Effects of Dispersant and Crude Oil Ingestion on Mallard Ducklings (Anas platyrhynchos)

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The use of chemicals to treat oil spills in international waters has become commonplace and accordingly, there has been considerable effort to develop more efficient products to disperse oil. Because oil spills affect marine life and aquatic birds, decisions concerning the application of dispersants to oil spills should be made by weighing the dispersing efficiency of the chemical against any increased potential for biological toxicity (TARZWELL 1970).

Tests of the toxic effects of dispersants have been performed on marine invertebrates and fish. PORTMANN & CONNER (1968) found solvent emulsifiers highly toxic to marine crustaceans, mollusks, and fish; but solvent emulsifiers in combination with oil usually were less toxic. Comparatively high concentrations of Corexit 7664 (Esso) were needed to induce mortality in subtropical marine crustaceans and fish (MCMANUS & CONNELL 1972), but no comparisons were made with effects of oil or of oil plus dispersants. It was also shown that crustacean larvae were more susceptible than adults to dispersants (PORTMANN & CONNER 1968) and that Baltic herring larvae were 50-100 times more sensitive to oil plus dispersant than to oil alone (LINDEN 1975). BALDINI & CUGURRA (1974) studied ichthyotoxic effects of dispersants and oil in both artificial seawater and freshwater systems and concluded that Corexit 8666 and Corexit 7664 had the lowest toxicities of the 14 products tested.

Sublethal concentrations of an oil dispersant (Gulf Agent 1009) alone were shown to have an arrestive effect on swimming in winter flounder (WILDASH 1974). Corexit 9527 has been reported to have deleterious effects on fertilization and embryonic development in several species of marine fishes. Furthermore, the combination of this dispersant with oil had greater effects than either component alone (LONNING AND HAGSTROM 1976), although the dispersant concentrations used may not have been environmentally realistic (CANEVARI & LINDBLOM 1976).

Whereas numerous investigators have studied the impact of dispersants on marine animals, the effects on aquatic avian species are relatively unknown. Corexit 9527 and dispersant-treated crude oil applied to eggshells of developing mallard embryos significantly reduced hatching success (ALBERS 1979). External exposure of incubating mallard hens indicated that Corexit 9527 alone or in combination with crude oil had no effect on egg hatchability, whereas oil alone significantly reduced hatchability (ALBERS 1980).

The present study was undertaken to measure effects of ingesting Corexit 9527 on growth and blood chemistries in mallard ducklings.

MATERIALS AND METHODS

Animals.

Day-old mallard ducklings were obtained from a commercial source (Whistling Wings, Hanover, IL), were banded and weighed, and randomly divided into four groups (n=25/group). The ducklings were initially housed in a brooder unit (Petersime, Gettysburg, OH) maintained at approximately $35 \pm 1^{\circ}\text{C}$, and at 11 days of age the ducklings were moved to $0.9 \times 0.9 \times 0.6$ m high vinyl-coated wire mesh cages (8-9 per cage) where the ambient temperature averaged $24 \pm 2^{\circ}\text{C}$. Water and commercial duck starter mash were provided ad libitum.

Treatment.

At 4 days of age the ducklings were started on the experimental diets. Prudhoe Bay crude oil (PBCO), the dispersant, or both were mixed with duck starter mash in the following concentrations: 0, 0.15% PBCO, 0.15% H_2O + 0.015% Corexit 9527 (Exxon), and 0.15% PBCO + 0.015% Corexit 9527.

Measurements.

Ducklings were weighed at 4 days of age (initial weight) and subsequently each week for 9 weeks. A 1-2 ml blood sample was withdrawn from each duckling by heart puncture at 3 week intervals through 9 weeks. Birds were fasted overnight before blood sampling. After hematocrit (HCT) determinations, plasma was separated and stored frozen at -5°C for subsequent analysis of blood chemistries. Biochemical measurements included glucose, total protein, triglycerides, cholesterol, sodium, and the activities of ornithine carbamyl transferase (OCT) (E.C.2.1.3.3) and alanine aminotransferase (ALT) (E.C.2.6.1.2).

Statistical Analyses.

