IRRI

Rice research highlights
GCP synergy

Rice projects sponsored wholely or partly by GCP

- SP1
  - Mini-core collections
  - SNP discovery (genome-wide SNP and Haplotypes)
  - Chromosomal Segmental Substitution Lines
  - Unlocking diversity in “wild genomes”
- SP2
  - Mutant applications
  - Drought response: reproductive (grain) and vegetative (tissue growth)
  - Stress transcriptome
  - QTL analysis--disease resistance, drought, salinity, P-deficiency
- SP3
  - MAS for drought
  - Improving new African rice (NERICA)

Questions

Are we closer to knowing genetic control of complex traits (e.g., drought tolerance) in rice?

Any results for applications and generalization?

“GCP factor”: mobilizing resources and collaboration to maximize synergy

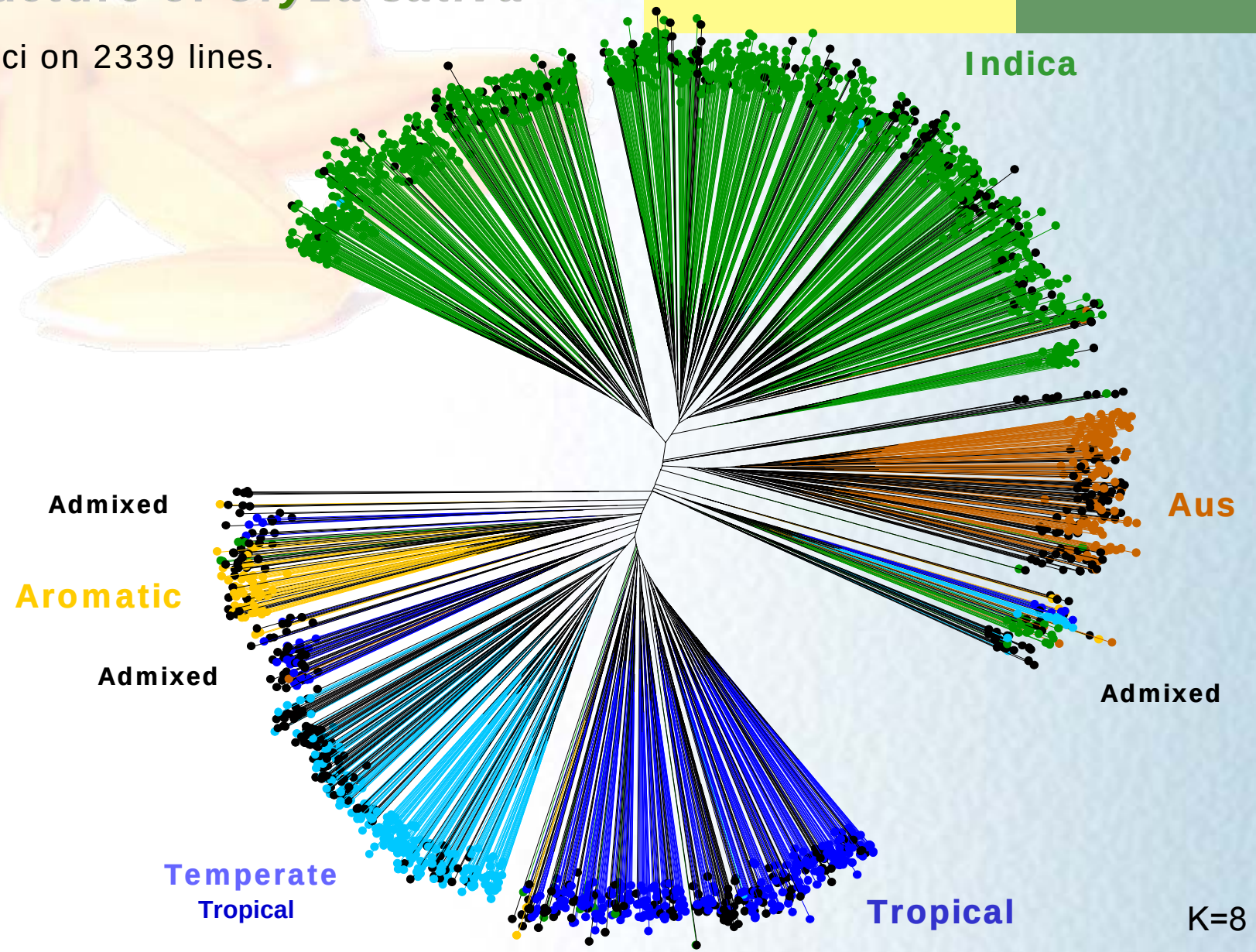
Problems and Challenges

Highlights

- **Genetic diversity platform**
- **QTLs large and small**
  - **Salinity, P-deficiency, submergence, drought**
- **Integration of expression signatures, NIL introgression, and QTL alignment**

