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Running Title: Rosaceae genomics

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TITLE: Multiple models for Rosaceae genomics

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Abstract

The plant family Rosaceae consists of over 100 genera and 3,000 species that include many important fruit, nut, ornamental, and wood crops. Members of this family provide high-value nutritional foods and contribute desirable aesthetic and industrial products. Most rosaceous crops have been enhanced by human intervention through sexual hybridization, asexual propagation, and genetic improvement since ancient times, 4000-5000 B.C. Modern breeding programs have contributed to the selection and release of numerous cultivars having significant economic impact on the U.S. and world market. In recent years, the Rosaceae community, both in the U.S. and internationally, has benefited from newfound organization and collaboration that has hastened progress in developing genetic and genomic resources for representative crops such as apple, peach, and strawberry. These resources, including ESTs, BAC libraries, physical and genetic maps, and molecular markers, combined with genetic transformation protocols and bioinformatics tools, have rendered various rosaceous crops highly amenable to comparative and functional genomics studies. This report serves as a synopsis of the resources and initiatives of the Rosaceae community, recent developments in Rosaceae genomics, and plans to apply newly accumulated knowledge and resources toward breeding and crop improvement.

Rosaceous crops for human health and nutrition

Rosaceae, comprised of over 100 genera and 3,000 species, is the third most economically important plant family in temperate regions (Dirlewanger et al., 2002). The U.S. is a leading producer of almond, apple, plum, peach, pear, raspberry, sour cherry, sweet cherry, and strawberry. Other non-edible species with almost exclusively ornamental value include rose, hawthorn, potentilla, cotoneaster, and pyracantha. The products of this family are in high demand for their nutritional and aesthetic values, and their cultivation drives regional economies (Fig. 1).

The Rosaceous tree genera, including *Malus*, *Pyrus*, and *Prunus*, are predominantly grown for their fruit, and originated in regions extending from Asia west to the Caucasus (Vavilov, 1951). The early *Malus* and *Pyrus* lineages gave rise to the cultivated apple (*M.* ×*domestica*), considered to be an ancient allopolyploid, with domesticated European and Asian pears representing multiple species. The fruit-producing *Prunus* also diverged into multiple species. Plums hold the central position within this genus with numerous species having originated in Europe, Asia, China, and North America (Smartt and Simmonds, 1995). The cherry subgenera evolved in Asia Minor, particularly Iran, Iraq, and Syria (Vavilov, 1951), whereas both peach and apricot have a Chinese center of origin. Pots recovered from cave dwellings in Europe contained cherry pits dated to 5,000-4,000 B.C. (Marshall, 1954).

During ancient times, *Malus*, *Pyrus*, and *Prunus* populated native human habitats, and fruits from these species were valuable sources of food. Subsequent centuries of selection and domestication produced the dramatic increase in fleshy fruit size that distinguishes today's commercial fruit from their wild relatives.

The economic importance of edible rosaceous crops derives from their flavorful fruits and nuts that provide unique contributions to dietary choices of consumers and overall human health. Rosaceous fruits are consumed in multiple forms, including fresh, dried, juice, and processed products. The variety of flavors, textures, and levels of sweetness and acidity offered by these fruits satisfies diverse consumer tastes and choices. Rosaceae fruits are also a major human dietary source of phytochemicals, such as flavonoids and other phenolic compounds, cyanogenic glucosides, phytoestrogens (Mazur et al., 2000), and phenols that could potentially yield health and disease-fighting advantages (Macheix et al., 1991; Swanson, 1998; Selmar, 1999). L-ascorbic acid, quercetin, kaempferol, myricetin, p-coumaric acid, gallic acid, and ellagic acid are

well-known antioxidants and/or cancer-inhibiting compounds that have been identified in these fruits.

Epidemiological evidence suggests that diets rich in fruit and vegetables significantly reduce cancer risk (Caragay, 1992; Ziegler et al., 1996). *In vitro* and *in vivo* studies with animal models provide evidence that fruit and leaf extracts from many Rosaceae species inhibit some cancers or have strong antioxidant activities (Yau et al., 2002). Ellagic acid, present in strawberry, red raspberry, arctic bramble, cloudberry, and other rosaceous berries (Mandal and Stoner, 1990; Hakkinen et al., 1998; Masuda et al., 1999; Hakkinen and Torronen, 2000; Harris et al., 2001; Cordenunsi et al., 2002)¸ has been shown to affect cell proliferation and apoptosis, suggesting a potential anticancer role. Some strawberry cultivars have the highest concentrations of L-ascorbic acid among all fruits (Haffner and Vestrheim, 1997). Other phenolic compounds belonging to distinct chemical classes, many of which are also potential antioxidants and anticancer agents (Eichholzer et al., 2001; Schieber et al., 2001), have been isolated from rosaceous fruits (Macheix et al., 1991).

Throughout the past century agricultural research has focused on increasing crop production to feed the growing human population. However, as malnutrition and infectious and nutrition-related diseases remain common, the research focus has begun to shift to include improving the dietary value of different foods and identifying novel compounds with pharmacological properties (Kishore and Shewmaker, 1999). Traditionally, new traits for improvement of crop plants have been primarily derived from their related wild species. However, genomics and bioinformatics advances over the past decade have provided new options for identifying useful compounds in plants and for manipulating encoding genes responsible for their production.

The profitable production of desirable fruits and nuts can be challenging as rosaceous crop producers must balance stringent quality expectations with yield requirements, cost efficiency and shipping/marketing constraints. A further challenge is to maintain crop quality following harvest while avoiding loss due to chilling injury disorders, decay, chemical contamination, and over-ripening. Thus, post-harvest and consumer-valued qualities and traits serve as ideal targets for crop product improvement. Complementary to the inescapable economic realities of crop production and distribution, consumer perception of quality characteristics deserves recognition as a primary driving force for cultivar development and

establishment of research priorities. Consumers make quality judgments based on sensory perception (including smell, taste, and appearance) as well as perceived nutritional value. The flavor quality of fruits and nuts is determined by many criteria including firmness, juiciness, sweetness, acidity, aroma, and texture.

Recent consumer interests in purchasing locally produced fruits and vegetables have highlighted the importance of rosaceous fruits and invigorated their production in areas having relative proximity to large population centers. Rosaceous crop products are valued precisely because of their nutritional and aesthetic qualities, thereby offering opportunities for varietal improvement aimed at both problem-solving and market expansion. The impressive gains achieved by conventional breeding notwithstanding, the translation of structural and functional genomics research into breeding tools and broadened research perspectives offers unprecedented opportunities for rapid and sustainable enhancement of value to all stakeholders.

Rosaceae phylogeny and diversity dictate the need for multiple models

The Rosaceae family has been traditionally divided into four subfamilies, grouped by fruit type. These include Rosoideae (*Rosa*, *Fragaria*, *Potentilla*, and *Rubus*; fruit = achene; x = 7, 8 or 9), Prunoideae (*Prunus*; fruit = drupe; x = 8), Spiraeoideae (*Spirea*; fruit = follicle or capsule; x = 9), and Maloideae (*Malus*, *Pyrus*, and *Cotoneaster*; fruit = pome; x = 17) (Potter et al., 2002). Recent phylogenetic analyses of combined sequence data from six nuclear and four chloroplast loci provided strong support for the division of Rosaceae into three subfamilies (Potter et al., 2007) (Fig. 2). These include Dryadoideae (including *Cercocarpus*, *Dryas*, and *Purshia*; x=9), Rosoideae (including *Fragaria*,*Potentilla*, *Rosa*, *Rubus*, and others; x=7), and Spiraeoideae (including *Kerria*, *Spiraea* and others; x = 8, 9, 15, or 17). Each of the latter two subfamilies has been further divided into supertribes, tribes, and subtribes. With less emphasis on fruit type, which is clearly homoplasious in the family (Fig. 2), the taxa formerly included within Maloideae and Prunoideae have now been reclassified into Spiraeoideae, with *Malus* placed in the tribe Pyreae and *Prunus* in the tribe Amygdaleae (Potter et al., 2007). The pome-bearing members of the family, such as *Malus* and *Pyrus*, as well as closely related genera that bear polyprenous drupes (e.g., *Crataegus*) have been classified in subtribe Pyrinae, which corresponds to the former subfamily Maloideae.

Patterns of diversification within the Rosaceae, combined with the need for rapid translation of genomics research into agronomic practice, suggest that multiple rosaceous species must be designated as reference models for acquisition of genomic information. Currently, the best-developed model species for Rosaceae include apple (*Malus* ×*domestica* Borkh.), peach (*Prunus persica* L. Batsch), and diploid strawberry (*Fragaria vesca* L.) (Table 1). There is no widely accepted model for Dryadoideae, and because there are no major commercial crops within this subfamily it will not be a focus of this report. Each of the three designated models represents a distant subtaxon within Rosaceae, and each has unique features making it well suited for targeted genomic research. Combined efforts toward developing integrated genetic and genomic resources in three diverse and representative species will advance genomic research in all rosaceous species.

Apple

Apple, *M.* ×*domestica* Borkh., a pome fruit in which the floral receptacle is the fleshy edible tissue, is the most important deciduous tree fruit crop grown in the U.S. and around the world. The genus *Malus* has more than 25 species and hybrids; most apple cultivars are diploid ($2n = 2x = 34$), self-incompatible, and have a juvenile period of 3 to 7 years (Korban and Chen, 1992; Janick et al., 1996).

The apple genome, at 1.54 pg DNA/2C nucleus or 750 Mb per haploid genome complement, is approximately the same size as that of tomato (Tatum et al., 2005). The apple molecular map currently has more than 900 markers on 17 linkage groups (Maliepaard et al., 1998; Liebhard et al., 2002; Xu and Korban, 2002b; Liebhard et al., 2003a; Liebhard et al., 2003b; Naik et al., 2006). Several efforts are underway to develop a substantial resource of molecular markers for genotyping and marker-assisted selection (Tartarini et al., 2000; Liebhard et al., 2002; Liebhard et al., 2003a; Liebhard et al., 2003b; Silfverberg-Dilworth et al., 2006). Transgenic apple has been obtained via *Agrobacterium*-mediated transformation of cultivars (Ko et al., 1998; Bolar et al., 2000; Defilippi et al., 2004; Teo et al., 2006), leading to promising transgenic lines with enhanced resistance to serious orchard diseases such as apple scab (*Venturia inaequalis*) and fire blight (*Erwinia amylovora*) (Ko et al., 2000; Bolar et al., 2001), and lines with suppressed ethylene and volatile esters in the fruit (Defilippi et al., 2004; Defilippi et al., 2005). Antisense RNA technology was used to induce post-transcriptional gene silencing (PTGS) and dsRNA-

mediated PTGS used to engineer resistance to crown gall (Escobar et al., 2003). Over 300,000 apple ESTs have been generated from different tissues, conditions, and genotypes (Newcomb et al., 2006; Gasic et al., 2007). Over 22, 600 EST clones from nine apple cDNA libraries, subjected to temperature or drought stress, were clustered and annotated, then used to study gene expression in response to abiotic stress {Wisniewski, 2008 #431. Park et al. (2006) analyzed apple EST sequences available in public databases using statistical algorithms to identify genes associated with flavor and aroma components in mature fruit. Sung et al. (1998) generated ESTs from young fruit, peels of mature fruit, and carpels of the Fuji apple and found that most ESTs isolated from young fruit were preferentially expressed in reproductive organs, suggesting their important roles during reproductive organ development.

Two bacterial artificial chromosome (BAC) libraries were constructed (Xu et al., 2001; Xu and Korban, 2002a) and used for map-based positional cloning of a cluster of receptor-like genes within the scab resistance *Vf* locus in apple (Xu and Korban, 2002a). Other apple BAC libraries (from both fruiting cultivars and rootstocks) were constructed and are being used for gene cloning and mapping (Vinatzer et al., 1998; Vinatzer et al., 2001). Recently, a scaffold physical map was constructed using BAC fingerprinting (Han et al., 2007). A complete genome sequence of the apple is underway, and it will be completed by mid-2008 (R. Velascco, Pers. Comm.)

Peach

Prunus has characteristic stone fruits or drupes in which seeds are encased in a hard, lignified endocarp (the stone), and the edible portion is a juicy mesocarp. Agriculturally important stone fruit species include *P. persica* (peach, nectarine), *P. domestica* L. (European or prune plum), *P. salicina* Lindl. (Japanese plum), *P. cerasus* L. (sour cherry), *P. avium* L. (sweet cherry), *P. armeniaca* L. (apricot), and *P. amygdalus* Batsch (almond). The peach karyotype ($x = 8$) has a clearly identifiable, large submetacentric chromosome and seven smaller chromosomes, two of which are acrocentric (Jelenkovic and Harrington, 1972; Salesses and Mouras, 1977). Little is known about the chromosomal location and organization of gene sequences in peach, but recent fluorescence *in situ* hybridization (FISH) in the closely related almond (*P. dulcis*) has enabled detection of individual chromosomes based on length and position of ribosomal DNA (rDNA) genes (Corredor et al., 2004). Peach chromosomal organization is probably similar to that of

other *Amygdalus* species, as crosses between peach and those closely related species (*P. ferganensis, P. mira, P. davidiana, P. kansuensis,* and the cultivated almond) are possible, and produce fertile hybrids. Crosses with species of other subgenera (*Prunophora* and *Cerasus*) such as apricot (*P. armeniaca*), Myrobolan plum (*P. cerasifera*), European plum (*P. domestica*), and Japanese plum (*P. salicina*) are also possible, but fertile hybrids are only occasionally produced (Scorza and Sherman, 1996). However, as predicted by their divergent evolutionary origin, sweet and sour cherry are considered the commercially important *Prunus* fruit species most distant from peach.

