IRRI Genotyping Service Laboratory Galaxy: bioinformatics for rice scientists

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Presented in behalf of my co-authors from IRRI

Lead Scientists

- Kenneth L. McNally Genebank resequencing
- Nickolai Alexandrov rice informatics consortium
- Michael Thomson Genotyping Service Laboratory
- Hei Leung Program Leader

Laboratory, software team

- Venice Margaret Juanillas
- Christine Jade Dilla-Ermita



Outline

- Introduction to IRRI & it's research agenda
- Bioinformatics support to molecular rice breeding at IRRI: IRRI GSL Galaxy
- Bioinformatics support to efforts for harnessing Rice Genetic Diversity
- Future activity: International Rice Informatics Consortium



INTERNATIONAL RICE RESEARCH INSTITUTE

Los Baños, Philippines

Mission:

Reduce poverty and hunger,

Improve the health of rice farmers and consumers,

Ensure environmental sustainability

All done through research, partnerships



Home of the Rice Green Revolution
Established 1960
www.irri.org

Aims to help rice farmers improve the yield and quality of their rice by developing..

- New rice varieties
- •Rice crop management techniques



A single strategic work plan for global rice research...

Global Rice Science Partnership: GRiSP

- Core: 3 international research centers
- Numerous research partners
- NEED TO SHARE RESEARCH SOLUTIONS







Many more...







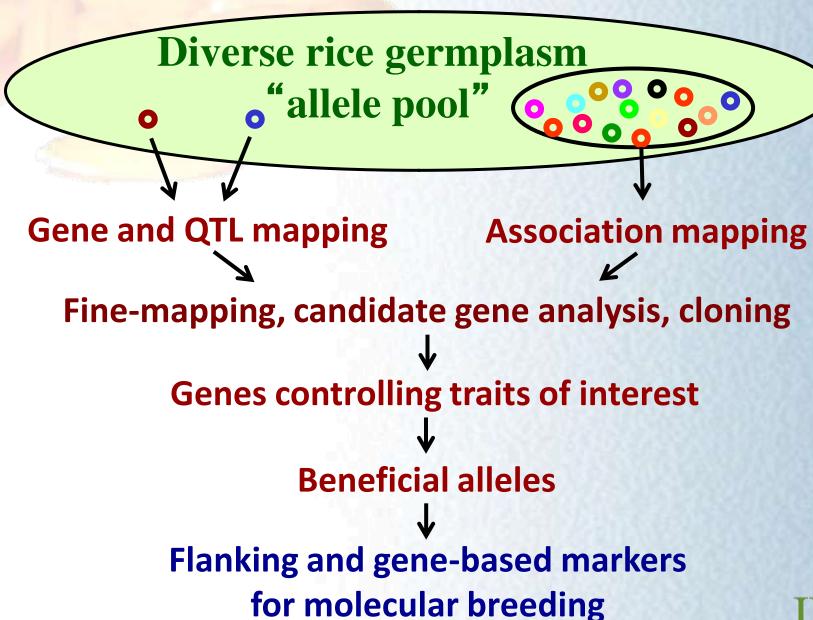
First GRiSP Research Theme with heavy bioinformatics ...

Accelerating the development, delivery, and adoption of improved rice varieties

 2.1. Breeding informatics, highthroughput marker applications, and multi-environment testing

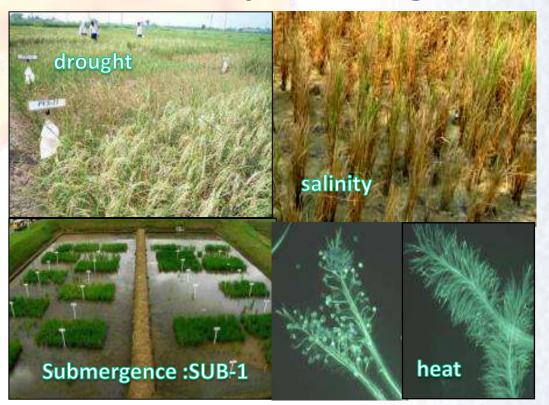


Allele mining for crop improvement



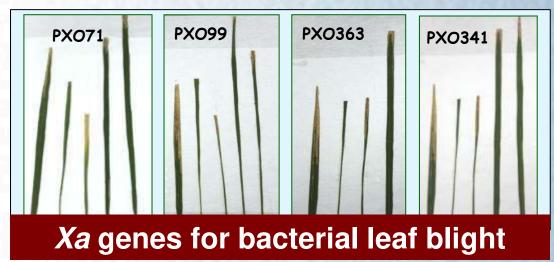


Major QTLs/genes for breeding



- QTLs and major genes for stress tolerance and disease resistance are known
- Flanking SSRs and genebased STS markers have been used to transfer these major QTLs
- Move to SNP markers for Marker Assisted Backcrossing (MABC). Marker Assisted Selection (MAS), Genomic Selection (GS)





Challenges for IRRI scientists/breeders

- Not familiar with SNP-based genotyping
 - o How do I score the alleles? (no gel image!!!)
 - o Data does not fit my spreadsheet (run out of columns, rows)...
 - o Cannot even view the data file using "ordinary" apps
 - Computer runs out of memory when I load the dataset...
 - Trusted analysis software crashes inexplicably...
- We need to
 - o enable field/bench researchers for bioinformatics
 - Share solutions openly across GRiSP partners, with rice research community as a whole



Galaxy has features that fit our needs

Open, web-based platform for <u>accessible</u>, <u>reproducible</u>, and <u>transparent</u> computational biomedical research.

