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### A STUDY OF SCHISTOSOMIASIS IN THE MIZURATA AREA, LIBYA

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by

Abubaker I. Swehli

A Thesis Submitted to the Faculty of The Graduate College in partial fulfillment of the Degree of Master of Arts

Western Michigan University Kalamazoo, Michigan August 1979

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Abubaker I. Swehli

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### INTRODUCTION AND LITERATURE REVIEW

#### Libyan Geography

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Libya is situated in North Africa (Fig. 1) and has a Mediterranean Coastline of 1900 kilometres. It extends over a vast territory inland from the Central Mediterranean Coast of North Africa to the highlands of North Central Africa. To the North it is bounded by the Mediterranean Sea, to the east by Egypt and Sudan. To the south it borders Niger, Chad, and Sudan. In parts of the south and west it touches Algeria and in the northwest Tunisia. Libya has a particularly strategic position, for it is a link between Africa and Europe as a potential dispersal route for schistosomes and their vectors.

The size of Libya is 1,760,000 square kilometers. It is the fourth largest country on the African continent. This area is composed of coastal plains, hills, mountains and deserts. The Libyan coastal plain has the most fertile lands and highest density of population.

The climatic conditions are influenced by the Mediterranean Sea to the north and the Sahara in the south. The country has a temperate climate, but the temperature is higher toward the south and is lower in the mountainous regions.

The Libyan population is small for so large an area. According to the 1973 census, the population was 2,257,000. The number rose to 2,683,000 by mid-1975.





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#### History of Schistosomiasis in Libya

Kadiki and Khan (1972) reported that 168,275 persons in Libya were infected with schistosomiasis. Schistosomiasis has existed in the southern region of the country for a long time. The first case of schistosomiasis as well as the presence of <u>Bulinus contortus</u> near Ghat (Kadiki and Khan 1972) was recorded in 1925 (Fig. 2).

Zavattari (1932) in a general survey of the southern part of Libya reported on the distribution of <u>Bulinus</u> <u>contortus</u> and urinary schistosomiasis. The same year, Lodato (1932) reported the presence of <u>Bulinus contortus</u> and three occurences of <u>S</u>. <u>haematobium</u> in different areas of the same region.

In (1935) Giardano reported a case of urinary schistosomiasis in a new area (El Barket). Impallomeni (1937) mentioned the presence of twenty cases of urinary schistosomiasis at Brak and Ubari (Fig. 3).

Scaduto (1937) reported the same disease at Morzuk. Nastasi (1938) made a good survey in the southern region. He was the first one to find <u>Biomphalaria</u> snails in the Fezzan area. Vermeil and Tournoux (1952) confirmed the presence of <u>Biomphalaria</u> snails at Fezzan, and he did an excellent survey of <u>Bulinus contortus</u>.

Godwin (1957) reported the presence of the disease in another new areas. Berry (1963) reported two species of <u>Bulinus</u> in the Fezzan area including the previously noted B. contortus.

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### MAP OF LIBYA



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Taworga is very well known as an area with intestinal schistosomiasis. Kadiki and Khan (1972) said that Muller and Feugain 1952 were the first to report the presence of <u>Biomphalaria</u> snails in the Taworga area. Godwin (1957) made a survey and identified the snails as <u>Biomphalaria</u>. He also examined the stool samples of 126 persons, and found that eight were infected with <u>Schistosoma mansoni</u> (7% infection rate). At the same time he examined the urine for urinary schistosomiasis but results were negative. He was the first to report the presence of <u>Schistosoma mansoni</u> in this area.

Berry (1963), and Yasuraoka (1966) collected <u>Biom</u>-<u>phalaria</u> snails, and determined that they were <u>Biomphalaria</u> <u>alexandrina</u>.

Halawani (1966) examined residents of the area and found that the infection rate with <u>Schistosoma mansoni</u> was about 30%. This may not be precise due to the small number of people tested (63 persons). In 1967 Halawani made another survey. He found <u>S. mansoni</u> eggs in thirtyone percent (31%) of stool samples from sixty-three persons.

In 1971 El-Gindy made a population survey for rectal schistosomiasis of 2,723 people and found 643 infected (24.7%).

In 1972 Kadiki and Khan, indicated an infection rate of seventeen percent (17%) for <u>S. mansoni</u> in the Taworga area. Amin (1975) mentioned in his report that the use of Byluscide as a molluskacide treatment was unsuccessful. He recommended building a fence around Taworga spring to prevent personal contact.

Derna is another focus of schistosomiasis, where the first cases of urinary schistosomiasis were reported in 1955 as diagnosed by the central laboratory of Derna hospital. Berry (1963) reported the presence of <u>S</u>. <u>haematobium</u> without referring to the snail host. Hamami (1965) found 20 cases of urinary schistosomiasis, and he found <u>Bulinus</u> snail in Ain Dabusia. Halawani (1966) examined many populations for the disease, and he found a few were positive. He also found <u>Bulinus</u> in a second location which he did not specify.

#### Schistosomiasis

Schistosomiasis is often called bilharziasis. It is a parasitic disease caused by a flatworm which uses humans as final, definitive hosts. The number of people infected with this disease ranges from 114 million to 200 million. The infected people live mostly in agricultural communities in many different parts of the world. There is no known way of making people immune to the disease.

