

gressive development of hardness throughout the autumn period in the hardy varieties, while the tender varieties show no such change. At the end of November, the differences in exosmosis as measured by this method amount to several hundred per cent. between the very tender and the most hardy varieties. Colorimetric tests for chlorides and nitrates in the exudate from the frozen roots correlate very well with the conductivity measurements, and regeneration of growth in the greenhouse indicates that injury by freezing may be closely estimated by this method. The author plans to investigate its application to other plants and to perfect details of technique. The precise experimental method and results will be published soon.

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#### A SOURCE OF DIASTASE

EXPERIMENTS designed to illustrate digestion are not uncommon in beginning courses in botany, the action of diastase or amylase on starch paste being a common method of procedure. Ptyalin, contained in the saliva, may be successfully employed, but for classes involving large numbers of students this is difficult to obtain in sufficient quantity. Malt diastase may be prepared, if facilities are available, but the extraction must be performed with care, and the yield is small unless large-scale operations are employed. The ordinary commercial preparations, available on the market under various trade names, are unsuited, since they contain starch or sugar or both and thus vitiate the results before the experiments are started. After a considerable canvass of the situation, a source of diastase has been found which meets the requirements of a starch- and sugar-free enzyme which is not only entirely suitable for experiments of the

nature indicated, but which is useful in a number of physiological and biological procedures as well. Since it seems apparent that teachers generally are unaware of an easily obtainable enzyme, this note may not be out of place.

The Digestive Ferments Company manufactures and sells a digestive ferment under the trade name of "Pangestin." The manufacturers claim that it is capable of converting eighty parts of potato starch into water soluble substances in five minutes in accordance with the U. S. P. X Revision test for pancreatin. Pangestin has good solubility, with a small amount of extraneous material not identified. Both the dry enzyme and aqueous solution give a negative test for starch. Freshly made aqueous solution and the same after standing fifteen hours give a negative test for glucose. The digestive power has been tried on potato, corn and other starches in 2 per cent. aqueous paste form. Ten minutes at room temperature, and without an activator, give very positive tests for sugar with Fehling's solution. Cornstarch is apparently acted upon more rapidly than potato starch.

Pangestin is not a simple enzyme. Not only is it strongly amylolytic, but, because it is of animal origin, it is also proteolytic, as shown by its power to digest completely the white of egg, and lipolytic, as shown by the hydrolysis of oils, such as cottonseed and olive. It is therefore a mixture of enzymes. This does not, however, detract from its usefulness, but rather enhances it. So far as the writer has been able to ascertain, Pangestin is the only trade product made in America conforming to the requirements of a starch- and sugar-free enzyme, although several other chemical companies have signified their interest in the production of a purified product. One German product submitted has also been tested out, but has been found unsatisfactory.

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## SPECIAL ARTICLES

#### NITROGEN FIXATION BY BLUE-GREEN ALGAE

NITROGEN fixation, a property common to several species of bacteria and a few fungi, has long been suspected as being a property of algae also. Considerable research by several investigators with green algae (*Chlorophyceae*) has failed to demonstrate that any one of these possesses the ability to use free nitrogen gas. The few claims that have been made to the contrary have been disproved, to the satisfaction of probably all the workers in this field. The

writers have recently had occasion to check this point further. Three cultures of green algae, namely, *Chlamydomonas*, *Chlorella* and *Scenedesmus*, obtained from Cornell University through the courtesy of Dr. F. B. Wann, and a *Pleurococcus*, isolated from a local soil, were repeatedly tested for nitrogen-fixing powers, with negative results.

What are the facts regarding the nitrogen-fixing ability of the other common group of algae, known as the blue-greens (*Cyanophyceae*)? Many articles have appeared during the past forty years in which references are made to the fact that under various