbed by mud and aquatic vegetation and this factor may account for its lack of downstream penetration.

N-Tritylmorpholine is highly toxic to various species of aquatic snail, but has a low mammalian toxicity and negligible phytotoxicity.¹ It is available as an emulsifiable concentrate, which is easy to handle and suitable for use with simple, drip-feed applicators. It is stable except in water with a low pH and is not readily absorbed by mud and aquatic vegetation. It was therefore considered suitable for

application to irrigation systems by a prolonged low-concentration technique. Although highly toxic to aquatic snails, *N*-tritylmorpholine is less toxic to their eggs. However, the eggs will hatch out and the hatchlings be killed during a long application period and therefore lack of ovicidal action may not be very important. Furthermore, preliminary estimations of the quantities of molluscicide required suggested that even a very prolonged application of a low concentration would not be uneconomic.

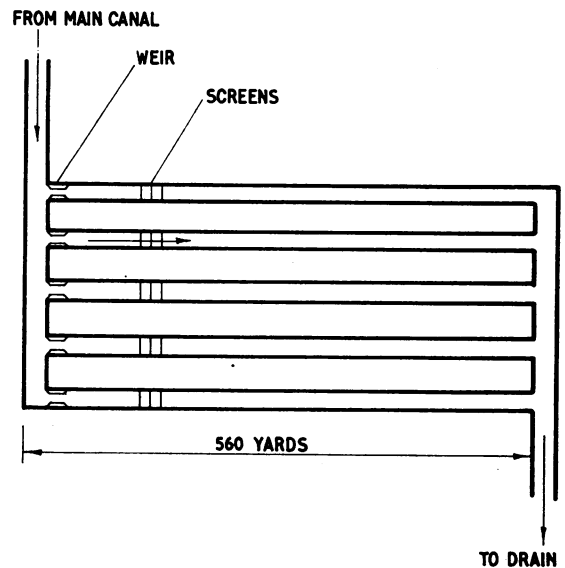
PILOT TRIALS

Materials and methods

Pilot trials were carried out at Arusha Chini, Tanzania, in a series of experimental canals that were specially constructed for field screening of molluscicides. Each was 560 yd (512 m) long, 3 ft-6 ft (0.9 m-1.8 m) wide and about 12 in-18 in (30.5 cm-45.7 cm) deep. The average gradient was about 1 in 5000 and the velocity of the water 220 yd/h (200 m/h). The layout of the experimental block is shown in Fig. 1.

The canals were designed and built to support dense populations of snails, which included the following species: *Biomphalaria pfeifferi*, *Bulinus tropicus*, *Lymnaea natalensis*, *Melanoides tuberculata* and *Cleopatra ferruginea*. The water was very clear and slightly alkaline, with an average pH of 8.0.

FIG. 1
LAYOUT OF EXPERIMENTAL BLOCK OF CANALS



¹ See the papers by Boyce et al., Brown et al. and Chapman on pages 1, 13, 73 and 43 of this issue.

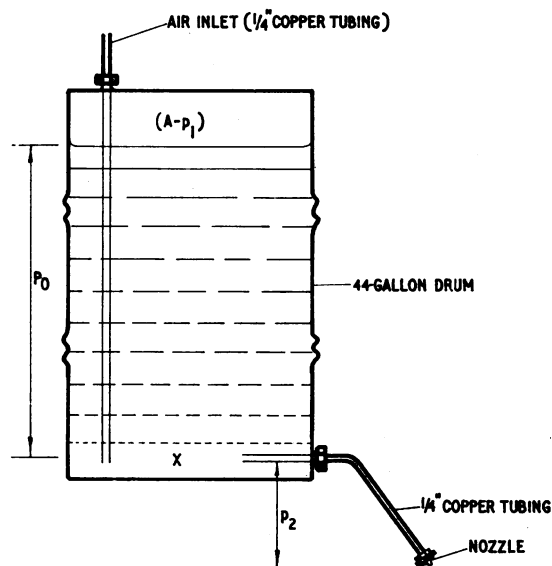
Abundant aquatic weed provided shelter and egg-laying sites for the snails and consisted mainly of the following species: *Potamogeton schweinfurthii*, *Chara kraussiana*, *Elodea canadensis*, *Myriophyllum* sp. and *Lemna* sp. Small measuring weirs (Parshall flumes) were built at the upstream end of each canal to provide an accurate and quick means of measuring discharges, which varied from about 0.2 ft³/s to 0.3 ft³/s (5.6 l/s-8.4 l/s). Three mesh screens, approximately 16, 400 and 2000 mesh per in² (2.5, 62 and 310 per cm²) were placed 25 yd (23 m) downstream of each weir to prevent reinvasion by snails after treatment.

The density of snail populations was estimated by a mud-sampling technique that has been described in full elsewhere (Crossland, 1962). Briefly, a plug of mud 10.5 cm in diameter is removed from the bottom of the canal with the aid of a tube sampler; the mud is washed through a sieve and the snails are sorted and counted. The effectiveness of molluscicide treatments may then be estimated by comparing pretreatment and post-treatment counts, after making due allowance for mortality in a similar untreated canal.

In addition to the estimates by the mud-sampling technique, it was desirable to estimate snail mortalities at more frequent intervals. Snails were therefore confined to cages and examined at daily intervals. The cages were made with wooden frames, 1 ft (30 cm) square and 9 in (23 cm) deep, covered with good-quality nylon bolting cloth, mesh size 48 GG. Four cages, each containing 25 *Biom. pfeifferi* and 25 *Bul. tropicus*, were placed in the treated canals at distances of 25 yd, 125 yd, 325 yd and 425 yd (23 m, 114 m, 297 m and 389 m) from the point of application of the molluscicide, and a similar series of controls was placed in an untreated canal. The cages were put in position 7 days before the treatments started and any snails that died before treatment began were replaced with freshly collected snails.

N-Tritylmorpholine was applied at a constant rate with the aid of the simple apparatus shown diagrammatically in Fig. 2. When the molluscicide starts to run out of the drum there is a drop in the level of the contained liquid; the air trapped in the space above the liquid expands and its pressure drops in inverse proportion to the amount of expansion. As the pressure of the air in the drum decreases, the pressure inside the liquid at a point X decreases by an equivalent amount; air is therefore sucked down the air inlet and begins to bubble through the liquid

FIG. 2
SIMPLE APPLICATOR FOR DISPENSING LOW DOSAGES OF *N*-TRITYLMORPHOLINE OVER LONG PERIODS

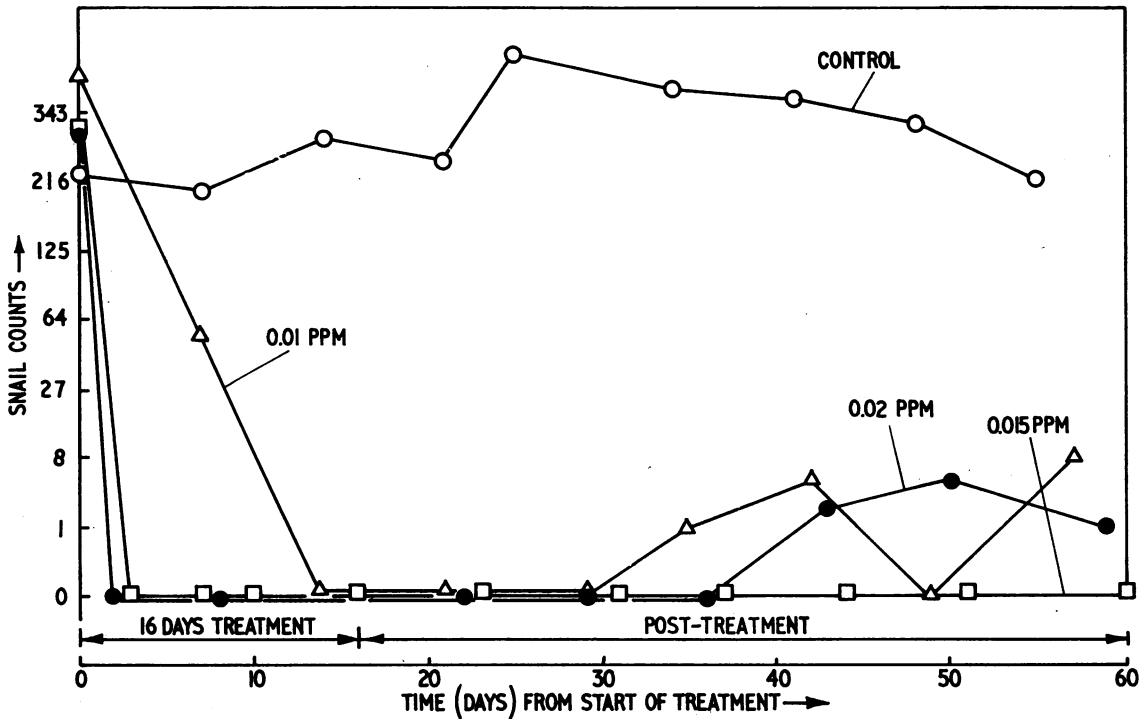


when $p_1 = p_0$. The pressure at X is then equal to $(A - p_1) + p_0$, i.e., atmospheric pressure (since $p_0 = p_1$). The flow rate is therefore dependent only on the height of the outlet tube (p_2) and the size of the nozzle. By choosing a suitable combination of values for p_2 and nozzle size, it is possible to achieve any desired emission rate.

