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NTA and Mercury in Artificial Stream Systems

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NTA AND MERCURY IN ARTIFICIAL STREAM SYSTEMS

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Project 16050 GQQ
Program Element 1BA023

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Prepared for

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ABSTRACT

Studies were conducted in six artificial stream channels to determine the fate of NTA (nitrilotriacetic acid, trisodium salt) added with and without sodium phosphate to these systems. In the two hour period required for a given water mass to traverse the channels, there was no appreciable removal of NTA or phosphate, even after a one month period of continuous input. Visible biological differences were noted between the various treatments. These differences may have been a result of pH alteration caused by the addition of the trisodium phosphate and NTA.

In anticipation of a long term program involving the fate of mercury and possible mercury-NTA interactions, several modifications were incorporated into the artificial stream system. Based on the results of laboratory studies, a mercury removal system utilizing shredded rubber tires as obtained from commercial tire recapping firms, was constructed. Laboratory studies indicated that NTA did not influence the uptake of mercuric ion by the rubber. The presence of NTA did alter the uptake pattern and final concentration by mosquitofish, *Gambusia affinis*.

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SECTION I

CONCLUSIONS

1. There is no detectable degradation of NTA, either added alone or with trisodium phosphate, during a two hour period in artificial stream systems.

2. Biological differences between treatments, as determined by visual observations and bacterial counts, may have been the result of treatment - induced pH changes in these soft water systems.

3. Granulated vulcanized rubber, a waste product of commercial tire recapping operations, provides a convenient and economical method of removing inorganic mercury from water. Uptake is not affected by NTA.

4. NTA does affect the uptake of mercury from water by mosquitofish, *Gambusia affinis*.

SECTION II

RECOMMENDATIONS

1. The interaction of NTA with mercury and other metals needs intensive investigation before NTA is released in large quantities to the environment. Especially needed are studies to determine NTA effects on heavy metal movement between components of aquatic ecosystems and how these are influenced by other additions (thermal, organic, chemical, etc.) to the aquatic habitat.

2. The use of vulcanized rubber scrap as a mercury removal agent should be futher investigated. Decontamination of problem areas in the environment may be feasible.

SECTION III

INTRODUCTION

Phosphates have been implicated in the increased rate of eutrophication in some streams and lakes. Since a great portion of the phosphates introduced by man into the environment result from the use of detergents, a great effort has been made to find phosphate substitutes. One compound that is now in use in some countries is the trisodium salt of nitrilotriacetic acid, usually referred to as NTA. This compound was voluntarily removed by manufacturers from detergents in the United States at the request of the Surgeon-General. The request was made because of the many unknown factors involved in the release of great quantities of a relatively unknown compound into the environment. Two areas of general concern were the possible production of carcinogenic products under certain conditions of NTA degradation, and the interaction of NTA, a well known chelating agent, with heavy metals in the environment.

The work described in this report consists of two phases. The first phase, an attempt to determine the fate of NTA introduced into artificial stream systems, was carried out in conjunction with personnel from the Environmental Protection Agency's National Pollutants Fate Research Program, Southeast Water Laboratory, Athens, Georgia. The second phase involved modifications of the stream facility and preliminary experiments with mercury and mercury-NTA interactions in preparation for a long term mercury study. The mercury work is presently underway.

SECTION IV

ANALYTICAL METHODS

Analyses for NTA, total organic carbon, total inorganic carbon, orthophosphate and nitrate were carried out by personnel of the Southeast Water Laboratory on samples prepared at the stream site and transported on ice to Athens, Georgia.

NTA was determined by the Zinc-Zincon semi-automatic procedure for the Technicon Autoanalyzer as described by Thompson and Duthie (1968). The minimum detectable concentration of this method is 0.2 mg/liter as Na_3NTA. A hydrogen form cation exchange resin was used to remove cations that might have interferred.

Total organic carbon was determined from acidified nitrogen-purged samples on a Beckman Model 915 Total Carbon Analyzer coupled to a Beckman Model 215A Infrared Analyzer.

Total inorganic carbon was determined by the procedure described by Kerr *et al*. (1970) utilizing a Varian Aerograph Gas Analyzer.

Orthophosphate and nitrate concentrations were determined on a Technicon Autoanalyzer according to procedures described in *FWCPA Methods for Chemical Analysis of Water and Wastes*, (1969).

Suspended bacteria were counted by serial dilution pour plates made with tryptone glucose extract agar.

All pH measurements were made with an Orion Specific Ion Meter and a Corning combination electrode.

Mercury determinations were made on a Coleman MAS 50 Analyzer modified by the addition of a digital read-out system. Fish were digested using the procedure described by Uthe *et al*. (1970). Analytical instructions provided with the Coleman Analyzer were followed.

SECTION V

EXPERIMENTAL RESULTS

Phase one

The artificial stream facility used in this study was constructed the summer of 1970 at the Atomic Energy Commission's Savannah River Plant. The particular site was chosen because of the availability of an adequate water supply and a level concrete platform of sufficient size. The facility consists of six concrete block channels 300 feet long, 2 feet wide and one foot deep with pools, 10 feet long, five feet wide and two feet deep, located at both ends. Figure 1 shows the six head pools, flow adjust valves, V-notch weirs, chemical feed systems and initial 30 feet of the stream channels. The system is arranged so that water can be input either into the pools or directly into the channels.

For this phase of the work, the entire system was lined with 11 mil transparent polyethylene sheeting, a covering selected because it was immediately available. Washed quartz builder's sand was placed in the channels to a uniform depth of 2 inches.

Water supplied from a deep well was input to the head pools and the valves adjusted to provide 25 gallons per minute into each channel as measured by weirs (Figure 2). The quality of the well water has been periodically monitored by E. I. duPont personnel since 1952 and the measured parameters vary little in time. A typical analysis is given in Table 1. Adjustable plates at the ends of the channels were set to maintain a water depth of eight inches. With this depth and an input of 25 gallons per minute, retention times averaged two hours in the channels and thirty minutes in the pools.

Water flow throught the channels was started in late August of 1970. Material collected from local ponds and streams originating from artesian outflows from the same aquifer supplying the artificial streams, was