Structure of *Oryza sativa*

50 Loci on 2339 lines.

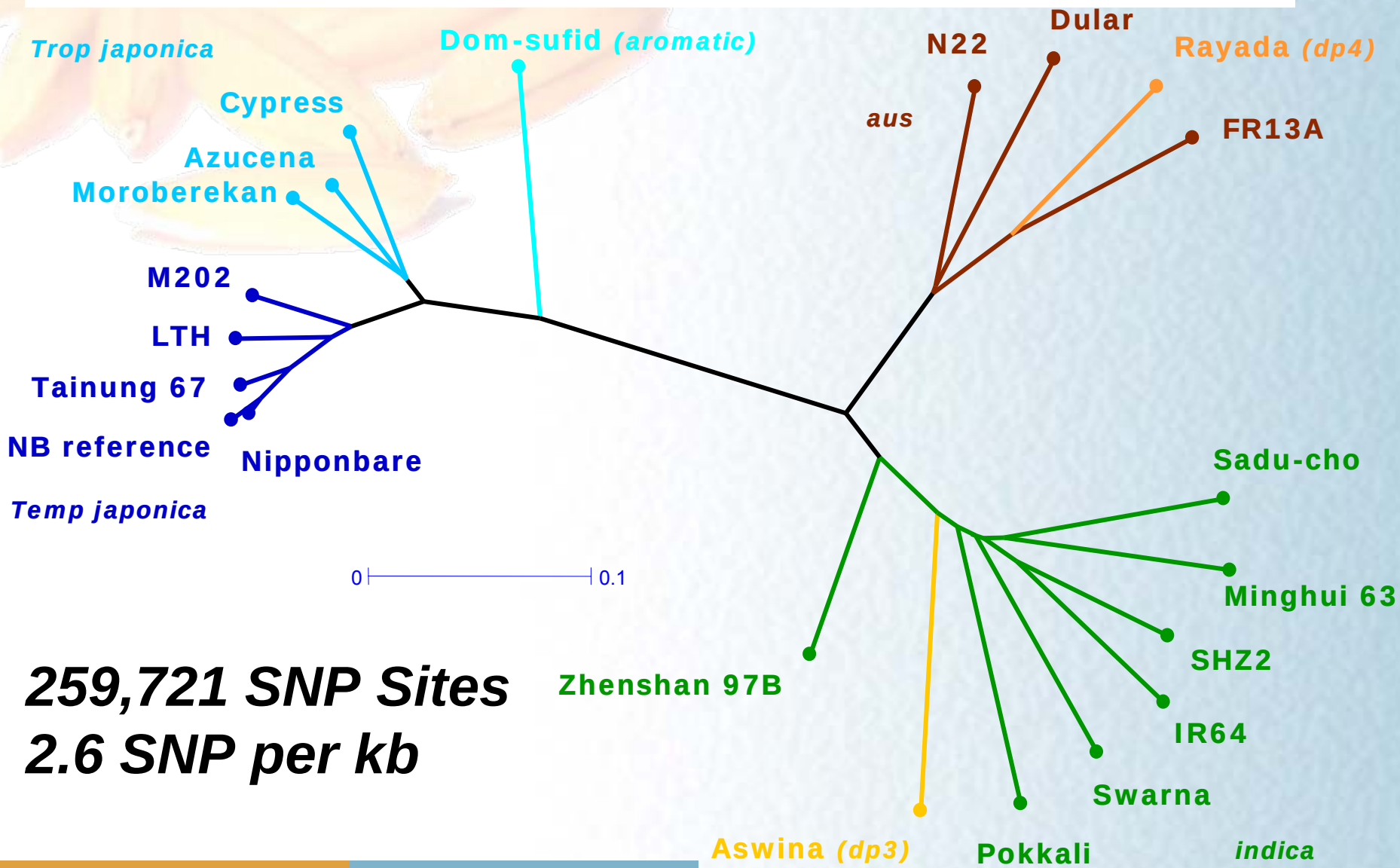


McNally et al IRRI

Genetic populations with special relevance to drought research

Population	Feature	Produced and accessible at	GCP
CSSL	2 *sativa* x *glabberima* libraries 4 wild inter-specific libraries	CIAT	+++
	4 japonica x indica libraries	NIAS, Japan	
RIL (tolerant x sensitive, including DH lines)	~ 6 commonly used populations	IRRI, IRD	
Breeding populations	Advanced lowland and upland breeding lines, NERICA series	IRRI, WARDA	+
Introgression lines	*glabberima* x *sativa*	CIAT, IRRI, WARDA	
Mutants	Insertons/activations Deletions	International Rice Functional Genomics Consortium	+

Oryza SNP Project: *Sequencing Multiple and Diverse Rice Varieties: Connecting Whole-Genome Variation with Phenotype*



259,721 SNP Sites

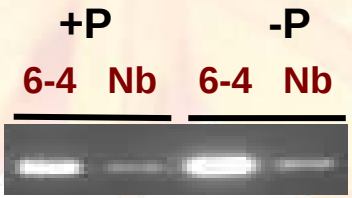
2.6 SNP per kb

McNally et al IRRI

Use of genome-wide SNP data

- Tag SNPs to characterize germplasm
  - High-resolution mapping and association analysis
- “universal” gene discovery chips
  - High-resolution genotyping (SNP, indels)
  - Transcriptome analysis

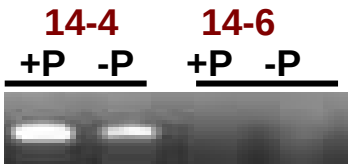
Analysis of candidate genes in NIL- Pup1 lines under +P & -P in the field



#4: fatty acid oxygenase

#18: dirigent-like

#26: hypothetical protein



#43: protein kinase

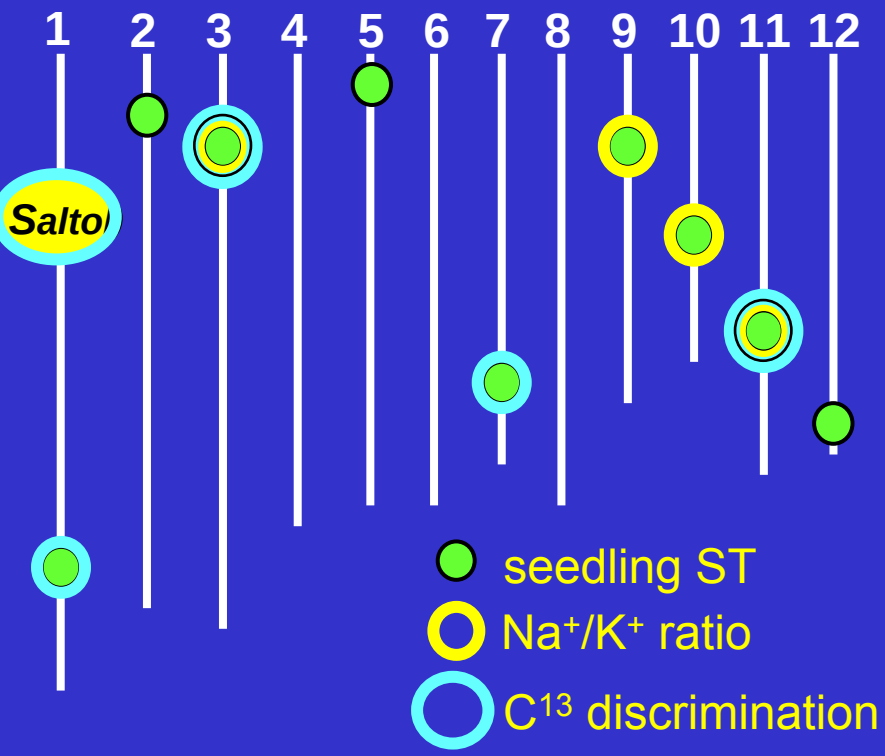
NILs 6-4/14-4: +Pup1; 14-6: -Pup1; Nb=Nipponbare.

Further analysis of these genes is ongoing (RNAi and overexpression)

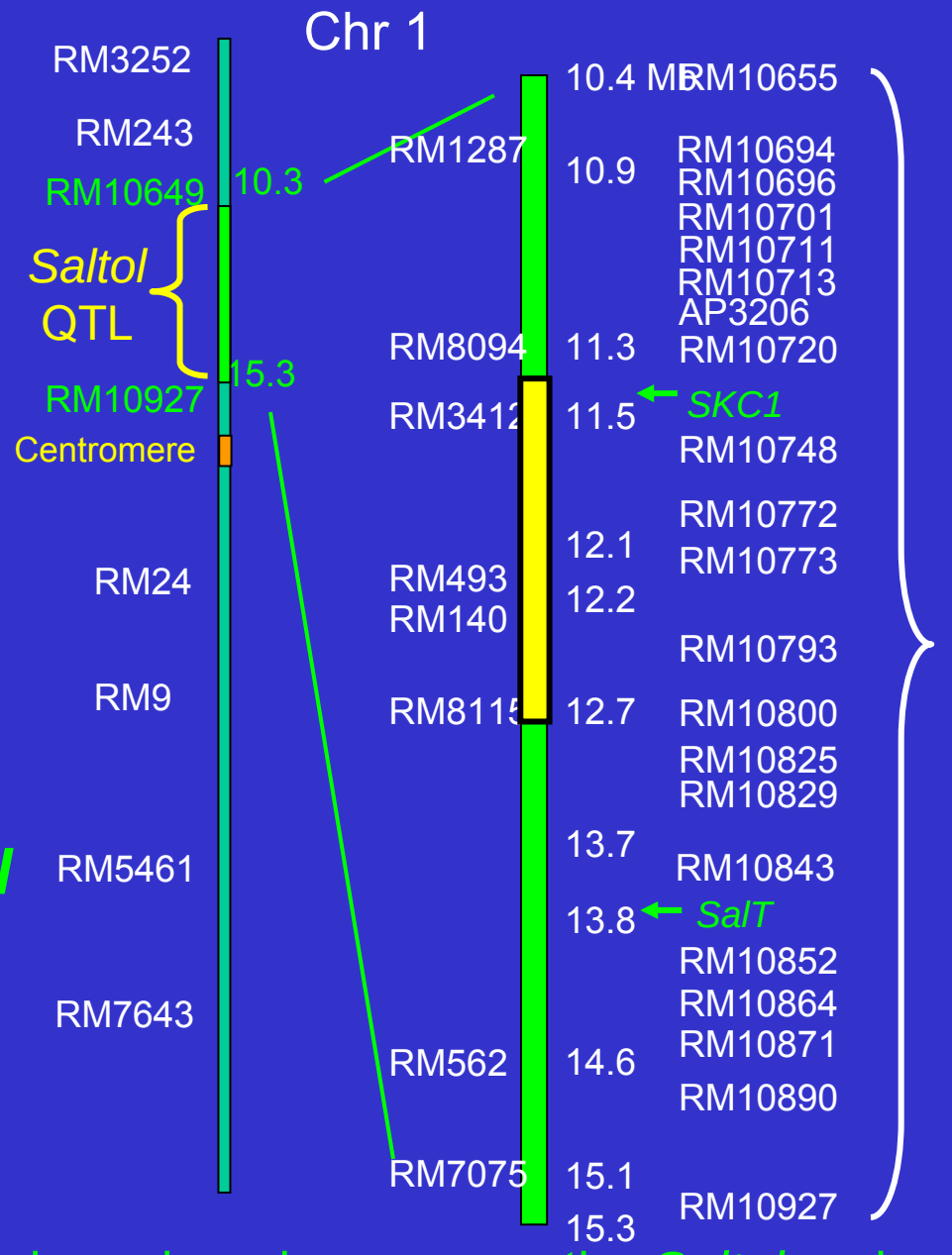
RNAi Trasgenic plants of The four candidate genes

