

## DEVELOPMENT OF DNA MARKERS FOR ROOT-KNOT NEMATODE RESISTANCE IN COTTON

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### Introduction

Southern root-knot nematodes (*Meloidogyne incognita*; RKN) reduce profits from Georgia cotton producers through yield loss, due directly to RKN or indirectly due to other diseases associated with it such as seedling diseases and fusarium wilt (*Fusarium oxysporum*), and increased production cost by nematicide applications. Although nematicides are effective in controlling RKN, they do not provide season-long protection. Also, the future availability of nematicides is uncertain due to environmental concerns. Further, in fields below threshold levels of RKN, small yield losses not justifying cost of nematicide application can occur. With costs of cotton production increasing and prices of fiber at a historical low, any loss of yield can be considered economically significant.

The development and use of cultivars with resistance to RKN offers the best management tool for RKN. However, progress in developing RKN resistant cultivars has been slow because the current screening process to identify resistant genotypes is tedious, time consuming, and destructive. Molecular markers offer an alternative screening process for identifying resistant genotypes in breeding programs. The development of diagnostic markers for genes conditioning RKN resistance will accelerate the transfer of these genes among genotypes or germplasm for new cultivar development. The objective of this study is to develop diagnostic DNA markers for genes conditioning RKN resistance in cotton.

### Materials And Methods

The RKN-resistant line M-120 RNR was crossed with Pima S-6, a susceptible *Gossypium barbadense* cotton line, to develop an F<sub>2</sub> population. The resistance of M-120 RNR comes from Auburn 623 RNR via Auburn 634 RNR, which was backcrossed to Coker 201 (Shepherd, 1982). Six plants of each parent, six F<sub>1</sub> plants, and 241 F<sub>2</sub> plants were inoculated with nematodes in a greenhouse. Variables measured were galling (rated on a 0-10 scale, where 0 = no galls and 10 = 91-100% galled), number of eggs extracted per root system, and eggs per gram of root. DNA extractions were obtained from F<sub>2</sub> plants. Approximately 200 restriction fragment length polymorphism (RFLP) markers were selected 20-25 centimorgans apart to cover the entire cotton genome. These markers were used to screen the 16 most resistant and 16 most susceptible F<sub>2</sub> plants. Regression analysis was utilized to test associations between the scores for RFLP markers and the phenotypic variables measured. The markers showing

a significant ( $P < 0.05$ ) association in this preliminary screening were used to screen the whole population in order to confirm the association.

### **Results And Discussion**

As expected, M-120 had significantly lower galling, number of eggs, and eggs per gram of root than Pima S-6 (Table 1). The  $F_1$  was highly resistant and was not significantly different from M-120 for any of the three variables, suggesting that dominant genes are involved in the resistance to RKN. Coefficients of correlation among variables were calculated using the  $F_2$  data. The correlation coefficients were significant (Table 2), suggesting that the three variables measure similar genetic factors. Galling is the easiest and fastest way of measuring resistance to RKN. The phenotypic distribution of the  $F_2$  plants for galling was skewed towards the resistant parent (Figure 1), suggesting that only few genes control RKN resistance.

The screening of over 130 RFLP markers has been completed, covering more than 70% of the genome, with the remaining markers to be screened in the next few months. Statistical analyses performed using the marker data available to-date have detected seven putative chromosomal regions to be significantly associated with the resistant phenotype, suggesting that a resistance gene may be present in these regions, although random sampling or scoring errors can not be ruled out at this point. The markers that showed significant association in the preliminary screening are currently being tested on the whole population to confirm the association. The chromosome regions around these markers are being investigated in more detail by testing additional markers that are closely linked to them.

### **Acknowledgements**

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### **References**

Shepherd, R.L. 1982. Registration of three germplasm lines of cotton. *Crop Sci.* 22:692.

Table 1. Means of the parental lines and the F<sub>1</sub> for galling, number of eggs, and eggs per gram of root.

