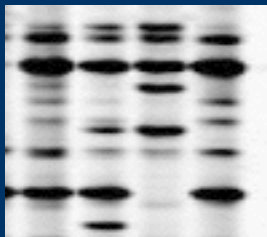
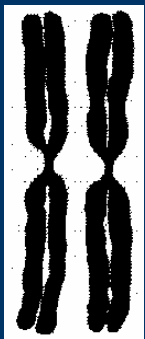


**Reniform Nematode Resistance from
Gossypium longicalyx - Cytogenetics,
Breeding & Molecular Marker
Development**

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Objectives



- INTROGRESSION (selective):
 - Resistance to reniform
 - NOT undesirable traits
- CYTOGENETICS:
 - Chromosomal constitutions
 - Numbers and kinds of aberrations
 - Recombination
 - Alien segment size
- MARKERS:
 - Analyses
 - MAS

Triple Species Hybrids

(Bell and Robinson, USDA)

G. hirsutum

$(A_h D_h)^2$ (4x)

x

G. longicalyx

$(F_1)^2$ (2x)

$A_h D_h F_1$

(3x)

$(A_h D_h F_1)^2$

(6x)

x

G. armournianum $(D_{2-1})^2$

(2x)

$A_h F_1 D_h D_{2-1}$

(4x)

Two Triple-species Hybrids

(Bell and Robinson, USDA)

TWO resistance sources:

- 1) $(G. \textit{hirsutum} \times \underline{G. \textit{longicalyx}})^2 \times \underline{G. \textit{armourianum}}$ (HLA)



- 2) $(G. \textit{hirsutum} \times \underline{G. \textit{herbaceum}})^2 \times \underline{G. \textit{longicalyx}}$ (HHL)