Pop1 phenotype expressed better under drought & overlaps with a major QTL for yield under water stress

IR29/Pokkali ST QTLs



Saltol fine-map



Diagnostic markers for *Saltol*

- SSRs
- Gene/allele specific
- SFPs

30 SSRs and gene-based markers across the *Saltol* region

Rice mutant lines with altered response to salinity

5,000 IR64 mutants screened for seedling tolerance & sensitivity

- **5 mutants selected as more tolerant than IR64**
- **3 mutants selected as more sensitive than IR64**

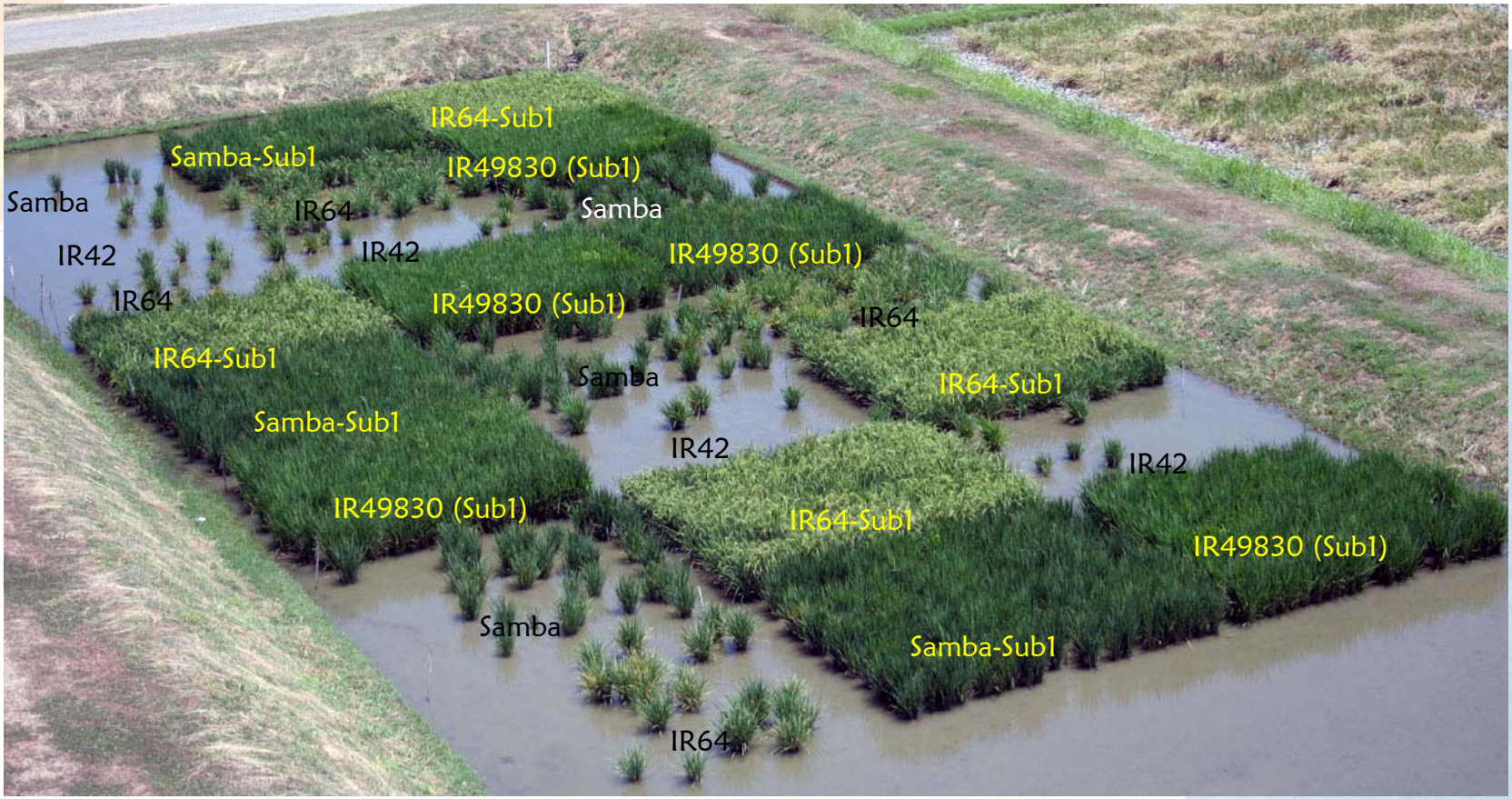
3 weeks @ EC = 18 dSm^{-1}

Babak Nakhoda, Abdel Ismail, IRRI

Introgression of *Saltol* and *Pup1* into popular varieties using MAB

- SSRs, gene-specific and SFP markers were developed for both *Pup1* and *Saltol*
- Sets of foreground and recombinant markers were selected for both QTLs
- MAB is being used to introduce *Pup1* into 3 Indonesian upland varieties and *Saltol* into two Bangladeshi varieties

Submergence of 17 days in the field at IRRI



LETTERS

Sub1A is an ethylene-response-factor-like gene that confers submergence tolerance to rice

Kenong Xu[1], Xia Xu[1], Takeshi Fukao[2], Patrick Canlas[1], Reycel Maghirang-Rodriguez[3], Sigrid Heuer[3], Abdelbagi M. Ismail[3], Julia Bailey-Serres[2], Pamela C. Ronald[1] & David J. Mackill[3]