All commercial peach cultivars belong to *P. persica,* including nectarines - which differ from peach by the absence of pubescence on the fruit surface. This characteristic segregates as a simple trait, and it is presumably controlled by a single gene or a few closely-linked genes. Unlike most congeneric species that exhibit gametophytic self-incompatibility, the peach is self-compatible. Breeding cultivars through self-pollination (Miller et al., 1989) combined with recent genetic bottlenecks (Scorza et al., 1985) have resulted in lower genetic variability in peach as compared with other *Prunus* crops (Byrne, 1990). Peach is the genetic and genomic reference species for *Prunus* because of its high economic value, self-compatibility allowing for development of F_2 progenies, availability of homozygous doubled haploids (Pooler and Scorza, 1997), and possibility of shortening its juvenile period to 1-2 years following planting (Scorza and Sherman, 1996). However, one drawback is that peach transformation remains inefficient.

Various *Prunus* maps connected with anchor markers can be found at the Genome Database for Rosaceae (GDR – see below), including more than 2,000 markers, the backbone of which is the reference *Prunus* map. This consensus map has been constructed with an interspecific almond × peach F_2 population ('Texas' × 'Earlygold' or T × E) and currently has 827 transportable markers covering a total distance of 524 cM (average density of 0.63 cM/marker) (Howad et al., 2005). Twenty eight major morphological, quality, and agronomic characters with simple Mendelian inheritance (Dirlewanger et al., 2004b) and 28 QTLs (Arús et al., 2003), most of them polymorphic within the peach genome, have been placed on the T x E consensus map. The peach doubled haploid 'Lovell' has been selected by the DOE Joint Genomics Institute's Community Sequencing Program for shotgun sequencing (8x genome coverage), which is currently slated to begin in 2008.

Strawberry

The cultivated strawberry with its accessory fruit comprised of a fleshy receptacle bearing many achenes (the "true" fruit) is genomically complex due to its octoploid (2*n*=8*x*=56) composition (Davis et al., 2007). Currently, 23 species are recognized in the genus *Fragaria*, including 13 diploids (2*n* = 2*x* = 14), four tetraploids, one hexaploid, and four octoploids (Folta and Davis, 2006). Accessions of the cultivars' two octoploid progenitor species, *F. chiloensis* and *F. virginiana*, are valued by strawberry breeders as sources of novel traits, especially pest resistance and abiotic stress tolerance. Because strawberry is a relatively new crop, dating to the 1700s (Darrow, 1966), as few as three introgressive backcrosses can yield selections of cultivar quality (J. Hancock, Pers. Comm.).

The current, provisional model of the *Fragaria* octoploid genome constitution is AAA'A'BBB'B' (Bringhurst, 1990). This suggests that the octoploids are diploidized allopolyploids that have descended from four diploid ancestors. Although the diploid ancestry of the octoploid has yet to be definitively established, the leading ancestral candidate diploids are *F. vesca* and *F. iinumae*, with *F. bucharica* and *F. mandshurica* as additional intriguing possibilities (Folta and Davis, 2006).

One ancestral diploid strawberry, *F. vesca*, has emerged as an attractive system for both structural and functional genomics due to its many favorable features (Folta and Davis, 2006). Plants are compact enough to be grown on a small scale in a laboratory, easy to propagate vegetatively, self-compatible, and produce many seeds per plant (Darrow, 1966) (Table 1). *F. vesca* has one of the smallest genomes among cultivated plants (206 Mbp *vs.* 157 Mbp in *Arabidopsis*) (Akiyama et al., 2001) as adjusted by Folta and Davis (2006).

An efficient transformation protocol renders *F. vesca* amenable to genetic manipulation (El Mansouri et al., 1996; Haymes and Davis, 1998; Alsheikh et al., 2002; Oosumi et al., 2006). The diploid model complements the development of transformation systems for the octoploid cultivated strawberry (Folta and Davis, 2006; Folta and Dhingra, 2006; Mezzetti and Costantini, 2006) allowing direct ingress of transgenes into breeding populations. The availability of robust transformation and reverse genetic systems in strawberry provide excellent resources for the broader Rosaceae genomics community by uniquely complementing the abundant DNA sequence resources available in apple (*M.* ×*domestica*) and peach (*P. persica*).

Diploid strawberry has several additional advantages over other widely used model species. Unlike the dry siliques of *Arabidopsis*, the fleshy strawberry fruit allows for studying the molecular mechanisms of fruit development and ripening. Furthermore, strawberry fruit has several unique properties, including non-climacteric ripening and unusual metabolites that cannot be studied in traditional fruit models, such as tomato.

Recent evidence, based on nucleotide sequence data, has revealed a close phylogenetic relationship between *Fragaria* and *Rosa* (Potter et al., 2007), thus suggesting that genomic resources for either genus may be readily applicable to the other. Genomics resources for roses include a bacterial artificial chromosome (BAC) library for *R. rugosa* consisting of 27,264 clones, 0.5 pg or 500 Mb, and corresponding to 5.2 × haploid genome equivalents (Kaufmann et al., 2003). This BAC library has been used to develop a fine-scale physical map of the *Rdr1* region to assemble a 400 kb tiling path of six BAC clones, and covering the *Rdr1* locus which confers resistance to blackspot. Genes involved in ethylene perception, due to their role in flower initiation and senescence, and floral scent, among the most valuable traits in roses, have been of particular interest in rose research. Molecular studies have been conducted to understand ethylene perception and signal transduction pathways in rose by taking advantage of knowledge obtained from alterations in the triple response in ethylene insensitive mutants in Arabidopsis (Guterman et al., 2002; Lavid et al., 2002). Wang et al. (2004) isolated seven putative 1-aminocyclopropane-1carboxylate (ACC) synthase clones from a cDNA library of aging rose petals of *R. hybrida* a cv. Kardinal. cDNA libraries from petals of the two tetraploid *R. hybrida* cvs. Fragrant Cloud and Golden Gate have been constructed and over 3,000 cDNA clones sequenced from these libraries. An annotated petal expressed sequence tag (EST) database of ~2100 unique genes has been generated, and used to create a microarray. Scalliet et al. (2006) explored the evolutionary pathway of scent production in European (*R. gallica officinalis* and *R. damascena*) and Chinese rose (*R. chinensis* and *R. gigantea*) species, both progenitors of the modern hybrid rose, *R. hybrida.* As genomic resources continue to develop for strawberry and rose, their applicability to each other and other Rosaceae will be more clearly defined.

Rosaceae genomics

The Rosaceae genomics initiative, like those of other crop groups, exploits and extends the fundamental knowledge generated in the *Arabidopsis thaliana* model system, which has dramatically advanced our understanding of the physiology, biochemistry, genetics, and evolution of plants. Arabidopsis is characterized by its small genome, ease of transformation, rapid regeneration time, and sexual fecundity. As extensive forward and reverse-genetic

resources are available, this renders Arabidopsis an agile system for exploring fundamental biological questions. The genomic information generated directly from Arabidopsis has proven to translate well to other Brassicaceae. However, Arabidopsis shares only partial functional overlaps with many crop plants in more phylogenetically remote plant families. Here, other suitable research-friendly species have been established as family-specific models, including *Medicago truncatula* and *Lotus japonicus* for Fabaceae, rice and *Brachypodium* for Poaceae, and tomato and potato for Solanaceae. For families with valuable crop species, genomics has progressed by selecting a representative model species for the family and focusing research efforts on developing genomic tools for that system. Findings are then translated directly to other related species of agricultural importance.

In recent years, the application of molecular technologies has steadily increased for Rosaceae (Hokanson, 2001). Concerted efforts have been undertaken by the Rosaceae community to develop genomics tools for this economically important family. Coordination within the community led to the establishment of a dedicated bioinformatics portal (the Genome Database for Rosaceae: GDR) and the production of a national Rosaceae white paper as well as a USDA request for proposals that eventually funded thirteen new projects to advance Rosaceae genomics (www.rosaceaewhitepaper.com). As these projects have unfolded, the need for developing multiple, subfamily-specific model systems within the Rosaceae has become apparent, focusing on peach (*Prunus* – Amygdeloideae), apple (*Malus* – Maloideae), and strawberry (*Fragaria* – Rosoideae), and their relative community strengths in the areas, respectively, of structural genomics, functional genomics, and reverse genetics. Initial translational genomic efforts have been directed toward developing high-density molecular linkage maps to accelerate breeding through marker-assisted selection (Dirlewanger et al., 2004b).

Genetic maps. Linkage mapping in Rosaceae has been conducted on a wide range of germplasm including most of the important cultivated species. Genetic maps have been developed for different species of *Prunus*, including peach {Dirlewanger, 2006 #390; Yamamoto, 2005 #234}, almond (Foolad et al., 1995; Viruel et al., 1995; Joobeur et al., 2000), and apricot (Hurtado et al., 2002; Lambert et al., 2004; Dondini et al., 2007). Partial maps for sour cherry (Wang et al., 1998), sweet cherry (Dirlewanger et al., 2004b), and interspecific crosses between peach and other species; e.g., almond (Joobeur et al., 1998; Bliss et al., 2002; Aranzana et al., 2003; Dirlewanger et al., 2004b), *P. davidiana* (Foulongne et al., 2003), *P.*

cerasifera (Dirlewanger et al., 2004a), and *P. ferganensis* (Verde et al., 2005) have been developed. Maps for apple (Hemmat et al., 1994; Maliepaard et al., 1998; Liebhard et al., 2003a; Liebhard et al., 2003b; Liebhard et al., 2003c; Kenis and Keulemans, 2005; Naik et al., 2006; Silfverberg-Dilworth et al., 2006), pear (Yamamoto et al., 2004), raspberry (Graham et al., 2004), diploid strawberry (Davis and Yu, 1997; Sargent et al., 2004; Cipriani et al., 2006; Sargent et al., 2006), cultivated octoploid strawberry (Lerceteau-Kohler et al., 2003; Weebadde et al.), and rose (Debener and Mattiesch, 1999; Debener et al., 2001; Rajapakse et al., 2001; Yan et al., 2005) are also available. Many of these genetic maps were constructed for particular breeding goals using specific mapping populations, resulting in multiple genetic maps of various lengths that vary in degree of saturation and type of markers. Efforts are underway to use high-density molecular markers and anchor loci to consolidate this information into a single genetic consensus or reference map for each species. Characteristic genetic linkage maps for three model rosaceous species (apple, peach, and strawberry) are described in Table 2.

Marker-assisted breeding Markers tightly linked to major genes for important traits, such as disease and pest resistances, fruit or nut quality, and self-incompatibility have been developed in *Malus* (Bus et al., 2000; Tartarini et al., 2000; Huaracha et al., 2004; Tahmatsidou et al., 2006; Gardiner et al., 2007), *Prunus*, and *Fragaria* (T. Sjulin, personal communication), and are currently being used for marker-assisted selection in both public and private breeding programs (Dirlewanger et al., 2004b)

Most rosaceous crops are characterized by long generation cycle and large plant size, hence the ability to eliminate undesirable progenies in breeding populations through marker-assisted selection (MAS) reduces cost, and allowing breeders to focus on populations comprised of individuals carrying desirable alleles of genes of interest. In peach, Etienne et al. (2002) and Quilot et al. (2004) mapped several major genes and QTLs related to fruit quality in intra and inter-specific peach progenies. Blenda et al. (2006) identified an AFLP marker significantly associated with peach tree short life (PTSL), a complex disease syndrome. This identification could lead to a MAS application in peach. Lalli et al. (2005) generated a resistance map for *Prunus* with 42 map locations for putative resistance regions distributed among seven of eight linkage groups, and both Lu et al. (1998) and Dirlewanger et al. (2004) identified markers linked to genes determining root knot nematode resistance. Selection for self-compatibility alleles can be performed in *Prunus* self-incompatible species using markers based on genes coding for this

character (Habu et al., 2006; López et al., 2006). In strawberry, a series of markers has been developed for traits of economic importance, namely for anthracnose resistance (Lerceteau-Kohler et al., 2003) and seasonal flowering (Albani et al., 2004; Sugimoto et al., 2005). In addition, a sequence-characterized amplified region (SCAR) marker, SCAR $R1_A$, has been linked to locus *Rpf_1* which confers resistance to a race of *Phytophthora fragariae* C.J. Hickman var. *fragariae*, which is the causal organism of red stele root rot disease in cultivated octoploid strawberry (Haymes et al., 2000).

The development of additional markers for MAS is dependent on the ability to perform genome scans of a progeny from a population segregating for a trait of interest, and then validating the trait-marker association(s) in alternate populations. However, in many rosaceous species, a genome scan is currently not possible due to a lack of evenly distributed markers that are polymorphic in the respective breeding population. Thus, marker development remains a top priority in most, if not all, rosaceous crops.

