American Journal of Botany
Celebrating 100 years 1914–2014

American Journal of Botany 101(10): 1686–1699, 2014.

SPECIAL INVITED PAPER

FRAGARIA: A GENUS WITH DEEP HISTORICAL ROOTS AND RIPE FOR EVOLUTIONARY AND ECOLOGICAL INSIGHTS[1]

AARON LISTON[2,5], RICHARD CRONN[3], AND TIA-LYNN ASHMAN[4]

[2]Department of Botany and Plant Pathology, Oregon State University, Corvallis, Oregon 97331 USA; [3]Pacific Northwest Research Station, USDA Forest Service, Corvallis, Oregon 97331 USA; and [4]Department of Biological Sciences, University of Pittsburgh, Pittsburgh, Pennsylvania 15260 USA

The cultivated strawberry, *Fragaria* ×*ananassa*, is one of the youngest domesticated plants. Its 18th century origin via hybridization in Europe between the North American *F. virginiana* and the South American *F. chiloensis* was documented by the botanist Antoine Nicolas Duchesne. His 1766 "Natural History of Strawberries" is an extraordinary work that integrates fundamental discoveries on the biology, ecology, and phylogeny of *Fragaria* with applied information on cultivation and ethnobotanical uses, serving as an inspiration for current research in the genus. *Fragaria* species exhibit the full range of sexual systems in the gynodioecy pathway from hermaphroditism to dioecy (and back again), as well as variation in self-compatibility, and evidence of sex chromosomes with female heterogamety. The genus is also characterized by interspecific hybridization and polyploidy, with a natural range of ploidy levels from diploids to decaploids. This biological diversity, combined with the availability of genomic resources and the ease of growing and experimenting with the plants, makes *Fragaria* a very attractive system for ecological and evolutionary genomics. The goal of this review is to introduce *Fragaria* as a model genus and to provide a roadmap for future integrative research. These research directions will deepen our understanding of the ecological and evolutionary context that shaped the ancestors of the cultivated strawberry, not only providing information that can be applied to efforts to shape the future of this important fruit crop but also our understanding of key transitions in plant evolution.

Key words: genomics; model organism; Rosaceae; strawberry.

Strawberries are an important food plant. Global strawberry production is twice the amount of all other berry crops combined (Stewart, 2011) and U.S. per capita consumption doubled between 1970 and 2004 (Boriss et al., 2006). Today, fresh consumption of strawberries trails only banana, apple, watermelon, and grape (USDA Economic Research Service, 2012). While primarily valued for their flavor, strawberries also have potential health benefits. Medicinal claims for strawberries have been made for centuries (Duchesne, 1766). Strawberries are high in the nutrients vitamin C, folate, and manganese (Giampieri et al., 2012). They are also rich in phenolic compounds, including anthocyanins, hydrolyzable tannins, and phenolic acids (Giampieri et al., 2012). Vitamin C and phenolic compounds are efficient antioxidants, and the potential role of strawberry consumption in human health and disease prevention is an active research area (Romandini et al., 2013). Tempering the enthusiasm for strawberry consumption is their prominent position on the list of foods with the highest pesticide residues (Environmental Working Group, 2013). Resistance to a *Fragaria*-specific powdery mildew (*Podosphaera macularis*) has been demonstrated in *F.* ×*ananassa* transformed with a peach locus (Jiwan et al., 2013), and the cultivation of such transgenic plants could reduce pesticide usage in strawberry. While transgenic strawberries are an important research tool (Folta, 2013), current prospects for their commercialization are limited, due to public resistance, a lack of industry support, and concerns over gene flow to the wild species of *Fragaria* (Mezzetti, 2009; Hummer et al., 2011).

An educationally important use of strawberries is for demonstrating DNA extraction (Gonda et al., in press). An advantage of using strawberries in this application is that they can be placed in a plastic bag and easily liquefied by hand. The cells are lysed with detergent (dish washing liquid or shampoo), filtered through a coffee filter, and DNA is precipitated in the presence of salt and ethanol (GenomeTV, 2010). The octoploid genome of the cultivated strawberry likely contributes to the abundant amounts of DNA obtained from this simple procedure.

Wild *Fragaria* (Rosaceae) species occur across the northern hemisphere and disjunctly in southern South America (Fig. 1, Table 1). Humans have likely consumed the fruits of wild *Fragaria* species for millennia. While archaeobotanical evidence is limited (Walshaw, 2009), *Fragaria* achenes have been documented from two pre-Columbian sites in eastern North America (Gremillion and Sobolik, 1996; Pauketat et al., 2002). *Fragaria chiloensis* was domesticated by the Picunche and

[1]Manuscript received 27 March 2014; revision accepted 1 July 2014.

The authors thank Vivian Lee Bowden, Michel Chauvet, Michelle Coyne, and Alice Telka for strawberry information; Michel Chauvet and Ariadne Luh for assistance with French translation; Branko Bakan, Gerald Carr, Luxi Chen, and Shiro Tsuyuzaki for permission to reproduce their photographs; and Matt Koski, Jacob Tennessen, Dan Potter, and an anonymous reviewer for comments on a draft of the manuscript. Our strawberry research is supported by the National Science Foundation (DEB 1020523 and DEB 1241006 to T.L.A., DEB 1020271 to A.L., and DEB 1241217 to A.L. and R.C.).

[5]Author for correspondence (e-mail: aaron.liston@oregonstate.edu)

doi:10.3732/ajb.1400140

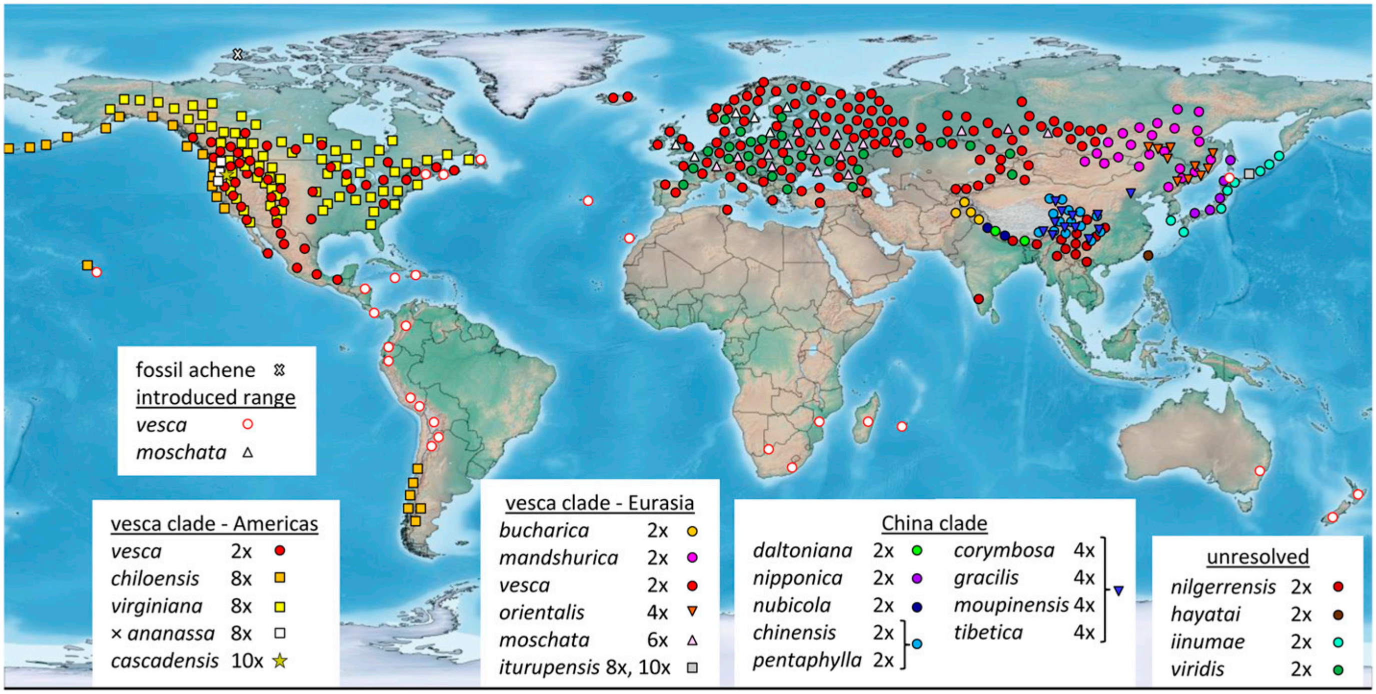

Fig. 1. Approximate geographic distribution of *Fragaria* species and ploidy. Due to uncertainty over species boundaries, the six endemic Chinese species are designated as diploids (*F. chinensis*, *F. pentaphylla*) or tetraploids (*F. corymbosa*, *F. gracilis*, *F. moupinensis*, *F. tibetica*). Data sources include the GBIF data portal (GBIF, 2014), the “Wild Strawberry” Dimensions of Biodiversity US-China project website (Ashman et al., 2014), published distribution maps (Staudt, 1999a, 1999b, 2003b, 2005, 2006, 2008; Staudt and Dickoré, 2001; Chukhina, 2008; Staudt and Olbricht, 2008; Rousseau-Gueutin et al., 2009), and base map (Shorthouse, 2010).

Mapuche people of Chile over 1000 yr ago (Finn et al., 2013). In Europe, *F. vesca* has been grown in gardens at least since the time of the Romans, and *F. moschata* since the 16th century (Wilhelm & Sagen, 1974). The modern cultivated strawberry, *Fragaria* ×*ananassa* originated in the 18th century in Europe from hybridization between two species imported from North and South America (Duchesne, 1766). The parental species, *F. virginiana* and *F. chiloensis*, also hybridize naturally in northwestern North America (Luby et al., 1992; Salamone et al., 2013), but there is no evidence that they were ever cultivated by the native Americans in this area. However, these hybrids represent an untapped opportunity to study the genetic equivalent of the domesticated strawberry in a natural ecological context (Table 2).

The sequencing and annotation of a genome provides access to the genes, their putative functions, and their genomic locations, as well as a reference that can be used to assay nucleotide variation and gene expression across individuals and conditions (Bevan and Uauy, 2013). The genome sequence of *Fragaria vesca* was a scientific milestone, representing the first published plant genome obtained solely with next-generation sequencing technology (Shulaev et al., 2011). *Fragaria vesca* has several attributes that made it an attractive target for genome sequencing. It is a diploid with a relatively small 206-Mbp genome (Longhi et al., 2014). Compared with most other cultivated perennials, the plants are compact, easily grown under controlled conditions, and can be propagated both sexually and asexually (Slovin and Michael, 2011). An efficient transformation system for *F. vesca* has been developed (Oosumi et al., 2006) and transposon-tagged mutants are available for reverse genetics applications (Veilleux et al., 2012). The genome sequence of *F. vesca* has already facilitated significant progress in functional genomics of flowering (Mouhu et al., 2013; Hollender et al., 2014) and fruiting (Kang et al., 2013), and these advances will ultimately be applied to crop improvement efforts (Folta, 2013; Longhi et al., 2014). More immediate progress is anticipated from the vast numbers of genetic markers developed from the genome sequence of *F. vesca* that can be used in genetic linkage mapping (Tennessen et al., 2013) and high-throughput assays for marker-assisted breeding (Chambers et al., 2013). The cultivated strawberry *F.* ×*ananassa* (which, like *F. vesca,* is readily transformed genetically; Shulaev et al., 2008) has also been used as a “taxonomic surrogate” for peach (a species not amenable to tissue culture) in a recent study of a mildew resistance locus isolated from peach and shown to be functional in strawberry (Jiwan et al., 2013). These results illustrate the kind of “cross-fertilization” that can occur with the development of genomic resources across Rosaceae (Shulaev et al., 2008; Longhi et al., 2014).