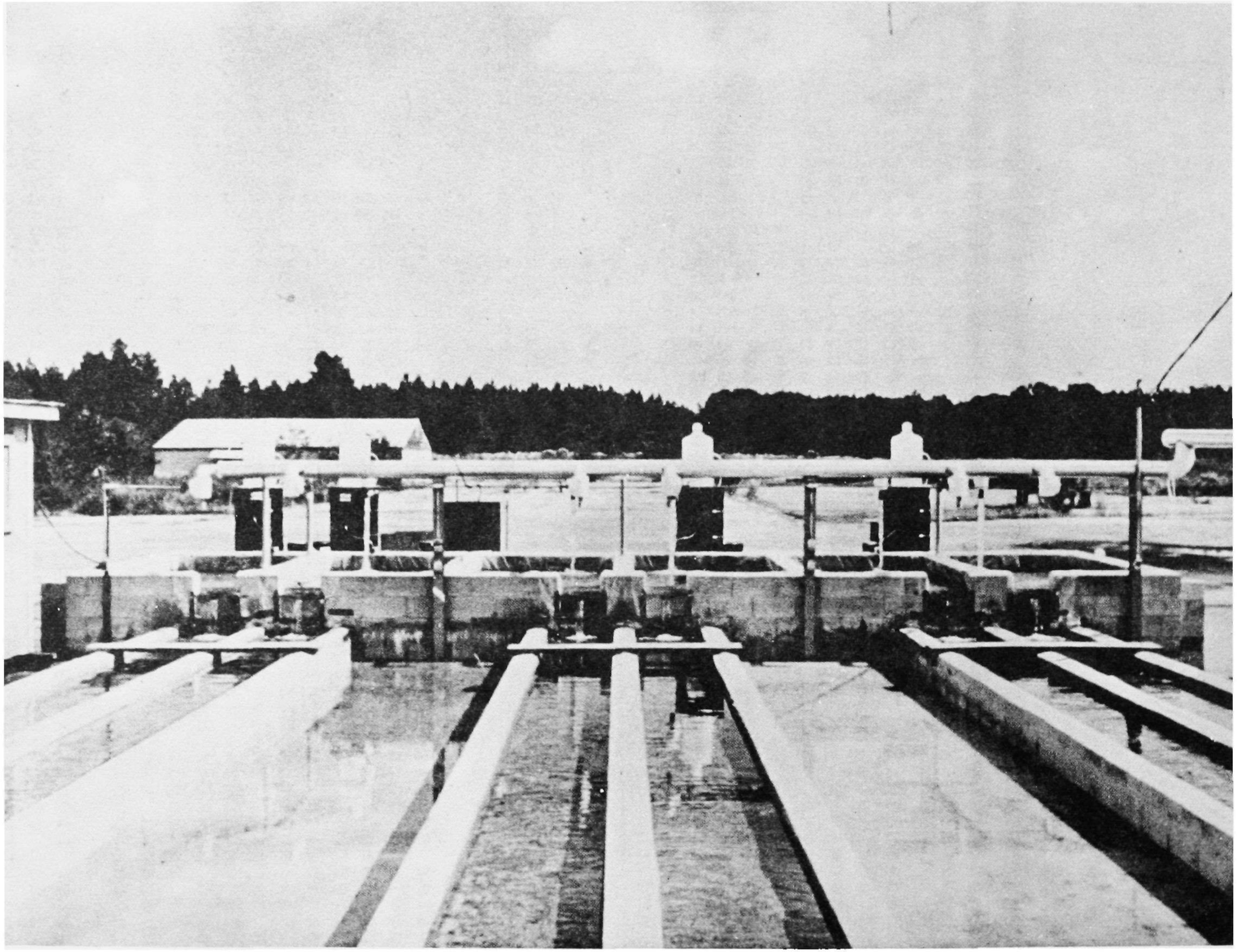

Figure 1. View of upper portion of stream channels, head pools, and water input system.

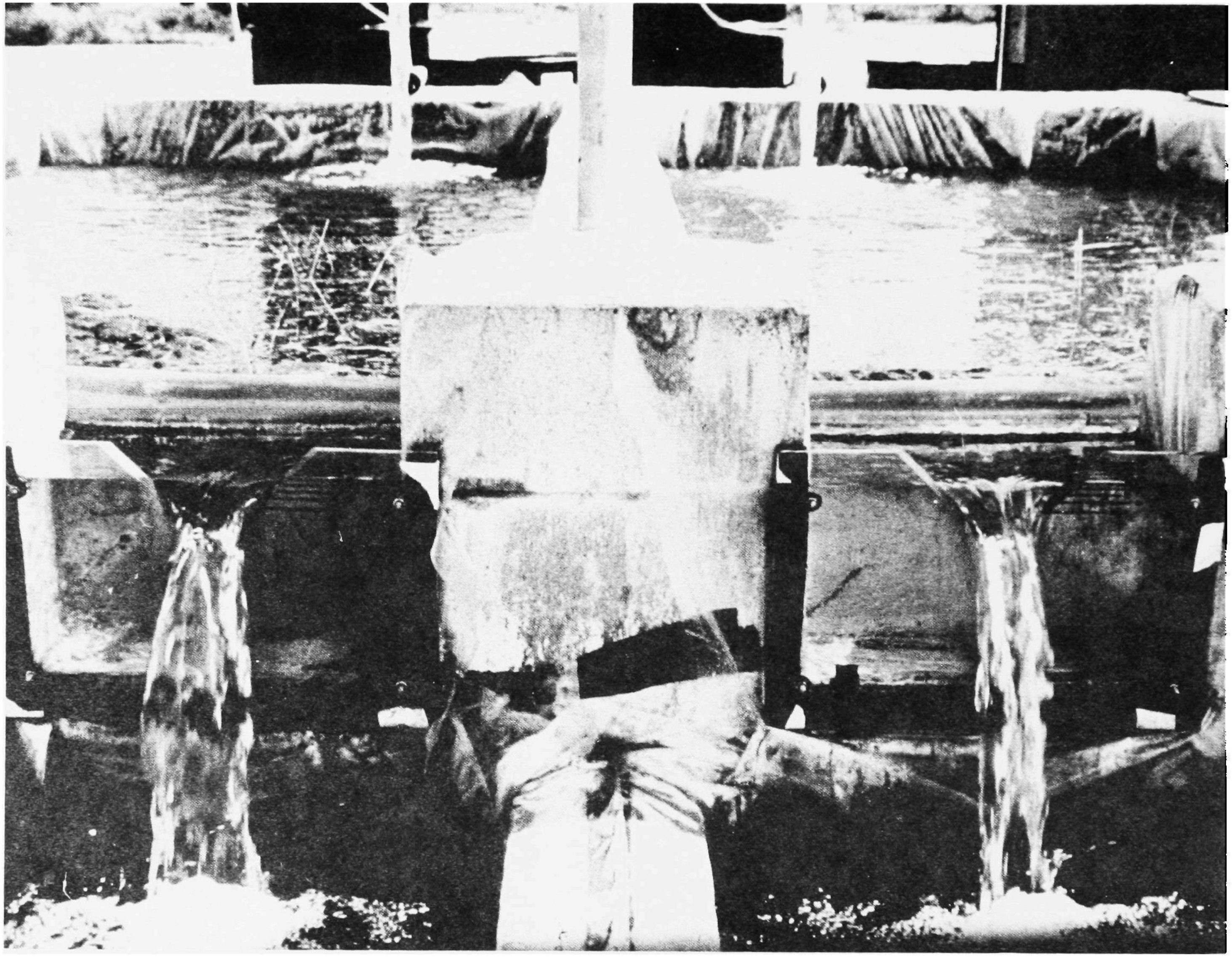

Figure 2. Weirs by which flows into the channels are measured.

TABLE I

WELL WATER ANALYSIS

Total Organic Carbon	3.2	mg/liter
Inorganic Carbon as CO2	24.75	mg/liter
Nitrate	.190	mg/liter
Orthophosphate	.0015	mg/liter
Hardness Ca CO_3	5.0	mg/liter
Dissolved Oxygen	9.5	mg/liter
Temperature	21.0°C	
pH	4.7	

introduced on several occasions into the head pools and channels for seeding purposes. Unsuccessful attempts were made to introduce fish (Gambusia affinis and Semotilus atromaculatus) into the channels. The last seeding was in mid September when 10 gallons of material were placed in each head pool, and the water flow reduced for a period of 24 hours to allow organisms to settle. The channels were then not disturbed for one month except for periodic adjustment of the flows. After this period, an intensive diel study was conducted to establish pre-dosing conditions and optimum sampling times. At that time large floating mats (algae and bacteria) had formed in all the head pools. Filamentous growth was visible on the walls and bottoms of both pools and channels. The growth was greatest near the input ends of the channels. The channels appeared to be very similar.