Structural genomics

BAC libraries. Large-insert BAC and cosmid/fosmid libraries have been constructed for peach and apricot (Georgi et al., 2002; Vilanova et al., 2003), apple (Vinatzer et al., 1998; Xu et al., 2001; Xu and Korban, 2002a), strawberry (Davis et al., 2007; Davis et al., 2008), rose (Kaufmann et al., 2003), and other rosaceous species.

Physical maps. Physical maps are important resources for positional cloning, marker development, QTL mapping, and cloning. They serve as useful platforms for whole genome sequencing efforts. Physical maps are available for several Rosaceae species, including peach and apple. The current physical map of peach is estimated to cover 287.1 Mb of the approximate peach genome size of 290 Mb (Baird et al., 1994), and it is comprised of 1,899 contigs, of which 239 contigs are anchored to eight linkage groups of the *Prunus* reference map. The peach map is a second generation map (Zhebentyayeva et al., 2006), expanded by HICF fingerprinting of additional BAC library clones from the existing BAC libraries of peach. A total of 2,511 markers have been incorporated into the physical framework. Due to the abundance of hybridization data, the initial physical map for peach was biased toward expressed genome regions. According to a conservative estimate the map covers the entire euchromatic peach genome. This map is publicly available at www.rosaceae.org.

A BAC-based physical map of the apple genome from 74,281 BAC clones representing ~10.5x haploid equivalents has been constructed (Han et al., 2007), and a physical map of *F. vesca* is under construction using a large-insert BAC library (Shulaev and Abbott, personal communication)

Genome sequencing. Full genome sequencing is an invaluable tool for plant researchers. The Rosaceae community will benefit greatly from sequencing genomes of the representative models for each the three subfamilies. At present, the largest publicly available genomic sequence resource in Rosaceae is that of *F. vesca*, from which ~1.75 Mb of genomic sequence derived from 50 genomic sites (as fosmid clones) has been deposited in GenBank under accession numbers EU024823-EU024872. Preliminary analysis of these sequences has revealed a gene density of about 1 gene per 6 kb and an SSR density of about one SSR locus per 5 kb (Folta and Davis, 2006). The available *Fragaria* genomic sequence database will provide a valuable resource in relation to assembly and annotation of other rosaceous genomes.

Peach is a prime candidate for complete sequencing among Rosaceae models because it has a comparatively small genome and has a close structural genomic relationship to many other important fruit crops (Dirlewanger et al., 2004b). The full genome sequence of peach will provide information on upstream regions of genes (promoters and enhancers) that are important for gene regulation, aid in identifying genes not discovered through EST sequencing projects, and provide a reference genome for comparative genomics in Rosaceae. The combination of the intrinsic advantages of peach as a model organism, the advent and application of high-throughput technologies for genetic and genomic analyses, and ongoing research collaborations are generating a wealth of genomic information. An initiative to sequence the peach genome was announced by the Joint Genome Institute (JGI) at the 2007 Animal and Plant Genome Meeting in San Diego. Complete sequencing of the peach genome will provide a road map for future sequencing of other model genomes, including apple, strawberry, and other representatives of the family. A group at the Istituto Agrario di San Michele All'Adige in Italy (Dr. Riccardo Velasco, Pers. Comm.) is pursuing shotgun sequencing of the apple genome using a combination of Sanger (4× genome coverage) and 454 (7× genome coverage) sequencing.

Functional genomics

Complete sequencing of several plant genomes, including *Arabidopsis* and rice, has identified tens of thousands of genes. About half are of unknown function. A major challenge for genomics is to identify the functions of unassigned genes and use this knowledge to improve economically important agronomic and quality traits in major crops.

Expressed sequence tags. EST sequencing projects are underway for several rosaceous species. ESTs enable gene and marker discovery, aid genome annotation and gene structure identification, guide single nucleotide polymorphism (SNP) characterization and aid proteome analysis (Nagaraj et al., 2007). Over 400,000 ESTs are currently available for Rosaceae species via NCBI dbEST and the Genome Database for Roscaeae (GDR, www.rosaceae.org).. With National Science Foundation (NSF Award #0321701; Schuyler Korban, PI) funding, over 104,000 5' end apple EST sequences have been generated, and a 40,000 unigene set has been identified (Gasic et al., 2007). Another ~150,000 5' end apple ESTs have been deposited in the National Center for Biotechnology Information (NCBI) by HortResearch in New Zealand (Newcomb et al., 2006). The combined publicly available apple ESTs add up to over 260,000 sequences, resulting in a 17,000 unigene set (NCBI, apple).

EST sequencing projects in peach are being conducted in several laboratories worldwide. Many of these projects have focused on fruit development, so other tissues are underrepresented in the database. All publicly available ESTs for peach have been downloaded from NCBI and housed in GDR. There are currently ~87,751 ESTs with a unigene set comprised of 23,721 different sequences. These ESTs have come predominantly from laboratories of the *Prunus* Genome Mapping consortium, supported by the USDA-IFAFS program (Albert Abbott), ESTree (Carlo Pozzi), and the Chile Consortium (Ariel Orellano).

Approximately 50,000 EST sequences from *F. vesca* and several thousand from *F. ×ananassa* have been deposited in GenBank by several contributing laboratories. From *F. vesca*, cDNA libraries have sampled cold, heat, and salt-stressed seedlings (Slovin and Rabinowitz, manuscript in preparation), and flower buds (Brese and Davis, unpublished data). These sequences have proven invaluable for annotation of *Fragaria* genomic sequences and as a source of candidate gene and random clones for use in reverse genetics investigations. Nevertheless, when EST sequence resources are considered, development of libraries and strawberry sequencing has lagged behind other major fruit crops and warrants further investment in genomic resource development. *F. ×ananassa* represents a potential diverse source of alleles that may be

directly relevant to selected traits of interest. Furthermore, an accounting of alleles may further illuminate the ancient diploid contributors to the octoploid strawberry genome.

The GDR development team has assembled and analyzed genera specific unigenes for ~370,000 Rosaceae ESTs available at the GDR website (Jung et al., 2008). The analysis includes identification of putative functions by pairwise comparisons against protein datasets, with SSR and SNP detection. Sequence comparison with 14 of the major plant EST datasets suggests that up to 14% of the Rosaceae unigenes may be unique to the family. Many of these genes are being functionally characterized in strawberry (K. Folta, Pers comm.). The availability of full genome sequence is expected to verify these numbers and provide a more refined gene catalog of Rosaceae species.

Reverse and forward genetics Two fundamentally different approaches are currently being used to elucidate gene function. A forward genetics approach begins with the observation of a unique phenotype and seeks to identify the gene(s) responsible for that phenotype. Conversely, reverse genetics begins with a candidate gene and describes the mutant phenotypes that result upon its disruption. Despite the fact that small genomes render forward genetics a potentially useful strategy for gene discovery, currently there are few forward-genetic populations available for rosaceous crops.

One approach to generating mutant phenotypes in plants is to inactivate a gene by inserting foreign DNA via *Agrobacterium*-mediated transformation (Krysan et al., 1999). This approach can be used for both forward and reverse genetics. An advantage of using insertional mutagenesis to disrupt gene function is that all mutations are tagged by a T-DNA sequence, enabling rapid direct identification of the gene related to the observed phenotype. In plants with completely sequenced genomes, identification of a short sequence of genomic DNA flanking the T-DNA insertion allows unambiguous identification of the mutated gene and linkage of the insertion to a chromosomal location. For plants that have not been completely sequenced, molecular tools such as EST collections and BAC libraries may allow the successful cloning and characterizing of a gene of interest.

Whereas T-DNA mutants may produce loss-of-function phenotypes, comparable studies have utilized a gain-of-function approach through activation tagging (Kardailsky et al., 1999; Weigel et al., 2000). Here, a set of transcriptional enhancers (typically from the CaMV 35S promoter/enhancer) is integrated into the genome using *Agrobacterium*-mediated transformation.

Upon generally random integration the enhancers greatly increase the basal transcription rates of nearby promoters. This approach is particularly useful in identifying gene function from members of multigene families where an effective knockout strategy is precluded by genetic redundancy.

F. vesca is an excellent candidate to generate a collection of T-DNA or transposon insertion or activation tagged mutants. With its small genome, *F. vesca* will require a minimal number of independent mutant lines to cover the complete genome. Based on the ~200 Mb genome size of *F. vesca* and assuming random T-DNA integration and an average gene length of 2 kb, we calculate that 255,000 or 391,000 independent T-DNA transformed lines must be produced to mutate any single gene with 95% or 99% probability, respectively (Krysan et al., 1999). Gene content and spacing in *Fragaria* are similar to those of Arabidopsis (Folta and Davis, unpublished), suggesting that these goals are realistic. A collection comprised of both T-DNA insertional mutants and *AcDs* activation tag lines is currently under development, with over 1,000 independently regenerated; these are now being characterized at Virginia Tech. Fewer activation tagged lines will be needed because one tag may affect several kb of gene space. Sequencing flanking regions adjacent to T-DNA integrations will provide resources both for reverse genetics and many markers for future mapping effort while revealing genes that affect important traits and interesting phenotypes.

<u>*RNAi*</u> Computational analysis of approximately 120,000 ESTs from *M. ×domestica* has identified ten distinct sequences that can be classified as representatives of seven conserved plant miRNA families, and secondary structure predictions reveal that these sequences possess characteristic fold-back structures of precursor miRNAs (Schaffer et al., 2007). Gleave et al. (2008) analyzed approximately 120,000 *M. ×domestica* cv. Royal Gala ESTs to identify ten distinct sequences that could be classified into seven conserved plant miRNA families. Three of these miRNAs were expressed constitutively in all tissues tested whereas others showed more restricted patterns of expression, A candidate gene approach coupled with RNAi silencing is being used to determine the function of apple ESTs in resistance to the bacterial fire blight disease (Norelli et al., 2007). Bioinformatics is used to identify publicly available apple ESTs either uniquely associated with *Erwinia amylovora* infected apple or similar to *Arabidopsis* ESTs associated with *Pseudomonas syringae* pv. *tomato* infection. An efficient transformation system for apple has been developed in which only a single EST-silencing gene is inserted per

transgenic line to select for apple RNAi mutants. The system uses a multi-vector transformation approach and PCR primers developed for pHellsgate8-derived vectors that detect presence of single or multiple EST silencing insertions in RNAi transgenic line and provide a sequencing template for determining the EST contained in the silencing insert. Additional candidate ESTs are currently being selected based upon cDNA suppression subtractive hybridization and cDNA-AFLP analyses (Norelli et al., 2007).

Neither identification of a mutant phenotype nor identification of a gene sequence alone will explain the molecular function of a gene. Modern functional genomics relies heavily on high throughput profiling methodologies like gene expression profiling, proteomics, metabolomics, and bioinformatics for detailed gene characterization. Most importantly, readily available transformation systems provide means for validating gene function in vivo.

Transcriptomics. Apple has the largest collection of ESTs and microarrays, and will provide the foundation for rosaceous microarray and gene expression analyses. Several apple microarrays are currently available or under construction and are available on different platforms, including Nimblegen, Invitrogen, and Affymetrix. Pichler et al. (2007) have demonstrated that field microarray experiments provide a viable approach for measuring seasonal changes in gene expression during apple bud development. Teamed with transgenic plants, custom apple arrays have also revealed that ethylene controls substantial production of flavor-associated volatiles (Schaffer et al., 2007), and other studies have investigated early fruit development (Newcomb et al., 2006; Lee et al., 2007). Some of the most fruitful early plant microarray experiments have been performed in strawberry. Using custom arrays, transcripts associated with ripening (Schwab et al., 2001), stress and auxin-induced gene expression (Aharoni et al., 2002), volatile production (Aharoni et al., 2004), fruit development (Aharoni and O'Connell, 2002), and flavor (Aharoni et al., 2000) have been identified. These seminal studies represent well-designed and highly productive approaches that answered fundamental questions in plant biology. More importantly, research suggests that contemporary studies with larger probe sets may lead to important discoveries for a multitude of plant responses.

Proteomics. Proteomics studies in the family are limited to date. A proteomic approach was applied to study flesh browning in stored 'Conference' pears (Pedreschi et al., 2007) and to identify novel isoforms of Pru av 1, the major cherry allergen in fruit (Reuter et al., 2005). Other studies to investigate the roles of dehydrins, among others, in cold temperature stress responses

of peach and apple using proteomic approaches are ongoing (Renaut et al., 2006; Renaut et al., 2008).

*Metabolomics***.** Rosaceous plants are extremely rich in specialized metabolites, many of which have documented utility in human health and nutrition. Chemistry and biosynthetic pathways of flavonoids, anthocyanins, and phenolics have been extensively studied in numerous rosaceous fruits with targeted analytical assays. Recent developments in metabolomics allow for global analysis and interrogation of metabolic networks. An untargeted metabolomics approach based on Fourier Transform Ion Cyclotron Mass Spectrometry was used to study four consecutive stages of strawberry fruit development and identified novel information on the metabolic transition from immature to ripe fruit (Aharoni and O'Connell, 2002). Metabolomic profiles of apple fruit revealed changes provoked by UV-white light irradiation and cold storage in primary and secondary pathways associated with ethylene synthesis, acid metabolism, flavonoid pigment synthesis, and fruit texture (Rudell et al., 2008). Broader use of metabolomics in Rosaceae will speed the discovery of novel gene functions in primary and secondary metabolism and will provide comprehensive data sets necessary to model metabolic networks related to human health-promoting metabolites.

