A Natural Chimera of Douglas-fir[1])

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In gymnosperms, the number of chromosomes is usually constant for most genera. In the genus Pinus for example, the haploid number (n) of chromosomes is 12, and the somatic number 2n = 24. In the genus Pseudotsuga, however, observations of three species showed differences in the number of chromosomes in the haploid sets. Douglas-fir (Pseudotsuga *menziesii* [MIRB.] FRANCO) has 13, but bigcone Douglas-fir (P. macrocarpa MAYR) and Formosan Douglas-fir (P. wilsoniana HAYATA) both have 12 chromosomes (CHRISTIANSEN 1963, THOMAS and CHING 1968). Reports of natural aneuploidy are rare in coniferous species. The occurrence of an abnormal slash pine (Pinus elliottii ENGELM.) seedling with several aneuploid cells with chromosome counts of 54 was reported by MERGEN (1958) and an aneuploid Norway spruce (Picea abies [L.] KARST.) with 2n = ±28 chromosomes was found by KIELLANDER (1950). The existence of chromosomal aberrations in Douglas fir was also revealed recently by OWENS (1967). This note reports the discovery of one sectional chimera of Douglas-fir, with separate parts of the plant having diploid (2n) and trisomic (2n + 1) tissue.

Materials and Methods

When the 2-0 mixoploid Douglas-fir was first observed in the nursery, it was only 12.3 cm in height, about one-half the height of normal Douglas-fir seedlings. A few normal-appearing needles were attached to the lower portion of the stem, but the rest of the crown had 14 branches bearing short, thick, abnormal needles. In its fourth growing season, the plant doubled in height and produced 31 branches. Only the three nearest the base had normal needles (Figure 1).

Fig. 1. — The Douglas-fir chimera in 1959, showing a normal branch growing from the base, and the rest of the plant with aberrant needles.

The cytological preparations were made from vegetative buds. The buds were placed in a saturated aqueous solution of monobromo naphthalene (O'MARA 1948) for 4 hours to shorten the chromosomes, and then killed and fixed in FARMER'S solution (100% alcohol-acetic acid, 3 : 1) for 24 hours. The buds were then hydrolyzed in 1.0 N hydrochloric acid, Feulgen stained, and squashed in a drop of acetocarmine (THOMAS and CHING 1968) and a drop of Hoyer's mounting medium (BEEKS 1955). Observations and photographs were made with a phase-contrast microscope.

Results

Twenty-seven chromosomes were counted in each of 10 cells from vegetative buds of the trisomic branches. Chromosomes of these cells differ from the normal specimens of Douglas-fir (BARNER and CHRISTIANSEN 1962, THOMAS and CHING 1967) in having five metacentric pairs, seven submetacentric pairs, one pair of telocentric chromosomes, and one extra-short metacentric chromosome. In contrast, the normal Douglas-fir has five metacentric pairs, six submetacentric pairs, and two pairs of telocentric chromosomes.

Although bud tissues taken from the normal-appearing shoots of the chimeric plant showed cells with 26 chromosomes, the karyotype was also different from the normal Douglas-fir in having one additional pair of submetacen-

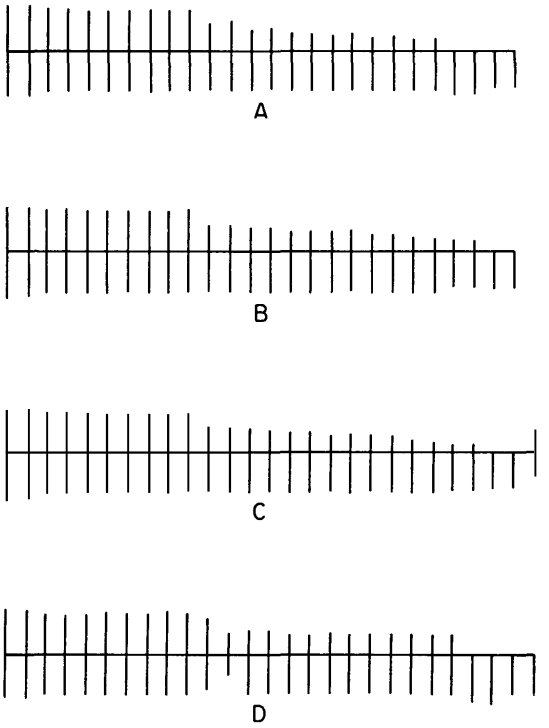

Fig. 2. — Idiogams of a normal and three aberrant Douglas-firs: A, as reported by THOMAS and CHING (1967); B, from a normal branch of the chimeric seedling; C, from an aberrant branch of the chimera; and, D, as described by OWENS (1967).

