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COASTAL WATER QUALITY CONTROL IN THE MEDITERRANEAN

Final report on the Joint WHO/UNEP Coordinated Pilot Project
(MED VII) (1976-1980)



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The first part of the document discusses the importance of maintaining accurate records of all transactions. It emphasizes that every entry, no matter how small, should be recorded to ensure the integrity of the financial statements. This includes not only sales and purchases but also expenses, income, and any other financial activity. The document also highlights the need for regular reconciliation of accounts to identify any discrepancies early on.

Next, it covers the various methods used for recording transactions, such as the double-entry system. This system ensures that every transaction is recorded in two accounts, one as a debit and one as a credit, which helps in maintaining the balance of the books. The document provides a detailed explanation of how these entries are made and how they affect the different components of the accounting equation.

The document then moves on to discuss the classification of transactions. It explains how transactions are categorized into different types, such as sales, purchases, and expenses, and how these are recorded in the appropriate accounts. It also discusses the importance of using the correct accounting principles and standards to ensure that the records are accurate and reliable.

Finally, the document concludes by emphasizing the role of the accountant in maintaining these records. It states that the accountant is responsible for ensuring that all transactions are recorded correctly and that the financial statements are prepared in accordance with the relevant accounting standards. This is a crucial part of the business's financial management and is essential for making informed decisions.

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1. INTRODUCTION

A series of conferences was convened in 1974 and 1975 to discuss the state of pollution in the Mediterranean and ways and means of providing the pollution abatement measures required. Some of the proposed programmes were adopted by the UNEP Intergovernmental Meeting on the Protection of the Mediterranean (Barcelona, January 1975) and endorsed by UNEP.

The UNEP Coordinated Mediterranean Pollution Monitoring and Research Programme resulted from the decisions made at the above meeting. It consists basically of seven pilot projects which are generally concerned with baseline studies and monitoring of various pollutants and deals mainly with coastal waters. Research into the effects of pollutants on marine organisms and on marine communities and ecosystems, problems of the coastal transport of pollutants and the sanitary quality of beaches and coastal waters also forms part of these projects.

One of these seven projects is the joint WHO/UNEP Coordinated Pilot Project on the Coastal Water Quality Control in the Mediterranean (MED VII). Its main objective is the assessment of the potential health hazards connected with the coastal waters of the Mediterranean, needed for the rational design and efficient implementation of national programmes for the control of coastal pollution from land-based sources in the area.

In 1975 an operational document was developed at the WHO/UNEP Expert Consultation on Coastal Water Quality Control Programme in the Mediterranean (Geneva, 1975), ((1) reference list), it constituted the programme basis of project MED VII and initiated its operational phase. The project was implemented in close collaboration with UNEP and was scheduled to end in March 1981.

The main elements of the programme of work are as follows:

- a. the monitoring of selected coastal areas;
- b. the initiation and promotion of scientific studies on the epidemiological evidence of health effects caused by pollution in coastal areas;
- c. principles and guidelines for coastal water pollution management;
- d. the development and propagation of relevant technical documentation;
- e. training and technical assistance.

The present document is intended to analyse, evaluate and discuss the practical and theoretical scientific results of the work carried out and implemented since the actual initiation of the project MED VII in July 1976 up to December 1980. Special attention is given to the monitoring programme which constitutes the larger and main scientific activity of the project.

2. THE MONITORING OF SELECTED COASTAL AREAS

2.1 National laboratories and areas monitored

The monitoring was implemented through a network of collaborating national laboratories nominated by their governments, and the selection and designation of coastal areas to be monitored was undertaken by the responsible national authorities. Figure I shows the geographical distribution of the 30 collaborating institutions nominated by their governments and approximately their respective monitoring areas.

The monitoring included two aspects:

- a. the surveillance of beaches and bathing waters;
- b. the surveillance of shellfish culture waters and shellfish flesh.

All the above 30 collaborating laboratories nominated by the Mediterranean coastal states signed a contract with WHO which set out the details of their work and studies within project MED VII.

The aspects of work each agreed to cover are as follows:

- three collaborating laboratories are monitoring shellfish only;
- eleven collaborating laboratories are monitoring recreational waters as well as shellfish;
- sixteen collaborating laboratories are only monitoring recreational waters.

Figure I indicates the collaborating laboratories covering one or the other of the above aspects as well as the collaborating laboratories covering both aspects.

The collaborating laboratories represent 14 Mediterranean countries out of a total of 18 (figure II). Thus the coastal areas of four countries were not covered by the monitoring programme. Moreover, the distribution of the collaborating institutions did not represent a well balanced representation of all the coastal areas.

2.2 Methods and material used

2.2.1. Monitoring of beaches and recreational waters

Scope

A standard pattern of surveillance representing a minimum programme was applied as a compulsory approach by all collaborating laboratories. This minimum programme could be extended to a larger one at the discretion, and according to the need and interest, of the individual collaborating laboratories.

The minimum programme aimed at promoting similarity and comparability between the results arrived at by the collaborating laboratories in the various study areas.

The surveillance covered beaches, recreational waters and sediments using physical, chemical, biological and microbiological parameters.

Parameters

Parameters in the minimum programme measured on individual samples were as follows:

- for beaches: the parameter of visual appearance following a uniform visual classification scheme (modified version of the Garber classification system) was applied;
- for recreational waters: microbiological tests were limited to the most common indicators of faecal pollution, i.e. total coliforms, faecal coliforms and faecal streptococci;
- for sediments: microbiological tests were the same as those for recreational waters;
- for effluents and rivers: major effluents and rivers discharging into the study area were expected to be examined at least for BOD₅, settleable solids and the volume of discharge. Sampling was supposed to be carried out on the same days as for coastal waters.

Parameters in the minimum programme describing general conditions in the monitoring area at the time of sampling were as follows:

- hydrographic conditions (sea temperature, salinity, oxygen, turbidity);
- dynamic conditions (state of tides, state of waves, current measurements, drifts);
- visual observation of pollution traces (colour, discoloration, unusual turbidities, odour, slicks, foams, floatable etc.).

Monitoring procedures

Common principles and procedures were developed for sampling and analysis in the monitoring areas in order to further promote harmonization and comparability.

Location of sampling points

Sampling points and reference points were defined for the water body following an agreed pattern. They were identified and kept throughout the project period.

Frequency of sampling was based on various factors of prime importance.

Analytical methods

Recognizing the need to use common methodology and analytical techniques which would promote comparability of results, guidelines and reference methods were planned and developed, and used during the project period. Already within the operational document some provision was made in that direction. For example, it was specified that for the determination of E. coli in water, only the membrane filtration method would be applied during the implementation of the project.

2.2.2. Monitoring of shellfish

Scope

Here also a standard pattern of surveillance representing a minimum compulsory programme was applied by all collaborating laboratories involved in the monitoring of shellfish.

The surveillance covered shellfish culture sea-waters, shellfish flesh and sediments from shellfish beds.

Parameters

They were the same for culture waters, shellfish flesh and sediments from shellfish beds.

The following parameters constituted the minimum compulsory programme: total coliforms, faecal coliforms, faecal streptococci and total heterotrophic bacteria.

Monitoring procedures

Here again the use of harmonized procedures in order to promote comparability of results was aimed at. All collaborating laboratories involved in shellfish monitoring were expected basically to use the Guide to shellfish hygiene by P.C. Wood, WHO, Geneva 1976, ((2) reference list).

Analytical methods

Similarly, the methods of analysis in the above guide were provided as examples. However, more specifically, a compulsory methodology was developed and followed.

Sampling frequency was dependent on the time of the year, the legal season for consumption and the period of maximum demand. Here again a harmonized approach was followed.

2.3 Results and their interpretation

2.3.1 Parameters investigated

a. Recreational waters

The great majority of collaborating laboratories used the three bacteriological indicators specified: i.e. total coliforms, faecal coliforms and faecal streptococci included in the minimum compulsory programme.

Similarly, but to a lesser extent, the collaborating laboratories monitored, in addition to the bacteriological data, air, sea and sample temperatures during transport, and salinity.

However, only a relatively small number of laboratories monitored some of the parameters describing general conditions in the monitoring area at the time of sampling. These parameters concerned meteorological, hydrographic and dynamic conditions and visual observation of pollution traces. The main reason for this deficiency was the lack of means, material and personnel required for carrying out elementary measurements in the absence of specialized meteorological services, or the non availability of such information.

