Biochemical changes in susceptible and resistant black gram cultivars induced by root-knot nematode, *Meloidogyne incognita*

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ABSTRACT

Biochemical changes in black gram varieties inoculated with root-knot nematode, *Meloidogyne incognita* were investigated. Observations were recorded in the biochemical modifications relating to various parameters like total chlorophyll, total sugar contents, protein, and proline content during post infection periods. The variation in total chlorophyll, total protein, proline and total sugar content in six cultivars i.e. PU 09-36(S), MU-44(S), VBG 11-031(R), VBG 11-016(R), KUG-715 (R)and NUL-205(R) were studied 45 days after inoculation. Reduced percentage of total chlorophyll contents were observed in inoculated samples than the healthy counterparts. However, an increase in amount of total protein, proline and total sugar contents was observed in the diseased tissues.

Key words: Black gram, Biochemical modifications, *Meloidogyne incognita*.

INTRODUCTION

Root-knot nematodes like many other obligate parasites are capable of disturbing the host metabolism. The changes in the physiological and biochemical processes of infected host as consequences of disturbed metabolism decide whether the host becomes susceptible or resistant to nematode attack (Krusberg, 1963). In this context an intimate knowledge of nematode physiology and biochemistry along with its host is absolutely essential for developing plant resistance against the nematodes. In the recent past some progress has also been made in this direction to understand the basic biochemical mechanism of plant-nematode interactions by several workers (Ganguly and Dasgupta, 1983; Howell and Krusberg, 1966; Mohanty, et al., 1995; Nayak, 2015). Considering the importance of the subject, the present investigation was undertaken to find the changes if any, in total sugar content, chlorophyll contents, protein and proline contents in relation to black gram inoculated with root-knot nematode, *Meloidogyne incognita*.

MATERIALS AND METHODS

In order to understand the basis of nematode resistance six varieties namely PU 09-36, MU-44 (Highly susceptible check) and KUG-715, NUL-205, VBG 11-031, VBG-11-016 (Resistant check) were grown in earthen pots in the greenhouse. These plants were washed 30 days after nematode inoculation and the following chemical compositions were estimated.

- Chlorophyll content (Spectro-colourimeter Method)
- Total protein content (Lowry Method)
- Total sugar content (Spectro-colourimeter Method)
- Total proline content (Spectro-colourimeter Method)

Seeds of black gram cultivars were sown in 15cm diameter earthen pots filled with steam sterilized soil. A week after germination, one seedling per pot was retained and inoculated with axenised suspension of approximately 1000 J2 of *M. incognita* per seedling per pot. A set of plants was left uninoculated to serve as control. Healthy and inoculated plants were harvested at 45 days after inoculation. The harvested roots were washed thoroughly under running tap water to remove the adhering soil particles. Then the shoots were surface sterilized in 1% HgCl2 solution followed by repeated washing with distilled water. The harvested shoots were dried on blotting paper and kept separately for biochemical analysis. The total chlorophyll content was estimated by Spectro-colourimeter method. Total sugar and proline contents were estimated by following Spectro-colourimeter methods. Modified micro Kjeldahls method was used for estimation of Crude protein and nitrogen content of shoot were estimated by following the procedure of Mahadevan and Sridhar (1986).

RESULTS AND DISCUSSION

Many kinds of chemicals are present in the plant system that influences the metabolism of the pests fed upon them. The chemicals may consists of simple nitrogen, phosphorus, potassium, carbohydrates and fats to complex proteins, enzymes, phenolic compounds etc., which may nourish, starve or kill the feeding organisms upon them. In the course of feeding some chemicals may be depleted or some others may be synthesized, de nova, that may be detrimental to the pest. In order to know the chemical and genetic basis of resistance, six varieties were chosen for a
detailed analysis. These plants were grown with utmost care, both in inoculated and control conditions. One set of each uninoculated (healthy) and inoculated (infected) plants were analysed to test the effects of root-knot nematode infection on the growth and vigour of the plants and their root-system.

**Effect of nematode infection on chlorophyll content:** It was observed that the chlorophyll ‘a’ content reduced from 15.253 to 14.500 mg/g due to nematode infection in variety PU-09-35 (Table 1). Likewise in other varieties MU-44, VBG 11-031, VBG 11-016, NUL-205 chlorophyll ‘a’ content reduced from 18.765 to 12.561, 19.897 to 14.257, 17.415 to 12.561 and 18.182 to 12.695 mg/g of leaf respectively. Only in variety KUG-715 chlorophyll ‘a’ content increased from 13.458 to 15.173 mg/g of leaf.

In case of chlorophyll ‘b’ content reduction from healthy to infected plants recorded were 11.294 to 10.498, 13.820 to 9.752, 14.694 to 10.690, 13.115 to 9.196, 13.750 to 9.626 mg/g in varieties PU 09-36, MU-44, VBG 11-031, VBG 11-016 and NUL-205 respectively. Only in variety KUG-715 the increase in chlorophyll ‘b’ content was marked from 13.458 to 15.173 mg/g of leaf.

Similarly total chlorophyll content was decreased from 11.435 to 10.660 in variety PU 09-36, 15.901 to 14.002 mg/g in variety MU-44, 14.882 to 10.806 mg/g in variety VBG 11-016, 13.250 to 9.324 mg/g in variety VBG 11-016 and 13.884 to 9.717 mg/g in variety NUL-205 except in variety KUG-715 there was increase of total chlorophyll ranged from 13.612 to 15.310 mg/g.

