# NEW FIELD DEVICE FOR QUANTITATIVE RECOVERY OF SCHISTOSOMA MANSONI CERCARIAE

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ANY EQUIPMENT that will permit direct quantitative recovery of *Schistosoma mansoni* cercariae from natural waters is an important tool for studying schistosome epidemiology.

A device utilizing the positive phototropism exhibited by the cercariae of S. mansoni was developed by Klock in 1955 (1). It has the disadvantage of requiring intense artificial light necessitating use of current from public utility lines or a portable generator. Therefore, its use under field conditions is limited.

An apparatus developed by Rowan in 1956 (2, 3) was based on the filtration principle, first as pressure filtration and later modified to the Buchner funnel vacuum filtration process (4). The cercariae were collected on filter paper which was then placed in a small amount of ninhydrin dye. The paper was steamed until almost dry, and the stained cercariae on the drv paper were counted in the laboratory. When the water was so turbid that cercariae were obscured on the filter paper, water had to be collected in large containers and flocculated, using sodium bicarbonate and filter alum. This procedure necessitated an additional hour of field time and considerably more equipment and chemicals.

It had been observed that light material suspended in the turbid water stopped the filter process. Continuous-flow centrifugation was indicated as a means of collecting cercariae and also allowing the major light suspended ma-

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### Description

A simple hand centrifuge, A, is the basic component of the new unit (see figure). In place of the nut that normally holds the fourtest-tube centrifuge head in position, a special cup of brass, B, is used. The inside of the cup is an inverted cone just as is the outside.

The bottom of the cup is drilled at its exact center and tapped with threads so that it can replace the nut. Four holes are drilled through the cup at the bottom outside edge exactly in line with the four test tube holders, C, and flexible plastic tubes, D, approximately 3 inches long with  $\frac{1}{8}$ -inch inside diameter, are fitted into each of these four holes. These plastic tubes from the special cup can be bent and the opposite ends put into the inside of the plastic centrifuge tubes in the holders.

When the centrifuge is rotated and the test tubes are swinging straight out, the plastic tubing is straight from the brass cup into the centrifuge tubes for about 2 inches. When water is poured into the top of the cup, it is forced out through the plastic tubes into the centrifuge tubes. When the centrifuge tubes are full, the excess water spills over their top edges and is thrown out.

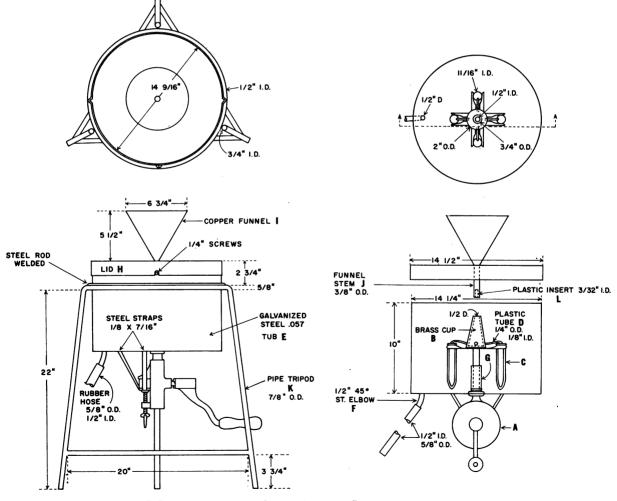
A pan or catching tub, E, is designed so that the centrifuge head can rotate inside it and the excess water can be caught and drained off from one small hole in the bottom through an elbow, F. This tub resembles an angelfood cake pan, with the revolving centrifuge head inside the pan, the vertical rotating shaft extending through the pipe, G, in the center of the pan, and the gear box and handle of the centrifuge below the pan.

A lid, H, with a funnel, I, fitted to its exact center is so designed that the actual funnel is on top, but its  $3\frac{1}{4}$ -inch stem, J, is on the underside. When the lid is placed on the catching tub, the funnel stem extends 2 inches inside the cup fitted to the revolving centrifuge head. A simple pipe tripod stand, K, holds and supports the entire mechanism; however, any type of support can be used.

The key element in the device and its use is that the opening of the input funnel stem must be calibrated to let into the cup only that amount of water which can be carried off by the four plastic tubes going to the centrifuge tubes. If the amount of water is in excess of this, it will run over the top of the cup and part of the sample will be lost. Therefore, each unit must be individually calibrated with its own funnel, cup, and tubing. This particular unit required a plastic insert with a  $\frac{5}{64}$ -inch inside diameter orifice, L.

#### **Procedures and Results**

A 5-gallon sample of water from an infected stream can be put through this device in 20 to 30 minutes. After the sample of water has passed and the head of the centrifuge is still spinning, 10 percent formalin with picric acid is added. The formalin-picric acid solution kills, preserves, and stains the cercariae while they are held by centrifugal force in the bottom of the centrifuge tubes. The total count of the cercariae is made from the solution remaining in the test tubes, and they can be examined mi-



Schematic view of continuous-flow cercariometer

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croscopically later because they are preserved. The apparatus provides the first practical means for field sampling of large volumes of turbid water for the presence of cercariae with a minimum amount of equipment.

This device was used during a study of the seasonal fluctuation of the density of cercariae in a natural stream in Puerto Rico. Five samples of 5 gallons each were taken at the same location on each Tuesday and Thursday from February 4 through December 2, 1965. On the 87 days when 435 samples were taken, the stream was found positive for cercariae on 26 days in 52 positive samples. The cercarial density in these positive samples ranged from 0.2 to 29.0 cercariae per gallon. Complete data from this field study will be reported subsequently.

#### Summary

A new device based on the continuous-flow centrifuge technique has been used successfully to recover *Schistosoma mansoni* cercariae from both clear and turbid natural waters. A 5gallon sample of water can be put through the device in 20 to 30 minutes. While the head of the centrifuge is still spinning, addition of 10 percent formalin with picric acid kills, preserves, and stains the cercariae for subsequent counting.

#### REFERENCES

- Klock, J. W.: A method for the direct quantitative recovery of *Schistosoma mansoni* cercariae from natural waters of Puerto Rico. Bull WHO 25: 738-740 (1961).
- Rowan, W. B.: A simple device for determining the population density of *Schistosoma mansoni* cercariae in infected waters. J Parasit 43: 696-697 (1957).
- (3) Rowan, W. B.: Daily periodicity of Schistosoma mansoni cercariae in Puerto Rican waters. Amer J Trop Med 7: 374–381 (1958).
- (4) Rowan, W. B., and Gram, A. L., Jr.: Relation of water velocity to Schistosoma mansoni infection in mice. Amer J Trop Med 8: 630-634 (1959).

## **Measles Film**

"Spot Prevention," a film to support and promote measles immunization, has been produced by the Public Health Service Audiovisual Facility. This fast-moving, humorous film, showing the chase and capture of measles "germ" and his "conversion" to protective vaccine, is animated, with a live introduction and close. Geared toward children from preschool to second grade but enjoyable to adults, the film is par-



ticularly adaptable for use by schools, churches, civic organizations, and television.

The 16-mm. color film, with sound, runs 13½ minutes. It is available for free short-term loans from the Public Health Service Audiovisual Facility, Atlanta, Ga. 30333, Attn: Distribution Unit. It can be purchased from Du Art Film Laboratories, Inc., 245 West 55th Street, New York 10019.