In 1851 Dr. Theodore Bilharz, who was Professor of Anatomy at the medical school in Cairo, was the first to

discover this disease. The name bilharziasis commemorates this discovery. In 1858, a Dr. Weinland of Harvard University renamed the worm <u>Schistosoma</u>, which means split body. The body of the male worm is split, divided, or folded over to form a groove or channel in which the female lies throughout its adult life. Today, the disease is known as schistosomiasis in South America and in the Far East, and as bilharziasis in Africa.

#### Human Schistomomiasis

The human schistosomes, or blood flukes, are digenetic trematodes. Three different types of schistosomes can cause disease in humans. <u>Schistosoma haematobium</u> is the urinary type of disease and is distributed widely in Africa. <u>Schistosoma mansoni</u> is the intestinal type of the disease and is also endemic in many parts of Africa. <u>Schistosoma japonicum</u> is the eastern type and causes the disease in China, Japan, and other countries of the far East but is not established in Africa.

A schistosome trematode needs an intermediate host, a snail, for completion of its life cycle. The snail host is different for each type of schistosome. In the case of <u>Schistosoma haematobium</u> (the urinary type) it needs any species of the genus <u>Bulinus</u>. <u>Schistosoma mansoni</u> (the intestinal type) requires a species of <u>Biomphalaria</u>. In the case of <u>Schistosoma japonicum</u> the intermediate host is a species of <u>Oncomelania</u>. <u>Schistosoma haematobium</u> and <u>S. mansoni</u> are widely distributed in North Africa including Morocco, Algeria, Tunisia, Libya, and Egypt. Both species are found in other African countries, but <u>S. haematobium</u> is more widely distributed.

#### A. Schistosoma mansoni

In 1902, Manson found lateral spined eggs in the feces of a patient from the West Indies. He mentioned the existence of a second species of blood fluke. Looss (1905) disagreed and explained that the lateral-spined eggs were <u>S. haematobium</u> eggs produced parthenogenetically in the absence of males. Sambon (1907) opposed the hypothesis of Looss and named the new species after Manson. Leiper (1915) discovered the new intermediate host in Egypt.

### B. Schistosoma haematobium

This species produces a chronic disease characterized by the presence of blood in the urine, and disease of the bladder. This has occurred in Egypt since the time of the Pharoahs as shown by the calcified ova found in the kidneys of two Egyptian mummies. The symptoms of the disease were found among the French troops during their invasion of Egypt in the beginning of the 18th Century.

Leiper (1915) identified the intermediate host of <u>S. haematobium</u> in Egypt by exposing mice to cercariae from <u>Bulinus</u>, and demonstrated that the mice developed worms which produced terminal-spined eggs. The River

Nile was the original center of <u>S</u>. <u>haematobium</u> distribution and from there the infection has spread to new regions.

### C. Schistosoma japonicum

Fujii (1847) was the first one who described the disease in Hiroshima, but Warren (1972) in his book mentioned that in 1883, Baelz, who worked in Japan, was the first one who mentioned this kind of disease in a language other than Chinese or Japanese. Yamagiwa (1890) found ova in a human brain. Katsurado (1904) named the species <u>Schistosoma japonicum</u>, and gave an accurate description of the eggs found in humans and cats. He found that dogs, horses, cattle, and cats were capable of being hosts of the parasite. Faust and Meleney (1924) discovered the intermediate host, <u>Oncomelania</u>, in China.

Kuntz et al (1976) studied the morphology of the adult by using electron microscopy, and observed a difference in the surface structures of the male and the female. The surface of the male is rough while that of the female is smooth. There are spines in the oral suckers and the acetabulum of both sexes which help them to attach to the blood vessels of the definitive host. The gynecophoral canal is also roughened by minute spines. The Life Cycle

The adult male and female are generally paired. The adult female lies in the gynecophoral channel of the male,

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into which sperm seem to be extruded through a pore below the ventral sucker. The sperm can get into the female duct and accumulate near the ovary until fertilization of eggs occurs (Kitajima et al 1976).

The female lays chains of small bead-like eggs, in the mesenteric or pelvic veins of the human. These eggs, containing larvae, go into the bladder or intestine, and according to the type of the worm are passed out of the body with urine or feces.

In warm water after from one minute to sixteen hours each egg hatches into a miracidium. Light plays an important role in the hatching process (Bair and Etges, 1973). Kassim and Gilbertson (1976), however, found that there is no significant difference in hatching in light and in darkness, and that the ionic composition of the medium does not have an effect on hatching.

The miracidium is a larval form with hair-like projections, cilia, that enable it to move through water. The miracidia swim and search for a particular kind of snail where they may continue development. They have just 24 hours to find one. Mason and Fripo(1977) indicate that changes of the reactions of miracidia to light are related to changes in temperature. Wright (1971) found that the movement of the miracidia to the water surface is a positive response to light and a negative response gravity. Objects in the water which reflect sunlight

will attract the miracidia to their submerged edges (Cherin, 1974). The motility of miracidia is dependent on the presence of Na<sup>+</sup> in the medium (Kassim and Gilbertson, 1976).

