

# Role of Pulse Crops in Achieving Food and Nutritional Security

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Dry Grain Pulses CRSP  
GLOBAL PULSE RESEARCHERS MEETING  
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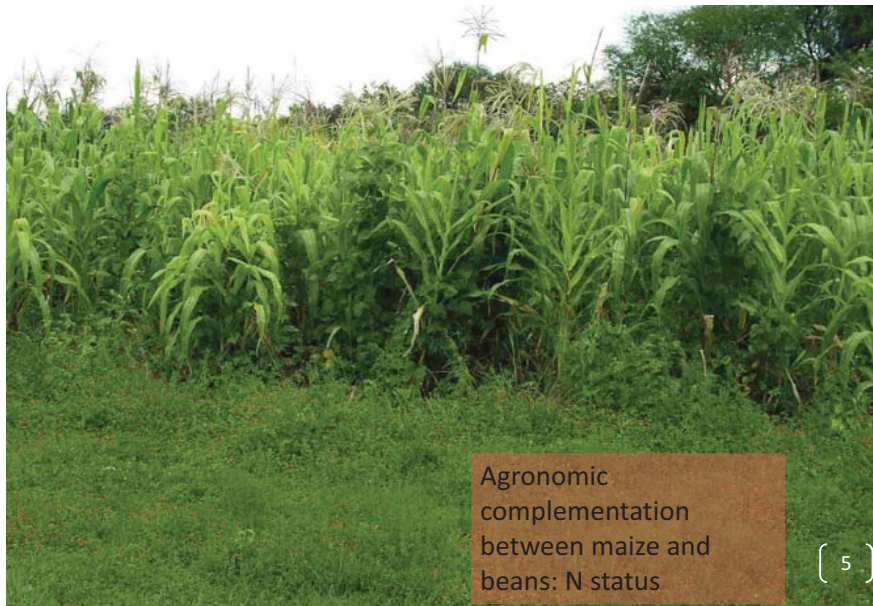
## Outline

- Role of Grain Legumes
- Goals of Grain Legume Improvement
- Key Ingredients
  - Biodiversity:
    - Farmer role in conservation
    - Ecological genomics
  - Breeding methods & genomics
    - Marker-assisted selection
    - Participatory breeding
  - The role of phenotyping in breeding for abiotic stresses
  - Funding
- Conclusions

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## Role of Grain Legumes or Pulses

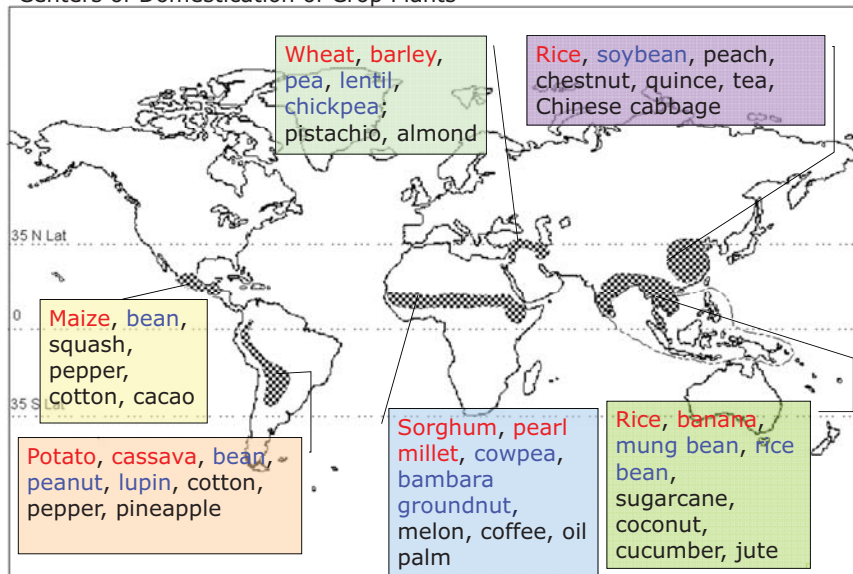
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San Agustín del Pulque, MICH, MEX (2004)



Centers of Domestication of Crop Plants



Modified from Gepts 2004

Goals of Grain Legume (Pulse) Improvement

## Goals

- Maximize yield within existing phenological constraints and for target environment
- Minimize inputs and maximize input use efficiency
- Improved human nutrition
- Farmer acceptance
- Consumer acceptance and marketability
- Any approach is fine, regardless of technology!

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## Key Ingredients: Biodiversity

### 1 Farmer Role in Conservation

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## Role of Farmers?