Only two laboratories carried out visual appearance tests for beaches. A few collaborating laboratories monitored additional parameters with a view to collecting more specific information or to developing new parameters more sensitive and more representative of the pollution by faecal material. In these laboratories pathogens like Salmonella, Shigella, Vibrio cholerae and enteric viruses, other micro-organisms such as bacteriophages, bacteria that grow on nutrient agar, V. parahaemolyticus, dinoflagellates, and total bacteria count, are being investigated (Table 1).

From the results so far obtained it appears that higher counts have been experienced for total coliforms than for faecal coliforms and enterococci. But the enterococci count was also reported to invariably give the lowest count values with respect to the other two compulsory parameters. It is questionable if the density of faecal streptococci in some coastal waters is not high enough to make a good parameter for assessing faecal contamination.

Although the concentration of total coliforms might not be the best indicator of contamination of sea-water with faecal material, some results obtained show a close correlation with the location of outfalls which discharge faecal material into the sea, or material which seems to enhance the survival and reproduction of micro-organisms in the sea.

Faecal coliforms, on the other hand, seem to be the most sensitive indicator of the degree of faecal contamination from warm-blooded animals in sewage pollution and dispersion around points of sewage discharge.

The ratio of faecal streptococci to faecal coliforms may indicate more clearly the nature of the contamination. This ratio can often be as high as 0 to 1. The recognized density differences between faecal coliforms and faecal streptococci in faecal material is a unique relationship and can be used in defining possible sources of pollution. Faecal coliform to faecal streptococci ratios in human faeces and domestic wastes may be expected to be greater than 4, whereas these same faecal coliforms to faecal streptococci ratios for the faeces of farm animals, cats, dogs and rodents are less than 0.7. Ratios less than 0.7 are also characteristic of drainage from farmland, cattle-feed-lot operations and storm water run-offs.

From the experience so far acquired it appears that the faecal coliform test may largely replace the traditional total coliform parameter for water quality measurements in recreational waters.

However, some results show that faecal streptococci are a good indicator of pollution. Their survival and frequency of discovery are similar to those of enteroviruses with $r = 0.9113$ and peaks of $r = 0.94$. It was also noted that when warm, industrial, alkaline wastes are present, large numbers of enterococci may be isolated while coliforms may be absent.

In one of the studies, the correlation analysis between the various bacteria and the enteroviruses was carried out. The results with the details of the regressing line are given in figures IV, V and VI. A significant correlation was found between the ratios of total coliforms to faecal coliforms, total coliforms to faecal streptococci and faecal coliforms to faecal streptococci.

However in another study no correlation was found between total coliforms or faecal coliforms and faecal streptococci (Table 2).

In general the enteric bacteria are reduced in the sea relatively more rapidly than enteroviruses. However faecal streptococci seem to display a die-away rate similar to that of enteroviruses (figure III). This may point to faecal streptococci as a useful indicator of the degree of viral contamination in the sea.

Considering that the selected indicators of pollution are relatively sensitive to the marine environment, it appears advisable to investigate and eventually to use a more suitable organism. To this end, some studies are being carried out on Salmonella, E. coli, bacteriophages and enteric viruses. No definite results are yet available.

While in some studies it appeared that the microbial concentration level in the sea was low in the summer as compared to other seasons, the contrary was observed for sediments. This is apparently due to increased daylight and solar radiation which may not reach sediments.

However, in other studies, higher mean values were noted during the summer season. This was attributed to the increase in coastal pollution due to the number of summer tourists accommodated along the coast.

The operational document proposed the following as compulsory parameters for the identification of effluents or outfalls in a minimum programme as mentioned in section 2.2: biochemical oxygen demand (BOD_5); settleable solids (SS); and the volume of the discharge.

These parameters were not widely applied, owing to the fact that in some of the monitored areas there were no effluents or outfalls of importance nearby. However, it was considered by some of the collaborating laboratories that the above parameters were insufficient for the assessment of the pollution in rivers and outfalls. They proposed to include pH, COD and TSS (total suspended solids) as well.

b. Shellfish areas

Here again, the great majority of collaborating laboratories used the four bacteriological indicators, i.e. total coliforms, E. coli, Streptococcus faecalis and total heterotrophic bacteria, included in the minimum compulsory programme.

To a lesser degree, the collaborating laboratories included physical and chemical parameters such as temperature, dissolved oxygen and meteorological factors in the parameters monitored. Once more, this was mainly due to the absence of the necessary facilities, equipment and personnel.

In the results of one of the collaborating laboratories, it was found that the positivity of V. parahaemolyticus in shellfish increased during the warmer months and decreased markedly during the cold ones. This seems to confirm the well-established view that V. parahaemolyticus is a natural inhabitant of sea-water and does not correlate with pollution.

From the experience gained during the project MED VII, it appears that the most important parameters in shellfish monitoring are faecal coliforms and faecal streptococci. Their importance is related to the hygienic and sanitary aspects of the control of shellfish. However, it has been found that although faecal streptococci and faecal coliforms are both indicators of faecal pollution, the value of their analyses cannot always be correlated. Thus, high values of enterococci were found in shellfish with few E. coli per gram of flesh.

Testing for Salmonella in shellfish flesh is receiving some attention, and consideration is under way to include it among the compulsory parameters.

Temperature of sea-water, salinity and rainfall with reference to date of sampling are considered to be the most important accompanying parameters and should be included among the compulsory parameters.

c. Fish

Although not specifically referred to in the operational document some investigations are being carried out on fish. In polluted waters, fish might have a concentration of bacteria and viruses from the water in their organs and muscles and become a public health hazard. The result of a preliminary study carried out by one of the collaborating laboratories indicated that when the concentration of coliforms in the water was $10^6/100\text{ml}$ and the total bacteria count was $1 \times 10^7/\text{ml}$, coliform bacteria were recovered from all the organs of fish as well as the muscles. The bacterial concentration in the organs was higher than in the water in which the fish lived. In view of the correlation between the concentration of bacteria in water and the recovery of bacteria from fish organs, the question arises as to whether fish grown in waters with recommended concentrations of faecal coliforms might not constitute a public health hazard.

2.3.2 Methodology and nutrients

a. Recreational waters

Guidelines

Realizing the importance of developing and applying similar methodologies and materials for the analysis of the various parameters, the project MED VII included in its first activities the preparation and distribution of a document entitled Guidelines for health related monitoring of coastal water quality, ((3) reference list). This was published under the joint sponsorship of UNEP and WHO. It was intended to help in promoting the steady improvement of skills, comparability of results and more efficient water quality control. The principal aim was to develop uniform reference methodologies for the monitoring aspects of project MED VII, and further promote comparability of results between laboratories and eventually between controlling agencies, nationally and internationally.

In addition to these Guidelines, the collaborating laboratories agreed to apply a more specific methodology for each of the compulsory microbiological parameters, as follows:

- (i) the same m-FC medium should be used for both coliform counts and faecal coliform counts. The suggested incubation temperature is $35^\circ + 0.5^\circ\text{C}$ for the total coliform count and $44.5^\circ + 0.2^\circ\text{C}$ (incubation in water bath) for the faecal coliforms count.

- (ii) liquid m-FC, as opposed to agar, should be used (Difco m-FC broth base (No.0883)) and rosolic acid; the rosolic acid is to be freshly prepared.
- (iii) the HA millipore filter (0.45 μ m, 47 mm) should be given preference.
- (iv) M-enterococcus agar (Difco should be used for faecal streptococci at an incubation temperature of $35^{\circ} + 0.5^{\circ}\text{C}$).

In order to give an additional impetus to apply the above methodology and use the suggested material, each laboratory was provided with nutrient and basic equipment relevant to the membrane filtration method (Table 3 outlines the material provided).

The adoption of the membrane filtration (MF) method brought some difficulties to the laboratories which were routinely using the most probable number (MPN) method; but some decided to run the two methods, in parallel, thus giving themselves the opportunity of comparing them.

Up to now, from the above exercise various contradictory results emerged. In one study it appeared that the two methods did not differ significantly. However, in other studies, the MPN results were giving higher counts. In yet another study, the comparison of the MPN with the MF method showed a fairly good correlation ($r = 0.8$). Table 4 lists the collaborating laboratories that run the MPN and the MF methods in parallel. Further investigation and results are necessary before any accurate appraisal of one or the other method can be made.

Another point that was raised by the collaborating laboratories concerned the suitability of the proposed nutrients for the microbiological parameters. From the experience gathered during the implementation of project MED VII by some collaborating laboratories, it appeared that the adopted m-FC medium incubated at 35°C for the determination of total coliforms was not giving satisfactory results. In this connection it was suggested that the M-Endo broth might be used. Here again the need for further experience was recognized before any decision could be taken in the matter.

