PN-ABA-55

AGENCY FOR INTERNATIONAL DEVELOPMENT
WASHINGTON D.C. 20523

DATE: 4/15/88

MEMORANDUM

TO: AID/PPC/CDIE/DI, room 209 SA-18

FROM: AID/SCI, Victoria Ose

SUBJECT: Transmittal of AID/SCI Progress Report(s)

Attached for permanent retention/proper disposition is the following:

AID/SCI Progress Report No. 6, 202
PR # 2 7/1/87 - 12/31/87

Attachment

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6. c202

USAID PROGRAM IN SCIENCE AND TECHNOLOGY COOPERATION

AID/SCI Proposal 6.202: A Small-Scale Field Testing of *Echinostoma liei* as a Biocontrol Agent against *Schistosoma mansoni* in Egypt.

Project No.	: 936 - 5542
Name of Grantee	: Theodor Bilharz Research Institute, Embaba, Egypt.
Principal Investigator	: Prof. Dr. Fouad Yousif.
Duration of the Project	: 3 Years.
Starting Date	: Jan. 1,1987.

Progress Report No. 2

(July 1, 87 - Dec. 31, 87)

Rec'd in SCI. MAR 2 0 1989

Project Personnel

Egyptian team (Theodor Bilharz Res. Inst., Medical Malacology Dept.)

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AID / SCI proposal 6.202: A small Scale Field Testing of *Echinostoma liei* as a Biocontrol Agent against *Schistosoma mansoni* in Egypt (Project No. 936 - 5542)

Progress Report No. 2

(July 1, 1987 - Dec. 31, 1987)

Summary

This project consists of a test of the trematode *Echinostoma liei* in the biological control of *Schistosoma mansoni* under simulated and natural field conditions by obtaining data on : a) the effect *E. liei* may have on the population density of *Biomphalaria alexandrina*, the snail host of *S. mansoni*, b) the effect *E. liei* may exhibit on the infection rate of *B. alexandrina* with *S. mansoni* and on the cercariae production by this snail.

The first year of the project was planned to be a preparatory phase. The activities which were invisaged for the second six months are: performance of 3 pilot experiments for the determination of the optimal number of *Biomphalaria* snails and *Schistosoma* and *Echinostoma* eggs to be used in the intervention experiment, developing of the life cycles of both parasites for the production of large materials needed for this experiment, collecting of ecological data in the experimental ditches, and visit of the principal investigator to U S A.

All these activities were carried out as envisaged. Thus, the 3 pilot experiments were performed. Results obtained indicated that 4000 snails or more per ditch section, 500,000 *Schistosoma* eggs and 5,000,000 *Echinostoma* eggs may be most suitable for the intervention experiment. The life cycles of both parasites were developed as planned and the ecological data of the ditches including temperature, water level, water analysis, pH, conductivity, fauna, flora and plankton were regularly collected.

Some problems were encountered during this work such as difficulty in using the modified Ekman grab for snail sampling as the vegatation became dense, unconvenience of one ditch for unknown reasons, the techniques used for introducing eggs of the parasites into ditches need to be improved, and explosion of algae in several ditches and trials were carried out to slove them.

The visit of the principal investigator to USA included Tulane, San Francisco, Johns Hopkins and Lowell Universities and was very useful and fruitful.

For the next six months, it is planned to carry out the main intervention experiment for testing *E.liei* against *S. mansoni* under simulated field conditions and to collect the base line date of four chosen natural canals which will be used in the third year of the project for the same testing under natural conditions.

1- Activities which were envisaged to be conducted during the second six months of the project (as given in the proposal) :

In this project, it is planned to test Echinostoma liei against Schistosoma mansoni under simulated field conditions then in a small scale in the field. The project is divided into 2 phases : a preparation phase for one year and intervention and evaluation phase for 2 years.

The preparation phase (first year) includes the preparing of the experimental area, purchasing of the equipment, training of personnel, standardization of techniques, preparing of materials(Biomphalaria alexandrina snails and eggs of the two parasites), collecting of ecological data in the experimental ditches, and carrying out three pilot experiments to get necessary information for the next phase. In the first six months (Jan - June, 1987) activities envisaged to be carried out in this period were mostly performed as shown in Report No. 1.

Three pilot experiments were set up in the period May - June 87:

1- The first experiment was for the determination of the optimal snail number which should be introduced in each section of ditches for the intervention experiment.
2- The second experiment was for the determination of the optimal number of Schistosoma eggs needed for each ditch section to induce a moderate infection in snails.
3- The third experiment was for the determination of the optimal number of Echinostoma eggs needed for each ditch section to induce a high rate of infection in snails.

The activities which were envisaged to be conducted during the second six months of the project (July - Dec., 1987) are as follows:

1- Maintaining of the life cycles of S. mansoni and E. liei in the Central Laboratory for the production of the large quantity of materials needed for the intervention experiments.

2- Collecting of ecological data in experimental ditches during the period of this report.

3- Performance of pilot experiments and analysis of the data.

2- Activities already carried out during the second 6 months of the project (July - Dec., 1987):

2.1- Maintaining of the life cycle of both parasites:

The life cycles of S. mansoni and E. liei were continuously maintained in the laboratory and were developed for providing the large quantities of materials needed for the pilot and intervention experiments as well as for some laboratory experiments.

After returning from USA, cultivation of snails in the Central Laboratory was changed from breeding in aquaria to be on mud and Nostoc algae. The latter method proved to be more successful.

2.2- Collecting of ecological data in experimental ditches:

The ecological data in the experimental ditches were reported regularly starting from April 87 to Dec. 87. These data include continuous recording of water temperature using a portable thermograph (six months Ryan), daily or every other day recording of the fluctuation of water level, surface temperature,

pH, electric conductivity. Analysis of water was carried out monthly till Sept. 87. Monthly reporting of biological data including the phyto and zooplankton, aquatic fauna and flora was also carried out. A summery of the data is attached.

2.3- Pilot experiments

2.3.1- Determination of the optimal snail number to be introduced in each section of ditches:

This experiment was started in 25 May 87 by placing 3 test numbers of Biomophalaria alexandrina snails, (1000, 2000, 4000), 2 replicates each, in 2 ditches (6 sections). Snail population, size distribution and trematode infection were followed up regularly for 7 months (till Dec., 1987). Sampling of snails was carried out fortnightly, using concurrently a modified Ekman grab and the dipnet. However the modified grab was used for one month only; it was not possible to continue with it because of the heavy growth of weeds which hindered its processing.

The conditions in one of the two ditches (3 sections) were unfortunately not convenient for unknown reasons, as Biomphalaria snails decreased rapidly and almost disappeared as early as mid September. A summary of data obtained from the other ditch (3 sections) is attached.

2.3.2.- Determination of the optimal number of Schistosoma eggs needed to induce a moderate rate of infection in snails:

This experiment was started in June 29, 87.
Eight sections (in 3 ditches) were used.
Median - sized 4000 B. alexandrina snails were placed in each section. Moreover two 30 - L plastic cages, each containing 300 snails were placed in each section to give more information about survivorship and infection of snails.

In July 1 st,three test doses of Schistosoma eggs (20,000 50,000, 100,000) were introduced into six sections, two replicates for each test number, two other sections were used as control. Introducing of eggs was carried out in one - litre glass jars which were placed 20 cm under water surface and left for 24 hour. The hatchability of the ova was determined in the laboratory and was found to be 77.2%.

Snail population, number of infected snails were followed up till Dec. 1987. Weekly sampling of snails began 3 weeks from the time of introducing eggs. Snails obtained from each section were counted and examined for cercarial shedding. All snails were replaced into their sites of collection after marking positive snails.

The number of cercariae obtained from each shedding snail (after exposure to sunlight for 30 minutes) was determined. A summary of the obtained data is attached.

2.3.3- Determination of the optimal number of Echinostoma eggs needed to induce a high rate of infection in snails:

This experiment was started in June 9, 1987. Median-sized 4000 B. alexandrina snails were placed in each ditch section and eight sections were used. This was followed, in June 23, , by introducing of developed Echinostoma eggs into sections (600,000, 700,000 and 1,000,000) two replicates for each test number. Two sections were used as control. This experiment is basically similar to the previous one and it was followed up till December 1987. Data obtained are attached.

2.4 - Certain laboratory experiments were carried out in the period of this report but results are not yet available : for studying egg production of E. liei from various doses of metacercariae/ rat,and for studying effect of water depth on hatchability of eggs of E. liei and S. mansoni located at various levels.

1- Balance Sheet:

Budget item	Total budget of the project(3 year) US $	Expenditures during the first year	Residue from total budget in Dec. 31, 1987
Salaries	75,700	12,044	61,656
Equipment	4,600	3,151	1,449
Materials & Supplies	10,000	3,154	6,846
Consultation	5,200	546	4,654
Travel	21,500	5,536	15,964
Other direct costs	4,500	1,136	3,364
Indirect costs	11,500	2,511	8,989
Grand Total	133,000	30,078	102,922

4- Deviation from original plan:

Some problems were encountered during this work and lead to some deviation of the original plan, however trials to solve it were carried out.

4.1- Difficulty with using the modified Ekman grab for snail sampling: This happened as a result of the dense growth of algae and vegetation in most ditches. The dense vegetation prevented the dipping of the grab into the water and the closure of its jaws. Therefore it was necessary to depend on the dip net for snail sampling.

An experiment for studying the efficiency of the dip net in comparison with the modified Ekman grab and the drag scoop, under the present semi-field conditions, was carried out. Thus, 18 separate ditch sections, were provided with three different densities; 1000, 2000 and 4000 Biomphalaria snails, 6 sections for each density. Three sizes were used (2-4mm, 4-9mm, >13 mm shell diameter) with known proportions of each size in each density. Snails were distributed in each section at random and 3 hours later each of the three tools were used twice in each snail density for comparative evaluation of these tools. The results obtained are attached.

4.2- Explosion of algae in ditches: A few weeks after the setting up of the pilot experiments in the ditches, various types of algae (see results) started to grow heavily in several ditches and soon the algae became very dense and hindered the process of snail sampling by the modified Ekman grab. To overcome this problem the algae were cleared partially at regular intervals. This was carried out manually which may have affected the snail population. Moreover, the dense algae may have also affected the infection of snails with miracidia. But with the onset of autumn, the growth of algae became naturally reduced. Analysis of water showed that it is rish in NO_3 , NO_2 and H_2S and this may explain the dense growth of algae. It appears that the pipe of underground water should be deepened.

4.3- Lack of a suitable method of introducing Schistosoma and Echinostoma eggs: Schistosoma and Echinostoma eggs were introduced into ditches by putting it in one-litre glass jars. Three jars were used for each section of ditches. They were placed 20 cm under the water surface. From the results, it appears that this method may be not satisfactory especially in the case of Echinostoma. Therefore, several other types of containers were tested in the laboratory for being used for introducing of eggs in the ditches water. It appears that the containers should be smaller in size, obaque and placed nearer to water surface and to each other than has been used inthe pilot experiments.

5 Plan for the next six months period:

5.1- Preparation of the experimentatl area for performing the principal intervention experiment (Jan - March, 88): This includes drying out of the ditches (10) for about 6 weeks to get rid of snails remaining from the pilot experiments, clearing out vegetation then providing the ditches with water and maintaining them for about 2 months for natural balance. Extention of the sunshade over 2 other ditches will be carried out. The partitions between ditch sections (20) will be supported with concrete bases.

5.2- Preparation of the materials needed for the principal intervention experiment (till May 88) : This includes collecting of thousands of Biomphalaria snails from the field, sorting for the size required and examing for natural trematode infection espically Schistosoma and

Echinostoma; 120,000 median - sized snails will be needed for the intervention experiment (= 4000 X 30). Millions of Schistosoma eggs (500,000 X 18 = 9,000,000) and Echinostoma eggs (5,000,000 X 18 = 90,000,000) will be prepared in the Central Laboratory.

5.3- Standardization of techniques : (April - May, 88)
Better standardization for snail sampling techniques will be tried. A system for quality control regarding snail measuring, introducing of eggs in ditches, collection of ecological data will be applied.

5.4- Training of staff (Jan - April, 88)
More research assistants (3), technical assistants (2) and laborers (2) will be trained for variable periods on malacological and parasitological techniques, collecting of ecological data in the field, snail sampling, sorting, measuring, etc.

5.5- Setting up of the main intervention experiment:
It is planned to set up the main intervention experiment in April, 1988. This requires collection of 120,000 median - sized Biomphalaria snails from natural canals and producing millions of Schistosoma and Echinostoma eggs as mentioned above. In this experiment, 10 ditches (30 sections) will be used, divided into 5 groups 6 sections each.

All sections will be provided randomly each with 4000 snails of the same size. A group of sections (S) will be provided with Schistosoma mansoni eggs only , a second group (E) will be provided with Echinostoma eggs only, two other groups of sections (S+E) and (S'+E') will be provided with Schistosoma eggs and 3 weeks later with Echinostoma eggs for a single application and simultaneously with several applications respectively and the fifth group of sections (C) will be left.

without introducing of infection, as control, Snails number, size, number of infected snails, number of produced cercariae will be determined fortnightly in each case.

5.6- Collecting of base line data in certain natural canals:

Four natural canals will be selected for testing Echinostoma against Schistosoma in them. In this year, the base line data including snail population, ecological data, infection etc. will be collected. Such data will be used for comparison after introducing Echinostoma eggs in these canalsnext year.

Principal Investigator

Cairo, Feb. 10,1988

(Prof. Dr. Fouad Yousif).

PROGRAMME AND PROCEDURES OF THE ECOLOGICAL STUDIES OF SCHISTOSOME SNAILS

PHYSICAL AND CHEMICAL PARAMETERS

(1) Temperature

- Alcotholic or mercuric thermometers, of 0°C - 40°C range, were used for measuring water surface temperature by direct immersion. Water temperature was measured daily at each ditch between 11.30 a.m. and 12.30 p.m.
- A thermograph, of Peabody Ryan type - Model "J" was used for measuring and recording continuous vertical profiles of subsurface temperatures. The chart width and span length of the thermograph provide for longer periods of recording in time up to 180 days continuously. The thermograph was used throughout the whole of pilot experiments.

(2) pH (Alkalinity and Acidity)

- The pH was measured by a portable electrometer connected to electrode (pH meter).
- pH measuring was done by direct immersion of the electrode into water, at depth of 25 cm of the water column.
- pH was measured daily at each ditch , between 11.30 a.m. and 12.30 p.m.

(3) Conductivity

- The conductivity was measured by a portable conductivity meter connected to electrode. This was done by direct immersion of the electrode into water, at depth of 25 cm of water column.
- Conductivity was measured daily at each ditch , between 11.30 a.m. and 12.30 p.m.