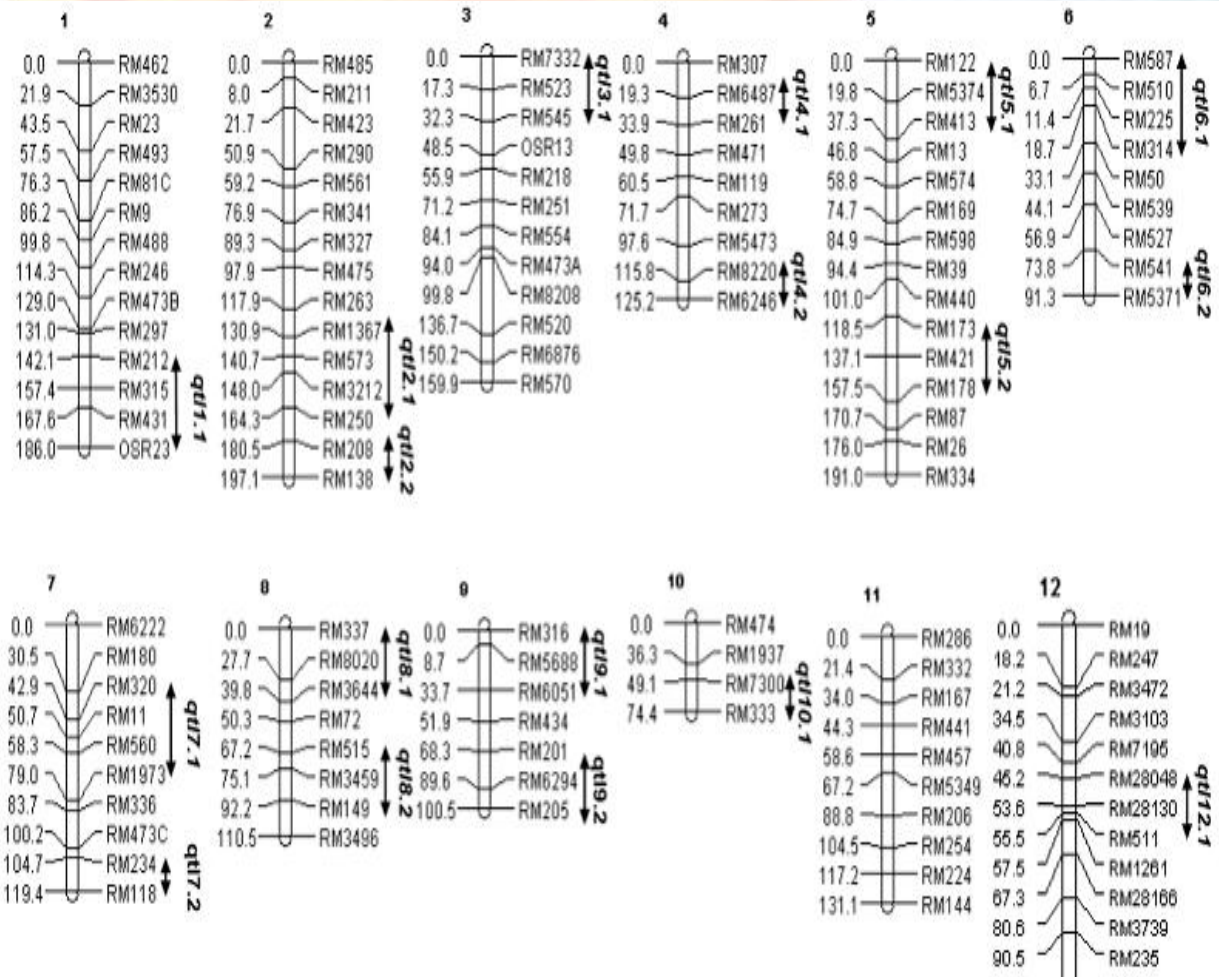
On DROUGHT

Words of wisdom from a drought breeder:

1. Smart conventional breeding will give fast results
   - Identify and disseminate donors
   - Promote direct selection
   - Identify traits and tools to improve screening
2. Link molecular genetic research tightly to crop improvement
   - Forward genetics first
   - Focus on big effects, and use big populations
   - Expression studies using near-isogenic lines to identify genes and deliver MAS-ready systems
3. Be patient, and stay the course

From G. Atlin's seminar (Oct. 2006)

Mapping major QTLs for yield under drought



QTL intervals identified under drought stress and NS conditions in Vandana\Way Rarem

Bernier et al. Crop Sci, 2007

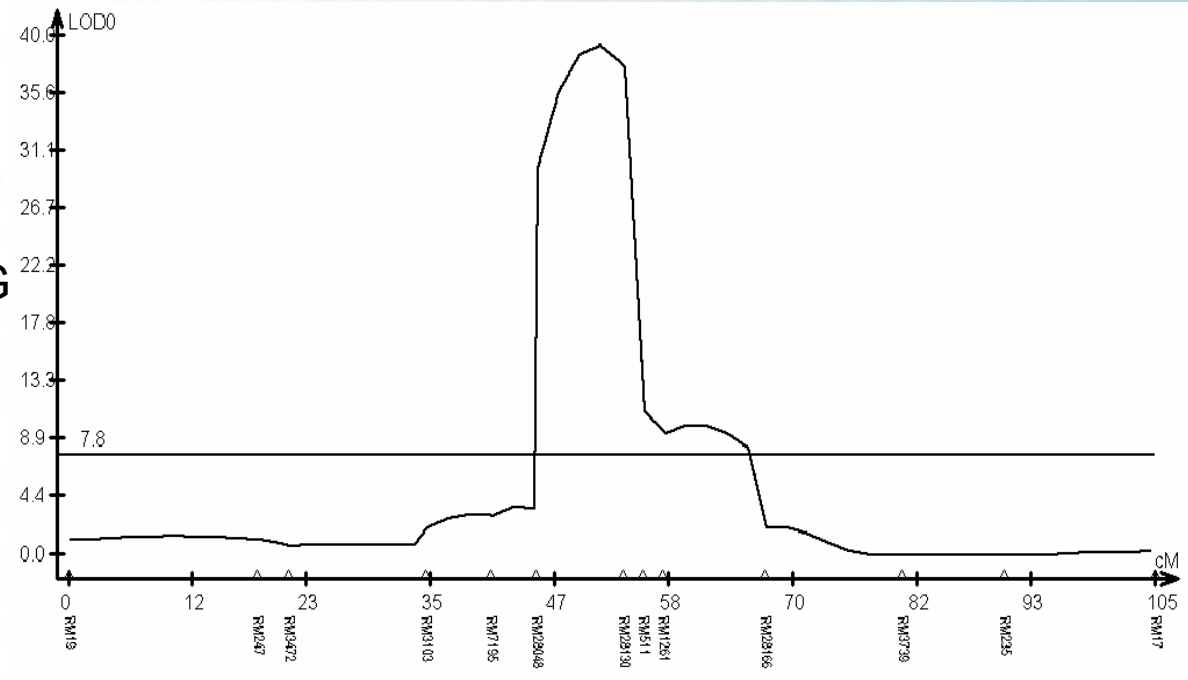
G. Atlin et al.

- **Vandana/Way Rarem: chr.12 (RM511) - 50% of variance for yield under stress**
- **Apo/IR64^2: chr. 9 (RM201) - 20%**
- **Apo/Swarna^2: chr. 6 (RM510) - up to 50%**

Major QTL for yield under drought

- **Vandana/WayRarem**

- Chr 12 (49cM)
- Large effect on grain yield under drought (R^2=0.33)
- Effect similar in 2 years (low G x E)
- Up to 51% of σ^2_G



- No effect under control conditions

J. Bernier et al., Crop Sci., 2007

Near-isogenic rice lines derived from IR64^{4}/Aday Sel

Lowland stress after re-irrigation
IR77298-5-6 NILs

- Sister-lines with contrasting performance to drought stress but no obvious polymorphism
- Genome scan (55K Affymetrix Chip) suggest 6 genomic regions different between two sister lines

Lowland stress 5-6 & 12-7 NILs

Region	Chr	Start	End	Mb
1	4	16,166,620	16,162,540	0.00408
2	5	27,035,050	29,363,633	2.328583
3	6	22,033,705	23,069,148	1.035443
4	9	15,522,889	16,077,435	0.554546
5	10	15,838,595	22,246,842	6.408247
6	12	23,636,569	23,967,787	0.331218

Venuprasad Ramaiak et al. IRRI

Improvement of water use efficiency in rice by expression of *HARDY*, an *Arabidopsis* drought and salt tolerance gene

Aarati Karaba*†, Shital Dixit*, Raffaella Greco*‡, Asaph Aharoni*§, Kurniawan R. Trijatmiko*¶, Nayelli Marsch-Martinez*‖, Arjun Krishnan**, Karaba N. Nataraja†, Makarla Udayakumar†, and Andy Pereira*,**††