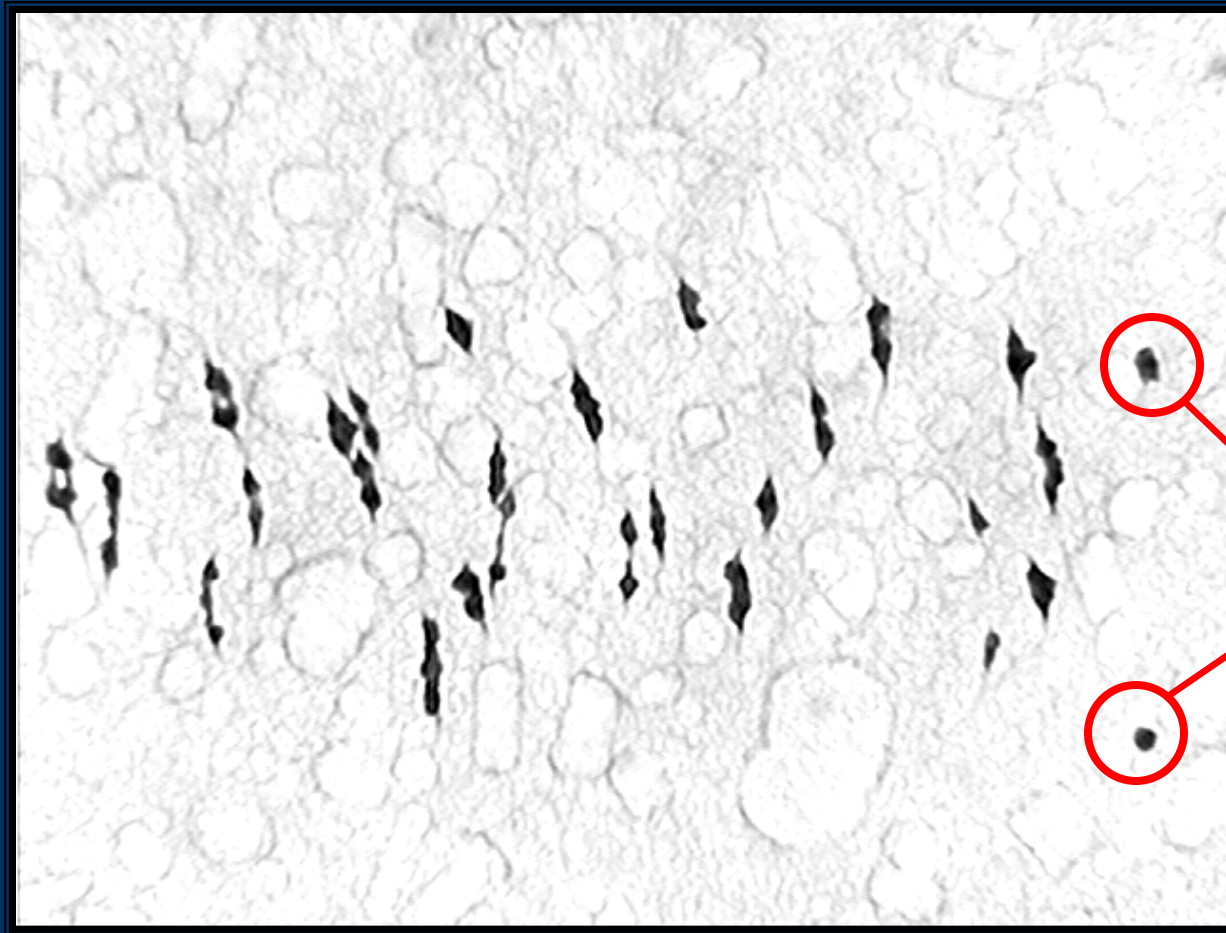
Cytogenetics Data Summary

Generation	# Plants Analyzed
BC 1F1	38
BC 1S1	31
BC 2F1	94
BC 3F1	23
BC 4F1	15
BC 5F1	2
Total	203

Chromosome numbers

Chrom. Gener.	51 Chr.	52 Chr.	53 Chr.
BC1F1	(5%) 2	(66%) 25	(29%) 11
BC1S1	(3%) 1	(84%) 26	(13%) 4
BC2F1	(22%) 21	(61%) 57	(17%) 16
BC3F1	0	(100%) 23	0
BC4F1	0	(100%) 15	0
BC5F1	0	(100%) 2	0