A chemical feed system was installed near the head pools to provide a continuous input of NTA and phosphate solutions. This system (Figure 3) consisted of four 54 gallon polyethylene-lined drums, air lift pumps which maintained a constant liquid level in four polyethylene bottles located on the drums, and four solenoid valves which were electrically pulsed to maintain the desired input rate. Flows from the solenoid valves entered the head pools near where the water was input so as to provide instant mixing.

Of the six channels, two were maintained as controls (1 and 4, looking from tail to head and counting left to right), two (3 and 5) received inputs of NTA and trisodium phosphate to provide 10 mg/liter of each compound, one (2) received NTA alone, and the last (6) received Na_3PO_4 alone, again to provide concentrations of 10 mg/liter. The chemical feed system was started on October 22, 1970. A continuous input was maintained until November 23, 1970 when subfreezing temperatures caused the feed system to stop and the experiment was terminated.

During the period of phosphate-NTA input, water samples were taken at 6 am and 2 pm daily from both ends of each channel. Temperature and pH were determined in situ. Water samples were fixed at the streams for titrating for dissolved oxygen. A single 150 ml water sample was taken for NTA, phosphate, and nitrate determinations. This water was filtered through a washed 0.45 micron sterile membrane filter into an acid rinsed sterile container and immediately refrigerated. Total organic carbon samples were taken in acid washed sterile containers, acidified immediately and refrigerated until analyzed. Inorganic carbon samples were filtered through 0.22 micron filters

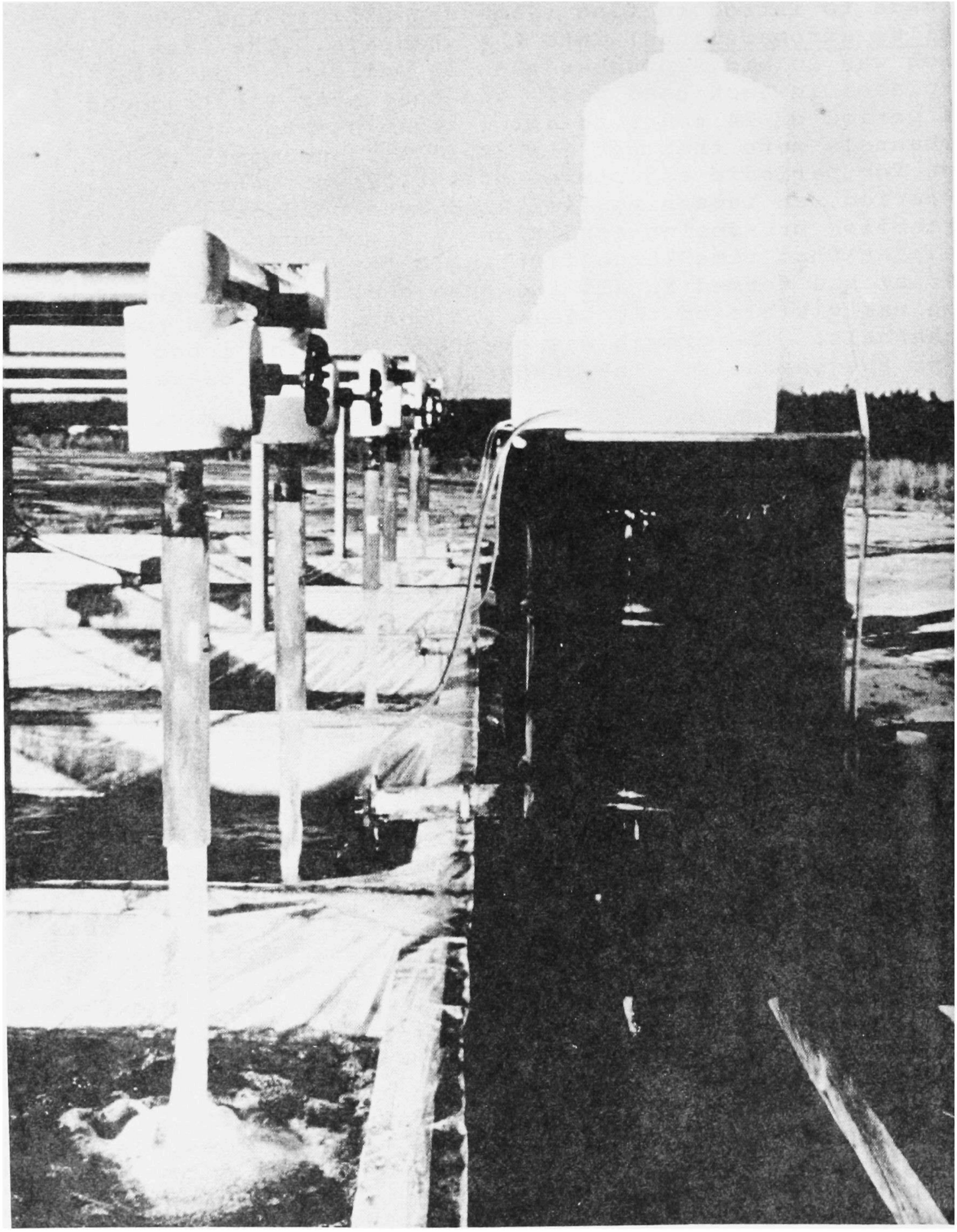

Figure 3. NTA and phosphate chemical feed system.

into air tight vials, sealed and refrigerated. Bacterial samples were taken in acid washed sterile containers, acidified immediately and refrigerated until analyzed. Inorganic carbon samples were filtered through 0.22 micron filters into air tight vials, sealed and refrigerated. Bacterial samples were taken in sterile dilution bottles and pour plates made within 15 minutes. The results of all chemical analyses and bacterial counts were punched on computer cards and computer programs written for data analysis by Southeast Water Laboratory personnel. A summary of results is given in the final report of Project 16050 GJB, Environmental Fate of NTA, (SEW, 1971). The variability of the data was such that tests for upstream-down-stream changes, and differences between treatments were very insensitive. No significant removal of NTA in any of the channels was detected. There were significant differences between treatments with respect to the bacterial counts, $Na_3PO_4 > Na_3PO_4$ + NTA > NTA controls, as determined by a Duncan's Multiple Range Test. This ranking corresponds to the average pH of the water in the channels, which was greatly influenced by the additions of the NTA and Na_3PO_4.

During the period of this study, several general observations were made that could not be considered quantitatively due to the lack of available personnel. At the start of the chemical input, there were extensive algal mats in all the head pools. The character of the mats other than the controls changed in time, apparently as a result of the chemical input. The mats in pools receiving NTA, either alone or with phosphate, became a brighter green, while the mat in the pool receiving only Na_3PO_4 was destroyed and transported over the weirs into the channels. The mats in the remaining pools were manually destroyed and allowed to pass downstream to maintain all channel systems in a similar condition. Microscopic examination of the mats shortly before this time showed no gross differences in qualitative species structure. Mats reformed quickly in the control pools, more slowly in all the pools receiving NTA, and did not form at all in the pool receiving only Na_3PO_4.

These observations suggest an effect of NTA on the biological communities, even though the results of chemical analyses did not indicate a significant removal, hence utilization, of this compound. Also indicated is some type of interaction between NTA and trisodium phosphate.

Phase two

The initial experiments with NTA demonstrated a number of problems with the artificial stream facility. No attempt was made to rectify most of these during the NTA work so that all personnel time could be devoted to data acquisition. The next use planned for the stream facility was a study to determine the fate of low levels of mercury introduced as mercuric ion into the channels and to determine biological effects resulting from the mercury. A great deal of preparation was necessary before this work could be started.

