

- Schubert, M. (1948) *Amer. J. trop. Med.*, **28**, 121-136
 Smithers, S. R. (1956) *Trans. roy. Soc. trop. Med. Hyg.*, **50**, 354-365
 Standen, O. D. (1949) *Ann. trop. Med. Parasit.*, **43**, 268-283
 Stirewalt, M. A. (1954) *Exp. Parasit.*, **3**, 504-516
 Sturrock, R. F. (1965) *Ann. trop. Med. Parasit.*, **59**, 1-9
 Teesdale, C. (1962) *Bull. Wld Hlth Org.*, **27**, 759-782
 Wright, C. A. (1963) *Trans. roy. Soc. trop. Med. Hyg.*, **57**, 142-147

Seasonal Variations of Cercarial Output from *Biomphalaria pfeifferi* and *Bulinus (Physopsis) globosus* in a Natural Habitat in Southern Rhodesia

by M. S. SHATTOCK, R. J. FRASER and P. A. GARNETT, *Bilharziasis Research Laboratory, Salisbury, Southern Rhodesia*

Pesigan et al. ^a in the Philippines and Pitchford & Visser ^{b, c} in South Africa have described the exposure of mice to bilharziasis by immersion in natural waters. From the lack of further information on this subject in the literature, it would appear that little further investigation has been carried out to determine any seasonal variations of cercarial output from infected snails. It was therefore decided that an attempt be made to discover what seasonal variations, if any, exist in the cercarial density of natural water-bodies.

A suitable site, 4 miles (7 km) east of Salisbury, Southern Rhodesia, on a small stream flowing into the Makabusi River, was chosen from which to collect a total of 100 snails on the same day of every alternate week, at approximately the same time of day and from the same place, for an entire year (April 1963 to March 1964). This particular site was selected because of its large snail population and the considerable human contact with the water.

The stream is fed from a fairly large catchment of a spongy nature and from a water reservoir, seepage from which keeps the stream barely flowing throughout the long, dry winter season.

The site of the sampling was a number of pools through which the water moves slowly, being held up by a series of rocky shelves, the stream trickling over bare granite surfaces into sedge-lined pools about 4-6 feet (1-2 m) wide and of the same length.

It was estimated that the dry-weather water-flow was approximately 0.25 ft³/sec (0.0762 m³/sec).

Within about 150 m of the site there is a large housing area used as transit accommodation for African labourers and their dependants going to and from Malawi, Zambia, and the Johannesburg gold mines. At this point, the stream is much used by the women for washing clothes and for personal ablution. It is obvious, from the state of the ground and of the vegetation on either bank of the stream, that the whole site is used as an outdoor latrine, particularly by children. The stream is perennial, with the vegetation on the banks and in the river-bed being fairly profuse and dense but not more than three feet (one metre) in height.

Materials and methods

A total of 100 snails, or as near to that number as could be found, was collected of *Biomphalaria pfeifferi* and *Bulinus (Physopsis) globosus*, in as near to equal numbers as possible, and placed in a polyethylene bag containing a little water. At the laboratory, they were transferred to a glass aquarium 12 inches (20.5 cm) in diameter and 6 inches (10.25 cm) high, containing matured water.^d A few pieces of dried lettuce, prepared according to the methods of Claugher,^d were offered as food.

On the following day, at about 10 a.m., the snails were removed from the tank and placed individually into 3-inch × 1-inch (75-mm × 25-mm) glass specimen tubes, each tube containing 10 ml of matured water. These tubes were then exposed to intense illumination for two hours, after which time the

^a Pesigan, T. P., Hairston, N. G., Jauregui, J. J., Garcia, E. G., Santos, E. T. & Besa, A. A. (1958) *Bull. Wld Hlth Org.*, **18**, 481.

^b Pitchford, R. J. & Visser, P. S. (1962) *Trans. roy. Soc. trop. Med. Hyg.*, **56**, 126.

^c Pitchford, R. J. & Visser, P. S. (1962) *Trans. roy. Soc. trop. Med. Hyg.*, **56**, 294.

^d Claugher, D. (1960) *Ann. trop. Med. Parasit.*, **54**, 333.

