

RGMIMS: a web-based Laboratory Information Management System for plant functional genomics research

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Abstract A robust Laboratory Information Management System (LIMS) is required for the efficient handling of data generated from large-scale insertional mutagenesis projects. The Rice Gene Machine Information Management System (RGMIMS), a web-based modular LIMS, developed in a rice functional genomics laboratory at CSIRO, currently has four core modules: Seed Management, Transformation Management, Plant/Progeny Management, Phenotype Management, and an ad hoc querying module. RGMIMS manages, preserves and tracks large inventories of transgenic germplasm and enables efficient and accurate record keeping of the large quantities of experimental data. RGMIMS automates and seamlessly integrates multi-step experimental processes. A web user interface, incorporating bar-coding utilities, enables rapid data capture and tracking of biological resources. Ontologies from

Gramene and Plant Ontology consortium are used to describe mutant phenotypes. RGMIMS supports generic research processes in plant mutagenesis and could readily be adapted to general LIMS for high-throughput plant research.

Keywords Insertional mutagenesis · Laboratory Information Management System (LIMS) · Ontology · Phenotype · Rice Gene Machine Information Management System (RGMIMS) · Rice · Transformation · Web application

Abbreviations

LIMS	Laboratory Information Management System
PACLIMS	Phenotype Assay Component LIMS
RGMIMS	Rice Gene Machine Information Management System
TPM	Transgenic plant monitor

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Results

Availability and contacts

RGMIMS is available freely to public-sector laboratories via a standard material transfer agreement with CSIRO. Demonstration software and supplementary information are available through <http://www.pi.csiro.au/fgrttpub/rgmims.htm>. Either the first or the last authors can be contacted for further information.

RGMIMS platform

RGMIMS is implemented in Java (<http://www.java.sun.com>), a platform-neutral open-source software, and developed on the Microsoft Windows's platform (<http://www.microsoft.com>). It has been tested using Windows XP and SUSE Linux operating systems, with Internet Explorer browser 6.0 and Mozilla

Firefox 2.0 client browsers. RGMIMS uses a client/server architecture: a rich browser client (a Java Applet and Java Server Pages, <http://java.sun.com>), Apache Tomcat server (with a Java servlet acting as a broker, <http://tomcat.apache.org>) to execute the business logic and mediate the communication between the client and the back-end relational database (Microsoft SQL Server 2000, <http://www.microsoft.com>).

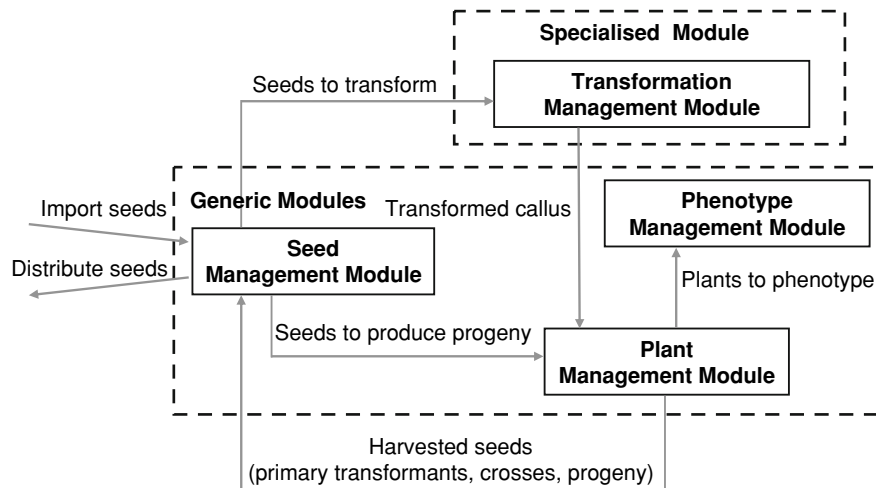


Fig. 1 RGMIMS has four core modules: Seed, Transformation, Plant and Phenotype. Each module supports a related set of functionality used at different stages of the operational insertional mutagenesis workflow. The modules are integrated and share information between them to reduce data redundancy

(the arrows show the interconnections between the modules and the flow of information). However, the modules are loosely coupled so that each can be used as a stand-alone such as the seed module, or in combination such as the seed and plant modules or the seed, plant and phenotype modules

