A MORPHOLOGICAL AND EXPERIMENTAL STUDY OF
SOME NEW DERMATITIS PRODUCING CERCARIAE
FROM CULTUS LAKE, B.C.

by
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ABSTRACT

Previous workers at Cultus Lake discovered a new species of Schistosome cercaria. In the present attempts to find this cercaria and to elucidate its life cycle, four completely different dermatitis-producing forms were discovered, two of which are considered new species. The other two may be new. Three of the cercariae are described in detail. These are the cercaria of *Trichobilharzia adamsi* n.sp., *Cercaria chilliwackensis* n.sp. and *Cercaria 10* which may be a new species. The description includes morphology, behavior, snail hosts, dermatitis-producing ability, and observations on the sporocysts of these forms. All were found to be capable of producing Schistosome dermatitis. Therefore two and possibly three more cercariae have been added to the list of known dermatitis-producers. The three cercariae were found in specific areas along the north-west shoreline of Cultus Lake.

Attempts were made to complete the life cycle of the three cercariae. Two approaches were adopted. Animals were trapped within the areas at which the cercariae were found, but examination showed that they did not possess adult Schistosomes. Laboratory experiments were carried out in which animals were subjected to the various cercariae and later examined for adult worms. An immature female worm was recovered from a domestic duck which had been exposed to cercariae. The worm, *Trichobilharzia adamsi*, is described as a new species. Part of the life cycle of this trematode is described. The domestic duck was established as a definitive host under experimental conditions. The specific definitive host of the trematode at Cultus Lake was not determined.

The cercaria of *T. adamsi* is placed in the "ocellata
group" on the basis of its morphology. *Cercaria chilli-wackensis* is placed along with *Cercaria douthitti* in the "douthitti group*. *Cercaria 10* is very similar to the cercaria of *Gigantobilharzia huronensis*, and is placed with the latter in the "spindalis group". All three of the cercariae described possess papillar spination. These spines have been described only once in the past as occurring on a dermatitis-producing cercaria. The papillar spination is described in detail. The spines are located in similar patterns on the bodies of the three cercariae. This pattern is carried through to the adult of *T. adamsi*. Evidence is given of the occurrence of papillar spination on cercariae elsewhere in the Province.

A modification of the perfusion technique of Yolles et al is described. This was used for the investigation of the experimental animals for adult worms. Further, a new technique using the side-arm flask of McMullen and Beaver is described. The apparatus concentrates Schistosome cercariae in large numbers in a relatively small area.
ACKNOWLEDGEMENTS

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INTRODUCTION

The dermal reaction to the penetration of certain Schistosome cercariae is known as Schistosome dermatitis. It has been the subject of much investigation in the past. The problem has been studied both from the point of view of Zoology and Medicine.

Schistosome dermatitis, more commonly known as "swimmer's itch", is characterized by an intense itching sensation which occurs from five to fifteen minutes after the skin has been subjected to the cercariae of certain Schistosomes. The irritation gradually subsides over a period of approximately one half hour, after which time minute macules may appear on the area of the skin which was exposed to the cercariae. In approximately twenty-four hours the itching again may become apparent and is often more intense than the first phase. By this time the macules have become distinctly papular and if sufficiently close together may have produced collectively a large erythema. The intensity of the itching sensation is very much increased with light rubbing on the area though initially the rubbing has no great effect. The phenomenon is one of sensitization and though first exposures of the skin to these cercariae result in no dermatitis, subsequent infections produce increasingly severe results.

From a medical standpoint the dermatitis seems important in that secondary infection may arise as the result of scratching of the irritating papules. Further, occasional severe allergic reactions to the foreign proteins of the cercariae occur, which are so intense as to incapac-
itate the victim. There is no evidence that these cercariae are able to mature in a human host, nor have they been demonstrated in the viscera of man. However, Penner (1941) showed that *Cercaria douthitti* may migrate in large numbers to the lungs of the rhesus monkey, and Olivier (1953) also demonstrated that the cercariae of some important dermatitis producers may reach the lungs of monkeys. Future experimentation with mammalian forms may reveal facts of more concern to medical science.

The term "Schistosome dermatitis" was first proposed by Cort (1928). His discovery of the relationship between aquatic snails and the dermatitis, in Michigan, was accidental. While sorting snails in a bucket he contracted an intense itch on the arm which was in contact with the water. The cercariae escaping from these snails were experimentally demonstrated to be the causative organisms of the dermatitis. They were supposedly identified as *Cercaria elvae* Miller, but at a later date this cercaria was recognized as having been erroneously identified and was found to have been *C. stagnicolae* Talbot. Cort predicted at this time that in all probability other related forms would be found which would be capable of producing a dermatitis, and that, as a result of this, the dermatitis would probably be found in many parts of the United States. At a later date Cort (1928 b) produced experimental dermatitis with three additional cercariae, *Cercaria douthitti* Cort 1914, a mammalian form, and two other cercariae which were at the time unnamed. In 1930 Vogel in Europe produced dermatitis experimentally with *Cercaria ocellata* La Valette St. George. This author described the phenomenon as "cercarial dermatitis" (cercarien-dermatitis). Cort however, believes that this term is too broad, since only the Schist-
Some cercariae are implicated in this type of dermatitis.

The two innominate cercariae mentioned by Cort in 1928 with reference to his work with experimental dermatitis remained unnamed until 1936 when Talbot described their morphology. They were named *Cercaria stagnicolae* and *Cercaria physellae*, and with *Cercaria elvae* they constitute the most important dermatitis-producers in the United States and Canada today (Cort, 1950). Also in 1936, Cort and Talbot described the behavior patterns of the four American Schistosomes which were known to be dermatitis-producers. The European form, *Cercaria ocellata*, was considered by Cort on the basis of behavior to be distinct from *C. elvae*.

In summary then, at the end of 1936 there were in all five species of Schistosome cercariae known to science which were capable of producing a dermatitis in man. However, some workers (Szidat and Wigand) at this time considered another cercaria, *C. pseudocellata*, as being very similar to but not synonymous with *C. ocellata*. Since that time a number of authors have insisted that *C. ocellata* is a complex and in reality consists of the following forms based on differences in size: *C. pseudocellata* Szidat and Wigand, *C. neocellata* Szidat and *C. parocellata* Szidat. The validity of these species is not unanimously agreed upon. However, Cort (1950) lists these cercariae tentatively, along with *C. ocellata*, as being distinct species. The questionable validity of size as a criterion of species differentiation has been emphasized by Neuhaus (1952) whose experimental work strongly supports his views.

In 1938 Buckley reported a dermatitis from Malaya called "sawah itch" and ascribed it to the cercaria of *Schistosoma spindalis* Montgomery 1906.
The number of known dermatitis-producing cercariae has been steadily increasing during the past thirty years. Cort's 1950 publication on the status of the subject listed all of the cercariae which were known and suspected at that time to be dermatitis-producers. Eighteen cercariae were included. Of these Cort states that many may be synonyms and the listing is tentative. Much work is necessary on life-cycle studies before the position of many of these cercariae can be definitely established. Of the forms listed only five are definitely described as being distinct species. The validity of the others is questionable. Since the publication of Cort's paper at least six more dermatitis-producing cercariae which are considered new, exclusive of those described in this work, have been found. Of these, two are found in marine snails. Najim (1951) worked out the life-cycle of *Gigantobilharzia huronensis* and described the cercaria as a dermatitis-producer. Simmons, Martin and Wagner (1951) reported that they had found two new dermatitis-producing cercariae in southern California, but no description of these was given. Wu (1953) has described the dermatitis-producing cercaria of *Trichobilharzia cameroni* and she has also elucidated the life-cycle of this form.

**Geographical Distribution of Schistosome Dermatitis.**

As interest in the problem of Schistosome dermatitis increased following Cort's initial discovery of the biological relationships involved, other workers far removed from the focus in Michigan began to undertake investigations of the problem.
Schistosome dermatitis has been reported from almost all regions of the world. Cort(1936 a) described the contemporary distribution of the dermatitis in Europe in 1936. A dermatitis coinciding with the type found in the United States was reported from Wales (where C. ocellata was definitely implicated). Other reports came from East Prussia, Switzerland, the Holstein Lake region south of Kiel, and France. Further, a dermatitis was reported from a lake in Haiti which was almost certainly caused by Schistosomes.

Cort, in his 1950 paper on the status of Schistosome dermatitis, summed up the distribution as was known at that time. From Africa there have been no confirmed reports of this dermatitis, but Porter(1938) found C. ocellata in the Orange Free State. It seems probable that the dermatitis exists in some regions on that continent. In Asia, Schistosome dermatitis exists as "sawah itch" in the Malay States and it has been reported as "kabure" from Japan. Two other unconfirmed reports indicate the presence of the dermatitis in India and Burma. It has been reported also from Australia and New Zealand. In the Western Hemisphere the dermatitis has been confirmed in North America only. However, there are reports of a similar dermatitis from Brazil. In Cuba it is known as "El Caribe". However, the position of Schistosome cercariae as the causative agents has not been definitely established. There have been unpublished reports of a similar dermatitis from Mexico. The author recently received a report of a dermatitis from Fairbanks, Alaska. Although it is not known for certain, this is probably caused by Schistosome cercariae.

At the time of Cort's publication in 1950 Schistosome dermatitis was also known from Minnesota, Wisconsin,
Iowa, North Dakota and other American States as far south as Florida. Hunter et al (1949) gave an account of the dermatitis found within the city limits of Seattle, Washington. Recently, Macy (1952) summarized our knowledge of dermatitis-producers in the States of the U.S. west coast.

Cort declares that Canada possesses the largest endemic areas of Schistosome dermatitis in the world. In 1936 he reported that Saunders found dermatitis near Saskatoon, Saskatchewan, and attributed it to *C. stagnicolae*. Saunders also reported that he had obtained cercariae of the "elvae group" from Paul Lake, B.C., and that he possessed substantiated reports of dermatitis in the Peace River district. He also claimed unconfirmed reports from Vancouver Island. McLeod (1940) and McLeod and Little (1942) reported that *Cercaria elvae*, *C. stagnicolae*, *C. physellae* and *C. dermolestes* were all responsible for local outbreaks of the dermatitis at Clear Lake, Manitoba. Wu (1953) published the lifecycle of *Trichobilharzia cameroni*, the cercariae of which were responsible for dermatitis outbreaks in the Province of Quebec.

**Background of the Problem:**

The frequency of reports of a high incidence of Schistosome dermatitis at Cultus Lake during the warm summer months led to the establishment of a research grant by the Dominion Department of Public Health, thereby enabling investigations to be carried out on a full-time basis. Work in 1951, under the direction of Dr. J. R. Adams, resulted in the collection of large numbers of snails from the resort area of Cultus Lake, by S. M. Sager and M. Thom. In the labor-
atory some of these snails gave off *Cercaria physellae* and *C. stagnicolae*, both dermatitis-producers. In addition to these forms another cercaria, a dermatitis-producer also, was found and was considered tentatively to be a new species. This cercaria, which possessed five pairs of flame cells in the body, was characterized mainly by the presence of distinctive and unique "papillar spines" on the body. These structures have previously been described only once on a dermatitis-producing Schistosome cercaria. On this basis and upon consideration of other morphological differences between the local form and other cercariae, the former was considered new to science.

During the summer of 1952 the survey work was taken over by the author. However, at this time attention was turned away from Cultus Lake in order to carry out a comprehensive survey of the Schistosome dermatitis problem in the Okanagan Valley. As a result further studies of the situation at Cultus Lake were not made until the completion of the Okanagan survey in the Fall of 1952.

With the knowledge of the probable existence of a new species of cercaria at Cultus Lake in mind the author began the investigation with the intention of working out if possible the life-cycle of this form. Attempts were made to trap mammals and to examine them for the presence of adult Schistosomes with the hope that the adult worm of the unknown cercaria would be discovered. All animals so examined were found to be negative in this respect.

With the coming of June, 1953, snails were being continuously collected and their larval trematode infections
studied. The situation was carefully watched for the appearance of the new cercaria. As the summer proceeded through July the cercariae described in this work appeared, but no cercaria was confirmed as being the previously described new form. Instead, more than one cercaria was found which possessed papillar spination, the characteristic which had been described as unique for the former unknown cercaria. Of these, four were confirmed to be capable of producing Schistosome dermatitis, and therefore each was considered to be potentially the unknown cercaria which had previously been described. Later comparisons seemed to indicate, however, that these four cercariae had not been seen before at Cultus Lake.

A discussion of the possible reasons for the failure to discover the original cercaria is given toward the end of the present work.
METHODS AND APPARATUS.

This study of the three Cultus Lake trematodes was divided into two categories:

(1) Morphological and host-relationship studies were made of the cercariae and sporocysts.

(2) Attempts were made to work out the life-cycle of the three Cultus Lake cercariae.

Methods of Study of the Larval Forms:

A—Collection and Culturing of Snail Intermediate Hosts:

All snail collections were made within a restricted area along the north-west shore of Cultus Lake (Plate IX). Due to the nature of the undergrowth along the shoreline in this region it was necessary to proceed from one point to another by means of a small boat. Examination of the beach for snails was greatly hindered by the presence in the shallow water of large numbers of dead trees and tangles of dead undergrowth. In many instances snails were found clinging to this submerged wood and in some areas they seemed to prefer this to the small rocks constituting the bottom.

During the early portion of the summer the surface of the lake was usually sufficiently calm to allow clear vision of the bottom in the shallow water. However, later in the summer the lake surface on many occasions was roughened by a brisk wind, making clear vision of the bottom impossible. This difficulty was overcome by the construction of a glass-bottomed viewer with a window $9\frac{5}{8} \times 6\frac{3}{4}$ inches. This gave a range of vision in two feet of water of approximately twice those dimensions. Admittedly, fewer snails
could be collected in this manner than could be collected when the surface was calm due to the limited degree of vision through the window.

The snails were collected into quart sealers with a maximum of 50 or 60 in each jar. They were safely kept in this concentration until the following day. The temperature of the lake water at the shore line was always taken with each collection of snails.

In the laboratory the snails were washed thoroughly in tap water and put into quart sealers in the concentration of five to six per jar. The sealers were half filled with tap water which had been allowed to stand approximately three or four days. It was found that water direct from the tap could not be used since it killed the snails within a few days. Some experiments were undertaken to determine the reason for this. It was learned from the authorities that chlorine is added to the city water in the form of a chloramine and this would not dissipate upon standing. Therefore some other explanation had to be found to explain why the tap water became tolerable to the snails upon standing. The results of the experiments showed that very probably the critical factor was one of pH and that the tap water was too acidic for snail survival. When the water was allowed to stand, the pH increased slightly to a neutral point. The role of the pH in affecting snails was verified by the addition of basic salts to water obtained directly from the tap in order to increase the pH to a point between 7 and 8. Juveniles, which are more susceptible to pH variations than adults, were kept successfully in water so treated.
In the past, collections of snails have been fed by adding lettuce to the jars (Sager, 1950). However, it was discovered that the lettuce caused the water in the jar to become quite murky after a period of a few days. Hence a substitute was found which proved to be much more practicable. *Stelaria media*, the common chick-weed, was used after it had been washed thoroughly in tap water. A small amount of this plant was added to each sealer and the snails fed quite readily upon it. While decaying in the jar these leaves do not increase the turbidity of the water.

Snails were maintained in the laboratory for a period of several weeks in the above manner. In June a number of juvenile snails was collected from the same area of Cultus Lake. These were kept in the laboratory in gallon jars and also in a five gallon aquarium. Most of these snails survived for a period of three months.