In this series of experiments the supply of molluscicide was replenished daily. With a 44-UK gal (200-litre) drum and 24-hour intervals between replenishments, the required emission rate was 140 ml/min. This was conveniently arranged by using a nozzle with a diameter of 0.05 in (0.13 cm) in combination with a head (p_2) of 9 in (23 cm). The required amount of a 16.5% (w/v) emulsifiable concentrate of *N*-tritylmorpholine was measured out and mixed with water to form an emulsion before being poured into the drum through a large funnel fitted with a fine gauze filter.

In preliminary laboratory tests with a prolonged, low-dosage technique it was found that a dose of 0.02 ppm of *N*-tritylmorpholine gave a complete kill of *Biom. pfeifferi* after only 2 days, whereas 0.01 ppm gave only a partial kill even after 7 days. In the field tests, therefore, it was decided to investigate the effects of prolonged doses of 0.01 ppm, 0.015 ppm and 0.02 ppm. An application period of 16 days was chosen because field data had shown that at Arusha

FIG. 3
EFFECT OF PROLONGED LOW-DOSAGE TREATMENT WITH *N*-TRITYLMORPHOLINE
ON CANAL POPULATIONS OF *BIOM. PFEIFFERI*



Chini the mean time from egg-laying to hatching was $12\frac{1}{2}$ days, with a maximum of 15 days, and thus it was expected that all snail eggs would hatch and the hatchlings would be killed before the end of the treatments.

An attempt was made to estimate the numbers of snail eggs in the canals by collecting them by hand for a specified time and recording the results as numbers collected per man-hour search. It was expected that this method would yield only crude estimates of the relative numbers of eggs in a series of successive observations. However, in addition to such estimates of the numbers of egg-masses, some relevant information on their age was obtained by classifying them according to their stage of embryonic development. The collections were therefore taken to the laboratory, where they were examined with the aid of a low-power binocular microscope. Egg-masses were counted as "late" if the eggs contained recognizable snails capable of crawling around inside their shells. Other embryonic stages were counted as "early". Egg-masses were counted

as dead only if all of the embryos within a given mass were dead. No attempt was made to distinguish between the egg-masses of *Biom. pfeifferi* and those of *Bul. tropicus*.

Methods for the determination of *N*-tritylmorpholine in water samples have been described by Beynon & Thomas.¹ However, since these were not available when the present work was carried out, a slightly different procedure was used. One-litre samples of water were extracted twice with 10 ml of redistilled isohexane, the water layer was run off and the isohexane extract dried through anhydrous sodium sulfate. Further fractions of 10 ml and 5 ml of isohexane were used to rinse out the separating funnel. The isohexane was back-extracted with 1 ml of concentrated sulfuric acid and the acid extract was diluted with 5 ml of 50:50 aqueous sulfuric acid. The absorbance associated with the yellow colour produced in the acid layer was measured on a Unicam SP 600 spectrophotometer at 435 nm and

¹ See the paper on page 47 of this issue.

the amount of *N*-tritylmorpholine equivalent to the net absorbance was calculated from a previously prepared calibration curve.

Water samples were taken from the upstream and downstream ends of the canals treated with doses of 0.01 ppm and 0.02 ppm of molluscicide, at 07.00 h and 19.00 h each day. The upstream sampling position was 25 yd (23 m) from the applicator and the downstream position 540 yd (495 m) from the applicator.

Results

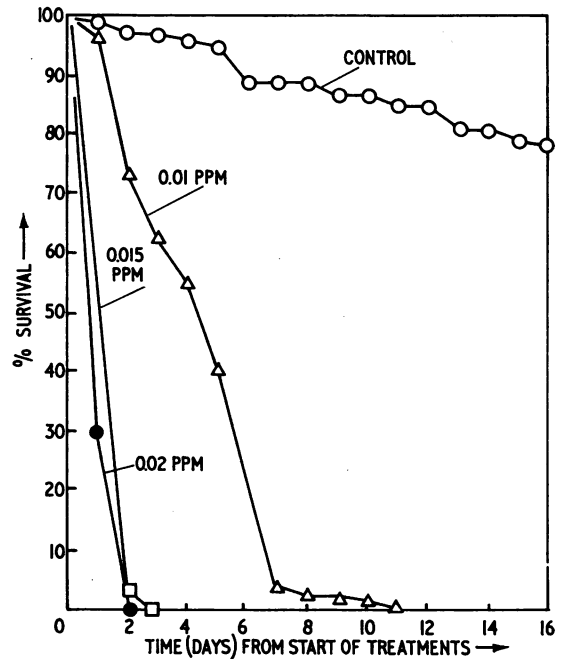
The effect of prolonged, low doses of *N*-tritylmorpholine on populations of *Biom. pfeifferi* is illustrated in Fig. 3, where each point represents the total number of snails collected in 168 mud samples, i.e., 3 samples per 10 linear yards (per 9.1 linear metres) of canal, 560 yd (512 m) long. On the ordinate the total counts of snails are plotted on a cube scale. This is preferred to an arithmetic scale because the transformation normalizes the data (Yeo, 1962) and removes that part of the sampling variation resulting from the aggregation of snails. Thus, the form of the distribution of the transformed data should be the same at all population densities.

In the untreated canal there was a trend towards an increase in density level until the 25th day, after which this trend was reversed. In the canal treated with a 0.01-ppm dose the numbers of *Biom. pfeifferi* decreased gradually to zero over a period of 14 days and thereafter no snails were found until the 35th day. In the canal treated with a 0.015-ppm dose the numbers of *Biom. pfeifferi* fell sharply to zero within the first 3 days and thereafter no snails were found until the 73rd day. In the canal treated with a 0.02-ppm dose there was a similar reduction in snail numbers, with snails reappearing in the samples on the 43rd day.

The mortalities among snails in 4 cages placed in each canal have been pooled and are presented in graphical form in Fig. 4, where each point represents the number of surviving snails out of a sample of 100. There is a striking similarity between these data and those of Fig. 3, showing that the mortality rate of the caged snails was similar to that for the natural population.

Similar data were collected for natural populations of *Bul. tropicus*, as well as for caged snails, and have been analysed in similar fashion. Fig. 5 shows the effect of the treatments on natural populations. Although none was completely effective, there was a very high mortality in all three treatments. In the

FIG. 4
EFFECT OF PROLONGED LOW-DOSAGE TREATMENT WITH *N*-TRITYLMORPHOLINE ON CAGED *BIOM. PFEIFFERI*



0.02-ppm treatment the population density was reduced to a very low level after 6 days, but in the 0.01-ppm and 0.015-ppm treatments there was a more gradual reduction in population density.

The data presented in Fig. 6 for caged snails are more revealing than those for natural populations because the observations were made at frequent intervals. In the 0.02-ppm treatment there were few deaths during the first 2 days, but in the next 5 days 80%-90% of the exposed snails died; the remaining snails died over a period of 4 days. This pattern of deaths results in a sigmoid curve, which closely parallels the curve given for *Biom. pfeifferi* in Fig. 4 for the 0.01-ppm treatment. The significance of these findings will be discussed later, after presentation of the results obtained in the large-scale trials.