Photos: D. Zizumbo

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## Location of Michoacán-Guanajuato experiments



Site	Distance* (m)
Jéruco	10
Cepio	15
Piñicuaro	20
Santa Ana Maya	20
Tupátaro	25
Yuriria	60
San Agustín del Pulque	110

\* Between wild and domesticated populations

Payró de la Cruz et al. Genet Res Crop Evol 2005

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## Wide range of diversity including improved cultivars and wild beans



	Average seed weight (g/100)
Wild	6
Weedy	20
Domesticated	39



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Environs of Yuriria, Guanajuato-Michoacán, Mexico

## Different Genetic Structure in W & D

	$H$	$h$	$G_{ST}$	$Nm$
Wild	0.24	0.13-0.18	0.40	0.78
Landraces	0.26	0.22-0.29	0.26	1.40
Breeding lines	--	0.03-0.06	--	--



- Wild: less diversity within populations, more among populations compared to Domesticated
- Higher gene flow in Domesticated:
  - Diversity within landraces: outcrosses
  - Seed exchange among farmers

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Zizumbo-Villarreal et al. 2005

## Study in Yuriria



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S. Barber & P. Gepts, unpubl. results

## Distinguishing two groups in a single region (Yuriria, GUA, MEX)

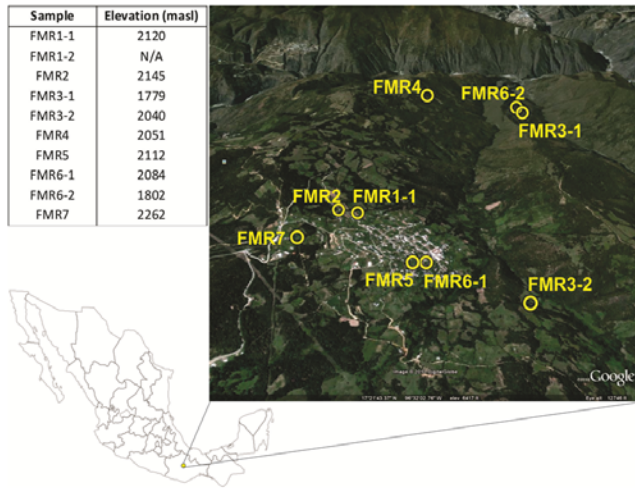
<i>Frijol</i>	<i>Frijola</i>
late harvest in Nov-Dec/ thicker seedcoats – longer to cook/ longer storage time	early harvest in Sep-Oct/ thinner seedcoats – shorter to cook/ shorter storage time
apetito morado	amapola delgada
cacahuate	color de rata
cafe	flor de mayo
higuerillo	flor de mayo gruesa
higuerillo delgado	flor de mayo delgada
higuerillo grueso	flor de junio
huamuchil	flor de junio - <i>Marcela</i>
morado bola	japonés de bola
morado grueso	morada (Rosita)
guindo (rojo)	ojo de cabra
palacio	ojo de liebre
pinto	peruano
viudo	de castilla

S. Barber & P. Gepts, unpubl. results

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## Study in Santa María Jaltianguis, OAX, MEX

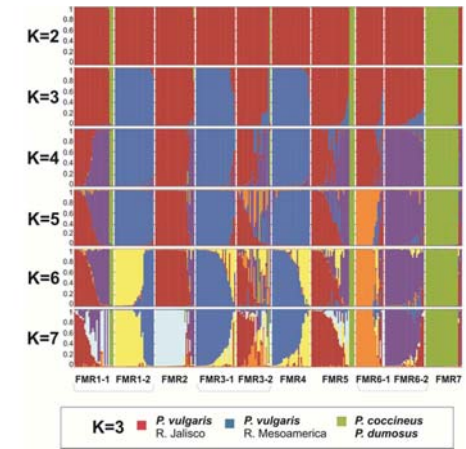
Sample	Elevation (masl)
FMR1-1	2120
FMR1-2	N/A
FMR2	2145
FMR3-1	1779
FMR3-2	2040
FMR4	2051
FMR5	2112
FMR6-1	2084
FMR6-2	1802
FMR7	2262



