What we still don't know about polyploidy

4 authors, including:

Douglas E Soltis
University of Florida
983 PUBLICATIONS   **54,388** CITATIONS

Richard Buggs
Queen Mary, University of London
121 PUBLICATIONS   **2,648** CITATIONS

Pamela S Soltis
University of Florida
625 PUBLICATIONS   **40,862** CITATIONS

Some of the authors of this publication are also working on these related projects:

Project   Systematics and evolution of Amaryllidaceae tribe Hippeastreae View project

Project   iDigBio View project

What we still don't know about polyploidy

Douglas E. Soltis,[1] Richard J.A. Buggs,[1,4] Jeff J. Doyle[2] & Pamela S. Soltis[3]

1 Department of Biology, University of Florida, Gainesville, Florida 32611, U.S.A.

2 L. H. Bailey Hortorium, Department of Plant Biology, Cornell University, Ithaca, New York 14853, U.S.A.

3 Florida Museum of Natural History, University of Florida, Gainesville, Florida 32611, U.S.A.

4 Present address: School of Biological and Chemical Sciences, Queen Mary University of London, London E1 4NS, U.K.

Author for correspondence: Dougles E. Soltis, dsoltis@ufl.edu

Abstract. During the past decade there has been a tremendous resurgence of interest in polyploidy that has in large part been stimulated by the development of increasingly powerful genetic and genomic tools. The result has been numerous new insights into the genomic and genetic consequences of polyploidy. The plethora of new discoveries has dramatically reshaped traditional views and concomitantly revealed that polyploidy is a highly dynamic and ubiquitous process. These recent advances in our understanding of polyploidy have stimulated numerous reviews, most focused on the various genetic, epigenetic, and genomic consequences of polyploid evolution. Whereas genetic and genomic attributes of polyploidization have received considerable attention, other crucial areas of polyploid evolution have received much less (e.g., ecology, pollination biology, physiology). The focus of this paper is not to review again recent discoveries, but to emphasize *what we do not yet know about polyploidy*, which despite all that has been learned about genome doubling is still an enormous amount. Our list is not meant to be comprehensive, but includes a range of topics that we have placed in several general categories, including mode of formation, ecological and physiological consequences, and genomic rules. Questions include: What is (are) the most frequent mechanism(s) of polyploidization? What factors promote/facilitate polyploidization? What factors favor autopolyploid vs. allopolyploid formation? Do multiple origins result in lineages with differing evolutionary trajectories and/or cryptic species? Our major goals are to stimulate discussion and promote further research.

Keywords please add

■ INTRODUCTION

Polyploidy (often referred to as whole genome duplication, WGD) has long been recognized as a major force in plant evolution. This interest has a long and storied history, beginning with the classic work of De Vries (Lutz, 1907; Gates, 1909), Kuwada (1911) and Winge (1917). A long series of major reviews of polyploidy in plants spans the mid-twentieth century: Müntzing (1936), Darlington (1937), Clausen & al. (1945), Löve & Löve (1949), Stebbins (1947, 1950, 1971), and Grant (1977, 1981). Due to work by these leading evolutionary biologists, polyploidy was a major focus of biosystematic research in plants for several decades.

1971?

Although the potential of molecular data for studying polyploidy was recognized early (e.g., Soltis & al., 1992), there was a lag in research effort on polyploidy as a process during the early phase of the modern era of molecular systematics, as phylogenetic tree reconstruction in the 1990s became the focal point of much of the plant systematic research effort (reviewed in Soltis & al., 2004). However, during the past decade there has been a tremendous resurgence of interest in polyploidy, in large part stimulated by the development of increasingly powerful genetic and genomic tools. The result has been numerous new insights into the genomic and genetic consequences of polyploidy.

Many of these discoveries have dramatically reshaped traditional views and concomitantly revealed that polyploidy is a highly dynamic and ubiquitous process. For example, genomic investigations have suggested that genome doubling is also a major evolutionary force in eukaryotic lineages other than plants, including yeast (Kellis & al., 2004) and many groups of vertebrates and invertebrates (reviewed in Levin, 2002; Gregory & Mable, 2005; Tate & al., 2005; Wendel & Doyle, 2005). Similarly, genomic studies have now revealed that ancient polyploidy may characterize all angiosperm lineages. For example, despite the small size of the *Arabidopsis thaliana* genome, studies of the age distribution and synteny of duplicated genes suggest two or more rounds of ancient genome duplication (Vision & al., 2000; Simillion & al., 2002; Bowers & al., 2003; Thomas & al., 2006). Investigations of the rice genome similarly suggested ancient polyploidy in the early history of the grass family (Poaceae) (Paterson & al., 2004; Yu & al., 2005). Evidence of ancient, genome-wide duplication events is present in many angiosperm genomes (Blanc & Wolfe, 2004a), including angiosperm lineages considered "basal" (Cui & al., 2006). Hence, the question being asked on a broad level is no longer the frequency of polyploidy across the angiosperms, but the number of polyploid events experienced by any given lineage. We stress, however, that it is still of interest to study the frequency of polyploidy in particular genera, families, or larger groups.

These recent advances in our understanding of polyploidy have resulted in many original papers and numerous reviews, most focused on the various genetic, epigenetic, and genomic consequences of polyploid evolution. In fact, reviews from just the past decade are too numerous to include (a partial list includes, D. Soltis & Soltis, 1999; P. Soltis & Soltis, 2000;

Wendel, 2000; Osborn & al., 2003; Liu & Wendel, 2003; volume edited by Leitch & al., 2004; Soltis & al., 2004; Adams & Wendel, 2005; Comai, 2005; Rapp & Wendel, 2005; Tate & al., 2005; Wendel & Doyle, 2005; Chen & Ni, 2006; Udall & Wendel, 2006; Adams, 2007; Chen, 2007; Chen & Tian, 2007; Paun & al., 2007; Rieseberg & Willis, 2007; Doyle & al., 2008; Hegarty & Hiscock, 2008; Leitch & Leitch, 2008; Flagel & Wendel, 2009; Freeling, 2009; Van de Peer & al., 2009; Parisod & al. 2010 and other papers in *New Phytologist* vol. 186 issue 1 devoted to polyploidy).

The focus of this paper is not to review again recent discoveries in the study of polyploidy, but to stress that despite considerable and rapid progress in our understanding of genome doubling in just the past 10–15 years, there is still an enormous amount that *we do not know about polyploidy*. We as a community need to review not only what has been done, but also what research could and should be done in the study of polyploid evolution. Doyle & al. (2008) recently took this approach in a review that focused on gene expression, epigenetics, diploidization, and the fate of duplicated genes in polyploids. Whereas the genetic and genomic attributes of polyploidization have received considerable attention, other crucial areas of research into polyploid evolution have received much less attention (e.g., ecology, pollination biology, and physiology). This limited research in some areas is perhaps not too surprising in that polyploidy is a complex process involving multiple mechanisms in many diverse lineages of organisms. What may be more surprising, however, is that some of the more important and less investigated issues pertaining to polyploid evolution are relatively inexpensive to investigate compared to genomics and other cutting-edge genetics initiatives. There is much still be learned about polyploid evolution "on the cheap."

Here we review what we consider to be some of the more important unanswered questions involving genome doubling. Our list is not meant to be comprehensive, but includes a range of topics that we have placed in several general categories, including mode of formation, ecological and physiological consequences, and genomic rules. Uninvestigated or underinvestigated research areas are posed as a series of questions with a brief discussion of each. We do not mean to play down in any way the importance of excellent work that has been done in less-investigated areas. Our major goals are to stimulate discussion and promote future research. Furthermore, we hope that other researchers will add to this list.

◼ FORMATION/EVOLUTION

How many categories or types of polyploids should be recognized, and what are the most useful criteria for distinguishing these types? — These questions have long been debated and perhaps still not successfully answered (for reviews regarding this debate, see Stebbins, 1950; Grant, 1981; Soltis & Rieseberg, 1986; Levin, 2003). Most researchers apparently consider autopolyploidy and allopolyploidy to be useful terms, given their frequency (often with no definition given) in the literature. There are two main classes of definitions (Stebbins,

1950; Grant, 1981; Jackson, 1982; Jackson & Casey, 1982; Ramsey & Schemske, 1998): genetic, in which cytological behavior provides the criteria for recognition, and taxonomic, in which one vs. two diploid species contributed genomes to the polyploid.

At issue is whether the interest of the researcher is primarily genetic or taxonomic (e.g., Doyle & Egan, 2010). If the concern is with such issues as the likelihood of genetic recombination or the preservation vs. segregational loss of parental loci, then what is most relevant is chromosome behavior—whether the segregational pattern of the polyploid is multisomic (genetic autopolyploidy) or disomic (genetic allopolyploidy). It matters much less to a geneticist whether one or multiple species were involved in the formation of the polyploid, but this issue is certainly of great interest to evolutionary biologists, for whom the presence or absence of hybridization is the key to the distinction of auto- vs. allopolyploidy. Significantly, geneticists and plant breeders use the term "hybrid" quite differently from systematists: for the former it includes the products of crosses between genotypes of the same species, whereas for the latter it is usually reserved for interspecies crosses.

Neither definition is easy to apply. To establish whether a plant is a genetic auto- or allopolyploid requires determining segregation ratios, preferably at many loci; even the easier, but still time-consuming expedient of studying meiotic behavior is inadequate, because the failure to observe multivalents at meiosis does not mean that chromosome pairing is not random among the four chromosomes of a tetraploid (Ramsey & Schemske, 2002). The possibility that a species could be a "segmental allopolyploid"—in which some chromosomes or individual loci segregate polysomically, and others disomically (Stebbins, 1950)—is also a problem for the genetic definition. A major unanswered question is whether such plants exist, at least in the long-term. Despite suggestions in the earlier literature (e.g., Sarvella, 1958), Sybenga (1996) stated the negative opinion quite forcefully: "An established polyploid is either an autopolyploid or an allopolyploid. In exceptional cases it is thinkable that a stable segmental allopolyploid arises, in which some sets of chromosomes are well differentiated and behave as in an allopolyploid, whereas other sets are not well differentiated and behave as in an autopolyploid. No clear cases have been found in the literature so far." But segmental allopolyploidy is commonly invoked nonetheless, and a recent modeling study has suggested that this intermediate condition could account for observed segregation ratios in nature (Stift & al., 2008).

In part for these reasons, Ramsey & Schemske (2002) preferred the taxonomic ("mode of origin") over the genetic ("cytological criteria") definition. But for anyone familiar with the controversy over species concepts in the evolutionary biology literature, the necessity of deciding whether the diploid progenitors of a polyploid belonged to one or two species seems almost as difficult for polyploids whose progenitors are known, as for those formed anciently from unknown (and even extinct) progenitors. Of course, the original cytological behavior of a paleopolyploid is also unknowable, and the process of diploidization, over thousands or millions of years, could shift

segregation patterns from polysomic to disomic, fuse chromosomes, and reduce chromosome numbers, so that what began as a genetic autopolyploid would be diagnosed as an allopolyploid. The existence of such cytological diploidization and the rate at which it could occur are unanswered genetic questions mentioned by Doyle & al. (2008). Other criteria that have been used to hypothesize allo- vs. autopolyploid origins are also problematical. A sister relationship between each of two duplicated sequences in a polyploid to orthologous sequences from different diploid progenitors is convincing evidence of taxonomic allopolyploidy, and methods have been developed for dealing with the complexities of high polyploids (Brysting & al., 2007). However, extinction of progenitor taxa can lead to a situation where homoeologues of a taxonomic allopolyploid are sister to one another, mimicking the condition of a taxonomic autopolyploid (Doyle & Egan, 2010). This is the likely situation in *Glycine*; in this genus the presence of two classes of centromeric heterochromatin repeats, localized to different sets of centromeres, has been used to hypothesize the existence of two subgenomes, as expected from an allopolyploid origin (Gill & al., 2009). Such non-phylogenetic criteria may be available in many taxa, but the complexity of repeated sequence evolution suggests caution in their interpretation.

The lack of a clear and readily applicable definition that fulfills the needs of diverse scientists is unsatisfying, but reflects the reality of the situation. It is probably not an issue that can be resolved by additional research, though it would be very useful to obtain segregation data from many loci in many polyploid species to determine the frequency of polyploids with mixed segregation patterns, the relationship between chromosomal and genetic segregation patterns, and the correlation between taxonomic and genetic allopolyploidy.

What is special about polyploidy? — A significant body of research, much of it discussed below, is aimed at discovering whether there are "rules" that govern the genetic, physiological, morphological, and evolutionary response to whole-genome duplication (WGD). Clearly, if such rules exist, then polyploidy deserves study as an important phenomenon in its own right. An alternative view is that what is most important about polyploidy is not WGD, which is unique to polyploids, but rather the joining of two diverged genomes or genotypes, which accompanies WGD in many polyploids; this is shared with hybrids, regardless of ploidy. We also need to distinguish between genome expansion in the broad sense, and WGD as a specific mechanism of genome expansion that involves the duplication of every gene. Phenomena such as increase in cell size with increasing ploidy may have less to do with genome doubling than with increasing genome size (e.g., Bennett, 1996), with polyploidy simply being the very common way in which genomes expand in plants.

From an evolutionary perspective, polyploidy is a speciation mechanism. In the simplest model of polyploid formation, genome doubling instantaneously creates a new species, reproductively isolated from its one or more diploid progenitors. As such, it is the only widely accepted mode of sympatric speciation (Hendry, 2009). That alone makes it of interest, along with the fact that it is such a common speciation mechanism

in plants (Soltis & al., 2007; Wood & al., 2009). However, as discussed below, the completeness of reproductive isolation between polyploids and their progenitors is one of many assumptions about polyploidy for which exceptions, and very few supporting data, exist.

Finally, it is worth noting at the outset of this review that there is already evidence that genetic differences, including genotypic differences within species, may at least counterbalance, if not outweigh, any generalizable effects of hybridity, genome doubling, or both, in polyploids (e.g., Riddle & al., 2006). This should not be a surprise, with all that has been learned about the degree of genotypic differentiation within at least some species (e.g., Fu & Dooner, 2002), but it suggests that it will require the investigation of many more polyploid species and their progenitors before patterns can be discerned against the background of genetic diversity.