The data for egg-masses (Table 1) are based on counts of a heterogeneous population of the eggs of *Biom. pfeifferi* and *Bul. tropicus*. This factor makes the results for the 0.01-ppm treatment difficult to interpret. Another complication is that the data for egg-masses should be related to those for the density of adult snails, given in Fig. 3 and 5. The decrease in the numbers of egg-masses until the 12th day should be related to the differential mortalities of the

FIG. 5.
EFFECT OF PROLONGED LOW-DOSAGE TREATMENT WITH *N*-TRITYLMORPHOLINE
ON CANAL POPULATIONS OF *BUL. TROPICUS*

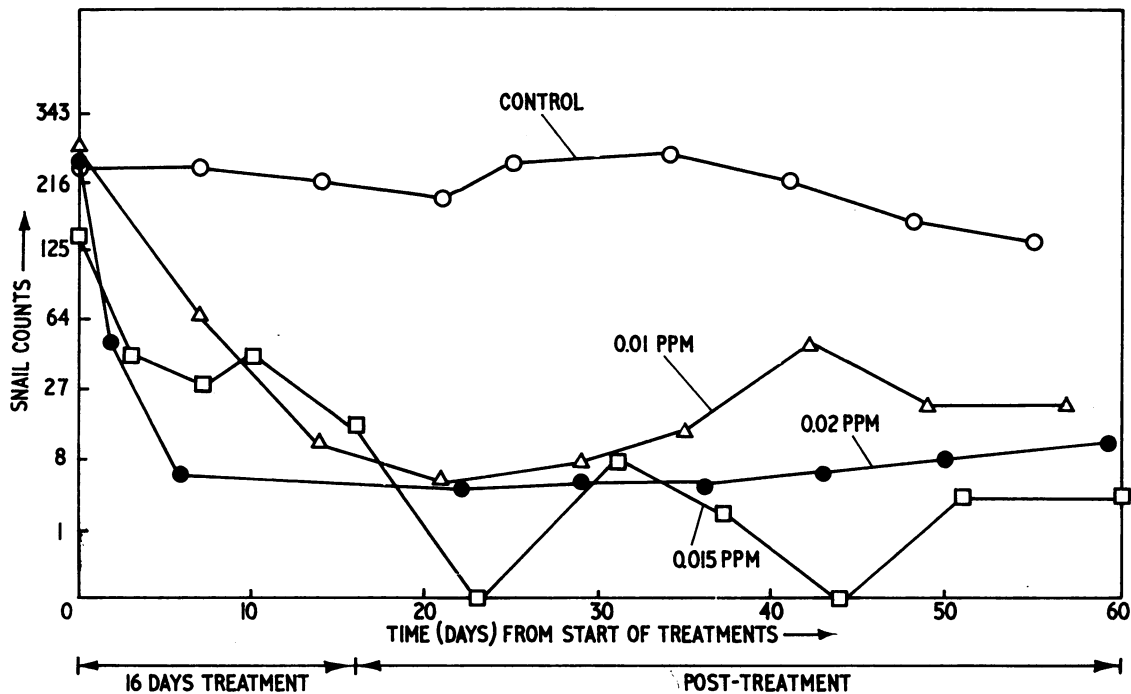


FIG. 6
EFFECT OF PROLONGED LOW-DOSAGE TREATMENT WITH *N*-TRITYLMORPHOLINE ON CAGED *BUL. TROPICUS*

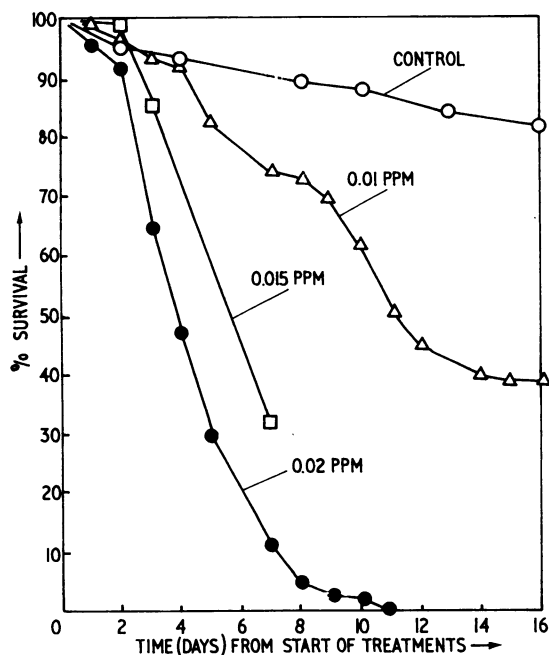


TABLE 1
NUMBER OF SNAIL EGG-MASSSES COUNTED
IN 3 MAN-HOURS^a

Concn of <i>N</i> -trityl-morpholine (ppm)	Time from start of treatment (days)	Egg-masses		
		Total no.	Dead (%)	"Early" ^b (% of total alive)
0.01	0-2	565	5.3	67.9
	3-7	277	32.9	50.0
	10-12	139	28.8	28.3
	14-16	164	23.8	51.2
	16	End of treatment		
23-25	286	8.0	87.1	
0.02	0-2	273	14.4	70.7
	3-7	254	48.4	32.1
	10-12	79	51.9	0
	14-16	23	87.0	0
	16	End of treatment		
	23-25	1	0	100
	36-38	0	0	0

^a Results obtained by pooling count in 1 man-hour's search on three successive days.

^b Approximately 0-7 days old.

adult populations of the two species. From the 14th day onwards it may be inferred that the egg-masses were mainly those of *Bul. tropicus*, because during this period no adult *Biom. pfeifferi* were found, whereas *Bul. tropicus* were always found, even if in relatively small numbers.

Interpretation of the results for the 0.02-ppm treatment is relatively easy. Between 0 and 2 days from the start, 273 egg-masses were collected; 14.4% of these were dead and 70.7% had been laid within the previous 7 days. Between 3 and 7 days, 254 egg-masses were collected, but 48.4% were dead and only 32.1% had been laid within the previous 7 days. From the 10th to the 12th days, 79 egg-masses were collected; 51.9% were dead and none had been laid during the previous 7 days. It follows that there was little or no egg-laying by either species of snail from the 3rd day onwards.

Determinations of *N*-tritylmorpholine in water samples are given in Table 2, together with the means and standard deviations for each series of samples. There is considerable variation in the data, partly as a result of difficulty in working at very low concentrations, the precision of the method at a concentration of 0.01 ppm being approximately ± 0.005 ppm, i.e., $\pm 50\%$. There is very little difference between the means of the morning and evening series of samples for any of the four sampling positions, suggesting a reasonably constant dosage rate. The standard deviations relative to the means are greater for the upstream samples than for the downstream samples, and this may be attributed to relatively poor mixing of the molluscicide at the upstream sampling positions. None of the means differs significantly from the nominal dose, although some of the individual determinations differ widely.

TABLE 2
ANALYTICAL DETERMINATION OF *N*-TRITYLMORPHOLINE IN WATER SAMPLES

Time (days) from start of treatment	Determined concentration of <i>N</i> -tritylmorpholine (ppm)							
	Nominal concentration 0.01 ppm				Nominal concentration 0.02 ppm			
	Upstream		Downstream		Upstream		Downstream	
	07.00 h	19.00 h	07.00 h	19.00 h	07.00 h	19.00 h	07.00 h	19.00 h
0	0.016	0.009		0.010	0.044	0.000		0.000
1	0.018	0.008	0.004	0.006	0.014	0.020	0.013	0.016
2	0.008	0.005	0.004	0.005	0.045	0.013	0.014	0.016
3	0.004	0.006	0.004	0.005	0.012	0.016	0.016	0.015
4	0.007		0.005				0.018	
5		0.006		0.008		0.000		0.011
6	0.030	0.013	0.004	0.005	0.004	0.009	0.006	0.000
7	0.017	0.007	0.004	0.000	0.007	0.017	0.009	0.013
8		0.019		0.011		0.034		0.018
9	0.007	0.013	0.008		0.015	0.024	0.014	0.018
10	0.009	0.016	0.008	0.011	0.028	0.023	0.018	0.026
11	0.012		0.011		0.036		0.025	
12		0.015		0.013		0.011		0.017
13	0.007	0.006	0.013	0.006	0.037	0.017	0.018	0.019
14	0.006	0.018	0.007	0.008	0.019	0.012	0.018	0.018
15	0.005	0.007	0.013	0.009	0.015	0.196	0.016	0.000
16	0.038		0.006		0.138		0.017	
Mean	0.013	0.011	0.0070	0.0075	0.032	0.028	0.016	0.0133
Standard deviation	0.010	0.0049	0.0033	0.0035	0.035	0.049	0.0047	0.0080

TABLE 3
MEAN MONTHLY WATER TEMPERATURES
IN EXPERIMENTAL CANALS

Month	Mean monthly water temperature (°C)			
	Upstream end of canals		Downstream end of canals	
	07.00 h	14.00 h	07.00 h	14.00 h
October	21.1	25.6	20.4	30.9
November	22.4	26.9	21.3	32.9
December	22.4	26.4	21.2	31.7