Sampling stations

In the operational document it is specified that the samples should be taken at a distance of 10m from the low tide mark. However, this presented some difficulties, especially to those laboratories usually concerned only with microbiological parameters and which did not have an appropriate boat for sampling. Various solutions were applied. It was agreed to maintain the current situation in which some collaborating laboratories sampled the coastal sea-water at the knee or the chest level. However, it was recognized that sampling carried out near the shore was exposed to a number of interferences, of a local nature, which affected the quality of the

coastal water tested. Therefore the results might be different from those obtained by the established procedures. It was proposed that the laboratories able to carry out sea sampling both on the shore and at a 10m distance should be encouraged to take up comparative studies and report the results in due course. It was understood that the sampling method applied should be described in great detail. Based on expected results the sampling location might be redefined and adjusted as appropriate.

It was recognized that the number of sampling points should not necessarily follow a pre-established pattern but should be limited to the strict necessary. Existing local conditions and results so far experienced could help to eliminate unnecessary sampling points and thus decrease the work load and reduce the financial burden.

b. Shellfish areas

Guidelines

Here too, in order to strengthen harmonization and comparability of results it was agreed by the collaborating laboratories to utilize the same methodology and the same nutrients more specifically for the analysis of the compulsory parameters. The following procedures were agreed on:

- for sea culture water: the same methods and nutrients agreed on for coastal sea-waters should be applied.
- for shellfish flesh: the MPN or the spread plate (SP) method should be applied for total coliforms and faecal coliforms and pour plate (PP) method for the faecal streptococci.

Adherence to the above standardization and methodology has unfortunately not been achieved by the majority of the collaborating institutes and any comparison of the results obtained is weakened by this deficiency.

While, in general, the proposed nutrients were used, various other media were also applied. More experience and results are required in order to further assess the relevant value of the proposed nutrients and consider the need to change them or not.

Sampling stations

The sampling networks lacked uniformity. Their density and direction, and the number of sampling points, were not in accordance with some of the general guidelines. Additional work and studies seem to be needed in order to develop these guidelines and arrive at the most economical arrangement which, at the same time, would provide the indispensable minimum data to permit an appropriate assessment and comparison of the quality of the investigated areas.

2.3.3 Quality criteria

a. Recreational waters

The water quality criterion is a quantifiable exposure-effect relationship between the density of an indicator in the water concerned and the potential human health risks involved in using that water.

On the other hand, the water quality protection standard is an accepted maximum level for the density of an indicator in the water associated with unacceptable health risks. The concept of acceptability implies that social, cultural, economic and political, as well as medical factors, are involved.

Recreational water quality criteria are therefore a dose-response type of relationship between the illness and water quality. They are translated into guidelines or standards after a decision is made on the "acceptable risks". The last decision is a political one where social, economic, political and health factors are taken into consideration.

Recreational water criteria are used for several purposes, of which some of the most important are:

- (i) decisions as to the acceptability of existing beaches;
- (ii) decisions on permits for the establishment of new recreational facilities and beaches;
- (iii) design criteria for treatment and disposal systems for liquid wastes.

In order to review the above health criteria for recreational waters the operational document provided for the organization of an Expert Consultation. This group was intended to undertake, in particular: the comparison of the various standards for the quality of recreational waters and a review of recommended health criteria for the quality of coastal bathing waters. It was convened in Athens in 1977 and its results have been published, ((4) reference list).

The Expert Consultation, after examining the available evidence, came to the conclusion that there was as yet no basis for recommending changes in the conclusion reached by the WHO Working Group on Guides and Criteria for Recreational Quality of Beaches and Coastal Waters (Bilthoven, 1974), namely:

"Highly satisfactory bathing areas should however show E. coli counts of consistently less than 100 per ml and, to be considered acceptable, bathing waters should not give counts consistently greater than 1 000 E. coli per 100 ml", ((5) reference list).

However, the Expert Consultation expanded the above criteria. It was felt that while it might be justified to use the more lenient criterion of 1 000 E. coli per 100ml for the quality control of recreational waters at existing facilities, new installations should be designed to ensure a higher recreational water quality. Moreover, decisions on criteria for the design of treatment and disposal systems for liquid wastes, involving large investments and having long-term implications for water quality, should be based as far as possible on the stricter criterion of 100 E. coli per 100ml.

The Expert Consultation also felt that the numerical criteria should be more closely defined statistically, and recommended that the criterion of 100 E. coli per 100ml should be defined as follows: no more than 10 per cent of at least ten consecutive samples collected during the bathing season should exceed 1 000 E. coli per 100ml.

In project MED VII the above were considered as interim criteria, to be consolidated or revised following appropriate epidemiological studies. The results of the monitoring programme of the areas studied in the project are, in the great majority, meeting these interim criteria, and have therefore been satisfactory.

Questions concerning epidemiological studies for health hazards related to coastal water pollution are discussed further in section 3 of this document.

b. Shellfish

There is extensive epidemiological evidence that the consumption of raw or insufficiently processed shellfish may cause a variety of enteric bacterial and viral infections.

For the objectives of the project MED VII, it was recognized that appropriate microbiological limits for the first two phases in assessing shellfish quality (the culture area and the shellfish in its natural surroundings) should be established. It is understood that for a full assessment of shellfish quality as a food product, shellfish should also be examined at subsequent phases of handling (transport, processing, marketing).

Considering the criteria already applied by several countries, the following interim standards have been recommended:

(i) for shellfish

- expressed as faecal coliforms per gram of shellfish flesh:

0-2 - sale permitted

3-10 - temporary prohibition of sale

above 10- sale prohibited

(ii) for water of shellfish culture areas

- less than 10 faecal coliforms/100ml in 80 per cent of samples
- less than 100 faecal coliforms/100ml in 20 per cent of samples

Here again the results so far obtained in the monitoring of shellfish are according to the interim criteria, giving satisfactory results to a great extent.

2.3.4. Statistical analysis

Appropriate standard forms, tables and graphs were developed for assisting the recording of the collected data and their handling in an orderly fashion.

In addition to classifying the primary data according to seasonal variations, currents, winds or other, some collaborating laboratories used statistical methods in order to assist in the summarizing process, in the sorting out of facts and in the assigning of significant probabilities. Such methods included the frequency distribution, the calculation of the average and empirical variance, the 't' test procedure and other.

One of the collaborating institutes carried out a study on "Statistical variation of microbiological quality of coastal waters: regulatory implication".

From this study it appears that among others:

- the microbiological quality of coastal waters can be adequately interpreted by a log normal probability distributing model;
- correct compliance with any statistically expressed water quality standard requires comparison of two probability distribution and not only two pairs of frequencies;
- the standard deviation of the concentrations of three indicators approaches quite closely that implied by the proposed interim coastal waters quality criteria;
- the standard deviation of a microbial indicator concentration, at a sampling station, is a useful and sensitive parameter for detecting discontinuous sources of pollution;
- standard deviation estimate derived from sets of 12-14 values which lies outside the 1 to 3 interval can be likely associated to a singular water sampling station.

2.4 Quality control pilot programme

The execution of project MED VII has highlighted the need to develop a

quality control programme with the aim of identifying a variety of laboratory variables and specifying essential quality control practices to assure the present and continued productions of reliable data. The objectives of the quality control programme will be:

- to assess laboratory performances and deficiency;
- to harmonize practices and promote their comparability.

It represents for bacteriological analysis the intercalibration which is applied to physical and chemical analysis.

Two consecutive stages are envisaged. The first corresponds to internal quality control (intralaboratory tests). The second covers inter-laboratory tests and comprises two phases. During phase I, known standard strains will be mixed with sea-water, either each strain separately, or in various percentages, and the recovery will be measured by standard techniques in the various laboratories. During phase II, sea-water samples will be mixed with unknown strains and recovery will again be measured by standard techniques in the different laboratories.

The Regional Activity Centre has agreed to undertake, on a contractual basis, the responsibility for this activity and to carry out the work and coordination involved, namely:

- preparation of standard strains and their dispatch to each laboratory;
- preparation of simulated polluted samples;
- analysis of simulated samples at collaborating laboratories;
- preparation and distribution of instructions to collaborating laboratories;
- collection of the results of collaborating laboratories;
- development of decay curves;
- comparison of the results received with its own results;
- preparation of final report.

The above exercise is intended to be carried out on a trial basis twice a year. It is expected to start as soon as required funds are available.

