and phosphatidyl ethanolamine indicating a general activity of the phospholipid pathways. Homogenates from fertilized eggs differed in several aspects. There was a more active conversion into triglyceride and there was a distinct increase of the incorporation of lipid into diglyceride 70 to 80 min after fertilization. Incorporation of the fatty acid into the phospholipids was similar to the unfertilized samples except for phosphatidyl ethanolamine which also showed increased radioactivity at the 70 to 80 minute stage.

It is expected that continued studies on the biochemical events occurring in the early and mid-mitotic stages will provide information for understanding the mechanism of cytokinesis.

The author gratefully acknowledges the materials and informative discussions provided by Drs. A. Rieck and R. Rappaport as well as the support of Dr. David P. Rall.

1967 #4

STUDIES OF ORGANIC ACID, SUGAR, AND AMINO ACID TRANSPORT IN ISOLATED PERFUSED FLOUNDER TUBULES

Maurice B. Burg and Peter Weller, National Heart Institute, Bethesda, Md.

Using methods previously described (The Bulletin 6:7, 1966) single proximal renal tubules of the flounder Pseudopleuronectes americanus were perfused in vitro. Steady state concentration of transported substrates were determined in the external bathing solution, tubule fluid, and renal cells.

In 10 experiments the addition of PAH-H^3 (2×10^{-5} M) to the external bath, resulted in secretion of PAH into the lumen at a rate (63.8×10^{-14} M min^{-1} mm^{-1} tubule length) which is approximately equal to that previously observed for the same concentration os iodopyracet (56×10^{-14}). The concentration of PAH in the tubule fluid was 21.6 times as great as in the bath, similar to the ratio for iodopyracet (17.2). In contrast, however, the concentration of PAH in the cells was much lower (12.7 times that in the bath) than for iodopyracet (133). Thus, the concentration profile of organic acids during transport by flounder tubules varies markedly, depending on the compound studied. For chlorphenol red the concentration is several orders of magnitude greater in the tubule fluid than in the cells, for PAH it is only twice as great, but for iodopyracet it ís lower in tubule fluid than in cells. These results are most simply explained by differences in the handling of various organic acids by two active transport mechanisms, one at the luminal and the other at the peritubular cell membrane. However, it is also possible that a portion of intracellular organic acid is bound and that this fraction differs depending on the compound studied.

When inulin-C^{14} was placed in the external bath very little appeared in the perfusion fluid (final concentration in the perfusion fluid in 3 experiments .04 times that in the bath) indicating low permeability to inulin. Also, when inulin-C^{14} was placed in the perfusion fluid there was little change in its concentration (ratio of .90 collected/perfused in 9 experiments) indicating little net fluid movement.

When PAH-H^3 (3×10^{-4} M) was placed in the perfusion fluid, there was some loss from the tubule lumen (mean PAH/inulin ratio in the collected fluid was .79 in 7 experiments). The PAH permeability measured by this efflux from the lumen is small, however, compared to the rapid

rate of secretion into the lumen. Concentrations in the tissue of inulin and PAH originating from the lumen were also measured. The PAH concentration in the tissue was .57 times that in the perfusion fluid, whereas inulin in the tissue was .04 times that in the perfusion fluid. Thus, both the luminal and pertibular membranes provide significant barriers to PAH loss from the lumen.

Glycine transport was studied in 8 experiments using glycine-H^3 and glycine-C^{14}. Equal concentrations (5×10^{-5} M) of glycine were placed in the perfusion fluid and external bath with a different radioactive label in each solution. Glycine concentration (estimated as the sum of C^{14} and H^3 activities) was .31 times as high in the collected as in the perfused fluid, consistent with net glycine absorption from the lumen. Movement of glycine from bath to lumen was small (concentration ratio .11, tubule fluid/bath). Tissue glycine concentration was 25 times greater than in the bath (or original perfusion fluid) suggesting that active transport into the cells from the lumen is a step in amino acid absorption. However, a large fraction of the glycine in the tissue (76%) originated from the bath suggesting that there is also transport into the cells across the peritubular border.

Glucose transport was studied in a similar manner. With 1.4 to 5.5 mM glucose in both the bath and perfusion fluid, glucose concentration in the fluid collected from the tubule lumen was .50 times that perfused, indicating net absorption (8 experiments). Movement of glucose from bath to lumen was small (concentration ratio .11, tubule fluid/bath). In contrast to the findings with glycine, however, concentrations of radioactivity in the tissue were relatively low and differed according to the position of the radioactive label in the glucose perfused. When uniformly labeled glucose was initially present in the perfusion fluid, total concentration of radioactivity in the tissue was 4.9 times that in the surrounding fluids (3 experiments), but with C-1 or C-6 labeled glucose this ratio was only 1.8 (5 experiments). The difference is due to the retention of products of glucose metabolism in the tissue from the uniformly labeled glucose, as was confirmed by paper chromatography.

When an isotonic Na-free perfusion fluid ($MgSO_4$ plus $CaSO_4$) was used, there was no inhibition of either glycine or glucose transport. This finding, however, does not rule out the possibility that the transport processes are Na-dependent since Na entered the tubule lumen from the bathing medium in large quantities during perfusion. This was indicated by measurements with Na^{22} and also by the occurrence of net fluid movement into the lumen under these conditions (inulin ratio .67 collected/perfused in 4 experiments). This high Na permeability is comparable to that found in mammalian proximal tubules.

1967 #5

SOME PARAMETERS FOR THE DOGFISH, Squalus acanthias

J. Wendell-Burger, Trinity College, Hartford, Conn.

Some general parameters for the spiny dogfish as determined here at the Laboratory are presented in three tables. Table 1 is self-explanatory, although one is cautioned that values may vary widely. Table 2 is an effort to organize the dogfish in terms of organ weights, tissue water, and organ water pools. In Column 1 are the organ weights for a single fish. No effort was made to idealize the fish or select an archtype. Weights from two other fish are given for comparison