Pigment analysis studies with reference to leaf reddening in Bt cotton

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Received : July, 2010; Accepted : August, 2010

SUMMARY
Healthy cotton leaves have maximum chlorophyll content as compared to all reddening affected leaves. The reduction of chlorophyll content in reddening affected leaf was maximum in MRC-6301 Bt cotton variety. Anthocynine content in reddening affected leaves was maximum in all the Bt cotton varieties as compared to healthy leaves of Bt cotton varieties at all the growth stages. Anthocynine content increased in reddening affected leaves in the range of 4.94 to 5.06% at square formation stage and in general similar position was observed at rest of the growth stages.

Key words : Reddening, Chlorophyll, Anthocynine

In Bt. Cotton (Gossypium hirsutum) the reddening of leaves may appear at the square formation stage to boll development stage leading to great loss in yield. The yield reduction are reported by 30-60% depending upon the variety and reddening intensity (Bhatt and Patil, 1976). The information on pigment analysis is meagre. Attempts were made here to estimate what are the changes occur in chlorophyll content and in antocynine content in leaf during leaf reddening.

MATERIALS AND METHODS
Present investigation was undertaken on the field of Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola during the Kharif season of 2006 – 2007. Three cotton varieties, which were studied under this investigation, were TCHH-4, MRC-6301 and NCS-145. In these three varieties Bt gene was introduced, therefore, Bt gene introduced varieties were treated as Bt.

The observations were recorded at three stages i.e. square formation stage, boll development stage and boll bursting stage. Depending upon the reddening leaves were classified into two categories viz., healthy and reddening affected leaves as suggested by snell and snell (1959). For these observations top fifth leaf was taken.

The chlorophyll content in leaf were estimated with DMSO (Dimethyl sulphoxide) method by adopting the procedure given by Hiscox and Israelston (1979).

Fresh leaf discs weighting 37.5 mg were placed in 10ml of extractant and held for 2 hr. at 60°C. The supernant was used for estimation of pigments. The optical density of the aliquot was measured on spectronic- 20 at the wavelength of 652nm with red filter. The total chlorophyll content of leaves was calculated as per, Arnon (1949) equations and was expressed in (mg/g).

Anthocynine content in leaf (ppm):

Fully developed red leaf was selected for the extraction of anthocynine content of in leaf of Bt and Non-Bt cotton varieties. The extraction of anthocynine from fully developed red leaf was done with DMSO (Dimethyl sulphoxide) method by adopting the procedure given by Hiscox and Israelstan (1979). Stock solution of pure anthocynine having the strength of 25 ppm was prepared and by using these 25 ppm stock solutions of different ppm concentrations of 10 ml volume were prepared in the range of 1 ppm, 5 ppm, 10 ppm and 15 ppm. The optical density of these known concentration solutions was measured on Spectronic – 20 and accordingly standard curve was prepared by using these standard curve of unknown solutions.

RESULTS AND DISCUSSION

The results obtained from the present investigation as well as relevant discussion have been presented under following heads:

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Total chlorophyll content of leaf (mg/g):
Changes in chlorophyll content of the leaves are given in the Table 1. Pigment analysis of the leaves from Bt. Cotton varieties viz., TCHH-4, MRC-6301, and NSC-145 at different growth stages at square formation stage, boll development stage and boll bursting stage of healthy and reddening affected leaves indicated that there was abrupt decrease in chlorophyll content with the succeeding growth stages. Initially there was steep fall in the chlorophyll content of the healthy as well as the reddening affected leaves at boll development stage. While at the boll bursting stage drastic reduction in the chlorophyll content of leaves was observed in all the Bt. Cotton varieties. The reduction of chlorophyll content in reddening affected leaves was maximum in MRC-6301.

In general progressive chlorophyll reduction in full red leaves was observed from 35 to 46%. Taneja et al. (1984) also observed the progressive chlorophyll reduction from 26 – 54%. Similar results were also obtained by Chimmad et al. (1998).

Anthocynine content of leaf (ppm):
Anthocynine content in reddening affected leaves was maximum in all the varieties as compared to healthy leaves at all the growth stages (Table 2). Anthocynine content increased in reddening affected leaves in the range of 4.94 to 5.06% at square formation stage and in general similar position was observed at rest of the growth stages, therefore, it can be concluded that as the anthocynine level increase in green leaf of Bt cotton genotypes turn into red colour appearance. However, the anthocynine content in reddening affected leaves of MRC-6301 cotton variety was maximum as compared to reddening affected leaves of NSC-145 cotton variety. This finding is in agreement with Perumal and Subramaniam (1979), Chimmad et al. (1998), Chimmad and Panchal (1998) and Edreva et al. (2002).

### Table 1: Chlorophyll content of leaf (mg/g)

<table>
<thead>
<tr>
<th>Stages</th>
<th>TCHH-4 H</th>
<th>RA</th>
<th>MRC-6301 H</th>
<th>RA</th>
<th>NSC-145 H</th>
<th>RA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Square formation stage</td>
<td>2.07</td>
<td>1.19</td>
<td>2.03</td>
<td>1.18</td>
<td>2.09</td>
<td>1.20</td>
</tr>
<tr>
<td>Boll development stage</td>
<td>1.53</td>
<td>0.96</td>
<td>1.51</td>
<td>0.94</td>
<td>1.57</td>
<td>0.98</td>
</tr>
<tr>
<td>Boll bursting stage</td>
<td>1.36</td>
<td>0.71</td>
<td>1.32</td>
<td>0.67</td>
<td>1.37</td>
<td>0.73</td>
</tr>
</tbody>
</table>

### Table 2: Anthocynine content of leaf (ppm)

<table>
<thead>
<tr>
<th>Stages</th>
<th>TCHH-4 H</th>
<th>RA</th>
<th>MRC-6301 H</th>
<th>RA</th>
<th>NSC-145 H</th>
<th>RA</th>
</tr>
</thead>
<tbody>
<tr>
<td>Square formation stage</td>
<td>3.85</td>
<td>8.91</td>
<td>3.90</td>
<td>8.96</td>
<td>3.76</td>
<td>8.77</td>
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<tr>
<td>Boll development stage</td>
<td>4.92</td>
<td>10.85</td>
<td>5.12</td>
<td>10.98</td>
<td>4.90</td>
<td>10.92</td>
</tr>
<tr>
<td>Boll bursting stage</td>
<td>5.92</td>
<td>12.72</td>
<td>5.97</td>
<td>13.40</td>
<td>5.88</td>
<td>12.52</td>
</tr>
</tbody>
</table>

**REFERENCES**