What is (are) the most frequent mechanism(s) of polyploidization? — The mechanism or mechanisms by which polyploid organisms most frequently form in natural populations remain(s) unclear. In plants, formation via the union of unreduced gametes has been viewed as the most likely method, rather than somatic doubling (Harlan and de Wet, 1975; de Wet, 1980). However, it is unclear how frequent a one-step process of formation is, involving fusion of unreduced egg and unreduced pollen, relative to a two-step process that involves formation of a triploid intermediate via fusion of one normal haploid gamete (e.g., a typical, haploid egg) with an unreduced gamete (e.g., sperm from unreduced or diploid pollen) (Ramsey & Schemske, 1998), followed by fusion of a triploid gamete (e.g., an egg) and a haploid gamete (e.g., a haploid sperm). The latter mechanism involving a triploid step toward tetraploid formation is referred to as a "triploid bridge." Some evidence suggests that this pathway may be an important step in the formation of some polyploids (Bretagnolle & Thompson, 1995; Ramsey & Schemske, 1998; Husband, 2004). But, does a triploid pathway actually predominate in natural systems, or do polyploids more often form instantly via the fusion of two unreduced gametes produced either by the same plant or by different plants?

Furthermore, does mode of origin make any evolutionary difference, or is it simply an interesting but academic question? The genetic attributes of the polyploid individual and ultimately species that result from these contrasting modes of formation are expected to vary with genetic diversity in the parental populations, breeding systems, etc. Studies modeling 1-step vs. 2-step modes of origin in species with different life histories (e.g., selfers vs. outcrossers; long-lived perennials vs. annuals) would be useful. What is the impact of the two modes on genetic diversity in the polyploid, for example, and how does this vary among species? Does one mode of origin allow a polyploid to incorporate a greater amount of diversity from its progenitor(s) into its genome than does the other? Does a triploid bridge model afford a greater opportunity for polyploid speciation, particularly in long-lived perennial taxa where triploids could persist for many years?

Given the important role attributed to unreduced gamete formation in the polyploidization process, it is noteworthy that fundamental questions regarding the frequency, causes,

and importance of unreduced gamete formation remain unanswered (e.g., Ramsey & Schemske, 1998). In fact, although relatively inexpensive, this has been a somewhat neglected area for several decades. Most progress in understanding the meiotic processes that give rise to unreduced gametes has been made in crop plants (reviewed in Ramanna & Jacobsen, 2003; Cai & Xu, 2007). As reviewed by Ramsey & Schemske (1998), based on the response to selection for unreduced gametes in crops (e.g., Parrott & Smith, 1986; Veronesi & al., 1986; Tavoletti & al., 1991; Calderini & Mariani, 1997), plant populations may possess heritable genetic variation for the capacity to produce unreduced gametes. It would be good to know for a well-characterized naturally occurring, diploid-polyploid system, if there is a strong genetic component to unreduced gamete formation. That is, are some diploid parental genotypes much more likely to produce unreduced gametes than others? If so, are those same genotypes much more likely than others to be involved in polyploidization events (a topic discussed later in this review)? Importantly, new insights about the genetic control of unreduced gamete formation (d'Erfurth & al., 2008; d'Erfurth, 2009) and the biology of triploids (Henry & al., 2005, 2009) have recently been reported for the model system *Arabidopsis*. For example, d'Erfurth & al. (2008) characterize the first gene (*AtPS1)* implicated in the formation of a high frequency of unreduced or "diplogametes" in plants. *AtPS1* mutants produce diploid pollen grains, and spontaneous triploid plants in the next generation, but female meiosis is not affected in the mutant. They showed that abnormal spindle orientation at male meiosis II leads to diplogamete formation.

In addition, what is the impact of the environment on unreduced gamete formation? Some of the early literature suggests an environmental component to unreduced gamete formation (reviewed in Ramsey & Schemske, 1998; see also Sax, 1936; McHale, 1983; Hagerup, 1932). If environmental stimuli do indeed trigger unreduced gamete formation, polyploids may be more likely to form in some portions of the ranges of the diploid progenitor species than in others. However, this topic, particularly in the context of geographic distribution and environmental variation, has not been pursued.

What is the extent of gene flow between diploids and polyploids? — Reproductive isolation between ploidal levels is often assumed as part of the model of speciation by polyploidy. However, as Stebbins (1971: 149) pointed out, there are two pathways by which gene flow could occur between two ploidal levels. First, if triploids occur as sporadic hybrids, these may produce tetraploid progeny, allowing gene flow between the polyploid and its progenitor population(s). Second, if unreduced ($2n = 2x$) gametes are at all common in the diploid, then new tetraploid progeny can be formed in crosses with reduced ($1n = 2x$) gametes of the tetraploid. It is notable that Stebbins (1971) predicted that such gene flow would always be unidirectional, from the diploid to the polyploid, providing a mechanism for ongoing incorporation of genetic diversity from the diploid population(s) into the polyploid. It has been suggested that triploids in *Arabidopsis* could provide a continuing genetic link between diploids and tetraploids (Henry & al., 2005).

Triploids may play a crucial role in *Chamerion*. Diploid and autotetraploid *Chamerion angustifolium* (fireweed) are largely allopatric, but in a contact zone, 59% of populations are of mixed cytotype (Husband & Schemske, 1998; Husband & Sabara, 2004). Reproductive isolation between diploids and tetraploids is pronounced and is the result of several different reproductive barriers. Nonetheless, triploids are usually present when diploids and tetraploids co-occur, ranging in frequency from 2% to 22% within populations. Triploids have low fertility (Burton & Husband, 2000), suggesting that post-zygotic isolation is also strong. However, triploids do produce some euploid gametes with one ($n = x$), two ($n = 2x$), or three ($n = 3x$) chromosome sets, and simulations suggest this may be sufficient to cause recurrent polyploid formation (Husband, 2004).

In *Sorghum*, there appears to be significant gene flow between the diploid crop species, *S. bicolor*, and the tetraploid weed, *S. halepense* (Arriola & Ellstrand, 1996). Continued inputs from diploids into a polyploid through gene flow have the same effect as multiple origins of a panmictic polyploid, allowing the polyploid to incorporate additional genetic variation from its progenitor(s). Such gene flow may also look like multiple origins, coupled with extensive crossing, to the molecular systematist, suggesting additional caution in interpreting the presence of multiple "diploid" haplotypes in a polyploid species (Stebbins, 1971). It would be interesting to explore whether there are any criteria that could distinguish between these two explanations for extensive genetic variation in a given polyploid; the problem is reminiscent of the introgression vs. lineage sorting problem as alternative explanations for incongruence between gene phylogenies. Simulations, modeling, and the development of rigorous testing methods would be beneficial.

In *Betula*, there is some morphological and molecular evidence that gene flow may occur in both directions between diploid *B. nana* and tetraploid *B. pubescens* (Thórsson & al., 2001), contrary to Stebbin's (1971) hypothesis that gene flow should be unidirectional. This gene flow appears to occur through triploid intermediates (Thórsson & al., 2001). The frequency and mechanism of the apparent gene flow from the tetraploid to the diploid needs further examination. Brown & Al-Dawoody (1979) suggested on the basis of observations of meiosis that *B. pubescens* is an octoploid and the species commonly thought to be diploid are in fact tetraploid: hence hybrids are hexaploid rather than triploid. This suggestion needs further testing as it might help to explain hybrid fertility.

What factors promote/facilitate polyploidization? — Some factors have long been considered of general importance in the formation of polyploids. For example, Grant (1981) indicated that polyploidy is promoted by a combination of what he considered "primary" factors: (1) long-lived organisms usually with a means of vegetative propagation; (2) primary speciation accompanied by chromosomal repatterning; (3) the common occurrence of natural, interspecific hybridization. But within a clade of diploid species, what factors (other than geographic proximity) determine if a polyploid will form between species? While this section primarily focuses on allopolyploid formation, following a taxonomic definition of allopolyploidy (i.e., a polyploid formed via interspecific hybridization and genome

doubling), the same question can be posed for autopolyploids (i.e., polyploids formed within a single species): what factors promote polyploidization?

Clearly there are many clades within which some congeneric species come into contact and produce polyploids, whereas others have the opportunity to form polyploids, but do not. Grant (1981) also clearly recognized that a genotypic component may be involved in the proclivity to produce polyploids. Not only is reduced chromosome pairing (in hybrids) due to chromosomal rearrangements between the parents an important factor in allopolyploid formation, but chromosomal pairing in meiosis is also under genetic control. The genetic control of chromosome pairing in tetraploid wheats (the *Ph* gene) has been well investigated (e.g., Dhaliwal, 1977; Martínez-Pérez & al., 1999). Genetic control of homoeologous pairing is now also known from *Brassica* (Nicolas & al., 2009), but the extent to which similar genetic systems exist in other taxa, and whether the mechanisms involved are homologous, remain open questions (Cifuentes & al., 2010). Hence, genotypic factors alone could influence pairing in the absence of chromosomal rearrangement.

Darlington (1937) proposed an inverse relationship between the fertility of a diploid hybrid and that of a tetraploid to which it gives rise. He reasoned that at low parental divergences, homoploid hybrids will be fertile because chromosomes will be able to pair at meiosis, but allopolyploids will be of low fertility because pairing will occur between both duplicated chromosomes and homeologous chromosomes from each parent, causing uneven segregation. At high parental divergences, homoploid hybrids will be sterile due to failure of chromosome pairing, but allopolyploids will be fertile due to consistent bivalent formation at meiosis. This is sometimes referred to as "Darlington's rule." Clearly, this has significant bearing on the issue of the frequency of auto- vs. allopolyploidy, and highlights the terminological and theoretical difficulties of these terms. The genetic definitions (multisomic vs. disomic pairing) are more appropriate in such a discussion, but the preponderance of the work cited below more often presupposes the taxonomic definitions (formation involving one vs. more species). An unanswered question in the polyploidy literature is how often these two definitions coincide; it is also unknown (and perhaps unknowable) how many current polyploids with disomic chromosome segregation began their existence as genetic autopolyploids, with no true homoeologues and tetrasomic behavior.

Importantly, early investigators also noted that some parental genotypic combinations seem better at producing allopolyploids than others. Based in part on research with *Layia* (Asteraceae), Clausen & al. (1949) suggested that "the success and constancy of amphiploids (allopolyploids) is linked with the degree of relationship found between their parents." Similar observations were reported for leafy-stemmed *Gilia* (Polemoniaceae) (Grant, 1981) and species of *Phlox* (Polemoniacae) from eastern North America (Levin, 1968).

Three studies (Chapman & Burke, 2007; Buggs & al., 2008; Paun & al., 2009) recently re-visited Darlington's rule, using genetic or phylogenetic proxies as measures of chromosomal differences between progenitors of polyploids. Chapman & Burke (2007) compared the mean genetic distance between the parental species of 12 homoploid and 26 polyploid hybrid species and indicated that hybridization between genetically distant congeners rather than genetically similar ones appears to be more likely to result in the production of an allopolyploid in nature.

Buggs & al. (2008) formulated a null hypothesis that hybridization would occur successfully between all species of a genus (recognizing that a genus is an arbitrary unit and the model could be applied as well to supra- and infraspecific clades), regardless of their phylogenetic divergence, and found that the mean phylogenetic divergence between parents of polyploids in eight genera was not significantly different from the mean divergence expected under this null expectation. The same analyses were conducted on homoploid hybrids in the same genera; these revealed a lower divergence between the parents of homoploid hybrids than the null expectation, even when unstable hybrids were included. Buggs & al. (2008) concluded that "contrasting patterns of divergence between the parents of polyploids and homoploid hybrids are … determined by the restriction of homoploid hybrid formation to low parental divergence, rather than the restriction of polyploid formation to high parental divergence" (p. 87).

Paun & al. (2009), analyzing a larger dataset of 16 homoploid hybrids and 32 allopolyploids, calculated genetic distances between parental pairs using nuclear and/or chloroplast sequences, converted each of these to a "genetic divergence index" by dividing by the mean genetic distance between all species pairs in the genus, found higher divergence index scores for allopolyploids than homoploid hybrids, and concluded that "parental divergence drives ploidy."

Buggs & al. (2009b) addressed the results of these three analyses by conducting a two-tailed paired *t*-test on the genetic distances between allopolyploid parental pairs and the average genetic distance between all species pairs in their respective genera, using the data of Paun & al. (2009), and found no significant difference. This result agrees with that of Buggs & al. (2008)—polyploid formation fits a model of random hybridization, whereas homoploid hybrid formation tends to occur successfully at lower parental divergence.

This overview indicates that considerable attention has recently been given to Darlington's (1937) hypothesis. But more data points are needed. A major shortcoming of the studies noted is that autopolyploids (following the typical taxonomic definition) have not been included. As Buggs & al. (2009b) argue, the hypothesis that parental divergence drives polyploidy cannot be tested fully without including divergences at or close to zero (i.e., autopolyploids and allopolyploids derived from closely related species).

Multiple origins have been documented in many autopolyploids and allopolyploids (Soltis & Soltis, 1990, 1993) and appear to be the rule in polyploidization. What remains unclear is whether certain diploid parental genotypes are better suited or more likely to participate in the formation of a given polyploid than others. That is, of the many genotypes that may be present in a diploid parent, or parents, are only certain genotypes or combinations of particular genotypes involved in the repeated

formation of a given polyploid? Alternatively, perhaps genotype does not matter, and all parental genotypes are essentially equally likely to form a polyploid given the opportunity (e.g., sympatric occurrence in parental species in the case of allopolyploids).

Some evidence suggests that the underlying parental genotype may contribute to the likelihood of polyploid formation. Populations of a given species may differ in their ability to produce unreduced gametes (see above; reviewed in Ramsey & Schemske, 1998), and this variation may exist within populations as well. Microsatellite data for populations of the recently and recurrently formed allopolyploids *Tragopogon mirus* and *T. miscellus* in fact indicate that only a few of the numerous genotypes currently detected in populations of one of the parental diploids (*T. dubius*) have actually been involved in polyploid formation (Symonds & al., 2010). These data are consistent with the hypothesis that only a few genotypes produced unreduced gametes or that only a few combinations of genotypes yielded polyploids (Tate & al., 2009a; Symonds & al., 2010). Alternatively, perhaps only a few genotypes of *T. dubius* were present at the actual time of polyploidy formation; and other genotypes have subsequently migrated into the area post-polyploid formation.