It must be noted that if the snails were kept in the laboratory for a period exceeding one week, the water was buffered every few days by adding to the jars and aquaria a small amount (approximately one-eighth teaspoon) of powdered limestone. A quantity of powdered limestone was also added to the battery-jars in which the tap water was allowed to stand.

The jars of snails were examined several times each day with a hand lens to determine whether cercariae were emerging. If cercariae were seen, the snails were taken from the jar and washed thoroughly in tap water to remove any possibility of the trematodes clinging to the shells. The snails were then isolated by placing each one
in a 1 x 2 inch vial with tap water which had been standing for at least four days. The cercariae seen initially in the quart sealer would be found later within one or more of the vials. The snail responsible for them was thus singled out. Emergence of the cercariae could be accelerated by changing the water in the vial or by shining a strong light into it.

When the snails giving off the cercariae were isolated, each was placed in a larger container with a small amount of chick-weed. These were then placed in a shallow trough which contained running water, to lower the temperature of the snail's water. The lower temperature both retarded decomposition of the chick-weed and slowed down the emergence of the cercariae until they were needed for examination and life-cycle experiments.

B-Methods of Studying the Cercariae:

All cercariae were studied after natural emergence from the snail host. On some occasions it was found that cercarial emergence could be induced by changing the water in the jar. This phenomenon was reported by Taylor and Baylis in 1930 (Cort and Talbot, 1936) with reference to the emergence of *Cercaria ocellata*. Further, it was discovered that emergence could be effected by shining a beam of light through the jar containing the snail host. In one instance, the number of *Cercaria chilliwackensis* present in the snail jar at mid-afternoon, after having remained constant for three hours, was increased from about six to approximately three dozen within a half hour by directing the beam of a microscope lamp through the jar. It was not determined in this instance whether the emergence was the result of a
light intensity increase or merely the result of a possible rise in temperature of the water in the jar. The effect of light upon the emergence of cercariae was studied by Olivier (1951) at which time he noted a definite correlation between cercarial emergence and the hours of daylight for the cercariae of *Schistosomatium douthitti*.

The cercariae were studied and described in detail. They were examined with various powers of the microscope in order to ascertain the morphological details and to obtain measurements. Examination under oil-immersion was necessary in order to observe the organ systems and to locate and describe, particularly, the papillar spination. Certain details of the behavior of these cercariae could be determined by microscopic examination. Other details such as proportional appearances of the various parts of the cercariae were observed. After studying these larvae a number of times sufficient experience was gained to allow recognition of them upon low magnification.

The cercariae were also studied from the point of view of their free-swimming behavior. The importance of swimming behavior as a specific characteristic was established by Talbot (1936). A vial containing fair numbers of the Cultus Lake cercariae was examined in a beam of light in a dark room to ascertain the degree of reaction, if any, to light. Their reaction to gravity was also noted, as well as the body position while resting. The behavior of the cercariae was observed while they were under a cover-glass in a wet mount preparation. The taxes of the cercariae and their characteristic swimming behavior were most clearly studied by utilization of the side-arm flask "concentrator"
described below. By this means fairly large numbers of the cercariae could be easily studied "en masse". As with the case of the morphology of the cercariae familiarity resulted in the ability to recognize one or two of these species by means of their swimming behavior.

The dermatitis-producing abilities of these cercariae were studied. For a detailed analysis of the techniques used see under "Schistosome dermatitis Experiments".

Cercariae were preserved by adding to the water an equal amount of hot 30% formalin.

Methods of Studying the Sporocysts:

When an infected snail died it was dissected as soon as possible under the binocular microscope. The sporocysts were measured and photographed under high magnification with a monocular microscope. In two instances the snail was dissected "in vivo" in order to observe the sporocysts and cercariae in the live state. Cercariae were thus observed emerging from the sporocysts. Before live sporocysts could be measured or photographed they had to be killed to stop all motion. This was effected by touching to the sporocyst mass a drop of 30% formalin on the end of a dissecting needle.

It was found more expedient to photograph the sporocysts than to draw them. In the snail tissue they were always found in a tangled mass and it was very difficult to trace any one sporocyst along its length for any appreciable distance.
Methods Used In Life-cycle Studies:

In attempting to solve the life-cycle of the Cultus Lake trematodes the cycle was attacked from two different points. Therefore these developmental studies may be considered under two headings:

1. Investigations of animals in the field which could possibly act as the definitive host.
2. The carrying out of infection experiments in the laboratory in which animals were subjected to the cercariae.

In the first instance the possible host is known while the potential parasite is unknown. In the second case the reverse condition exists, the parasite being the known factor.

A-Trapping:

Since the unknown, 1951 cercaria possessed five pairs of flame cells in the body it was similar in this respect to Cercaria douthitti Cort 1914. Price (1931) worked out the life-cycle of C. douthitti in small mammals, i.e. mice. Schistosomatium douthitti became the only dermatitis producer which was known to occur in mammals. It was therefore considered likely that the Cultus Lake cercaria would be found to undergo development in small mammals and traps were set out for these. The first series of traps were set out during the fall of 1952 and the spring of 1953. The area covered was named by the author the "beach" area for the sake of differentiation (Plate K). All traps used were mouse-traps, with the exception of a group of larger spring-traps used to catch Mink. The mouse-traps were baited with walnuts. The larger spring-traps were arranged in a small
circle with a dead fish suspended above the center of the circle approximately three feet from the ground. All traps were set within a few yards of the shoreline, except in one instance. In the latter case the traps were extended to approximately 30 feet from shore in an attempt to vary the yield.

The trapping attempts outlined above yielded 49 small mammals. These included 35 *Peromyscus* spp., 2 *Micromus* spp., 11 *Sorex* spp. and one Mink. The animals were examined in the laboratory for adult Schistosomes. All were found to be negative in this respect. Two methods of examination were adopted:

1. The intestinal mesenteries were stretched under a binocular microscope and examined; the liver was macerated and examined.

2. The faeces or intestinal contents were examined for ova by using the side-arm flask technique.

During the summer months of 1953 the author learned that the Schistosome cercariae which possess papillar spination occured in snails located farther southward along the beach (Plate X). None whatever emerged from snails collected from the "beach" area. In view of this, traps were set on August 31, 1953, in Area B from which the greatest number of papillated cercariae were recovered. The traps yielded only 4 *Peromyscus* and 2 *Sorex* spp. Examination proved these to be devoid of adult Schistosomes.

No adult Schistosomes were found in the animals caught in the field, and as a result the natural host of the parasites could not be established.
Infection experiments were greatly hampered by a lack of material. This was due to the extremely small percentage of snails infected with the cercariae in question. Methods had to be adopted which would increase the number of cercariae in the water used for infection experiments.

**Side-arm Flask Concentrator:**

Initial attempts to infect white mice with cercariae were made by holding the mouse manually, with the tail immersed in a vial of water containing the organisms. However, in this position the mouse was usually very active and disturbed the water in the vial with movements of the hind feet and tail, as well as almost invariably defecating into the vial. The animal could not be maintained in this position for long periods of time. Olivier and Stirewalt (1952) outlined a system for restraining mice for infection through the tail. In the present instance however, a method was sought which would not only restrain the movements of the mouse, but which at the same time would allow for a greater concentration of cercariae to be present near the exposed area.

Concentration of the cercariae was effected as follows. A 500 ml. side-arm flask (Plate 1), such as is used in the McMullen and Beaver technique, was first filled with water to the level D. Snails were then put into it which were giving off the same species of cercaria. A cork E was pushed into the mouth of the flask to make it air-
PLATE 1

A-flask
B-snails
C-paper bag, double
D-first water level
E-cork, air tight
F-second water level
G-light source
H-cardboard tube
I-cork plunger
J-laboratory mouse
K-clamp
L-clamp stand
M-tail-clamp, padded
APPARATUS FOR EXPOSURE OF MICE TO CERCARIAE

PLATE 1
tight. The level of water in the flask was then raised to the second level \( F \) by adding more water through the side-arm. The entire flask was enclosed under a paper bag of double thickness with a hole in it to allow the side-arm to protrude through. A strong lamp was shone into the arm. The apparatus would now utilize the characteristic negative geotaxis and positive phototaxis of the cercariae emerging from the snails. These became concentrated in the side-arm.

Mice were subjected to the cercariae by the following method. The mouse was put into a cardboard tube with a small hole at the bottom allowing the tail to protrude through. A cork plunger was then pushed through the tube on top of the mouse to such a point that the animal became sufficiently confined to restrain its movements. Movements of the tail were reduced to a minimum by applying a small clamp \( M \) to the base of the tail. The clamp was padded on the inside with absorbent cotton. The mouse and tube were held in place above the side-arm by means of a clamp stand, and the tail of the animal was placed within the side-arm of the flask.

This apparatus allowed longer periods of motionless exposure of the mouse to the cercariae and the apparatus could be left for periods of time without attention. Another obvious advantage of this technique lies in the fact that the cercariae become concentrated in the region of intended exposure of the mouse, due to the taxes of the former.

The apparatus may be used very efficiently for separating double infections (i.e., Schistosomes and Strigeids) where the degree of response to light differs between the two types of cercariae.
Animals Used In Experiments:

All animals used were known to be free of Schistosomiasis infections. They had been raised under known conditions previous to the experiments. After the mice had been subjected to cercariae faecal checks revealed that these animals were parasitized by a tape-worm, *Hymenolepis*. This however, would not affect in any way the validity of the results.

Pekin ducklings ranging in age from one week to one month were exposed by allowing them to stand in water containing the cercariae. This method was adopted by Talbot (1936) in his attempts to infect ducks with *Cercaria elvae*, *C.stagnicola* and *C.physella*. Talbot used a greater amount of water than did the author.

Two pigeons were exposed to cercariae by standing them in shallow water containing the organisms. The water was approximately one and one half inches in depth. Further, one of the pigeons was given a few cercariae by intraperitoneal injection.

White mice were exposed to the cercariae through the tail, as mentioned on page 17.

One Guinea pig was used in the experiments. Cercariae were applied to the inquinal region of the animal and a few were administered orally.
Methods of Examination of Animals:

The animals which had been subjected to cercariae were examined for the presence of adult worms while still living, and when they had been killed for dissection. At various intervals after initial exposure faecal checks were made to determine whether Schistosome ova were present in the faeces. This was done by using the side-arm flask technique (McMullen and Beaver, 1945) or by adding some faeces to tap water in a vial. Occasionally the DCF method of flotation was adopted, as outlined by Chandler ("Introduction to Parasitology"). Examinations of faecal smears were also made. The faecal material was collected by allowing the animal to defecate in a large glass jar.

All animals were killed with intraperitoneal Nembutal. The optimal dosage for the mice was found to be .1 ml. of Veterinary Nembutal. The amount given to the ducklings increased in proportion to their weight. In one case a duck weighing approximately twelve pounds was given eight mls. before it succumbed.

Experimental animals were dissected and examined for adult Schistosomes. Price (1931) dissected adults of Schistosomatium douthittii from the hepatic portal veins of mice. McLeod and Little (1942) outlined a method of dissecting adult Schistosomes from the serosa of the gut. Ova of S. pathlocopticum were found in the liver of infected mice by Tanabe (1923). These three sites are implicated in adult Schistosome infections. Therefore these regions were examined in the animals which had been subjected to the cercariae in the present experiments. Livers of animals other than mice
were examined by comminuting them in citrated saline. The extricated liver was cut into two or three large pieces and each was put into a Petri dish containing saline. Here, the larger piece was teased apart with dissecting needles. The mass thus formed was allowed to stand for approximately one hour. Each piece of tissue was then removed with forceps and rinsed in another dish containing citrated saline. The initial dishes plus the rinse dish were examined under a binocular microscope for adult worms. This technique revealed the one adult Schistosome described in this work.

The livers of the mice were perfused using a modification of the Yolles technique (1947). In all cases the liver only was perfused. It was very difficult to remove the liver of the mouse while leaving the aorta completely intact. Therefore, the liver was ligatured off, extracted and perfused, while examination of the intestinal mesenteries and serosa was carried out later. Much more experience with this technique would be necessary in order to effect both a successful perfusion of the liver and the intestinal blood vessels.

Since there existed no source of air-pressure in the laboratory the necessary pressure was supplied by means of a hand-bulb. A manometer was constructed which was connected to the perfusion apparatus to give an indication of the amount of pressure within the apparatus during perfusion and also to effect a constant pressure for all experiments. At first it was found necessary for a second person to use the hand-bulb to maintain the pressure. However as experience was gained it was found that this operation could be performed by the author while doing the perfusing. This
was done by the following method. The ligature on the vena cava behind the diaphragm was left long and when the liver was removed the latter was partially suspended by fastening the free end of the thread in a clamp on a stand. This allowed the left hand to be free to pump the bulb while the right hand inserted the needle into the hepatic sinus just below the point of attachment of the long thread. The system worked admirably and in a very short time the liver turned creamy white. The flow through the needle was regulated by squeezing the rubber hose with the right hand and by observing the liquid level within the manometer.

Ligaturing of the blood vessels proved difficult at first since these are very small and they may easily be torn with forceps or probe. This problem was overcome by using an ordinary darning needle of medium size. Two threads (for double ligatures) approximately one foot long were put through the eye of the needle and this was passed beneath the blood vessel to be ligatured, while the latter was held suspended with a blunt probe. After the threads were pulled through about half way the needle was removed and the threads were tied as far apart as possible. The vessel was then cut between the ligatures.

It was found that the liver could be partially suspended by a very slight pull upon the ligatures on the portal vein and vena cava (anterior to the diaphragm). This was useful in removing the liver from the body cavity and for general manipulation of it. The method was used since it was found that the diaphragm often tore too readily when it was used for manipulating the liver.
The intestinal mesenteries and serosa of all animals were examined by removing the intestine and viewing its surface with a binocular microscope. The mesenteries could be clearly examined by stretching them from the intestine with dissecting needles.
MORPHOLOGY OF THE CULTUS LAKE TREMATODES

TRICHOBILHARZIA ADAMSI

CERCARIA (Plate 111)

Morphology:

The cercaria of *Trichobilharzia adamsi* is an aphanyngeal, furcocercous, distome and resembles very closely the cercariae of the "ocellata group". Measurements were made of 25 cercariae which were fixed in 15% formalin. The Means are given with the Standard error: body length .2301 ± .0069 mm; body width .0802 ± .0014 mm; tail-stem length .3413 ± .0023 mm; tail-stem width, proximal .0438 ± .00082 mm, distal .0371 ± .002 mm; furcal length .2007 ± .0054 mm; furcal papilla length .0173 ± .00077 mm; oral sucker width .0515 ± .00071 mm; ventral sucker diameter (for 20 specimens) .0372 ± .00062 mm; distance from center of ventral sucker to posterior end of body .0966 ± .059 mm.

The tail-stem of the cercaria is slightly longer than the body. The furcae are greater than half the length of the tail-stem. Dorso-ventral fin-folds are present on the furcae.

The entire body, tail-stem and furcae are uniformly covered with minute retrorse spines. On the body these are most obvious on the anterior half and they seem to diminish in size as one approaches the posterior end of the body, making them difficult to observe. Those spines on the tail-stem and furcae are spaced farther apart and appear
somewhat more coarse or sturdy.

Papillar spines are present on the cuticle of this cercaria. On each side, a spine is located about midway between the most anterior point of the body and the junction of the oral organ with the body cuticle laterally; two are located fairly close together immediately posterior to the above junction; one is situated slightly anterior to the level of the eyespot. In addition to these some papillar spines were seen at the level of the ventral sucker. In this region two were seen on the left side of the cercaria and one in the corresponding position on the opposite side of the body. It is probable that these more posterior papillar spines are arranged bilaterally symmetrically and that some of them were not seen at the time of observation. The papillae located in the region of the ventral sucker are much less distinct than those near the oral sucker. In some instances the minute hair located on the papilla nearest the eyespot and on the two papillae near the junction of the oral organ and lateral cuticle were very long and distinct. Of especial importance is the fact that the groups of papillar spines are not all in the same plane of focus.

