



FEED THE FUTURE

The U.S. Government's Global Hunger & Food Security Initiative

Genetic improvement of cowpea to overcome biotic stress and drought constraints to grain productivity



Phil Roberts

University of California - Riverside



Legume Innovation Lab

Feed the Future Innovation
Lab for Collaborative
Research on Grain Legumes



USAID
FROM THE AMERICAN PEOPLE

Project Personnel:

University of California, Riverside

Tim Close

Bao-Lam Huynh

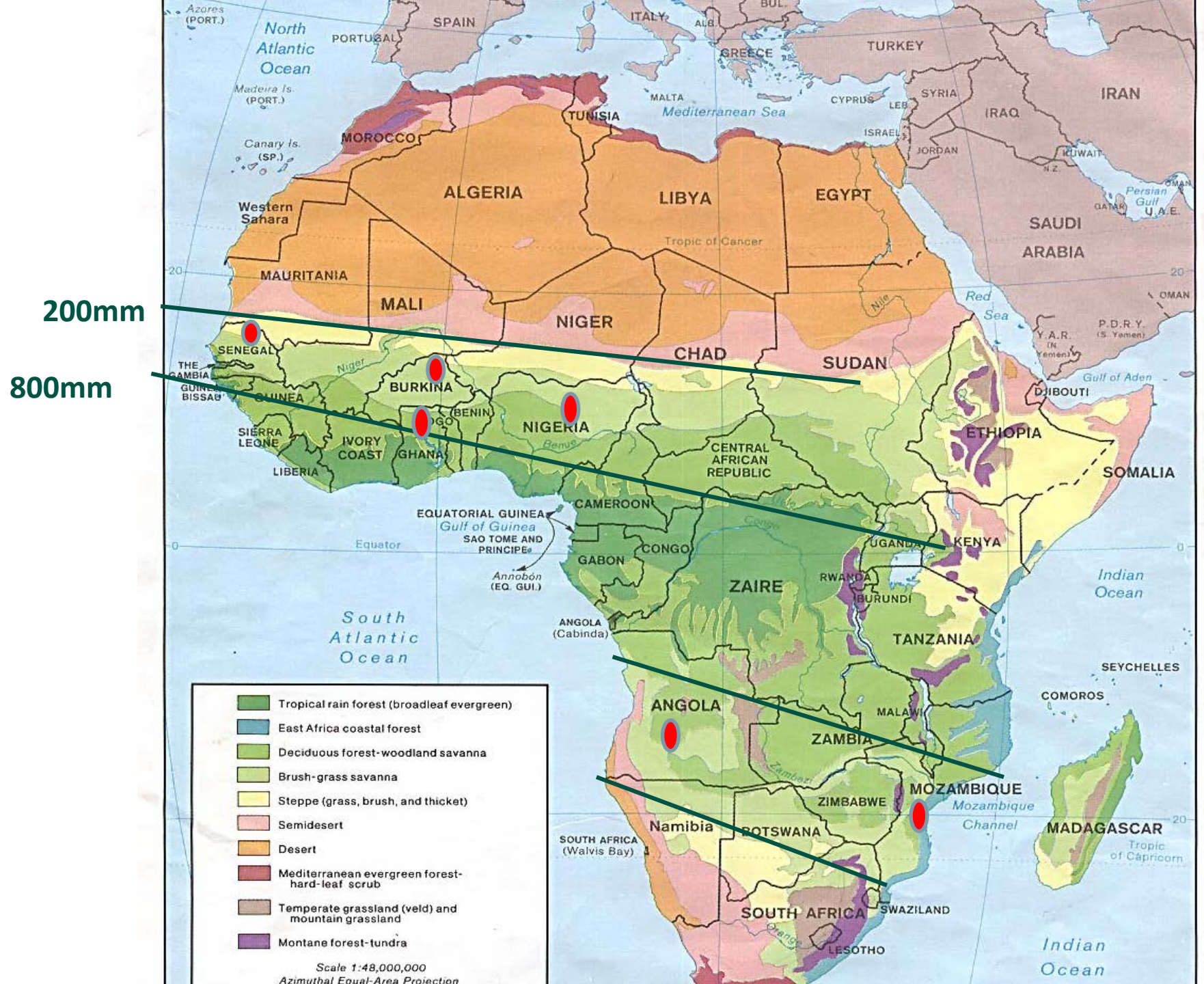
Host Countries (Africa)

Francis Kusi & Ibrahim Atokple, Savannah Agricultural Research Institute, Ghana (SARI)

Ndiaga Cisse, Institut Senegalais de la Recherches Agricole, Senegal (ISRA)

Joseph Batieno & Issa Drabo, Institut de l'Environnement et des Recherches Agricole, Burkina Faso (INERA)

Trainees: Arsenio Ndeve (Mozambique); Mame Penda Sarr (Senegal); Sassoum Lo (Senegal); Mitchell Lucas (CA-USA); plus 10 MS and PhD trainees based in HC programs



Objective 1: Discover QTL for insect resistance and apply in molecular breeding for target regions in West Africa and the US

Collaborators:

Clementine Dabire, INERA, Burkina Faso

Barry Pittendrigh, MSU, USA

Manu Tamo, IITA, Benin

Christian Fatokun, IITA, Nigeria

Ousmane Boukar, IITA, Nigeria

Ibrahima Sarr, ISRA, Senegal

- Aphid, Flower thrips and Pod sucking bug resistance
- Populations phenotyped, SNP genotyped, QTL mapped
- Marker selection to introgress R traits into advanced drought tolerant breeding lines.

Objective 1: Discover QTL for insect resistance and apply in molecular breeding for West Africa and the US

Resource:

Aphid resistance – multiple sources

Questions:

Which resistance is effective in each target area?

Do the aphid populations vary for response to resistance source (biotypes)?

Actions:

Increase seed of resistance source panel

Uniform tests of resistance panel in multiple locations

Genotype aphid populations (B. Pittendrigh)

QTL mapping and markers for R loci

MABC for aphid resistance into elite varieties

Cowpea aphid

Aphis craccivora



Assessment of cowpea yield loss due to Aphids in Ghana (*Kusi et al, 2014*)

Resistant:

Improved Zaayura 3.8 %

SARC 1-57 4.9 %

SARC 1-91-1 9.8 %



Partially resistant:

IT97K-499-35 17.1 %

Padituya 16.1 %

Susceptible:

Susc BC progeny 32.8 %

IT99K-573-1-1 32.1 %

Apagbaala 30.3 %



Variation in aphid resistance among the world cowpea core collection grown in California, 2013

Valuable resource of resistance traits



Big Buff

Big Buff

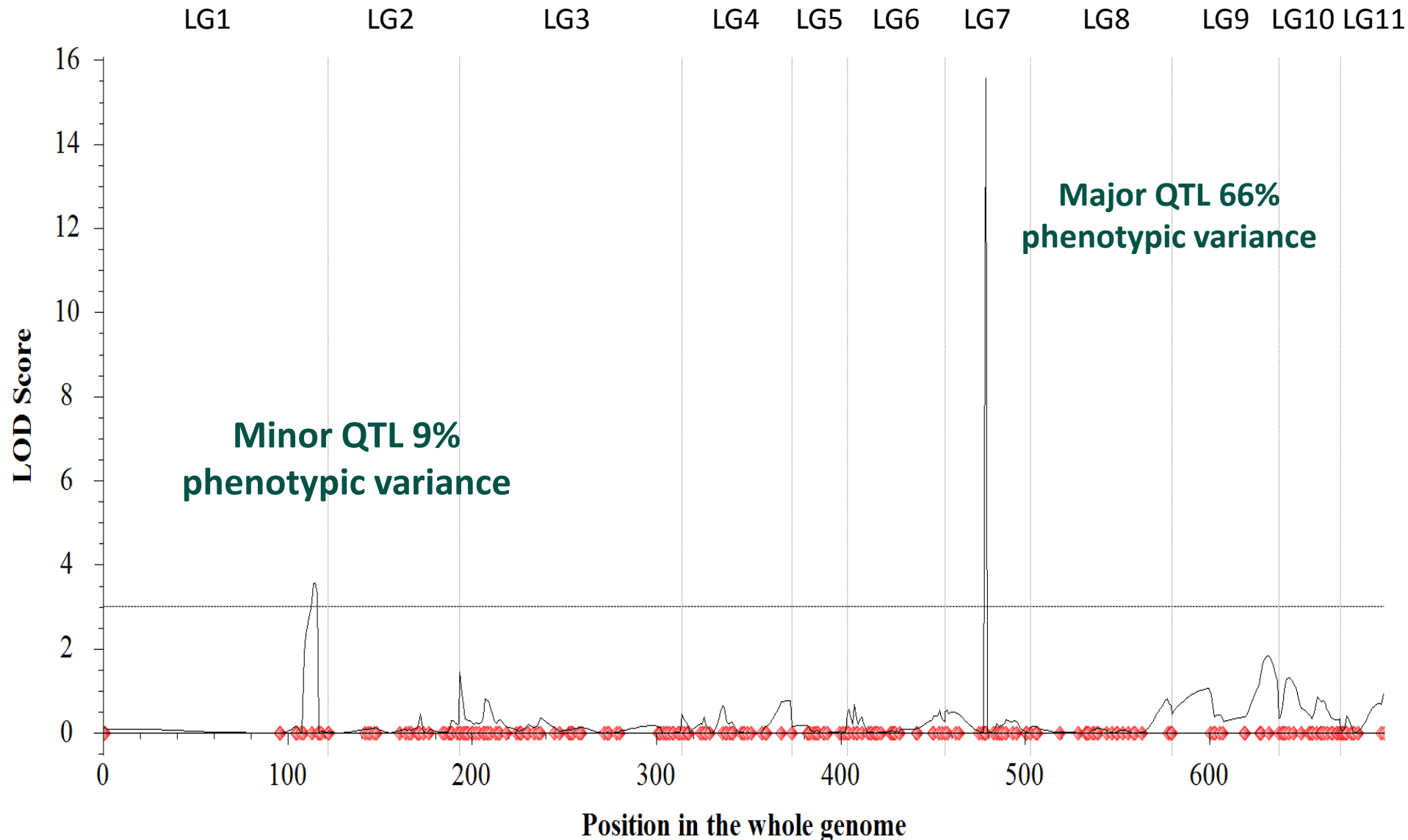
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Objective: Develop improved versions of elite cowpea varieties with effective aphid resistance from diverse sources

