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EFFECTS OF CRUDE OILS ON THE GASTROINTESTINAL PARASITES OF TWO SPECIES OF MARINE FISH

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ABSTRACT: To assess the effect of crude oil on selected gastrointestinal parasites of fish, winter flounder (*Pseudopleuronectes americanus*) naturally infected with a digenetic trematode (*Steringophorus furciger*), were exposed to contaminated sediment or water soluble fractions of Venezuelan crude oil for 34 and 160 days respectively. Similarly, Atlantic cod (*Gadus morhua*) harboring an acanthocephalan (*Echinorhynchus gadi*), were treated with extracts of Hibernia and Venezuelan oils for periods of 81 to 140 days. In all cases prevalence and intensity of parasitic infections were lower in oil-treated fish. The effect appeared to be more pronounced within fish exposed to water soluble extracts than to oil-contaminated sediment. Fewer parasites present in fish exposed to oil might be attributed to direct toxicity induced by drinking water soluble fractions of crude oil and/or modification of the gut environment brought about by changes in host physiology.

INTRODUCTION

Sindermann (1979), in a review of pollution-associated diseases, remarked that fish taken in polluted areas showed increased abnormalities. The same author stressed that there was a paucity of information on the interaction of pollutants and parasites in fish hosts. There are reports of increased parasitic infections in fish from polluted areas (Overstreet and Howse, 1977; Haensly et al., 1982). Boyce and Yamada (1977) provided evidence that parasitized fish were more susceptible to zinc than noninfected controls while Pascoe and Cram (1977), and Pascoe and Woodworth (1980) reported that exposure of parasitized fish to heavy metals caused mortality. In contrast, exposure of the longhorn sculpin (*Myoxocephalus octodecemspinosus*) infected with a trypanosome to water soluble fractions of crude oil produced minimal changes (Kiceniuk et al., 1982).

During long term exposure to water soluble extracts of Venezuelan crude oil, winter flounder and Atlantic cod consumed less feed, resulting in reduced condition factors (Fletcher et al., 1981; Kiceniuk and Khan, 1983). Both cod and flounder are naturally infected with metazoan parasites (Linton, 1940; Heller, 1949; Dollfus, 1953; Polyanskii, 1955; Scott, 1975, 1976). Since a relationship can exist between environmental pollutants and the degree of parasitism (Haensly et al., 1982), this study was undertaken to ascertain the effect of crude oils on selected parasites occurring naturally in the lumen of the gastrointestinal tract of these fish.

MATERIALS AND METHODS

All fish used in this study originated from Conception Bay, Newfoundland. The flounder, adults only, were collected by divers equipped with SCUBA in shallow water (1–2 m) during summer 1980 at Long Pond (*vide* Kennedy and Steele, 1971, for geographical location). Atlantic cod, subadults and adult (~4–7 yr old), were captured in a cod trap set at 10 m deep about 2 km north of Long Pond. The fish were held prior to and during the experiments in tanks through which sea water flowed continuously. In the first experiment that involved flounder, two groups of experimental fish were exposed to freshly prepared and 1 yr old weathered sediment as reported previously (Fletcher et al., 1981). Control fish were held in a tank with uncontaminated sediment (Table 1). Duplicate samples of sediment were analysed for hydrocarbon levels by fluorescence spectroscopy (Keizer and Gordon, 1973) one week after the beginning and at the conclusion of the experiment. All groups of flounder were fed freshly thawed caplin (*Mallotus villosus*) ad libitum thrice weekly. In the second experiment, the flounder were not fed and the experimental group was exposed to water soluble fractions of crude oil. The latter was prepared by adding 60 ml of crude oil to a head tank (80 liters) daily allowing it to mix with a constant spray of sea water and then drawing off the bottom contents (270 liters/day). A second supply (bypass) into the fish tank was adjusted to provide a total flow of 5 liters/min.

In the experiments (III and IV) which involved cod, 60 ml of crude oil were added to the head tank three times weekly. The flow rate from this tank was adjusted also to 2.5 liters/min and a bypass supplied an additional flow of 2.5 liters/min. Hydrocarbon

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TABLE 1. Experimental protocol involved in exposing marine fish to water soluble petroleum components (WSOF) or petroleum contaminated sediment in flow through sea water systems.

	Experiment number									
	I			II		III			IV	
	C[a]	O.O	N.O	C	V	C	V	H	C	V
Fish species		Flounder		Flounder			Cod		Cod	
No. examined	14	12	16	19	18	21	21	21	10	10
Mean length (cm)	33	32	31			48	47	48	67	63
(range)	(25–38)	(27–39)	(27–35)	—	—	(40–57)	(42–53)	(40–57)	(49–78)	(48–78)
Mean weight (g)	420	469	375			903	825	958	2,940	2,410
	(185–749)	(234–760)	(234–623)	—	—	(587–1,132)	(693–1,443)	(493–1,633)	(1,394–4,584)	(1,063–3,699)
Time of year		July–Dec '80		Sept–Oct '80			Aug–Oct '80		Nov '80–May '81	
Period exposed (days)		160		34			81		140	
Hours of light		Natural photoperiod		Natural photoperiod			Natural photoperiod		12	
Water temp. (C)		4–13		8–12			7–12		0–4	
Tank size (liters)		300		300			3,000		3,000	
Food		Caplin		—			Caplin		Caplin	
Sediment/WSOF[b]		Sediment			WSOF[b]		WSOF		WSOF	
Concentration (μg/liter) (total hydrocarbon)	<50	2,600	2,700	0	~100	0	50–100[c]	50–100	0	50–100
Salinity (‰)		~31.4		~31.4			~31.4		~32.0	
Oxygen		Saturated		Saturated			Saturated		Saturated	