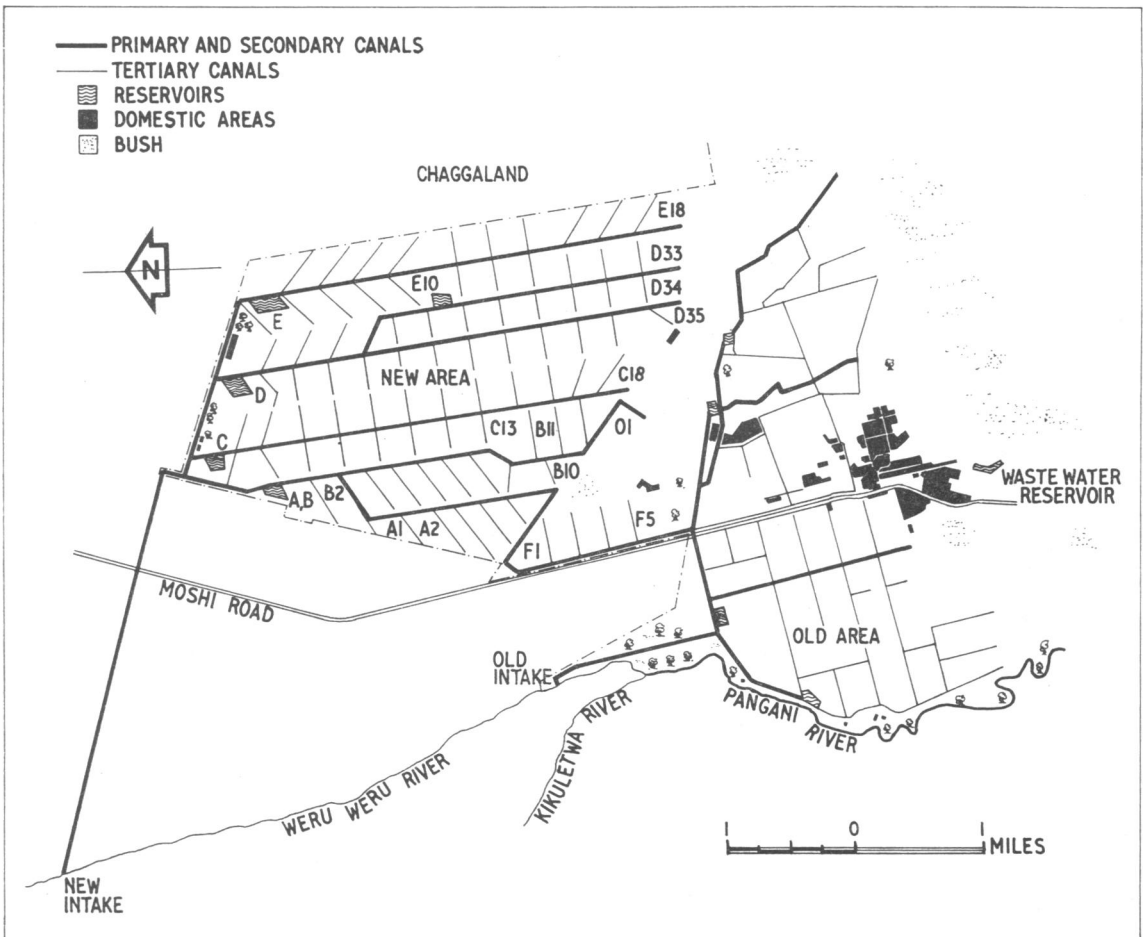
Daily water temperatures were recorded, and the monthly means of these are given in Table 3. There was a diurnal variation of about 4 deg C at the upstream end of the canals but at the downstream end, where there was less water movement, the variation was 10 deg C to 11 deg C.

LARGE-SCALE TRIALS

The experimental area

A large-scale trial was carried out at the Tanganyika Planting Company's sugar estate, situated in a low-rainfall area (yearly average 18 in (46 cm)) near Moshi in the Kilimanjaro region of Tanzania

FIG. 7
ARUSHA CHINI ESTATE OF THE TANGANYIKA PLANTING COMPANY



at an altitude of 2000 ft (610 m) (see Fig. 7). About 25 000 tons of sugar are produced annually from 9000 acres (3640 ha) of sugar cane, split into two areas of 5000 acres (2020 ha) and 4000 acres (1620 ha), which are called the New Area and the Old Area, respectively.

The New Area was chosen for the experiment because the pretreatment snail density was higher and road conditions were better than in the Old Area. Bilharziasis is the most important communicable disease on the estate. The prevalence of *Schistosoma mansoni* among the labour force is about 50% and among schoolchildren nearly 100%. *S. haematobium*, although common in other areas of Tanzania, is not of any importance at Arusha Chini. Snail-control measures during the last few years appear to have had some effect on the number of people infected with *S. mansoni*, but the African population has been largely transitory and data are inconclusive.

The estate is supplied with water from the Weru Weru River via two main canals with separate intakes. One of these canals normally has a discharge of 40 ft³/s (1132 l/s) and supplies water to the Old Area, while the other supplies water to the New Area and has a discharge of 60 ft³/s (1698 l/s). The company's water right is limited to an offtake of 100 ft³/s (2831 l/s) and water is therefore used up to its maximum availability. To make the best use of the available water the company has built a series of reservoirs so that water can be stored when not being used directly for irrigation. In the New Area, i.e., the experimental area, the main canal is about 4 miles (6.4 km) long and supplies water to 5 reservoirs with capacities ranging from 1 to 5 million ft³ (28 000 m³ to 144 000 m³). From the reservoirs secondary canals with carrying capacities ranging from 15 ft³/s to 30 ft³/s (424 l/s-849 l/s) carry water to an extensive system (about 100 miles, or 160 km) of tertiary canals, which run alongside the fields of sugar cane. Plastic siphons are used to feed the water from the tertiary canals to the rows of sugar cane. In the cane fields the water soaks into the soil or is lost by transpiration and evaporation, and therefore little drainage is required.

Snails are usually absent from the Weru Weru River because it is normally wide, deep and swift-flowing and there are no suitable habitats except during exceptionally long dry seasons. The main canal is also a poor habitat. The reservoirs, however, contain very large numbers of snails. The secondary canals also contain many snails, but in the tertiary

canals snails are found only at the upstream ends, where there is usually some permanent water.

The only medically important snail in the area is *Biom. pfeifferi*, intermediate host of *S. mansoni*. The most numerous snail is *Bul. tropicus*, itself of little medical importance but related to other species, e.g., *Bul. globosus* and *Bul. truncatus*, that are intermediate hosts of *S. haematobium*. Its susceptibility to the treatment was therefore of interest as an indicator of the possible susceptibility of other bulinids. *Lymnaea natalensis*, intermediate host of *Fasciola gigantica*, is also very common.

Application of molluscicide

Treating the measured quantity of water (70 ft³/s; 1982 l/s) with a dose of 0.025 ppm required 4.2 kg of active material (am) per 24 hours, or 25 litres of FX 28 (an emulsifiable concentrate that contains 16.5% *N*-tritylmorpholine). Two 44-UK gal (200-litre) drums were used, and 12.5 litres of FX 28 were added to each drum after mixing with water. The mixing was carried out in 4-UK gal (18-litre) containers and the mixture was then poured into the drums. The total volume in each drum was 200 litres and the mixture was therefore equivalent to 6.25% (v/v) of FX 28 in water.

The dispenser was similar to that already described and illustrated in Fig. 2, but two drums were needed to cope with the amount of molluscicide required. To prevent any possible interference with the dispenser, a concrete shelter was built for it and the outlet was protected by ½-in (1.25-cm) diameter lead piping cemented into a concrete wall. The nozzle size and the length of the outlet pipe were preset to give a constant emission rate of 280 ml/min.

The discharge of the canal was sometimes altered and in these instances the amount of molluscicide added to the drums was altered accordingly. The quantity of water taken into the main canal from the river was accurately measured ($\pm 5\%$) at a Cipoletti measuring weir and readings were taken daily before replenishing the drums with molluscicide. In addition to these daily readings an automatic water recorder was available, from which it was possible to check any diurnal variations. Once the dispensing routine had been established it required only 2 people, 1 junior technician and 1 labourer, to spend half-an-hour each day recharging the drums.

Experimental techniques

The Tanganyika Planting Company employs 4 men who systematically search the whole of the

irrigation system on a routine basis and report on the number of snails they find. Records are kept by the hospital staff, who use the data as a basis for judging the success of snail-control measures and for deciding how often to apply molluscicides. For the experiment with *N*-tritylmorpholine these data were useful, but to obtain more precise information it was necessary to examine part of the area in much greater detail. Two of the 5 reservoirs in the New Area and 7 of the secondary canals were therefore selected for detailed study.

