PERSPECTIVE AND HYPOTHESIS

Polyploidy Is Genetic Hence May Cause Non-Adaptive Radiations, Whereas Pseudopolyploidy Is Genomic Hence May Cause Adaptive Non-Radiations

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ABSTRACT

There are two ways eukaryotes double number of chromosomes: (1) whole genome duplication (polyploidy), in which all nuclear DNA is replicated, and (2) karyotypic fission (pseudopolyploidy), in which all chromosomes are physically bifurcated. We contrast polyploidy with pseudopolyploidy, highlighting when it is crucial to look at genetic vs. genomic levels. We review history of pseudopolyploidy, including recent mechanisms by which chromosomal bifurcation may occur and outline methods for detecting such genomic changes. We then delve into the evolutionary implications, with particular focus on adaptive potential, of these two forms of doubling chromosome numbers. We address the common assertion that polyploidy induces adaptive radiations, which contains three fallacies. First, while polyploidy causes quantum speciation, evolutionary theory implies that these radiations should be non-adaptive. Polyploidy causes reproductive isolation, minute effective population sizes, and increased mutation rates, which all imply a diminished role for selection. Second, due to lack of karyotyping in recent decades and lack of distinction between genomic and genetic effects, it is usually impossible to detect pseudopolyploids. Third, pseudopolyploids lack minority cytotype exclusion because they readily backcross with their progenitors, which thereby means no reproductive isolation for newly formed pseudopolyploids. Pseudopolyploidy will thereby not result in radiations until pseudopolyploid descendants undergo subsequent chromosome rearrangements or grow new centromeres. Pseudopolyploids may have a modest selective advantage over their progenitors due to diminished linkage disequilibrium. Thus, pseudopolyploidy may induce adaptive non-radiations. We encourage a renaissance of karyotyping to distinguish between these two mechanisms and a renaissance in genomic perspectives in evolution. *J. Exp. Zool. (Mol. Dev. Evol.) 320B: 286–294, 2013.* © 2013 Wiley Periodicals, Inc.

J. Exp. Zool. (Mol. Dev. Evol.) 320B:286–294, 2013

How to cite this article: Gorelick R, Olson K. 2013. Polyploidy is genetic hence may cause non-adaptive radiations, whereas pseudopolyploidy is genomic hence may cause adaptive non-radiations. J. Exp. Zool. (Mol. Dev. Evol.) 320B:286–294.

In our personal lives, when looking for housing, we focus on large-scale aspects of architecture, such as number of bedrooms and bathrooms and whether or not the unit is attached to others on the sides (townhouse) or above and below (apartment building). Small-scale aspects of architectures still concern us, but less so, for example we may care marginally about whether the exterior of the building is clothed in aluminum, vinyl, brick, or adobe. But one of us (Root) still opted to live in a building clothed in what we

This article was published online on 29 May 2013. Subsequently, it was determined that Table 1 had been edited incorrectly, and the correction was published on 7 June 2013.

Grant sponsor: Natural Sciences and Engineering Research Council of Canada (NSERC).

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Received 1 October 2012; Accepted 12 March 2013
DOI: 10.1002/jez.b.22499
Published online in Wiley Online Library (wileyonlinelibrary.com).

consider to be the ugliest of gray bricks. Likewise, when examining the DNA of an organism, we can focus on large-scale aspects, such as karyotypes that have been acknowledged to be evolutionarily important at least since the 1880s (Boveri, 1889; Wilson, 1896), or small-scale aspects, such as the DNA sequences elucidated by Watson and Crick ('53).

Large-scale aspects of nuclear DNA, dealing with pieces of chromosomal material that can be seen under a light microscope, are known as genomic. Small-scale aspects of nuclear DNA, dealing with small numbers of nucleotides that can only be quantified with sequencing or other fine-scale techniques, are known as genetic. While evolutionary biologists have largely focused on genetic aspects since the advent of sequencing and PCR (Kreitman, '83; Mullis, '90), we should not forget about the evolutionary implications of changes in genome architecture. While there exists plenty of work on evolutionary and developmental aspects of crossing-over recombination, which is a genomic phenomenon, in this paper we focus on an even more substantial change in genome architecture, namely doubling the number of chromosomes.

There are substantial qualitative differences between genomic and genetic views of evolution (Heng, 2009). Lynch's (2007b) book, "shows that it is only by looking at the details of genome architecture and associated population genetics that we can really see how important non-adaptive evolutionary explanations can be" (Gorelick, 2008, p. 165). Evolution of cancer, including susceptibilities to cancer, can only be understood from a genomic perspective, as demonstrated by the high incidences of chromosomal rearrangements and aneuploidy in cancer cells (Heng et al., 2009; Heng et al., 2011). We showed that only by looking at the genome level can the functions and origins of sex be understood (Gorelick and Heng, 2011). We also showed that radiations in plants may largely be a result of polyploidy, aka whole genome duplication, which (antithetically, as we show below) is definitely a genetic rather than a genomic phenomenon (Gorelick and Olson, 2011).

In our personal lives, what happens when twice as many people as expected show up for supper? We can go back into the kitchen and cook twice as much food. Alternatively, we could simply serve each person half as much food. The same two alternatives exist for genomic architecture of eukaryotes that double their number of chromosomes: replicate all nucleotides in each nucleus or chop each chromosome in half. The former tactic, known as whole genome duplication or polyploidy, results in double the weight of DNA per nucleus. The latter tactic, known as karyotypic fission or pseudopolyploidy, results in no change in weight of DNA per nucleus. This paper is a unification of a long-recognized but oft-forgotten pattern (pseudopolyploidy; since at least Vandel, '37) with a recently explicated molecular mechanism (relative timing of centromere replication; Kolnicki, 2000; Perry et al., 2004), and a nascent evolution theory (genome theory; outlined herein), distinct from polyploidy via whole genome duplication. We discuss how a nucleus may end up with double the number of chromosomes of its ancestors via different mechanisms and how one can distinguish between these two mechanisms when examining the contents of a nucleus. We also contrast the very different evolutionary implications of polyploidy and pseudopolyploidy with regard to adaptive and radiative potential, even though the two mechanisms are often erroneously conflated.