The small genome size in *Fragaria* and the availability of a reference sequence also facilitate comparative genomics, and a recent study has included whole genome sequencing of the cultivated strawberry and four wild relatives (Hirakawa et al., 2014). The biological diversity of wild *Fragaria* (Table 1) has been regularly used in breeding efforts as a source of novel genetic variation that can be introgressed into the cultivated strawberry (Hancock, 1999; Chambers et al., 2013). It is less appreciated that the biological diversity of *Fragaria*, especially with respect to sexual system and polyploidy, also makes it a very attractive system for ecological and evolutionary genomics. The goal of this review is to introduce *Fragaria* as a model genus and to provide a roadmap for future integrative research in these and other areas.

TABLE 1. *Fragaria* species and their ploidy level, mating system and geographic distribution.

Taxon	Ploidy	Mating system	Distribution
Vesca clade			
F. ×*ananassa* Duchesne subsp. *ananassa*	8x	Subdioecious (modern cultivars: hermaphroditic)	cultivated
F. ×*ananassa* Duchesne subsp. *cuneifolia* (Nutt. ex Howell) Staudt	8x	Subdioecious	NW N. America
F. bucharica Losinsk.	2x	Hermaphrodite SI	W Himalayas
F. cascadensis Hummer	10x	Subdioecious	Oregon, USA
F. chiloensis (L.) Duchesne	8x	Subdioecious	Alaska-California; Hawaii; Chile, Argentina
F. iturupensis Staudt	8x, 10x	Subdioecious	Iturup Island
F. mandshurica Staudt	2x	Hermaphrodite SI	NE Asia
F. moschata Duchesne	6x	Dioecious	W Eurasia
F. orientalis Losinsk.	4x	Dioecious	NE Asia
F. vesca L. subsp. *americana* (Porter) Staudt	2x	Hermaphrodite SC	NE N. America
F. vesca L. subsp. *bracteata* (A. Heller) Staudt	2x	Gynodioecious or Hermaphrodite SC	W N. America
F. vesca L. subsp. *californica* (Cham. & Schldl.) Staudt	2x	Gynodioecious or Hermaphrodite SC	SW N. America
F. vesca L. subsp. *vesca* L.	2x	Hermaphrodite SC	W Eurasia, also introduced (see Fig. 1)
F. virginiana Duchesne	8x	Subdioecious	N. America
China clade			
F. chinensis Losinsk.	2x	Hermaphrodite SI	China
F. corymbosa Losinsk.	4x	Dioecious	China
F. daltoniana J. Gay	2x	Hermaphrodite SC	Nepal, adjacent China
F. gracilis Losinsk.	4x	Dioecious	China
F. moupinensis (Franch.) Cardot	4x	Dioecious	China
F. nipponica Makino	2x	Hermaphrodite SI	Japan
F. nubicola Lindl.	2x	Hermaphrodite SI	Himalayas
F. pentaphylla Losinsk.	2x	Hermaphrodite SI	China
F. tibetica Staudt & Dickore	4x	Dioecious	China
Unresolved phylogenetic position			
F. hayatai Makino	2x	Hermaphrodite SC?	Taiwan
F. iinumae Makino	2x	Hermaphrodite SC	Japan
F. nilgerrensis Schltdl. ex J. Gay	2x	Hermaphrodite SC	SE Asia
F. viridis Duchesne	2x	Hermaphrodite SI	W Eurasia

Notes: SI = self-incompatible; SC = self-compatible. Subspecies are not listed for *F. chiloensis* and *F. virginiana* (Staudt, 1999a) nor *F. nipponica* (Staudt and Olbricht, 2008).

MORPHOLOGY, SPECIES, AND PHYLOGENY OF *FRAGARIA*

Technically, the strawberry is an aggregate accessory fruit. The botanical fruits are dry achenes that are embedded in a fleshy receptacle. This morphological combination is shared by all species of *Fragaria* and has apparently evolved in parallel three times in Rosaceae tribe Potentilleae (Eriksson et al., 1998). The other occurrences are in *Comarum palustre* and in two species of *Potentilla* that were formerly segregated as the genus *Duchesnea*. The coordinated development of seed maturation in the achene with the softening and expansion of the receptacle in *Fragaria* provides a unique opportunity to investigate signaling between plant organs (Kang et al., 2013). It would be instructive to extend this study of gene expression in a developmental context to the other two cases of this unusual fruit morphology to determine whether the same gene expression patterns occur.

The same basic life history is shared by all *Fragaria* species: they are insect-pollinated, low-growing, herbaceous perennials capable of clonal growth, and have animal-dispersed fleshy accessory fruits (Johnson et al., 2014). The floral and vegetative morphology of *Fragaria* is also relatively uniform. The leaves are usually evergreen (*F. iinumae* is deciduous), and generally trifolioliate. Five leaflets are typical of some Chinese species, while 4–5 leaflets are rarely observed in *F. virginiana* (Kellerman, 1892) and *F. cascadensis* (Hummer, 2012). Flowers are always actinomorphic, white (sometimes tinged with pink), and usually 5-petalled (*F. iinumae* has 6–8 petals, Fig. 2D). In some species, staminate and pistillate flowers are readily distinguished (Fig. 2A, B), but in others (e.g., gynodioecious *Fragaria vesca* subsp. *bracteata*) the pistillate flowers have anthers and are very similar to the bisexual ones (Li et al., 2012; Ashman et al., 2012). The mature receptacles ("fruits") are more diverse and can be used to differentiate species (Fig. 2E–I). However, they are also quite variable within a species, and because their characteristic features (color, shape, and achene and calyx positions at maturity) are "inevitably destroyed" (Staudt, 1999a) in herbarium specimens, they are not well documented in the botanical literature. For this reason, identification keys tend to focus on details of leaf texture, dentation, venation, and pubescence. Further complicating identification is extensive phenotypic plasticity within species (Chabot and Chabot, 1977; Jurik et al., 1982). The morphology of the stolons is also a reliable diagnostic character (Staudt, 2008), with some species having monopodial stolons (after the first sterile internode, a plantlet can develop at every internode) and others

TABLE 2. Potential research areas that can be addressed in *Fragaria* through a combination of phylogenetic and population genomics/genetic approaches, and ecological, physiological or developmental biology.

Research Area/Ecological question	Example of *Fragaria* species that can be used to address issue	Current understanding in *Fragaria*	Exemplar references
A) Sexual and mating system evolution			
How do sex chromosomes evolve?	Octoploid clade	Evidence for proto-sex chromosomes; dynamic sex determining regions.	Spigler et al., 2008, 2010; Goldberg et al., 2010; Tennessen et al., 2013
How do separate sexes evolve from hermaphroditism?	Genus-wide	Ecological drivers include inbreeding depression, stressful habitats and antagonists.	Ashman, 1999, 2005; Ashman and Penet, 2007
What genetic mechanisms underlie male (female) sterility?	Gynodioecious *F. vesca* subsp. *bracteata*	Novel form of nuclear dominant male sterility.	Tennessen et al., 2013
How does the polyploidy–dioecy association arise?	Diploid–polyploid ancestor–descendent pairs	Dioecy is not associated with loss of SI, but polyploidization can alter sex allocation.	Njuguna et al., 2013; Ashman et al., 2013; Kwok, 2013
Is self incompatibility reversible? Is selfing a dead end?	SI species/SC species	Both loss and gain are possible in *Fragaria*; genetic dissection suggests multiple S-loci.	Bošković et al., 2010; Njuguna et al., 2013
B) Speciation, hybridization, polyploidy and range expansion			
How does polyploidy contribute to ecological and evolutionary amplitude?	Diploid–polyploid ancestor–descendent pairs	Based on current understanding range size is related to species age but not ploidy.	Johnson et al., 2014
Are polyploids the result of single or multiple origins?	Diploid–polyploid ancestor–descendent pairs	Two octoploid ancestors identified, ancestry of other polyploids. uncertain; single or multiple origins untested for all polyploids.	Rousseau-Gueutin et al., 2009; Njuguna et al., 2013
To what degree are polyploid subgenomes repatterned?	Diploid–polyploid ancestor–descendent pairs	Sex locus is in different locations in *F. chiloensis* and *F. virginiana*.	Spigler et al., 2008, 2010; Goldberg et al., 2010; Tennessen et al., 2013
How reproductively isolated are species? Is the hybrid zone stable, or does hybridization contribute to adaptation?	Natural hybrids *F.* ×*ananassa* subsp. *cuneifolia* *F.* ×*bifera*	Pattern of introgression in subsp. *cuneifolia* suggests geographic limitations that may be reinforced by selection via bioclimate.	Salamone et al., 2013
How do speciation genes arise? What are the molecular mechanisms of reproductive isolation?	Diploid species	Variation in interfertility among diploid species, yet unrelated to genetic distance.	Bors and Sullivan, 2005; Nosrati et al., 2011b
What contributes to successful invasion? What are the molecular mechanisms of weediness?	*F. vesca* subsp. *vesca* established outside of native range	Evidence of establishment in multiple independent locations (Hawaii, Andes, Azores, La Réunion, Madagascar, New Zealand).	Staudt, 1999a; Plantwise Knowledge Bank, 2014
C) Life-history transitions			
How does flowering time evolve?	Wide latitudinal range in *F. vesca* subsp. *vesca*	Central genetic components of flowering time pathways, but with novel regulatory mechanisms, have been identified.	Mouhu et al., 2009
How does drought tolerance evolve and by what genetic or functional pathways?	*F. chiloensis* vs. *F. virginiana*	Potential candidate genes in control of plant responses to drought stress identified.	Razavi et al., 2011; Šurbanovski et al., 2013
How does frost tolerance evolve and by what genetic or functional pathways?	Wide latitudinal range in *F. vesca* subsp. *vesca*	Clinal variation in frost hardiness with latitude of population origin.	Sønsteby and Heide, 2011
What genetic or functional pathways control allocation between sexual/asexual reproduction or alternative strategies for clonal growth?	Sympodial and monopodial species and runnerless variants of *F. vesca*	Genetic dissection of regulation of flowering time in response to temperature and photoperiod. Clonal growth strategies enhance fitness via resource sharing.	Alpert, 1991; Hytönen et al., 2009; Koskela et al., 2012
How do fleshy fruits evolve from dry fruits?	*Fragaria* vs. other members of Potentilleae	Transcriptomic profiling of early fruit development.	Hollender et al., 2014
How does woodiness evolve from herbaceousness (or vice versa)?	*Fragaria* vs. *Dasiphora fruticosa*	Transcriptome comparison.	T. Poulson and D. Hearn, Towson University, unpublished manuscript

sympodial (plantlets and new stolons develop at alternate internodes, Fig. 2C). Like fruits and flower sexuality, this trait is best observed in living specimens. Achene size is known to differ between diploid and tetraploid plants of *F. mandshurica* and *F. orientalis* (Staudt, 2003b), and between diploids of *F. vesca* and the North American octoploids (Staudt, 1999a). However, its reliability for ploidy estimation across a diversity of species and populations remains to be determined.

Current species concepts in *Fragaria* are predominantly based on the systematic studies of Günter Staudt (1926–2008). His research incorporated crossing studies, cytogenetics, and the taxonomic revision of most species (Table 1). With one

Fig. 2. Morphological diversity of *Fragaria.* (A) *F. chiloensis*, staminate flower. (B) *F. chiloensis*, pistillate flower. (C) *F. chiloensis*, sympodial stolons, with sterile node (red arrow) alternating with fertile node (yellow arrow). (D) *F. iinumae,* flower with 7 petals, 6–9 petals are characteristic of this species. (E) *F. iinumae,* achenes in shallow pits on the receptacle, also found in the octoploid clade. Also note the glaucous leaflets, a characeteristic shared with *F. virginiana.* (F) *F. vesca,* achenes raised above the surface of the receptacle. (G) Tetraploid *F. moupinensis* (back) and diploid *F. pentaphylla* (front). Note that *F. moupinensis* is larger, and the central leaflet overlaps the two lateral leaflets. (H) *F. daltoniana*, elongated mature receptacle and small, coriaceous leaves. One of the most distinctive species of the China clade. (I) *F. viridis,* the reflexed calyx and mature receptacle that detaches from the calyx with an audible "click" are characteristics of this species. Photographer and Location: (A, B) Gerald Carr (Oregon, USA); (C) Aaron Liston (Oregon, USA); (D, E) public domain (Japan); (F) Oxfordian Kissuth (Germany); (G) Aaron Liston (Sichuan, China); (H) Luxi Chen (Tibet, China); (I) Branko Bakan (Slovenia).

exception (*F. iturupensis*), intraspecific variation in ploidy is not known, and thus chromosome number is a reliable way to distinguish *Fragaria* species. The use of flow cytometry in conjunction with an 8× standard for *F.* ×*ananassa* is an efficient method for determining ploidy (R. Cronn, unpublished data).

