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EFFECT OF HYDROLYTIC ENZYMES IN THE PATHOGENESIS OF SCLEROTIAL ROOT ROT OF PEANUT*

P. AXANDAVALLY AMMA, N. SHANMUGAM and M. CHANDRASEKHARAN NAIR
College of Agriculture, Coimbatore, Tamil Nadu

The root rot of peanut (*Arachis hypogaea* L.) caused by *Sclerotium rolfsii* Sacc. is of common occurrence in many parts of India. The cell wall macerating activity of this pathogen was attributed to the high potency of its endopolygalacturonase activity (Bateman 1968, 1972). An important aspect which affects the role of polysaccharide degrading enzymes in plant pathogenesis is the control of the production of such enzymes (Albersheim *et al.* 1969). The type and concentration of mono and polysaccharides in the immediate vicinity of the site of action of the pathogen affects both quantitatively and qualitatively the secretion of such enzymes. In the present study the production of endopolygalacturonase and cellulase (Cx) by the pathogen isolated from peanuts was studied both *in vitro* and *in vivo*. The *in vitro* production in a medium with cell wall material as sole carbon source was also studied,

Materials and Methods

The enzyme extract from both the culture filtrate and from infected plant materials were prepared by following the methods of Bateman (1963). Cell wall material was prepared from 10 days old and 30 days old hypocotyl tissues of peanut following the method of Nevins *et al.* (1967). The viscometric methods described by Uritani and Stahmann (1961) for endopolygalacturonase and Barker and Walker (1962) for cellulase (Cx) were followed in the present study also without any modifications. Each detection was repeated three times and the averages are presented in the tables.

Results and Discussion

Production of *endopolygalacturonase (Endo-PG)*

The isolate S. *rolfsii* from peanut produced the enzyme endo-PG both in culture as well as in the infecled peanut hypocotyl tissues. The *in vitro* and *in vivo* production of the same during different incubation periods are presented

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in Table 1. The endo-PG activity was found to be more during the early stages of pathogenesis. In the culture also this enzyme was secreted in the early periods of incubation. As the disease advanced the enzyme activity declined.

Table 1

***In vitro* production of endopolygalacturonase by S. *rolfsii* in culture during different incubation periods and its *in vivo* production in inoculated hypocotyl tissues during different periods after inoculation**

Interval in days	Production of enzyme-expressed as percentage reduction in viscosity from that of control	
	In vitro	*In vivo*
4	62.85	36.90
8	47.90	36.15
12	44.80	24.05
16	30.25	20.42
20	18.60	17.40
In vitro activity	S. E. — 1.38,	C. D. = 4.25
In vivo activity	S. E. = 0.90,	C. D. - 2.77

Table 2

In *vitro* production of cellulase (Cx) by S. *rolfsii* in culture during different incubation periods and its production *in vivo* in inoculated peanut hypocotyl tissues during different periods after inoculation

Interval in days	Production of cellulase (Cx) expressed as percentage reduction in viscosity from that of control	
	In vitro	*In vivo*
4	42.20	11.74
8	56.39	19.45
12	42.89	20.27
16	33.35	9 45
20	31.40	9.37
In vitro activity	C. D. = 1.94	S. E. = 0.63
In vivo activity	C. D. = 0.68	S. E. = 1.24

Cellulase (Cx) activity

Cellulase activity could also be detected both *in vitro* and *in vivo* (Table 2). The maximum *in vitro* activity was noted on the 8th day of incu. bation. In the inoculated hypocotyl tissue the activity was found to increase abruptly on the 8th day after inoculation registering the maximum on the 12th day. Effect of cell wall material as sole carbon source in enzyme secretion,

The pathogen was found to secrete both endo-PG and cellulase (Cx) in media containing cell wall material isolated from the peanut hypocotyl tissue as sole carbon source. Activites of both endo-PG and cellulase was maximum in the medium with the cell wall materials from the 10 day old hypocotyl tissues than from those of 30 days old (Table 3)

Table 3

Enzyme production in media with cell wall material as sole carbon source (expressed as percentage reduction in viscosity than that of the control)

Name of enzyme	Production in media with cell wall materfai of different ages	
	10 days old tissue	30 days old tissue
Endo-PG	45.03	38,72
Cellulase (Cx>	31.36	22.75

The mechanism of pathogenesis of *Sderotium rolfsii* have been investigated in detail and the role of enzymes on the same is a well documented phenomenon (Bateman 1968; Bateman and **Beer** 1965; Van Etten and Bateman 1969). However, reports about the production and role of the same by the peanut isolate was lacking. In the present study the peanut isolate was found to have good endopolygalacturonase (endo-PG) and cellulase (Cx activity. The organism was also found to secrete these enzymes in media with the peanut hypocotyl cell wall material as the sole carbon source. The hypocotyl region is the most vulnerable site of infection in peanuts by this organism. The secretion of relatively large quantities of these enzymes, when the fungus was cultured on cell walls isolated from this highly susceptible region, constitute on pathogenesis. The main macerating agent of this pathogen is known to be endopolygalacturonase (Bateman 1968). Very high endo-PG obtained in the initial stages in culture as well as in the inoculated hypocotyl tissues showed that this is the main enzyme involved in the early stages of pathogenesis of sclerotial root rot disease of peanut. Cellulase was present only at a later stage of pathogenesis which has been shown to be true for *S. rolfsii* infections of bean also by Bateman (1969),

When cell wall material was used as a sole carbon source the enzyme secretion was found to be maximum with younger tissues. It is a common observation with this disease that the younger plants are more susceptible than older ones to infection by this organism (Subramanian 1960) The present finding that enzyme secretion is more with cell wall materials from younger tissues explains; why younger plants are more susceptible to infection by the pathogen than the older ones.

Summary

An isolate of *Sclerotium rolfsii* Sacc. from peanut was found to produce the enzymes endopolygalacturonase (endo-PG) and cellulase (Cxi both in culture as well as in the inoculated peanut hypocotyl tissues. Endo-PG secretion was more during the early stages of growth and pathogenesis. On the other hand high cellulase activity was recorded during advanced stages of pathogenesis and in old cultures. These enzymes were also secreted with cell wall material isolated from the hypocotyl tissues as sole carbon source. The results are discussed as proof of the role of these enzymes in the sclerotial root rot disease of peanuts.

സംഗ്രഹം

സ്ക്ലീറോഷിയം എന്ന കുമിളിൽ കാണുന്ന 'എൻസോ പോളിഗാലക്ടുണേസ്', 'സെല്ലുലേസ്' എന്നീ എൻസൈമുകളെ കുറിച്ചുള്ള വിശദപഠനങ്ങളിൽ ഇവ പീനട്ടിന്റെ വേര് ചീയൽ രോഗത്തിനു കാരണമാണെന്നു തെളിഞ്ഞു.

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