- Accessible: Users w/o programming experience can easily specify parameters and run tools and workflows.
- Reproducible: Galaxy captures info so that any user can repeat and understand a complete computational analysis.
- Transparent: Users <u>share and publish analyses</u> via the web and create interactive, web-based documents that describe a complete analysis.



Integration of Galaxy to Genotyping Service Lab workflow

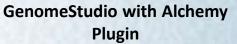
Illumina BeadXpress Genotyping

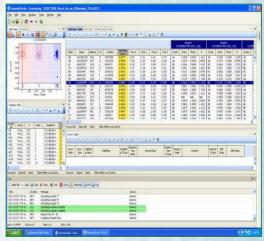




Infinium Custom 6k chip

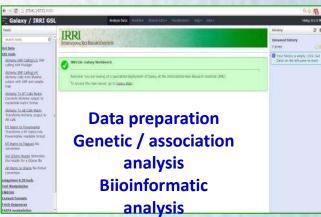








Software tools on IRRI GSL-Galaxy



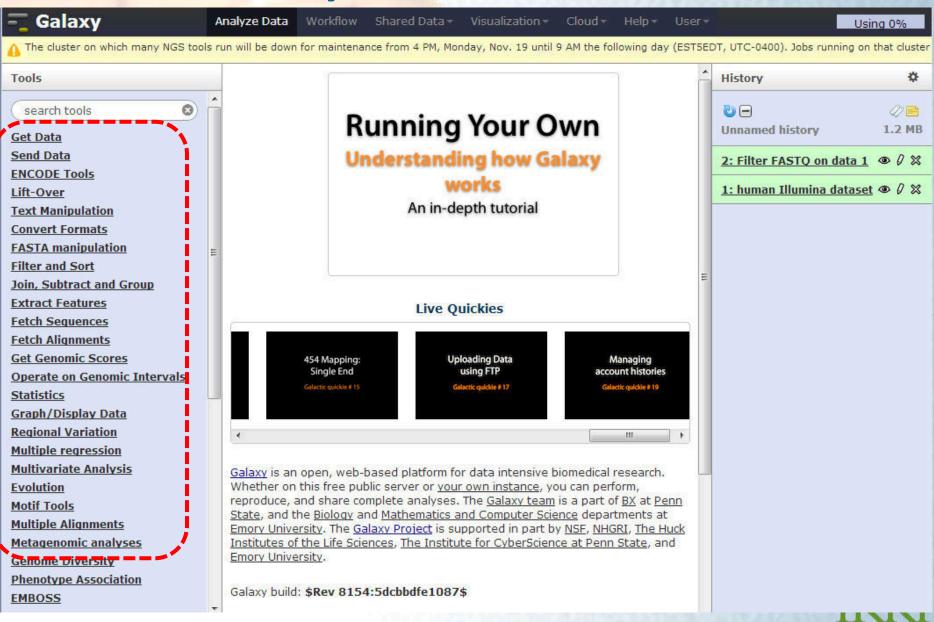




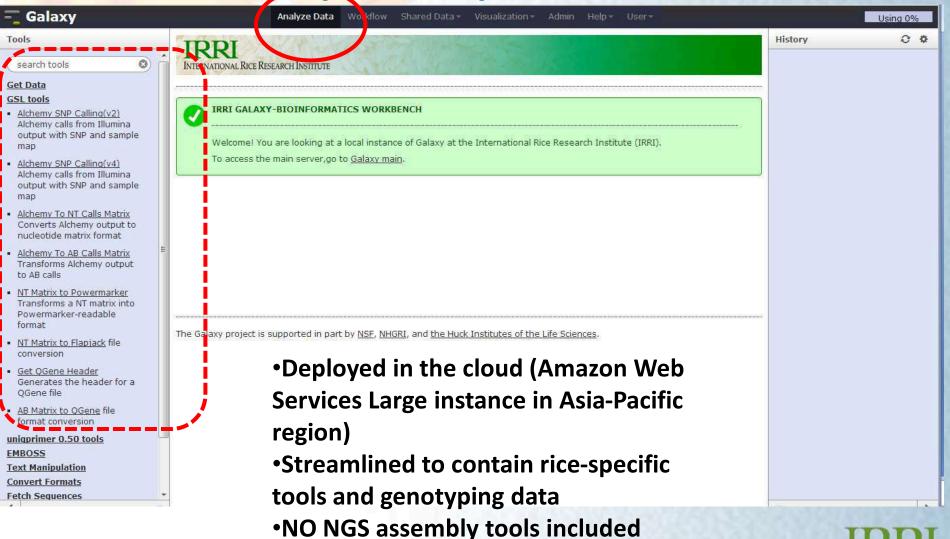
Fluidigm EP1Genotyping



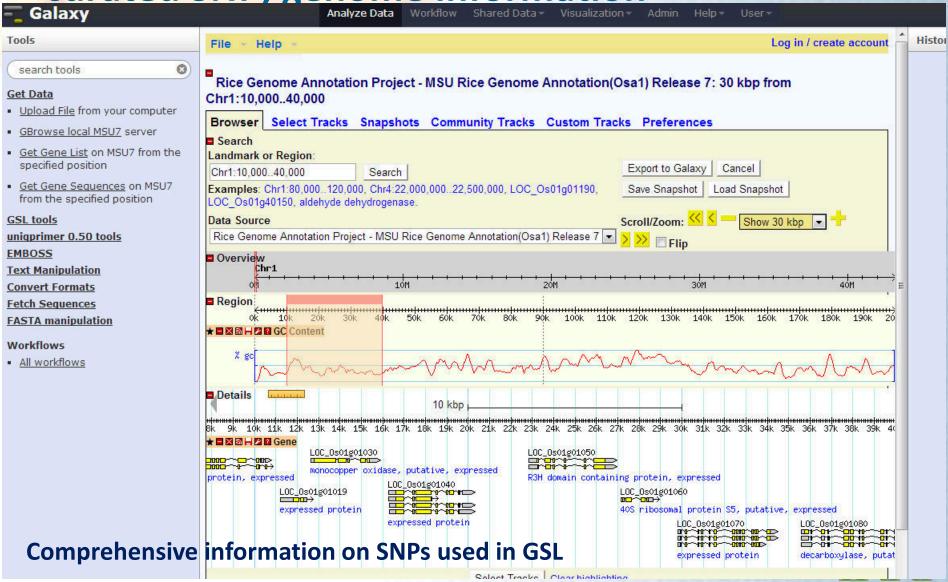
Standard Galaxy release



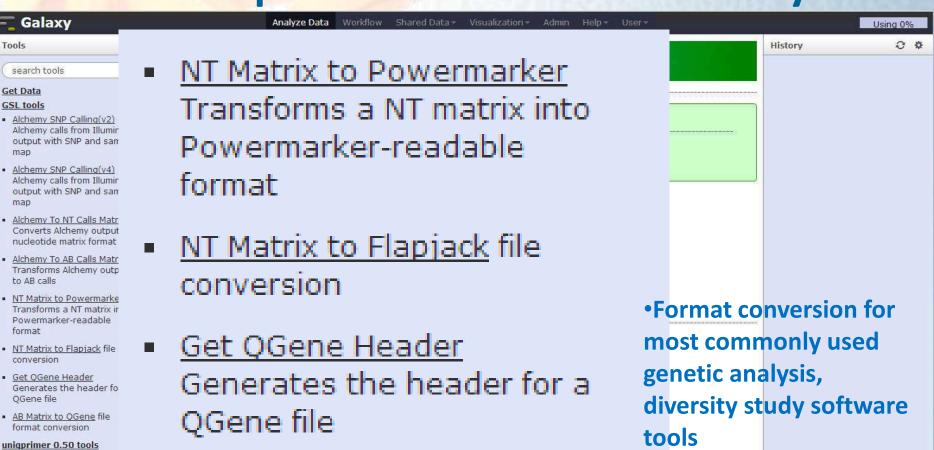
IRRI GALAXY (current)