Molecular phylogenetic analyses based on two nuclear genes (Rousseau-Gueutin et al., 2009) and chloroplast genomes (Njuguna et al., 2013) consistently resolve two clades: the "China clade" comprising nine diploid and tetraploid species—eight of which are distributed in China and adjacent Himalayan countries, one of which is endemic to Japan; and the "vesca clade" comprising 11 species ranging from diploid to decaploid and distributed in northern Eurasia, North and South America, and Hawaii. Monopodial stolons are characteristic of all species in the China clade plus *F. viridis* (Staudt, 2008). The phylogenetic positions of three diploid species, Eurasian *F. viridis*, southeast Asian *F. nilgerrensis*, and Japanese *F. iinumae*, currently remain unresolved. *Fragaria hayatai*, endemic to Taiwan, was recently recognized as distinct from *F. nilgerrensis* based on the presence of spinules on the pollen exine (Staudt, 2008). It has not been included in any genetic or phylogenetic studies of the genus. The vesca clade includes *Fragaria chiloensis* and *F. virginiana,* the octoploid ancestors of the cultivated strawberry, *F.* ×*ananassa* subsp. *ananassa.* Phylogenetic analyses of nuclear genes (Rousseau-Gueutin et al., 2009; DiMeglio et al., 2014) implicate *F. iinumae* and *F. vesca* s.l. (including *F. mandshurica*) as progenitors of these two octoploids as well as the octoploid/decaploid *F. iturupensis.* Plastome phylogenetics (Njuguna et al., 2013) resolve these three species as monophyletic and sister to *F. vesca* subsp. *bracteata*, suggesting a shared cytoplasm from a western North American ancestor.

Conflicting evidence exists for the origin of the tetraploid *F. orientalis* and hexaploid *F. moschata.* Nuclear genes are consistent with an allotetraploid origin of *F. orientalis* from *F. vesca* and *F. mandshurica* (Rousseau-Gueutin et al., 2009), while these nuclear data and two chloroplast intergenic spacers (Lin and Davis, 2000) support *F. viridis* as the maternal progenitor of *F. moschata.* In contrast, plastome evidence supported a common cytoplasmic ancestry for *F. moschata* and tetraploid *F. orientalis*, and no evidence for *F. viridis* maternal ancestry in *F. moschata* (Njuguna et al., 2013). An important caveat is that these phylogenetic studies included a single accession of each species and very few genes; thus, increased sampling of both should resolve these discrepancies.

Two different decaploids are known in the vesca clade. *Fragaria iturupensis* is only known from Iturup Island (in the southern part of the Kuril archipelago) and octoploidy has been observed in two accessions (Staudt and Olbricht, 2008). Subsequently, a decaploid accession of *F. iturupensis* was described (Hummer et al., 2009). Thus, it seems likely that both 8*x* and 10*x* individuals exist in the narrow geographic range of this species. The decaploid *Fragaria cascadensis* was recently described from populations in the Cascade range of western Oregon (Hummer, 2012). This taxon is morphologically similar to *F. virginiana* and was previously confused with this taxon. The phylogenetic origins of these two decaploids have not been determined.

The China clade includes four tetraploids; however, only one species, *F. tibetica*, has been the subject of a taxonomic revision (Staudt and Dickoré, 2001), and the taxonomic status of the other three (*F. corymbosa, F. gracilis, F. moupinensis*) remains uncertain. Likewise, published phylogenetic analyses (Rousseau-Gueutin et al., 2009; Njuguna et al., 2013; DiMeglio et al., 2014) have not resolved the diploid ancestors of these species, nor whether they have single or multiple origins.

The extant species of *Fragaria* are estimated to have last shared a common ancestor between 1.0 and 4.1 million years ago (mya), while the octoploid clade originated between 0.370 and 2.05 mya (Njuguna et al., 2013). These estimates are consistent with the only fossil evidence of the genus, a single achene (A. Telka, retired, Geological Survey of Canada, personal communication) dating to 2.9 ± 0.4 mya (Matthews and Ovenden, 1990; Töpel et al., 2012). The fossil was collected on Prince Patrick Island, Canada at 76°–77° N latitude, representing a remnant of Pliocene-Pleistocene Beringia. Although this sample predates the estimated age of the octoploid clade, the current distribution of their known ancestors (*F. iinumae* in Japan and *F. vesca* subsp. *bracteata* in Northwest North America) suggests a possible octoploid origin in Beringia. Unfortunately, the specimen is "not currently discoverable" (M. Coyne, Canada Centre for Mapping and Earth Observation, Ottawa, ON, personal communication) and thus unavailable for further study of its morphology and species identity.

HISTORICAL BACKGROUND

Two major topics of *Fragaria* research, sex and polyploidy, can be traced back to the extraordinary work of Antoine Nicolas Duchesne (1747–1827). In 1766, he published *Histoire Naturelle des Fraisiers* (*Natural History of Strawberries*). The book's subtitle, "*les vues d'économie réunies à la botanique*", can be translated as "an integration of economic and botanical perspectives." This aim of 250 yr ago resonates with the theme of this special issue (*plus ça change…*). The fact that the author was 19 yr old when his book was published is only one of its several remarkable features. The historical significance of Duchesne's work has been previously described (Paris, 2000; Chauvet, 2003; Ratcliff, 2007). In addition to precise botanical descriptions, the history, cultivation, flowering and fruiting seasons, and effects of climate on each species are described. In "a truly unusual attempt for this century" (Lee, 1966), Duchesne also describes his ideas on the relationships of the species, arranges them by this system, and even includes a drawing of a genealogical tree. This diagram is considered the second published phylogenetic network (Morrison, 2014) and shows his novel (and subsequently proven) hypothesis that *Fragaria* ×*ananassa* originated as a hybrid between *F. chiloensis* and *F. virginiana.* The first section concludes with a discussion of strawberry cultivation and utilization for food, medicine, and other uses (e.g., stolons for dental floss!).

Part two of his book, titled *Remarques Particulières*, includes the first description of separate sexes in strawberry. Not only did Duchesne observe the somewhat cryptic floral differences, but he also conducted experiments that proved both male and female plants are required for successful fruit production in the dioecious, cultivated species *F. moschata* and *F. chiloensis.* Duchesne's discovery of strawberry sex had immediate practical benefits: it explained why the common practice of culling "sterile" male plants from plantings of *F. moschata* was a bad idea. He also discovered the key to fruit production in *F. chiloensis*. Only female plants were introduced to Europe from South America, and fruit production was very sporadic. In 1764, Duchesne successfully pollinated female plants of *F. chiloensis* with pollen from *F. moschata.* The large, beautiful strawberries produced led to an audience with King Louis XV and served as the impetus for the production of his book (Lee, 1966)[6].

---

[6] A collection of 73 drawings of *Fragaria* prepared by Duchesne was published in 2003 (Staudt, 2003a). The originals were discovered in the Library of the Paris Natural History Museum in 1962 by Vivian Lee, an undergraduate at Stanford University (Lee, 1964, V. L. Bowden, personal communication). She wrote four chapters on the history of strawberry (Lee in Darrow, 1966). Her contribution is clearly acknowledged in the preface, but not on the title page, and most subsequent authors have misattributed the chapters to George M. Darrow, a USDA strawberry geneticist.

Duchesne also attempted to cross the European *F. vesca* with the South American *F. chiloensis,* without success. A century and a half later, the discovery of a polyploid series in *Fragaria* (Ichijima, 1926; Longley, 1926) would explain why this cross failed. Differences in chromosome number between the diploid *F. vesca* and the octoploid *F. chiloensis* is the cause of the reproductive isolation observed by Duchesne. Likewise, the vigorous F1 hybrid that Duchesne made between hexaploid *F. moschata* and octoploid *F. chiloensis* is also completely sterile (Mangelsdorf and East, 1927). Duchesne was apparently aware that interspecific hybrids were often sterile, as he wrote in 1766 that he was still waiting to see if his hybrid plants would produce fertile seed (Lee, 1966).

Duchesne's interest in strawberry can be traced to his 1761 discovery in the gardens of Versaille (where his father was superintendent of the king's buildings) of a unifoliolate plant in a bed of the typical, trifolioliate *Fragaria vesca*. He sent specimens to Linnaeus, who published it as the species *Fragaria monophylla*. This experience, and his observations of variable reproductive isolation among *Fragaria,* led Duchesne to ponder the nature of plant species, and the final section of his book includes a thorough exposition of what became known two centuries later as the biological species concept (Paris, 2000). In fact, this is why Duchesne used the term "race" instead of species in his 1766 treatment of *Fragaria*.

Fragaria vesca was used in some of the earliest plant genetic studies following the rediscovery of Mendel's laws. Richardson (1914) cited Duchesne's (1766) observations of "*F. monophylla*" and reported Mendelian segregation of these leaf traits (as well as red vs. white fruit color and stolon presence/absence) in *F. vesca.* Likewise, the polyploid series in *Fragaria* was discovered in the early years of cytogenetics (Ichijima, 1926; Longley, 1926). In general, polyploid species of *Fragaria* are larger, more vigorous, and more variable than the diploids (Schiemann, 1932). Octoploidy has likely contributed to the large increases in fruit size (Sherman et al., 1966) and probably other aspects of yield (fruit number, plant vigor, disease resistance, Hancock, 1999) obtained in the cultivated strawberry.

From an applied perspective, the presence of different ploidy levels among *Fragaria* species has functioned as an impediment to the incorporation of desirable traits from the diploid *F. vesca* and hexaploid *F. moschata* into the the octoploid *F. ×ananassa* (Evans, 1977). However, efforts to overcome this barrier have provided a rich data set on chromosome behavior in interspecific crosses (Senanayake and Bringhurst, 1967), the occurrence of unreduced gametes (Bringhurst and Gill, 1970) and varying levels of fertility among ploidy levels in *Fragaria* (Bors, 2000). The presence of separate sexes in *F. ×ananassa* was also treated as an undesirable trait in early cultivars (Darrow, 1966), and their absence in modern cultivars apparently represents "unconscious artificial selection" by strawberry breeders (Schiemann, 1932). However, both of these features, sexual system diversity and polyploidy, are what make *Fragaria* a unique system for answering basic questions about two fundamental and universal evolutionary transitions in flowering plants.

SEXUAL AND MATING SYSTEM EVOLUTION

While cultivated *Fragaria ×ananassa* is a self-compatible hermaphrodite, it is derived from the hybridization of two wild species that show gender dimorphism, dioecious (males and females) *F. chiloensis* and subdioecious (hermaphrodites, males, and females) *F. virginiana*. Gender dimorphism is common among *Fragaria* species (Fig. 2A, B; Table 1), and the genus is proving to be an exceptional model system for understanding sexual system and sex chromosome evolution, not only because it hosts the full range of sexual systems in the gynodioecy pathway from hermaphroditism to dioecy (and back again), but also because it has variation in self-compatibility, and evidence of sex chromosomes with female heterogamety (Kihara, 1930). Knowledge of the genetics of sex determination in *Fragaria* is also an important tool for crop development as wild species continue to be resources for genetic improvement of the cultivar (reviewed in Hancock, 1999). In addition, this knowledge will provide genetic insight into a key transition in plant evolution.