When the miracidia reach suitable snails they attach themselves to the soft tissues of the head, foot, or tentacles, bore into the snails, and migrate to the digestive glands. There they grow into sporocysts which are hollow, sac-like organisms with germ cells on the outer wall. Each sporocyst buds off a number of secondary sporocysts (about 30) similar to the first. After a while, each secondary sporocyst migrates to the snail's liver where it produces about 40 forked-tail cercariae. The cercariae leave the snail and swim in the water. They have 24 to 36 hours to find a human host before they die. The cycle from egg to cercaria takes place in from 2 to 12 weeks. Each egg can potentially produce approximately 1200 cercariae.

When the cercaria find their final host, they attach themselves to the skin by suckers: and shake off their tails. About 15 minutes is needed to pass into the blood vessels under the skin. Then they migrate to the liver where they become adults. From the liver they migrate to the veins of the bladder or intestines where they lay eggs, and thus start the cycle again.

The Snail Host

<u>Biomphalaria</u>, the intermediate host for <u>S. mansoni</u>, is a fresh water snail with a flat discoidal shell of from four to six whorls. It's blood is red with dissolved haemoglobin. The snail is hermaphroditic and does not need to copulate to reproduce since self fertilization can occur. This snail can survive in the muddy bottom of a spring or an empty pool. Then, when it refills, the hiding snail can establish a new population.

The shell, discoidal and biconcave, forms a disc varying in height with a diameter between 7 - 22 mm. The whorl numbers vary with different species from three and a half to seven. When few whorls are present the whorls increase in width rapidly. But in those forms with many whorls, they are more closely coiled and gradually increase in width. The whorls may be rounded or angular. The umbilicus under the center of the whorls may be wide and shallow or narrow and deep.

The color of the shell is variable, but in general it is brown, white, and in some cases reddish. The shell is slightly sculptured with curved, transversely arranged growth lines.

The genus <u>Biomphalaria</u> can be divided into four species, <u>B. pfeifferi</u>, <u>B. alexandrina</u>, <u>B. choanomphala</u>, and <u>B. sudanica</u>. The two species that are found in Libya are <u>B. pfeifferi</u>, and <u>B. alexandrina</u>.

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<u>Biomphalaria</u> <u>alexandrina</u> (Ehrenberg): The shell is about 15 mm in diameter when full grown. It consists of five to five and a half whorls, rounded or angular.

<u>Biomphalaria pfeifferi</u> (Krauss): The shell is less than 15 mm in diameter when full grown with less than five whorls, which are angular or convex on both sides. The diameter of the shell in most cases is about two and a half times the height of the last whorl. The diameter of the umbilicus is usually smaller than the height of the last whorl.

Charles (1977) reported that increasing <u>Schistosoma</u> infection in <u>Biomphalaria</u> snails is due to age rather than multiple exposure. Newton (1953) noted the occurrence of a higher frequency of infection with <u>S. mansoni</u> in juvenile rather than adult <u>Biomphalaria</u> snails.

Liang (1974) found that if pH exceeded 7.8, then the snail egg-laying ceased. At the same time, he didn't find any effect of calcium or magnesium on the growth rate of the snails. Harrison and Shiff (1966) had indicated that a high level of calcium or magnesium can lower the egg-laying potential.

Harry et al (1957) found that <u>Biomphalaria</u> snails had an optimum chloride concentration of between 20 - 110 ppm. Malek (1958) observed that <u>Bulinus</u> was less tolerant of salinity than Biomphalaria.

Harry et al (1957) found that the optimum concentration of sulfate for <u>Biomphalaria</u> snails was 10 - 80 ppm (as  $SO_4$ ).

A three year study by Van der Schalie and Berry (1973), on the effect of temperature on pulmonate snails, indicates that no reproduction occurs when the temperature exceeds  $30^{\circ}$ C.

#### MATERIAL AND METHOD

#### The Study Area:

The areas where a total of fourteen springs were examined in the Mizurata region include:

- 1. Taworga Area: The main spring Fig. 3
- 2. El Heesha Area:
- Ain El halumia
- Ain Zreik
- Ain Emheide
- Ain Eljemal
- Ain Omelefyone
- Ain Omeletheil
- Ain Esala
- Ain Hassan Fig. 4
- Ain Zayed
- Ain Slaia

3. El Mafruth Area:

- Ain Buhashesh
- Ain Elmalha
- Ain Naji

(Note: Ain = Spring in Arabic)

# FIG. 3

TAWORGA SPRING







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#### Collecting Technique

The main water supply in Taworga is a large old spring, (Fig. 3) with warm water all the year round, from which one main river and a few small canals originate. The main river is also divided into several streams supplying many villages with water, and the streams have many branches which end in a vast area of marshes.

Snails were collected in large numbers from the shallow borders of the spring with an aquatic dip net. A boat was used for access at the inlet of the main rivers, and for some stream and marsh sampling.

Because water is taken during day time for irrigation, most of the snails were found under rocks along the shore of the spring. At night or in the morning, the water level in the spring reaches maximum, and drops during the day, leaving snails behind under rocks where they are then found.

