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Biological Variables, Especially Skeletal Deformities in Fish, for Monitoring Marine Pollution [and Discussion]

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Phil. Trans. R. Soc. Lond. B 1979 **286**, 457-464
doi: 10.1098/rstb.1979.0040

Phil. Trans. R. Soc. Lond. B. **286**, 457–464 (1979)
Printed in Great Britain

Biological variables, especially skeletal deformities in fish, for monitoring marine pollution

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When considering principles for selection of indicators, i.e. biological variables, for monitoring marine pollution, it must be regarded as important to search for effects on the highest possible level of organization. For a global monitoring programme there are, however, many practical limitations in the number of useful indicators. The paper suggests skeletal deformities in fish as one possible indicator for this purpose in the future and gives a review on the occurrence, effects, causative factors and possible mechanisms of skeletal deformities in fish.

Introduction

In 1976, at the 8th Session of Gesamp (Group of experts on the scientific aspects of marine pollution), Unesco expressed its interest in forming a working group to reexamine the feasibility of monitoring biological variables of marine pollution. Background information was presented at the 9th Session of Gesamp, which was held in New York in 1977.

At the 9th Session of Gesamp, it was recommended that such a working group should be set up, and the following terms of reference were submitted to be considered by the group:

(*a*) to formulate a rationale for monitoring biological variables, taking into account existing and planned regional and global monitoring programmes, paying particular attention to those which include monitoring of biological variables;

(*b*) to determine the scientific requirements for monitoring relevant biological variables;

(*c*) to assess the feasibility of establishing practical procedures for the monitoring of biological variables related to marine pollution.

At its first meeting, at Aberdeen in December 1977, the working group concluded that there had been several previous working groups that had been working with the same, or similar problems. Two of those were S.C.O.R. working group 29 (Continuous Monitoring in Biological Oceanography; established 1968, reported 1973), and I.C.E.S. Subgroup on the Feasibility of Effects Monitoring (established 1975, reported 1978). This led to a discussion of whether or not the new group would be able to contribute something new about this problem. It was agreed, however, that the members of the group, by intersessional work, should undertake a preparation of documents for the second meeting and give comments upon different kinds of possible indicators. The outcome of this work would then be presented at Gesamp 10. The present document is a part of this work, substantially dealing with skeletal deformities in fish as a possible indicator of marine pollution.

Principles for selection of indicators

In our ambition to protect the marine environment, the strategy must be to search for the effects on the highest possible level of organization. It would thus be advantageous to work with ecosystems, communities, or, at least, populations. Effects on individuals, organs, tissues, cells and organelles would therefore be of a lower priority in our search for useful methods. One of the major problems, however, is the fact that the natural biological background against which the responses to human influences have to be detected is far from stable. Populations, communities and ecosystems change in various ways, directionally, cyclically and on time scales that may range from hours to centuries. On the community level, for instance, the length of time required by an assemblage of a population to complete a response may be very long. Some commercially important aquatic species may take 25 or more years to complete their lives, and five or more years to reach harvestable size. Computer simulations of the behaviour of populations have shown that time lags of 10 or more years are required before fisheries might expect to feel the beneficial effects of a change in management policies (Hackney & Minns 1974).

On a more immediately relevant level, many cyclical changes are responses to 'normal' levels of abiotic variations that have diurnal, circadian, lunar, seasonal or annual periodicities. Such biological changes constitute 'background noise' that complicates the analysis of eventual anthropogenic effects on aquatic life. But virtually all abiotic factors that influence aquatic life also fluctuate with a degree of irregularity that poses important problems. Consequently, no biological measure that we wish to use will behave as a 'baseline', which would be desirable. We can realize that such a concept is an oversimplification of the problem.

Pollution can usefully be regarded as simply another environmental variable which may induce, especially on higher levels of organization, effects that are difficult to distinguish from those caused by natural stressors. However, the identification of situations where effects occur as 'hot spots' would allow the location of more detailed studies including experimental programmes designed to investigate causative agents (I.C.E.S. 1978).

