

RESPIRATORY AND ENZYMATIC CHANGES IN THE SCLEROTIAL
ROOT ROT OF GROUNDNUT*

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Sclerotium rolfsii Sacc. causes a serious root rot disease of groundnut (*Arachis hypogaea* L.) in almost all parts of the world wherever groundnut is grown. The effect of infection on the respiratory rate and changes in the terminal oxidative enzymes in the hypocotyl region of groundnut plants are presented in this paper.

Materials and Methods

Groundnut plants of the variety TMV-2 and a highly pathogenic isolate of *S. rolfsii* isolated from groundnut was used as the host-pathogen combination throughout the investigations. Seedlings were raised and inoculated following the method of Wu (1967). Seeds were surface sterilised with 0.5 per cent mercuric chloride solution, washed repeatedly, finally soaked in sterile distilled water for one hour and then placed in dishes containing 100 ml of 1 per cent water, allowed to germinate and grow at room temperature ($26 \pm 2^\circ\text{C}$). Inoculation of these seedlings were done when they were 10 days old by placing uniform blocks of 3 day old oatmeal agar culture of the fungus in the hypocotyl region. The inoculated area became necrotic in 12 hours and sunken due to collapse of the invaded cells. In 48 hours the lesion increased in size and the hypocotyl tissues were completely dead in 8 days. The respiratory rate of healthy and diseased tissues were measured on the 1st day, 2nd day, 4th day and 8th day after inoculation by the direct method of Umbreit *et al.* (1964), using Warburg's manometer with side arm reaction vessels. The tissues were permitted to respire for 3 hours and readings were taken every half an hour.

Polyphenol oxidase activity was detected according to the method of Matta and Dimond (1963). Peroxidase by the method of Mudd *et al.* (1959), catalase activity by the method of Beers and Sizer (1952) and Ascorbic acid oxidase by the method of Oberbacher and Vines (1963).

Results and Discussion

A slight increase in the respiratory rate was observed immediately after inoculation (Table 1). It then gradually increased and reached the

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maximum on the 4th day after inoculation. The oxygen uptake was then decreased and was 27.4 per cent lower than the healthy tissues at the end of the 8th day after inoculation.

All the oxidative enzymes detected were also found to be on the increase in the infected tissues. The percentage increase of all of them are given in Table 2.

The maximum enhanced activity in the case of all the oxidative enzymes were found to occur on the 4th day after inoculation, there after it decreased. The change in polyphenol oxidase activity was abrupt when compared to other enzymes studied.

Table 1

Respiratory rate of **healthy and inoculated hypocotyl tissues** of groundnutplants

Sl No	Days after inoculation	U of oxygen consumed per g. of tissues (dry weight) per hour		Percentage of increase (+) or decrease (—) over healthy
		Healthy	Inoculated	
1	1	92.3	97.4	+ 5.52
2	2	99.53	114.63	+ 15.1
3	3	106.56	172.4	+ 61.7
4	4	117.66	85.33	— 27.4

Respiration occupying a central place in metabolism provides energy to support cellular processes. It is a common observation that respiration increase in plants in response to infection. In the present study the respiratory rate reached the maximum by the time of external symptom development and afterwards it declined. A similar trend in increase of respiratory activity was reported in bean hypocotyl due to infection by *R. solani* *Ceratocystis fimbriata* (Uritani and Akazawa, 1959; Wood 1967) after critically reviewing, this phenomenon of increased respiratory rate in infected plants have stated that in almost all the cases the respiration increased sharply at the time of first appearance of the symptoms and then it declined as necrosis progressed, The results of the present study also were in agreement with this statement. Increase in respiratory rate is often attributed to an alteration in the terminal oxidase or change in the usual pathway of carbohydrate breakdown. The

groundnut hypocotyl tissue registered a fairly high increase in oxidative enzymes concomitant with increase in respiration. Similar results were also obtained in *R. Solani* infection of bean (Maxwell and Bateman, 1967). Thus it is clear that the augmented respiration associated with symptom development in the sclerotial root rot of groundnut is brought about by changes in the terminal oxidative pathway.

