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VIRAL & MICROBIAL POLLUTION OF WATER: DETECTION AND CONTROL:

A Visit to the Environmental Health Laboratory, Hebrew University

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13. ABSTRACT

Projects of possible interest to the U.S. Army were discussed in written correspondence and during a visit to the Environmental Health Laboratory, Hebrew University, Jerusalem. These had to do with detection and assay of bacteria and viruses in water, with processing contaminated water for human use, and with study of the physiological effects of ions present in brackish and desalinated water. A new scheme for rapid determination and identification of viruses in water might prove to have sufficient originality to qualify for overseas Army support. An existing facility for routine detection and assay of viruses in water might be of value to U.S. Army or Navy elements in the Mediterranean area interested in purchasing such services.

Key words: (U) Water Pollution; Water, recovery from Waste; Water, detection of viral contamination; Water, sterilization; Water, brackish: physiological properties.

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TECHNICAL REPORT

VIRAL AND MICROBIAL POLLUTION OF WATER: DETECTION AND CONTROL:

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by

J.B. BATEMAN

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I. INTRODUCTION

Projects originating in the Environmental Health Laboratory were brought to our attention by the Director, Professor Hillel I. Shuval, about a year ago with the thought that they might be suitable for support by the U.S. Army. These projects became therefore the subject of enquiries directed to the Army Research Office and to the Army Medical Research and Development Command.

The first project was entitled "High rate stabilization for the treatment and renovation of waste water for base camps in small communities." Work on this project was being sponsored in the United States by the Ecology Development Corporation (1) and in view of this, combined with the fact that the basic research phases had been succeeded by pilot and field tests, no further consideration was given to the possibility of ERO support. At this point Professor Shuval (2) proposed for our consideration

> "the possibility of expanding and extending our present study on the basic high rate stabilization pond systems sponsored by EDC. We have named this method the Accelerated Photosynthetic System (APS) and are in the process of applying for patents on certain of its aspects. Our proposal is to include a study of the efficiency of such treatment systems in the total removal of pathogenic bacteria, viruses, and protozoa. This question may be particularly pertinent in the case of Army installations found in areas with high endemic rates of enteric diseases. The efficiency of the waste water treatment system in the removal of pathogens must be given major emphasis if waste water is to be reused for irrigation of recreational areas such as golf courses, or for the irrigation of vegetable crops."

Enquiry revealed, however, that this extension of the project is likewise at an advanced stage. An illustrated flier (3) is circulated by Accelerated Photosynthetic System (APS), Inc., a subsidiary of EDC. The flier contains the following description:

> "The Accelerated Photosynthetic System is a low cost wastewater treatment process in which a combination of algae and bacteria, mechanical aeration and chemical clarification provides a high quality tertiary effluent suitable for use in agriculture and industry.
>
> The APS process is particularly advantageous in regions of ample sunshine which will maintain high levels of photosynthetic algal activity. The photosynthetic activity of algae provides a source of dissolved oxygen while mechanical aeration is used for short periods during night-time hours or extremely cloudy hours during daytime.
>
> The process was developed in Israel where conditions required a low cost efficient wastewater treatment facility with the capability of producing a reclaimed wastewater for intensive agricultural use and industrial purposes. The APS process was further intensified and modified in Israel using the concepts of high rate algal processes in California."

and the statement that

> "the APS wastewater treatment and wastewater reclamation process is exclusively marketed and manufactured throughout the world by Accelerated Photosynthetic System (APS), Inc., ..."

A further extension would involve conversion of the tertiary effluent to a final product suitable for unrestricted domestic use, using reverse osmosis and adsorption by activated carbon, taking advantage of the special competence in the field of reverse osmosis available at the Weizmann Institute. It was made clear that any arrangement involving the U.S. Army would have to be made by joint sponsorship with the Ecology Development Corporation and it was concluded that provision for such an arrangement did not exist in the mission of the European Research Office. The initiative should come from home-based elements, possibly from the Corps of Engineers.

A second project was later outlined in the following terms by Professor Shuval (4):

"A number of years ago, Dr. Levine of the United States developed a rapid method for detecting coliform bacteria in water with the help of carbon 14 tagged lactose. He was never able to get his system to work properly, but in theory, the method is very attractive since it can give results in two to four hours.

A colleague of mine has done some innovative work on this and we think that we have overcome most of the problems that Levine encountered and are interested in producing a prototype automatic bacteriological continuance water testing device which will take hourly samples and give results within about a two-hour period. Such a device might be of considerable interest in civil defense activities. Of course, it would have numerous applications in direct civilian work.