[a] C = control; O.O = old oil; N.O. = new oil; V = Venezuelan; H = Hibernia.
[b] Water soluble oil fraction.
[c] Measured duplicate samples at the beginning (high values) and at end of the oiling regime.

TABLE 2. Comparison of the effects of crude oil administered as oil-contaminated sediment or water soluble fractions of crude oil on natural infections of the parasites *Steringophorus furciger* in winter flounder (experiments I and II) and *Echinorhynchus gadi* in Atlantic cod (experiments III and IV).

Expt. no.	Group	No. of fish	No. (%) parasitized	$\bar{x}$ parasites/fish	Condition (K) factor (SD)[a]
I	Control	14	12 (86)	6.8 (1.1–16.9)[c]	1.53 ± 0.213
	Old oil[b]	12	8 (67)[g]	3.9 (0.1–12.0)	1.58 ± 0.175
	New oil	16	8 (50)[g]	1.5 (0.3–3.1)	1.41 ± 0.226
II	Control	19	18 (94)	15.4 (7.4–26.1)	—
	Oil-treated	18	12 (67)[g]	5.9[d,e] (1.7–12.4)	—
III	Control	21	21 (100)	4.9 (3.1–7.0)	0.93 ± 0.084
	Venezuelan	21	17 (81)[g]	2.8 (1.4–4.7)	1.01 ± 0.140
	Hibernia	21	15 (71)[g]	2.0[d,e] (1.0–3.4)	0.85[f] ± 0.122
IV	Control	10	10 (100)	3.9 (1.4–7.7)	0.88 ± 0.120
	Oil-treated	10	7 (70)[g]	2.0 (0.6–4.0)	0.82 ± 0.063

[a] SD is the standard deviation.
[b] Oil refers to Venezuelan crude used in all experiments except in experiment III when Hibernia was also used.
[c] Means and 95% confidence intervals calculated on $\sqrt{\ }$ transformed data, normal distribution confirmed by Kolmogorov-Smirnov goodness of fit test in all cases except controls in experiments III and IV.
[d] $P < 0.05$ in Kruskal-Wallis one-way ANOVA.
[e] $P < 0.05$ in one-way ANOVA of the $\sqrt{\ }$ transformed data.
[f] $P < 0.05$ in ANOVA.
[g] $P < 0.01$ in Fisher's exact test of prevalence of infection in treatment group compared to its respective control.

analyses were performed daily for 1 wk at approximately monthly intervals.

In three experiments (I, III, IV) where feeding was involved, the food was weighed prior to and after feeding. Total body weight of each fish was monitored also at the start and at the conclusion of the experiments (experiment II excepted). Observations were made on the general behavior of the fish. At the conclusion of each experiment, the fish were necropsied and parasites from the lumen of the gastrointestinal tract were fixed in formalin-alcohol-acetic acid solution. The consistency and color of the intestinal contents were also recorded. In addition, 35 flounder and 35 cod were necropsied following capture to determine the prevalence and intensity of intestinal parasites. Means and 95% confidence intervals were calculated using the square root transformed data. Tests of significance included Kolmogorov-Smirnov goodness of fit test to compare the distribution of the transformed data to a normal distribution. One way analysis of variance was done on the transformed data but since the data for the control groups in experiments III and IV departed significantly from the normal, the Kruskal-Wallis' one way ANOVA was performed also on the untransformed data. Fisher's exact test (Sokal and Rohlf, 1969) was used to compare the prevalence of infection in each treatment group with the respective control for each experiment.

RESULTS AND DISCUSSION

Experiments with winter flounder

The parasite most commonly encountered in the lumen of the digestive tract (stomach to rectum) of winter flounder is a digenetic trematode *Steringophorus furciger* (Olsson, 1868; Odhner, 1905) (syn. *Fellodistomum furcigerum*). Sporocysts and cercaria of the parasite occur in bivalves and its definitive hosts include a variety of fish (Køie, 1979). Ninety-four percent (33) of 35 fish collected from Conception Bay harbored an average of 11.5 (6.5–17.8) individuals of *S. furciger* host.