(4) Water level

A woody graduated stand was fixed at each ditch. A standard point for the water level was determined at each ditch,and is located at the lower level of the pipe which outflow the water from each ditch.

Water level fluctuation from this point was recorded daily, at each channel, between 11.30 a.m. and 12.30 p.m.

BIOLOGICAL PARAMETERS

1- Phytoplankton

Detailed analysis of phytoplanktonic population requires estimation of number and volume of each species. Samples were taken from the top 30 cm of the water column by simply hand dipping one-liter sample bottle. Phytoplankton samples were fixed by the addition of 1% of Lygal's solution or 70% ethyl alcohol.

Identification and enumeration were done under a compound microscope. Phytoplankton consists of individual cells, filaments and colonies, and it is best to count cells, although this procedure is impractical in the case of many multicellular colonies.

Counting organisms was most commonly done in the Sedgwick Rafter (S-R) cell by counting along horizontal strips the length of the cell. Usually 2 to 4 strips are counted at a maginfication of at lest 200X.

- Phytoplankton samples were taken every 2 weeks, one sample from each ditch.
- Time of collection was between 11 a.m. - 1 p.m.

2- Zooplankton

Zooplankton organisms were almost distributed throughout the ditches. A fine mesh was used to filter the organisms from the water. The volume of filtered water was calculated.

Zooplankton samples were preserved with enough neutralized formalin, to produce a final concentration of about 4%.

If the number of individuals was relatively small, it was best to settle the entire sample and count all organisms with an inverted microscope. But, if the number was very large, it

was necessary to take subsamples to facilitate counting. However, the number of zooplankton per unit volume of ditch water was estimated within statistical limits by counting the number of individuals in several subsamples.

- Zooplankton samples were taken every 2 weeks (one sample for each ditch).
- Time of collection was within 11 a.m. - 1 p.m.

3 Benthic Fauna

Quantitative estimations of benthic fauna require effective sampling procedures, separation of the orgaisms from the substratum, identification and enumberation of the different species.

Modified Ekman grab (Fig.) and a dip net were used for sampling benthic fauna. Sieves were used for separation of the organisms from the sediments. When much organic debris is present, the sample was placed into shallow white enamel tray and the organisms were sorted manually.

Flotation methods were widely used to assist in the separation of benthic organisms from sediments. Floating organisms were removed quickly with forceps or a small strainer.

- Benthic samples were taken every 2 weeks.
- Time of collection was between 11 a.m. - 1 p.m.

Ecological Data

- Water analysis (Table 1) and laboratory experiments show that the underground water used in the ditches is convenient for snail breeding as well as survivorship and infectivity of miracidia of both *Schistosoma* and *Echinostoma*.
- Conductivity and pH of the water in all ditches throughout the period of pilot experiments (May - Dec.,1987) were within normal scales (Table 2, Fig. 2, 3, 4 & 5).
- The maximum water temperature observed was 28.5 °C in August, at 5 cm from water surface. While the minimum temperature was 12.5 °C in December, at 20 cm from the water surface (Fig. 1).
- Plankton organisms, benthos and vegetation associated with *Biomphalaria alexandrina* throughout the period of pilot experiments are presented in tables 3-11, qualitatively and quantitatively.
- This proofes that natural biological balance was attained in the experimental ditches.

Table (1) Chemical analysis of underground pump water

Date of analysis : 9-4-1987

Colour : clear PH : 7.5

Temp. : 20 °C Conductivity : 710 µmhos

Parameter (ppm)	Result	Parameter (ppm)	Result
Sulfides (S^{--})	0.1	Iron (Fe)	0.6
Dissolved Oxygen (O_2)	0.7	Manganese (Mn)	1.5
Biochemical Oxygen demand (B.O.D)	4.2	Total alkalinity ($CaCo_3$)	290
		Total hardness ($CaCo_3$)	284
Chemical Oxygen demand (C.O.D)	8.0	Calcium hardness ($CaCo_3$)	188
Total dissolved solids (T.D.S)	600	Magnesium hardness	116
Ash of dissolved solids	562	Calcium (Ca)	76.2
Suspended solids	66	Magnesium (Mg)	27.80
Ash of suspended solids	40	Chlorides (Cl)	132
Ammonia (N)	1.37	Silicates (SiO_2)	28.1
Nitrites (N)	0.03	Phosphates (Po_4)	0.7
Nitrates (N)	0.51	Copper (Cu)	Nil
Oils and Fats	Nil	Lead (pb)	0.05
		Faecal Coliforms /100 c.c	24

Table (2) Chemical analysis of ditches water during the period of pilot experiments

Parameters (ppm)	Ditch No.							
	2	3	4	5	6	7	8	9
Sulfides	0.58	0.44	0.36	0.32	0.16	0.36	0.36	0.36
B.O.D	9.4	2.0	3.5	2.2	2.2	4.6	2.9	2.7
C.O.D	<15	<15	<15	<15	16	<15	<15	<15
TDS			486			496	502	503
Ammonia (N)	0.025	0.01	0.014	0.02	0.02	0.02	0.01	0.02
Nitrites (N)	0.237	0.175	0.20	0.09	0.09	0.66	0.07	0.95
Nitrates (N)	0.03	Nil.	0.01	Nil.	Nil.	Nil.	2.1	Nil.
Iron (Fe)			0.5	0.6	0.5	0.65	0.55	0.65
Manganese (Mn)			0.8	1.4	0.9	0.9	0.75	1.2
Total Alkalinity ($CaCo_3$)			232			248	214	228
Total Hardness ($CaCo_3$)			226			302	272	300
Calcium Hardness($CaCo_3$)			110			176	164	164
Magnasium Hardness			116			126	108	136
Calcium (Ca)			60.44			70.4	65.6	65.6
Magnisium (Mg)			27.8			30.2	25.92	32.6
Chlorides (Cl)			136			136	136	136
Silicates (SiO_2)			0.4					
Faecal Coliforms/100cc	122	128	34	12	18			

Table (3)

Other Snail populations associated with Biomphalaria alexandrina in pilot experiment (A) (ditch #2)

Date	Number of Snails / 20 dips							
	Bulinus	Physa	Lanistes	Cleopatra	Lymnaea	Gabiella	Planorbis	Bellamya
			Section (C) / 1000 Snails					
11-6-87	15	326	1	6	-	-	-	-
24-6	9	191	10	8	-	18	-	-
7-7	20	490	35	25	-	61	-	-
21-7	15	443	92	8	-	83	-	-
1-8	13	370	82	14	1	57	6	21
18-8	5	223	50	1	-	18	3	-
1-9	17	223	31	11	-	-	-	1
15-9	35	148	18	-	-	28	-	1
29-9	13	81	37	-	-	18	-	-
14-10	17	84	52	5	-	40	-	-
27-10	16	78	50	-	-	39	-	-
11-11	5	40	55	13	-	8	-	-
25-11	-	33	6	-	-	-	-	-
9-12	18	26	6	-	-	1	-	-
20-12	3	20	23	1	-	-	-	-
			Section (A) / 2000 Snails					
11-6-87	2	87	4	3	-	19	-	-
24-6	10	200	20	4	1	42	-	-
7-7	28	183	121	4	3	30	9	-
21-7	18	124	77	21	-	31	-	-
1-8	10	159	31	9	-	27	1	-
18-8	13	91	44	22	1	98	-	-
1-9	3	136	-	-	40	-	-	4
15-9	-	65	2	2	-	26	-	-
29-9	10	34	28	3	-	97	-	-
14-10	6	36	51	6	-	151	4	-
27-10	5	58	51	12	-	122	-	-
11-11	1	38	31	28	-	20	-	-
25-11	-	9	18	1	-	-	-	-
9-12	9	1	21	-	-	-	-	-
20-12	18	23	9	-	-	7	-	-
			Section (B) / 4000 Snails					
11-6	2	122	3	1	-	1	-	-
24-6	13	173	-	3	-	35	-	-
7-7	52	198	38	7	-	54	-	-
21-7	33	91	32	8	-	91	-	-
1-8	23	94	18	18	-	58	-	-
18-8	11	140	72	5	-	102	2	-
1-9	7	107	61	15	-	82	1	-
15-9	32	73	71	-	-	39	-	-
29-9	4	79	40	-	-	59	-	18
14-10	4	42	56	14	-	106	-	-
27-10	10	47	29	3	-	19	-	-
11-11	1	38	32	5	-	29	-	-
25-11	2	20	10	-	-	3	-	-
9-12	5	14	6	1	-	-	-	-
20-12	2	32	11	2	-	-	-	-

Table (4)

Other snail populations associated with *Biomphalaria alexandrina* in pilot experiment (3)

ditch (6)

Date of sampling	No. of snails / 20 dips				
	Bulinus	Physa	Cabiella	Lanistes	Bellamya
	Section (A)				
28-7-87	1	13	-	-	-
11-8-87	1	11	2	1	3
30-8-87	-	31	8	7	2
14-9-87	-	5	1	-	-
30-9-87	-	8	2	6	21
12-10-87	-	15	3	2	7
	Section (B)				
28-7-87	-	15	7	6	-
11-8-87	-	6	3	1	1
30-8-87	1	13	6	1	2
14-9-87	-	52	5	5	-
30-9-87	2	18	-	4	-
12-10-87	2	35	2	5	-

Table (5)

Other snail populations associated with *Biomphalaria alexandrina* in pilot experiment (3)

ditch (4)

Date of sampling	No. of snails / 20 dips				
	Bulinus	Physa	Cabiella	Lanistes	Bellamya
	Section (A)				
28-7-87	-	26	1	-	-
11-8-87	-	45	1	1	-
30-8-87	11	500	18	5	2
14-9-87	10	185	20	-	-
30-9-87	10	130	2	5	-
12-10-87	15	180	5	5	-
	Section (B)				
28-7-87	2	43	2	-	-
11-8-87	-	12	-	2	-
30-8-87	4	300	3	-	-
14-9-87	5	96	5	-	-
30-9-87	3	66	-	2	-
12-10-87	3	152	2	2	-
	Section (C)				
28-7-87	2	52	2	2	-
11-8-87	-	13	-	1	2
30-8-87	-	167	3	1	2
14-9-87	2	76	8	1	-
30-9-87	-	35	1	-	-
12-10-87	-	70	1	-	-

Table (6)

Other snail populations associated with *Biomphalaria alexandrina* in pilot experiment (3)

ditch (5)

Date of sampling	No. of snails / 20 dips				
	Bulinus	Physa	Cabiella	Lanistes	Bellamya
	Section (A)				
28-7-87	-	49	6	-	-
11-8-87	-	22	2	3	3
30-8-87	-	7	-	-	1
14-9-87	-	1	1	3	-
30-9-87	-	-	-	3	-
12-10-87	-	30	-	5	-
	Section (B)				
28-7-87	-	61	-	2	-
11-8-87	-	35	-	1	-
30-8-87	4	207	-	3	-
14-9-87	-	-	-	1	1
30-9-87	2	4	-	4	1
12-10-87	-	44	-	3	-
	Section (C)				
28-7-87	1	16	1	2	-
11-8-87	-	6	-	1	-
30-8-87	-	90	2	3	-
14-9-87	-	2	-	-	-
30-9-87	-	2	-	1	-
12-10-87	-	28	-	5	-

Table (7) Seasonal abundance of benthos in ditch No. 7 during the observation period *

Section (A)

Date of sampling	Insecta				Mollusca						Amphibia	Fish
	Lethocerus sp	Dolomedes sp	Anisops sp	Anisoptera nuaphs	Bulinus truncatus	Physa acuta	Lanistes carinatus	cleopatra bulimoides	Lymnaea cauillaudi	Bellamya unicolar	Tadepoles of Bufo sp	Tilapia nilotica
25-7-87	-	-	-	-	9	13	7	8	-	6	-	-
16-8-87	-	-	-	-	11	18	5	18	-	8	2	12
30-8-87	-	-	-	-	6	5	6	28	5	12	-	-
13-9-87	4	-	-	-	-	6	5	25	5	-	-	-
28-9-87	-	-	-	-	2	6	6	20	5	-	1	3
11-10-87	-	-	-	-	7	4	2	3	-	-	-	5
25-10-87	-	-	-	-	15	-	2	36	-	-	-	4

Section (B)

25-7-87	1	-	-	-	8	24	3	10	-	2	-	3
16-8-87	-	-	-	-	-	40	15	35	2	10	1	-
30-8-87	-	-	1	1	14	6	6	30	4	5	2	-
13-9-87	-	-	-	-	9	5	3	10	4	-	2	2
28-9-87	2	-	-	-	6	5	3	15	4	-	-	2
11-10-87	-	-	-	-	4	9	1	3	-	-	1	-
25-10-87	-	-	-	-	12	1	3	21	-	-	-	3

Section (C)

25-7-87	-	-	-	-	4	22	-	16	-	3	2	5
16-8-87	-	-	-	-	21	44	11	55	-	19	1	4
30-8-87	-	-	-	-	14	8	2	18	4	-	2	-
13-9-87	1	1	-	-	10	7	4	5	2	-	3	12
28-9-87	-	-	-	-	12	8	5	16	6	-	-	4
11-10-87	-	-	-	-	3	8	1	11	-	4	-	-
25-10-87	-	-	-	-	13	2	-	19	-	-	-	-

* Number per 10 dips

*

Table (8) Seasonal abundance of benthos in ditch No. 8 during the observation period

Section (A)

Date of sampling	Insecta				Mollusca						Amphibia	Fish
	Lethocerus sp	Dolomedes sp	Anisops sp	Anisoptera numphs	Bulinus truncatus	Physa acuta	Lanistes carinatus	Cleopatra bulimoides	Lymnaea cauillaudi	Bellamya unicolar	Tadpoles of Bufo sp	Tilapia nilotica
25-7-87	-	-	-	-	-	65	2	-	-	1	4	5
16-8-87	3	-	-	1	-	32	2	4	-	2	3	17
30-8-87	-	-	-	-	10	6	3	15	-	2	1	19
13-9-87	1	-	-	10	5	2	5	-	4	-	-	13
28-9-87	-	-	-	-	4	23	6	16	3	-	2	14
11-10-87	-	-	1	-	4	10	5	6	3	-	-	15
25-10-87	1	1	1	-	27	15	-	9	-	-	-	35

Section (B)

25-7-87	9	1	-	-	-	20	5	3	-	6	1	4
16-8-87	1	1	6	-	7	23	2	8	-	-	-	15
30-8-87	-	-	-	-	11	10	5	14	2	3	1	2
13-9-87	2	-	-	1	3	108	24	-	-	-	-	16
28-9-87	-	-	-	-	3	34	5	15	1	-	2	-
11-10-87	3	-	-	-	5	11	-	4	-	-	-	3
25-10-87	-	-	-	-	8	11	-	-	-	-	-	-