[1]) This study was supported in part by a grant from the Bureau of Land Management, U.S. Department of the Interior. The authors are, respectively, Professor of Forest Genetics and Research Assistant, School of Forestry, Oregon State University. — Paper 682, School of Forestry, Oregon State University, Corvallis, Oregon 97331.

Table 1. — Comparison of the relative unit lengths of chromosomes for normal and aberrant Douglas-fir.

Reference	Chromosome number in haploid set													
	1	2	3	4	5	6	7	8	9	10	11	12	13	—
Normal Douglas-fir (*P. menziesii* var. *menziesii*)	144	136	128	128	120	99	92	87	83	80	77	69	57	
Chimeric Douglas-fir														
Normal-looking branch	144	136	130	126	120	98	92	89	84	79	75	70	55	extra
Aberrant branch	146	135	133	128	127	100	96	92	87	82	76	69	58	69

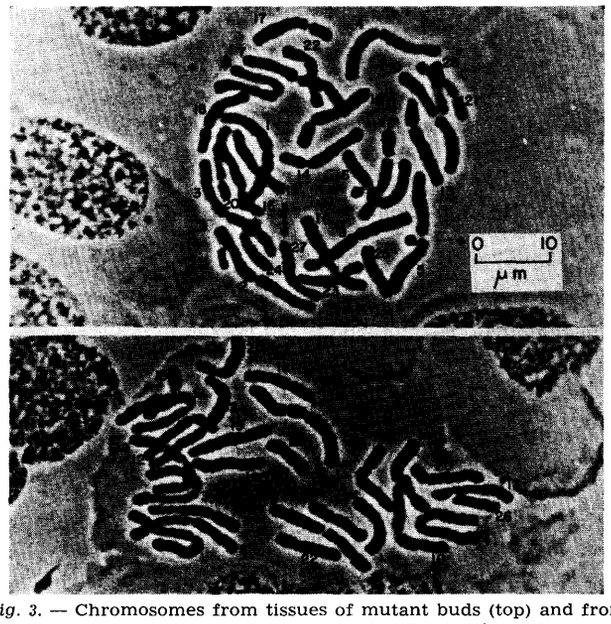

Fig. 3. — Chromosomes from tissues of mutant buds (top) and from a normal-appearing branch of the Douglas-fir chimera. The arrow indicates the extra chromosome.

trics and one fewer pair of telocentrics. *Table 1* and *Figure 2* compare the relative lengths of chromosomes of the haploid set and the diploid idiogram of the chimera with that reported by several investigators for normal and aberrant Douglas-fir chromosomes. *Figure 3* contains photomicrographs of chromosomes from bud tissues of the trisomic and the normal-looking branches of the studied plant.

Discussion

Another type of trisomic Douglas-fir was recently reported by Owens (1967). It also has 27 chromosomes (2n + 1), which include 11 long metacentric, 11 submetacentric, 4 short telocentric chromosomes and a single short metacentric chromosome, contrasted to the accepted karyotype of normal Douglas-fir with the 10-12-4 sequence. Owens suggested that a trisomic could be produced either by a misdivision of the centromere in a subtelocentric chromosome or by a reciprocal translocation between two chromosomes with subterminal centromeres, followed by a nondisjunction of the altered chromosomes and fusion with a normal gamete.

In the karyotype of the trisomic tissue of the chimera in our study, besides the extra metacentric chromosome, there were 10 long metacentric, 14 submetacentric, and 2 telocentric chromosomes, which was quite different from that of Owens. Evidently many types of chromosomal aberrations are possible in nature, possibly caused by inversion, nondisjunction, or other phenomena.

This and other chimeras collected in subsequent years by us may possibly be used for development of dwarfing rootstock and bonzai culture. Further studies on mitotic behavior and metabolic changes of these aberrant specimens will undoubtedly shed more light on somatic segregation in long-lived organisms.

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