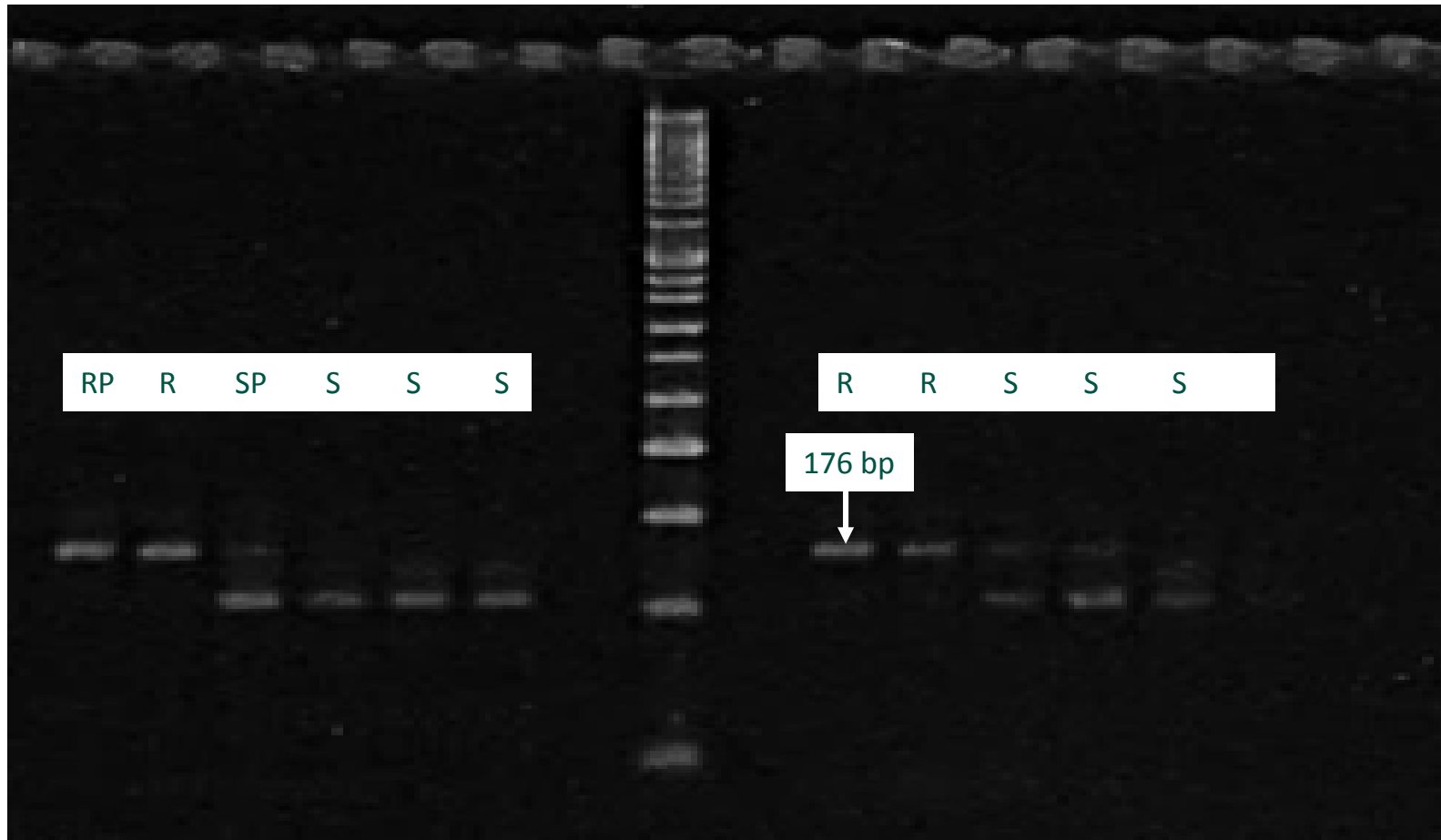
Example: New blackeyes in California with aphid resistance from IT97K-556-6



QTLs for aphid resistance in CB27 x IT97K-556-6



Combining SSR and SNP marker genotyping for expedited backcrossing (SARI Kusi & Agyare)



SSR marker CP 171F/172R linked to
aphid resistance locus in SARC 1-57-2

SSR marker CP171F/172R

23.55 cM on LG10

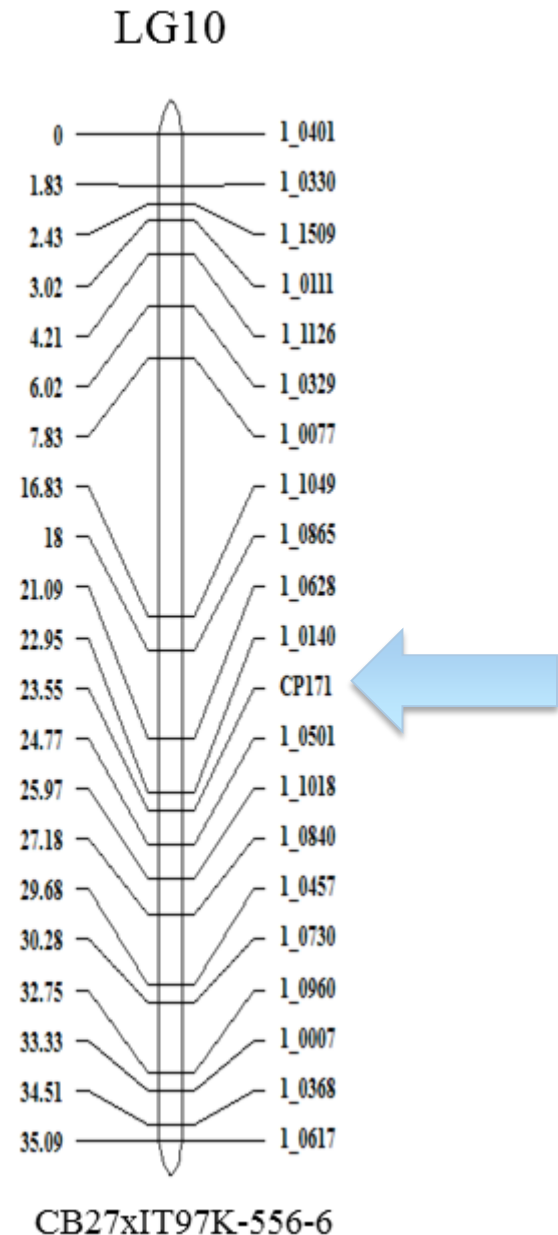
RIL mapping population CB27 x IT97K-556-6

(Richard Agyare, SARI Ghana)

Thus far:

