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INCREASE IN POLYPHENOL OXIDASE AND PEROXIDASE WITH HIGHER INTENSITIES OF COCONUT ROOT (WILT) DISEASE

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ABSTRACT

The levels of polyphenol oxidase and peroxidase were found to increase with intensity of coconut root (wilt) disease. The correlation between phenol oxidising enzymes and coconut root (wilt) index formulated on the basis of foliar symptoms was positive.

INTRODUCTION

VASCULAR discolouration of roots is one of the characteristic symptoms of coconut root (wilt) disease (Menon and Nair, 1949). The involvement of polyphenols in vascular browning formed the basis of our earlier investigations on the estimation of total phenols in the roots of coconut palms in relation to the disease (Joseph and Jayasankar, 1973). The significant decrease in the total phenol content of the coconut roots with increase in intensities of root (wilt) disease was of interest and prompted studies on the levels of phenol oxidising enzymes. Results presented in this paper provide additional support to the previous findings (Joseph and Jayasankar, 1973).

MATERIALS AND METHODS

The disease index of the palms was recorded using the formula of George and Radha (1973).

Freshly emerging roots were collected from coconut palms and stored in the freezer overnight for facilitating extraction of enzymes. Approximately 15g of sliced root samples were macerated in a Waring blender with 250 ml of cold citrate-phosphate buffer (pH 7·0). The extract was filtered through cheese cloth and added to two volumes of chilled acetone with gentle stirring. The precipitated acetone powder preparations were dried in a vacuum dessicator in an air-conditioned room at 22–24° C and kept in a refrigerator. 250 mg of dried material extracted in 10 ml of citrate-phosphate buffer (pH 7·0) was used for enzyme assays.

Polyphenol oxidase activity was measured by a method similar to that of Matta and Dimond (1963). The reaction mixture consisted of 3·0 ml catechol (0·5%), 0·75 ml citrate phosphate buffer (pH7·0), and 0·25 ml enzyme extract in a cuvet. The optical density of the reaction product was measured at 400 mμ in a Beckman DU_2 model spectrophotometer after 5 min.

Peroxidase was estimated by the pyrogallol test (Colowick and Kaplan, 1955). The

TABLE I

Specific activity of polyphenol oxidase an peroxidase

Disease index	No. of palms examined	Polyphenol oxidase	Peroxidase
		OD/mg protein/5 min.	
0-10	27	5·18	5·15
11-50	14	10·47**	10·55
51-60	9	13·47*	11·45

* Significant at 5% level.
** Significant at 1% level.

assay system contained 2·0 ml pyrogallol solution (40 μm) 1·5, ml citrate phosphate buffer (pH 7·0), 0·5 ml hydrogen peroxide (3%), and 1.0 ml enzyme preparation. The reaction was checked by the addition of 5 ml 5 N sulphuric acid after incubation for 5 min at 37° C. The coloured purpurogallin formed was extracted twice with 5 ml each of solvent ether. The ether was evaporated at room temperature, the residue was dissolved in 5 ml ethanol, and the optical density of the solution was measured at 430 mμ in a Beckman DU_2 model spectrophotometer.

Protein was determined by the method of Lowry *et al.* (1951).

RESULTS AND DISCUSSION

The levels of polyphenol oxidase and peroxidase increased with intensity of disease. Pathogenic infection and external injuries contribute to an increase in the phenol oxidising enzymes (Table I). Similar increase also occurs in response to toxins or pectinolytic enzymes. Association of several pathogens with root (wilt) disease has been reported (Shanta and Radha, 1975). How far these factors contribute to the phenomenon remains to be elucidated.

The correlation between polyphenol oxidase and disease index in the range of 11–50 was highly significant and positive. In this range the activity of peroxidase also increased with increase in disease index but did not reach the significant level at 5%. Within the disease index of 51–60, the correlation coefficient between polyphenol oxidase and disease index was positive and significant at 5% level. The correlation between the phenol oxidising enzymes and disease index of the individual palms in the range of 0–60 (Fig. 1) is suggestive of the possibility of correlating vascular browning with foliar symptoms of coconut root (wilt) disease under more defined conditions.

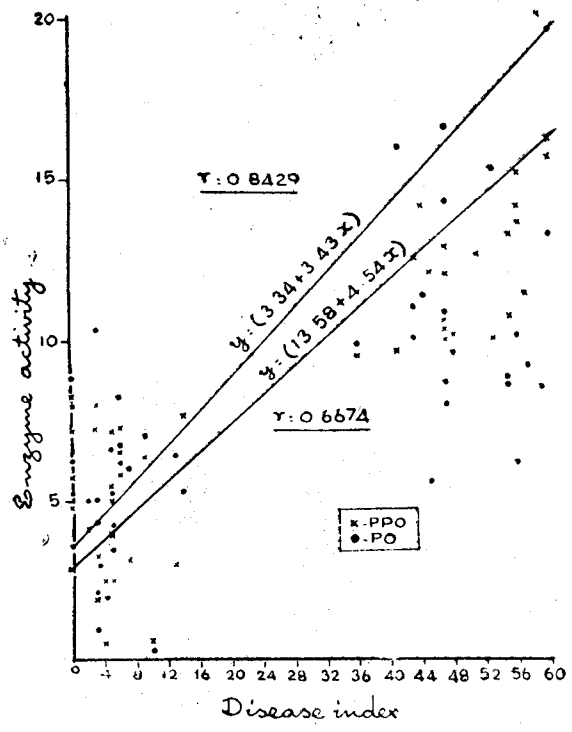

Figure 1. Correlation between phenol oxidising enzymes and disease index in the range of 0–60.

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REFERENCES

COLOWICK, S. P. AND KAPLAN, N. O. 1955. The pyrogallol test. p. 773. in *Methods in Enzymology*. Vol. II. Academic Press, New York.

GEORGE, M. V. AND RADHA, K. 1973. Computation of disease index of root (wilt) disease of coconut. *Indian J. Agric. Sci.* **43**: 366–370.

JOSEPH, K. V. AND JAYASANKAR, N. P. 1973. Polyphenol content in coconut roots in relation to root (wilt) disease. *J. Plant. Crops* **1** (Suppl.) 99–101.

LOWRY, O. H., ROSEBROUGH, N. J., FARR, A. L., AND RANDALL, R. J. 1951. Protein measurement with Folin—phenol reagent. *J. Biol. Chem.* **193**: 265–275.

MATTA, A. AND DIMAND, A. E. 1963. Symptoms of fusarium wilt in relation to quantity of fungus and enzyme activity in tomato stems. *Phytopathology* **53**: 574–578.

MENON, K. P. V. AND NAIR, U. K. 1949. The wilt (root disease) disease of coconuts in Travancore and Cochin. *Indian Coconut J.* **3**: 5–10 and 40–44.

SHANTA, P. AND RADHA, K. 1975. Recent studies on the root (wilt) disease. Paper presented at the Fourth Session of the FAO Technical Working Party on Coconut Production, Protection and Processing, Kingston, Jamaica.