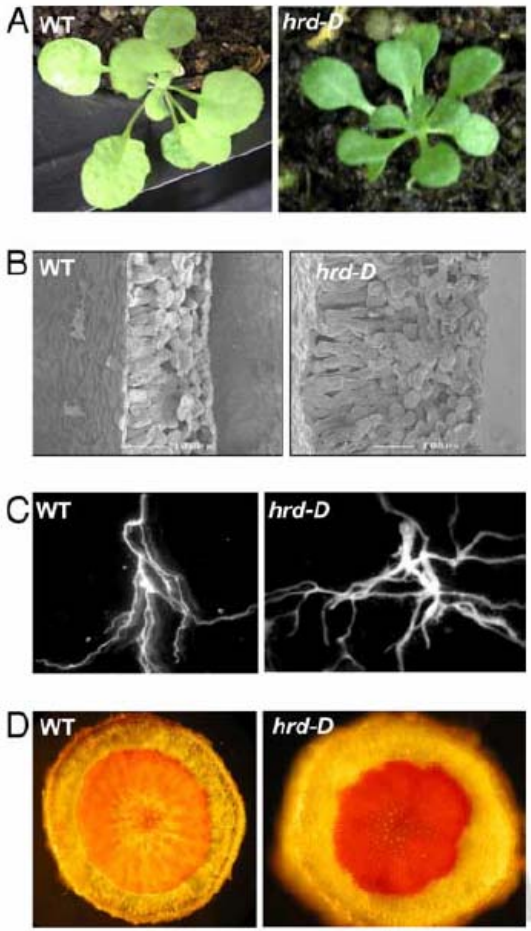


Fig. 1. The *hrd-D* mutant phenotype in *Arabidopsis*. (*A*) Rosette leaf phenotype of WT and *hrd-D* mutant with smaller slightly curled thicker deep

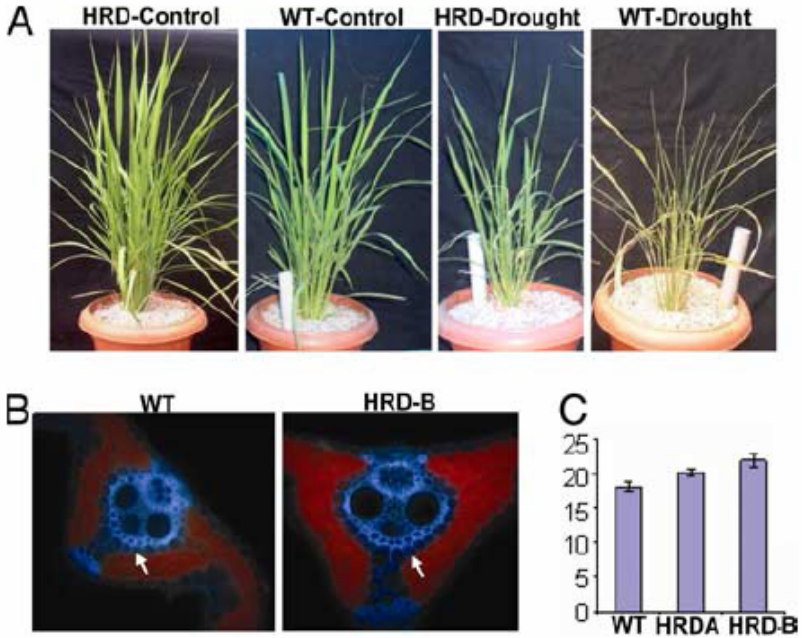
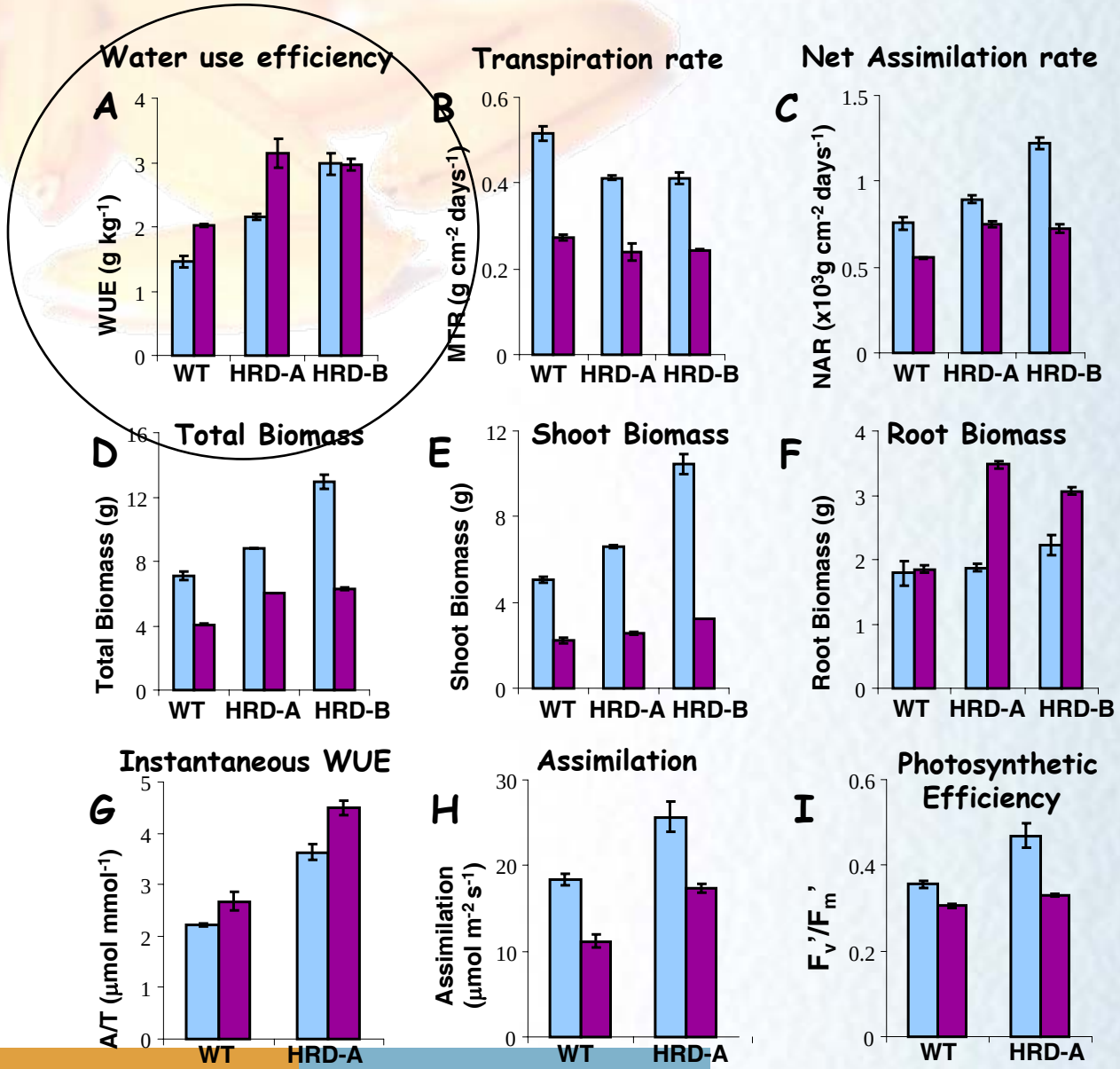


Fig. 4. Phenotype of *HRD* overexpression in rice. (*A*) Rice *HRD* overexpression line compared with WT Nipponbare under well watered (control) and water-stress (70% field capacity) conditions. (*B*) Leaf cross-section of WT and *HRD* overexpression lines, observed under fluorescence microscope, revealing red chlorophyll fluorescence and blue vascular bundles surrounded by the bundle sheath cells marked with an arrow. (*C*) Number of bundle sheath cells in WT compared with *HRD* overexpressors, which show significant increase ($n > 5$, $p = 7.5^{-10}$).