3 independent resistance loci

LG 1, 7, 10



Aphid resistance breeding in Ghana using foreground and background MAS

Francis Kusi



Damage on
susceptible seedlings

Aphid infested
Zaayura seedlings

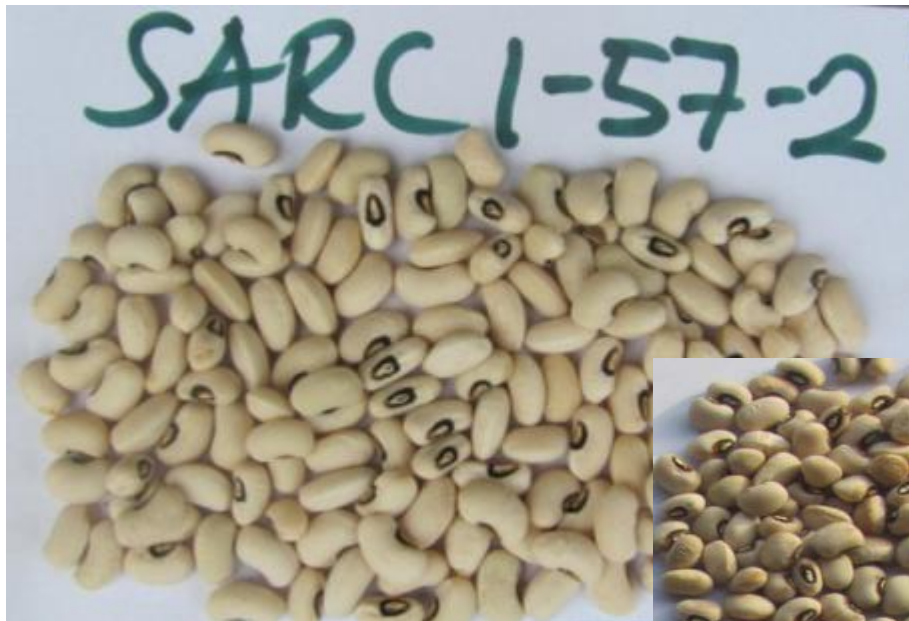


SARC-1-57-2
surviving aphid
attack



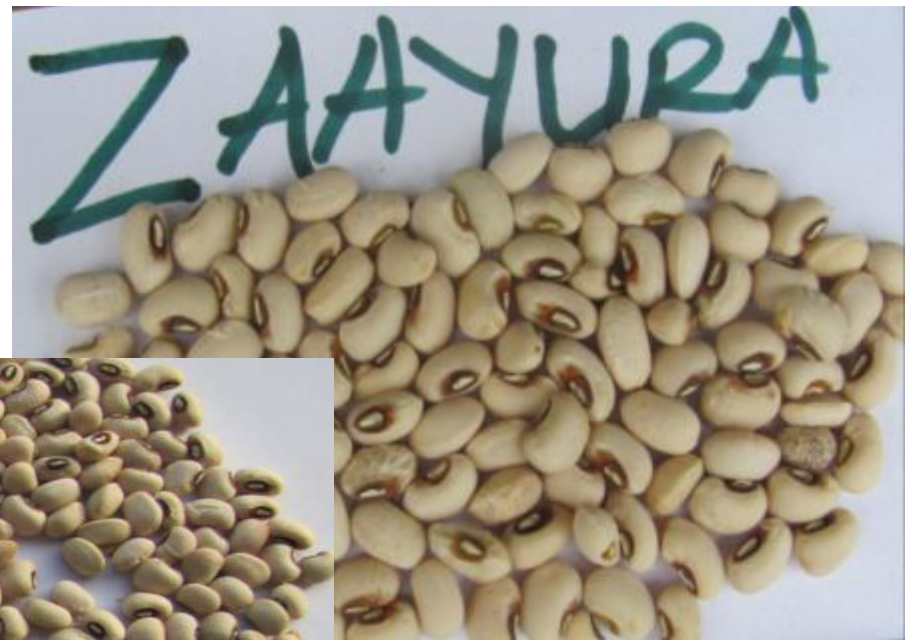
Aphid resistance breeding in Ghana using foreground and background MAS

Francis Kusi and Ibrahim Atokple



Aphid resistance donor

BC4F3 derived by MAS
for aphid resistance



SARI released variety
(recurrent susceptible)



Cowpea aphid resistance panel for determining resistance uniqueness and aphid biotypes

Cowpea genotype	Burkina Faso	Senegal	Ghana	Nigeria	Cal-USA
58-77	MR	R	R	R	MR
INIA19	R	R	--	S	R
IT97K-556-6	R	--	R	R	R
KN1	R	R	--	S	S
KvX-295-2-124-99	R	R	R	S	R
SARC-1-57-2	R	MR	R	R	S
TVNu-1158	--	--	--	R	R
APAGBAALA	S	S	S	MR	S
BAMBEY21	S	S	S	MR	S
CB27	S	R	R	S	S
IT82E-18	S	S	S	S	S



Contents lists available at ScienceDirect

Agri Gene

journal homepage: www.elsevier.com/locate/aggene



Comparison of complete mitochondrial DNA sequences between old and new world strains of the cowpea aphid, *Aphis craccivora* (Hemiptera: Aphididae)



Weilin Sun ^a, Bao-Lam Huynh ^b, James Adebayo Ojo ^c, Brad S. Coates ^d, Francis Kusi ^e, Philip A. Roberts ^b, Barry R. Pittendrigh ^{a,*}

- Mitogenome: 15,308 and 15,305 bp shotgun Illumina Hi seq2500 sequence reads
- 99.7% sequence identity with 57 and 11 substitutions predicted for NW and OW strains
- Divergence around 108,000 -188,000 years ago

Issues with linkage drag

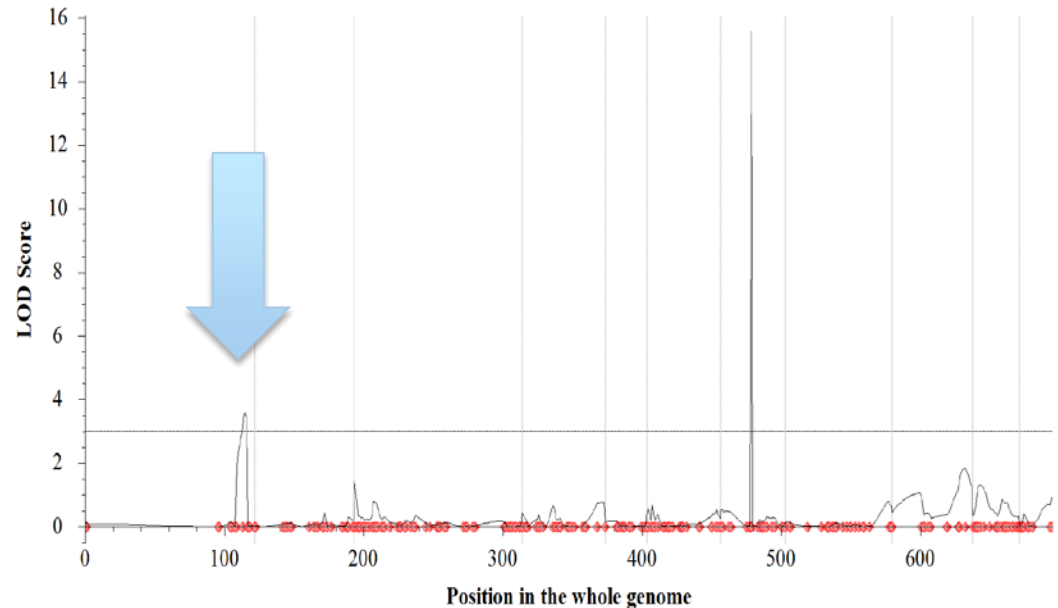
aphid resistance QTL on LG1 dragged brown eye
required two more generations to break linkage



