

TABLE I
COMPARISON OF O¹⁸ IN 4,5-DIMETHYLCATECHOL FORMED ENZYMICALLY FROM 3,4-DIMETHYLPHENOL^a IN O¹⁸₂^b AND H₂O AND IN O₂ AND H₂O^{18c}

| Experiment | Found | Atom % excess O ^{18d} theoretical for uptake of one atom | No uptake |
|---|-------|---|--------------|
| O ¹⁸ ₂ + H ₂ O | 0.52 | 0.59 | 0.00 |
| | .51 | | |
| | .56 | | |
| O ₂ + H ₂ O ¹⁸ | .00 | 0.59 | 0.00 |
| | .00 | | |

^a Twenty-five ml. reaction volumes contained 0.3 mmole ascorbic acid, 1.3 mmole KH₂PO₄, 2.15 mmole K₂HPO₄, 0.45 mmole 3,4-dimethylphenol and 4.0 mg. purified^b mushroom phenolase having 20-80 cresolase⁶ and ca. 1000 catecholase⁷ units/mg. dry wt. 4,5-Dimethylcatechol (30-50% yield) was isolated through its lead salt, from an ether extract of the reaction mixture, m.p. 84-86°. No hydroxylation occurred in the system when heat-denatured enzyme was substituted for active protein. ^b Prepared electrolytically. ^c Obtained from the Stuart Oxygen Company, containing 1.4 atom % O¹⁸. ^d Mass spectrometry was performed by the Consolidated Engineering Corporation on carbon dioxide samples obtained by Unterzaucher pyrolysis⁸ of 4,5-dimethylcatechol samples. Oxygen recovery was quantitative.

Since the phenolase complex is a cuprous protein^{9,10,11,12} which is in the cupric form after each hydroxylation¹³ and which combines with inhibitor

(5) M. F. Mallette and C. R. Dawson, *Arch. Biochem.*, **23**, 29 (1949).
(6) M. F. Mallette and C. R. Dawson, *THIS JOURNAL*, **64**, 2344 (1942).

(7) W. H. Miller, M. F. Mallette, L. J. Roth and C. R. Dawson, *ibid.*, **66**, 514 (1944).

(8) W. E. Doering and E. Dorfman, *ibid.*, **75**, 5595 (1953).

(9) F. Kubowitz, *Biochem. Z.*, **292**, 221 (1937); **299**, 32 (1938). *cf.* D. Keilin, *Proc. Roy. Soc. (London)*, **104B**, 206 (1929); D. Keilin and T. Mann, *ibid.*, **125B**, 187 (1938).

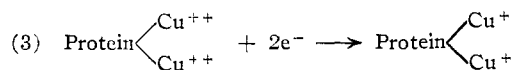
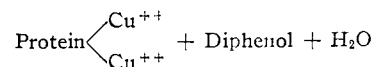
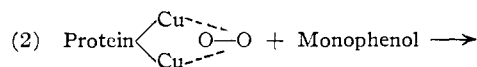
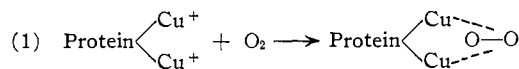
(10) J. Doskocil, *Collection Czechoslov. Chem. Commun.*, **15**, 614 (1950).

(11) A. B. Lerner, "Advances in Enzymology," Vol. XIV, F. F. Nord, ed., 1953, p. 73.

(12) H. S. Mason, "Advances in Enzymology," Vol. XVI, F. F. Nord, ed., 1955, p. 105.

(13) Hydroxylation does not proceed in the absence of reducing agents: *cf.* R. C. Behm and J. M. Nelson, *THIS JOURNAL*, **66**, 711 (1944); M. Suda, N. Kimoto and S. Naono, *J. Biochem. Soc. (Japan)*, **26**, 603 (1954); A. B. Lerner, T. B. Fitzpatrick, E. Calkins and W. H.

CO in the ratio 2 Cu⁺/CO,⁹ hydroxylation by this enzyme system is describable as



The hydroxylative function of phenolase (eq. 1 and 2) is thus coupled to an electron source (eq. 3), i.e., oxidation of *o*-diphenol to *o*-quinone, which may be linked in turn to the common pathways of metabolism through TPNH⁺¹⁴ or DPNH⁺¹⁵, possibly by quinone reductase.¹⁶ The function of the phenolase complex as a terminal oxidase will be in demand during the biosynthesis of *o*-diphenols from monophenols. We propose that these *o*-diphenols are subsequently utilized to form flavonoids, lignins, tannins, cuticulation diphenols of arthropods, melanoproteins of chordates, and possibly adrenaline and noradrenaline.¹² Some instances of light-irreversible inhibition of terminal respiration by carbon monoxide^{17,18} may be accounted for in these terms.

Summerson, *J. Biol. Chem.*, **191**, 799 (1951); L. P. Kendal, *Biochem. J.*, **44**, 442 (1949).

(14) F. Kubowitz, *Biochem. Z.*, **293**, 308 (1937).

(15) E. A. H. Roberts and D. J. Wood, *Biochem. J.*, **53**, 332 (1953).

(16) W. D. Wosilait and A. Nason, *J. Biol. Chem.*, **206**, 255 (1954); **208**, 785 (1954).

(17) G. K. K. Link and R. M. Klein, *Bot. Gaz.*, **133**, 190 (1951).

(18) G. C. Webster, *Plant Physiol.*, **29**, 399 (1954).

DEPARTMENT OF BIOCHEMISTRY
UNIVERSITY OF OREGON MEDICAL SCHOOL
PORTLAND, OREGON

H. S. MASON
W. L. FOWLKS
E. PETERSON

RECEIVED APRIL 11, 1955

BOOK REVIEWS

Annual Review of Physical Chemistry. Volume 5. G. K. ROLLEFSON, Editor, University of California, and R. E. POWELL, Associate Editor, University of California. Annual Reviews, Inc., Stanford, California. 1954. ix + 540 pp. 16 X 23 cm. Price, \$7.00.

The fifth volume of these reviews maintains the excellence of former years. This series is now becoming very well known. Every chemistry and physics library must have these volumes on their shelves. Not only physical chemists and chemical physicists, but spectroscopists, nuclear, radiation and solid state physicists as well as biologists will do well to add this set to their book collection. These surveys offer the best possible means for keeping abreast in the many fields covered. The present volume includes the following topics: Thermochemistry and the Thermodynamic Properties of Substances, Heterogeneous Equilibria and Phase Diagrams, Solutions of Electrolytes, Solutions of Nonelectrolytes, Isotopes, Radioactivity and Nuclear Structure, Radiation Chemistry, Theory of Molecular Structure and Spectra, Spectroscopy, The Solid State, Kinetics

of Reactions in Solution, Kinetics of Reactions in Gases, Properties of Macromolecules in Solution, Colloid Chemistry, Cryogenics, Nuclear Magnetic Resonance, Crystallography, Surface Chemistry and Catalysis, The Microwave Spectra of Gases, Experimental Molecular Structure, Ion Exchange, Statistical Mechanics of Transport and Non-equilibrium Processes, Modern Aspects of Electrode Kinetics.

The literature survey covers the year 1953. Some reviewers have chosen a few important papers, which are discussed at greater length. A complete coverage of all papers for the year would take too much space. The total number of literature citations is 3422! Naturally the style and mode of presentation differs for the various reviews. However, all of them are quite readable.

The authors and editors are to be congratulated upon this excellent compilation.

OFFICE OF ORDNANCE RESEARCH
BOX CM, DUKE STATION
DURHAM, N. C.

GEORGE GLOCKLER