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REPORT OF THE FAO/UNEP/IOC WORKSHOP ON THE
BIOLOGICAL EFFECTS OF POLLUTANTS ON
MARINE ORGANISMS

Malta, 10-14 September 1991

Convened in the framework of the MED POL Programme and
organised jointly with the Euro-Mediterranean Centre
on Marine Contamination Hazards (Council of Europe)

PRELIMINARY

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TABLE OF CONTENTS

	<u>Page No.</u>
Introduction	1
1. Opening of the meeting (Agenda item 1)	1
2. Background and scope (Agenda item 2)	2
3. Election of officers (Agenda item 3)	3
4. Adoption of the agenda (Agenda item 4)	3
5. Organization of the work (Agenda item 5)	3
6. Presentation of papers (Agenda item 6)	3
7. The applicability of the techniques in field studies on a routine basis and their significance and interpretation vis-à-vis marine pollution risk assessment (Agenda item 7)	5
8. Future research needs (Agenda item 8)	7
8.1 Toxicity testing procedures	8
8.2 Choice of testing conditions and substrates	9
9. Recommendations (Agenda item 9)	10
10. Any other matter (Agenda item 10)	11
11. Adoption of the report (Agenda item 11)	12
12. Closure of the meeting (Agenda item 12)	12
ANNEX I : List of participants	13
ANNEX II : Agenda of the meeting	27
ANNEX III : List of papers presented	29
ANNEX IV : Papers presented at the Workshop	31

Introduction

The present Workshop on the Biological Effects of Pollutants on Marine Organisms was convened by FAO, UNEP and IOC and organized jointly with the Euro-Mediterranean Centre on Marine Contamination Hazards (Council of Europe) in the framework of the Long-term Programme for Pollution Monitoring and Research in the Mediterranean (MED POL - Phase II) which constitutes the scientific and technical component of the Mediterranean Action Plan. One of the objectives of the MED POL programme is to generate information that can be used for the development of environmental quality objectives in the context of the technical implementation of the Protocol for the Protection of the Mediterranean Sea against Pollution from Land-Based Sources, which has already been ratified by almost all the Mediterranean States. The study of the biological effects of pollutants on marine organisms is essential in assessing marine pollution risks and establishing water criteria.

The Workshop was hosted by the Foundation for International Studies, Valletta, Malta, which is an autonomous self-governing organization devoted to the pursuit of research and training at international level in a number of fields including environmental matters. It took place from 10 - 14 September 1991 and was attended by 50 participants from Algeria, Bulgaria, Canada, Egypt, France, Greece, Italy, Malta, Romania, Spain, Tunisia, Turkey, U.K. and Yugoslavia, as well as by representatives of FAO, IOC/GEEP and the Foundation. A number of participants was also attending the Second Intensive Training course on the Applications of Ecotoxicology in the Monitoring, Regulation and Control of Marine Pollution in the Mediterranean, which took place in Malta from 3 - 14 September 1991. A list of all participants appears as Annex I.

1. Opening of the meeting (Agenda item 1)

The Workshop was opened on Tuesday morning by Mr. G.P. Gabrielides, FAO Senior Fishery Officer (Marine Pollution), on behalf of the Food and Agriculture Organization of the United Nations and by Prof. S. Busuttil, Director-General of the Foundation for International Studies, Malta, and Executive Chairman of the Euro-Mediterranean Centre. Mr. Gabrielides welcomed the participants to the Workshop, thanked the Foundation for accepting to host it and pointed out the excellent co-operation which exists between the Co-ordinating Unit for the Mediterranean Action Plan and the Euro-Mediterranean Centre on Marine Contamination Hazards. He also expressed the wish that the Workshop would motivate and encourage Mediterranean

scientists interested in ecotoxicology to experiment with some promising biochemical and physiological response techniques being used in northern Europe for assessing marine pollution effects. He reiterated the intention of his Organization to suggest, in the future, the introduction of biological effects monitoring in the framework of the MED POL programme.