Most toxic substances exert their effects on a basic level in the organism by reacting with enzymes or by affecting membranes and other functional components of the cells. Biochemical and physiological techniques are commonly used in laboratories to measure such effects and together with histological, histochemical and haematological studies they can contribute most fruitfully to reveal the toxic mechanism of a single or a group of substances. However, the interpretation can often be hard to make, as observed effects may, for instance, reflect a general adaptation syndrome, rather than damage to a particular organ or function. Furthermore, effects at these levels of organization are also less useful in predicting how the whole animal will manage in a complex field situation, i.e. for an evaluation of whether the observed symptoms are ecologically significant.

As far as possible, the search for effects should therefore be concentrated on irreversible damage that could be judged to cause significant impairment in the animal's fitness to survive. An ideal approach, when trying to find a good biological variable to detect marine pollution, can be described in the following way. An irreversible, easily detectable, effect on the highest possible level of organization in the system should be used. The effect should, at least in the first step, be expressed in clearly contrasting forms, and ideally in binary (yes–no) form. It is also important to take into account that this kind of monitoring should be feasible also for

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

developing countries, lacking highly trained laboratory personnel and sophisticated equipment.

With this in mind, and facing the fact that our knowledge is unsatisfactory within most of the areas that could be applicable to our purpose, there are very few variables left to discuss, and this is of course also one of the reasons that biological monitoring for pollution is so rare.

For the purpose of listing feasible monitoring variables for studies in the Oslo Commission areas and the north west Atlantic, a document was prepared by an I.C.E.S. working group which reported in 1978 (I.C.E.S. 1978). The document discussed selected recent literature on the effects of pollutants on marine organisms under six headings: biochemical, morphological, physiological, behavioural, population/community and genetic. It also examined the ways in which these effects are measured experimentally, including the use of bioassay procedures. It was suggested that five topics would be useful and applicable to a pollution effects monitoring programme. The topics were: (1) liver/somatic and gonad/somatic index; (2) skeletal deformities; (3) tumours, lesions, etc.; (4) gill damage; (5) general observations on morphology.

In the search for biological variables to be used in a global monitoring programme, the I.C.E.S. document is quite useful. When extending the ambition from designing a local programme to a global one, there will, however, be practical limitations in the number of useful topics, or in the way they can be used. This is so mainly because of the fact that a global programme must be adapted to the practical possibilities of developing countries. It was thus suggested at Gesamp 9 that a 'simple form' should be attained. This, together with the above mentioned prerequisities (irreversible damage, expressed in binary form, etc.) drew the author's attention to skeletal deformities in fish as having some evident advantages, compared with other possible variables. In what follows, other variables are therefore omitted and priority is given to a discussion on occurrence, etiology and other relevant aspects of skeletal deformities. It is worth mentioning, however, that many other variables might be regarded as equally or even more useful in the future.

Occurrence of skeletal deformities

Pugheadness, i.e. abnormal formation of the skull, and spinal and vertebral anomalies, is often evident by an external inspection of the fish or is revealed with the aid of dissection or radiography. The investigations can be performed on material in a variety of conditions (for instance, on fish from commercial catches, routine samples, frozen material or material preserved in neutral preservatives).

The deformities occur in varying degrees of severity and in spinal and vertebral deformities they may manifest themselves as dorso-ventral flexures (Rosenthal & Rosenthal 1950) or body humps, termed lordosis or a 'humpbacked condition' (Hickey 1972). Lateral flexures and abnormal backward spinal curvature are termed scoliosis and kyphosis respectively. Such damages are reported in great numbers from marine, brackish and freshwater areas by many authors, too many to be listed here. Single reports on vertebral and spinal abnormalities are very old, and some recent papers describe comparatively high frequencies of damage in wild populations of fish. The frequency and mode of occurrence have, in many cases, given rise to the suspicion of a connection with water pollution (see, for example, Wunder 1975; van de Kamp 1977).

Gill & Fisk (1966) reported a frequency of 9% of vertebral anomalies in a population of sockeye salmon (*Oncorhynchus nerka*) and Orska (1962) found 11% in pike (*Esox lucius*).

