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Influence of Organic Salts on the Action of Tyrosinase

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With the object of finding an analogy to the inhibition phenomena observed by us in precipitin reactions with azoproteins[1] we examined the effect of the sodium salts of organic acids on the action of the oxidase of potatoes and mushrooms on tyrosin. Many were found to have an inhibiting effect upon the ferment action, but the

TABLE I.

0.1 cc. of potato extract (1 gram of potato was extracted with 10 cc. of water and the solution was filtered) was added to a mixture of 0.4 cc. of a solution of tyrosin (10 milligrams in 10 cc. water) and 0.05 cc. of the solutions containing 1 millimole of the sodium salt of the acid in 10 cc. of water. The tests were kept in the room for 2½ hours. To determine the intensity of color, the tests were compared with a series of dilutions made from a solution equal in color to the control tube containing tyrosine and ferment only.

Substances used: 1 acetic acid, 2 propionic acid, 3 butyric acid, 4 valeric acid, 5 heptylic acid, 6 chloracetic acid, 7 trichloracetic acid, 8 brombutyric acid, 9 cyanacetic acid, 10 tartaric acid, 11 citric acid, 12 acrylic acid, 13 crotonic acid, 14 fumaric acid, 15 ethylsulphonic acid, 16 benzolsulphonic acid, 17 cyclohexane carboxylic acid, 18 benzoic acid, 19 α naphtoic acid, 20 β naphtoic acid, 21 furancarboxylic acid, 22 thiophene carboxylic acid, 23 picolinic acid, 24 phenylactic acid, 25 cinnamic acid, 26 hydrocinnamic acid, 27 orthomethylbenzoic acid, 28 metamethylbenzoic acid, 29 paramethylbenzoic acid, 30 orthohydroxybenzoic acid, 31 metahydroxybenzoic acid, 32 parahydroxybenzoic acid, 33 orthochlorbenzoic acid, 34 metachlorbenzoic acid, 35 parachlorbenzoic acid, 36 orthobrombenzoic acid, 37 metabrombenzoic acid, 38 parabrombenzoic acid, 39 orthonitrobenzoic acid, 40 metanitrobenzoic acid, 41 paranitrobenzoic acid.

Numbers of the substances	Control	1	2	3	4	5	6	7	8	9
Intensity of color	10	9½	8½	9	8½	8½	9½	9	1½	10

Numbers of the substances	10	11	12	13	14	15	16	17	18	19
Intensity of color	9½	9	9	9	9	9	10	4½	0	1½

Numbers of the substances	20	21	22	23	24	25	26	27	28	29
Intensity of color	0	8	1½	0	8½	2½	8	5	0	0

Numbers of the substances	30	31	32	33	34	35	36	37	38	39
Intensity of color	½	0	½	4	0	0	7	0	0	9

Numbers of the substances	40	41
Intensity of color	3	0

degree of inhibition varied greatly with the different acids. In a general way it can be stated that the carboxylic acids of benzene and of other cyclic compounds acted more strongly than fatty acids, and that stronger inhibition was caused by the meta and para substituted acids than by the ortho substituted. The tests were made with solutions neutral to litmus, and also in the presence of buffers. Most of the substances acted similarly on mushroom extract and on potato extract, but in some instances differences were observed.

A part of the results is presented in Table I, the figures indicating the degree of oxidation as judged from the intensity of the color.

Somewhat analogous phenomena were noticed when in place of ferment, iron salts and H_2O_2 were used for the oxidation of tyrosin.

A similar difference in the action of ortho, meta and para substituted aromatic acids was also seen in their inhibiting effect upon the color reaction of sodiumsalicylate with ferric chloride. Apparently this was due to the formation of iron compounds which separated in the form of precipitates, but the effect was the same when the precipitation was prevented by addition of a solution of gelatin.

This is a preliminary report.

[1] Landsteiner, K., *Biochem. Z.*, 1920, civ, 280.

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Further Studies on the Heterogenetic Haptene.

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In continuation of the studies recorded previously[1] the following observations were made.

The crude material obtained from horse kidney by extraction with alcohol, and freed from ether soluble substances in the manner described previously, was extracted with boiling benzene. From the solution on standing a precipitate separated out which was removed by centrifugalization. The solution was concentrated. On addition of an equal volume of alcohol a precipitate formed. This was dissolved in benzene and precipitated with alcohol, and the procedure was repeated many times until there appeared in the benzene solution a substance giving a distinct copper orcinol reaction. The