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The influence of oil pollution on three copepods at Helsinki, Finland

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Some experiments were conducted in which three copepods: *Eurytemora hirundoides, Thermocyclops oithonoides* (planktonic species) and *Halectinosoma curticorne* (bottom dweller) were exposed to different kinds of oil, the oil (0.1 ml/l) being dropped into Petri dishes. *Thermocyclops* and *Halectinosoma* behaved in a different way to *Eurytemora* when exposed. *Eurytemora* individuals died much faster than *Thermocyclops* and *Halectinosoma,* and hence swam away from the oil in order to survive. *Eurytemora* individuals exposed to fuel oil fell into reversible narcosis, waking up when the oil slick was removed.

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1. Introduction

The most damaging component of oil and detergents is their soluble fraction, itself a mixture of hydrocarbons usually rich in low aromatics (Nelson-Smith 1972). Around the 1970s a lot of experiments were done testing the toxicity of oil to planktonic organisms. Concerning copepods, the most quoted are the investigations made by Mironov (1968, 1969), who found that concentrations of 0.1 ml oil/l sea water were catastrophic to *Acartia* and *Calanus.* McCauley (1966) had observed that *Gammarus* disappeared from an oil-polluted river, whereas *Cyclops* sp. survived. In fact, it has been observed that copepods can feed on 'tar balls' at the surface of the sea (Morris & Butler 1973) once the poisonous fractions have disappeared.

Some useful summaries of the effect of oil on plankton organisms have been given by Nelson-Smith (1970, 1972), Corner (1978) and Davenport (1982). This study is concerned with oil and its effect on three copepod species: *Eurytemora hirundoides* Nordqv., *Thermocyclops oithonoides* G.O. Sars ad *Halectinosoma curticorne* Boeck. The main emphasis is laid on the behaviour of the three species when exposed to oil.

2. Methods

The copepods were sampled at the bridge between Kuusisaari and Helsinki in May–July 1984 and 1985. The material was brought to the laboratory in thermoflasks and preserved, when needed, in a refrigerator at +6°C and at a salinity of 3‰.

For the experiments 10 ml Petri dishes were filled with sea water, 100 animals were introduced into each dish, and a concentration of 0.001 ml oil/dish was dropped on the surface of the water. Thus, a basic concentration of 0.1 ml oil/l water was used. The following types of experiments were carried out:

Control experiments

For all three species tests were run in order to check the reaction of the animals to the experimental situation. This was because one possible reason for the death of the animals is, namely, crowding.

Experiment 1

The escape movements of the three copepod species were tested in a test chamber constructed as reported in Gyllenberg & Lundqvist (1976).

The chamber was constructed from two microscope slides and a piece of silicon tubing bent to form the shorter sides and the bottom. Within the chamber the animals were able to move in two directions: horizontally and vertically.

After definite time periods the places of the animals in the test chamber were checked and recorded, and then compared to a random distribution in Table 1.

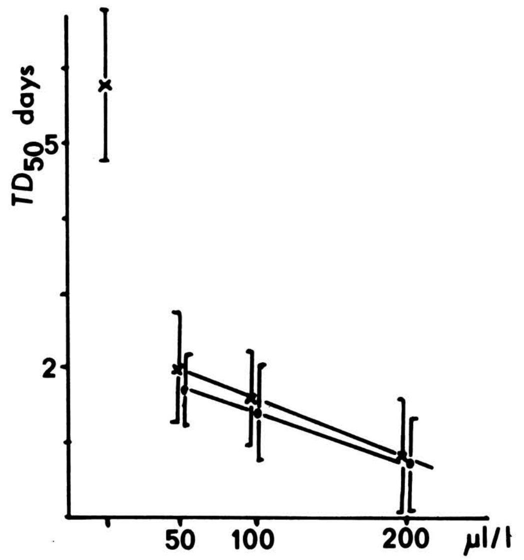

Fig. 1. *Eurytemora.* Rate of death of 50 per cent (TD_{50}) as a function of the concentration of oil (in µl oil/l sea water). Mean and standard deviation are given ($N=3$). Crosses = exposed to crude oil for 1 day, circles = exposed to diesel oil for 1 day.