* Number per 10 dips

Table (9) Seasonal abundance of benthos in ditch No. 9 during the observation period *

Section (A)

Date of sampling	Insecta				Mollusca						Amphibia	Fish
	Lethocerus sp	Dolomedes sp	Anisops sp	Anisoptera nymphs	Bulinus truncatus	Physa acuta	Lanistes carinatus	cleopatra bulimoides	Lymnaea cauillaudi	Bellamya unicolar	Tadepoles of Bufo sp	Tilapia nilotica
25-7-87	4	-	-	-	-	10	2	-	-	14	1	30
16-8-87	-	-	-	-	2	13	10	15	-	7	2	27
30-8-87	1	-	-	1	15	10	9	20	-	1	1	28
13-9-87	-	-	2	-	3	4	-	-	-	-	3	-
28-9-87	-	-	-	-	5	15	-	17	4	-	-	7
11-10-87	-	3	2	-	1	-	-	-	-	-	-	-
25-10-87	1	-	-	-	2	-	-	-	-	-	-	-

Section (B)

25-7-87	4	-	-	-	-	10	-	-	-	3	1	11
16-8-87	1	-	-	-	-	17	2	7	-	-	2	3
30-8-87	1	-	-	1	12	15	5	25	-	4	-	4
13-9-87	-	-	-	-	2	2	-	-	-	-	-	2
28-9-87	-	1	2	-	10	2	-	19	-	-	1	9
11-10-87	-	-	-	-	-	-	-	1	2	-	-	-
25-10-87	-	-	-	-	4	2	-	-	-	-	-	-

Section (C)

25-7-87	-	-	-	-	-	15	4	2	-	5	1	4
16-8-87	-	1	-	-	-	28	-	18	2	2	1	13
30-8-87	1	-	1	-	13	17	4	20	-	-	-	5
13-9-87	-	-	-	-	3	-	-	-	-	-	2	3
28-9-87	-	2	-	2	9	4	-	12	5	-	-	12
11-10-87	1	-	-	-	-	2	-	-	-	1	-	-
25-10-87	-	-	-	-	5	2	-	-	-	-	-	-

* Number per 10 dips

Table (10)

Plankton

Phytoplankton

Species Composition

Division : Chlorophyta

Pediastrum sp.
Scenedesmus sp.
Oedogonium sp.
Closterium sp.
Spirogyra sp.
Hydrodictyon sp.
Ankistrodesmus sp.

Division : Cyanophyta

Oscillatoria sp.
Spirulina sp.
Lyngbya sp.
Anabaena sp.
Nostoc sp.

Division : Chrysophyta

Navicula sp.
Nitzschia sp.
Coscinodiscus sp.
Synedra sp.
Gyrosigma sp.
Gomphonema sp.

Zooplankton

Species Composition

Copepoda : *Cyclops sp.*
Nauplius larva

Rotifera : *Keratella sp.*
Brachionus sp.
Asplanchna sp.

Nematoda : *Larvae of Nematoda*

Cladocera: *Bosmina sp.*

Insecta : *Culex larvae & pupae*
Chironomus larva

Table (11)

Benthos

Species Composition

Insecta

Lethocerus sp.
Dolomedes sp.
Anisops sp.
Anisoptera nymphs

Mollusca

Bulinus truncatus
Physa acuta
Lanistes carinatus
Cleopatra bulimoides
Lymnaea cauillaudi
Bellamya unicolar
Gabbiolla senaariansis

Amphibia

Tadepoles of Bufo sp.

Fish

Tilapia nilotica

Aquatic Macrophytes

Cyperus dives
Phragmites communis
Phragmites australis
Typha angustata
Paspalum paspaloides
Ceratophyllum demersum
Chara sp.

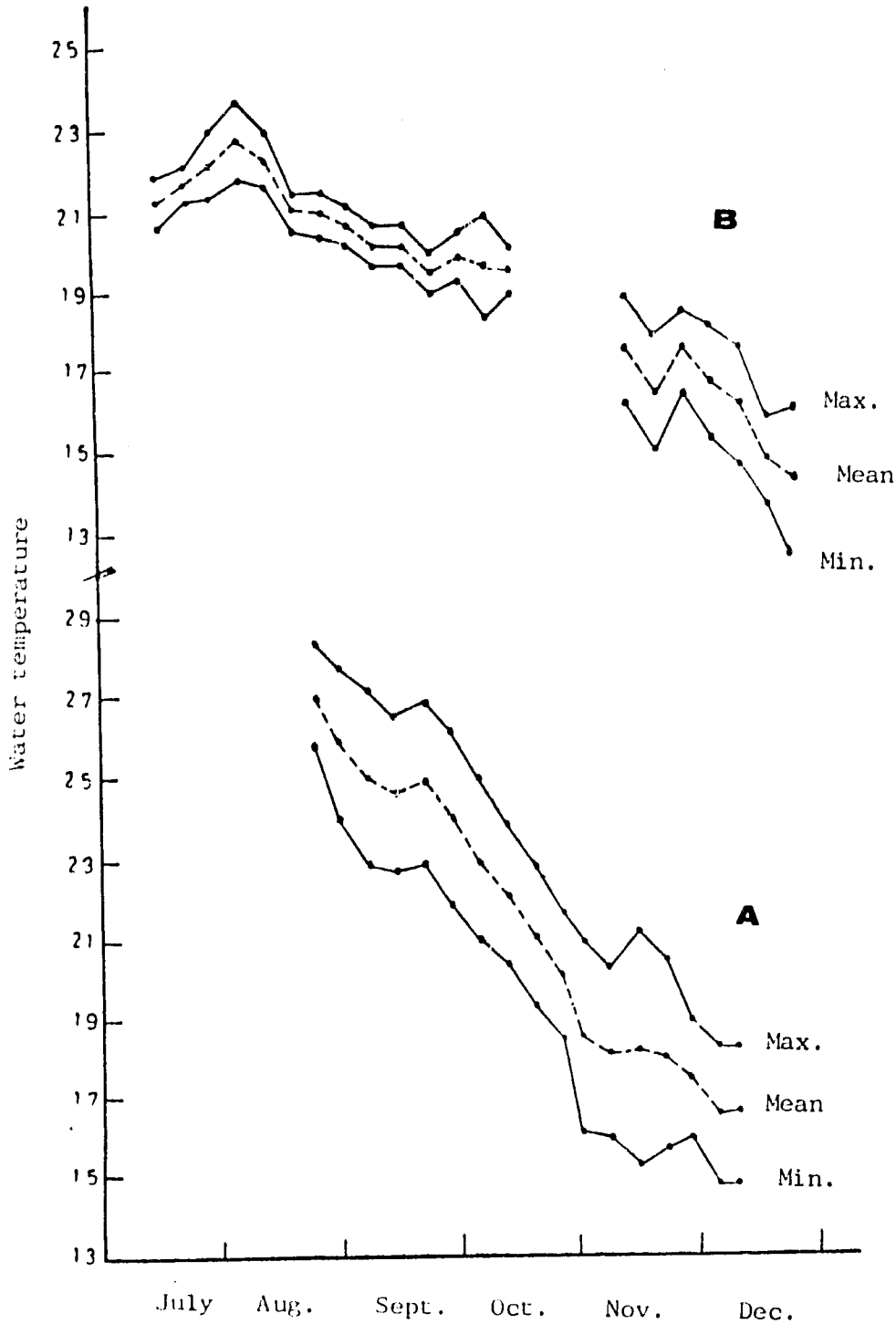


Fig. (1) Maximum, Minimum, and mean of water temperature (Recorded by Ryan thermograph) at different levels.
A-at 5cm from the surface.
B-at 20cm from the surface.

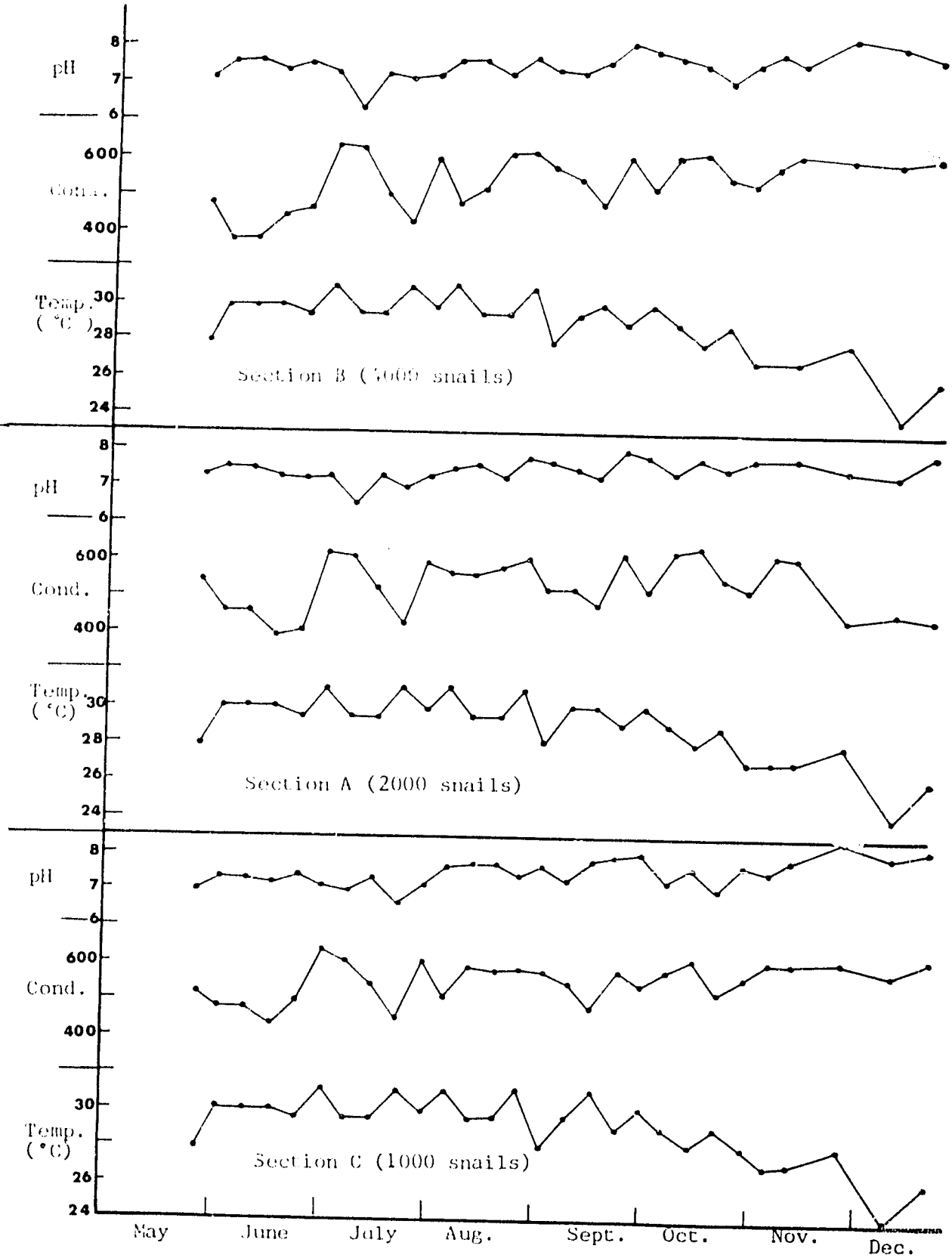


Fig. (2) Surface water temperature , conductivity and pH in Ditch 2 during the pilot experiment No.1.

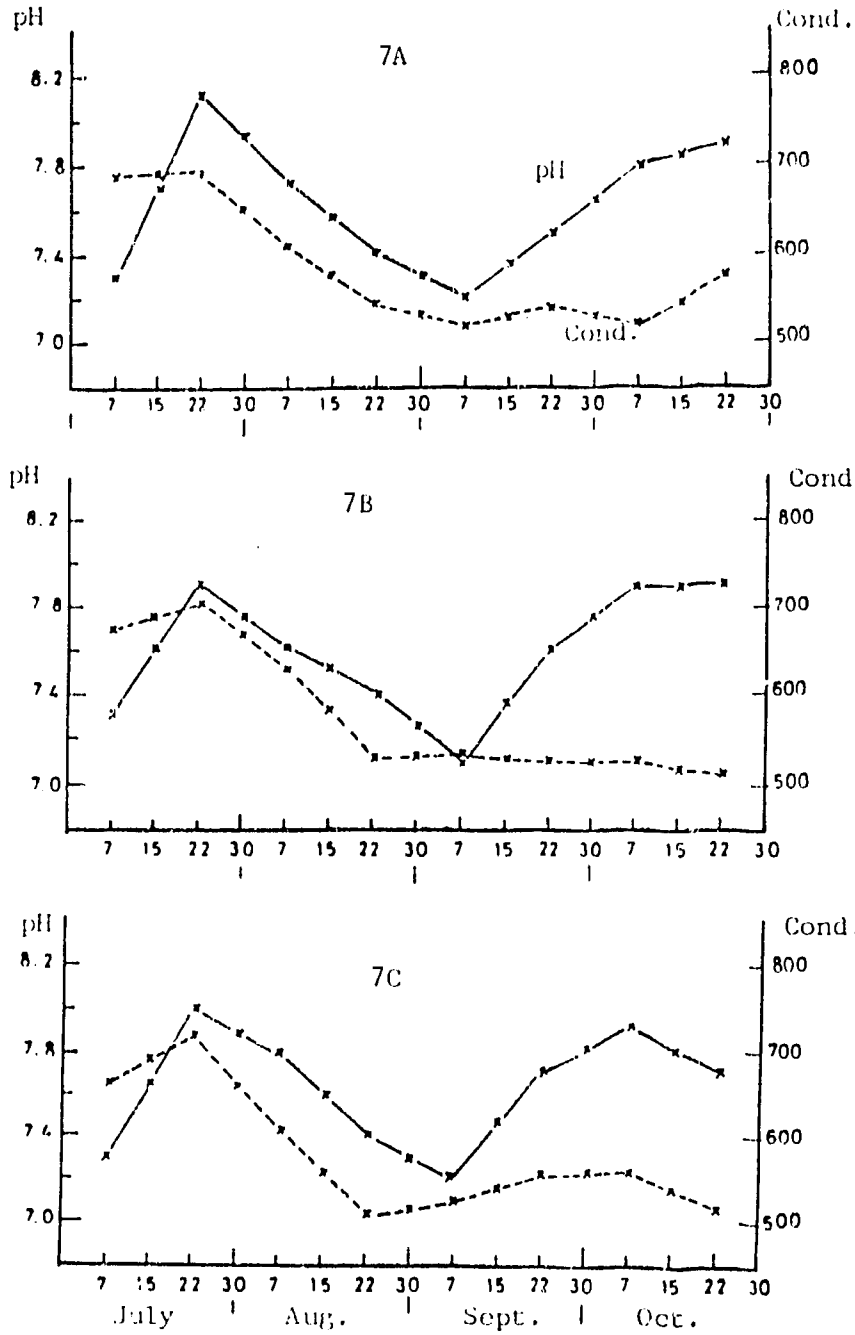


Fig. (3) Seasonal changes in conductivity and pH in ditch No.7.

