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Acute Toxicity of Organochlorined Pesticides to the European Eel, *Anguilla anguilla:* The Dependency on Exposure Time and Temperature

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The problems of pollution in relation to fish and their environment have received increasing attention during past decades. Many investigations have been made to measure acute toxicity of pollutants on aquatic organisms. As a result, the use of fish in toxicity testing is well established in many countries by law. A great number of different test methods have been described (Sprague 1969; Mc Lean 1980). Also in the Valencian community, the increasing use of pesticides has led us to study the toxic effect of two organochlorined pesticides in frequent use, lindane and endosulfan, on a teleostean fish that is known to be very resistent to aquatic contamination, *Anguilla anguilla*.

Lindane is the gamma isomer of hexachlorocyclohexane (HCH). It is a neurotoxicant whose effects are normally seen within hours, and result in increased activity, trembling, and convulsions leading to prostration (Ware 1983). Lindane toxicity exhibits a negative temperature correlation. It is more important as an insecticide (Ramade 1976). The cyclodienes like endosulfan, are persistent insecticides which are stable in soil and relatively stable to the ultraviolet action of sunlight. They are neurotoxicants that have effects similar to those of DDT and HCH. The cyclodienes have a positive temperature correlation; that is, their toxicity increases with increases in the surrounding temperature (Ware 1983).

It is well-known that temperature plays an important role in the toxicity of pollutants in environmental water. Cairns et al. (1975) reviewed the effect of temperature on the toxicity of pollutants, but a general picture could not be postulated. Kovacs and Leduc (1982) have found in juvenile rainbow trout that the LC50 (96h) for cyanide was lowest at 6ºC and higher at 12ºC. Then, a higher sensitivity is found at lower temperature. Smith and Heath (1979) showed in several species of fish that usually the LC50 decreased with increasing temperature. We accept that toxicity of insecticides is strongly affected by changing temperatures, generally in one of two ways:toxicity may increase with increasing temperature and so display a positive temperature coefficient. Insecticides belonging to the organophosphorus, carbamate, and cyclodiene classes are usually considered to have positive temperature coefficients (Warwick 1985). The toxicities of DDT, Lindane and most pyrethroids, however, are negatively correlated with temperature (Warwick 1985; Gluth 1983; Kovacs 1982). Pollutant toxicity to fish has mostly been measured by using lethal concentrations and determining LC_{50} (lethal concentration for 50% of individuals) within 4 days (Sprague 1969).

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This paper reports the effects of three temperatures (15, 22 and 29°C) and four times on the toxicity of some organochlorine pesticides to *Anguilla anguilla*.

MATERIAL AND METHODS

Eels of specie Anguilla anguilla, purchased from a fish farm were used for all toxicity tests. They measured 16-20 cm in length, and weighed 20-30 g. Before they were used for experiments, fish were kept in 300-L glass tanks for about 2 weeks. The tanks were supplied with a continuous flow of tap water (temperature, 20ºC; total hardness, 250 ppm as $CaCO_3$; pH 7.9 ± 0.2; alkalinity 4.1 mmol/L). A 12h photoperiod (7.00 to 19.00 h) was maintained. This acclimatization system is described in Gluth and Hanke (1983).

After acclimatization to laboratory conditions, they were kept in the well-aerated test aquaria which were similar in size to the experimental set, and during that time the fish were not fed.

The insecticides used were lindane and endosulfan, both are persistent insecticides and are stable in soil and water. Stock solutions were prepared by dissolving lindane in acetone and endosulfan in water; appropriate quantities of this solution were pipetted into glass aquaria (40L) containing 35 L of test solution and ten fish. Ten more eels used as controls were kept in 35 L of clean water. The test jars were checked every 24 hours to 96 h to record and remove dead animals. The lack of movement and rigidity was used as the criterion for animal death.

Test temperatures (15, 22 and 29ºC ± 0.5ºC) were mantained by constant temperature water baths surrounding the aquaria.

Three replicates were done at each dosage and temperature combination with one of the two different insecticides.

Methods used in the acute lethality tests followed the static test procedures of Committee on Methods for Toxicity Tests with Aquatic Organisms (1975). The animals were acclimatized to the test conditions for 2 days at each temperature.

The percentages of mortality were calculated in each concentration after 24, 48, 72 and 96 h of exposure and converted to probits (Fisher and Yates 1963), the lindane and endosulfan concentrations were converted to logs. The concentration causing 50 % mortality of the test animals (LC50) and their 95 % confidence limits were calculated using the methods of Litchfield and Wilcoxon (1949).

RESULTS AND DISCUSSION

The acute toxicity of lindane in experimental medium at various temperatures is given in Table 1, showing the 24, 48, 72 and 96-h LC50 values with 95% confidence limits.

Lindane, applied to *Anguilla anguilla* at three temperatures, 15, 22 and 29ºC, exhibited a negative temperature coefficient of toxicity in this experimental medium. For this reason, the percentages of mortality are greater at 15ºC than at 22ºC for the same concentration of toxic. But, on the other hand, at 29ºC the percentages of mortality are higher than at 22ºC. Perhaps, this fish is more sensitive to high temperatures and an increase in the respiratory coefficient facilitates the entry of the toxin into its organism.

