

for one gland was -88 mV and that for the other was -77 mV). The electrical potential difference recorded by the Cl-selective microelectrode in 10 successful impalements averaged  $-52 \pm 1$  mV and also was remarkably consistent. The intracellular Cl activity calculated from these values as described previously (Duffey et al. J. Memb. Biol. 42:229-245, 1978) is 60 mM, a value nearly 9 times that which would be predicted by the Nernst equation for an equilibrium distribution in the presence of an extracellular activity of 206 mM ( $290 \text{ mM} \times 0.71 =$  ).

Silva et al. (unpublished observations) have found that the intracellular concentration of Cl in shark rectal gland averages  $91 \pm 8$  mM (16 glands). Thus, the apparent activity coefficient of cell Cl is  $(60/91) = 0.66$ , a value only slightly lower than 0.78, the activity coefficient of an aqueous solution of 100 mM NaCl. This suggests that, given the uncertainties involved, there is not likely to be much sequestration or binding of cell Cl.

### Conclusion

Silva et al. have proposed a model for active Cl secretion by dogfish rectal gland which involves the uphill movement of Cl into the cell across the contraluminal membrane coupled to the downhill entry of Na. The present finding that cell Cl is, in fact, well above the value predicted for an equilibrium distribution is consistent with this model. Additional studies designed to explore the Na-dependency of Cl accumulation are clearly indicated. Supported by research grants to R.A.F. and S.G.S. from the NIH-NIAMDD (AM-16275 and AM-18199) and the Wechsler Research Foundation and by research grants to F.H.E. and P.S. from the National Science Foundation (PCM 77-01146) and the NIH-NIAMDD (AM-18078).

### EFFECTS OF CRUDE OIL FRACTIONS ON HERRING GULL CHICKS (*Larus argentatus*)

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Perhaps the most alarming effect of crude oil on young marine birds (i.e., gulls and guillemots) is the prolonged reduction in growth observed after ingestion of a single small dose (Science 199: 315-317, 1978; Bull. MDIBL 17:40-42, 1977, and this volume). The specific compound(s) responsible for this effect is unknown. Crude oil is a complex mixture of hundreds of organic compounds, and a reasonable approach to solving this problem appears to be testing of oil fractions in the sensitive gull chick system. We present here results of such experiments with South Louisiana crude oil (SLC).

Herring gull chicks were collected from either Old Man Island or Little Duck Island, Maine, and maintained as previously described (Science 199:315-317, 1978). Experimentals received a single oral dose (stomach tube) of 1 ml SLC or that amount of a given fraction found in 1 ml of whole oil; controls were not dosed. Birds were weighed daily after dosing. At day 7 birds were sacrificed and tissues were weighed and frozen for residue analyses (data not yet available). Two crude oils were used, both supplied as SLC by the American Petroleum Institute; one oil was obtained in 1976 (SLC-76; used in previous studies) and the other in 1978 (SLC-78). Oils were fractionated on alumina into aliphatic and aromatic fractions.

Neither SLC-78 nor its fractions had any effect on chick growth. In contrast, SLC-76 and its aromatic fraction did significantly inhibit growth (Table 1). Although 7 day body weights for the SLC-76 aliphatic gulls were somewhat lower than controls, the difference was not significant. Adrenal and nasal salt gland weights for aliphatic dosed gulls were significantly higher than controls, but were significantly lower ( $p < 0.05$ ) than either whole oil or aromatic dosed birds. Analysis of oil samples by capillary gas-liquid chromatography and mass spectrography showed that SLC-78 lacked

TABLE 1

Effect of SLC-76, SLC-76-aliphatic or SLC-76-aromatic fractions on herring gull chick body weight (BW) and organ weights

	BW (g)		(g/kg BW)	Organ weight	
	At dosing	7 d after dosing		Nasal gland (mg/kg BW)	Adrenals (mg/kg BW)
Control (9)	373±30	697±23	41.1±2.5	574±16	62.5±2.4
SLC-76 (6)	365±17	475±33**	49.1±4.0	860±37**	102.8±8.6**
SLC-76 (6) aliphatic	366±27	570±40	43.3±4.9	671±30*	79.1±3.4*
SLC-76 (6) aromatic	353±23	515±37**	46.4±3.0	833±35**	99.0±6.3**

Data given is mean ± SE with the number of birds in each group in parentheses.

\* Significantly different from controls,  $P < 0.06$ .

\*\* Significantly different from controls,  $P < 0.01$ .

polynuclear aromatics with 3 or more rings whereas many of this class of compound were present in SLC-76 (Hallett, unpublished data). These findings suggest that polynuclear aromatics are the most potent inhibitors of gull growth present in this crude oil. Since we do not yet have residue data, we cannot begin to speculate on the target organs that are involved. However, preliminary experiments with adult Leach's storm petrels (*Oceanodroma leucorhoa*) that were dosed with  $^3\text{H}$ -labeled oil and sacrificed 1 day later indicate that about 90% of the oil is retained by the bird and that high levels are found in the gastrointestinal system, blood, muscle and liver. Supported by USPHS Grant ES 00920.

#### NA AND CL UPTAKE BY DISPERSED RECTAL GLAND TUBULES

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Active Cl secretion by shark rectal gland is Na-dependent and can be inhibited by ouabain and furosemide (Silva et al. Am. J. Physiol. 232:F298). Measurements of intracellular Cl activity and electric potential indicate that Cl accumulates well above electrochemical equilibrium (see Duffey et al., this volume). These observations suggest that the uphill step in Cl transport is an obligatory co-transport with Na across the basolateral membrane. Because of its complex architecture, the intact, perfused gland is not suitable for direct measurements of NaCl co-transport. We therefore attempted to examine cellular uptake of  $^{22}\text{Na}$  and  $^{36}\text{Cl}$  in isolated rectal gland tubules. If the lumen of the isolated tubule is normally collapsed, initial accumulation should occur almost exclusively from the basolateral side.

After cannulating the vein and artery, the rectal gland was cleared of blood by perfusing with Ca-free dogfish Ringer (see Silva et al Am. J. Physiol. 232:F298, for composition of Ringer). The vein was then clamped and the gland distended with 0.1 mM Ca-dogfish Ringer containing 8 mg/ml of hyaluronidase (Type 5, Sigma) and 5 mg/ml of crude collagenase (Worthington). Thin slices were cut with a Stadie-Riggs microtome and these were incubated for 20 min at ambient temperature in the above enzyme mixture with magnetic stirring and gassing with 1%  $\text{CO}_2$  in  $\text{O}_2$ . The suspension was then filtered through