**Effect of nematode infection on protein content:** Due to infection of root-knot nematode, the protein content increases in infected cultivars as 0.103, 0.231, 0.351, 0.239, 0.151 and 0.202 mg/100g in varieties PU 0-36, MU-44, VBG 11-031, VBG 11-016, KUG-715, and NUL-205 compared to 0.069, 0.106, 0.330, 0.178, 0.092 and 0.141 in varieties PU 0-36, MU-44, VBG 11-031, VBG 11-016, KUG-715, and NUL-205 and also the protein percentage was highest 64.13 percent in case of KUG-715 (Table 2).

**Effect of nematode infection on total sugar content:** The amount of sugar present in the inoculated plants were recorded as 0.132, 0.081, 0.095, 0.050, 0.048 and 0.077 percent in the varieties PU 0-36, MU-44, VBG 11-031, VBG 11-016, KUG-715, and NUL-205 respectively on fresh weight basis (Table 3). Conversely this amount was decreased in all the cases in healthy plants i.e., 0.089, 0.054, 0.077, 0.038, 0.027, 0.073 percent in the shoot portion of these varieties.

**Effect of nematode infection on proline contents:** The amount of proline in the uninoculated plants were recorded as 19, 35, 45, 49, 52, and 43 µg/g on fresh weight basis in varieties PU 0-36, MU-44, VBG 11-031, VBG 11-016, KUG-715, and NUL-205 (Table 4). while this was increased in inoculated infected plants as 24, 47, 52, 58, 64 and 47 respectively.

### Table 1: Chlorophyll content (a, b, total) of black gram varieties due to the infection of root knot nematode

<table>
<thead>
<tr>
<th>Variety</th>
<th>Chlorophyll ‘a’ content mg/g of leaf</th>
<th>Chlorophyll ‘b’ content mg/g of leaf</th>
<th>Total chlorophyll in leaf mg/g</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Healthy (H)</td>
<td>Infected (I)</td>
<td>Mean</td>
</tr>
<tr>
<td>CD (0.05)</td>
<td>0.010</td>
<td>0.434</td>
<td>0.043</td>
</tr>
</tbody>
</table>

### Table 2: Protein content of *M. incognita* infected (I) and healthy (H) black gram varieties.

<table>
<thead>
<tr>
<th>Variety</th>
<th>Protein content (mg/100g) % on dry weight basis</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Healthy leaves</td>
</tr>
<tr>
<td>PU 09-36</td>
<td>0.069</td>
</tr>
<tr>
<td>MU-44</td>
<td>0.106</td>
</tr>
<tr>
<td>VBG 11-031</td>
<td>0.330</td>
</tr>
<tr>
<td>VBG 11-016</td>
<td>0.178</td>
</tr>
<tr>
<td>KUG-715</td>
<td>0.092</td>
</tr>
<tr>
<td>NUL-205</td>
<td>0.141</td>
</tr>
<tr>
<td>CD(0.05)</td>
<td>0.009</td>
</tr>
</tbody>
</table>
Table 3: Total sugar content in leaves of Blackgram varieties infected by root-knot nematode as compared to control plants.

<table>
<thead>
<tr>
<th>Variety</th>
<th>Healthy leaves (H)</th>
<th>Infected leaves (I)</th>
<th>Mean</th>
<th>% increase (+) decrease (-) over Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>PU 09-36</td>
<td>0.089</td>
<td>0.132</td>
<td>0.155</td>
<td>48.31</td>
</tr>
<tr>
<td>MU-44</td>
<td>0.054</td>
<td>0.081</td>
<td>0.108</td>
<td>50.00</td>
</tr>
<tr>
<td>VBG 11-031</td>
<td>0.077</td>
<td>0.095</td>
<td>0.133</td>
<td>23.37</td>
</tr>
<tr>
<td>VBG 11-016</td>
<td>0.038</td>
<td>0.050</td>
<td>0.063</td>
<td>31.57</td>
</tr>
<tr>
<td>KUG-715</td>
<td>0.037</td>
<td>0.048</td>
<td>0.042</td>
<td>29.72</td>
</tr>
<tr>
<td>NUL-205</td>
<td>0.073</td>
<td>0.077</td>
<td>0.075</td>
<td>5.47</td>
</tr>
<tr>
<td>CD(0.05)</td>
<td><strong>0.0076</strong></td>
<td><strong>0.0074</strong></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Table 4: Proline content in the leaves of Blackgram varieties infected (I) with root knot nematodes as compared to control (H)

<table>
<thead>
<tr>
<th>Variety</th>
<th>Healthy leaves (H)</th>
<th>Infected leaves (I)</th>
<th>Mean</th>
<th>% increase (+) decrease (-) over Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>PU 09-36</td>
<td>19</td>
<td>24</td>
<td>33.50</td>
<td>26.31</td>
</tr>
<tr>
<td>MU-44</td>
<td>35</td>
<td>47</td>
<td>41</td>
<td>34.2</td>
</tr>
<tr>
<td>VBG 11-031</td>
<td>45</td>
<td>52</td>
<td>48.5</td>
<td>15.5</td>
</tr>
<tr>
<td>VBG 11-016</td>
<td>49</td>
<td>58</td>
<td>53.5</td>
<td>18.36</td>
</tr>
<tr>
<td>KUG-715</td>
<td>52</td>
<td>64</td>
<td>58</td>
<td>23.07</td>
</tr>
<tr>
<td>NUL-205</td>
<td>43</td>
<td>47</td>
<td>45</td>
<td>9.30</td>
</tr>
<tr>
<td>CD(0.05)</td>
<td><strong>4.74</strong></td>
<td><strong>5.50</strong></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

CONCLUSION

The present investigation clearly indicated that *Meloidogyne incognita* played a key role in altering the normal physiology and biochemical processes of the tested host plant. Further, it is opinion that the basic information provided in this investigation will certainly be helpful to understand the complicated areas of the biochemical mechanisms of plant nematode-interaction in blackgram relating to root-knot and other plant parasitic nematodes.

REFERENCES


