15 years of GDR: New data and functionality in the Genome Database for Rosaceae

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ABSTRACT

The Genome Database for Rosaceae (GDR, https://www.rosaceae.org) is an integrated web-based community database resource providing access to publicly available genomics, genetics and breeding data and data-mining tools to facilitate basic, translational and applied research in Rosaceae. The volume of data in GDR has increased greatly over the last 5 years. The GDR now houses multiple versions of whole genome assembly and annotation data from 14 species, made available by recent advances in sequencing technology. Annotated and searchable reference transcriptomes, RefTrans, combining peer-reviewed published RNA-Seq as well as EST datasets, are newly available for major crop species. Significantly more quantitative trait loci, genetic maps and markers are available in MapViewer, a new visualization tool that better integrates with other pages in GDR. Pathways can be accessed through the new GDR Cyc Pathways databases, and synteny among the newest genome assemblies from eight species can be viewed through the new synteny browser, SynView. Collated single-nucleotide polymorphism diversity data and phenotypic data from publicly available breeding datasets are integrated with other relevant data. Also, the new Breeding Information Management System allows breeders to upload, manage and analyze their private breeding data within the secure GDR server with an option to release data publicly.

INTRODUCTION

The Genome Database for Rosaceae (GDR, https://www.rosaceae.org) is the central repository and data-mining resource for genomics, genetics and breeding data of Rosaceae, an economically and nutritionally important crop family that includes almond, apple, apricot, blackberry, cherry, peach, pear, plum, raspberry, rose and strawbery. Composed of species with a wide variety of form, habit, function and ploidy, the Rosaceae family also provides a useful system for studying plant biology (1). First established in 2003, GDR initially provided web interfaces and analysis tools for emerging genomic and genetic data such as genus-specific EST unigene sets, linkage maps and genetic markers (2,3). With the availability of whole genome sequence data and large-scale phenotypic and genotypic data, GDR, as reported in our year 10 update, markedly expanded with data and functionality (4). In the past 5 years we have witnessed another big change in the volume and type of data generated by the Rosaceae research
community (Table 1). These data include multiple versions of whole genome assemblies and annotations for each major crop species, increased RNA-Seq data, multiple single-nucleotide polymorphism (SNP) arrays for major crops, increased publications on quantitative trait loci (QTLs) and genetic maps, especially using SNPs and more breeding programs and projects that utilize SNP genotyping. We have performed new types of analyses, developed the Rosaceae Trait Ontology (TO) (5) and focused on integration of data across databases, organisms and data types. The value-added efforts undertaken in data analyses, curation and integration were combined with development and enhancement of new and existing search interfaces and tools to enable more efficient sharing and reuse of the pivotal data generated by the community. This report describes the GDR with a focus on new data and functionalities accumulated and developed in the past 5 years.

DATA AND WEB INTERFACE

The GDR interface has been re-designed to provide easier access points to data and tools such as the Major Genera Quick Start and Tools Quick Start featured on the home-page. The Major Genera Quick Start allows users to view which types of data are available for a genus of interest and provides links to access these data. Similarly, Species pages under the species navigation menu provide the same information for major species. The Tools Quick Start is organized into genomics, genetics and breeding sections; each section provides links to appropriate pages to access available data and tools. New features that can quickly familiarize users to GDR data and functionality include the dynamic data overview page where users can browse the current data types and numbers in GDR and short video tutorials. Tutorials are available for site overview, species pages, Breeding Information Management System (BIMS) and all of the search pages. GDR is implemented in Tripal (https://tripal.info), an open-source, resource efficient database platform (6,7). Below, we describe the currently available data and interfaces, with a focus on new features.