High-throughput transformation and gene function validation. High-throughput profiling platforms and bioinformatics data mining generate numerous biological hypotheses about candidate gene functions that must be tested in subsequent experiments. These follow-up experiments rely upon the availability of a robust transformation protocol. Transformation systems have been developed in many rosaceous crops, including apple, peach, rose, and strawberry. Using degenerate primers designed from conserved regions of transcription factor genes, Ban et al. (2007) identified a transcription factor from apple skin responsible for red coloration; its function was verified in transient transformation of apple seedlings as well as in transgenic tobacco. A similar candidate gene approach has been used to identify a transcription factor responsible for red flesh and foliage color in apple (Chagné et al., 2007; Espley et al., 2007).

Rosaceae comparative genomics

A phylogenetic view provides a refined basis for the inference of homology, orthology, and functional conservation with well-studied plant species. Common markers between maps

constructed from intra- and interspecific populations of *Prunus* allow for comparisons among genomes of seven species, including peach, almond, apricot, cherry, *P. cerasifera*, *P. davidiana*, and *P. ferganenesis*. All comparisons are essentially syntenic and colinear (Arús et al., 2003; Dirlewanger et al., 2004b), thus strongly suggesting that the *Prunus* genome is similar within the genus and in agreement with patterns of frequent intercrossability and interspecific hybrid fertility known between many species of this genus. Only two major chromosomal rearrangements, both reciprocal translocations, have been reported. One was reported in the F_2 of a cross between 'Garfi' almond and the red leaf peach rootstock 'Nemared' and involved linkage groups 6 and 8 (G6 and G8) (Jauregui et al., 2001). This translocation was also found in the cross 'Akame' × 'Juseitou', where 'Akame' is also a red-leafed genotype (Yamamoto et al., 2005). The other reciprocal translocation was found in a genotype of myrobolan plum (*P. cerasifera*) and affecting G3 and G5 (Lambert et al., 2004). These results suggested that reciprocal translocations might occur, and were maintained within a given *Prunus* species characterizing monophyletic transects (i.e., red-leafed peaches) although the majority of individuals within a species were within the general *Prunus* genome configuration.

Pear maps constructed by Dondini et al. (2004), Pierantoni et al.(2004), and Yamamoto et al. (2002; 2004) have sufficient SSRs to enable alignment of all pear and apple linkage groups. Map comparisons suggest that genome organization is conserved between apple and pear.

A comparison between apple and peach genomes (Fig. 3) is possible based on 30 common loci (24 RFLPs and 6 isozymes) found between the 'Texas' × 'Earlygold' *Prunus* map and the 'Prima' × 'Fiesta' *Malus* map (Dirlewanger et al., 2004a). *Prunus* linkage groups 3 and 4 (G3 and G4) have three or more markers in common with the *Malus* map; each *Prunus* group has detected a pair of homoeologous apple groups with the same marker order. Each of two additional *Prunus* groups (G2 and G7) has two markers in common with one *Malus* linkage group, and G1 has eight markers in common with *Malus*. G1 is the longest and most populated linkage group with markers in T × E and most other *Prunus* maps, suggesting that it corresponds with chromosome 1. *Malus* does not have a similar long chromosome. Four markers found in the upper part of *Prunus* G1 correspond to two homoeologous groups of *Malus*, and the four remaining markers map to a different *Malus* linkage group. These results indicate that the long *Prunus* chromosome may be split into two in *Malus* or in the original species that formed the *Malus* amphidiploid.

Results of the diploid strawberry map of *F. vesca* × *F. nubicola* (Sargent et al., 2006) indirectly suggest that both genomes are highly conserved as markers mapped on their F_2 generate the expected seven linkage groups. Findings by Denoyes-Rothan et al. (2006) indicate that linkage groups found in the octoploid *F.* ×*ananassa* map can be aligned into seven homoeologous groups, each with four linkage groups. Each homoeologous group corresponds to one of the diploid *Fragaria* linkage groups. A study between diploid *Fragaria* and tetraploid *Rosa* suggests that synteny conservation is high, but chromosomal rearrangements may have occurred between these two genera (markers from two *Rosa* linkage groups are located in one *Fragaria* linkage group) (Rousseau et al., 2006).

A comparison between the diploid strawberry and peach genomes based on more than ten markers per *Fragaria* linkage group is currently in progress. Results suggest that these two distant genomes still conserve synteny, but have undergone reshuffling since their divergence from a common ancestor. Synteny is revealed by considering the eight *Prunus* linkage groups and noting that five contain most or all markers from only one group of *Fragaria* and the other three include markers of two groups. Thus, reshuffling must have occurred to a large extent with various fission/fusion, translocation, and inversion events.

The *Arabidopsis* sequence has been compared with the map positions of: a) RFLPs mapped in T × E, most of them detected with probes based on Rosaceae or *Arabidopsis* EST sequences (Dominguez et al., 2003); b) peach ESTs anchored to the *Prunus* consensus map (Jung et al., 2006); c) peach ESTs physically located in *Prunus* BAC contigs (Jung et al., 2006); d) complete sequence of one and partial sequences of two peach BAC clones (Georgi et al., 2003); and e) complete sequence of a single myrobolan plum (*P. cerasifera*) BAC clone (Claverie et al., 2004b). In all cases, syntenic regions can be found, but these are limited to small DNA fragments. This suggests an overall view of partial genome conservation. The largest *Prunus*-Arabidopsis conserved region identified by Dominguez et al. (2003) comprised 25 cM in LG2 of *Prunus* corresponding to 5.4 Mbp of Arabidopsis chromosome 5. These results suggest that comparative mapping for gene discovery outside of the family may not be feasible, underscoring the importance of complete genomic characterization of representative species in the family.

A novel marker concept based upon “gene pair” polymorphisms provides a promising approach to comparative genomics (Davis et al., 2008). This concept exploits the small genome sizes and the concomitantly small intergenic distances characteristic of rosaceous species. As

demonstrated in *F. vesca*, the commonality of small (2-5 kb) intergenic distances facilitates the amplification of gene pair PCR products in which a forward primer is targeted to conserved coding sequence in one gene and the reverse primer is similarly targeted in an adjacent gene. By mining the abundance of polymorphism in a respective intergenic region, gene pair PCR products yield conveniently detectable polymorphisms of multiple types, enabling the respective loci to be utilized as robust anchor loci for comparative mapping in the various rosaceous species (Davis et al., 2008).

Rosaceae bioinformatics and data integration

Several important bioinformatics resources have been developed for various rosaceous crops over the last few years. Sequence databases like ESTree db (www.itb.cnr.it/estree/) with EST information for peach (Lazzari et al., 2005), the *F. vesca* EST database (FVdbEST, http://www.vbi.vt.edu/~estap), and a candidate gene database (http://www.bioinfo.wsu.edu/gdr/projects/prunus/abbott/PP_LEa/) and transcript map for peach (Horn et al., 2005) provide important repositories for Rosaceae sequence information and have advanced genomics research in the family.

The "Plant Databases: A Needs Assessment White Paper" (Beavis et al., 2005) stressed that access to curated, integrated, and searchable genome databases is essential to fully leverage the wealth of genetic, genomic and molecular data generated worldwide by plant genome projects. Such data integration gateways have been created for many model species [i.e., The Arabidopsis Information Resource (TAIR) (Rhee et al., 2003), Oryzabase - An Integrated Biological and Genome Information Database for Rice (Kurata and Yamazaki, 2006) and MaizeGDB – the community database for maize genetics and genomics data (Lawrence et al., 2007)] and for plant families [i.e., Gramene – A Resource for Comparative Mapping in the Grasses (Jaiswal et al., 2006), SGN - the Solanaceae Genomics Network (Mueller et al., 2005), and LIS – the Legume Information System (Gonzales et al., 2005)].

The creation in 2003 of the federally funded Genome Database for Rosaceae (GDR) (www.bioinfo.wsu.edu/gdr) was an important step towards integrating all Rosaceae structural and functional genomics initiatives (Jung et al., 2004; Jung et al., 2008). Initiated in response to the growing availability of genomic data for peach, GDR now contains comprehensive data for the genetically anchored peach physical map, Rosaceae maps, markers and traits, and all publicly

available Rosaceae ESTs. The ESTs are assembled to produce unigene sets of each genus and the entire Rosaceae. Other annotations include putative function, microsatellites, open reading frames, single nucleotide polymorphisms, plant and gene ontology terms, and anchored map position where applicable. Most of the published Rosaceae genetic maps can be viewed and compared through CMap (Ware et al., 2002), the comparative map viewer. The peach physical map can be viewed using WebFPC/WebChrom and also through an integrated GDR map viewer, which provides a graphical interface to the combined genetic, transcriptome and physical mapping information categorized by taxonomy, tissue type, putative function and gene ontology. The GDR datasets are available to download or search in batch mode using web-based BLAST (Altschul et al., 1990) or FASTA (Pearson and Lipman, 1988) servers. Other web-based tools include a sequence assembly server using CAP3 (Huang and Madan, 1999) and an SSR server for identifying microsatellites and primers in sequence data sets.

The Rosaceae research community consistently uses GDR as a portal for disseminating and mining data, coordinating collaborations and workgroups, and sharing important news and publications. Usage figures show 218,409 visits and 2,070,880 pages accessed between August 1, 2006 and July 31, 2007. GDR will continue to curate, integrate and visualize new Rosaceae genomics and genetics data as they become available. These will include genetic maps, markers and traits, phenotype and genotype data, apple physical map, microarray data, and genome sequences from apple, peach, and strawberry.

Rosaceae community effort and the future of Rosaceae genomics

The Rosaceae Genomics Initiative (RosIGI, http://www.bioinfo.wsu.edu/gdr/community/international/) was established to link and coordinate cross-Rosaceae genomics efforts internationally. These include EST and genome sequencing, genetic and physical mapping, molecular marker discovery, development of a microarray gene expression analysis platform, forward and reverse genetic tools and high throughput gene validation**.** In the United States these efforts are coordinated by the US Rosaceae Genomics, Genetics, and Breeding Executive Committee (RosEXEC). Rather than selecting a single model and focusing all resources on developing a full array of resources and tools for it, the Rosaceae community adopted an alternative strategy of developing family-wide resources and leveraging the most appropriate system to maximize efficiency. This strategy has

been effective in other plant and microbial research communities (i.e., legume and *Phytophthora*). A white paper outlining long- and short-term strategies has been prepared by the community and is available at the GDR website (www.rosaceaewhitepaper.com).

Beyond the horizon

Genomics efforts in the Rosaceae family represent more than simply another set of organism sequences. The accelerating accumulation of genomics-level information in this particular family brings great advantages. Over the past five years, while various "revolutionary" genomics tools have come and gone, a validated set of proven microarray platforms, marker development methods and high-throughput cloning systems has been established. These proven tools are ripe for application in questions applicable to rosaceous plant biology and crop production. New high-throughput sequencing techniques, means for quantitative gene expression analyses and novel phenotyping platforms are in their infancy and will mature around economically useful species rather than model systems. The crop species within Rosaceae are well positioned to benefit from these emerging technologies.

Being a late arrival to the party has its advantages. The codified Rosaceae community can coalesce around standardized techniques, plant model systems, and computational tools residing in a central database, while strengthening comparative studies and fortifying meaningful meta-analyses. Most importantly, in commitment to the translational genomics paradigm, the Rosaceae genomics community is uniquely poised to incorporate breeders' and growers' input into genomics-level experimental design, bringing greater direct impact to basic science studies while resisting the allure of generating knowledge only for knowledge's sake. With versatile, relevant plant systems, emerging models and a concerted approach, ongoing studies in Rosaceae genomics will contribute to facets of basic plant science while bringing higher-quality products to the consumer with lower environmental impacts.