I cannot reveal the details of this development since we are applying for a patent, but if your office is at all interested in the possibility of supporting the development of such an instrument, we would be happy to hear from you."

to which a staff member of the U.S. Army Medical Research and Development Command replied that

"for Army field use...the aspects of simplicity and size cannot be minimized,"

adding that

"We would be interested in learning more about the device if it can meet, or has the potential for meeting the following requirements:

a. Provide valid results in one to two hours.

b. Not weigh more than the present Millipore apparatus.

c. Not require the handling or storage of hazardous materials.

d. Have a simple procedure, easy to read results, and not require expensive, sophisticated equipment."

The ensuing correspondence with Professor Shuval made it clear that these projects constituted only a fraction of a departmental program of research on water quality management and water pollution control. The visit about to be reported was prompted by Professor Shuval's suggestion that items of interest to the U.S. Army could perhaps be found within this program or could emerge as feasible modifications or extensions.

It may be well to mention here that in July 1970 (5) we reported a talk with Dr. Y. Goldshmid of the Mekaroth Water Company, Tel Aviv, about the need for investigation of the physiological effects of ions present in desalinated and brackish water when these are mixed in various proportions, as they are in Eilat on the Red Sea. Israeli physiologists were to be approached by Dr. Goldshmid and a plan for further study worked out. No news of progress along these lines was received until Shuval wrote (6) expressing interest in the project:

> "Dr. Y. Goldsmith (sic) of the Mekorot Water Company, who drew up the proposal, has now left that organization. At the time, Dr. Goldsmith consulted with our group in the drafting of the proposal and we have continued our interest in this very important subject.
>
> Our group at the Hebrew University, as well as others concerned with water quality research in Israel, are particularly interested in this problem since we may find that desalinated water will become, in time, a major source of drinking water in the country. Very little work has been done in evaluating the physiological effects of supplying desalinated water.
>
> One particular aspect of this problem is the variation in total dissolved solids in the water supply of a community which is desalinating brackish water. For example in Eilat, the desalination plant works at a constant rate and supplies almost completely desalinated water in the winter, but in summer months with increased water supply demands, the desalinated water is mixed with brackish water and the community receives water with a high dissolved solids content. The physiological impact of such seasonal changes has not, to the best of our knowledge, been studied. A committee of the

Israel National Council for Research and Development, of which I am a member, has been appointed to draw up the research needs in this area. If the United States Army is interested in supporting work on this problem, I would be happy to arrange that specific proposals be submitted to you.

Another similar problem is the physiological and toxicological effect of using reclaimed waste water for drinking water. We are interested in carrying out a chronic feeding study with experimental animals, using highly purified waste water meeting the most rigorous chemical and bacteriological health standards. We feel that such a study would be a desirable first step in obtaining approval from health authorities for the use of reclaimed waste water for community water supplies.

Since the United States Army may at times be in a position of re-using waste water for water supply purposes even before this becomes necessary in most American cities, it might be an appropriate area of study for your group."

In the belief that physiological studies of this kind, important as they are, could be carried out in the United States, and remembering also that the specific problems likely to be encountered in the United States would be somewhat different from those at Eilat, the offer to submit formal proposals was not accepted.

II. THE ENVIRONMENTAL HEALTH LABORATORY

The Environmental Health Laboratory, founded in 1966, is housed in the old University buildings in the Russian Cathedral complex, near the Old City. The academic staff, of whom four are permanent and 15 are temporary employees, includes specialists in algology (aquatic biology), toxicology (biochemistry), virology and bacteriology (environmental microbiology), environmental chemistry, and environmental engineering. The staff have teaching responsibilities toward the Faculties of Medicine and Science, a total of 20 courses being offered. Their research activities have been summarized (7):

"The environmental health laboratory serves as an interdisciplinary base for teaching and research programs in the evaluation and control of hostile biological, chemical, and physical factors in the environment of man including water, air, land and food pollution.

Current research activities in the field of water quality management and water pollution control include studies on the detection and control of water-borne pathogenic bacteria and viruses; toxicology of nitrites and nitrates in water, air and food; marine pollution; biological and physical-chemical processes of waste water purification and renovation; self-purification processes and ecology of streams; anti-viral activity of sea water; health effects associated with the reuse of waste-water.

The Laboratory has been designated by the World Health Organization as a collaborating Institution of the International Reference Centers on Community Water Supply and Waste Treatment."