In contrast, at El Heesha, and El Mafruth springs, most of the snails were found beneath the surface film of the spring, where algae form a surface mat or scum (Fig. 5). At each spring ten to fifteen minutes were spent in each of three locations. All snails in the net or on the objects examined were collected. Thus data on snail numbers should reflect their proportional abundance within springs, and should approximate their relative abundance in comparing springs, even though density per



SNAILS CONCENTRATED FROM SWEEPS WITH A COLLECTING NET

FIG. 5

unit area was not determined. Samples of plants were also collected for identification.

Snails were put in containers with a small amount of water from the spring for live transport to Tripoli for laboratory examination.

### Examination of Snails for Cercariae:

Snails were examined in the following manner in the laboratory:

They were washed, and <u>Biomphalaria</u> snails were separated from the other species and put in a wide-mouthed container. The container was covered, and water was changed at two days intervals and the snails were fed with fish food. The containers were examined for cercariae in the morning, afternoon, evening, and at night. When cercariae were found at the surface, snails from the containers were crushed gently, the broken shells removed, with dissecting needles under a dissecting microscope, and a search for the presence of additional cercariae was made in the macerated bodies.

#### Physical and Chemical Observation:

Observations of temperature, pH, turbidity, dissolved oxygen, and conductivity were made. The mean of five measurements, was obtained at each spring. The measurements were from locations near the surface at four areas around the edges, and one in the middle.

Air temperature averages reported for each area, and

water temperatures were measured with a laboratory type mercury thermometer.

pH measurements were made with a portable pH meter.

Turbidity, dissolved oxygen, and conductivity were measured with a portable Japanese Martek Hariba instrument, provided by the Center for Marine Research in Tripoli.

Additional water analysis was done on water samples from Ain Taworga and Ain Zayed, which yielded the largest collections of Biomphalaria snails.

A gallon (approx. 3.8 liters) of water from each of the two springs was taken to the Central Laboratory, Ministry of Public Health, in Tripoli, where  $Ca^{++}$ ,  $Mg^{++}$ ,  $Na^+$ ,  $K^+$ ,  $Cl^-$ ,  $F^-$ ,  $So_4^-$ ,  $NO_3^-$ , total hardness, and alkalinity were determined by standard chemical methods.

### Biological Observation:

<u>Biomphalaria</u> snails were measured, using the method of Mandahl-Barth (1957). The position of these measurements is shown in Fig. 6. Species determinations were based on the collections in Table 5, derived from the work of Gelfand (1967), and from consultation with Dr. A. Clarke in the National History Museum, Washington D.C, and Mr. Hans Wurzinger in the Museum of the University of Michigan.

#### Statistical Evaluation

A corner test for association (Dixon and Massey



1957) was used to check for significant relationships between percent or number of <u>Biomphalaria</u> snails and several other environmental variables. The test is a simple non-perametric procedure which is readily applied to data that can be ranked, using two variables at right angles to each other. A sign is applied alternately, positive, then negative, to the four quadrants formed by the vertical and horizontal lines through the median points of the rankings, counts are made along each axis and from each direction until data points of changed sign are encountered. The sum of the four counts is compared to a critical value for the sample number at the desired level of significance. Sums exceeding the critical value indicate a significant correlation, either positive or negative as indicated by the sign.

The critical value at 95% level with a sample size of 14 is 11.

T tests of mean infection rate at different ages was performed on the Western Michigan University Computer Center computer using their Statpack program.

### Epidemiological Study:

Stool samples were collected and examined for eggs to determine presence of infection with schistosomiasis.

Each person was given an empty container with a lid. The people were instructed to pass the stool outside and place about 10  $\text{cm}^3$  of stool from only one person in the

container and close it firmly. Each container was labeled with the name of the person, and the date of collection. Handling Samples:

# A. <u>Direct smears of stool samples were perpared as</u> <u>follows</u>:

The slide was placed on filter paper, then two drops of saline solution were added in the center of the slide. A small piece of the fecal sample was stirred into the saline, and the gross particles were removed with a needle. A cover-slip was added prior to examination for eggs under the microscope at 100 X and 430 X.

#### B. Sedimentation

About one gram of the stool sample was placed in a 50 ml beaker and 2 ml of normal saline solution were added. Then it was beaten until the stool sample was broken into small particles. The contents of the beaker were poured through a sieve while stirring, and then funneled into a test tube.

More saline solution was added until the test tube was filled. It was then allowed to sediment for half an hour, then the supernatant was decanted. The sediment was poured to a petri-dish, the test tube was washed with saline, and the washings were added to the petri dish. The solution was then allowed to settle for five minutes and examined under a dissecting binocular microscope. Each stool sample was examined twice, first by direct smear and then by sedimentation in normal saline. Description of the Epidemological Study Area:

Mizurata city is 207 kilometers to the east of Tripoli, and is known as an endemic focus for <u>Schistosoma</u> <u>mansoni</u>. The focus of the infected area appears to be Taworga and El Heesha springs, because of the presence of <u>Biomphalaria</u> snails in those areas. These snails are the intermediate host of <u>Schistosoma mansoni</u>.

The small village of Taworga is about 50 kilometers south of Mizurata city. It is reached by following the highway toward Benghazi city for thirty five kilometers, then turning left and proceeding fifteen kilometers (Fig. 7).

At the present time there is an Egyptian Land Reclamation Company (El Bahaira) working on an agriculture project to transfer the water of the old spring fifteen kilometers to the west for irrigation of an area of 8000 acres (320 hectares) that will contain 150 farms. Most of the farmers in the labor force are from Egypt.