**Black-eye
recurrent
parent**

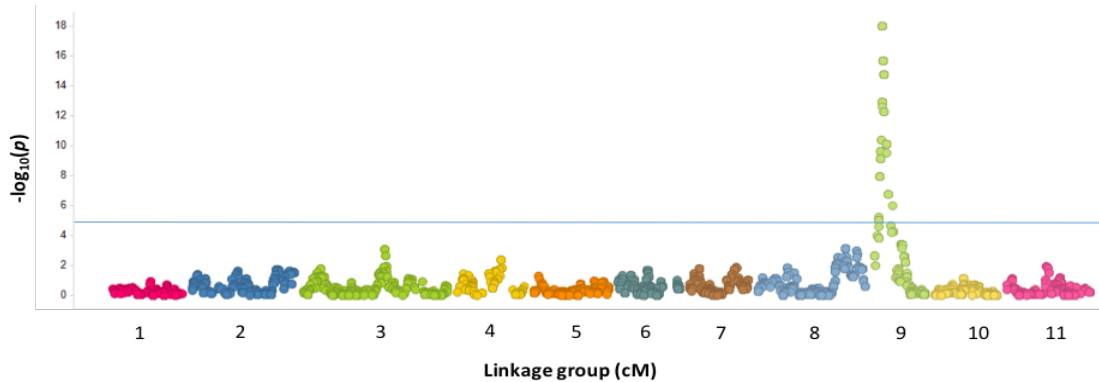


**Aphid
resistant
with
Brown-eye**

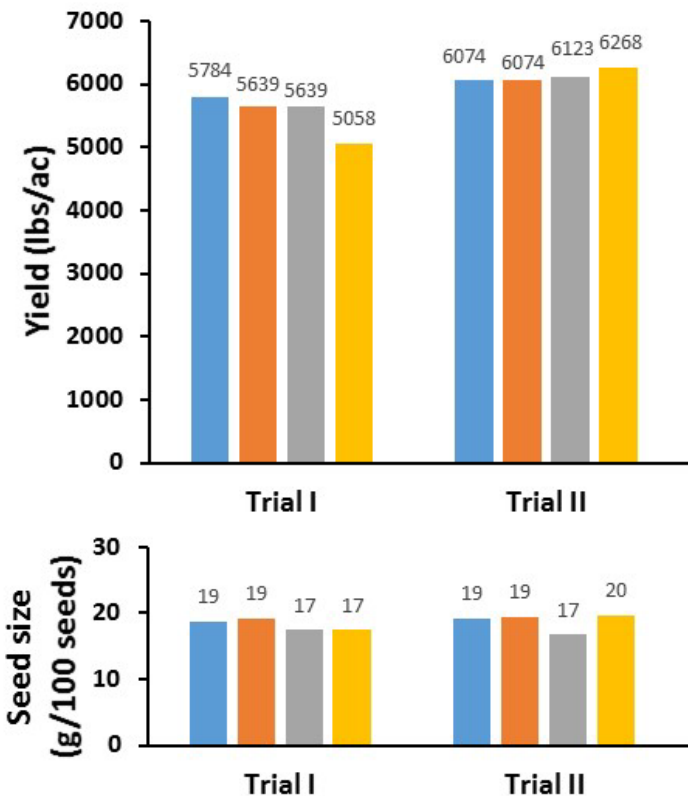


Linkage about 1cM: at cycle 3 no recombinants, but a few at cycle 5

M. javanica resistance on VuLG9 associated with small seed size IT84S-2049 donor



**Average of 15% reduction in seed size
compared to CB46
Releasing as a canning variety**



Flower thrips (*Megalurothrips sjostedti*)



Adult female (L) and 2nd instar (R)
Credit; G Goergen, IITA-Benin



Wilted flower buds (L) and adult thrips
(R) on cowpeas in Ghana (from I
Atokple)

Flower thrips damage in field screening trial, Senegal

Sorghum field at back is mature (80-90 days)

Cowpeas in screening trial have no pod-set, remain vegetative.

Normally harvested at 65-70 days



Field screening trial



Flower thrips symptoms

Parents of RIL mapping population under pressure from flower thrips

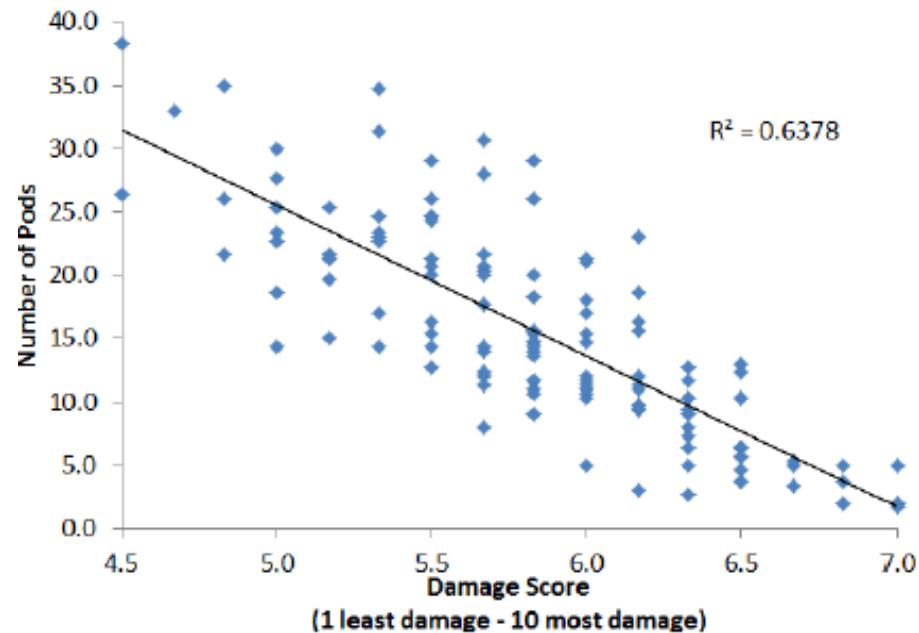
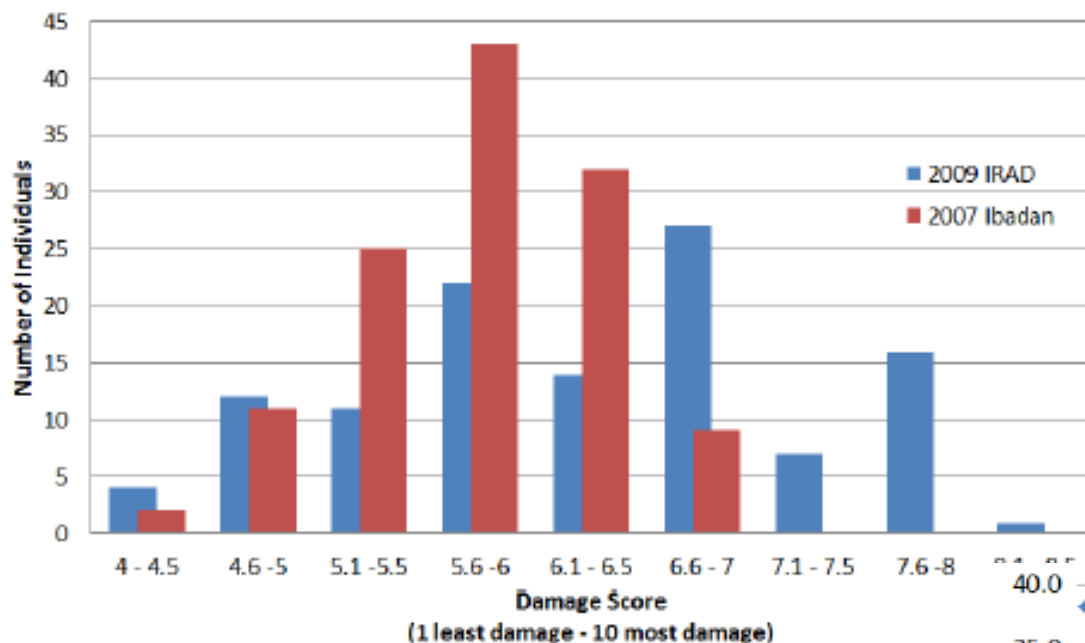


Sanzi (resistant)