Multivariate analysis of variance was used to identify treatment effects on weekly body weight, and the Bonferroni method of multiple comparisons was used for pair-wise comparisons of the treatment effects. Body weight data were randomly balanced and incomplete sets of observations were excluded from this analysis. The effects of treatment on HCT, plasma activities of ALT and OCT, and triglyceride concentrations were compared by analysis of variance for repeated measures (treatment and sampling time) and plasma cholesterol concentration was analyzed by one-way analysis of variance. Significant treatment effects were analyzed by Tukey's HSD method of multiple comparisons.

RESULTS

Five ducklings died during the study, two in the group fed crude oil and one in each of the other groups. These mortalities

were probably due to sampling technique and did not appear related to oil or dispersant exposure. Mean body weight, plasma OCT activity, and glucose concentration did not differ among groups during the nine week study. Analysis of variance indicated a significant interaction between treatment and sampling time (P<0.025) for ALT activity. No differences in ALT activity were seen among groups until week nine when the activity in the dispersant group was elevated compared to controls (Table 1). A comparison across time indicated that ALT activities increased in treatment groups at week nine (Table 1).

Analysis of variance revealed significant effects of treatment (P<0.01) and sampling time (P<0.0025) on hematocrit, but no interaction between treatment and sampling time. Treatment means pooled across time were lower (P<0.05) in the oil-fed group when compared to the controls (Table 2). Sampling time means pooled across treatment were less (P<0.05) at week 3 than at weeks 6 and 9 (\bar{X} + SEM: wk 3 = 36.8 + 0.29, wk 6 = 37.6 + 0.28, week 9 = 37.9 + 0.29).

Table 1. Effect of ingesting 0.15% Prudhoe Bay crude oil (oil), 0.015% Corexit 9527 (dispersant), or 0.15% oil + 0.015% dispersant (oil + dispersant) on plasma activity of alanine aminotransferase (ALT).

Treatment	3 weeks	IU/L 6 weeks	9 weeks ²
Control n	14.33 (1.261) 19	18.43 (1.101) 24	17.44 ^B (0.810)
0i1 ³	15.70 ^a (1.096) 24	18.72 ^a (0.860) 24	23.19 ^{A,B,b} (1.205)
Dispersant ³	17.71 ^a (1.185) 24	17.36 ^a (1.231) 23	28.73 ^{A,b} (5.240)
Oil + Dispersant ³	17.38 ^a (1.440)	17.33 ^a (0.090)	23.17 ^A , ^B , ^b (2.212)

Values given are means (SEM).

Treatment group means at week 9 with different capital letter superscripts are significantly different (P<0.05).</p>

 $^{^3}$ Sets of treatment group means were compared across time (week 3 vs 6 vs 9) and means with different lower case letters are significantly different (P<0.05).

Effect of ingesting 0.15% Prudhoe Bay crude oil (oil), 0.015% Corexit 0527 (dispersant) or 0.15% oil + 0.015% dispersant (oil + dispersant) on hematocrit and plasma concentrations of triglycerides, cholesterol, and sodium Table 2.

Control $37.8^{\rm A}$ (0.33) $145.3^{\rm B}$ (8.60) $232.5^{\rm A}$ (12.03) $129.6^{\rm B}$ (2.26) 0.11	reatment	Hematocrit 2 % n = 17-24 birds	crit ² 4 birds	Triglycerides ³ mg/dl n = 20-25 birds	rrides ³ birds	Cholesterol ⁴ mg/dl n = 18 birds	erol ⁴ 1 birds	Sodium ⁴ mEq/L n = 21-23 birds	m L 3 birds
36.2^{B} (0.35) 201.5^{A} (12.96) 193.5^{B} (5.00) $37.7^{A,8}$ (0.29) 158.6^{B} (10.36) $223.9^{A,8}$ (5.90) 38.1^{A} (0.31) $177.4^{A,8}$ (12.09) 229.3^{A} (8.40)	0]	37.8 ^A	(0.33)	145.3 ⁸	(8.60)	232.5 ^A	(12.03)	129.6 ^B	(2.26)
37.7 ^A , ^B (0.29) 158.6 ^B (10.36) 223.9 ^A , ^B (5.90) 38.1 ^A (0.31) 177.4 ^A , ^B (12.09) 229.3 ^A (8.40)		36.2 ^B	(0.35)	201.5 ^A	(12.96)	193.5 ^B	(2.00)	140.2 ^A	(2.17)
38.1^{A} (0.31) 177.4^{A} , (12.09) 229.3^{A} (8.40)	Dispersant	37.7 ^A ,B	(0.29)	158.6 ^B	(10.36)	223.9 ^A ,B	(2.90)	135.9 ^{A,B}	(2.83)
	. Dispersant	38.1 ^A	(0.31)	177.4 ^{A,B}	(12.09)	229.3 ^A	(8.40)	141.4 ^A	(0.92)

Values shown are means (SEM).