When the NTA study was over, all sand was removed from the channels and the plastic lining, which was already beginning to deteriorate, was removed. Based on the recommendations of experts familiar with reservoir and irrigation channel linings, a 20 mil black PVC (polyvinyl chloride) film was selected for lining material. This plastic was purchased so that each channel lining would be a single piece. After the streams and pools were covered, a two inch layer of new sand was placed on the bottoms of the channels. To cut down on the input of organic material into channels 1 and 6 (insects, mice, spiders and millepedes were often found in these channels), a PVC lined moat about six inches deep was constructed on each side of the channel system (Figure 4). The moat next to channel one also served to eliminate the extra heat input to the outside wall of channel one by direct sunlight. The areas between the channel pairs were partitioned and flooded to somewhat alleviate excessive heating of channel three and channel five walls and, secondarily, provide additional experimental chambers and conditioning areas. These are also visible in Figure 4.

Continuous recording water quality monitors were installed at the head and tail ends of the channels. These units, Schneider Instrument Company RM25 Robot Monitors, have provisions for recording dissolved oxygen, pH, conductivity, temperature and solar radiation input. A switching system was designed so that each channel is monitored for ten minutes each hour. The submersible pumps and building housing the monitor at the head end of the channels are shown in Figure 5.

When stream operation was first begun as described in Phase one of this report, a number of attempts were made to introduce fish, especially mosquitofish (Gambusia affinis) into the channels. No amount of conditioning was sufficient to allow these animals to survive for more than several days. Twenty yards from the ends of the channels in the effluent stream, however, there appeared a breeding mosquitofish population

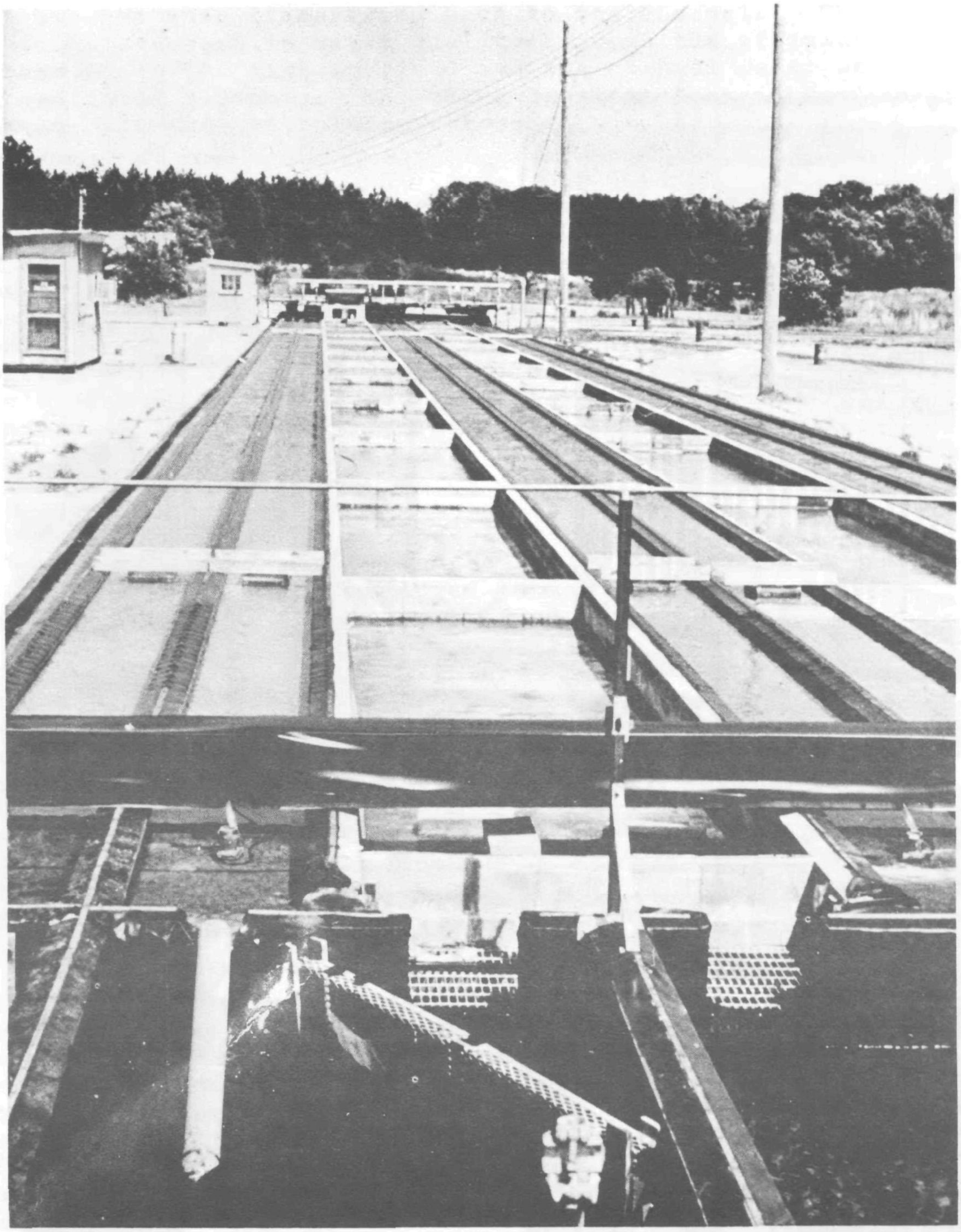

Figure 4. View of PVC covered channels showing moat at extreme left and pump system feeding water quality monitors.