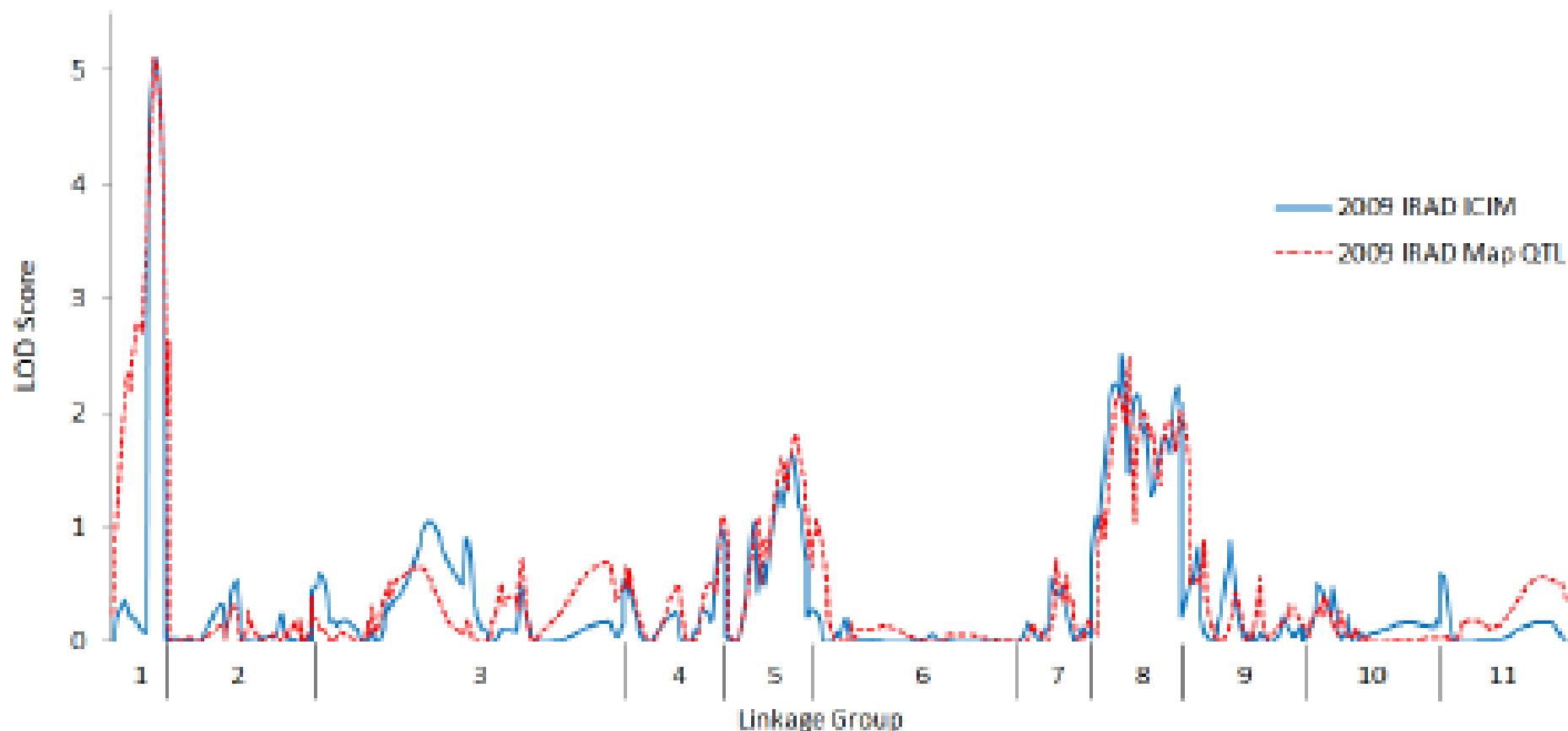
Vita 7 (susceptible)

Phenotype scores of flower thrips damage in Sanzi and Vita 7 RIL. (IRAD – blue; Ibadan – red)

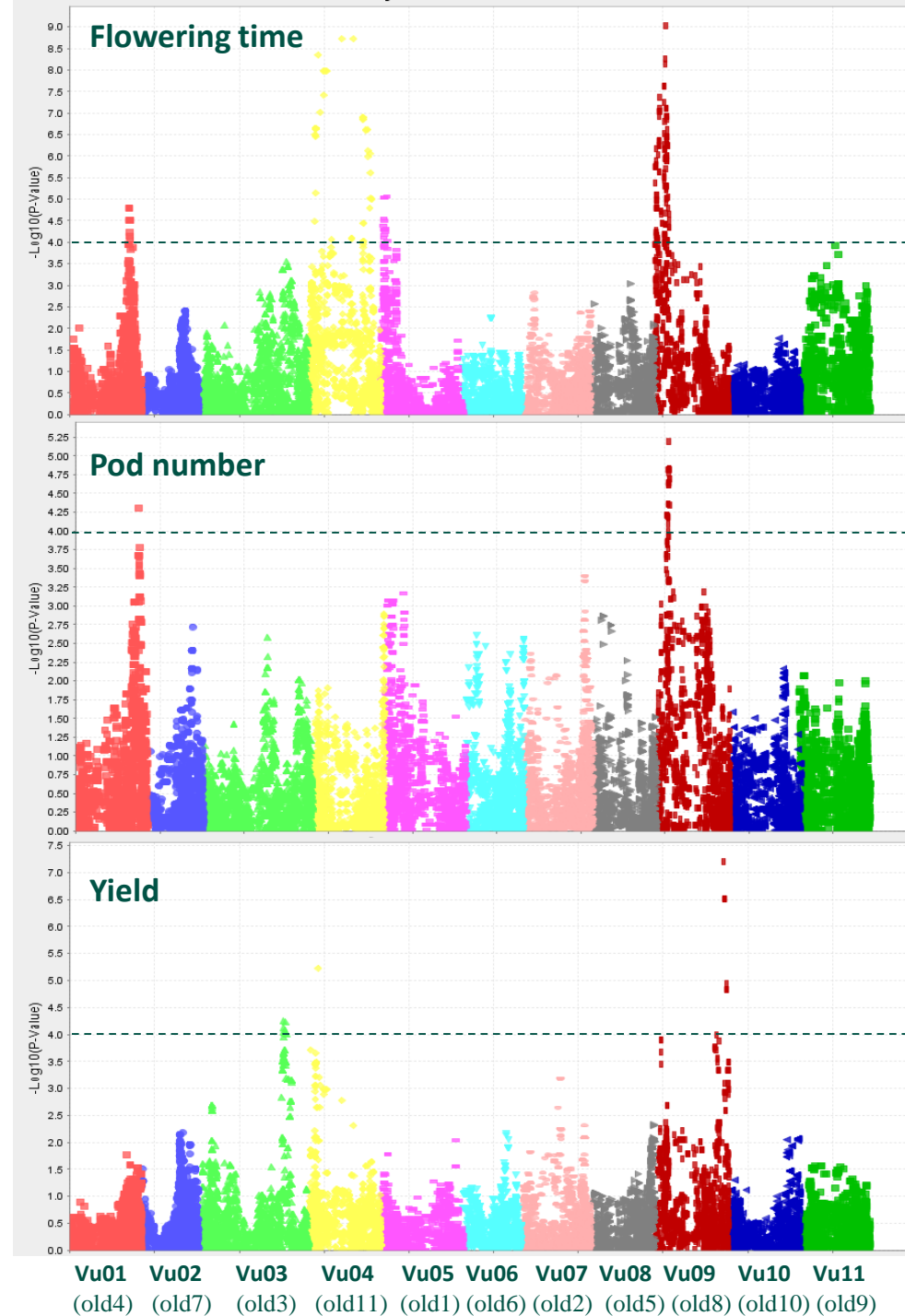


QTL for flower thrips resistance in Sanzi x Vita 7 RIL, phenotyped at IRAD, Cameroon

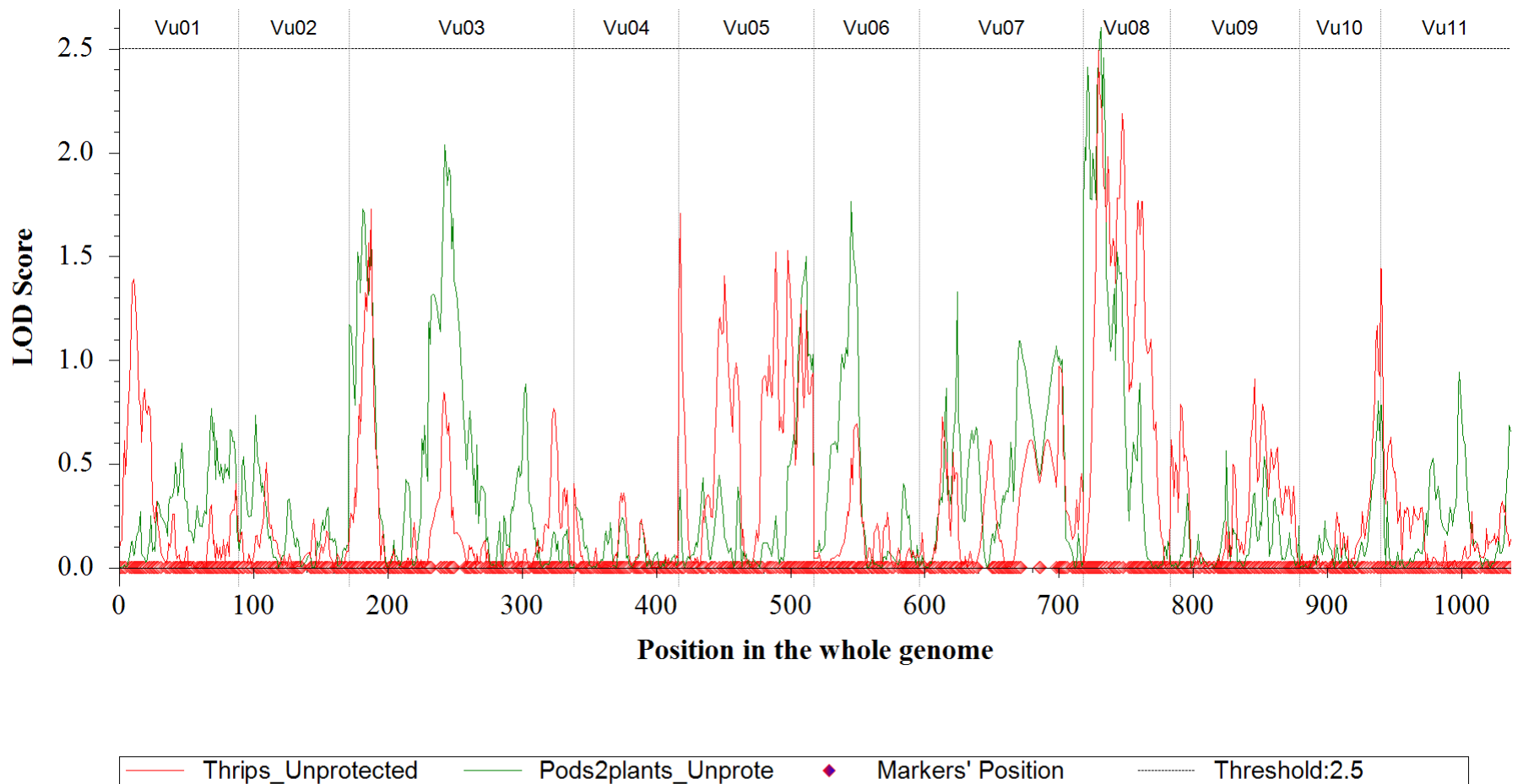
QTL identified on LG 1 (5 and 8?). ICIM and Map QTL