Genomics data

Whole genome sequence and associated data. Currently available in GDR are multiple versions of whole genome assemblies and annotations from seven major crops and 14 species: Fragaria vesca, Fragaria × ananassa, Fragaria trinervis, Fragaria nipponica, Fragaria nubicola, Citrus orientalis, Malus × domestica, Prunus avium, Prunus persica, Potentilla micrantha, Pyrus communis, Rosa multiflora, Rosa chinensis and Rubus occidentalis. To standardize the names of genome assemblies and annotations, GDR uses and recommends a naming protocol: [Genus] [species] genome [v[assembly version],a[annotation-version]], for both the F. vesca genome v1.1 (10) and v2.0 (11), additional annotations are available: v1.1.a2 (12) and v2.0.a2 (13), respectively. The genes from v1.1.a2 have also been aligned to the v2.0 genome, so all three gene annotations are available. The draft genome of P. micrantha (14), a species that does not develop fleshy fruit but is closely related to Fragaria, is also available. For Malus × domestica, de novo assembly of a ‘Golden Delicious’ doubled-haploid apple tree (GDDH13) with 17 pseudomolecules is now available (15) along with three versions of assemblies for the heterozygous apple genome (16). For Prunus, in addition to two assemblies of peach, P. persica genome v1.0 (17) and v2.0 (18), the whole genome assembly of sweet cherry P. avium genome v1.0 (19) is available. For Pyrus, the draft genome of the European pear P. communis genome v1.0 (20) is available. For Rosa, two new assemblies from a double haploid R. chinensis plant (21,22) are available as well as a R. multiflora draft genome v1.0 (23). For Rubus, the chromosome-scale whole genome assembly of black raspberry R. occidentalis genome v1.1 (24) as well as the draft genome v1.0 (25) are available.

Additional data provided by the GDR team on these assemblies include computational annotation of predicted genes with homology to genes of closely related or model plant species and assignment of InterPro protein domains (26) and GO terms (27,28). The GDR team also performs synteny analysis to find conserved syntenic regions among the newest versions of all publicly available Rosaceae genomes using MCScanX (29). Some of the Rosaceae whole genome assembly data are available in NCBI with NCBI’s own gene annotation and naming convention. To help researchers compare two different gene annotation sets, GDR performs BLAT analysis between NCBI annotated genes and genes from the original genome assemblies.

Assemblies annotated as above and GDR functional and synteny annotations can be accessed in their respective genome pages, gene search pages, BLAST server and using graphical viewers such as JBrowse (30) and the Tripal Synteny Viewer (https://github.com/tripal/tripal-synview). Each species page provides a summary for the species along with a resources sidebar with hyperlinks to various data and tools for the species, and has a genome subsection that lists all genome assemblies for the species. Individual genome pages provide downloadable files, including Generic File Format and FASTA formats, for an assembly that include annotated gene predictions, homology and positions of repeats and generic markers including SNPs. Additionally, lists of annotated functional terms and Microsoft Excel files of protein homologs mapped via BLAST+ (31) are available for downloading. These files contain hyperlinks to external databases as well as to GDR pages including JBrowse and gene or marker detail pages when applicable. The Search Genes and Transcripts page allows users to search for specific genes and sequences in the above assemblies and transcriptome datasets. The new search interface allows users to conduct various searches, by genus, species, dataset, gene/transcript name, genomic location and association with computationally inferred functionality such as protein names, GO terms, InterPro domains and KEGG pathway terms, all in one page. This search interface allows users to perform a query such as ‘Return all genes anno-
tated with the word ‘resistance’ between 1.0 and 3.5 Mb on chromosome Pp02 of the peach genome v2.0’. Using the search site, users can download the results or proceed to the gene details page within the GDR. Recently added functionality in the Search Genes and Transcripts page includes ‘customized output’. This option allows users to customize the result table and the downloadable Excel file to include various functional annotation results. The gene details page has several links in the resources sidebar to display the sequence and its motif annotations, genome alignments and homologies to sequences of other species in the GDR and other databases. The alignment details provide links to view a gene in JBrowse. The new synteny section lists all the orthologs and paralogs in other genomes discovered by synteny analyses and provides hyperlinks to the gene pages and Synteny Viewer. Via the Synteny Viewer page (Figure 1A), accessible from the tools navigation menu, users can choose a scaffold/chromosome of one reference genome and choose multiple other genomes for comparison. The viewer then displays a clickable circular image, as well as a table, showing all the syntenic blocks between them (Figure 1B). Choosing one block either from the image or the table leads to a page where all the syntenic genes within the block are shown with the Expect value (E-value) of their homology (Figure 1C). Gene names are linked to a specific gene page (Figure 1D) where orthologs/paralogs in other genomes are available among other information on the genes such as associated function and genomic location with a link to JBrowse (Figure 1E). Conserved synteny data made available in the GDR thus allows users starting with one rosaceous genome to explore genes, anchored trait loci and genetic markers within orthologous regions of another rosaceous genome. Using JBrowse, users can view all the genomic features aligned to the genome, such as gene models, transcripts, repeats, SNPs, other genetic markers and genes from other model plant species. In addition, the multivariant viewer JBrowse plugin has been implemented to allow users to view SNPs present among sequence data from multiple germplasm individuals. For example, RosBREED resequencing data (23 accessions) aligned to the peach v1.0 genome is available in JBrowse through the plugin. The GDR has upgraded to the Tri-pal BLAST module (http://tripal.info/extensions/modules/tripal-blast-analysis), replacing the old BLAST and batch BLAST tools. The new BLAST enables results to link to the genome scaffolds in JBrowse and to the gene/transcript details in the GDR and on the NCBI. Predicted genes from whole genome sequences were used in the construction of PlantCyc (metabolic pathway) databases (32) using PathwayTools (33). Currently, four PlantCyc databases are available in the GDR: PeachCyc, AppleCyc, FragariaCyc and RubusCyc.