LITERATURE CITED

Aharoni A, Giri AP, Verstappen FWA, Bertea CM, Sevenier R, Sun ZK, Jongsma MA, Schwab W, Bouwmeester HJ (2004) Gain and loss of fruit flavor compounds produced by wild and cultivated strawberry species. Plant Cell **16:** 3110-3131

Aharoni A, Keizer LCP, Bouwmeester HJ, Sun ZK, Alvarez-Huerta M, Verhoeven HA, Blaas J, van Houwelingen A, De Vos RCH, van der Voet H, Jansen RC, Guis M, Mol J, Davis RW, Schena M, van Tunen AJ, O'Connell AP (2000) Identification of the *SAAT* gene involved in strawberry flavor biogenesis by use of DNA microarrays. Plant Cell **12:** 647-661

Aharoni A, Keizer LCP, Van den Broeck HC, Blanco-Portales R, Munoz-Blanco J, Bois G, Smit P, De Vos RCH, O'Connell AP (2002) Novel insight into vascular, stress, and auxin-dependent and -independent gene expression programs in strawberry, a non-climacteric fruit. Plant Physiol. **129:** 1019-1031

Aharoni A, O'Connell AP (2002) Gene expression analysis of strawberry achene and receptacle maturation using DNA microarrays. J. Exp. Bot. **53:** 2073-2087

Akiyama Y, Yamamoto Y, Ohmido N, Ohshima M, Fukui K (2001) Estimation of the nuclear DNA content of strawberries (*Fragaria* spp.) compared with *Arabidopsis thaliana* by using dual-step flow cytometry. Cytologia **66:** 431-436

Albani MC, Battey NH, Wilkinson MJ (2004) The development of ISSR-derived SCAR markers around the SEASONAL FLOWERING LOCUS (SFL) in *Fragaria vesca*. Theor. Appl. Genet. **109:** 571-579

Alsheikh MK, Suso HP, Robson M, Battey NH, Wetten A (2002) Appropriate choice of antibiotic and *Agrobacterium* strain improves transformation of anti biotic-sensitive *Fragaria vesca* and *F.v. semperflorens*. Plant Cell Rep. **20:** 1173-1180

Altschul SF, Gish W, Miller W, Myers EW, Lipman DJ (1990) Basic local alignment search tool. J. Mol. Biol. **215:** 403-410

Aranzana MJ, Pineda A, Cosson P, Dirlewanger E, Ascasibar J, Cipriani G, Ryder CD, Testolin R, Abbott A, King GJ, Iezzoni AF, Arús P (2003) A set of simple-sequence repeat (SSR) markers covering the *Prunus* genome. Theor. Appl. Genet. **106:** 819-825

Arús P, Howad W, Mnejja M (2003) Marker development and marker-assisted selection in temperate fruit trees. *In* R Tuberosa, RL Phillips, M Gale, eds, Proc. Intern. Congr. *In the wake of the double helix: from the green revolution to the gene revolution*. Avenue Media, Bologna, Italy, pp 309-325

Baird WV, Estager AS, Wells JK (1994) Estimating nuclear DNA content in peach and related diploid species using laser flow cytometry and DNA hybridization. J. Am. Soc. Hort. Sci. **119:** 1312-1316

Baldi P, Patocchi A, Zini E, Toller C, Velasco R, Komjanc M (2004) Cloning and linkage mapping of resistance gene homologues in apple. Theor. Appl. Genet. **109:** 231-239

Ban Y, Honda C, Hatsuyama Y, Igarashi M, Bessho H, Moriguchi T (2007) Isolation and functional analysis of a *MYB* transcription factor gene that is a key regulator for the development of red coloration in apple skin. Plant Cell Physiol. **48:** 958-970

Beavis W, Gessler D, Rhee S, Rokhsar D, Main D, Mueller L, Eva Huala E, Stein L, Lawrence CJ (2005) Plant Biology Databases: A Needs Assessment - White Paper. *In* http://www.gramene.org/resources/plant_databases.pdf,

Blenda AV, Wechter WP, Reighard GL, Baird WV, Abbott AG (2006) Development and characterisation of diagnostic AFLP markers in *Prunus persica* for its response to peach tree short life syndrome. J. Hort. Sci. Biotechnol. **81:** 281-288

Bliss FA, Arulsekar S, Foolad MR, Becerra V, Gillen AM, Warburton ML, Dandekar AM, Kocsisne GM, Mydin KK (2002) An expanded genetic linkage map of *Prunus* based on an interspecific cross between almond and peach. Genome **45:** 520-529

Bolar JP, Norelli JL, Harman GE, Brown SK, Aldwinckle HS (2001) Synergistic activity of endochitinase and exochitinase from *Trichoderma atroviride* (*T. harzianum*) against the pathogenic fungus (*Venturia inaequalis*) in transgenic apple plants. Transgenic Res. **10:** 533-543

Bolar JP, Norelli JL, Wong KW, Hayes CK, Harman GE, Aldwinckle HS (2000) Expression of endochitinase from *Trichoderma harzianum* in transgenic apple increases resistance to apple scab and reduces vigor. Phytopathology **90:** 72-77

Bringhurst RS (1990) Cytogenetics and evolution in American *Fragaria*. HortScience **25:** 879-881

Bus V, Ranatunga C, Gardiner S, Bassett H, Rikkerink E (2000) Marker assisted selection for pest and disease resistance in the New Zealand apple breeding programme. Acta Hort. **538:** 541-547

Byrne DH (1990) Isozyme variability in four diploid stone fruits compared with other woody perennial plants. J. Hered. **81:** 68-71

Calenge F, Van der Linden CG, Van de Weg E, Schouten HJ, Van Arkel G, Denance C, Durel CE (2005) Resistance gene analogues identified through the NBS-profiling method map close to major genes and QTL for disease resistance in apple. Theor. Appl. Genet. **110:** 660-668

Caragay AB (1992) Cancer-preventive foods and ingredients. Food Technology **46:** 65-68

Chagné D, Carlisle CM, Blond C, Volz RK, Whitworth CJ, Oraguzie NC, Crowhurst RN, Allan AC, Espley RV, Hellens RP, Gardiner SE (2007) Mapping a candidate gene (*MdMYB10*) for red flesh and foliage colour in apple. BMC Genomics **8**

Chaparro JX, Werner DJ, Omalley D, Sederoff RR (1994) Targeted mapping and linkage analysis of morphological isozyme, and RAPD markers in peach. Theor. Appl. Genet. **87:** 805-815

Cipriani G, Pinosa F, Bonoli M, Faedi W (2006) A new set of microsatellite markers for *Fragaria* species and their application in linkage analysis. J. Hort. Sci. Biotechnol. **81:** 668-675

Claverie M, Dirlewanger E, Cosson P, Bosselut N, Lecouls AC, Voisin R, Kleinhentz M, Lafargue B, Caboche M, Chalhoub B, Esmenjaud D (2004b) High-resolution mapping and chromosome landing at the root-knot nematode resistance locus *Ma* from Myrobalan plum using a large-insert BAC DNA library. Theor. Appl. Genet. **109:** 1318-1327

Cordenunsi BR, do Nascimento JRO, Genovese MI, Lajolo FM (2002) Influence of cultivar on quality parameters and chemical composition of strawberry fruits grown in Brazil. J. Agric. Food Chem. **50:** 2581-2586

Corredor E, Roman M, Garcia E, Perera E, Arús P, Naranjo T (2004) Physical mapping of rDNA genes establishes the karyotype of almond. Ann. Appl. Biol. **144:** 219-222

Darrow GM (1966) The strawberry: history, breeding and physiology. Holt, Rinehart and Winston, New York

Davis TM, Denoyes-Rothan B, Lerceteau-Kohler E (2007) Strawberry. *In* C Kole, ed, Genome Mapping & Molecular Breeding in Plants IV: Fruits and Nuts. Springer, Berlin, pp 189-206

Davis TM, Folta KM, Shields M, Zhang Q (2007) Gene pair markers: An innovative tool for comparative linkage mapping. *In* Proc. 6th North American Strawberry Symp., p (In Press)

Davis TM, Folta KM, Shields M, Zhang Q (2008) Gene pair markers: An innovative tool for comparative linkage mapping. *In* Proc. 6th North American Strawberry Symp., p (In Press)

Davis TM, Yu H (1997) A linkage map of the diploid strawberry, *Fragaria vesca*. J. Hered. **88:** 215-221

Debener T, Mattiesch L (1999) Construction of a genetic linkage map for roses using RAPD and AFLP markers. Theor. Appl. Genet. **99:** 891-899

Debener T, Mattiesch L, Vosman B (2001) A molecular marker map for roses. Proceedings of the Third International Symposium on Rose Research and Cultivation**:** 283-287

Defilippi BG, Dandekar AM, Kader AA (2004) Impact of suppression of ethylene action or biosynthesis on flavor metabolites in apple (*Malus domestica* Borkh) fruits. J. Agric. Food Chem. **52:** 5694-5701

Defilippi BG, Kader AA, Dandekar AM (2005) Apple aroma: alcohol acyltransferase, a rate limiting step for ester biosynthesis, is regulated by ethylene. Plant Sci. **168:** 1199-1210

Denoyes-Rothan B, Rousseau BM, Barrot L, Lerceteau-Köhler E, Sargent D, Simpson D, Monfort A, Arús P, Guerin G (2006) Comparing the linkage maps of two levels of ploidy in *Fragaria*. *In* 3rd International Rosaceae Genomics Conference, 19-22 March 2006, Napier, New Zealand

Dettori MT, Quarta R, Verde I (2001) A peach linkage map integrating RFLPs, SSRs, RAPDs, and morphological markers. Genome **44:** 783-790

Dirlewanger E, Cosson P, Bouderhi K, Renaud C, Capdeville G, Tauzin Y, Laigret F, Moing A (2006) Development of a second-generation genetic linkage map for peach [*Prunus persica* (L.) Batsch] and characterization of morphological traits affecting flower and fruit. Tree Genet Genomes **3:** 1-13

Dirlewanger E, Cosson P, Howad W, Capdeville G, Bosselut N, Claverie M, Voisin R, Poizat C, Lafargue B, Baron O, Laigret F, Kleinhentz M, Arús P, Esmenjaud D (2004) Microsatellite genetic linkage maps of myrobalan plum and an almond-peach hybrid - location of root-knot nematode resistance genes. Theor. Appl. Genet. **109:** 827-838

Dirlewanger E, Cosson P, Howad W, Capdeville G, Bosselut N, Claverie M, Voisin R, Poizat C, Lafargue B, Baron O, Laigret F, Kleinhentz M, Arús P, Esmenjaud D (2004a) Microsatellite genetic linkage maps of myrobalan plum and an almond-peach hybrid - location of root-knot nematode resistance genes. Theor. Appl. Genet. **109:** 827-838

Dirlewanger E, Cosson P, Tavaud M, Aranzana MJ, Poizat C, Zanetto A, Arús P, Laigret F (2002) Development of microsatellite markers in peach [*Prunus persica* (L.) Batsch] and their use in genetic diversity analysis in peach and sweet cherry (*Prunus avium* L.). Theor. Appl. Genet. **105:** 127-138

Dirlewanger E, Graziano E, Joobeur T, Garriga-Caldere F, Cosson P, Howad W, Arús P (2004b) Comparative mapping and marker-assisted selection in Rosaceae fruit crops. Proc. Natl. Acad. Sci. U. S. A. **101:** 9891-9896

Dominguez I, Graziano E, Gebhardt C, Barakat A, Berry S, Arús P, Delseny M, Barnes S (2003) Plant genome archaeology: evidence for conserved ancestral chromosome segments in dicotyledonous plant species. Plant Biotechnol J **1:** 91-99

Dondini L, Lain O, Geuna F, Banfi R, Gaiotti F, Tartarini S, Bassi D, Testolin R (2007) Development of a new SSR-based linkage map in apricot and analysis of synteny with existing *Prunus* maps. Tree Genet. Genomes **3:** 239-249

Dondini L, Pierantoni L, Gaiotti F, Chiondini R, Tartarini S, Bazzi C, Sansavini S (2004) Identifying QTLs for fire-blight resistance via a European pear (*Pyrus communis* L.) genetic linkage map. Mol. Breed. **14:** 407-418

Eichholzer M, Luthy J, Gutzwiller F, Stahelin HB (2001) The role of folate, antioxidant vitamins and other constitutents in fruit and vegetables in the prevention of cardiovascular disease: The epidemiological evidence. Int. J. Vit. Nutri. Res. **71:** 5-17

El Mansouri I, Mercado JA, Valpuesta V, Lopez-Aranda JM, Pliego-Alfaro F, Quesada MA (1996) Shoot regeneration and *Agrobacterium*-mediated transformation of *Fragaria vesca* L. Plant Cell Rep. **15:** 642-646

Escobar MA, Civerolo EL, Polito VS, Pinney KA, Dandekar AM (2003) Characterization of oncogene-silenced transgenic plants: implications for Agrobacterium biology and post-transcriptional gene silencing. Mol. Plant Pathol. **4:** 57-65

Espley RV, Hellens RP, Putterill J, Stevenson DE, Kutty-Amma S, Allan AC (2007) Red colouration in apple fruit is due to the activity of the *MYB* transcription factor, *MdMYB10*. Plant J. **49:** 414-427

Etienne C, Rothan C, Moing A, Plomion C, Bodenes C, Svanella-Dumas L, Cosson P, Pronier V, Monet R, Dirlewanger E (2002) Candidate genes and QTLs for sugar and organic acid content in peach [*Prunus persica* (L.) Batsch]. Theor. Appl. Genet. **105:** 145-159

Folta KM, Davis TM (2006) Strawberry genes and genomics. Crit. Rev. Plant Sci. **25:** 399-415

Folta KM, Dhingra A (2006) Transformation of strawberry: The basis for translational genomics in Rosaceae. In Vitro Cell. Dev. Biol. Plant **42:** 482-490

Foolad MR, Arulsekar S, Becerra V, Bliss FA (1995) A genetic map of *Prunus* based on an interspecific cross between peach and almond. Theor. Appl. Genet. **91:** 262-269

Foulongne M, Pascal T, Arús P, Kervella J (2003) The potential of *Prunus davidiana* for introgression into peach [*Prunus persica* (L.) Batsch] assessed by comparative mapping. Theor. Appl. Genet. **107:** 227-238

Gardiner S, Bus V, Rusholme R, Chagné D, Rikkerink E (2007) Apple. *In* C Kole, ed, Genome Mapping and Molecular Breeding, Vol 4. Springer-Verlag, Heidelberg, pp 1-62

