

sides the extracellular digestion in the intestine there is also an intracellular digestion. However, since the life span of the absorbing cells is only 3 days⁸ autophagic lysosomes may also be present.

The small bodies are generally negative and structurally they conform to microbodies. Such bodies are frequently found in renal and hepatic cells. Recently, structures probably representing microbodies have been demonstrated also in other organs (epididymis, Sertoli cells) by AFZELIUS and NICANDER⁹. This points to a greater ubiquity of these bodies than had been assumed earlier, and their possible presence in the absorbing intestinal epithelium is, therefore, not surprising.

Zusammenfassung. Zytoplasmatische Körperchen absorbierender Zylinderzellen (Saugferkeldarm) wurden licht- und elektronenmikroskopisch auf saure Phosphatase (SP) untersucht: Zytoplasma enthält mehrere SP-

positive Körnchen (offenbar lysosomales System) sowie kleinere, SP-negative Körperchen, als «Microbodies» gedeutet. Den heterophagischen Lysosomen wird eine intrazelluläre Verdauungsfähigkeit zugesprochen.

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⁷ CH. DE DUVE and R. WATTIAUX, A. Rev. Physiol. 28, 435 (1966).

⁸ H. A. PADYKULA, Fedn. Proc. Fedn. Am. Soc. exp. Biol. 27, 873 (1962).

⁹ B. AFZELIUS and L. NICANDER, J. Ultrastruct. Res., in press.

Early Detection of Infection of Sugarcane by Ratoon-Stunting Disease (RSD)

RSD which depresses the growth of the ratoon crop of sugarcane is believed to be a virus disease and is difficult to detect before it exerts its growth-retarding effect¹. VAN KAMMEN and BROUWER² described an increase in levels of polyphenoloxidase in tobacco leaves inoculated with tobacco mosaic virus; a similar observation was recorded for maize infected with maize rough dwarf virus by HARPAZ and KLEIN³. We attempted to use this procedure for the detection of RSD in sugarcane.

Healthy leaves of sugarcane about 130 cm long were cut into sections 33 cm long and split along the midrib. One half of the split section was inoculated by dusting it lightly with carborundum and rubbing it on either side with each of two fingers wetted with inoculant. The inoculant for the test half was juice squeezed from RSD-infected cane stalks, that for the control half juice from healthy stalks of the same variety. Inoculum was rinsed off with running water, the bottom 1 cm of the half-leaves was cut off under water and the split halves were put into tubes (33 cm by 28 mm) containing 30 ml distilled water. The tops were closed with 'parafilm' and the tubes, in racks, were incubated at 24°C for various periods up to 7 days under fluorescent lights (13.5 h day). At the end of the incubation the half-leaves were removed from the tubes, blotted dry, cut up with scissors and representative samples of 1–1.5 g were ground⁴ with 15 ml McIlvaine buffer² for 10 sec; a second grinding with 7 ml for 5 sec served to rinse the machine and disperse any fibres on the head. The dispersion was made up to 25 ml with buffer, stirred in the presence of a few drops of iso-octanol and centrifuged for 20 min at 8000 g and 4°C. Polyphenoloxidase was determined with chlorogenic acid as substrate by a spectrophotometric method similar to that described in ², except that the reaction was carried out in a Perkin-Elmer 4000 A recording spectrophotometer and that the extinction values from the recordings were plotted against time on semilogarithmic paper. A straight-line relationship between log extinction

and time held for 2–3 min after the reaction was started by addition of chlorogenic acid to the test sample and the slope of this line was taken as a measure of enzyme activity. The ratio of activity/g wet weight for the test extract to the activity/g wet weight for the control extract, called the R value, was found to vary considerably from one test to the next. For 151 tests the mean R value was 1.23 and this was significantly greater than unity ($P < 0.001$). Although many modifications of each step of the method were tried we were unable to obtain more reproducible results. Thus the minimum number of replicate samples which would have to be analysed to give an accuracy in diagnosis of 9/10 and 99/100, respectively, were 29 (23) and 70 (56) for 2 groups of experimental results. In view of the large number of replicate analyses required, the method in its present form is not suitable for routine detection of RSD.

Zusammenfassung. Im Einklang mit Erfahrungen bei Tabak- und Maispflanzen konnte in Blättern des Zuckerrohres nach Impfung mit Saft von an «Ratoon Stunting Disease» erkranktem Zuckerrohr eine Erhöhung des Polyphenoloxidasen-Spiegels festgestellt werden. Eine praktische Anwendung dieser Methode zur frühen Diagnose dieser Viruserkrankung ist noch nicht möglich.

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¹ D. R. L. STEINDL, Proc. int. Soc. Sug. Cane Technol., 7th Congr. 457 (1950).

² A. VAN KAMMEN and D. BROUWER, Virology 22, 9 (1964).

³ I. HARPAZ and M. KLEIN, Experientia 20, 274 (1964).

⁴ The grinding was carried out in 50 ml polypropylene tubes (Nalge Co., Rochester, N.Y., USA) using an Ultraturax TP 18/2 (Janke & Kunkel KG, Staufen I. Br., West Germany).