Even though dioecy is rare in angiosperms (6%), its evolution from hermaphroditism has occurred in over half of all plant families (Renner and Ricklefs, 1995) suggesting strong convergent evolution. On the basis of character-state mapping on the chloroplast phylogeny of *Fragaria*, male sterility evolved in both major clades independently, and there may have been as many as six transitions from hermaphroditism to male sterility (Njuguna et al., 2013). Interestingly, female function was also lost several times, and female sterility was associated with increases in ploidy. Comparisons between closely related taxa that differ in sexual system offer powerful means for understanding the origin of sex chromosomes from autosomes, as well as provide evidence for the initial steps in sex chromosome evolution, including linkage of and recombination suppression between sex function "loci" (Charlesworth et al., 2005).

Sex determination in octoploid strawberries was previously thought to be determined by a single Mendelian locus consisting of either a single gene or a gene complex with femaleness dominant to maleness, the latter of which was recessive to hermaphroditism (Ahmadi and Bringhurst, 1991). However, a reassessment via genetic mapping in *F. virginiana* led to the discovery of the earliest stage of a sex chromosome, the linkage of two gene regions (one for male function and one for female function), but with recombination between them (Spigler et al., 2008, 2010). Moreover, comparative mapping in the sibling species *F. chiloensis* (Goldberg et al., 2010) also showed dominant male sterility was linked in coupling to female fertility, but here there was no evidence of recombination between them, suggestive of the next stage of the stepwise evolution of a sex chromosome (Charlesworth et al., 2005). However, the location of the sex determining region (SDR) was not syntenous in the two species, the SDR mapped to the bottom of chromosome VI-A in *F. chiloensis* but the top of Vl-C in *F. virginiana*. This result suggests that either the two do not share the same sex determining locus as previously thought (Ahmadi and Bringhurst, 1991) or that the SDR has been translocated between chromosomes in the same homeologous group. Both dynamics have been documented in other taxa (e.g., sticklebacks, Ross et al., 2009). The development of genomic resources in *Fragaria* is providing ingress to tests of these alternatives as well as determining whether the SDR show the hallmarks of evolving sex chromosomes (sex-linkage of sexually antagonistic genes, degeneration of genes, the accumulation of repetitive elements, and low density of genes [Ming and Moore, 2007; Otto et al., 2011; Spigler et al., 2011]). Extending these approaches to *F. moschata*, where ZW sex chromosomes have been observed (Kihara, 1930) will also make *Fragaria* an ideal system for answering the open question of whether ZW sex determination systems follow the same dynamics as XY systems (Bachtrog et al., 2011).

In addition to the underlying genetics of sex expression, population genetic models identify the central role of selfing rate, inbreeding depression, and relative seed and pollen fitness gain curves in affecting sex ratio and sex system evolution (reviewed by Webb, 1999). Empirical work in a number of systems has demonstrated the importance of plant interactions with both abiotic (resources) and biotic (pollinators and enemies) factors in modifying these (reviewed by Ashman, 2002, 2006; Spigler and Ashman, 2012). The combination of observational and experimental studies of *F. virginiana*, in particular, have provided the most complete evaluation of interplay of ecological and genetic forces determining the location of a population along the hermaphroditism–dioecy continuum. For instance, despite strong resource-based selection on female frequency, i.e., the sex ratio (Ashman, 1999), the evolution of full dioecy in this subdioecious species is retarded by selection against males via florivore damage (Ashman et al., 2004; Ashman and Penet, 2007), and gene flow from other populations where sex allocation plasticity and/or the colonizing ability in hermaphrodites is beneficial (Spigler and Ashman, 2011). Furthermore, studies of phenotypic selection combined with a growing understanding of the genetic architecture has provided unparalleled insight into how and when sexual dimorphism evolves during the evolution of separate sexes (Morgan and Ashman, 2003; Ashman, 2005; Spigler and Ashman, 2011, 2012). Moreover, comparative analyses across species has confirmed the existence of greater sexual dimorphism in dioecious species compared with subdioecious and gynodioecious species (Ashman et al., 2013), though hybridization can lead to the breakdown of these associations (Govindarajulu et al., 2013). Hybridization may in fact fuel transitions in sex determining regions in this genus (Govindarajulu et al., 2013). Developing molecular tools will be used to confirm this and to add a mechanistic understanding of the control of sexual dimorphism (Spigler et al., 2011).

Finally, although many ecological and genetic factors have been associated with the transition to separate sexes in plants (Givnish, 1980; Vamosi et al., 2003; Ashman, 2006; Spigler and Ashman, 2012), one of the most bewildering relationships is that between dioecy and an increase in ploidy (Baker, 1984; Miller and Venable, 2000; Ashman et al., 2013). The genus *Fragaria* is amenable to answering the question of how genome doubling leads to the initiation of dimorphic sexual systems (Ashman et al., 2013). The prominent hypothesis that invokes loss of self-incompatibility and concomitant increased selfing and inbreeding depression following whole genome duplication (Miller and Venable, 2000) is not supported in *Fragaria* because although polyploid dioecious taxa are often sister to diploid hermaphroditic taxa, these diploids are not always self-incompatible (Njuguna et al., 2013). As a result, the door is open for tests of other possible pathways in *Fragaria* given its high rate of unreduced gamete formation, ongoing natural polyploid formation, and ease of synthetic polyploid creation (Ashman et al., 2013; Kwok, 2013).

SPECIATION, HYBRIDIZATION, POLYPLOIDY, AND RANGE EXPANSION

Fragaria species occur across a broad range of temperate habitats and elevations (Fig. 1) from sea level sand dunes to moist, productive meadows to high, dry mountain summits (Johnson et al., 2014). In addition, considerable ecological amplitude is found within and between many of the species (Hancock and Bringhurst, 1978, 1979, 1981; Hancock and Luby, 1993). This variation represents a potential source of genetic variation for climatic tolerance, disease/pest resistance, and yield-associated traits (Hancock and Luby, 1993; Stewart and Folta, 2010). Furthermore, this natural variation (Table 2) provides the opportunity to investigate the ecological genomics of traits such as flowering time (Mouhu et al., 2009), drought (Razavi etal., 2011; Šurbanovski et al., 2013), and frost tolerance (Sønsteby and Heide, 2011).

Despite sharing the same life history traits, only one species, *F. vesca* subsp. *vesca*, is known to have become established in natural environments outside of its native range (Staudt, 1999a; Plantwise Knowledge Bank, 2014). In fact, the sequenced genome (Shulaev et al., 2011) was obtained from a U.S. Department of Agriculture (USDA) accession "Hawaii 4" originally collected as a naturalized plant near Volcanoes National Park in Hawaii. It is also established on other oceanic islands and in the South American Andes (Table 2). The available genomic resources for this subspecies make it a promising candidate for characterizing the molecular mechanisms of weediness, as well as the impact of isolation on genetic drift.

Most, if not all, flowering plants are polyploids, having undergone one or more whole genome duplications, making polyploidy the single most important mechanism of speciation in land plants (Otto and Whitton, 2000; Wood et al., 2009; Jiao et al., 2011). Polyploidy is a revolutionary event that induces drastic changes in both genome organization and ecological properties that can lead not only to speciation, but biological adaptation and range expansion (Stebbins, 1985; Vamosi and Dickinson, 2006; Pandit et al., 2011; Parisod, 2012)—perhaps especially in response to rapid environmental change (Dynesius and Jansson, 2000; Fawcett et al., 2009).

Despite long-held views that polyploids are more successful because they have enhanced tolerance to abiotic stress and greater ecological amplitude, this issue remains controversial (Martin and Husband, 2009; Soltis et al., 2010; Madlung, 2013). A recent phylogenetically controlled analysis of bioclimatic niche and range size for 21 *Fragaria* species (Johnson et al., 2014) shows high levels of divergence in bioclimatic niches, and even significant overdispersion along some bioclimatic gradients, suggesting extensive evolutionary lability in physiology and climate tolerance among species. Indeed, the absence of niche conservativism and/or high selfing ability could contribute to wide range breadth and even weediness of the diploid *F. vesca* subsp. *vesca* (Table 2). Moreover, although polyploids had larger mean area and more northerly position of their ranges than diploids, there was no significant difference between the two groups, possibly due to the opposing forces of breeding system and polyploidy or the complicating factors of species age and geographic barriers (Johnson et al., 2014).

While bioclimatic studies can provide a snapshot of interspecific distributions at a global scale, only integrated genetic, functional, and population studies can determine whether the proposed mechanisms that follow polyploidization/hybridity contribute to ecological amplitude and stress tolerance of polyploids independent of the contributions of multiple origins or age of species on range size. The potential role of the proposed mechanisms for polyploid fitness (e.g., increased gene dosage, biochemical activity, cell and organ size, increased gene or biochemical diversity or plasticity, genetic novelty arising from transgressive segregation or genome restructuring; reviewed by Soltis et al. [2010]) can all be readily evaluated in *Fragaria* (see following section and Table 2).

Genomic repatterning that follows whole genome duplication is also thought to be an important source of genetic diversity that fuels adaptive evolution in polyploids (Hegarty and Hiscock, 2008). As a result, intensive focus has been placed on characterizing the effects of allopolyploidy on genome structure or gene expression in a handful of taxa (e.g., *Brassica*, *Glycine* [soy], *Gossypium* [cotton], *Tragopogon, Triticum* [wheat]). This effort has revealed significant structural changes, including deletions, translocations, and transpositions in both paleo- and neopolyploids (Song et al., 1995; Ozkan et al., 2001; Chen and Ni, 2006; Gaeta and Pires, 2010). Linkage mapping results to date in *Fragaria* suggests a high degree of synteny among the octoploid subgenomes in *F. virginiana* (Spigler et al., 2010) and *F.* ×*ananassa* (Isobe et al., 2013). However, evidence for inversions confined to single homoeologs has been reported in *F.* ×*ananassa* (Sargent et al., 2012; van Dijk et al., 2014). These linkage maps were all created with microsatellite markers. The utilization of targeted sequence capture (Tennessen et al., 2013) for genetic linkage mapping has the advantage of also providing sequence data from each mapped locus. These sequences can be used to discriminate among octoploid subgenomes, providing for the first time the opportunity to quantify aspects of genome repatterning in high polyploid *Fragaria* (J. A. Tennessen et al., Oregon State University, unpublished manuscript).

Diploid and polyploid *Fragaria* species often grow sympatrically (Hancock and Bringhurst, 1981; Fig. 2G); thus, the potential for interspecific hybridization exists (Table 2). Naturally occurring pentaploids have been reported from California (*F. vesca* × *F. chiloensis,* Bringhurst and Senanayake, 1966) and Northeast China (presumably *F. mandshurica* × *F. orientalis*; Lei et al., 2005). In Europe, both 5*x* and 7*x* individuals of *F. vesca* × *F. moschata* been reported (Nosrati et al., 2011a, 2013), while a single 9*x* individual of *F. vesca* × *F. chiloensis* has been observed in California (Bringhurst and Senanayake, 1966). In each of these cases, the odd polyploid hybrids co-occur with the parental species, and their persistence is likely attributable to clonal reproduction. For this reason, they are not included as "species" in Table 1.

Interspecific hybridization also occurs within a ploidy level. In Europe, hybrids of diploid *F. vesca* and *F. viridis* have been named *F.* ×*bifera*, and probably can occur wherever the two parents are present (Staudt et al., 2003). In northwestern North America, a large zone of introgression occurs between the octoploids *F. chiloensis* and *F. virginiana* (Luby et al., 1992; Salamone et al., 2013). In the case of *F.* ×*bifera*, the frequency and ecological context of hybridization has not been quantified. In the case of the octoploid hybrids (*F.* ×*ananassa* subsp. *cuneifolia*), the stability and potential adaptive significance of the introgression zone remains unexplored. In general, varying degrees of reproductive isolation within and among ploidy levels (Bors and Sullivan, 2005; Nosrati et al., 2011b) provides a rich opportunity to investigate the molecular mechanisms of reproductive isolation and the potential role of speciation genes (Table 2).