TWO WAYS TO DOUBLE THE NUMBER OF CHROMOSOMES

Ploidy levels are seemingly simple enough to calculate. Count the number of chromosomes per nucleus for taxa along a phylogeny. If a derived taxon has double the number of chromosomes per nucleus than one of its putative ancestors, then infer that polyploidy has occurred. For taxa with one nucleus per cell, this amounts to counting chromosomes per cell. Even for the numerous taxa with endoploidy, it is simple enough to look for the base (minimum) number of chromosomes per diploid or haploid nucleus. The problem is that there are two ways to double the number of chromosomes per nucleus, each of which have very different evolutionary consequences.

Usually it is assumed that polyploidy means that the entire nuclear genome has duplicated (e.g., Sobel et al., 2010). Whole genome duplication can itself occur in two ways: via fusion of two "unreduced gametes" or via endomitosis, which is duplication of all chromosomes without the subsequent nuclear division. (Being that gametes and gametophytes are stages with reduced ploidy, the phrase "unreduced gametes" is an overused oxymoron, but we will not offer up a better alternative.) As has been well known at least since Stebbins ('71), polyploidy results in reproductive isolation and allows for tinkering of the redundant portions of the genome via neo-functionalization and sub-functionalization (Force et al., '99). But polyploidy probably does not result in adaptive radiations (Schluter, 2000). We will elaborate on the evolutionary consequences of polyploidy, but first describe another way that numbers of chromosomes can double.

Number of chromosomes per nucleus can double when all chromosomes are simultaneously divided in two at or near the centromere(s), a process known as agmatoploidy, karyotypic fission, and/or our preferred term, pseudopolyploidy (Vandel, '37; Nordenskiöld, '51; Tobias, '53; Mello-Sampayo, '61; Todd, '70). Chromosomal fission, akin to aneuploidy, is the step-wise process of increasing the chromosome number by dividing individual chromosomes rather than the entire karyotypic complement. Nonetheless, this process can also double or nearly double the number of chromosomes in a nucleus (Olson and Gorelick, 2011).

Pseudopolyploidy is probably more common than generally believed, but has been seldom investigated and, even then, is often dismissed for specious reasons. For example, pseudopolyploidy is not mentioned in Rieseberg's (2001) review of chromosomal rearrangements. At least in plants, chromosomal fission (and likely pseudopolyploidy) is more prevalent than the literature

suggests, but its evidence is often obscured by further changes to individual chromosomes or by subsequent polyploidy (Jones, '98; also refer to the review by Perry et al., 2004). Perhaps the existence of pseudopolyploidy may not have been taken seriously because there did not exist any mechanisms by which all chromosomes could be simultaneously fissioned, although pseudopolyploidy has been well documented since at least Vandel ('37) and was further highlighted in both plants and animals (Nordenskiöld, '51; Tobias, '53; Todd, '70; Giusto and Margulis, '81; Kolnicki, '99). Pseudopolyploidy could be taken more seriously once Kolnicki (2000) and Perry et al. (2004) elucidated heterochronic mechanisms by which it could occur. The first step in almost any chromosomal replication in any eukaryote is for the centromeres to be duplicated (Earnshaw and Tomkiel, '92). If replication of the remainder of the chromosomes is inhibited, but the rest of the cell cycle continues, then the resulting daughter cells will have twice as many chromosomes (Kolnicki, 2000). The mechanism is referred to as centric duplication-fission (Perry et al., 2004). However, note that all the newly fissioned chromosomes will be telocentric (unless isochromosomes are formed; see Perry et al., 2004). Therefore, one larger metacentric (i.e., bi-armed) chromosome is divided into two smaller telocentric (i.e., single-armed) chromosomes (refer to Fig. 1). It is also possible to get the same result but via fission of centromeres (in lieu of duplication of centromeres). Regardless of the means, the end is the same: double the number of chromosomes with no genome replication.

Note that pseudopolyploidy will only double the number of chromosomes that are not strictly telocentric. If the centromere appears at one end of the chromosome, neither centromeric duplication nor centromeric fission will result in a new chromosome. For example, for a nucleus containing ten pairs of metacentric chromosomes and five pairs of telocentric chromosomes, fission would yield daughter nuclei with 25 telocentric chromosomes. Also note that a similar process, called agmatoploidy, occurs in organisms with holocentric centromeres, those which span the entire length of the chromosome rather than occur in a discrete locale.

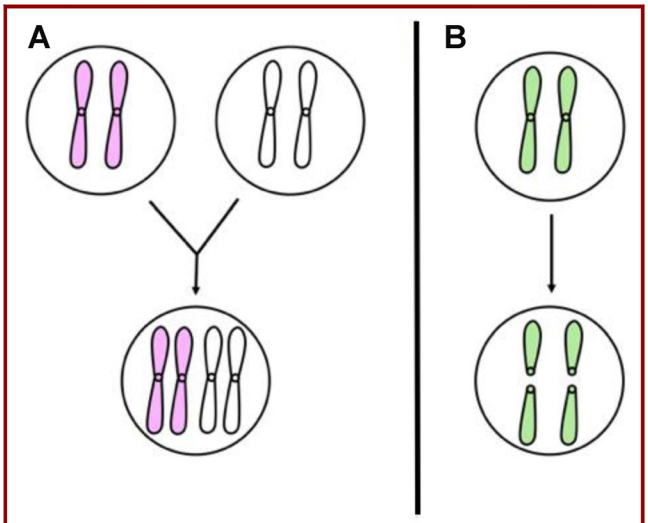

Figure 1. A: Polyploidy (whole genome duplication): each mother nucleus contains two chromatids with a ploidy level of 2n. Daughter nucleus contains four chromatids. B: Pseudopolyploidy (karyotypic fission): the mother nucleus has two chromatids, while the daughter nucleus contains four chromatids.

Given that endopolyploidy is now recognized as being increasingly common, including during meiosis ("pre-meiotic doubling"), in retrospect, it should not be too surprising that recently duplicated centromeres in meiosis I are either separated or fissioned, resulting in pseudopolyploidy. What we now need are ways to distinguish pseudopolyploidy from polyploidy and the evolutionary implications of pseudopolyploidy.

