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Evaluation of Pentron D-90® Toxicity to Juvenile Estuarine Fish and Blue Crabs

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Pentron D-90® is a low molecular weight polyacrylamide based ferrous sulfate compound which has been proposed for use at steam electric generating facilities to temporarily alleviate condenser tube corrosion problems (patent applied for by Apollo Chemical Corp., Whippany, New Jersey). Penton D-90® is to be introduced into the condenser system at a dosage rate of 5 mg/l (total compound:volume) for 1 hour/day for 7 days and subsequently 3 mg/l for 1 hour/day until the corrosion problem is corrected. Approximately 80% of the initial compound will be retained in the condenser tubes followed by slow degradation of the compound and its derivatives.

This study was initiated to provide baseline information on the possible toxicity of Pentron D-90® to selected estuarine organisms. A maximum concentration of 5 mg/l was used since the entire compound and its derivatives will ultimately reach the environment. Juvenile Atlantic menhaden (*Brevoortia tyrannus*), spot (*Leiostomus xanthurus*) and blue crab (*Callinectes sapidus*) were used as representative test organisms because of their importance as commercial and sport fishery species, as well as their ubiquitous distribution in the Chesapeake Bay and other estuaries (HILDEBRAND & SCHROEDER 1927 and WILLIAMS 1974).

MATERIALS AND METHODS

The toxicity of Pentron D-90® was evaluated by standard 96-hour static bioassay procedures outlined in *Standard Methods* (AMERICAN PUBLIC HEALTH ASSOCIATION et al. 1976). This included recommendations for the maintenance of stock organisms as well as the testing of the compound. Some minor modifications of procedures outlined in *Standard Methods* were made because it was felt the modifications gave more meaningful results. These changes are discussed where appropriate. Acclimation and testing were both performed at the Benedict Estuarine Research Laboratory's Aquatic Toxicology Facility using Patuxent River water.

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Collection and Maintenance of Organisms

Juvenile Atlantic menhaden, spot and blue crabs were seined from the Patuxent River between Benedict and Sheridan Point, Maryland. The three species were held in 560 liter aquaria which were supplied with continuous flows of well-aerated Patuxent River water. The basic chemical characteristics of the water used during acclimation and for the tests are outlined in Table 1. All groups were maintained in the laboratory at 25°C (± 1°C) for a minimum of 2 weeks before the studies were conducted. Less than 1% mortality occurred in the holding tanks after the first week. A cyclic artificial photoperiod (incandescent light) of 16L:8D with a 45-minute transition between light periods was used. All fish were fed TetraMenu 4 in 1 Flakes (Tetra Sales Corp., Haywood, California) daily. All animals were fed up to 24 hours before the initiation of the test experiments. No animals were fed during the 96-hour testing periods. The average weight and total length of menhaden and spot and the average weight and total width of blue crabs are presented in Table 2.

TABLE 1

Basic water quality used for acclimation and testing.

Parameter	Mean
pH[a]	7.8
Salinity (°/oo)[b]	6.8
Chloride (mg/l)[a]	5535
Sodium (mg/l)[c]	1872
Sulfate (mg/l)[a]	510
Magnesium (mg/l)[a]	385
Calcium (mg/l)[a]	119
Potassium (mg/l)[a]	81
Total Iron (mg/l)[c]	0.15
Temperature (°C)	25 (± 1)°C
Dissolved Oxygen (mg/l)[a]	>5.8 at all times

[a]Analyzed by procedures in AMERICAN PUBLIC HEALTH ASSOCIATION et al. (1976).

[b]Analyzed by procedures in STRICKLAND & PARSONS (1972).

[c]Analyzed on a Perkin-Elmer Model 503 atomic absorption spectrophotometer (Perkin-Elmer Corp., Norwalk, Connecticut).

TABLE 2

Total number of animals used per tank, mean weight (± S.D.), mean total length (± S.D.) of fish and mean width (± S.D.) of crabs.

Test Number	Species	Number of Animals	Mean Weight (±S.D.) (g)	Mean Total Length (±S.D.) (mm)	Mean Width (±S.D.) (mm)
1- Control	*Brevoortia tyrannus*	20	5.5(±1.66)	89.6(±9.91)	
2- Experimental	*Brevoortia tyrannus*	20	4.9(±1.27)	83.7(±7.52)	
3- Control	*Leiostomus xanthurus*	20	3.2(±0.93)	72.7(±5.90)	
4- Experimental	*Leiostomus xanthurus*	20	4.2(±1.97)	77.2(±10.11)	
5- Control	*Callinectes sapidus*	20	5.0(±2.99)		41.0(±9.51)
6- Experimental	*Callinectes sapidus*	20	4.5(±2.28)		39.1(±7.79)
7- Pentron® D-90 Reference	None				

Experimental Procedures

Twenty control and 20 experimental animals of each species were tested in separate tanks. A seventh tank (labeled Pentron D-90® reference tank) was used to monitor changes in Pentron D-90® concentration because preliminary tests showed that the compound did not remain in solution because of flocculation. Total iron was used to monitor the concentration of Pentron D-90® since it comprised approximately 25% of the compound (see Table 3). Iron concentrations were determined by atomic absorption spectrophotometry (Model 503, Perkin-Elmer Corp., Norwalk, Connecticut). At the beginning of the study, Pentron D-90® was placed into the experimental tanks and vigorously stirred approximately 0.5 hours before the test animals were introduced. Just before the animals were introduced into the experimental tanks, 200 ml aliquots were removed from the tank and acidified with 12 M HCl to a pH of 3.9. Acidification was necessary to prevent the formation of insoluble iron hydroxides. After 96 hours the animals were removed from the tanks and the entire content of each tank was acidified to pH 3.9, vigorously stirred and a 200 ml aliquot removed for iron determination. The three tanks holding the control animals and the Pentron D-90® reference tank were treated in the same manner described above at time 0 and 96 hours.