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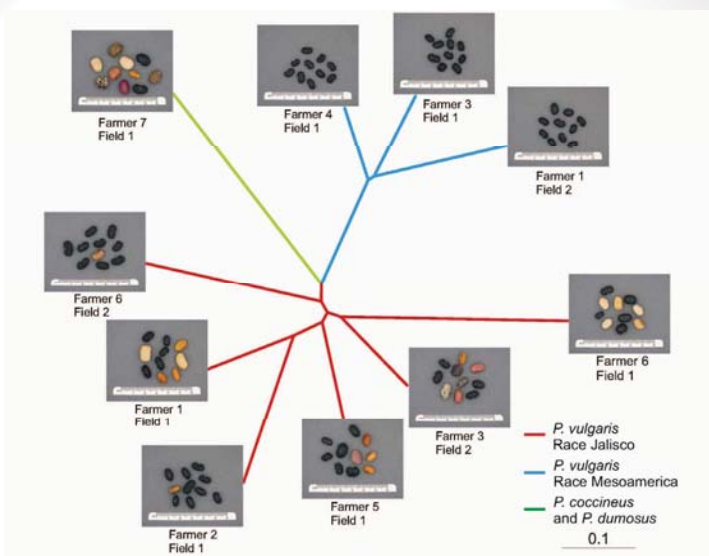
M. Worthington, D. Soleri, & P. Gepts, submitted

## Farmer selection according to species & adaptation



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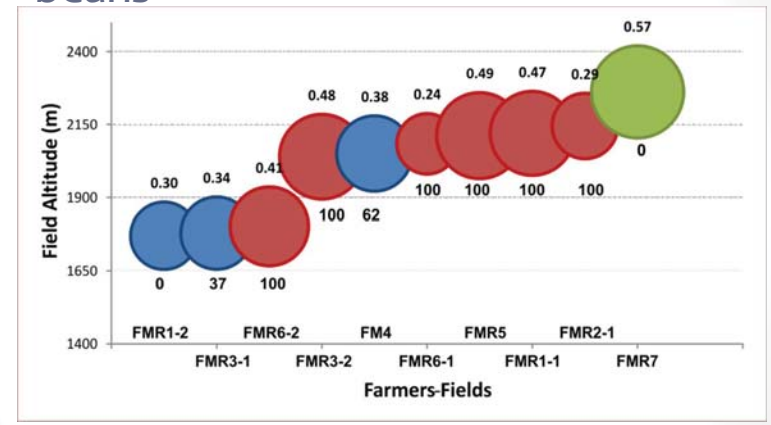
M. Worthington, D. Soleri, & P. Gepts, submitted



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M. Worthington, D. Soleri, & P. Gepts, submitted

## Altitude distribution of different beans



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M. Worthington, D. Soleri, & P. Gepts, submitted

## Conclusions: Farmer-Involvement in farm biodiversity management

- Awareness of adaptation of germplasm segments
  - Climate
  - Phenology
- What type of selection?
  - Stabilizing selection: Maintenance of type
  - Directional selection? Yield?
  - Maintain genetic diversity; Dynamic system: not a gene bank nor a museum
- Niche for intervention by participatory breeding
  - Improved germplasm
  - Selection procedure

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## Key Ingredients: Biodiversity

### ② Ecological Genomics

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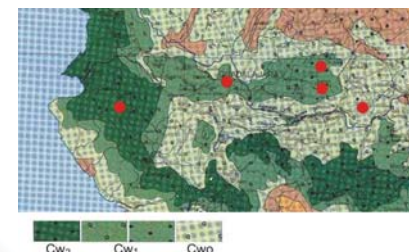
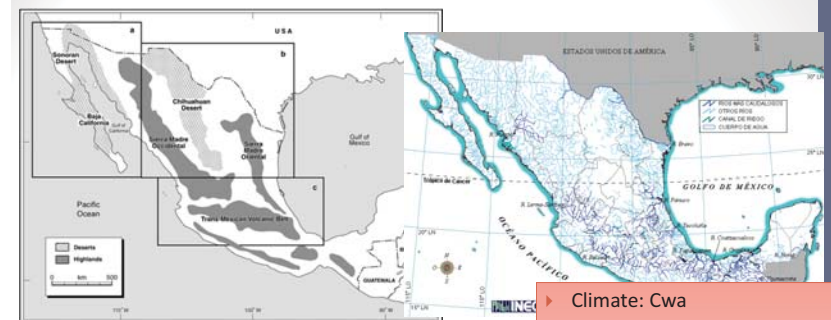
## THE SUGGESTED DOMESTICATION CENTER OF COMMON BEAN IN MEXICO



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M. Kwak, J. Kami & P. Gepts, Crop Sci., March 2009

## WHY THE LERMA-SANTIAGO BASIN?



- ▶ Climate: Cwa
  - ▶ Subtropical:  $t^{\circ}$  coldest month: 5-18  $^{\circ}$ C
  - ▶ Subhumid: 4-6 months of humidity in summer
  - ▶ Semi-warm: average annual  $t^{\circ}$ : 18-22  $^{\circ}$ C
- ▶ Vegetation:
  - ▶ Dry deciduous forest to drier thorn forest

