

Cowpea Genome Resources Developed in Support of Breeding

- Cowpea Context
- UC Riverside - Africa Linkage
- SNP Map & Synteny
- Physical Map & Genome Sequence
- A Smorgasbord of Traits

Poster 464
Workshop 272

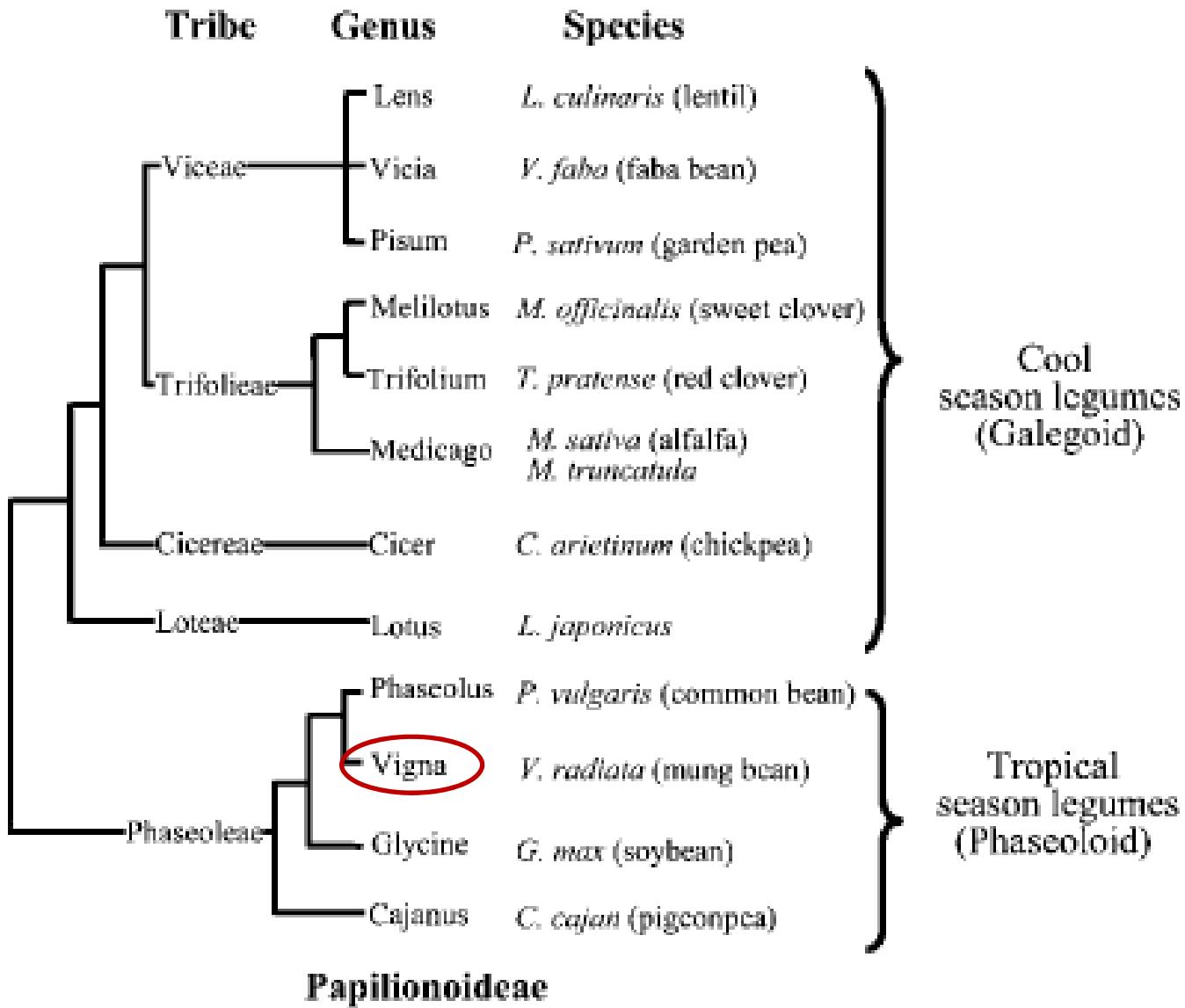
Timothy Close
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Cowpea

- *Vigna unguiculata* L. Walp
- Genome size ~623 Mb
- $2n=2x=22$
- Origin - native to Africa
- Importance - food, fodder, helps increase nitrogen availability in soil
- Well adapted to water-limited & hot environments



Fabaceae Family



Choi et al. 2004b

Rich and Diverse Germplasm Available

<u>Collection</u>	<u>Accessions</u>	<u>Countries</u>
IITA-Nigeria	14,500	65
USDA-USA	6,841	50
UC Riverside	5,200	41

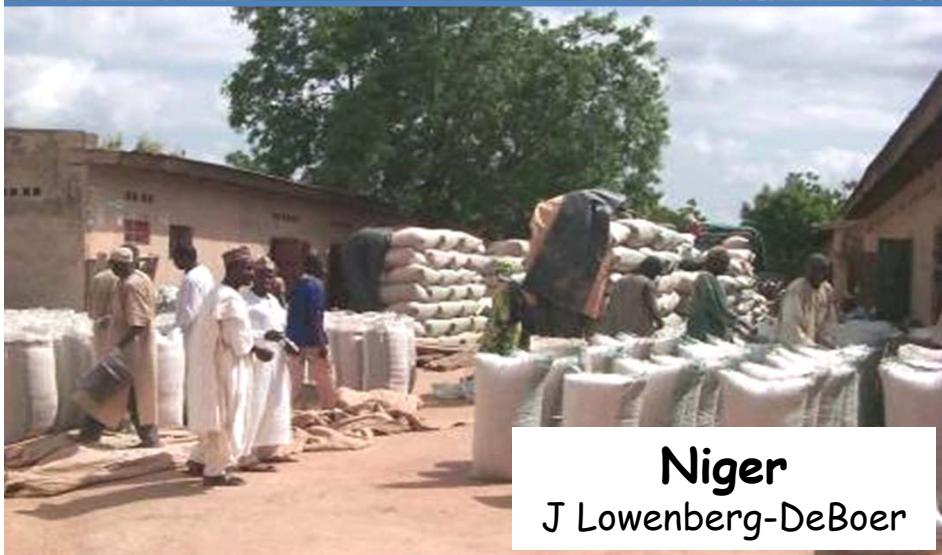


Cowpea is critically important to diets, economy and cropping systems of Africa

Senegal
J Ehlers



Nigeria
J Lowenberg-DeBoer



Niger
J Lowenberg-DeBoer



Burkina Faso
J Lowenberg-DeBoer

Partners in semi-arid cowpea zone:

IITA-Kano, Nigeria

ISRA, Senegal

INERA, Burkina Faso

Eduardo Mondlane University, Mozambique

200 mm

600 mm

Senegal

Burkina Faso

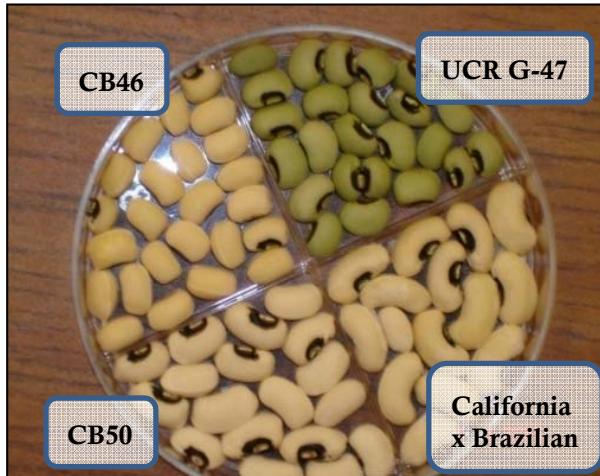
Nigeria

Mozambique

- [Green square] Tropical rain forest (broadleaf evergreen)
- [Blue square] East Africa coastal forest
- [Light green square] Deciduous forest-woodland savanna
- [Light yellow square] Brush-grass savanna
- [Yellow square] Steppe (grass, brush, and thicket)
- [Pink square] Semidesert
- [Orange square] Desert
- [Dark red square] Mediterranean evergreen forest-hard-leaf scrub
- [Brown square] Temperate grassland (veld) and mountain grassland
- [Purple square] Montane forest-tundra

Scale 1:48,000,000
Azimuthal Equal-Area Projection

Cowpea - Breeding for Trait Improvement



California

- Seed quality (larger grain & whiteness of seed coat)
- Nematode resistance
- Fusarium resistance
- Aphid resistance
- Lygus resistance
- Yield



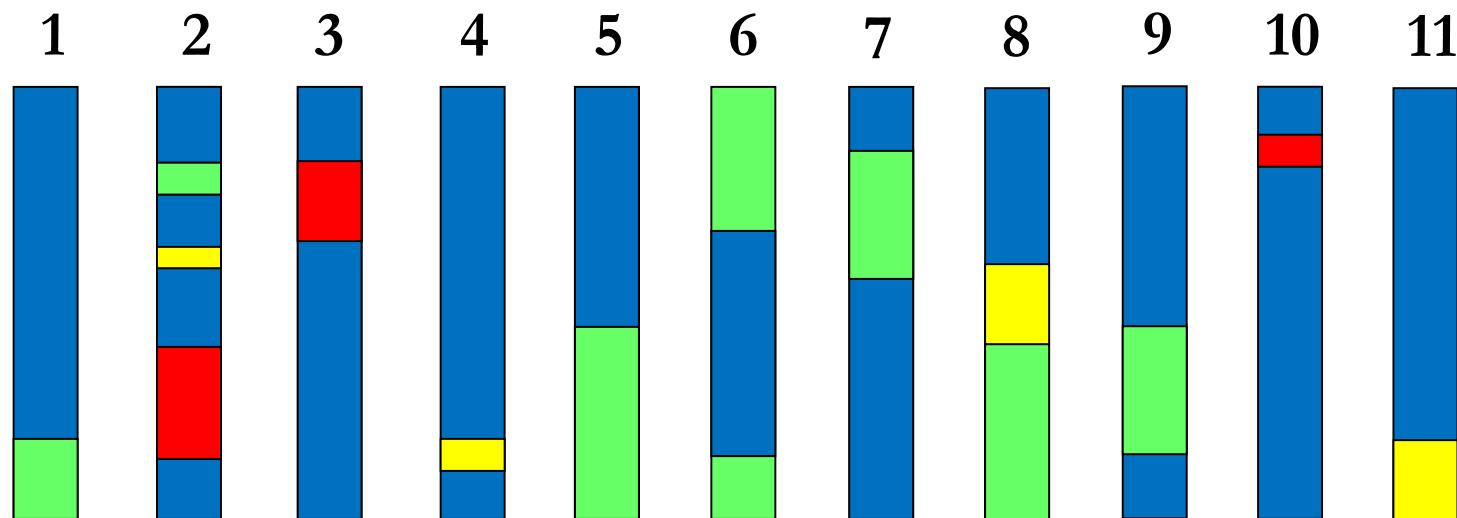
Africa

- Drought tolerance
- Yield
- Early Maturity
- Nematode resistance
- Striga resistance
- Thrips resistance
- Lygus resistance
- Macrophomina resistance