How do new polyploids establish and then persist in natural populations? — Not only is the formation of polyploids unclear, but their subsequent establishment and persistence in natural populations are poorly understood as well. New polyploids must overcome competition with their parents both for abiotic resources and for reproductive opportunities (Levin, 1975). The latter problem is known as minority cytotype exclusion, and was proposed by Levin (1975) on the basis that polyploids usually possess only post-zygotic mechanisms of reproductive isolation from their diploid progenitors, so that most polyploid (2*x*) gametes are lost by union with the much more numerous 1*x* gametes of the diploid, leading to the formation of sterile triploids. This phenomenon was demonstrated in experimental populations of *Chamerion* by Husband (2000). Theoretical models suggest it may be overcome by increased unreduced gamete formation in the diploid (and a concomitant higher frequency of polyploid individuals), increased hybrid fitness, slight niche separation, lower inbreeding depression, and higher self-fertilization in the tetraploid (Fowler & Levin, 1984; Felber 1991; Rodriguez, 1996; Felber & Bever, 1997; Rausch & Morgan, 2005). Few studies from natural systems test these theoretical models, though Buggs & Pannell (2006) showed reduced minority cytotype exclusion of hexaploid *Mercurialis annua* due to self-fertilization. Baack & Stanton (2005) showed experimentally that neither niche differentiation nor tetraploid competitive superiority appears sufficient to explain tetraploid establishment in *Ranunculus adoneus*. They suggested that rare dispersal events may be responsible for the establishment of neo-polyploids. Bursts of unreduced gamete formation caused by environmental stress (Hagerup, 1932; Mable, 2004) may contribute to establishment of polyploids through increased frequency of polyploid formation, but, as noted above, the role of environmental stimuli in triggering unreduced gamete formation has not been conclusively shown in nature. Other studies

have examined differences in ecology between diploids and related polyploids in nature (see section below: "What are the ecological consequences of polyploidy?"). There is much still to be learned, and more to be demonstrated in natural systems, about how neopolyploids establish in nature.

What factors favor autopolyploid vs. allopolyploid formation? — Autopolyploidy seems prevalent in several angiosperm families, such as Saxifragaceae (reviewed in Soltis & al., 2007) and Cactaceae (e.g., Remski, 1954; Pinkava & al., 1985; Sahley, 1996; Fleming & al., 1998; Hamrick & al., 2002). In these families, many if not most, of the documented polyploids seem to be genetic autopolyploids. In contrast, only allopolyploids are known from other genera and families (e.g., *Nicotiana*, *Brassica*). Some families (e.g., Poaceae) appear to harbor both categories. What factors determine whether autopolyploids or allopolyploids form? Is chromosome size a factor? Would small chromosomes be less likely to form complex multivalents that might promote meiotic chromosome pairing abnormalities and ultimately sterility? Saxifragaceae and Cactaceae both have small chromosomes, but there are groups with small chromosomes that are not known to form autopolyploids, or rarely form autopolyploids (e.g., Brassicaceae). This is an area for which basically nothing is known.

Are allopolyploids really much more prevalent than autopolyploids? — It is essentially dogma that allopolyploidy is much more prevalent than autopolyploidy in nature. This sentiment dates to the longstanding traditional view that autopolyploidy is rare and perhaps maladaptive (Stebbins, 1950; Grant, 1971). For example, Stebbins (1950) maintained that the only unambiguous example of autopolyploidy in nature is *Galax aphylla* (now *Galax urceolata*). The fact that these views were held by giants in the field of angiosperm genetics and evolutionary biology (Stebbins, 1950, 1971; Grant, 1981) certainly set back by decades acceptance of the counterview that autopolyploidy was also important in natural populations (Lewis, 1980). Adding to the complexity is that autopolyploids and allopolyploids are simply endpoints; in natural populations there is a genetic continuum between autopolyploids and allopolyploids. Intermediate types, in which segregation is disomic for some loci and multisomic for others, have sometimes been referred to as "segmental allopolyploids" (Stebbins, 1950; see discussion of this topic below). But clearly the possible existence of intermediates between clear autopolyploids and clear allopolyploids can also make it difficult to estimate the frequency of auto- vs. allopolyploids.

The widespread availability of easily scored genetic markers led to studies that indicated that tetrasomic inheritance did indeed operate in many suspected autotetraploids (e.g., Soltis & Soltis, 1990). It quickly became apparent that autopolyploidy was much more prevalent than traditionally maintained (Soltis & Soltis, 1990, 1993, 1999). But how common is autopolyploidy relative to allopolyploidy? Ramsey & Schemske (1998) made the noteworthy calculation that the rate of autopolyploid formation in natural systems is actually high (see also Lewis, 1980). Although most such autopolyploid "experiments" ultimately fail in nature, with even a very small success rate, the number of naturally occurring autopolyploids is likely to be quite high.

not in lit.cit.

One problem in recognizing autopolyploids is that this is the "stealth" mechanism of polyploid evolution—autopolyploids are very difficult to detect and document (Soltis & al., 2007). As a result, the frequency and importance of autopolyploidy in nature have certainly been underestimated. Whereas allopolyploids typically involve morphologically well-differentiated parents and are thus more readily detected and distinguished as hybrids, autopolyploids may be nearly indistinguishable from their diploid parents. Autopolyploidy in several genera (*Tolmiea*, *Heuchera*) was discovered completely by chance (e.g., Soltis, 1984; Wolf & al., 1989), either via routine chromosome counting or allozyme surveys of what were presumed diploid populations. Large-scale surveys of ploidy via flow cytometry are helping to clarify the frequency of autopolyploids relative to their diploid progenitors (e.g., Husband & Sabara, 2004; Balao & al., 2009), but the actual discovery of autopolyploids remains largely a chance phenomenon.

Many species have multiple cytotypes that are essentially indistinguishable morphologically, but rarely have these been investigated in any detail. If many or most of these cytotypes are autopolyploids, the frequency of autopolyploidy in nature must be very high (Soltis & al., 2007). Using the California Flora as a database, of 2647 species from 346 genera in 62 angiosperm families, 334 species (13%) have multiple cytotypes (i.e., clear 3x, 4x, or higher multiples of the base chromosome number for the genus). Most of these are presumed to be autopolyploids, but all would require study to determine if they, in fact, are genetic and/or taxonomic autopolyploids. Because some of these 334 chromosomally polymorphic taxonomic species actually have more than two cytotypes, the total number of putative autopolyploid events is much higher (unpublished data of Ramsey & al.; reviewed in Soltis & al., 2007). Finally, autopolyploids are rarely considered distinct species from their diploid parents, further masking the importance of autopolyploidy. Autopolyploids are typically recognizable as species distinct from their diploid progenitors following any of several species concepts and should therefore be so designated (Soltis & al., 2007).

Do multiple origins result in lineages with differing evolutionary trajectories and/or ultimately cryptic species? — Although multiple or recurrent origins of polyploids are common in natural populations (Soltis & Soltis, 1993, 1999), the long-term evolutionary impact of repeated formations is unclear. At the level of genetic variation, it is logical to postulate that recurrent formations from genetically distinct diploid parental populations would introduce genetic variation into the polyploid and recent genetic studies bolster this hypothesis (e.g., Doyle & al., 1999; Segraves & al., 1999; Meimberg & al., 2009; Symonds & al., 2010). The presence or absence of gene flow between individuals resulting from separate origins of a polyphyletic polyploid species should have a significant effect on the subsequent evolutionary trajectory of the species, determining whether the species will remain as a collection of isolated and genetically depauperate populations or a single, panmictic species enriched by lineage recombination from its different origins. How this genetic variation may influence ecological or life-history attributes remains to be investigated, however.

Some evidence suggests that repeated formations can lead to cryptic species that are reproductively isolated to varying degrees. Six of the eight allopolyploid species of *Glycine* subg. *Glycine* (Fabaceae) had multiple origins, but among these, some show evidence of homogenization through lineage recombination whereas others do not, instead appearing to form distinct lineages (Doyle & al., 2004). Populations of arctic allopolyploids of separate origin are often morphologically distinct with varying degrees of interfertility (Brochmann & al., 2004).

Reproductive isolation among populations of a polyploid may arise through time, perhaps even in the absence of genetic differences resulting from multiple origins. Werth & Windham (1991) provocatively suggested that if alternative homeologs were silenced across an allopolyploid genome (reciprocal silencing), reproductive isolation and incipient speciation could ensue. Lynch & Force (2000) independently elaborated on this same hypothesis, and recent evidence shows reciprocal silencing to contribute to isolation between strains of diploid *Arabidopsis thaliana* (Bikard & al., 2009). Of particular interest in such scenarios are the ultimate fates of polyploid populations of independent origin. Such populations reflect the genetic contributions of different genotypes of the diploid parents; hence, distinct allopolyploid populations may be initiated with distinct genetic and cytogenetic signatures (Soltis & Soltis, 1999), leading to greater effects of reciprocal silencing.

Reciprocal formations of a given polyploid represent a special case of multiple origins. The allotetraploid *Tragopogon miscellus* (Asteraceae) has formed reciprocally (Ownbey & McCollum, 1953; Soltis & Soltis, 1989). This change in maternal vs. paternal parentage not only has yielded an obvious and dramatic difference in inflorescence morphology, but may also have resulted in reproductive isolation between the two separate formations. Attempts to cross these forms of *T. miscellus* failed (Ownbey & McCollum, 1953), but more work is needed. If further experimentation supports this reproductive isolation, then these reciprocal formations represent distinct biological species. At least three of the eight *Glycine* allopolyploids possess chloroplast genomes from both diploid progenitors (Doyle & al., 2004), indicating reciprocal formations. It is unclear, however, just how common reciprocal formations are in nature; this is an important issue, given the potential significance of cytoplasmic effects in plant evolution (Levin, 2003).

Are polyploids more successful than diploid progenitors? — It is generally assumed that polyploids are more successful than their diploid parents, at least in some habitats, particularly novel or disturbed ones (e.g., Levin, 1983; Stebbins, 1985; Otto & Whitton, 2000). On a broader scale of "success," Fawcett & al. (2009) proposed that genome doubling actually helped numerous plant lineages survive the K-T mass extinction (see also commentary of Soltis & Burleigh, 2009). However, the issue of success raises a suite of general questions: What is "success" in evolution, and how should it be assessed? Is it assessed as abundance within the range? Or perhaps as the number of ecological niches occupied? Should the criterion be the overall range, or the ability to colonize new regions? To evaluate polyploid "success", do we consider only the short term (e.g., the geographic range or niche of a polyploid compared

to its parent)? Or do we consider much longer evolutionary timeframes (e.g., number of more inclusive clades tracing their origin to an ancient polyploid event)?

Despite long-held views that polyploids may have broader ecological amplitudes than their diploid progenitors and may therefore have higher fitness, at least in the short term (e.g., Levin, 1983), very few experimental studies have actually addressed this issue. Superior fitness of a polyploid relative to its diploid progenitor(s) may ultimately be manifested as increased diversification of polyploid lineages. Polyploidy could be widespread in occurrence, but of little subsequent evolutionary importance, or polyploidy may stimulate diversification. It can be questioned, however, what is meant by this; *Homo sapiens* has yet to produce any new lineages, and perhaps may become extinct without doing so; if this means that humans are a dead end, does it diminish from what is, arguably, our current success?

Using a phylogenetic approach, Wood & al. (2009) estimated that 15% of angiosperm speciation events and 31% of fern speciation events involved polyploidy. They stressed that these estimates are four times higher than previous estimates. But they also reported that despite the high incidence of polyploid speciation, they found no evidence of subsequent increases in diversification in polyploid lineages. However, the Wood & al. (2009) analyses are focused primarily at species within genera. What about the role of polyploidy in diversification if deeper levels are considered?

A different approach is required to address the question at deep levels, one method being to seek to pinpoint ancient genome duplication events on a phylogeny. Using an interdisciplinary approach involving phylogenetics and genomics, it may be possible to determine whether ancient polyploidy events have made a major contribution to lineage diversification. The number of species in those lineages that arose following an ancient polyploidy event can then be compared to a sister lineage that did not experience that genome doubling event. An initial analysis of several prominent hypotheses of ancient polyploidy events in angiosperms (reviewed in Soltis & al., 2009) suggests that there is increased diversification after genome duplications. However, more work is needed to pinpoint exactly when in angiosperm phylogenetic history these events occurred.

Many extant taxa appear to have experienced one or more rounds of genome duplication in their evolutionary history. Examples include *Arabidopsis* (Bowers & al., 2003), rice (Paterson & al., 2004), and several basal angiosperms, including *Persea* and *Liriodendron* (Cui & al., 2006). The actual timing of the ancient polyploidy events and the placement of these events on phylogenetic trees is not always clear for various reasons (Fawcett & al., 2009; Doyle & Egan, 2010), but there is some evidence to support the claim that polyploidy has resulted in diversification of lineages. For example, the highly species-rich grass family (Poaceae) appears to be derived from a polyploid ancestor (Paterson & al., 2004). Barker & al. (2008) identified three polyploidy events in Compositae (Asteraceae), a family that includes around 10% of all angiosperm species, but were circumspect about whether polyploidy is causally related to this diversity, although additional data may support or refute

a causal relationship. A polyploidy event also marks the papilionoid legumes, the largest radiation in the third largest family of flowering plants (Pfeil & al., 2005). Similarly, Brassicales may also be derived from an ancient polyploidy event (reviewed in Soltis & al., 2009); in addition, genomic data suggest that an ancient polyploidization may coincide with the origin of a large eudicot clade that comprises 75% of all angiosperms (Bowers & al., 2003; Tang & al., 2008; Freeling, 2009; Cenci & al., 2010).