Genotype	Galling	No. of eggs	Eggs/g root
M-120	1.3 a <sup>†</sup>	550 a	56 a
Pima S-6	5.7 b	91650 b	4061 b
F <sub>1</sub>	1.0 a	3120 a	113 a

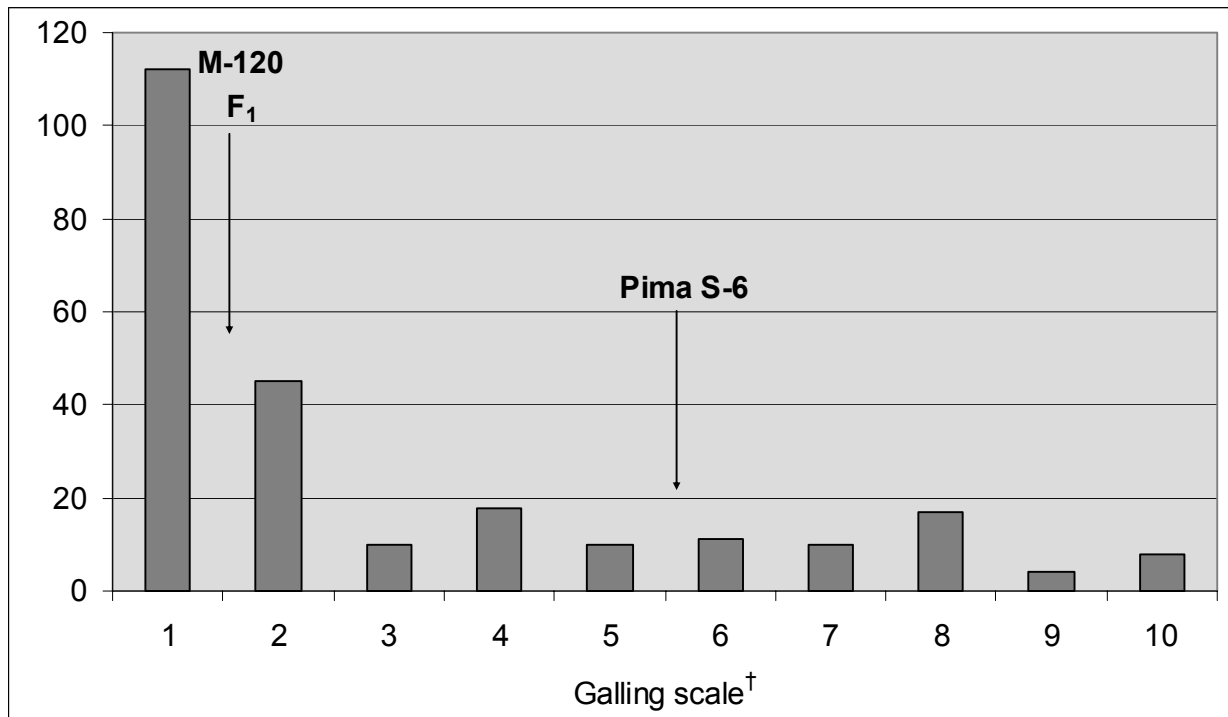
<sup>†</sup> Means followed by the same letter are not significantly different at  $P = 0.05$ .

Table 2. Correlation coefficients among variables.

	Galling	No. of Eggs	Egg/g root
Galling	1.00	-	-
Eggs	0.53*	1.00	-
Eggs/g root	0.57*	0.91*	1.00

\* Significant at  $P = 0.05$ .

Figure 1. Frequency distribution of F<sub>2</sub> plants from the cross M-120 x Pima S-6 for galling.



<sup>†</sup> 0 = no galls; 1 = 1-10% galled; 2 = 11-20% galled; 3 = 21-30% galled; 4 = 31-40% galled; 5 = 41-50% galled; 6 = 51-60% galled; 7 = 61-70% galled; 8 = 71-80% galled; 9 = 81-90% galled; 10 = 91-100% galled.