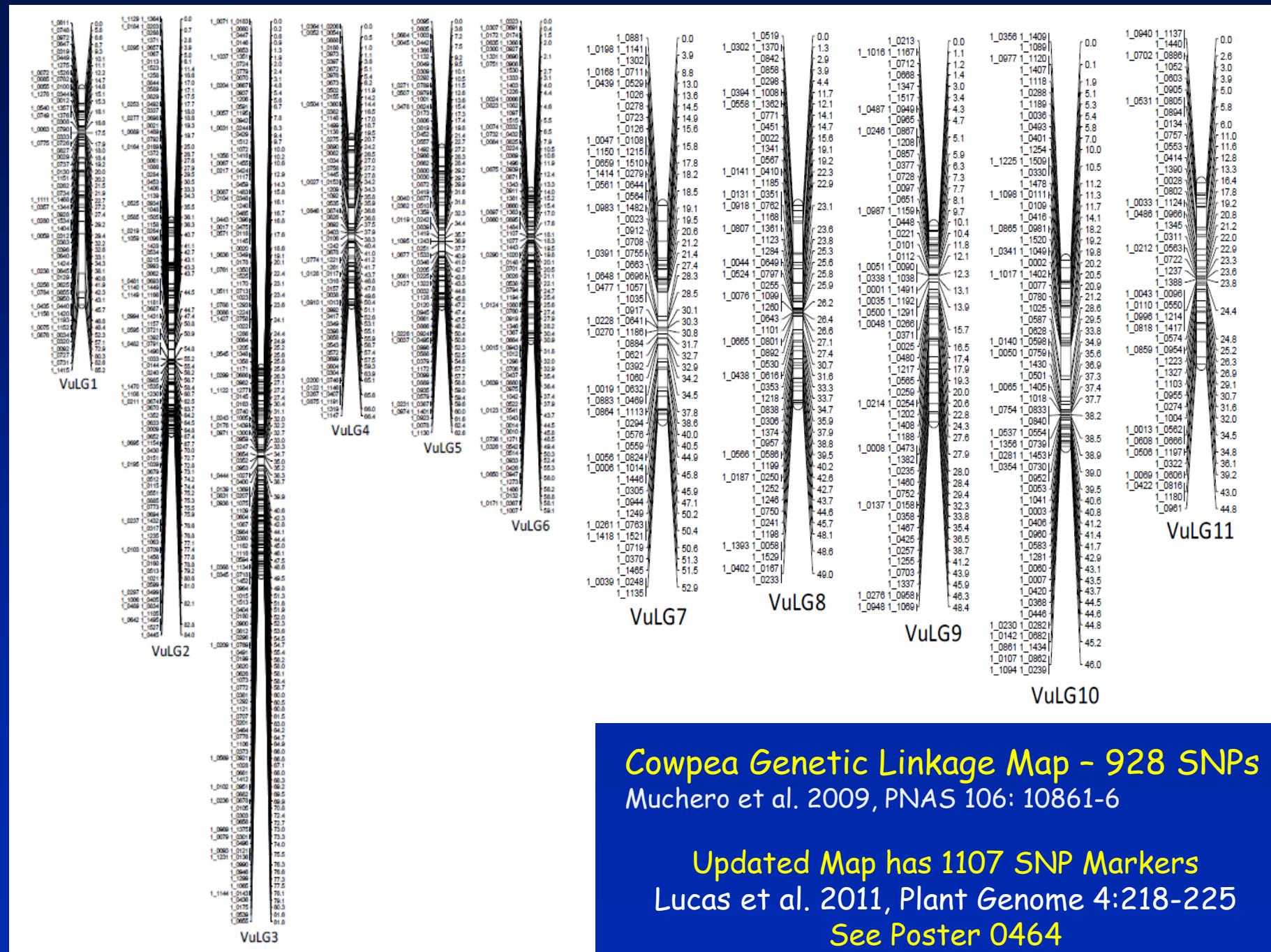
Cowpea Genetic Ideotype Breeding

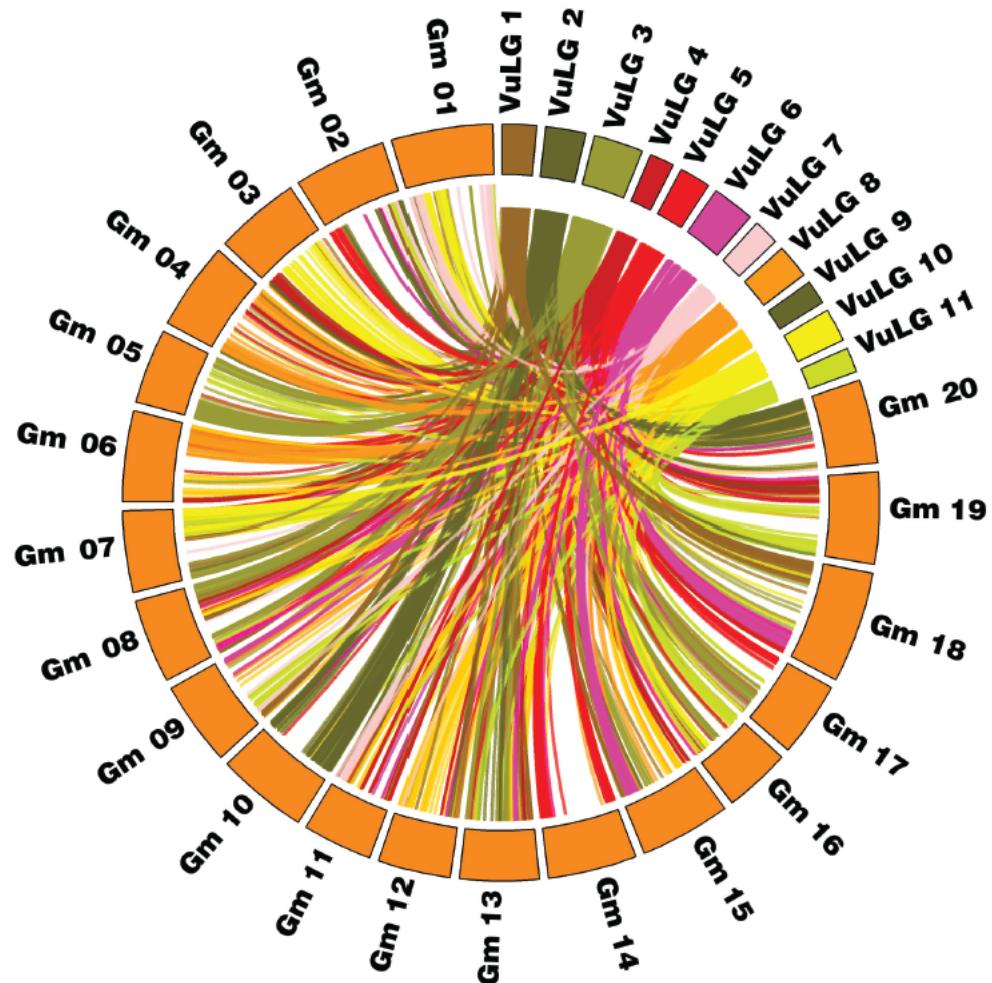
(use markers to combine loci from multiple parents while keeping population sizes low)