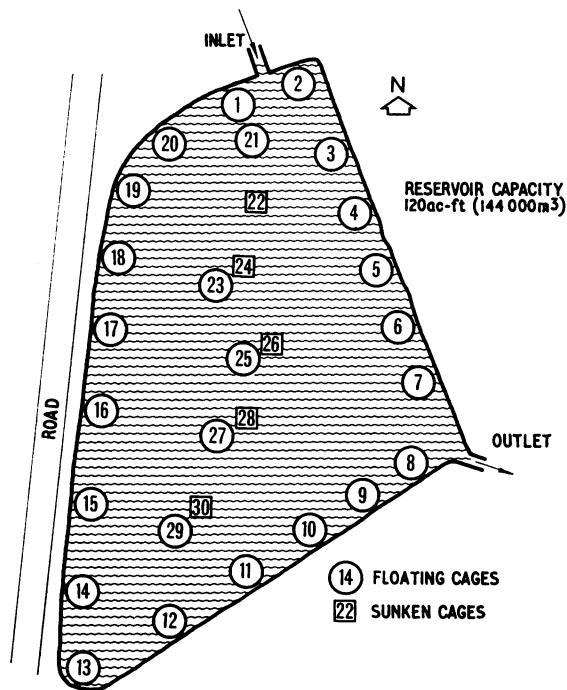
In these places drag scoops were used for snail sampling. Three scoops were taken at each sampling position and the numbers of snails collected per 3 scoops were pooled and counted as one sample. One hundred and fifty scoops, i.e., 50 samples, were taken each week from reservoir AB and also from each of the 7 canals, but because of dense undergrowth around the banks of reservoir C, paths had to be cut through to each sampling position and the number of samples was restricted to 26 (distributed at regular intervals around the circumference).

Seventy snail cages were placed at intervals throughout the treated area but, as with the snail-sampling programme, most effort was concentrated on reservoirs AB and C and their associated canals. In reservoir AB, 20 cages were placed at intervals around the sides and 5 floating cages plus 5 sunken cages were attached to stakes in deeper water in the main body of the reservoir (Fig. 8). Ten cages were placed around the sides of reservoir C (Fig. 9), 18 in the secondary and tertiary canals associated with reservoirs AB and C, and 5 in canal E, the canal farthest from the application point, i.e., 5 miles to 7 miles (8 km to 11 km). One week before the treatment was started, 25 *Biom. pfeifferi* and 25 *Bul. tropicus* were put into each cage. The cages were examined a day or two before the start of the treatment and any dead snails were removed and replaced by live ones. They were then examined at frequent intervals during the course of the treatment.

For the analytical determination of *N*-tritylmorpholine in water samples the field procedure for low dosage levels, as described by Beynon & Thomas,¹ was used with a battery-operated Unicam SP 1300 colorimeter. Calibration curves were derived by plotting the net absorbance against standard concentrations of *N*-tritylmorpholine within the range 0.01 ppm to 0.05 ppm, using the irrigation water. High blank values were sometimes obtained and

¹ See the paper on page 47 of this issue.

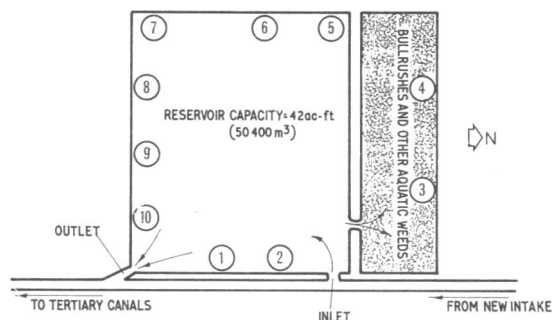
FIG. 8
POSITION OF THE CAGES IN RESERVOIR AB^a



^a 25 *Biom. pfeifferi* and 25 *Bul. tropicus* were placed in each cage a few days before treatment started.

these were attributed to an unusually high organic content, but in most cases the blank value was low and the results were reproducible. A few standards were made up at the start of each day's work and compared with previously prepared calibration curves. Whenever possible, water samples for analysis were taken from near the snail cages.

FIG. 9
POSITION OF THE CAGES IN RESERVOIR C^a



^a 25 *Biom. pfeifferi* and 25 *Bul. tropicus* were placed in each cage a few days before treatment started.

Results

The results of sampling for *Biom. pfeifferi*, *L. natalensis* and *Bul. tropicus* are given in Tables 4, 5 and 6. The results of the analytical determinations and the mortalities of caged snails are presented together in Tables 7, 8 and 9 to facilitate comparison between the biological and the chemical results.

Six water samples were collected from the main canal about 1 mile (1.6 km) downstream of the application point (Table 9); determinations of 5 of

these gave results at or near the nominal dose of 0.025 ppm, but there was one result of 0.01 ppm.

The next sampling positions were in reservoir AB, about 4 miles (6.4 km) downstream from the point of application. At the start of the treatment the reservoir was about two-thirds full. Water was taken in night and day at a rate of 10 ft³/s to 15 ft³/s (283 l/s to 424 l/s) and released from the outlet via a sluice gate between 05.00 h and 14.00 h, at a rate of 25 ft³/s to 30 ft³/s (707 l/s to 849 l/s). During the

TABLE 4
RESULTS OF SAMPLING FOR *BIOMPHALARIA PFEIFFERI*^a

Date	Number of snails taken from									Total
	Reservoir AB	Canal B1-B2	Canal B7-B8	Canal A2-A3	Reservoir C	Canal C4-C5	Canal C13-C14	Canal O	Canal E4-E5	
1965										
1 Feb.	181	607	412	165	63	246	102	610	99	2 485
10 Feb.	82	782	608	76	70	550	108	612	285	3 173
11 Feb.	Treatment started									
17 Feb.	0	0	0	0	65	0	0	0	0	65
24 Feb.	0	0	0	0	2	0	0	0	0	2
3 March	0	0	0	0	0	0	0	0	0	0
10 March	0	0	0	0	0	0	0	0	0	0
10 March	Treatment ended									
17 March	0	0	0	0	0	0	0	0	0	0
24 March	0	0	0	0	0	0	0	0	0	0
31 March	0	0	0	0	0	0	0	0	0	0
7 April	0	0	0	0	0	0	0	0	0	0
14 April										
21 April	0	0	0	0		0	0	0	0	0
28 April	0	0	0	0		0	0	0	0	0
5 May	0	0	0	1		0	0	2	0	3
12 May	0	0	0	1	Empty and dry from 20 April to 10 July	0	0	3	0	4
19 May	0	0	0	5		0	0	7	0	12
27 May	0	0	0	3		0	0	15	0	18
2 June	0	3	1	4		0	0	11	0	19
9 June	13	0	0	7		0	0	20	4	44
15 June	16	0	0			0				
23 June	19	0	5	19		0	3	15	0	61
1 July	16	2	5	30		0	4	20	3	80
8 July	29	4	6	14		0	4	21	0	78
15 July	27	6	11	23	0	0	0	43	0	110

^a 426 samples; 50 from each locality except reservoir C, where 26 samples were taken.

2nd day of treatment 6 water samples were collected from various places; the estimated molluscicide concentrations ranged between 0.015 ppm and 0.02 ppm (Table 7).

On the 3rd day, 21 out of 30 cages in the reservoir were examined and all *Biom. pfeifferi* were dead, although the mortality of *Bul. tropicus* was only 7.4%. By the 5th day the molluscicide concentration had reached 0.025 ppm. On the 6th day the water level had fallen and it was then possible to examine

all 30 cages, some of which had previously been submerged. All *Biom. pfeifferi* had been killed. There was an over-all mortality of 51.5% of *Bul. tropicus*, but the dead snails were not distributed evenly, there being fewer dead snails in cages 5-14 than in cages 1-4 and 15-30. Reference to Fig. 8 shows that these cages were on the perimeter of the reservoir farthest from the inlet and, therefore, after treatment for 5 to 6 days, had been subjected to molluscicide for a shorter period than the others.