The main water resource in Taworga is derived from an underground origin consisting two internal spring areas near each other which form the one big spring called Ain Taworga.

The depth of the spring, in the area away from the upwelling, is up to 10 meters, whereas the internal spring



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areas have depths of 300 to 400 meters. The output of the spring is over  $3.0 \text{ m}^3/\text{second}$ . The temperature of the spring ranges from  $25^{\circ}\text{C}$  to  $27.1^{\circ}\text{C}$ . The spring is heavily vegetated.

Water from the spring is pumped into high cement canals to irrigate the agricultural project. Fifty percent of the water is used in irrigating an old agricultural area for the growth of the <u>Juncus</u> plant. People living around the spring take their water supply directly from the small streams originating from the spring. Most of the streams crossing the small villages are heavily populated with <u>Biomphalaria</u> snails. Children and women stay for long times in the water close to the spring, using it for washing dishes, washing clothes, and other domestic purposes.

Each stream gives off many branches as it crosses villages, and the branches then break to form several small canals to irrigate palm-trees. The marshy plant, <u>Juncus</u>, which is the main source of income, grows along most of the wet land resulting from this irrigation. The adults collect the <u>Juncus</u> and the women process them into mats and some other hand-crafted articles (Fig. 8, 9, 10). This, and raising sheep, goats, donkeys, and camels, are the principal agricultural activities.

The Taworga area is inhabited by many tribes. The majority are settled tribes, living in houses built of

parts of palm-trees. A few nomadic tribes tent and move from one place to another with their animals as they look for <u>Juncus</u> plants, which they call "deese". The total population of the Taworga area was 11,389 persons in the 1964 census, but the estimated population of the area in 1971 was about 16,000 persons. The number of tribes and population in the area are not constant because of their movement.

El Heesha, the second location studied (Fig. 7), is an oasis reached by following the highway from Mizurata to Benghazi for 95 kilometers, then turning left and proceeding for 17 kilometers on a difficult road. The area is marshy, with salty soil. It lies below sea level, with many salt lakes. Many fresh water springs are distributed in this area. The inhabitants use these springs for many domestic purposes including drinking water, washing, swimming, washing dishes, and children playing (Fig. 11 & 12). There are a few houses made from palm-tree materials that grew in the area.

The inhabitants in this area number about 699 persons (1964 census). They also make mats from the <u>Juncus</u> which is abundant around the springs. Many of these people migrate from one place to another to cultivate the <u>Juncus</u> plant. By their movement with their animals they can infect another spring with <u>Biomphalaria</u>, and these snails may carry schistosomiasis if it is not already there.

# A WOMAN TRANSPORTING JUNCUS PLANT









FIG. 10 THE USE OF JUNCUS IN HAND-CRAFTS



CHILDREN WADING IN THE INFECTED WATER OF AIN ZAYED



FIG. 12

DISHES LEFT BY PICTURE-SHY WOMAN WHO WAS CLEANING THEM IN THE SPRING



El Mafruth area is about 8 km south of El Heesha, and has many springs, but no houses. The people live in many tents around the springs (Fig. 13). Few of them come from Taworga; most come from the south of the country.

There is a project for building houses for all the nomadic people. But it receives little acceptance by them because they believe by moving into houses they will loose their freedom in life. They want to be near their animals, and move with them anywhere they choose to move.



# FIG. 13

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#### RESULTS

Ten infected springs contained <u>Biomphalaria</u> snails (Table 1), six had <u>Biomphalaria</u> <u>alexandrina</u> and four <u>Biomphalaria</u> <u>pfeifferia</u>. Most of these also had large quantities of <u>Melanoides</u> <u>tuberculata</u>. A few springs contained <u>Physa</u> <u>acuta</u>.

All springs had at least one species of snail present (Table 1). Numbers of individuals of <u>Biomphalaria</u> were not significantly correlated with numbers of other snails as measured with a corner test for association (Table 2, Fig. 14 ).

The plants in Taworga spring included: <u>Juncus</u>, filamentous algae, <u>Chara vulgaris</u>, and <u>Zanichella</u>. The plants from springs of the El Heesha area were: <u>Juncus</u>, <u>Chara</u> <u>vulgaris</u> and <u>Phragmitis</u>. No plant collections were made in the El Mafruth area.

#### Incidence of Cercaria in Snails

Snails from three springs from areas most heavily infected with <u>Biomphalaria</u> snails (Taworga spring, and from the El Heesha area, Ain Zayad and Ain Hassan) were examined for cercariae.

1. <u>Taworga Area</u>:

One hundred five of the <u>Biomphalaria</u> snails collected from the main spring (Ain Taworga) were placed into five containers, each with twenty or twenty five snails. These containers were checked four times a day for cercariae.

