that all of HCO_3^- accumulation in aqueous is due to formation of the ion rather than HCO_3^- transport as such. We showed many years ago (Comp. Biochem. Physiol. Vol. 5, p. 201, 1962) that carbonic anhydrase inhibition reduced the concentration of cold HCO_3^- in the aqueous humor of *S. acanthias.*

A general formulation of the process of aqueous humor formation in the elasmobranch follows: HCO_3^- formation from CO_2 is the most rapid process; Na^+ in part travels with it as a necessary counter-ion. Sodium entry may also be mediated by Na^+-K^+ ATPase, but clear evidence on this point will be difficult to obtain since a large portion of isotopic Na^+ (and Cl^-) entry is by exchange diffusion. Flow of aqueous must be at least in part dependent upon HCO_3^- formation and Na^+ transfer.

The significant fact emerges that these properties and quantitative relations in a major subclass of fish are also found in the rabbit and presumably in man. Slowing of aqueous humor flow by inhibition of the carbonic anhydrase catalyzed formation of HCO_3^- and the associated Na^+ transfer is the mechanism by which acetazolamide, methazolamide, and other sulfonamides reduce intra-ocular pressure in glaucoma. We conclude that the chemistry of aqueous humor formation is remarkably stable throughout the vertebrate phyla. (This work was supportedy by NIH grant GM 16934.)

1973 #31

MINIMAL DDE-DISRUPTION OF OSMOREGULATION IN MALLARD TYPE DUCKS *Anas platyrhynchos*

David S. Miller, Allyn Seymour, Jr., David Shoemaker, Gilbert Maeda, David B. Peakall, Robert W. Risebrough, and William B. Kinter, Mount Desert Island Biological Laboratory; Cornell University, Ithaca, New York; and Canadian Wildlife Service, Ottawa, Canada

Studies reporting that organochlorine contaminants inhibit ATPases (Cutkomp *et. al.*, Chem.-Biol. Interactions, 3,439, 1971) and disrupt osmoregulatory events in marine fish (Kinter *et al.*, Environ. Health Perspectives, 1, 169, 1972) have led us to consider organochlorine inhibition of avian nasal gland Na-K-ATPase as a contributing factor in the recent large kills of young seabirds that have been reported on both sides of the Atlantic. Recently Friend and coworkers (Bull. Environ. Contam. Toxicol., 9, 49, 1973) studied the effect of DDE feeding (10-1000 ppm) on nasal gland secretion in the mallard and found decreased nasal gland function in response to an intravenous salt load in pesticide treated animals maintained on fresh water. These authors however did not observe this effect in mallards maintained on a one percent sodium chloride solution.

As our preliminary experiments with ducks showed a large variability in nasal gland function in response to intravenously administered salt, we elected to examine the effects of DDE feeding on nasal gland Na-K-ATPase and plasma electrolytes. To this end white Pekin ducks (hatched and raised at the laboratory) and mallards of a local strain (both *Anas platyrhynchos*) about three months of age were divided into paired flocks of about five birds each and fed *ad lib* duck breeder pellets (kindly donated by Agway, Inc.). The experimental flock had 100 ppm p,p'-DDE (Aldrich

Chemical Co.) added to mash before pelletization; this level was selected to equal or exceed the maximal DDE contamination which might be found in the environment. Tap water was provided *ad lib*. After one week the flocks were moved to covered sheds and the tap water was replaced with 100 percent sea water (434 meq/1 Na, 8.8 meq/1 K, 965 mosm/kg). The animals were weighed and blood samples were taken prior to the experiment after one week on fresh water and several times during the period of exposure to sea water. Towards the end of long sea water periods animals exhibited signs of weakness. At the end of the experiment the ducks were decapitated and the nasal glands and kidneys excised. Samples were set aside for DDE residue analysis (data not yet available) and freeze-dried tissue homogenates were assayed for ATPase activity using a modification of the procedure of Bonting (Membranes and Ion Transport, E.E. Bittar, editor, Wiley-Interscience, New York, 1971, pp. 257-363). The Lowry procedure was used for protein determinations (Lowry *et al.*, J. Biol. Chem., 193, 265, 1951).

During the initial one-week period (fresh water) we observed no change in body weight or plasma electrolyte levels in either experimental or control birds. When exposed to sea water both groups of ducks lost weight rapidly and exhibited progressively increasing plasma osmolalities and sodium concentrations. Potassium concentrations remained constant over the course of the experiments. A representative experiment is shown in Figure 1. Although DDE feeding did tend to increase