As genomic data become available for more angiosperms and as phylogenetic hypotheses of the angiosperm tree of life become better resolved at deep nodes, it should be possible to place or pinpoint with more confidence ancient genome duplication events on the angiosperm phylogenetic tree. In so doing, we can better assess whether ancient polyploidy events are typically associated with the formation of highly species-rich lineages as has been suggested for other species-rich groups such as yeast (Scannel & al., 2006).

■ GENETIC/GENOMIC

Doyle & al. (2008) stressed the importance of the question of "are there genetic rules in polyploid formation?" But they considered this issue only at very deep phylogenetic levels. As background, following a WGD, a newly formed polyploid organism is immediately left with a duplicate copy of each gene. Genomic studies indicate that not all genes are retained in duplicate over the course of subsequent genome evolution; some genes are returned to singleton status. But here we want to ask this question at a finer scale: Do genomic rules begin to operate immediately in new polyploids, or are these rules enforced only after longer periods of time. Do "rules" operate at the level of individuals, and perhaps in multiple origins of the same polyploid species?

Does genomic/genetic evolution repeat itself in polyploids of multiple origin? — Gould (1994) suggested that, on a broad evolutionary scale, if we could replay the evolutionary tape of life on Earth, it would play differently—"history involves too much chaos," and too many chance events are involved for the evolutionary process to be repetitive. In contrast, Conway Morris (2003) has argued that the broad trajectory of evolution is repetitive and predictable. However, what is true on a finer scale? Are certain aspects of the polyploidy process "hard-wired"?

Comparisons of completely sequenced plant genomes (e.g., *Arabidopsis* and *Oryza*) suggest that there may be "rules" to gene evolution across genomes over large spans of evolutionary time and involving not only plants but also vertebrates and yeast. Studies at deep levels suggest that preservation of duplicated gene copies following genome duplication is far from random, with specific functional categories preferentially retained (Blanc & Wolfe, 2004b) and reduplicated in subsequent polyploidizations (Seoighe & Gehring, 2004; Chapman & al., 2006). Paterson & al. (2006) showed that many gene and domain families actually have convergent fates following independent WGD events in *Arabidopsis*, *Oryza*, *Saccharomyces* and *Tetraodon*. This has also been found in comparison

of *Arabidopsis* and five tetraploid cotton species (Rong & al., 2010). Similarly, within *Arabidopsis* itself, several episodes of genome duplication have been inferred (Bowers & al., 2003); through these multiple rounds of genome duplication, some genes seem to be repeatedly returned to singleton status, whereas other genes are maintained as duplicates (Chapman & al., 2006). Collectively, these observations indicate that there *may* exist certain "principles" that govern the fates of gene and genome duplications.

Surely it is to be expected that such patterns of polyploid evolution will have a strong genetic component: the more similar the genetic backgrounds of independently formed polyploids, the more similar should be their response to polyploidy. In support of this logic, Barker & al. (2008) noted that in the case of independent events in Asteraceae, "paleopolyploidy can yield strikingly consistent signatures of gene retention in plant genomes despite extensive lineage radiations and recurrent genome duplications but that these patterns vary substantially among higher taxonomic categories." It would therefore not be surprising to find many more similarities in patterns of gene retention among the eight independently and recently formed *Glycine* allopolyploids (Doyle & al., 2004), or between *Tragopogon mirus* and *T. dubius* (Soltis & al., 2004) than between *Arabidopsis* and yeast. Studies at such lower hierarchical levels thus offer important insights into the existence of polyploid evolutionary "rules."

At an even finer scale, multiple origins of the same polyploid species permit an evaluation of this question at a very recent time frame. There is some evidence that immediate aspects of genome change in polyploids are repeated. For example, in the wheat group (*Aegilops*, *Triticum*), elimination of non-coding chromosome- and genome-specific sequences occurs early in synthetic polyploids, beginning as early as the first generation and completed by the second or third allopolyploid generation (Feldman & al., 1997; Ozkan & al., 2001). The loss of these repetitive elements was not random but instead was reproducible among the synthetic lines, suggesting a directed basis for genome restructuring (Ozkan & al., 2001). However, more recent studies suggest that such restructuring is genotype-dependent (Mestiri & al., 2010). Data for populations of independent origin in the recently formed allotetraploids *T. mirus* and *T. miscellus* (reviewed below) also suggest that some aspects of genome evolution may be "hard-wired" and that indeed molecular evolution is repeated to some degree. In both *T. mirus* and *T. miscellus* rapid loss of one copy of individual DNA segments (homoeologs) has been documented following polyploidization (Tate & al., 2006, 2009; Buggs & al., 2009a, 2010; Koh & al., 2010). Individuals from multiple populations consistently exhibited homoeolog loss at certain loci, whereas other genes never displayed loss, suggesting a "repeated" pattern of evolution. However, in another sense the losses are stochastic at this early stage (40 generations post-polyploid formation) in that populations are not fixed for any given loss; rather, losses were detected in only some individuals.

Additional studies are needed to test further the generalities of these findings and to address how quickly such changes may occur. Studies of additional polyploid groups will also permit

further testing of hypotheses that account for why some genes are retained preferentially as duplicates and others as singletons following polyploidy (Freeling, 2009; Birchler & Veitia, 2009).

Searching for "rules" at the morphological, anatomical, and physiological levels, and then working toward the genome may also be a fruitful enterprise. There appear to be some predictable effects of polyploidy, or at least of increase in genome size, codified in the "nucleotype theory" of Bennett (1996), reflected in such widespread phenomena as increase in guard cell size with increasing genome size (reviewed in Beaulieu & al., 2008). At the physiological level, polyploidy often leads to higher photosynthetic capacity on a per-cell basis (Warner & Edwards, 1993).

Are there winners and losers when diploid genomes are combined? — When divergent diploid genomes are combined to form an allopolyploid genome, and subsequent genomic/genetic changes occur (homoeolog loss, gene silencing), are such processes evenly spread across both parental genomes, or is one parent a "winner" and the other a "loser"; that is, are most changes in one direction, favoring one parent over the other? If so, what determines who wins and loses? Recent analyses of patterns of gene expression in allotetraploid cotton and the allotetraploid *Arabidopsis suecica* suggest that one diploid parent may be a "winner" and the other a "loser" in terms of gene expression. Wang & al. (2004) examined gene expression in synthetic autotetraploid *A. thaliana* and in multiple independent synthetic and natural lines of *A. suecica*, an allotetraploid derived from *A. thaliana* and *A. arenosa*, and found that some progenitor genes are differentially expressed in early generations, whereas other genes are silenced in later generations or among different siblings within a selfing generation, suggesting that the gene silencing is "rapidly and/or stochastically" established. More recently, microarrays revealed gene expression in the allopolyploid *A. suecica* was much more similar to the pattern in one parent: 65% of the non-additively expressed genes in *A. suecica* were repressed, and 94% of the repressed genes match those genes that were expressed at higher levels in *A. thaliana* than in *A. arenosa* (Wang & al., 2006). These results were considered "consistent with the silencing of *A. thaliana* rRNA genes and with overall suppression of the *A. thaliana* phenotype in synthetic allotetraploids and natural *A. suecica*."

In two cotton synthetic polyploids ("A" × "D" and "A" × "G" genomes), Rapp & al. (2009) recently discovered "genomic dominance" by which gene expression for thousands of genes closely mirrors that of only one of the two parents, both when genes from that parent are up-regulated and down-regulated relative to the other parent. In the AD polyploid, the D genome was "dominant", and in the AG polyploid the A genome was dominant. The mechanistic basis for this phenomenon is unknown. This experiment did not distinguish between homeologs, but another experiment, conducted on the "A" × "D" synthetic allopolyploid, distinguished between the expression of homeologous genes from the two parental diploids. There was a general tendency for homeologs from the "D" genome to be silenced more often than those from the "A" genome in F_1 hybrids, synthetic allopolyploids, and an allopolyploid crop variety (Chaudhary & al., 2009).

[margin note left: 2009 a or b?]

[margin note right: not in lit.cit.]

Research on *Tragopogon* (Asteraceae) similarly suggests that one diploid parent may consistently be a "loser" genome. Concerted evolution of the rDNA cistron has been documented in several allopolyploids (reviewed in Kovarik & al., 2005). These studies suggest the possibility of a "loser" genome in terms of rDNA copy number. In the recently and repeatedly formed *Tragopogon* allopolyploids (*T. mirus* and *T. miscellus*), for example, concerted evolution of rDNA consistently occurs towards either *T. pratensis* (in the case of *T. miscellus*) or *T. porrifolius* (in the case of *T. mirus*). This same directionality is observed in populations of *T. miscellus* and *T. mirus* of independent origin—that is, evolution repeats itself (Kovarik & al., 2005). Homogenization has not occurred to completion in any population, however; a small number of copies of rDNA of the shared diploid parent (*T. dubius*) remain in all populations of the allopolyploids studied. Interestingly, expression studies reveal that the major RNA type present in both *T. mirus* and *T. miscellus* is actually that of *T. dubius*—that is, there is actually an inverse relationships between gene copy number and expression in the allopolyploids (Matyášek & al., 2007). This is not a general rule in allopolyploids, however. In *Glycine*, rDNA repeat number was found to be highly skewed toward one parent in most of the 46 individuals of six allopolyploid species studied by Joly & al. (2004). Nearly all plants expressed both parental repeat types, though often at very low levels, but only three individuals expressed the minority repeat type at a higher level than the majority parental class (Joly & al., 2004).

In several allopolyploids, rapid loss of DNA occurs following polyploidization (e.g., *Triticum*, Ozkan & al., 2001; *Tragopogon*, Tate & al., 2006, 2009b; Buggs & al., 2009a, 2010). In *Tragopogon miscellus*, studies that surveyed duplicate gene pairs showed a slight bias towards loss of *T. dubius* homeologs, rather than *T. pratensis* homeologs (Tate & al., 2006, 2009; Buggs & al., 2009a, 2010). In one *Glycine* allotetraploid (*G. dolichocarpa*), mRNA levels of thousands of genes comprising the leaf transcriptome are more strongly correlated with expression levels in one parent (*G. tomentella* D3) than the other (*G. syndetika*). There is also some bias towards expression of the *G. tomentella* D3 homeolog at many of these genes (D. Ilut & al., unpub. data). However, of genes that show silencing of one homeolog, a greater overall percentage silence the *G. tomentella* copy (J.E. Coate & al., unpub. data).

Over longer periods of time, silenced genes are likely to be lost. In *Arabidopsis* this has led to homeologous blocks with very different degrees of gene retention, suggesting that there are "winners and losers" in the long run (Thomas & al., 2006). In taxa whose genomes have experienced multiple polyploidy events, one can ask whether patterns of gene loss are replicated in different events. In *Glycine*, which has experienced two rounds of WGD within the last ca. 50 million years (Shoemaker & al., 2006; Doyle & Egan, 2010), this does not seem to be the case for photosynthesis gene families: gene lineages that retained both homeologs following the ca. 50 million years event are no more likely to retain duplicates after the <10 million years event than are lineages that failed to retain both early homeologs (J.E. Coate & J.J. Doyle, unpub. data). What does appear to be replicated, however, is a propensity

2009 a or b?

for functional groups of genes to expand by polyploid vs. non-polyploid duplication: Photosystem II gene families expand by mostly polyploidy both across different taxa (*Glycine*, *Medicago*, *Arabidopsis*) and in nested WGD events within a species, whereas Calvin cycle gene families tend to expand by non-polyploid duplications; this is perhaps due to the greater opportunity for functional differentiation of Calvin cycle genes and selection on gene balance in Photosystem II families (Coate & Doyle, unpub. data).

What is the fate of duplicate genes? — Population genetic theory predicts that the consequences of genome duplication should be sensitive to differences in effective population size (Ne) (Lynch & Conery, 2003b; Lynch, 2006). For example, neofunctionalization (acquisition of new expression patterns by pre-existing genes) is more likely to occur in large populations. In contrast, subfunctionalization (dividing the ancestral expression profile between duplicated gene copies: Lynch & Force, 2000) is an unlikely outcome in large populations because drift is so slow that a partially subfunctionalized allele (the first step in the process) is likely to be fully silenced by secondary mutations before fixation occurs (Lynch & al., 2001), and confers additional mutational cost (Lynch, 2006). Even in yeast, basic questions about duplicated gene fates remain controversial (Gu & al., 2003; Kellis & al., 2004; Wolfe, 2004; He & Zhang, 2005).

not in lit.cit. (also no "a")

How widespread are tissue-specific changes in gene expression? — Adams & al. (2003) and Chaudhary & al. (2009) have demonstrated that duplicate genes may be differentially expressed in different tissues of polyploid cotton. Similar results have been found in *Tragopogon* polyploids (Buggs & al., in prep.). This differential expression may be a potentially important factor contributing to the success of polyploids. It remains unclear how widespread this differential expression of duplicates is in polyploids, but its finding in both recent and older polyploids suggests that it may be common. It is also unclear if this differential expression is evolutionarily meaningful. What is the utility of having two copies of a gene that are expressed in different tissues, but that differ only slightly in DNA sequence? While providing a convenient explanation for the maintenance of duplicate genes over evolutionary time, such subfunctionalization may contribute little to the subsequent evolution of the species, unless it results in escape from adaptive conflict, a process in which duplication enables paralogous copies to partition and subsequently optimize both ancestral function and a novel function that evolved in the progenitor gene (Des Marais & Rausher, 2008).

not in lit.cit.

Understandably, protein-coding and ribosomal RNA genes have received most of the attention in the polyploidy literature. However, much of the plant genome is composed of other sequences, notably repeated sequences of various classes. Centromeric heterochromatin repeats have already been mentioned in the context of hypothesizing polyploid origins (Gill & al., 2009), and their evolution following genome merger in allopolyploids is of great interest given the importance of centromeres in the evolution of homeologous chromosomes.

Because of their known or hypothesized roles in genome and gene evolution, transposable elements (TEs) are of considerable interest and importance in the study of polyploidy.