An additional feature of *Fragaria* biology is the ability for clonal growth. Ecological (Alpert, 1991) and molecular genetic (Hytönen et al., 2009; Koskela et al., 2012) studies have provided insights into the functional pathways that control allocation between sexual and asexual reproduction. Resource transfer among ramets has also been demonstrated to have a genetic basis (Alpert et al., 2003). Clonality may also play a role in facilitating the survival and establishment of new polyploids in *Fragaria.* An initially sterile hybrid of *F. chiloensis* × *F. virginiana* (no flowers between 1923 and 1939 and male-sterile flowers from 1939 to 1951) produced its first fertile flowers after nearly 30 yr (Schiemann, 1958). Not mentioned in the paper is the miraculous survival of this clone in Berlin throughout the Second World War! The responsible genetic mechanism remains unknown, and the experiment would be a challenge to replicate.

GENE EXPRESSION AND TRANSCRIPTOMICS

With the publication of the genome of *F. vesca* (Shulaev et al., 2011), a comprehensive catalog of expressed genes are available for *Fragaria.* Through a combination of gene prediction models and transcriptome evidence, the diploid *F. vesca* genome is estimated to contain 34 809 predicted protein coding genes, and annotations are available for 25 050 genes (Shulaev et al., 2011). Over 90% of predicted genes are supported by transcript evidence, and extensive efforts to define organ- and tissue-specific expression have drawn from a large number of independent libraries (Bombarely et al., 2010; Folta et al., 2010; Darwish et al., 2013). Nearly 1125 different non-translated RNAs have also been characterized, including transfer RNAs, ribosomal RNAs, small RNAs, and microRNAs, with most efforts focused on diploid *F. vesca* (Shulaev et al., 2011) and octoploid *Fragaria* ×*ananassa* (Ge et al., 2013). Short RNAs are exceedingly diverse in *F.* × *ananassa* (Ge et al., 2013), and they far outnumber the known classes of small RNAs, accounting for 96.4% of the sRNA pool. Given the recent discovery of diverse phenotypes ascribed to small ORFs in *Arabidopsis* (Hanada et al., 2013), this uncharacterized pool of RNAs may be an untapped resource for understanding the genetic control of agronomic traits, leaf and flower morphogenesis, and different attributes of fitness.

The vast majority of gene expression and transcriptomic studies in *Fragaria* have focused on agronomic traits, although increasing attention is being given to the transcriptomic (and related proteomic and metabolomics) components of climate sensing and climatic adaptation. Fruit traits are an important focal point in domesticated strawberry, and several transcriptomic studies have focused on aspects of fruit development, including the role of hormone signaling in receptacle enlargement (Kang et al., 2013), the role of MYBs transcription factors and phenylpropanoids in fruit development and ripening (Muñoz et al., 2011; Medina-Puche et al., 2014), and transcript coexpression during receptacle ripening (Hollender et al., 2014). Stress-related expression responses have been measured under a number of agronomically relevant treatments (osmotic stress, water saturation, high temperature, cold temperature), and a number of pathways have been implicated in these different responses (Rivarola et al., 2011).

The process of cold acclimation, winter hardiness and desiccation tolerance, and the timing of spring growth and fall dormancy are all key components of yield and fitness, and they are likely to show strong gradients in natural populations of widespread *Fragaria* species. To date, transcriptomic approaches have not been used to study gene expression over the span of a complete growing cycle. Focused studies on diploid *F. vesca* have dissected the role of key seasonal "integration" genes (e.g., SOC1; Mouhu et al., 2013) that regulate yearly cycles of vegetative and reproductive development. Detailed studies of gene expression during the induction of dormancy show that phytohormones promote changes in DNA methylation (Zhang et al., 2012), and these types of transcriptomic responses are amenable to transcriptomic dissection. Recent examinations of metabolite (Rohloff et al., 2012) and proteomic changes (Koehler et al., 2012) during cold acclimation have identified the proteins and metabolites that participate in cold acclimation, and genotypic differences between populations and cultivars in these gene products. These types of quantitative phenotypes are easily merged with transcriptomic studies, and they have the potential to reveal the order of steps behind the orchestration of perennial cycles of dormancy and growth.

With the ability to create synthetic polyploids (Kwok, 2013; Zhang et al., 2014) and to measure environmental responses with genetically identical (clonal) individuals across diverse environments, combined with the availability of genetic and genomic resources, *Fragaria* is an ideal system for exploring the relationship between ploidy formation, ploidy level, and the coordination of transcriptomic control. The processes and rules that coordinate gene expression in *Fragaria* polyploid genomes remain largely unexplored, and the contribution of multiple genomes and multiplicative sets of genes—perhaps as many as 140 000 protein coding genes in the octoploid species—to reproductive fitness and climatic adaptation are equally obscure. The myriad of genetic and epigenetic processes known to modify gene expression patterns in polyploids is diverse and includes structural rearrangement and gene conversion (Ozkan et al., 2001; Salmon et al., 2010; Buggs et al., 2012), epigenetic repatterning (Madlung et al., 2002), subgenomic expression dominance (Adams et al., 2003; Grover et al., 2012; Xu et al., 2014), and possible interactions with small RNAs (Ha et al., 2009). The extent to which these mechanisms operate in polyploid *Fragaria* is unknown, but the availability of synthetic and natural polyploids offers the ability to examine the importance of these mechanisms to *Fragaria* polyploids overall, and to determine whether specific mechanisms are correlated with successive ploidy levels. In light of the young age of the genus, high sequence similarity across the genomes of *Fragaria* species (and subgenomes of polyploid species) will make it difficult to discriminate among "alloalleles" and identify the subgenomic origin for specific transcripts. However, even if only one subgenome can be reliably identified in a polyploid, it still provides a testable scenario for the null hypothesis of equal contribution of subgenomes to the transcript pool. In this context, experiments focused on highly divergent sequence regions, such as 3′ untranslatable regions (UTRs) (Beck et al., 2010) or known alloallelic single nucleotide polymorphisms (SNPs) (Tennessen et al., 2013), seem the most likely to identify transcripts to a genome of origin.

CONCLUSION

In contrast to more established "model plants" in ecology and evolution, such as *Mimulus* (Wu et al., 2008), *Silen*e (Bernasconi et al., 2009), *Aquilegia* (Kramer, 2009), and *Boechera* (Rushworth et al., 2011), *Fragaria* includes a species of agricultural importance. Research involving wild strawberry species is far from unique in being able to capitalize on the genetic and genomic resources obtained for plants of economic importance. Studies of the diverse wild relatives of cultivated *Helianthus* (sunflower) have made enormous contributions to our knowledge of hybrid speciation (Rieseberg et al., 2003), while our understanding of polyploidy has been greatly illuminated by research on wild species of *Glycine* and *Gossypium* (Doyle et al., 2004; Adams and Wendel, 2005). Ecological and evolutionary genomics in *Fragaria* has the potential to provide further insights into both hybridization and polyploidy, while uniquely presenting the opportunity to investigate sexual system evolution. In addition to benefiting from the efforts of a large research community focused on the cultivated strawberry, the familiarity of strawberries provides an opportunity to engage and educate the public about botanical research (e.g., Gonda et al., in press). Ultimately, the increased understanding of plant evolution in an ecological context obtained through the study of wild relatives of crop species can contribute essential information that will be needed to adapt agriculture to a changing global environment.

LITERATURE CITED

Adams, K. L., R. Cronn, R. Percifield, and J. F. Wendel. 2003. Genes duplicated by polyploidy show unequal contributions to the transcriptome and organ-specific reciprocal silencing. *Proceedings of the National Academy of Sciences, USA* 100: 4649–4654.

Adams, K. L., and J. F. Wendel. 2005. Polyploidy and genome evolution in plants. *Current Opinion in Plant Biology* 8: 135–141.

Ahmadi, H., and R. S. Bringhurst. 1991. Genetics of sex expression in *Fragaria* species. *American Journal of Botany* 78: 504–514.

Alpert, P. 1991. Nitrogen sharing among ramets increases clonal growth in *Fragaria chiloensis*. *Ecology* 72: 69–80.

Alpert, P., C. Holzapfel, and C. Slominski. 2003. Differences in performance between genotypes of *Fragaria chiloensis* with different degrees of resource sharing. *Journal of Ecology* 91: 27–35.

Ashman, T.-L. 1999. Determinants of sex allocation in a gynodioecious wild strawberry: Implications for the evolution of dioecy and sexual dimorphism. *Journal of Evolutionary Biology* 12: 648–661.

Ashman, T.-L. 2002. The role of herbivores in the evolution of separate sexes from hermaphroditism. *Ecology* 83: 1175–1184.

Ashman, T.-L. 2005. The limits on sexual dimorphism in vegetative traits in a gynodioecious plant. *American Naturalist* 166: S5–S16.

Ashman, T.-L. 2006. The evolution of separate sexes: A focus on the ecological context. *In* L. D. Harder and S. C. H. Barrett [eds.], The ecology and evolution of flowers, 204–222. Oxford University Press, Oxford, UK.

Ashman, T.-L., D. Cole, and M. Bradburn. 2004. Sex-differential resistance and tolerance to a flower-bud clipping weevil in a gynodioecious wild strawberry: Implications for floral and sexual system evolution. *Ecology* 85: 2550–2559.

Ashman, T.-L., R. Cronn, A. Liston, J. Li, and M. Dong. 2014. Wild strawberry: Dimensions of biodiversity US–China project. Website http://wildstrawberry.org/ [accessed 03 March 2014].

Ashman, T.-L., A. Kwak, and B. C. Husband. 2013. Revisiting the dioecy–polyploidy association: Alternate pathways and research opportunities. *Cytogenetic and Genome Research* 140: 241–255.

Ashman, T.-L., and L. Penet. 2007. Direct and indirect effects of a sex-biased antagonist on male and female fertility: Consequences for reproductive trait evolution in a gender dimorphic plant. *American Naturalist* 169: 595–608.

Ashman, T.-L., R. B. Spigler, M. T. Goldberg, and R. Govindarajulu. 2012. *Fragaria*: A polyploid lineage for understanding sex chromosome evolution. *In* R. Navajas-Pérez [ed.], New insights on plant sex chromosomes, 67–90. Nova Science, Hauppauge, New York, USA.

Bachtrog, D., M. Kirkpatrick, J. Mank, J. C. Pires, W. Rice, and N. Valenzuela. 2011. Are all sex chromosomes created equal? *Trends in Genetics* 27: 350–357.

Baker, H. G. 1984. Some functions of dioecy in seed plants. *American Naturalist* 124: 149–158.

Beck, A. H., Z. Weng, D. M. Witten, S. Zhu, J. W. Foley, P. Lacroute, C. L. Smith, et al. 2010. 3′-end sequencing for expression quantification (3SEQ) from archival tumor samples. *PLOS ONE* 5: e8768.

Bernasconi, G., J. Antonovics, A. Biere, D. Charlesworth, L. F. Delph, D. Filatov, T. Giraud, et al. 2009. *Silene* as a model system in ecology and evolution. *Heredity* 103: 5–14.

Bevan, M. W., and C. Uauy. 2013. Genomics reveals new landscapes for crop improvement. *Genome Biology* 14: 206.

Bombarely, A., C. Merchante, F. Csukasi, E. Cruz-Rus, J. L. Caballero, N. Medina-Escobar, R. Blanco-Portales, et al. 2010. Generation and analysis of ESTs from strawberry (*Fragaria ×ananassa*) fruits and evaluation of their utility in genetic and molecular studies. *BMC Genomics* 11: 503.