■ IT93K-503-1 ■ Mouride ■ Melakh ■ 58-77



Graphical genotype of desired outcome





13 mapping populations

- 11 sets of RILs
- 2 F4 breeding populations

1293 individuals

1107 EST-derived SNPs

11 linkage groups

856 marker bins

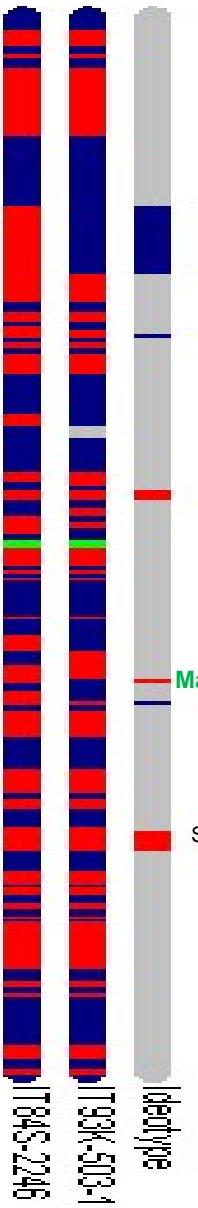
680 cM total map

New map in HarvEST:Cowpea v1.27, available from <http://harvest.ucr.edu>

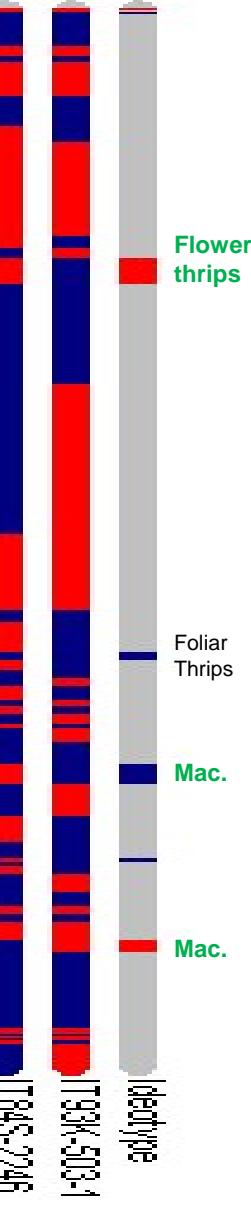
See also Poster 0464

Lucas et al. 2011. Cowpea-Soybean Synteny Clarified Through an Improved Genetic Map. The Plant Genome 4:218-225

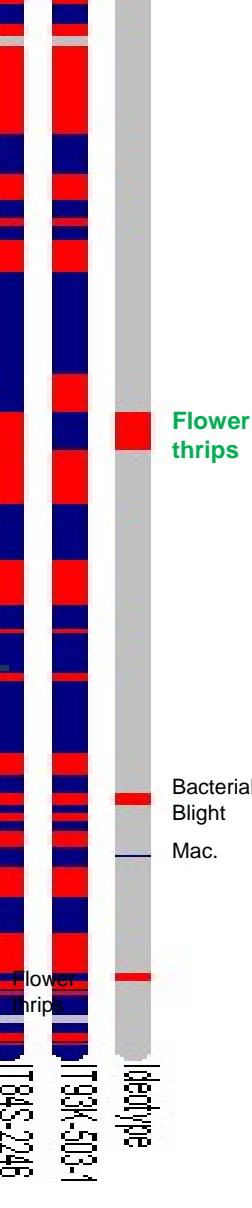
LG2



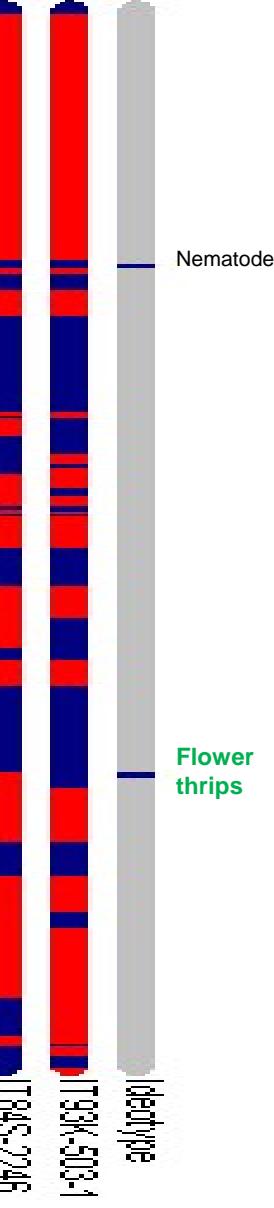
LG4



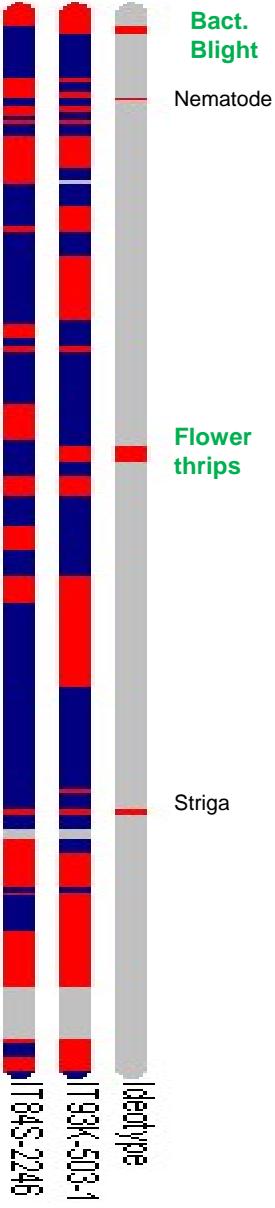
LG5



LG8



LG9



BreedIt'

- Developed a prototype program to choose a marker set for a given cross
 - Input SNP data table (genotype or locus file)
 - Input trait (QTL, markers) and map files
 - Program Options
 - Choose 2 parents
 - Choose cM interval for background selection
 - Choose cM interval for markers flanking traits
 - Output list of SNPs for customized assay
 - Can quickly compare cost versus marker density