Presented are the treatment means that were calculated from the observations at week 3, 6, and 9. Means with different letter superscripts are significantly different (P<0.05).

Presented are the treatment means that were calculated from the observations at week 6 and 9. Means with different letter superscripts are significantly different (P<0.05).

Means with different letter superscripts are 4 Values presented are the treatment means at week 9. significantly different (P<0.05). Significant effects of treatment (P<0.005) and sampling time (P<0.0001) were detected for plasma triglyceride concentration. Treatment means were higher (P<0.05) in the oil-fed group than in the control group (Table 2), and triglyceride concentration increased (P<0.05) at week 9 (\overline{X} + SEM: wk 6 = 145.7 + 5.38, wk 9 = 198.8 + 9.76). Plasma cholesterol concentration was only depressed in the oil group (P<0.05), when compared to the control. The concentration of plasma sodium at week 9 was elevated (P<0.05) in the oil- and oil + dispersant-fed birds compared to the controls. A significant interaction between treatment and sampling time (P<0.0025) was observed for total plasma protein. There was a significant increase (P<0.05) in plasma protein between weeks 6 and 9, with the exception of the oil-fed birds, but there were no treatment-related effects at either week 6 or 9.

DISCUSSION

Chemical dispersants are used to reduce oil/water interfacial tension. Product information (Exxon Chemical Company, 1977) states that these chemicals when applied to oil spills will reduce the adherence of oil to beaches and other surfaces, and will accelerate a quicker dissipation of the spill. The efficacy of these products notwithstanding, it is crucial to know the biological effects of dispersants alone and in combination with crude oils before sanctioning their application.

Body weight, one of the variables employed to assess toxic exposure in growing birds, was not affected by treatment. Plasma ALT and OCT activities were used to monitor any ensuing histopathological changes in the liver and kidney. The only significant alteration was an increase in ALT activity at week 9 in the dispersant group. No other effects of dispersants were observed, which indicates that at prescribed concentrations these chemicals are relatively innocuous to waterfowl.

The exposure of birds to 0.15% PBCO reduced hematocrit and cholesterol concentration, and elevated plasma levels of triglycerides and sodium. The reduction in hematocrit has been consistently observed in growing birds receiving oil in their food (RATTNER & EASTIN 1981). Single oral doses of crude oils given to immature guillemots (Cepphus grylle) and gulls (Larus argentatus) produced a dose-dependent chronic inhibition of growth and a transient rise in plasma sodium concentration (MILLER et al. 1977). Single doses of crude oil affected osmoregulatory ability in ducks (Anas platyrhynchos) maintained on 100% saltwater, but not in ducks given 60% saltwater or freshwater (MILLER et al. 1976). Although the ducks in the present study were not saltwater-stressed, an increase in plasma sodium suggests that the chronic ingestion of crude oil for 9 weeks had an apparent effect on osmoregulatory function. Reduced cholesterol concentrations in oil-fed birds could be related to impaired intestinal transport or to synthesis. whereas the elevation of triglycerides after the overnight fast may indicate a more rapid onset of fat mobilization and gluconeogenesis in oiled birds. When crude oil was fed in combination with dispersant, the dispersant appeared to diminish some of the toxic effects of the oil.

In conclusion, mallard ducklings can probably ingest low levels of dispersant alone, or in combination with crude oil, for 9 weeks without overt or marked biochemical indications of toxicity. Although the present study provides supportive data for the use of Corexit 9527, the effects of dispersants on marine invertebrates and fish (BALDINI & CUGURRA 1974; LONNING & HAGSTROM 1976), and on avian eggs (ALBERS 1979), warrants further investigation of dispersants.

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