Gasic K, Gonzales DO, Thimmapuram J, Malnoy M, Vodkin LO, Liu L, Aldwinckle HS, Carroll N, Orvis K, Goldsbrough P, Clifton S, Fulton L, Hou S, Korban SS (2007) Comparative genomic analysis of a large apple EST collection. *In* Plant & Animal Genome Conference XV, Vol P-226, San Diego, p 158

Georgi LL, Wang Y, Reighard GL, Mao L, Wing RA, Abbott AG (2003) Comparison of peach and Arabidopsis genomic sequences: fragmentary conservation of gene neighborhoods. Genome **46:** 268-276

Georgi LL, Wang Y, Yvergniaux D, Ormsbee T, Inigo M, Reighard G, Abbott AG (2002) Construction of a BAC library and its application to the identification of simple sequence repeats in peach [*Prunus persica* (L.) Batsch]. Theor. Appl. Genet. **105:** 1151-1158

Gleave AP, Ampomah-Dwamena C, Berthold S, Dejnoprat S, Karunairetnam S, Nain B, Wang YY, Crowhurst RN, MacDiarmid RM (2008) Identification and characterisation of primary microRNAs from apple (*Malus domestica* cv. Royal Gala) expressed sequence tags. Tree Genet. Genomes **4:** 343-358

Gonzales MD, Archuleta E, Farmer A, Gajendran K, Grant D, Shoemaker R, Beavis WD, Waugh ME (2005) The Legume Information System (LIS): an integrated information resource for comparative legume biology. Nucleic Acids Res. **33:** D660-D665

Graham J, Smith K, MacKenzie K, Jorgenson L, Hackett C, Powell W (2004) The construction of a genetic linkage map of red raspberry (*Rubus idaeus* subsp. *idaeus*) based on AFLPs, genomic-SSR and EST-SSR markers. Theor. Appl. Genet. **109:** 740-749

Guterman I, Shalit M, Menda N, Piestun D, Dafny-Yelin M, Shalev G, Bar E, Davydov O, Ovadis M, Emanuel M, Wang J, Adam Z, Pichersky E, Lewinsohn E, Zamir D, Vainstein A, Weiss D (2002) Rose scent: Genomics approach to discovering novel floral fragrance-related genes. Plant Cell **14:** 2325-2338

Habu T, Kishida H, Morikita M, Kitajima A, Yamada T, Tao R (2006) A simple and rapid procedure for the detection of self-compatible individuals in Japanese apricot (*Prunus mume* Sieb. et Zucc.) using the loop-mediated isothermal amplification (LAMP) method. HortScience **41:** 1156-1158

Haffner K, Vestrheim S (1997) Fruit quality of strawberry cultivars. Acta Hort. **439:** 325-332

Hakkinen SH, Karenlampi SO, Heinonen IM, Mykkanen HM, Torronen AR (1998) HPLC method for screening of flavonoids and phenolic acids in berries. J. Sci. Food. Agric. **77:** 543-551

Hakkinen SH, Torronen AR (2000) Content of flavonols and selected phenolic acids in strawberries and *Vaccinium* species: influence of cultivar, cultivation site and technique. Food Res. Int. **33:** 517-524

Han Y, Gasic K, Marron B, Beever JE, Korban SS (2007) A BAC-based physical map of the apple genome. Genomics **89:** 630-637

Harris GK, Gupta A, Nines RG, Kresty LA, Habib SG, Frankel WL, LaPerle K, Gallaher DD, Schwartz SJ, Stoner GD (2001) Effects of lyophilized black raspberries on azoxymethane-induced colon cancer and 8-hydroxy-2 '-deoxyguanosine levels in the Fischer 344 rat. Nutr. Cancer **40:** 125-133

Haymes KM, Davis TM (1998) *Agrobacterium* mediated transformation of 'Alpine' *Fragaria vesca*, and transmission of transgenes to R_1 progeny. Plant Cell Rep. **17:** 279-283

Haymes KM, Van de Weg WE, Arens P, Maas JL, Vosman B, Den Nijs APM (2000) Development of SCAR markers linked to a *Phytophthora fragariae* resistance gene and their assessment in European and North American strawberry genotypes. J. Am. Soc. Hort. Sci. **125:** 330-339

Hemmat M, Weeden NF, Brown SK (2003) Mapping and evaluation of *Malus* ×*domestica* microsatellites in apple and pear. J. Am. Soc. Hort. Sci. **128:** 515-520

Hemmat M, Weeden NF, Manganaris AG, Lawson DM (1994) Molecular marker linkage map for apple. J. Hered. **85:** 4-11

Hokanson SC (2001) SNiPs, chips, BACs, and YACs: Are small fruits part of the party mix? HortScience **36:** 859-871

Horn R, Lecouls AC, Callahan A, Dandekar A, Garay L, McCord P, Howad W, Chan H, Verde I, Main D, Jung S, Georgi L, Forrest S, Mook J, Zhebentyayeva T, Yu YS, Kim HR, Jesudurai C, Sosinski B, Arús P, Baird V, Parfitt D, Reighard G, Scorza R, Tomkins J, Wing R, Abbott AG (2005) Candidate gene database and transcript map for peach, a model species for fruit trees. Theor. Appl. Genet. **110:** 1419-1428

Howad W, Yamamoto T, Dirlewanger E, Testolin R, Cosson P, Cipriani G, Monforte AJ, Georgi L, Abbott AG, Arús P (2005) Mapping with a few plants: Using selective mapping for microsatellite saturation of the *Prunus* reference map. Genetics **171:** 1305-1309

Huang X, Madan A (1999) CAP3: A DNA sequence assembly program. Genome Res **9:** 868-877

Huaracha E, Xu M, Korban SS (2004) Narrowing down the region of the *Vf* locus for scab resistance in apple using AFLP-derived SCARs. Theor. Appl. Genet. **108:** 274-279

Hurtado MA, Romero C, Vilanova S, Abbott AG, Llacer G, Badenes ML (2002) Genetic linkage maps of two apricot cultivars (*Prunus armeniaca* L.), and mapping of PPV (sharka) resistance. Theor. Appl. Genet. **105:** 182-191

Jaiswal P, Ni J, Yap I, Ware D, Spooner W, Youens-Clark K, Ren L, Liang C, Zhao W, Ratnapu K, Faga B, Canaran P, Fogleman M, Hebbard C, Avraham S, Schmidt S, Casstevens TM, Buckler ES, Stein L, McCouch S (2006) Gramene: a bird's eye view of cereal genomes. Nucleic Acids Res. **34:** D717-723

Janick J, Cummins JN, Brown SK, Hemmat M (1996) Apples. *In* J Janick, ed, Fruit breeding. Volume I. Tree and tropical fruits, Vol I. John Wiley and Sons, New York

Jauregui B, de Vicente MC, Messeguer R, Felipe A, Bonnet A, Salesses G, Arús P (2001) A reciprocal translocation between 'Garfi' almond and 'Nemared' peach. Theor. Appl. Genet. **102:** 1169-1176

Jelenkovic G, Harrington E (1972) Morphology of the pachytene chromosomes in *Prunus persica*. Can. J. Genet. Cytol. **14:** 317-324

Joobeur T, Periam N, Vicente MC, King GJ, Arús P (2000) Development of a second generation linkage map for almond using RAPD and SSR markers. Genome **43:** 649-655

Joobeur T, Viruel MA, de Vicente MC, Jauregui B, Ballester J, Dettori MT, Verde I, Truco MJ, Messeguer R, Batlle I, Quarta R, Dirlewanger E, Arús P (1998) Construction of a saturated linkage map for *Prunus* using an almond x peach F_2 progeny. Theor. Appl. Genet. **97:** 1034-1041

Jung S, Jesudurai C, Staton M, Du ZD, Ficklin S, Cho IH, Abbott A, Tomkins J, Main D (2004) GDR (Genome Database for Rosaceae): integrated web resources for Rosaceae genomics and genetics research. BMC Bioinformatics **5**

Jung S, Main D, Staton M, Cho I, Zhebentyayeva T, Arús P, Abbott A (2006) Synteny conservation between the Prunus genome and both the present and ancestral Arabidopsis genomes. BMC Genomics **7**

Jung S, Staton M, Lee T, Blenda A, Svancara R, Abbott A, Main D (2008) GDR (Genome Database for Rosaceae): integrated web-database for Rosaceae genomics and genetics data. Nucleic Acids Res. **36:** D1034-D1040

Kardailsky I, Shukla VK, Ahn JH, Dagenais N, Christensen SK, Nguyen JT, Chory J, Harrison MJ, Weigel D (1999) Activation tagging of the floral inducer FT. Science **286:** 1962-1965

Kaufmann H, Mattiesch L, Lorz H, Debener T (2003) Construction of a BAC library of *Rosa rugosa* Thunb. and assembly of a contig spanning *Rdr1*, a gene that confers resistance to blackspot. Mol. Genet. Genomics **268:** 666-674

Kenis K, Keulemans J (2005) Genetic linkage maps of two apple cultivars (*Malus* ×*domestica* Borkh.) based on AFLP and microsatellite markers. Mol. Breed. **15:** 205-219

Kishore GM, Shewmaker C (1999) Biotechnology: Enhancing human nutrition in developing and developed worlds. Proc. Natl. Acad. Sci. U. S. A. **96:** 5968-5972

Ko K, Brown SK, Norelli JL, Aldwinckle HS (1998) Alterations in *nptII* and *gus* expression following micropropagation of transgenic M7 apple rootstock lines. J. Am. Soc. Hort. Sci. **123:** 11-18

Ko KS, Norelli JL, Reynoird JP, Boresjza-Wysocka E, Brown SK, Aldwinckle HS (2000) Effect of untranslated leader sequence of AMV RNA 4 and signal peptide of pathogenesis-related protein 1b on attacin gene expression, and resistance to fire blight in transgenic apple. Biotechnol. Lett. **22:** 373-381

Korban SS, Chen H (1992) Apples. *In* F Hammerschlag, R Litz, eds, Biotechnology of Fruit Tree Crops. C.A.B. Intl, Oxnard, CA, pp 203-227

Krysan PJ, Young JC, Sussman MR (1999) T-DNA as an insertional mutagen in Arabidopsis. Plant Cell **11:** 2283-2290

Kurata N, Yamazaki Y (2006) Oryzabase. An integrated biological and genome information database for rice. Plant Physiol. **140:** 12-17

Lalli DA, Decroocq V, Blenda AV, Schurdi-Levraud V, Garay L, Le Gall O, Damsteegt V, Reighard GL, Abbott AG (2005) Identification and mapping of resistance gene analogs (RGAs) in *Prunus*: a resistance map for *Prunus.* Theor. Appl. Genet. **111:** 1504-1513

Lambert P, Hagen LS, Arús P, Audergon JM (2004) Genetic linkage maps of two apricot cultivars (*Prunus armeniaca* L.) compared with the almond Texas x peach Earlygold reference map for Prunus. Theor. Appl. Genet. **108:** 1120-1130

Lavid N, Wang JH, Shalit M, Guterman I, Bar E, Beuerle T, Menda N, Shafir S, Zamir D, Adam Z, Vainstein A, Weiss D, Pichersky E, Lewinsohn E (2002) O-methyltransferases involved in the biosynthesis of volatile phenolic derivatives in rose petals. Plant Physiol. **129:** 1899-1907

Lawrence CJ, Schaeffer ML, Seigfried TE, Campbell DA, Harper LC (2007) MaizeGDB's new data types, resources and activities. Nucleic Acids Res. **35:** D895-900

Lazzari B, Caprera A, Vecchietti A, Stella A, Milanesi L, Pozzi C (2005) ESTree db: a tool for peach functional genomics. BMC Bioinformatics **6:** Article no. S16

Lee YP, Yu GH, Seo YS, Han SE, Choi YO, Kim D, Mok IG, Kim WT, Sung SK (2007) Microarray analysis of apple gene expression engaged in early fruit development. Plant Cell Rep. **26:** 917-926

Lerceteau-Kohler E, Guerin G, Laigret F, Denoyes-Rothan B (2003) Characterization of mixed disomic and polysomic inheritance in the octoploid strawberry (*Fragaria* x*ananassa*) using AFLP mapping. Theor. Appl. Genet. **107:** 619-628

Liebhard R, Gianfranceschi L, Koller B, Ryder CD, Tarchini R, Van de Weg E, Gessler C (2002) Development and characterisation of 140 new microsatellites in apple (*Malus* ×*domestica* Borkh.). Mol. Breed. **10:** 217-241

Liebhard R, Kellerhals M, Pfammatter W, Jertmini M, Gessler C (2003a) Mapping quantitative physiological traits in apple (*Malus* x*domestica* Borkh.). Plant Mol. Biol. **52:** 511-526

Liebhard R, Koller B, Gianfranceschi L, Gessler C (2003c) Creating a saturated reference map for the apple (*Malus* ×*domestica* Borkh.) genome. Theor. Appl. Genet. **106:** 1497-1508

Liebhard R, Koller B, Patocchi A, Kellerhals M, Pfammatter W, Jermini M, Gessler C (2003b) Mapping quantitative field resistance against apple scab in a 'Fiesta' x 'Discovery' progeny. Phytopathology **93:** 493-501

López M, Vargas FJ, Batlle I (2006) Self-(in)compatibility almond genotypes: A review. Euphytica **150:** 1-16