Karaba et al PNAS, in press

HRD overexpression improves WUE in rice

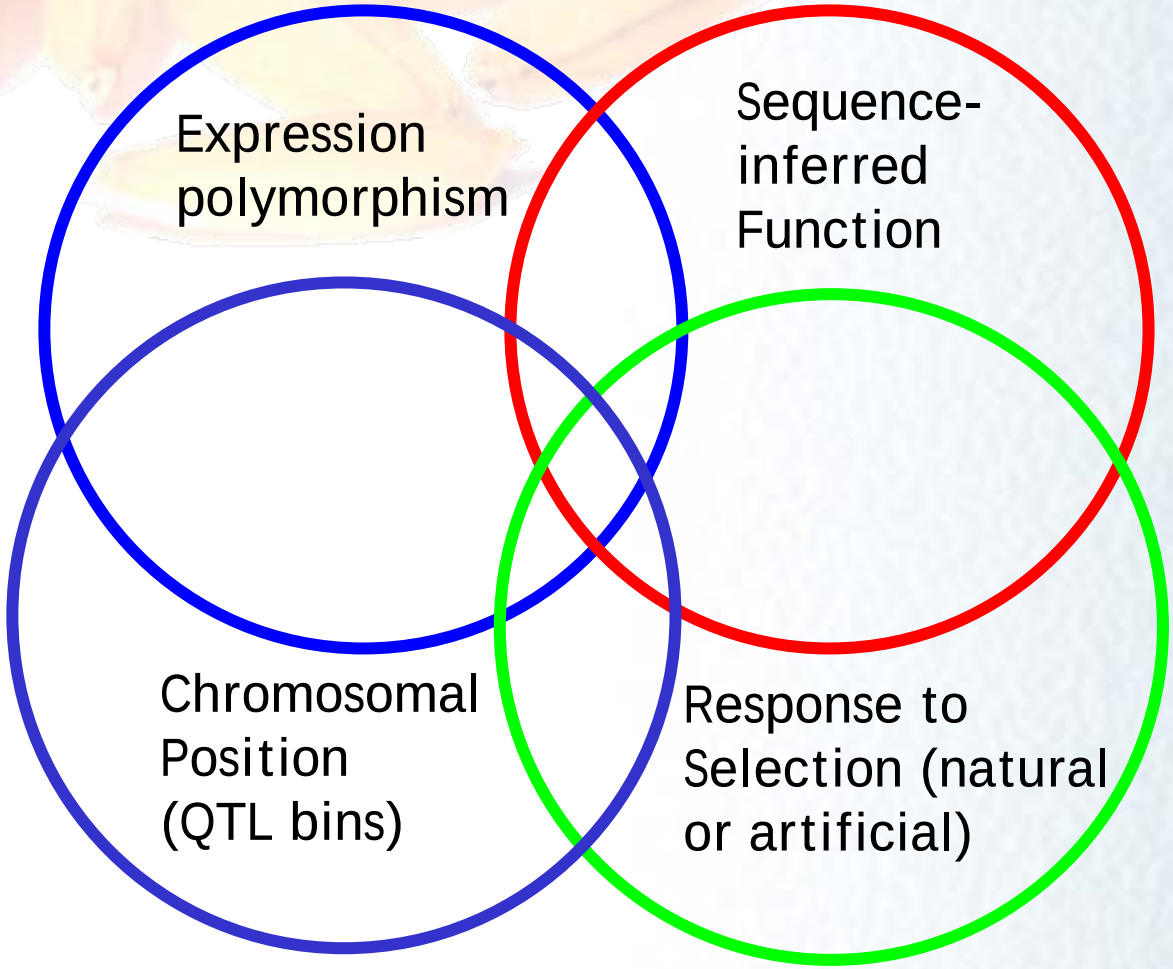


Field capacity
Well-watered

70%
field capacity
Drought

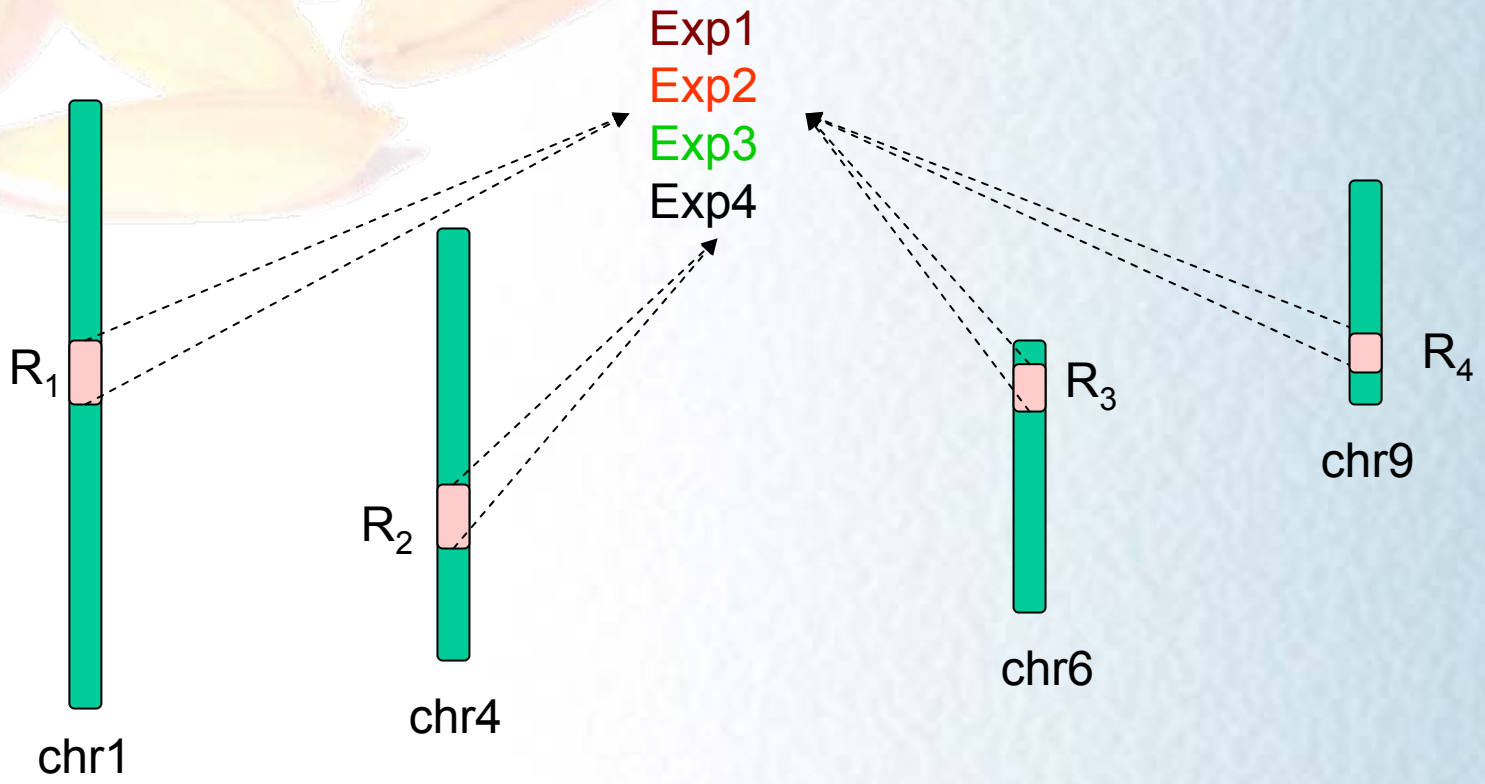
Karaba et al PNAS, in press

Integrate expression and mapping data to narrow candidate regions



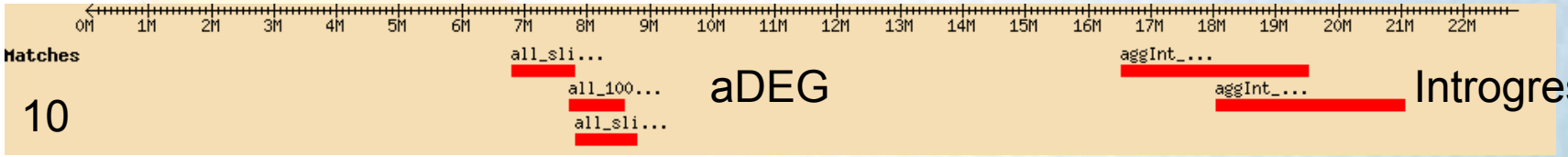
- ✓ **Expression signatures under different stresses**
- ✓**Introgression of NIL detected by whole-genome chips**
- ✓**QTL alignment**