GWAS for MAGIC population under unprotected thrips attack in Senegal (60K SNP iSelect genotyping)



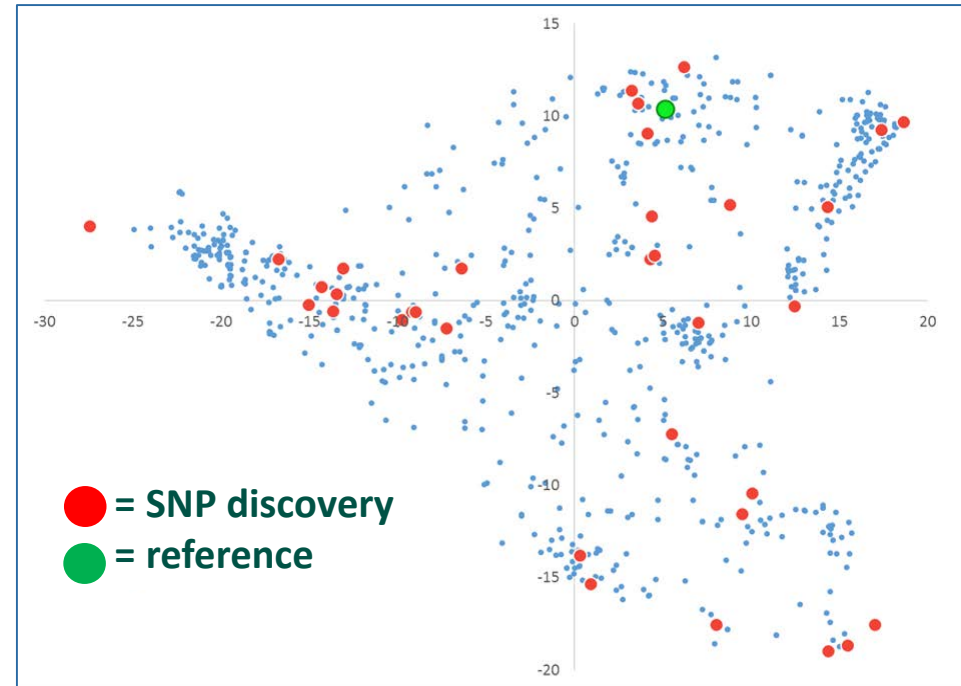
QTL on VuLG8 affecting flower thrips index and pod number in unprotected Yacine x 58-77 RIL population



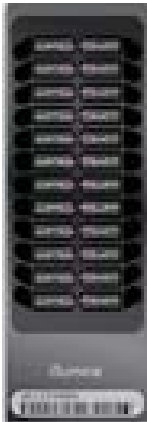
New resources -- high-density SNP assay

Illumina Cowpea iSelect Consortium Array

- ❑ **WGS sequencing of 36 accessions** (12.5x coverage), representing geographic, phenotypic and genetic diversity
- ❑ The **WGS assembly from IT97K-499-35** used as **reference** to discover SNPs



PCA Plot



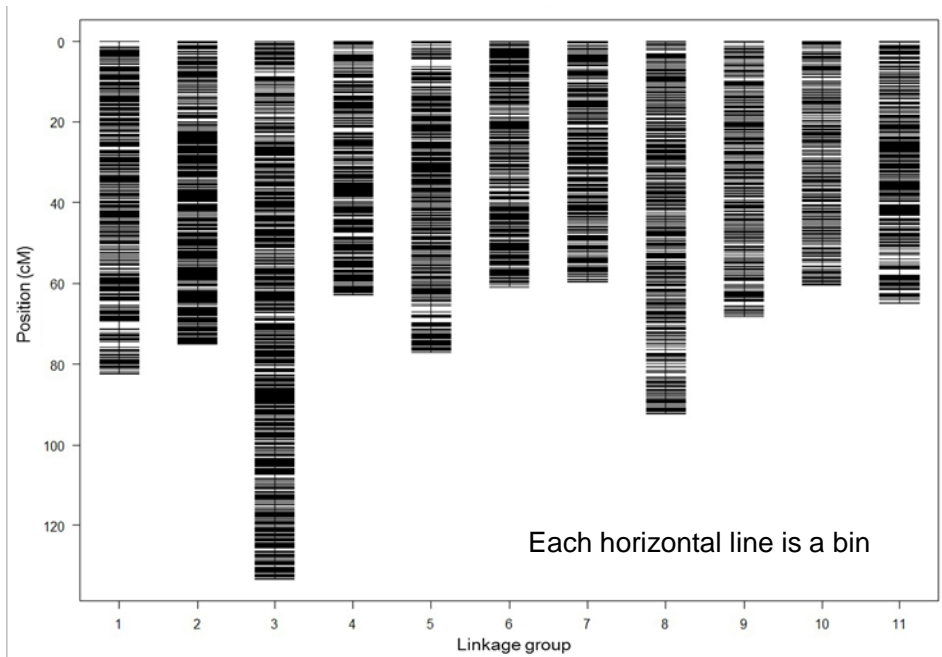
**Targeted gene models and
largest sequence contigs**

- 56,719 intended SNPs (60,000 assays)
- **51,128 SNPs** on the array (includes 1,040 from GoldenGate Assay)
- More than 49,000 SNPs provide consistent data (96%)

New resources -- Consensus Genetic Map

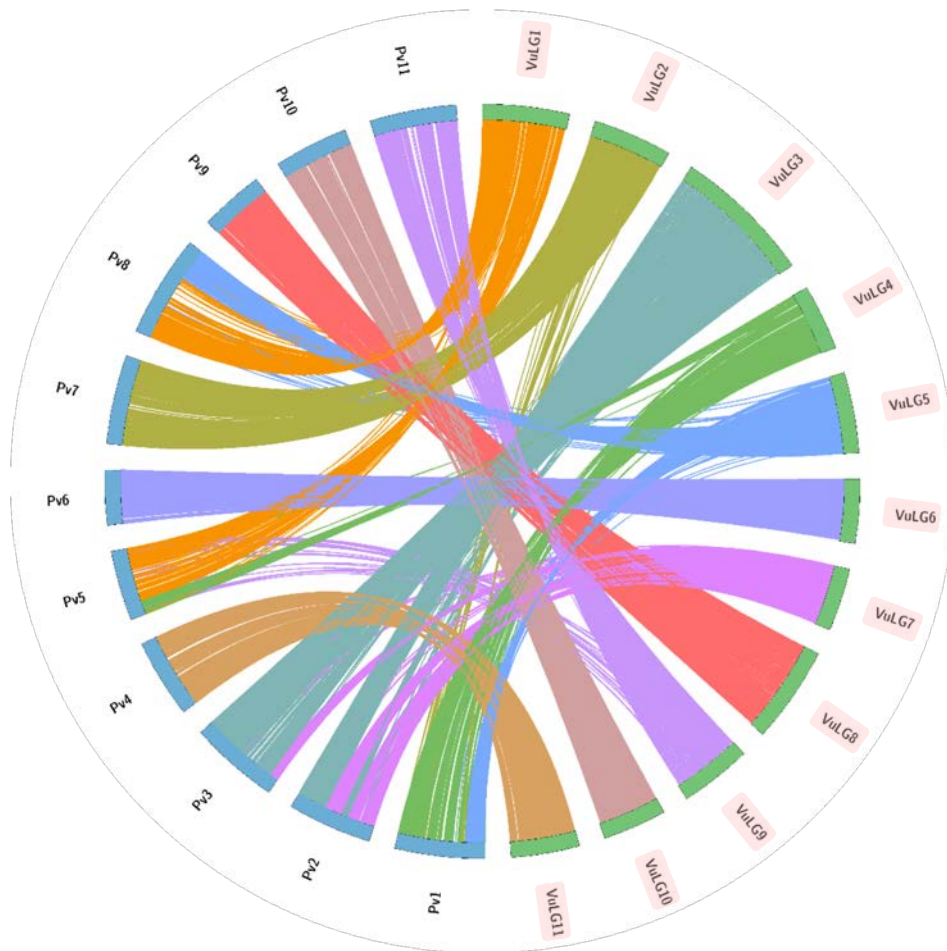
- ❑ SNP genotyping 5 RIL populations
- ❑ Construction individual genetic maps (MSTmap)
- ❑ Individual maps merged into consensus (MergeMap).