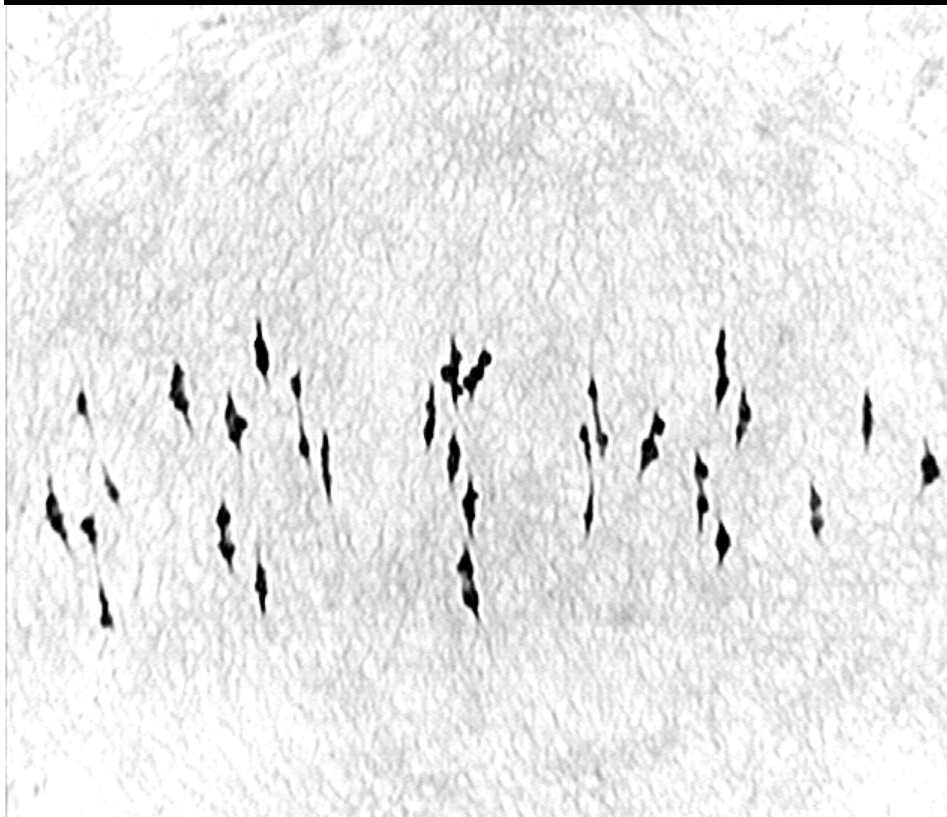
Model Type in BC2



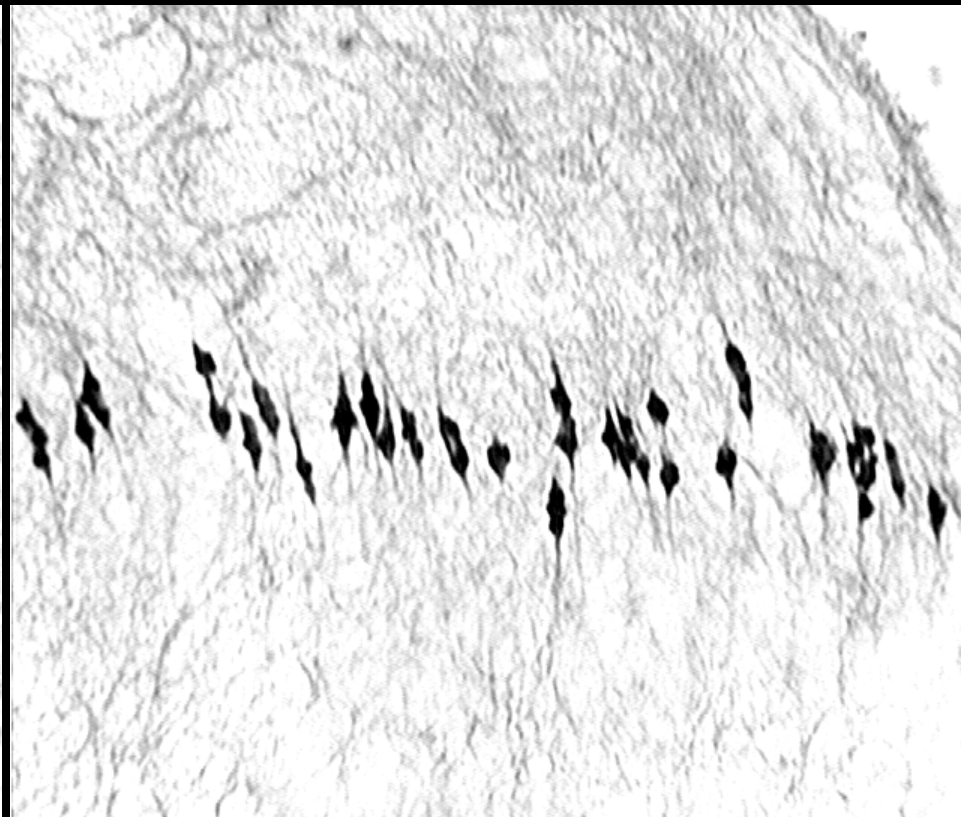
(Not
recombinant)

25II+2I (Res-BC2-129)

Modal Type in Resistant BC3, BC4 and BC5 Plants



26II



26II

Recombination

Proximal and distal
XOs are too distant.



Proximal XO is okay.
Distal XO is too distant.



Proximal XO is too
distant. Distal XO is
okay.



Proximal and distal XOs
are okay.