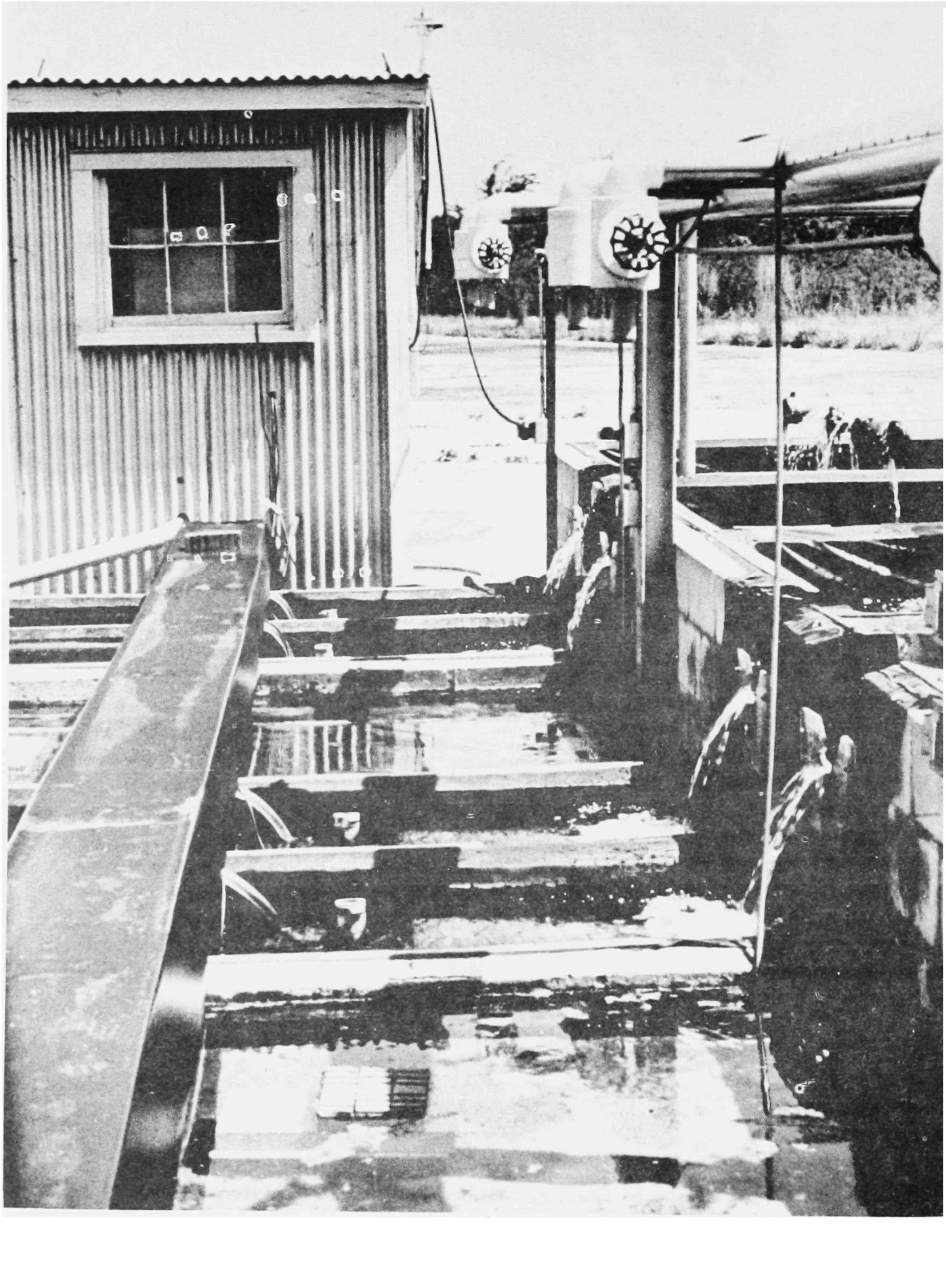

Figure 5. Pumping system and water quality monitor at head end of streams.

resulting from animals that had escaped. Even these could not survive when transferred back to the channels. The major differences in water quality between the effluent stream where the fish survived and the channel water were pH and total hardness. The water leaving the tail pools passes with much turbulence through a poured concrete channel.

A water treatment system was installed at the head pools to make the water entering the channels more like what is found in natural streams in this area. The cost of a single large system to treat all the water, the most desirable approach, was too high and six separate systems were constructed. Each consists of three PVC sewer pipes filled with limestone chips through which the water must pass before entering the head pools (Figure 6). We have found that the limestone must be changed at least monthly to maintain a relatively constant water quality. Fish survive with no difficulty in the treated water.

After the installation of the water treatment facility, the water was turned on, adjusted to a flow of 25 gallons per minute, and the streams allowed to seed naturally. Four hundred mosquitofish were introduced into each channel.

A major source of variability in the NTA-phosphate data was due to erratic functioning of the chemical feed system. The pulsed solenoid valve technique, which has been used so successfully in laboratory studies at the Southeast Water Laboratory, behaved poorly in the field because of the highly alkaline phosphate solutions and the extreme temperature fluctuations. For the mercury study, a four channel peristaltic tubing pump was obtained (Figure 7) and procedures established whereby the pump is recalibrated daily. The mercury solutions are input directly into the channels at a depth selected from the results of a series of dye dispersal studies.

Because of the sensitive position of the Savannah River Plant with respect to environmental contamination, there was some concern over our proposed experiments which would result in releases of mercury to the environment. A method was sought whereby mercury in water from the channels would be removed before the water was released into natural streams. A number of approaches were suggested but the most feasible appeared to be a system that would utilize the ability of vulcanized rubber to tightly bind mercury. This property of rubber was pointed out by Mr. Edwin R. Russell of the Savannah River Laboratory (SRL) and, indeed, forms the basis for a patent application by Mr. Russell and others from SRL.

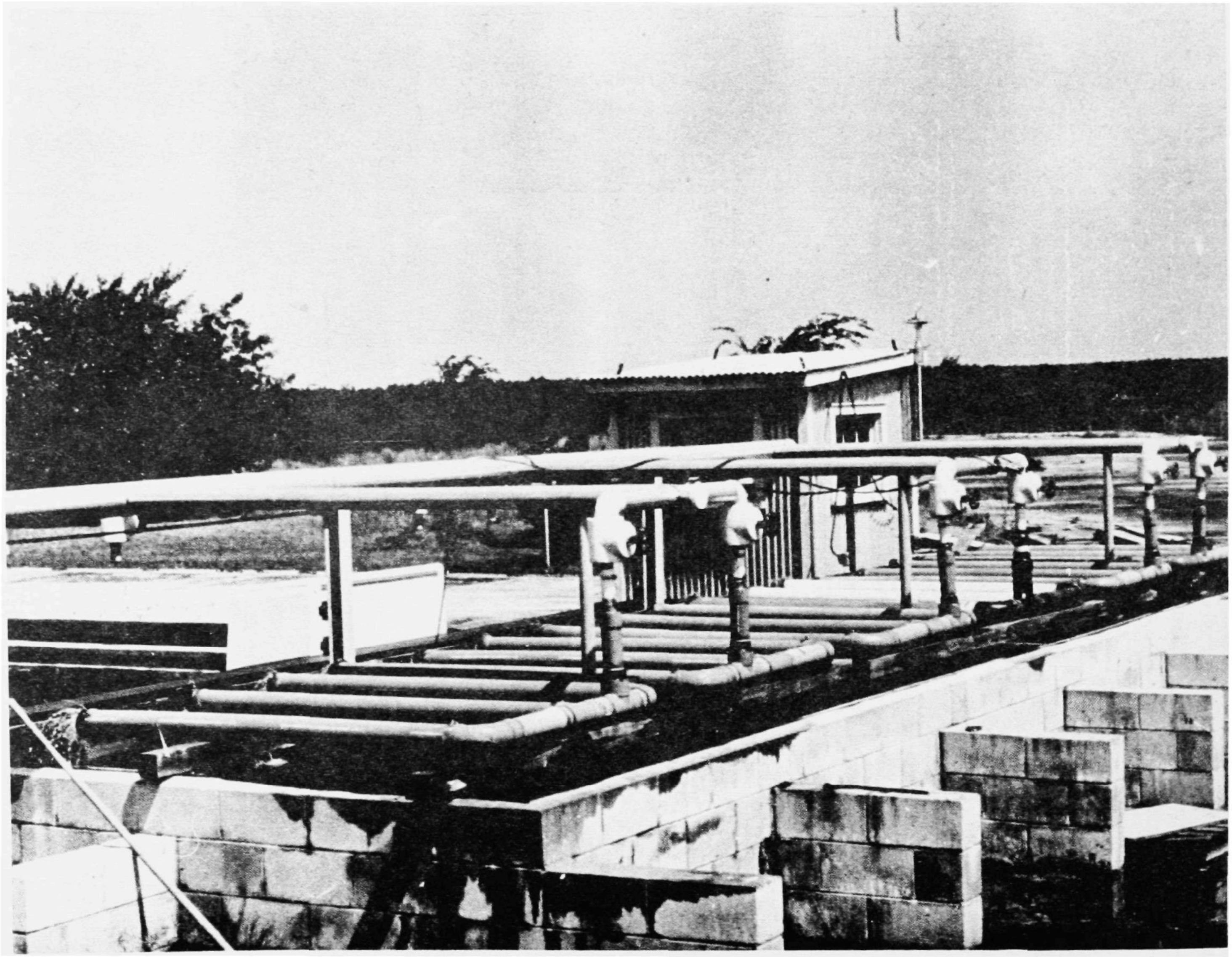

Figure 6. Water treatment facility consisting of limestone filled tubes.