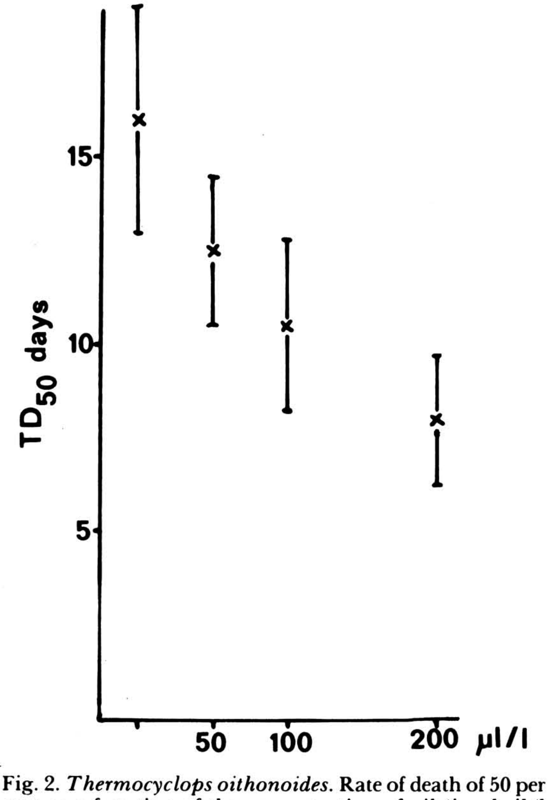

Fig. 2. *Thermocyclops oithonoides.* Rate of death of 50 per cent as a function of the concentration of oil (in µl oil/l sea water). Mean and standard deviation are given (N=6).

Experiment 2

Different species were tested for survival in different concentrations of crude oil. *Eurytemora hirundoides* was mainly used, but in some experiments *Thermocyclops oithonoides* and *Halectinosoma curticorne* were also tested in 0.1 ml oil/l sea water. Test values are found from the TD_{50} experiments.

Experiment 3

Different types of oil were used. In the main Neste crude oil (Russian oil, Ventspils) was employed, but in additon to this, different concentrations of Neste diesel oil (Winter quality) were used in testing the survival rate of *Eurytemora hirundoides.* Experiments with combustible (fuel) oil (Viscosity 230) proved to be unsatisfactory, since the animals revived after a while, and no definite TD_{50} could be measured.

3. Results

In Fig. 1 the rate of death of 50 per cent of *Eurytemora* was drawn as a function of concentration of oil µl/l sea water. It can be seen that *Eurytemora* normally perished after about 5–6 days when not exposed to any oil, but that survival was significantly shorter for individuals exposed to oil for 1 day ($P<0.01$**). The short survival period could be due to overcrowding. The TD_{50} for diesel oil was also somewhat lower than that for crude oil x, but not significantly so ($t = 0.57-0.20$ corresponding to $0.9>P>0.6$).

Fig. 2 and Fig. 3 give the rate of death of 50 per cent of *Thermocyclops* and *Halectinosoma* individuals. Both species survive longer in pure sea water, and therefore react differently to the experimental situation than *Eurytemora.* In some experiments with pure sea water the *Thermocyclops* individuals even reproduced, in spite of the absence of food. Therefore, in order to test the effect of oil pollution, TD_{50} for oil exposed species was divided by TD_{50} for the test situation in Fig. 4. It appears that *Eurytemora* die faster when exposed to oil ($P<0.05$*), but only when they cannot escape from the oil slick by swimming away (Table 1). This table reveals the fact that *Eurytemora* always swim away from the source of pollution, whereas *Cyclops* and *Halectinosoma* move around randomly.