Prof. Busuttil welcomed the participants to Malta and expressed his pleasure that his Foundation was able to host the Workshop and co-operate with the Mediterranean Action Plan. Mr. V. Axiak, Professor of Biology at the Malta University and Chairman of the Programme Co-ordinating Board of the Euro-Mediterranean Centre on Marine Contamination Hazards, and Mr. A. Micallef, Director of the Centre, associated themselves with the welcome remarks and briefly reviewed some of the activities of the Centre.

2. Background and scope (Agenda item 2)

Mr. Gabrielides outlined the background and scope of the Workshop. In doing so, he referred to the previous MED POL activities in this field and specifically to the two pilot studies which took place in Phase I concerning the effect of pollutants on organisms, communities and ecosystems and the two research activities in Phase II, namely, activity G dealing with the toxicity, persistence, bioaccumulation, carcinogenicity and mutagenicity of chemical contaminants and activity I, dealing with ecosystem modifications in areas influenced by pollutants. Recently, the research component was revised and these activities now take place in the framework of Research Area III - "Effects".

The present Workshop would concentrate on techniques to study the sublethal toxic effects at the "individual" level of organisation or below. There exist a multitude of toxicological tests at the sublethal level which consider the pollutant stress on the biochemistry, physiology, behaviour, genetics and other aspects of biological activities of the individual organism. However, the utility and relevance of many of these techniques is still being assessed. The IOC/UNEP/IMO Group of Experts on the Effects of Pollutants (GEEP) has so far organized three practical workshops to compare and evaluate a number of techniques currently available or proposed for measuring the biological effects of pollutants at levels from the cell to the community.

The aim of the present Workshop is to provide a platform for Mediterranean scientists working in this field:

- a) to present their work relevant to biological effects;
- b) to exchange views and information on the various problems involved;

- c) to discuss the applicability of the techniques in field studies on a routine basis and their significance and interpretation vis-a-vis marine pollution risk assessment;
- d) to make recommendations on research needs and other matters.

3. Election of officers (Agenda item 3)

The Workshop unanimously elected Mr. Victor Axiak, Professor of Biology at the University of Malta, as Chairperson, Dr. Marc Lafaurie, Maître de Conférence at the University of Nice, as Vice-Chairperson and Dr. Giovanni Pagano, Senior Research Officer at the Istituto Nazionale Tumori, Naples, as Rapporteur. Mr. Gabriel P. Gabrielides, FAO Senior Fishery Officer (Marine Pollution) at the Co-ordinating Unit for the Mediterranean Action Plan, Athens, acted as Technical Secretary of the Workshop.

4. Adoption of the agenda (Agenda item 4)

The provisional agenda as proposed by the Technical Secretariat was accepted without modifications. It appears as Annex II.

5. Organization of the work (Agenda item 5)

The Workshop agreed to devote the first two days for the presentation of papers. It would work in plenary only but, if necessary, drafting groups could be formed for specific tasks assigned by the Chairperson. The working hours would be 09.00 - 12.30 hours for the morning session and 14.30 - 18.00 hours for the afternoon session. Friday afternoon was reserved for the writing up of the report.

6. Presentation of papers (Agenda item 6)

Twenty one papers were presented to the Workshop, and these dealt with a range of biological responses of different test species to marine contaminants (see Annex III). During the first half of the session devoted to paper presentations, Dr. Ken Renton, in his key-note paper, gave an overview of the molecular indicators of pollution, in which the utility and limitations of the present biomarkers were assessed and the need for the incorporation of biochemical indicators in pollution monitoring programmes was discussed. Lafaurie *et al.* presented results on the application of a range of biochemical markers in the assessment of environmental quality of sediments, water and

biota in the Northern Mediterranean. This work illustrated the need for use of a range of biochemical markers for the evaluation of biological impact of complex mixtures of contaminants in the field.