Transcriptome data. The GDR team now regularly performs an analysis that combines published RNA-Seq and dbEST datasets to create a reference transcriptome (RefTrans) for major species or genera and provides putative gene function identified by homology to known proteins. The RNA-Seq sequences from peer-reviewed publications were downloaded from the NCBI Short Read Archive (SRA) and subject to quality control using the Trimmomatic (v0.32, default parameters, 34) and custom Perl scripts. The remaining RNA-Seq reads were assembled de novo with Trinity (v2.6.6, 35) using default assembly parameters and a minimum coding length of 200 bases. Qual-

| Number of entries by year | Data type | 2008 | 2013 | 2018 | Details |
|---------------------------|-----------|------|------|------|---------|
| Genome                    | 0         | 5    | 21   |      | Multiple versions of whole genome assemblies and annotations from seven major crops and 14 species |
| Gene and mRNA Transcript  | 0         | 236191 genes | 528890 genes and 585574 mRNAs |      | Genes and mRNAs from whole genome assemblies and parsed from NCBI nucleotide sequences |
| Marker                    | 1700      | 2229311 | 3285775 |      | RefTrans for six genus and unigene V5 for six genus and the rosaceae family |
| Phenotype data            | 0         | 8613  | 14411 |      | Including 127877 for Fragaria, 2613842 for Malus, 470747 for Prunus, 1593 for Pyrus, 7116 for Rosa and 8743 for Rubus |
| Germplasm                 | 0         | 8163  | 14411 |      | Including 27 for Fragaria, 108 for Malus, 168 for Prunus, 9 for Pyrus, 15 for Rosa and 13 for Rubus |
| Genotype data             | 0         | 578 568 | 878426 |      | QTL and MTL associated with 392 agronomic traits |
| Publication               | 2447      | 5182 | 7449 |      | Data available for 1967 species, specific species pages with hyperlinks to various data and tools for 13 major species |