Van de Kamp (1977) gave figures for herring (*Clupea harengus*), caught around the British Isles during 1950 to 1973, showing a mean frequency of 1.3–3.8 %. During one single year, as much as 8.4 % of the herrings in a sample from the southern North Sea demonstrated vertebral deformities. Wunder (1975) reported 6 % spinal deformities (toxic osteosclerosis) in a population of whitefish (Coregonidae) from the Bodensee.

Effects on the individual

Different possible effects on the individual due to spinal or vertebral damage are described in the literature: (*a*) impaired swimming performance (Poston 1967; Patten 1968; Kroger & Guthrie 1971; Bengtsson 1974*a*; Weis & Weis 1976); (*b*) decreased ability to escape (Kroger & Guthrie 1971); (*c*) hindrance in catching food (Kroger & Guthrie 1971). Further effects were also suggested by Hickey (1972), such as decreased capability for territorial defence, reduced ability to compete for a sexual partner, and weakness due to associated physiological conditions, which can be detrimental during periods of stress. Bengtsson (1974*b*) found vertebral damage to cause effects on adjacent neural tissue in minnows (*Phoxinus phoxinus*).

Causative factors

There are a great many observations made in the laboratory that point to a number of different causative factors, some of which are related to water pollution. The list in table 1 is partly based on a review by Bengtsson (1975*a*).

A number of toxic substances are also reported to cause vertebral or spinal deformations in fish. Some of these are shown in table 2.

Table 1

causative factor	references
hereditary factors	Rosenthal & Rosenthal (1950), Gordon (1954), Takeushi (1966)
defective embryonic development	Templeman (1965), Dahlberg (1970), Sinderman (1970)
unsuitable water temperature	Seymor (1959), Brungs (1971), John (1959), Hubbs (1959), Milton (1971), Lee & Williams (1970)
low levels of dissolved oxygen	Blaxter (1969), Turner & Farley (1971)
radiation (X- and u.v.-irradiation)	Solberg (1938), Hinricks (1925)
dietary vitamin deficiencies	Kitamura *et al.* (1965), Poston (1967), Halver *et al.* (1969), Wilson & Poe (1973)
parasitic infection	Hoffman *et al.* (1962), Reichenbach-Klinke & Elkan (1965)
electric current	Hauck (1949), Spencer (1967)
trauma	Blaxter (1969)

Table 2

causative substance	references
chlorinated hydrocarbons (toxaphene, DDT, Kepone)	Mehrle & Mayer (1975*a*), Smith & Cole (1973), Couch *et al.* (1977), Hansen *et al.* (1977)
organophosphorus pesticides (e.g. malathion, parathion, phosalone, demeton)	Meyer (1966), McCann & Jasper (1972), Weis & Weis (1976)
dispersed crude oil	Lindén (1976)
heavy metals (Zn, Cd, Pb)	Bengtsson (1974*b*, 1975*a*), Bengtsson *et al.* (1975), Nakamura (1974), Koyama & Itazawa (1977), Davies & Everhart (1973)

Possible mechanisms

Spinal deformations may occur at different developmental stages, but it is probable that in field populations most of the damage is done during the egg stage or immediately afterwards. Some hypotheses have been stated on the possible mechanisms caused by pollutants in adult fish. Bengtsson (1974*b*, 1975*b*) suggested that vertebral damage caused by zinc might be attributed to the effect of zinc on the muscle action potential and thus to an effect on the neuromuscular system. A similar action of cadmium was also suggested by Bengtsson *et al.* (1975). The theory was further developed by Larsson (1975), who noticed ionic imbalances, hypocalcaemia and hypokalaemia in plasma from fish exposed to cadmium. A possible decalcification of the skeletal bone was also suspected since such an effect (renal osteomalacia) is well documented in connection with chronic cadmium poisoning in mammals (e.g. the Itai-itai disease in man). Thus, a possible weakness of the bone tissue, together with neuromuscular effects, caused by hypocalcaemia and/or hypokalaemia, might explain the occurrence of skeletal damage in cadmium-exposed fish. This theory was further supported by Koyama & Itazawa (1977), from experiments with fish given oral doses of cadmium.