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## WESTERNMOST PUTATIVE DOMESTICATION LOCATION, MASCOTA-AMECA BASIN



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## WESTERNMOST PUTATIVE DOMESTICATION LOCATION, MASCOTA-AMECA BASIN



Dec. 5, 2008

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## Eco-geographic variation of bean landraces in Brazil

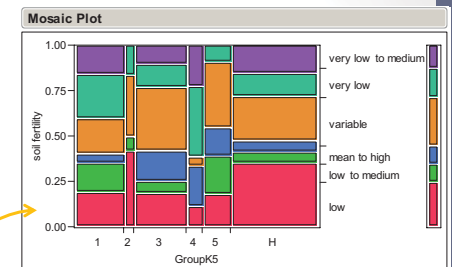
- Biome: mainly semi-deciduous forest, pine forest
- Only difference between A and M?
  - Altitude: ~ 100m
  - Yearly average T°: 23C
  - Average rainfall growing season: 549 mm



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## Differences among K = 5 STRUCTURE groups and market classes

Eco-geographic variables	Prob>ChiSq
Biome	0.0086
Geomorphology	0.0639
Soil slope	0.0009
Soil fertility	<0.0001

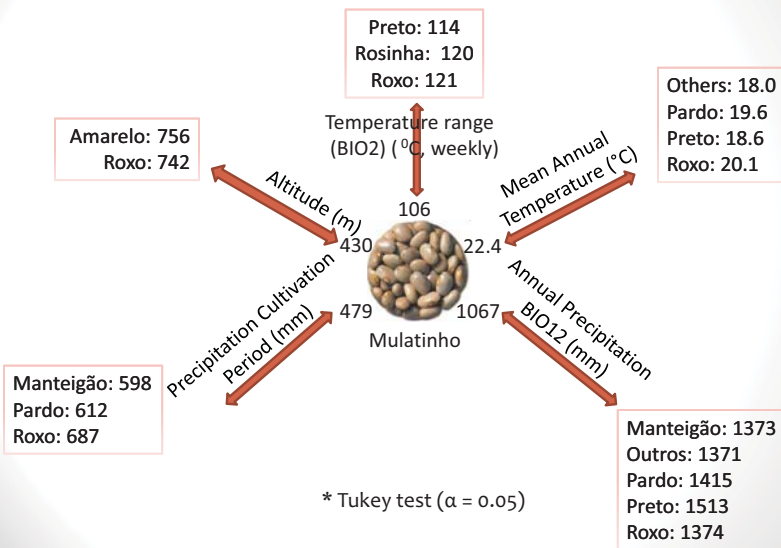


ANOVA: 'Mulatinho' market class: lower altitudes, precipitation; higher temperature



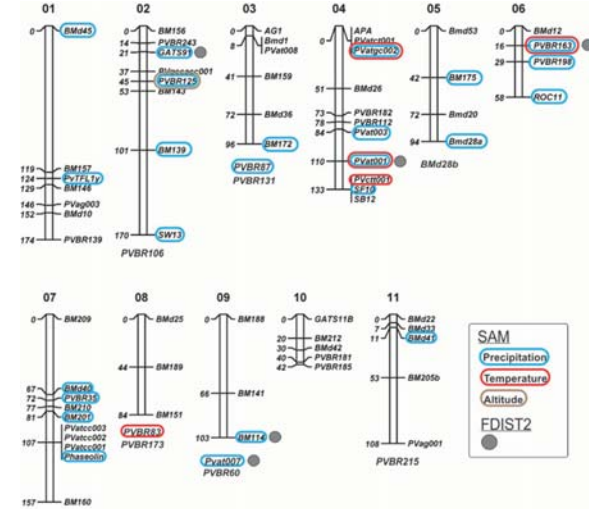
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## Significant\* Contrasts between Mulatinhos and Other Commercial Types



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## Molecular Linkage Map of Common Bean: Significant Correlations with $T^\circ$ , P, and Altitude



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## What is next?