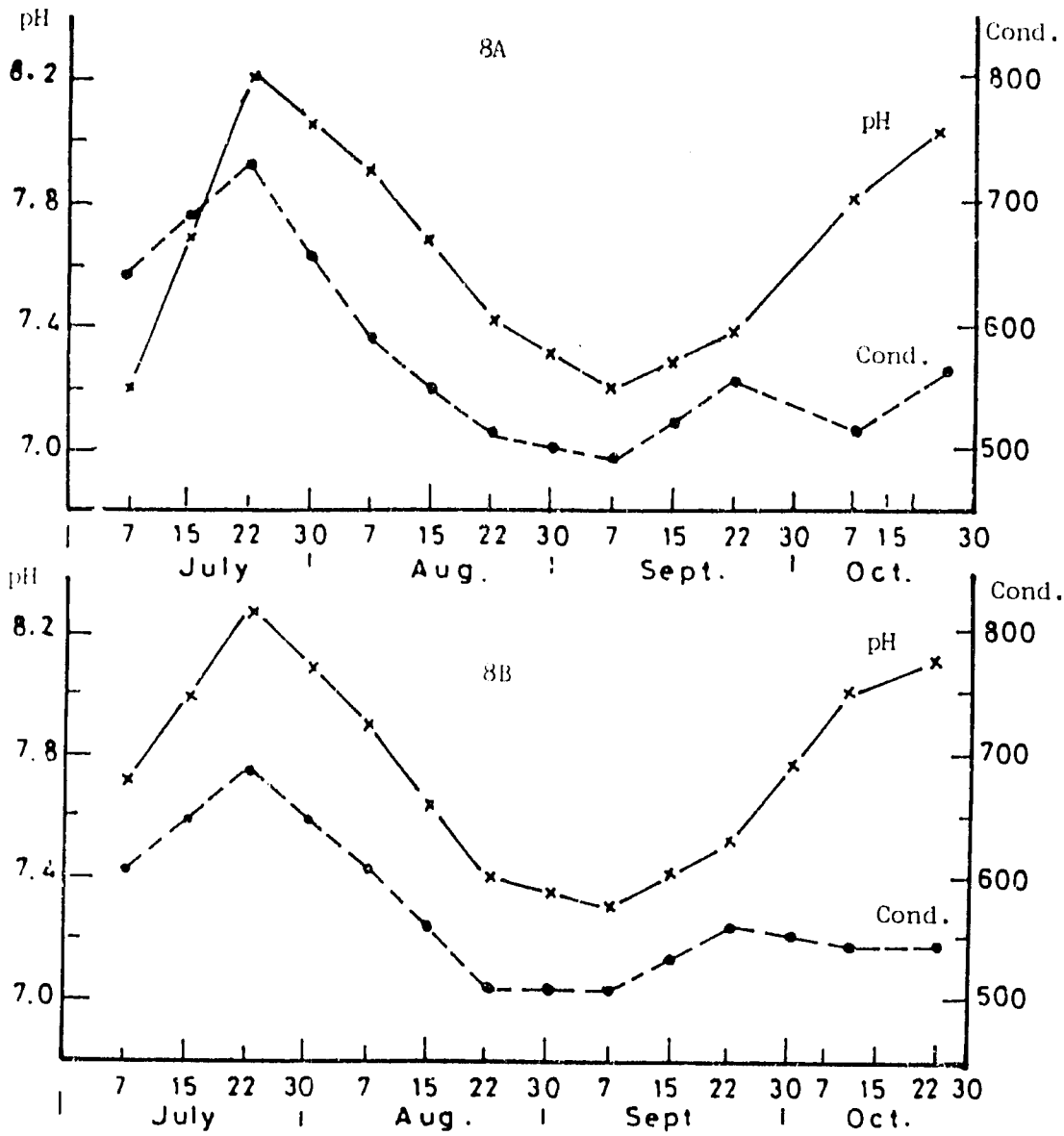


Fig. (4). Seasonal changes in conductivity and pH in ditch No.8.

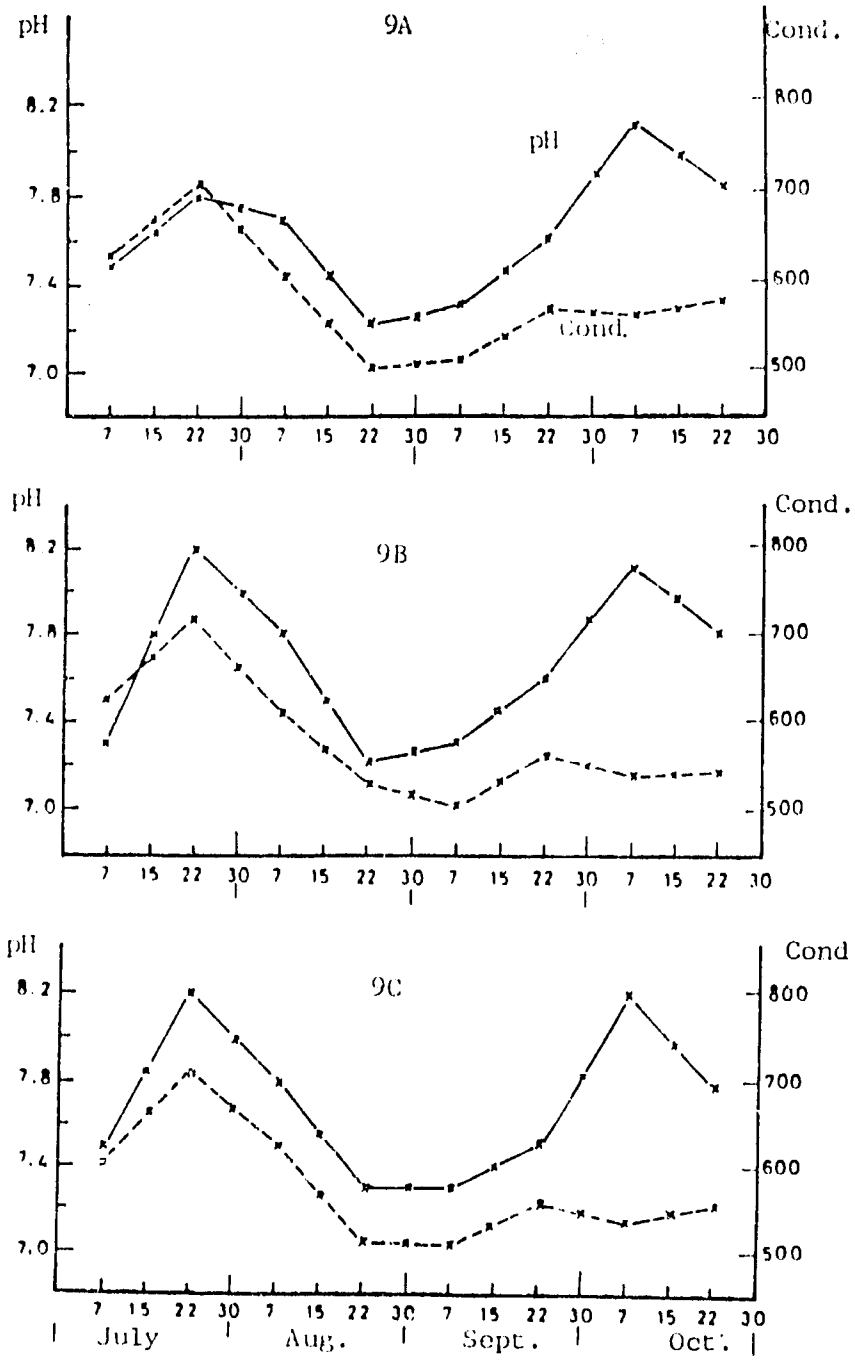


Fig. (**5**). Seasonal changes in conductivity and pH in ditch No.9

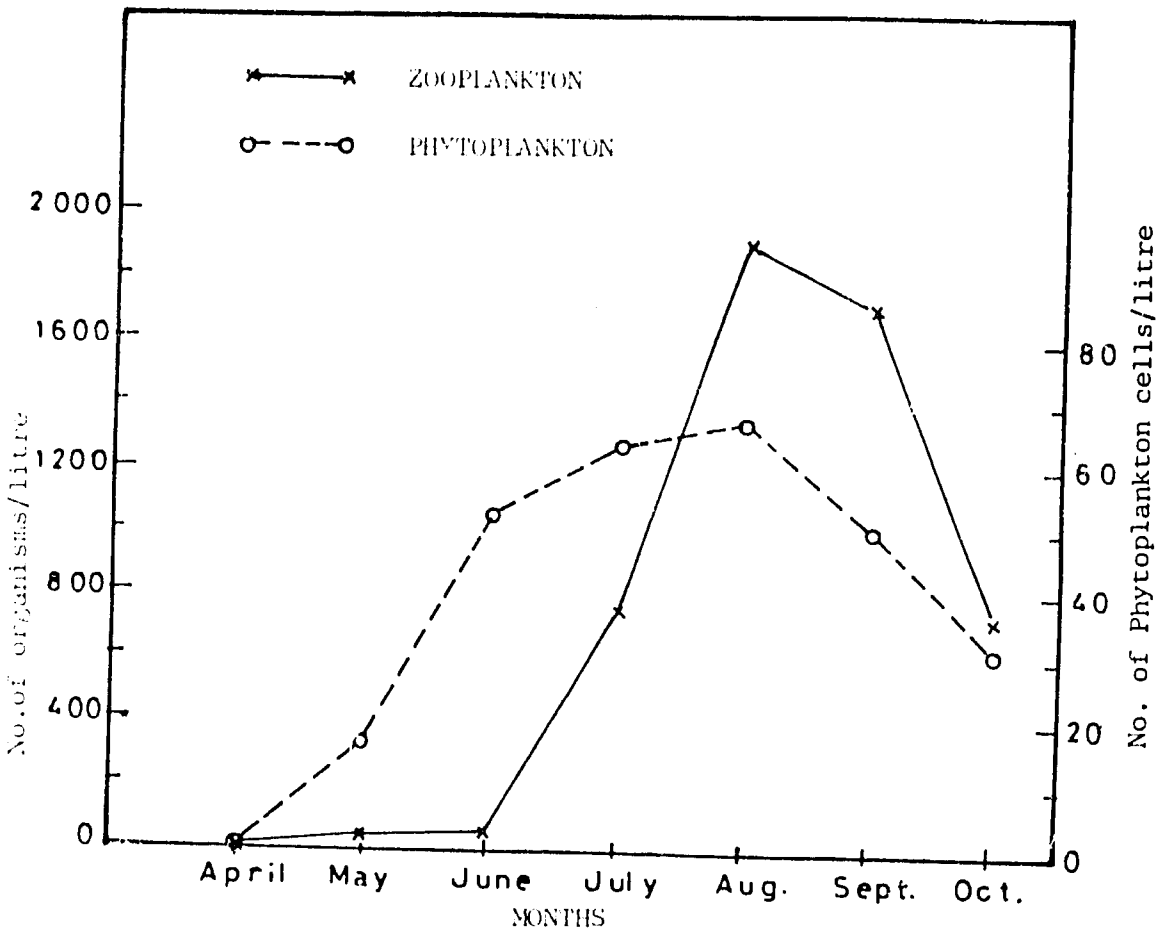


Fig.(6). Seasonal abundance of phyto-and zooplankton organisms in ditch No.6

PILOT EXPERIMENT NO. 1

DETERMINATION OF THE OPTIMAL SNAIL NUMBER WHICH SHOULD BE INTRODUCED IN EACH SECTION OF DITCHES FOR THE INTERVENTION EXPERIMENT

Comments :

- The introduced *Biomphalaria alexandrina* snails survived in the three sections, with 1000, 2000 and 4000 snails, for variable periods of time reaching more than 16 weeks (Fig.7) Thus, the number of of surviving snails decreased gradually with time in all cases of test densities. This decrease is not similar; being more in the case of 4000 snails /section than in the other two test numbers. This reduction in the number of surviving snails was gradual and slow for about 8 weeks and became more rapid in July.
- New generations of snails started to appear a week after introducing snails in the sections. These snails increased gradually and became dominant after 10 weeks from setting up of the experiment.
- Consequently, 4000 snails or more may be convenient for being used in the intervention experiment.