The percentage response (from no. of dead animals out of 100) was transformed to probits according to the table given on p. 286 in Saunders & Fleming (1971). Thereafter the dose was transformed to log (concentration) and drawn as an x-axis. The corresponding

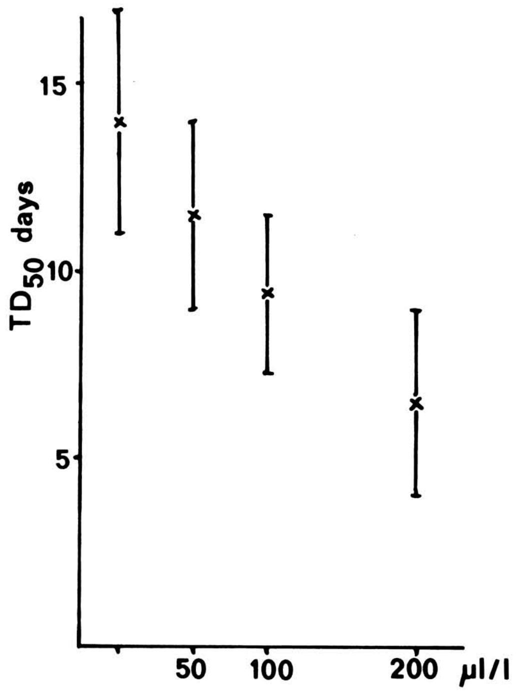

Fig. 3. *Halectinosoma curticorne.* Rate of death of 50 per cent as a function of the concentration of oil (in µl oil/l sea water). Mean and standard deviation are given ($N=6$).

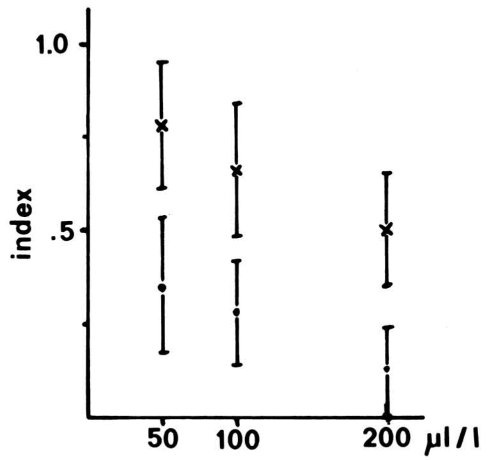

Fig. 4. TD_{50} (for oil exposed animals) divided by TD_{50} (test animals in pure sea water), as a function of the concentration of oil ($N=4$). *Thermocyclops oithonoides* = crosses, *Eurytemora hirundoides* = circles.

MLD was read from the intersection point of the x-value with a probit value of 5.0. The concentration was given as µl oil/l sea water.

The values obtained from the probit analyses were used to draw the curves in Fig. 5. *Eurytemora* (crosses) were significantly different from *Thermocyclops* and *Halectinosoma* ($P<0.01$**), whereas the curves for *Thermocyclops* and *Halectinosoma* were not significantly different ($0.6<P<0.3$). Thus it seems that *Thermocyclops* and *Halectinosoma* can endure doses about 100 times larger than those for *Eurytemora.* Even when the experiments were extended to 30 days about 10% of the *Thermocyclops* and *Halectinosoma* survived in 0.1 ml oil/l sea water.

In some experiments it was found that narcosis complicates tests with fuel oil, and these results lead to difficulties of interpretation in all experiments carried out with fuel oil.

4. Discussion

Apparently some differences exist in the responses of the three copepod species to oil. *Eurytemora* dies comparatively faster when

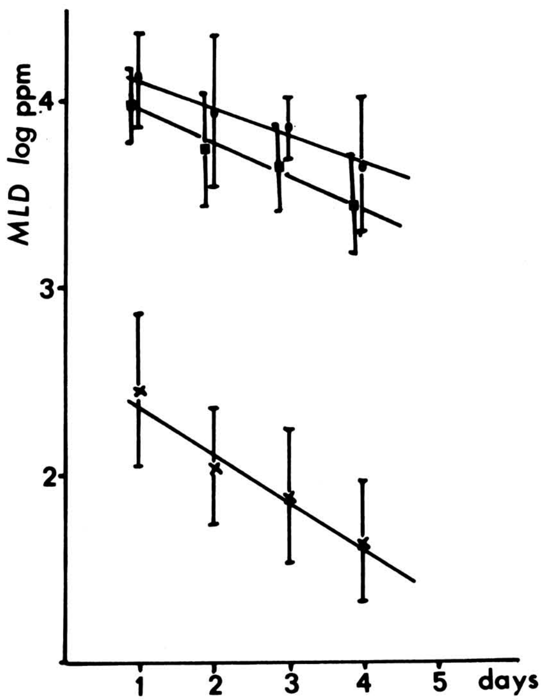

Fig. 5. MLD (in log ppm) as a function of days after the oil was dropped on the surface. Mean and standard deviation are given ($N=3$). The following species were tested: *Eurytemora hirundoides* (crosses), *Halectinosoma curticorne* (squares) and *Thermocyclops oithonoides* (circles). Time of exposure to oil 1 day.

