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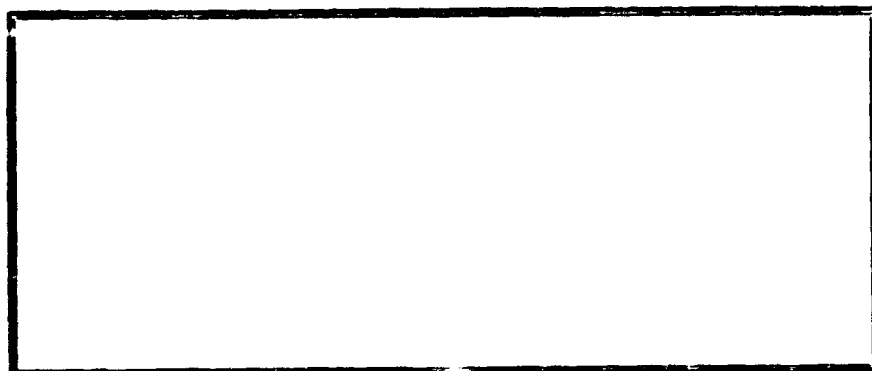
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U. S. NAVAL MEDICAL RESEARCH LABORATORY
U. S. Naval Submarine Base
New London, Connecticut

MEMORANDUM REPORT NO. 58-2
LIMITED FIELD EVALUATION OF
THE MILLIPORE FIELD MONITOR KIT
ABOARD SUBMARINES

NM 24 01 20.04.03

19 March 1958

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SUMMARY PAGE

THE PROBLEM

To determine whether or not the Millipore Field Monitor Kit, a kit designed for rapid bacteriological analyses of water, can be used successfully aboard submarines.

FINDINGS

The kit was found to have some disadvantages, the chief one of which was that corpsmen must receive sufficient indoctrination in its use. It is felt that the disadvantages noted are far outweighed by the advantages of the kit.

APPLICATIONS

The Millipore Field Monitor Kit can be used in all surface vessels of the U. S. Navy as well as submarines.

ADMINISTRATIVE INFORMATION

This investigation was undertaken as a part of Bureau of Medicine and Surgery Research Project NM 24 00 00 - Physiology of the Undersea Environment Including Habitability of and Escape from Submarines. The present report is Report No. 3 on Subtask 4 (Field Evaluation of Products and Equipment Affecting Submarine Habitability), of Task 24 01 20. It was approved for publication on 19 March 1958.

Published by the U. S. Naval Medical Research Laboratory
For Official Use

ABSTRACT

A limited evaluation of the use of the Millipore Field Monitor Kit was made aboard four operating submarines. It was found that the chief drawback to the kit was that the hospital corpsmen must be thoroughly indoctrinated in its use. Self indoctrination is not possible. A few minor technical difficulties were encountered in the kits but have since been corrected by the manufacturer. The advantages are as follows: Kit is compact and portable, simple to operate, tests can be completely performed aboard submarines, results can be obtained within 24 hours, the test requires no expensive equipment and the test is relatively inexpensive to perform.

INTRODUCTION

It is a well founded public health fact that the potability of water cannot be assured without regular bacteriological examinations of the water. To conduct these examinations, in the past, has required laboratory facilities and trained personnel and for this reason such tests could not be done aboard a submarine. The hospital corpsmen had to rely on having the tests conducted ashore at the nearest medical facility capable of doing them. Unfortunately this was not so simple. Most medical department laboratories are concerned primarily with clinical tests and because of their work loads they frequently cannot always do water bacteriology. This, combined with the operational schedules of the submarines, usually results in the tests not being done. Water sanitation is therefore compromised.

A test kit for bacteriological analyses of water has been developed by the Millipore Filter Corporation, Watertown, Massachusetts, - one which is simple to conduct and one which does not require cumbersome laboratory materials or techniques which involve extensive laboratory training. The test kit, known as the Millipore Field Monitor Kit, appears to be ideally suited for use by submarine hospital corpsmen. It is inexpensive, compact, and disposable, and its use would make the submarine self-sufficient in regard to determining drinking water potability. The test makes use of a relatively new bacteriological tool - the membrane filter which is a thin, porous sheet containing microscopic holes smaller than bacteria.

The testing of water by the membrane filter technique has been approved by the U. S. Public Health Service as an alternate method to the standard methods of examining water.

U. S. Navy Preventive Medicine Unit No. 2, Norfolk, Virginia, conducted limited studies ⁽¹⁾ of this kit and have determined it to be fast, reliable and simple to operate. The United States Air Force has adopted the membrane filter technique for bacteriological analyses of water in the field. ⁽²⁾ No field evaluation by Naval Operating Forces had been done, however, and it was decided therefore that a limited trial aboard operating submarines would be of value.

The Millipore Field Monitor Kit was brought to the attention of the Officer-in-Charge, Naval Medical Research Laboratory, who assigned evaluation of this device to LT Louis R. Kaufman, MSC, USN, who serves as the Laboratory's Public Health Officer and Director of its School of Submarine Medicine Technique.

Description of Kit and its Operation

The Field Monitor Kit (figure 1) consists of a disposable plastic cell into which is fixed a membrane filter and filter pad, all sterilized during manufacture. There are two openings: one on the top of the cell and the other on the bottom which are covered with removable rubber caps to maintain sterility. Disposable, pre-sterilized plastic tubes fit into each opening and by fitting the proper one to a metal syringe water is drawn through the filter. Bacteria are collected on the surface of the filter. A selective growth media is supplied in ampules which is instilled into the filter pad after the sample water has been drawn through (figure 2). The Monitor cell is incubated for 12-24 hours and bacterial colonies showing the metallic sheen of coliform organisms are then counted (figure 3). No special magnifying device is necessary to see the colonies but sometimes a small hand magnifier is helpful.