Arinç and Şen presented results of a laboratory investigation on the induction of liver 7-ethoxyresorufin-O-deethylase (EROD) activity by exposure of fish to polycyclic hydrocarbons, while Pavičić *et al.* assessed the use of methallothionein-like proteins as metal pollution indicators in mussels. Kurelec proposed a multi-xenobiotic resistance mechanism in mussels and sponges, similar to the mechanism of multi-drug resistance found in tumor cells, supporting his proposals by a number of laboratory investigations. Krajnović-Ozretić and Ozretić investigated the use of liver toxicity biomarkers, plasma proteins and haematological parameters in assessing exposure of fish to contaminants, while Bolognesi *et al.* assessed the use of genotoxicity indicators in mussels exposed to contaminated sites from the Ligurian Sea.

Chassard-Bouchaud illustrated the use of micro-analytical techniques in the cellular and sub-cellular analysis of the localization and damage induced by uranium, and other radionuclides in three bivalves. Galdies and Axiak presented results of bio-kinetic studies of lead in another bivalve at the cellular and sub-cellular levels.

The second half of the session was devoted to whole-organismic responses to marine contamination. Dr. John Widdows, in his key-note paper, gave a review of the attributes of physiological energetic responses and their role in biological monitoring programmes. Investigations on the applicability of larval and developmental bioassays were presented by Quiniou and Toularastel on bivalves, and by Pagano *et al.* on echinoderms, respectively. Baldi and Pepi presented a review of chromium toxicity to a range of microorganisms, including tolerance phenomena.

Three papers on phytoplanktonic responses to contaminants as measured in the laboratory were presented. These included: effects of exposure to copper and mercury on growth rates in phytoflagellates (Gotsis-Skretas and Christaki); on diatoms by exposure to the herbicides, 2,4-D and trifluralin (Ünsal); and on other unicellular algal species on exposure to lindane (Peñalva and Fernandez).

Verriopoulos presented results of a laboratory investigation of the sublethal effects of zinc, chromium and copper on the survival, fertility, feeding and respiration rates of two copepods, while El-Komi and Tayel provided lethal toxicity data on exposure to organophosphorus pesticides of adult and larval stages of barnacles. Abdel-Moati reported on laboratory and *in situ* investigations on the accumulation of mercury and lead by mussels as well as

on the effects on protein and carbohydrate body contents and on the filtration rates of these test species.

Two papers reported on toxicological investigations carried out on cultured fish. Papoutsoglou and Tziha showed how low oxygen levels, low pH levels and high total ammonia levels which may result from organic pollution affected the blood chemistry and the body chemical composition of sea bream and sea bass, while Glamuzina investigated the effects of water soluble fractions of crude oil on the eggs, larvae and postlarvae of these same species. All the papers presented appear in full, in Annex IV of the report.

7. The applicability of the techniques in field studies on a routine basis and their significance and interpretation vis-a-vis marine pollution risk assessment (Agenda item 7)

In opening the discussion section on the use of ecotoxicological tests for the regulation and control of marine pollution, Mr. Lloyd stressed the need to be clear about the definition of pollution. The GESAMP definition refers to the protection of marine resources from harm; this is an anthropocentric definition which focuses on the need for man's use of the marine resources to be protected. A second use of the term pollution is the presence of man-made chemicals, or man-made increases in natural chemicals, in the marine environment. This definition is based on the fact that all chemicals, at whatever concentration, will have some effect (even though imperceptible at very low concentrations) on marine organisms; it is therefore an ecocentric view. Whether water is polluted or not depends on the sensitivity of chemical and biological analysis to measure the presence of such chemicals.

Pollution control measures reflect to some extent the definition of pollution used. Protection of a resource can be achieved by setting water quality standards or selective controls applied to chemical products. Such procedures require good ecotoxicological concentration/response data for use in hazard and risk assessments. Removal of man-made chemicals from the environment can be achieved by using the best available technology (within economic feasibility) for waste treatment and by the control of chemical products. These procedures can be used independently of ecotoxicological hazard and risk assessments and they satisfy the need to reduce all chemical inputs as far as possible. It may not be a cost-effective approach.