Table 1. The distance (mm) reached by the test animals from the outlet point (P) of the crude oil, measured as the length of a vector in the polar coordinate system with P in origo. The empirical test values were compared (χ^2-analysis) with the expected frequency distribution (assuming random movement) for different frequency classes, and for the time periods indicated. Only the values for *E. hirundoides* after 20−120 min differ significantly from the expected. 30 animals were used in each experiment.

Distance from P	Distribution (min. after start of experiment) 0	5	20	60	120	240	600	Expected distribution
E. hirundoides								
1−14	7	3	1	1	0			8
15−22	11	10	11	9	7			9
29−42	6	10	10	10	13			7
43−56	6	7	8	10	10			6
T. oithonoides								
1−14	7	5	9	6	−	5	5	8
15−28	7	10	9	10	−	11	11	9
29−42	10	10	5	5	−	8	7	7
43−56	6	5	7	9	−	6	7	6
H. curticorne								
1−14	7	6	5	6	−	5	5	8
15−28	8	10	8	7	−	11	10	9
29−42	9	6	10	9	−	8	9	7
43−56	6	8	7	8	−	6	6	6

exposed to oil than both *Thermocyclops* and *Halectinosoma*. The death of *Eurytemora* does not depend solely on the effect of oil, since other factors such as over crowding in a small Petri dish, may also affect the mortality of *Eurytemora*. Nevertheless, Fig. 5 supports the idea that *Eurytemora* dies faster when exposed to oil than both *Thermocyclops* and *Halectinosoma*. The endurance of *Cyclops* to oil in the field has also been investigated by McCauley (1966), who found that *Cyclops* sp. in an oil-polluted river survived the pollution.

The strategy of *Eurytemora* is to avoid oil pollution by swimming away from the source, as is shown in Table 1. Gyllenberg & Lundqvist (1976) also found a distinct difference in the behaviour of the two species *Acartia* and *Thermocyclops* when an emulsifier was dropped in the upper corner of a test chamber. *Acartia* moved away from the source of pollution, whereas *Thermocyclops* swam around randomly in the chamber.

As fuel oil is tar-like in shape and probably does not contain so many soluble aromatics, the effect of this type of oil on animals is largely reversible. This is evident even after an exposure time of a day or more. On the other hand, crude oil or diesel oil always affected the animals in an irreversible way if removed after a day's exposure. Probably the effect is due to the aromatic soluble substances present in oil (Nelson-Smith 1970).

There are two ways oil and hydrocarbons can enter zooplankton: either by direct uptake from solution in sea water, or by assimilation from particulate diets (Corner 1978). At least where alkanes of higher chain lengths and some aromatics are concerned it is obvious that the latter way is more usual. Gyllenberg (1981) found that *Eurytemora hirundoides* actually eat ^{14}C-1-naphthalene when this is provided in seawater. Again Lee (1975) found that copepods rapidly took up ^{3}H-benzpyrene, ^{14}C-benzpyrene, ^{3}H-methylcholanthrene and ^{14}C-naphthalene from sea water. These hydrocarbons were metabolized by crustaceans to various hydroxylated and more polar metabolites.

Pure toxicity tests with copepods have not been extensively carried out. Mironov (1968, 1969) found that the copepods *Acartia* and *Calanus* and other zooplankton perished within 24 hours in 0.1 ml oil/sea water, and that concentrations of 0.1−0.001 ml/l had pronounced toxic effects on the larval stages of crabs and shrimps. It seems like 0.1 ml/l is the crucial concentration also for *Eurytemora*, although some individuals survived for several days. The set-up with Petri dishes containing 10 ml sea water also correspond to oil pollution near the sea surface, and as both *Acartia* and *Eurytemora* can swim away from the source of pollution, it seems likely they are never endangered by the light fractions of the oil.