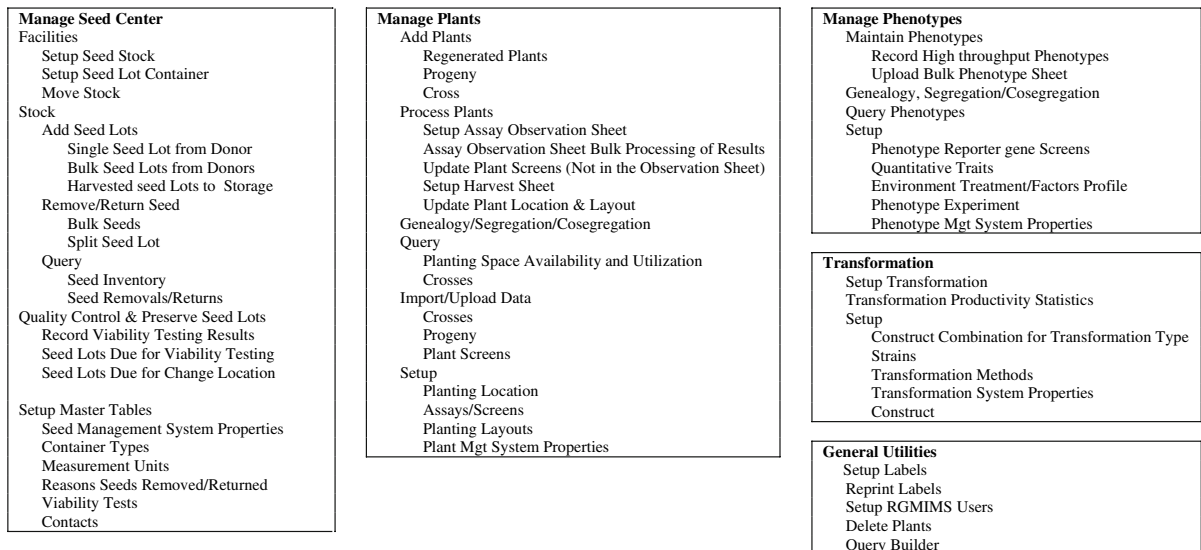


Fig. 2 Menu structure of integrated modules of RGMIMS

Key features of RGMIMS

RGMIMS has four core modules—Seed Management Module, Transformation Management Module, Plant Management Module and Phenotype Management Module (Fig. 1). The available menu structure is presented in Fig. 2.

Seed management module

This module manages information about seed receipt, handling, storage, inventory levels, quality and preservation. Seed acquisition is recorded and seeds are

assigned a storage location (Fig. 3). Accession number, passport information, taxonomy of imported seeds are captured. NCBI's taxonomy (<http://www.ncbi.nlm.nih.gov>) is used to standardise the description of plant species. Seed storage facilities are organised hierarchically in terms of stores, containers and packets. The facilities can be reorganised graphically using “drag and drop” features. The seed inventory provides information on seed availability and an audit trail of seed movements. Seed lots can be split into sub-lots, each with a unique accession and storage location. Seed viability review results are recorded to ensure the quality of seed in storage; RGMIMS automatically flags any seed lots due for

Figure 3 consists of four screenshots from the RGMIMS software interface:

- (a) Add Single Seed Lot from Donor:** A form for recording seed lot details. Fields include Preferred/Inhouse Line Name (R36), Critical Seed Threshold (10), Donor Line Name (Junkown), and various identification numbers (Quarantine Despatch, CPI, AUS, Harvest Date). It also includes a section for Notes and Seed Store information.
- (b) NCBI Taxonomy Browser:** A window showing a hierarchical tree of species. The tree is expanded to show 'Oryza sativa (indica cultivar-group)' selected.
- (c) Setup Contacts:** A window for managing contacts. It shows a list of contacts on the left and detailed information for 'Brar, Darshan' on the right, including name, phone, email, and address.
- (d) List of Seed Stores and Storage Containers:** A window for selecting storage locations. It shows a list of stores (Cold room, Cold Storage CSU, working stock) and a list of containers (freezer box, Box 1, Box 2, Box 3).

