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Chimeric aneuploids and their role in the evolution of early generation synthesized Arabidopsis allopolyploids

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Chimeric aneuploids and their role in the evolution of early generation synthesized *Arabidopsis* allopolyploids

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ABSTRACT

More than 90% of extant angiosperms show evidence of polyploidization (whole genome duplication) events in their evolutionary history. Polyploidization leads to stochastic disturbances in genomic structure, gene regulation, and chromosome maintenance and, thus, introduces diversity into a population of neopolyploids and promotes evolutionary change. To understand the basic mechanisms of polyploid-induced variation, we investigated chromosomal changes in sibling lines of synthetically formed *Arabidopsis* allopolyploids. Centromeric fluorescence *in-situ* hybridization (FISH) probes were then applied to chromosome spreads to track cases of aneuploidy (loss/gain of chromosomes).

We hypothesized that variations in the degree of aneuploidy between sibling lines of neoallopolyploids could produce enough genetic diversity to induce speciation. Our data indicate that allopolyploidization has lead to rapid karyotypic changes, phenotypic variations, and variable viability between the sibling lines.

INTRODUCTION

Biodiversity has become an increasingly important field of study, as it is key to the maintenance and survival of any population, ecosystem, or biosphere. Genetic variability within a population can lead to evolutionary change and is vital to the future of all biological systems (Groom et al., 2006). It is one thing to know that variation exists, but another to understand its scientific origins; therefore, it is in our interest to study the mechanisms that lead to genetic and thus potential evolutionary diversity. To that end, our lab uses allopolyploids of *Arabidopsis* to study molecular, genetic, and cytogenetic mechanisms that lead to variation within an allopolyploid population to better understand the evolutionary forces behind polyploid induced speciation.

OBJECTIVES

- Compare **chromosome composition** between 6 sibling lines of a newly formed population of allohexaploid (derived from a cross between 2x *A. thaliana* X 4x *A. suecica*, which itself is an allotetraploid of 2x *A. thaliana* and 2x *A. arenosa*) to determine the karyotypic divergence in the population.
- Correlate **abnormal phenotypes** (flower size, leaf morphology, flowering time, etc.) with levels of aneuploidy in each plant.
- Compare **fertility levels** between siblings and parental species.

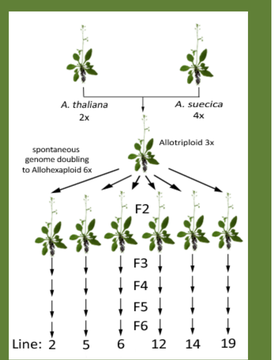


Figure 1. Pedigree of the *Arabidopsis* allohexaploid.

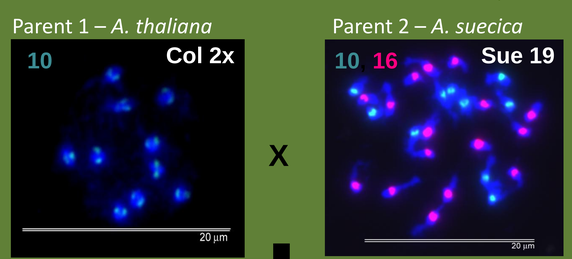


Figure 2. Chromosome composition of the *Arabidopsis* allohexaploid parents: *A. thaliana* (10 AT chromosomes) and *A. suecica* (10 AT chromosomes, 16 AA chromosomes).