Rice genome browser installed as data source for curated SNP, genome information



Data manipulation tools in GSL Galaxy



AB Matrix to QGene file

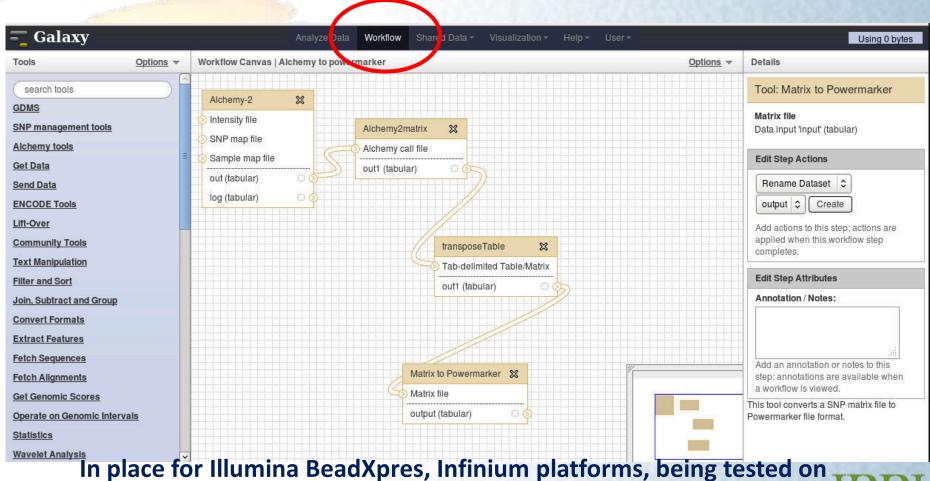
format conversion

EMBOSS
Text Manipulation

Convert Formats
Fetch Sequences

IRRI

Workflows for rice data analysis already available



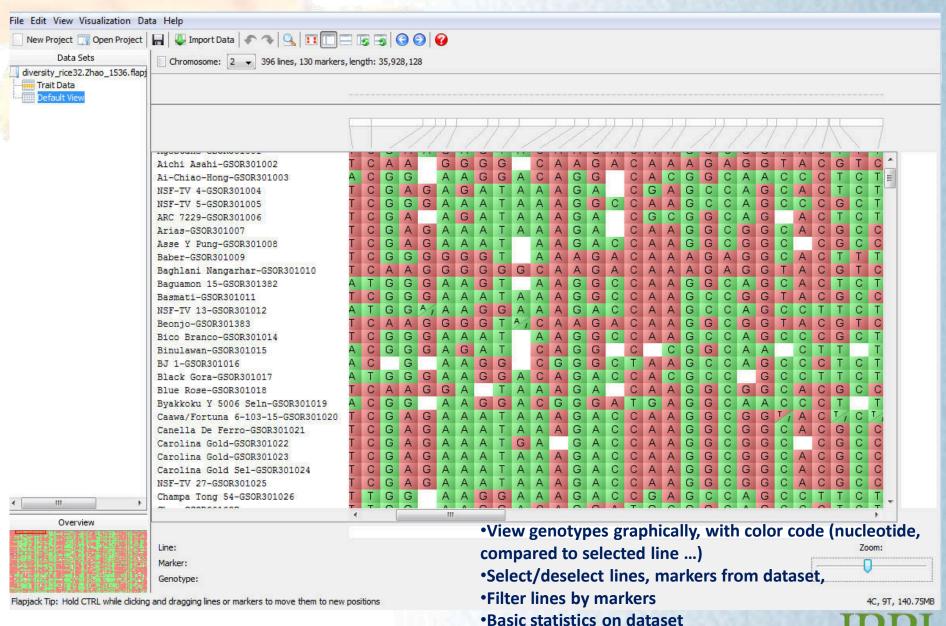
Fluidigm system...

Software Tools for SNP analysis

- SNP calling: Alchemy (Wright et al 2010)
- SNP data exploration, visualization: Flapjack, TASSEL
- Genetic linkage mapping: Mapmanager QTX, R/QTL
- QTL analysis: R/QTL, Qgene, MPMap (for multi-parent crosses)
- GWA analysis: TASSEL
- Population structure / diversity analysis : Powermarker,
 Structure



Flapjack: visualize, manipulate SNP data



http://bioinf.scri.ac.uk/flapjack/index.shtml



A powerful software for marker data analysis

Home	Downloads	Analysis	Screenshots	FAQ	Support
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What's new?

- Version 3.25 released on 2/5/2006. This is a permanent version!! Please cite the Bioinformatics paper.
 Choose 'How To Cite' from Help menu to see more details.
- Version 3.09 released on 7/14/2004. Support 120 dpi settings.
- Version 3.08 released on 6/14/2004. Fixed a bug in core set selection.
- Version 3.07 released on 3/20/2004. (1) New SNP identification tool (2) Fix several minor bugs.
- Version 3.03 is available for download. (1) Fix a bug in batch export. (2) New functionality for single haplotype phase assignment. (3) Allow for all-missing individuals for most analyses.
- PowerMarker V3.0 was officially released on January 30th, 2004. Full documentation is included.
- The following functions are disabled in PowerMarker: (1) Marker selection for haplotype data, genotype data and trio data (2)
 Population structure inference based on EM algorithm (3) Logistic regression and least angle regression for association study.
 The author is stilling working on the publications. The algorithms have been implemented and tested with simulated data.

 Please contact the author if you are interested in these algorithms.

What's PowerMarker?

PowerMarker is a comprehensive set of statistical methods for genetic marker data analysis, designed especially for SSR/SNP data analysis. PowerMarker builds a <u>powerful user interface</u> around both new and traditional statistical methods for population genetic analysis. See <u>analysis</u> to check out the versatility of PowerMarker. PowerMarker is also a 2D Viewer - which was used intensively for visualizing linkage disequilibria results. <u>Download PowerMarker now and speed up your data analysis!</u>

http://statgen.ncsu.edu/powermarker/





TASSEL (Buckler Laboratory, Cornell University): a software package to evaluate traits associations, evolutionary patterns, and linkage disequilibrium.