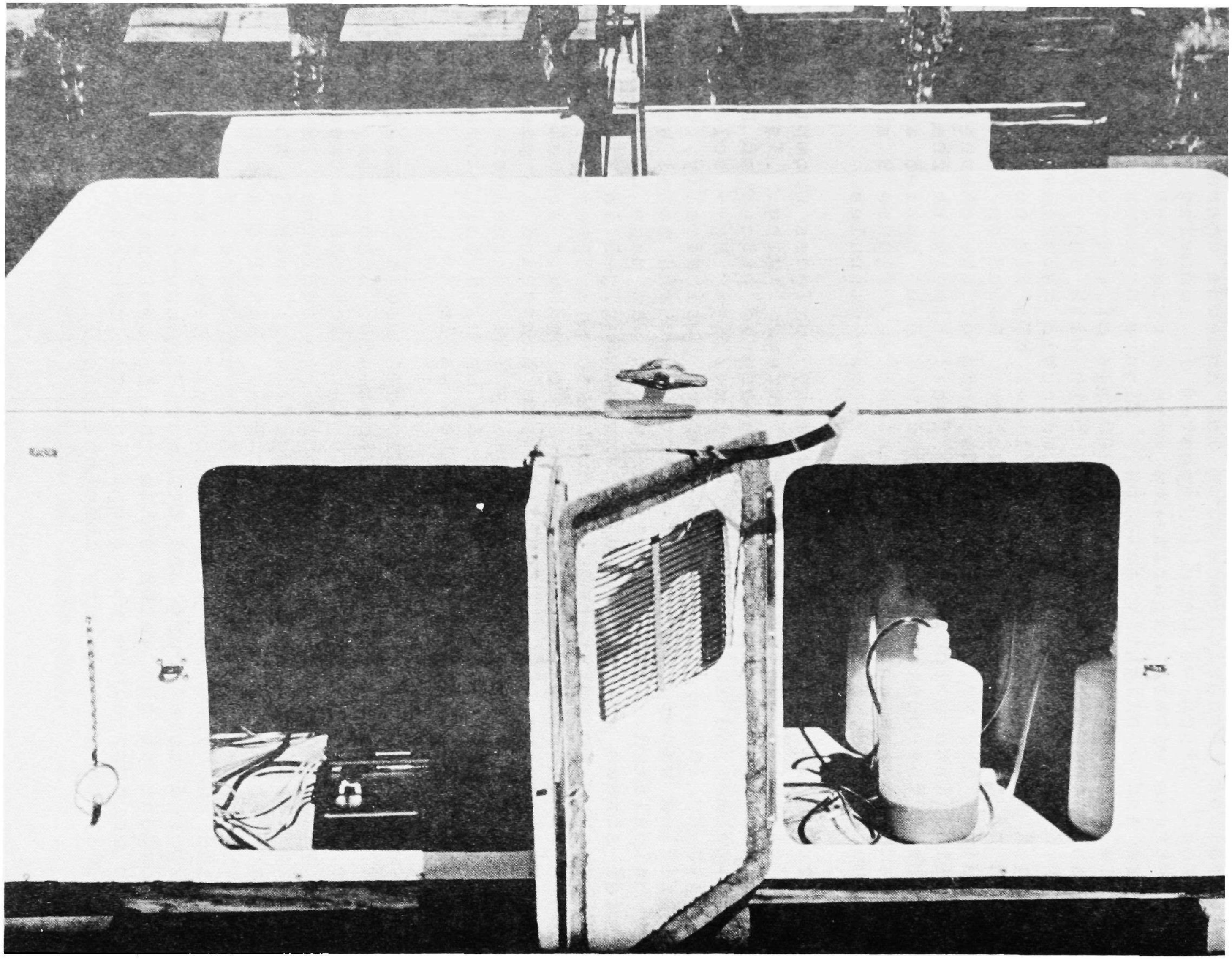

Figure 7. Mercury input feed system with four channel peristaltic pump.

Granulated vulcanized rubber is available as a waste product resulting from commercial tire recapping operations. We tested rubber samples obtained locally for mercury uptake and, while results were somewhat influenced by the source of the material and the amount of pre-soaking in water, found very good removal of mercuric ions from low aqueous concentrations in all cases. Since we were interested in removing mercury from our stream water, experiments were carried out at room temperature and a slightly acid pH. Figure 8 shows a typical result. In this particular case, 200 mls of pre-soaked (6 weeks) granulated rubber was added, both with and without continuous mixing, to 800 mls of a mercuric chloride solution containing 1 mg Hg++/liter. 10 mg/liter NTA did not affect the uptake of the mercury by the rubber. Based on these laboratory studies, the tail pools at the ends of the channels were modified into mercury recovery facilities with the understanding that the contaminated rubber would be buried as radioactive waste at the end of the experiments. Figure 9 shows two systems already filled with the rubber granules.

The ability of mercury to adsorb to many surfaces prompted a study to determine uptake by the PVC stream lining. Isotope tracer techniques were used. Small measured pieces of PVC film were submersed in one liter of 1 mg/liter Hg++ solution spiked with Hg 203. Pieces were removed periodically, rinsed with distilled water, counted for activity, and total attached mercury calculated. The mercury solution was made up using artificial stream water. Results are summarized in Figure 10. The PVC appears to saturate at about 3250 micrograms of mercury per square meter of exposed surface. To measure actual uptake in the streams, 80 PVC strips were suspended in each of the channels to be removed periodically for Hg analysis (Figure 11). These same strips are to be used for biomass data to estimate growth on the channel walls.

At a time when it still seemed likely NTA would be immediately introduced on a large scale into detergents, laboratory studies were begun to consider some possible interactions between NTA and mercury. Mosquitofish were exposed to sublethal levels of mercuric ion (0.05, 0.10, 0.20 mgs/liter) both in the presence and absence of 10 mg/liter NTA. Results are presented in Figure 12. After 18 hours of exposure, fish were digested and analyzed for total mercury. Body concentrations of fish exposed to mercury and NTA were independent of the mercury content of the water while the concentrations of those exposed to mercury alone was significantly correlated ($r = 0.72$, $p < 0.01$) to the mercury concentration.