In allopolyploid *Gossypium*, some but not all transposable element families were shown to move between homeologous genomes (Zhao & al., 1998). Lim & al. (2007) used genomic in situ hybridization (GISH) to document the progressive process of homeologous repeated element homogenization across genomes in *Nicotiana* allopolyploids of increasing age. In the case of older polyploids, particularly those whose progenitors are unknown, differences in the composition of TEs in homeologous genomic regions cannot be assumed to be due to insertion following (or due to) polyploidy, because it is possible that the differences were already present in the progenitors. In soybean, for example, striking differences in the number of TEs were observed between a 1 megabase region and its several-fold larger homeolog, which could have been due to accumulation following polyploidy. However, the larger homeolog resides in a pericentromeric region, so it is possible that the difference between the two homeologs has more to do with the dynamics of different regions of the genome than with polyploidy, and that the different locations could have predated genome merger (Innes & al., 2008).

Transposable elements have been implicated in the restructuring of genomes in early stages of allopolyploid evolution, but the degree to which this is true varies among species, and the underlying mechanisms have been unclear. Parisod & al. (2010) reviewed the role of transposable elements in allopolyploid evolution, and concluded that "the effect of allopolyploidy on TE genome fractions may be more complex than generally assumed. As TEs are abundant and dispersed throughout genomes, they predispose a young allopolyploid genome to rapid shuffling, participating in natural genetic engineering and producing abundant raw material for adaptive evolution at a crucial moment. These effects of TEs warrant more detailed study in the future."

■ ECOLOGY/PHYSIOLOGY

What are the ecological consequences of polyploidy? — This has been a major question in polyploid research for many years (see Levin, 1983), but, unfortunately, the recent explosion of new data on the genetic and genomic consequences of polyploidy has not been accompanied by a similar leap in our knowledge of the ecological consequences of polyploidy. Historically, many generalizations were proposed about the distribution and abundance of polyploids, based on a few genera and a wealth of anecdotal data. Many of these generalizations have not held up to modern scrutiny, and a firm ecological context is still lacking for polyploidy. Most studies on patterns of distribution of different ploidal levels are based only on field observations, rather than a combination of observations and experimentation. Although there is much to do, very few investigators are pursuing ecologically focused research on auto- or allopolyploids in natural populations, or analyzing data from a large number of taxa. We review a few pertinent papers below as exemplars.

A recent study by Martin & Husband (2009) examined sources of variation between two diploid and one polyploid species (without attempting to discriminate between auto- and allopolyploidy) from each of 144 angiosperm genera in North America and found no differences in the range breadth or range climatological position between diploid and polyploid congeners. In contrast to an early and well-known hypothesis, polyploids were not more likely to occupy northern latitudinal positions and were no more displaced from diploids than other congeneric diploids. The authors point out that their result agrees well with a study by Bowden (1940), who planted 100 species of diploid and polyploid plants and found no consistent differences in their degree of cold tolerance. Martin & Husband (2009) suggest that the most interesting sources of differentiation may be those that confer the ability of different ploidal levels to overlap in their range. A similar broadly sampled study is needed to examine the differences between ploidal levels at a more local habitat scale (e.g., Lewis & al., 1967; Lumaret & al., 1987; Gauthier & al., 1988; Ståhlberg, 2009).

The hypothesis that niche differentiation is common between ploidal levels would be most convincingly and effectively revealed by reciprocal transplant experiments, rather than purely correlative studies. Such reciprocal transplant experiments have recently been undertaken in four diploid-polyploid comparisons (Flegrová & Krahulec, 1999; Baack & Stanton, 2005; Buggs & Pannell, 2007; Raabová & al., 2008). With the exception of Flegrová & Krahulec (1999) these studies have not provided strong support for the hypothesis that polyploidy has given rise to adaptive differentiation. More reciprocal transplant experiments are needed, particularly of recently formed polyploids. It is also important to distinguish evolutionary effects of polyploidy from effects that arose in a polyploid after its formation. For example, Maherali & al. (2009) conclude that differences in water use efficiency between diploid and tetraploid *Chamerion* arose after, and not as a consequence of, polyploidy.

Correlations between polyploidy and the sexual system have often been suggested (reviewed in Mable, 2004; Pannell & al., 2004). Two recent studies have tested these correlations on a broad taxonomic scale (Barringer, 2007; Husband & al., 2008). Barringer (2007) found support for the hypothesis that polyploids self-fertilize more than diploids in data from 235 species of angiosperms, in both phylogenetically independent contrasts and cross-species analyses. Husband & al. (2008) analyzed variation in outcrossing rates among ten diploid-polyploid species pairs, finding that polyploids had lower outcrossing rates than diploids, also confirming the long-standing hypothesis. Interestingly, allopolyploids tended to have higher selfing rates than autopolyploids.

Thompson & al. (2004) noted that, "Until recently almost nothing was known about the effects of plant polyploidy on interactions with herbivores and pollinators." This sentence summarizes well a gaping hole in our understanding of the ecological consequences of both auto- and allopolyploidy. In a model study of the interaction of diploids, autopolyploids, and their pollinators and herbivores, Thompson & al. (2004) showed that autotetraploid plants of *Heuchera grossulariifolia* (Saxifragaceae) have higher levels of attack by the moth *Greya politella* than diploid plants of this species. Furthermore, diploid and

autotetraploid plants occurring in sympatry also differ in the overall set of pollinators that they attract. As Thompson & al. (2004) conclude, "Hence, the evolution of polyploid populations has the potential to change significantly the evolutionary ecology of interactions with herbivores and pollinators." Since 2004, a few more studies have shown that polyploidy can play an important ecological role in the diversification of plant–animal interactions and community organization, though their interactions are often complex and perhaps hard to generalize (Nuismer & Cunningham, 2005; Kennedy & al., 2006; Münzbergová, 2006; Pearse & al., 2006; Arvanitis & al., 2007; Oswald & Nuismer, 2007; Halverson & al., 2008a; Leena & al., 2008; Nuismer & Ridenhour, 2008; Thompson & Merg, 2008).

One common explanation for the evolutionary success of polyploid plants is that polyploids often, but not always, occupy a wider range of environments than do their diploid ancestor or ancestors (Otto & Whitton, 2000). Two explanations for this pattern are: (1) a wider plasticity in polyploid species than in the diploid parents and (2) the existence of a range of locally adapted types in the polyploid, perhaps due to multiple origins. Münzbergová (2007) investigated the performance and plasticity of diploid and hexaploid cytotypes of *Aster amellus* (Asteraceae) in a common garden. The hexaploid plants occurred in habitats with a wider range of competition intensity. Plants were grown from seeds from these populations with and without competition in a common garden. The results of the Münzbergová (2007) study provide no support for either of the two suggested explanations for the wider range of habitats occupied by hexaploid plants, leading her to conclude that other explanations must now be proposed and pursued. Similarly, Bretagnolle & Thompson (2001) found no evidence for consistent differences in plasticity between diploid and related tetraploid *Dactylis glomerata*. In a broader study, Petit & Thompson (1999) examined 451 angiosperm species in 50 genera and 22 families in the Pyrenees. Individual polyploid species were found to have a smaller ecological range than diploids. Their results suggested that polyploidy may be a means of ecological diversification via taxonomic diversification, rather than through larger ecological amplitudes of polyploids *per se*. Although the higher plasticity of higher ploidal levels is frequently assumed, this may not be the case. Clearly more studies (e.g., common garden, coupled with phylogenetically paired species) are needed to test this hypothesis.

Diploids and polyploids may also differ in pathogen resistance. Using mathematical models, Oswald & Nuismer (2007) explored the possibility that new polyploids are initially more resistant to pathogens than their diploid progenitors. Significantly, newly formed polyploid populations may be more resistant to pathogens than their diploid progenitors. This effect can be strong; in the case of perennials with recurrent polyploid formation, the effect may last indefinitely. This interesting idea potentially provides another general explanation for the successful establishment of new polyploids. This hypothesis for the success of polyploids certainly requires testing in natural populations. Aspects of the hypothesis, including the long-term effects of genome merger and doubling, can also be studied at the genomic level, by analyzing genes involved in disease resistance (R-genes). A comparison of paleopolyploid *Glycine* with diploid *Phaseolus* found that the R-genes were partitioned between homeologous regions rather than doubled relative to the diploid condition (Innes & al., 2008 and unpublished data). Thus, it is possible that polyploidy might initially double the number of resistance genes, but that R-genes are subject to diploidization.

Recent progress has been made in our understanding of the ecological consequences of polyploidy, though this has not occurred at the same rate as the growth in our knowledge of the genomic consequences. It is to be hoped that in future the two fields may be more closely linked, and the ecological consequences of the various genomic consequences of polyploidy will be studied in the field. Some of our progress in understanding the ecology of polyploidy has overturned older generalizations, or shown that generalization is not yet possible. Our current state of knowledge may currently be less than what we thought it was a decade ago.

■ PROSPECTUS

Our knowledge of polyploidy as an important historical and ongoing biological process is clearly growing at an unparalleled rate. However, as is typical in scientific research, the more we learn, the more we realize we don't know. Entire new areas of research, such as genome-based studies of ancient polyploidy and analyses of the fates of homeologous genes, have been developed within only the past few years as technological and informatic breakthroughs have revolutionized the field. Early observations on the duplicated nature of the *Arabidopsis thaliana* genome and the loss of homeologous genes in recent allotetraploids in *Tragopogon*, for example, have raised entirely new and unanticipated questions, such as, "are all angiosperms polyploid?", "are whole-genome duplications associated with survival of the K-T boundary and with clade diversification?", "are rapid changes in genome composition biased toward one or the other parent of the polyploid?", and so on. As a result, the field of polyploid research is attracting scientists from disciplines other than those traditionally concerned with it.

Polyploidy has attracted at least its share of anecdotes, particularly with regard to the ecology of polyploids. Despite a fundamental and widely accepted inference that allopolyploids have broader ecological amplitudes than their diploid parents, until recently, few studies have applied either quantitative or experimental approaches to analyses of polyploid habitats or distributions. The explicitly biotic components of ecology—e.g., pollinators, pathogens, herbivores—have received even less attention. Observations that many apomictic plants are odd-ploids have long been reported in the literature, but what environmental conditions select for this combination of traits, at times to the exclusion of diploid or other lower-ploid individuals? Current methods of rapid ploidy assessment, coupled with quantitative, hypothesis-driven ecological analyses, will be instrumental in helping us to understand the ecology of polyploidy.

Likewise, some theory exists for understanding and predicting the dynamics of mating system evolution in polyploid systems and for anticipating the fates of homeologous genes with respect to population size. However, data addressing both questions are limited, in part because of an historic shortage of genetic markers that could be used to either infer mating systems or follow homeologs through populations and time. Again, given current tools, and what has been learned in the recent past, the field is poised to ask and answer questions that have only rarely, if ever, been addressed. Furthermore, the time is perfect for the development of additional mathematical theory to describe other aspects of polyploidy, such as formation, establishment, ecology, and evolution. A notable recent effort in this area is that of Mayrose & al. (2010) on chromosome number evolution.

Through the topics raised in this non-traditional review—focused on those areas about which we know very little, rather than those that have been amply studied—we hope to stimulate new research. Both conceptual and mathematical theory on the biology of polyploids is needed, and data to test both new and existing theory would be extremely valuable. Such integration of theory and data—across levels of biological organization, from genes to the genome to the ecosystem, and across clades—will catapult the study of polyploidy into new arenas and will perhaps answer at least a few of things that we don't know about polyploidy.

■ LITERATURE CITED

Adams, K.L. 2007. Evolution of duplicated gene expression in polyploid and hybrid plants. *J. Heredity* 98: 136–141

Adams, K.L. & Wendel, J.F. 2005. Polyploidy and genome evolution in plants. *Curr. Opin. Pl. Biol.* 8: 135–141.

Arriola, P.E. & Ellstrand, N.C. 1996. Crop-to-weed gene flow in the genus *Sorghum* (Poaceae): Spontaneous interspecific hybridization between Johnsongrass, *Sorghum halepense*, and crop sorghum, *S. bicolor. Amer. J. Bot.* 83: 1153–1160.

Arvanitis, L., Wiklund, C. & Ehrlén, J. 2007. Butterfly seed predation: Effects of landscape characteristics, plant ploidy level and population structure. *Oecologia* 152: 275–285.

Baack, E.J. & Stanton, M.L. 2005. Ecological factors influencing tetraploid speciation in snow buttercups (*Ranunculus adoneus*): Niche differentiation and tetraploid establishment. *Evolution* 59: 1936–1944.

Balao, F., Casimiro-Soriguer, R., Talavera, M., Herrera, J. & Talavera, S. 2009. Distribution and diversity of cytotypes in *Dianthus broteri* as evidenced by genome size variations. *Ann. Bot.* 104: 965–973.

Barker, M.S., Kane, N.C., Matvienko, M., Kozik, A., Michelmore, R.W., Knapp, S.J. & Rieseberg, L.H. 2008. Multiple paleopolyploidizations during the evolution of the Compositae reveal parallel patterns of duplicate gene retention after millions of years. *Molec. Biol. Evol.* 25: 2445–2455.

Barringer, B.C. 2007. Polyploidy and self-fertilization in flowering plants. *Amer. J. Bot.* 94: 1527–1533.

Beaulieu, J.M., Leitch, I.J., Patel, S., Pendharkar, A. & Knight, C.A. 2008. Genome size is a strong predictor of cell size and stomatal density in angiosperms. *New Phytol.* 179: 975–986.

Bennett, M.D. 1996. The nucleotype, the natural karyotype and the ancestral genome. *Symp. Soc. Exp. Biol.* 50: 45–52.

Bikard, D., Patel, D., Le Metté, C., Giorgi, V., Camilleri, C., Bennett, M.J. & Loudet, O. 2009. Divergent evolution of duplicate genes leads to genetic incompatibilities within *A. thaliana. Science* 323: 623–626.

Birchler, J.A. & Veitia, R.A. 2009. The gene balance hypothesis: Implications for gene regulation, quantitative traits and evolution. *New Phytol.* Early View, Nov. 2009, http://www3.interscience.wiley.com/cgi-bin/fulltext/123188160/PDFSTART.