TABLE 5
RESULTS OF SAMPLING FOR *LYMNAEA NATALENSIS*^a

Date	Number of snails taken from									Total
	Reservoir AB	Canal B1-B2	Canal B7-B8	Canal A2-A3	Reservoir C	Canal C4-C5	Canal C13-C14	Canal O	Canal E4-E5	
1965										
1 Feb.	64	253	116	11	1	28	31	288	9	801
10 Feb.	32	109	116	11	10	214	39	307	44	882
11 Feb.	Treatment started									
17 Feb.	0	0	0	0	0	0	0	0	0	0
24 Feb.	0	0	0	0	0	0	0	0	0	0
3 March	0	0	0	0	0	0	0	0	0	0
10 March	0	0	0	0	1	0	0	0	0	1
10 March	Treatment ended									
17 March	0	0	0	0	0	0	0	0	0	0
24 March	0	0	0	0	0	0	0	0	0	0
31 March	0	0	0	0	0	0	0	0	0	0
7 April	0	0	0	0	0	0	0	0	0	0
14 April										
21 April	0	0	0	0		0	0	0	0	0
28 April	0	0	0	0		0	0	0	0	0
5 May	0	0	0	0		0	0	0	0	0
12 May	0	0	0	0	Empty and dry from 20 April to 10 July	0	0	4	0	4
19 May	0	0	0	0		0	0	0	0	0
27 May	0	0	0	0		0	0	6	0	6
2 June	8	0	0	0		0	0	17	2	27
9 June	13	2	1	2		0	0	37	21	76
15 June	7	10	4	0		0	0			
23 June	16	8	7	0		0	0	31	10	72
1 July	17	8	6	2		0	0	13	3	49
8 July	30	11	9	6		0	23	13	27	122
15 July	49	4	8	10		0	4	9	37	127

^a 426 samples; 50 from each locality except reservoir C, where 26 samples were taken.

On the 7th day a series of 10 water samples was analysed and the results were all near to the nominal concentration. The results of samples 27-30 show that there was no difference in concentration between surface and bottom strata. The observations on caged *Bul. tropicus* were discontinued after 20 days, by which time there was a mortality of 96.1%. However, observations on the natural population were continued (Table 6) and after 30 days' treatment the population of *Bul. tropicus* in reservoir AB was eliminated.

Reservoir C was situated only a few hundred yards from reservoir AB but was of a very different character. The inlet and outlet were both situated on the eastern side and an earthen bank divided it into two parts, which were connected by a single narrow channel. The smaller part was completely overgrown with bull-rushes and other aquatic vegetation (Fig. 9). Within 2 days of the start of the treatment the concentration of molluscicide near the inlet was 0.025 ppm and shortly afterwards large numbers of dead snails, mainly *Biom. pfeifferi*, were seen floating on the surface.

TABLE 6
RESULTS OF SAMPLING FOR *BULINUS TROPICUS*^a

Date	Number of snails taken from									Total
	Reservoir AB	Canal B1-B2	Canal B7-B8	Canal A2-A3	Reservoir C	Canal C4-C5	Canal C13-C14	Canal O	Canal E4-E5	
1965										
1 Feb.	142	27	199	223	432	254	459	96	584	2 416
10 Feb.	91	15	173	27	363	288	582	68	607	2 214
11 Feb.	Treatment started									
17 Feb.	17	17	16	25	251	123	393	46	208	1 096
24 Feb.	9	14	15	21	413	97	417	31	9	1 026
3 March	3	9	7	15	167	24	125	17	4	371
10 March	0	2	0	3	124	22	103	0	0	254
10 March	Treatment ended									
17 March	0	0	0	0	129	24	83	0	2	238
24 March	0	0	0	0	474	20	108	0	9	611
31 March	0	0	0	0	266	24	116	0	11	417
7 April	0	0	0	0		27	117	6	13	163
14 April										
21 April	9	0	0	0		19	67	8	17	120
28 April	13	0	0	5		22	60	31	25	156
5 May	14	0	0	12		23	37	39	39	164
12 May	15	2	8	8	Empty and dry from 20 April to 10 July	32	58	120	52	295
19 May	16	9	11	28		57	108	105	59	393
27 May	15	17	8	22		52	106	188	57	465
2 June	23	22	26	29		21	59	185	24	389
9 June	57	18	27	62		47	265	235	115	826
15 June	79	36	52							
23 June	86	31	39	107		222	260	215	50	1 010
1 July	135	57	66	134		249	282	220	70	1 213
8 July	126	62	80	60		97	190	288	44	947
15 July	124	119	123	104	0	147	290	259	71	1 337

^a 426 samples; 50 from each locality except reservoir C, where 26 samples were taken.

TABLE 7
 NUMBER OF SURVIVING SNAILS IN CAGES IN RESERVOIR AB AND RESULTS
 OF ANALYTICAL DETERMINATIONS

Location of cages (see Fig. 8)	Number of surviving snails at time (days) from start of treatment								Concn of <i>N</i> -tritylmorpholine (ppm) after		
	<i>Biom. pfeifferi</i> ^a		<i>Bul. tropicus</i> ^a						2 days	5 days	7 days
	3 days	6 days	3 days	6 days	9 days	12 days	16 days	20 days			
1	0		22	8	1	0			>0.015		0.02
2	0		24	6	3	0			>0.015		
3	0		23	9	9	4	0				
4	0		21	8	4	2	1	0			
5	0		25	18	7	0					
6	0		24	17	7	Lost					>0.02
7	0		24	21	10	6	5	Lost	0.02	0.025	
8	0		23	23	12	9	8	4	0.02		
9	0		24	21	12	7	6	5			
10	0		21	12	5	5	1	0			
11	0		24	21	7	7	6	3			>0.02
12	0		24	20	5	3	2	0			
13	0		25	22	6	6	6	3	0.015	0.025	
14	0		24	24	7	7	4	4			
15	0		21	11	4	4	3	3			
16	0		24	13	5	3	3	0	0.02		
17		0		12	1	1	1	1			
18	0		24	6	2	2	2	0			>0.025
19	0		23	10	1	1	1	0			
20		0		6	1	1	1	0			
21	0		23	3	0						
22		0		4	0						
23		0		13	2	0					0.025
24		0		6	2	0					>0.02
25		0		6	5	1	0				
26		0		12	1	1	1	1			
27		0		6	6	3	2	0			0.025
28		0		15	4	2	2	1			0.025
29	0		23	9	9	8	7	2			>0.02
30	0		20	5	5	2	0				>0.02
Cumulative mortality (%)	100	100	7.4	51.1	80.9	88.3	91.4	96.1			

^a Each cage contained 25 *Biom. pfeifferi* and 25 *Bul. tropicus* when treatment started.

TABLE 8. NUMBER OF SURVIVING SNAILS IN CAGES IN RESERVOIR C AND RESULTS OF ANALYTICAL DETERMINATIONS

Location of cages (see Fig. 9)	Number of surviving snails at time (days) from start of treatment														Concn of N-triphenylmorpholine (ppm) after					
	<i>Biom. pfeifferi</i> ^a							<i>Bul. tropicus</i> ^a							2 days	4 days	5 days	6 days	7 days	8 days
	3 days	5 days	7 days	11 days	13 days	15 days	21 days	3 days	5 days	7 days	11 days	13 days	15 days	21 days						
1	0							20	14	4	1	1	0	0.025						
2	0							5	1	1	0									
3	1	0						2	0											
4	21	19	17	15	9	6	0	22	22	21	21	20	20	0	<0.01	<0.01	>0.025			
5	2	1	1	0				19	15	15	8	8	8	8	<0.01	0	0.015	0	0	
6	15	8	5	4	1	1	1	24	23	23	23	23	22	22	<0.01	>0.015	>0.015	>0.015	0	
7	16	3	2	0				25	23	23	19	19	19	16	<0.01	0.015	0.02	0.02		
8	6	0						21	21	19	13	13	13	13	0.01	0.01	0.02			
9	0							22	21	18	18	18	16	15						
10	0							19	15	14	10	10	9	8						
Cumulative mortality (%)	75.6	87.6	90.0	92.4	96.0	97.2	99.6	28.4	38.0	44.8	54.8	55.2	57.2	67.2						

^a Each cage contained 25 *Biom. pfeifferi* and 25 *Bul. tropicus* when treatment started.

In those cages placed near the inlet (cages 1 and 2 in Fig. 9) or near the outlet (cages 9 and 10) there was 100% mortality of *Biom. pfeifferi* within 3 days. In cages 3 and 8, which were farther from the main flow of water, there was 100% mortality after 5 days, and in cages 5 and 7, still farther from the main flow, there was 100% mortality after 11 days. In cage 4, which was placed in an overgrown corner far from any discernible water movement, some snails survived for 20 days. In cage 6, also in a backwater, one snail survived even longer.

Water samples were taken on day 4 from positions 4, 5, 6, 7 and 8; the determined molluscicide concentrations were all 0.01 ppm or less. Further samples taken from these places on days 5 and 6 indicated a gradual increase in concentration except in the far corner of the overgrown part of the reservoir.

In the secondary canal fed from reservoir C there was 100% mortality of caged *Biom. pfeifferi* within a few days, showing that the molluscicide passed through the reservoir with little or no loss, even though it had not penetrated into certain places. There was a similar effect on the natural population of *Biom. pfeifferi*. Seventy snails were collected in 26 samples (Table 4) during the week before treatment and 65 in a similar series 6 days from the start, but, whereas snails were found in 23 out of 26 pre-treatment samples, on day 6 they were found in only 6 of the samples, all from the western perimeter of the reservoir. After 3 weeks no more *Biom. pfeifferi* were found. The toxicological effect on the population of *Bul. tropicus* in reservoir C and its associated canals was very much less than in the AB system.

