

## SHORT COMMUNICATION

# DOES TREHALOSE OCCUR IN ANGIOSPERMAE?\*

ARNOLD E. S. GUSSIN

Department of the Biological Sciences, Clark Science Center, Smith College, Northampton,  
Mass. 01060, U.S.A.

(Received 12 October 1971)

**Abstract**—Two previous reports on the identification of trehalose in Angiospermae could not be corroborated.

TREHALOSE (1- $\alpha$ -D-glucopyranosyl- $\alpha$ -D-glucopyranoside) occurs widely in invertebrate phyla, in fungi, in many algae, and in occasional lower vascular plants.<sup>1</sup> In 1967 Oesch and Meier<sup>2</sup> isolated trehalose from the 'cells of the cambial zone and the youngest xylem cells' of the stem of an 80-yr-old beech tree, *Fagus silvatica* L. The only other flowering plant in which trehalose has been identified, albeit tentatively, is the hypocotyl of cabbage, *Brassica oleracea* L. var. *capitata* L.<sup>3</sup>

In my continuing study of trehalose and its hydrolytic enzyme trehalase (trehalose 1-glucohydrolase, E.C. 3.2.1.28) in insects<sup>4</sup> and in pollen,<sup>5,6</sup> it was convenient to attempt to corroborate the above findings. Indeed, my discovery of trehalose in pollen<sup>5</sup> and in other plant parts, and my concomitant inability to isolate its substrate, made the work of Oesch and Meier<sup>2</sup> and Keen and Williams<sup>3</sup> all the more intriguing.

Isolations of trehalose were carried out exactly as reported,<sup>2,3</sup> except that 10 times the amount of stated material was used. The beech limbs *Fagus silvatica* L. var. *atropunicea* West were cut in July in order to duplicate as closely as possible the experiments of Oesch and Meier.<sup>2</sup> *Brassica oleracea* L. var. Eastern Ballhead hort. (hypocotyls, 14 and 25 days after sowing) was the other plant examined. Identification of the disaccharide was attempted with TLC<sup>7</sup> and by enzymatic procedures<sup>8</sup> routinely used in my laboratory. The trehalase employed was a pure housefly preparation (1 band on polyacrylamide gel electrophoresis) made according to Lefebvre and Huber.<sup>9</sup> The enzyme is specific for substrate, hydrolytically splitting 1 mol of trehalose into 2 mol of glucose. Thus, the presence of trehalose in a sample incubated with trehalase is indicated by a colorimetric glucose increment in experimental tubes compared to boiled controls.<sup>5</sup> No trehalose was found in extracts from either angiosperm.

\* Contribution No. 42 from the Smith College Department of the Biological Sciences.

<sup>1</sup> R. M. ROBERTS and K. C. TOVEY, *Arch. Biochem. Biophys.* **133**, 408 (1969).

<sup>2</sup> F. OESCH and H. MEIER, *Phytochem.* **6**, 1147 (1967).

<sup>3</sup> N. T. KEEN and P. H. WILLIAMS, *Plant Physiol.* **44**, 748 (1969).

<sup>4</sup> A. E. S. GUSSIN and G. R. WYATT, *Arch. Biochem. Biophys.* **112**, 626 (1965).

<sup>5</sup> A. E. S. GUSSIN, J. H. MCCORMACK, L. Y. L. WAUNG and D. S. GLUCKIN, *Plant Physiol.* **44**, 1163 (1969).

<sup>6</sup> A. E. S. GUSSIN and J. H. MCCORMACK, *Phytochem.* **9**, 1915 (1970).

<sup>7</sup> V. A. DESTEPHANIS and J. G. PONTE, JR., *J. Chromatog.* **34**, 116 (1969).

<sup>8</sup> S. FRIEDMAN, *Arch. Biochem. Biophys.* **87**, 252 (1960).

<sup>9</sup> Y. A. LEFEBVRE and R. E. HUBER, *Arch. Biochem. Biophys.* **140**, 514 (1970).

BioRad Ag501-X8 (D) is routinely used to deionize sugar extracts prior to chromatography. This resin tenaciously binds trehalose; the disaccharide can be removed from the resin only with prolonged washing. Cabbage hypocotyls infected with clubroot fungus, *Plasmodiophora brassicae* Wor., were studied by Keen and Williams<sup>3</sup> along with uninfected hypocotyls. The former hypocotyls might have been the source of the tentatively identified trehalose in the latter, especially if virgin resin was not used for every deionization.

Oesch and Meier<sup>2</sup> suggested, but did not prove, that the trehalose they isolated was not the result of bacterial contamination, but was a metabolic product of the tree itself. In view of the absence of trehalose in the beech tree I studied, the isolation and identification of the disaccharide should be extended to additional specimens of beech, and to other flowering plants. The suggestion that the trehalose-trehalase system might mediate hexose transport in plant<sup>10</sup> and animal<sup>11</sup> organs points to the necessity for the existence of a transient trehalose. It is conceivable that Oesch and Meier's<sup>2</sup> trehalose could be a transient molecule, but the high concentration they noted (0.8–0.9% of the neutral sugars) would probably obviate this possibility.

*Acknowledgements*—The staff of the Smith College Botanical Gardens generously provided the experimental materials. Mr. Edwin J. Hudson was a dedicated assistant and Dr. Philip Reid read the manuscript. The research was supported by grants from the National Science Foundation.

<sup>10</sup> K. T. GLASZIOU and K. R. GAYLER, *Planta* **85**, 299 (1969).

<sup>11</sup> B. SACKTOR, *Proc. Natl. Acad. Sci. U.S.A.* **60**, 1007 (1968).

*Key Word Index*—Angiospermae; trehalose.