- Evaluation of plant materials (a.k.a. phenotyping)
  - Yield and component traits
  - Germplasm
  - Locations/years
- High density of markers
  - Fragments (SSRs, indels), SNPs
  - Number: the more the better
- Other sources of genomic/genetic information
  - Transcriptomics
  - Metabolomics

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## Key Ingredients: Breeding Methods & Genomics

- 1 Marker-assisted selection

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## Progress in CBB resistance breeding

Miklas et al. 2006

Table 1. Representative dry bean lines and cultivars with resistance to common bacterial blight (CBB) derived from individual or combined sources

1970, 1994, 1973 Lines & cultivars	Sources				Disease score (1-9) <sup>a</sup>
	<i>P. vulgaris</i>		<i>P. coccinea</i>	<i>P. acutifolius</i> (Tepary markers)	
	Montana No. 5	PI 207262			
Jules, Chase, Montcalm	×				6, 7, 8
XAN 112, BAT 93	×	×			4, 5, 6
XR 235-1-1			×		5, 6
USPT-CBB-1	×		×		4, 5, 6
OAC 88-1				× (SU91)	4, 5, 6
HR 67				× (BC420)	3, 4, 5
XAN 159, CBB-Teebus				× (SU91 and BC420)	2, 3, 4
ABCP-8, USDR-CBB-15	×			× (SU91)	3, 4, 5
Wilkinson 2	×		×	× (SU91 and BC420)	2, 3, 4
XAN 309, VAX lines 3-6	×	×		× (SU91)	1, 2

<sup>a</sup>Relative average disease scores (1-9, where 1 is no visible symptoms and 9 is completely diseased) compiled from published and unpublished data.

1999



- 1) improved screening methods;
- 2) identified resistant *P. vulgaris*, *P. coccinea*, and *P. acutifolius* germplasm; e.g., Miklas et al. 2003; Singh & Nuñez 1999; etc.
- 3) molecular mapping to tag resistance loci (Nodari et al. 1993; Miklas et al. 1996; Jung et al. 1997; Miklas et al. 2000); and
- 4) pyramided resistance genes (Miklas et al. 2006b; Mutulu et al. 2005).

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## Where do we go from here?

- Other diseases?
  - White mold (Miklas et al. 2009; Soule et al. 2011; Mkwaila et al. 2011; Pérez-Vega et al. 2012)
  - Web blight (Godoy-Lutz et al. 2003; Takegami et al. 2004; Beaver et al. 2012)
- Generalize use of MAS?
  - Remains a tool and not a goal
  - Under what conditions is MAS applicable or not as a tool?
  - Other traits and genetic architecture (QTLs)
- Coordination among “players”: Kirkhouse Trust ABC and WAC project, CIAT
  - Development of new markers: whole-genome sequencing and PhaseolusGenes database
- Other species: e.g., *P. lunatus*

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KIRKHOUSE TRUST ABC ANNUAL MEETING  
17/05/2010 - 21/05/2010 - ICE - SUA MOROGORO - TANZANIA

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### Kirkhouse Trust: African Bean Consortium (ABC) Project

- Main Goal: Introduce MAS capability in East African bean breeding programs
  - Focused on BCMV, ALS, ANT, CBB, Pythium RR → Preferred variety + 2-3 resistances
- Institutional improvement focused on Africa
  - National programs; education within Africa
- Main strategic elements
  - Human infrastructure: markers, plant pathology (CIAT-Kawanda)
  - Physical infrastructure: DNA fragment analysis; SNPs?
- At UC Davis:
  - 1x methyl-filtrated BAT93 sequencing
  - PhaseolusGenes database
  - Training of African students (applied bioinformatics, lab analysis, PhaseolusGenes)

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# PhaseolusGenes

Bean Breeder's Molecular Marker Database

ABOUT/METHODS GENOME BROWSER CMAP DOWNLOAD

**PhaseolusGenes**

PhaseolusGenes is a web resource for identifying and exploring markers, quantitative-trait loci (QTL), and simple sequence repeat (SSR) region information for *Phaseolus vulgaris*. Use this database to find marker information and explore them through the associated UCSC Genome Browser and Cmap implementations.

To use PhaseolusGenes, enter a search term in the right or click below for advanced options. You may also enter a sequence to use our online blast tool to blast a sequence against the soybean genome.

**Explore PhaseolusGenes**

**Markers**  
Explore data from various traits, as well as searching sequences.

**QTL**  
Search PhaseolusGenes' QTL list by entering traits in the search box to the right. Alternatively, you can see an entire list of searchable traits.

**SSR**  
We have a number of confirmed and predicted SSR regions in the database, accessible by searching to the right, or blasting suspected SSR regions against the soybean genome with our Blast Search feature.