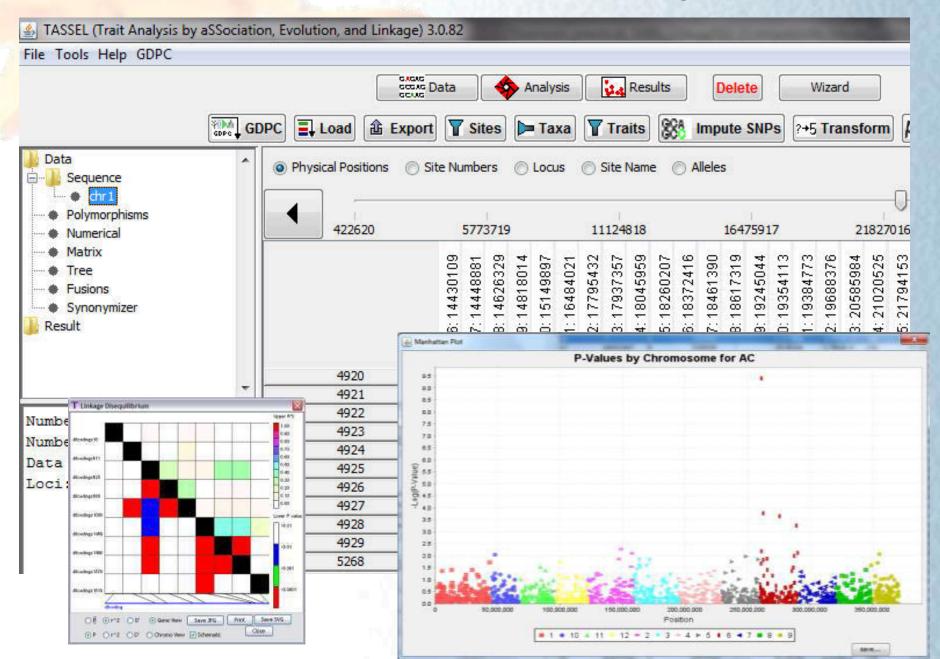
Three areas of strength:

- 1. Integrates with various diversity databases (Panzea, Gramene, Sorghum, and GRIN)
- 2. Provides new and powerful statistical approaches to association mapping eg. General Linear Model (GLM) and Mixed Linear Model (MLM).
- 3. handles a wide range of indels (insertion & deletions) which is the most common type of polymorphism in maize.

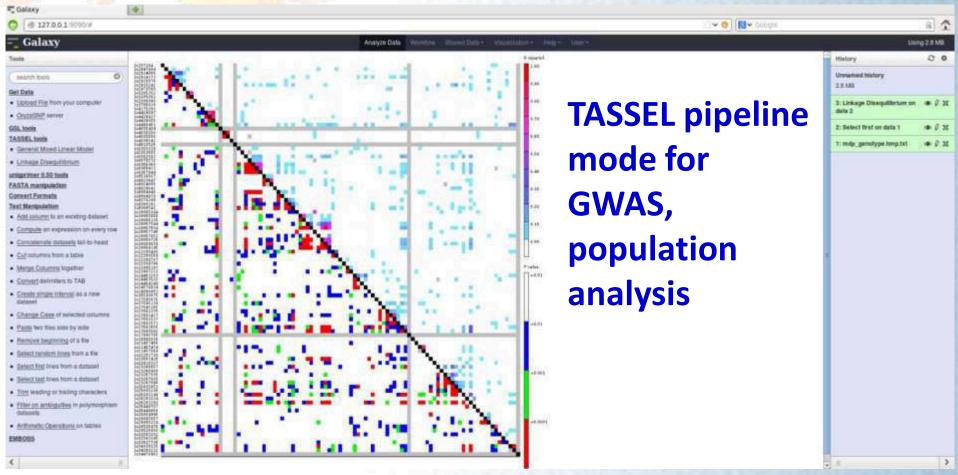
www.maizegenetics.net/tassel



Tassel stand-alone has lots of analysis tools...

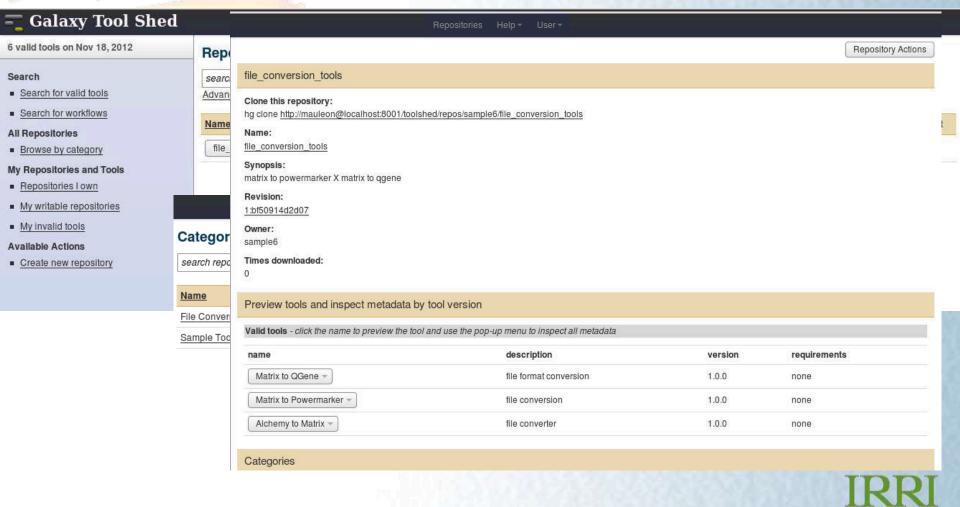


TASSEL analysis tools are being incorporated into Galaxy ...