Boriss, H., H. Brunke, and M. Kreith. 2006. Commodity profile: Strawberries. Agricultural Issues Center, University of California, Davis, California, USA. Website http://aic.ucdavis.edu/profiles/Strawberries-2006.pdf [accessed 25 March 2014].

Bors, R. H. 2000. A streamlined synthetic octoploid system that emphasizes *Fragaria vesca* as a bridge species. Ph.D. dissertation, University of Guelph, Guelph, Ontario, Canada.

Bors, R. H., and J. A. Sullivan. 2005. Interspecific hybridization of *Fragaria moschata* with two diploid species, *F. nubicola* and *F. viridis. Euphytica* 143: 201–207.

Bošković, R. I., D. J. Sargent, and K. R. Tobutt. 2010. Genetic evidence that two independent *S*-loci control RNase-based self-incompatibility in diploid strawberry. *Journal of Experimental Botany* 61: 755–763.

Bringhurst, R. S., and T. Gill. 1970. Origin of *Fragaria* polyploids. II. Unreduced and doubled-unreduced gametes. *American Journal of Botany* 57: 969–976.

Bringhurst, R. S., and Y. D. A. Senanayake. 1966. The evolutionary significance of natural *Fragaria chiloensis* × *F. vesca* hybrids resulting from unreduced gametes. *American Journal of Botany* 53: 1000–1006.

Buggs, R. J. A., S. Chamala, W. Wu, J. A. Tate, P. S. Schnable, D. E. Soltis, P. S. Soltis, and W. B. Barbazuk. 2012. Rapid, repeated, and clustered loss of duplicate genes in allopolyploid plant populations of independent origin. *Current Biology* 22: 248–252.

Chabot, B. F., and J. F. Chabot. 1977. Effects of light and temperature on leaf anatomy and photosynthesis in *Fragaria vesca. Oecologia* 26: 363–377.

Chambers, A., S. Carle, W. Njuguna, S. Chamala, N. Bassil, V. M. Whitaker, W. B. Barbazuk, and K. M. Folta. 2013. A genome-enabled, high-throughput, and multiplexed fingerprinting platform for strawberry (*Fragaria* L.). *Molecular Breeding* 31: 615–629.

Charlesworth, D., B. Charlesworth, and G. Marais. 2005. Steps in the evolution of heteromorphic sex chromosomes. *Heredity* 95: 118–128.

Chauvet, M. 2003. Foreword. *In* G. Staudt, Les dessins d'Antoine Nicolas Duchesne pour son Histoire naturelle des fraisiers, 16–23. National Museum of Natural History, Paris. Also at website http://uses.plantnet-project.org/en/Foreword_to_Duchesne%27s_strawberry_drawings [accessed 10 March 2014].

Chen, Z. J., and Z. Ni. 2006. Mechanisms of genomic rearrangements and gene expression changes in plant polyploids. *BioEssays* 28: 240–252.

Chukhina, I. G. 2008. *Fragaria. In* A. N. Afonin, S. L. Greene, N. I. Dzyubenko, and A. N. Frolov [eds.], Interactive agricultural ecological atlas of Russia and neighboring countries [online]. Website http://www.agroatlas.ru [accessed 29 March 2014].

Darrow, G. M. 1966. Strawberry: History, breeding and physiology. Holt, Rinehart & Winston, New York, New York, USA. Also at website http://specialcollections.nal.usda.gov/speccoll/collectionsguide/darrow/Darrow_TheStrawberry.pdf.

Darwish, O., J. P. Slovin, C. Kang, C. A. Hollender, A. Geretz, S. Houston, Z. Liu, and N. W. Alkharouf. 2013. SGR: An online genomic resource for the woodland strawberry. *BMC Plant Biology* 13: 223.

DiMeglio, L. M., G. Staudt, H. Yu, and T. M. Davis. 2014. A phylogenetic analysis of the genus *Fragaria* (strawberry) using intron-containing sequence from the *ADH*-1 gene. *PLOS ONE* 9: e102237.

Doyle, J. J., J. L. Doyle, J. T. Rauscher, and A. H. D. Brown. 2004. Diploid and polyploid reticulate evolution throughout the history of the perennial soybeans (*Glycine* subgenus *Glycine*). *New Phytologist* 161: 121–132.

Duchesne, A. N. 1766. Histoire naturelle des fraisiers. Didot le jeune, Paris, France. Also at website http://reader.digitale-sammlungen.de/resolve/display/bsb10301393.html.

Dynesius, M., and R. Jansson. 2000. Evolutionary consequences of changes in species' geographical distributions driven by Milankovitch climate oscillations. *Proceedings of the National Academy of Sciences, USA* 97: 9115–9120.

Environmental Working Group. 2013. EWG's 2013 Shopper's guide to pesticides in produce. Website http://www.ewg.org/foodnews/summary.php [accessed 26 March 2014].

Eriksson, T., M. J. Donoghue, and M. S. Hibbs. 1998. Phylogenetic analysis of *Potentilla* using DNA sequences of nuclear ribosomal internal transcribed spacers (ITS), and implications for the classification of Rosoideae (Rosaceae). *Plant Systematics and Evolution* 211: 155–179.

Evans, W. D. 1977. The use of synthetic octoploids in strawberry breeding. *Euphytica* 26: 497–503.

Fawcett, J. A., S. Maere, and Y. de Peer. 2009. Plants with double genomes might have had a better chance to survive the Cretaceous Tertiary extinction event. *Proceedings of the National Academy of Sciences, USA* 106: 5737–5742.

Finn, C. E., J. B. Retamales, G. A. Lobos, and J. F. Hancock. 2013. The Chilean strawberry (*Fragaria chiloensis*): Over 1000 years of domestication. *HortScience* 48: 418–421.

Folta, K. M. 2013. Functionalizing the strawberry genome—A review. *International Journal of Fruit Science* 13: 162–174.

Folta, K. M., M. A. Clancy, S. Chamala, A. M. Brunings, A. Dhingra, L. Gomide, R. J. Kulathinal, et al. 2010. A transcript accounting from diverse tissues of a cultivated strawberry. *Plant Genome* 3: 90–105.

Gaeta, R. T., and J. C. Pires. 2010. Homoeologous recombination in allopolyploids: The polyploid ratchet. *New Phytologist* 186: 18–28.

GBIF [Global Biodiversity Information Facility]. 2014. GBIF Data Portal. Website http://www.gbif.org [accessed 03 March 2014].

Ge, A., L. Shangguan, X. Zhang, Q. Dong, J. Han, H. Liu, X. Wang, and J. Fang. 2013. Deep sequencing discovery of novel and conserved microRNAs in strawberry (*Fragaria×ananassa*). *Physiologia Plantarum* 148: 387–396.

GenomeTV. 2010. How to extract DNA from strawberries. Website http://www.youtube.com/watch?v=hOpu4iN5Bh4 [accessed 26 March 2014].

Giampieri, F., S. Tulipani, J. M. Alvarez-Suarez, J. L. Quiles, B. Mezzetti, and M. Battino. 2012. The strawberry: Composition, nutritional quality, and impact on human health. *Nutrition* 28: 9–19.

Givnish, T. J. 1980. Ecological constraints on the evolution of breeding systems in seed plants: Dioecy and dispersal in gymnosperms. *Evolution* 34: 959–972.

Goldberg, M. T., R. B. Spigler, and T. L. Ashman. 2010. Comparative genetic mapping points to different sex chromosomes in sibling species of wild strawberry (*Fragaria*). *Genetics* 186: 1425–1433.

Gonda, R., K. DeHart, T.-L. Ashman, and A. Slinsky-Legg. In press. The strawberry caper: Using scenario-based problem solving to integrate middle school science topics. *American Biology Teacher*.

Govindarajulu, R., A. Liston, and T. L. Ashman. 2013. Sex-determining chromosomes and sexual dimorphism: Insights from genetic mapping of sex expression in a natural hybrid *Fragaria ×ananassa* subsp. *cuneifolia. Heredity* 110: 430–438.

Gremillion, K. J., and K. D. Sobolik. 1996. Dietary variability among prehistoric forager-farmers of Eastern North America. *Current Anthropology* 37: 529–539.

Grover, C. E., J. P. Gallagher, E. P. Szadkowski, M. J. Yoo, L. E. Flagel, and J. F. Wendel. 2012. Homoeolog expression bias and expression level dominance in allopolyploids. *New Phytologist* 196: 966–971.

Ha, M., J. Lu, L. Tian, V. Ramachandran, K. D. Kasschau, E. J. Chapman, J. C. Carrington, et al. 2009. Small RNAs serve as a genetic buffer against genomic shock in *Arabidopsis* interspecific hybrids and allopolyploids. *Proceedings of the National Academy of Sciences, USA* 106: 17835–17840.

Hanada, K., M. Higuchi-Takeuchi, M. Okamoto, T. Yoshizumi, M. Shimizu, K. Nakaminami, R. Nishi, et al. 2013. Small open reading frames associated with morphogenesis are hidden in plant genomes. *Proceedings of the National Academy of Sciences, USA* 110: 2395–2400.

Hancock, J. F. 1999. Strawberries. CAB International, Wallingford, UK.

Hancock, J. F., and R. S. Bringhurst. 1978. Inter-populational differentiation and adaptation in the perennial, diploid species *Fragaria vesca* L. *American Journal of Botany* 65: 795–803.

Hancock, J. F., and R. S. Bringhurst. 1979. Ecological differentiation in perennial octoploid species of *Fragaria. American Journal of Botany* 66: 367–375.

Hancock, J. F., and R. S. Bringhurst. 1981. Evolution in California populations of diploid and octoploid *Fragaria* (Rosaceae): A comparison. *American Journal of Botany* 68: 1–5.

Hancock, J. F., and J. J. Luby. 1993. Genetic resources at our doorstep: The wild strawberries. *BioScience* 43: 141–147.

Hegarty, M. J., and S. J. Hiscock. 2008. Genomic clues to the evolutionary success of polyploid plants. *Current Biology* 18: R435–R444.

Hirakawa, H., K. Shirasawa, S. Kosugi, K. Tashiro, S. Nakayama, M. Yamada, M. Kohara, et al. 2014. Dissection of the octoploid strawberry genome by deep sequencing of the genomes of *Fragaria* species. *DNA Research* 21: 169–181.

Hollender, C.A., C. Kang, O. Darwish, A. Geretz, B.F. Matthews, J. Slovin, N. Alkharouf, and Z. Liu. 2014. Floral transcriptomes in woodland strawberry uncover developing receptacle and anther gene networks. *Plant Physiology* 165: 1062–1075.

Hummer, K. E. 2012. A new species of *Fragaria* (Rosaceae) from Oregon. *Journal of the Botanical Research Institute of Texas* 6: 9–15.

Hummer, K. E., N. Bassil, and W. Njuguna. 2011. Fragaria. *In* C. Kole [ed.], Wild crop relatives: Genomic and breeding resources, 17–44. Springer, Berlin, Germany.

Hummer, K. E., P. Nathewet, and T. Yanagi. 2009. Decaploidy in *Fragaria iturupensis* (Rosaceae). *American Journal of Botany* 96: 713–716.

Hytönen, T., P. Elomaa, T. Moritz, and O. Junttila. 2009. Gibberellin mediates daylength-controlled differentiation of vegetative meristems in strawberry (*Fragaria* × *ananassa* Duch). *BMC Plant Biology* 9: 18.

Ichijima, K. 1926. Cytological and genetic studies on *Fragaria. Genetics* 11: 590–604.

Isobe, S. N., H. Hirakawa, S. Sato, F. Maeda, M. Ishikawa, T. Mori, Y. Yamamoto, et al. 2013. Construction of an integrated high density simple sequence repeat linkage map in cultivated strawberry (*Fragaria* × *ananassa*) and its applicability. *DNA Research* 20: 79–92.

Jiao, Y., N. J. Wickett, S. Ayyampalayam, A. S. Chanderbali, L. Landherr, P. E. Ralph, L. P. Tomsho, et al. 2011. Ancestral polyploidy in seed plants and angiosperms. *Nature* 473: 97–100.

