

## BIVALENT PAIRING IN AN INDUCED TETRAPLOID OF TRADESCANTIA

GEORGE W. SKIRM

*Arnold Arboretum, Jamaica Plain, Massachusetts*

Received June 25, 1942

THE tetraploid forms of any genus are usually separated into two groups, auto- and allopolyploid, depending on the behavior of the chromosomes at meiosis. The meiotic behavior is also used as an indication of phylogenetic history. Those forms that produce bivalents at meiosis I usually are assumed to be the result of interspecific hybridization followed by chromosome doubling, while those forms that exhibit multiple configurations at meiosis I are assumed to be intraspecific hybrids followed by chromosome doubling. DARLINGTON (1937) proposes an alternative to this by postulating that an autotetraploid may show bivalent pairing if enough time has elapsed to allow the accumulation of internal rearrangements so that there no longer remains the necessary attraction between the four homologues to result in quadrivalent formation. The induced tetraploids in *Tradescantia* show that other factors—namely, the time of chromosome doubling and the degree of structural heterozygosity in the parents—may also influence the behavior of the chromosomes of the induced tetraploid at the first meiotic division.

An attempt was made to induce polyploidy in the genus *Tradescantia* by means of thermal shock, following the methods of RANDOLPH (1932). The equipment used for this work has been described by SAX (1937). The experiment was designed to produce a controlled polyploid and to compare this with the known parents. The parent plants were two diploid clonal lines of natural hybrids between *Tradescantia canaliculata* and *T. humilis* originally collected by DR. EDGAR ANDERSON at Austin, Texas, and propagated under his accession numbers of Oak Hill 5 and Oak Hill 13. These plants are completely self-sterile and set no open-pollinated seed under greenhouse conditions. When these plants are hand cross-pollinated, each capsule contains an average of 3.5 seed.

SAX found that sudden changes in temperature were as effective as extremes of temperature for the production of polyploid tissue; thus the parent plants were preconditioned at 39–40°F for a few hours immediately following inter-pollination and then were quickly shifted to the temperature of 95–97°F for varying lengths of time. The treatments were repeated daily until the first pollinated flowers had begun to mature the hybrid seed. The time elapsing between pollination and the administering of the thermal shock was varied considerably, in an attempt to induce chromosome doubling at the first division of the hybrid zygote. The ar-

range of the flower clusters is such that the treatments given developing zygotes would also effect developing gametes at the two meiotic divisions. At maturity the seeds were collected and bulked. Germination was poor, and less than fifty percent of the seeds produced plants. The plants were grown to maturity under field conditions. The chromosome number of each plant was determined from the haploid microspore division using the acetocarmine technic. Many of the plants died or did not bloom, but among the 338 plants which flowered 311 were diploids, 18 were triploids, six were tetraploids, and three were aneuploids with either 17 or 19 chromosomes.

Oak Hill 5 and 13 are normal diploids with good cytological behavior. Each of the parents possesses a single centric fragment, which appears to be inherited through the female side and which is slightly deleterious to the rate of development of the pollen tube. There were very few segregates containing more than one fragment. The parents also possess a series of genes for chlorophyll deficiency, which appear to be complementary rather than allelic. There were various degrees of chlorophyll deficiency, varying from true lethal albinos to barely perceptible virescence. Associated with the chlorophyll deficiency is a less vigorous growth rate. There was no association between the chlorophyll deficiency and the presence or absence of fragments which would indicate that the chlorophyll deficient genes were borne on the major chromosomes. Occasionally unbalanced gametes functioned as indicated by the aneuploid types produced. The parents also possessed a number of genes effecting sterility, because the sterility of the progeny varied from 5 to 100 percent, and most of it could not be accounted for by chromosomal irregularities.

A random sample of the diploid segregates was examined for meiotic behavior. Chromosome pairing was normal, with only occasional univalents and inversion bridges which are characteristic for most species of *Tradescantia*. The triploid segregates showed the typical trivalent, bivalent, and univalent distribution found in spontaneous triploids. Five of the six tetraploid plants were similar to normal tetraploid species in their meiotic behavior. From 50 to 60 percent of the chromosomes were paired as quadrivalents, with the remaining chromosomes forming trivalents, bivalents, and univalents (*cf.* ANDERSON and SAX 1934).