*Table 1. Comparison of number of GDR entries between 2008, 2013 and 2018 by data type*
ity control of the ESTs included vector sequence screening (UniVec_Core, ftp://ftp.ncbi.nih.gov/pub/UniVec/) using cross_match (36), removal of tRNA/rRNA/snRNA sequences identified using tblastx (37) and Poly-A tail trimming. The filtered ESTs were assembled using the CAP3 program (P −90, 38). Bowtie (v 2.3.3) (39) was applied to multi-map the RNA-Seq reads and ESTs back to the assembled contigs and singlets. The contigs and singlets were clustered into genes using CH-HIT (v4.6.4, 40) and Corset (v1.0.7) (41) with default parameters. The longest isoform greater than 500 nt was selected to represent each Corset cluster and create the RefTrans sequences. The RefTrans sequences are functionally characterized by pairwise comparison using the BLASTX algorithm against the Swiss-Prot and TrEMBL protein databases. Information on the top ten matches with an E-value of ≤ 1E-06 are recorded and stored in GDR together with the RefTrans sequences. InterPro domains and Gene Ontology assignments are made using InterProScan at the EBI through Blast2GO. Transcriptomes and their associated annotation are available to download in the transcriptome page that can be accessed from each species page, to search and download in the Search Genes and Transcripts page, to view on the genome in JBrowse and to perform similarity searches in the BLAST server. Currently available datasets include F. vesca GDR RefTrans v1.0, P. avium GDR RefTrans v1.0, P. persica GDR RefTrans v1.0, Malus × domestica GDR RefTrans v1.0 and Rubus GDR RefTrans v2.0. The GDR continues to provide unigene v5.0 for each genus (Prunus, Malus, Fragaria, Rosa and Pyrus), composed of singlets and contigs assembled from the publicly available Rosaceae ESTs downloaded from dbEST at NCBI. Unigene v5.0 for the entire Rosaceae is composed of the assembled contigs and singlets for the five genera.

GDR also hosts user-provided assembled transcripts such as a rose transcriptome, which are orthologous open reading frame sequences of roses used to construct the WagRhSNP 68K Axiom SNP array (42), black cherry Prunus serotina unigene v1.0 from Pennsylvania State University, and assembled transcript sequences of red raspberry, Rubus idaeus (43). In addition, the GDR provides links to RNA-Seq and DNA datasets available in the NCBI SRA for Rosaceae genus or major species.

Genetic maps, genetic diversity data, markers and trait loci

Genetic maps. With continuous effort on curating peer-reviewed, published data, GDR now contains 313 genetic maps for Rosaceae species, including 168 for Prunus, 108 for Malus, 27 for Fragaria, 15 for Rosa, 13 for Rubus and 9 for Pyrus. The data associated with genetic maps include mapped positions of molecular markers, QTLs and heritable phenotypic markers, as well as mapping population(s) and publication(s). GDR has a new graphic interface, MapViewer (http://tripal.info/extensions/modules/tripalmap) to view genetic maps. MapViewer allows users to view and compare maps from different populations and species, facilitating information transfer from well-studied species to less-studied ones. These comparisons are espe-
cially useful in Rosaceae due to well conserved synteny among the genomes of Rosaceae genera (1,44). While the functionality of MapViewer is similar to CMap (45), a commonly used tool in biological databases including the GDR, MapViewer is much more integrated with other pages, such as the map, marker and QTL pages. In addition, MapViewer allows users to zoom into specific regions of a linkage group, choose types of markers to be displayed and change the colors of the markers that are displayed.

**Genetic diversity data.** The GDR contains DNA polymorphism data from various published genetic diversity studies and public breeding projects such as RosBREED (46). Currently, data from 24 diversity projects are available: thirteen from Prunus, ten from Malus/Pyrus and one from Malacomeles (false serviceberry). The GDR provides separate search pages for the SNP and the SSR genotypic data to provide appropriate query and downloadable result tables depending on the data type. There are 19 and 5 datasets available for SSR and SNP genotype search pages, respectively. In the SNP genotype search page, users can filter results by dataset name, species, germplasm name, SNP name, genomic location and/or gene name (Figure 2A). Users can also upload a file with germplasm names. This filtering allows users to perform tailored querying, such as finding SNP polymorphisms around a gene of interest in a chosen set of germplasm. The results table provides SNP name, genomic location, allele and genomic data of all of the germplasm chosen in the order of SNP location in the genome, so that users can view the genotype of each germplasm along the chromosome (Figure 2B). Users can download the genotypes for all markers displayed in the results page or the genotypes for only the markers that are polymorphic within the germplasm set chosen (Figure 2B).