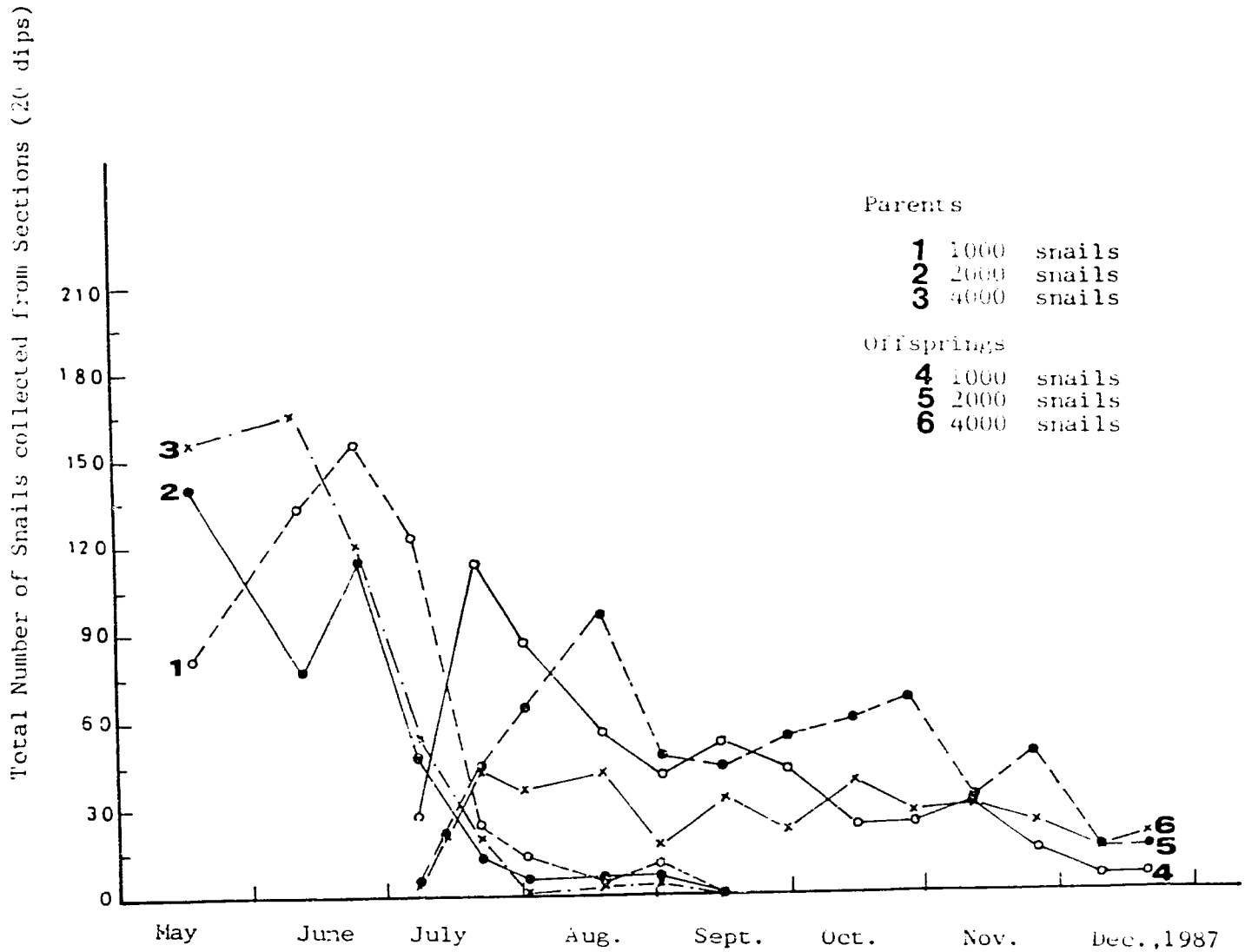


Fig.(7) Population Dynamics of snails (Pilot Experiment No.1).

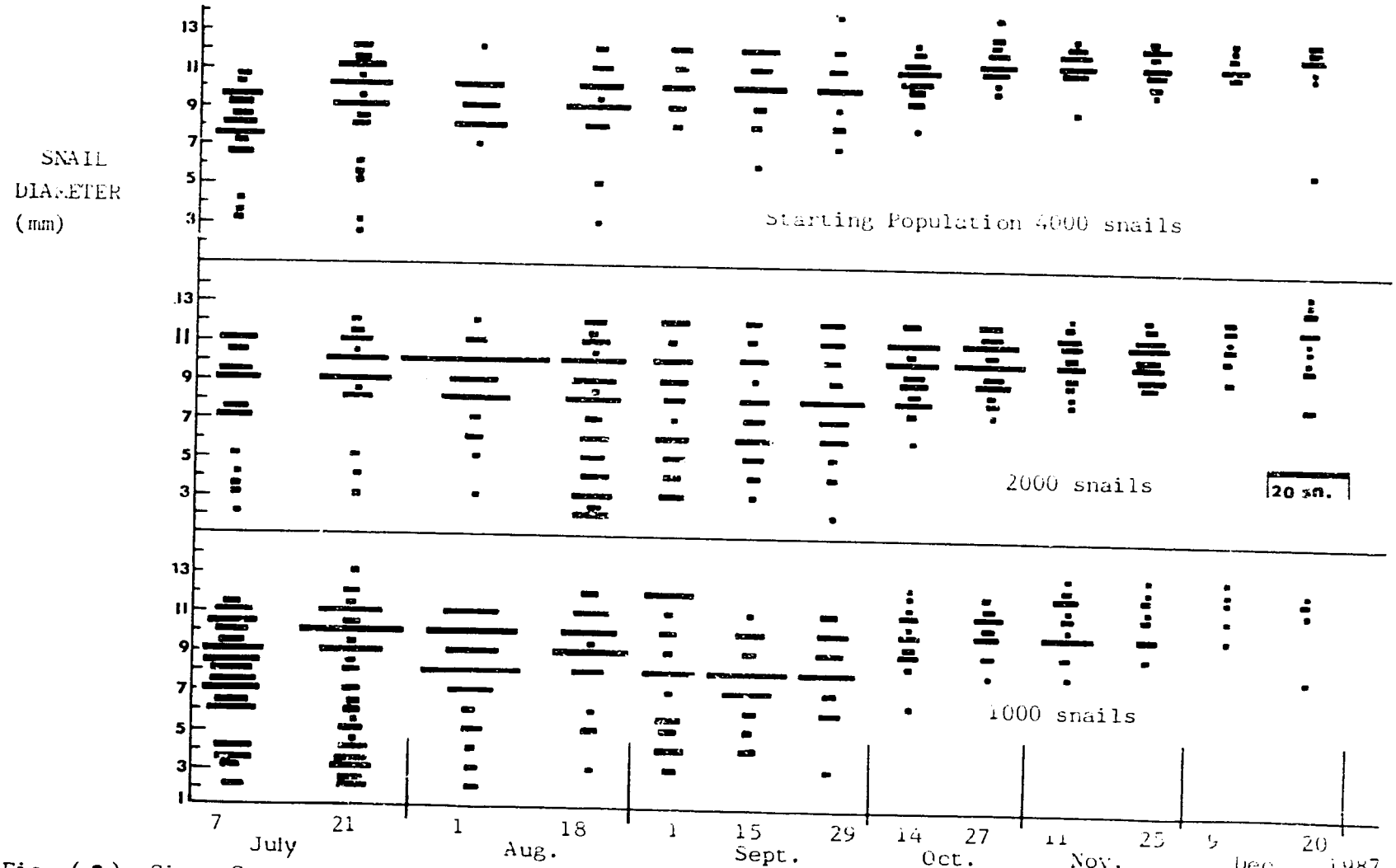


Fig. (8) Size Structure of Biomphalaria alexandrina populations during the period of pilot experiment No.1

Pilot Experiment No. 2

DETERMINATION OF THE OPTIMAL NUMBER OF *SCHISTOSOMA* EGGS NEEDED FOR EACH DITCH SECTION TO INDUCE MODERATE INFECTION IN SNAILS.

Comments

- The cumulative number of infected snails increased with the increase of introduced *Schistosoma* eggs reaching to a mean of 35.5 snails (19.5 %) in the case of 100.000 eggs / ditch section (table 14). This result gives indication that it is needed to increase the number of introduced eggs per ditch section to about 500.000 to get moderate rate of snail infection in the intervention experiment.
- The cercarial output of shedding snails increased also with the increase of ova number / ditch. It appears that there is a direct relation between cercarial output and ova dosage (Table 18).
- The first appearance of shedding snails found at 42 days post introducing of eggs. This was the same in all section regardless the ova test numbers.
- Shedding snails were obtained till 91 days after introducing of eggs (100,000) and 56 days after introducing (50,006 and 20,000 densities).
- The maximum number of shedding snails was obtained after 56 days in the case of (100,000 eggs) and 42 days in the case of (50,000 and 20,000 eggs).
- Mean cumulative number of shedding snails / section was proportional to the ova dose being 35.5 snails (100,000), 11.5 snails (50,000) and 6 snails (20,000).

- The rate of infection of snails with S. mansoni is less than expected. This result may be attributed to one or all of the following factors:

1- Dense vegtation and algae.

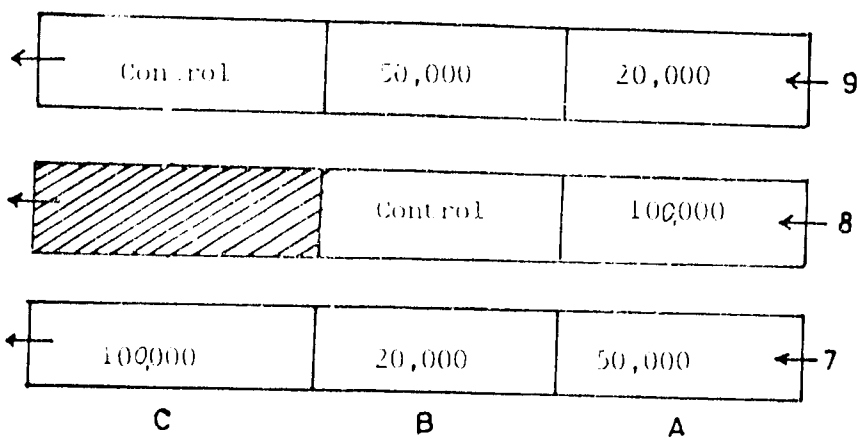
2- Unsuitable method of introducing eggs.

3- Chemistry of water; at least in some sections after algae explosion.

Snails in Cages

- Shedding snails were first obtained at 35 days from introducing the eggs.
- Maximum number of shedding snails was obtained after 45 days in case of 100,000 eggs / section, and 42 days in the case of 50,000 and 20,000 eggs / section.
- No shedding snails were obtained after 70 days from the date of introducing eggs.
- In the case of caged snails, rate of infection relative to surviving snails at the time of first shedding was found to decrease with the increase of ova number. This result may be explained by the higher mortality of snails in the groups exposed to higher doses.

Fig.(9). Diagramatic plan of ditches showing the distribution of ova doses of Schistosoma mansoni in various sections. (Pilot experiment No.2).



*4000 snails have been placed in each section.

Table (12) Survivorship and infection of free *Biomphalaria alexandrina* snails exposed to 20,000 *Schistosoma mansoni* eggs / ditch section

Time (Days Post-exposure)	No. of collected snails *			No. of snails shedding cercariae			Cumulative number of infected snails		
	1	2	Mean	1	2	Mean	1	2	Mean
35	18	28	23	-	-	-	-	-	-
42	23	36	29.5	-	8	4	-	8	4
49	23	15	19	2	-	1	2	8	5
56	18	6	12	2	-	1	4	8	6
63	16	6	11	-	-	-	4	8	6
70	12	1	6.5	-	-	-	4	8	6
77	12	-	6	-	-	-	4	8	6
84	6	-	3	-	-	-	4	8	6
91	1	-	0.5	-	-	-	4	8	6
98	2	-	1	-	-	-	4	8	6
105	-	-	-	-	-	-	4	8	6
112	-	-	-	-	-	-	4	8	6

*per 10 dips

Table (13) Survivorship and infection of free *Biomphalaria alexandrina* snails exposed to 50,000 *Schistosoma mansoni* eggs / ditch section

Time (Days Post-exposure)	No. of collected snails*			No. of snails shedding cercariae			Cumulative number of infected snails		
	1	2	Mean	1	2	Mean	1	2	Mean
35	33	29	31	-	-	-	-	-	-
42	11	45	28	1	18	9.5	1	18	9.5
49	23	15	19	2	-	1	3	18	10.5
56	18	6	12	2	-	1	5	18	11.5
63	16	6	11	-	-	-	5	18	11.5
70	12	1	6.5	-	-	-	5	18	11.5
77	12	-	6	-	-	-	5	18	11.5
84	6	-	3	-	-	-	5	18	11.5
91	1	-	0.5	-	-	-	5	18	11.5
98	2	-	1	-	-	-	5	18	11.5
105	-	-	-	-	-	-	5	18	11.5
112	-	-	-	-	-	-	5	18	11.5

*per 10 dips

Table (14) Survivorship and infection of free *Biomphalaria alexandrina* snails exposed to 100,000 *Schistosoma mansoni* eggs / ditch section

Time (Days Post-exposure)	No. of collected snails *			No. of snails shedding cercariae			Cumulative number of infected snails		
	1	2	Mean	1	2	Mean	1	2	Mean
35	25	66	45.5	-	-	-	-	-	-
42	24	26	25	-	14	7	-	14	7
49	12	24	18	1	3	2	1	17	9
56	11	36	23.5	1	22	11.5	2	39	20.5
63	14	20	17	2	5	3.5	4	44	24
70	7	22	14.5	1	6	3.5	5	50	27.5
77	2	16	9	-	10	5	5	60	32.5
84	1	12	6.5	-	3	1.5	5	63	33.5
91	2	14	8	-	2	1	5	65	35.5
98	3	10	6.5	-	-	-	5	65	35.5
105	1	8	4.5	-	-	-	5	65	35.5
112	1	7	4	-	-	-	5	65	35.5
119	-	1	0.5	-	-	-	5	65	35.5

*per 10 dips

Table (15) Survivorship and infection of caged *Biomphalaria alexandrina* snails exposed to 20.000 *Schistosoma mansoni* eggs / ditch section

Time (Days Post-exposure)	No. of surviving snails			No. of snails shedding cercariae			Cumulative number of infected snails		
	1	2	Mean	1	2	Mean	1	2	Mean
35	174	185	179.5	4	5	4.5	4	5	4.5
42	164	112	138	14	4	9	18	9	13.5
49	150	106	128	6	2	4	24	11	17.5
56	89	72	80.5	2	6	4	26	17	21.5
63	36	48	42	3	2	2.5	29	19	24
70	16	28	22	1	8	4.5	30	27	28.5
77	10	17	13.5	-	-	-	30	27	28.5
84	9	9	9	-	-	-	30	27	28.5
91	7	5	6	-	-	-	30	27	28.5
98	5	4	4.5	-	3	1.5	30	30	30
105	4	2	3	-	-	-	30	30	30
112	3	1	2	-	-	-	30	30	30

Table (16) Survivorship and infection of caged *Biomphalaria alexandrina* snails exposed to 50.000 *Schistosoma mansoni* eggs / ditch section

Time (Days Post-exposure)	No. of surviving snails			No. of snails shedding cercariae			Cumulative number of infected snails		
	1	2	Mean	1	2	Mean	1	2	Mean
35	210	150	180	2	4	3	2	4	3
42	186	95	139.5	8	10	9	10	14	12
49	150	85	117.5	4	3	3.5	14	17	15.5
56	118	22	70	-	3	1.5	14	20	17
63	110	15	66.5	2	4	3	16	24	20
70	8	12	10	-	3	1.5	16	27	21.5
77	4	6	5	-	-	-	16	27	21.5
84	3	5	4	-	-	-	16	27	21.5
91	5	3	4	-	-	-	16	27	21.5
98	4	1	2.5	-	-	-	16	27	21.5
105	2	1	1.5	-	-	-	16	27	21.5
112	2	1	1.5	-	-	-	16	27	21.5

Table (17) Survivorship and infection of caged *Biomphalaria alexandrina* snails exposed to 100,000 *Schistosoma mansoni* eggs / ditch section