## TABLE 1

SNAILS FROM SPRINGS OF MIZURATA AREA

Name of the Spring	<u>Biompha</u> - laria alexand- rina	Biompha- laria pfeif- feri	<u>Mela-</u> noides tuber- culata	Physa acuta	Lymnaea	*Others	Total
1. Taworga Area							
- The Main Spring	589	-	35	20	6		650
2.El Heesha Area							
- Ain El Halumia	4		36	-	-		40
- Ain Zreik		13	117	-	-		130
- Ain Em Heide	15	-	70	-	-		85
- Ain Eljemal	40	-	56	<b>e</b> 7	-		96
- Ain Omelefyone	-	-	278	11			289
- Ain Omeletheil	-	-	294	47	-		341
- Ain Esala	-	14	48	-	-		62
- Ain Hassan	310	-	80	-	-		390
- Ain Zayed	550	-	90	35	-		675
- Ain Slaia	-	168	150	-	-		318
3. El Mafruth Area							
- Ain Buhashesha	-	-	-	disa)	-	34	34
- Ain Elmalha	-	-	134		-	16	150
- Ain Naji	-	7	57	4	-		68
Total	1,508	202	1,445	117	6	50	3,328

\*These snails belongs to Family Ellobiidae.

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### TABLE 2

# NUMBER OF SNAILS FROM MIZURATA AREA

Name of Spring	No.of Snails Collected	No.of Biomphala- ria Snails	No.of Other Snails	% of <u>Biompha</u> - laria
Ain Taworga	650	589	61	90.6
Ain Elhalumia	40	4	36	10.0
Ain Zreik	130	13	117	10.0
Ain Emheide	85	15	70	17.6
Ain Eljemal	96	40	56	41.6
Ain Omelefyone	289	0	289	0.0
Ain Omeletheil	341	0	341	0.0
Ain Esala	62	14	48	22.5
Ain Hassan	390	310	80	79.4
Ain Zayed	675	550	125	81.4
Ain Slaia	318	168	150	52.8
Ain Buhashesha	34	0	34	0.0
Ain Elmalha	150	0	150	0.0
Ain Naji	68	7	61	10.2
	i	1		



The quadrant sum (S) for this test does not reach the value of 11 required to show significant association at the 5% level for a sample size of 14.

FIG. 14

The cercariae appeared in only two containers, while the others were negative. When the snails were crushed, only two snails released cercariae, one from one positive container and one from a negative container. The resulting 1.9% rate must be a conservative underestimate of infection rates.

2. El Heesha Area:

a. One hundred forty <u>Biomphalaria</u> snails were collected from Ain Zayed and divided into five containers, twenty-eight snails per container. Cercaria appeared at the surface in only one container. When the snails were crushed, however, two were found to be infected, a rate of 1.43%.

b. One hundred thirty-two <u>Biomphalaria</u> snails were collected from Ain Hassan, and divided into five containers. Cercariae appeared at the surface of only one container. When they were crushed only one infected snail was found, a rate of 0.75%.

From the physical observations reported in Table 3 it can be seen that Ain Esala was the most alkaline spring in the areas whereas Ain Emheide was the least alkaline. The total number of snails in springs was not significantly correlated with pH when tested by a corner test for assiciation. A positive correlation was found for pH and percent <u>Biomphalaria</u> using the corner test (Fig. 15).

The springs all have adequate amounts of oxygen.

# TABLE 3

## PHYSICAL-CHEMICAL DATA FOR SPRINGS OF THE MIZURATA AREA

			~			
Name of the Spring	Air Tempera- ture	Water Tempera- ture	.∵pH	Turbi- dity	Oxygen dissolved in water	Conducti- vity
1. <u>Taworga Area</u>	32.05					
- The Main Spring		26.7	7.3	83	9	4.4
2. El Heesha Area	31.32					:
- Ain El Halumia		26.1	7.4	66	12.1	6.2
- Ain Zreik		27.1	7.1	46	9.5	6.5
- Ain Emheide		29.2	5.9	128	10.8	16.1
- Ain Eljemal		25.5	7.4	47	9.5	5.8
- Ain Omelefyone		26.9	7.1	33	11.4	6.7
- Ain Omeletheil		27.1	6.9	51	9.8	6.9
- Ain Esala		27;3	7.8	51	12.8	6.6
- Ain Hassan		27.0	7.3	18	9.5	6.3
- Ain Zayed		27.3	7.6	77	10.4	5.8
- Ain Slaia	i	27.7	7.6	64	12.3	5.9
3. El Mafruth Area	33.92					
- Ain Buhashesha		26.9	6.8	134	11.5	14.9
- Ain Elmalha		27.0	7.0	138	11.8	16.6
- Ain Naji		26.8	6.8	141	12.5	16.7



## A CORNER TEST FOR pH VERSUS PERCENT <u>BIOMPHALARIA</u>



The quadrant sum (S) value for this test exceeds the value of 11 required to show significant association at the 5% level for a sample size of 14.

Dissolved oxygen ranged from 9 to 12.8 ppm. Highest turbidity was in the El Mafruth area springs. Results of the corner test show that the percent of <u>Biomphalaria</u> is not significantly correlated with turbidity (Fig. 15) nor with dissolved oxygen.

Chemical observations are listed in Table 4 and 5. The amount of calcium (necessary for mineralization of snail shells) are second only to sodium in amounts for the metallic ions. The total hardness of the water was high. The amount of chloride is approximately double the sulfate level. The total ionic concentration is measured by conductivity. The corner test showed a significant negative association of percent <u>Biomphalaria</u> with level of conductivity (Fig. 17). The same data were tested with a Mann-Whitney non-parametric test. A U of 41 equaled the tabled critical value for the 0.05 level in Brower and Zar (1977) for the sample size, confirming the results of the corner test.