BreedIt output file for marker selection

SNP	SNP_ID	LG	cM	IT93K-503-1	IT84S-2246	Polymorphic	Trait	Selected	Reason	Distance
1_0190	16480_663	4	6.83	AA	AA	0		0		
1_0502	5268_412	4	10.27	BB	AA	1		1	Every 5 cM	
1_1148	10853_451	4	10.36	AA	AA	0		0		
1_0382	6867_337	4	10.66	AA	AA	0		0		
1_1360	12393_305	4	12.23	BB	AA	1		0		
1_0504	14303_873	4	12.76	AA	BB	1		0		
1_1499	12324_917	4	13.69	AA	BB	1		1	Flanking	3.41
1_1138	17303_348	4	14.09	BB	AA	1	Flower Thr	1	Trait	0.4
1_0155	474_351	4	15.56	BB	BB	0		0		
1_0082	13873_544	4	17.97	AA	AA	0		0		
1_0756	14622_249	4	18.37	AA	AA	0		0		
1_1413	1078_282	4	18.38	BB	AA	1		1	Flanking	4.29
1_1445	13794_319	4	19.1	BB	BB	0		0		
1_0847	8899_1022	4	19.48	--	--	0		0		
1_0774	16646_118	4	20.16	BB	AA	1		0		
1_1221	1202_1215	4	20.16	BB	AA	1		1	Flanking	1.77
1_1242	4217_685	4	20.72	BB	BB	0	Foliar Thrip	0		
1_0027	3683_549	4	21.49	BB	AA	1		1	Flanking	1.34
1_0153	14462_171	4	21.49	BB	AA	1		0		
1_0535	1808_342	4	22.85	AA	BB	1		0		
1_1261	11736_560	4	22.92	BB	BB	0		0		
1_1092	5061_428	4	23.66	BB	AA	1		0		
1_0646	12854_535	4	24.12	BB	AA	1		0		
1_0874	7102_965	4	24.4	AA	BB	1		0		
1_1264	11709_707	4	24.43	--	--	0		0		
1_0826	9147_1655	4	24.55	BB	AA	1		0		
1_0692	8273_1205	4	25	AA	BB	1		0		
1_0403	4774_665	4	25.31	--	AA	0		0		
1_0106	8625_1231	4	25.57	AA	BB	1		0		
1_0678	13269_270	4	27.6	BB	AA	1		1	Every 5 cM	6.11

Cowpea Physical Map

<http://phymap.ucdavis.edu/cowpea/>

- African breeding genotype IT97K-499-35
- 2 BAC clone libraries (*Hind*III and *Mbo*I)
- 60,000 BACs were DNA fingerprinted; 30,000 BES
- Final physical map = assembly of 43,717 BACs with a depth of 10x genome coverage; one in two BACs has BES
- Total of 790 contigs

PhyMap
Cowpea
Genome

Home
PhyMap Databases
GBrowse

WheatDB
Physical Mapping
Databases

Generation
A CGIAR Challenge Program

Last modified on
June 10, 2009



Physical Map of Cowpea

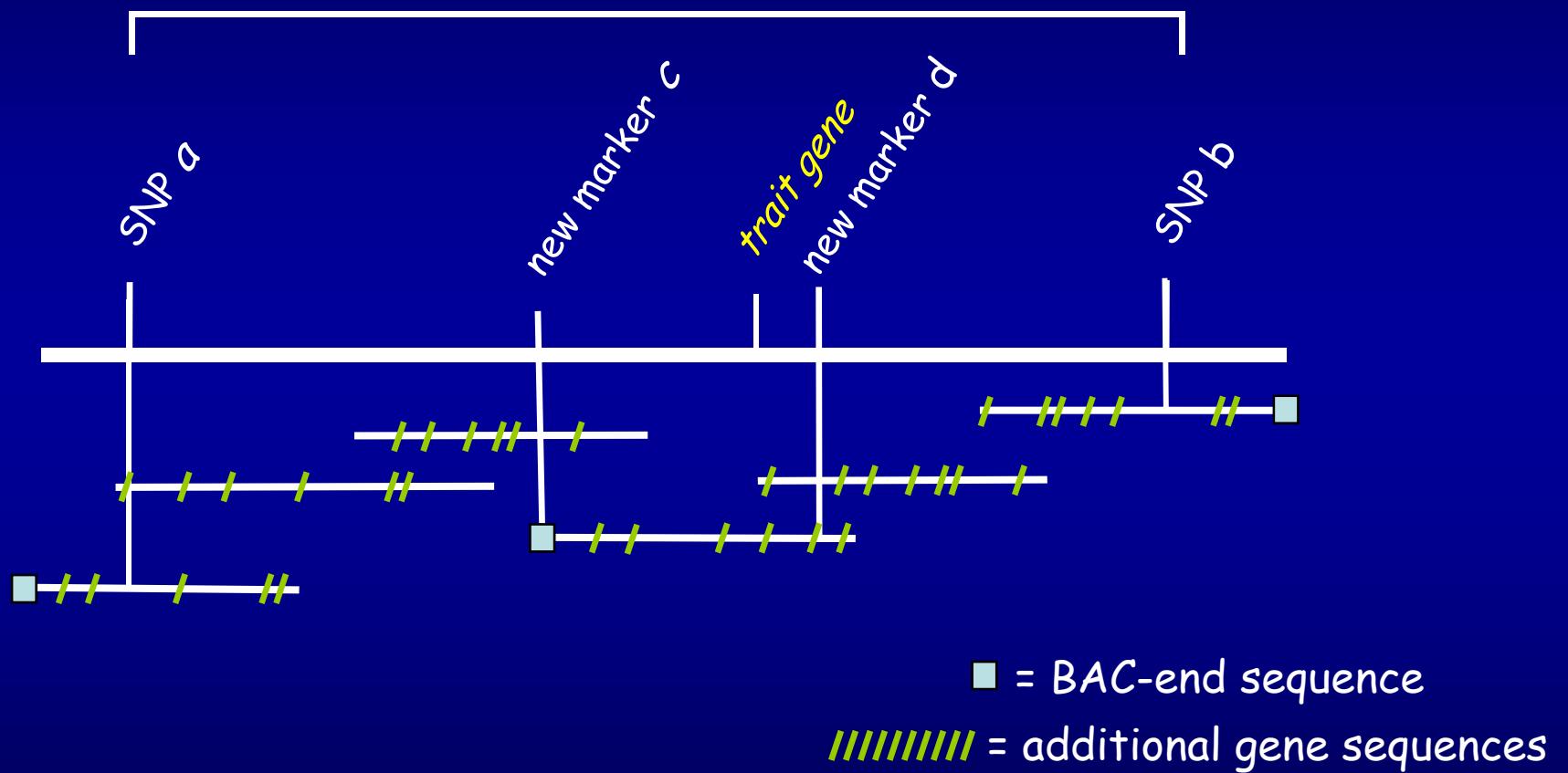
This database is an outcome of the cowpea component of the CGIAR Generation Challenge Program Tropical Legumes I project, "Improving tropical legume productivity for marginal environments in sub-Saharan Africa". This work has been supported since May 2007.

