tween the direction of the motion accompanied by the unconditioned stimulus in the horizontal position (up or downwards) and the motion in the opposite direction during which the unconditioned stimulus was omitted. Thus static as well as kinetic conditioned reactions could be developed. These reactions appeared in the majority of our observations before the horizontal position was reached; in other words, they were mainly anticipatory in nature. In these cases the first appearance of conditioned reactions could be observed without omitting the unconditioned stimulus. Conditioned reactions in exactly the horizontal position were, however, also observed. The inference that we have here to do with conditioned reactions is based upon the fact that these reactions did not exist before the animals were trained, and that they showed typical characteristics of conditioned reactions, such as inhibition by fortuitous external stimuli, or extinction after repeated application of the conditioned stimulus without reinforcement by the unconditioned one. The strength of these conditioned reactions may sometimes exceed that of the unconditioned defense reflexes.

The effect of various peripheral and central lesions (elimination of labyrinthine and other afferent impulses, destruction of cortical areas) upon these conditioned reactions will be reported later.

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Sugar Alcohols VIII. The Oxidative Specificity of *Acetobacter Suboxydans.*

K. PIERRE DOZOIS, C. JELLEFF CARR AND JOHN C. KRANTZ, JR.

From the Departments of Bacteriology and Pharmacology, School of Medicine, University of Maryland, Baltimore, Md.

Neuberg and Hoffmann[1] have observed that cultures of *Acetobacter suboxydans* in killed yeast medium containing 1% of glycerin will quantitatively oxidize the glycerin to dihydroxyacetone. In our studies[2] on the relationship between chemical constitution and utilization by bacteria of the sugar alcohols we became interested in determining whether the *Acetobacter suboxydans* oxidized specifically the secondary alcohol group in glycerin or, if its oxidative power was general for secondary alcohols.

1 Neuberg, C., and Hoffmann, E., *Biochem. Z.*, 1935, **279**, 318.

2 Dozois, K. P., Hachtel, F., Carr, C. J., and Krantz, J. C., Jr., *J. Bact.*, 1935, **30**, 190.

To a wide evaporating dish containing 500 cc. of boiling water 400 gm. of fresh brewers' yeast were added. The yeast was thoroughly ground with water until a smooth, thick paste-like mass was obtained. The mass was then transferred to a 3-liter flask and sufficient boiling water was added to make the total 2000 cc. The mixture was then boiled for 20 minutes, cooled to room temperature and placed in an ice-box for 48 hours. The supernatant fluid was siphoned off and filtered through paper until a clear, amber-colored liquid was obtained. This liquid was again boiled for 20 minutes, cooled and put in the ice-box for 48 hours and filtered. About 1200 cc. of the final product was obtained; this was placed in flasks, 200 cc. in each and sterilized in an autoclave at 15 pounds pressure. After sterilization the medium was stored at room temperature for 12 hours and the desired carbohydrate added to make a 1% solution. This medium was then inoculated with a culture of *Acetobacter suboxydans* and incubated at room temperature, in the dark for 7 days. The compounds studied were methyl alcohol, ethyl alcohol, propylene glycol, isopropyl alcohol, glycerin, trimethylene glycol, erythritol and mannitol.

Chemical tests for the products of oxidation of these compounds are well established and were employed to detect their presence or absence. None of the compounds studied was oxidized by the *Acetobacter suboxydans* culture except glycerin, which after a few days showed the capacity to reduce promptly Fehling's solution in the cold (dihydroxyacetone). It is surprising that substituting a methyl group for the primary alcohol grouping (CH_2OH) in glycerin with the formation of propylene glycol frustrates the capacity of the organism to oxidize the secondary alcohol group, which is common to each molecule, to a ketone. However, it has been demonstrated that the mycoderma aceti and sorbose bacteria[3] will oxidize propylene glycol to monohydroxyacetone. In addition, the placing of the secondary alcohol grouping between 2 methyl groups, as it appears in isopropyl alcohol, renders it refractory to the organism which cannot oxidize the compound to acetone. The secondary alcohol groupings of erythritol and mannitol, having a primary alcohol grouping attached to one adjacent carbon atom and another secondary alcohol grouping to the other, are likewise recalcitrant to the oxidative influence of the *Acetobacter suboxydans*.

Conclusion. From the compounds studied it is apparent that a secondary alcohol, to be oxidized to a ketone by the *Acetobacter suboxydans* by this procedure, requires a primary alcohol grouping

[3] Piloty, O., *Ber.*, 1897, **30**, 316.

on each adjacent carbon atom. The secondary alcohol group in glycerin only, meets these requirements; therefore, it is concluded that the *Acetobacter suboxydans* exhibits an oxidative specificity for the trihydric alcohol, glycerin.

9310 P

Correlation of *in vitro* Activity of Normal Human Gastric Juice on Casein at pH 7.4 with Gastric Intrinsic Factor.

F. H. L. TAYLOR, W. B. CASTLE, ROBERT W. HEINLE AND MARGARET A. ADAMS. (Introduced by R. N. Nye.)

From the Thorndike Memorial Laboratory, Boston City Hospital, and Harvard Medical School, Boston.

The administration of mixtures of normal human gastric juice (intrinsic factor) and beef muscle (extrinsic factor) at pH 5 or 7 to patients with pernicious anemia results in increased blood production and clinical improvement.[1, 2]

In 1930, in association with Dr. C. W. Heath, unsuccessful attempts were made to show that gastric juice at pH 7.4 caused the production of amino acid from beef muscle. Griffiths[3] showed that gastric juice incubated with beef muscle globulin at pH 6 produced certain chemical changes. Emerson and Helmer[4] could not confirm his results. However, the present observations show that when casein was substituted for beef muscle, gastric juice at pH 7.4 did produce progressive changes in the casein.

Normal human gastric juice was obtained free from bile after injection of histamine, filtered through gauze and placed in the ice box. A one percent solution of A. H. Thomas & Company washed casein was prepared at pH 7.4, avoiding excess of acid or alkali. To 50 ml. of this solution were added 50 ml. of normal human gastric juice at pH 7.4 and 2 ml. of toluol. The mixture was incubated at pH 7.4 for 24 hours at 37.5°C., the pH remaining essentially constant.

Ten ml. samples were removed at 4 hours and 24 hours for formol

[1] Castle, W. B., Townsend, W. C., and Heath, C. W., *Am. J. Med. Sc.*, 1930, **180**, 305.

[2] Castle, W. B., *Science*, 1935, **82**, 159.

[3] Griffiths, W. J., *Biochem. J.*, 1934, **28**, 671.

[4] Emerson, C. P., and Helmer, O. M., *Am. J. Digestive Dis. and Nutrition*, 1936, **3**, 735.