such as those identified in the fragment amplified with the AB-nested primers have been reported to account for a substantial fraction of intraspecific cpDNA variation (Zurawski and Clegg 1987), presumably because such length changes can be tolerated in noncoding regions. When such sequence variation is assessed only by length comparisons as with the AB fragment, it is not possible to relate the observed length differences to specific mutational events. By sequencing 200 bp of the approximately 1000 bp AB fragment, the insertion/deletion events that resulted in the two length polymorphisms were defined. However, since seven length variants were identified for the entire AB fragment, complete sequence information for these seven fragments would likely provide useful information to further discriminate among the chloroplast types. If these seven AB length variants had been included in the haplotype analysis, 11 different haplotypes would have been identified.

From the Los Alamos National Laboratory, Los Alamos, New Mexico (Brettin) and the Department of Horticulture, Michigan State University, East Lansing, MI 48824 (Karle. Crowe, and lezziou). The authors thank Dr. Frederique Santi for generously providing leaves of the French sweet cherry selections. Address correspondence to Dr. Amy lezziou at the address above or e-mail: lezziou@pilot.msu.edu.

© 2000 The American Genetic Association

References


Received March 30, 1999
Accepted August 18, 1999

Corresponding Editor: David B. Wagner

A Gene for Leaf Necrosis in Chickpea (Cicer arietinum L.)

P. M. Gaur and V. K. Gour

Necrosis of leaves was observed in the glabrous mutant (ICC 15566) of desi chickpea (Cicer arietinum L.). It was characterized by drying of leaflet margins to drying of complete leaflets of older leaves. The oldest leaves were the most affected and the intensity of necrosis decreased toward the apical meristem. A single recessive gene, designated nec, was found to govern the necrotic characteristic. The nec locus was linked to gl (glabrous shoots) with a map distance of 16 ± 3 cM. The loci slv (simple leaves), mlv (multipinate leaves), nlv (narrow leaflets), hg (prostrate growth habit), P (pink corolla), and shp (round seed shape) segregated independently of nec.

A glabrous mutant of chickpea (Cicer ar-
free from necrosis. The necrotic plants produced only a few pods.

All F₁ plants were normal, healthy, and vigorous. In the F₂, the frequencies of healthy and necrotic plants closely fit a 3:1 Mendelian ratio (Table 1), indicating that necrosis is governed by a single recessive nuclear gene. The gene symbol nec is proposed for this trait.

The linkage relations of nec were determined with several other morphological trait loci including Gl/gl, pubescent/glabrous shoot (Pundir and Reddy 1989); St/lv, normal pinnate/simple leaf (Ekbote 1942); Mlv/mlv, normal pinnate/multipinnate leaf (Pundir et al. 1990); Ntv/ntv, normal/narrow leaflets (Ramanujam and Singh 1945); Hg/hg, erect/prostrate plant growth habit (Argulkar and D'Cruz 1963); P/p, pink/white corolla (Khan and Akhtar 1934); Sl/sI, single-podded/double-podded (D'Cruz and Tendulkar 1970); and Sph/shp, angular/round seed shape (Kazan et al. 1993). Our results confirm the previously reported mode of inheritance at these loci. The contingency chi-square test indicated linkage between nec and gl in the F₂ of all five crosses (Table 2). An average distance of 15.8 ± 3.0 CM was estimated between these two loci. Healthy glabrous plants and necrotic pubescent plants were recovered as recombinants in the F₂, further ruling out the possibility of the occurrence of necrosis in glabrous plants due to their susceptibility to insect damage.

A study of joint segregation of the nec and slf loci from the cross ICC 10034 × ICC 15566 indicated linkage between these loci (data not presented). This linkage, however, needs further confirmation as failure of expression of double poddedness in necrotic plants may be due to their poor vegetative growth. The loci slf, mlv, ntv, hg, P, and shp segregated independently of the nec locus (data not presented).