The cowpea [*Vigna unguiculata* (L.) Walp.] genome has a size of about 600 million base pairs. During the past two years, considerable progress has been made in the establishment of core genome resources for cowpea. These resources include cDNA libraries and more than 180,000 partial cDNA sequences (Expressed Sequence Tags; ESTs), single nucleotide polymorphisms (SNPs) and high throughput SNP genotyping, several recombinant inbred line (RIL) populations, a consensus genetic map containing more than 1000 SNP markers, genomic bacterial artificial chromosome (BAC) libraries and BAC-end sequences (BES). The BAC library has been used to generate a physical map of cowpea, which has been anchored to the genetic map via SNP loci. These resources facilitate access of researchers to the cowpea genome. The unifying objective is to accelerate a transition to marker assisted breeding for genetic improvement resulting in yield gain in sub-Saharan Africa. The BAC physical map and associated BES provide opportunities for the development of additional marker discovery, fine mapping, trait analysis and gene cloning. The cowpea genome, via this online resource, now provides a powerful enabling



Example of Cowpea SNP & Physical Map

0.6 cM average distance between SNP markers



BACs anchored to markers provide a sequence resource for additional marker development

Cowpea Genome Sequencing in Progress

~4300 MTP BACs, estimated completion
mid-2012, NSF project

First draft shotgun assembly available

- 60x coverage (Illumina)
- 97% of known genes in portion assembled using SOAPdenovo
- BLAST through www.harvest-blast.org

Fusarium oxysporum f.sp. *tracheiphilum*

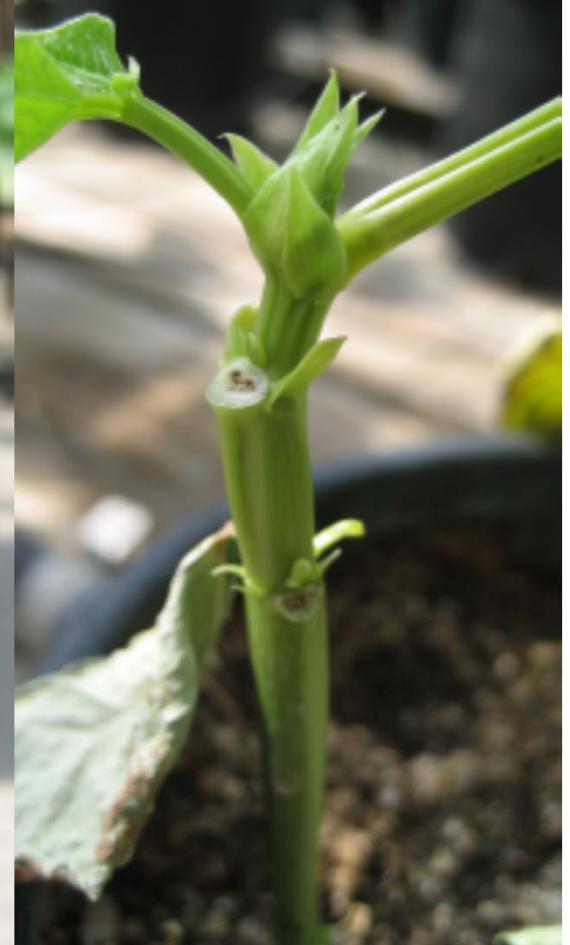
Marti Pottoroff, UC Riverside PhD thesis project