3. INITIATION AND PROMOTION OF EPIDEMIOLOGICAL STUDIES

The hazards to human health from bathing waters arise from the swallowing of polluted water and from direct contact with the skin. There is circumstantial evidence of association between disease and bathing in

polluted water. However, additional epidemiological studies are required in order to better ascertain and establish the relationship of cause and effect. These studies should aim at providing the necessary data base for evaluating health effects and developing water quality criteria for the recreational use of coastal waters in the Mediterranean.

To this end the Expert Consultation mentioned under 2.3.3 was assigned the following objectives:

- study the epidemiological and other evidence on which various studies for recreational waters around the Mediterranean are based;
- recommend long-term and short-term research programmes aimed at providing reliable data for management decisions.

The meeting made recommendations for long- and short-term epidemiological studies. It developed a design for the conduct of "week-end type" prospective epidemiological-microbiological studies, and a second design for the conduct of similar studies on "organized vacation groups".

The meeting further recommended that three to four epidemiological studies be carried out in representative areas of the Mediterranean and, pending the results from these studies, that the proposed interim criteria should be applied.

Unfortunately the recommended studies did not materialize due to the absence of the necessary financial support. Under these circumstances no revision of the interim water quality criteria can be undertaken at this stage.

4. PRINCIPLES AND GUIDELINES FOR COASTAL WATER POLLUTION CONTROL

The operational document recognized the importance, complexity and need of coastal water pollution control. Effective marine pollution control will ultimately depend on modification, reduction and dispersion of wastes discharged and dumped into the sea. It will be necessary to institute and execute a series of local and regional pollution abatement programmes covering the major population and industrial centres around the Mediterranean. Such programmes should aim at the development of long-range plans covering large geographical areas.

To develop the necessary long-term comprehensive programmes, as well as taking into consideration their short-term impact, requires a multi-level approach. This will lead to the establishment of a hierarchy of individual projects and programmes of a similar nature, which will only vary in scope, depth and detail. It is also necessary to assure effective and prompt action at the local level and at the same time to provide the required framework for coordination of activities.

In response to the priority needs of the Mediterranean countries, the development of principles and guidelines for coastal water quality management has been included in project MED VII as one of its main objectives.

A Workshop was convened on Coastal Water Pollution Control (Athens, 1977), ((6 reference list), in order to examine the methodology for marine pollution control planning and to outline a plan of action leading to the development of a model code of practice for the disposal of liquid wastes into the Mediterranean.

The Workshop, among other proposals, suggested an outline of contents of a code of practice for coastal pollution control in the Mediterranean, intended as a guide to the preparation of the different sections of the code.

Considering the priority importance of pollution from land-based sources, UNEP, in collaboration with WHO and with the assistance of national Mediterranean experts, developed a Protocol for the protection of the Mediterranean Sea against pollution from land-based sources, which was agreed and signed at a conference of Plenipotentiaries in Athens, in May 1980, ((7) reference list).

To assist responsible national authorities in the negotiation and eventual implementation of the Protocol, a publication entitled Principles and guidelines for the discharge of wastes into the marine environment was prepared and published under the joint sponsorship of UNEP and WHO, ((8) reference list).

The material presented in the above publication may be used in various ongoing and planned activities of the Mediterranean Action Plan and in particular for the eventual development of a model code of practice for the management of waste from land-based sources. In fact, the content of the document referred to above covers part of the outline of a code of practice for coastal pollution control suggested by the Workshop.

Following the agreement on the Protocol, it is expected that the Principles and guidelines will constitute a first useful tool for its implementation.

5. DEVELOPMENT AND PROPAGATION OF TECHNICAL DOCUMENTATION

Much work has been undertaken in recent years on various aspects of marine pollution management such as criteria, standards, monitoring, control, etc. Relevant documentation was brought to the attention of the Principal Investigators participating in project MED VII as well as to other institutes and public services in the Mediterranean area. However, special care was taken not to copy blindly the results of the work undertaken in various places, but to try to adapt the experience gained to the conditions and specific needs of the area.

Annual meetings of Principal Investigators

During the entire operation of project MED VII, annual meetings with the Principal Investigators were organized. They proved to be a most efficient forum where the analysis of existing knowledge and experience and their adaptation to the needs and conditions of the Mediterranean area could take place with outstanding success.

In addition to the above selective action for the promotion of scientific knowledge, the periodic meetings became an efficient means for the propagation and exchange of knowledge and experience. Moreover, contact among the Principal Investigators was not confined to the meetings, being expanded to include individual and personal contact on technical matters. It became the first practical step towards the indispensable close collaboration among the Mediterranean states in order to succeed in the harmonized assessment and protection of the marine environment and the coastal area.

The educative character of these yearly meetings was no less important. They provided a forum where questions were raised and answers provided and discussed, where problems were presented and their solutions worked out.

In these meetings, the work under way was presented, discussed, analysed and evaluated. Directives for the future of the project were considered and accordingly developed. Plans, not only for the immediate future, but also for future years were among their endeavours. Finally they provided a platform where any problems relevant to the quality control of coastal areas could be discussed and where recommendations on any of the appropriate subjects could be made. Some priority recommendations will be discussed in the last section of the report, "Discussion and conclusions".

The reports on the annual meetings of Principal Investigators jointly convened by WHO and UNEP were printed and circulated, ((9,10,11,12) reference list).

Technical meetings and relevant documentation

Below is a consolidated list of various technical meetings and publications which have already been referred to in the appropriate sections.

- (i) Group of Experts on Guidelines for Health Related Monitoring of Coastal Water Quality (Rovinj, 1977), ((3) reference list). See section 2.3.2(a).

The development of the Guidelines took place to complement the operational document and to meet the needs of the participants in project MED VII in carrying out the monitoring activities.

The first draft was prepared by a WHO consultant, reviewed and adapted by the Group of Experts, and the mid-term review meeting for the project, ((9) reference list). The Guidelines are provisional and should be revised when further experience gathered may justify relevant changes.

- (ii) Group of Experts on Health Criteria and Epidemiological Studies Related to Coastal Water Pollution (Athens, 1977), ((4) reference list). See section 2.3.3(a).

The main objectives of the activity were to review health effects criteria, guidelines and standards related to recreational coastal waters, to develop

recommendations for the design of long-and short-term epidemiological studies in order to produce the required criteria, and to propose interim criteria.

The proposed work was carried out by the Group of Experts. The main results of this meeting were the development of interim criteria for the quality of recreational coastal waters, which were used during the project period, and the development of two types of protocols for the conduct of epidemiological studies.

The interim criteria were not revised, as the proposed epidemiological studies were not completed and therefore no data were available for such a revision.

- (iii) Workshop on Coastal Water Pollution Control (Athens, 1977), ((6) reference list). See section 4.

The Workshop recognized that there is an urgent need to provide governments with information about existing experience and proven methods to assist in national decision making.

Moreover, it recognized that coastal pollution problems in the Mediterranean have certain common factors which justify the development of a shared philosophy and approach in relation to objectives and ways and means of achieving them. Harmonization of efforts should be ensured through the development of a model code of practice for the disposal of liquid wastes into the Mediterranean.

- (iv) Principles and Guidelines for the discharge of wastes into the marine environment, ((8) reference list).

This publication was intended to assist responsible national authorities in the implementation of the Protocol for the Protection of the Mediterranean Sea Against Pollution from Land-based Sources.

Periodic bulletin

Although not a purely technical document, this publication, issued by the Regional Activity Centre has contributed substantially to the propagation of technical and other information. It is a useful addition to the contacts achieved by the annual meetings and to the occasional correspondence with or between the collaborating laboratories.

6. TRAINING AND TECHNICAL ASSISTANCE

Training and technical assistance were part of the activities intended for inclusion in the work of project MED VII. Their contribution to the scientific results, although indirect, was none the less important and effective.

On technical grounds, the form of training that was more agreeable, useful and acceptable to the laboratories was the "on-the-job" training. There, the direct contact between the parties concerned, the concrete aspects for

discussion and the direct field approach were positive points with regard to the efficiency of the on-the-job training approach. Moreover this form of training strengthened and multiplied contacts between laboratories and scientists.

The training aspects included in the annual meetings of Principal Investigators were also important and successful on scientific grounds. The exchange of views and knowledge, the discussion on the summary reports of the collaborating laboratories, the opinions of the majority, the conflicting results of laboratory analyses included a training component of great value. This appraisal was shared by the collaborating laboratories who, when a choice was given, expressed a preference for holding a seminar rather of a training course.

