OCCURRENCE OF COMMON ANTIGENS IN JUTE AND COLLETOTRICUM CORCHORI

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ANTIGENIC similarities exist between susceptible hosts and their respective parasites\(^1\)\(^,\)\(^2\). The possible involvement of common antigens as a basic compatibility factor has been implicated\(^3\)\(^,\)\(^4\), with a corollary that resistant or incompatible reaction occurs only when common antigens are absent between the host and the pathogen.

An attempt was therefore made to determine the serological relationship between Colletotrichum corchori Pavgi and Singh, a virulent foliar parasite of jute and susceptible and resistant jute cultivars.

Antigens of an isolate of C. corchori, a susceptible cultivar (JRC112) and a resistant cultivar (JRC 321) of jute were prepared\(^6\). Fungal mycelium, 50 g (fresh weight) was crushed with sea sand and the soluble protein extracted with 0.05 M Tris-HCl buffer pH 7.4. The cell debris was removed by centrifugation at 8500 g for 20 min. followed by centrifugation at 105,000 g for 1 hr. The protein separated by precipitation with ammonium sulphate at 30%, 60% and 90% saturation levels. The precipitated protein was purified by dialysis against the same buffer and quantity of protein present was measured\(^6\). For the preparation of plant antigens, 30 g (fresh weight) of jute leaves were used. Antiserum was also prepared against fungal antigen by intramuscular injection to albino rabbit with 1 mg fungal antigen/ml, emulsified with the same volume of Freund’s complete adjuvant. The dose was repeated at 7-day interval with Freund’s incomplete adjuvant for four consecutive weeks. The animal was bled two days after the last injection from border vein of the ear, serum collected, centrifuged at 5000 r.p.m. for 10 min at 4\(^\circ\)C and the supernatant was used as fungal antiserum. Normal serum was also obtained from the rabbit’s ear before immunisation.

Double immuno-diffusion tests in agar gel was carried out\(^7\). Clear agar medium (1% agar, 0.1% Na\(_2\)SO\(_4\), dissolved in 0.05M barbitone buffer, pH 8.4) was poured in 5 cm Petri dishes (5 mL/petri dish) and allowed to solidify. Wells (3 mm diameter) were cut in the medium using a sterile cork borer at a distance of 4 mm from each other. Finally undiluted antiserum of the pathogen was tested against both homologous and heterologous antigens. Titre values of the antiserum and antigen of the pathogen were 8 and 16, respectively. Cross reactivity test was carried out for 72 hr at 30\(^\circ\)C. The results are presented in Table 1 and figure 1.

**Table 1** Immunodiffusion tests with fungal (C. corchori) and plant antigens (jute cultivars JRC 212 and JRC 321) and fungal antiserum

<table>
<thead>
<tr>
<th>Antigens of host/pathogen</th>
<th>Antiserum Normal serum (NS)</th>
<th>Antiserum Fungal serum (FS)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Fungal antigen (Fa) (C. corchori)</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Plant (Jute) antigen</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Susceptible cv JRC212 (Sa)</td>
<td>-</td>
<td>+</td>
</tr>
<tr>
<td>Resistant cv JRC321 (Ra)</td>
<td>-</td>
<td>-</td>
</tr>
</tbody>
</table>

+ presence of precipitation reaction; - absence of precipitation.

**Figure 1.** Agar-gel double diffusion test using antigens of host and parasite and antiserum of parasite. NS—normal serum, FA—fungal antiserum; Fa—fungal antigen; Sa—antigen of susceptible host (cv JRC 212); Ra—antigen of resistant host (cv JRC 321).
Distinct precipitin reactions occurred when fungal antiserum was reacted against its own antigen and antigen of susceptible cultivar (JRC 212). However, no precipitin line developed when the antiserum was tested against antigen of the resistant cultivar (JRC 321). The normal serum (collected before immunisation) showed no precipitin band with any of the antigens tested.

The results of agar-gel double diffusion test confirm that common antigenic relationship exists between the susceptible jute cultivar and the foliar pathogen C. corchori. Pathogenicity test of the isolate of C. corchori also revealed that it is virulent on cv. JRC 212 while JRC 321 is moderately resistant. The precipitin reaction is usually observed within 24 hr. In the case of jute, however, the reaction occurred only after 72 hr. The delayed response may be attributed to the slow rate of diffusion of the host antigen because precipitation was observed within 24 hr when reactivity was tested between the fungal antigen and its own antiserum.

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PROTEOLYTIC ACTIVITY IN THE GUT OF
RED COTTON BUG, DYSDERCUS
CINGULATUS FABR. AND ITS PREDATOR,
ANTILOCHUS COQUEBERTII (FABR.)
(HETEROPTERA: PYRRHOCORIDAE)

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The role of digestive proteinases of insects has been extensively studied and reviewed by many workers. Insects generally contain trypsin-like enzymes which act at neutral or alkaline optima. The occurrence of pepsin-like enzymes is rare among insects.

The present communication describes the nature of proteolytic enzymes of two species of bugs having phytophagous and carnivorous feeding habits.

The red cotton bug Dysdercus cingulatus and its predator Antiolochus coquebertii were collected from silk cotton tree Bombax ceiba Linn and reared in the laboratory at room temperature (32–34°C) and humidity (60–75% R.H.) D. cingulatus was fed on soaked cotton (Gossypium sp) seeds. All nymphal instars and adults of A. coquebertii were given eggs, nymphs and adults of Dysdercus as food. The newly emerged males and females of the two species were sorted out age wise and kept separately. Both fed and starved individuals were used for the experiments. For starved lot, both sexes were allowed to starve for 48 hr. Another lot was fed daily for comparison.

Insects were immobilized by freezing and dissected out in chilled insect physiological saline. At least 16 guts were pooled and used for a single observation. The tissues were weighed and homogenized in phosphate buffer pH 7.0 and 0.1 M KCl-HCl pH 2.0. The enzyme activity and the total proteins of the homogenates were determined. To determine acidic and neutral proteolytic activity, assays were performed at pH 2.0 and pH 7.2 respectively using haemoglobin and casein as substrate.

Results obtained on proteinase activity in the gut of D. cingulatus and A. coquebertii are summarized in Table 1. Highest specific activity was recorded at pH 2.0 in the starved males of D. cingulatus (Table 1). It was higher than that of the females. The proteinase activity in D. cingulatus recorded at pH 7.0 is roughly half of that found at pH 2.0.

In A. coquebertii, enzyme activity is recorded only at pH 2.0 in both the sexes and it is higher in the males than in females. No proteinase activity was found at pH 7.0 in A. coquebertii (Table 1).