DISTINGUISHING POLYPLOIDY (DUPLICATION) FROM PSEUDOPOLYPLOIDY (FISSIONING)

Prior to the 1980s, whenever people collected and published karyotypes, genome duplication and genome fissioning were easy to distinguish. We know of one botanist that still does chromosome counts in the field (Baker et al., 2009), but such instances are a rarity in modern times. Most of the volumes in the series "Animal Cytogenetics" were published in 1974, although the final volumes on amphibians and birds were published in 1990 (Olsen, '66; Borganonkar, '74; Egozcue, '74; Fregda, '74; Gustavsson, '74; Hayman and Martin, '74; Ohno, '74; Patton, '74; Crozier, '75; White and Webb, '76; Smith and Virkki, '78; Hewitt, '79; Ueshima, '79; Matuszewski, '82; Christidis, '90; King, '90). The journal Cytogenetics changed its name to Cytogenetics and Cell Genetics in 1973 and changed again to Cytogenetics and Genome Research in 2002 to reflect emphasis away from classical karyotyping. Currently, instead of pulling out a microscope to count chromosomes in squashed cells, many researchers count single-copy genes or run flow cytometry or microdensitometry.

For decades, the Missouri Botanical Garden published the Index to Plant Chromosome Numbers and made the data publicly available online (Goldblatt and Johnson, 2008). While very useful at a glance, this database provides only number of chromosomes, not ploidy level nor whether increased chromosome number was due to polyploidy or pseudopolyploidy. Additionally, many cited records do not include a karyotype image, which makes interpretation of the numbers nigh impossible. We do not know of similar databases for animal, fungal, or other eukaryotic chromosome numbers.

Databases exist that report the mass of chromosomes per nucleus (C-values, in pg) in plants, animals and fungi, largely based on flow cytometric and microdensimetric measurements (Bennett and Leitch, 2005; Kullman et al., 2005; Gregory, 2010). If chromosome numbers double and C-values double for the same

pair of species, then we can infer that whole genome duplication has occurred. If chromosome numbers double and C-values remain unchanged, then we can infer that chromosomal fissioning has occurred. Apparently nobody has yet tried merging these two databases to distinguish polyploidy from pseudopolyploidy, an omission that may be due to non-overlapping taxa in the databases for chromosome number and C-value. Only a few taxa exist with both chromosome number and C-value because much work is done without cell squashes. Even if each database contained the same taxa, there are mechanisms (accumulation of transposable elements and satellites) that substantially alter C-values without altering ploidy, which confound inferring ploidy via combining chromosome numbers and C-values. For example, the congeneric palms *Pinanga coronata* and *P. subintegra* have 16 pairs of chromosomes (Sarkar, '70), but a huge difference in C-values, 35.42 vs. 55.62 pg (Röser et al., '97). Similar differences in C-content without differences in chromosome numbers are found in kangaroo rats, genus *Dipodomys* (John and Miklos, '79) due to differences in satellite DNA, and a twofold difference in C-values in the rice genus *Oryza* due to transposable elements (Chen and Wu, '82; Piegu et al., 2006).

To best infer whether polyploidy or pseudopolyploidy has occurred is to examine karyotypes. This way, it will be possible to also detect aneuploidy, pericentric inversions, Robertsonian fusions, etc.

Having defined polyploidy and pseudopolyploidy and outlined ways to distinguish the dramatically different mechanisms with empirical data, we now address the evolutionary implications of each. Of particular note, neither polyploidy nor pseudopolyploidy is expected to cause adaptive radiations. Rather, evolutionary theory predicts that polyploidy will produce non-adaptive radiations and pseudopolyploidy will produce adaptive non-radiations.

POLYPLOIDY CAUSES RADIATIONS, BUT IS NOT ADAPTIVE

There is no doubt that progeny formed via polyploidy—regardless of whether they are due to autopolyploidy, allopolyploidy, or endomitotic reduplication—are reproductively isolated from their parents. Backcrosses of parent and offspring form individuals with an odd number of each homologous chromosome. While triploids are not evolutionary dead-ends, they seldom introgress with either their diploid or tetraploid parents. Most polyploid populations probably go extinct due to what Levin ('75) called minority cytotype disadvantage. However, occasionally, independent segregation of triploid nuclei will produce a haploid or diploid gamete, but the probability of this grows exponentially small as the number of homologous chromosomes increases. Theoretically, some of the most likely routes out of triploid sterility are (1) production of "unreduced" (triploid) gametes plus syngamy to form a hexaploid, (2) production of "unreduced" triploid gametes that backcross with haploid gametes to form tetraploid offspring, the so-called triploid bridge, (3) endomitosis to produce a hexaploid, and (4) hybridogenetic reduction or parasex to remove either one or two sets of homologous chromosomes to produce either diploid or haploid gametes or gametophytes (Diaz et al., '96; Ramsey and Schemske, '98; Ali et al., 2001; Albertin and Marullo, 2012). Hexaploids are still reproductively isolated from their diploid or tetraploid ancestors, tetraploids are still reproductively isolated from their diploid ancestors, while hybridogenetic reduction or parasex usually completely eliminates the entire genome of either the diploid or tetraploid ancestor. There are thus some chances for genetic leakage via triploids, but not many. Polyploidy forms a reproductive isolating barrier, albeit not an entirely complete one (Ramsey and Schemske, '98; Slotte et al., 2008). By the biological species concept, neopolyploids are therefore new species. Repeated polyploidization, even if created via fusion of unreduced gametes (Segraves et al., '99; Vanichanon et al., 2003) consequently can cause radiations (Soltis and Soltis, '93).

Numerous well respected authors suggest that polyploidy is adaptive (e.g., Rieseberg and Willis, 2007; Fawcett et al., 2009; Maherali et al., 2009; Ouarda et al., 2009; Soltis et al., 2009; Albertin and Marullo, 2012). Molecular mechanisms have been proposed for how polyploid formation induces adaptation, such as reciprocal epigenetic silencing that results from polyploidization, at least after a few generations (Chen, 2007; Vyas et al., 2007; Flagel and Wendel, 2009), especially for allopolyploids (Salmon et al., 2005). While we have no doubt that these epigenetic changes occur and may even have some selective advantage, they do not necessarily lead to adaptation.

