

added glucose were compared with those without added glucose. This indicates, therefore, that at least in tumor tissue under anerobic conditions and in the presence of glucose, incorporation of the major portion of the radiophosphate may depend upon glycolysis. However, the evidence presented does not permit one to rule out the possibility that the remaining radiophosphate incorporation in liver and kidney slices under anerobic conditions is an "exchange" phenomenon independent of oxidative energy.

Summary. Phosphate incorporation into nucleic acid and phosphoprotein of liver, kidney and tumor tissue slices shows some dependence upon the presence of oxygen.

The inhibition of radiophosphate incorporation into nucleic acid and phosphoprotein of tumor slices by nitrogen and its partial reversal by the addition of glucose suggest that, at least in tumor tissue, glycolysis can also serve as a source of energy for the incorporation.

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17290. Low Speed Microtomy for the Electron Microscope.

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The beam of a 50-kv electron microscope does not penetrate specimens that are thicker than $0.1\ \mu$. Thus, in order to study biological specimens with this type of microscope we have further developed the technic described by Pease and Baker,¹ who altered a Spencer Rotary Microtome (model 820) by adding a wedge to the mechanism for forward movement. We reduced the angle of the inclined plane surface by a factor of 10 to 1 so that each step is $0.1\ \mu$ rather than $1.0\ \mu$.

Since forward movement results from a pin in sliding contact with an inclined plane surface, it is important that the surface of the plane be as nearly perfectly flat as possible. The accompanying photograph, Fig. 1, illustrates the means by which the problem of a flat surface for even forward movement was solved. At the left of the picture (A) the original plane can be seen. The reduced angle is shown by D. The light gray triangular area (B) is an angle-reducing block of steel, to which is fastened an optical flat (C). A brass disc (E) instead of a pin is used for the feed screw tip. Horizontal movement of the disc is responsible for forward movement of the specimen. Vertical movement of the

flat and the mechanism to which it is attached causes the slicing action of the microtome. Since the disc and flat are held firmly in contact by a spring, any irregularities in the surface of the plane will be evident on the sliced section.

Since the ordinary microtome knife cannot

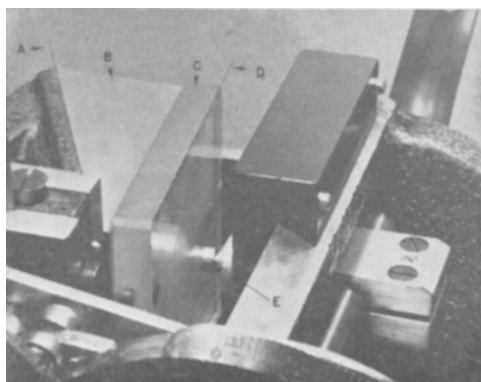


FIG. 1.
Microtome.

A. Original inclined plane surface of Spencer Microtome.

B. Steel wedge reducing from 25° to 2.5° the angle of the original plane surface with respect to direction of the feed screw movement.

C. Optical flat upon which cross feed disc rides.

D. Altered inclined plane surface, making an angle of 2.5° with direction of cross feed movement.

E. Cross feed disc.

¹Pease, D., and Baker, R., *PROC. SOC. EXP. BIOL. AND MED.*, 1948, 67, 470.

be used to cut sections as thin as $0.1\ \mu$, ours was especially made,* hollow ground on both sides, with a long bevel.

Fixing and Mounting Technics. Several technics have been used to prepare tissues for microtomy. Fixation in about 4% neutral formalin, dehydration through graded alcohols, clearing in xylene, and imbedding in paraffin (85°C melting point) has proved to be one of the most successful. With this method it is not necessary to "double imbed"; that is, to use both celloidin and paraffin.

The process of mounting a tissue section on a grid for electron microscopy varies according to the specimen and the operator, but in general the following technic has been successful. The section is transferred directly from the knife to a glass slide. A dissecting needle with a microscopic point is used to lift and move the sections. The point may be inserted slightly into the edge of the paraffin of the first section of a "ribbon" and the "ribbon" pulled out somewhat. A drop of warm water added to the section on the slide will further "spread" the section. When the water has dried, a drop of xylene may be added, or the whole slide may be immersed in xylene, to dissolve the paraffin from the tissue.

When the tissue is thoroughly dry, the slide is immersed in 2% collodion in amyl acetate which is thin dried in an even film. Lines are scored around the specimen; the slide is

breathed upon and immersed gently into water. The section adheres to the collodion film which strips from the glass slide and floats free. A grid is brought up beneath the specimen and both are lifted from the water. Thus the section and the supporting collodion film may be centered on the grid ready for electron observation.

Fig. 2 illustrates some results obtained with the equipment and technics described.

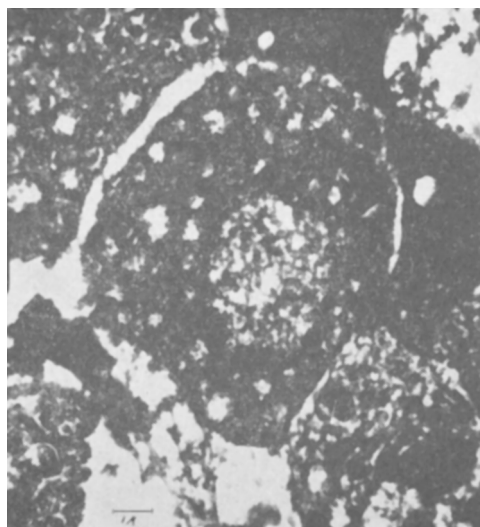


FIG. 2.

Section of rat intestine, sectioned at $0.1\ \mu$. The greater part of the field is filled with a single cell. The nucleus appears less dense than structures seen in the cytoplasm. Individual structures have not been identified. $\times 13,700$.

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17291. The Rate and Total Loss of Body Water on the Survival Time of Adrenalectomized Frogs.*†

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One aspect of the adrenal problem is the relation of the adrenal cortex to the regulation

of water and electrolytes in body fluids. The water content of certain tissues and, more pertinently, of certain cells (eviscerated carcass and liver of rats;¹ skeletal muscles of

* This investigation was aided by the Comly-Coleman Fund of the Ohio State University.

† This work was originally initiated at our suggestion by Martin W. Williams.

¹ Silvette, H., and Britton, S. W., *Am. J. Physiol.*, 1933, 104, 399.