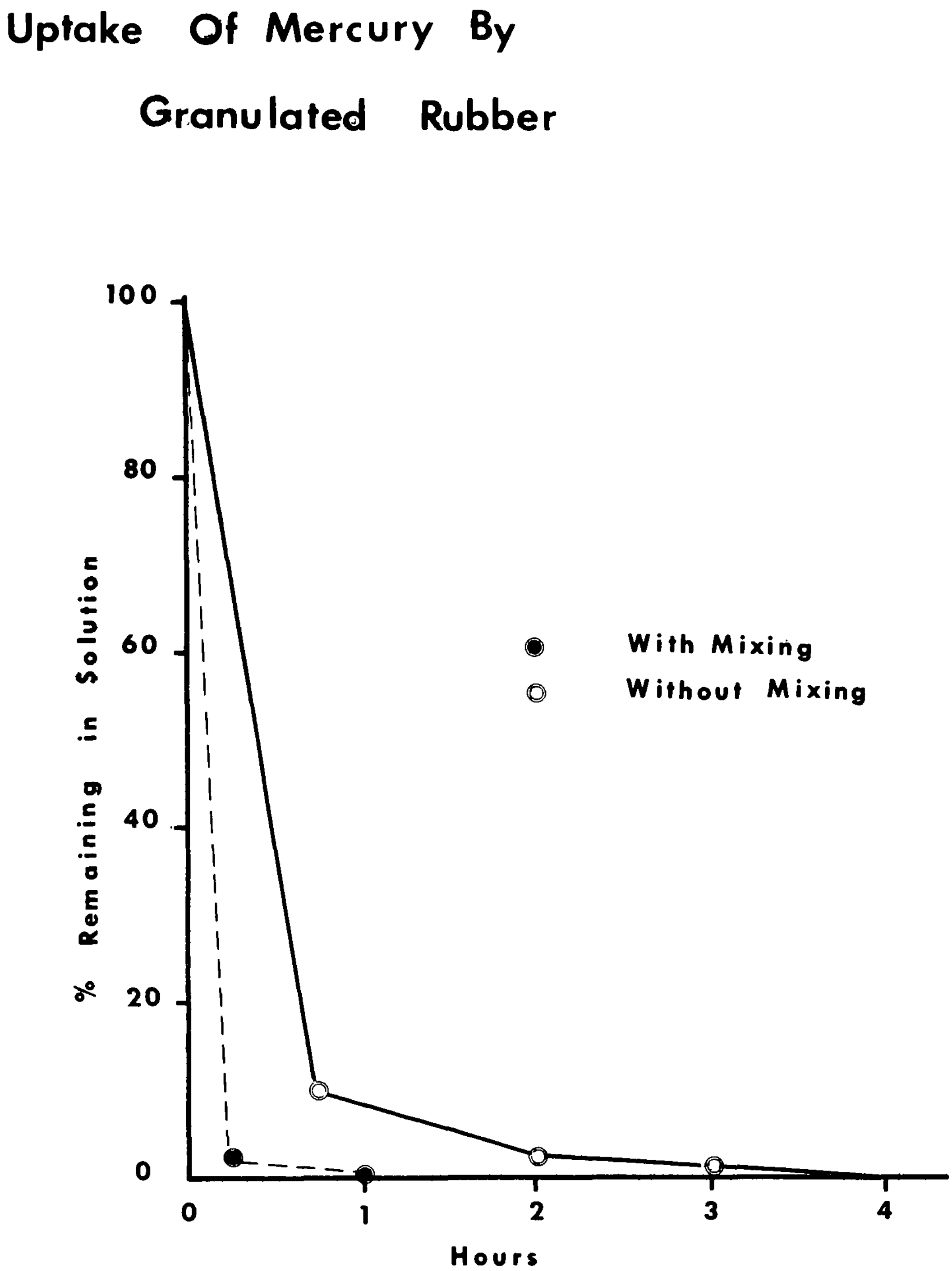

Figure 8. Results of rubber uptake.

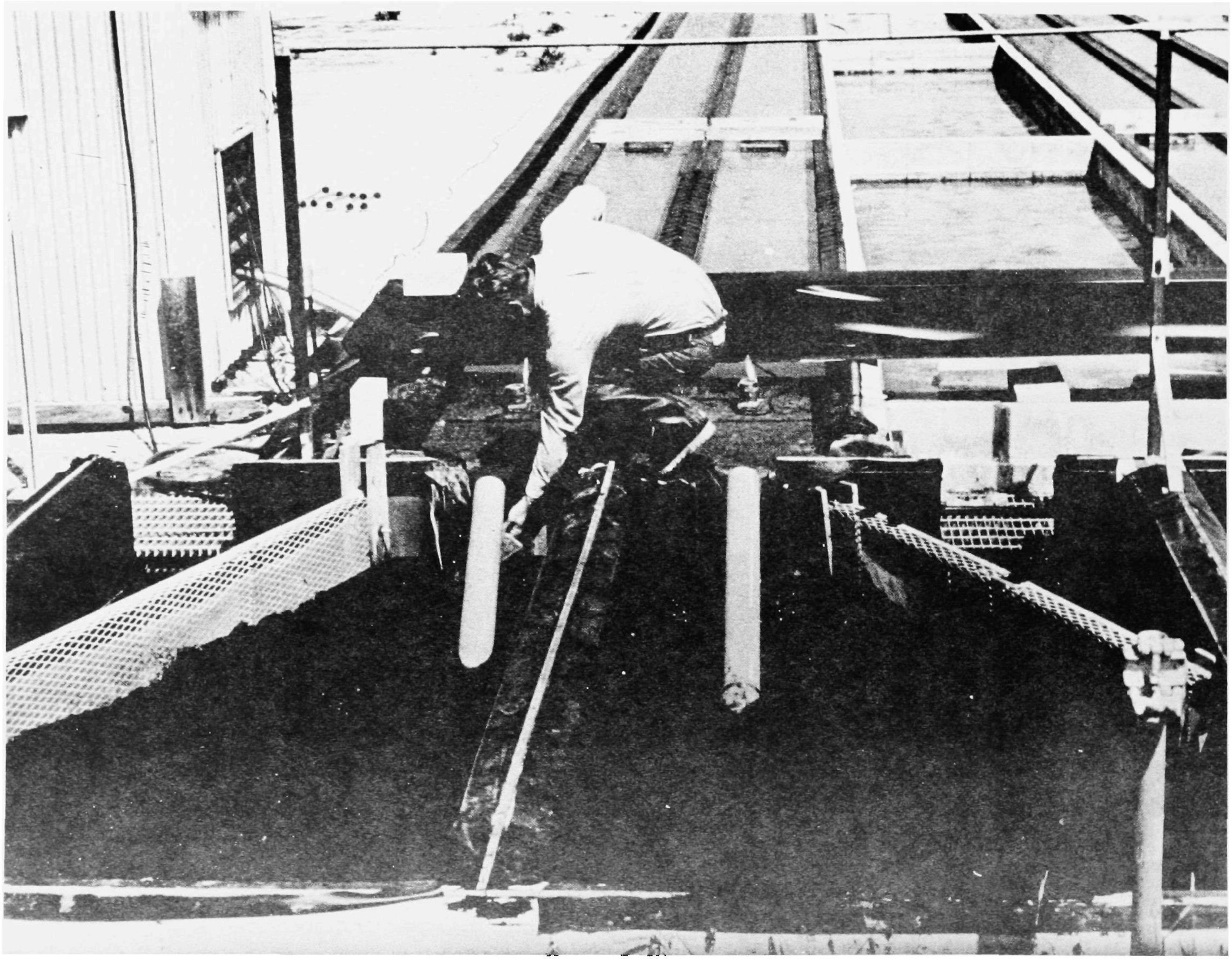

Figure 9. Rubber beds in tail pools for removal of mercury from artificial stream water.