SNAILS COLLECTED AND FOUND INFECTED AND CERCARIAL OUTPUT MEASURED BY EXAMINATION
FOR TWO HOURS ON TWO SUCCESSIVE DAYS

Date snails collected	<i>Biomphalaria pfeifferi</i>				<i>Bulinus (Physopsis) globosus</i>			
	Number collected	Average No. infected	Percentage infected	Average No. cercariae/infected snail/2 hrs	Number collected	Average No. infected	Percentage infected	Average No. cercariae/infected snail/2 hrs
2 April 1963	75	3.5	5	423	15	5.5	37	502
15 April 1963				Flooding				
2 May 1963	61	8.0	13	347	27	13.0	48	96
15 May 1963	63	3.0	5	160	39	20.0	51	124
5 June 1963	32	7.5	23	197	66	29.0	44	91
13 June 1963	70	9.5	14	424	30	15.5	51	195
25 June 1963	53	4.5	8	150	47	5.0	11	20
2 July 1963	52	2.2	4	116	48	0.0	0	0
17 July 1963	66	0.0	0	0	34	3.0	9	93
30 July 1963	66	1.0	1	7	34	0.0	0	0
13 Aug. 1963	63	0.0	0	0	37	0.0	0	0
27 Aug. 1963	76	2.5	3	90	24	0.0	0	0
10 Sept. 1963	74	1.5	2	297	24	0.0	0	0
25 Sept. 1963	30	1.0	3	515	31	14.0	4	170
8 Oct. 1963	32	3.0	9	58	34	18.0	53	444
22 Oct. 1963	20	2.0	10	240	15	12.5	83	342
Nov. 1963				No snails collected				
10 Dec. 1963	24	3.0	12	1415	1	0.0	0	0
Dec. 1963 to March 1964				Flooding				

lights were switched off and the snails removed from the tubes and replaced in the glass tank.

The number of tubes containing cercariae were counted separately for each species and recorded. All cercariae shed from *B. pfeifferi* were pooled in a glass beaker, and the same was done for *B. (Ph.) globosus*. The cercariae, in measured aliquots from the beaker, were counted under a dissecting microscope, and it was thus possible to estimate the total number of cercariae shed in two hours by the infected snails of each species.

Twenty-four hours later, the procedure was repeated with the same snails, and the number found infected and the cercarial output were again recorded.

From the two sets of figures obtained, the average number of infected snails and the average number

of cercariae being shed per infected snail in a two-hour exposure period were calculated.

All surviving snails were returned to the original habitat. From April to August, all snails collected were returned, but in September and October a few snails of both species died during the 48 hours of examination.

Discussion

The accompanying table sets out the data obtained. It is of interest to find that *B. pfeifferi* were more numerous in this habitat than *B. (Ph.) globosus*, and also that collections of 100 snails from a rather restricted habitat could be made throughout the dry season until mid-September, when the temperature rose and showers of rain occurred.

The number of infected snails, particularly *B. (Ph.) globosus*, found during the period of the experiment showed marked variations. A proportion of infected *B. pfeifferi* persisted throughout the winter months, but infected *B. (Ph.) globosus*, on the other hand, disappeared during this period. The average cercarial output per infected snail was also at a minimum during the latter part of the winter months of July to September. V. de V. Clarke (unpublished) and Schiff^e have also mentioned that seasonal variation exists in the transmission of bilharziasis.

During the early winter months of May and June, although the cercarial output was high, the lowered seasonal temperatures limited the breeding of snails. At the same time, contact with natural water-bodies by the human population was probably also limited. During July and August, when snails were beginning to recover from the cold, so few cercariae were liberated and so little human contact with natural water took place that little or no infection was likely. However, during October and November, the hottest months of the year, cercarial output was very high and, at the same time, human contact with natural water was frequent, with the result that, during this period, opportunities for transmission were the highest in the year. From December to March, heavy rains fell and floods occurred that washed away so many snails that negligible infection could have taken place. From April, when stable weather conditions returned, the snail populations began to build up until the cold weather again set in.

There was a seasonal periodicity of infectivity of

the water-body, with two peak periods: from April to June and from October to December, at which times there were more vector snails infected and shedding cercariae. During October, the proportion of infected snails collected was high and, because of the hot weather, human contact with natural water was frequent. Thus it is probable that, in Southern Rhodesia, most infections are contracted by humans during this period. The high infection rates for snails and the high cercarial output of infected snails would continue from November through March but for the heavy, flushing rains, which have a catastrophic effect on snail existence in natural watercourses.

Simultaneously with the foregoing experiments, the immersion of eight mice was carried out, using methods similar to those of Pitchford & Visser,^{b, c} every week for nine months, from 1 April 1963 to 6 January 1964, at the same site from which the snails were collected. The mice were kept for an incubation period of at least two months and then killed and perfused. The procedure for perfusion was modified from the methods of Yolles and his co-workers.^f It has been demonstrated in this laboratory, in other experimental work, that this technique ensures nearly total recovery of worms from mice. However, no worms were recovered from any of these mice, despite the high cercarial output from both *B. pfeifferi* and *B. (Ph.) globosus*. This negative result was surprising in view of the excellent results achieved with this method by other workers.

^e Schiff, C. J. (1964) *Ann. trop. Med. Parasit.*, **58**, 240.

^f Yolles, T. K. Moore, D. V., Giusti, D. L., de, Ripsom, C. A. & Meleney, H. E. (1947) *J. Parasit.*, **33**, 419.