UNSATURATED FAT OXIDASE: DISTRIBUTION, FUNCTION AND HISTOCHEMICAL IDENTIFICATION IN PLANT TISSUES

Sir:

Tauber[1] and Strain[2] reported that the enzyme associated with unsaturated fats in soy meal is an "unsaturated fat oxidase." "Carotene oxidase"[3] has been renamed "unsaturated fat oxidase" by these authors because the oxidation of carotene in soy meal is a result of the enzymatic oxidation of unsaturated fat, and it is further apparent that the "lipoxidase" demonstrated earlier[4,5] is also the same as unsaturated fat oxidase.

By means of some new methods of observation recorded here it is possible to confirm the work of Tauber and Strain and to reveal at the same time the function and intimate association of unsaturated fat oxidase with specific tissues in numerous crop plants. The presence of unsaturated fats in microscopic preparations may be detected by means of Nile blue sulfate, osmium tetroxide, Sudan III and IV and Sudan black B. The oxidation and oxidase system of these fats is best studied by using the leuco form of methylene blue, leuco indigo carmine, guaiacol, and "Nadi" reagent. By means of the above indicators the location and function of the unsaturated fat oxidase may be demonstrated *in vivo*.[6,7]

Recent preliminary observations indicate that the unsaturated fat oxidases are activated in regions of the plant that are alkaline or neutral, where water loss is taking place, beneath wound surfaces, and where inhibitols or antioxidants are absent or inactivated. Dilute solutions (0.0012–0.00028%) of sodium selenite, an antioxidant to many respiration systems, were applied to sustaining sections and it was found that the general oxidase reaction is halted and that the oxidation of indicators is confined to those tissues (epidermis, hypodermis, etc.) which have an accumulation of unsaturated fats. Sodium selenite, as an alkaline salt, depressed oxidase activity in tissues (cortex and xylem) that are not fatty in nature and where the oxidases are activated only at a *p*H of 4.8–5.8. Sodium selenite and alkaline buffer systems activated the oxidation of unsaturated fats by apparently freeing the lipoids and the associated oxidase system from naturally occurring alkali-labile inhibitols and loose addition compounds introduced (iodine, carbonic acid, etc.) into sustaining sections. Root and stem tissues from six species of crop plants grown in buffered media at a *p*H of 7.2–7.6 had a high oxidase activity in fatty tissues at the seedling stage, but at the end of five weeks the unsaturated fat supply was very low and the fatty tissues were characterized by a low oxidase activity and the presence of fatty degradational products. The reverse of this condition was obtained in the case of plants grown in buffered media at a *p*H of 4.8–5.6. Plants grown in relatively acid media had a higher fatty accumulation and a low oxidase activity until treated in section with alkaline buffered solutions. It is thus possible to visually isolate and activate oxidase systems in sustaining sections and in growing plants by means of alkaline salts which activate unsaturated fat oxidase systems but depress the oxidases associated with non-fatty tissues.

(1) H. Tauber, THIS JOURNAL, **62**, 2251 (1940).
(2) H. H. Strain, *ibid.*, **63**, 3542 (1941).
(3) J. B. Sumner and R. J. Sumner, *J. Biol. Chem.*, **134**, 531–533 (1940).
(4) E. Andre and K. Hon, *Compt. rend.*, **195**, 172–174 (1932).
(5) C. H. Lea, *Rep. Food Invest. Bd., Lond.*, 55–56 (1937).
(6) D. S. Van Fleet, *Am. J. Bot.*, **29**, 1–15 (1942).
(7) D. S. Van Fleet, *ibid.*, **29**, 747–755 (1942).

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THE MUTAROTATION OF β-D-ALTROSE

Sir:

When D-altrose was first isolated in crystalline form[1] we reported that mutarotation was not observed, the first reading being taken 3.8 minutes after solution of the sugar in water at 20°. Austin and Humoller[2] had reported a small mutarotation, $[\alpha]^{20-25}D$ $-28.75 \rightarrow -32.30°$ in water, for the antipodal L-altrose, and accordingly had designated it as the β-modification.