Why is there a misconception that polyploidy is adaptive? We take a conservative approach and will naively assume that an adaptive radiation means rapid speciation in which newly formed species have a selective advantage over their ancestors, although a few studies have shown selective disadvantages of polyploidy (e.g., D'Souza et al., 2005). To demonstrate adaption is much more onerous than just showing a fitness advantage (Williams, '66; Schluter, 2000). We must be clear that we are not implying that polyploidy is maladaptive. Rather, we show below that genome theory predicts non-adaptive outcomes following a polyploidization event. Non-adaptive could mean that phenotypes are selectively neutral or that effects of selection are masked by, say, hitchhiking.

If polyploids are formed via autoploidy or endomitosis, there is little reason to suspect that the polyploids will be any better adapted than their ancestors insofar as they all have the same genotype. While neo-functionalization of homeologues is possible, it is rare, especially compared with non-functionalization and sub-functionalization (Force et al., '99; Lynch, 2007b). Unlike neo-functionalization, sub-functionalization is not itself adaptive, but may induce neo-functionalization and may reduce linkage disequilibrium. Silencing of homeologous genes seems to be reciprocal, but otherwise random (Adams et al., 2003). While autoploids may have phenotypic divergence from their diploid

progenitors, there is no evidence that shift in phenotype is advantageous (Oswald and Nuismer, 2011). Seemingly the only easy way to get adaptive polyploidy is via allopolyploidy, with fitness increases due to heterozygote advantage at many loci. Polyploidy may generate increased phenotypic plasticity (Gorelick, 2005), affecting the interactions of plants and their pollinators, herbivores or pathogens, perhaps yielding adaptive change (Segraves et al., '99; Thompson et al., 2004), although there exists some empirical evidence suggesting that polyploidy does not increase plasticity (Munzbergova, 2007). Regardless of plasticity we argue in the next three paragraphs that polyploid formation should result in increased drift, linkage, and mutation, tilting the balance away from selection in both mutation-selection balance and drift-selection balance.

Because of their reproductive isolation, neopolyploids have effective population sizes that are very small (Holmes et al., 2009). Small effective population sizes are more subject to drift: even highly advantageous mutations may be lost from the population by chance, while highly deleterious mutations can accumulate (Rieseberg, 2001; Lynch, 2006). Gorelick (2009) outlined the foundational role of drift in cactus evolution. Based on the population genetic structure of the group, including small effective population sizes and relatively long generation times, the standard adaptive stories give way to the random non-adaptive action of drift. Given the dramatic decrease in effective population size following polyploidy and increased generation time due to genome expansion (sensu Gregory, 2002), drift will therefore dominate selection in the population genetics of newly formed polyploids.

Larger genomes have higher mutation rates (measured in base pairs/genome/generation). This correlation is evident when examining correlations of mutation rates and genome size across all eukaryotes. Lynch (2006) found that doubling genome size results in 1.72 times the number of mutations. Higher mutation rate is also expected for neopolyploids insofar as huge portions (~25%) of their homeologous chromosomes are silenced, probably via cytosine methylation (Adams et al., 2003). Methylated cytosines are by far the richest source of transitions and transversions, in the form of $C \rightarrow T$ point mutations (Gorelick, 2003 and references therein). Mutation-selection balance models yield allele frequencies as a function of the ratio of mutation rate divided by selection coefficient, and there is little a priori reason to believe that polyploidy will cause anywhere near as large a change in selection coefficient as in mutation rate.

Earlier, we alluded to the fact that sub-functionalization induced by polyploidy should reduce linkage disequilibrium, thereby slightly increasing multi-locus selection coefficients, that is reducing genetic hitchhiking (Bürger, 2000). This slight increase in size of selection coefficients due to polyploidy, however, is a minor effect, especially when compared with the doubling or tripling of mutation rates due to the same polyploidy. Furthermore, in the long-run, linkage disequilibrium should in fact increase for polyploid lineages that eventually undergo diploidization, as is usually believed to occur (Ma and Gustafson, 2005; Chen et al., 2007; Sémon and Wolfe, 2007; Berjano et al., 2009; Ozkan and Feldman, 2009).

Overall, there should be large increases in drift and mutation in polyploid lineages, with little or no change in selection (Table 1). This not a scenario in which adaptation would ever be expected (Lynch, 2007a, b). For further discussion of how polyploidy probably causes radiations that are not adaptive see Gorelick (2009) and Gorelick and Olson (2011).

Note that each of the arguments regarding drift, mutation, and reciprocal epigenetic silencing are genetic arguments, focusing on individual genetic loci. Thus, while a naïve perspective would indicate that polyploidy is a genomic phenomenon due to whole genome duplication, in fact, most of the effects of polyploidy are genetic. The only evolutionary consequence of polyploidy that seems genomic is reproductive isolation notwithstanding triploid bridges. By contrast, pseudopolyploidy seems entirely genomic, with nary a single effect at a single locus, unless one is solely concerned with centromeres.

PSEUDOPOLYPLOIDY MAY BE ADAPTIVE, BUT WILL NOT CAUSE RADIATIONS

Unlike with polyploidy, pseudopolyploidy does not lead to quantum speciation. Pseudopolyploids can and often do backcross with their unfissioned progenitors and their descendants. Except

Table 1. Evolutionary implications of polyploidy (whole genome duplication) vs. pseudopolyploidy (karyotypic fission).

	Effective population size (N_e)	Mutations/site per generation (μ)	Subfunctionalization	Linkage disequilibrium (LD)
Polyploidy	$N_e \approx 0$ non-adaptive	$\mu \uparrow$ non-adaptive	Yes usually non-adaptive	LD $\uparrow$ non-adaptive
Pseudo-polyploidy	$\Delta N_e = 0$ –	$\Delta \mu = 0$ –	No –	LD $\downarrow$ adaptive?