Two heavily pigmented eyespots are located approximately one third of the distance from the oral to the ventral sucker. The eyespots possess a lens which is directed laterad.

The body contains five pairs of penetration glands. The two anterior pair are more granular in appearance than those farther posterior. They are arranged with the first pair anterior to the ventral sucker and the second pair
postero-lateral to it. Farther posteriorly the remaining three pair of glands are arranged somewhat linearly on each side of the body. The ducts of the penetration glands were seen in cross-section at a flexure within the oral sucker and here five individual ducts were distinguished. No papillae were seen at the point of exit of the ducts through the anterior cuticle, although these may have been present.

An oesophagus beginning within the oral sucker at a point somewhat posterior to the anterior end runs back between the anterior pair of penetration glands, bifurcating behind the pair of eyespots. The caeca were obscured by the penetration glands and their termination was not observed.

The excretory system consists of six pairs of flame cells within the body and one pair in the proximal portion of the tail-stem. All lead into a system of collecting tubules. On both sides of the body, there is one flame cell on each side of the eyespot and one other between the eyespot and the bifurcation of the main excretory collecting tubule. Three flame cells are located posterior to the above bifurcation on each side of the body. Thus the flame cell pattern of this cercaria is 2(3+/3+1/). One departure from the above pattern of this cercaria was noted. In this instance the flame cell located immediately posterior to the eyespot in other cercariae was seen to be adjacent to flame cell number one, and its junction with the anterior tubule was also anterior to the eyespot. This situation existed on one side of the cercaria only. No reason can be given for this departure from the apparently normal condition and it must therefore be considered an anomaly.
Najim (1951) observed similar anomalies in the excretory system of the cercaria of *Gigantobilharzia huronensis*. The main collecting tubule runs into an excretory bladder posteriorly. Two ciliated patches are located near the bifurcation of the main collecting tubule on each side of the body. One of these is situated in the main collecting tubule near the point of bifurcation. The position of the other patch is not entirely certain. It is located either in the proximal portion of the posterior lateral excretory tubule or in the main collecting tubule. An excretory tube runs from the bladder medially down the length of the tail-stem and bifurcates. One branch passes through each furca, opening terminally through an excretory papilla. No Island of Cort could be seen near the excretory bladder. The bladder was very obscure in outline and could not be observed clearly.

**Behavior:**

The cercaria of *T. adamsi* is positively phototactic and the cercariae collect quite rapidly in the side-arm of the "concentrator". Some of them attach by the ventral sucker to the side of the glass on the light side. In this position they form a U shape. Others remain free swimming on this side. The cercariae do not congregate at the surface film but are usually found in all parts of a vial containing them. When resting the furcae are usually held together. Under a cover-glass the cercariae are rarely observed lying in a dorso-ventral position due to the degree of protrusion of the ventral sucker. They sometimes hitch along under a cover-glass by alternate attachment of the ventral and oral suckers.
Dermatitis-Producing Ability:

The cercaria of *T. adamsi* is a dermatitis-producer, as determined by experiment. This section is discussed more fully under "Schistosome dermatitis Experiments".

Snail Host and Percent Infection:

The cercaria emerged from a snail identified as *Physa c.f. coniformis*. This snail was collected from Area B, C16, on August 3, 1953, and was the only snail collected from which the cercariae were obtained. On the above date 25 *Physa* were taken. Since the cercariae were obtained only this once the percent infection must be based only upon this collection. The infection percentage can be considered only during the period of time at which the cercariae are emerging--snails infected were determined only by cercarial emergence. Since, in the case of the cercaria of *T. adamsi*, the only period of emergence which we can be sure of is the date which the infected snail was collected, the percent infection must be calculated upon the basis of the number of snails collected at that time. The possibility exists that the cercaria was emerging during other visits to this area and was missed in the collections. This cannot be known for certain, however. The percent infection, since one of the 25 *Physa* gave off cercariae, is 4%.

Comparison With Known Forms:

With the increased recognition of Schistosome dermatitis in the last few decades it is doubtful if a Schistosome cercaria has been described without an investi-
gation into its ability to produce a dermatitis. It becomes necessary then to compare the cercaria of \textit{T. adamsi} with other dermatitis-producers for the possibility of synonymity. Further, this cercaria need only be compared with others possessing the same flame cell pattern—that is, six pairs of flame cells in the body and one pair in the tail-stem.

Cort (1950) states that the chief reason for separation of \textit{Cercaria dermolestes} from \textit{C. stagnicolae} is that the snail host of the former is \textit{Stagnicola palustris}, while the host of \textit{C. stagnicolae} is \textit{S. emarginata}. This is a separation based on differences between hosts of the same genus \textit{Stagnicola}. A high degree of host specificity is usually found in the Schistosomes. It is reasonable therefore, to conclude that cercariae emerging from snails of different families need not be compared critically for the possibility of synonymity. All dermatitis-producers known to date parasitize snails of one family only. On this basis comparison will be made between the cercaria of \textit{T. adamsi} and other dermatitis-producers which parasitize snails of the family \textit{Physidae}.

\textit{Cercaria oregonensis} MacFarlane and Macy (1946) is a dermatitis-producer which is found in \textit{Physa} sp. In the past a certain amount of doubt has existed as to whether \textit{C. oregonensis} is actually distinct enough from other cercariae of the "ocellata group" to be considered a separate species. However, Macy and Moore (1953) claim to have established the adult of this cercaria and they give data which seems to support the view that the cercaria is a separate species. The measurements of the cercaria of \textit{T. adamsi} do not come close to the measurements of \textit{C. oregonensis}. The
two anterior pairs of penetration glands of the latter are located well in front of the ventral sucker, whereas these glands in our cercaria are distributed about the ventral sucker. Differences in the positions of the excretory structures of the two cercariae exist, the most anterior flame cell of *C. oregonensis* lying medio-posteriorly to the eye-spot. Further, the bifurcation of the main collecting tubule in the latter does not coincide with that of our cercaria.

The cercaria of *Trichobilharzia cameroni* Wu(1953) very closely resembles that of *T. adamsi*. However, a significant difference exists between the two forms in the ratio of the body length to the tail-stem length. In the former this ratio is .86, while in the Cultus Lake cercaria the ratio of the body length to the tail-stem length is .67. The most obvious points of difference between these two species are seen upon comparison of the adults.

*Cercaria physellae* is very similar to the cercaria of *T. adamsi* morphologically and the snail hosts belong to the same family, i.e. the *Physidae*. Differences in the measurements of these two forms exist and these are given in Table 1. It is easier to compare the adults of these two species since the cercariae are so similar in every respect.

Comparisons of the cercaria of *T. adamsi* with other forms has not been considered on the basis of the possession by this cercaria of papillar spination. The possibility does exist that this type of spination has been overlooked by other workers in their observations of the cercariae now known.
TABLE 1
Comparison of Measurements of the Cercaria of *T.adamsi* and Cercaria *physellae*.

<table>
<thead>
<tr>
<th>Dimension</th>
<th>Cercaria of <em>T.adamsi</em></th>
<th>C.<em>physellae</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Body length</td>
<td>.2301 ± .0069 mm</td>
<td>.265 ± .0084 mm</td>
</tr>
<tr>
<td>Body width</td>
<td>.0802 ± .0014 mm</td>
<td>.060 ± .0046 mm</td>
</tr>
<tr>
<td>T.S.length</td>
<td>.3413 ± .0023 mm</td>
<td>.374 ± .0106 mm</td>
</tr>
<tr>
<td>T.S.width</td>
<td>.0438 ± .00082 mm</td>
<td>.040 ± .0036 mm</td>
</tr>
<tr>
<td>Furc.length</td>
<td>.2007 ± .0054 mm</td>
<td>.196 ± .0078 mm</td>
</tr>
<tr>
<td>Oral suc.dia.</td>
<td>.0515 ± .00071 mm</td>
<td>.039 ± .0020 mm</td>
</tr>
<tr>
<td>Vent.suc.dia.</td>
<td>.0372 ± .00062 mm</td>
<td>.029 ± .0024 mm</td>
</tr>
</tbody>
</table>

* The measurements of *C.physellae* were taken from the description by Talbot(1936).
Since worm eggs were never found in faecal examinations of the duck it is not possible at this time to described the morphology of the ovum or miracidium larva of *T. adamsi*. The lack of eggs in the faeces of the duck was in all probability due to the worm being too immature at that particular time. On the basis of this reasoning the companion duck, subjected to the same cercariae at the same time, was allowed to live for an additional 30 days with the hope that any worms which might be present within it would mature. However, as previously mentioned, this duck was later found to be uninfected.

**SPOROCYSTS** (Plate 1V)

On the morning of August 17 the snail infected with the cercaria of *T. adamsi* was found to be dead. It was dissected immediately. The sporocysts within the snail tissue had not yet reached a state of disintegration which would render them useless for study.

Dissection revealed a fairly heavy infestation of sporocysts within the digestive gland. It was impossible to untangle the sporocysts sufficiently to allow an appreciation of their length and to draw them with any degree of accuracy. A part of the tangle was therefore separated and this was photographed to record some of the details of the sporocysts' macroscopic morphology.

It was possible to measure the width variations
of these sporocysts without having to untangle them. The measurements obtained are given in Table 2. The width of the sporocysts varied from a minimum of .035 mm to a maximum of .175 mm. The large degree of variation is due to the fact that the sporocyst consists for the most part of an alternation of fairly large bulges and extremely narrow portions along its length. Since the sporocysts had been dead for a short time prior to the dissection of the snail the developing cercariae within them could not be seen clearly in outline. It is reasonable to assume that the bulges along the sporocyst's length contained groups of the developing cercariae, the narrow portions being too small for these cercariae to be present within.

### TABLE 2

Measurements of Width of *T. adamsi* Sporocysts.

<table>
<thead>
<tr>
<th>Measurements of Width (mm)</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>.063</td>
<td></td>
</tr>
<tr>
<td>.056</td>
<td>.035-.175 mm</td>
</tr>
<tr>
<td>.154</td>
<td></td>
</tr>
<tr>
<td>.140</td>
<td>Average</td>
</tr>
<tr>
<td>.135</td>
<td></td>
</tr>
<tr>
<td>.140</td>
<td></td>
</tr>
<tr>
<td>.175</td>
<td>.108 mm</td>
</tr>
<tr>
<td>.133</td>
<td></td>
</tr>
<tr>
<td>.168</td>
<td></td>
</tr>
<tr>
<td>.035</td>
<td></td>
</tr>
<tr>
<td>.035</td>
<td></td>
</tr>
<tr>
<td>.147</td>
<td></td>
</tr>
<tr>
<td>.119</td>
<td></td>
</tr>
</tbody>
</table>
The adult of *Trichobilharzia adamsi* was initially suspected as being a parasite of birds, on the basis of the flame cell pattern of the cercaria. The latter possesses six pairs of flame cells in the body, a condition existing in most other dermatitis-producing cercariae found to mature in birds. Possessing the cercaria of *T. adamsi*, experimental attempts were made to raise adult worms in two Pekin ducklings (*Anser domesticus*) aged approximately one month. One was subjected to the cercariae on August 5 and the other on the following day. The birds were made to stand in a jar containing 1.5 cms. of water with approximately one to two dozen cercariae, for a period of 15 to 20 minutes. During this time the ducklings frequently drank the water and splashed about considerably, thus increasing the chance of their becoming infected.

The first duck was sacrificed on September 16, 42 days after exposure to the cercariae. Examination of the visceras revealed one immature adult female Schistosome in the liver. The worm was considered immature since no eggs were present within the uterus and previous faecal examinations had not revealed ova.

Since the one worm found in the first duck was thought to be immature it was considered advisable to allow the remaining duck, subjected to the same cercariae at approximately the same time, to live for a longer period. This would give any worms which might be present a chance to mature. The second duck was sacrificed on October 7, exactly 70 days from the time of exposure to the cercariae.
Examination of this duck failed to reveal any worms in the viscera.

The worm found in the first duck was studied while living. It was then examined after it had been killed with formalin and finally from a permanent mount. For mounting it was stained with Acetic-alum-carmine and counter-stained with Light Green.

**Behavior:**

The worm was discovered in the liver residue one-half to three-quarters of an hour after the liver had been removed from the duck. At this time the worm was quite active, though the duck had been dead for approximately one hour. It did not "hitch along" in the dish but the anterior end performed apparently aimless twisting motions. When brought beneath a cover-glass on a slide the worm did not at any time stretch out into a straight line. This made measurements of the body length very difficult to obtain.

**External Morphology:**

The entire body is distinctly flattened, especially the posterior half. The posterior end of the body is truncate and consists of three lobes, two lateral and the other making up the posterior termination of the body. The worm possesses both an oral sucker and a ventral sucker, the latter of which is at first seen only with difficulty.

Coarse spines cover the anterior portion of the body. These are arranged in lines which run transversely
over the cuticular surface. The anterior coarse spines merge more posteriorly into spines which are less obvious at about the level of the posterior edge of the ventral sucker. The spination is present on the entire body surface and is more easily seen near the posterior region of the body than along the middle.

Minute papillar spines, seen only "in vivo", are located on the lateral cuticle near the extreme anterior end of the worm. A description of these spines is given more fully under a separate heading. Three bilateral pairs were seen, in addition to one spine further posterior than the others which probably possessed an unseen mate on the opposite side. On each side a spine is located approximately mid-way between the most anterior end of the worm and the junction of the oral sucker with the lateral cuticle; two are located fairly close together immediately posterior to the above junction. One spine was seen on the right side a slight distance posterior to the previous pair. This spine is the one which probably was duplicated on the opposite side.

The description of the papillar spination reveals that the relative positions of these spines in the adult is identical with the relative positions of the anterior groups of spines noted on the cercaria.

The ventral sucker has an almost globular appearance and it is probable that the organ is used not as a sucker but as a means of eroding the surrounding tissue for attachment. Very long radially arranged spines are present on the convex surface of the organ.
Internal Structure:

A narrow, somewhat twisted oesophagus leads from the oral sucker posteriorly to a level slightly anterior to the ventral sucker. At this point the oesophagus bifurcates forming two pouch-like tubes laterally and immediately anterior to the ventral sucker. The two tubes, or intestinal caeca, then approach the mid-line again and disappear in the region of the ventral sucker. The posterior portion of the oesophagus and especially the two lateral pouches of the caeca contain masses of yellowish granules which are probably the remains of ingested erythrocytes. Between the ventral sucker and the posterior end of the ovary the intestinal tract is either completely invisible or extremely indistinct. Between the sucker and the anterior end of the ovary there appears a thick twisted tube which is either double or possessing a very large lumen. In the region of the ovary the course of the intestinal tract is in doubt. Posterior to the ovary the intestine continues its course as a single unbranched tube to a point near the flattened posterior end. The intestine undulates in a horizontal plane between the ovary and the posterior end of the body. It could not be determined with any degree of certainty whether the lateral caeca bifurcated anterior to the ventral sucker again unite in the immediate vicinity of the ventral sucker, or whether this union occurs further posteriorly in the region of the ovary. This latter condition exists in the closely similar forms, i.e. T. ocellata, T. stagnicola and T. physellae (McMullen and Beaver, 1945). However, in the area between the ventral sucker and the ovary the thickened tube seen does not appear obviously double. Further, the manner in which the two lateral pouch-like enlargements anterior to the ventral sucker approach the mid-line seems to suggest in its very
abruptness a fusion of these tubes in the immediate vicinity of the ventral sucker. The question must for the moment remain an open one until further evidence is accumulated concerning this point.