Mehrle & Mayer (1975*a*) reported that toxaphene might cause a functional decrease of vitamin C, affecting the collagen synthesis of the skeleton in fathead minnows (*Pimephales promelas*). This hypothesis was also supported in experiments with brook trout (*Salvelinus fontinalis*), which resulted in a decreased concentration of collagen after treatment with toxaphene (Mehrle & Mayer 1975*b*). There are thus good experimental indications of a connection between spinal and vertebral deformations in fish and water pollution. So far it has not been possible, however, to tie observed high frequencies of spinal and vertebral deformities in wild populations of fish to water pollution. In a paper by van de Kamp (1977), which evaluates the usefulness of vertebral deformities in herring for a pollution monitoring programme, it is concluded that the highest percentages of deformities were found in areas which probably had the highest degree of pollution. In these areas, the percentage of deformities also showed a slight increase over a number of years.

Recommendations

It may be asked whether we need to wait for more experimental work to be performed within this field before a monitoring programme could be started. There is no doubt that much further experimental research on the aetiology of vertebral damage is needed before we can analyse the causal relation between pollution and deformities to a desirable extent. The start of a monitoring programme would, however, besides identifying 'hot spots' with unusually high frequencies of damage, also initiate further experimental research.

In monitoring for effects, there is a need to document the variability of these effects in natural populations, for example by using a number of measurement sites to establish the natural range of the factors studied. Sites should be chosen to represent the observed or predicted gradients of contamination, and the greater the number of sites, the better the possibility of distinguishing natural variability (the 'noise') from real pollution effects (the 'signal') (I.C.E.S. 1978).

On this level, it is not possible to establish practical procedures for a monitoring programme on skeletal deformities in fish. It is suggested, however, that the topic is worthy of urgent consideration and that a simple form of it should be linked to already existing monitoring programmes. For this purpose, it is also necessary to discuss and give recommendations on suitable indicator organisms. Such an organism must meet several prerequisites, including wide geographic distribution, abundance, absence of extensive migratory patterns, easy availability and survival in polluted waters. It is possible that the best choice would be a species belonging to the clupeid family of fish. Such suggestions, on choice of species, complementary measures and other practical procedures should, however, be critically assessed in respect of their value, advantages and shortcomings.

It can be concluded that spinal or vertebral deformities seem to meet the majority of the prerequisites mentioned above. Of particular importance is that it seems possible to apply the technique in the existing practical situation in developing countries and that the answer might, as a first step, be expressed in a simple binary (yes–no) form.

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PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES

Discussion

T. L. Coombs (*Institute of Marine Biochemistry*, *Aberdeen*, *U.K.*). Could the speaker expand on the time scale of skeletal effects: are they immediate or do they occur after a long period, such as several years?

B.-E. Bengtsson. It is not possible to give one single answer to this question. Depending on the stage of development of the fish and which causative factor is working, the time scale might be very different. With electric current, for instance, the damage might occur within a few seconds. For some substances (some organophosphorus pesticides and chlorinated hydrocarbons) the damage might occur within a few hours or a few days (e.g. with zinc). Other causative factors might take a much longer time, perhaps years, before vertebral damage is manifested. It is also important to note that fish embryos or fry are usually very sensitive and may demonstrate vertebral damage or spinal curvature after a few hours of exposure to unfavourable conditions.

M. Waldichuk (*Pacific Environment Institute*, 4160 *Marine Drive*, *West Vancouver*, *B.C.*, *V7V* 1*N*6, *Canada*). How many fish never survive their vertebral deformities in early stages of life, and therefore distort the statistics on the distribution of fish with deformities when they are monitored?

B.-E. Bengtsson. In my own experiments, using zinc and cadmium on the minnow, I have never found vertebral damage to be a lethal factor *per se*. In the sea, however, it must be suspected that damaged fish are eliminated by predators, competition, etc. to such an extent that an underestimation of the frequency of damage might result. This is, however, not unique for this kind of damage but the risk is always there when it is required to monitor effects that cause significant impairment in the animal's fitness to survive.

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B BIOLOGICAL SCIENCES