

concurrent wastage by such attack as shown by Nozaki and Bartlett.³

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1. Goldberg, Hohensten and Mark, *J. Poly. Sci.*, 1947, 2, 503. 2. Price, *Reactions at Carbon-Carbon Double Bond*, p. 103, Interscience Publishers, New York, 1946. 3. Nozaki and Bartlett, *J. Amer. Chem. Soc.*, 1946, 68, 1686.

PYROGENIC DECOMPOSITION OF CARENE IN THE PRESENCE OF COPPER AND ALUMINIUM CATALYSTS

ON passing the vapours of carene (b.p. 163-68° C./745 mm., d_{4}^{20} : 0.8468, n_D^{20} : 1.4716, from Indian turpentine, *P. longifolia*) through copper turnings heated to 100° ± 15° C.¹ in the pyrogenic unit previously described,² at an hourly liquid space velocity: 0.14, the terpene hydrocarbon was decomposed. Among the reaction products were 8.8% gases and 89.2% oil. 20.3% of the pyrolysate distilled between 173-78° C./745 mm. (d_{4}^{15} : 0.8697, n_D^{15} : 1.4775) and contained p-cymene.

With aluminium turnings, the gases amounted to 12.3% and oil 83.7%. The yield of the 173-78° C./745 mm. fraction was reduced to 18.5% (d_{4}^{15} : 0.8699, n_D^{15} : 1.4797).

The experiments suggest that a furnace of copper³ or aluminium will have a gentle accelerating effect on the disproportionation of carene to p-cymene.

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December 21, 1948. M. L. JOSHI.

1. See Orlov, "Pyrogenic oxidation of turpentine in the presence of a copper catalyst," *Ukrainski Khem. Zhurnal*, 1926, 2, 1; *Chem. Zentr.*, 1926, II, 660. 2. Sondhi, Bhushan, Gulati and Joshi, *J. Indian Chem. Soc. (Ind. and News Ed.)*, 1947, 10, Nos. 1 & 2, 17. 3. See Kirkpatrick (to Hercules Powder Co.), U.S. Pat. June 25, 1946, 2, 402, 898.

ON THE NATURE OF INHIBITION OF ERYTHROCYTE PYROPHOSPHATASE BY VERONAL-ACETATE BUFFER

It was previously reported¹ that the erythrocyte pyrophosphatase is greatly inactivated by incubation for ½ hr. with M/35 veronal-acetate buffer alone, prior to the addition of the substrate and the activator; and that the presence of the activator (Mg^{++} ion) protect the enzyme from such inactivation to a certain extent. On further study on the nature of the inactivation it was discovered that both the buffer constituents, viz., veronal and acetate, are themselves responsible for the inactivation to a great extent, the inactivation due to heat (38° C.) being comparatively small (Table).

1 ml. of 1 in 20 haemolysate (human erythrocytes) was incubated with 3 ml. of the inhibitor of different concentrations for varying periods of time, and then the enzyme activity was determined by adding 1 ml. of 0.1 M $MgCl_2$ and 0.5 ml. of 0.01 M sodium pyrophosphate. Period of hydrolysis 15 mins. pH -7. Temp. -38° C. Percentage of inhibitions were calculated from the orthophosphate content of the trichloroacetic acid filtrates.

TABLE

Enzyme incubated with	Per cent. inhibition produced		
	preliminary incubation period		
	15 mins.	30 mins.	60 mins.
0.2 M Sodium acetate	60	80	..
0.1 M " "	..	68	87
0.04 M " "	..	33	62
0.04 M Sodium veronal	..	76	88
0.02 M " "	..	42	62
0.01 M " "	..	22	34
Water	4	6	12

It was further observed that the pyrophosphate ion affords better protection of the enzyme than the Mg ion against the inhibition due to the buffer constituents.

A number of substances related to the buffer constituents were studied and varying degrees of inhibition were observed. All the solutions were adjusted to pH 7, and after incubating 1 ml. of the enzyme with 3 ml. of the inhibitor of varying concentrations for different time periods, the

activity was determined in *unbuffered* aqueous medium, the period of hydrolysis being reduced to 15 mins.

Among the narcotics studied, luminal is found to be more inhibitory than veronal. The inhibitor effect of acetate is increased by substituting acid groups like halogen or carboxyl (iodoacetate or malonate), while substitution of the basic amino group (glycine) almost abolished the inhibitor effect of the acetate. Alanine and phenyl-alanine produced no inhibition, but tryptophane, tyrosine and cystine produced inhibition.

Besides the inhibitors already reported viz., (1) formaldehyde, alloxan, iodoacetate, oxalate, malonate, and citrate, the following produced more than 50% (in some cases almost complete) inhibition after incubating the enzyme with decimolar solutions of the inhibitors for 30mins: sodium acetate, sodium monochloracetate, sodium butyrate, sodium lactate, sodium mandalate, sodium phenoxyacetate, sodium pyruvate, sodium maleate, sodium malate, sodium fumarate, sodium succinate, sodium aspartate, sodium tartarate, sodium glutarate, sodium glutamate, acetaldehyde, thiourea, guanidine, and creatine. Higher concentrations are required in the case of the following inhibitors:—acetamide and urethane (M), ethanol, methanol, and urea (2 M), and acetone (3 M).

The pyrophosphatases of optimum pH 7.6 of several animal tissues (liver, kidney, intestinal mucosa, brain, testes, spleen, and muscle of guinea pig) showed identity to the erythrocyte enzyme not only in their property of being inactivated by calcium, fluoride, formaldehyde, and ethanol, but also in being inhibited by $\frac{1}{2}$ hr. incubation with veronal-acetate buffer.

Since the pyrophosphate ion protects the enzyme against the buffer inactivation it may be permissible to surmise that the buffer constituents as well as the substances related to them produce inhibitions by blocking the active centres of the enzyme from reacting with the substrate; the inhibitor effect depending upon a particular molecular structure and the extent of inhibition depending upon the nature of the groups in the molecule. Similar observations were reported in the cases of dehydrogenases² and lipases.^{3,4}

Further work is in progress.

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TRICOTYLY IN *CAPSICUM ANNUUM* VAR. *GROSSA* SENDT.

THE occurrence of two cotyledonary leaves in all dicotyledons is well known though instances are on record where recognisable seed-leaves are wanting or there is only one (Sargent, 1903). In the latter it is assumed that the two cotyledons were completely or partially fused. Compton (1913) has made a detailed study of syncotyly in several dicotyledons. While polycotyly is usual among Gymnosperms (Coulter and Chamberlain, 1910), it is rare in Angiosperms. The following note relates to such a condition noted by the writer recently in *Capsicum annuum* var. *grossa*, commonly known as red pepper.

In a culture of seedlings of *Capsicum annuum* var. *grossa*, which were raised for cytological work a solitary seedling showed the presence of three cotyledons instead of the normal two. The cotyledons were compared with those of a normally developing specimen of the same age. The cotyledons in the latter (Fig. 1) measured 3.2×0.8 cm. each and the angle of divergence between the point of insertion was 180° . In the tricotyledonary seedling (Fig. 2) one of the three cotyledons measured 2.3×0.6 cm. while the other two were 2.3×0.45 cm. each; and the angle of divergence between the larger cotyledon and the other two was much greater than the angle between the larger cotyledon and the other two was much greater than the angle between the latter.

Anatomical observations were made in the tricotyledonary seedling and compared with a normal one. In a normal seedling the root in the early stages is diarch (Fig. 3). A little higher in the hypocotyledonary