One of the tetraploids was distinctly different from the other tetraploid segregates. In this plant, designated as #91, most of the chromosomes were paired as bivalents. Only 5.6 percent of the chromosomes were associated as quadrivalents, and the occasional univalent—2.5 percent—were distributed regularly, suggesting precocious terminalization of pre-existing chiasmata. The number of chiasmata per chromosome was 0.9. The frequency of inversion bridges was 3.7 percent. Pollen sterility was relatively

low—8.4 percent. A comparison of the parents and the tetraploid is shown in table 1.

The cytological behavior of the induced tetraploids indicates two types of origin. Since most *Tradescantias* are heterozygous in respect to chromosome structure (SWANSON 1940), the production of a tetraploid by meiotic irregularities resulting in diploid gametes would produce a plant in which no two of the four genomes are identical. The lack of preferential pairing would favor quadrivalent formation. In plant 91, however, the meiotic behavior suggests that chromosome doubling occurred at the time of fertilization or during embryonic development. Somatic doubling in a plant known to be heterozygous in chromosome structure would result in two sets of duplicated genomes. At meiosis there would be preferential pairing of duplicated genomes, and only occasionally would chromosomes of unlike genomes pair to form quadrivalents or pair as bivalents and pro-

TABLE 1  
*Comparison of induced tetraploid #91 with its diploid parents.*

	OAK HILL 5	OAK HILL 13	#91
Diameter of PMC at metaphase I	29.5 $\mu$	30.8 $\mu$	44.6 $\mu$
Width of mature pollen	33.7 $\mu$	34.0 $\mu$	43.3 $\mu$
Resting nucleus measured in long axis of root cell	17.4 $\mu$	15.3 $\mu$	25.2 $\mu$
Length of stomata guard cell	48.8 $\mu$	56.4 $\mu$	70.3 $\mu$
# stomata/mm <sup>2</sup> of leaf	92.3	110.3	60.6
# xta. per cell	10.7	10.8	21.4
# xta. per chromosome	0.89	0.90	0.89
% inversion bridges at anaphase I	5.8%	6.5%	3.7%
% pollen sterility (abnormal)	21.0%	17.0%	8.4%

duce inversion bridges. Most of the bivalents would consist of identical chromosomes.

Since the temperature shock was given at various times during floral development, the induced polyploids could be derived from chromosome doubling in the gametes or from chromosome duplication in the fertilized egg or during embryonic development. The production of diploid gametes could result either from premeiotic polyploidy or from meiotic irregularities. The triploid segregates must have been derived from diploid gametes produced by one of the parental plants. The origin of the tetraploid segregates is indicated by their meiotic behavior.

The interpretation of bivalent formation in an autopolyploid is supported by the evidence from other sources. Thus in the diploid form of *Primula floribunda*  $\times$  *P. verticellata* the chromosomes pair as bivalents, but in the somatically derived tetraploid the chromosomes still pair as bivalents. This pairing is primarily between chromosomes of similar

genomes with only a slight tendency for cross pairing or the formation of quadrivalents (DARLINGTON 1937). Preferential pairing also may occur within a species. In the triploid *Drosophila* and *Rumex* (DARLINGTON 1937) and in the tetraploid *Melandrium* (WARMKE and BLAKESLEE 1940) the X chromosomes show preferential pairing, although the X and the Y chromosomes pair regularly in the normal diploid forms. Preferential pairing also occurs when the opposing chromosomes come from the same species as has been demonstrated by many workers. Chromosomes tend to pair more frequently with their duplicated identical partners than with the partially homologous partners with which they previously paired. In the presence of identical chromosomes, the latent attraction between the semi-homologous chromosomes is neglected and only bivalents are formed as a rule. Among the species with a higher chromosome number there remains, however, a minor tendency for the residual attraction to be expressed, and this results in the formation of an occasional multivalent (*cf.* DARLINGTON on polyploid species of an intermediate type; and KATTERMAN (1931) on *Bromus erectus*).

In one species, *Allium Schoenoprasum*, LEVAN (1935) has shown that two distinct cytological types occur. One is a giant autotetraploid by definition and the other is a smaller allotetraploid. This has been interpreted by DARLINGTON as an example of the change of an allotetraploid into an autotetraploid by the progressive elimination of all of those characters by which one parent of the allotetraploid was distinguished from the other parent; or perhaps the converse has occurred and the species arose as an autotetraploid with free pairing among the homologues resulting in quadrivalents. The latter condition still remains in some of the forms. The others have changed in several ways and have lost their gigantism as well as the autopolyploid pairing due to cytological differentiation arising between the different pairs of the haploid sets. A method by which this latter type of change could come about without any genetic change that is physiologically expressed is by inversions coming into the chromosomes.