Recombination

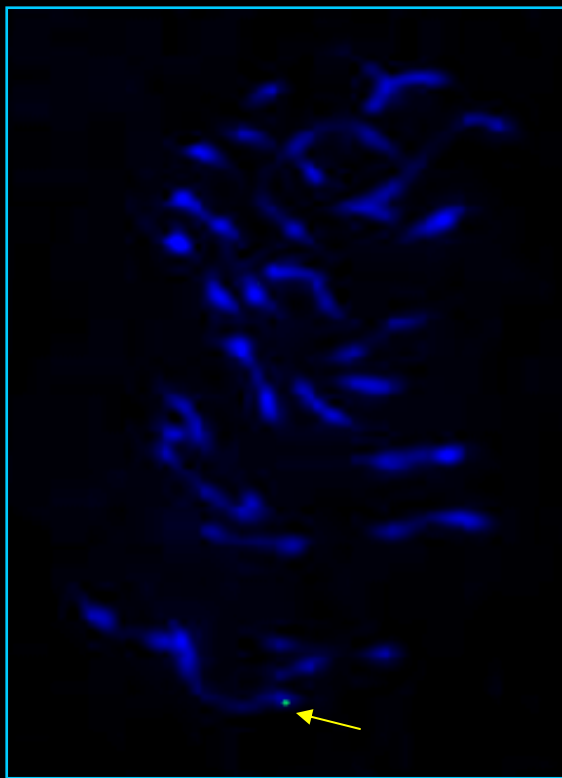
* Estimate of Recombination in BC2

- 2/20 Resistant Plants Analyzed from either 26II OR 1IV+24II
- At least ~10% Recombination in BC2

* Estimate of Recombination in BC3 and BC4

- Approx. 85% of the Resistant Plants Analyzed from 26II OR 1IV+24II

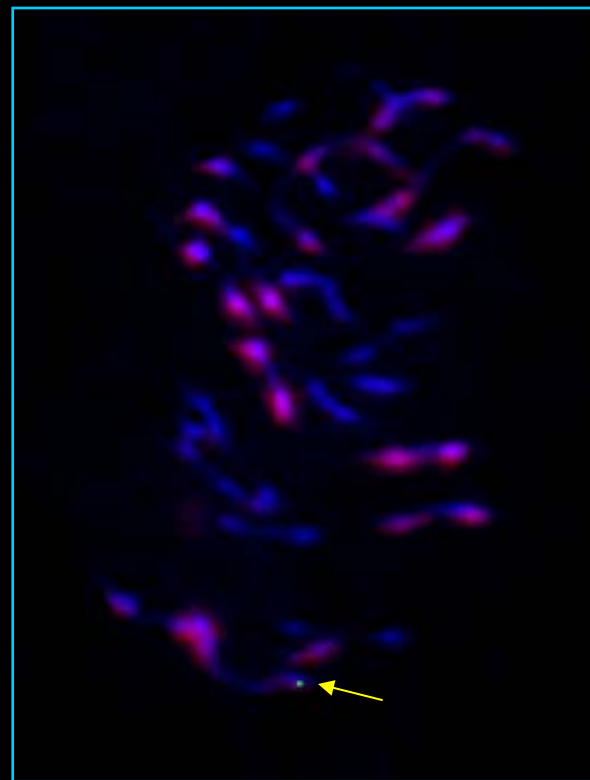
GISH (Genomic in situ hybridization) on BC3 1-6 (Resistant)



