

within 30 minutes. Estimation of the amount of reducing sugars formed indicates that in the great majority of these artificial bean-oil preparations the undigested residue which is responsible for the positive starch-iodine test and which resists digestion for some hours after the control preparations are digested does not exceed 5 per cent.; however, in some cases this fraction was observed to be much greater. Sodium chloride was added to obtain maximum activity of the enzyme.

The reason for this variation and the relation of the ether soluble factor in the intact bean to the delayed digestion of bean starch is being studied further. The relative inaccessibility of the bean starch to *in vitro* digestion by pancreatic amylase is apparent when soaked whole beans are heated and crushed under conditions which more than suffice to render the starch of potatoes quite available. When digested as described with equal starch concentrations the difference in the amount of reducing sugars formed in the two cases is very striking. The starch of beans which are finely ground before cooking appears to be more easily digested than that of the beans which are cooked whole and then mashed.

Procedures other than prolonged heating employed in compensating for the delayed digestion include the well-known action of acids upon insoluble starch as well as treatment with yeast and enzymes of barley malt. While the yeast has many obvious advantages in adding desirable nutritional factors simple preliminary treatment with acid with subsequent neutralization appears to be more practical and efficient than the latter in accelerating the digestion *in vitro*.

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DEMONSTRATION OF THE FORMATION OF A FILAMENT FROM TISSUE CON- STITUENTS IN VITRO

It has been possible during the past four years to entertain visitors to this laboratory with an *in vitro* demonstration of what we have extravagantly termed a "nerve regeneration." A perfectly formed spiral filament can be precipitated from a phosphate extract of brain tissue when a direct current is passed through the material confined in a glass tube of small diameter. This demonstration has served to mystify the skeptics and impress the more gullible of my less scientific collaborators. The interest expressed justifies the recording of this phenomenon, which was first observed during the course of some experiments on the cataphoresis of a mixture of brain proteins.

The demonstration may be repeated as follows: 12

inches of glass tubing with an external diameter of approximately 3 mm (internal diameter may vary between 1 and 2 mm) is bent into a "U" tube approximately 5 inches tall. The tissue extract is prepared by taking one volume of brain cortex (freed of the more obvious blood vessels) and mixing with two volumes of .02 M phosphate buffer, pH 7.3, in a small mortar and pestle, where it is ground until it is quite homogenous. This mixture is centrifuged at 1,500 r.p.m. for approximately 15 minutes, or until the gross particles are separated from the supernatant extract. It is not necessary to centrifuge until the supernatant is perfectly clear. Some of this extract is transferred to the glass "U" tube with the aid of suction and 110 volts of direct current are applied to this mixture with the aid of platinum electrodes inserted in both ends of the "U" tube. The platinum electrodes are made from pieces of platinum wire, gauge No. 22. Contact between the solution and the platinum is made by inserting the wire a distance of 1/16 inch below the surface of the mixture. Shortly after the current is applied a small precipitate begins to form at the anode (acid-forming pole). This precipitate extends itself to form a thin filament which spirals down the tube to form what looks like a fine spring. The thickness of the filament will vary depending upon the strength of the current and the density of the tissue extract. Reversing the polarity of the current causes solution of the filament by the alkali formed at the cathode.

It has been possible to make these spiral filaments from the brains of rats, hogs, cattle, rabbits and man. Attempts to duplicate this phenomenon with tissue extracts from other organs have not been successful in the few trials attempted nor has it been possible to date to duplicate the experiment with solutions of purified proteins.

The filament formed has low tensile strength but with care can be removed from the glass tube. It is not pure protein but is a mixture of most of the components of the original extract.

The explanation of this phenomenon may depend in part upon the fact that the protein in solution in the brain extract is precipitated when the acid is formed at the end of the anode. The orientation of this precipitate into a spiral may be influenced by the shape of the vessel in which it is formed, the electrical field of force about the precipitate and the movement of the fluid in the container.

The biological significance of this demonstration can be almost anything the imaginative reader cares to imply. Additional work is indicated.

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