Temperature of the springs showed no significant correlation with percent <u>Biomphalaria</u> over the slight range found in the study (Fig. 18).

#### Snail Species Determination:

The mean of ten measurements was obtained from random samples of <u>Biomphalaria</u> snails from each spring containing them. The height of the snails ranged from 3.8 mm to 5 mm, while the diameter ranged from 10.3 mm to 16 mm.



The quadrant sum (S) value for this test does not reach the value of 11 required to show significant association at 5% level for a sample size of 14.

TABLE	4
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## WATER ANALYSIS FOR TAWORGA SPRING

ions	m. eg./L	mg/L
Calcium	16.46	, 328.64
Magnesium	7.58	92.34
Sodium	19.57	450
Potassium	1.00	40
Chloride	29.24	1036.6
Fluoride	-	1.9
Sulfate	10.68	512.82
Nitrate	0.34	25
Total hardness		1200
Alkalinity		205

.

## TABLE 5

### WATER ANALYSIS FOR ZAYED SPRING (AIN ZAYED)

ions	m. eg./L	mg/L
Calcium	19.69	392.8
Magnesium	16.18	196.8
Sodium	36.97	850
Potassium	1.12	45
Chloride	44.67	1760.8
Fluoride	-	1.9
Sulpate	20.28	974.0
Nitrate	0.39	25
Total Hardness		1800 mg/L
Alkalinity		165 mg/L

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### A CORNER TEST FOR CONDUCTIVITY VERSUS PERCENT <u>BIOMPHALARIA</u>

FIG. 17

	CONDUCT	IVITY						
\$	8	-16.7 -16.6 -16.1 •						
•		-14.9						
BIOMPHALAR 0 10	IA 10.2	- 6.9 - 6.7 - 6.6 17	•	41	52	79	81	90.5
6 @		- 6.5 - 6.3 - 6.2 - 5.9 - 5.8		٠	8	6	9	
		- 4:4						•

The quadrant sum (S) value for this test does reach the value of 11 required to show significant association at the 5% level for a sample size of 14. 44

•

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The quadrant sum (S) value for this test does not reach the value of 11 required to show significant association at the 5% level for a sample size of 14.

FIG. 18

# TABLE 6

# MEASUREMENT MEANS OF BIOMPHALARIA SNAILS FROM THE MIZURATA AREA

Name of the Spring	Height of the snail in mm	Diameter mm	Number of Whorls	Color of Shell	Name of the Species
Taworga Spring	3.8	10.3	3.5	Brown	<u>Biomphalaria</u> <u>alexandrina</u>
Ain El Halumia	4.5	15.7	5	Brown	<u>Biomphalaria</u> <u>alexandrina</u>
Ain Zreik	4.0	14.4	4	Brown	<u>Biomphalaria</u> pfeifferi
Ain Emheide	5	15.8	5	Brown	<u>Biomphalaria</u> <u>alexandrina</u>
Ain Eljemal	4.5	15.5	5	Brown	<u>Biomphalaria</u> <u>alexandrina</u>
Ain Esala	3.9	14.1	4	Brown	<u>Biomphalaria pfeifferi</u>
Ain Hassan	5	16	5	Brown	<u>Biomphalaria</u> <u>alexandrina</u>
Ain Zayed	5	15.8	5	Brown	<u>Biomphalaria</u> <u>alexandrina</u>
Ain Slaia	3.5	13.9	3	Brown	<u>Biomphalaria</u> pfeifferi
Ain Nąji	3.6	13.8	3	Brown	<u>Biomphalaria</u> <u>pfeifferi</u>

.

The number of whorls was from 3 to 5. All snails were brown shelled (Table 6).

The identification cannot be based directly on size determination of the snails collected live because some springs had received recent molluscicidal treatment, and the living specimens present were predominantly juveniles. In order to be certain about the identity of the <u>Biomphalaria</u>, dissections of related specimens must be made. In Taworga spring most of the snails were juvenile because most of the control activities concentrated on this spring, and the use of Byluscide destroyed the adults. All of the snails present were a new generation, so species determination in this spring is based on previous studies. The color of the snails was brown when dried, but many of them were greenish and white when the collection was made. The authorities consulted agreed that the species determinations used here were probably correct.

### Human Infection Rate Determination:

Measurement of human infection was made in June 1977 by stool examination. The stool samples were collected between 10 A.M. and 3 P.M. and examined within 24 hours. These samples were collected in Sayed Aisha Religious School. It was chosen because it was the nearest school to the main source of water in the Taworga area. It is situated on the south side of the spring, 250 meters away from the spring. Most of the students on their way to

school play in and with the water which is heavily populated with Biomphalaria snails.

In a few cases the direct smear was positive for samples that were negative in the sedimentation test.

Eighty-nine students were tested. One hundred percent of 11, 12, and 13 year olds were infected (Table 7). No infection was found in the 6 and 8 year olds. The overall infection rate among children of all ages was 86.5%.

A T-test performed using the Statpack program for T-tests on the Western Michigan University Computer Center computer showed that 6 to 9 year olds had significantly lower infection rates than 10 to 15 year olds.