RIL Population	# Lines	# Mapped SNPs	# Marker bins
Tvu-14676 x IT84S-2246-4	135	14,660	1,216
Sanzi x Vita7	121	15,619	1,265
LB30 x LB1162	119	7,964	697
CB46 x IT93K-503-1	106	16,578	1,083
CB27 x IT82E-18	94	16,566	977



- ✓ **37,372 SNPs** at **3,280** unique positions (bins) ☞ 34-fold increase in density and 4-fold increase in resolution
- ✓ **837.11 cM** ☞ 1 bin per 0.26 cM
- ✓ Largest gap = 1.85 cM

Synteny with Common Bean (*Phaseolus vulgaris* L.)



Pv: *P. vulgaris* pseudomolecules.

VuLG: *V. unguiculata* linkage groups.

✓ Cowpea iSelect SNP design sequences were related to *P. vulgaris* pseudomolecules by MUMmer.

✓ Extensive synteny:

- 6 cowpea LGs are syntenic with single common bean pseudomolecules.

VuLG2 = Pv7

VuLG9 = Pv11

VuLG6 = Pv6

VuLG10 = Pv10

VuLG8 = Pv9

VuLG11 = Pv4

✓ Common chromosome numbering is needed for both, and other *Vigna* species.

New resource 8-parent MAGIC population 305 RI lines

			High Yielding Under Drought in:				
Genotype	Source	Diversity Group	Senegal	Burkina Faso	Mozambique	Nigeria	Key Traits
CB27	UCR	A			Yes		Heat tolerance
IT00K-1263	IITA	F			Yes	Yes	<i>Striga</i> (races 1,2,3,4,5) resistance
IT82E-18	IITA	B			Yes		Broadly adapted, high yield
IT84S-2049	IITA	C		Yes			Aphid, bacterial blight, CAbMV Root-knot nematode resistance
IT84S-2246	IITA	G		Yes	Yes		Aphid, bacterial blight, CAbMV Root-knot nematode resistance
IT89KD-288	IITA	D		Yes		Yes	Yield, grain quality
IT93K-503-1	IITA	H	Yes				Drought tolerance, <i>Macrophomina</i> , <i>Striga</i> (races 1,3,4) resistances
SuVita 2	INERA	E	Yes	Yes	Yes		<i>Striga</i> (races 1,2,4) resistant, drought tolerance, high yield

8-parent MAGIC population

UC Riverside, California – Summer 2015



Photoperiod, maturity,
pod load, growth habit

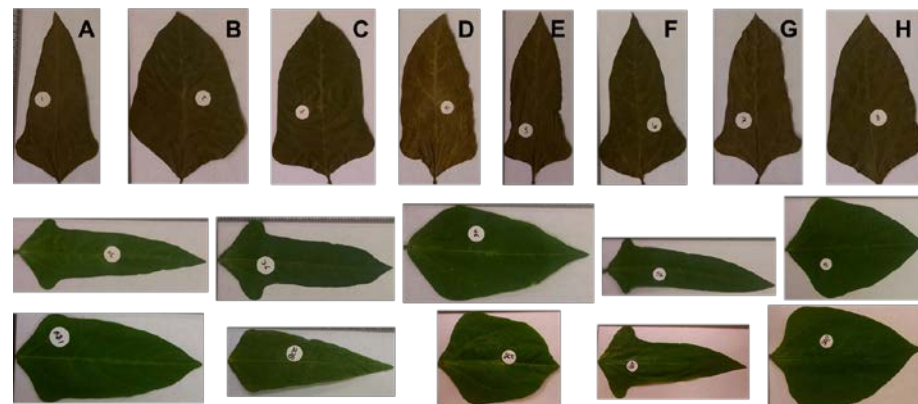
Seed type
(parents
top panel)



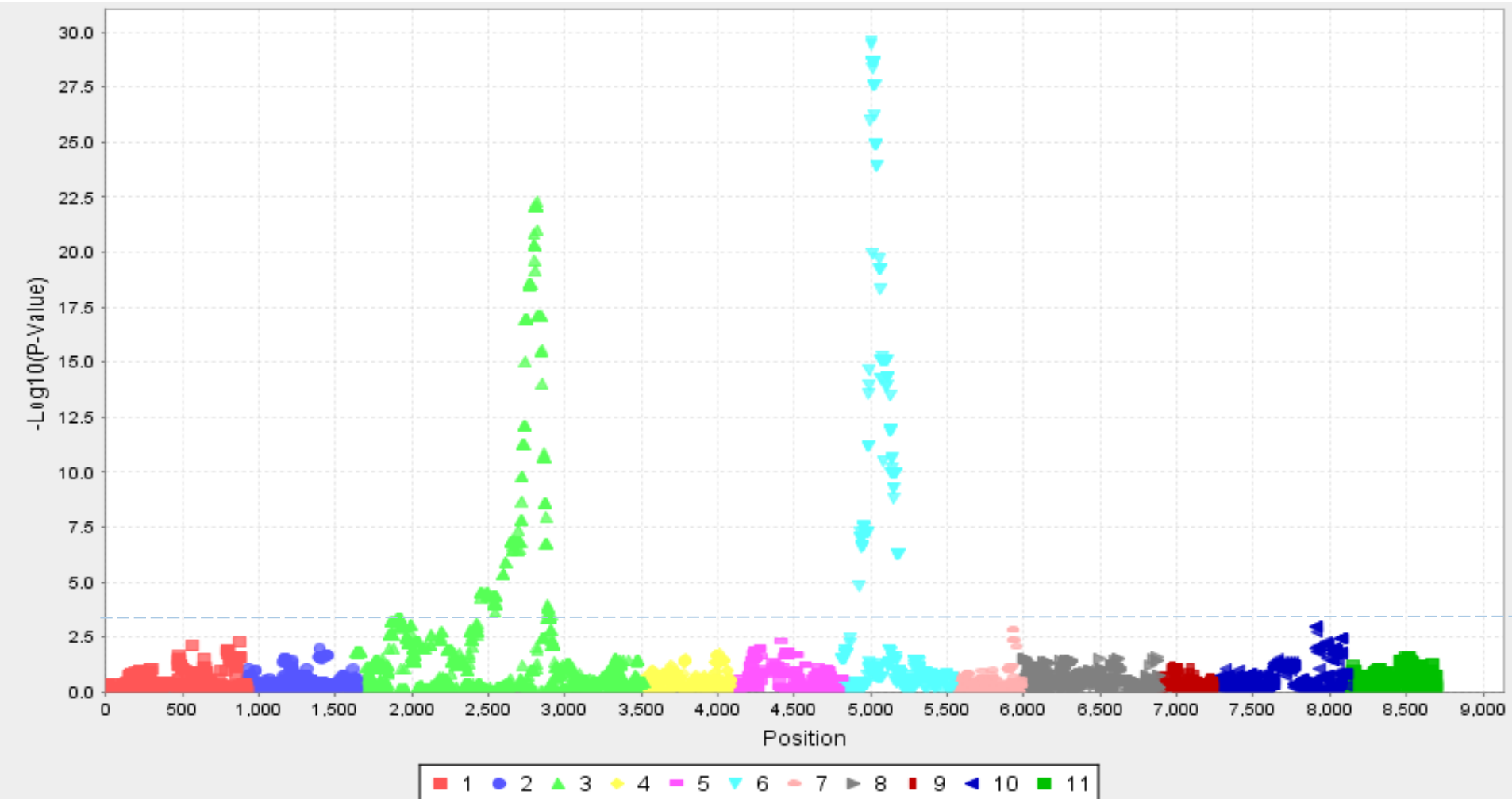
Flower color
(parents top panel)



Leaf shape
(parents
top panel)



MAGIC: Genome scan for Fusarium resistance



Peaks coincided with QTLs mapped
in bi-parent RILs Pottorff et al. (2014)
BMC Genomics 15:328

Positive alleles from
the resistant parents
CB27 and IT00K-1263

Objective 2: Complete release and validation of advanced cowpea lines developed under the Pulse CRSP in Burkina Faso, Senegal, and US.