Polyploidy causes non-adaptive radiations.
Pseudopolyploidy causes adaptive non-radiations.

for maybe the centromere, which is composed almost entirely of repetitive elements (Jiang et al., 2003), every gene on the fissioned genome has a homologue on the unfissioned genome. Each unfissioned metacentric chromosome can synapsis with two fissioned telocentric chromosomes (Todd, '70; Margulis and Sagan, 2002). Meiosis then proceeds as usual in the backcross. In subsequent backcrosses to either fissioned or unfissioned individuals, the resulting diploid offspring can be a mélange of fissioned and unfissioned chromosomes. There is therefore no obvious minority cytotype exclusion in pseudopolyploids.

Eventually some of the fissioned (telocentric) chromosomes may once again become metacentric via incipient formation of a centromere from transposable elements (Kidwell and Lisch, 2001) or through subsequent pericentric inversion (Todd, '70; Kolnicki, '99, 2000). At this juncture backcrosses may become incapable of meiosis due to failure to synapse. However, this is not believed to happen often, hence we expect some, but not rampant, speciation.

Empirical surveys show that pseudopolyploidy can cause modest diversifications, but not the explosive diversifications known as radiations. Vandel ('37) reviews several examples of apparent pseudopolyploidy in nematodes, platyhelminthes, annelids, arthropods, and even some vertebrates. Nordenskiöld ('51) showed that pseudopolyploidy exists in plants, albeit only in one genus of sedge. And ironically sedges have diffuse centromeres, that is holocentric chromosomes. Tobias ('53) provided much more definitive evidence for pseudopolyploid-induced modest diversification, albeit only in mammals. Todd ('70) independently provided even more evidence for pseudopolyploidy and modest diversification, albeit also only in mammals. Giusto and Margulis ('81) and Kolnicki ('99) followed Todd ('70) with evidence of pseudopolyploidy in additional mammalian taxa. Olson and Gorelick (2011) provide evidence for pseudopolyploidy and a modest radiation of the family Zamiaceae in the otherwise depauperate gymnosperm order Cycadales.

Because pseudopolyploidy does not change genome size (except for possibly a slight increase in size of centromeres), but simply partitions the existing genome into smaller chromosomes, there are no theoretical reasons nor empirical evidence for pseudopolyploidy causing any changes in drift or mutation. Effective population sizes should remain unchanged due to meiotic recognition of both fissioned and unfissioned homologues. Chromosomal fission—because it does not involve any transpositions, insertions, deletions, or duplications—should not cause any changes in cytosine methylation. The only question is whether pseudopolyploidy induces changes in selection.

There appears to be no empirical studies on whether pseudopolyploidy alters drift or mutation rates because pseudopolyploidy has been so poorly studied and/or conflated with polyploidy. We must rely on theory here. In a similar vein, we question studies showing correlation (or lack thereof) between polyploidy and genetic variance in populations because the term polyploidy has sometimes included a mix of both forms of chromosome number doubling: whole genome duplication and pseudopolyploidy. It is also difficult assessing the roles of drift and mutation rates in pseudopolyploids because of lack of controlling for population size (usually large in diploids and small in polyploids with whole genome duplication) when comparing genetic variances of populations.

Chromosomal fission always causes an unequivocal reduction in linkage disequilibrium. Linkage groups that could previously only be broken with crossing-over recombination, can now be broken with segregation (Todd, '70). This may be adaptive if the previously linked loci were an example of deleterious genetic hitchhiking. On the other hand, it may be maladaptive if a co-adapted gene complex is more easily broken. Instead of focusing on whether fission is adaptive; we merely want to know whether selection is affected (not worrying if fitness increases or decreases). By reducing linkage disequilibrium, multi-locus selection will increase insofar as selection can act at loci that were previously shielded by hitchhiking (Levin, 2002). It is therefore at least possible that pseudopolyploidy might be adaptive in some circumstances.

There are, however, instances when pseudopolyploidy (and polyploidy) may be deleterious, when the number of chromosomes grows so large that there is a high probability of aneuploidy. Once the number of chromosomes becomes sufficiently high, aneuploidy becomes common, as seen in taxa with over 250 pairs of homologous chromosomes, for example, the pteridophyte genus *Ophioglossum*, the monocot genus *Voanioala*, and the eudicot genus *Echeveria* (Löve et al., '77; Johnson et al., '89; Uhl, 2007). True ferns (i.e., other than those in the Ophioglossales) are likely the product of an ancestral karyotypic fission event followed by one or more rounds of whole genome duplication, with karyotypes dominated by telocentric chromosomes (Soltis and Soltis, '87). The eudicot genus *Echeveria* seems to be the product of both chromosomal fission and polyploidy (Uhl, '92, 2007). Regardless, exceedingly high chromosome numbers often incur missteps in segregation.

Note that pseudopolyploidy is genomic, not genetic. Pseudopolyploidy diminishes linkage by physically separating linked loci on what were originally the same chromosome. Pseudopolyploidy does not affect any single locus ("genetic") phenomena: pseudopolyploidy does not change per locus mutation rates, per locus selection coefficients. Compare this with polyploidy, as summarized in Table 1.

CONCLUDING REMARKS

There exist two fundamentally different ways of doubling the number of chromosomes: (1) duplicating all DNA within each nucleus or (2) chopping each chromosome into two pieces at or near the centromere(s). While both are often called polyploidy, only whole genome duplication should bear this name because of

the radically different evolutionary implications of these two processes.

Whole genome duplication causes quantum speciation, albeit with the price of drastic reductions in effective population size and increased mutation rates, both of which effectively mask selection. Increased drift and mutation of polyploids are a prediction of classical genetic (including epigenetic) and population genetic models. Sub-functionalization of neopolyploids is likewise a typical genetic phenomenon that is best investigated at small scales, that is by examining a small number of DNA nucleotides. This classical form of polyploidy, whole genome duplication, should thus cause non-adaptive radiations.

