

Figure 2. Effect of a pH-gradient on sodium uptake by isolated brush border vesicles. Preincubation median and dilution and wash solution were those indicated in the legend to Figure 1. The incubation media contained 100 mM mannitol, 0.5 mM $^{22}\text{NaCl}$, 99.5 mM choline chloride, 2 mM Ca^{++} -gluconate and either 20 mM HEPES-Tris (pH 8.2) or 20 mM MES-Tris (pH 6.2).

INHIBITION OF CALCIUM-DEPENDENT ATPase FROM MOLLUSC MANTLE TISSUE

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It has been well documented that in many species of birds, exposure to organochlorine pesticides leads to the formation of thin eggshells which are easily broken, thus leading to reproductive failure (Nature 224:44-46, 1969; Nature 224:47-48, 1969; Comp. Gen. Pharmacol. 4:305-313, 1973). An enzymatic basis for the thinning effect has been proposed by Miller et al. (Nature 259:122-124, 1976). They have shown that eggshell-thinning in ducks was accompanied by decreased activity of the calcium-transporting enzyme, Ca-ATPase, in the shell gland.

The process of shell formation by the mantle tissue in molluscs can be considered analogous to that occurring in the shell gland of birds. The purpose of this study was to determine if DDT, *in vitro* also inhibited the Ca-ATPase from mantle tissue of several species of molluscs.

Clams, *Mya arenaria*, and mussels, *Mytilus edulis*, were collected on Laboratory Beach, Salsbury Cove, Maine. Sea scallops, *Placopecten magellanicus*, were kindly supplied by Dan Schick, Department of Marine Resources, Boothbay Harbor, Maine. Ca-ATPase activity was determined by a modification of the method of Miller et al. (Nature 259:122-124, 1976). The final concentrations in the assay medium (1.5 ml) were 10 mM CaCl_2 , 2 mM disodium ATP, and 92 mM Tris (pH 7.4). DDT was added to the assay medium in dimethyl sulfoxide. The final solvent concentration was 1.6%.

The results from these experiments are shown in Figure 1. Baseline control values for clam and scallop Ca-ATPase were 3.45 and 2.65 $\mu\text{moles P}_i/\text{mg protein/hr}$, respectively. In contrast, the mussel has a much

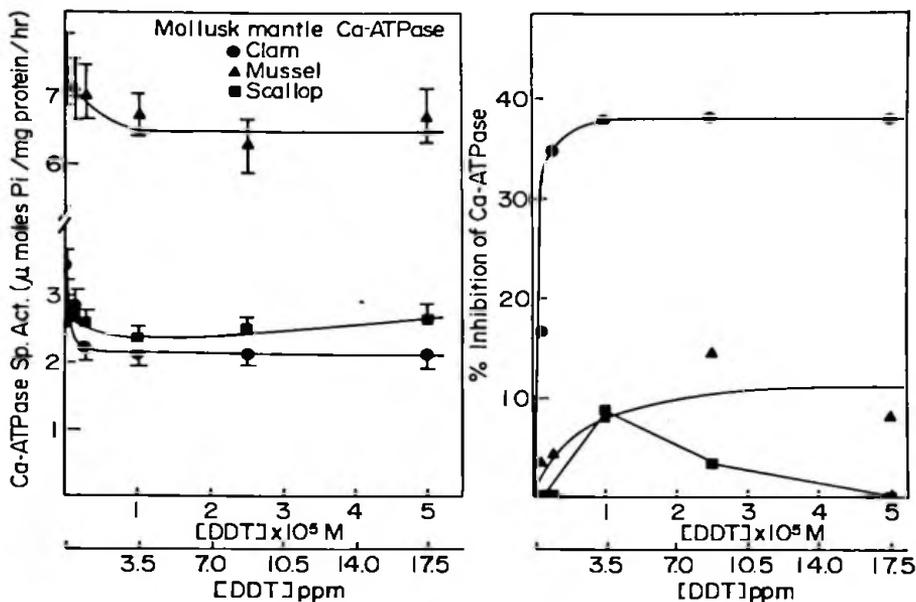


Figure 1. Effect of DDT on Ca-ATPase *in vitro*. The assay was performed at 30°C. Each value represents the mean \pm SE of five separate determinations.

higher value, 7.5 μ moles P_i /mg protein/hr, indicative of greater transport capacity. The Ca-ATPase from the clam was highly sensitive to DDT with significant inhibition at 0.5 ppm (1.43 μ M) and nearly maximal inhibition at 1.0 ppm (2.68 μ M). In contrast, mussel Ca-ATPase showed minimal inhibition; there was no significant effect on scallop Ca-ATPase.

In summary, DDT shows significant inhibition of clam mantle Ca-ATPase *in vitro*; Ca-ATPase from mussel and scallop was only minimally affected. Although no *in vivo* studies have yet been made, DDT appears to be a potential threat for normal growth and development in sensitive species of molluscs. Furthermore, it is suggested that measurement of calcium fluxes in the mantle tissue of a sensitive species of mollusc, such as the clam, might provide a model system for examining the mechanism whereby the organochlorine pesticides alter calcium transport.

BILE SECRETORY FUNCTION IN ISOLATED PERFUSED LIVER OF THE LITTLE SKATE, *Raja erinacea*.

III. OXYGEN CONSUMPTION

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In livers isolated from 1.0 Kg male skates and perfused at 12-15°C with well oxygenated, red cell free elasmobranch Ringers, there is a linear relationship between portal cannula pressure and perfusate flow over a 0.5 to 10.0 cm H_2O range in hydrostatic pressure. Flow increases approximately 0.3 ml $min^{-1} g^{-1}$ liver for each 1.0 cm H_2O elevation in portal pressure. To establish optimal flow rates for adequate oxygen delivery in this isolated organ, we measured hepatic capacity to consume oxygen in eight experiments in which O_2 extraction was measured. Mean flow rates ranged from 0.16 to 3.03 ml $min^{-1} g^{-1}$ liver and were achieved by varying perfusate pressure. The perfusate was oxygenated by bubbling 99% O_2 , 1% CO_2 through an air stone in the pre-liver reservoir which achieved a pO_2 between 600-800 mm Hg. Simultaneous perfusate