**Brought to you by:**

**Data Contributors**

- Jan Brazdil & Peter Pauly, Guelph University
- Jim Kelly, Michigan State University
- Phil McClean, North Dakota State University
- Phil Malen & Richard Lorenz, USDA-ARS, Prosser, WA
- Thiago L. P. O. de Souza, Evandro de Barros & Maurício Moreira, Universidade Federal de Viçosa, Brazil
- Kangfu Yu, Agriculture Canada, Greenhouse and Processing Crops Research Centre, Ontario, Canada

**Funding**

- The Kirkhouse Trust, <http://www.kirkhousetrust.org/>
- The United States Department of Agriculture, National Institute of Food and Agriculture, <http://www.nres.usda.gov/>

Search markers

Learn to use PhaseolusGenes

Search term:   
Search field: All fields

Blast search (soybean)

(Blast w/ e-value cutoff 0.0001)

Enter one sequence in simple text

Phaseolusgenes.bioinformatics.ucdavis.edu

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# PhaseolusGenes: QTLs

ABOUT/METHODS GENOME BROWSER CMAP DOWNLOAD

Address: <http://phaseolusgenes.bioinformatics.ucdavis.edu/>

**QTL trait list**

Back to main page

**QTL Trait list**

- %I-3 score
- %I-4 score
- 100 seed weight (g)
- Adventitious root (HIGH PHOSPHORUS) Biomass (greenhouse)
- Adventitious root (HIGH PHOSPHORUS) Length (field)
- Adventitious root (HIGH PHOSPHORUS) Length (greenhouse)
- Adventitious root (HIGH PHOSPHORUS) Number (field)
- Adventitious root (HIGH PHOSPHORUS) Specific root length (field)
- Adventitious root (HIGH PHOSPHORUS) Specific root length (greenhouse)
- Adventitious root (LOW PHOSPHORUS) Biomass(field)
- Adventitious root (LOW PHOSPHORUS) Number (field)
- Adventitious root (LOW PHOSPHORUS) Specific root length (field)
- Adventitious root (LOW PHOSPHORUS) Specific root length (greenhouse)
- Angle (degrees)
- Anthracnose
- Anthracnose Strain 45 (LEAF)
- Anthracnose Strain 45 (STEM)
- Anthracnose Strain 45 (STEM)
- Anthracnose Strain A7 (LEAF)
- Anthracnose Strain A7 (STEM)
- Anthracnose Strain A7 (STEM)
- Auly Stem Blight
- Average root diameter (mm)(+45)
- Average root diameter (mm)(+42/45)
- Bacterial Brown Spot (BBS) Low nucleation temperature mean, °C
- Bacterial Brown Spot (BBS) Lesion no.
- Bacterial Brown Spot (BBS) Lesion no.
- Bacterial Brown Spot (BBS) Low nucleation temperature mean, °C
- Bacterial Brown Spot (BBS) Leaves Prosen at -2 °C
- Bacterial Brown Spot (BBS) Season no.
- Bacterial Brown Spot (Field Resistance)
- Basal root dry weight (g)plant-Greenhouse
- Basal root growth angle (SRGA)
- Basal root length (cm)plant-Greenhouse
- Basal root responsiveness to IAA

Search markers

Learn to use PhaseolusGenes

Search term:   
Search field: All fields

Blast search (soybean)

(Blast w/ e-value cutoff 0.0001)

Enter one sequence in simple text

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# PhaseolusGenes: Genome Browser

Home Genomes Tables DNA PDF/PS Session Help

UCSC Genome Browser on G. max Sep 2008 Assembly (glyMax1)

move <<< < < < > > > >> zoom in 1.5x 3x 10x base zoom out 1.5x 3x 10x

position/search Gm01:1-55,915,595   size 55,915,595 bp

move start    Click on a feature for details. Click or drag in the base position track to zoom in. Click side bars for track options. Drag side bars or labels up or down to reorder tracks. Drag tracks left or right to new position.

move end

default tracks



Use drop-down controls below and press refresh to alter tracks displayed.

Tracks with title of items will automatically be displayed in mouse click/pop-up menu.