Regions of correlated expression (RCEs) across different microarray experiments



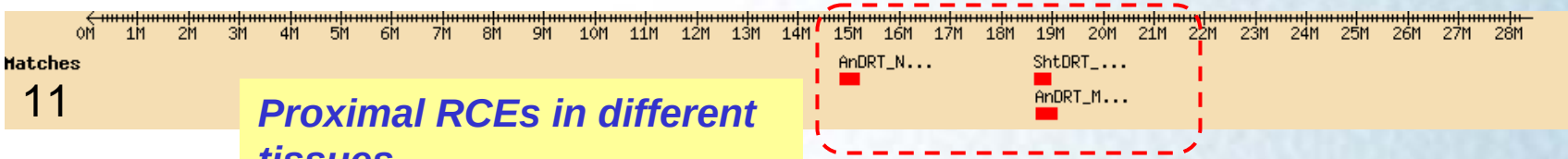
Systematic computation of gene expression correlation R (averaged across experiments) for a given genome block size has shown that some regions (of varying size) have significantly correlated expression (Spellman & Rubin 1998, Mauleon et al. in prep).

Integrated view of analysis results in GBrowse

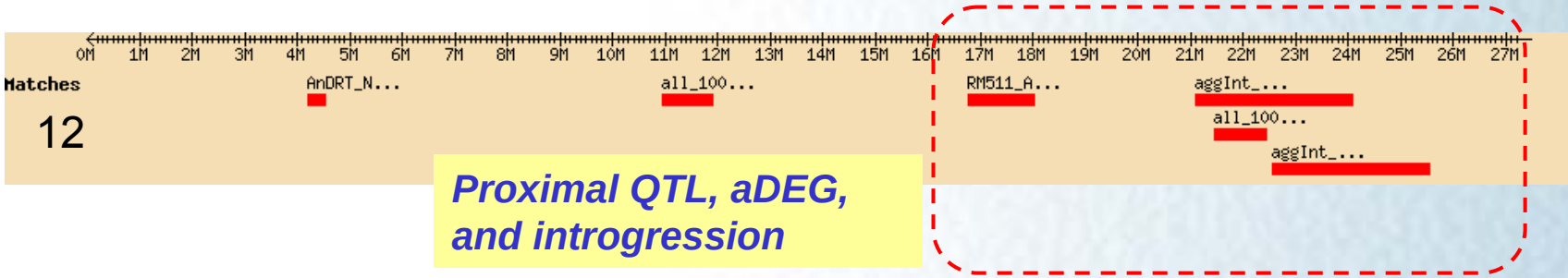


aDEG

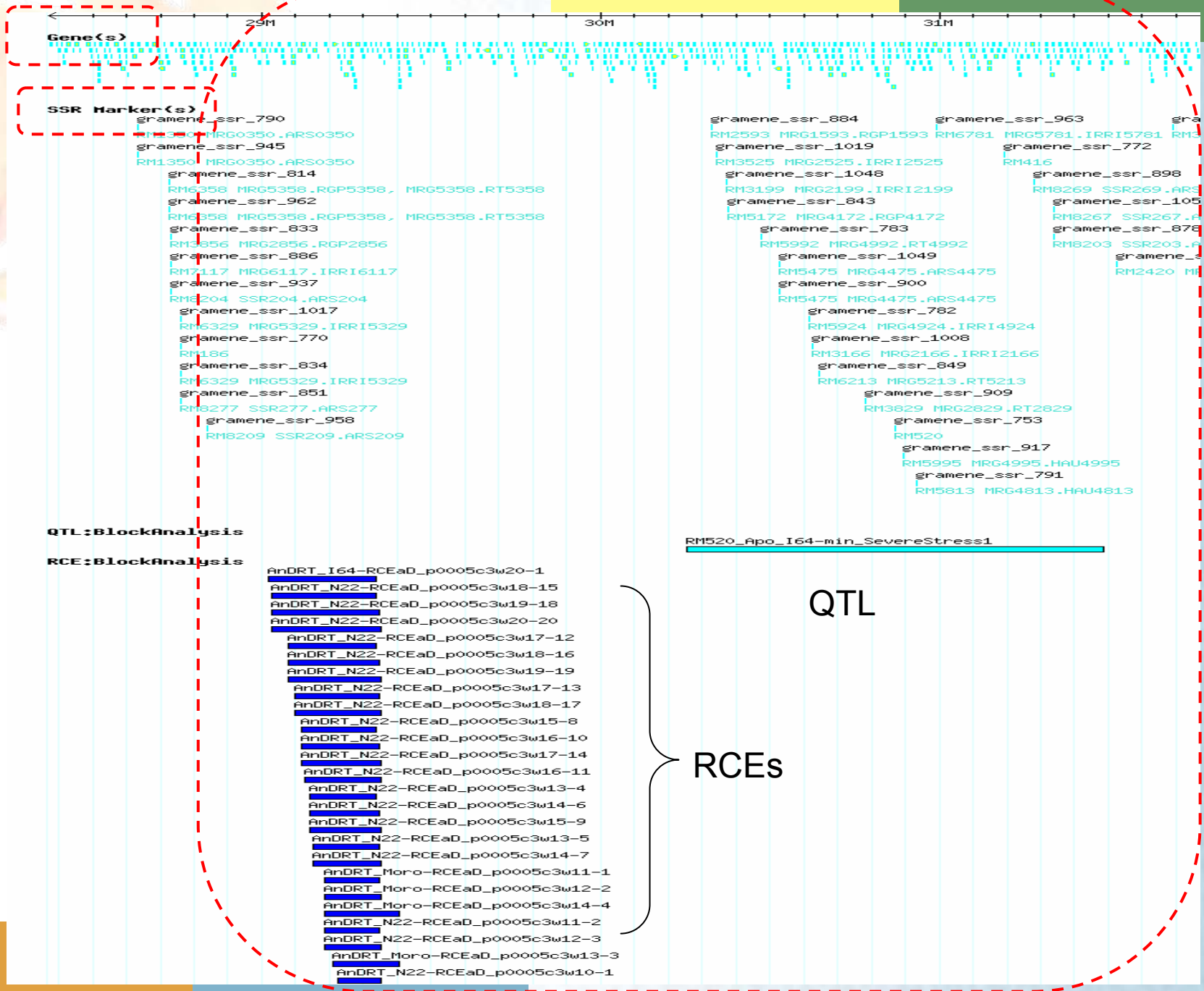
Introgression



Proximal RCEs in different tissues



Proximal QTL, aDEG, and introgression


29M
30M
31M
Gene(s)
SSR Marker(s)
gramene_ssr_790
RM1350 MRG0350.ARS0350
gramene_ssr_945
RM1350 MRG0350.ARS0350
gramene_ssr_814
RM6358 MRG5358.RGP5358, MRG5358.RT5358
gramene_ssr_962
RM6358 MRG5358.RGP5358, MRG5358.RT5358
gramene_ssr_833
RM2856 MRG2856.RGP2856
gramene_ssr_886
RM7117 MRG6117.IRRI6117
gramene_ssr_937
RM8204 SSR204.ARS204
gramene_ssr_1017
RM6329 MRG5329.IRRI5329
gramene_ssr_770
RM186
gramene_ssr_834
RM6329 MRG5329.IRRI5329
gramene_ssr_851
RM8277 SSR277.ARS277
gramene_ssr_958
RM8209 SSR209.ARS209
gramene_ssr_884
RM2593 MRG1593.RGP1593
gramene_ssr_1019
RM3525 MRG2525.IRRI2525
gramene_ssr_1048
RM3199 MRG2199.IRRI2199
gramene_ssr_843
RM5172 MRG4172.RGP4172
gramene_ssr_783
RM5992 MRG4992.RT4992
gramene_ssr_1049
RM5475 MRG4475.ARS4475
gramene_ssr_900
RM5475 MRG4475.ARS4475
gramene_ssr_782
RM5924 MRG4924.IRRI4924
gramene_ssr_1008
RM3166 MRG2166.IRRI2166
gramene_ssr_849
RM6213 MRG5213.RT5213
gramene_ssr_909
RM3829 MRG2829.RT2829
gramene_ssr_753
RM520
gramene_ssr_917
RM5995 MRG4995.HAU4995
gramene_ssr_791
RM5813 MRG4813.HAU4813
gramene_ssr_963
RM6781 MRG5781.IRRI5781
gramene_ssr_772
RM416
gramene_ssr_898
RM8269 SSR269.ARS
gramene_ssr_105
RM8267 SSR267.A
gramene_ssr_878
RM8203 SSR203.A
RM2420 M
QTL:BlockAnalysis
RCE:BlockAnalysis
RM520_Apo_I64-min_SevereStress1
QTL
AnDRT_I64-RCEaD_p0005c3w20-1