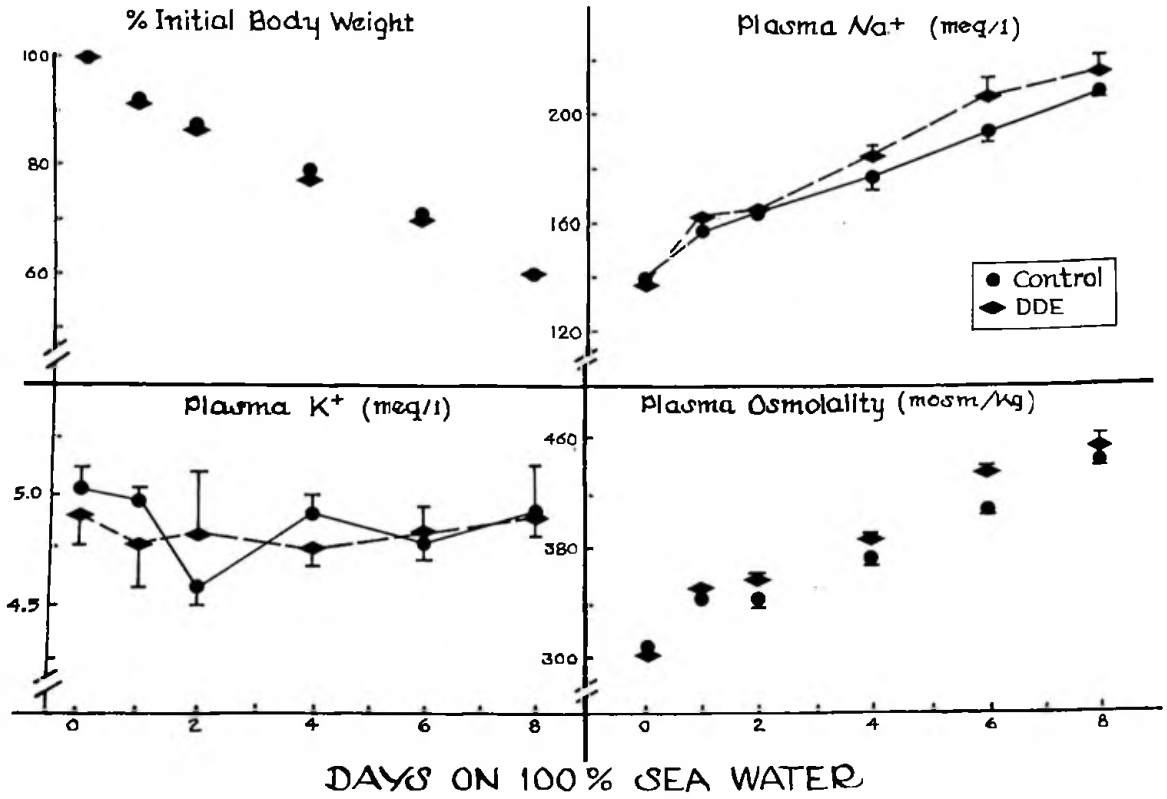

Figure 1. Effect of drinking sea water on body weight and plasma composition in a paired flock experiment with control and DDE fed (100 ppm) white Pekin ducks. Each point represents the mean value derived from five birds; when large enough, variability is indicated by SE bars. Statistical comparisons: $P < 0.02$ for plasma Na 1 day on SW and for plasma osmolality six days on SW; all other differences between control and DDE values are not significant ($P > 0.05$).

slightly weight loss and rise in plasma osmolality and sodium concentration during the sea water period, the differences between individual control and experimental values were rarely large enough to be statistically significant. A comparison of nasal gland and kidney Na-K-ATPase activities over the time course of the experiments also showed minimal differences between the experimental and control groups (Table 1). Noteworthy in the mallard DDE feeding caused a small (22 percent) but significant decrease in nasal gland Na-K-ATPase activity after 19 days on sea water.

TABLE 1

EFFECTS OF DIETARY DDE (100 ppm) ON NASAL GLAND AND KIDNEY Na-K-ATPase IN DUCKS MAINTAINED ON FRESH WATER (FW) AND 100% SEA WATER (SW)*

Maintenance	Control	DDE Fed
	(μmoles P_i/mg protein X hr)	
WHITE PEKIN		
Nasal Gland		
FW	16.3 ± 1.5 (4)	17.0 ± 2.9 (4)
2 days SW	20.2 ± 3.8 (4)	22.6 ± 2.8 (4)
3 days SW	45.4, 36.4 (2)	49.5, 25.7 (2)
8 days SW	31.6 ± 2.5 (4)	30.5 ± 1.9 (5)
Kidney		
8 days SW	4.6 ± 0.2 (5)	3.5 ± 0.5 (4)
MALLARD		
Nasal Gland		
FW	16.7 ± 1.1 (4)	--
19 days SW	41.4 ± 4.4 (4)	32.4 ± 1.3 (4)†
Kidney		
FW	2.7 ± 0.5 (4)	--
19 days SW	3.6 ± 0.7 (4)	3.5 ± 0.4 (4)

*Values generally mean + SE (n), where n is the number of ducks

†Significantly different from SW control ($P < 0.05$).

In conclusion neither the white Pekin duck nor its wild forebear the mallard are sensitive enough to demonstrate conclusive effects of DDE, at least at expected environmental levels on nasal gland function or plasma osmoregulation.

This work was supported primarily by USPHS Grant ES 00920.