**Genetic marker and SNP array data.** The GDR provides details on more than 3 million genetic markers used in genetic map development, genetic diversity studies, genome wide association studies and SNP array development. Marker annotations include marker aliases, source germplasm, source description, primer sequences, polymerase chain reaction conditions, literature references and map position, where available. For SNPs, the marker details also include SNP array name, SNP array ID, dbSNP ID, alleles, flanking sequences and probes. SNP marker data available from GDR include those from array development projects such as the 9K (47), 20K (48) and 480K (49) arrays for apple, IRSC cherry 6K array (50), IPSC peach 9K array (51), 90K array for cultivated strawberry (52) and 68K array for rose (42). The SNP array data are available to download in Microsoft Excel format from the genome pages, to view in JBrowse as well as to query in the Marker Search page. The GDR provides a new ‘SNP Marker Search’ page in addition to the existing ‘Marker Search’ and ‘Search Nearby Markers’ pages. The search filter feature in the ‘Marker Search’ page includes marker name, marker type, the species from which the marker is developed, the species to which the marker is mapped, trait name and map position in the genetic map and genome. Filtering by trait name is a new feature that allows users to search for markers that are near and/or within QTLs using the associated trait name. The table in the results page shows marker name, alias, marker type, species, genetic map location and genome location. The downloaded file contains the same information, as well as the citation. The ‘SNP Marker Search’ is designed so that users can filter using array information as well as SNP name and genomic location. The results table is also specific for SNP, with alleles, SNP array information, genome location and flanking sequences. In both search pages, users can upload a file of marker names for querying. Another search interface, ‘Search Nearby Markers’, allows users to find markers near a targeted locus.

**Trait locus data and Rosaceae Trait Ontology.** The GDR contains detailed data on published trait loci: QTLs and MTLs (Mendelian Trait Loci). These data include 3799 QTLs and 103 MTLs that are associated with 392 horticultural traits such as powdery mildew disease resistance, fruit skin color, ripening time and volatile organic compound content. These trait terms constitute the Rosaceae Trait Ontology. The Rosaceae Trait Ontology has been built to standardize trait names and abbreviations for all trait data entered into the GDR and to connect trait terms to TO (5), with the goal of data integration across databases, organisms and data types. Each of these trait terms is either an existing TO term or a child term of one. One trait term can belong to multiple Root TO terms. New Rosaceae Trait Ontology terms, which do not exist in TO, have been submitted to the Trait Ontology consortium for inclusion. In addition to the Rosaceae Trait Ontology, QTLs are annotated in the GDR with aliases, curator-assigned QTL labels, published symbols, trait names, taxa, trait descriptions, screening methods, map positions, associated markers, statistical values, datasets, contact information and references. The search page for trait loci allows searching by trait locus type (QTL or MTL), species, trait category (root TO term), trait name, published symbol and GDR-assigned label.