The fairly conspicuous ovary is located with its anterior end .175 mm posterior to the edge of the ventral sucker. The ovary is slightly convoluted possessing four and one half loops along its length. No trace could be seen of the continuation of the ovary into an oviduct, nor of a seminal receptical behind the ovary. In fact, no other trace of the genital tract could be discerned. It is probable however, that the oviduct continues somewhat anterior to its point of departure from the ovary.

A vitelline duct is seen superimposed upon the surface of the ovary and extending further posteriorly along the intestinal tract. The anterior and posterior terminations of this duct were quite indistinct. Numerous vitellaria are scattered further posteriorly on either side of the intestinal tract and these continue to a point slightly anterior to the beginning of the posterior lobation of the body.

A particularly outstanding characteristic of this worm is the possession of clusters of yellow, glistening globules in the anterior portion of the body (anterior to the ovary). These globules appear to be very close beneath the surface of the body cuticle and are most concentrated in the region anterior to the ventral sucker. The globules are very similar in appearance to the minute bodies within the intestinal caeca and they possess the same yellow hue.
However, they seem to be more regular in their outline than are the latter.

Another feature of this worm, which does not appear to have been described for any other similar form, is the occurrence "in vivo" seemingly on the surface of the ventral cuticle near the posterior of the body, of longitudinal striations which give the appearance of parallel ducts. These fine lines are extremely difficult to observe. Eleven of them were seen as illustrated in Plate 11.

Comparison of T. adamsi With Known Forms:

Comparison with other forms indicates that the Schistosome now being dealt with is a member of the genus Trichobilharzia, as redescribed by McMullen and Beaver in 1945. A more detailed comparison between this and other members of the Schistosomatidae is given under "Taxonomic Discussion".

The adult Schistosome resembling most closely Trichobilharzia adamsi is T. physellae. In Table 3 the measurements of T. adamsi are compared with those of T. physellae. Since the measurements of T. adamsi were made under different conditions a separate column is provided stating the conditions under which each measurement was obtained.

T. physellae is similar to T. adamsi on the basis of morphology and host relationships. Both of these forms undergo development in a Physa in the intermediate stage of the life-cycle. All members of the "ocellata group" for which the adult has been described undergo development in
Lymnaeid snails excepting T.physellae.

From Table 2 it can be seen that T.adamsi is considerably wider than T.physellae especially in the region near the ventral sucker. This difference is significant in view of the probable immaturity of the one T.adamsi measured. The greater width of T.adamsi is reflected in the width of the oral sucker of this form as compared with the sucker width of T.physellae. Due to the degree of flexure of the body of the Cultus Lake form the body length had to be measured by a summation of the lengths of the flexes. This method is inaccurate but it will give a value within plus or minus ten calibrations, or ± .19 mm at the magnification used for this observation. The body thickness of T.physellae is between one-half and two-thirds of the width. Measurements of the permanent mount of T.adamsi indicate that the thickness is about two-fifths of the width immediately posterior to the ventral sucker. It is probable that the worm was slightly compressed under the cover-glass however, effecting a greater degree of flattening. Nevertheless, the difference in thickness between these two forms is significant. McMullen and Beaver(1945) indicated that in some cases T.physellae appeared to possess "extremely fine spines" which were especially obvious near the middle of the body of the males of this species. The body spines of T.adamsi are readily seen and are especially obvious anterior to the ventral sucker. Further, T.adamsi possesses the papillar spination which is unique and has not been described on any other adult dermatitis-producer. No spherical yellow globules are described as occurring in T.physellae.

The above differences would seem to eliminate the
# TABLE 3
Comparison of Measurements of *T. adamsi* with *T. physellae*

<table>
<thead>
<tr>
<th>Dimension</th>
<th>Condition Under Which Measurement Made of <em>T. adamsi</em></th>
<th>Measurements of <em>T. adamsi</em></th>
<th>Corresponding Measurement for <em>T. physellae</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Body Width:</td>
<td>in vivo</td>
<td>.105 mm</td>
<td>---</td>
</tr>
<tr>
<td>- across v.s.</td>
<td></td>
<td>.093 mm</td>
<td>.04-.08 mm</td>
</tr>
<tr>
<td>- further post.</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Body Length</td>
<td>15% formalin</td>
<td>3.61 mm</td>
<td>up to 4.4 mm</td>
</tr>
<tr>
<td>Ventral Sucker:</td>
<td>15% formalin</td>
<td>.064 mm</td>
<td>.024-.032 mm retracted, up to .04 mm protruding.</td>
</tr>
<tr>
<td>- width</td>
<td></td>
<td>.026 mm</td>
<td></td>
</tr>
<tr>
<td>- length</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Oral Sucker:</td>
<td>15% formalin</td>
<td>.036 mm</td>
<td>.024-.028 mm</td>
</tr>
<tr>
<td>- width</td>
<td></td>
<td>.039 mm</td>
<td>.024-.044 mm</td>
</tr>
<tr>
<td>- length</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Cuticle (Lat.)</td>
<td>15% formalin</td>
<td>.004 mm</td>
<td>---</td>
</tr>
<tr>
<td>Vent. Suc. - Oral</td>
<td>permanent mount</td>
<td>.111 mm</td>
<td>.2-.3 mm extended, .14 contracted <strong>wmm</strong></td>
</tr>
<tr>
<td>Vent. Suc. to Oesoph. Biofurca.</td>
<td>permanent mount</td>
<td>.026 mm</td>
<td>.024-.060 mm</td>
</tr>
<tr>
<td>Ovary to back of Ventral Sucker</td>
<td>permanent mount</td>
<td>.175 mm</td>
<td>.1-.3 mm</td>
</tr>
</tbody>
</table>
possibility of synonymity between these two forms. The cercariae of *T. physellae* differ from those of *T. adamsi*, principally in their measurements. This aspect was discussed under the description of the cercaria of *T. adamsi*.

Recently, another species of *Trichobilharzia* was added to the list of the three previously known dermatitis-producing members of this genus. This was *Trichobilharzia cameroni* Wu(1953). This worm is described as being from .052 to .059 mm in width in the region of the ovary. The measurement is to be contrasted with that of *T. adamsi* which, as indicated in Table 3, measured .0933 mm "in vivo". Wu described the intestinal caeca of *T. cameroni* as reuniting "anterior to the reproductive organs". Although it is not known for certain, this situation seems also to exist in *T. adamsi* and the caeca may actually unite in the region of the ventral sucker. The ventral sucker of *T. cameroni* is described as quite prominent. The organ in *T. adamsi* is not prominent and this region of the worm must be scrutinized fairly carefully before the sucker can be seen. Another point of differentiation between the two worms lies in the fact that the former possesses no body spination. *T. adamsi* possesses body spination and in addition the papillar spines on the anterior region of the body.
PLATE 11

Immature adult female of *Trichobilharzia adamsi*.

--The drawing was made free-hand--
PLATE III

Fig.1-Cercaria of T.adamsi with body twisted to side to show details of excretory system.

Fig.2-Cercaria of T.adamsi, dorso-ventral view. X denotes a papillar spine.

--Drawings made free-hand--
PLATE IV

Photomicrographs of *Trichobilharzia adamsi* sporocysts.
--The pointers should be ignored--

Figs.1 and 2- X 40.
Fig.3- X 100.
CERCARIA 16A

Cercaria 16A is mentioned briefly at this time since it possesses the papillar spination described as characteristic of the other cercariae in this work. Of further interest is the fact that this cercaria occurred in a double infection.

On September 4 Cercaria 16A was discovered emerging from one of the snails, Lymnaea emarginata, in the laboratory. The snail had been collected from Area B (Plate X.) on August 29. The larva appeared to be very similar to the cercaria of Trichobilharzia adamsi on the basis of morphology, especially in the arrangement of the organ systems. So marked was this resemblance that initially this cercaria, now designated as Cercaria 16A, was considered as being synonymous with the other form. Measurements revealed that C.16A possessed a slightly longer tail-stem than that of the other cercaria and this coupled with the fact that the snail host was of a different family than that of the host of the other form led to the conclusion that the cercaria was probably a different type. Further, the cercaria did not attach as readily to glass as did the cercaria of T.adamsi.

Since Cercaria 16A was at first believed to be synonymous with the cercaria of T.adamsi no infection experiments were carried out with this cercaria. The snail host was dissected before it was recognized as constituting a separate species.

Measurements were made of a few cercariae which had been fixed in 15% hot formalin. This was done in order to
effect a rough comparison with those measurements obtained for the cercaria of *T. adamsi*. Comparison of these measurements is given in Table 4. The number in brackets following the measurements of *Cercaria 16A* indicates the number of cercariae measured.

**TABLE 4**

Measurements of *Cercaria 16A* and the C. of *T. adamsi*

<table>
<thead>
<tr>
<th>Dimension</th>
<th>Cercaria 16A</th>
<th>C. of <em>T. adamsi</em></th>
</tr>
</thead>
<tbody>
<tr>
<td>Body length</td>
<td>.273 mm (3)</td>
<td>.230 mm</td>
</tr>
<tr>
<td>Body width</td>
<td>.069 mm (2)</td>
<td>.080 mm</td>
</tr>
<tr>
<td>T.S. length</td>
<td>.446 mm (5)</td>
<td>.341 mm</td>
</tr>
<tr>
<td>T.S. width</td>
<td>.042 mm (4)</td>
<td>.044 mm</td>
</tr>
<tr>
<td>Oral suc. wid.</td>
<td>.051 mm (4)</td>
<td>.052 mm</td>
</tr>
<tr>
<td>Furcal length</td>
<td>.206 mm (5)</td>
<td>.201 mm</td>
</tr>
</tbody>
</table>

Although the number of *Cercaria 16A* measured is not sufficiently great for an adequate specific diagnosis of this cercaria the average measurements given above will serve for comparison. It can be seen from the table that the tail-stem of *Cercaria 16A* is appreciably longer than that of the other cercaria. The other measurements given coincide quite closely and serve to illustrate to some extent the degree of similarity of these two cercariae.

Considering the organ systems, the two cercariae differ to the greatest extent in the degree of flexure of the main collecting tubules near the point of bifurcation laterally. In the cercaria of *T. adamsi* this flexure is slightly looped (Plate 111) whereas the flexure of *Cercaria 16A* is much more marked and the tubule in this region is somewhat convoluted.
In all other respects the two cercariae are very similar.

Papillar Spination:

Minute papillar spines were seen on the anterior region of this cercaria and their position corresponded with that of the spines on the cercaria of T. adamsi, with two exceptions. On Cercaria 16A immediately posterior to the junction of the oral sucker with the lateral cuticle two papillar spines were seen close together, as on the cercaria of T. adamsi, and in addition another was seen further ventrad between these two by focusing upwards slightly. These three papillar spines formed a triangle with two of them on the same horizontal plane and with the "apex" pointing ventrad.

In addition to the spine seen slightly ventrad to the lateral plane of focus another was seen on the dorsal surface of the cercaria immediately posterior to the junction of the oral sucker with the dorsal body cuticle. The cercaria was lying on its right side when this spine was observed. It appears almost impossible to observe such a spine while viewing it "end on" and therefore the spine was not seen while the cercaria was lying dorso-ventrally.

Double Infection:

Cercaria 16A was discovered escaping from the snail host along with a Strigeid cercaria. This represented the only case of a double infection for any of the cercariae described in this work. The two cercariae could be readily
distinguished on the basis of size and behavior. The Schistosome is comparatively large and swims somewhat lethargically. When at rest the latter holds the furcae at right angles to each other and slowly drifts downward. It is slightly positively phototactic but does not attach readily to glass. The Strigeid on the other hand was very small and swam quite actively for fairly long periods of time. When both of these cercariae were placed together in the "concentrator" (page 17) the Schistosomes alone were found in the side-arm, attracted by the light. This constitutes an excellent method of separating the two cercariae, since the water in the side-arm may readily be drawn off containing the Schistosomes.
CERCARIA CHILLIWACKENSIS (Plate V)

Morphology:

This is an apharyngeal, furcocercous distome cercaria of the "douthitti group". The following measurements (with the exception of the body width) were made of 25 cercariae fixed in 15% hot formalin. Since most specimens after fixation were bent slightly dorso-ventrally it was difficult to obtain measurements of the body width and only seven such measurements were taken. The means of the measurements are given with their standard error: body length .2668 ± .0084 mm; body width (7 specimens) .0782 ± .0027 mm; tail-stem length .3606 ± .0026 mm; tail-stem width, proximal .0436 ± .00041 mm, distal .0360 ± .00071 mm; furcal length .2058 ± .0042 mm; furcal papilla length .0145 ± .00029 mm; ventral sucker diameter .0350 ± .00041 mm; oral sucker length .0864 ± .0032 mm.

The ratio of the body length to the tail-stem length is approximately the same as that of the cercaria of T. adamsi. The furcae of Cercaria chilliwackensis are longer than half the length of the tail-stem. Dorso-ventral fin-folds are present on the quite narrow furcae.

The entire body, tail-stem and furcae are covered with minute retrorse spines. Those covering the body are much more prominent on the anterior half. Farther posterior on the body the spines are difficult to see, probably because they are shorter. Spines on the tail-stem and furcae are spaced considerably farther apart and are shorter than those found on the anterior half of the body. Spines are
present on the ventral sucker and these are arranged radially.

Papillar spines are present on the cuticle of the body. These were seen on the lateral cuticle of the cercaria and those near the anterior end of the body occupy much the same position as do those on the other cercariae described in this work (Plate V). The papillar spines located posterior to the eyespots on the other hand, are arranged in a distinctly different pattern and seem to be more spread out with relation to one another. On one side of the body papillar spines were seen which divided the space between the eyespot and the ventral sucker roughly into thirds. Another was seen on this side at the level of the ventral sucker or slightly posterior to its diameter. A papilla which corresponds to this latter position was seen on the opposite side of the body and another was seen on the other side corresponding to the papilla immediately posterior to the eyespot described above. It is probable that the papillar spination is bilaterally symmetrical and that all the spines on the other side were not seen. The most prominent of the papillar spines on *Cercaria ehilliwackensis* are the pair located immediately posterior to the junction of the oral sucker with the lateral body cuticle. The spines of this cercaria are not necessarily all in the same plane of focus. It is perhaps of especial significance that in some instances the small hair-like projection on the papillae was seen to move or vibrate and just prior to the death point of the cercaria this projection was observed to bend over and disintegrate.

The two eyespots are heavily pigmented and each possesses a "lens" which is directed laterad. The eyespots are
located between the oral and ventral suckers, slightly anterior to the mid-way point between these two organs.

The body of the cercaria possesses five pairs of penetration glands, the anterior two pairs of which are somewhat more granular and darker in appearance than the posterior glands. The four anterior glands surround the ventral sucker, one pair being anterior to this organ and one pair posterior to it. A nucleus was seen in each of them. Three pairs of smaller glands lie farther posteriorly.

The ducts of the penetration glands, which take a somewhat twisted course to the anterior end of the body, were seen in cross-section at one of the more anterior flexures. The duct bundle was observed to be composed of five separate ducts on each side. The ducts are slightly constricted at the point where they pass into the oral sucker. No papillae were observed at the termination of the ducts on the anterior end of the body, but these structures may be present.

A fairly long, thin oesophagus was seen beginning within the oral sucker at a point somewhat posterior from the tip of this organ and extending backward between the ducts of the penetration glands. A bifurcation of the oesophagus could not be discerned.