Fig. 3 Seed acquisition from donors. (a) Details of the imported seed lot are recorded, such as donor accession number, passport information and imported quantity; (b) a taxonomy is selected from the NCBI Taxonomy browser to describe the species of the imported seeds; (c) a donor is

selected from an electronic diary of contacts; (d) a storage location is assigned to the seeds from the pick list of customised stores and containers. A measurement unit is selected from the pick list of customised units (not shown)

quality control based on seed age and previous viability results. To preserve seeds in storage and prevent their deterioration and loss, the system automatically flags seed lots due for movement to more ideal store rooms, based on seed age and the customised period for each store.

Transformation management module

This module handles information about the transformation systems used to produce genetic modifications in the plants under study (Fig. 4). It records details of tissue culture when it is a component of the transformation strategy. Details and results of transformation protocols are recorded, including gene constructs, selected callus/plant lines. The efficiency of the tissue culture processes and transformations

can be easily monitored, from the initiation of callus to the number of transgenic lines obtained.

Plant management module

This module manages information about the generation, screening, monitoring and harvesting of the transgenic as well as non-transgenic plants. The production of regenerated lines and the propagation of plants through crossing and selfing are recorded. An ‘observation sheet’ is used to screen and record the results of a variety of assays that are applied to the lines. A ‘harvest sheet’ is used to monitor plant harvest, to print barcode seed packet tags for harvested plants and to record the yields of seed lots from single or pooled plants. A pedigree tree is dynamically generated showing plants, parents,

(a) Add New Transformation

Name: New Transformation
 Gene Transfer Method: Agrobacterium
 Transformation Type: DsE_single
 Transform Date:
 Group ID: 26
 Construct_1: pSk100
 Strain: AGL1
 Transformed Callus Seed
 Seed Lot Barcode:
 Total Calli Used:
 Comments:
 Completed:
 Persons Handling This Transformation: UserID: upa001
 Resistant Calli:

Callus Line	Number of Regenerated Callus
1	9

(b) Retrieve Transformations

All Transformations
 Transformations For: upa001
 List of Transformations:
 TT317
 TT318
 TT319
 TT320
TT321
 TT322
 TT323
 TT324
 TT325
 TT326
 TT327
 TT328
 TT329
 TT330
 TT331
 TT332
 TT333
 TT334
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 TT343
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 TT345
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 TT351
 TT352
 TT353
 TT354
 TT355
 TT356
 TT357

(c) Transformation Productivity Statistics

Specify Search Criteria:
 Transformation: TT321
 Start Date:
 End Date:
 Handler:
 All Types Transformed Callus Seed Plant
 Search On Specified Constructs:

Construct Number	Construct Absent	Construct Name

Results based on search Criteria:
 Transformation Productivity Stats

Transformation Name	Total Resistant PreRegenerated	Total Regenerated Callus Lines	Total Regenerated Plants	No Callus Produced	Constructs Used	Date Transformed	Person Handling
TT321	22	7	48	40	pM139362; pM1400	11/01/2004	je2213

Total Transformations: 1
 Total Calli Used: 40
 Total Resistant Calli: 22
 Total Regenerated Calli: 7
 Total Regenerated Plants: 48
 Productivity of Calli Transformation (%): 57.5

Fig. 4 Transformation management. (a) New transformation details (constructs, transformation method, target tissue/quantity) are recorded with logged user as “person handling” as default; (b) once entered, existing transformation can be

retrieved to update/edit transformation progress; (c) productivity statistics can be retrieved for different transformations, callus/plant lines, persons, dates or constructs