Pseudopolyploidy, aka karyotypic fission, is a very poorly known phenomenon, although we are not altogether sure why it is unknown other than lack of karyotyping in recent decades (but see Faraut, 2008; Ouarda et al., 2009). Both early proponents of pseudopolyploidy were famous, so their papers should not have been lost to obscurity. Albert Vandel was famous for first proposing geographical parthenogenesis (Vandel, '28). Phillip Tobias, who passed away in 2012, was famous for uncovering the Piltdown Man hoax (Tobias, '92), describing *Homo habilis* from Olduvai Gorge (Leakey et al., '64), and being a very early white South African opponent of apartheid. Unfortunately mechanisms underlying pseudopolyploidy (fissioning all chromosomes at centromeres during meiosis) was not explicated until after biologists eschewed karyotyping and genomics for the more modern world of DNA sequencing and genetics. Due to lack of karyotyping, we do not know how common pseudopolyploidy is in plants or animals. We suspect that many minor diversifications of lineages are due to pseudopolyploidy, especially when systematists use karyotypes for distinguishing species. Pseudopolyploidy should not cause rampant speciation because backcrosses of pseudopolyploids and their unfissioned relatives are perfectly viable and fertile. However, unlike with whole genome duplication, pseudopolyploidy should not increase drift or mutation and may even provide a slight selective advantage through physical bifurcation of the chromosomes. Therefore, pseudopolyploidy should result in adaptive non-radiations.

Polyploidy is very much a genetic phenomenon, while pseudopolyploidy is a genomic phenomenon that is best studied by looking at whole chromosomes or large portions thereof. A genetic phenomenon, such as polyploidy, should therefore have radically different evolutionary implications than a genomic phenomenon, such as pseudopolyploidy, Furthermore, based on evolutionary theory, neither polyploidy nor pseudopolyploidy is expected to produce adaptive radiations. However, the action of polyploidy on the evolutionary landscape of eukaryotes cannot be denied, while the effects of pseudopolyploidy requires further study, especially empirical study. Thus, not only do we encourage evolutionary biologists to once again start karyotyping (as well as measuring C-values) but also supplement their evolutionary genetics with some evolutionary genomics.

ACKNOWLEDGMENT

We thank Natural Sciences and Engineering Research Council of Canada (NSERC) for a Discovery Grant to R.G.

LITERATURE CITED

Adams KL, Cronn R, Percifield R, Wendel JF. 2003. Genes duplicated by polyploidy show unequal contributions to the transcriptome and organ-specific reciprocal silencing. Proc Natl Acad Sci USA 100:4649–4654.

Albertin W, Marullo P. 2012. Polyploidy in fungi: evolution after whole-genome duplication. Proc R Soc B Biol Sci 279:2497–2509.

Ali SNH, Huigen DJ, Ramanna MS, Jacobsen E, Visser RGF. 2001. Genomic *in situ* hybridization analysis of a trigenomic hybrid involving *Solanum* and *Lycopersicon* species. Genome 44:299–304.

Baker MA, Rebman JP, Parfitt BD, Pinkava DJ, Zimmerman AD. 2009. Chromosome numbers in sone cacti of western North America—VIII. Haseltonia 1:117–134.

Bennett MD, Leitch IJ. 2005. Angiosperm DNA C-values database (release 6.0, Oct. 2005) Available online at: http://www.rbgkew.org.uk/cval/homepage.html

Berjano R, Roa F, Talavera S, Guerra M. 2009. Cytotaxonomy of diploid and polyploid *Aristolochia* (Aristolochiaceae) species based on the distribution of CMA/DAPI bands and 5S and 45S rDNA sites. Plant Syst Evol 280:219–227.

Borganonkar D. 1974. Animal cytogenetics: Chordata 4: Mammalia II: Placentalia: 4. Insectivora and Chiroptera. Stuttgart: Gebrüder Borntraeger.

Boveri T. 1889. Ein geschlechtlich erzeugter organismus ohne mütterliche eigenschaften. Sitz Gesel Morph u Physiol Müchen 5:73–83.

Bürger R. 2000. The mathematical theory of selection, recombination, and mutation. Chichester: Wiley.

Chen ZJ. 2007. Genetic and epigenetic mechanisms for gene expression and phenotypic variation in plant polyploids. Annu Rev Plant Biol 58:377–406.

Chen Y-J, Wu H-K. 1982. A comparison of karyotypes among six *Oryza* species. Bot Bull Acad Sin 23:163–183.

Chen LZ, Lou QF, Zhuang Y, et al. 2007. Cytological diploidization and rapid genome changes of the newly synthesized allotetraploids *Cucumis* × *hytivus*. Planta 225:603–614.

Christidis L. 1990. Animal cytogenetics: Chordata 3b: Aves. Stuttgart: Gebrüder Borntraeger.

Crozier RH. 1975. Animal cytogenetics: Insecta 7: Hymenoptera. Berlin: Gebrüder Borntraeger.

Diaz J, Schmiediche P, Austin DF. 1996. Polygon of crossability between eleven species of *Ipomoea*: section Batatas (Convolvulaceae). Euphytica 88:189–200.

D'Souza TG, Storhas M, Michiels NK. 2005. The effect of ploidy level on fitness in parthenogenetic flatworms. Biol J Linn Soc 85:191–198.

Earnshaw WC, Tomkiel JE. 1992. Centromere and kinetochore structure. Curr Opin Cell Biol 4:86–93.

Egozcue J. 1974. Animal cytogenetics: Chordata 4: Mammalia II: Placentalia: 5. Primates. Stuttgart: Gebrüder Borntraeger.

Faraut T. 2008. Addressing chromosome evolution in the whole-genome sequence era. Chromosome Res 16:5–16.

Fawcett JA, 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 USA 106:5737–5742.

Flagel LE, Wendel JF. 2009. Gene duplication and evolutionary novelty in plants. New Phytol 183:557–564.

Force A, Lynch M, Pickett FB, et al. 1999. Preservation of duplicate genes by complementary, degenerative mutations. Genetics 151:1531–1545.

Fregda K. 1974. Animal cytogenetics: Chordata 4: Mammalia II: Placentalia: 2. Carnivora. Stuttgart: Gebrüder Borntraeger.