The excretory system of *Cercaria chilliwackensis* is composed of five pairs of flame cells in the body and one pair in the proximal portion of the tail-stem, all leading into a series of collecting tubules. Within the body three pairs of flame cells are located anterior to the ventral sucker and two pairs are posterior to it. These lead
into the anterior lateral and posterior lateral collecting tubules respectively. The flame cell formula is therefore $2(3^{+}/2^{+}1^{/})$. On each side of the body the anterior and posterior lateral collecting tubules join in the region of the ventral sucker to form a main collecting tubule. This runs posteriorly into an excretory bladder located at the junction of the body with the tail-stem. Two patches of cilia are present on each side of the body. One of these is located in the main collecting tubule near the point of bifurcation. The position of the other is not certain. It is located either in the main collecting tubule or in the anterior lateral collecting tubule. The outline of the bladder could not be seen clearly. From the bladder a central excretory tubule runs medially down the entire length of the tail-stem and bifurcates. Each branch runs through a furca and opens at the tip through an excretory papilla.

**Behavior:**

*Cercaria chilliwackensis* exhibits a mild response to a light stimulus moving in a positive direction. The degree of negative geotaxis is slight and the cercariae are found in all regions of the vial. However, when the vial is left undisturbed the cercariae usually cluster near the meniscus, the majority remaining free swimming. Under a strong light stimulus in the "concentrator" approximately one quarter of the cercariae attach to the jar on the positive side, forming a U shape with the body and tail-stem. Other cercariae drifting in the water move intermittantly, tail first, and then while resting drift slowly downward with the body below the tail. If the cercaria is swimming horizontally it will sink when resting, with the body
slowly pointing downward. The cercariae do not gather in clouds at the surface.

The emergence time of the cercaria is quite difficult to determine since any change of the water seems to induce more of the cercariae to emerge. As far as could be determined the cercariae emerge after 10 a.m. and the emergence continues until approximately 2 or 3 p.m. At 9.30 a.m. there are very few, if any, cercariae present in the vial.

**Dermatitis-Producing Abilities:**

*Cercaria chilliwackensis* is a dermatitis-producer as determined by experiment. This characteristic is discussed more fully under "Schistosome dermatitis Experiments".

**Life-cycle Experiments:**

Eight albino mice, two pigeons aged approximately one month, and one duckling were exposed to *Cercaria chilliwackensis* in the laboratory. Four of the mice were exposed by holding them with the tail suspended within a small vial containing a few cercariae in water. The remaining four were exposed with the "concentrator". One of the pigeons was made to stand with its feet in a cup of water containing from 100 to 200 cercariae. The other was given an intraperitoneal injection of 1.5 mls. of water containing from fifteen to thirty cercariae. The duckling was exposed by immersing the feet in a cup of water, one-quarter filled, containing up to two dozen cercariae. The mice which had been exposed to the cercariae manually were sacrificed 42
43 and 44 days after initial exposure. Those mice which had been put into the "concentrator" were sacrificed 55 and 59 days after exposure. Of the two pigeons used in the experiments, one died one month after exposure of an unknown cause and the other was sacrificed 45 days after exposure. The pigeon which died prematurely was the one which had been given an intraperitoneal injection and the death of the bird may have been related to the injection.

All attempts to infect laboratory animals with *Cercaria chilliwackensis* were unsuccessful. The explanation for this may be that so few cercariae were available for the experiments. Cercariae used for the experiments by other workers in the past numbered in the thousands; in spite of this, the experiments frequently were unsuccessful (Talbot, 1936).

**Sporocysts:**

The sporocysts from which *Cercaria chilliwackensis* was emerging were dissected from a snail giving off these cercariae. The snail had been dead for approximately one hour. Dissection revealed that the sporocysts were present as a tangled mass. It was found impossible to dissect any one of these structures out intact in order to measure its length. The macroscopic appearance of the sporocysts was recorded by photographing them (Plate VI). Measurements of the widths of the sporocysts were made. Extreme variations in the widths occurred and the sporocysts appeared very similar to those of *T. adamsi*. The widths varied from a minimum of .042 mm. to a maximum of .175 mm. The measurements obtained are given in Table 5.
TABLE 5
Measurements of Width of Cercaria chilliwackensis Sporocysts.

<table>
<thead>
<tr>
<th>Measurements of Width (mm)</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>.007</td>
<td></td>
</tr>
<tr>
<td>.063</td>
<td>.042-.175 mm.</td>
</tr>
<tr>
<td>.126</td>
<td></td>
</tr>
<tr>
<td>.098</td>
<td>.042</td>
</tr>
<tr>
<td>.042</td>
<td>Average</td>
</tr>
<tr>
<td>.168</td>
<td>.114 mm.</td>
</tr>
<tr>
<td>.070</td>
<td></td>
</tr>
<tr>
<td>.168</td>
<td></td>
</tr>
<tr>
<td>.070</td>
<td></td>
</tr>
<tr>
<td>.105</td>
<td></td>
</tr>
<tr>
<td>.168</td>
<td></td>
</tr>
</tbody>
</table>

Snail Host:

On July 7, 1953, cercariae which appeared identical with Cercaria chilliwackensis were found emerging from a snail identified as Physa c.f. coniformis. All cercariae were used in experimental infection attempts and the snail host died before any of them could be preserved for future study. On July 17 another snail, also Physa c.f. coniformis, was discovered in the laboratory to be giving off the cercariae described above as Cercaria chilliwackensis. The only means of comparison between the latter cercaria and the former were drawings of the first cercaria. On this basis, then, the cercariae were compared and considered identical. The first snail was collected on July 16, 1953, from Area A.
The latter snail was collected on July 16, 1953, from the area "South end of Beach" (Plate X).

Percent Infections:

Because *Cercaria chilliwackensis* emerged from the snails collected both from Area A and "South end of Beach" and since these snails were collected at different dates, it is necessary to set the date limits upon which the percentages are based to coincide with the dates of collection. The percent infection of snails collected in the two areas is therefore based upon the period from July 4 to July 16. Further, the percentages are based upon the number of *Physa* collected because these snails seem to be the only genus implicated. Only one collection was made in the area "South end of Beach" during the period from July 4 to July 16 and therefore percentage calculations are based on the number of *Physa* collected during this one visit. The number of snails collected during the above period is given in Table 6.

**TABLE 6**

Number of Snails Collected in Areas A and South end of Beach, With Dates.

<table>
<thead>
<tr>
<th>Area A</th>
<th>South end of Beach</th>
</tr>
</thead>
<tbody>
<tr>
<td>Date Collected</td>
<td>Number</td>
</tr>
<tr>
<td>July 4</td>
<td>125</td>
</tr>
<tr>
<td>July 16</td>
<td>89</td>
</tr>
<tr>
<td>Totals</td>
<td>214</td>
</tr>
</tbody>
</table>

Area A: *C. chilliwackensis* in \( \frac{1}{214} \times 100 = .467 \% \) of *Physas*
South end of Beach: \( C.\text{chilliwackensis} \) in

\[
\frac{1}{38} \times 100 = 2.63\% \text{ of } \text{Physa}.
\]

The percent infection based on the total number of snails (\text{Physa}) collected from both of these areas during the period from July 4 to July 16 is expressed as:

\[
\frac{2}{214 + 38} \times 100 = .794\% \text{ of } \text{Physa}.
\]

Comparison With Known Forms:

\textit{Cercaria chilliwackensis} possesses an excretory system with one pair of flame cells less than the seven pairs characteristic of the "ocellata group". It is therefore necessary to compare this cercaria with others possessing five pairs of body flame cells. In addition, \( C.\text{chilliwackensis} \) will be compared with cercariae which are known dermatitis-producers.

\textit{Cercaria tuckerensis} Miller(1927) exhibits the greatest degree of similarity to \( C.\text{chilliwackensis} \) morphologically: \( C.\text{tuckerensis} \) is somewhat larger and narrower than \( C.\text{chilliwackensis} \). A comparison of the measurements of these two cercariae is given in Table 7. Measurements of \( C.\text{tuckerensis} \) were obtained from Miller(1927). Perhaps the greatest point of difference between \( C.\text{tuckerensis} \) and \( C.\text{chilliwackensis} \) is the fact that the snail hosts of these two larvae belong to different families. \( C.\text{tuckerensis} \) is found in \text{Planorbis} sp.while, as has already been stated, \( C.\text{chilliwackensis} \) is found in \text{Physa}.
TABLE 7
Comparison of Measurements of C.chilliwackensis and C.tuckerensis

<table>
<thead>
<tr>
<th>Dimension</th>
<th>C.chilliwackensis</th>
<th>C.tuckerensis</th>
</tr>
</thead>
<tbody>
<tr>
<td>Body length</td>
<td>.267 mm</td>
<td>.325 mm</td>
</tr>
<tr>
<td>Body width</td>
<td>.078 mm</td>
<td>.08 mm</td>
</tr>
<tr>
<td>T.S.length</td>
<td>.361 mm</td>
<td>.4 mm</td>
</tr>
<tr>
<td>T.S.width</td>
<td>.044 mm</td>
<td>.04 mm</td>
</tr>
<tr>
<td>Furca length</td>
<td>.206 mm</td>
<td>.325 mm</td>
</tr>
<tr>
<td>Oral suc.len.</td>
<td>.086 mm</td>
<td>.1 mm</td>
</tr>
</tbody>
</table>

Cercaria douthitti Cort, as described by Price(1931), resembles C.chilliwackensis in that it possesses five pairs of flame cells in the body and is a known dermatitis-producer. This form however, has no fin-folds on the furcae and in addition it is found in Lymnaeidae.

Cercaria elongata Brackett has five flame cells on each side of the body. This pattern differs nevertheless from that of C.chilliwackensis. C.elongata possesses two flame cells connected to the anterior lateral collecting tubule and three connecting with the posterior tubule. This condition is reversed in C.chilliwackensis. Other differences between the two cercariae are the exceptionally long tail-stem of C.elongata and the occurrence of this cercaria in the snail Gyraulus parvus.

Comparison between C.chilliwackensis and Cercaria jaensbhi Johnstone and Cleland(1937) reveals that whereas the former possesses at least an oesophagus, the digestive system is entirely lacking in C.jaenschi. The latter cerc-
aria also attaches to the light side of a container while *C. chilliwackensis* does not attach to any great degree. *C. jaenschii* is found in *Amerianna* spp.

The above comprise the total number of dermatitis-producing cercariae known at the present time which possess five pairs of flame cells in the body. *Cercaria C* of Kemp (1919) and the cercaria of *Schistosomatium pathlocopticum* Tanabe (1923) are forms in which the body flame cells number five pair. These were not described as dermatitis-producers and they also differ from *C. chilliwackensis* in the type of snail host. Penner (1942) undertook to clarify the position of *Cercaria douthitti* and the cercaria of *S. pathlocopticum*. He concluded that the two forms were, in reality, synonyms. However, for the sake of the present comparison the cercaria of *S. pathlocopticum* will be considered separately. In *Cercaria C* of Kemp the penetration glands are arranged in a different manner about the ventral sucker, while the cercaria of *S. pathlocopticum* possesses no fin-folds. As far as can be determined these last two forms complete the list of known Schistosome cercariae, dermatitis-producers or otherwise, which possess an excretory system comprising five pairs of flame cells in the body and one pair in the tail-stem. On the basis of the comparisons made above, *Cercaria chilliwackensis* is described as a species distinct from any known form.
PLATE V

Fig. 1 - *cercaria chilliwackensis*, showing position normally attained under a cover-glass.

Fig. 2 - Protrusion of ventral sucker of *C. chilliwackensis*.

Fig. 3 - *C. chilliwackensis*, dorso-ventral view, illustrating morphology. The excretory system is shown on one side only.

X denotes papillar spines.

--All drawings were made free-hand--
PLATE VI

Fig. 1-Photomicrograph of freshly killed *Cercaria chilliwackensis*. X 24

Fig. 2-Cercaria of Fig. 1 enlarged. X 60

Fig. 3-Sporocysts of *C. chilliwackensis*. X 100
CERCARIA 10 (Plate VII)

Morphology:

This is an aphyaryngeal, brevifurcate, distome cercaria of the "spindalis group". All measurements were made of twenty-five specimens fixed in 15% hot formalin. The Means are given with the Standard error: body length .1985 ± .0094 mm; body width .0779 ± .002 mm; tail-stem length .2092 ± .0045 mm; tail-stem width, proximal .0366 ± .0014 mm, distal .0260 ± .00057 mm; furcal length .0955 ± .0049 mm; furcal papilla length .0120 ± .00026 mm; oral sucker width .0537 ± .0011 mm; ventral sucker .0295 ± .00057 mm, by .0216 ± .00082 mm.

The tail-stem of Cercaria 10 is very slightly longer than the body. While alive the tail-stem of this cercaria exhibits a marked "wrinkled" appearance. That is, the cuticle of the tail-stem is not straight, as it is in most of the other Schistosome cercariae, but a series of indentations occurs down its length on both sides. The furcae are extremely contractile. Occasionally they were observed to contract to mere lobes, extending at other times to a much greater length. The body of Cercaria 10 is "spade-shaped" and is narrowed at the posterior end where it joins the tail-stem. The proximal portion of the tail-stem is greater in width than the narrow posterior region of the body. The oral sucker of the cercaria is quite protrusable and when viewed alive this organ is continually being withdrawn into the body and pushed out anteriorly.

The entire body, tail-stem and furcae are covered
with minute retrorse spines. Those on the body are of uniform size and are as easily seen posterior to the ventral sucker as anterior to it. The spines on the tail are spaced farther apart than those on the body.

**Cercaria 10** possesses papillar spines on the body cuticle. The pattern of the spines anterior to the eyespots coincides with that of the cercaria of *T. adamsi* and **Cercaria chilliwackensis** and they are bilaterally symmetrical. One is situated on the oral sucker between the most anterior point of the body and the junction of the oral sucker with the lateral body cuticle. Two papillar spines are located quite close together immediately posterior to the above junction. Another is found on the lateral cuticle slightly anterior to the level of the eyespots. A papillar spine was seen slightly anterior to the level of the ventral sucker on the same side of the body as the ones described above and another was observed on the opposite side of the body slightly posterior to the diameter of the ventral sucker. The papillar spines of **Cercaria 10** are readily seen and are more obvious than those of either the cercaria of *T. adamsi* or **Cercaria chilliwackensis**.

Two granular eyespots are present in this cercaria. These are located at a point just anterior to the mid-point between the oral and ventral suckers. The eyespots are not enclosed within a definite delimiting structure and in this respect **Cercaria 10** differs markedly from the other cercariae described in this work. Minute pigmentation granules may be seen scattered somewhat from the principal body of the eyespot. The ill-defined nature of the eyespots of this cercaria is marked and may be easily discerned at first glance.
The body of *Cercaria 10* contains five pairs of penetration glands. The most anterior gland on each side is lobed and is in this respect quite unique. Initially it was believed that there were three pairs of anterior glands, so distinct is the lobation. Closer examination however, and a count of the number of ducts leading from the glands, indicated that there were only two pairs of anterior glands, the first pair being lobed. It was very difficult to determine whether gland one or gland two was the lobed one, but very close examination seemed to indicate that the most anterior pair were the lobed ones. The anterior two pairs surround the ventral sucker, with pair number two lying directly posterior to this organ. The posterior three pairs of glands are arranged linearly on each side and lie immediately posterior to pair number two. A nucleus was observed within each gland. The ducts from the glands extend anteriorly as a compact but large bundle. The connection of the ducts with the most anterior pair of penetration glands is dorsad. The number of ducts making up each bundle was observed to be five, in a cross-sectional view at a flexure. The ducts take a somewhat twisted course and open at the anterior end of the body. At this termination five papillae were observed but it could not be determined whether the ducts opened at the tip of the papillae or at their base.