Davenport (1982) is of the opinion that the concentrations used by research workers in performing toxicity tests are typical only of water in intimate contact with oil slicks. This factor stems partly from the wish of the experimentator to obtain 'positive' results, and it would seem desirable that far more studies should be performed in the future at a total hydrocarbon concentration of below 1 μg/g.

It is obvious that more work is needed, especially regarding the sublethal effects of oil. A few studies have demonstrated oil effects on the behaviour of planktonic organisms. Feeding tends to be depressed and food selection altered at hydrocarbon concentrations of around 250 ng/g hydrocarbons in the copepod *Acartia clausi* and *Acartia tonsa* (Berman & Heinle 1980). Blumer (1969) suggested that oil suppressed mate selection and escape responses as well. This latter statement is in accordance with the findings of Gyllenberg & Lundqvist (1976) and this paper, showing that the emulsifiers and oil may inhibit natural escape movements of *Thermocyclops oithoinoides* and *Halectinosoma curticorne*.

It thus seems that the copepods are able to absorb small amounts of the extremely damaging components of oil, namely the aromatic hydrocarbons. They thereafter undergo biosynthesis into other components, and these components become part of their lipid pool and are not necessarily concentrated in a specific organ (Blumer et al. 1970). The copepods are therefore predestined to become carriers of hydrocarbons when the sea is polluted with oil, and can carry the hydrocarbons with them to the next step in the food chain.

References

Berman, M. S. & Heinle, D. R. 1980: Modification of the feeding behaviour of marine copepods by sublethal concentrations of water accomodated fuel oil. — Mar. Biol. 56:59—64.

Blumer, M. 1969: Oil pollution of the ocean. — In: Hoult, D. P. (ed.), Oil on the sea, pp. 5—13. New York. Plenum Press.

Blumer, M., Souza, G. & Sass, J. 1970: Hydrocarbon pollution of edible shellfish by an oil spill. — Mar. Biol. 5:195—202.

Corner, E. D. S. 1978: Pollution studies with marine plankton. Part 1. Petroleum hydrocarbons and related compounds. — Adv. Mar. Biol. 15:289—380.

Davenport, J. 1982: Oil and planktonic ecosystems. — Phil. Trans. R. Soc. Lond. B 297:369—384.

Gyllenberg, G. 1981: Ingestion and turnover of oil and petroleum hydrocarbons by two planktonic copepods in the Gulf of Finland. — Ann. Zool. Fennici 18:225—228.

Gyllenberg, G. & Lundqvist, G. 1976: Some effects of emulsifier and oil on two copepod species. — Acta Zool. Fennica 148:1—24.

Lee, R. F. 1975: Fate of petroleum hydrocarbons in marine zooplankton. — Proc. 1975 Conf. Prevention Control Oil Poll. San Fransisco, California March 25—27: 549—553. American Petroleum Institute, Washington D.C.

McCauley, R. N. 1966: The biological effects of oil pollution in a river. — Limnol. Oceanogr. 11:475—486.

Mironov, O. G. 1968: Hydrocarbon pollution of the sea and its influence on marine organisms. — Helgoländer Wiss. Meeresunters. 17:335—339.

—"— 1969: The effect of oil pollution upon some representatives of the Black Sea zooplankton (in Russian with English summary). — Zool. Zh. 48:980—984.

Morris, B. F. & Butler, J. N. 1973: Petroleum residues in the Sargasso Sea and on Bermuda beaches. — Proc. Joint Conf. Prevention Control Oil spills. Washington D. C. March 13—15. 521—529. American Petroleum Institute, Washington D. C.

Nelson-Smith, A. 1970: The problem of oil pollution of the sea. — In: Russel, F. S. & Yonge, M. (eds.), Adv. Mar. Biol. 8:215—306. London, Ac. Press.

—"— 1972: Oil pollution and marine ecology. — Paul Elek (Scientific Books) Ltd. London.

Saunders, L. & Fleming, R. 1971: Mathematics and statistics. 2nd ed. — The Pharmaceutical Press. London.

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