Time (Days Post-exposure)	No. of surviving snails			No. of snails shedding cercariae			Cumulative number of infected snails		
	1	2	Mean	1	2	Mean	1	2	Mean
35	193	98	145.5	-	1	0.5	-	1	0.5
42	171	74	121.5	-	2	1	-	3	1.5
49	95	64	79.5	5	3	4	5	6	5.5
56	35	40	37.5	3	1	2	8	7	7.5
63	23	36	29.5	2	4	3	10	11	10.5
70	14	15	14.5	2	4	3	12	15	13.5
77	7	11	9	-	-	-	12	15	13.5
84	2	7	4.5	-	-	-	12	15	13.5
91	2	7	4.5	-	-	-	12	15	13.5
98	1	4	2.5	-	-	-	12	15	13.5
105	-	3	1.5	-	-	-	12	15	13.5
112	-	2	1	-	-	-	12	15	13.5

Table (18) Periodic cercarial output per stimulant period (30 m) of infected snails exposed to various doses of *Schistosoma mansoni* eggs / ditch section

No of cercariae per snail ova dose/section	Range	Mean
20.000	9 - 136	80
50.000	16 - 193	156
100.000	11 - 317	203

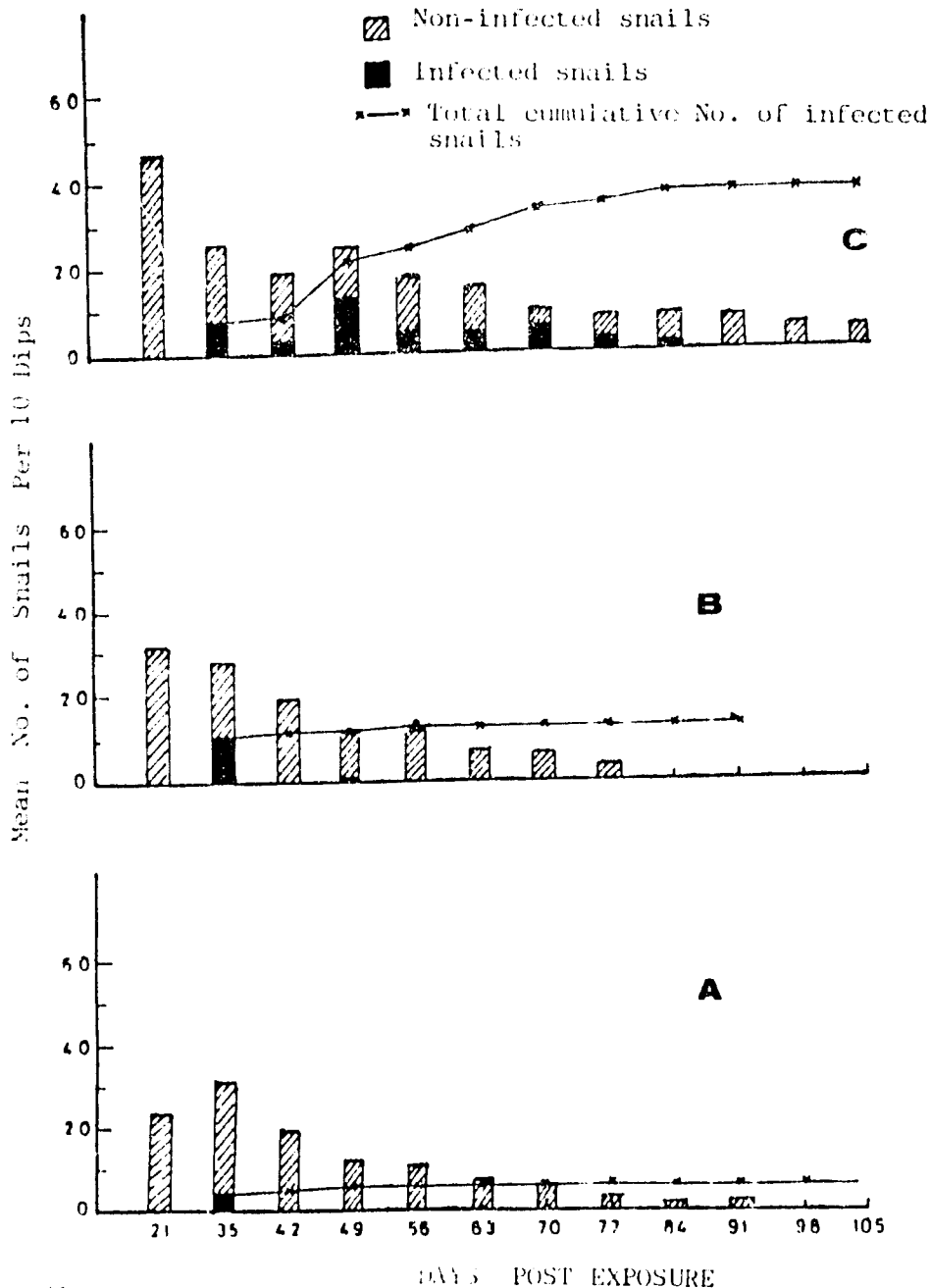


Fig.(10) Survivorship and infection of free Biomphalaria alexandrina snails exposed to various numbers of schistosoma mansoni eggs.

A. 20,0000 eggs/ditch section
B. 50,0000 eggs/ditch section
C. 100,000 eggs/ditch section

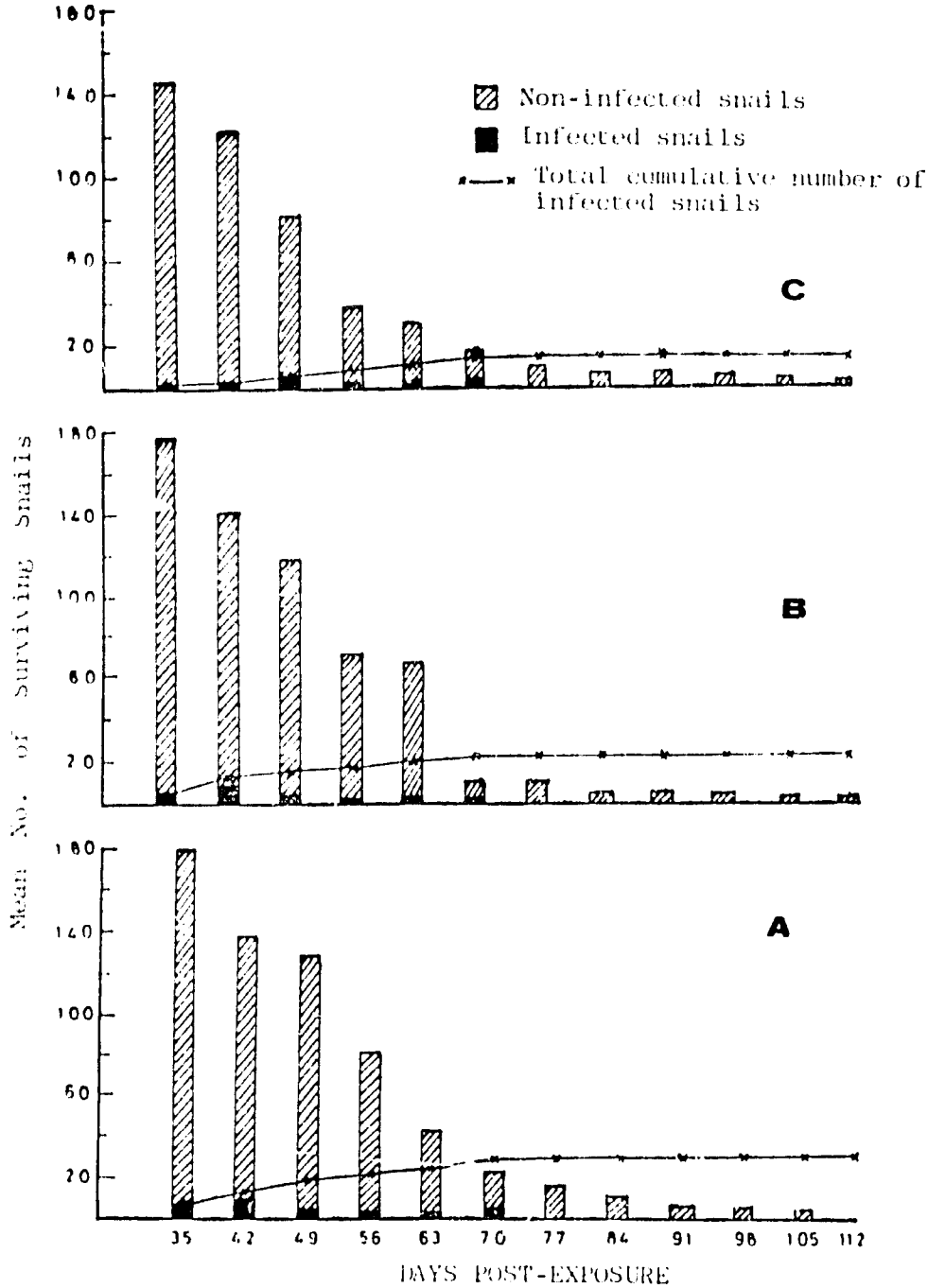


Fig. (11) Survivorship and infection of caged Biomphalaria alexandrina snails exposed to various numbers of Schistosoma mansoni eggs.

A. 20,000 eggs/ditch section
B. 50,000 eggs/ditch section
C. 100,000 eggs/ditch section

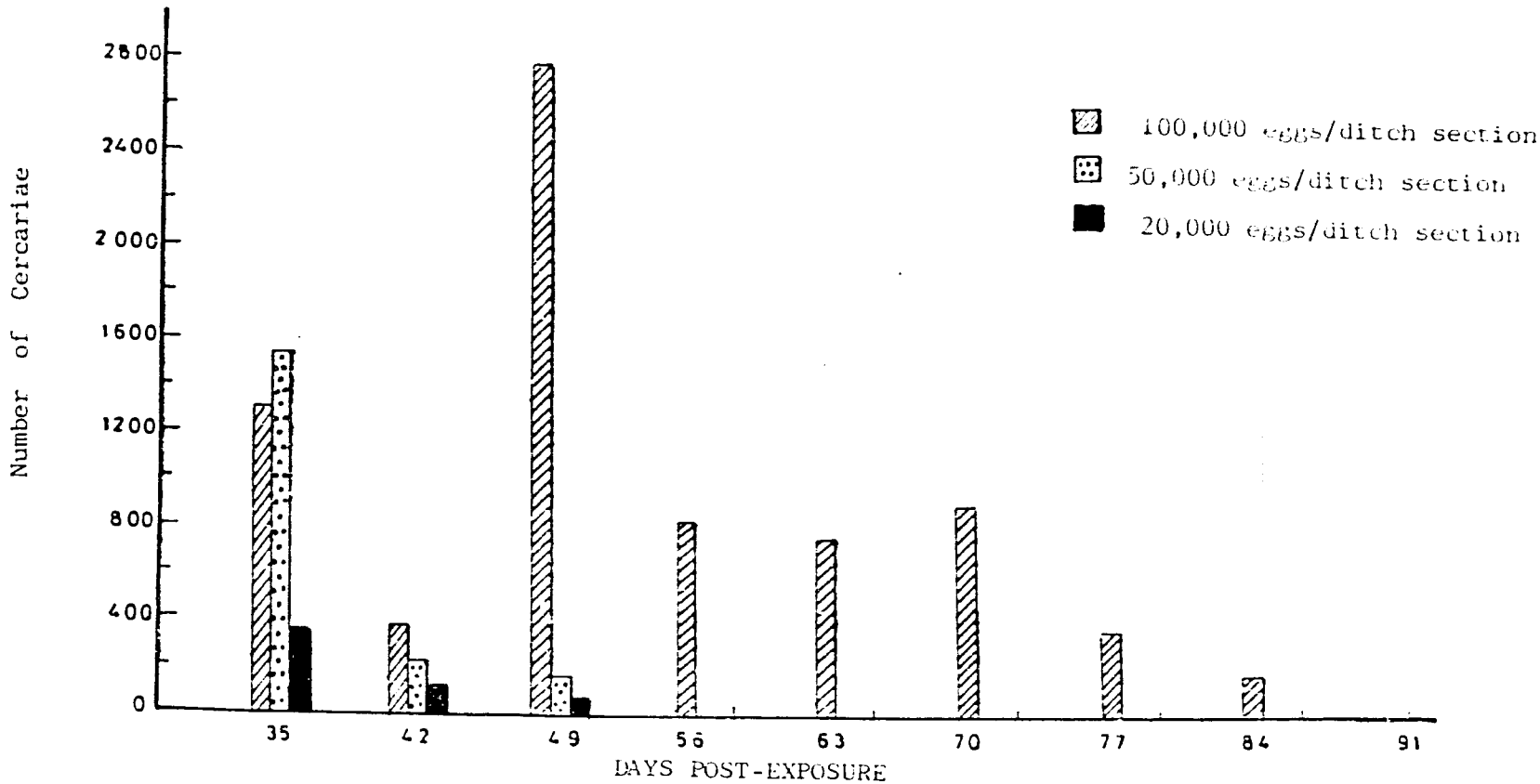


Fig. (12) Total Periodic Cercarial Production of free Biomphalaria alexandrina snails exposed to various numbers of Schistosoma mansoni eggs throughout the observation period.