Blanc, G. & Wolfe, K.H. 2004a. Widespread paleopolyploidy in model plant species inferred from age distributions of duplicate genes. *Pl. Cell* 16: 1667–1678.

Blanc, G. & Wolfe, K.H. 2004b. Functional divergence of duplicated genes formed by polyploidy during *Arabidopsis* evolution. *Pl. Cell* 16: 1679–1691.

Bowden, W.M. 1940. Diploidy, polyploidy, and winter hardiness relationships in the flowering plants. *Amer. J. Bot.* 27: 357–370.

Bowers, J.E., Chapman, B.A., Rong, J.-K. & Paterson, A.H. 2003. Unravelling angiosperm chromosome evolution by phylogenetic analysis of chromosomal duplication events. *Nature* 422: 433–438.

Bretagnolle, F. & Thompson, J.D. 1996. An experimental study of ecological differences in winter growth between sympatric diploid and autotetraploid *Dactylis glomerata. J. Ecol.* 84: 343–351.

Brochmnan, C., Brysting, A.K., Alsos, I.G., Borgen, L., Grundt, H.H., Scheen, A.-C. & Elven, R. 2004. Polyploidy in arctic plants. *Biol. J. Linn. Soc.* 82: 521–536.

Brown, I.R. & Al-Dawoody, D. 1979. Observations on meiosis in three cytotypes of *Betula alba* L. *New Phytol.* 83: 801–811.

Brysting, A.K., Oxelman, B., Huber, K.T., Moulton, V. & Brochmann, C. 2007. Untangling complex histories of genome mergings in high polyploids. *Syst. Biol.* 56: 467–476.

Buggs, R.J.A. & Pannell, J.R. 2006. Rapid displacement of a monoecious plant lineage is due to pollen swamping by a dioecious relative. *Curr. Biol.* 16: 996–1000.

Buggs, R.J.A. & Pannell, J.R. 2007. Ecological differentiation and diploid superiority across a moving ploidy contact zone. *Evolution* 61: 125–140.

Buggs, R.J.A., Chamala, S., Wu, W., Gao, L., May, G.D., Schnable, P.S., Soltis, D.E., Soltis, P.S. & Barbazuk, W.B. 2010. Characterization of duplicate gene evolution in the recent natural allopolyploid *Tragopogon miscellus* by next-generation sequencing and Sequenom iPLEX genotyping. *Molec. Ecol.* 19 (Suppl. 1): 1–15.

Buggs, R.J.A., Doust, A.N., Tate, J.A., Koh, J., Soltis, K., Feltus, F.A., Paterson, A.H., Soltis, P.S. & Soltis, D.E. 2009a. Gene loss and silencing in *Tragopogon miscellus* (Asteraceae): Comparison of natural and synthetic allotetraploids. *Heredity* 24: 1–9.

Buggs, R.J.A., Soltis, P.S., Mavrodiev, E.V., Symonds, V.V. & Soltis, D.E. 2008. Does phylogenetic distance between parental genomes govern the success of polyploids? *Castanea* 73: 74–93.

Buggs, R.J.A., Soltis, P.S. & Soltis, D.E. 2009b. Does hybridization between divergent progenitors drive whole-genome duplication? *Molec. Ecol.* 18: 3334–3339.

Burton, T.L. & Husband, B.C. 2000. Fitness differences among diploids, tetraploids and their triploid hybrids in *Chamerion angustifolium* (Onagraceae): Mechanisms of inviability and implications for polyploid evolution. *Evolution* 54: 1182–1191.

Cai, X. & Xu, S.S. 2007. Meiosis-driven genome variation in plants. *Curr. Genomics* 8: 151–161.

Calderini, O. & Mariani, A. 1997. Increasing 2*n* gamete production in diploid alfalfa by cycles of phenotypic recurrent selection. *Euphytica* 93: 113–118.

Cenci, A., Combes, M.C. & Lashermes, P. 2010. Comparative sequence analyses indicate that *Coffea* (Asterids) and *Vitis* (Rosids) derive from the same paleo-hexaploid ancestral genome. *Molec. Genet. Genomics* 283: 493–501.

Chapman, B.A., Bowers, J.E., Feltus, F.A. & Paterson, A.H. 2006. Buffering of crucial functions by paleologous duplicated genes may contribute cyclicality to angiosperm genome duplication. *Proc. Natl. Acad. Sci. U.S.A.* 103: 2730–2735.

Chapman, M.A. & Burke, J.M. 2007. Genetic divergence and hybrid speciation. *Evolution* 61: 1773–1780.

Chaudhary, B., Flagel, L., Stupar, R.M., Udall, J.A., Verma, N., Springer, N.M. & Wendel, J.F. 2009. Reciprocal silencing, transcriptional bias and functional divergence of homeologs in polyploid cotton (*Gossypium*). *Genetics* 182: 503–517.

Chen, Z.J. 2007. Genetic and epigenetic mechanisms for gene expression and phenotypic variation in plant polyploids. *Annual Rev. Pl. Biol.* 58: 377–406.

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

Chen, Z.J. & Tian, L. 2007. Roles of dynamic and reversible histone acetylation in plant development and polyploidy. *Biochim. Biophys. Acta* 1769: 295–307.

Cifuentes, M., Grandont, L., Moore, G., Chèvre, A.M. & Jenczewski, E. 2010. Genetic regulation of meiosis in polyploid species: New insights into an old question. *New Phytol.* 186: 29–36.

Clausen, J., Keck, D.D. & Hiesey, W.M. 1945. *Experimental studies on the nature of species*, vol. 2, *Plant evolution through amphiploidy and autopolyploidy, with examples from the Madiinae.* Washington, D.C.: Carnegie Institute of Washington.

Comai, L. 2005. The advantages and disadvantages of being polyploid. *Nature Rev. Genet.* 6: 836–846.

Conway Morris, S. 2003. *Life's solution: Inevitable humans in a lonely universe.* Cambridge: Cambridge Univ. Press.

Cui, L., Wall, P.K., Leebens-Mack, J., Lindsay, B.G., Soltis, D.E., Doyle, J.J., Soltis, P.S., Carlson, J., Arumuganathan, A., Barakat, A., Albert, V., Ma, H. & DePamphilis, C.W. 2006. Widespread genome duplications throughout the history of flowering plants. *Genome Res.* 16: 738–749.

Darlington, C.D. 1937. *Recent advances in cytology*, 2nd ed. Philadelphia: P. Blakiston's Son and Co.

D'Erfurth, I., Jolivet, S., Froger, N., Catrice, O., Novatchkova, M. & Mercier, R. 2009. Turning meiosis into mitosis. *PLoS Biol.* 7(6): e1000124M doi: 10.1371/journal.pbio.1000124.

D'Erfurth, I., Jolivet S., Froger N., Catrice, O., Novatchkova, M., Simon, M., Jenczewski, E. & Mercier, R. 2008. Mutations in *AtPS1* (*Arabidopsis thaliana* parallel spindle 1) lead to the production of diploid pollen grains. *PLoS Genet.* 4: e1000274.

De Wet, J.M.J. 1980. Origins of polyploids. Pp. 3–15 in: Lewis, W.H. (ed.), *Polyploidy: Biological relevance.* New York: Plenum Press.

Des Marais, D.L. & Rausher, M.D. 2008. Escape from adaptive conflict after duplication in an anthocyanin pathway gene. *Nature* 454: 762–765.

Dhaliwal, H.S. 1977. The *Ph* gene and the origin of tetraploid wheats. *Genetica* 47: 177–182.

Doyle, J.J., Doyle, J.L. & Brown, A.H.D. 1999. Origins, colonization, and lineage recombination in a widespread perennial soybean polyploid complex. *Proc. Natl. Acad. Sci. U.S.A.* 96: 10741–10745.

Doyle, J.J., Doyle, J.L., Rauscher, J.T. & Brown, A.H.D. 2004. Evolution of the perennial soybean polyploid complex (*Glycine* subgenus *Glycine*): A study of contrasts. *Biol. J. Linn. Soc.* 82: 583–597.

Doyle, J.J. & Egan, A.N. 2010. Dating the origins of polyploidy events. *New Phytol.* 186: 73–85.

Doyle, J.J., Flagel, L.E., Paterson, A.H., Rapp, R.A., Soltis, D.E., Soltis, P.S. & Wendel, J.F. 2008. Evolutionary genetics of genome merger and doubling in plants. *Annual Rev. Genet.* 42: 443–461.

(not in text) **Duarte, J.M., Cui, L., Wall, P.K., Zhang, Q., Zhang, X., Leebens-Mack, J., Ma, H., Altman, N. & dePamphilis, C.W.** 2006. Expression pattern shifts following duplication indicative of subfunctionalization and neofunctionalization in regulatory genes of *arabidopsis. Molec. Biol. Evol.* 23: 469–478.

Fawcett, J.A., Maere, S. & Van de Peer, Y. 2009. Plants with double genomes might have had a better chance to survive the Cretaceous-Tertiary extinction event. *Proc. Natl. Acad. Sci. U.S.A.* 106: 5737–5742.

Felber, F. 1991. Establishment of a tetraploid cytotype in a diploid population: Effect of relative fitness of the cytotypes. *J. Evol. Biol.* 4: 195–207.

Felber, F. & Bever, J.D. 1997. Effect of triploid fitness on the coexistence of diploids and tetraploids. *Biol. J. Linn. Soc.* 60: 95–106.

Feldman, M., Liu, B., Segal, G., Abbo, S., Levy, A.A. & Vega, J.M. 1997. Rapid elimination of low-copy DNA sequences in polyploid wheat: A possible mechanism for differentiation of homoeologous chromosomes. *Genetics* 147: 1381–1387.

Flagel, L.E. & Wendel, J.F. 2009. Gene duplication and evolutionary novelty in plants. *New Phytol.* 183: 557–564.

Flegrová, M. & Krahulec, F. 1999. *Anthoxanthum odoratum* and *A. alpinum*: Life history parameters at two different altitudes. *Folia Geobot.* 34: 19–31.

Fleming, T.H., Maurice, S. & Hamrick, J.L. 1998. Geographic variation in the breeding system and the evolutionary stability of trioecy in *Pachycereus pringlei* (Cactaceae). *Evol. Ecol.* 12: 279–289.

Fowler, N.L. & Levin, D.A. 1984. Ecological constraints on the establishment of a novel polyploid in competition with its diploid progenitor. *Amer. Naturalist* 124: 703–711.

Freeling, M. 2009. Bias in plant gene content following different sorts of duplication: Tandem, whole-genome, segmental, or by transposition. *Annual Rev. Pl. Biol.* 60: 433–453.

Fu, H. & Dooner, H.K. 2002. Intraspecific violation of genetic colinearity and its implications in maize. *Proc. Natl. Acad. Sci. U.S.A.* 99: 9573–9578.

Gates, R.R. 1909. The stature and chromosomes of *Oenothera gigas* De Vries. *Arch. Zellf.* 3: 525–552.

Gauthier, P., Lumaret, R. & Bedecarrats, A. 1998. Ecotype differentiation and coexistance of two parapatric tetraploid subspecies of cocksfoot (*Dactylis glomerata* L.) in the Alps. *New Phytol.* 139: 741–750.

(here and in text changed from "in press") **Gill, N., Findley, S., Walling, J.G., Hans, C., Ma, J., Doyle, J.J., Stacey, G. & Jackson, S.A.** 2009. Molecular and chromosomal evidence for allopolyploidy in soybean, *Glycine max* (L.) Merr. *Pl. Physiol.* 151: 1167–1174.

Gould, S.J. 1994 The evolution of life on Earth. *Sci. Amer.* 271: 85–86.

Grant, V. 1971. *Plant speciation*. New York: Columbia Univ. Press.

Grant, V. 1981. *Plant speciation*, 2nd ed. New York: Columbia Univ. Press.

Gregory, T.R. & Mable, B.K. 2005. Polyploidy in animals. Pp. 428–501 in: Gregory, T.R. (ed.), *The evolution of the genome*. San Diego: Elsevier.

Gu, Z., Steinmetz, L.M., Gu, X., Scharfe, C., Davis., R.W. & Li, W.-H. 2003. Role of duplicate genes in genetic robustness against null mutations. *Nature* 421: 63–66.

Hagerup O. 1932. Über Polyploidie in Beziehung zu Klima, Ökologie und Phylogenie. *Hereditas* 16: 19–40.

Halverson, K., Heard, S., Nason, J. & Stireman, J. 2008. Differential attack on diploid, tetraploid, and hexaploid *Solidago altissima* L. by five insect gallmakers. *Oecologia* 154: 755–761.

Hamrick, J.L., Nason, J.D., Fleming, T.H. & Nassar, J.M. 2002. Genetic diversity in columnar cacti. Pp. XX-XX in: Fleming, T.H. & Valiente-Banuet, A. (eds.), *Columnar cacti and their mutualists: Evolution, ecology and conservation*. Tucson: Univ. of Arizona Press.

Harlan, J.R. & de Wet, J.M.J. 1975. On Ö. Winge and a prayer: The origins of polyploidy. *Bot. Rev.* 41: 361–390.

He, X. & Zhang, J. 2005. Rapid subfunctionalization accompanied by prolonged and substantial neofunctionalization in duplicate gene evolution. *Genetics* 169: 1157–1164.

Hegarty, M.J. & Hiscock, S.J. 2008. Genomic clues to the evolutionary success of polyploid plants *Curr. Biol.* 18: R435–R444.

Hendry, A.P. 2009. Evolutionary biology: Speciation. *Nature* 458: 162–164.

Henry, I.M., Dilkes, B.P., Tyagi, A.P., Lin, H.Y. & Comai, L. 2009. Dosage and parent-of-origin effects shaping aneuploid swarms in *A. thaliana. Heredity* 103: 458–468.

Henry, I.M., Dilkes, B.P., Young, K., Watson, B., Wu, H. & Comai, L. 2005. Aneuploidy and genetic variation in the *Arabidopsis thaliana* triploid response. *Genetics* 170: 1979–1988.

Husband, B.C. 2004. The role of triploid hybrids in the evolutionary dynamics of mixed-ploidy populations. *Biol. J. Linn. Soc.* 82: 537–546.