Lu ZX, Sosinski B, Reighard GL, Baird WV, Abbott AG (1998) Construction of a genetic linkage map and identification of AFLP markers for resistance to root-knot nematodes in peach rootstocks. Genome **41:** 199-207

Macheix JJ, Sapis JC, Fleuriet A (1991) Phenolic compounds and polyphenoloxidase in relation to browning in grapes and wines. Crit. Rev. Food Sci. Nutr. **30:** 441-486

Maliepaard C, Alston FH, van Arkel G, Brown LM, Chevreau E, Dunemann F, Evans KM, Gardiner S, Guilford P, van Heusden AW, Janse J, Laurens F, Lynn JR, Manganaris AG, den Nijs APM, Periam N, Rikkerink E, Roche P, Ryder C, Sansavini S, Schmidt H, Tartarini S, Verhaegh JJ, Vrielink-van Ginkel M, King GJ (1998) Aligning male and female linkage maps of apple (*Malus pumila* Mill.) using multi-allelic markers. Theor. Appl. Genet. **97:** 60-73

Mandal S, Stoner GD (1990) Inhibition of N-nitrosobenzylmethylamine-induced esophageal tumorigenesis in rats by ellagic acid. Carcinogenesis **11:** 55-61

Marshall RE (1954) Cherries and cherry products, Vol Vol. 5. Interscience Publishers Ltd., New York and London

Masuda T, Yonemori S, Oyama Y, Takeda Y, Tanaka T, Andoh T, Shinohara A, Nakata M (1999) Evaluation of the antioxidant activity of environmental plants: Activity of the leaf extracts from seashore plants. J. Agric. Food Chem. **47:** 1749-1754

Mazur WM, Uehara M, Wahala K, Adlercreutz H (2000) Phyto-oestrogen content of berries, and plasma concentrationsand urinary excretion of enterolactone after asingle strawberry-meal in human subjects. Br. J. Nutr. **83:** 381-387

Mezzetti B, Costantini E (2006) Strawberry (*Fragaria* ×*ananassa*). Methods Mol. Biol. **344:** 287-295

Miller PJ, Parfitt DE, Weinbaum SA (1989) Outcrossing in peach. HortScience **24:** 359-360

Mueller LA, Solow TH, Taylor N, Skwarecki B, Buels R, Binns J, Lin C, Wright MH, Ahrens R, Wang Y, Herbst EV, Keyder ER, Menda N, Zamir D, Tanksley SD (2005) The SOL Genomics Network: a comparative resource for Solanaceae biology and beyond. Plant Physiol. **138:** 1310-1317

Nagaraj SH, Gasser RB, Ranganathan S (2007) A hitchhiker's guide to expressed sequence tag (EST) analysis. Brief. Bioinformatics **8:** 6-21

Naik S, Hampson C, Gasic K, Bakkeren G, Korban SS (2006) Development and linkage mapping of E-STS and RGA markers for functional gene homologues in apple. Genome **49:** 959-968

Newcomb RD, Crowhurst RN, Gleave AP, Rikkerink EHA, Allan AC, Beuning LL, Bowen JH, Gera E, Jamieson KR, Janssen BJ, Laing WA, McArtney S, Nain B, Ross GS,

Snowden KC, Souleyre EJF, Walton EF, Yauk YK (2006) Analyses of expressed sequence tags from apple. Plant Physiol. **141:** 147-166

Nier S, Simpson DW, Tobutt KR, Sargent DJ (2006) A genetic linkage map of an inter-specific diploid *Fragaria* BC_1 mapping population and its comparison with the *Fragaria reference* map (FV X FN). J. Hort. Sci. Biotechnol. **81:** 645-650

Norelli JL, Borejsza-Wysocka E, Baldo AM, Aldwinckle HS, Bassett CL, Farrell RE, Malnoy M, Lalli DA, Korban SS, Gasic K, Wisniewski ME (2007) Functional genomic analysis of apple (*Malus*) ESTs associated with fire blight (*Erwinia amylovora*). Phytopathology **97:** S185-S185

Oosumi T, Gruszewski HA, Blischak LA, Baxter AJ, Wadl PA, Shuman JL, Veilleux RE, Shulaev V (2006) High-efficiency transformation of the diploid strawberry (*Fragaria vesca*) for functional genomics. Planta **223:** 1219-1230

Park S, Sugimoto N, Larson MD, Beaudry R, van Nocker S (2006) Identification of genes with potential roles in apple fruit development and biochemistry through large-scale statistical analysis of expressed sequence tags. Plant Physiol. **141:** 811-824

Pearson WR, Lipman DJ (1988) Improved tools for biological sequence comparison. Proc. Natl. Acad. Sci. U. S. A. **85:** 2444-2448

Pedreschi R, Vanstreels E, Carpentier S, Hertog M, Lammertyn J, Robben J, Noben JP, Swennen R, Vanderleyden J, Nicolai BM (2007) Proteomic analysis of core breakdown disorder in Conference pears (*Pyrus communis* L.). Proteomics **7:** 2083-2099

Pichler FB, Walton EF, Davy M, Triggs C, Janssen B, Wunsche JN, Putterill J, Schaffer RJ (2007) Relative developmental, environmental, and tree-to-tree variability in buds from field-grown apple trees. Tree Genet. Genomes **3:** 329-339

Pierantoni L, Cho K-H, Shin I-S, Chiodini R, Tartarini S, Dondini L, Kang S-J, Sansavini S (2004) Characterisation and transferability of apple SSRs to two European pear F_1 populations. Theor. Appl. Genet. **109:** 1519-1524

Pooler MR, Scorza R (1997) Irradiation and heat affect peach pollen germination and fertility. HortScience **32:** 290-291

Potter D, Eriksson T, Evans RC, Oh S, Smedmark JEE, Morgan DR, Kerr M, Robertson KR, Arsenault M, Dickinson TA, Campbell CS (2007) Phylogeny and classification of Rosaceae. Plant Syst. Evol. **266:** 5-43

Potter D, Gao F, Bortiri PE, Oh SH, Baggett S (2002) Phylogenetic relationships in Rosaceae inferred from chloroplast *matK* and *trnL-trnF* nucleotide sequence data. Plant Syst. Evol. **231:** 77-89

Quilot B, Wu BH, Kervella J, Genard M, Foulongne M, Moreau K (2004) QTL analysis of quality traits in an advanced backcross between *Prunus persica* cultivars and the wild relative species *P. davidiana*. Theor. Appl. Genet. **109:** 884-897

Rajapakse S, Byrne DH, Zhang L, Anderson N, Arumuganathan K, Ballard RE (2001) Two genetic linkage maps of tetraploid roses. Theor. Appl. Genet. **103:** 575-583

Renaut J, Hausman J-F, Bassett C, Artlip T, Cauchie H-M, Witters E, Wisniewski M (2008) Quantitative proteomic analysis of short photoperiod and low-temperature responses in bark tissues of peach (*Prunus persica* L. Batsch). Tree Genet Genomes **DOI 10:** 1007/s11295-11008-10134-11294

Renaut J, Hausman JF, Wisniewski ME (2006) Proteomics and low-temperature studies: bridging the gap between gene expression and metabolism. Physiol. Plant. **126:** 97-109

Reuter A, Fortunato D, Garoffo LP, Napolitano L, Scheurer S, Giuffrida MG, Vieths S, Conti AD (2005) Novel isoforms of *Pru av 1* with diverging immunoglobulin E binding properties identified by a synergistic combination of molecular biology and proteomics. Proteomics **5:** 282-289

Rhee SY, Beavis W, Berardini TZ, Chen G, Dixon D, Doyle A, Garcia-Hernandez M, Huala E, Lander G, Montoya M, Miller N, Mueller LA, Mundodi S, Reiser L, Tacklind J, Weems DC, Wu Y, Xu I, Yoo D, Yoon J, Zhang P (2003) The Arabidopsis Information Resource (TAIR): a model organism database providing a centralized, curated gateway to Arabidopsis biology, research materials and community. Nucleic Acids Res. **31:** 224-228

Rousseau M, Hibrand Saint Oyant L, Foucher F, Barrot L, Lalanne D, Sargent D, Simpson D, Laigret F, Denoyes-Rothan B (2006) Comparative mapping in the Rosoideae tribe: Rosa and Fragaria. . *In* 3rd International Rosaceae Genomics Conference, 19-22 March 2006, Napier

Rudell DR, Mattheis JP, Curry FA (2008) Prestorage ultraviolet-white light irradation alters apple peel metabolome. J. Agric. Food Chem. **56:** 1138-1147

Salesses G, Mouras A (1977) Use of protoplasts for *Prunus* chromosomes study. Ann. Amelior. Plant. **27:** 363-368

Sargent DJ, Clarke J, Simpson DW, Tobutt KR, Arús P, Monfort A, Vilanova S, Denoyes-Rothan B, Rousseau M, Folta KM, Bassil NV, Battey NH (2006) An enhanced microsatellite map of diploid *Fragaria*. Theor. Appl. Genet. **112:** 1349-1359

Sargent DJ, Davis TM, Tobutt KR, Wilkinson MJ, Battey NH, Simpson DW (2004) A genetic linkage map of microsatellite, gene-specific and morphological markers in diploid *Fragaria*. Theor. Appl. Genet. **109:** 1385-1391

Sargent DJ, Rys A, Nier S, Simpson DW, Tobutt KR (2007) The development and mapping of functional markers in *Fragaria* and their transferability and potential for mapping in other genera. Theor. Appl. Genet. **114:** 373-384

Scalliet G, Lionnet C, Le Bechec M, Dutron L, Magnard JL, Baudino S, Bergougnoux V, Jullien F, Chambrier P, Vergne P, Dumas C, Cock JM, Hugueney P (2006) Role of petal-specific orcinol O-methyltransferases in the evolution of rose scent. Plant Physiol. **140:** 18-29

Schaffer RJ, Friel EN, Souleyre EJF, Bolitho K, Thodey K, Ledger S, Bowen JH, Ma JH, Nain B, Cohen D, Gleave AP, Crowhurst RN, Janssen BJ, Yao JL, Newcomb RD (2007) A Genomics approach reveals that aroma production in apple is controlled by ethylene predominantly at the final step in each biosynthetic pathway([w]). Plant Physiol. **144:** 1899-1912

Schieber A, Stintzing FC, Carle R (2001) By-products of plant food processing as a source of functional compounds - recent developments. Trends Food Sci Technol. **12:** 401-413

Schwab W, Aharoni A, Raab T, Perez AG, Sanz C (2001) Cytosolic aldolase is a ripening related enzyme in strawberry fruits (*Fragaria* x *ananassa*). Phytochemistry **56:** 407-415

Scorza R, Mehlenbacher SA, Lightner GW (1985) Inbreeding and coancestry of freestone peach cultivars of the eastern United States and implications for peach germplasm improvement. J. Am. Soc. Hort. Sci. **110:** 547-552

Scorza R, Sherman WB (1996) Peaches. *In* J Janick, JN Moore, eds, Fruit breeding. Volume I. Tree and tropical fruits. John Wiley and Sons, New York

Selmar D (1999) Cyanide in foods: Biology of cyanogenic glucosides and related nutritional problems. *In* JT Romeo, ed, Phytochemicals in human health protection, nutrition, and plant defense, Vol 33 Kluwer Academic, New York, pp 369-392

Silfverberg-Dilworth E, Matasci CL, Van de Weg WE, Van Kaauwen MPW, Walser M, Kodde LP, Soglio V, Gianfranceschi L, Durel CE, Costa F, Yamamoto T, Koller B, Gessler C, Patocchi A (2006) Microsatellite markers spanning the apple (*Malus* ×*domestica* Borkh.) genome. Tree Genet. Genomes **2:** 202-224

Silfverberg-Dilworth E, Matasci CL, Van de Weg WE, Van Kaauwen MPW, Walser M, Kodde LP, Soglio V, Gianfranceschi L, Durel CE, Costa F, Yamamoto T, Koller B, Gessler C, Patocchi A (2006) Microsatellite markers spanning the apple (*Malus* x*domestica* Borkh.) genome. Tree Genet. Genomes **2:** 202-224

Smartt J, Simmonds NW (1995) Evolution of crop plants, Ed 2nd edn. Longman Scientific & Technical, Harlow, UK

Sugimoto T, Tamaki K, Matsumoto J, Yamamoto Y, Shiwaku K, Watanabe K (2005) Detection of RAPD markers linked to the everbearing gene in Japanese cultivated strawberry. Plant Breed. **124:** 498-501

Sung SK, Jeong DH, Nam JM, Kim SH, Kim SR, An G (1998) Expressed sequence tags of fruits, peels, and carpels and analysis of mRNA expression levels of the tagged cDNAs of fruits from the Fuji apple. Mol. Cells **8:** 565-577

Swanson CA (1998) Vegetables, fruits, and cancer risk: the role of phytochemicals. *In* WR Bidlack, ST Omaye, MS Meskin, DKW Topham, eds, Phytochemicals. A New Paradigm. CRC Press L.L.C., Boca Raton, pp 1-12

Tahmatsidou V, O'Sullivan J, Cassells AC, Voyiatzis D, Paroussi G (2006) Comparison of AMF and PGPR inoculants for the suppression of Verticillium wilt of strawberry (*Fragaria* ×*ananassa* cv. Selva). Appl. Soil Ecol. **32:** 316-324