Ecotoxicological hazard and risk assessments require data on concentration/effect relationships. These can be used to classify the hazardous properties of a chemical, to provide environmental quality standards for substances that can occur in the environment at potentially harmful concentrations, in the evaluation of biological monitoring procedures, and for bioassays.

Bioassays use measurements of biological effects to indicate the presence of chemicals in the water at significant concentrations. The main use of such techniques is to measure the concentrations of substances for which chemical analysis is insufficiently sensitive, and to indicate the presence of chemicals which are not included in chemical monitoring programmes. This requires that the tests should have a high degree of reproducibility and precision. Bioassays should not attempt to duplicate or replace chemical monitoring but should be integrated into such programmes. Results of such bioassays cannot be used alone to predict the risk of damage to living aquatic resources.

For those substances which are shown to be present or have the potential to be present in the marine environment, specific tests to derive concentrations/response relationships were required to make risk assessments and, where necessary, establish water quality standards to give adequate protection to marine communities. It is essential that these toxicological tests should incorporate a proper control of the chemical conditions of exposure; the concentrations should be kept as constant as possible, the chemical state of the substance should be identified, and the exposure conditions should be relevant to those encountered in the environment. Lack of such control destroys the usefulness of the data for hazard and risk assessments, and therefore for pollution control. There is a better need for harmful effects to be related to tissue concentrations, and the state of these levels should be identified, i.e. whether they are in stores of fat (lipophilic substances) or in inert granules (metals).

The effects that are measured should be capable of extrapolation to predict the risk of harm to aquatic communities. This cannot be done for biochemical effects but is possible for measurements of growth potential and embryonic development. The ability to obtain good concentration/response data diminishes with tests on higher levels of biological organisation i.e. on mesocosms containing complex communities.

Finally, ecotoxicological risk assessments should be incorporated into economic cost-benefit analyses in order to obtain a rational regulation of pollution that takes priorities into account. There is a need for a better education of the public in the scientific assessment of pollution risk in order to influence their risk perception and so contribute to rational political decisions. Disagreement among the scientific community as to what constitutes a risk has led to a general lack of confidence in ecotoxicological predictions.

After the presentation of Mr. Lloyd, the discussion concentrated on the suitability of biological effects techniques for marine pollution assessment purposes stressing their limitations. The main limitation identified was the fact

that some of these techniques are undertaken under unrealistic experimental conditions and subsequent claims of the importance of the derived information may not be justifiable.

Many of these techniques are applicable only to certain species and to a small group of chemicals and therefore the physiological background of each species should be known before undertaking any toxicological experiments.

The idea was put forward that these techniques could be used to identify "hot-spot" areas where chemical monitoring should be initiated.

As the process of evaluation is still going on, intercalibration exercises are needed to ensure improved quality and comparability of results. Practical workshops taking place elsewhere could also be repeated in the Mediterranean region.

It was pointed out that the suggestions of committees entrusted with the recommendation of specific procedures to be used in monitoring programmes may be biased by the personal views of the individual committee members and such subjective judgement should be thoroughly discouraged.

Ecotoxicological risk assessment should also be incorporated into more general approaches such as control management and environmental policy analysis.

The view was expressed that more effort should be devoted to useful monitoring which could help in making proper decisions for pollution control purposes. The views of decision-makers are often influenced by public opinion pressure groups and, as a result, certain decisions taken are based on emotion and perception rather than on actual scientific facts.

8. Future research needs (Agenda item 8)

The Workshop recognized an urgent need for marine research in environmental toxicology to be directed more specifically to the solution of perceived pollution problems. In particular, the role of ecotoxicology in the monitoring, regulation and control of pollution has potentially enormous benefits in optimising the allocation of scarce resources. Unless, however, agreement is reached on a number of issues, it is likely that national and international agencies will move as a result of public pressure towards pollution control strategies based wholly upon best technological means and/or control of pollution at source. While such strategies will result in some environmental improvement, they are not cost-effective, and therefore not an optimal solution

for an economically deprived area such as the Mediterranean.