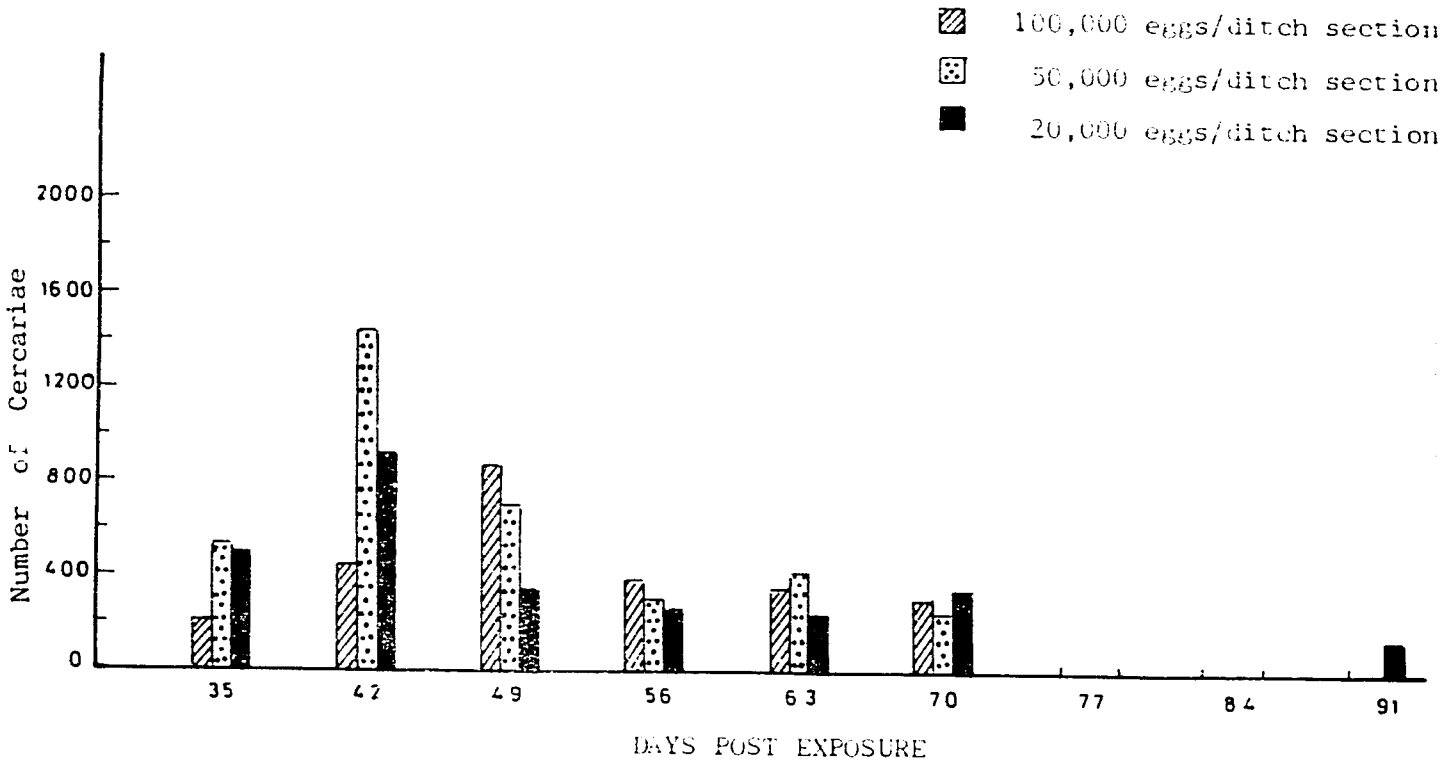


Fig. (**13**) Total Periodic Cercarial production of Caged Biomphalaria alexandrina snails exposed to various numbers of Schistosoma mansoni eggs throughout the observation period

Pilot Experiment No 3

DETERMINATION OF THE OPTIMAL NUMBER OF ECHINOSTOMA EGGS NEEDED FOR EACH DITCH SECTION TO INDUCE HIGH RATE OF INFECTION IN SNAILS.

Comments

- The cumulative number of infected snails increased with the increase of Echinostoma eggs reaching a mean of 16 snails in the case of 1,000,000 eggs / section (Table 19).
- This result gives indication that it is needed to increase the number of introduced eggs / section to about 5,000,000 to get high rate snail infection in the intervention experiment.

Table (19) Survivorship and infection of *Biomphalaria alexandrina* snails exposed to various numbers of *Echinostoma liei* eggs in ditch sections

Number of eggs ditch section	600,000			700,000			1,000,000		
Time (Days post exposure)	No. of collected snails	No. of snails shedding cercariae	Cumulative number of infected snails	No. of collected snails	No. of snails shedding cercariae	Cumulative number of infected snails	No. of collected snails	No. of snails shedding cercariae	Cumulative number of infected snails
46	309	0	0	457	0	0	324	11	11
58	340	0	0	416	2	2	372	7	12
64	304	2	2	467	4	6	269	6	12
72	327	6	8	320	6	11	292	8	16
86	209	0	8	231	2	11	171	1	16
95	117	0	8	100	1	11	75	0	16
102	85	0	8	167	1	12	44	0	16
115	35	0	8	79	0	12	23	0	16
120	77	0	8	71	0	12	32	0	16

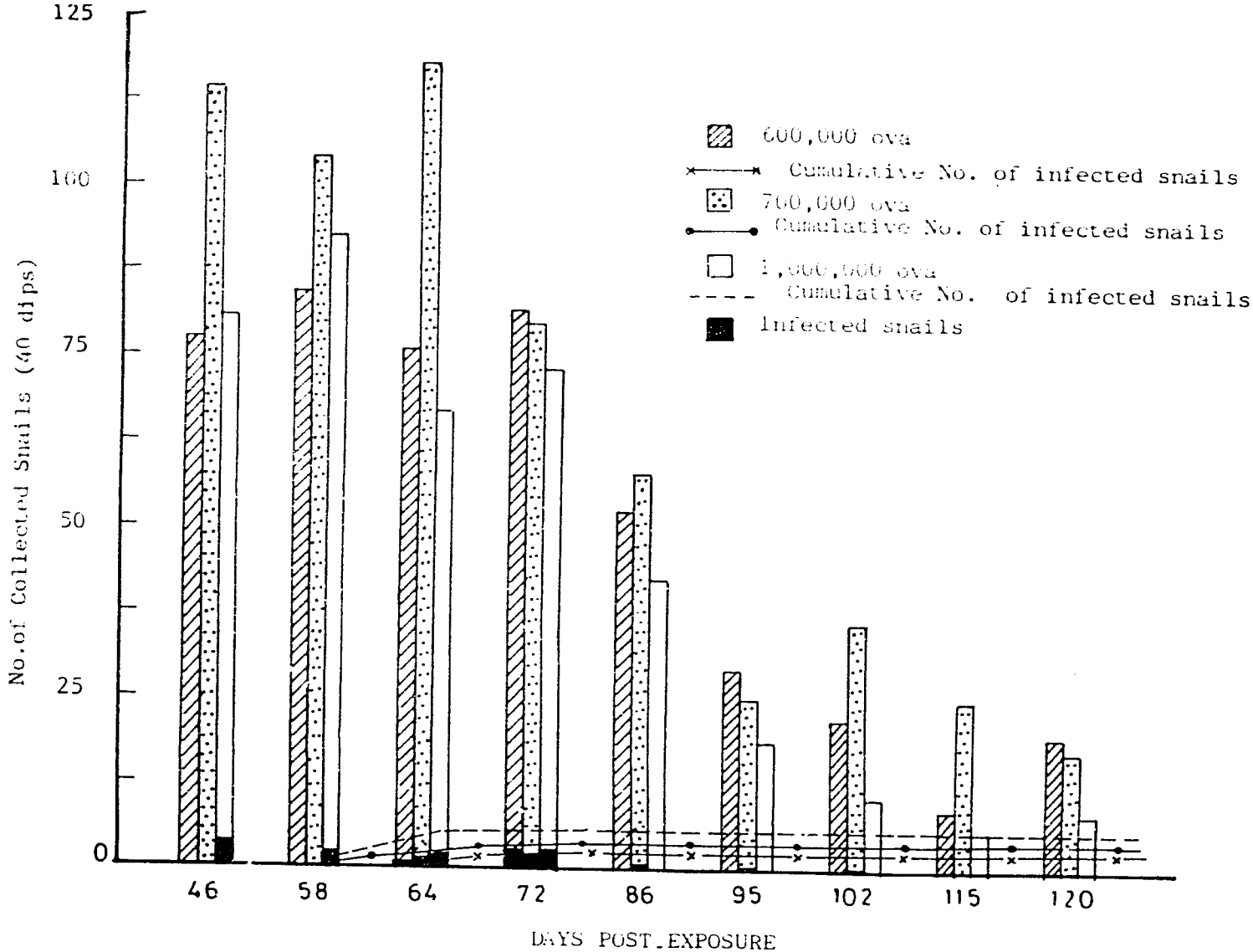


Fig. (14) Survivorship and infection of Biomphalaria alexandrina snails exposed to various numbers of Echinostoma liei eggs in ditch sections.

EFFICIENCY OF CERTAIN TOOLS (DIP NET, DRAG SCOOP AND MODIFIED EKMAN GRAB) IN SNAIL SAMPLING FROM THE DITCHES

<u>Comments</u>

- A thorough comparison of the efficiency of the three sampling tools is present in tables 20 and 21
- The dip net appears to be the easiest tool for snail sampiling. Its main advantage in comparison to other tools is the short time required to obtain a sample. This period is about 20 % the time taken by the drag scoop and 9% the time of Ekman grab to get a sample.
- The drag scoop is the best tool to get the greatest number of snails per dip and comparing the recovery rate of snails, from various population densities, it proved to be the most efficient in representing the real density.
- From the age - structure, it is possible to estimate many important parameters as reproduction and mortality rates. So, data can be made much more valuable by determining the age - structure of the population. Table (21) shows the efficiency of the three tools in representing all size - groups of the population. The present results indicate that no tools tested here provide the same proportion of different size - groups as the real size structure of the population, except the Ekman grab in the case of 4000 snails / section (the highest density). But generally, the size structures of populations collected by the 3

tools are not bad and are not far away from the real structre of population.

- In evaluation the efficiency of the 3 tools, mean recovery rate or 0.4' with 95% confidence of ± 0.089 was found for both the drag scoop and Ekman grab. While this rate for dip net was only 0.29 with confidence of ± 0.27.

Table (20) Efficiency of certain tools (dip net, drag scoop and modified Ekman grab) in snails sampling from the ditches.

Snail density	Collecting tool	Dip net		Drag scoop		Ekman grab	
		Mean	S.d	Mean	S.d	Mean	S.d
	Number of dips	40		40		24	
1000/ditch (222/cu.m)	No. of snails/dip	3.3	6.3	5.1	5	3.2	2.8
	No. of snails /cu.m	58.94	59.18	92.4	98.79	88.98	67.65
	Time/dip (sec.)	9.4	7.2	61.4	12.2	156	72.10
	Time/cu.m (min)	2.8	1.35	18.27	4.64	70.3	24.16
	No. of snails/cu.m/min.	22.8	23.69	4.6	4.44	1.26	0.89
	Recovery rate to population tested.%.	26.5		41.6		40.08	
2000/ditch (444/cu.m)	No. of snails/dip	9.9	10	12.2	10.6	8.3	7.1
	No. of snails /cu.m	178.13	199.28	217.8	204.67	224.1	234.07
	Time/dip (sec.)	14.4	13.7	84.7	19.7	176.8	98.3
	Time/cu.m (min)	4.29	4.17	25.2	7.27	79.6	52.33
	No. of snails/cu.m/min.	38.4	25.27	8.2	6.78	3.3	3.17
	Recovery rate to population tested.%.	40.1		49		50.4	
4000/ditch (888/cu.m)	No. of snails/dip	10	9.7	17.5	12.8	10.8	10.7
	No. of snails /cu.m	179.02	178.69	314	243.6	293.9	350.3
	Time/dip (sec.)	22.7	3.81	92.6	21.3	181.7	87.2
	Time/cu.m (min)	6.77	0.94	27.57	8.45	81.87	31.67
	No. of snails/cu.m/min.	26	25.48	10.96	7.23	3.25	2.45
	Recovery rate to population tested.%.	20.2		35.4		33.1	
	Mean recovery rate to population tested	29 %		42 %		41.2 %	

Table (21) Efficiency of the sampling tools in representing size structure of the snail population.

Tools	Snails density / ditch								
	1000			2000			4000		
	Size groups of snails *								
	Small	Med.	Large	Small	Med.	Large	Small	Med.	Large
Original ratios of different size groups (%)	5	70	25	8	68	25	8	68	25
	Recovered ratios of different size groups.%.								
Dip net	13	60	27	21	62	17	13	77	10
Drag Scoop	15	68	17	18	64	18	7	83	10
Ekman grab	23	53	24	20	56	24	7	65	28

* Small gp. : 2 - 4 mm shell diameter
Medium gp. : 4 - 9 mm shell diameter
Large gp. : 9 - 13 mm shell diameter

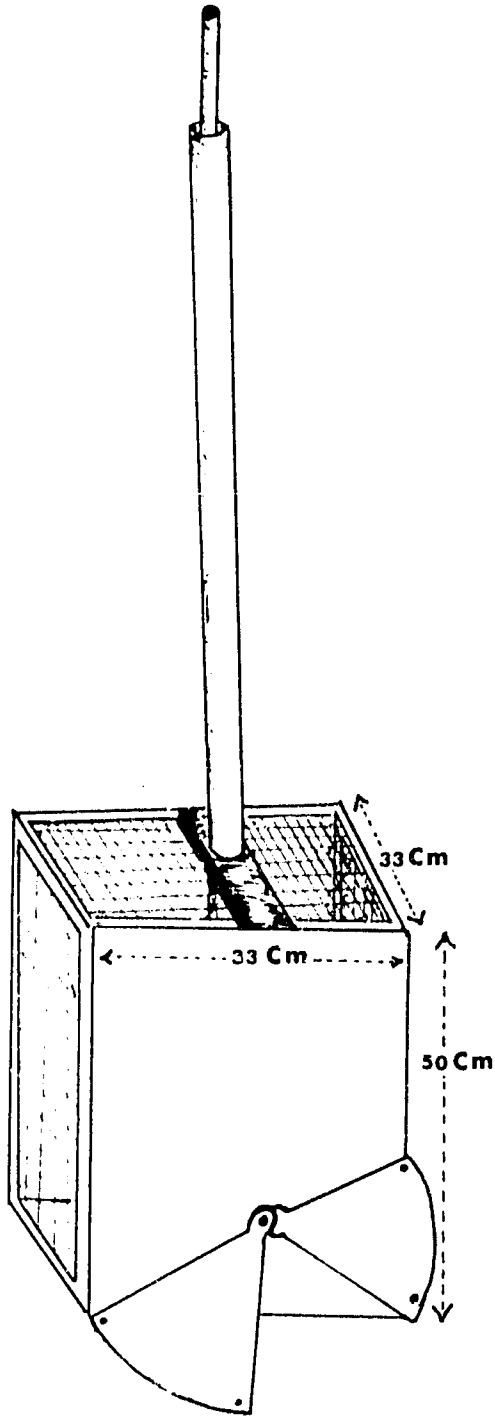


Fig (15) Diagramatic drawing for the modified Ekman grab

EFFECT OF MIRACIDIAL DOSE AND HABITAT VOLUME ON INFECTION OF BIOMPHALARIA ALEXANDRINA WITH SCHISTOSOMA MANSONI

- To detect the proper miracidial dose needed for getting a suitable rate of infection in B. alexandrina with S. mansoni in the ditches, it was necessary to carry out some laboratory experiments for the effect of water volume and miracidial and snail densities on infection.

- In this experiment, four different volumes of water were used; 4,8,20 and 40 litres. Three miracidial densities were tested namely 100, 500 and 1000/L of water volume. In all cases the snail density was 10 /L.