IRRI Galaxy Toolshed ("APPS STORE") is under development



Genotyping data management

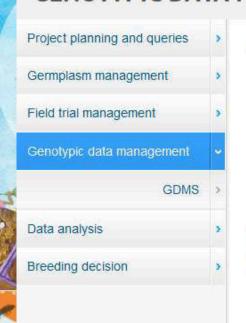
IRRI GSL manages data of customers ...

- Customer declares as private retained in GSL Galaxy account of customer
- Customer declares data as public loaded into Genotyping Data Management System; shared with research community





GENOTYPIC DATA MANAGEMENT



Genotypic Data Management System

Category Data management

Usability Easy to use

Platform







Version Beta 1

Developers Trushar Shah (ICRISAT)

Genotypic Data Management System (GDMS) enables users to store, search and retrieve molecular marker information, genotyping data and genetic maps.



This tool is in development

Login or register to post comments: Genotypic data management







GENOTYPING DATA MANAGEMENT SYSTEM



Home Upload

Retrieve

Delete

Logout

Welcome

Username:	public		
Password:			
S	Submit Clear		

The **Genotyping Data Management System** aims to provide a comprehensive public repository for genotype, linkage map and QTL data from crop species relevant to the GCP's aim to boost crop productivity and resilience for smallholders in drought-prone environments.

This system is developed in Java and the database is MySQL. The initial release records details of current genotype datasets generated for GCP mandate crops along with details of molecular markers and related metadata. The Retrieve tab on banner is a good starting point to browse or query the database contents. The datasets available for each crop species can be queried. Access to datasets requires user login.

Data may be currently exported to the following formats: 2x2 matrix and Flapjack formats. Data submission is through templates; upload templates are available for genotype, QTL and map data (type of markers - SSR, SNP and DArT).

Contact

Login Successful!

Now you can <u>Upload</u>, <u>Retrieve</u> or <u>Delete</u> Data





(Data can be uploaded using provided templates.

To upload, select button, browse & upload template containing data.)

Please upload Marker Information before uploading Genotyping Data

Marker Information

Genotyping Data

Maps/QTLs

SSR Marker

SNP Marker

CISR Marker

CAP Marker

SSR Marker Sample Template

SNP Marker Sample Template

CISR Marker Sample Template

CAP Marker Sample Template

Choose File No file chosen

Submit





GENOTYPING DATA MANAGEMENT SYSTEM



Home Upload Retrieve Delete Logout





Genotyping Data Retrieval

• Genotypin	ng Matrix	Polymorphic Markers	[©] Map/QTL Data	
Retrieve using :	◎ GIDs	GermplasmNames	Markers	Dataset
Select the Dataset	: 11862_SNP	set 🔻		
Choose Data Export For Genotyping X Marker Ma Flapjack Please select the map		/iew ▼		
	Flapjack data fFlapjack Map fi			



Genotyping Data Retrieval

Genotyping Matrix

Polymorphic Markers

Map/QTL Data

Retrieve using:

O GIDs

GermplasmNames

Markers

Operation of the contract o

Select the Dataset

11862_SNPset ▼

Choose Data Export Format You Would Like to View

Genotyping X Marker Matrix

Flapjack

2016 Germplasm ID(s) 384 Marker(s)

Data Export Formats

Genotyping X Marker Matrix





Genotype data matrix ...

```
fd17
                               fd6 fd7 fd8 fd9 id10007384 id1004256 id1018329 id11000133 id11008929 id1
-40000 CK00QJ008-RiceSNP-Plate5 R001 C001 T/T C/C T/T N/N N/N G/G A/A G/G A/A G/G C/C A/A G/G C/C G/G C/C T/T A/A C/C
-40001 CK00QJ008-RiceSNP-Plate5 R002 C001 T/T C/C T/T N/N N/N G/G A/A G/G A/A G/G C/C C/C G/G C/C T/T A/A C/C
-40002 CK00QJ008-RiceSNP-Plate5 R003 C001 T/T T/T T/T N/N N/N G/G C/C A/A T/T A/A T/T A/A T/T C/C A/A T/T A/A G/G T/T
-40003 CK00QJ008-RiceSNP-Plate5 R004 C001 T/T C/C T/T N/N N/N G/G A/A A/A A/A G/G C/C A/A G/G C/C G/G C/C T/T A/A C/C
-40004 CK00QJ008-RiceSNP-Plate5 R005 C001 T/T C/C T/T N/N N/N T/T A/A G/G A/A G/G C/C C/C G/G C/C T/T A/A T/T
-40005 CK00QJ008-RiceSNP-Plate5 R006 C001 T/T C/C T/T N/N N/N G/G A/A G/G A/A G/G C/C C/C G/G C/C G/G T/T T/T A/A C/C
-40007 CK00QJ008-RiceSNP-Plate5 R008 C001 T/T T/T T/T N/N N/N T/T A/A A/A T/T A/A T/T A/A T/T C/C A/A T/T A/A G/G T/T
-40008 CK00QJ008-RiceSNP-Plate5 R001 C002 T/T C/C T/T N/N N/N G/G C/C A/A A/A G/G C/C C/C G/G C/C G/G C/C A/A A/A C/C
-40009 CK00QJ008-RiceSNP-Plate5 R002 C002 T/T T/T T/T N/N N/N G/G C/C A/A T/T A/A T/T A/A T/T C/C A/A T/T A/A G/G T/T
-40010 CK00QJ008-RiceSNP-Plate5 R003 C002 T/T T/T T/T N/N N/N G/G C/C A/A T/T A/A T/T A/A T/T C/C A/A T/T A/A G/G T/T
-40011 CK00QJ008-RiceSNP-Plate5 R004 C002 T/T T/T T/T N/N N/N G/G C/C A/A T/T A/A T/T A/A T/T C/C A/A T/T A/A G/G T/T
-40012 CK00QJ008-RiceSNP-Plate5 R005 C002 T/T T/T T/T N/N N/N T/T A/A A/A T/T A/A T/T A/A G/G T/T A/A T/T A/A A/A T/T
-40016 CK00QJ008-RiceSNP-Plate5 R001 C003 T/T T/T T/T N/N N/N G/G A/A G/G A/A G/G C/C C/C G/G C/C T/T A/A C/C
-40017 CK00QJ008-RiceSNP-Plate5 R002 C003 T/T C/C T/T N/N N/N G/G A/A G/G A/A G/G C/C C/C G/G C/C T/T A/A C/C
-40018 CK00QJ008-RiceSNP-Plate5 R003 C003 T/T C/C T/T N/N N/N G/G A/A A/A A/A G/G C/C C/C G/G C/C T/T A/A C/C
-40019 CK00QJ008-RiceSNP-Plate5 R004 C003 T/T C/C T/T N/N N/N G/G A/A G/G A/A G/G C/C A/A G/G C/C G/G C/C T/T A/A C/C
-40020 CK00QJ008-RiceSNP-Plate5 R005 C003 T/T C/C T/T N/N N/N G/G A/A G/G A/A G/G C/C C/C G/G C/C T/T A/A C/C
```



Genotyping Data Retrieval





Submit

$\textbf{'127'} \ \text{markers are polymorphic between } \textbf{'CK001BB042-VBP_R001_C001'} \ \textbf{\& 'CK00YR020-VBP_R001_C001'}$

Marker	Marker	Marker	Marker	Marker
dd3000535	fd13	fd8	fd9	id10001250
id10001624	id10003555	id1003559	id1008267	id1008821
id1016436	id1021920	id11003281	id11003924	id11004215
id11004240	id11004398	id11004812	id11005646	id11007840
id11009201	id11009456	id11010245	id11010335	id12001224
id12001321	id12001996	id12002728	id12003803	id12004047
id12005205	id12008557	id12010050	id2000096	id2001102
id2001565	id2001831	id2002229	id2006486	id2006621
id2007526	id2009319	id2010102	id2010969	id2014034
id2016108	id3000111	id3001992	id3004190	id3004633
id3005145	id3005216	id3005817	id3006808	id3007932
id3008957	id3009433	id3010106	id3011048	id3013669
id3015399	id3017762	id3017899	id4002852	id4002913
id4003259	id4003973	id4004493	id4005120	id4007105
id4009024	id4010238	id4010621	id4011016	id4011774
id5000015	id5000759	id5003785	id5004668	id5007536
id5007714	id5010661	id5010992	id5012152	id5013231
id5014338	id5014589	id6000402	id6003000	id6004481
id6005661	id6011429	id6012426	id6013529	id6016941
id7000871	id7001478	id7001628	id7002260	id7002427
id7002784	id7002859	id7004429	id7004442	id8001908
id8002314	id8002662	id8004838	id8007472	id9002014
id9002497	id9003188	id9003471	id9004727	id9005086
id9006377	id9006988	id9007356	id9007763	ud1000711
ud2002015	ud4000438	ud8001072	wd11001469	wd2000409
wd5000542	wd8003200			

Second GRiSP Research Theme with heavy bioinformatics ...

Harnessing genetic diversity to chart new productivity, quality, and health horizons

- 1.2. Characterizing genetic diversity and creating novel gene pools (SNP genotypes, whole genome sequencing, phenotypes)
- 1.3. Genes and allelic diversity conferring stress tolerance and enhanced nutrition (candidate genes)



IRGC - the International Rice Genebank Collection @ IRRI

World's largest collection of rice germplasm held in trust for the world community and source countries (www.irri.org/GRC)



Rice exhibits deep population structure.

Phylogenetic tree for 200K SNPs on 3,000 lines McNally et al., 2013 unpublished

Unpublished data removed



The Rice 3,000 Genomes Project: Sequencing for Crop Improvement

Kenneth McNally, Nickolai Alexandrov, Ramil Mauleon, Chengzhi Liang, Ruaraidh Sackville Hamilton,

Zhikang Li, Ren Wang, Hongliang Chen, Gengyun Zhang, Hongsheng Liang, Hei Leung, Achim Dobermann, Robert Zeigler







+ Many Analysis Partners



Bioinformatics challenges of the project...