In order to make the most rapid progress towards routine applications, the choice of techniques (test species, experimental and monitoring procedure) should be based on a few selected approaches which are well-established and documented. A major research activity should therefore be to develop and apply these techniques within the Mediterranean, bearing in mind both the oceanographic and the biotic peculiarities of this area as well as laboratory organization and availability of resources.

8.1 Toxicity testing procedures

No single test procedure can be appropriate for all circumstances. It is therefore necessary to establish a repertoire of procedures which would meet the requirements of the various pollution situations which exist. In particular, the Workshop concentrated on a range of biochemical and physiological approaches. Some of these are reasonably well established and are ready for universal application in the short-term. These may include:

a) Molecular biomarkers, such as:

- mixed-function oxidase (MFO) especially 7-ethoxy-resorufin-O-deethylase (EROD);
- acetylcholinesterase (AChE);
- metallothioneins (MTh);
- DNA alterations.

b) Toxicity testing on early life stages, including:

- fertilization toxicity (both in vitro and in vivo), and
- developmental toxicity (embryo/larval stages in bivalves, echinoids and fish).

c) Bioenergetics (Scope For Growth) in mussels (or other species where appropriate).

d) Cytogenetic testing, such as:

- micronucleus induction, and

- cytogenetic analysis (primarily in mussels and sea urchins).

Where these techniques are used as bioassays and in biomonitoring procedures, they should be fully integrated into chemical monitoring programmes, so that they can identify the occurrence of unknown chemicals present at biologically significant concentrations in the marine environment. The limitations, as well as the value, of these tests should be recognized; these should include evaluation of the reproducibility and precision of the results obtained.

Any additional set of procedures appearing to be of interest at the present time cannot be recommended for universal application. They may,

however, be worth investigating in a few laboratories which have specific expertise, or may be susceptible of further development until their routine utilization may be recommended.

These approaches include, e.g.:

- a) the cellular/subcellular distribution of pollutants;
- b) the multi-drug resistance (MDR) system, as a possible indicator of adaptation to a polluted environment;
- c) the occurrence of neoplastic and preneoplastic lesions in fish populations (preferably non-migratory, benthic species);
- d) the use of remote sensing as a monitoring tool.
- e) toxicity testing techniques designed to investigate the effects of pollutants on primary producer organisms, particularly if they are sufficiently sensitive to be ecologically relevant

The Workshop also recognized that studies carried out within the framework of aquaculture programmes may provide useful data within an ecotoxicological context.

8.2 Choice of testing conditions and substrates

It must be emphasized that environmentally relevant conditions (e.g. concentration, route of administration and duration of experiment) should be invariably adopted in toxicity testing. There remains scope for the use of "classical" toxicological techniques such as determinations of LC 50 or EC 50 using a variety of organisms such as phytoplankton, zooplankton, polychaetes,

etc., provided that the purposes of the experiments are clearly defined and their limitations are clearly understood. In particular, the use of lethal toxicity tests or other tests involving the exposure of organisms or cells to high concentrations of toxicants for short periods of time may be viewed as a part of a more comprehensive risk assessment for regulatory purposes.

By utilizing environmentally realistic levels of toxicants, investigators should be aware, however, that in a few cases they may encounter a positive stimulation of the biological event being considered (e.g. growth, fertilization, etc.). This phenomenon is currently termed "hormesis" which has been reported extensively in the literature. If this phenomenon may be confirmed in the environment, hormesis might reflect potential alterations at the community level.