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# Population-specific Genomics in Plant Breeding & Genotyping by Sequencing

Sequencing of P1, P2 BLAST onto bean and soybean genomes --> physical map  
Use RI population for genetic map and QTL analysis  
Identify polymorphisms --> potential markers

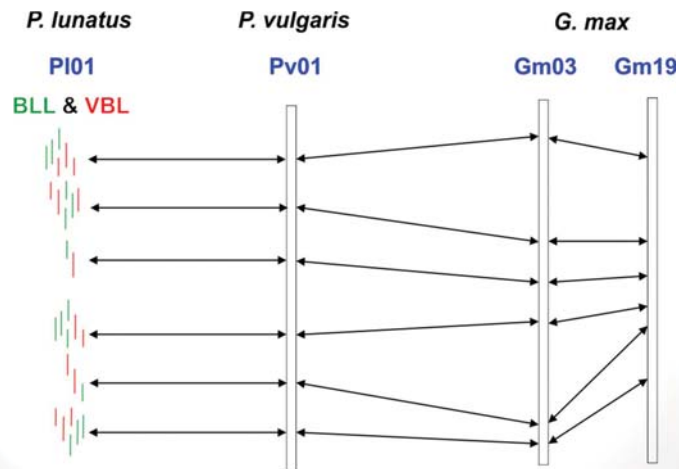
Large Lima (Andean)  Baby Lima (Mesoamerican) 

UC 92 Bush (P1) x UC Haskell Viny (P2) = F1

F1 leads to:  
 - Recombinant Inbred population (Genetic Analysis)  
 - Inbred Backcross population (Field Breeding)  
 - Other breeding populations

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## “Instant” Mapping in Lima Bean through Synteny With Common Bean and Soybean



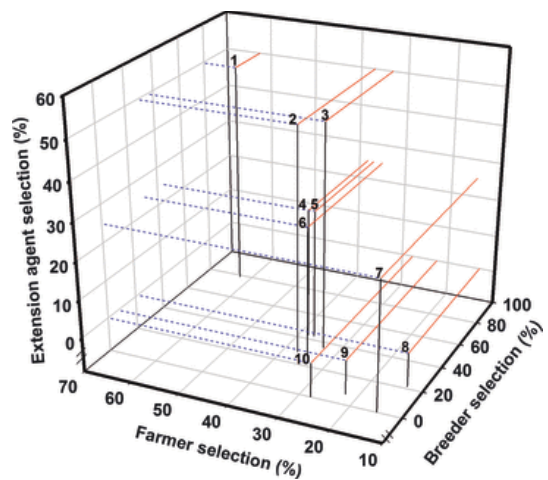
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## Key Ingredients: Breeding Methods & Genomics

### 2 Participatory Breeding

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## Participatory approach in common bean (*Phaseolus vulgaris* L.) breeding for drought tolerance for southern Ethiopia



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## Why “do” participatory breeding?

### Gender Differences

Trait	Top 6 ranking traits	
	Men	Women
Earliness	1	1
Fast-to-cook		2
Marketability	2	5
Drought tolerance	4	3
Grain yield	3	4
Germination	5	
Taste		6
Specific adaptation	6	

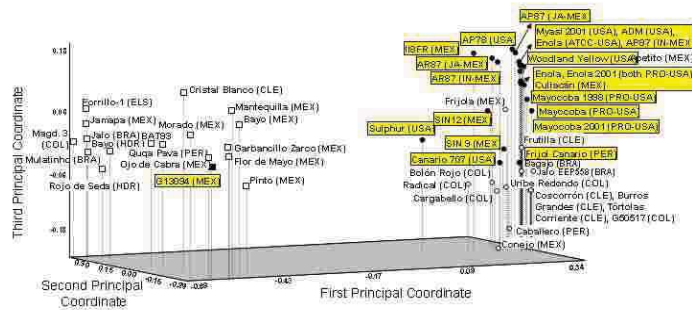
- Cultural and gender differences
- Environmental differences: broad vs. narrow adaptation
- G x E
- Novelties & flexibility

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## Results of Breeding for a “New” Bean Type: Azufrado Peruano: Azufrado (M) x Canario (A)

Effect of selection on organization of genetic diversity in breeding gene pools

- Distribution of recombinants in genome
- “Haplotype blocks” in relation to agronomically important genes

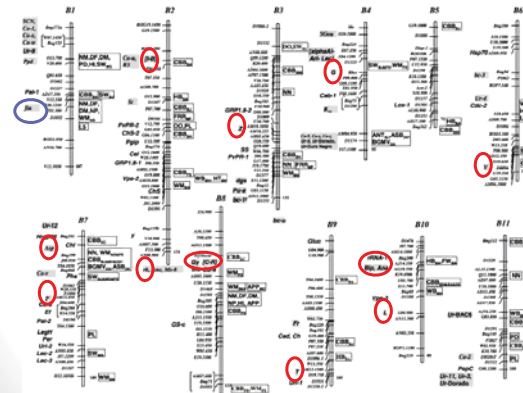


Principal Coordinate Analysis

Pallottini et al. 2004

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## Some Help from Genetic Features of Common Bean