The assistance in material and equipment consisted mainly of basic supplies necessary to carry out the MF method of microbial analysis by all the collaborating laboratories (Table 4). This was of substantial help in promoting the use of the same methodology and nutrient, thereby facilitating comparability. Moreover, it stimulated the execution of parallel runs with different methodologies and their relevant comparison.

7. DISCUSSION AND CONCLUSIONS

7.1 The monitoring of selected coastal areas

The nomination by the Mediterranean states of collaborating institutes and the subsequent selection of coastal areas to be monitored, did not provide a balanced representation of either the Mediterranean countries or their surrounding coastal areas. These deficiencies should be remedied for any planned follow-up to monitoring activity in any future programme.

Areas to be monitored should respond to well-defined characteristics in order to fulfil the following basic objectives:

- (i) provide a balanced picture of the Mediterranean area as a whole;
- (ii) cover mainly places affected by comparatively large concentrations of population and industry;
- (iii) cover adequately coastal areas used by large numbers of people for recreational purposes and exposed to pollution from land-based sources;
- (iv) cover large shellfish culture areas exposed to pollution from land-based sources.

Any proposed monitoring of quality of coastal areas should not be limited to bacteriological quality, as in the case of project MED VII, but should cover all relevant and necessary physical, chemical, bacteriological and other parameters of quality.

In proposing a future monitoring programme on quality of coastal areas, due attention should be given to including monitoring of land-based sources of pollution, i.e. rivers, outfalls from major municipal sewage discharges, outfalls from industrial units and any others which may affect substantially the selected monitoring area.

7.2 Methods and materials

While it was agreed by the Principal Investigators that the compulsory parameters utilized up to now for recreational coastal waters should be maintained, they suggested that the total number of coliforms should be discontinued in the future for routine monitoring. Notwithstanding, total coliforms might be used as an indicator for cleaner waters. Thus, basically faecal coliforms and faecal streptococci should be retained as compulsory bacteriological parameters.

As far as shellfish monitoring is concerned, it is considered that the four compulsory bacteriological parameters, namely total coliforms, faecal coliforms, faecal streptococci and heterotrophic bacteria, could be reduced to two: faecal coliforms and faecal streptococci.

Considering the difficulty experienced by a number of laboratories in monitoring all the meteorological, hydrographic, dynamic and visual parameters included in the minimum compulsory programme, further studies are required for simplifying them or limiting them to a necessary strict minimum.

As far as shellfish culture areas are concerned, physical and chemical parameters may not be of great importance for routine monitoring. However, this view requires additional investigation as it is not shared by all the collaborating institutes. The importance of testing for Salmonella in shellfish and its inclusion in the minimum programme is being considered. However, further study is still needed before recommending it for inclusion in the minimum compulsory programme.

The results of the comparison of the compulsory MF method with the MPN method, now being undertaken by a number of laboratories, will clarify this issue. From preliminary readings, results appear to be comparable.

As far as media are concerned, there is some evidence that the m-FC broth for the detection of total coliforms in sea-water presents some difficulties in interpreting the results. A comparative study using m-Endo broth or agar is under way in order to resolve this controversy.

For the time being, no changes have been agreed on regarding the compulsory methods and culture media established for the testing of total coliforms, faecal coliforms and streptococci.

Monitoring of water sediments, although an important indicator of pollution, is not included in the compulsory minimum programme. This is due to difficulties encountered by collaborating laboratories in the collection of such samples. However, they are encouraged to undertake sediment collection and analysis.

Sampling stations located near the sea-shore present some problems. A proposed comparative study to be undertaken by the collaborating laboratories may provide a solution regarding the present inconsistencies in sampling, which is currently made at knee-height, or at chest-height, or at 10 metres' distance from the low tide mark.

Project MED VII was not a "one-shot" exercise intended to provide rigid rules and procedures for the establishment and management of a monitoring network, required for the assessment of pollution and for providing information for its appraisal and control. On the contrary, it was intended to develop a dynamic and at the same time elastic approach that would permit improved efficiency and quality, and expansion of the work carried out. To this end, the elementary research component included in the pilot phase should be strengthened and continued in any proposed follow-up long-term programme.

On considering the above matter, the participants in project MED VII recommended the following research activities as being of priority importance:

- (i) isolation of pathogenic bacteria and human enteroviruses and their survival in sea-water;
- (ii) fish monitoring for the presence of pathogenic bacteria;
- (iii) study of certain micro-organisms such as Pseudomonas aeruginosa and Vibrio parahaemolyticus as new parameters;
- (iv) monitoring of pathogens as indicator bacteria in shellfish and culture areas.

7.3 Criteria and epidemiological studies

Project MED VII did not go further than to establish two interim criteria, one for recreational water and the other for shellfish (culture areas and shellfish flesh). These should be translated into guidelines and standards. To this end, a decision as to "acceptable risk" of symptoms of varying degrees of severity or of specific diseases should be made. Such decisions on the pollution control programmes to achieve these standards are essentially political. However, the interim criteria should be consolidated or amended, following epidemiological studies, which should give the dose-response type relationship between illness and water quality.

Lack of relevant support did not allow the implementation of the proposed epidemiological studies. However, these are indispensable and should be included in future programmes in order to give the interim criteria their correct value and permit the consideration of standards.

Further work on the formulation of selected environmental quality criteria should be included in any proposed follow-up long-term programme.

7.4 Principles and guidelines for coastal water pollution management and control

Following the progress made in the monitoring and assessment of pollution the time has come to proceed from monitoring to pollution control. As mentioned, a Protocol for the protection of the Mediterranean Sea against pollution from land-based sources, ((7) reference list), has recently been agreed by the Mediterranean countries. This provides the legal basis for the development and implementation of a pollution control programme.

The recommended plan of action for the development of a model code of practice could provide the basis for such a control programme.

The following items should be dealt with in the code of practice:

- (i) information systems for coastal pollution control;
- (ii) criteria for the design of collection, treatment and disposal systems for waste waters, including reuse;
- (iii) design for collection, treatment and disposal systems;
- (iv) environmental impact assessment;
- (v) administrative, financial and allied aspects.

Part of the model code of practice has already been prepared and included in the Principles and guidelines for the discharge of wastes into the marine environment

Further studies and activities should be promoted for pollution control in the Mediterranean and should be included in a proposed long-term programme. Local and national activities should be stimulated, which will assist in the study and assessment of methods of control of coastal water pollution due to municipal sewerage. Attention should also be given to present deficiencies in the required qualified personnel and their training.

The development of appropriate administrative procedures which will ensure the necessary coordination should be studied by the interested countries and an efficient solution applied. Relevant manpower needs and structure should equally be considered. Administrative practices such as the issuing of appropriate individual licences should be developed and introduced wherever appropriate. These should be in accordance with the requirements of the Protocol.

The potential environmental impact of new installations around the Mediterranean Sea should be considered. A number of the above-mentioned tasks may involve international cooperation when the proposed relevant programmes are to be undertaken jointly by the interested countries.

Such programmes may concern:

- establishment of detailed pollution source inventories and the assessment of waste loads reaching the Mediterranean (common methodology would ensure the comparability of results);
- preparation of sampling schemes and analytical procedures for monitoring of municipal and industrial effluents;
- monitoring of effluents of rivers, in particular the more polluted and the establishment of standardized measurement methodology, as well as data handling and analytical quality control.

The development of a model code of practice already mentioned may also fall into this category.

The development of technical documentation and its diffusion should be continued and even strengthened. In fact, the agreement on the Protocol will increase substantially the activities in the field of control and management. A number of guides, codes of practice and similar manuals will be required in order to promote harmonization and a standardized approach, thus increasing the comparability of results.

7.5 Training

Although successful, the training component was too small. It should be enlarged substantially, considering also that the Protocol referred to above will undoubtedly require the training of a great number of technical personnel at various levels and belonging to various disciplines.

The substantial results, and the successful multiple role that the occasional meetings achieved, advocate their continuation in the future. However, in view of the wide character that a proposed long-term programme should have, it would be advisable to have the various disciplines represented in the periodical meetings.

References

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4. WHO/UNEP - Health criteria and epidemiological studies related to coastal water pollution. Copenhagen, 1977.
5. WHO Regional Office for Europe - Guidelines and criteria for recreational quality of beaches and coastal waters. Copenhagen, 1975.
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7. UNEP - Conference of Plenipotentiaries of the coastal states of the Mediterranean region for the protection of the Mediterranean Sea against pollution from land-based sources (Athens, 1980). Final Act and Protocol. New York, United Nations, 1980.
8. WHO/UNEP - Principles and guidelines for the discharge of wastes into the marine environment. Copenhagen, 1979.
9. WHO Regional Office for Europe - Mid-term review of the joint WHO/UNEP coordinated pilot project on coastal water quality control in the Mediterranean (MED VII). Copenhagen, 1977.
10. WHO Regional Office for Europe - First report on monitoring of recreational coastal water quality and shellfish culture areas. Copenhagen, 1978.
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12. WHO Regional Office for Europe - Third report on coastal quality monitoring of recreational and shellfish areas (MED VII). Copenhagen, 1980.