Collaborators:

Dr. Ousmane Boukar, IITA, Nigeria

Dr. Samba Thiaw, ISRA, Senegal

Dr. Mywish Maredia, Michigan State U., USA

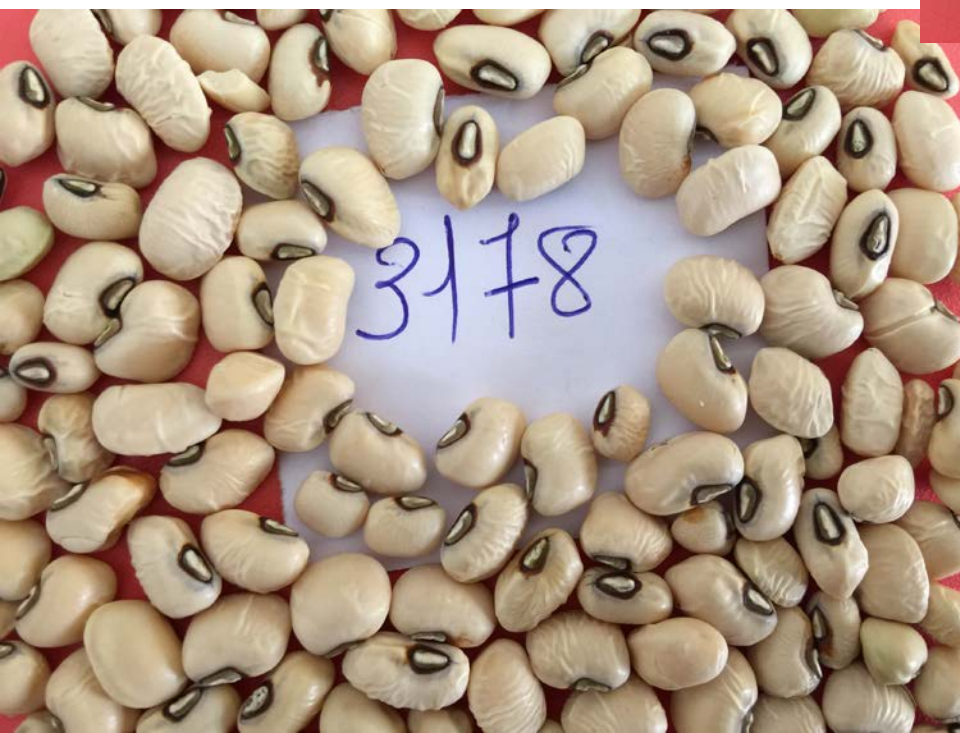
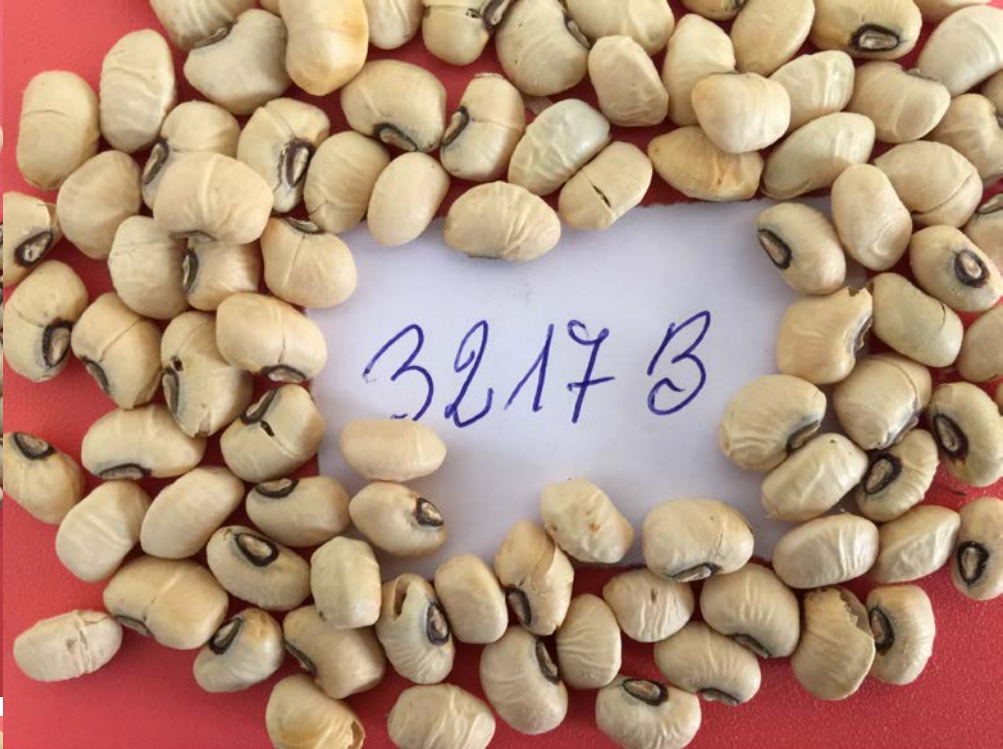
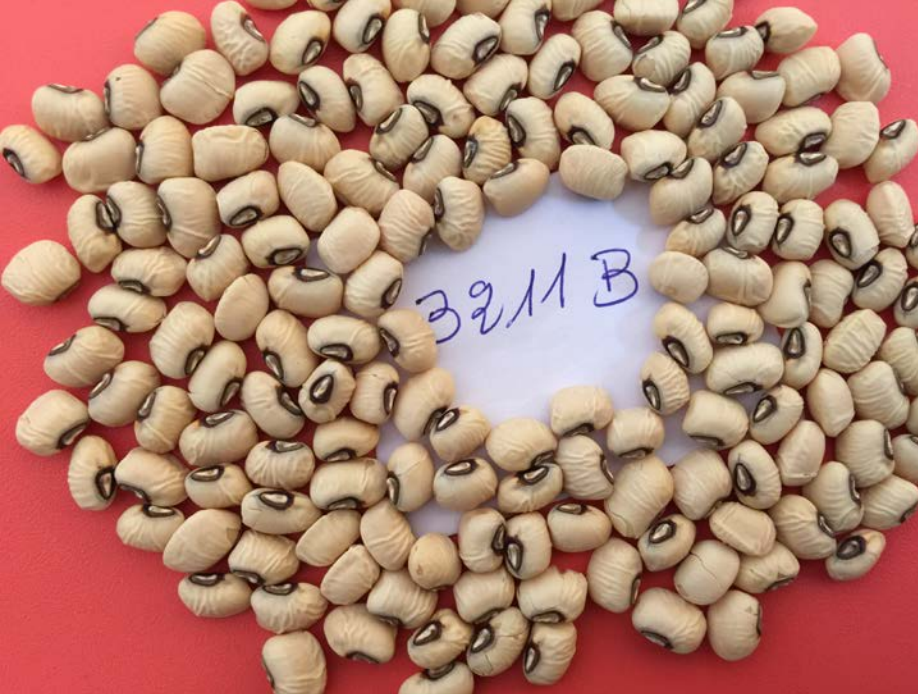
Burkina Faso: 4 released CRSP lines

Senegal: 5 large white released CRSP lines

USA: Lygus, Fusarium and nematode resistant blackeyes

Large-seeded white grain types for Senegal release:
Montiero source crossed into Senegal elite Melakh (N. Cisse)
All lines *Bacterial Blight* and *CpMv* resistant; line 3217 *Amsacta* tolerant

Lines	Yield10 Station	Yield12 On-farm	Yield13, On- farm	Days to Maturity	100 Seed-wt
3178	1767	859	606	59.8	26.8
3217	1871	824	687	59.5	25.8
3211	1360	739	512	60.5	25.8
3205	1551	709		62.8	26.5
MELAKH	1455	698	627	60.0	20.3
3201	1441	670		59.8	26.3



Pre-release white grain types for Burkina Faso (I. Drabo)

Striga resistant; range of disease resistances; high biomass yield







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Thankyou



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