Giusto JP, Margulis L. 1981. Karyotypic fission theory and the evolution of old world monkeys and apes. BioSystems 13:267–302.

Goldblatt P, Johnson DE, editors. 2008. Index to plant chromosome numbers, 2004–2006. St. Louis: Missouri Botanical Garden. Available online at: http://www.tropicos.org/Project/IPCN [accessed 9 April 2013]

Gorelick R. 2003. Evolution of dioecy and sex chromosomes via methylation driving Muller's ratchet. Biol J Linn Soc 80:353–368.

Gorelick R. 2005. Environmentally-alterable additive genetic variance. Evol Ecol Res 7:371–379.

Gorelick R. 2008. Review of "The origins of genome architecture" by Michael Lynch. Plant Sci Bull 54:165.

Gorelick R. 2009. Evolution of cacti is largely driven by genetic drift, not selection. Bradleya 27:41–45.

Gorelick R, Heng HHQ. 2011. Sex reduces genetic variation: a multidisciplinary review. Evolution 65:1088–1098.

Gorelick R, Olson K. 2011. Is lack of cycad (Cycadales) diversity a result of a lack of polyploidy? Bot J Linn Soc 165:156–167.

Gregory TR. 2002. Genome size and developmental complexity. Genetica 115:131–146.

Gregory TR. 2010. Animal genome size database. Available online at: http://www.genomesize.com [accessed 9 April 2013]

Gustavsson I. 1974. Animal cytogenetics: Chordata 4: Mammalia II: Placentalia: 3. Ungulata. Stuttgart: Gebrüder Borntraeger.

Hayman DL, Martin PG. 1974. Animal cytogenetics: Chordata 4: Mammalia I: Monotremata and Marsupialia. Stuttgart: Gebrüder Borntraeger.

Heng HHQ. 2009. The genome-centric concept: resynthesis of evolutionary theory. BioEssays 31:512–525.

Heng HHQ, Bremer SW, Stevens JB, et al. 2009. Genetic and epigenetic heterogeneity in cancer: a genome-centric perspective. J Cell Physiol 220:538–547.

Heng HHQ, Liu G, Stevens JB, et al. 2011. Decoding the genome beyond sequencing: the new phase of genomic research. Genomics 98:242–252.

Hewitt GM. 1979. Animal cytogenetics: Insecta 1: Orthoptera: grasshoppers and crickets. Berlin: Gebrüder Borntraeger.

Holmes GD, James EA, Hoffmann AA. 2009. Divergent levels of genetic variation and ploidy among populations of the rare shrub, *Grevillea repens* (Proteaceae). Conserv Genet 10:827–837.

Jiang JM, Birchler JA, Parrott WA, Dawe RK. 2003. A molecular view of plant centromeres. Trends Plant Sci 8:570–575.

John B, Miklos GLG. 1979. Functional aspects of satellite DNA and heterochromatin. Int Rev Cytol 58:1–114.

Johnson MAT, Kenton AY, Bennett MD, Brandham PE. 1989. *Voanioala gerardii* has the highest known chromosome number in the monocotyledons. Genome 32:328–333.

Jones K. 1998. Robertsonian fusion and centric fission in karyotype evolution of higher plants. Bot Rev 64:273–289.

Kidwell MG, Lisch DR. 2001. Transposable elements, parasitic DNA, and genome evolution. Evolution 55:1–24.

King M. 1990. Animal cytogenetics: Chordata 2: Amphibia. Stuttgart: Gebrüder Borntraeger.

Kolnicki RL. 1999. Karyotypic fission theory applied: kinetochore reproduction and lemur evolution. Symbiosis 26:123–141.

Kolnicki RL. 2000. Kinetochore reproduction in animal evolution: cell biological explanation of karyotypic fission theory. Proc Natl Acad Sci USA 97:9493–9497.

Kreitman M. 1983. Nucleotide polymorphism at the alcohol dehydrogenase locus of *Drosophila melanogaster*. Nature 304: 412–417.

Kullman B, Tamm H, Kullman K. 2005. Fungal genome size database. Available online at: http://www.zbi.ee/fungal-genomesize/ [accessed 9 April 2013]

Leakey LSB, Tobias PV, Napier JR. 1964. A new species of the genus *Homo* from Olduvai Gorge. Nature 202:7–9.

Levin DA. 1975. Minority cytotyoe exclusion in local plant populations. Taxon 24:35–43.

Levin DA. 2002. The role of chromosomal change in plant evolution. Oxford: Oxford University Press.

Löve A, Löve D, Pichi-Sermolli REG. 1977. Cytotaxonomical atlas of the Pteridophyta. Vaduz, Liechtenstein: J. Cramer.

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

Lynch M. 2007a. The frailty of adaptive hypotheses for the origins of organismal complexity. Proc Natl Acad Sci USA 104:8597–8604.

Lynch M. 2007b. The origins of genome architecture. Sunderland: Sinauer.

Ma XF, Gustafson JP. 2005. Genome evolution of allopolyploids: a process of cytological and genetic diploidization. Cytogenet Genome Res 109:236–249.

Maherali H, Walden AE, Husband BC. 2009. Genome duplication and the evolution of physiological responses to water stress. New Phytol 184:721–731.

Margulis L, Sagan D. 2002. Acquiring genomes: a theory of the origin of species. New York: Basic Books.

Matuszewski B. 1982. Animal cytogenetics: Insecta 3: Diptera I: Cecidomyiidae. Berlin: Gebrüder Borntraeger.

Mello-Sampayo T. 1961. Differential polyteny and karyotype evolution in *Luzula*: a critical interpretation of morphological and cytophotometric data. Genet Iberica 13:1–22.

Mullis KB. 1990. The unusual origin of the polymerase chain reaction. Sci Am 262:56–61; 64–65.

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

Nordenskiöld H. 1951. Cyto-taxonomical studies in the genus *Luzula*. I. Somatic chromosomes and chromosome numbers. Hereditas 37:325–355.

Ohno S. 1974. Animal cytogenetics: Chordata I: Protochordata, Cyclostomata and Pisces. Stuttgart: Gebrüder Borntraeger.