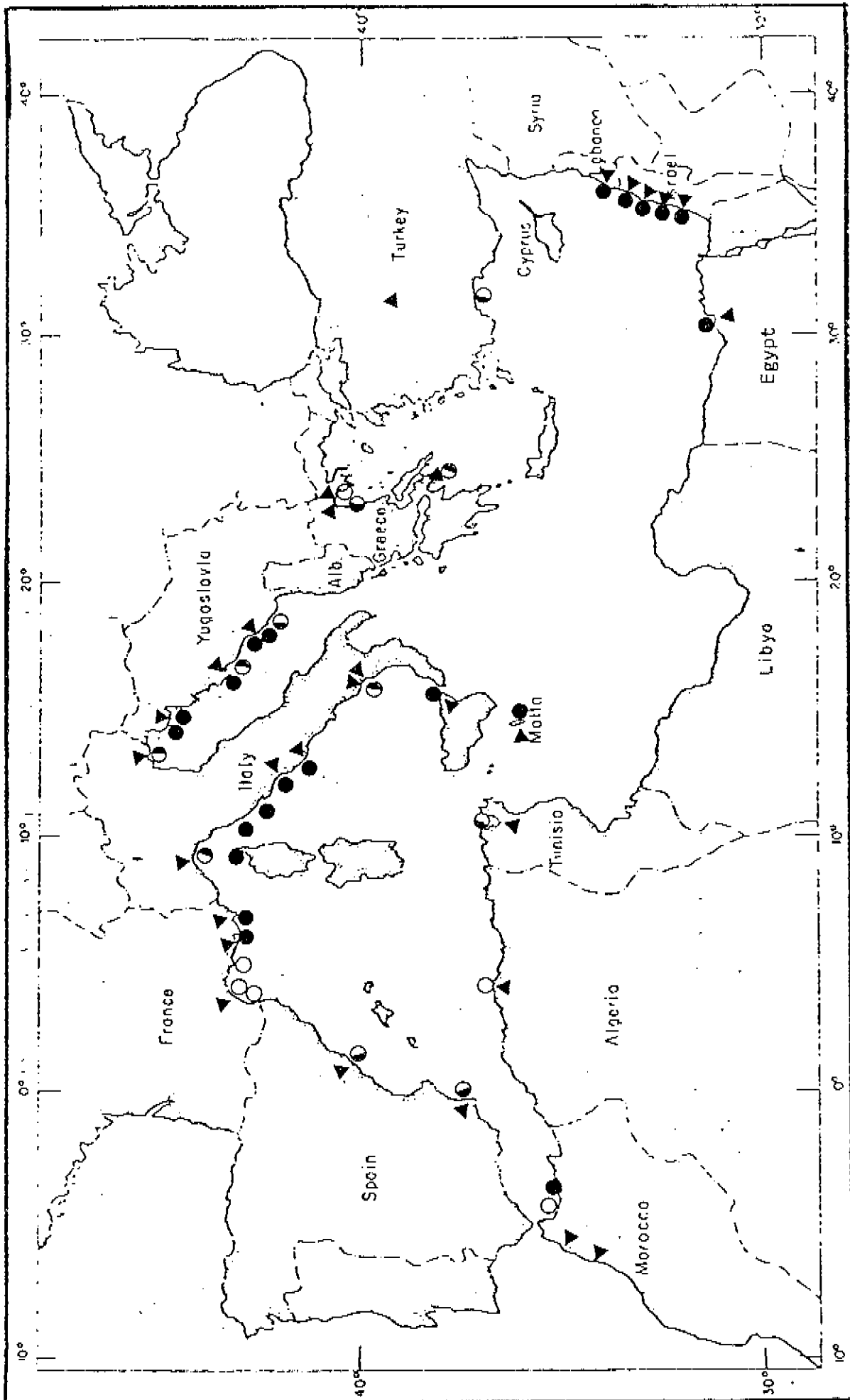


Figure I Collaborating Laboratories and monitored areas

Areas monitored for:

- Shellfish
- Recreational waters
- Shellfish and recreational waters
- ▲ Collaborating Laboratories