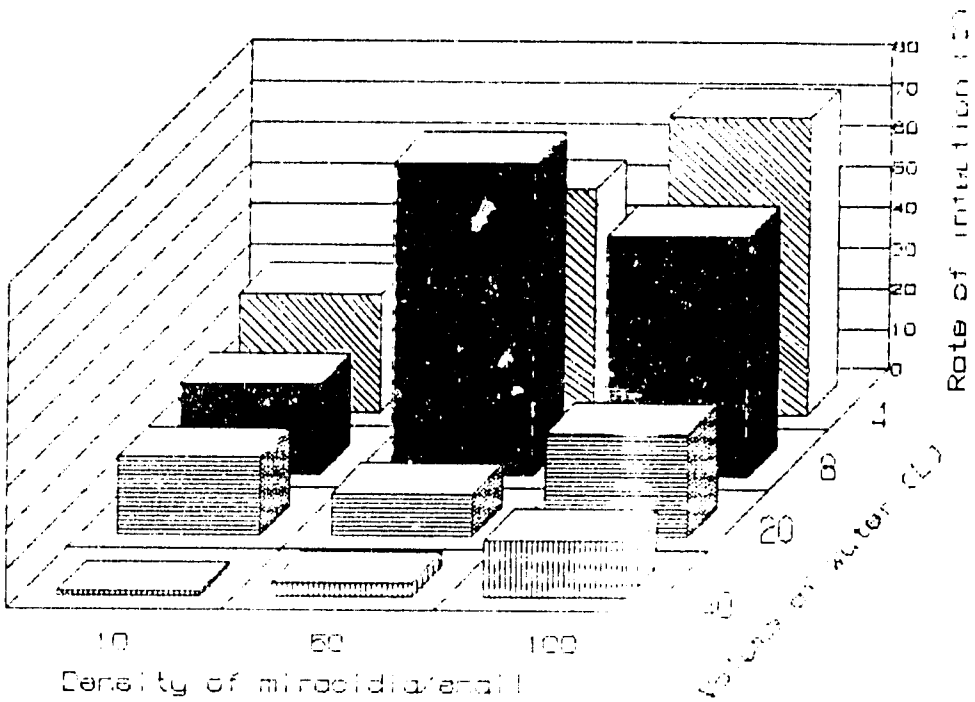
- The number of surviving snails at first shedding and the number of infected snails relative to surviving ones were determined. Results are summerized in table 22 and Fig. 16.

- It appears from these results that infection rate of snails decreases markedly with increase in habitat volume and with decrease of miracidial density. Ten fold increase of habitat volume gave rise to about 1/40, 1/17 and 1/5 of the infection rate with miracidial / snail densities of 10, 50 and 100, respectively.

Table 22: Effect of miracidial dose and volume of water on infection of *Bicephalaria alexandrina* with *schistosoma mansoni*

Volume of water (L)	Miracidial dose 10 / snail			50 / snail			100 / snail		
	survived snails at 1st shedding	shedding snails		survived snails at 1st shedding	shedding snails		survived snails at 1st shedding	shedding snails	
		cumulative number	%		cumulative number	%		cumulative number	%
4	38	11	29	29	16	55.2	37	27	72.9
8	72	16	21.9	77	59	76.6	34	20	58.8
20	165	31	18.8	139	14	10.1	131	33	25.2
40	142	1	0.7	91	3	3.3	187	26	13.9

Fig. (**16**). Effect of miracidial dose and volume of water on infection of *Biomphalaria alexandrina* with *Schistosoma mansoni*.



Appendix of Photographic Plates

Plate (1) Experimental area showing field lab and underground water.

Plate (2) Experimental area showing the wooden sunshade of the ditches, main feeding channel and water inlet to a ditch covered with a sieve.

Plate (3) Ditch partitions with windows opened, cages containing snails and foam plates for sampling of snails egg masses.

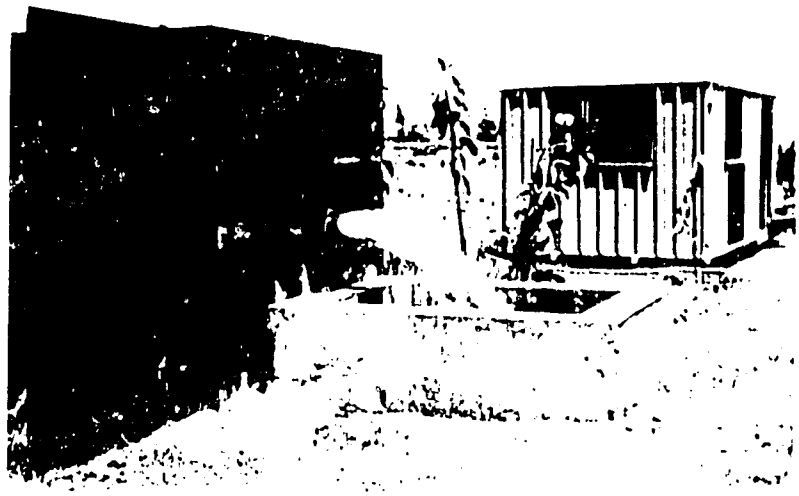
Plate (4) Snail sampling with the drag scoop and modified Ekman grab.

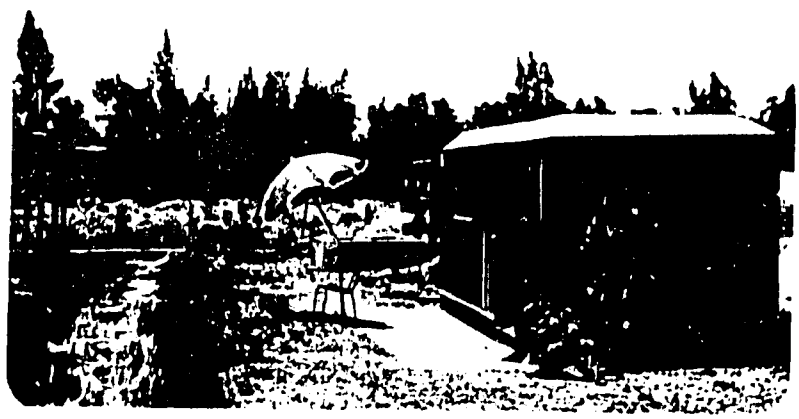
Plate (5) Explosion of algae and aquatic vegetation.

Plate (6) Setting the "Ryan" thermograph apparatus and measuring of water pH.

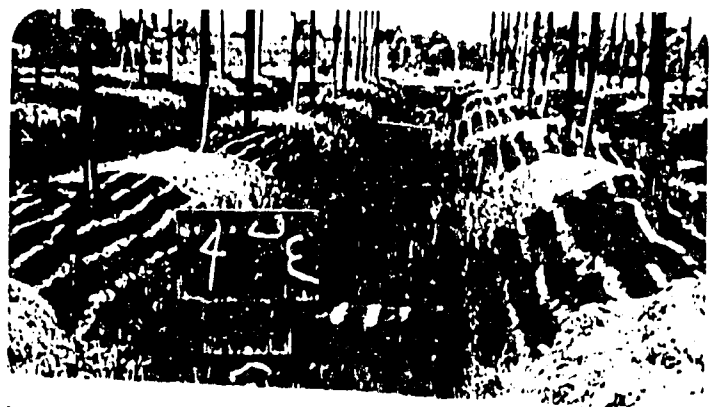
Plate (7) Natural habitats from which *Biomphalaria* snails were collected.

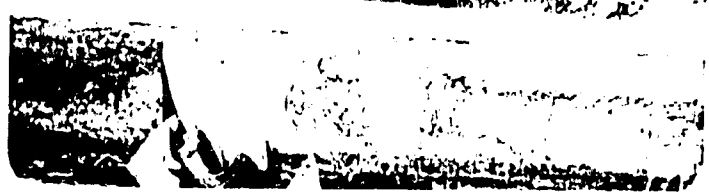
PL.1





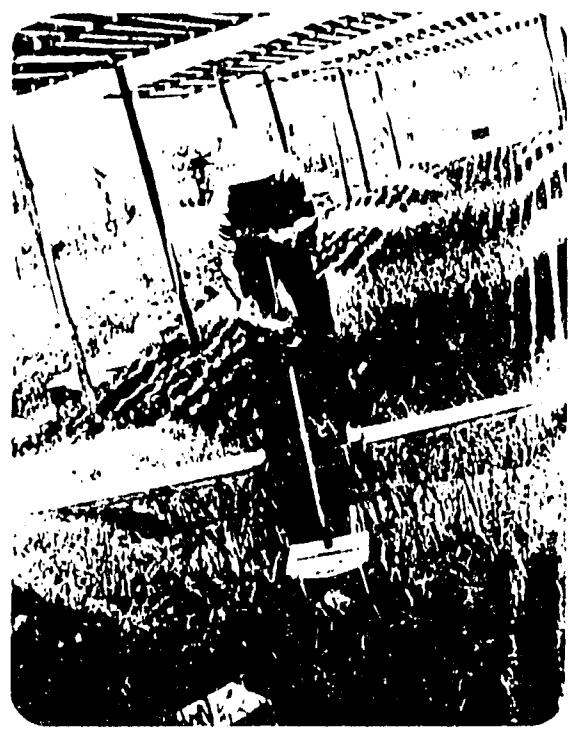
PL. 2





PL. 3

PL. 4



PL.5

PL.6

PL.7

USAID PROGRAM IN SCIENCE AND TECHNOLOGY COOPERATION

Report on the Visit of Dr. E. Malek
to Research Project No. 936-5542
Cairo, Egypt

Duration of the visit: 14 May - 27 May 1987

The project involves the testing of Echinostoma revolutum (=E. liei) against Schistosoma mansoni under simulated and natural field conditions. It has two phases: the preparation phase which lasts for one year, and the intervention and evaluation phase which is projected for two additional years.

The visit coincided with the beginning of the preparation phase and the following activities were carried out.

1. Visit to the experimental area

The experimental area is at about 25 kilometers north of Cairo and had been constructed in a research farm belonging to the Theodor Bilharz Research Institute.

The experimental area was described in the grant proposal. Eight of the 18 parallel ditches will be used in the project. All ditches are already divided by partitions into 3 sections each. Instead of the experimental ditches being supplied with Nile water from a main irrigation canal underground water will be used. A pump has already been installed shortly before the visit, and underground water is being used.

Also during the visit laborers were in the process of installing shades for the ditches. This steel and wood shading is much higher than the previous straw shading which was described in the grant proposal.

2. Discussions of Ongoing Work:

Discussions were held with Drs. Yousif, Roushdy, El Emam, and Twefik concerning the ongoing work with respect to the project. These discussions involved the maintenance of the life cycle of Schistosoma mansoni and the maximum numbers of eggs which will become available for the experiments; the maintenance of the life cycle of Echinostoma liei and the maximum numbers of eggs which can be obtained; the collection of Biomphalaria alexandrina snails from field sources and their sorting out by size and the examination of snails for their natural infections.

It seems that all laboratory procedures are very satisfactory and will ensure the provision of eggs of S. mansoni, eggs of E. liei and snails in numbers large enough to be used in the experimental work.

3. Discussions of Studies for the Coming Year.

For the coming year some experiments will be conducted to obtain basic information for the second phase of the project. Among these experiments are those to determine the optimal snail numbers to be introduced in each of the 3 sections of the ditches e.g. 1000, 2000 or 4000; to determine the optimal number of schistosome eggs to produce a moderate rate of infection in the snails and to determine the optimal number of echinostome eggs needed to produce a high rate of infection in the snails.

4. Lecture Delivered:

A 2-hour lecture was delivered to the Malacology Section of the Theodor Bilharz Research Institute. The lecture covered various aspects of malacology as well as biological control in general in particular the research project i.e. the intramolluscan intertrematode competition and predation.

5. Individuals Visited in Connection with the Project

Dr. Fouad Yousif, Project Principal Investigator

Theodor Bilharz Research Institute (TBRI)

Dr. Ahmed El-Garem, Director TBRI

Dr. Mohamed El Eman, TBRI

Dr. Magdy Twefik (Ain Shams University)

Dr. Menriet Zaki Roushdy, TBRI

Dr. Nawal Haroun, TBRI

Mr. Lawrence Irwin, U.S. Agency for International Development (USAID)

Director, Science and Technology Office

Dr. Shereef Areef, USAID

USAID PROGRAM IN SCIENCE AND TECHNOLOGY COOPERATION

Report on the visit of Dr. Fouad Yousif to USA
through the Research Project No. 936 - 5542

Duration of the visit : 29 July - 27 August 1987

The program of the visit included :

1- Dept. of Tropical Medicine, Tulane University, New Orleans (Prof. Dr. Emile Malek, the American Collaborator in the Project) from 30 July - 14 August: During this period, there was an opportunity for visiting the laboratory of Medical Malacology of Prof. Malek and for discussing the progress of the project and the problems encountered. Meanwhile a thorough review of the literature was made as well as photocopying of many interesting papers. Moreover, a type of sieve, glass microfibre filters and two books with relevant contents to the project were purchased. Cataloques for materials and equipment were consulted. Nets to be used in the experimental area at Quanater, Egypt were purchased.

A special seminar was presented to the Faculty and students at the Department of Tropical Medicine, Tulane University. The seminar covered various asperts of the research project and during the seminar there was exchange of views and ideas about progress of the project and problems encountered in the field.

2- Dept. Of Epidemiology, University of California, San Francisco (Dr. Lie and Dr. Heyneman) (15 - 19 August)

During this period there was an opportunity to make a long meeting with Dr. Lie and Dr. Heyneman, to discuss with them various aspects of the research project and to exchange views.

Dr. Lie and Dr. Heyneman have a previous long experience with testing Echinostoma spp in the field, therefore their comments and ideas were very costructive and of practical help. I had the chance to review the library of the Deptartment and to get many reprints and photocopies of interesting publications from Dr. Heyneman.

3- Dept. of Immunology & Infections Diseases, Johns Hopkins University, Baltimore and USAID (Washington D.C.) (20 - 24 August)

During this period there was an opportunity to meet Dr. Clive Shiff in his office in USAID Washington and also to visit his Department in the Johns Hopkins University, Baltimore, to discuss with him problems met with in the semifield experiments, especially the problem of algae explosion, difficulties of sampling with the grab; Dr. Shiff kindly provided me with some ideas for the control of algae and for snail sampling under the conditions of the project.

4- University of Lowell : Centre for Tropical Diseases, Lowell (25 - 27 August, 87)

During this visit there was an opportuity to visit the laboratories of this centre, to meet Prof. Dr. John Bruce and his coworkers especially Dr. Liang and to study the techniques for laboratary cultivation of schistosome vector snails and maintenance of the life cycles of different spp. Of Schistosoma on a large scale .

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