Jiwan, D., E. H. Roalson, D. Main, and A. Dhingra. 2013. Antisense expression of peach mildew resistance locus O (*PpMlo1*) gene confers cross-species resistance to powdery mildew in *Fragaria* × *ananassa. Transgenic Research* 22: 1119–1131.

Johnson, A. L., R. Govindarajulu, and T.-L. Ashman. 2014. Bioclimatic evaluation of range in *Fragaria* (Rosaceae): Consequences of variation in mating system, ploidy and species age. *Botanical Journal of the Linnean Society* 176: 99–114.

Jurik, T. W., J. F. Chabot, and B. F. Chabot. 1982. Effects of light and nutrients on leaf size, CO_2 exchange, and anatomy in wild strawberry (*Fragaria virginiana*). *Plant Physiology* 70: 1044–1048.

Kang, C., O. Darwish, A. Geretz, R. Shahan, N. Alkharouf, and Z. Liu. 2013. Genome-scale transcriptomic insights into early-stage fruit development in woodland strawberry *Fragaria vesca. Plant Cell Online* 25: 1960–1978.

Kellerman, W. A. 1892. Interesting variations of the strawberry leaf. *Botanical Gazette* 17: 257–258.

Kihara, H. 1930. Karyologische Studien an *Fragaria* mit besonderer Beruksichtigung der Geschlechtschromosomen. *Cytologia* 1: 345–357.

Koehler, G., R. C. Wilson, J. V. Goodpaster, A. Sønsteby, X. Lai, F. A. Witzmann, J.-S. You, et al. 2012. Proteomic study of low-temperature responses in strawberry cultivars (*Fragaria* × *ananassa*) that differ in cold tolerance. *Plant Physiology* 159: 1787–1805.

Koskela, E. A., K. Mouhu, M. C. Albani, T. Kurokura, M. Rantanen, D. J. Sargent, N. H. Battey, et al. 2012. Mutation in *TERMINAL FLOWER1* reverses the photoperiodic requirement for flowering in the wild strawberry *Fragaria vesca. Plant Physiology* 159: 1043–1054.

Kramer, E. M. 2009. *Aquilegia*: A new model for plant development, ecology, and evolution. *Annual Review of Plant Biology* 60: 261–277.

Kwok, A. 2013. The role of polyploidy in the evolution of gender dimorphism: An experimental approach using *Fragaria vesca*. Ph.D. dissertation, University of Guelph, Guelph, Ontario, Canada.

Lee, V. 1964. Antoine Nicolas Duchesne—First strawberry hybridist. *American Horticultural Magazine* 43: 80–88.

Lee, V. 1966. Duchesne and his work. *In* G. M. Darrow [ed.], Strawberry: History, breeding and physiology, 40–72. Holt, Rinehart & Winston, New York, New York, USA. Website http://specialcollections.nal.usda.gov/speccoll/collectionsguide/darrow/Darrow_TheStrawberry.pdf.

Lei, J., Y. Li, G. Du, H. Dai, and M. Deng. 2005. A natural pentaploid strawberry genotype from the Changbai mountains in northeast China. *HortScience* 40: 1194–1195.

Li, J., M. H. Koski, and T.-L. Ashman. 2012. Functional characterization of gynodioecy in *Fragaria vesca* ssp. *bracteata* (Rosaceae). *Annals of Botany* 109: 545–552.

Lin, J., and T. M. Davis. 2000. S1 analysis of long PCR heteroduplexes: Detection of chloroplast indel polymorphisms in *Fragaria. Theoretical and Applied Genetics* 101: 415–420.

Longhi, S., L. Giongo, M. Buti, N. Surbanovski, R. Viola, R. Velasco, J. A. Ward, and D. J. Sargent. 2014. Molecular genetics and genomics of the Rosoideae: state of the art and future perspectives. *Horticulture Research* 1: 1.

Longley, A. E. 1926. Chromosomes and their significance in strawberry classification. *Journal of Agricultural Research* 32: 559–568.

Luby, J. J., J. F. Hancock, and J. R. Ballington. 1992. Collection of native strawberry germplasm in the Pacific Northwest and Northern Rocky Mountains of the United States. *HortScience* 27: 12–17.

Madlung, A. 2013. Polyploidy and its effect on evolutionary success: Old questions revisited with new tools. *Heredity* 110: 99–104.

Madlung, A., R. W. Masuelli, B. Watson, S. H. Reynolds, J. Davison, and L. Comai. 2002. Remodeling of DNA methylation and phenotypic and transcriptional changes in synthetic *Arabidopsis* allotetraploids. *Plant Physiology* 129: 733–746.

Mangelsdorf, A. J., and E. M. East. 1927. Studies on the genetics of *Fragaria. Genetics* 12: 307–339.

Martin, S. L., and B. C. Husband. 2009. Influence of phylogeny and ploidy on species ranges of North American angiosperms. *Journal of Ecology* 97: 913–922.

Matthews, J. V. Jr. and L. E. Ovenden. 1990. Late Tertiary plant macrofossils from localities in arctic/subarctic North America: A review of the data. *Arctic* 43: 364–392.

Medina-Puche, L., G. Cumplido-Laso, F. Amil-Ruiz, T. Hoffmann, L. Ring, A. Rodríguez-Franco, J. L. Caballero, et al. 2014. MYB10 plays a major role in the regulation of flavonoid/phenylpropanoid metabolism during ripening of *Fragaria* × *ananassa* fruits. *Journal of Experimental Botany* 65: 401–417.

Mezzetti, B. 2009. GMO strawberry: Methods, risk and benefits. *In* K. M. Folta and S. E. Gardner [eds.], Genetics and genomics of Rosaceae, 487–506. Springer, Berlin, Germany.

Miller, J. S., and D. L. Venable. 2000. Polyploidy and the evolution of gender dimorphism in plants. *Science* 289: 2335–2338.

Ming, R., and P. H. Moore. 2007. Genomics of sex chromosomes. *Current Opinion in Plant Biology* 10: 123–130.

Morgan, M. T., and T.-L. Ashman. 2003. Quantitative character evolution under complicated sexual systems, illustrated in gynodioecious *Fragaria virginiana. American Naturalist* 162: 257–264.

Morrison, D. A. 2014. Is the tree of life the best metaphor, model, or heuristic for phylogenetics? *Systematic Biology* 63: 628–638.

Mouhu, K., T. Hytönen, K. Folta, M. Rantanen, L. Paulin, P. Auvinen, and P. Elomaa. 2009. Identification of flowering genes in strawberry, a perennial SD plant. *BMC Plant Biology* 9: 122.

Mouhu, K., T. Kurokura, E. A. Koskela, V. A. Albert, P. Elomaa, and T. Hytönen. 2013. The *Fragaria vesca* homolog of suppressor of overexpression of *constans1* represses flowering and promotes vegetative growth. *The Plant Cell* 25: 3296–3310.

Muñoz, C., J. F. Sánchez-Sevilla, M. A. Botella, T. Hoffmann, W. Schwab, and V. Valpuesta. 2011. Polyphenol composition in the ripe fruits of *Fragaria* species and transcriptional analyses of key genes in the pathway. *Journal of Agricultural and Food Chemistry* 59: 12598–12604.

Njuguna, W., A. Liston, R. Cronn, T. L. Ashman, and N. Bassil. 2013. Insights into phylogeny, sex function and age of *Fragaria* based on whole chloroplast genome sequencing. *Molecular Phylogenetics and Evolution* 66: 17–29.

Nosrati, H., A. H. Price, P. Gerstberger, and C. C. Wilcock. 2011a. Identification of a natural allopentaploid hybrid *Fragaria* (Rosaceae), new to Europe. *New Journal of Botany* 1: 88–92.

Nosrati, H., A. H. Price, P. Gerstberger, and C. C. Wilcock. 2013. The first report of an alloheptaploid from the genus *Fragaria* (Rosaceae). *New Journal of Botany* 3: 205–209.

Nosrati, H., A. H. Price, and C. C. Wilcock. 2011b. Relationship between genetic distances and postzygotic reproductive isolation in diploid *Fragaria* (Rosaceae). *Biological Journal of the Linnean Society* 104: 510–526.

Oosumi, T., H. A. Gruszewski, L. A. Blischak, A. J. Baxter, P. A. Wadl, J. L. Shuman, R. E. Veilleux, and V. Shulaev. 2006. High-efficiency transformation of the diploid strawberry (*Fragaria vesca*) for functional genomics. *Planta* 223: 1219–1230.

Otto, S. P., J. Pannell, C. L. Peichel, T.-L. Ashman, D. Charlesworth, A. K. Chippendale, L. F. Delph, et al. 2011. About PAR: The distinct evolutionary dynamics of the Pseudoautosomal Region. *Trends in Genetics* 27: 358–367.

Otto, S. P., and J. Whitton. 2000. Polyploid incidence and evolution. *Annual Review of Genetics* 34: 401–437.

Ozkan, H., A. A. Levy, and M. Feldman. 2001. Allopolyploidy-induced rapid genome evolution in the wheat (*Aegilops–Triticum*) group. *Plant Cell* 13: 1735–1747.

Pandit, M. K., M. J. O. Pocock, and W. E. Kunin. 2011. Ploidy influences rarity and invasiveness in plants. *Journal of Ecology* 99: 1108–1115.

Paris, H. 2000. Paintings (1769–1774) by A. N. Duchesne and the history of *Cucurbita pepo. Annals of Botany* 85: 815–830.

Parisod, C. 2012. Polyploids integrate genomic changes and ecological shifts. *New Phytologist* 193: 297–300.

Pauketat, T. R., L. S. Kelly, G. J. Fritz, N. H. Lopinot, S. Elias, and E. Hargrave. 2002. The residues of feasting and public ritual at Early Cahokia. *American Antiquity* 67: 257–279.

Plantwise Knowledge Bank. 2014. *Fragaria vesca.* Website http://www.plantwise.org/KnowledgeBank/Datasheet.aspx?dsid=24409 [accessed 26 March 2014].

Ratcliff, M. J. 2007. Duchesne's strawberries: Between growers' practices and academic knowledge. *In* S. Müller-Wille and H.-J. Rheinberger [eds.], Heredity produced: At the crossroads of biology, politics, and culture, 1500–1870, 205–228. MIT Press, Cambridge, Massachusetts, USA.

Razavi, F., E. De Keyser, J. De Riek, and M. C. Van Labeke. 2011. A method for testing drought tolerance in *Fragaria* based on fast screening for water deficit response and use of associated AFLP and EST candidate gene markers. *Euphytica* 180: 385–409.

Renner, S. S., and R. E. Ricklefs. 1995. Dioecy and its correlates in the flowering plants. *American Journal of Botany* 82: 596–606.

Richardson, C. W. 1914. A preliminary note on the genetics of *Fragaria. Journal of Genetics* 3: 171–177.

Rieseberg, L. H., O. Raymond, D. M. Rosenthal, Z. Lai, K. Livingstone, T. Nakazato, J. L. Durphy, et al. 2003. Major ecological transitions in wild sunflowers facilitated by hybridization. *Science* 301: 1211–1216.

Rivarola, M., A. P. Chan, D. E. Liebke, A. Melake-Berhan, H. Quan, F. Cheung, S. Ouyang, et al. 2011. Abiotic stress-related expressed sequence tags from the diploid strawberry *Fragaria vesca* f. *semperflorens. Plant Genome* 4: 12–23.

Rohloff, J., J. Kopka, A. Erban, P. Winge, R. C. Wilson, A. M. Bones, J. Davik, et al. 2012. Metabolite profiling reveals novel multi-level cold responses in the diploid model > *Fragaria vesca* (woodland strawberry). *Phytochemistry* 77: 99–109.