The glabrous mutant of chickpea may prove useful in entomological and pathological studies because of its high susceptibility to insects (Pundir and Reddy 1989). For example, the glabrous mutant would be a good medium for rearing large populations of black aphids required for studies of stunt disease transmission. We found that the usefulness of the glabrous mutant was adversely affected by its heritable necrosis. Necrosis reduces the photosynthetic area of the plant by partial or complete drying of leaves and leads to poor vegetative growth. We have recovered healthy glabrous recombinant plants that will be more useful in this respect than the original necrotic glabrous mutant. Moreover, the glabrous trait is now available in the background of different leaf types, namely, simple leaves, multipinnate leaves, and narrow leaflets.

Even though necrosis is an agronomically undesirable character, it is a valuable genetic marker for gene mapping. The linkage between nec and slf loci is a good addition to the information on genetic linkage in chickpea. Attempts are being made using isozyme markers to assign this linkage to the previously reported isozyme linkage groups of chickpea (Gaur and Slinkard 1990a,b; Kazan et al. 1993).

The genera Cicer and Pisum are closely related and have several common linkage groups consisting of homologous genes (Gaur and Slinkard 1990a,b; Kazan et al. 1993; Simon and Muehlbauer 1997). A mutant characterized by brownish, papery, necrotic margins of the leaflets and stipules, designated “burnt leaf,” has been reported in Pisum (Sharma 1973). Initially only the tips of leaflets and stipules show a burnt appearance which subsequently spreads to whole leaflets and stipules. A recessive gene, designated bul, was found to control this mutation. The bul gene of Pisum does not appear homologous to the nec gene of Cicer as the bul gene affects the growing tips more severely than the lower part of the plant, whereas the opposite is the case with the nec gene of Cicer. The bul gene has been mapped on chromosome 3 of Pisum close to st for reduced stipules and chi-6 for light green to yellowish green plant (Marx 1980). The location of the gl-nec linkage on the gene map of Cicer is yet to be determined.

Table 2. Contingency χ² tests for joint segregation of nec (necrosis) and gl (glabrous) loci in F₂ of chickpea

<table>
<thead>
<tr>
<th>F₂ phenotype</th>
<th>PU/HL</th>
<th>GL/HL</th>
<th>PU/NC</th>
<th>GL/NC</th>
<th>χ²</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>ICC 5316 × ICC 15566</td>
<td>157</td>
<td>13</td>
<td>12</td>
<td>30</td>
<td>84.7</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>ICC 5434 × ICC 15566</td>
<td>138</td>
<td>20</td>
<td>14</td>
<td>36</td>
<td>68.0</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>ICC 10034 × ICC 15566</td>
<td>145</td>
<td>9</td>
<td>11</td>
<td>24</td>
<td>77.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>ICC 10301 × ICC 15566</td>
<td>188</td>
<td>19</td>
<td>25</td>
<td>41</td>
<td>81.8</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>JG 315 × ICC 15566</td>
<td>81</td>
<td>8</td>
<td>11</td>
<td>28</td>
<td>52.9</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Pooled data</td>
<td>364.0</td>
<td></td>
<td></td>
<td>1.3</td>
<td>0.860</td>
<td></td>
</tr>
</tbody>
</table>

References


© 2000 The American Genetic Association
Table 1. Meliotic configurations reported in Coffea arabica, in tetraploid C. canephora and in interspecific arabusta hybrids (C. arabica x C. canephora 4x)

