Effects of Prolonged Exposure to Sevin® on an Estuarine Fish, *Leiostomus xanthurus* Lacépède

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Introduction

Sevin is the registered trademark of the synthetic carbamate insecticide, 1-naphthyl N-methylcarbamate. It is also known as carbaryl. This insecticide is toxic to a wide variety of insects as both a contact and a stomach poison. It is reported to be effective against many insects which have become resistant to chlorinated hydrocarbon and phosphate insecticides. Sevin is also toxic to crustaceans and has been used to a limited extent in marine environments to control burrowing shrimp, Callianassa

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californiensis and Upogebia pugettensis (1). The digging activities of these crustaceans make the substrate in some areas unsuitable for oyster production.

Sevin has a relatively low degree of mammalian toxicity and is generally classified among the safer pesticides now in use (2). This comparatively low acute toxicity also applies to fish. The amount of Sevin required to produce a 48-hr. LC_{50} (concentration lethal to 50% of test population) in rainbow trout, Salmo gairdneri, and longnose killifish, Fundulus similis, is about 250 times greater than that of DDT (3). Katz (4) tested 95% Sevin and reported the 48-hr. TL_m (median tolerance limit) for coho salmon (Oncorhynchus kisutch), rainbow trout, bluegill (Lepomis macrochirus), and threespine stickleback (Gasterosteus aculeatus) to be 0.99, 1.35, 5.30, and 10.45 p.p.m., respectively. Stewart, Millemann, and Breese (5) reported 80% Sevin to give a 24-hr. TL_m of 6.7 p.p.m. for the threespine stickleback. They also found 1-naphthol, a hydrolytic product of Sevin, to be more toxic than Sevin to fish.

In the present study juvenile spot, Leiostomus xanthurus, survived 5 months of continuous exposure to 0.1 p.p.m. Sevin in flowing sea water. I am unaware of previous studies in which marine fish were chronically exposed to Sevin or other carbamate insecticides.

Materials and Methods

This laboratory experiment was conducted in a constant-flow system in which fish were exposed to continuously renewed solutions of Sevin in sea water. I put 200 juvenile spot, averaging 18 mm. standard length, in each of two fiberglass-lined wooden tanks. These tanks had a capacity of about 180 liters and measured 30 cm. in width, 25 cm. in depth, and 244 cm. in length. A constant flow of 100 liters of unfiltered sea water per hour passed through each of the tanks. Average monthly water temperatures during the exposure period (March 24 - August 29, 1966) varied from 16° to 29°C. and average monthly salinities ranged from 24 to 30 p.p.t. The fish were fed daily on a mixture of ground fish and dog food.

A Sigmamotor[1/] pump equipped with a Zero-Max speed changer continuously metered (2 ml./min.) a stock solution of Sevin into one of the tanks to make a solution of 0.1 p.p.m. (milligrams per liter) Sevin in sea water. The other tank of fish served as a control. The 0.1 p.p.m. concentration was selected on the basis of results from preliminary screening tests in which 1.0 p.p.m. Sevin killed 60% of a population of spot after 12 days of

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1/Trade names referred to in this publication do not imply endorsement of commercial products

continuous exposure. I prepared 18 liters of the stock solution weekly from 1.5 grams of Sevin (98% purity), 0.5 ℓ. of acetone, and 17.5 ℓ. of tap water (pH 7.2). The small amount of acetone helped dissolve the Sevin. The reported solubility of Sevin in water is less than 99 mg./ℓ.

Fish surviving the chronic exposure to Sevin were examined for possible pathology and changes in enzyme levels. Other survivors were subjected to drastic salinity changes to test their osmoregulatory ability.

Growth and Mortality

The fish that survived the chronic exposure to Sevin exhibited no symptoms of pesticide poisoning during the 5 months. Mortality was about 65% in both control and experimental groups. I believe that this high mortality was due to overcrowding and the limited amount of sea water flowing through the tanks. I was forced to terminate this experiment after 5 months because of a failure in the salt-water system which resulted in the death of the remaining control fish. Growth rate of the Sevin-exposed fish was comparable to that of controls.

Histopathological Examination of Fish

Five spot that survived the 5-month exposure to 0.1 p.p.m. Sevin were examined by Dr. E. M. Wood, consulting pathologist,

to identify possible pathological changes. He also examined unexposed spot and five "positive control" spot from the survivors (50% of a test population) of a 13-day exposure to 1.0 p.p.m. Sevin.

Dr. Wood's report was negative in most respects. The positive controls showed no histologic lesions and he could not separate them from the unexposed fish. The report on the chronically exposed fish was as follows:

"I am able to separate the group on 0.1 part per million of Sevin from both of the two previous groups, and this separation is based on central nervous system lesions. Unfortunately, there is an excellent possibility that these lesions have nothing to do with the Sevin exposure. As you will recall from my report of your fish exposed to 2,4-D, parasitism was described in the positive controls consisting of a sporozoan-like organism in the brain tissue. In the present experiment, all five of the fish exposed to Sevin for 5 months show this brain parasitism which is completely absent in the other two groups. In these fish, some tissue reaction has occurred to these parasites consisting of an inflammatory reaction involving in some cases both the meninges and the substance of the brain itself. I would infer from these changes

that you may observe symptoms suggesting central nervous system damage and, in addition, mortality which you may incorrectly identify with the Sevin treatment. It is possible, of course, that the exposure to Sevin has in some way reduced the resistance of this group to infestation by this parasite. The alternative indicates that the absence of these organisms in your control groups makes questionable the validity of the controls, as such.

"We have previously examined rainbow trout exposed to Sevin up to 1250 parts per billion for 21 days. These fish showed nonspecific changes in both gills and visceral adipose tissue. Such changes are lacking in your specimens."

It is unfortunate that specimens of the true control fish were not available for histological comparison with the Sevin-exposed fish.

Enzyme Activity

Some of the carbamate insecticides, like the organophosphates, are cholinesterase inhibitors. O'Brien (6) reported that the carbamates with anticholinesterase activity appear to fall into two well defined groups. Those which are ionic or strongly basic are very toxic to mammals but almost completely ineffective against insects, whereas those which are not ionic or ionizable may have good insect toxicity and are often less toxic

to mammals than to insects. Sevin is in the latter group.

Weiss (7) showed that fish-brain acetylcholinesterase is inhibited *in vivo* by many of the organophosphorus insecticides at concentrations of 0.1 mg./ℓ. and lower. To determine the brain-enzyme activity of the fish used in this experiment, I sacrificed five spot from each tank after about 2 1/2 months and at the end of the 5-month exposure. Brain cholinesterase (ChE) levels of the spot were measured by the technique described by Holland and Lowe (8). Brain ChE of the fish after 2 1/2 months was only slightly inhibited (about 83% of normal), and at the end of 5 months was within the normal range of activity for spot. Brain ChE of spot exposed to a near-lethal concentration (1.0 p.p.m.) of Sevin for 13 days was 68% of normal. Compared to many of the organophosphorus insecticides, Sevin appears to be a rather mild cholinesterase inhibitor in fish.

Salinity Tolerance

Spot surviving the chronic exposure to Sevin were subjected to rapid increases and decreases in salinity to determine any change in their osmoregulatory ability. Unexposed spot received the same treatment. We observed no differences in reactions between the exposed and unexposed fish. Both groups of fish tolerated a direct transfer from water of 30 p.p.t. salinity

to water of 50 p.p.t. salinity. Other specimens were transferred directly from water of 28 p.p.t. salinity to tap water. Neither group tolerated the tap water longer than a few hours.

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