Husband, B.C., Ozimec, B., Martin, S.L. & Pollock, L. 2008. Mating consequences of polyploid evolution in flowering plants: Current trends and insights from synthetic polyploids. *Int. J Pl. Sci.* 169: 195–206.

Husband, B. & Sabara, H.A. 2004. Reproductive isolation between autotetraploids and their diploid progenitors in fireweed, *Chamerion angustifolium* (Onagraceae). *New Phytol.* 161: 703–713.

Husband, B.C. & Schemske, D.W. 1998. Cytotype distribution at a diploid-tetraploid contact zone in *Chamerion* (*Epilobium*) *angustifolium* (Onagraceae). *Amer. J. Bot.* 85: 1688–1694.

Innes, R.W., Ameline-Torregrosa, C., Ashfield, T., Cannon, E., Cannon, S.B., Chacko, B., Chen, N.W.G., Couloux, A.,Dalwani, A., Denny, R., Deshpande, S., Egan, A.N., Glover, N., Hans, C.S., Howell, S., Ilut, D., Jackson, S., Lai, H., Mammadov, J., Martin del Campo, S., Metcalf, M., Nguyen, A., O'Bleness, M., Pfeil, B., Podicheti, R., Ratnaparkhe, M.B., Samain, S., Sanders, I., Ségurens, B., Sévignac, M., Sherman-Broyles, S., Thareau, V., Tucker, D.M., Walling, J., Wawrzynski, A., Yi, J., Doyle, J.J., Geffroy, V., Roe, B.A., Saghai Maroof, M.A. & Young, N.D. 2008. Differential accumulation of retroelements and diversification of NB-LRR disease resistance genes in duplicated regions following polyploidy in the ancestor of soybean. *Pl. Physiol.* 148: 1740–1759.

Jackson, R.C. 1982. Polyploidy and diploidy: New perspectives on chromosome pairing and its evolutionary implications. *Amer. J. Bot.* 69: 1512–1523.

Jackson, R.C. & Casey, J. 1982. Cytogenetic analyses of autopolyploids: Models and methods for triploids to octoploids. *Amer. J. Bot.* 69: 487–501.

Joly, S., Rauscher, J.T., Sherman-Broyles, S.L., Brown, A.H.D. & Doyle, J.J. 2004. Evolutionary dynamics and preferential expression of homoeologous 18S-5.8S-26S nuclear ribosomal genes in natural and artificial *Glycine* allopolyploids. *Molec. Biol. Evol.* 21: 1409–1421.

Kellis, M., Birren, B.W. & Lander, E.S. 2004. Proof and evolutionary analysis of ancient genome duplication in the yeast *Saccharomyces cereviseae*. *Nature* 428: 617–624.

Kennedy, B.F., Sabara, H.A., Haydon, D. & Husband, B.C. 2006. Pollinator-mediated assortative mating in mixed ploidy populations of *Chamerion angustifolium* (Onagraceae). *Oecologia* 150: 398–408.

Koh, J., Soltis, P.S. & Soltis, D.E. 2010. Homeolog loss and expression changes in natural populations of the recently formed allotetraploid *Tragopogon mirus* (Asteraceae). *BMC Biol.* 11: 97.

Kovarik, A., Pires, J.C., Leitch, A.R., Lim, K.Y., Sherwood, A.M., Matyasek, R., Rocca, J., Soltis, D.E. & Soltis, P.S. 2005. Rapid concerted evolution of nuclear Ribosomal DNA in two allopolyploids of recent and recurrent evolution. *Genetics* 169: 931–944.

Kuwada, Y. 1911. Meiosis in the pollen mother cells of *Zea mays* L. *Bot. Mag.* 25: 163–181.

Leena, A., Christer, W. & Johan E. 2008. Plant ploidy level influences selection by butterfly seed predators. *Oikos* 117: 1020–1025.

Leitch, A.R. & Leitch, I.J. 2008. Genomic plasticity and the diversity of polyploid plants. *Science* 320: 481–483.

Leitch, A.R., Soltis, D.E., Soltis, P.S., Leitch, I.J. & Pires, J.C. (eds.). 2004. Biological relevance of polyploidy: Ecology to genomics. *Biol. J. Linn. Soc.* 82: 409–700.

Levin, D.A. 1968. The genome constitutions of eastern North Amerian *Phlox* amphiploids. *Evolution* 22: 612–632.

Levin, D.A. 1975. Minority cytotype exclusion in local plant populations. *Taxon* 24: 35–43.

Levin, D.A. 1983. Polyploidy and novelty in flowering plants. *Amer. Naturalist* 122: 1–25.

Levin, D.A. 2002. *The role of chromosomal change in plant evolution.* New York: Oxford Univ. Press.

Levin, D.A. 2003. The cytoplasmic factor in plant speciation. *Syst. Bot.* 28: 5–11.

Lewis, W.H. 1980. Polyploidy in species populations. Pp. 103–144 in: Lewis, W.H. (ed.), *Polyploidy: Biological relevance.* New York: Plenum Press

Lewis, W.H., Suda, Y. & Macbryde, B. 1967. Chromosome numbers of *Claytonia virginica* in the St. Louis, Missouri area. *Ann. Missouri Bot. Gard.* 54: 147–152.

Lim, K.Y., Kovarik, A., Matyasek, R., Chase, M.W., Clarkson, J.J., Grandbastien, M.A. & Leitch, A.R. 2007. Sequence of events leading to near-complete genome turnover in allopolyploid *Nicotiana* within five million years. *New Phytol.* 175: 756–763.

Liu, B. & Wendel, J.F. 2003. Epigenetic phenomena and the evolution of plant allopolyploids. *Molec. Phylog. Evol.* 29: 365–379.

Löve, A. & Löve, D. 1949. The geobotanical significance of polyploidy. I. Polyploidy and latitude. *Portugaliae Acta Biol., Sér. A* **volume?** 273–352.

Lumaret, R., Guillerm, J.-L., Delay, J., Ait Lhaj Loutfi, A., Izco, J. & Jay, M. 1987. Polyploidy and habitat differentiation in *Dactylis glomerata* L. from Galicia (Spain). *Oecologia* 73: 436–446.

Lutz, A.M. 1907. A preliminary note on the chromosomes of *Oenothera lamarckiana* and one of its mutants, *O. gigas. Science* 26: 151–152.

Lynch, M. 2006. The origins of eukaryotic gene structure. *Molec. Biol. Evol.* 23: 450–468.

Lynch M. & Force, A. 2000 The probability of duplicate gene preservation by subfunctionalization. *Genetics* 154: 459–473.

Lynch, M., O'Hely, M., Walsh, B. & Force, A. 2001. The probability of fixation of a newly arisen gene duplicate. *Genetics* 159: 1789–1804.

Mable, B.K. 2004. 'Why polyploidy is rarer in animals than in plants': Myths and mechanisms. *Biol. J. Linn. Soc.* 82: 453–466.

Maherali, H., Walden, A.E. & Husband, B.C. 2009. Genome duplication and the evolution of physiological responses to water stress. *New Phytol.* 184: 721–731.

Martin, S.L. & Husband, B.C. 2009. Influence of phylogeny and ploidy on species ranges of north american angiosperms. *J. Ecol.* 97: 913–922.

Martínez-Pérez, E., Shaw, P., Reader, S., Aragón-Alcaide, L., Miller, T. & Moore, G. 1999. Homologous chromosome pairing in wheat. *J. Cell Sci.* 112: 1761–1769.

Matyášek, R., Tate, J.A., Lim, Y.K., Šrubařová, H., Leitch, A.R. Soltis, D.E., Soltis, P.S. & Kovařík, A. 2007. Concerted evolution of rDNA in recently formed *Tragopogon* allotetraploids is typically associated with an inverse correlation between gene copy number and expression. *Genetics* 176: 2509–2519.

Mayrose I., Barker, M.S. & Otto, S.P. 2010. Probabilistic models of chromosome number evolution and the inference of polyploidy. *Syst Biol.* 59: 132–144.

McHale, N.A. 1983. Environmental induction of high frequency 2*n* pollen formation in diploid *Solanum. Canad. J. Genet. Cytol.* 25: 609–615.

Meimberg, H., Rice, K.J., Milan, N.F., Njoku, C.C. & McKay, J.K. 2009. Multiple origins promote the ecological amplitude of allopolyploid *Aegilops* (Poaceae). *Amer. J. Bot.* 96: 1262–1273.

Mestiri, I., Chagué, V., Tanguy, A.-M., Huneau, C., Huteau, V., Harry, B., Coriton, O., Chalhoub, B. & Jahier, J. 2010. Newly synthesized wheat allohexaploids display progenitor-dependent meiotic stability and aneuploidy but structural genomic additivity. *New Phytol.*: in press.

Müntzing, A. 1936. The evolutionary significance of autopolyploidy. *Hereditas* 21: 263–378.

Münzbergová, Z. 2006. Ploidy level interacts with population size and habitat conditions to determine the degree of herbivory damage in plant populations. *Oikos* 115: 443–452.

Münzbergová, Z. 2007. No effect of ploidy level in plant response to competition in a common garden experiment. *Biol. J. Linn. Soc.* 92: 211–219.

Nicolas, S.D., Leflon, M., Monod, H., Eber, F., Coriton, O., Huteau, V., Chèvre, A.M. & Jenczewski, E. 2009. Genetic regulation of meiotic cross-overs between related genomes in *Brassica napus* haploids and hybrids. *Pl. Cell* 21: 373–385.

Nuismer, S.L. & Cunningham, B.M. 2005. Selection for phenotypic divergence between diploid and autotetraploid *Heuchera grossulariifolia. Evolution* 59: 1928–1935.

Nuismer, S.L. & Ridenhour, B.J. 2008. The contribution of parasitism to selection on floral traits in *Heuchera grossulariifolia. J. Evol. Biol.* 21: 958–965.

Ohno, S. 1970. *Evolution by gene duplication.* New York: Springer.

Osborn, T.C., Pires, J.C. Birchler, J.A., Auger, D.L., Chen, Z.J., Lee, H.-S., Comai, L., Madlung, A., Doerge, R.W., Colot, V. & Martienssen, R.A. 2003. Understanding mechanisms of novel gene expression in polyploids. *Trends Genet.* 19: 141–147.

Oswald, B.P. & Nuismer, S.L. 2007. Neopolyploidy and pathogen resistance. *Proc. Roy. Soc. London, Ser. B., Biol. Sci.* 274: 2393–2397.

Otto, S.P. & Whitton, J. 2000. Polyploid incidence and evolution. *Ann. Rev. Genet.* 34: 401–437.

Ownbey, M. & McCollum, G.D. 1953. Cytoplasmic inheritance and reciprocal amphiploidy in *Tragopogon. Amer. J. Bot.* 40: 788–796.

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

Pannell, J.R., Obbard, D.J. & Buggs, R.J.A. 2004. Polyploidy and the sexual system: What can we learn from *Mercurialis annua*? *Biol. J. Linn. Soc.* 82: 547–560.

Parisod, C., Alix, K., Just, J., Petit, M., Sarilar, V., Mhiri, C., Ainouche, M., Chalhoub, B. & Grandbastien, M.-A. 2010. Impact of transposable elements on the organization and function of allopolyploid genomes. *New Phytol.* 186: 37–45.

Parrott, W.A. & Smith, R.R. 1986. Recurrent selection for 2*n* pollen formation in red clover. *Crop Sci.* 26: 1132–1135.

Paterson, A.H., Bowers, J.E. & Chapman, B.A. 2004. Ancient polyploidization predating divergence of the cereals, and its consequences for comparative genomics. *Proc. Natl. Acad. Sci. U.S.A.* 101: 9903–9908.

Paterson, A.H., Chapman, B.A., Kissinger, J.C., Bowers, J.E., Feltus, F.A. & Estill, J.C. 2006. Many gene and domain families have convergent fates following independent whole-genome duplication events in *Arabidopsis, Oryza, Saccharomyces* and *Tetraodon. Trends Genet.* 22: 597–560.

Paun, O., Fay, M., Soltis, D.E. & Chase, M.W. 2007. Genetic and epigenetic alterations after hybridization and genome doubling. *Taxon* 56: 649–656.

Pearse, I.S., Krugel, T. & Baldwin, I.T. 2006. Innovation in anti-herbivore defense systems during neopolypoloidy—the functional consequences of instantaneous speciation. *Pl. J.* 47: 196–210.

Petit, C., Bretagnolle, F. & Felber, F. 1999. Evolutionary consequences of diploid-polyploid hybrid zones in wild species. *Trends Ecol. Evol.* 14: 306–311.

Pfeil B.E., Schlueter, J.A., Shoemaker, R.C. & Doyle, J.J. 2005. Placing paleopolyploidy in relation to taxon divergence: A phylogenetic analysis in legumes using 39 gene families. *Syst. Biol.* 54: 441–454.

Pinkava, D.J., Baker, M.A., Parfitt, B.D. & Mohlenbrock, M.W. 1985. Chromosome numbers in some cacti of western North America – V. *Syst. Bot.* 10: 471–483.

Raabová, J., Fischer, M. & Münzbergová, Z. 2008. Niche differentiation between diploid and hexaploid *Aster amellus. Oecologia* 158: 463–472.

Ramanna, M.S. & Jacobsen, E. 2003. Relevance of sexual polyploidization for crop improvement – a review. *Euphytica* 133: 3–8.

Ramsey, J. & Schemske, D.W. 1998. Pathways, mechanisms, and rates of polyploid formation in flowering plants. *Annual Rev. Ecol. Syst.* 29: 467–501.

Ramsey, J. & Schemske, D.W. 2002. Neopolyploidy in flowering plants. *Annual Rev. Ecol. Syst.* 33: 589–639.

Rapp, R.A. & Wendel, J.F. 2005. Epigenetics and plant evolution. *New Phytol.* 168: 81–91.

Rausch, J.H. & Morgan, M.T. 2005. The effect of self-fertilization, inbreeding depression, and population size on autopolyploid establishment. *Evolution* 59: 1867–1875.

Remski, M.F. 1954. Cytological investigations in *Mammillaria* and some associated genera. *Bot. Gaz.* 116: 163–171.