Table 1. This table shows the LC50 values, expressed in mg/L, 95% confidence limits (in brackets)

	Lindane **LC50** (mg/L)			
Tª (ºC)	**24h**	**48h**	**72h**	**96h**
15	0.34 (0.30-0.39)	0.33 (0.29-0.38)	0.32 (0.27-0.38)	0.32 (0.27-0.38)
22	0.67 (0.64-0.70)	0.67 (0.64-0.70)	0.67 (0.64-0.70)	0.67 (0.64-0.70)
29	0.48 (0.36-0.64)	0.46 (0.40-0.52)	0.46 (0.40-0.52)	0.45 (0.39-0.52)

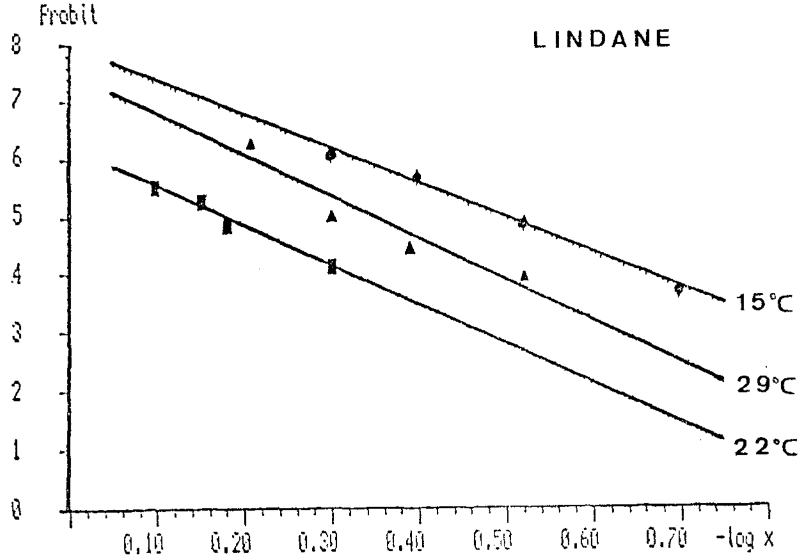
Endosulfan, applied to *A. anguilla*, exhibits a positive temperature correlation, the percentages of mortality are higher at 29ºC than at 22 ºC for the same concentration of toxic.

Table 2. This table shows the LC50 values, expressed in mg/L, 95% confidence limits (in brackets).

	Endosulfan **LC50** (mg/L)			
Tª (ºC)	**24h**	**48h**	**72h**	**96h**
15	0.040 (0.037-0.043)	0.039 (0.036-0.042)	0.038 (0.034-0.042)	0.038 (0.034-0.042)
22	0.042 (0.033-0.052)	0.042 (0.033-0.052)	0.042 (0.033-0.052)	0.042 (0.033-0.052)
29	0.023 (0.020-0.027)	0.022 (0.018-0.025)	0.020 (0.017-0.023)	0.020 (0.017-0.023)

Results from these tables suggest that endosulfan is more toxic than lindane for this aquatic organism, *Anguilla anguilla*.
In Tables 1 and 2 the differences that exist between 24, 48, 72 and 96-h LC50 values can be seen. The LC50 values are similar or identical for these four times in both pesticides at three temperatures. The results suggest that the greatest efficiency was obtained with 24-hr LC50 values.This data is in agreement with Canyurt (1983), who proposed the greatest effect in a time between 24 and 48 hours.

In this study on eels an increase in organochlorined pesticide toxicity was observed associated with increasing and decreasing water temperature (Fig. 1). Gluth (1984) proposed the same effect on carp to subjected to several organochlorined pesticides like aldrin, DDT, endrin and lindane, and to diferent temperatures, and he proved that the carp were more stressed by the high and low temperatures. There is no simple, obvious explanation for these differences. Presumably they reflect variations in the interactions of the factors that contribute to the net toxic effect. These include rates of absorption, distribution,



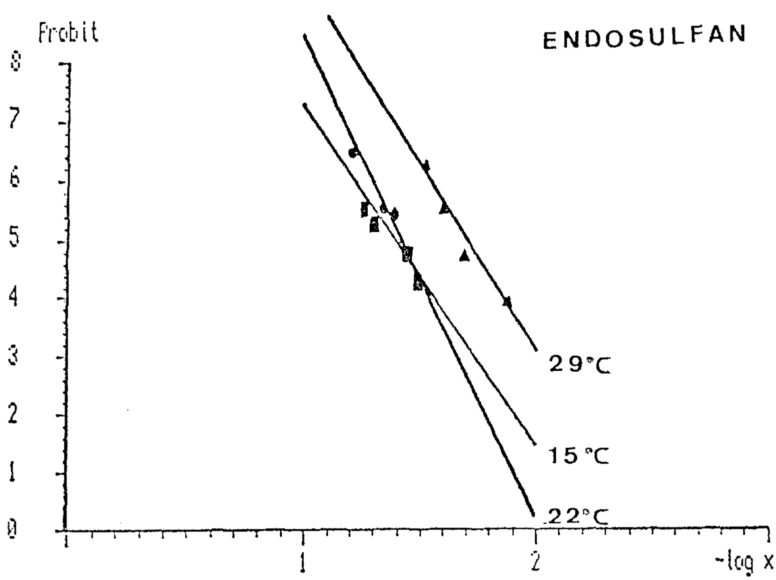


Figure 1. Influence of Lindane and Endosulfan concentration at three temperature intervals on the percentages of mortality.

penetration of organs, detoxification, excretion and differential target site interactions (Canyurt 1983).

The principal site of action of organochlorined pesticides in the vertebrate organism is the central nervous system, and there is a growing body of evidence that shows that they act through interference with nerve transmission (Ware 1983). That interference occurs as a result of alterations in ion transport across the excitable membrane (De Fernicola 1985).

Although application of laboratory data to field conditions is always difficult, the results of this study suggest that the application of lindane and endosulfan at high and low environmental temperatures would be more hazardous to *Anguilla anguilla*.

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