the effect of succinate was pronounced and was, in fact, comparable with glutamate. We may conclude that succinate, which is formed under the conditions of our experiments as an end product of glucose breakdown in the old culture, is a building brick for syntheses in the new; and that, of the amino-acids eventually synthesized from it, glutamate is the most important. The development of these views has prompted us to investigate the following problems.

The necessity of carbon dioxide for bacterial growth. When basal media are thoroughly freed from carbon dioxide by a stream of purified air, the growth of *Bact. lactis aerogenes*[2] and other bacteria is delayed indefinitely[3]. Work by Elsden[4] has indicated that carbon dioxide can be assimilated to form succinic acid, and Werkman and his colleagues[5] using isotopic carbon confirm the suggestion. In view of the role of succinic acid in the lag phase of *Bact. lactis aerogenes* to which we have directed attention, it is significant that growth of this strain begins almost immediately if about ten parts per million of succinic acid is added to the basal glucose medium containing a little pyruvate, even though thoroughly aerated with a carbon dioxide-free stream. Growth has been reported in rich media aerated by a carbon dioxide-free stream on previous occasions and has been attributed to the production of sufficient carbon dioxide for growth, by fermentation. An alternative explanation, suggested by our experiments, is that the rich medium supplies nutrients the synthesis of which, had they been absent, would involve the participation of carbon dioxide.

Metabolism of sulphate ions. Experiments with mutant strains of *Esch. coli*[6] indicate that bacteria metabolize sulphate according to the following sequence[7]:

$$\rightarrow SO''_4 \rightarrow SO''_3 \rightarrow S'' \rightarrow \text{cysteine} \rightarrow \text{cystathionine} \rightarrow \text{homocysteine} \rightarrow \text{methionine}.$$

We have shown that several of the compounds in this sequence reduce 'early lag' in basal media where the sole source of sulphur is sulphate. It is of interest to note that sodium sulphide can remove a small but definite fraction of lag at a concentration of a few parts per million.

Action of phenol and other drugs. When cells of the present strain are taken at the point of minimum lag and inoculated into a basal medium also containing 0·4 gm./litre of phenol, a lag period of several hours develops; but the growth-rate and stationary phase remain largely unaffected. Small amounts of glutamate, succinate and methionine cause large reductions in this type of lag, and we may assume that phenol owes its bacteriostatic action to its ability to inhibit these initial syntheses. Similar results have been obtained when the lag was produced by alcohols, ketones, esters and other narcotics. Other amino-acids and organic anions which reduce 'early lag' in different degrees do not, however, reduce lag produced by phenol; in fact, they increase it; and it may be supposed that the phenol also inhibits the processes by which these other amino-acids are converted into glutamate.

Work is still in progress on these problems, and a full account will be published elsewhere.

Addition in proof. During the progress of these investigations, S. J. Ajl and C. H. Werkman (*Arch. Biochem.*, **19**, 493; 1948) published a full account of work with *Esch. coli* in which they demonstrated that several compounds taking part in the Krebs's oxidation cycle, including succinic acid, removed the necessity for carbon dioxide in a basal medium.

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Effect of Histamine on Hyaluronidase Activity

According to modern investigations, the rheumatic and allergic inflammations are characterized by the pathologically increased activity of hyaluronidase. This enzyme hydrolyses the amorphous mucoids of the connective tissue and increases its permeability[1]. But it has not yet been possible to establish why the activity of hyaluronidase increases.

Bearing in mind the histamine theory of the rheumatic and allergic inflammations, we have investigated whether histamine might be responsible for the increase of hyaluronidase action. The activity of hyaluronidase—measured *in vitro* by viscosimetric assay—was not influenced at all by histamine or by synthetic antihistamines. These results are inconsistent with the experiments of Swyer[2], and Mayer and Kull[3], who found that *in vivo* histamine increases, and antihistamines oppose, the spreading effect of hyaluronidase.

Further, we investigated the cause of the difference between the *in vivo* and *in vitro* effects of hyaluronidase. It is known that heparin inhibits hyaluronidase[4], and that histamine opposes certain effects of heparin. Therefore we investigated whether histamine hinders the hyaluronidase-inhibiting effect of heparin. Hyaluronidase action was measured again by the viscosimetric method. When hyaluronidase was inhibited by heparin, histamine opposed this inhibition, and the effect re-appeared with its original speed.

From these results we envisage the mechanism of the increase of hyaluronidase action as follows: histamine, released during rheumatic inflammation, opposes the hyaluronidase-inhibiting action of heparin, and therefore increases the activity of hyaluronidase.

Further work is in progress to establish what kinds of pharmacons are able to prevent the histamine–heparin antagonism, and hence to deal with this important symptom of rheumatic inflammation, namely, the increase of hyaluronidase activity and of permeability.

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