A digestive tract was observed between the penetration gland ducts. This consisted of an oesophagus leading from the posterior end of the oral sucker backward to a point approximately half-way between the eyespots and the ventral sucker. Here the oesophagus bifurcated into two very short caeca. The outline of the caeca was best seen when the cercaria had been subjected to a very dilute solution of neutral
red. When this technique had been employed numerous small red granules could be seen within the intestinal caeca.

The excretory system of Cercaria 10 is one of its most unique features. It consists of only four pairs of flame cells in the body and one pair in the proximal portion of the tail-stem, all running into a series of collecting tubules. Two pairs of flame cells lead into the anterior collecting tubules (one pair on either side of the eyespots) and two pairs lead into the posterior collecting tubule. Thus, the flame cell pattern of this cercaria is \(2(2+/2+1/)\).

The anterior and posterior lateral collecting tubules join at a point level with the anterior end of the ventral sucker, where they form a main collecting tubule on either side of the body. The proximal portion of the main collecting tubule is characterized by two distinct regions of convolution and the actual course of the tubule cannot be accurately followed through these areas. A ciliated patch is located near the point of bifurcation of the main collecting tubule and also within the coils of the more posterior convolution. The main collecting tubule runs posteriorly where it joins an excretory bladder near the junction of the body with the tail-stem. The outline of the bladder could not be clearly observed. A central excretory tubule leaves this bladder and runs medially down the length of the tail-stem. It bifurcates near the furcae and a tubule leads through each furca to open through an excretory papilla at the furcal tip.

Associated with the eyespots of Cercaria 10 structures were observed which may represent portions of the nervous system. An extremely fine ring surrounds each eyespot and this tends to become extended toward the midline (Plate VII,
Fig. 1). This structure was noticed about the eyespots of the cercaria of *T. adamsi* and *Cercaria chilliwackensis* but it is much more obvious and easily seen in *Cercaria 10*, probably because of the ill-defined nature of the eyespots of the latter. Leading laterally from each ring and crossing the anterior collecting tubule of the excretory system is a very fine structure which may be a nerve tract. This bifurcates laterally into an anterior and posterior "line". No other details in this respect could be observed. The structures described consist merely of fine refractile lines and are very difficult to observe.

**Behavior:**

*Cercaria 10* is markedly negatively geotactic. In general, the majority of the cercariae in a vial at any one time will be found clinging to the surface film by the ventral sucker. Usually the cercariae cannot be seen in numbers by looking through the side of the vial into the water, the container appearing quite devoid of these forms. It is necessary to view the surface of the water, with a lens if possible, to appreciate the numbers present within the vial. While clinging to the surface many of the cercariae remain motionless. They become active is disturbed. A few of the cercariae attach to the side of the vial. *Cercaria 10* also exhibits a slow, positive reaction to a light stimulus. When a strong light is shone into the vial the cercariae will attach to the side of the vial where the stimulus is strongest. A strong light stimulus seems to obliterate to some extent the intense negative geotaxis. When free swimming in a vial the entire cercaria swims with a spiral motion describing a somewhat corkscrew path.
The cercariae may be concentrated in very large numbers in the side-arm of the "concentrator" described under "Methods and Apparatus".

Under a cover-glass the cercariae hitch along by alternate attachments of the oral and ventral suckers. The body of this cercaria exhibits an amazing ability to withstand unfavorable conditions and usually remains viable long after the tail-stem has become disjoined from the body. In one instance the body remained viable when the water under the cover-glass had nearly completely dried, the dry spots closely surrounding it. The flame cells could be studied with ease.

**Dermatitis-Producing Abilities:**

*Cercaria 10* is a dermatitis-producer, as determined experimentally. This characteristic is discussed more fully under "Schistosome Dermatitis Experiments".

**Life-cycle Experiments:**

Four white mice, one Guinea pig aged approximately one month, and two ducklings were exposed to *Cercaria 10* in attempts to complete or elucidate the life-cycle of this trematode. Two of the mice were exposed both with the "concentrator" and manually, while two were exposed with the "concentrator" alone. The Guinea pig was shaved in the inquinal region and drops of water containing cercariae were applied to this area. Considerable difficulty was experienced in retaining the water on this region for any length of time. The Guinea pig was also given approximately two dozen cercariae orally. The ducklings were exposed in a manner similar to
that used in experiments with the cercaria of *T. adamsi*, and were held in place for ten minutes. Three of the mice were sacrificed sixty and sixty-one days after exposure. One mouse died of an unknown cause on the forty-fourth day. The Guinea pig was sacrificed eighty-two days after exposure to the cercariae. The ducklings were killed fifty-one days after they had been subjected to the cercariae.

All attempts to infect laboratory animals with *Cercaria 10* were unsuccessful. A definite explanation cannot be put forth to account for this. With the aid of the "concentrator" a fairly large number of cercariae could be brought near the exposed area of the mice. Admittedly, Talbot (1936) in his experimental attempts to infect water fowl with *C. elvae, C. physellae* and *C. stagnicolae*, used large numbers of cercariae and repeated exposures without results. In the experiments with *Cercaria 10* the reason for the failure of adult worms to become established in the animals used must fall into one of two categories. Either the conditions for cercarial penetration were inadequate (i.e. insufficient cercariae, exposure time too short, wrong area used for exposure) or the animals used in the experiments were incapable of acting in the capacity of the adult host.

More experiments would be necessary using larger numbers of cercariae over a greater period of time before mice could be eliminated as a possible adult host for this parasite.

**Sporocysts:**

Sporocysts of *Cercaria 10* were observed both within
a snail host of this cercaria which died prior to dissection and within a snail which was dissected "in vivo". Measurements were made of the sporocysts in the latter snail after the larvae had been killed with a drop of formalin. The width varied considerably along its length. Widths at the widest points varied from .098 mm to .133 mm for six measurements. The minimum width values were .042 mm and .049 mm. The exact length of the sporocysts could not be determined since the organisms were very much intertwined with each other and they could not be dissected apart without a tear occurring somewhere along the length. However, the length probably exceeds 16 times the width at the largest point.

The measurements of the widths are given in Table 8.

<table>
<thead>
<tr>
<th>Width (mm)</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>.133</td>
<td>.042-.133 mm</td>
</tr>
<tr>
<td>.098</td>
<td>Average</td>
</tr>
<tr>
<td>.119</td>
<td>.098 mm</td>
</tr>
<tr>
<td>.105</td>
<td></td>
</tr>
<tr>
<td>.126</td>
<td></td>
</tr>
<tr>
<td>.112</td>
<td></td>
</tr>
<tr>
<td>.042</td>
<td></td>
</tr>
<tr>
<td>.049</td>
<td></td>
</tr>
</tbody>
</table>

Since one of the snail hosts of Cercaria 10 was dissected alive, it was possible to observe the living sporocysts. This allowed the cercariae to be observed while escaping from the sporocysts. The escape was effected by
the cercaria hitching along up to the long thin end of the sporocyst and wriggling vigorously until it had squeezed through the long narrow "tubule" (Plate VII, Fig. 2).

Snail Host and Percent Infections:

Cercaria 10 emerged from a total of four snails, all of which have been identified as Physa c.f. coniformis. Three of these snails, found on July 4, were collected from Area B. The other was collected on July 29 from Area A. The latter snail died one day after the cercariae were first observed emerging and dissection revealed a juvenile leech within the shell. The leech had been collected along with the snail; beneath the shell of a snail the molluscan leeches are completely hidden and are brought in with the snail from the field.

Because Cercaria 10 was found in both Area A and Area B at different times it is necessary to consider the percent infection based on the period delimited by the dates of these two collections. Cercariae were discovered from Area B first on July 4 and lastly from Area A on July 29. The percent infection will therefore be considered on the basis of the number of snails collected on and between these dates. Further, the percents are given only on the basis of the number of Physa infected. The number of snails collected from each area is given with the dates of collection in Table 9.
### TABLE 9

Number of Snails Collected From Areas A and B, With Dates.

<table>
<thead>
<tr>
<th>Area A</th>
<th>Area B</th>
</tr>
</thead>
<tbody>
<tr>
<td>Date Collected</td>
<td>Number</td>
</tr>
<tr>
<td>July 4</td>
<td>125</td>
</tr>
<tr>
<td>July 16</td>
<td>89</td>
</tr>
<tr>
<td>July 25</td>
<td>32</td>
</tr>
<tr>
<td>July 29</td>
<td>19</td>
</tr>
<tr>
<td>Total Physas</td>
<td>265</td>
</tr>
</tbody>
</table>

During the period of emergence three snails were found infected with *Cercaria 10* from Area B and one from Area A. The percentage of *Physas* infected with *Cercaria 10* for these two areas is then:

**Area B:** *Cercaria 10* in \( \frac{3}{109} \times 100 = 2.75\% \) of *Physas*

**Area A:** *Cercaria 10* in \( \frac{1}{265} \times 100 = 0.377\% \) of *Physas*

Both Areas Together: *Cercaria 10* in \( \frac{4}{374} \times 100 = 1.07\% \) of *Physas*

The above calculations are valid for the period between July 4 and July 29, inclusive.

**Comparison With Known Forms:**

At the time of writing, only two dermatitis-producing cercariae have been described which possess an excretory system consisting of four pairs of flame cells in the
body. The earliest described is the cercaria of *Schistosoma spindalis* Montgomery 1906 (Soparkar, 1921) and it has been described only from India and Malaya (Soparkar, 1921; Buckley, 1938) and South Africa (Porter, 1938). The other dermatitis-producing cercariae possessing four pairs of flame cells in the body is the cercaria of *Gigantobilharzia huronensis* Najim (1951). As far as can be determined, only three other Schistosome cercariae are known which possess an excretory system of this kind and these are non-dermatitis-producers. *Cercariae indicae XLVII* Sewell (1922) has been described from Achur and Wynaad in India. *Cercaria B* of Kemp (1919) was described from Hamun-i-Helmand, Seistan in India. Annie Porter reported the discovery in Africa of a Schistosome cercaria which was photophobic. This cercaria was designated as *C. elandsiae* and it possessed four pairs of body flame cells. The cercaria of *G. huronensis* is the only cercaria with this type of excretory system which has been found in the past in the Western Hemisphere. A comparison will be made between the above forms and *Cercaria 10*.

The cercaria of *S. spindalis* produces a dermatitis which is contracted by workers in the padis of Malaya, where it is known as "sawah itch". Buckley described the dermatitis as one which is contracted in the morning and becomes evident in the evening. The cercaria of *S. spindalis* is similar in macroscopic appearance to *Cercaria 10*. The furcae are comparatively short. The body spination, as described by Soparkar, is especially marked at the anterior portion of the body. Further, the oral organ is protrusable. The intestinal caeca of this cercaria contain granular material which stains with intravitam neutral red. The cercaria varies in other respects from *Cercaria 10* to such a degree
that possible synonymity is eliminated. The cercaria of *S. spindalis* possesses no eyespots. The tail-stem is much longer proportionally than the body as compared with the same ratio in *Cercaria 10*. In addition, the anterior two pairs of penetration glands are both located in front of the ventral sucker in the cercaria of *S. spindalis*. The snail host of the latter is *Planorbis exustis* which is not a Physid snail. The behavior pattern of the cercaria of *S. spindalis* is quite different from that of *Cercaria 10*. The latter are found clinging to the surface film while the former hang in the water with the body downward and gradually sink, only to swim upwards again intermittantly.

The cercaria which most closely resembles *Cercaria 10* is that of *Gigantobilharzia huronensis* Najim(1951). So close is the resemblance that the two may in future be found to be synonymous. The behavior pattern of the two cercariae is essentially the same. The cercaria of *G. huronensis* is found in *Physa gyrina*. The body proportions are similar to those of *Cercaria 10*, the furcae being shorter than half the length of the tail-stem and the tail-stem length very slightly exceeding that of the body. Najim described the cercaria as possessing papillar spines on the side of the body. However, only three of these were apparently seen on one side, with one on the opposite side. They are located in positions corresponding with some of the structures found on *Cercaria 10*. The relative positions of the papillar spination on the two cercariae are illustrated below.
Within the body further similarities between the two cercariae become apparent. Both possess five pairs of penetration glands. Najim stated that the second pair of glands is tri-lobed. Cercaria 10 is described as possessing lobation in the first pair of glands. Either one of the cercariae has been incorrectly described, or the cercariae are two separate forms. The question must, for the moment, remain an open one. The excretory system of the cercaria of *G. huronensis* is essentially the same as that of Cercaria 10. However, the two regions of marked convolution in the main collecting tubules of Cercaria 10 are not described as occurring in Najim's cercaria. He does state however, that the elements of the excretory system of his cercaria were found occasionally to vary considerably in position. In the author's own words, "Variations in the position of the flame cells and the looping of the tubules were noticed quite often." The number of flame cells he states, were even found to vary, an "extra" one being noted at times posterior to the ventral sucker. It is possible that a cil-
iated patch was mistaken for a flame cell in this latter case, but it is equally possible that the cercaria actually possessed a greater number of flame cells than were described by Najim. This author also mentions that the third and fourth flame cells "might come closer together in position". In view of this high degree of "variation" it seems undesirable to eliminate the possibility of there being more than four pairs of body flame cells in this cercaria. The cercaria of *G. gyrauli* Brackett (1940), belonging to the same genus as Najim describes for his organism, possesses six pairs of flame cells in the body.

The adult worm of *Gigantobilharzia huronensis* was experimentally established in chickens and canaries. It was found in naturally infected goldfinches and cardinals.

The principal points of difference between *Cercaria 10* and the cercaria of *G. huronensis* are as follows: Najim's cercaria is described as possessing a fairly large "brain" with the lateral lobes surrounding the eyespots. This structure was not observed to be so extensive in *Cercaria 10*. In the latter cercaria the nervous system, already described, encloses the eyespots very closely (Plate VII, Fig. 1). This is perhaps one of the most obvious differences between the two cercariae. Najim states that the bifurcation of the oesophagus of his cercaria is incomplete or bulb-like. The bifurcation of the oesophagus of *Cercaria 10* was observed to be distinct and a number of granules were seen within the short caeca. Further, Najim described relatively few papillar spines on the body of his cercaria. More of these structures were observed on *Cercaria 10*. 
It seems probable that *Cercaria 10* falls within the genus *Gigantobilharzia* on the basis of comparison with the cercaria of *G. huronensis* although this cannot be definitely established until the adult of *Cercaria 10* is known. Only two other members of the genus *Gigantobilharzia* have been described. These were reported by Brackett (1942). This author described the life-cycle of *G. gyrauli* and the adults of *G. lawayi*. As previously mentioned, the cercaria of *G. gyrauli* possesses six pairs of flame cells in the body. The possibility of synonymity between Najim's cercaria and *Cercaria 10* cannot be overlooked. It is therefore advisable to leave the question an open one until future work can be carried out with *Cercaria 10*.

The other species described with four body flame cells are not dermatitis-producers, or rather they have not been described as such to date. However, a comparison will be made at this time between these forms and *Cercaria 10* since the number of cercariae known to possess this type of flame cell pattern is so small. *Cercariae indicae XLVII* possesses no furcal fin-folds and in this respect differs from *Cercaria 10* to the greatest extent. The macroscopic appearance of this cercaria is strikingly similar to that of *Cercaria 10*. The appearance of the eyespots is similar to that of the Cultus Lake cercaria in that there is a marked degree of granulation. Further, the penetration glands, although possessing no lobation, are arranged in a similar pattern about the ventral sucker, with respect to those of *Cercaria 10*. The posterior portion of the body is somewhat bulbous, narrowing down greatly at the point of junction with the tail-stem.