AnDRT_N22-RCEaD_p0005c3w18-15
AnDRT_N22-RCEaD_p0005c3w19-18
AnDRT_N22-RCEaD_p0005c3w20-20
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AnDRT_N22-RCEaD_p0005c3w13-5
AnDRT_N22-RCEaD_p0005c3w14-7
AnDRT_Moro-RCEaD_p0005c3w11-1
AnDRT_Moro-RCEaD_p0005c3w12-2
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RCEs

Questions

Are we closer to knowing genetic control of complex traits (e.g., drought tolerance) in rice?

Any results for applications and generalization?

“GCP factor”: mobilizing resources and collaboration to maximize synergy

Problems and Challenges

Gene discovery and applications in rice

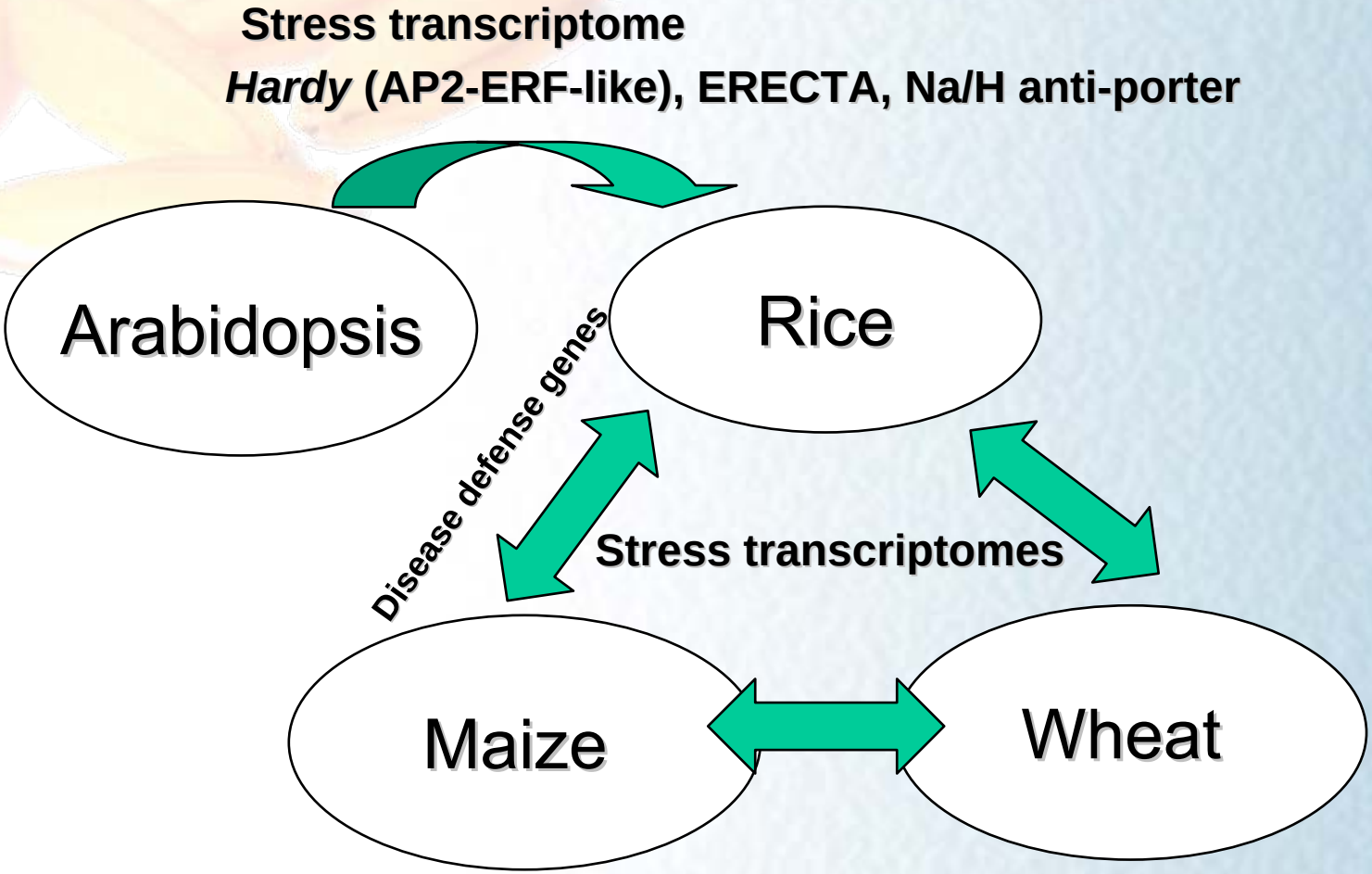
Potential/Power:

- **Interplay of forward and reverse genetics approaches**
- **Using smart genetic populations**
- **Whole-genome genotyping and expression**
- **Comparative biology**

Challenges:

- **The right alleles or allelic combinations that add tolerance to existing breeding lines/varieties**
- **Uptake in breeding programs—rapid trait conversion (MAS, MAB)**
-

Comparative biology between species



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- **Different PIs from Competitive and Commissioned Grants**
- **Projects and colleagues not supported by GCP**

Slides/ideas provided by

- **Abdel Ismail, Arvind Kumar, Ramil Mauleon, Rachid Serraj, Ken McNally**
- **Andy Pereira, Shoshi Kikuchi, Mathias Lorieux, Rebecca Nelson**

Oryza SNP Project

Sequencing Multiple and Diverse Rice Varieties: Connecting Whole-Genome Variation with Phenotype

Goal: To generate a multi-varietal, genome-wide SNP (single nucleotide polymorphism) database for rice to allow association between whole-genome variation and phenotype.

IRRI

Colorado State University
Knowledge to Go Places

PERLEGEN SCIENCES

TIGR

Generation
Cultivating Plant Diversity for the Resource-Poor

CSREES

International Rice Functional Genomics Consortium

http://www.oryzasnp.org/