As for substrate choice in toxicity testing, in-field investigations may either focus on water or sediment toxicity, or both. Since it is recognized that marine contaminants of whatever class are rapidly deposited in sediments, it is essential that techniques be developed for studying the response of benthic organisms to contaminated sediments. On the other hand, the measurements of effects in the water column is made difficult by the low concentrations of chemicals present; this could be overcome if techniques to concentrate these substances (eg. absorption/elution) could be developed so that tests can be made on the concentrates. As for the utilization of sediment samples, it is currently controversial whether sediment toxicity should be tested on either solid phase or water extracts (elutriate), or both. Efforts should be made to determine the most appropriate techniques for studying sediment toxicity.

9. Recommendations (Agenda item 9)

The Workshop made the following recommendations:

- a) Ecotoxicological effect measurements in the Mediterranean should be directed more specifically to the solution or amelioration of existing pollution problems.
- b) Toxicity testing procedures should be developed and applied based on (i) existing knowledge and techniques directed primarily towards monitoring and regulation of pollution and (ii) the specific oceanographic and biotic conditions in the Mediterranean.
- c) Those techniques and approaches which are listed in section 8.1 should be promoted and applied as widely as possible. It is recommended that further aspects of ecotoxicological studies (such as responses at community level and multispecies toxicity tests) will be discussed and assessed in other further international meetings.

- d) The proper choice, control and description of the chemical conditions of exposure within toxicity tests and the integration of chemical and biological monitoring programmes need to be given a much greater emphasis.
- e) There remains a continuing need for training in specific techniques and in the development of relevant research strategies. It is recommended that a workshop is organized as early as possible to train Mediterranean scientists in a number of biological monitoring techniques.
- f) The Workshop recommends the establishment of a working group which will formulate a pilot biomonitoring exercise for implementation by selected Mediterranean institutions in their respective areas. The purpose will be to evaluate the environmental impact of pollution sources and to correlate the results with chemical monitoring data. Moreover, this group recommended necessary training and intercalibration exercises.
- g) MED POL should increase its support to enhance communication and collaboration between laboratories especially between those in developed and developing countries.
- h) MED POL should seek ways to initiate informal co-operation with laboratories in the Black Sea countries.

10. Any other matter (Agenda item 10)

The publication of the proceedings was brought up under this agenda item and the Technical Secretary explained that these would be published in the MAP Technical Reports Series unless otherwise decided by the Workshop.

After an extensive discussion, it was decided that the best solution would be to publish all the papers presented at the Workshop in the MAP Technical Reports Series. However, before publication, the papers should be reviewed by other participants of the Workshop. If the authors would not accept any of the comments of the referee, then this comment, together with the author's reply, would appear at the bottom of the paper. In the meantime, participants were also encouraged to publish their papers in international scientific journals mentioning that the paper was presented at the FAO/UNEP/IOC Workshop on the Biological Effects of Pollutants on Marine Organisms (Malta, 10-14 September 1991).

Authors wishing to submit a revised version of their paper for the review, should do so as soon as possible but not later than the end of October 1991.

11. Adoption of the report (Agenda item 11)

The present report was adopted by the Workshop on Saturday, 14 September 1991.

12. Closure of the meeting (Agenda item 12)

The Hon. Minister of Education and Interior Dr. Ugo Misfud Bonnici, responsible for environment, stressed, in his speech, the importance of environmental protection in man's development and well-being and expressed his confidence that the deliberations of the workshop would be useful in formulating control measures for the protection of the Mediterranean sea.

The Technical Secretary, in his closing remarks, expressed satisfaction for the results of the workshop and thanked the participants for the constructive spirit in which it was conducted. He also thanked the officers of the workshop, the guest-speakers and everybody else who contributed directly or indirectly to the success of the workshop. The work of the Director and staff of the Euro-Mediterranean Centre on Marine Contamination Hazards was gratefully acknowledged and appreciation was expressed for the warm hospitality.

After the usual exchange of courtesies the Chairperson closed the workshop .