Blue---All the chromosomes

Red----A1-genome

Green—F1-genome



BC3 1-6

Highly Resistant plant

26II Chrom. Config

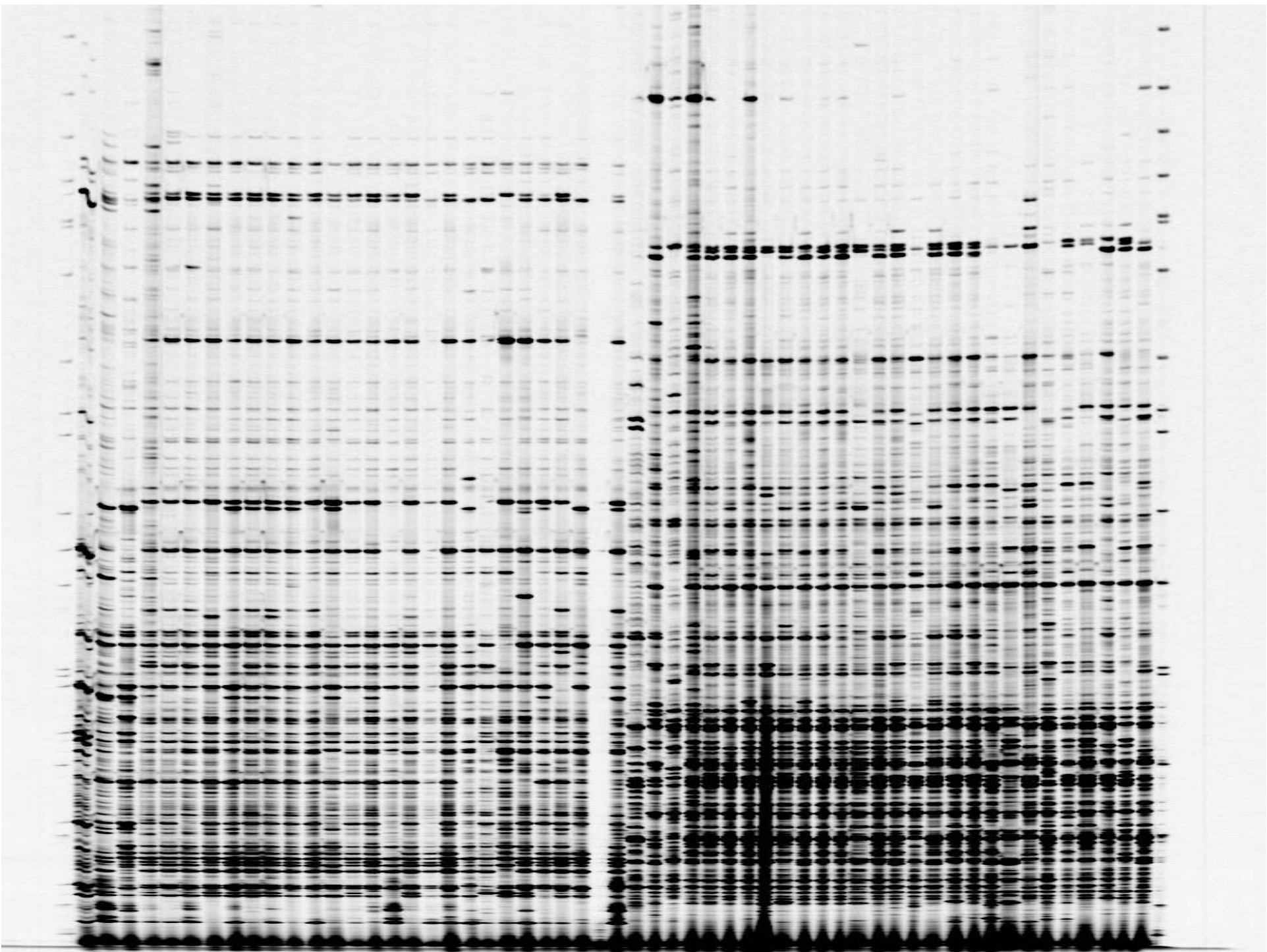
BC4F1 Segregating 1:1 (5R:5S)

MARKER DEVELOPMENT

- “PHASE-I” MARKERS: alien AFLPs

- SCREENING PANEL:
 - *G. hirsutum* (AD₁)
 - *G. longicalyx* (F₁) and *G. armourianum* (D₂₋₂)
 - Resistant BC₁ parents (Bulked)
 - 12 most resistant plants of 186 BC₂ plants
 - 12 most susceptible plants of 186 BC₂ plants