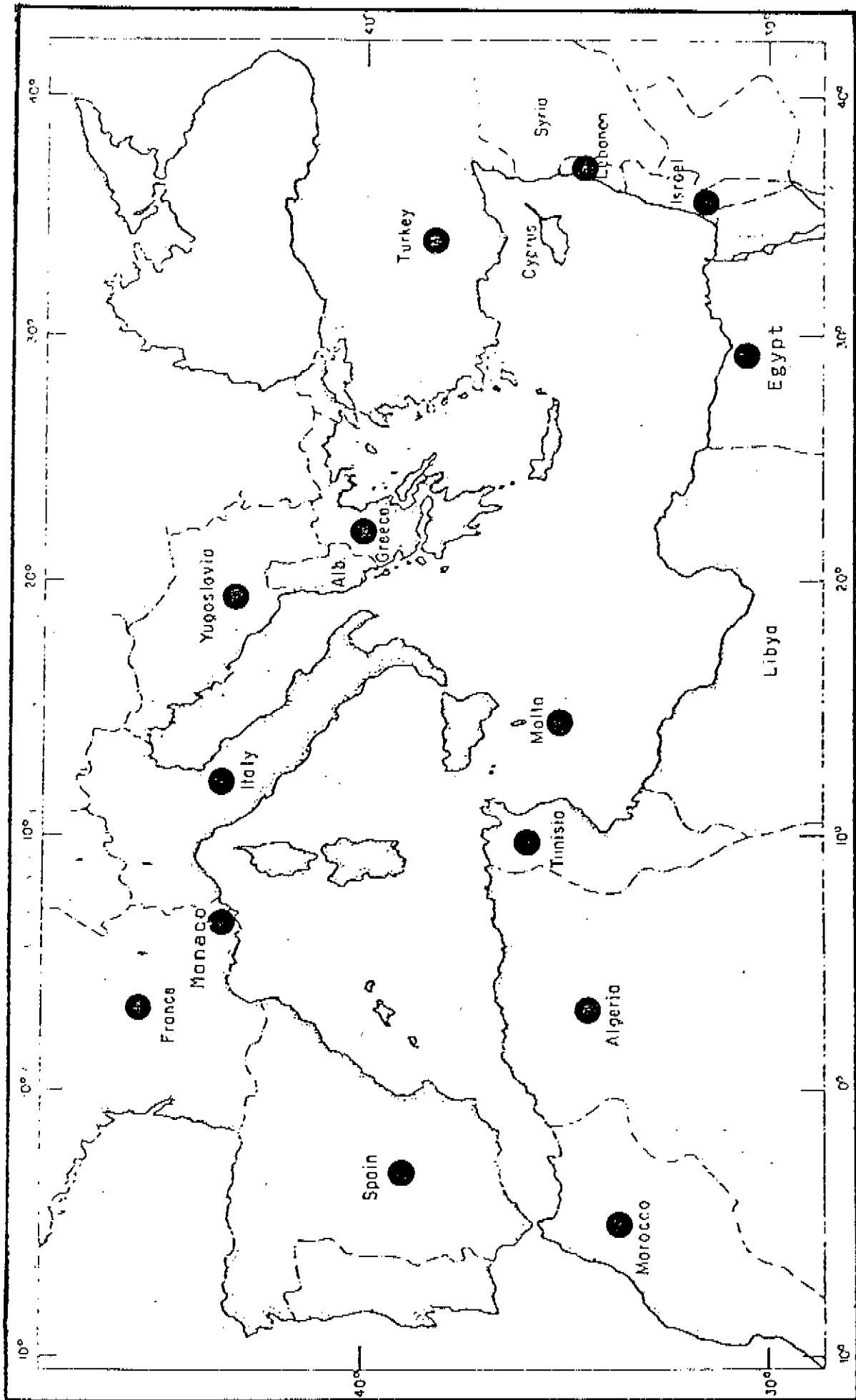


Figure II: Countries participating in Project ALE VII

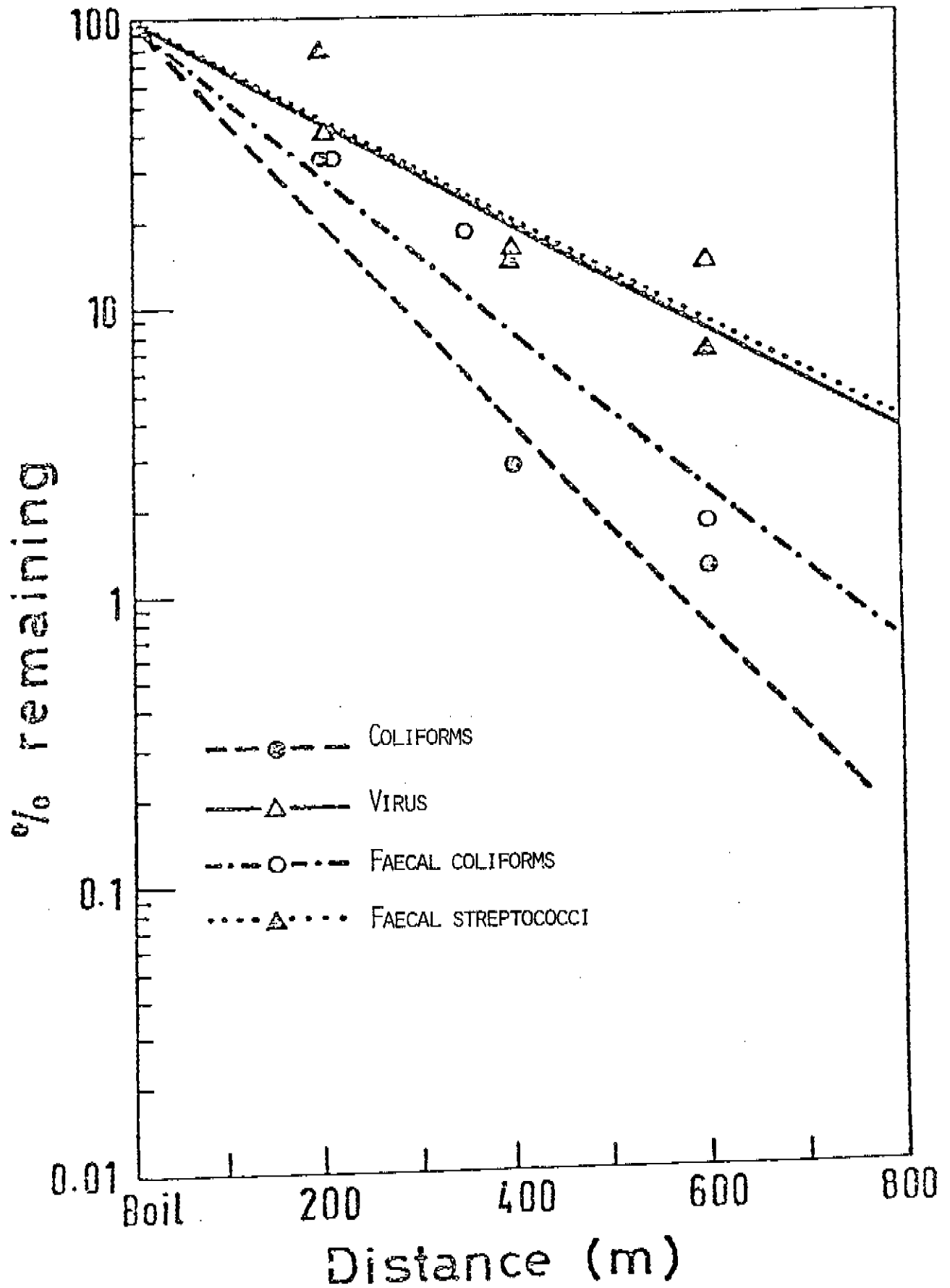


Figure III: Disappearance of enteric micro-organisms at various distances from the point of discharge

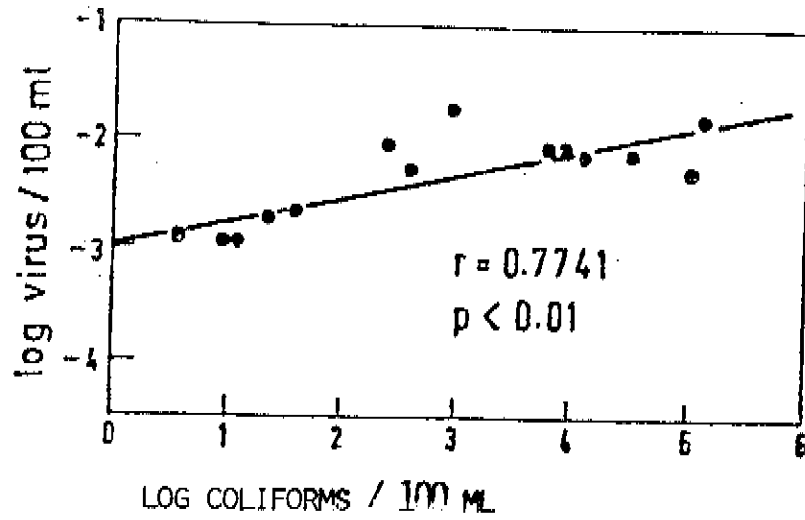


Figure IV: Correlation between Coliforms and viruses found at beaches

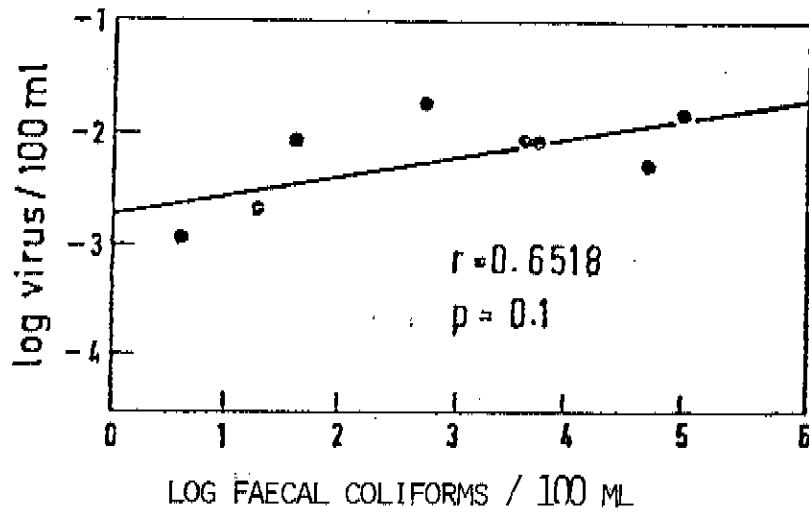


Figure V : Correlation between Faecal coliforms and viruses found at beaches

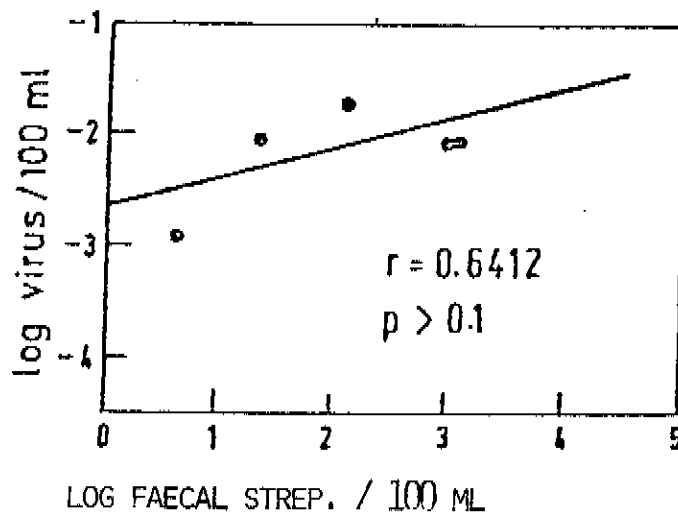


Figure VI: Correlation between Faecal streptococci and viruses found at beaches

Table 1

Optional parameters, applied by some of the institutes
collaborating in project MED VII

Code No.	Microbiological	Chemical	Biological
27.02			
35.04			
35.08		Nutrients (occasionally) BOD ₅ , COD, DO	
36.06	<u>V. parahaemolyticus</u> , <u>Salmonella</u> , heterotrophic bacteria	BOD ₅ -DO	
36.07	<u>V. parahaemolyticus</u> , <u>Salmonella</u> <u>Shigella</u> , fungi, sulphite reducer, <u>Clostridium</u> , coagulase-positive staphylococci, enterococci	BOD ₅ -pH-DO	
36.11	Total and faecal coliforms, faecal streptococci in the sediments, fungi in the sand - <u>V. parahaemolyticus</u> <u>Salmonella</u> , <u>Shigella</u> , <u>Vibrio</u> <u>cholerae</u>	DO	
47.02	Enterovirus		