Following exposure to contaminated sediment, a decrease in food consumption, by approximately one third, was noted during week 4 onwards in the flounder exposed to freshly oiled sediment. This was apparent also after week 5 among flounder exposed to the weathered oil sediment. These trends continued until week 13 when all three groups ate progressively less food as the water temperature decreased from 13 to 7 C. Both groups of oil-treated fish at this time displayed erratic swimming. At necropsy, intestinal contents of oil-treated fish were generally pale fluid green in contrast to the dark semi-solid material in the controls. In the latter, the prevalence of infection with *S. furciger* was higher and the mean number of parasites recovered per host was greater than in either of the fish groups exposed to oil-contaminated sediment (Table 2). It was observed also that the prevalence and the mean number of worms per fish were greater in fish exposed to weathered than to freshly oiled sediment. In experiment II, similar results were observed (Table 2). The mean number and prevalence of parasites were significantly higher in control flounder than in oil-treated fish.

Experiments with Atlantic cod

Two groups of subadult cod were exposed to 50–100 ppb of Venezuelan and Hibernia crude oils for 81 days duration. About 3 wk after exposure, some of the cod in the Hibernia group displayed an unusual behavioral pattern, rotating 360 degrees as they swam. This behavior was observed also 5 wk onwards among the Venezuelan-exposed group. Fish in both oil-treated groups showed a tendency to remain close to the surface where there was a thin film of oil. These fish developed an intense dark pigmentation. They were unusually hyperactive when disturbed. Food consumption in both groups of oil-treated fish decreased so that in the last week of the experiment, the controls ate almost twice that of the oil-treated fish. Control cod showed a mean weight gain during the experiment of 66% in contrast to 44% in Venezuelan-treated and 33% in Hibernia-treated fish. Emaciation was apparent in the cod exposed to Hibernia crude oil as is indicated by the significantly lower condition factor (Table 2). At necropsy, more than 50% in both oil-treated groups contained fluid-green intestinal contents in contrast to dark semi-solid feces in the controls.

The acanthocephalan *E. gadi* was the most common parasite observed in the lumen of the digestive tract of Atlantic cod originating from Conception Bay. Amphipods serve as intermediate hosts while a number of fish species act as final hosts (Ekbaum, 1938). Ninety-four percent (33) of 35 cod necropsied prior to experimentation harbored a mean of 11.3 (11.1–11.6) individuals of *E. gadi*/fish. In the third experiment, fewer [71% (15)] Hibernia-treated fish were parasitized and the number of parasites was significantly lower in the latter than among the controls (Table 2). The Venezuelan oil-treated group also had a significantly lower prevalence of infection than the control cod.

In experiment IV, the controls consumed only slightly more food (10%) than the oil-treated fish but at the conclusion, the mean weight loss was significantly greater (2×) in the oil-exposed fish (Kiceniuk and Khan, 1983). There was no food in the stomach of oil-treated fish and the intestinal contents were fluid light-green.

At necropsy, all control cod harbored the parasite *E. gadi* in contrast to 70% of the oil-treated fish (Table 2). The mean parasite level was also significantly greater than that of oil-treated fish.

In the present study, the behavior, appearance and condition of the oil-exposed fish were consistent with the observations from other studies which indicate that exposure to oil induces stress in fish (Hawkes, 1977; Hodgins et al., 1977; Patten, 1977; Wang and Nicol, 1977; McCain et al., 1978; Payne et al., 1978; Fletcher et al., 1981; Solangi and Overstreet, 1982). Although fewer parasites occurred in the experimental fish in contrast to those necropsied prior to the present studies, prevalences and number of parasites were consistently lower in oil-treated fish. This might be attributed to lack of nutrients in the hosts' intestinal tract, direct toxicity of the ingested crude oil to the parasites and/or modification of the gut environment as a result of oil induced changes in the physiology of the fish. The tegument of digenean and acanthocephalan parasites is actively involved in uptake of nutrients from digested intestinal contents of their hosts (Uglem and Read, 1973; Asch and Read, 1975). However, it is unlikely that lack of nutrients is the cause of the low numbers of parasites in oil-treated fish since in experiment IV both groups ate about the same amount of food during the experiment. Marine fish must drink to osmoregulate. Some of the hydrocarbons ingested are toxic (Malins, 1977) and might likely cause the helminths to be voided from their hosts. It is known that fish excrete metabolites of hydrocarbons in bile (Varanasi and

Malins, 1977) and it has been found recently that the bile acid composition is changed following exposure of cod to oil (Kiceniuk, unpubl. data). Therefore, oil-induced changes in the physiology of fish could produce changes in the gut environment of the parasite. The significantly lower numbers of parasites noted in the oil-exposed fish might be attributed to the direct or indirect effect of the ingested oil fractions.

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CORRECTION . . .

The citation for Wiggins, J. P. (1977) in the article:

STROHLEIN, D. A., AND B. M. CHRISTENSEN. 1983. Metazoan parasites of the eastern cottontail rabbit in western Kentucky. J. Wildl. Dis. 19: 20–23.

is incorrect and should read:

WIGGINS, J. P. 1977. Parasites and related pathology of the cottontail rabbit (*Sylvilagus floridanus*) in central Pennsylvania. Ph.D. Dissertation. Pennsylvania State University, University Park, Pennsylvania, 91 pp.