<table>
<thead>
<tr>
<th>Species/accessions</th>
<th>References</th>
<th>I</th>
<th>II</th>
<th>III</th>
<th>IV</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. arabica cv. SL28</td>
<td>Owuor (1985)</td>
<td>1.8</td>
<td>19.9</td>
<td>0.1</td>
<td>0.5</td>
</tr>
<tr>
<td>C. arabica cv Catura,</td>
<td>Grassias and Kammacher (1975)</td>
<td>0.5</td>
<td>21.5</td>
<td>0.0</td>
<td>0.1</td>
</tr>
<tr>
<td>Tetraploid C. canephora</td>
<td>Grassias (1980)</td>
<td>1.6</td>
<td>0.4</td>
<td>14.0</td>
<td>10.0</td>
</tr>
<tr>
<td>Tetraploid C. canephora</td>
<td>Boaventura (1990)</td>
<td>3.6</td>
<td>15.2</td>
<td>0.7</td>
<td>1.9</td>
</tr>
<tr>
<td>Tetraploid C. canephora</td>
<td>Owuor (1985)</td>
<td>2.4</td>
<td>1.8</td>
<td>14.9</td>
<td>0.9</td>
</tr>
<tr>
<td>Arabusta hybrids</td>
<td>Owuor (1985)</td>
<td>6.1</td>
<td>0.8</td>
<td>17.4</td>
<td>0.4</td>
</tr>
<tr>
<td>Arabusta hybrids</td>
<td>Grassias (1980)</td>
<td>5.8</td>
<td>8.5</td>
<td>13.4</td>
<td>15.9</td>
</tr>
</tbody>
</table>

* l = univalent; II = bivalent; III = trivalent; IV = tetravalent.

In spite of its economic importance, genetic research devoted to Coffea arabica (2n = 4x = 44), the only polyploid species in the genus, has been rather limited. According to recent molecular cytogenetic investigations (Lashermes et al. 1999), C. arabica is an amphidiploid formed from the hybridization between two closely related diploid species (2n = 22), C. canephora and C. eugenioides. The evidence suggests recent speciation and low divergence between the two constitutive genomes of C. arabica and those of its progenitor species. In spite of the close relationship between the two constitutive genomes, C. arabica is considered to display diploidlike meiotic behavior (Krug and Mendes 1940; Table 1).

Polyplody is a major process of genome evolution that can promote rapid speciation (deWet 1980). In particular, allotetraploidy arising as a result of combining related, but not completely homologous, genomes is very common among angiosperms. The stabilization of allopolyploids requires a restriction of pairing and genetic recombination between the different parental chromosomes related by ancestral homology (Moore 1998). Thus allopolyploids are characterized by bivalent formation during meiosis and disomic inheritance (Burnham 1962; Stebbins 1950). On the other hand, the ability to transfer useful traits from a relative species to the cultivated form by conventional methods depends on the genomic affinity between the two species (Kimber 1984). For instance, little or no exchange between the homologous genomes would be expected if there is no interspecies chromosome pairing.

Analysis of the mode of inheritance can provide an indication of the degree of intergenome chromosome pairing and therefore an assessment of recombination potential between the different genomes. The mode of inheritance can be deduced from examinations of segregation of alleles at a number of individual loci. Until recently, analyses of this type in coffee trees have not been feasible because of a lack of suitable genetic markers. The development of restriction fragment length polymorphism (RFLP) markers representing various loci with multiple codominant alleles means that such analysis can now be envisaged.

In the our study, RFLP markers were used to determine the mode of inheritance in C. arabica and in a tetraploid interspecific hybrid called arabusta (Capot 1972) between C. arabica and an autotetraploid form of one of its diploid progenitors, C. canephora. Results are discussed in relation to the chromosome meiotic behavior and the possibility of gene exchange between the homologous genomes in C. arabica, as well as in interspecific hybrids between C. arabica and diploid relatives.

**Materials and Methods**

The segregating material surveyed consisted of 14 F2 plants of C. arabica obtained by controlled selfing of the F1 hybrid (Et 30 x Catura), and 70 plants resulting from the backcross of a tetraploid interspecific arabusta F1 hybrid (Et 30 x IF 181T) to C. arabica (accession Et 30). The arabusta hybrid resulted from a cross between a plant of C. arabica (accession Et 30) used as the female parent, and a tetraploid plant of C. canephora, IF