- Primary data analysis: SNP calls, reference genome refinement, phylogenetic analysis, genotype → phenotype association, etc...
- Efficient <u>database</u> system that allows the integration of the genebank information with phenotypic, breeding, genomic, and IPR data
- Development of <u>toolkits/workbenches</u> for use by research scientists and rice breeders
- Make these databases, tools, & analyses results <u>publicly accessible</u> (& constantly updated)



More analysis ...

Can we assemble new references?
Find important SNPs (merging with current GWAS/QTL results)

- in CDSs
- in promoters and other regulatory motifs
 Reconstruct large deletions/insertions/inversions in genome

Find correlated SNPs

Focus on known genes associated with traits

Find conserved genome regions selected by breeders

- Need for speed
- Need for collaborations



IRIC: International Rice Informatics Consortium

PAG 2013: first introduction of the initiative to the scientific community





IRIC Portal is a central point of IRIC



Bioinformatics Institute

A*STAR Singapore

NIAS
MIPS
CAS
Academia Sinica
EMBRAPA
CSHL

Cornell
Cirad
CAAS
MPI
AGI
Gramene

TGAC
IRD
KZI
Wageningen UR
Plant Onto
Uni Queensland



Initial Contact Organizations

GRiSP Centers (4)

IRRI CIAT IRD

Cirad

Breeding Companies (7)

Bayer CropSciences

Biogemma Mahyco

Mars Food Global

Pioneer RiceTec Syngenta

Foundations (5)

Gates Foundation NSF, U.S.A. Sloan Foundation

USAID, U.S.A. USDA-CREES

Universities (14)

Arizona Genomics Institute

Cornell University

Federal University of Pelotas, Brazil

Huazhong AU, China

Katsetsart University, Thailand

Kyung Hee University, Korea

Louisiana State University

Michigan State University

Oregon State University

Perpignan University, France

UC-Riverside

University of Delaware

University of Queensland, Australia

Wageningen UR, Netherlands

Others (5)

GigaScience Journal

GigaDB.org

iPlant Collaborative, U.S.A.

Gramene

Plant & Trait Ontology

Institutions (16)

Academia Sinica, Taiwan

CAAS, Beijing & Shenzhen

CAS, Beijing

Cold Spring Harbor Laboratory

EMBL-EBI, U.K.

EMBRAPA, Brazil

ICAR, India

INRA, France

Kunming Zoo Institute, China

MIPS, Germany

MPI-Tuebingen, Germany

NCGR-CAS, SIBS, Shanghai

NIAS, Japan

The Genome Analysis Centre, U.K.

USDA-Research

NCGR, Sante Fe, NM

Tech Companies (3)

BGI-Shenzhen Affymetrix

Pacific Biosciences



Existing rice portals

MSU Rice Genome Annotation Project http://rice.plantbiology.msu.edu/

RAP-DB http://rapdb.dna.affrc.go.jp/



Rice Genome Annotation Project Funded by the NSF





The Rice Annotation Project Database

BGI-RIS Rice Information System

http://rice.genomics.org.cn/rice/index



to 2010 Update



Rice Information Syst ***

PlantGDB

http://www.plantgdb.org/OsGDB/





Gramene

http://www.gramene.org/





IRIC portal content

- Sequences and analysis of 3,000 genomes*
 - SNPs
 - assemblies
 - phylogenetic trees
 - o genes associated with traits
 - regulatory motifs
 - most significant variations
- Other available rice genome sequences (~2,000 rice entries in SRA)
- Sequences of rice microorganisms
- Sequences of other grasses (e.g. for C4 project)
- Genotyping results from GBS, 44K and 700K affy chips
- Phenotypic data
- Gene expression data
- Gene functions and networks
- Analysis tools
- Linked to rice seeds database
- Linked to other IRRI databases and portals

*Total amount of genotyping data: ~3K*20Mio = 60Bio



IRIC portal development team @ IRRI

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Jeffrey Detras
Roven Rommel Fuentes
Ramil Mauleon
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Visionary input
Achim Dobermann

Technological advices
Marco van den Berg

Management team
Hei Leung
Ruaraidh Sackville Hamilton
Kenneth McNally
Ramil Mauleon
Nickolai Alexandrov



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- Now Hiring: Two (2) post-doctoral positions for computational biology / bioinformatics based at IRRI
- http://irri.org

Join Us - The IRRI Job Board

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Postdoctoral Fellow - Computational Biology (PDF-2013-11-KM)	10/31/2013	Philippines	Fixed-term (Renewable), National	PhD in Genetics, Bioinformatics, Statistics or closely related disciplines.	Apply now
Postdoctoral Fellow - Bioinformatics (PDF-2013-10-NA)	10/27/2013	Philippines	Fixed-term (Renewable), International	PhD in Bioinformatics, Computer Science, (Bio) Statistics	Apply now