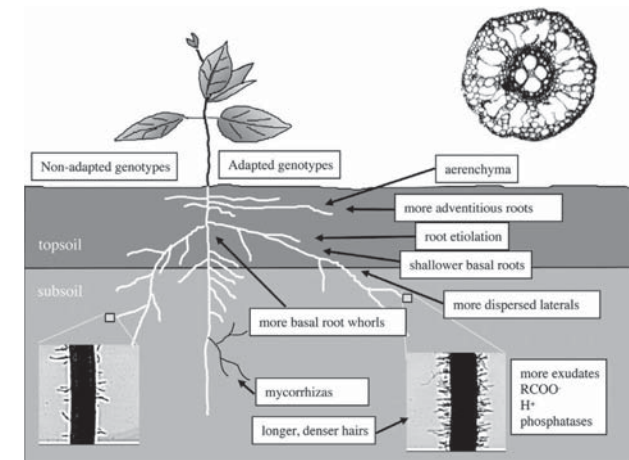


Freyre et al. 1998; Kelly et al. 2003

- Predominant selfing
- Seed color and growth habit gene distribution
- Markers
- Panel of pedigreed accessions
- Gene pool or race panels

## Key Ingredients: The Role of Phenotyping in Breeding for Abiotic Stresses

## Phenological traits enhancing P acquisition



Lynch 2011

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## Drought (Beebe et al. 2008)

- Breeding for drought tolerance in Mesoamerican market classes (red, black, carioca, mulatinho)
- Contrasting selection environments
- **Visual selection for well-filled pods (effective partitioning of photosynthates into seeds), in addition to yield per se**
- Under drought stress: substantial improvement (~ 20-100% > controls) in all color classes
- Under well-watered conditions: some classes and locations: up to ~15-20% > control
- Some lines **also had tolerance to low soil P** → can combine two important stresses

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## Biological nitrogen fixation

- Legume-Rhizobium specificity (or lack thereof) vs. N fixing ability
- Progress has been made towards understanding the initial steps of symbiosis at the molecular level:
  - initial recognition of Rhizobium by host ~ functionally conserved among legumes
- Limited progress towards understanding subsequent molecular steps:
  - Bacterial effector proteins and molecular targets in host cells; evolutionary diverged
- Supra-molecular processes: Rhizobial adaptation, especially to low fertility

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Den Herder & Parniske 2009

## Key Ingredients: Funding

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## Funding deficit for grain legume research

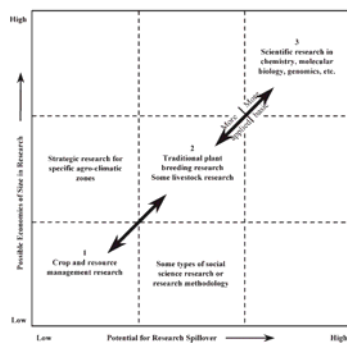
Comparison of Funding among Legume and Cereal CRP3s<sup>a</sup>

CRP 3 (Number of Species)	Years	Amount (M USD)	Total Amounts (M USD)
Legumes (8)	2011-13	138	138
Dryland Cereals (5)	3	79	1138
Maize (1)	3	238	
Rice (1 or 2)	5	593	
Wheat (1)	3	228	

<sup>a</sup> Information from various documents or web pages on [http://www.cgiarfund.org/cgiarfund/research\\_portfolio](http://www.cgiarfund.org/cgiarfund/research_portfolio)

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## Coordination among different players



- Transition between breeding and genetics/genomics
- Breeding on station and participatory breeding, including social sciences
- Trait-based breeding and physiology

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From Dalrymple 2008

See also: Feed the Future:

[http://www.feedthefuture.gov/sites/default/files/resource/files/FTF\\_research\\_strategy.pdf](http://www.feedthefuture.gov/sites/default/files/resource/files/FTF_research_strategy.pdf)

## Conclusions

- Improved management and utilization of germplasm:
  - Joint analysis of agronomic, molecular, and GIS information (ecological genomics and germplasm conservation and utilization)
  - Farmers play a role in maintenance of diversity and practice “stabilizing selection”
- Molecular markers
  - Understanding of inheritance of traits
  - Marker-assisted selection? Subordinated to phenotypic selection
    - Species, crop, markers (high-throughput), technology
- Participatory or distributed breeding
- Specific phenotypic traits informed by physiology with overall yield becomes critically limiting (in contrast with DNA sequence)
- Nitrogen fixation:
  - Combine selectivity and high BNF
- Funding:
  - Take into account nitrogen, rotational, and nutritional contributions of legumes

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