Code No.	Microbiological	Chemical	Biological
47.03	<u>Salmonella</u> , enterovirus, bacterial count		
47.04	<u>V. parahaemolyticus</u> , <u>Salmonella</u>		
47.05	<u>V. parahaemolyticus</u> , <u>V. cholerae</u> , <u>Salmonella</u> , <u>Shigella</u>	COD, pH	
48.01			
48.06/48.12	Heterotrophic bacteria		
48.09	<u>Salmonella</u> , virus, enterococci	NH ₄ ⁺ , NO ₂ ⁻ , NO ₃ ⁻ , P, MBAS	
48.10	<u>Salmonella</u>	Nutrients	phytoplankton
48.11	Virus	Nutrients-pH	chlorophyll
48.15			
52.01	<u>Vibrio - Salmonella</u>	pH	
MA.03			
MO.01	<u>Salmonella</u>	Nutrients	
56.03			
29.05			

Code No.	Microbiological	Chemical	Biological
29.06			
88.03	sulphite reducer, <u>Clostridium</u> , total bacteria, <u>Pseudomonas</u> , <u>Aeromonas</u>		
89.05			
95.02	Heterotrophic bacteria	Nutrients	phytoplankton
95.04		BOD ₅ pH	
95.05		Nutrients	phytoplankton, chlorophyllie

Table 2

Correlation Between the Mandatory Parameters

Sampling points	r		r		r	
	TC	: FC	TC	: FS	FC	: FS
No. 1	0.995		-0.013		-0.003	
No. 2	0.889		0.348		0.401	
Nos. 1 & 2	<u>0.988</u>					
3	0.823		0.061		0.336	
4	0.748		0.278		-0.126	
Nos. 3 & 4	0.763					
No. 5	0.993		-0.187		-0.178	
No. 6	0.540		-0.805		0.159	
Nos. 5 & 6	0.790					
Control a for 1 & 2	0.485		-0.293		-0.299	
Control b for 5 & 6	0.924		-0.362		-0.408	
Total number of samplings = 69	<u>0.929</u>					

There is a good correlation between the total coliforms and faecal coliforms when the number of samples is adequate, but no correlation between any of them and the number of faecal streptococci.

Table 3

Basic Material and Equipment Provided by WHO/UNEP
to Every Collaborating Laboratory Joining
Project MED VII

Material and equipment	Quantity
Membrane filter holders. Teflon-faced Pyrex, 47 mm (Millipore XX10-047-20)	2 pcs.
Membrane filters (with grid), diameter of pore 0.45 um (Oxoid MF 47)	1000 pcs.
m-enterococcus agar (Difco 0746-01-8)	5 lb. (2.27 kg)
m-FC broth base (Difco 0883-01-1)	11 lb. (4.99 kg)
Rosalic acid (Difco 3228-09-1)	12 g.
Sterifil filter holders for 47 mm MF filter (Millipore XX11-047-10)	4 pcs.
Funnel cover, rubber (Millipore XX25-047-54)	1 pc.
Vacuum filtering flask, 1 litre (Millipore XX10-47-05)	2 pcs.
Vacuum pressure pump, 220 v 50 Hz (Millipore XX60-220-50)	1 pc.
Vacuum hose, gum rubber, 1.2 m. (Millipore XX71-000-04)	2 pcs.
Filter forceps (XX62-000-06)	1 pc.
Filter holder, hydrosol manifold, 6 places (Millipore XX25-047-00)	1 pc.

Table 4

Methods used for Analysing Compulsory Microbiological Parameters
by the Collaborating Laboratories

MPN - most probable number of multiple tube method
MF - membrane filtration method with recommended media
PP - pour plate method

Code No.	Country and designated institution	Bacteriological test			Remarks
		Total coliforms	Faecal coliforms	Faecal streptococci	
AL.01	<u>ALGERIA</u> 1. Centre de recherches Océanographiques et des Pêches, Alger	-	-	-	No information available
27.02	<u>EGYPT</u> 1. Centre for Postgraduate Studies and Research, Alexandria University	MF	MF	MF	
35.08	<u>FRANCE</u> 1. Scientific & Technical Institute for Sea Fisheries (ISTPM), Sète	MPN	MPN	MPN	
35.04	2. Research Centre for Biology & Medical Oceanography (CERBOM), Nice	MF	MF	MF	

Code No.	Country and designated institution	Bacteriological test			Remarks
		Total coliforms	Faecal coliforms	Faecal streptococci	
	<u>GREECE</u>				
36.07	1. Laboratory of Hygiene, Medical School, University of Thessaloniki	MF/MPN	MF/MPN	MF	
36.11	2. Environmental Pollution Control Control Project, Ministry of Social Services, Athens	MF/MPN	MF/MPN	MF	
36.06	3. Department of Food Hygiene, Veterinary faculty, University of Thessaloniki	MF/MPN	MF/MPN	MF	For shellfish only MPN method for coliforms and faecal coliforms and PP for faecal streptococci
	<u>ISRAEL</u>				
47.03	1. Environmental Engineering Laboratories, Technion, Haifa	MF	MF	MF	
47.02	2. Environmental Health Laboratory, Hadassah Medical School, Hebrew University, Jerusalem	MF	MF	MF	
47.04	3. A. Felix Public Health Laboratories, Ministry of Health, Tel Aviv	MPN/MF	MPN	MF	

Code No.	Country and designated institution	Bacteriological test			Remarks
		Total coliforms	Faecal coliforms	Faecal streptococci	
47.05	4. Public Health Laboratories, Ministry of Health, Haifa	MPN/MF	MPN	MF	
	<u>ITALY</u>				
48.10	1. Higher Institute of Health, Rome	MF	MF	MF	
48.12	2. Zoological Station, Naples				
48.06	3. Centre for Study and Research in Sanitary Engineering, Institute of Water Supply and Wastes Disposal, Naples	MF	MF	MF	
48.11	4. Institute of Water Research (CNR), Rome	MF	MF	MF	
48.15	5. Institute of Hygiene, Genoa	MPN/MF	MPN/MF	MPN/MF	
48.01	6. Institute of Hydrobiology and Fish Culture, Messina	MF	MF	MF	

Code No.	Country and designated institution	Bacteriological test			Remarks
		Total coliforms	Faecal coliforms	Faecal streptococci	
48.09	7. Institute of Hygiene, Trieste	MF	MF	MF	
52.01	<u>LEBANON</u> 1. Marine Research Centre, National Council for Scientific Research, Beirut	MF	MF	MF	
MA.03	<u>MALTA</u> 1. Public Health Laboratory, Ministry of Health & Environment, Valletta	MF/MPN	MF	MF	
MO.01	<u>MONACO</u> 1. Scientific Centre of Monaco, Monte-Carlo	MF	MF	MF	
56.03	<u>MOROCCO</u> 1. National Institute of Health Rabat 2. Scientific Institute of Marine Fisheries, Casablanca	MF	MF	MF	No information available

Code No.	Country and designated institution	Bacteriological test			Remarks
		Total coliforms	Faecal coliforms	Faecal streptococci	
29.06	<u>SPAIN</u> 1. Laboratories and Services of the Malaga Provincial Health Authority, Malaga	MPN/MF	MPN/MF	MPN/MF	
29.05	2. Laboratories and Services of Iarragona Provincial Health Authority, Iarragona	MPN/MF	MPN/MF	MPN/MF	
88.03	<u>TUNISIA</u> 1. Institut Pasteur de Tunis, Tunis	MF	MF	MF	
89.05	<u>TURKEY</u> 1. Environmental Engineering Department, Middle East Technical University, Ankara	MF	MF	MF	
95.04	<u>YUGOSLAVIA</u> 1. Rudjer Boskovic Institute, Centre for Marine Research, Rovinj	MF/MPN	MF/MPN	MF	

Code No.	Country and designated institution	Bacteriological test			Remarks
		Total coliforms	Faecal coliforms	Faecal streptococci	
95.02	2. Institute for Oceanography and Fisheries, Split	MF/MPN	MF/MPN	MF	
95.05	3. Marine Biological Station, Institute of Biology, University of Ljubljana, Portoroz	MF/MPN	MF/MPN	MF	MPN for total and faecal coliforms for shellfish