Tartarini S, Sansavini S, Vinatzer B, Gennari F, C D (2000) Efficiency of marker assisted selection (MAS) for the Vf scab resistance gene. Acta Hort. **538:** 549-552

Tatum TC, Stepanovic S, Biradar DP, Rayburn AL, Korban SS (2005) Variation in nuclear DNA content in *Malus* species and cultivated apples. Genome **48:** 924-930

Teo G, Suziki Y, Uratsu SL, Lampinen B, Ormonde N, Hu WK, DeJong TM, Dandekar AM (2006) Silencing leaf sorbitol synthesis alters long-distance partitioning and apple fruit quality. Proc. Natl. Acad. Sci. U. S. A. **103:** 18842-18847

Vavilov NI (1951) The origin, variation, immunity and breeding of cultivated plants. Selected writings of N.I. Vavilov, translated from Russian by K. Starr Chester. Chron. Bot. **13:** 1-366

Verde I, Lauria M, Dettori M, Vendramin E, Balconi C, Micali S, Wang Y, Marrazzo MT, Cipriani G, Hartings H, Testolin R, Abbott AG, Motto M, Quarta R (2005) Microsatellite and AFLP markers in the *Prunus persica* [L. (Batsch)] × *P. ferganensis* BC_1 linkage map: saturation and coverage improvement Theor. Appl. Genet. **111:** 1013-1021

Vilanova S, Romero C, Abernathy D, Abbott AG, Burgos L, Llacer G, Badenes ML (2003) Construction and application of a bacterial artificial chromosome (BAC) library of *Prunus armeniaca* L. for the identification of clones linked to the self-incompatibility locus. Mol. Genet. Genomics **269:** 685-691

Vinatzer BA, Patocchi A, Gianfranceschi L, Tartarini S, Zhang HB, Gessler C, Sansavini S (2001) Apple contains receptor-like genes homologous to the *Cladosporium fulvum*

resistance gene family of tomato with a cluster of genes cosegregating with *Vf* apple scab resistance. Mol. Plant-Microbe Interact. **14:** 508-515

Vinatzer BA, Zhang HB, Sansavini S (1998) Construction and characterization of a bacterial artificial chromosome library of apple. Theor. Appl. Genet. **97:** 1183-1190

Viruel MA, Messeguer R, Devicente MC, Garciamas J, Puigdomenech P, Vargas F, Arús P (1995) A linkage map with RFLP and isozyme markers for almond. Theor. Appl. Genet. **91:** 964-971

Wang D, Fan J, Ranu RS (2004) Cloning and expression of 1-aminocyclopropane-1-carboxylate synthase cDNA from rosa (*Rosa* x*hybrida*). Plant Cell Rep. **22:** 422-429

Wang D, Karle R, Brettin TS, Iezzoni AF (1998) Genetic linkage map in sour cherry using RFLP markers. Theor. Appl. Genet. **97:** 1217-1224

Ware DH, Jaiswal P, Ni J, Yap IV, Pan X, Clark KY, Teytelman L, Schmidt SC, Zhao W, Chang K, Cartinhour S, Stein LD, McCouch SR (2002) Gramene, a tool for grass genomics. Plant Physiol. **130:** 1606-1613

Weebadde CK, Wang D, Finn CE, Lewers KS, Luby JJ, Bushakra J, Sjulin TM, Hancock JF (2007) Using a linkage mapping approach to identify QTL for day-neutrality in the octoploid strawberry. Plant Breed. **0:** doi: 10.1111/j.1439-0523.2007.01430.x

Weigel D, Ahn JH, Blazquez MA, Borevitz JO, Christensen SK, Fankhauser C, Ferrandiz C, Kardailsky I, Malancharuvil EJ, Neff MM, Nguyen JT, Sato S, Wang ZY, Xia YJ, Dixon RA, Harrison MJ, Lamb CJ, Yanofsky MF, Chory J (2000) Activation tagging in Arabidopsis. Plant Physiol. **122:** 1003-1013

Xu M, Song J, Cheng Z, Jiang J, Korban SS (2001) A bacterial artificial chromosome (BAC) library of *Malus floribunda* 821 and contig construction for positional cloning of the apple scab resistance gene *Vf*. Genome **44:** 1104-1113

Xu ML, Korban SS (2002a) A cluster of four receptor-like genes resides in the *Vf* locus that confers resistance to apple scab disease. Genetics **162:** 1995-2006

Xu ML, Korban SS (2002b) AFLP-derived SCARs facilitate construction of a 1.1 Mb sequence-ready map of a region that spans the *Vf* locus in the apple genome. Plant Mol. Biol. **50:** 803-818

Yamamoto T, Kimura T, Saito T, Kotobuki K, Matsuta N, Liebhard R, Gessler C, van de Weg WE, Hayashi T (2004) Genetic linkage maps of Japanese and European pears aligned to the apple consensus map. . Acta Hort. **663:** 51-56

Yamamoto T, Kimura T, Shoda M, Imai T, Saito T, Sawamura Y, Kotobuki K, Hayashi T, Matsuta N (2002) Genetic linkage maps constructed by using an interspecific cross between Japanese and European pears. Theor. Appl. Genet. **106:** 9-18

Yamamoto T, Yamaguchi M, Hayashi T (2005) An integrated genetic linkage map of peach by SSR, STS, AFLP and RAPD. J. Jpn. Soc. Hort. Sci. **74:** 204-213

Yan Z, Denneboom C, Hattendorf A, Dolstra O, Debener T, Stam P, Visser PB (2005) Construction of an integrated map of rose with AFLP, SSR, PK, RGA, RFLP, SCAR and morphological markers. Theor. Appl. Genet. **110:** 766-777

Yau MH, Che CT, Liang SM, Kong YC, Fong WP (2002) An aqueous extract of *Rubus chingii* fruits protects primary rat hepatocytes against tert-butyl hydroperoxide induced oxidative stress. Life Sci. **72:** 329-338

Zhebentyayeva TN, Horn R, Mook J, Lecouls A, Georgi L, Abbott AG, Reighard GL, Swire-Clark G, Baird WV (2006) A physical framework for the peach genome. Acta Hort.**:** 83-88

Ziegler RG, Mayne ST, Swanson CA (1996) Nutrition and lung cancer. Cancer Causes Control **7:** 157-177

Figure legends

Figure 1. Representative rosaceous crops exhibiting diversity of fruit types: A. *Prunus persica*, peach, fleshy drupe; B. *P. armeniaca*, apricot, fleshy drupe; C. *Malus ×domestica*, apple, pome; D. *Fragaria ×annanassa*, strawberry, achenes; E. *Rosa ×hybrida*, rose, achene; F. *Pyrus communis*, pear, pome; G. *Prunus avium*, sweet cherry, fleshy drupe; H. *Prunus domestica*, plum, fleshy drupe; I. *Rubus idaeus*, raspberry, drupelets.

Figure 2. Phylogenetic tree of Rosaceae, based on the multi-gene analysis by Potter et al. (2007) and modified from their Figure 4. Positions of several economically important genera are indicated and an hypothesis for the evolution of fruit types, based on parsimony optimization of that character, are represented by the different colors of the branches. The three subfamilies recognized by Potter et al. (2007) are shown on the right hand margin; Dry=Dryadoideae. The tree was rooted with representatives of the family Rhamnaceae (Rh).

Figure 3. Map comparison between *Prunus* (Joobeur et al., 1998) and *Malus* (Maliepaard et al., 1998). *Prunus* linkage groups are noted with G and *Malus* groups as L followed by a number. Only linkage groups with two or more common markers have been included. Markers in parenthesis in *Prunus* correspond to approximate positions deduced from other maps. Incomplete linkage groups are indicated by two parallel oblique lines.

Table 1. Model plant species for Rosaceae genetics and genomic research, compared to Arabidopsis

	Malus ×domestica	*Prunus persica*	*Fragaria vesca*	*Arabidopsis thaliana*
Genome size (Mbp/C)	750	280	206	157
Chromosome number (2n)	$2n = 2x = 34$ (some triploid cultivars)	$2n = 2x = 16$	$2n = 2x = 14$	$2n = 2x = 10$
No. of species in the genus	~ 35	~ 53	~ 20	9
Generation time (seed to seed)	4-8 years	3-5 years	10-16 weeks	6-8 weeks
Life cycle	perennial	perennial	perennial	annual
Seed production/plant	~ 700 (5-10 per fruit)	~ 300 (1 per fruit)	>2,500	~5,000
Plants per sq. meter	0.67	0.67	~100	776
Fleshy fruit formation	pome	drupe	receptacle	no
Juvenile period	3-7 years	1-2 years	none	none
Vegetative propagation	yes, hard and softwood cuttings	yes, hard and softwood cuttings	yes, runners and crown divisions	No
Self-compatible	no	yes	yes, cultivars and model species	Yes
Inbreeding depression	Moderate to severe	moderate	none to moderate	none to moderate
Transformation	tissue culture	tissue culture	tissue culture	*in planta*
Transformation efficiency	80%	<1%	100%	~1%
ESTs	> 300,000	85,340	>45,000	> 1.2 million
Genome sequence	mid-2008	end of 2008	~1%	Yes
Physical map	yes	yes	in progress	Yes
Linkage maps	yes	yes	yes	Yes

Table 2. Most relevant published genetic linkage maps of three model species in Rosaceae					
F. vesca	Population	Markers	Linkage groups	Map length	Reference
	F_2 between cv. Baron Solemacher and a clone collected from the wild	75 RAPD 3 isozyme loci 2 morphological traits	7	445 cM	(Davis and Yu, 1997)
	F_2 between *F. vesc*a f. semperflorens and F. *nubicola*	68 SSRs 1 SCAR 6 gene-specific markers 3 morphological traits	7	448 cM	(Sargent et al., 2004)
	Additions to above	109 SSRs and EST-SSRs	7	424 cM	(Sargent et al., 2006)
	F_2 between *F. vesca* 91.333.2 and cv. Snovit	66 SSRs	8	NA	(Cipriani et al., 2006)
	BC (*F. vesca* × [*F. vesca* × *F. viridis*])	31 SSRs 2 gene-specific markers	7	NA	(Nier et al., 2006)
	F_2 (*F. vesca* × *F. nubicola*) and BC_1 (*F. vesca* × *F. viridis*)	29 gene specific primers	7	Added to existing map	(Sargent et al., 2007)
Peach	F_2 (NC174RL × 'Pillar')	83 RAPD 1 isozyme 4 phenotypic markers	15	NA	(Chaparro et al., 1994)
	F_2 (almond 'Texas' × peach 'Earlygold') (reference map for *Prunus*)	11 isozymes 361 RFLP 449 SSR 5 STS, 42 resistance-associated genes	8	524 cM	(Dirlewanger et al., 2004b; Howad et al., 2005); (Joobeur et al., 1998; Aranzana et al., 2003; Lalli et al., 2005)
	F_2 (Dwarf peach 54P455 × almond 'Padre')	143 RFLP 8 isozymes 4 phenotypic characters 4 SSRs 1 RAPD 1 CAP	8	1144 cM revision and addition to existing map	(Bliss et al., 2002)
	F_2 ('Ferjalou Jalousia '× 'Fantasia')	82 SSR 37 RFLP 61 AFLP 1 isozyme 6 phenotypic characters	7	621 cM	(Dirlewanger et al., 2006)
	F_2 ('Akame' × 'Juseitou')	94 SSR 34 AFLP 24 RAPD 3 ISSR 9 phenotypic characters	8	571 cM	(Yamamoto et al., 2005)
	'Lovell' × 'Nemared'	169 AFLP	15	1297 cM	(Lu et al., 1998)
	BC_1 (IF7310828 × *P. ferganensis*)	78 RFLP 63 AFLP 57 SSR 16 RAPD 2 phenotypic characters	8	665 cM	(Dettori et al., 2001; Verde et al., 2005)
Apple	F_1 'Rome Beauty' × 'White Angel'	Isozymes RFLP	24 (♀) 21 (♂)	950 cM	(Hemmat et al., 1994)
	F_1 'Prima' × 'Fiesta'	124 RFLP	17	842 cM (♀)	(Maliepaard et al.,

		133 RAPD 17 Isozyme 4 AFLP 1 SCAR 10 SSRs		984 cM (♂)	1998)
	F_1 'Fiesta' × 'Discovery'	217 RAPD 118 SSR markers	17	914 cM (♀) 1015 cM (♂)	(Liebhard et al., 2002)
	'Prima' x 'Fiesta'	200 SSRs, 25 AFLPs	17	~800 cM	(Liebhard et al., 2003c)
	F_1 'White Angel' x 'Rome Beauty'	41 SSR		Added to existing map	(Hemmat et al., 2003)
	F_1 'Fiesta' × 'Discovery'	3 CAPS 14 SSCP	14	Added to existing map	(Baldi et al., 2004)
	F_1 'Fiesta' × 'Discovery'	149 SSRs (current reference map)	17	Added to existing map	(Silfverberg-Dilworth et al., 2006)
	F_1 'Braeburn' ×'Telamon'	245 AFLP 19 SSRs	17	1039 cM (♀) 1245 cM (♂)	(Kenis and Keulemans, 2005)
	'Discovery' × 'TN10-8'	43 NBS markers	10	Added to existing map	(Calenge et al., 2005)