With a larger amount of D-altrose now available from the deacetylation of its α-pentaacetate, which was obtained by the acetolysis of 4,6-benzylidene-α-methyl-D-altroside,[3] we have found that this sugar exhibits a complex mutarotation. Thus, 2.000 g. of D-altrose in 50 ml. of water showed $[\alpha]^{20}D$ +11.7, 19.3, 25.1, 32.1 and 33.1° (final) at the end of 2.83, 4.10, 6.00, 17.8 and 31 minutes, respectively. From calculations of the velocity coefficients, derived from these and other intermediate measurements, it would appear that the mutarotation consists of a very rapid interconversion of the furanose and pyranose modi-

(1) Richtmyer and Hudson, THIS JOURNAL, **57**, 1720 (1935).
(2) Austin and Humoller, *ibid.*, **56**, 1154 (1934).
(3) Richtmyer and Hudson, *ibid.*, **63**, 1730 (1941).

fications, followed by a slower interconversion of α and β pyranose modifications. These velocity coefficients, $m_1 = 0.0792$ and $m_2 = 0.357$, are greater than any which have been reported previously in the sugar series.[4] The velocity coefficient for the rapid reaction is approached closest by the value $m_2 = 0.311$ for the compound D-mannose·$CaCl_2$·$4H_2O$ of Dale,[5] which appears from the evidence of Isbell[6] to be a furanose form. It is possible that our crystalline D-altrose also represents a furanose form. By extrapolation, the initial specific rotation of D-altrose would be about $-69°$; in accordance with the usual nomenclature it is designated tentatively as a β-modification.

In view of the extremely rapid change of rotation which occurs within the first three minutes after altrose is dissolved in water, and the resulting uncertainty in the calculated rotation at zero time, we plan to make additional measurements within that period, as well as at a lower temperature, the results to be reported in a subsequent communication.

(4) See F. J. Bates and Associates, "Polarimetry, Saccharimetry and the Sugars," United States Government Printing Office, Washington, D. C., 1942, pp. 442, 762.

(5) Dale, THIS JOURNAL, **51**, 2788 (1929).

(6) Isbell, *ibid.*, **55**, 2166 (1933); Isbell and Pigman, *J. Research Natl. Bur. Standards*, **18**, 141 (1937).

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NEW BOOKS

A Textbook of Biochemistry. By ROGER J. WILLIAMS, Ph.D., D.Sc., Professor of Chemistry, University of Texas. Second edition. D. Van Nostrand Company, Inc., 250 Fourth Avenue, New York, N. Y., 1942. x + 533 pp. Illustrated. 14 × 22 cm. Price, $4.00.

The first edition of this textbook was published in 1938. In the present revision, the author states in the preface that he has attempted to introduce the most outstanding advances of the past few years without altering in any essential manner the original outline of the text. The chapters on "Essential Food Constituents" and "Biochemical Catalysts" have undergone the most revision. Most of the other chapters remain largely unchanged. Errors in the first edition, pointed out by THIS JOURNAL'S reviewer, still remain. There is also carried over from the first edition the misleading statement (page 90) that hemoglobins may *unite* with oxygen to form methemoglobin.

It is disappointing to find that the chapter on intermediate protein metabolism presents so little of the data obtained by the use of isotopes which provides us with our current concepts in this field. The name of Schoenheimer does not occur once in this chapter. The synthesis of creatine in the body from glycine, arginine, and methionine is briefly stated without giving any of the beautiful evidence that has been obtained to warrant such a statement. Limitations of space undoubtedly make this necessary where there is so much ground to cover, and certainly the author does an excellent job of covering all the essential phases of biochemistry. The reviewer is, however, apprehensive that the growing trend in textbooks to present only the "cold dope" without the experimental evidence behind it tends to develop students lacking in the ability to critically judge and evaluate scientific information.

As users of the first edition can testify, the author possesses the ability to present the facts in a clear and lucid manner. This textbook can be recommended as a valuable aid for the introduction of the general student to the subject matter of biochemistry.

ERIC G. BALL

General Inorganic Chemistry. By M. CANNON SNEED, Professor of Chemistry, and J. Lewis Maynard, Assistant Professor of Chemistry, in the School of Chemistry, University of Minnesota. D. Van Nostrand Company, Inc., 250 Fourth Avenue, New York, N. Y., 1942. xviii + 1166 pp. 180 figs. 14.5 × 22 cm. Price, $4.50.

A partial examination of the book before looking at the preface left the reviewer in doubt as to whom the book was intended to serve. Chapter 1, "Some Fundamentals," gives simple definitions and elementary descriptions of a few typical chemical phenomena. Chapter 2, "Atomic Theory and Structure of the Atom" seems to take for granted a wide knowledge of descriptive chemistry, of electrical phenomena and the laws of physics, and it deduces chemical properties including ionic valence, covalence and coördinate covalence, from the assumed electron groupings. Chapter 3, "The Periodic System," deals with the historical development of the idea, without the reader having been given more than a hint that there are a number of elements. Chapter 4, "Oxygen and Ozone," is mostly conventional descriptive matter evidently, except for a few theoretical points, at the level of absolute beginners in science.

The whole book is one of 1166 pages, which is much larger than the usual college textbook of chemistry and