III. PROJECTS DISCUSSED WITH PROFESSOR SHUVAL AND DR. KATZENELSON

1. A Rapid Detector for Coliform Bacteria

This device, involving uptake of C14-labelled lactose, is capable of indicating the presence in water of two cells per 100 ml and of detecting contamination by sewage. In Shuval's view it is not likely to prove capable of modification for field use; this is his answer to the specifications laid down by the USAMRDC. Its principal value, if it proves amenable to complete automation, would be as a substitute for the conventional manual methods now essential for the continuous monitoring of municipal water supplies.

2. Routine Assay of Viruses in Water

Negative results of bacteriological tests for contamination do not necessarily prove absence of contamination. The enteroviruses are more stable and in Shuval's view assay of viral contamination will become of increasing importance. Several studies, done with support from the

USDHEW and the U.S. Environmental Protection Agency, as well as from Israeli domestic sources, illustrate the point. Sewage from one town, containing on the average 10^4 virus particles per liter, is discharged into the Jordan River where, although highly diluted, the viruses can still be detected 25 km downstream. In sea water, despite the presence of antiviral substances associated with specific marine bacteria, the 90% decay time of radioactive enteroviruses may be as long as 48 hours compared to 30 - 120 minutes for coliform bacteria and times somewhat greater than this for Salmonella spp. The increasingly prevalent hepatitis danger, both from polluted drinking water and possibly also from swimming in polluted water, likewise points to the need for reliable virological assay methods.

Shuval, Katzenelson and other members of the Hebrew University groups have studied several methods for concentrating and detecting small concentrations of enteroviruses in large volumes of water. A phase separation method will detect as few as 1 - 2 infectious units in a liter of water and another method, unspecified, under development increases this sensitivity one hundred-fold.

Accordingly Shuval feels able to offer the routine services of his laboratory on roughly the following terms: ten-liter water samples can be concentrated by a factor of about 10^3 overnight and within 10 days a figure for the number of enterovirus plaque-forming units can be obtained. About 200 such samples of fresh water or sea water could be assayed per year at a cost of roughly $\$10^4$, or $50 per sample. If virus typing is requested, the tests would take about a month for resolution into the polio-ECHO-rheo groupings and the cost would of course be greater.

3. Development of a New Method for Rapid Determination and Identification of Viruses in Water

Shuval outlined an ingenious four-stage procedure which has undergone preliminary tests. The results suggest that it is not unreasonable to aim for a 24-hour test. Shuval asked that the details be considered confidential, although in my view--and I said so--it is not unlikely that the individual steps have been tried in some form by other investigators. One, indeed, was attributed by Dr. Shuval to Somerville of the University of Glasgow*.

*Probably R.G. Somerville, Royal Infirmary, Glasgow, specializing on viral immunofluorescence; development of automated system for evaluation and quantitation. (8)

4. The Inactivation of Viruses with Ozone

In work supported by the U.S. Environmental Protection Agency it has been shown that ozone is more effective than chlorine in killing enteroviruses, and a study of the kinetics is providing the information needed for development of more effective ways of controlling viral pollutants. An ozonator for field use might be useful.

5. Detection and Inactivation of the Cholera Vibrio

Shuval says that current ideas about the contamination of water with cholera are based upon information obtained in India 30 or 40 years ago, much of it in need of re-evaluation or re-investigation. This would require that ways be devised of identifying the organism rapidly in water, of sampling and testing municipal sewage, and of suitably treating it. The serious outbreak of cholera in Israel in 1969, the first for many years, was traced to the illegal watering of vegetables with sewage. The organism is, apparently, difficult to remove except by scrubbing, which is not feasible with leafy vegetables such as lettuce. It was mentioned, incidentally, that a vibriophage was also found on leaves; whether the idea of biological control of the vibrio was being entertained, I do not know. The work actually accomplished by Shuval's group was done during the 1969 outbreak. Concentration of the organisms was done with gauze pads for the purpose of detection, while chlorine proved to be effective for disinfection.

IV. RECOMMENDATIONS

All of the five topics just discussed have some military relevance but it is not clear that the approaches used are of sufficient originality to justify support, nor are the local epidemiological circumstances sufficiently unusual. An exception may be made in the case of the projected 24-hour method for rapid determination and identification of viruses in water, the details of which are not divulged in this report. If sufficient interest exists, Prof. Shuval will be invited to submit a proposal for limited circulation on a confidential basis. We have been cautioned, however, that our ability to support overseas research depends upon the presence of some unique attribute that is not available within the United States.

The remaining projects might well be brought to the attention of the Corps of Engineers to whom a copy of this report will be sent.

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