In most of the cages placed in the canals there was 100% kill of *Biom. pfeifferi* within 2-3 days (Table 9), but in some, e.g., in B6 and in the cage placed in the overflow from reservoir C, snails lived for up to 11 days. In these places there was no permanent flow of water and the molluscicide took longer to penetrate. Cage B6 was placed in a pool at the top of a tertiary canal that was not being used for irrigation and this pool was fed only intermittently by seepage water. Similarly, the pool caused by the overflow from reservoir C was fed with water intermittently.

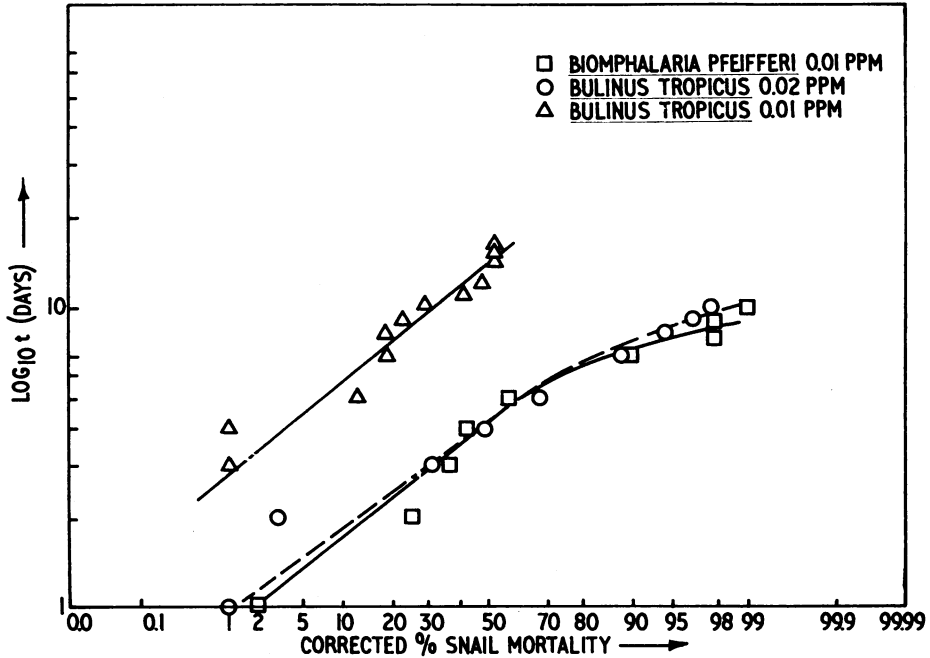
The treatment eliminated *Biom. pfeifferi* from the irrigation system for about 3 months. The first snails to be found after the treatment were towards the downstream end of the irrigation system in the secondary canals A and O. Small numbers of snails were recovered in the samples for a further period of a month, and then there was a gradual increase in numbers. *Biom. pfeifferi* were not found in reservoir

TABLE 9
 NUMBER OF SURVIVING SNAILS IN CAGES PLACED IN VARIOUS SECONDARY AND TERTIARY CANALS
 AND RESULTS OF ANALYTICAL DETERMINATIONS

Location of cages (see Fig. 7)	No. of surviving <i>Biom. pfeifferi</i> ^a after							Concn of <i>N</i> -triphenylmorpholine (ppm) after						
	3 days	5 days	7 days	11 days	13 days	15 days	20 days	2 days	4 days	5 days	6 days	7 days	8 days	9 days
Main canal								0.025	>0.025		0.025	0.01 (?)	>0.025	0.025
Reservoir D											>0.025			
Reservoir E1 Inlet													>0.02	
Outlet (1)													>0.02	>0.025
Outlet (2)											>0.01		>0.025	>0.015
Reservoir E10 Inlet								0.015		0.01				
Outlet								0.015		0.015				
Secondary canals														
01			0					0.015		0.01				
02			0											
03			0											
04			0											
05			0											
F1			0					0.02						
F2			0											
F3			0											
F4			0											
F5			0											
Tertiary canals														
B1								0.025						
B3	0													
B4	0								>0.015					
B5	0								0.015					
B6	18	15	6	0					?			>0.025		
B7	7	0							0.015					
C3	0													
C7	1	0												
C8	0													
E10			20	3	0									
E12			20	15	12	5	0							
E14			24	0										
E15			19	0										
E16			18	Lost				trace		0	0.015		trace	0.015
Overflow from reservoir C	15	8	0						0		>0.025			

^a Each cage contained 25 snails at the start.

FIG. 10

RELATION BETWEEN SUSCEPTIBILITY OF SNAILS TO LOW CONCENTRATIONS OF *N*-TRITYLMORPHOLINE AND DURATION OF EXPOSURE

AB for 4 months. Elsewhere snails were absent for periods varying from 3 to 5 months.

DISCUSSION

In the pilot trials the mortality rate of *Bul. tropicus* in the 0.02-ppm treatment was very similar to that of *Biom. pfeifferi* in the 0.01-ppm treatment (Fig. 4 and 6). There were few deaths during the first 2 days, then a period of 5 days when 80%-90% of the snails died, and finally a period of 4 days when the remaining 5%-10% died. The sigmoid shape of these curves is similar to that obtained with a variable concentration and constant exposure time. This suggested that the methods of probit analysis might be applicable and to test this possibility the data were plotted on logarithmic probability paper after correcting for control mortalities by Abbott's formula (Fig. 10). Between the limits of 20% and 70% mortality there is a good fit to straight regression lines, although for high mortalities a curvilinear trend is indicated. The curves for exposure of *Biom. pfeifferi* to a dose of 0.01 ppm and exposure of *Bul. tropicus* to 0.02 ppm are nearly identical. The curve

for exposure of *Bul. tropicus* to 0.01 ppm is parallel to the others but there are no data for mortalities greater than 52%.

The LT_{50} and LT_{90} have been estimated for concentrations of 0.01 ppm, 0.015 ppm, and 0.02 ppm and are given in Table 10. The LT_{50} increases three- to four-fold when the concentration decreases from 0.02 ppm to 0.01 ppm, i.e., the product of concentration and time (ct) for a given percentage mortality increases with time. It appears at first sight

TABLE 10
 LT_{50} AND LT_{90} FOR PROLONGED, LOW DOSAGES OF *N*-TRITYLMORPHOLINE

Snail species	LT_{50} and LT_{90} (days) at <i>N</i> -tritylmorpholine concn of					
	0.01 ppm		0.015 ppm		0.02 ppm	
	LT_{50}	LT_{90}	LT_{50}	LT_{90}	LT_{50}	LT_{90}
<i>Biom. pfeifferi</i>	4	7	<1	1.85	<1	1.0
<i>Bul. tropicus</i>	14	25	5.5	10	4	7

that it would be more economical to apply *N*-tritylmorpholine in high doses for short periods than in low doses for prolonged periods. This is almost certainly true for some situations but it is not so for irrigation systems because of the nature of the application problems.

These are best illustrated by reference to a particular example. Each case must be considered on its merits, but the general principles considered here are widely applicable. The New Area of the Arusha Chini estate (Fig. 7) was treated with Bayluscide in 1961 (Crossland, 1963), the aim being to treat the canals with 1 ppm for 8 hours and the reservoirs with 0.5 ppm for 24 hours or longer. During the week before the treatment started all the reservoirs were drained so that dilution would be minimal. The reservoirs were then filled with treated water; this took between 2 and 4 days. To achieve a final concentration of 0.5 ppm in the reservoirs, the incoming water was treated with 1 ppm for 12-hour periods, with intervals of 12 hours between treatments. The reservoirs were filled two at a time and then, after the treated water had been allowed to stand for 24 hours, it was released to the secondary and tertiary canals. By this time the concentration of molluscicide had fallen to 0.25 ppm and it was necessary to apply booster applications of 1 ppm for 8 hours at the reservoir outlets. Gangs of labourers were stationed downstream from each reservoir to ensure that treated water was channelled to each tertiary canal, and small earthen weirs were built near the top of the tertiaries to cause ponding of the treated water and thus prolong the effect of the molluscicide where it was most needed. The application took 6 days; 80 labourers were used to assist with the water management and the whole operation had to be carefully planned in advance, with the close collaboration of the irrigation department. The treatment schedule was certainly elaborate but it was the best way of using the techniques and materials that were available at the time.