**Breeding data**

Breeding data stored in the GDR include phenotypic, genotypic, germplasm and pedigree data from the RosBREED project (46), the Washington State University Apple Breeding Program (53) and private breeding programs. Publicly available breeding data can be queried using the ‘Search Trait Evaluation’ and ‘Search Genotype’ pages. The ‘Search Trait Evaluation’ page provides two tabs, for querying qualitative or quantitative traits. In each tab, users can filter the data by crop dataset name and trait cut-off values of up to three trait descriptors. The result table and downloadable file has germplasm name, species, the trait values chosen and the dataset name. The germplasm and the dataset name in the results table are linked to the detail page where other associated data can be accessed. The germplasm page has a resource sidebar for genotypic and phenotypic data, as well as an overview and associated images, where the data can be viewed and downloaded. In addition, the public breeding data can be queried and downloaded using the BIMS. BIMS is a new Tripal module (http://tripal.info/extensions/modules/tripal-bims) we developed to provide breeders and breeding project teams with tools to store, manage, archive and analyze their private or public breeding data. BIMS is
Figure 2. SNP Genotype Search Page in GDR. (A) Users can search SNP genotype data by dataset name, species, germplasm name, SNP name, genome location and/or gene name. Users can also upload a file with a list of germplasm names. (B) Search result table that shows SNP name, genome location, allele and the genotype data of all the germplasm chosen in the order of SNP location in the genome. The red square highlights the options to download the genotype for all the marker displayed in the result page or the genotype data that are polymorphic in the germplasm set chosen.

Community resources and data submission

The GDR continues to provide community-based resources under the community navigation menu, including pages for the US RosEXEC, RosIGI and conference, employment notices and mailing lists. The US RosEXEC (Rosaceae Genomics, Genetics and Breeding Executive Committee) and RosIGI (Rosaceae International Genomics Initiative) serve as communication and coordination focal points for the research community, with the former also operating as the Steering Committee for the GDR. The US RosEXEC and RosIGI pages provide official documents, minutes of quarterly meetings, membership lists and subcommittee information. Several mailing lists, in addition to the GDR mailing list, are available to serve the community with information for specific interests or purposes and the archives can be viewed through the message board sites.

While the GDR team actively curates data from publications, we encourage authors to officially archive their datasets by submitting their data at the time of manuscript submission. The Data Submission page under the Data navigation menu provides various data templates for genes, genetic maps, QTL maps, as well as genotypic and phenotypic data. In addition, the page has a link to a page where the recommended list of files for whole genome data submission are available.

CONCLUSION AND FUTURE DIRECTION

Recent availability of multiple genome assemblies and annotation data from seven major crops and 14 species in the Rosaceae family opened up the opportunity to investigate the evolution and biological basis of a wide range of plant forms and functions, as well as to share knowledge among major rosaceous crops to improve their performance. With the goal of facilitating these efforts, the GDR focused on integrating the wealth of the new genomic data with transcriptomic, genetic map, genetic marker, trait locus, phenotypic and genotypic data. To accommodate scientists’ data-
mining needs that came with these new types and large volume of data, we made various new web interfaces available. The new web interfaces are either new Tripal modules that we developed, such as MapViewer, BIMS, Chado Loader, Chado Data Display and Chado Search modules (55) or Tripal modules that other database teams developed such as the Synteny Viewer and Tripal BLAST. The open-source database platform Tripal, allows us to meet emerging demands for storage, querying and display of new data types more efficiently and quickly.

During the last 5 years, we also made advances in curating data with more ontologies and incorporating ontologies in data query pages to facilitate data-sharing across different data types, species and databases. Future effort will include further developing MapViewer to integrate genome data and genetic maps, providing an enhanced querying interface, expanding analysis capabilities in BIMS through access to additional functionality, integrating Galaxy analysis platform through Tripal Galaxy module (https://www.drupal.org/project/tripal_galaxy), providing access to High Performance Computing and enabling cross-Tripal databases querying capability using Tripal Elasticsearch (56) and Tripal Exchange (http://tripal.info/tutorials/v3.x/web-services).

Use of the GDR, the community resource for Rosaceae genomics, genetics and breeding research, has continued to grow over the last 5 years. Between September 1, 2013 and August 31, 2018, the GDR had 256075 visits by 95259 unique visitors from 190 countries, who accessed 1547877 pages. The GDR is part of AgBioData (57) (https://www.agbiodata.org), a consortium working to improve standards and sustainability of genomics, genetics and breeding databases and further enable agricultural science.

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