Duponol containing approximately 3 mg of diazo Red ITRN salt was added to  $150 \ \mu l$  of digest. The resulting color was read at 550 in a Lowry cuvette in a Coleman spectrophotometer using a special cuvette adapter.

Hydrolysis was proportional to time up to 75 minutes and independent of substrate concentration when less than 30% of the available substrate was hydrolysed at molarities of 0.01 to 0.04. Suitable blanks were run for preformed naphthol and non-enzymatic hydrolysis. In *M. obtusatus* the activity is quite high and there is no consistent rise during development. In *M. finmarchicus* the  $\mu$ g naphthol liberated per egg 15 minutes varies between 0.04 and 0.08 for the first 8 days. Between the 8th and 9th day a sudden (threefold) rise (0.26 ± .02  $\mu$ g) is characteristic. Maximal activity is 2 to 3 times this value and occurs with considerable variability by the 12th to 15th day. Esterase activity increases 10 - 15 times by the time of hatching. In embryos at 12-15 days the major store of esterase is in the gut and caecae and the activity of the contents is higher than that of the cells.

In *M. obtusatus* determinations of esterase activity of separated yolky vesicles and embryonic cells showed the esterase to be distributed in both regions. The embryonic cells usually had more than half the esterase content but comprised less than half the cell volume.

## Potassium Prevents Entry of Neutral Red into Yeast Cells from Acid Media

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We have investigated yeast and sea urchin eggs as laboratory material for teaching classical principles concerning the passive transfer of weak acids and bases across cell membranes. The weak base, neutral red, would be expected to traverse the membrane most rapidly as the lipid soluble non-ionized phase that is found most abundant in alkaline media. Once inside the cell, the penetrator non-dissociated dye would ionize and be trapped and would accumulate there as a relatively non-penetrator cation. Loss of dye from the medium can be detected colorimetrically when the dye is acidulated to a uniform pH.

The sea water medium for sea urchin eggs produced troublesome precipitates when alkalinized. Finally we turned exclusively to yeast which

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\*\*Guest lecturer at the Laboratory Symposium, summer 1956; present address, University of Michigan. has the added advantage of ready availability at inland laboratories and in all seasons.

Unlike the sea urchin eggs, yeast took up dye nearly as completely from acid media as from alkaline. We speculated that dye cations formed in the acid medium might adsorb to the negative outer cell wall which has been described by Rothstein. Addition of 100 mM KC1 (or for that matter, of NaC1) prevented fixation of the dye from acid media by competing with the dye cation for adsorption sites on the yeast.

External potassium competitively inhibits fixation of dye from media of fixed acidity over the range 6 to 100 mM KC1. Yeast, in media with K held at 100 mM, takes up hardly any dye between pH 2.3 and 3.0, while between 3.0 and 7.0 uptake increases progressively to equal that at 7.0 with no K at all. Further work is needed to exclude other possible mechanisms for the effect of external K.

## Studies On Bird Semantics

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In the United States, Eastern Crows (*Corvus b. brachyrhynchos* emit an assembly call which, when recorded and broadcast in nature, causes crows to approach the sound source. An alarm call, when broadcast similarly, causes the birds to fly away.

In France, three "crows" (C. monedula, C. frugilegus and C. c. corone) roost and feed together in winter. Broadcasts of recorded distress calls, emitted when the birds are restrained, cause the birds to approach the sound source at first and afterwards to disperse.

Using exchanged recordings, the distress calls of the French "crows" were broadcast to crows in the U. S. There were no observable reactions in Maine during the summer and in Pennsylvania during the winter. In early summer in Pensylvania, but not in Maine, the distress call of C. monedula induced crows to behave as the French "crows" did. In France, the "crows" did not respond to the recorded alarm of the American species, but reacted to the assembly call as they did to their own distress calls.

In the U. S. Herring Gulls (*Larus argentatus*) responded to their food-finding call, when recorded and broadcast, by approaching the sound source and to broadcasts of their alarm call by dispersal. In France, with the same species, the calls elicited no responses.

These observations suggest that bird develop communication signals within each species having features in common with those of related species, but with regional variations within the species. In cases in which individuals are exposed only to the signals of their own group, they may