Kemp, in his description of *Cercaria B* stated that
this form is closely related to *Cercariae indicae XLVII*. *Cercaria B* differs markedly from *Cercaria 10* in that the former possesses a pair of un-pigmented eyespots. Further, no trace of a digestive tract could be seen by Kemp in his observations. Kemp's description of this cercaria, although a valuable contribution to the field at the time of publication, is quite vague in comparison with the precise descriptions found in more modern works, and is highly inadequate for comparative purposes.

It is not difficult to distinguish between *Cercaria 10* and *C. elandsiae* Porter(1938). The former is markedly positively phototactic while the latter is photophobic. While the number of flame cells in the excretory system is the same for both, the position of these flame cells differs quite distinctly in the two forms. The course of the main collecting tubule of these cercariae varies with each case to a considerable degree.
PLATE VII

Fig. 1—Cercaria 10 showing nervous circum-ocular rings and structures which may be nerve tracts.

Fig. 2—Excretory papilla on furca tip of Cercaria 10

Fig. 3—End of sporocyst of Cercaria 10 showing method of escape of cercariae.

Fig. 4—Cercaria 10, dorso-ventral view, illustrating morphology. The excretory system is shown on one side only, with penetration glands shown only on opposite side for clarity. X denotes papillar spines.

—All drawings were made free-hand—
PLATE VIII

Fig.1-Photomicrograph of Cercaria 10 sporocysts. X 40
Fig.2-Sporocysts of Fig.1 enlarged. X 100
Fig.3-Photomicrograph of freshly killed Cercaria 10. X 100
FIG. 1

FIG. 2

FIG. 3

PLATE V111
PAPILLAR SPINATION.

As far as can be determined, papillar spination such as is found on the Cultus Lake trematodes has never been described in the past on adult Schistosomes. The occurrence of similar spines on Schistosome cercariae has been described only seldom in the past. Szidat in 1929 and Gordon in 1934 reported the occurrence of similar spines on Schistosome cercariae (Najim, 1951). The former observed what he called "sense-bristles" on the cercaria of B. polonica. Gordon reported spines on the cercaria of Schistosoma mansoni and S. haematobium. Wall (1941) reported the presence of similar spines on the cercaria of Spirorchis elephantis. This author described the cercaria as also possessing papillar spines on the tail-stem. Vercammen-Granjean (1951) published a paper in which he stated that he observed long spines with basal papillae on the body of the cercaria of Schistosoma mansoni. He included illustrations of this spination and these structures appear very similar to those observed on the three Cultus Lake forms. However, the hair-like projection on the papillae appears very much longer and finer than this structure on the Cultus Lake cercariae. Granjean's observations were made with a magnification of 1470X, a considerably higher magnification than the slightly less than 1000X used to observe the local forms. Granjean stated that it was of primary importance that the cercariae be studied "in vivo" and at the correct temperature (30°C). He also emphasized that the cercariae were studied in human serum. Twenty-four papillar spines were described on the body of the cercaria. These were not all in the same plane but were symmetrical in their arrangement. No spines were located on the extreme lateral "edge" of the body, being above and below this plane.
Granjean's observations were confirmed by J.J.C. Buckley of the School of Hygiene and Tropical Medicine of London. Further, the author stated that in a private correspondence Buckley informed him he had observed similar spines on the cercaria of *Schistosoma bovis*. Granjean also indicated that in 1934 Gordon, Davey and Peaston observed similar spines on the tail-stem of *S. mansoni*.

Najim (1951), in his description of the life history of *Gigantobilharzia huronensis*, reported the occurrence of papillate spines on the body of the cercaria of this species. Three spines were seen on one side and one on the other side, as illustrated in his drawing of the cercaria. These spines are located in positions which coincide with some of those on the Cultus Lake forms, but more spines were observed on the latter.

**Morphology of Spination on Cultus Lake Cercariae:**

The papillar spination of the Trematodes described in the present work can be viewed only with the use of oil-immersion. Further, it is necessary that the observations be made immediately after the cercariae are put beneath a cover-glass as a wet-mount. The spines tend to disintegrate very early during the course of the observation.

There are a number of points of similarity between this type of spination on the three Cultus Lake Trematodes. The papillar spines possess at their base a minute dome-like papilla projecting slightly from the cuticle. The size of these papillae varies. They are usually largest near the anterior end of the body. The spines are present as an ex-
tremely fine "hair" or projection at the apex. The projections vary in length from a minimum the same length as the papilla is high, to about four or more times the height of the papilla. The longer "hairs" or spines are very much thinner than the shorter ones and they appear almost as shadows. In general, the papillated spines are more obviously seen near the anterior portion of the body.

Twice while observing Cercaria chilliwackensis some of the hair-like projections on the papillae were observed to be undergoing a vibratory motion.

The papillar spines on the anterior half of the body, and probably also further posterior, are bilaterally symmetrical. On each side of the body the cercariae possess one spine between the most anterior point and the junction of the lateral cuticle with the oral sucker; two spines close together just posterior to the above junction; one spine between the above pair and the level of the eyespots; and further posterior in the region of the ventral sucker, other spines arranged in various patterns. The above pattern of papillar spination is common to the three types of cercariae described herein, regardless of the obvious dissimilarities of these cercariae in other respects. The position of the spines varies with the different cercariae in one respect only. Those papillar spines located posterior to the level of the eyespots are not located in similar positions in the three cercariae. The positions of these more posterior papillar spines are characteristic for each of the cercaria of Tadamisi, Cercaria chilliwackensis and Cercaria 10 as illustrated in Plates III, V and VII.
The papillar spines are not all located on the same plane. Much focusing is necessary in order that these spines may be seen and very rarely do more than two or three spines appear on the same plane of focus. On Cercaria 16A, described on page 45, a "triangle" formed by three papillar spines was observed near the junction of the oral sucker with the lateral cuticle. Further, a papillar spine was observed on the dorsal surface of this cercaria immediately adjacent to the posterior edge of the oral sucker. On a few occasions while viewing the ventral surface of Cercaria chilliwackensis, a papillar spine was observed on the ventral cuticle by focusing upwards. This spine was located midway between the lateral body cuticle and the ventral sucker, at a level with the posterior edge of the latter.

The spines situated upon the apex of the minute papillae disintegrate, considerably in advance of the death point of the cercaria. Occasionally the minute spine was observed to bend over and disintegrate prior to the death point. Just before the death point of the cercaria only the papillae are seen, while after the cercaria has died these papillae are located only with great difficulty and have usually disappeared completely.

The life-cycle experiments undertaken with the cercaria of T.adamsi revealed that the papillar spination is carried over into the adult stage of this form. The pattern of the more anterior spines of the cercaria is retained in the adult (Plate 11). The spines of the adult T.adamsi are slightly more difficult to observe than those of the cercaria.
During the survey of the Okanagan Valley in 1952 another cercaria was discovered which bore papillar spines in a very similar pattern to that of the Cultus Lake forms. The exact identity of the Okanagan cercaria has not been determined but it belongs to the "douthitti group" possessing as far as could be observed at the time, five pairs of flame cells in the body. The behavior pattern of the Okanagan cercaria coincided with the behavior described for the cercaria of *G. huronensis* Najim, *Cercaria douthitti* Cort and *Cercaria 10*, and indeed the cercaria may be synonymous with one of these forms.

The significance of the papillar spination is not at all certain. It is possible that these structures have been merely over-looked by other workers in the past. The possibility also exists that cercariae possessing this type of spination are entirely separate forms from those described without them, and are actually new species. To the author's mind, it seems highly unlikely that a worker who is familiar with cercariae and who has been studying them for some time could describe accurately the minute retrorse spination on the body of a cercaria and over-look the larger, more complex papillar spines. The only reasonable explanation for the possible failure of previous workers to observe papillar spines on cercariae seems to lie in the fact that the papillar spines disintegrate soon after the organisms have been placed under a cover-glass. Perhaps initial concentration by a worker on some other morphological aspect of the cercaria in view allowed any papillar spines which might have been present to disintegrate before the attention was focused on them.
TAXONOMIC DISCUSSION.

Taxonomy of the Adult *Trichobilharzia adamsi*.

Price (1930) in his description of the family *Schistosomatidae*, divided the group into two subfamilies—the *Schistosominae* and the *Bilharziellinae*. The *Schistosominae* includes female worms which are round in cross-section, while the worms of the *Bilharziellinae* are more or less flattened, especially in the more posterior regions of the body. Further, the intestinal caeca of the *Schistosominae* unite caudal of the equator of the body, while those of the other subfamily unite more anteriorly. It is apparent therefore that the adult Schistosome described in the present work is a member of the *Bilharziellinae*, on the basis of the flatness of the body and the morphology of the intestinal caeca.

The genera of the subfamily *Bilharziellinae* were listed by Price as *Bilharziella*, *Gigantobilharzia*, *Dendritobilharzia* and *Trichobilharzia*. The description of *Bilharziella* stated that the posterior of the body is distinctly flattened, thus inferring that the more anterior regions of the body were not as flattened. The description does not coincide with that of the local worm. Further, the *Bilharziella* species described by Price are not as greatly elongated with respect to their width as is the present worm. The worms of *Gigantobilharzia* are somewhat cylindrical and do not possess suckers. *Dendritobilharzia* possesses a cuticle devoid of spines and suckers are absent. In addition, these worms possess a digestive caecum with lateral caeca extending from it. Price's description of the genus *Trichobilharzia* was quite inadequate for comparison with other forms. Between the time
of publication of Price's paper and the McMullen and Beaver publication (1945) of the description of the adults of *C. stagnicae*, *C. physellae* and *C. elvae*, another genus became established by Ejsmont (Brackett, 1942). This was *Pseudobilharziella*. McMullen and Beaver revised the genus *Trichobilharzia* and included under this description the former genus *Pseudobilharziella*. For this reason, the latter genus is mentioned only briefly at this time. Of great importance was the description given by McMullen and Beaver of the specific diagnosis of the revised genus *Trichobilharzia*, and on the basis of this description an adequate comparison can be made between this generic diagnosis and the worm being described at present. On the basis of this comparison and comparison with illustrations of the adult *T. stagnicae*, *T. physellae* and *T. ocellata* our worm is diagnosed as *Trichobilharzia*.

**Taxonomy of the Cultus Lake Cercariae.**

The taxonomic position of cercariae has in the past been determined on the basis of a number of criteria. Faust (1924) stated that the position of the flukes should be determined on the basis of a system which is carried through the entire life-cycle with a minimum of change. The only system of which this can be said, as observed by Faust, is the excretory system. On the basis of embryological development and morphology the excretory system of furcocercous cercariae is probably the most primitive of all types (Hussey, 1941). Relatively little change occurs in the excretory structures throughout the life-cycle of the fluke. On the other hand, Dawes (1946) stated that the flame cell "formula" does not designate phylogenetic relationships between cercariae, while Harper (1929) was of the opinion that the excretory system
is not an infallible guide in cercarial diagnosis. The excretory system is usually extremely difficult to trace with any degree of accuracy. Further, Stunkard (1929) observed that certain additions to the excretory system do occur during the transition from cercaria to adult.

Most authors in the past have based much of their descriptive work upon the specific measurements of cercariae. Various cercariae have been separated and considered separate species because they were "smaller" or "larger" than another similar form. One great difficulty in this respect has been that the cercariae are very contractile and there is usually a wide range of measurements for any given dimension. Porter (1938) reported that she had found the cercaria of *S. spindalis* in South Africa, but the measurements of this cercaria were less than those of the Indian strain, particularly the length of the tail-stem. On this basis she proposed that the African cercaria be considered as a variety *africana*. Neuhaus (1952) in experiments with the cercaria of *Trichobilharzia szidati* found that "the cercariae of various intermediate host individuals of the same kind and the same origin are different". He also stated that cercarial modifications occur in each individual intermediate host under the influence of individual variations of the host, and the age of the host influences the developmental time of the parasite. Neuhaus observed that one could correlate the different development times with large differences in the cercariae. It would seem therefore, that measurements of the dimensions of cercariae do not form a reliable source of criteria upon which to base comparisons.

Miller (1926) outlined a system for grouping Schist-
osome cercariae based on a similar system previously put forth by Sewell. This grouping is based on the flame cell pattern of the cercariae, as well as upon other data, and is given as follows for the cercariae known in 1926.

**GROUP A.**-Human Schistosomes, possessing three pairs of flame cells in the body.

**GROUP B.**—Cercaria B of Kemp.
Cercaria of *Schistosoma spindalis*.
*Cercariae indicae XLVII*

This group is characterized by the presence of four pairs of flame cells in the body. The group was modified from the original of Sewell by the addition of *Cercaria B*, the excretory system of which was unknown when the group was formed.

**GROUP C.**—Cercaria douthitti.
*Cercaria C* of Kemp.
*Cercaria of Schistosomatium pathlocopticum*.

Originally Sewell created this group to also include *Cercaria elephantis* but Miller gave this a separate grouping. Group C is characterized by the presence of five pairs of flame cells in the body of the cercaria. Sewell (1922) stated that "no forms corresponding to this group have been found in India".

**GROUP D.**—Cercaria ocellata.
*Cercaria bombayensis no. 19*.
*Cercaria elvae*.
*Cercaria gigantea*.

This group is characterized by the presence of six pairs of flame cells in the body and by
the number of penetration glands. The "ocellata group", as constructed by Sewell (1922), is typed by *Cercaria ocellata*. The excretory system of this cercaria was unknown at the time the group was constructed by Sewell. The latter stated that the excretory pattern of the "ocellata group" was $2(4+1)$ and he included a new cercaria discovered by himself, *Cercaria indicae* XXV, within this group. This cercaria possesses an excretory pattern $2(3+/1+/1/)$. The body is described as being pear-shaped, being widest at the posterior end. The penetration glands were given as consisting of two anterior pairs plus a large amorphous mass posterior to the ventral sucker which was considered to be possibly other penetration glands. The tail-stem was described as being joined to the body distinctly ventro-posteriorly. The furcae were quite short and the tail-stem was nearly twice the length of the body.

Sewell also stated that the cercariae of this group differ "from nearly all other closely related forms" in that the penetration gland ducts open through papillae at the anterior end of the body instead of opening through conical hollow spines.

Many additions have been made to the above groups since 1926. Group D has received the greatest number of additions with the increasing number of known Schistosome dermatitis producers, and others. Group C, the "douthitti group", has received a few additions, also dermatitis-producers. As far as can be determined Group B has received but two additions to those listed by Miller. These are the cercaria of *G. huronensis* Najim and *Cercaria elandsiae* Porter.
Position of the Cultus Lake Cercariae:

Cercaria 10, on the basis of the flame cell pattern and other similarities, may be placed within Miller's Group B, or Sewell's "spindalis group". This cercaria although a dermatitis-producer similar to the cercaria of *S. spindalis* does not belong to the genus *Schistosoma*. The latter genus is characterized by the absence of eyespots and these structures are present on Cercaria 10. In fact, the very close similarity between the cercaria of *Gigantobilharzia huronensis* and Cercaria 10 constitutes strong evidence in favor of the assignment of the latter to this genus. Nothing further may be said concerning the classification of Cercaria 10 until the adult of this form is known.

Cercaria chilliwackensis possesses five pairs of flame cells in the body and therefore may be located in the "douthitti group", or Group C of Miller.