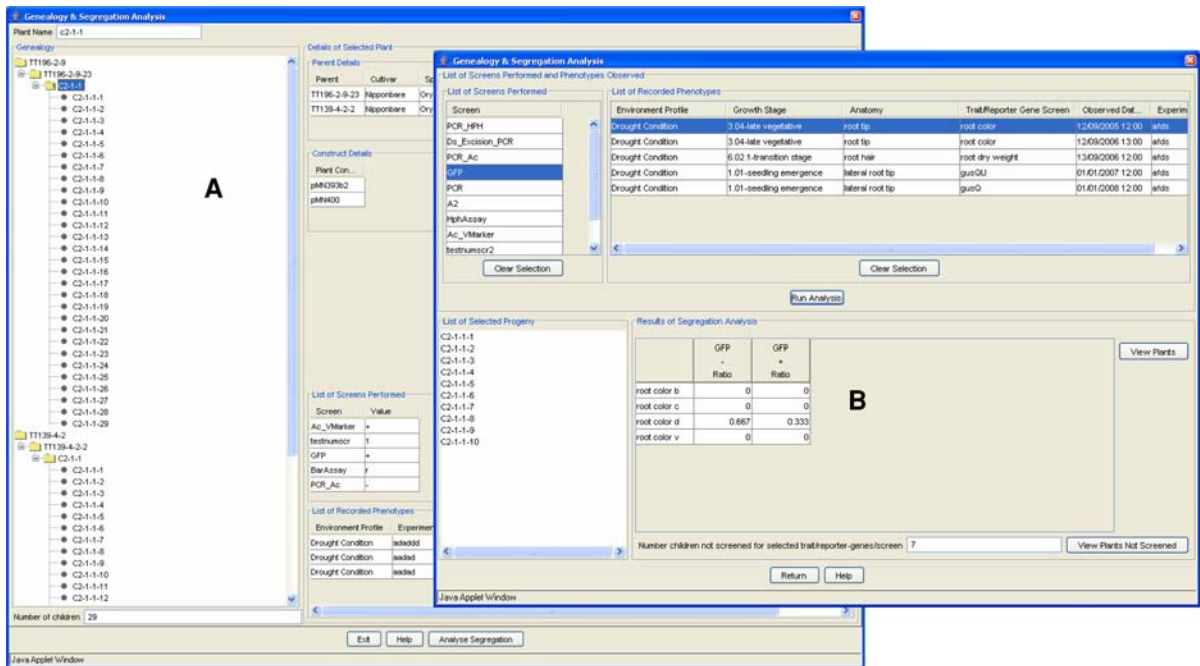


Fig. 5 A pedigree tree. (a) Parents of F_1 plants, sibling(s) and progeny are displayed on the left-hand panel and the details of molecular tests and phenotypic screening are displayed in the

right-hand panel; (b) the results of a co-segregation analysis of the selected progeny, involving a qualitative molecular screen and qualitative trait, is displayed

sibling(s) and progeny, as well as details of any molecular tests and phenotypic screening (Fig. 5). This tree permits segregation analysis where inheritance pattern is summarised.

RGMIMS captures the hierarchical organisation of the growth facilities in terms of geographical location, plots/glasshouses and rows/benches. RGMIMS enables efficient space usage of the growth facilities. For instance, newly produced lines can only be located in growth facilities with available space and the occupied space within the growth facilities is released automatically when plants are processed as harvested, dead or discarded. Reorganisation of plants within the growth facility is permitted, and including space reservation in advance.

Phenotype management module

This module handles the phenotypic characterisation of the transgenic plants, where many screens with a variety of parameters are carried out on every plant. To facilitate the sharing of information with collaborators and the wider research community, a

combination of generally accepted biological ontologies from Gramene (<http://www.gramene.org>; Jaiswal et al. 2006) and Plant Ontology Consortium (<http://www.plantontology.org>; The Plant OntologyTM Consortium 2002) are used to describe phenotypes in standardised terms. A ‘Phenotype sheet’, is used to characterise and evaluate transgenic plants (Fig. 6). It captures the results of plants screened for various physical and molecular traits, in different plant parts/organs at different developmental stages described using the relevant ontologies.

Discussion

Large-scale plant insertional mutagenesis projects, such as the CSIRO Plant Industry’s Rice Gene Machine Project (<http://www.pi.csiro.au/fgrttpub/>, Upadhyaya et al. 2006), produce vast amounts of diverse experimental data and resources. In our experience, data management with paper records, electronic spreadsheets and database tables (Microsoft AccessTM and SQLServer 2000TM) were inefficient, produced poor quality data and afforded

Panel A: Phenotype Sheet Data

Plant Name	Position No	1.01-seedling emergence root tip 10/07/2007 12:00 Value	1.01-seedling emergence root tip 10/07/2007 12:00 Lab Book Ref	1.01-seedling emergence root tip 10/07/2007 12:00 count Value	1.01-seedling emergence root length 10/07/2007 12:00 Lab Book Ref
B10-1-15-1	1633			2	lr2002
B10-1-15-2	1634	red			lr2002
B10-1-15-3	1635			3	lr2002
B10-1-15-4	1636	blue			lr2002
B10-1-15-5	1637				