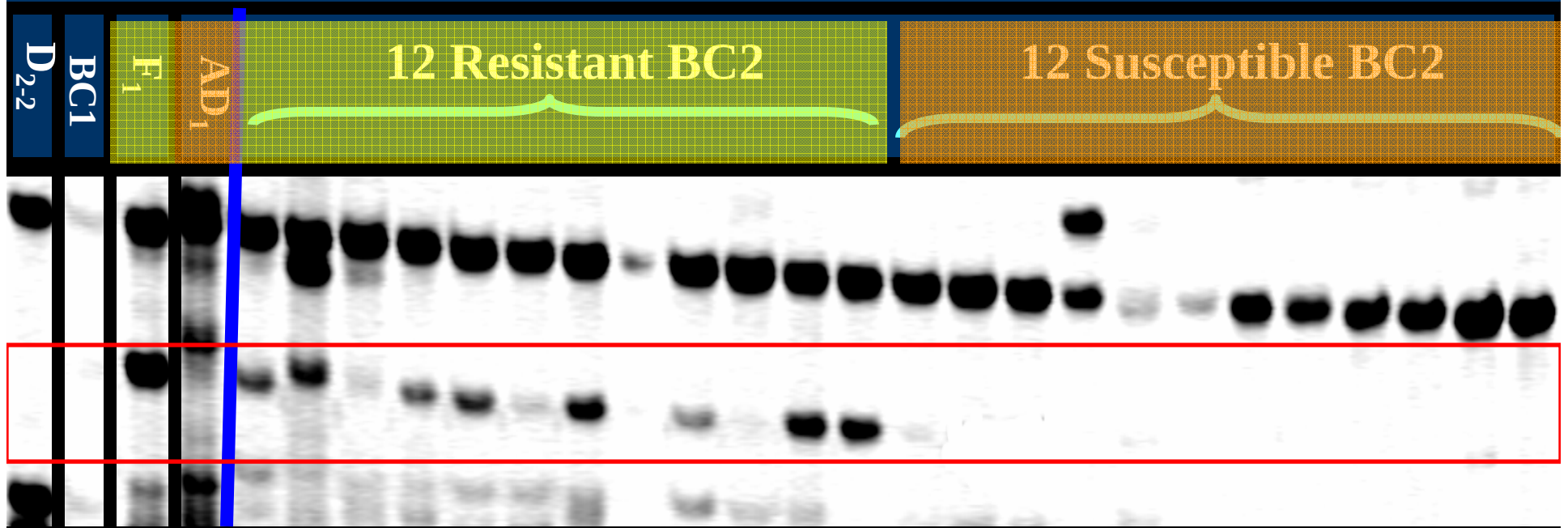
- Li-Cor System



Potential Markers



Potential Markers



Marker Development

Research in Progress..

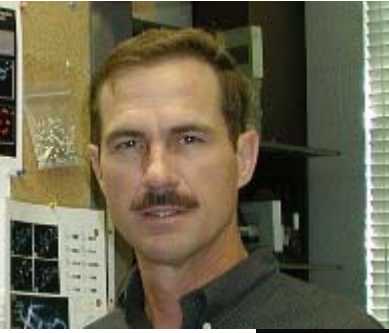
- Identify AFLP markers tightly linked to the reniform nematode resistance gene

THEN:

- “Phase-II” Markers -- e.g. SSRs
 - Identify linkage group (*using mapped loci*)
 - MAS, especially among inbreds (“Zygosity assays”)

STATUS SUMMARY

- **The existing data indicate successful introgression!!**
- **NEEDS:**
 - Markers for MAS
 - Backcrossing
 - Inbreeding
 - Eliminate unnecessary alien germplasm
 - GISH (physical size of alien segment)
 - Markers
 - Phenotypic screens
 - Yield and fiber assessments



Acknowledgments Cotton Inc.