The cercaria of *Trichobilharzia adamsi* belongs to the genus which is included in Group D. The cercariae of the known species of *Trichobilharzia* all belong to the "ocellata group".
Plate IX illustrates the relative position of the collecting area (C16) to other points at Cultus Lake. The map (Plate X) of the specific area under investigation shows the relative positions of the three areas from which snails were collected that produced the cercariae described in this work. These areas all consist of a fairly shallow beach which is similar in nature throughout. The bottom is composed of rocks which in places are covered with a light growth of algae. Snails were gathered in water not exceeding an arm's length in depth. The snails from these separate localities have all been identified as belonging to the same species. However, despite the apparent uniformity of conditions the occurrence of the cercariae described was confined to specific areas. It must be noted here that collections of snails were only occasionally made outside of the areas indicated and cercariae could quite conceivably have occurred outside of the boundaries of the areas. These boundaries cannot be precisely located about any given area and they are shown merely in an attempt to delimit somewhat the area in question.

The cercariae were found to be specific for a given area and only in one case, near Point alpha, did the specific areas overlap. Cercaria 10 was found in Area B and in Area A on July 4 and July 29 respectively. The concentration of these was three times as great in Area B (July 4) as in Area A (July 29). Cercaria chilliwackensis was found in Area A farther north on July 4, and in the Area "south end of beach" on July 17. Later in the summer, on August 3, the cercaria of T.adamsi was found in Area B. That is, the latter cercaria
seemed to have replaced *Cercaria 10* in this area, almost a month after *Cercaria 10* was found here. It is to be emphasized that at no time were the above three types of cercariae found in snails which had been located in close proximity.

For an explanation of this phenomenon it will be necessary to consider the adult hosts of the Trematodes. Of the three forms described in this work a host has been found for only one of them, the cercaria of *T. adamsi*. This was experimentally determined to be a water fowl. It may be suspected then, that a water fowl is acting as the adult host of the parasite at Cultus Lake. Since the cercaria has been found only in Area B it is likewise reasonable to suspect that these water fowl inhabit the region of the shore line of Area B. Large numbers of water fowl have been observed during the day far out toward the middle of the lake. None have ever been seen by this worker actually close to Area B during his daytime visits. It is perhaps conceivable that the birds come into the area during the night. However, no reason is suggested as to why the birds would choose this particular location. A study of the habits of the birds seen during the day is needed and Area B should be closely watched at night in order to determine whether the birds actually do come to this region at that time.

The remaining two cercariae found at an earlier date than the above possess a flame cell pattern perhaps indicative of mammalian forms, but the possibility of an avian host also exists. Since the life-cycle of *Cercaria chilliwackensis* and that of *Cercaria 10* is not known we can say nothing factual concerning the ecology of these parasites. The close similarity between *Cercaria 10* and the cercaria of *G. huron-
Najim seems to indicate that this cercaria is an avian form. It must be noted that Cercaria 10 was replaced later in the season by an avian parasite, T. adamsi, which proves that birds capable of harboring the Trematodes are either present or do visit the vicinity of this region. However, Cercaria 10 is also similar to the cercaria of S. spin-dalis which matures in the buffalo of Malaya.

An important question in the consideration of life-cycles is chronology. We can delimit the period of existence of the cercaria to a few hours, or possibly days, after it has emerged. It therefore follows that penetration of the cercaria into the definitive host occurs during this time. In the case of Trichobilharzia adamsi the adult worm becomes mature in approximately 50 days from the time of exposure of the host, under laboratory conditions. With other forms there may be a variation within approximately 20 days on either side of this figure. When the worm reaches maturity eggs are deposited if it is a female and these reach the water with the host faeces. It may take approximately three or four days for the eggs themselves to become fully mature before they are released by the female (Price, 1931). The egg gives forth a miracidium which may be free-swimming for a very short time (similar to the cercariae), and this penetrates a snail. Cercariae usually emerge from the snail from approximately 50 days after the mollusc has become infected. The above developmental times have been determined in the laboratory in the past by many workers.

However, under natural conditions in the field the life-cycles are not so direct. If we begin the cycle in the case of T. adamsi with the free-swimming cercaria on August 4,
on the basis of the above chronology the adult would be mature on approximately September 23 and miracidia would hatch three days later. The latter would infect snails and the cercariae, according to laboratory data of other workers, would escape on approximately November 15. This is not the case. Dermatitis-producing cercariae are found annually and their periods of emergence vary only slightly from year to year. It is obvious then that the cycle is retarded at some point. It remains now to consider the possible points of retardation.

The life-cycle of the Trematodes could be slowed down at two points:

1. development of the adult in the definitive host.

2. development of the sporocysts and cercariae within the snail intermediate host.

If the maturation of the adults in the definitive host is retarded the snails will become infected with miracidia during the spring, the adults "wintering" in the definitive host. Now, if the snail is to be infected in the spring and if normal development of the sporocysts and cercariae is to occur, the miracidia will have to penetrate the snail near the first part of June. This would allow the cercariae to emerge at the time at which they have been observed (July). It is possible that the snails become infected earlier than June and a certain amount of retardation of development occurs in the snail, to allow the cercariae to escape later in the summer.

If the larval development of the Trematodes is re-
tarded in the intermediate host stage of the life-cycle, the snails must become infected in the Fall. The infection would be carried over the winter in the snail host. However, all of the snails found to be infected with dermatitis-producing Schistosomes during the present investigations were snails which had not been carried over from the previous year. They had all hatched during the spring of the same year that they were found giving off cercariae. It is therefore obvious that they must have become infected at some time during the spring, prior to the first week of June.
PLATE IX

Contour map of Cultus Lake, B.C. illustrating the relative position of the collecting area C16 to important landmarks.
PLATE X

Enlargement of square C16 shown in Plate IX
illustrating relative positions of specific collecting areas.

- Cercaria 10; spacing indicates concentration of infection.
- Cercaria of *T. adumsi*.
- *Cercaria chilliwackensis*.
SCHISTOSOME-DERMATITIS EXPERIMENTS.

The cercaria of Trichobilharzia adamsi, Cercaria 10, Cercaria chilliwackensis and Cercaria 16A were investigated experimentally to determine their ability to produce Schistosome dermatitis. All were found to be dermatitis-producers. A description is given below of the procedure used to determine this characteristic of the various types of cercariae.

The Cercaria of T. adamsi.

On August 4, 1953, a few drops of water containing cercariae were applied with a dropper to the flexor surface of the author's right arm. Within five minutes a prickling was felt on this area which gradually increased in intensity. At this time the area on the surface of the arm was rubbed vigorously with the palm of the left hand and the "itching" subsided. One day later no macules were present. The same cercariae were applied to the left arm of a woman co-worker and she felt an intense itching within a few minutes. One day later eight small papules were present on this region of her arm. This woman in the past had always shown a very marked reaction to dermatitis-producing cercariae.

**In addition see Experiments with "Fend-E".

Cercaria 16A.

On September 4 a few drops of water containing these cercariae were applied to the flexor surface of the author's left forearm. The exposed region was held near a light. Within ten minutes an intense prickling sensation was felt
which lasted a few minutes. The next day a large red, raised area plus two red papules were present on this region and these itched when rubbed lightly, in a manner suggestive of mosquito bites. Four days after exposure the papules and raised portion had subsided leaving only a slight pigmentation and elevation of the skin.

**Cercaria 10.**

A few drops of water containing cercariae were applied to the flexor surface of the author's left forearm. Two such attempts were made to produce a reaction but neither resulted in any reaction whatever. The cercariae were then applied to the forearm of the co-worker mentioned above. Within a few minutes she reported a prickling sensation which increased in intensity. At this point she rubbed the area vigorously with formalin and no further reaction was noted.

Later, some drops of water containing these cercariae were applied to the author's right forearm and within five minutes a severe prickling sensation was felt which grew in intensity. Three hours later at least four macules were apparent on that region of the forearm. Simultaneously, the cercariae were applied to the forearm of the author's co-worker. An itch resulted which, two hours later, resulted in the formation of two papules.

One of the snails giving off Cercaria 10 was dissected and some of the cercariae thus obtained were applied to the right forearm of the author. Within ten minutes a prickling was felt and within one hour three macules were observed on this area. An occasional prickling was still noticeable a few
hours after application of the cercariae.

It is curious to note that a dermatitis was not produced on the author's left arm in the first experiment with this cercaria while later, using exactly the same method, the reaction was produced consistently on the other arm.

Twelve days after the above experiments with the cercaria had been performed a few small raised portions which were not pigmented were present on the flexor surface of the author's right forearm. These did not produce an irritation at this time.

*Cercaria chilliwackensis.*

The ability of this cercaria to produce a dermatitis was investigated by applying a number of the cercariae in a drop or two of water to the flexor surface of the forearm of the author and the author's co-worker. The cercariae produced a fairly intense itching on the co-worker's arm, followed by two papules which lasted for approximately ten days. The cercariae on the author's arm produced a fairly intense itching when the water had nearly completely dried. Two papules followed which lasted well over three weeks.

*Experiments With "Fend-E".*

This substance, which is sold as a dirt repellent for miners, was investigated to determine whether it would inhibit penetration of the dermatitis-producing cercariae. It was found to be capable of preventing a dermatitis when the cercaria of *T. adamsi* was applied to the region of the
arm which had been coated with it. However, it was discovered that the substance would wash off in running tap-water. Thus, it was considered impractical for use as a Schistosome dermatitis preventative. It was later learned that there exists a similar repellent on the market, "Fend-I", which is insoluble in water, and future experimentation with this substance may prove fruitful.
DISCUSSION.

As has been previously mentioned, an undescribed cercaria was discovered at Cultus Lake in 1951, and this led to the present investigation. It is necessary to consider at this time the possible reasons for the cercaria having failed to turn up during the course of the recent work. Three possibilities exist for the explanation of the situation:

1. Due to random selection of the snails collected in the areas, snails harboring the cercaria were by chance missed.

2. The 1951 cercaria was restricted to a small area. This area was not described specifically by previous workers and the 1953 collections failed to sample the area.

3. The cercaria was erroneously described, the description consisting in reality of a composite of two or more of the cercariae described in the present work.

The first two possible explanations infer that the cercaria previously described does in fact exist. If the first possibility outlined above is true, then certainly infections of snails by this cercaria are extremely scarce, more so even than were the infections described at the present time. However, S.M. Sager in his investigations of the area in 1951 reported the incidence of infection of this cercaria at that time to be of the order of 6%, a figure which is much in excess of the results of the present investigation.
The second possibility is perhaps more reasonable. The accounts of the discovery of the unknown cercaria of 1951 state that it was found in C16, the area "in toto" in which the present investigations were carried out. No trace of descriptions of specific sub-areas where this cercaria was found are available. In view of the fact that the Trematodes described in the present work were found to occur in specific limited areas, it seems probable that the 1951 cercaria could be easily missed. The sub-areas in which the present cercariae were found included a total area of no more than one third of the entire C16. Admittedly, snails were collected from most of the entire area in question, but those collected from regions north of the "South end of beach" area were for the most part juveniles and never gave off cercariae of any type.

The third and last possible explanation for the absence of the 1951 cercaria infers that the cercaria as described by the other workers does not exist. This would mean either that some other cercaria (possibly one of the three described in the present work) was incorrectly described, or that the description put forth was actually a composite of two or more different forms. The only possible explanation for this latter occurring, to the author's mind, is that the great amount of emphasis put upon the possession of papillar spination by this first cercaria may have led to the conclusion that only one species was being dealt with. However, the cercariae described at the present time do not appear closely similar morphologically, even at first glance. It seems unlikely that the previous workers would mistake them for a single species.
Upon consideration of all the factors involved, it seems most probable that either the first or second of the possible explanations outlined above explains the failure to uncover the 1951 cercaria during the summer of 1953. In other words, in the author's opinion, it is highly likely that the cercaria described as being tentatively new in 1951 still exists as such and has not been found again. Continued investigations of the types of Schistosome cercariae present in the area in question should be made in succeeding years and the 1951 cercaria should not be considered non-existent with any degree of certainty until two more such investigations representing two summers work have failed to reveal it.
SUMMARY.

Three dermatitis-producing cercariae, discovered at Cultus Lake during the summer of 1953, have been described. In addition reference has been made to the existence of another very similar cercaria in this region. Two of the Trematodes are described as new species, while two may be new. The position of the latter is left open to question due to a lack of description of the adults.

Attempts were made to discover the life-cycle of each of the three Trematodes. Initially only the cercarial stage was known. Two approaches were adopted. Animals were trapped in the field and examined for adult Schistosomes; and laboratory animals were exposed to the cercariae in attempts to infect them. All examinations of animals from the field proved to be negative.

The major portion of the life-cycle of one of the cercariae was established experimentally and the Trematode, described as a new species, has been named Trichobilharzia adamsi. In the laboratory two Pekin ducklings were exposed to the cercaria of T. adamsi and later one immature adult female was removed from the liver of one of them. Established in this life-cycle at present are the female adult, sporocyst and cercaria, as well as the two hosts involved (the natural adult host has not been determined). Trichobilharzia adamsi is a member of the "ocellata group".

Cercaria chilliwackensis, diagnosed as a new species, was exposed in the laboratory to white mice, pigeons and one Pekin duckling. Later examination proved that no cercariae had penetrated and reached the adult stage. Cercaria chilli-
wackensis is a member of the "douthitti group" and appears very similar to C. tuckerensis Miller 1927.

Cercaria 10 was exposed to white mice, one Guinea pig and two Pekin ducklings. All animals were found to be negative when later examined for the presence of the adult worm. Cercaria 10 is a member of the "spindalis group" and is very similar, if not identical with, the cercaria of Gigantobilharzia huronensis Najim 1951. No definite determination of the position of Cercaria 10 could be made. It was therefore deemed advisable to leave the matter open until the adult is known and further comparisons can be made.

All three cercariae, Cercaria 10, Cercaria chilliwackensis and the cercaria of T. adamsi possess a type of spination which is distinct from the usual type of small spination found on the body of Schistosome cercariae. This has been called "papillar spination" and has been previously described only by Najim (1951) on non-human dermatitis-producing Schistosome cercariae. The spines have been found very infrequently by certain other workers on some of the other Schistosomes. A description of this spination has been given in the present work. Two important phenomena were discovered with relation to the occurrence of the papillar spines on the Cultus Lake Trematodes. The papillar spines are present in a very similar pattern on the body of all of the cercariae described. Further, it was established that the spination is carried through from the cercaria to the adult of T. adamsi and for the most part, the location pattern is maintained in the adult. As far as can be determined, this represents the first time that papillar spination has been described as occurring on an adult Schistosome.
Ecological studies of the snail hosts of Cercaria 10, Cercaria chilliwackensis and the cercaria of T. adamsi revealed that the snails giving off the individual types of cercariae were confined to specific areas. Only in one instance did these overlap. In this case the occurrence of Cercaria 10 was replaced later in the season by the cercaria of T. adamsi. It is believed that the infection of the snail intermediate hosts of the Trematodes occurs at some time during the spring, prior to June.

An outline has been given of the methods adopted in determining the dermatitis-producing characteristics of the Cultus Lake cercariae described in this work.

A modification of the perfusion technique of Yolles et al, is described. This was used in attempts to recover any adult Schistosomes which might have been present in the experimental white mice. The reasons for the adoption of these modifications are given. They enabled the author to perform the necessary perfusions of the livers while at the same time maintaining a constant pressure in the perfusion apparatus. The latter was necessary since no source of constant air-pressure was available and a hand-bulb had to be used.

A new technique has been described for concentrating large numbers of Schistosome cercariae when the number available through ordinary means is limited. This technique involves the use of a side-arm flask and utilizes the taxes of the cercariae. Cercaria 10, which is especially responsive to a light stimulus, could be concentrated in extremely large numbers in the side-arm of the flask.


101.