wilting and stunting

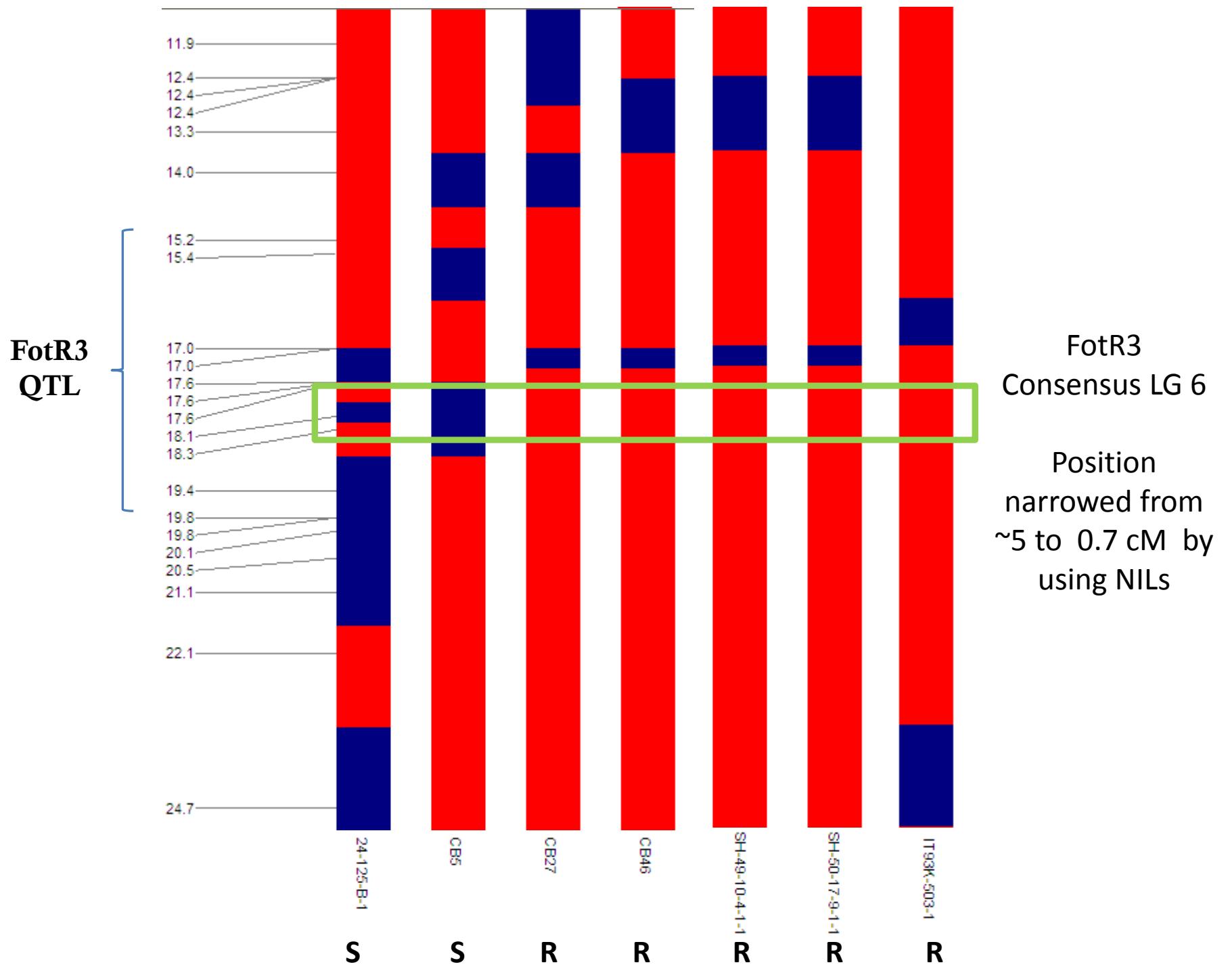


chlorosis



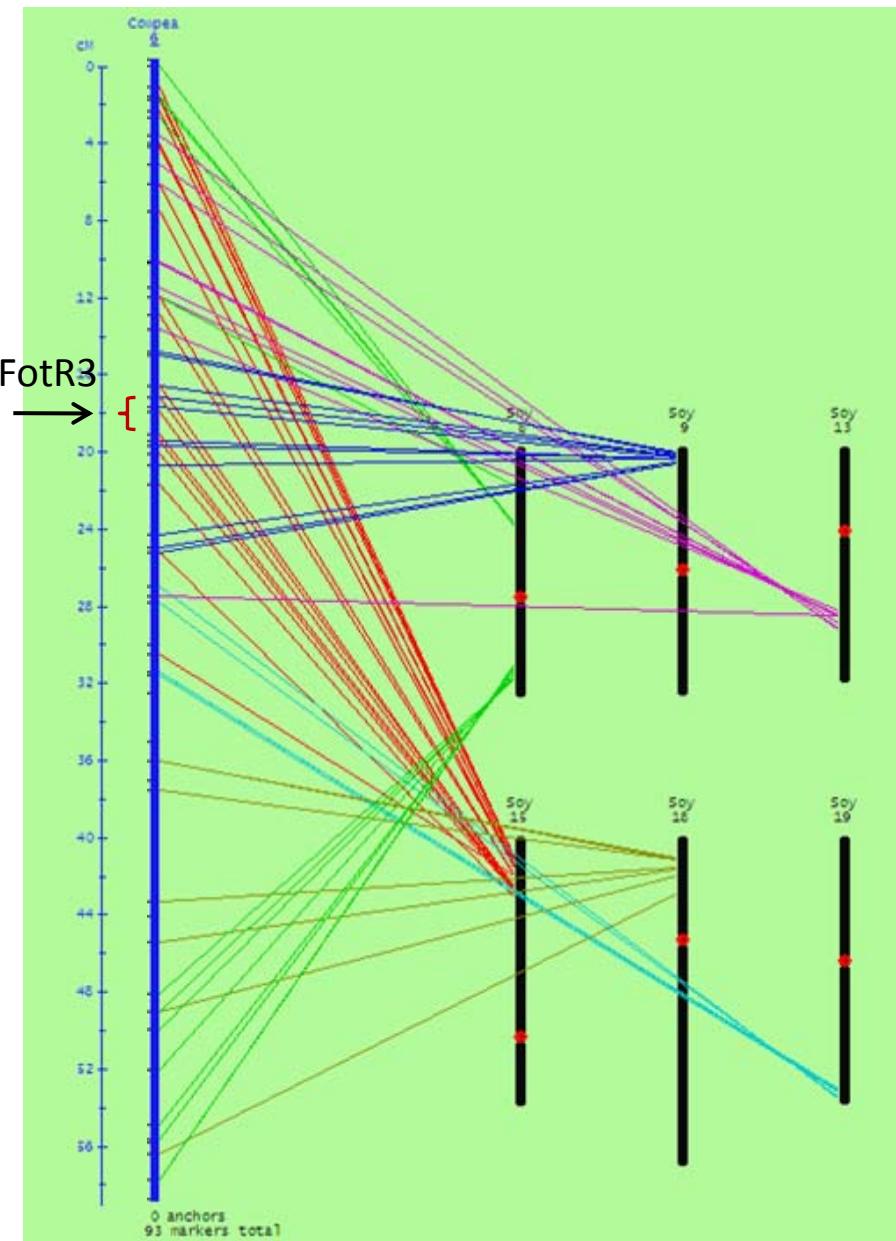
vascular
discoloration

Fusarium Disease Symptoms



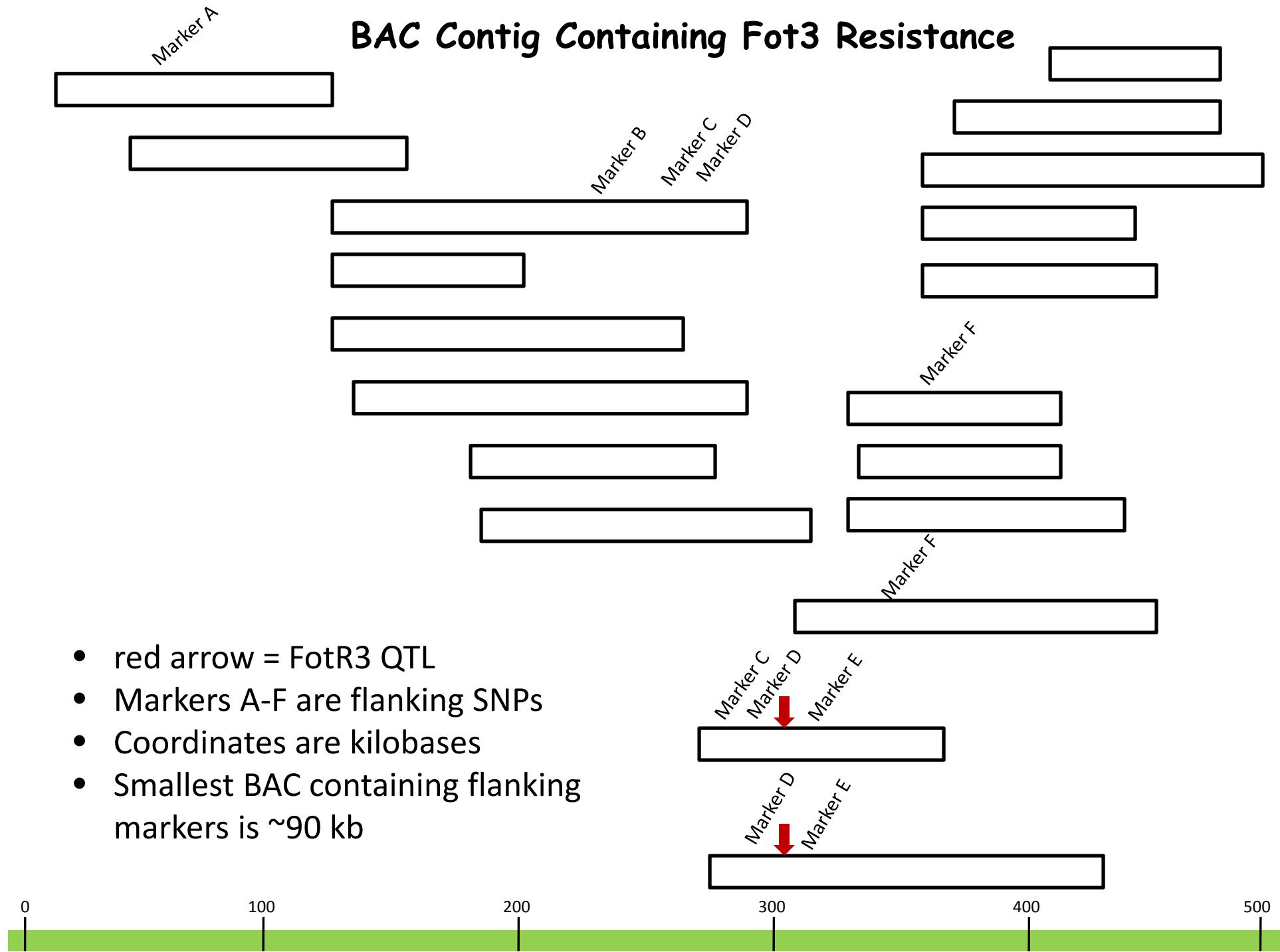
Synteny of Resistance Region with Soybean

Image from HarvEST:Cowpea
<http://harvest.ucr.edu>



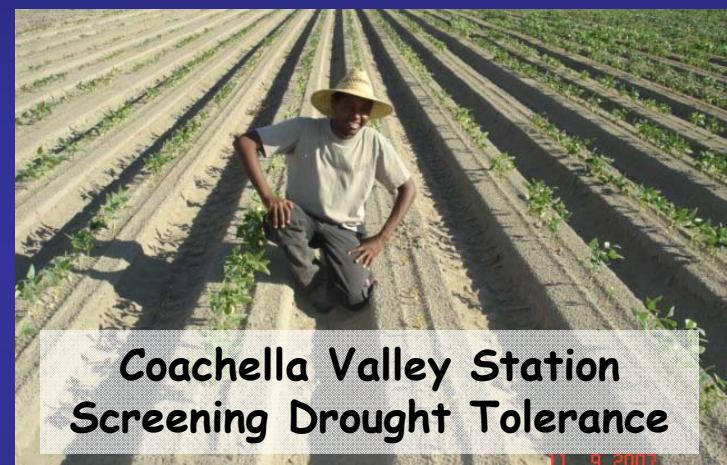
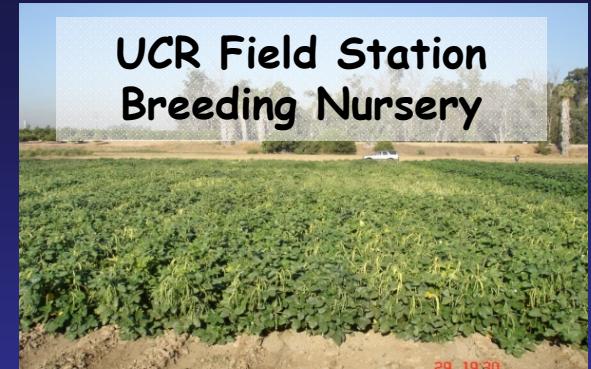
Leucine-rich repeat transmembrane protein kinase, putative
Leucine-rich repeat family protein / protein kinase family protein
Leucine-rich repeat transmembrane protein kinase, putative
Leucine-rich repeat family protein / protein kinase family protein
Similar to kinase-related [Arabidopsis thaliana
Similar to kinase-related [Arabidopsis thaliana
Disease resistance protein (NBS-LRR class), putative
Disease resistance protein (NBS-LRR class), putative
ATMLO1/MLO1 (MILDEW RESISTANCE LOCUS O 1)
RPM1 (RESISTANCE TO P. SYRINGAE PV MACULICOLA 1)
Disease resistance protein (NBS-LRR class), putative
Disease resistance protein (NBS-LRR class), putative
Disease resistance protein (NBS-LRR class), putative
NPR3 (NPR1-LIKE PROTEIN 3); protein binding

BAC Contig Containing Fot3 Resistance



Cowpea traits available for fine mapping, marker development, map-based cloning, mechanistic studies

- aphid resistance
- bacterial blight resistance
- chilling tolerance at seedling emergence
- drought tolerance
- Fusarium resistance
- flower & foliar thrips resistance
- grain weight
- growth habit
- heat tolerance at reproductive stages
- leaf shape
- lygus bug resistance
- maturity time
- Macrophomina resistance
- nematode resistance
- pod dehiscence
- root mucilage
- seed coat color & patterns
- virus resistance
- yield



Some Take-Home Points

- Cowpea is a critical food crop in sub-Saharan Africa
- Automated SNP genotyping & 1107-SNP map available for cowpea
- SNP map combines well with old & new phenotypic data to reveal many trait loci, supporting marker-assisted breeding
- Anchored physical map and sequences facilitate research on genes controlling traits
- Candidate genes emerge from synteny with reference legumes (soybean, etc.)
- Improvements in whole genome sequence are in progress

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Thank you!