Table 2

Changes in the activity of oxidative enzymes in the groundnut hypocotyl region due to infection by *S. rolfsii* (activity expressed as unit changes in optical density per 0.1 ml of extract, 0.001 = 1 unit)

Sl No	Days after inoculation	Type of tissue	Enzymes			
			Oxidative Polynuclear oxidase	Peroxidase	Catalase	oxidase
1	1	A	12	146	120	210
		B	19	148	140	270
		C	58.3	1.36	16.60	24.0
2	2	A	13	155	140	212
		B	30	190	160	230
		C	130.7	23.20	14.28	36.79
3	4	A	15	160	150	220
		B	40	200	190	320
		C	166.6	25.0	26.60	45.40
4	8	A	15	158	150	220
		B	10	190	180	248
		C	33.3	20.20	20.60	12.72

A = Healthy

B = Infected

C = Percentage increase over healthy

Summary

Infection of groundnut hypocotyl region by *Sclerotium rolfsii* was found to augment the respiratory rate of the host tissue. The respiratory rate reached the maximum during the symptom development stage and at this stage the activity of all the important terminal oxidative enzymes were also found to be enhanced. The results are discussed as proof of the role of increased oxidative enzyme activity in enhancing the respiratory rate of infected groundnut plants.

സംഗ്രഹം

സ്കൂളിറോഷ്യം റോലഫ്സി എന്ന ഫംഗസിന്റെ ആക്രമണം ഏറ്റവും നിലക്കടലയുടെ ബീജപത്രാധര കോശങ്ങളുടെ ശ്വാസോച്ഛ്വാസ തോത് വളരെ ഏറെ വർദ്ധിപ്പിക്കുകയുണ്ടായി. രോഗ ലക്ഷണങ്ങൾ പ്രത്യക്ഷപ്പെടുന്നതിനോടൊപ്പമാണ് ഈ വർദ്ധനവ് ഏറ്റവും മുൻപ് ധന്യത്തിൽ എത്തിയത്. ഇപ്രകാരം ശ്വാസോച്ഛ്വാസ തോത് വർദ്ധിപ്പിക്കുവാനുള്ള കാരണം ഓക്സീകരണ ഏൻസൈമുകളുടെ അധികരിച്ച പ്രവർത്തനമാണെന്ന് തെളിഞ്ഞു.

REFERENCES

- Bateman, D. F. and Daly, J. M. 1967. The respiratory pattern of *Rhizoctonia solani* infected bean hypocotyle in relation to lesion maturation. *Phytopathology* 57, 127-131.
- Beers, R. F. and Sizer, J. M. 1952. A spectrophotometric method for measuring the breakdown of hydrogen peroxide by catalase. *Biol. Chem.* 195, 133-140.
- Matta, A. and Dimond, A. E. 1963. Symptoms of Fusarium wilt in relation to quality of fungus and enzyme activation in tomato stem. *Phytopathology* 53, 574-575.
- Maxwell, D. P. and Bateman, D. F. 1967. Changes in activities of some oxidases in extracts of *Rhizoctonia* infected bean hypocotyl in relation to lesion maturation. *Phytopathology* 57, 132-136.
- Mudd, J. B., Johnson, B. G., Burris, R. M. and Buchholts, K. P. 1959. Oxidation of Ir.dole-acetic acid by quick grass rhizomes. *Plant Physiol.* 34, 144-148.
- Oberbacher, M. F. and Vines, H. M. 1963. Spectrophotometric assay of ascorbic acid oxidase. *Nature, Lond.* 97, 1203-1204.
- Umbreit, W. W. Burris, R. H. and Stauffer, J. F. 1964. *Manometric techniques*. Burges Publishing Co. Minnesota.
- Uritani, I. and Akazawa. 1959. Alteration of the respiratory pattern in infected plants. In *Plant Pathology* Vol. 1. (eds). Horsfall, J. G. and Dimond, A. E. pp 349-390. Academic Press New York.
- Wood, R. K. S. 1967. *Physiological Plant Pathology*. Blackwell Scientific Publications, Oxford and Edinburgh.
- Wu, L. C. 1967. Physiology of parasitisms. 2. Biochemical changes in the mung bean seedlings infected with *Rhizoctonia solani* Kühn. *Bot. Bull. Academia Sinia* 6, 144-152.