Olsen MW. 1966. Segregation and replication of chromsomes in turkey parthenogenesis. Nature 212:435–436.

Olson K, Gorelick R. 2011. Chromosomal fission accounts for small-scale radiations in *Zamia* (Zamiaceae; Cycadales). Bot J Linn Soc 165:168–185.

Oswald BP, Nuismer SL. 2011. Neoploidy and diversification in *Heuchera grossulariifoloia*. Evolution 65:1667–1679.

Ouarda HE, Walker DJ, Khouja ML, Correal E. 2009. Diversity analysis of *Acacia tortilis* (Forsk.) Hayne ssp *raddiana* (Savi) Brenan (Mimosaceae) using phenotypic traits, chromosome counting and DNA content approaches. Genet Resour Crop Evol 56:1001–1010.

Ozkan H, Feldman M. 2009. Rapid cytological diploidization in newly formed allopolyploids of the wheat (*Aegilops triticum*) group. Genome 52:926–934.

Patton JL. 1974. Animal cytogenetics: Chordata 4: Mammalia II: Placentalia: 1. Rodentia. Stuttgart: Gebrüder Borntraeger.

Perry J, Slater HR, Choo KHA. 2004. Centric fission—simple and complex mechanisms. Chromosome Res 12:627–640.

Piegu B, Guyot R, Picault N, et al. 2006. Doubling genome size without polyploidization: dynamics of retrotransposition-driven genomic expansions in *Oryza australiensis*, a wild relative of rice. Genome Res 16:1262–1269.

Ramsey J, Schemske DW. 1998. Pathways, mechanisms, and rates of polyploid formation in flowering plants. Annu Rev Ecol Syst 29:467–501.

Rieseberg LH. 2001. Chromosomal rearrangements and speciation. Trends Ecol Evol 16:351–358.

Rieseberg LH, Willis JH. 2007. Plant speciation. Science 317:910–914.

Röser M, Johnson MAT, Hanson L. 1997. Nuclear DNA amounts in palms (Arecaceae). Bot Acta 110:79–89.

Salmon A, Ainouche ML, Wendel JF. 2005. Genetic and epigenetic consequences of recent hybridization and polyploidy in *Spartina* (Poaceae). Mol Ecol 14:1163–1175.

Sarkar SK. 1970. Palmae. Res Bull Cytogen Lab (Dept Bot, Univ. Calcutta).

Schluter D. 2000. The ecology of adaptive radiation. Oxford: Oxford University Press.

Segraves KA, Thompson JN, Soltis PS, Soltis DE. 1999. Multiple origins of polyploidy and the geographic structure of *Heuchera grossulariifolia*. Mol Ecol 8:253–262.

Sémon M, Wolfe KH. 2007. Rearrangement rate following the whole-genome duplication in teleosts. Mol Biol Evol 24:860–867.

Slotte T, Huang HR, Lascoux M, Ceplitis A. 2008. Polyploid speciation did not confer instant reproductive isolation in *Capsella* (Brassicaceae). Mol Biol Evol 25:1472–1481.

Smith SG, Virkki N. 1978. Animal cytogenetics: Insecta 5: Coleoptera. Berlin: Gebrüder Borntraeger.

Sobel JM, Chen GF, Watt LR, Schemske DW. 2010. The biology of speciation. Evolution 64:295–315.

Soltis DE, Soltis PS. 1987. Polyploidy and breeding systems in homosporous Pteridophyta: a reevaluation. Am Nat 130:219–232.

Soltis DE, Soltis PS. 1993. Molecular data and the dynamic nature of polyploidy. Crit Rev Plant Sci 12:243–273.

Soltis DE, Albert VA, Leebens-Mack J, et al. 2009. Polyploidy and angiosperm diversification. Am J Bot 96:336–348.

Stebbins GL. 1971. Chromosomal evolution in higher plants. London: Edward Arnold.

Thompson JN, Nuismer SL, Merg K. 2004. Plant polyploidy and the evolutionary ecology of plant/animal interactions. Biol J Linn Soc 82:511–519.

Tobias PV. 1953. Trends in the evolution of mammalian chromosomes. S Afr J Sci 50:134–140.

Tobias PV. 1992. Piltdown: an appraisal of the case against Sir Arthur Keith. Curr Anthrol 33:243–294.

Todd NB. 1970. Karyotypic fissioning and canid phylogeny. J Theor Biol 26:445–480.

Ueshima N. 1979. Animal cytogenetics: Insecta 6: Hemipetera II: Heteroptera. Berlin: Gebrüder Borntraeger.

Uhl CH. 1992. Polyploidy, dysploidy, and chromosome pairing in *Echeveria* (Crassulaceae) and its hybrids. Am J Bot 79:556–566.

Uhl CH. 2007. Chromosomes and hybrids of *Echeveria*. XI. South American species of Series Racemosae. Haseltonia 13:3–22.

Vandel A. 1928. La parthenogenese geographique: contribution a l'etude biologique et cytologique de la parthenogenese naturelle. Bull Biol France Belg 62:164–281.

Vandel A. 1937. Chromosome number, polyploidy, and sex in the animal kingdom. Proc Zool Soc Lond A 107:519–541.

Vanichanon A, Blake NK, Sherman JD, Talbert LE. 2003. Multiple origins of allopolyploid *Aegilops triuncialis*. Theor Appl Genet 106:804–810.

Vyas P, Bisht MS, Miyazawa S, et al. 2007. Effects of polyploidy on photosynthetic properties and anatomy in leaves of *Phlox drummondii*. Funct Plant Biol 34:673–682.

Watson JD, Crick FHC. 1953. Molecular structure of nucleic acids. Nature 171:737–738.

White MJD, Webb GC. 1976. Animal cytogenetics: Insecta 2: Blattodea, Mantodea, Isoptera, Grylloblattodea, Phasmatodea, Dermaptera and Embioptera. Berlin: Gebrüder Borntraeger.

Williams GC. 1966. Adaptation and natural selection: a critique of some current evolutionary thought. Princeton: Princeton University Press.

Wilson EB. 1896. The cell in development and inheritance. 1st edition. New York: Macmillan.