In July 1978, stool samples were collected from the native inhabitants and the Egyptian workers at the agriculture project. Infection rates were 22% and 75.7% respectively (Table 8).

#### TABLE 7

SCHISTOSOMIASIS (S.MANSONI) IN SAYEDA AISHA RELIGIOUS SCHOOL\* ON 6.7.1977

Age in Years	No. of Students Tested	No. of Students Positive	% Positive
6	3	0	0
7	. 3	2	66
8	2	0	0
9	4	1	25
10	9	8	88.8
11	14	14	100
12	12 12		100
13	13 18		100
14	14 14		92.8
15	10	9	90
Total	89	77	86.5

\*Because Sayeda Aisha is a Religious School it was open at that date during summer vacation. This school is the nearest one in the area to the Spring. 49

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### TABLE 8

### ADULT SCHISTOSOMIASIS NEAR TAWORGA SPRING

	No.of People Tested	No. of People Positive	% Positi <b>v</b> e
Population	100	22	22
Egyptians	95	72*	75.7

\*23 cases of S. Mansoni and 44 cases of S. haematobium.

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#### DISCUSSION

The collection of snails in the field and their transportation to Tripoli caused many of them to die in transit. Others may have been killed in the laboratory by changes of water, or by unknown causes this may have caused a differential mortality, and lowered the estimated infection rate.

The lack of significant association between <u>Biompha-</u> <u>laria</u> and non-<u>Biomphalaria</u> snails (Table 2, Fig. 14) indicates competition is not a major factor in determining the distribution of <u>Biomphalaria</u> in the springs examined. A complete snail survey in springs, examining all the snails for <u>Schistosoma</u> infection, is very necessary to determine the proper molluscicide treatment.

Fortunately, in most parts of the country, the amount of snail habitat is limited and control of infection by the destruction of snails can give very good results in a short period of time.

The presence of cercaria in some snails not found to be liberating cercaria, and the failure to find cercaria in instances where cercaria had been released prior to crushing the snails for examination, shows that the percent infection figures must be considered to be low estimates. With the low infection rates of 1.32%, cryptic infections could increase the rate rapidly, e.g., five undetected infections would double the stated rate in this case to a value of 2.64%.

In Ain Emheide the pH was very low compared with other springs, but that does not appear to affect the population of snails because of deficiency in the amount of calcium which is necessary for shell building.

The ionic composition of the water is probably related to the proximity to the ocean, the below sea level elevation, and the consequent lack of drainage. That may have a direct effect on the high conductivity.

Van der Schalie and Berry (1973) indicated that higher temperature in water makes less dissolved oxygen available to aquatic animals. The prospective changes caused by elevated temperature have been also outlined by Cairns (1968): "Death through direct effects of heat; internal functional aberrations (reduced oxygen, disruption of food supply, decreased resistance to toxic substances); interference with spawning or other critical activities in the life cycle; competitive replacement by more tolerant species as a result of the above physiological effects." Application of these studies indicates that because of small spring sizes in the Mizurata area, the use of solar energy to warm the water could be useful to prevent snail reproduction, especially when the water temperature of the springs is in the range of  $27^{\circ}$ C.

For the epidemiological study, there were difficulties in collecting fecal samples from the females because of social factors. A female nurse is needed in any study or

project dealing with the female portion of the population. A house to house survey in Taworga and El Heesha area is very necessary for locating cases of schistosomiasis. Health education may increase the willingness of the local people to provide stools for examination and aid them in developing habits that will reduce the spread of schistosomiasis. Egyptians employed in the agriculture development project constitute a danger in the spread of the disease because of their greater mobility. It is highly recommended that anyone who is infected be prevented from working in this area.

It is also desirable to control movement to and from the endemic foci of people and materials containing live snails as much as possible. The introduction of infected snails or infected people to a new area can establish the disease cycle.

This study has shown that a correlation exists between percent <u>Biomphalaria</u> versus both pH and conductivity. These are added to other findings and suggestions of possible use in the control of schistosomiasis through disease dependence on vectors as tabulated in Table 9.

From the finding of the present study as well as from the results of previous studies, it can be concluded that schistosomiasis is an important public health problem in Libya because of the large area of the country and the distribution of schistosomiasis. Any project using springs

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# TABLE 9

### SOME SCHISTOSOMIASIS CONTROL POSSIBILITIES

Approach Through Human Host	Approach Through Molluscan Host Control		
	1)Physical Habitat	2)Chemical Control with molluscicides	3)Biological Control
a. Education with improve	a)Temperature (Van	a)Bayluscide	a)Predators (cray
sanitary conditions.	der Schalie and	(effective on	fish, ducks, etc)
	Berry 1973)	B. alexandrina)	
b. Screening and treatment	b)pH (this study)	b)Frescon (effective	b)Parasites (šchio-
program.	c)Conductivity	on <u>B. pfeifferi</u> )	myzid flies, other
	(this study)	c)Copper sulfate	trematodes, etc)
	d)Current velocity	(cumulative and	c)Competition with
		incompatible with	other snails and
		many human uses	genetically mani-
		of water)	pulated Biompha-
			laria.

must consider this problem because these constitute the only habitat of snails in many areas.

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