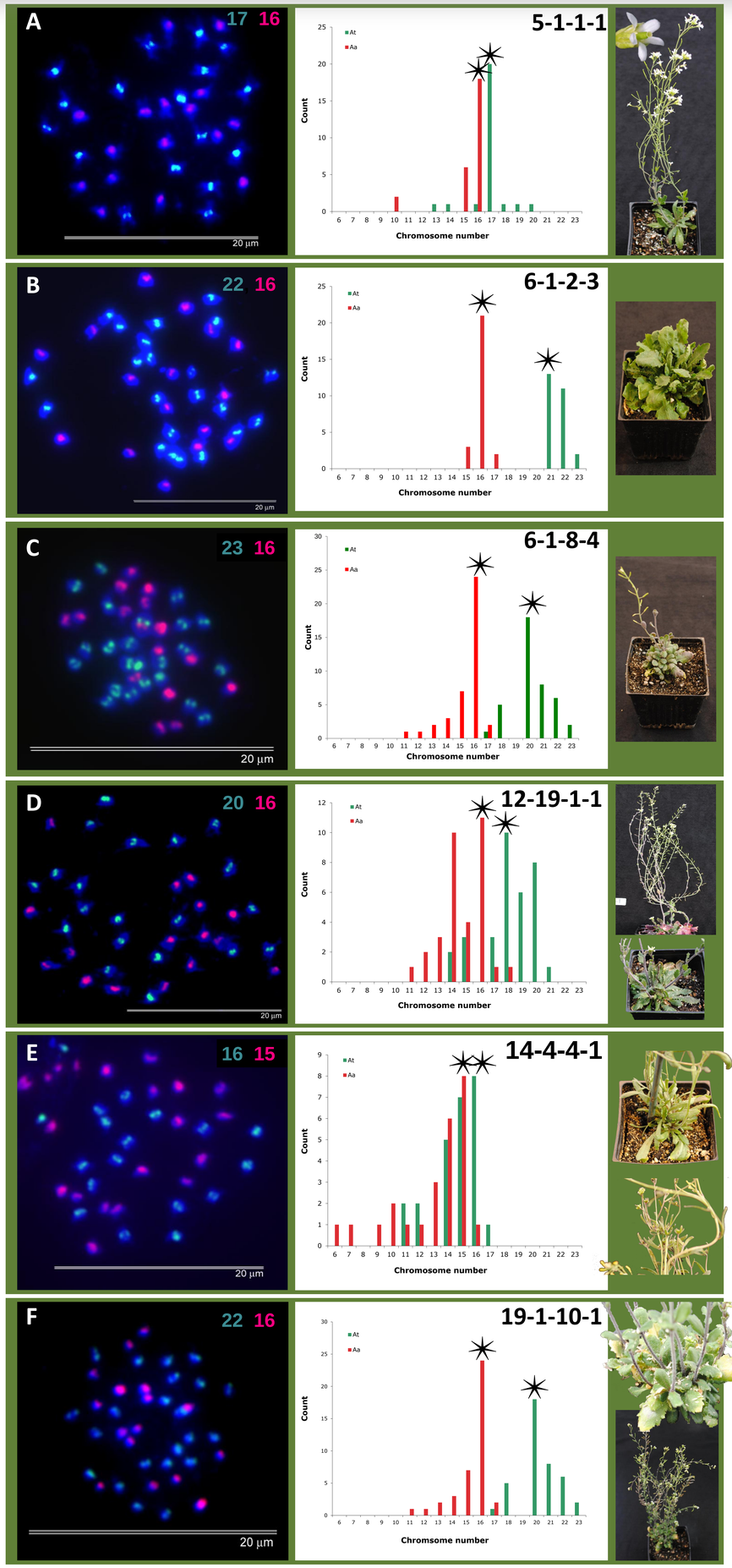


Figure 3. Allohexaploids from the F6 and F7 generations showing varying degrees of chimeric aneuploidy/euploidy and abnormal phenotypes. Chromosomes were labeled with species-specific centromeric FISH probes. Corresponding graphs to the right of each FISH picture show the frequency of each chromosome number per cell (✶ represents the modal At and Aa chromosome number). (**A**) F6 5-1-1-1 aneuploid cell with 17 *A. thaliana* (At) and 16 *A. arenosa* (Aa) chromosomes. The modal chromosome number of this plant was 17 and 16, At and Aa chromosomes, respectively (N=50). (**B**) F7 6-1-2-3 supernumerary aneuploid cell with 22 At and 16 Aa chromosomes. Mode: 21 At and 16 Aa chromosomes (N=21). (**C**) F7 6-1-8-4 with 23 At and 16 Aa chromosomes. Mode: 20 At and 16 Aa (N=28). (**D**) F7 12-19-1-1 cell with 20 At and 16 Aa. Mode: 18 At and 16 Aa (N=33). (**E**) F7 14-4-4-1 aneuploid cell with 16 At and 15 Aa, representing the modal chromosome number (N=25). (**F**) 19-1-10-1 cell with 22 and 16 At and Aa chromosomes. Mode: 20 At and 16 Aa chromosomes (N=40).

MATERIALS and METHODS

Fluorescence *in-situ* hybridization (FISH):

A. thaliana centromeric repeats (180bp) were labeled with Fluorescein-dUTP (GREEN), *A. arenosa* centromeric repeats (200bp) were labeled with Texas Red-dCTP (RED). The labeled cells of many individuals of the six sibling lines were analyzed using fluorescent microscopy at F3, F6 and F7 generations (**10-50 cells from each plant**).

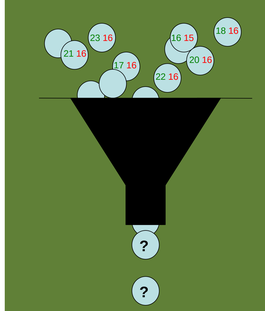
Morphological assays:

Pollen viability and morphological parameters were measured for each individual.

RESULTS

- F3 gen. siblings, although healthy, were genomically unstable (>96% deviation from expected chromosome number).
- Pollen of the F7 gen. was considerably less viable than either parent species. Seed production also decreased.
- The degree of chromosomal loss does not seem to correlate directly with the degree of pollen inviability.
- Aneuploidy can arise both from loss or gain of either parent species' chromosomes, but *A. arenosa* chromosomes were inherited more faithfully than *A. thaliana* (Figure 3).

CONCLUSION



Although distinct chromosomal and phenotypic differences between siblings indicate the possibility of emerging speciation, our data suggest that only some of the lines may retain a chromosomal composition that is consistent with long-term survival through the polyploidization bottleneck and eventual speciation.

Figure 4. The polyploidization bottleneck. Varying chromosome numbers may lead to changes in phenotype between siblings. However, long term survival and speciation are limited by the degree of chromosomal variation and environmental factors (the bottleneck).

CITATIONS

Groom, M.J., G.K. Meffe, and R.C. Carroll, and contributing authors. 2006. *Principles of Conservation Biology*, 3rd ed. Sinauer associates, Inc., MA, pp. 30.

ACKNOWLEDGMENTS

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