Panel B: Maintain Growth Stage, Anatomy, Trait and Reporter Screen Combination

Growth Stage	Anatomy	Trait	Unit Measurement	Date to Observe	Time to Observe
1.01-seedling emergence	root tip	root color		10/07/2007	12:00
1.01-seedling emergence	root tip	root number	count	10/07/2007	12:00

Panel C: Trait Ontology Search Results

Name	Accession	Definition	Synonym
root anatomy and morphology trait	TO:0000043		
root color	TO:0000065		RTCL
root dry weight	TO:0000078	The average dry weight of the root determined p...	RTDWT,total root dry w...
deep root dry weight	TO:0000081	The average dry weight of the root, (for the root ...	drwt,deep root dry w...
penetrated root number	TO:0000083	Its the total number of roots penetrating the hard ...	rpr,number of penetra...
root number	TO:0000084	Average sum of number of roots in a plant derive ...	RTNE,br
penetrated root thickness	TO:0000090	At 50 days after sowing (DAS), the thickness of ...	PENRTH,prt
deep root to shoot ratio	TO:0000092		DRTSR0,deep root r...
root pulling force	TO:0000093		rpt,RTPF
basal root thickness	TO:0000094	The thickness of the root present 2cm below the ...	BARTH,brt

Panel D: Trait Ontology Tree

- trait ontology
 - anatomy and morphology related trait
 - root anatomy and morphology trait
 - root number
 - penetrated root number
 - total root number
 - stress trait
 - abiotic stress trait
 - root number
 - penetrated root number
 - total root number

Fig. 6 A 'Phenotype sheet'. This sheet is used to perform large-scale characterisation and evaluation of transgenic plants: (a) The results of plants screened for various physical and molecular traits, at various anatomies, developmental stages and time points described using the relevant ontologies are

captured; (b) the traits and reporter screens to be observed at various growth stages and anatomy are set up; (c) the ontology describing the trait to be observed is selected from the Plant Ontology Consortium's 'Trait Ontology'; (d) the selected trait ontology can be visualised as a tree

limited accessibility and information sharing. Integrating and managing experimental data and resources present major challenges for these projects. Several Laboratory Information Management Systems (LIMS) have been developed to address these challenges, (Scott et al. 2003; McLaren et al. 2005; Donofrio et al. 2005); however, only Phenotype Assay Component LIMS (PACLIMS, Donofrio et al. 2005) offers a web-based, platform-independent solution. These systems do not appear to possess comprehensive functionality to manage the data and resources generated throughout the insertional mutagenesis process—from transformation and tissue-culture, through breeding, molecular screening, characterisation and harvesting, to the preservation

and quality control of resources, and integration of commonly-accepted ontologies describing phenotypes and taxonomy. The RGMIMS has been developed in close collaboration with its users: research scientists, laboratory managers and technicians. Via the web, RGMIMS manages and tracks hundreds of thousands of biological resources in a collective repository, provides comprehensive electronic record management, ensures easy access to a large body of data, and achieves automation and seamless integration of insertional mutagenesis workflow.

RGMIMS is a multi-user system with access controlled by username and password. Graphical user interfaces, integrated with barcode scanners and electronic weighing scales, were designed to increase

speed and quality of data entry. Online and printed help is available. Results from analytical instruments and data from legacy systems can be imported to, or exported from, RGMIMS via spreadsheet. Query tools provide ready access to relevant information; users can formulate ad hoc queries without recourse to traditional query languages.

There are scripts to automatically download ontologies and taxonomy from public FTP sites and to upload it into RGMIMS. The user could schedule the running of these scripts at predefined times.

Traceability is maintained to ensure regulatory compliance, e.g. transformations and mutant plant lines are traceable to their source seed lots. Resources are tracked rapidly through unique identifiers, automatically generated barcode labels, and their holding locations such as growth facilities and seed storage locations.

RGMIMS is configurable to specific laboratory requirements. It can be customised to user preferences including plant naming conventions, measurement units, growth facilities, and contact details of biological resource donors or recipients. As a platform, RGMIMS could be readily adapted to support a wide variety of plant functional genomics studies.

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