ANNEX I

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ANNEX II

AGENDA OF THE MEETING

1. Opening of the meeting
2. Background and scope
3. Election of officers
4. Adoption of the agenda
5. Organization of the work
6. Presentation of papers
7. The applicability of the techniques in field studies on a routine basis and their significance and interpretation vis-à-vis marine pollution risk assessment
8. Future research needs
9. Recommendations
10. Any other matter
11. Adoption of the report
12. Closure of the meeting

ANNEX III

LIST OF PAPERS PRESENTED

Key-note papers

1. Biochemical techniques for the assessment of chemical pollutant exposure in the marine environment, by **K.W. RENTON**
2. Role of physiological energetics in ecotoxicology and environmental pollution monitoring, by **J. WIDDOWS**

Other papers

3. Biochemical and physiological responses of Mytilus edulis to Hg and Pb in the coastal waters of the Alexandria region, by **M. ABDEL-MOATI**
4. Induction of liver 7-ethoxyresorufin O-deethylase in gilthead seabream by benzo(a)pyrene and its potential use in biochemical monitoring of environmental pollutants, by **E. ARINÇ and A. ŞEN**
5. Microbial response to Chromium toxicity, by **F. BALDI and M. PEPI**
6. Carcinogenic and mutagenic pollutants: impact on marine organisms, by **C. BOLOGNESI, M. PARRINI, P. ROGGIERI, C. ERCOLINI and C. PELLEGRINO**
7. Biological effects of uranium and transuranium nuclides on the marine bivalves, Mytilus edulis, Crassostrea gigas, and Cerastoderma edule; Microanalysis at the cellular and subcellular levels, by **C. CHASSARD-BOUCHAUD**
8. Influence of organophosphorus herbicides on barnacles (Crustacea: Cirripedia), by **M. EL-KOMI and F. TAYEL**
9. The fate of lead in a benthic bivalve, by **C. GALDIES and V. AXIAK**
10. The early fish stages as an object of toxicological studies; case of sea bass, Dicentrarchus labrax, and gilthead sea bream, Sparus aurata, by **B. GLAMUZINA**
11. Physiological responses of two marine phytoplanktonic species to heavy metal pollution, by **O. GOTSIS-SKRETAS and U. CHRISTAKI**

12. Detection and evaluation of hepatic intoxication in fish, by **M. KRAJNOVIĆ-OZRETIĆ** and **B. OZRETIĆ**
13. The multi-xenobiotic resistance mechanism in aquatic organisms, by **B. KURELEC**
14. Biochemical markers in pollution assessment: field studies at the North coast of the Mediterranean sea, by **M. LAFAURIE, A. MATHIEU, J-P. SALAUN, J-F. NARBONNE, F. GALGANI, M. ROMEO, J-L. MONOD** and **PH. GARRIGUES**
15. Sublethal toxicity testing in sea urchin fertilization and embryogenesis: a study of polluted water and sediment from two rivers in Campania, Italy, by **G. PAGANO, P.A. DINNEL, A. ESPOSITO, M. GUIDA, G. MELLUSO** and **N.M. TRIEFF**
16. Effects of water pollution, caused by organic material, on the physiology and body chemical composition of sea bream (*Sparus aurata*) and sea bass (*Dicentrarchus labrax*), by **S. PAPOUTSOGLU** and **G. TZIHA**
17. Metal binding proteins of *Mytilus galloprovincialis*, similar to metallothioneins, as a potential indicator of metal pollution, by **J. PAVIČIĆ, B. RASPOR** and **M. BRANICA**
18. Lindane effects on the growth, size and composition of two marine unicellular algae, by **S. PEÑALVA** and **F. FERNANDEZ**
19. Biological effects of contaminated water tested by marine bivalve embryo-biassay, by **F. QUINIOU** and **F. TOULARASTEL**
20. Effects of herbicides on the growth of marine phytoplankton, by **M. ÚNSAL**
21. Effects of sublethal concentrations of zinc, chromium and copper on the marine copepods *Tisbe holothuriae* and *Acartia clausi*, by **G. VERRIOPOULOS**

ANNEX IV

PAPERS PRESENTED AT THE WORKSHOP