If the history of Plant 91 were not known definitely, there would be a tendency for one to classify this type of polyploid as an allotetraploid, because chromosome pairing is predominantly in the form of bivalents. In a somatically double plant there is a certain amount of cross pairing between non-duplicated chromosomes and a low percentage of quadrivalent associations. The amount of cross pairing is a function of the frequency and size of the interstitial inversions in the original diploid form. Crossing over in the non-duplicated chromosomes and their subsequent segregation as chromatids would result in the formation of non-identical gametes. Irregular disjunction of the quadrivalents formed, while not seen in #91, might possibly lead to an upset of the condition necessary for bivalent formation

in subsequent generations. Species exhibiting this type of behavior would closely approximate DARLINGTON's definition of polyploid species of an intermediate type (*cf.* DARLINGTON 1937, table 30).

The duplicated type of chromosome behavior demonstrated by #91 is not found among the wild populations of *Tradescantia*. It cannot exist except by asexual propagation in a self-sterile species. In a natural colony the chances of two plants of an approximately similar cytological makeup simultaneously doubling their chromosomes in the somatic tissue to result in the formation of bivalents and then intercrossing is extremely remote. If crosses between a somatically doubled and a gametically doubled tetraploid form occurred, the resulting progeny, while tetraploid, would not have the necessary identical chromosomes to give the type of pairing found in #91. If only one plant were produced in a colony of diploids, the resulting progeny would be triploids. In either case the precise condition required for the type of chromosome association found in #91 would be lost irretrievably. This limitation may be removed in certain species where the induced tetraploid condition restores self-fertility (STOUT and CHANDLER 1941). Allo-syndesis in an autopolyploid also could occur in a structurally heterozygous species in which the plants are self-fertile although normally cross-pollinated.

The origin of an autopolyploid *Tradescantia* which behaves as an allopolyploid cytologically may explain in part the heretofore anomalous cytological behavior of those species which possess polyploid forms but show no evidence of polyspecific ancestry (MUNTZING 1936).

#### SUMMARY

Two types of tetraploids were derived from a diploid plant by temperature changes during gametic and embryonic development. Five of the tetraploids behaved as autopolyploids with somewhat more than half of the chromosomes pairing as quadrivalents. These presumably were derived from the union of diploid gametes. One of the induced tetraploids behaved as an allopolyploid with chromosomes pairing primarily as bivalents. The origin of this plant is attributed to chromosome doubling following fertilization. The difference in chromosome behavior is attributed to structural heterozygosity in the parental genomes.

#### LITERATURE CITED

- ANDERSON, E., and K. SAX, 1934 A cytological monograph of the American species of *Tradescantia*. *Bot. Gaz.* 97: 433-475.  
DARLINGTON, C. D., 1937 Recent advances in cytology. 2nd Edition. Philadelphia: Blakiston's.  
KATTERMAN, G., 1931 Über die Bildung polyvalentei Chromosomenverbannde bei einigen Gramineen. *Planta* 12: 732-744.  
LEVAN, A., 1935 Zytologische Studien in *Allium Schoenoprasum*. *Hereditas* 22: 1-128.

- MÜNTZING, A., 1936 The evolutionary significance of autopolyploidy. *Hereditas* **21**: 263-378.
- RANDOLPH, L. F., 1932 Some effects of high temperature on polyploidy and other variations in Maize. *Proc. Nat. Acad. Sci.* **18**: 222-229.
- SAX, K., 1937 Effect of variations in temperature on nuclear and cell division in *Tradescantia*. *Amer. J. Bot.* **24**: 218-225.
- STOUT, A. B., and C. CHANDLER, 1941 Change from self-incompatibility to self-compatibility accompanying change from diploidy to tetraploidy. *Science* **94**: 118.
- SWANSON, C. P., 1940 The distribution of inversions in *Tradescantia*. *Genetics* **25**: 438-465.
- WARMKE, H. E., and A. F. BLAKESLEE, 1940 The establishment of a 4n dioecious race in *Melandrium*. *Amer. J. Bot.* **37**: 751-762.