TABLE 3

Major chemical components of Pentron D-90®[a].

Parameter	Concentration (mg/kg)
Ammonia-N	285
Organic-N	5,400
Sulfate	31,500
Total Volative Solids	61,000
Sodium	51
Zinc	12
Copper	3
Chromium	6
Iron	250,000
Weight Loss at 100°C	33.95%

[a]Chemical composition data supplied by Mr. Larry Ramsey of the State of Maryland Water Resources Administration.

All tests were carried out in 95-liter nontoxic plastic tanks which contained 60 liters of Patuxent River water filtered through 45-micron filter bags to remove large suspended particles. Each tank was fitted with an aeration tube which extended to the bottom. Controlled artificial oxygenation of the experimental, control and reference tanks was performed before and during all tests to insure adequate oxygen concentrations above 5 mg/l at all times. This aeration procedure was used rather than changing the water every 24 hours to avoid excessive handling of the test organisms. Dissolved oxygen, pH, salinity and temperature were measured every 24 hours during the 96-hour observation period. No attempt was made to adjust basic water quality or to replenish test solutions with Pentron D-90® while the study was in progress.

Animals that died during the testing period were removed from the tank when death was first noted or at the end of each 24-hour period. The criteria for determining death of fish were the cessation of gill movement and the lack of response to mild mechanical stimuli. The criteria for determining death of crabs were the cessation of scaphognathite activity and the lack of response to mild mechanical stimuli. Wet weight and total length were determined for each fish. Wet weight and total width were determined for each crab. Animals that survived the 96-hour exposure were measured at the end of the 96-hour testing period.

RESULTS AND DISCUSSION

Pentron D-90® was not toxic to juvenile Atlantic menhaden, spot or blue crabs at an introduced concentration of 5 mg/l over a 96-hour period. One control menhaden died during the 96-hour observation period and one experimental blue crab was killed by predation when it moulted during the study. No other deaths occurred.

The total iron concentrations present in the control, experimental and Pentron D-90® reference tanks (no animals) at the start of the observation period and at the end of 96 hours are presented in Table 4. If one looks at the concentration of iron in the Pentron D-90® reference tank (test number 7 in Table 4) it can be seen that 0.85 mg/l could not be recovered at the end of the 96 hours after acidification to pH 3.9. Since iron is not volatile, most of the iron must have remained bound to the aquarium walls, or less likely, bound in hydroxides not soluble at pH 3.9. This means that even less iron was in solution during the study at pH 7.8.

TABLE 4

Concentration of total iron at time 0 hours and 96 hours in the control, experimental and Pentron D-90® reference tanks.

Test Number	Species	Total Iron (mg/l)	
		0 Hours	96 Hours
1- Control	*Brevoortia tyrannus*	0.16	0.15
2- Experimental	*Brevoortia tyrannus*	1.29	0.22
3- Control	*Leiostomus xanthurus*	0.15	0.15
4- Experimental	*Leiostomus xanthurus*	1.28	0.21
5- Control	*Callinectes sapidus*	0.15	0.15
6- Experimental	*Callinectes sapidus*	1.25	0.19
7- Pentron D-90® Reference	None	1.26	0.41

Many of the ferric and ferrous salts such as the chlorides are highly soluble in water, however, the ferrous ions are readily oxidized in natural surface waters to the ferric condition and form insoluble hydroxides (HEM & CROPPER 1959). These precipitates tend to allomerate, flocculate and settle or be absorbed on surfaces, therefore very little iron remains in solution to act as a toxicant (DOUDOROFF & KATZ 1953). However, if the concentration is sufficient and the water is not strongly buffered, the addition of iron salts may act as a toxicant. McKEE & WOLF (1971) have listed several references which show a wide range of concentrations that may be harmful or lethal to freshwater fish. The concentrations which were harmful to freshwater fish ranged from 100 mg/l up to 10,000 mg/l for 24-hour exposures. Since estuarine waters are highly buffered (STUMM & MORGAN 1970), the range of iron concentrations which may act as a toxicant would be of the same order or higher for estuarine fish.

Since a maximum concentration of 1.25 mg/l iron would be introduced at a steam electric generating facility when 5 mg/l of Pentron D-90® are used (iron comprises approximately 25% of Pentron D-90®; Table 3), no toxicity from iron would be expected to occur to fish and crabs. This study supports that contention.

This study also shows that no other component in Pentron D-90® is toxic to fish and crabs over a 96-hour period at the concentration planned for use at steam electric generating facilities.

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