It should now be clear that practical considerations are more important to the field worker than whether or not any particular value of *ct* is more effective than another. Of course, there are limitations even to this conclusion. For example, with *N*-tritylmorpholine a prolonged dose of 0.01 ppm would need to be applied for at least 30 days, assuming no downstream losses, to achieve satisfactory control of *Biom. pfeifferi*, whereas 0.02 ppm need be applied for only 4-5 days to give a similar effect. The most economical dosage regime may best be chosen by a com-

promise between practical necessity and the most effective use of known *ct* relationships.

If the LT_{50} for a given low dose is greater than about 3 days, the value of *ct* for a given percentage kill is so great that the dosage becomes uneconomic. In practice, the most suitable concentration for a particular snail species is that which gives an LT_{50} of about 24 hours. This is equivalent to the LC_{50} for a constant exposure period of 24 hours, a statistic that has been evaluated for a wide range of snail species. The most suitable application time is the shortest that is needed to give complete coverage of the irrigation system, but with a minimum of 3-4 days. These conclusions may be summed up in the following generalized statement for choosing dosage levels for irrigation systems:

(1) Evaluate the LC_{50} (*x* ppm) for a 24-hour exposure period for the particular species of snail(s).

(2) Estimate the maximum time (*y* days) for the water to travel from the intake to the extremities of the system (e.g., by a pilot trial of the low-dosage technique). If $y < 3$ the prolonged low-dosage technique may not be applicable and consideration should be given to the choice of another dosage regime.

(3) Where there is a continuous flow of irrigation water, apply the molluscicide in a concentration of *x* ppm for *y* days.

(4) Where the water flows in cycles, e.g., in the area of the Nile Delta, other factors may be of overriding importance. For example, if the cycle is 5 days open, followed by 10 days closed, the molluscicide should probably be applied for the whole of the 5-day "open" period, and the treatment repeated as necessary, depending on repopulation rates and climatic factors.

In the large-scale trial at Arusha Chini, which was the first of its kind, the application time was 30 days but *Biom. pfeifferi* were eliminated from all except a few places after only 4-5 days. For experimental purposes the natural population was allowed to recover from the effects of the treatment, but in later work a low dose was applied for 5 days once every month. This has given effective snail control and it seems likely that the interval between treatments could be extended. In some places on the estate the treatment has not been effective. All such places are backwaters or pools of standing water where the molluscicide cannot penetrate. However, since these are very few and easily identified, the snails can be eliminated by localized treatments.

After treatment of the New Area with *N*-tritylmorpholine, snails reappeared 3 months later and the first snails were found towards the downstream extremities of the irrigation system. Similar results have been observed after treating the area with other molluscicides. Although every effort has been made to treat all the snail habitats, which in this case are relatively well defined, repopulation has always taken place a few months, or even weeks, after the end of the treatment. The first snails are invariably collected towards the downstream end of the irrigation system. Further, it has been shown that relatively few snails are introduced from the river. Possibly, if snails were eliminated, they were reintroduced from outside but more probably some snails, or snail eggs, escaped or survived treatment whatever molluscicide or method of application was used and these survivors and their progeny were capable of rapid recolonization of the available habitats. Assessment of the relative importance of these methods of recolonization would be valuable, but the problem may be intractable, owing to the difficulty of detecting snails in very low densities.

In the 30 days' treatment of the New Area, 140 UK gal (640 litres) of FX 28 (a 16.5% emulsifiable concentrate) were used. This treatment cost about £560 (\$1570), which was appreciably less than the cost of treating the same area with other molluscicides. One treatment with any of these molluscicides is generally effective for 3-4 months. However, if *N*-tritylmorpholine is applied for only 5 days, instead of 30 days, the snails can be eliminated. The cost of control of snail eggs with this molluscicide was therefore 6 times that of the cost of control of snails.

On the other hand, it may be that the 30 days' treatment was unnecessarily prolonged and that the same degree of control could have been achieved by 15 days' treatment, together with localized treatment of places where the molluscicide failed to penetrate. Even so, the most effective use of molluscicide seems to be achieved by relatively frequent applications, e.g., once a month, designed to control snails, but not necessarily snail eggs.

Under some conditions, low-dosage treatment with *N*-tritylmorpholine has been highly selective. At Arusha Chini, no fish, tadpoles or other aquatic animals were killed. In a similar large-scale trial in Southern Rhodesia a dose of 0.03 ppm was applied for 10 days to an area that included 24 night-storage dams containing large numbers of fish. Although highly effective against *Biom. pfeifferi* and *Bul. globosus*, the treatment had no effect on the fish. On the other hand, in trials carried out in Egypt, a dose of 0.04 ppm gave a high mortality of fish. These results have stimulated a more intensive study of the toxicity of *N*-tritylmorpholine to fish. Shiff, Crossland & Miller (1967) carried out a series of carefully controlled trials in 6 identical concrete-lined fish ponds and found a remarkable difference in the susceptibility of two species of *Tilapia*. *T. melanopleura* was susceptible to molluscicidal doses but *T. mossambica* was not. They conclude that "this difference . . . is most important in relation to practical application. *Tilapia mossambica* is indigenous to Africa and can be cultured in most waterbodies. If this species is introduced into fish farms in areas where snail-control procedures are necessary, the problem of the accidental destruction of fish by molluscicide can be obviated".

POSTSCRIPT

The data presented in Table 1 indicate a surprisingly high mortality of snail eggs following exposure to a molluscicide which is reputedly "non-ovicidal". These data have prompted the author to a more detailed study of the effects of prolonged doses on snail embryos. Under laboratory conditions doses of 0.05 ppm or less have little effect even when maintained for a period of up to 7 days. On the other hand, concentrations of 0.1 ppm or greater are lethal to snail embryos if maintained for 48 hours or longer. Under such conditions of exposure the development of the embryo may be markedly retarded and it may die before hatching; or if development proceeds more or less normally the young snail dies soon after hatching.

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RÉSUMÉ

L'auteur décrit une technique d'application du molluscicide *N*-tritylmorpholine dans les systèmes d'irrigation. Au moyen d'un dispositif simple, le produit est introduit à l'origine du réseau sous des concentrations faibles, mais constantes, pendant des périodes prolongées.

Au cours d'essais préliminaires dans des canaux expérimentaux, le traitement par 0,01, 0,015 et 0,02 partie par million de *N*-tritylmorpholine a entraîné chez *Biomphalaria pfeifferi* une mortalité de 100%. Après 16 jours d'application du produit, les nouvelles générations de mollusques ont également été détruites; on peut donc estimer que cette technique permet de pallier dans une certaine mesure le manque d'action ovicide de la *N*-tritylmorpholine. L'expérimentation a été poursuivie sur une plus grande échelle en Tanzanie. Ici, le molluscicide, à la concentration de 0,025 partie par million, a été incorporé pendant 30 jours consécutifs, aux installations de départ d'un système d'irrigation couvrant 2000 hectares et comportant cinq grands réservoirs et un réseau étendu de canaux. Après 4 à 5 jours d'application, la mortalité chez *Biomphalaria pfeifferi* a été de 100% dans la quasi-totalité du secteur expérimental, le molluscicide ayant pénétré plus lentement dans quelques endroits où l'eau était à demi-stagnante. Pour *Lymnaea natalensis* et *Bulinus tropicus*, les taux de mortalité ont atteint 100% et 90-95%

respectivement. L'efficacité du traitement s'est maintenue pendant 3 à 4 mois. Ces résultats, comparables à ceux obtenus par d'autres méthodes utilisant d'autres composés, ont cependant été acquis à moindre frais et n'ont exigé que peu de main-d'œuvre. Le coût des opérations peut encore être réduit si l'on applique le molluscicide à faibles concentrations pendant des périodes de 4 à 5 jours toutes les 4 à 8 semaines, et si l'on y adjoint un traitement des collections d'eau stagnante où le produit ne pénètre qu'insuffisamment.

Deux autres essais importants ont eu lieu dans un système d'irrigation de Rhodésie. L'application de *N*-tritylmorpholine à la concentration de 0,03 partie par million pendant 10 jours a donné des résultats satisfaisants en ce qui concerne *Biomphalaria pfeifferi* et *Bulinus globosus*. On n'a noté aucune action piscicide à cette occasion, mais des tentatives analogues effectuées en Egypte ont amené la mort de nombreux poissons. Une expérimentation ultérieure, en Rhodésie, a mis en évidence une différence de sensibilité envers le molluscicide chez *Tilapia* selon les espèces.

Les problèmes théoriques et pratiques que pose la sélection des concentrations les plus favorables sont examinés. L'auteur suggère une formule générale destinée à faciliter ce choix.

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