Riddle, N.C., Kato, A. & Birchler, J.A. 2006. Genetic variation for the response to ploidy change in *Zea mays* L. *Theor. Appl. Genet.* 114: 101–111.

Rieseberg, L.H. & Willis, J.H. 2007. Plant speciation. *Science* 317: 910–914.

Rodriguez, D.J. 1996. A model for the establishment of polyploidy in plants: Viable but infertile hybrids, iteroparity, and demographic stochasticity. *J. Theor. Biol.* 180: 189–196.

Rong, J., Feltus, F.A., Liu L., Lin, L. & Paterson, A.H. 2010. Gene copy number evolution during tetraploid cotton radiation. *Heredity* Advance Online Publication 17 February 2010; doi: 10.1038/hdy.2009.192

Sahley, C.T. 1996. Bat and hummingbird pollination of an autotetraploid columnar cactus, *Weberbauereocereus weberbaueri* (Cactaceae). *Amer. J. Bot.* 83: 1329–1336.

Sarvella, P. 1958. Multivalent formation and genetic segregation in some allopolyploid *Gossypium* hybrids. *Genetics* 43: 601–619.

Sax, K. 1936. The experimental production of polyploidy. *J. Arnold Arbor.* 17: 153–159.

Scannell, D.R., Byrne, K.P., Gordon, J.L., Wong, S. & Wolfe, K.H. 2006. Multiple rounds of speciation associated with reciprocal gene loss in polyploid yeasts. *Nature* 440: 341–345.

Segraves, K.A., Thompson, J.N., Soltis, P.S. & Soltis, D.E. 1999. Multiple origins of polyploidy and the geographic structure of *Heuchera grossulariifolia. Molec. Biol.* 8: 253–262.

Shoemaker, R.C., Schlueter, J. & Doyle, J.J. 2006. Paleopolyploidy and gene duplication in soybean and other legumes. *Curr. Opin. Pl. Biol.* 9: 104–109.

Simillion, C., Vandepoele, K., Van Montagu, M.C., Zabeau, M. & Van de Peer, Y. 2002. The hidden duplication past of *Arabidopsis thaliana. Proc. Natl. Acad. Sci. U.S.A.* 99: 13627–13632.

Soltis, D.E. 1984. Autopolyploidy in *Tolmiea menziesii* (Saxifragaceae). *Amer. J. Bot.* 71: 1171–1174.

Soltis, D.E., Albert, V.A., Leebens-Mack, J., Bell, C.D., Paterson, A., Zheng, C., Sankoff, D., Wall, P.K. & Soltis, P.S. 2009. Polyploidy and angiosperm diversification. *Amer. J. Bot.* 96: 336–348.

Soltis, D.E. & Burleigh, J.G. 2009. Surviving the K-T mass extinction: new perspectives of polyploidization in angiosperms. *Proc. Natl. Acad. Sci. U.S.A.* 106: 5455–5456.

Soltis, D.E. & Rieseberg, L.H. 1986. Autopolyploidy in *Tolmiea menziesii* (Saxifragaceae): Evidence from enzyme electrophoresis. *Amer. J. Bot.* 73: 310–318.

Soltis, D.E. & Soltis, P.S. 1989. Allopolyploid speciation in *Tragopogon*: Insights from chloroplast DNA. *Amer. J. Bot.* 76: 1119–1124.

Soltis, D.E. & Soltis, P.S. 1990. Chloroplast DNA and nuclear rDNA variation: Insights into autopolyploid and allopolyploid evolution. Pp. 97–118 in: Kawano, S. (ed.), *Biological approaches and evolutionary trends in plants.* New York: Academic Press.

Soltis, D.E. & Soltis, P.S. 1993. Molecular data facilitate a reevaluation of traditional tenets of polyploid evolution. *Crit. Rev. Pl. Sci.* 12: 243–273.

Soltis, D.E. & Soltis, P.S. 1999. Polyploidy: Origins of species and genome evolution. *Trends Ecol. Evol.* 14: 349–351.

Soltis, D.E., Soltis, P.S., Pires, J.C., Kovarik, A. & Tate, J.A. 2004. Recent and recurrent polyploidy in *Tragopogon* (Asteraceae):

Genetic, genomic, and cytogenetic comparisons. *Biol. J. Linn. Soc.* 82: 485–501.

Soltis, D.E., Soltis, P.S., Schemske, D., Hancock, J., Thompson, J.N., Husband, B. & Judd, W.S. 2007. Autopolyploidy and sympatric speciation in angiosperms: Have we grossly underestimated the number of species? *Taxon* 56: 13–30.

Soltis, D.E., Soltis, P.S. & Tate, J.A. 2004. Advances in the study of polyploidy since *Plant Speciation*. *New Phytol.* 161: 173–191.

Soltis, P.S., Doyle, J.J. & Soltis, D.E. 1992. Molecular data and polyploid evolution in plants. Pp. 177–201 in: Soltis, P.S., Soltis, D.E. & Doyle, J.J. (eds.), *Molecular systematics of plants*. New York: Chapman and Hall.

Soltis, P.S. & Soltis, D.E. 2000. The role of genetic and genomic changes in the success of polyploids. *Proc. Natl. Acad. Sci. U.S.A.* 97: 7051–7057.

Ståhlberg D. 2009. Habitat differentiation, hybridization and gene flow patterns in mixed populations of diploid and autotetraploid *Dactylorhiza maculata* s.l. (Orchidaceae). *Evol. Ecol.* 23: 295–328.

Stebbins, G.L. 1947. Types of polyploids: Their classification and significance. *Advances Genet.* 1: 403–429.

Stebbins, G.L. 1950. *Variation and evolution in plants*. New York: Columbia Univ. Press.

Stebbins, G.L. 1971. *Chromosomal evolution in higher plants*. London: Addison-Wesley.

Stebbins, G.L. 1985. Polyploidy, hybridization, and the invasion of new habitats. *Ann. Missouri Bot. Gard.* 72: 824–832.

Stift, M., Berenos, C., Kuperus, P. & Van Tienderen, P.H. 2008. Segregation models for disomic, tetrasomic and intermediate inheritance in tetraploids: A general procedure applied to *Rorippa* (Yellow cress) microsatellite data. *Genetics* 179: 2113–2123.

Sybenga, J. 1996. Chromosome pairing affinity and quadrivalent formation in polyploids: Do segmental allopolyploids exist? *Genome* 39: 1176–1184.

Symonds, V.V., Soltis, P. & Soltis, D.E. 2010. Dynamics of polyploid formation in *Tragopogon* (Asteraceae): Recurrent formation, gene flow, and population structure. *Evolution* 64: 1984–2003.

Tang, H., Wang, X., Bowers, J.E., Ming, R., Alam, M. & Paterson, A.H. 2008b. Unraveling ancient hexaploidy through multiply-aligned angiosperm gene maps. *Genomic Res.* 18: 1944–1954.

Tate, J.A., Joshi, P., Soltis, K., Soltis, P.S. & Soltis, D.E. 2009b. On the road to diploidization? Homoeolog loss in independently formed populations of the allopolyploid *Tragopogon miscellus* (Asteraceae). *BMC Pl. Biol.* 9: 80.

Tate, J.A., Ni, Z., Scheen, A.C., Koh, J., Gilbert, C.A., Chen, Z.J., Soltis, P.S. & Soltis, D.E. 2006. Evolution and expression of homeologous loci in *Tragopogon miscellus* (Asteraceae), a recent and reciprocally formed allopolyploid. *Genetics* 173: 1599–1611.

Tate, J.A., Soltis, D.E. & Soltis, P.S. 2005. Polyploidy in plants. Pp. 371–426 in: Gregory, T.R. (ed.), *The evolution of the genome*. San Diego: Elsevier Academic Press.

Tate, J.A., Symonds, V.V., Doust, A.N., Buggs, R.J.A., Mavrodiev, E.V., Soltis, P.S. & Soltis, D.E. 2009a. Synthetic polyploids of *Tragopogon miscellus* and *T. mirus* (Asteraceae): 60 years after Ownbey's discovery. *Amer. J. Bot.* 96: 979–988.

Tavoletti S., Mariani, A. & Veronesi, F. 1991. Phenotypic recurrent selection for 2*n* pollen and 2*n* egg production in diploid alfalfa. *Euphytica* 57: 97–102.

Thomas, B.C, Pedersen, B. & Freeling, M. 2006. Following tetraploidy in an *Arabidopsis* ancestor, genes were removed preferentially from one homeolog leaving clusters enriched in dose-sensitive genes. *Genome Res.* 16: 934–946.

Thompson, J.N. & Merg, K.F. 2008. Evolution of polyploidy and the diversification of plant-pollinator interactions. *Ecology* 89: 2197–2206.

Thompson, J.N., Nuismer, S.L. & Merg, K. 2004. Plant polyploidy and the evolutionary ecology of plant/animal interactions. *Biol. J. Linn. Soc.* 82: 503–510.

Thórsson, A.E.T., Salmela, E. & Anamthawat-Josson, K. 2001. Morphological, cytogenetic, and molecular evidence for introgressive hybridization in birch. *J. Heredity* 92: 404–408.

Udall, J.A. & Wendel, J.F. 2006. Polyploidy and crop improvement. *Crop Sci.* 46 (suppl. *The Plant Genome*): S3–S14.

Van de Peer, Y., Maere, S. & Meyer, A. 2009. The evolutionary significance of ancient genome duplications. *Nat. Rev. Genet.* 10: 725–732.

Veronesi, F., Mariani, A. & Bingham, E.T. 1986. Unreduced gametes in diploid *Medicago* and their importance in alfalfa breeding. *Theor. Appl. Genet.* 72: 37–41.

Vision, T.J., Brown, D.G. & Tanksley, S.D. 2000. The origins of genomic duplications in *Arabidopsis*. *Science* 290: 2114–2117.

Wagner, W.H. 1970. Biosystematics and evolutionary noise. *Taxon* 19: 146–151.

Wang, J., Tian, L., Lee, H.-S., Wei, N.E., Jiang, H., Watson, B., Madlung, A., Osborn, T.C., Doerge, R.W., Comai, L. & Chen, Z.J. 2006. Genomewide nonadditive gene regulation in *Arabidopsis* allotetraploids. *Genetics* 172: 507—517.

Wang, J., Tian, L., Madlung, A., Lee, H.-S., Chen, M., Lee, J.J., Watson, B., Kagochi, T., Comai, L. & Chen, Z.J. 2004. Stochastic and epigenetic changes of gene expression in *Arabidopsis* polyploids. *Genetics* 167: 1961–1973.

Warner, D.A. & Edwards, G.E. 1993. Effects of polyploidy on photosynthesis. *Photosynthesis Res.* 35: 135–147.

Wendel, J.F. 2000. Genome evolution in polyploids. *Pl. Molec. Biol.* 42: 225–249.

Wendel, J.F. & Doyle, J.J. 2005. Polyploidy and evolution in plants. Pp. 97–117, in: Henry, R. (ed.), *Diversity and evolution in plants*. Oxon: CABI Publishing.

Werth, C.R. & Windham, M.D. 1991 A model for divergent, allopatric speciation of polyploid pteridophytes resulting from silencing of duplicate-gene expression. *Amer. Naturalist* 137: 515.

Winge, Ö. 1917. The chromosomes, their number and general importance. *Compt.-Rend. Trav. Carlsberg Lab.* 13: 131–275.

Wolf, P.G., Soltis, P.S. & Soltis, D.E. 1989. Tetrasomic inheritance and chromosome pairing behaviour in the naturally occurring autotetraploid *Heuchera grossulariifolia* (Saxifragaceae). *Genome* 35: 655–663.

Wood, T.E., Takebayashi, N., Barker, M.S., Mayrose, I., Greenspoon, P.B. & Rieseberg, L.H. 2009. The frequency of polyploidy speciation in plants. *Proc. Natl. Acad. Sci. U.S.A.* 106: 13875–13879.

Yu, J., Wang, J., Lin, W., Li, S., Li, H., Zhou, J., Ni, P., Dong, W., Hu, S., Zeng, C., Zhang, J., Zhang, Y., Li, R., Xu, Z., Li, S., Li, X., Zheng, H., Cong, L., Lin, L., Yin, J., Geng, J., Li, G., Shi, J., Liu, J., Lv, H., Li, J., Wang, J., Deng, Y., Ran, L., Shi, X., Wang, X., Wu, Q., Li, C., Ren, X., Wang, J., Wang, X., Li, D., Liu, D., Zhang, X., Ji, Z., Zhao, W., Sun, Y., Zhang, Z., Bao, J., Han, Y., Dong, L., Ji, J., Chen, P., Wu, S., Liu, J., Xiao, Y., Bu, D., Tan, J., Yang, L., Ye, C., Zhang, J., Xu, J., Zhou, Y., Yu, Y., Zhang, B., Zhuang, S., Wei, H., Liu, B., Lei, M., Yu, H., Li, Y., Xu, H., Wei, S., He, X., Fang, L., Zhang, Z., Zhang, Y., Huang, X., Su, Z., Tong, W., Li, J., Tong, Z., Li, S., Ye, J., Wang, L., Fang, L., Lei, T., Chen, C., Chen, H., Xu, Z., Li, H., Huang, H., Zhang, F., Xu, H., Li, N., Zhao, C., Li, S., Dong, L., Huang, Y., Li, L., Xi, Y., Qi, Q., Li, W., Zhang, B., Hu, W., Zhang, Y., Tian, X., Jiao, Y., Liang, X., Jin, J., Gao, L., Zheng, W., Hao, B., Liu, S., Wang, W., Yuan, L., Cao, M., McDermott, J., Samudrala, R., Wang, J., Wong, G.K., Yang, H. 2005. The genomes of *Oryza sativa*: A history of duplications. *PLoS Biol.* 3(2): e38.

Zhao, X.P., Si, Y., Hanson, R.E., Crane, C.F., Price, H.J., Stelly, D.M., Wendel, J.F. & Paterson, A.H. 1998. Dispersed repetitive DNA has spread to new genomes since polyploid formation in cotton. *Genome Res.* 8: 479–492.

17