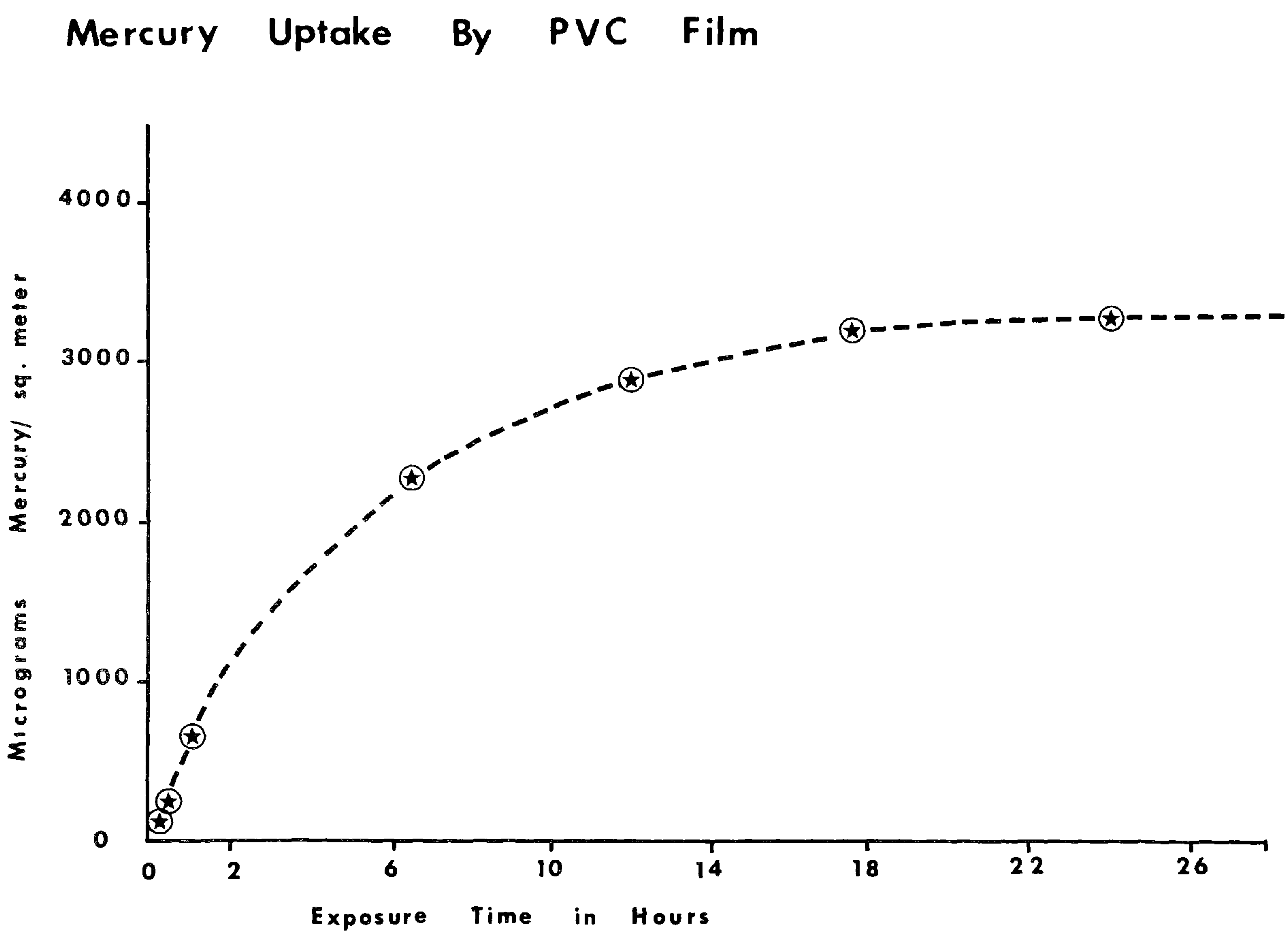

Figure 10. The uptake of mercury from a mercuric chloride solution by a polyvinyl chloride film.

Figure 11. Plastic strips suspended in streams.

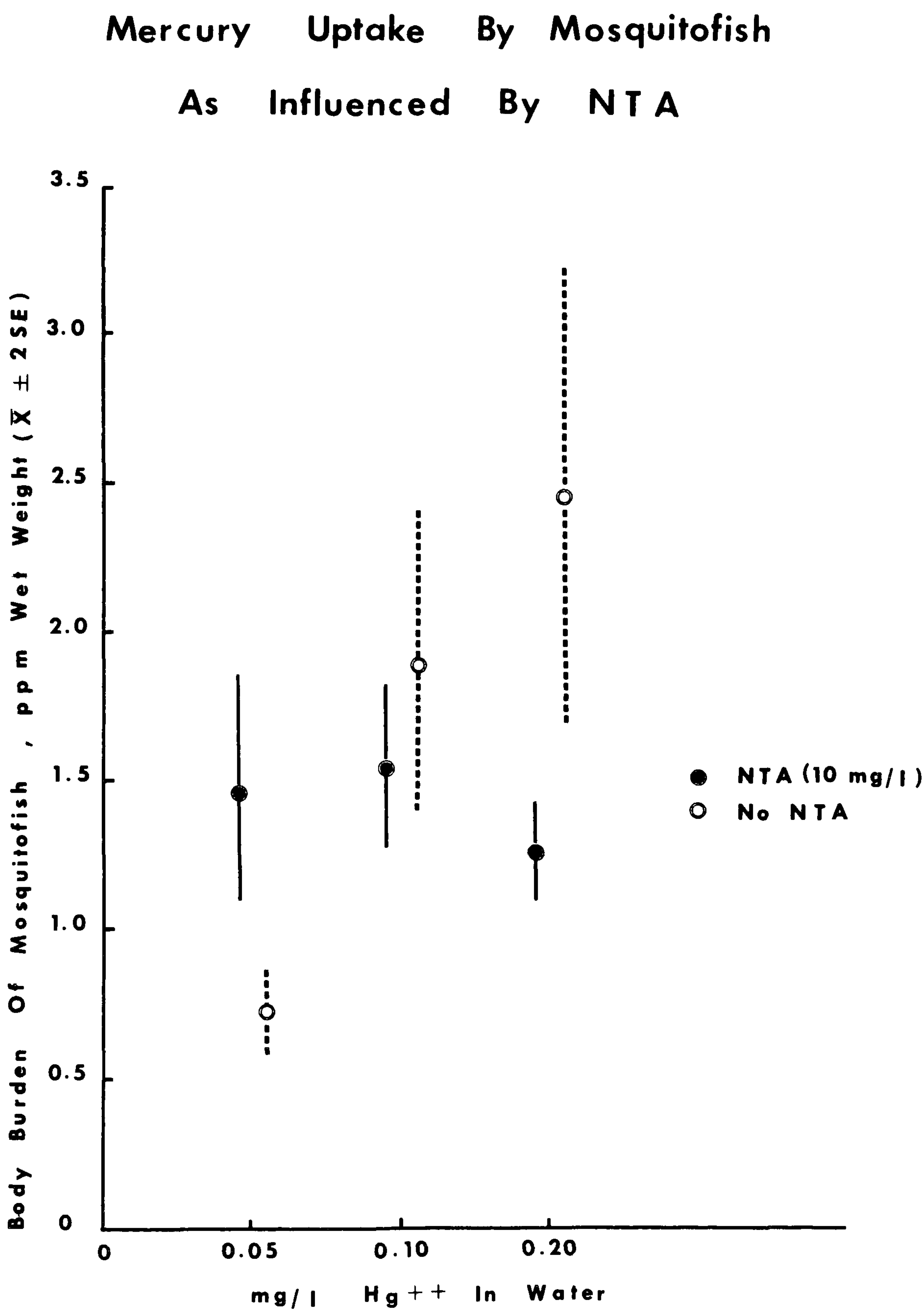

Figure 12. Influence of NTA on mercury uptake by mosquitofish, *Gambusia affinis*.

Further analysis of the data showed significant negative correlations between body weight and mercury concentration in the NTA +Hg treatments but not in the Hg treatments alone.

Several tests were made which showed the NTA in no way interferes with the mercury analyses as performed. Since it appears that NTA may yet become a major detergent constituent in this country, further work is needed on the interaction of this compound with mercury and other heavy metals.

SECTION VI

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16. *Abstract*

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