Procedure

Four submarines operating from New London were selected for participation in the evaluation and each was supplied with a metal syringe, sufficient Monitors, sampling tubes and media ampules to conduct 4 tests per week for 6 weeks. The hospital corpsmen on each of the submarines were given a 2 hour familiarization lecture with demonstrations and practical use of the Kit, a complete set of written instruction on how to conduct the tests and questionnaires to fill out at the time of each test.

Each hospital corpsman was instructed to collect duplicate samples twice a week from each of two tanks. It was suggested that the tank selected be rotated so that all four tanks would be tested weekly. One of the duplicate samples was collected by the

usual sampling method ⁽³⁾ and sent to the U. S. Navy Medical Research Laboratory within 4 hours for testing (10 ml. portions of samples in 5 lactose broth tubes) while its duplicate was tested with the Monitor Test Kit on board using 100 ml samples. The Monitor cells were incubated in various warm areas on the submarine having a temperature near 35°C for a period of 12-24 hours. Upon completion of the incubation, the bacterial colonies showing coliform sheen were counted and the number recorded on the questionnaire. Ten per cent glycerine was then added to the filter as a preservative, the filter removed from the cell and then attached to the questionnaire for reference.

Results

A total of 82 water samples were tested, 62 of which were 100 ml. duplicates. The remaining 20 samples were not duplicates.

Comparing the 5-tube fermentation test results it was found that 55% of the samples were in agreement (18 positives agree, 16 negatives agree) and 45% in disagreement (20 positives disagree, 3 negatives disagree). The agreement percentage is somewhat lower than was expected. According to Thomas ⁽⁴⁾ the coliform densities determined by membrane filter techniques average about 70% of the confirmed fermentation tube results. This variance is believed to be the result of inexperience on the part of the submarine hospital corpsmen in reading results. Only 1 of the 4 corpsmen had had any previous laboratory training and this did not include sanitary bacteriology. Since few hospital corpsmen entering submarine service have experience in sanitary bacteriology it will be necessary for these men to have a thorough indoctrination in the use of the Monitor Test Kit before the results of their using this method of water testing can be fully evaluated. It is understood that the Millipore Filter Corporation has prepared an instruction booklet, a film and a slide series for instructing personnel in the use of the Field Monitor Kit. This instruction could be given to new submarine hospital corpsmen in the School of Submarine Medicine Technique and to graduates already aboard submarines by establishing short courses in the home ports.

Two hospital corpsmen reported that they had difficulty detecting coliform sheen under fluorescent light. While this is somewhat of a problem it is believed that familiarity with use of the kit will correct this situation.

Difficulty was experienced in drawing the water through the filters. At times, the filtration would stop and no amount of effort could re-establish flow. The filters had to be discarded. According to the Millipore Corporation this was due to an air block forming from an incomplete filling of the Monitor Cell with water. The inlet hole on the cell has now been redesigned and the syringe valve modified so that the air block problem is essentially avoided. Some skill, however, will still be required on the part of the operator.

A problem was encountered with Monitor Cells popping open aboard some boats when snorkeling. This was due to unequal pressures between the outside and the inside of the cell and somewhat to lack of uniformity in fit between the two halves of the cells. According to the manufacturer the molds for making the cells have been changed and better fitting cells are being produced. Regardless, popping open can be avoided by leaving the cap off the lower opening during incubation for equalization of pressure.

Finding of an area aboard the submarines that would have a fairly constant temperature in the range of 35°C was a problem. Temperature fluctuations of 10°C was not uncommon and constituted a threat to reliability of results. Some of the areas tried were: galley overhead, bottom shelf of medical locker, radar transmitter and receiver stack, I.C. fuse box and over-head in crew's mess. A small compact 110 volt AC incubator about the size of a cigar box (to hold about 12 Monitor cells) is needed to assure uniformity of temperature during incubation.

Removing the filters intact for preservation and record purposes was very difficult due to the tendency of the filter to stick to the Monitor Cell. This problem has been corrected, according to the manufacturer, by molding the Monitors out of polystyrene instead of tenite.

Conclusions

Despite the disadvantages noted during the limited field test of the Field Monitor Kit, it is believed that its advantages are, by far, more numerous and more significant than the disadvantages. It is felt that the Kit should be included as part of the allowance of the submarine and that hospital corpsmen and Squadron Medical Officers should be indoctrinated in its use.

REFERENCES

1. U. S. Navy Preventive Medicine Unit No. 2. Evaluation of Millipore Field Monitor Kit. Official Report, October 1956.
2. Jones, Thomas C., Use of the Membrane Filter for Potable Water in the Air Force. U. S. Armed Forces Medical Journal 8:1495 - 1506, October 1957
3. Manual of Naval Preventive Medicine, NavMed P1510. Paragraph A-14
4. Thomas, Harold A., Jr., Woodward, Richard L. and Kabler, Paul W., Water Potability Control with Membrane Filters. Paper was presented at 49th Meeting National Research Council, Subcommittee on Waste Disposal and Subcommittee on Water Supply of the Committee on Sanitary Engineering and Environment, October 1955.



Figure 1. - Field Monitor Kit Assembled Showing Disposable
Monitor Cell and Plastic Tube



Figure 2. - Instilling Media into Monitor Cell from Ampule.



Figure 3. - Monitor Cell with Membrane Filter Enlarged Twice Natural Size. Bacteria colonies can be seen on surface of filter.