Romandini, S., L. Mazzoni, F. Giampieri, S. Tulipani, M. Gasparrini, N. Locorotondo, B. Mezzetti, and S. Bompadre. 2013. Effects of an acute strawberry (*Fragaria* × *ananassa*) consumption on the plasma antioxidant status of healthy subjects. *Journal of Berry Research* 3: 169–179.

Ross, J. A., J. R. Urton, J. Boland, M. D. Shapiro, and C. L. Peichel. 2009. Turnover of sex chromosomes in the stickleback fishes (Gasterosteidae). *PLOS Genetics* 5: e1000391.

Rousseau-Gueutin, M., A. Gaston, A. Aïnouche, M. L. Aïnouche, K. Olbricht, G. Staudt, L. Richard, and B. Denoyes-Rothan. 2009. Tracking the evolutionary history of polyploidy in *Fragaria* L. (strawberry): New insights from phylogenetic analyses of low-copy nuclear genes. *Molecular Phylogenetics and Evolution* 51: 515–530.

Rushworth, C. A., B.-H. Song, C.-R. Lee, and T. Mitchell-Olds. 2011. *Boechera*, a model system for ecological genomics. *Molecular Ecology* 20: 4843–4857.

Salamone, I., R. Govindarajulu, S. Falk, M. Parks, A. Liston, and T.-L. Ashman. 2013. Bioclimatic, ecological, and phenotypic intermediacy and high genetic admixture in a natural hybrid of octoploid strawberries. *American Journal of Botany* 100: 939–950.

Salmon, A., L. Flagel, B. Ying, J. A. Udall, and J. F. Wendel. 2010. Homoeologous nonreciprocal recombination in polyploid cotton. *New Phytologist* 186: 123–134.

Sargent, D. J., T. Passey, N. Surbanovski, E. Lopez Girona, P. Kuchta, J. Davik, R. Harrison, et al. 2012. A microsatellite linkage map for the cultivated strawberry (*Fragaria* × *ananassa*) suggests extensive regions of homozygosity in the genome that may have resulted from breeding and selection. *Theoretical and Applied Genetics* 124: 1229–1240.

Schiemann, E. 1932. Entstehung der Kulturpflanzen. Borntraeger, Berlin, Germany. Translated by E. Midgard. Work Projects Administration No. 3888.

Schiemann, E. 1958. Beobachtungen an einem Subfertilen *Fragaria*-Klon. *Planta* 52: 77–95.

Senanayake, Y. D. A., and R. S. Bringhurst. 1967. Origin of *Fragaria* polyploids. I. Cytological analysis. *American Journal of Botany* 54: 221–228.

Sherman, W. B., J. Janick, and H. T. Erickson. 1966. Inheritance of fruit size in strawberry. *Proceedings of the American Society for Horticultural Science* 89: 309–317.

Shorthouse, D. P. 2010. SimpleMappr, an online tool to produce publication-quality point maps. Website http://www.simplemappr.net [19 March, 2014].

Shulaev, V., S. S. Korban, B. Sosinski, A. G. Abbott, H. S. Aldwinckle, K. M. Folta, A. Iezzoni, et al. 2008. Multiple models for Rosaceae genomics. *Plant Physiology* 147: 985–1003.

Shulaev, V., D. J. Sargent, R. N. Crowhurst, T. C. Mockler, O. Folkerts, A. L. Delcher, P. Jaiswal, et al. 2011. The genome of woodland strawberry (*Fragaria vesca*). *Nature Genetics* 43: 109–116.

Slovin, J. P., and T. P. Michael. 2011. Structural and functional genomics. *In* K. M. Folta and C. Kole [eds.], Genetics, genomics and breeding of berries, 240–308. Science Publishers, Enfield New Hampshire, USA.

Soltis, D. E., R. J. A. Buggs, J. J. Doyle, and P. S. Soltis. 2010. What we still don't know about polyploidy. *Taxon* 59: 1387–1403.

Song, K., K. Tang, and T. C. Osborn. 1995. Rapid genome change in synthetic polyploids of *Brassica* and its implications for polyploidy evolution. *Proceedings of the National Academy of Sciences, USA* 92: 7719–7723.

Sønsteby, A., and O. M. Heide. 2011. Environmental regulation of dormancy and frost hardiness in Norwegian populations of wood strawberry (*Fragaria vesca* L.). *European Journal of Plant Science & Biotechnology* 5: 42–48.

Spigler, R. B., and T. L. Ashman. 2011. Sex ratio and subdioecy in *Fragaria virginiana*: The roles of plasticity and gene flow examined. *New Phytologist* 190: 1058–1068.

Spigler, R. B., and T. L. Ashman. 2012. Gynodioecy to dioecy: Are we there yet? *Annals of Botany* 109: 531–543.

Spigler, R. B., K. S. Lewers, and T. L. Ashman. 2011. Genetic architecture of sexual dimorphism in a subdioecious plant with a proto-sex chromosome. *Evolution* 65: 1114–1126.

Spigler, R. B., K. S. Lewers, A. L. Johnson, and T. L. Ashman. 2010. Comparative mapping reveals autosomal origin of sex chromosome in octoploid *Fragaria virginiana. Journal of Heredity* 101: S107–S117.

Spigler, R. B., K. S. Lewers, D. S. Main, and T. L. Ashman. 2008. Genetic mapping of sex determination in a wild strawberry, *Fragaria virginiana*, reveals earliest form of sex chromosome. *Heredity* 101: 507–517.

Staudt, G. 1999a. Systematics and geographic distribution of the American strawberry species. Taxonomic studies in the genus *Fragaria* (Rosaceae: Potentilleae). University of California Press, Berkeley, California, USA.

Staudt, G. 1999b. Notes on Asiatic *Fragaria* species: *Fragaria nilgerrensis* Schltdl. ex J. Gay. *Botanische Jahrbücher* 121: 297–310.

Staudt, G. 2003a. Les dessins d'Antoine Nicolas Duchesne pour son Histoire naturelle des fraisiers. National Museum of Natural History, Paris, France.

Staudt, G. 2003b. Notes on Asiatic *Fragaria* species: III. *Fragaria orientalis* Losinsk. and *Fragaria mandshurica* spec. nov. *Botanische Jahrbücher* 124: 397–419.

Staudt, G. 2005. Notes on Asiatic *Fragaria* species: IV. *Fragaria iinumae. Botanische Jahrbücher* 126: 163–175.

Staudt, G. 2006. Himalayan species of *Fragaria* (Rosaceae). *Botanische Jahrbücher* 126: 483–508.

Staudt, G. 2008. Strawberry biogeography, genetics and systematics. *Acta Horticulturae* 842: 71–84.

Staudt, G., and W. B. Dickoré. 2001. Notes on Asiatic *Fragaria* species: *Fragaria pentaphylla* Losinsk. and *Fragaria tibetica* spec. nov. *Botanische Jahrbücher* 123: 341–354.

Staudt, G., L. M. DiMeglio, T. M. Davis, and P. Gerstberger. 2003. *Fragaria* × *bifera* Duch.: Origin and taxonomy. *Botanische Jahrbücher* 125: 53–72.

Staudt, G., and K. Olbricht. 2008. Notes on Asiatic *Fragaria* species V: *F. nipponica* and *F. iturupensis. Botanische Jahrbücher* 127: 317–341.

Stebbins, G. L. 1985. Polyploidy, hybridization, and the invasion of new habitats. *Annals of the Missouri Botanical Garden* 72: 824–832.

Stewart, P. J. 2011. *Fragaria* history and breeding. *In* K. M. Folta and Kole [eds.], Genetics, genomics and breeding of berries, 114–137. Science Publishers, Enfield, New Hampshire, USA.

Stewart, P. J., and K. M. Folta. 2010. A review of photoperiodic flowering research in strawberry (*Fragaria* spp.). *Critical Reviews in Plant Sciences* 29: 1–13.

Šurbanovski, N., D. J. Sargent, M. A. Else, D. W. Simpson, H. Zhang, and O. M. Grant. 2013. Expression of *Fragaria vesca* PIP aquaporins in response to drought stress: PIP down-regulation correlates with the decline in substrate moisture content. *PLOS ONE* 8: e74945.

Tennessen, J. A., R. Govindarajulu, A. Liston, and T. Ashman. 2013. Targeted sequence capture provides insight into genome structure and genetics of male sterility in a gynodioecious diploid strawberry, *Fragaria vesca* ssp. *bracteata* (Rosaceae). *Genes|Genomes|Genetics* 3: 1341–1351.

Töpel, M., A. Antonelli, C. Yesson, and B. Eriksen. 2012. Past climate change and plant evolution in western North America: A case study in Rosaceae. *PLOS ONE* 7: e50358.

USDA Economic Research Service. 2012. Most commonly consumed fresh fruits among U.S. consumers. Website http://www.ers.usda.gov/data-products/chart-gallery/detail.aspx?chartId=30486#.UzIYqPldVKc [accessed 26 March 2014].

Vamosi, J. C., and T. A. Dickinson. 2006. Polyploidy and diversification: A phylogenetic investigation in Rosaceae. *International Journal of Plant Sciences* 167: 349–358.

Vamosi, J. C., S. P. Otto, and S. C. H. Barrett. 2003. Phylogenetic analysis of the ecological correlates of dioecy in angiosperms. *Journal of Evolutionary Biology* 16: 1006–1018.

Van Dijk, T., G. Pagliarani, A. Pikunova, Y. Noordijk, H. Yilmaz-Temel, B. Meulenbroek, R. G. Visser, and E. van de Weg. 2014. Genomic rearrangements and signatures of breeding in the allo-octoploid strawberry as revealed through an allele dose based SSR linkage map. *BMC Plant Biology* 14: 55.

Veilleux, R. E., K. P. Mills, A. J. Baxter, K. T. Upham, T. J. Ferguson, S. H. Holt, N. Lu, et al. 2012. Transposon tagging in diploid strawberry. *Plant Biotechnology Journal* 10: 985–994.

Walshaw, S. 2009. *Fragaria* spp. *In* G. Fritz [ed.], Laboratory guide to archaeological plant remains from eastern North America [online]. Website http://artsci.wustl.edu/~gjfritz/Fragaria_spp.html [accessed 26 March 2014].

Webb, C. J. 1999. Empirical studies: Evolution and maintenance of dimorphic breeding systems. *In* M. A. Geber, T. E. Dawson, and L. F. Delph [eds.], Gender and sexual dimorphism in flowering plants, 61–96. Springer, Berlin, Germany.

Wilhelm, S., and J. E. Sagen. 1974. History of the strawberry. Division of Agricultural Sciences, University of California, Berkeley, California, USA.

Wood, T. E., N. Takebayashi, M. S. Barker, I. Mayrose, P. B. Greenspoon, and L. H. Rieseberg. 2009. The frequency of polyploid speciation in vascular plants. *Proceedings of the National Academy of Sciences, USA* 106: 13875–13879.

Wu, C. A., D. B. Lowry, A. M. Cooley, K. M. Wright, Y. W. Lee, and J. H. Willis. 2008. *Mimulus* is an emerging model system for the integration of ecological and genomic studies. *Heredity* 100: 220–230.

Xu, C., Y. Bai, X. Lin, N. Zhao, L. Hu, Z. Gong, J. F. Wendel, and B. Liu. 2014. Genome-wide disruption of gene expression in allopolyploids but not hybrids of rice subspecies. *Molecular Biology and Evolution* 31: 1066–1076.

Zhang, L., Y. Wang, X. Zhang, M. Zhang, D. Han, C. Qiu, and Z. Han. 2012. Dynamics of phytohormone and DNA methylation patterns changes during dormancy induction in strawberry (*Fragaria* × *ananassa* Duch.). *Plant Cell Reports* 31: 155–165.

Zhang, Q., K. M. Folta, and T. M. Davis. 2014. Somatic embryogenesis, tetraploidy, and variant leaf morphology in transgenic diploid strawberry (*Fragaria vesca* subspecies *vesca* "Hawaii 4"). *BMC Plant Biology* 14: 23.