

iciency, the confidence levels of tests of significance, and confidence ranges for estimates of variables (COCHRAN 1947). When the F statistic or the two-tail t is used to test differences among means, the confidence level is affected very little by deviations of the magnitude usually encountered in experimental data (Box 1953, COCHRAN 1947). True significance levels usually range between 4 and 7 percent for the 5-percent level and between $\frac{1}{2}$ and 2 percent for the 1-percent level (COCHRAN 1947). The one-tail t test can be more strongly affected by deviations from normality. Tests among variances and variance components are sensitive to deviations from normality, and special caution is warranted for these. But for the most commonly used tests, i.e., F and two-tail t tests, the normality assumption is usually satisfied by biological data.

According to COCHRAN (1947), there are two principal effects of heterogeneity of error variation:

- (1) Loss of statistical efficiency, and
- (2) Loss of sensitivity of tests of significance.

The magnitudes of these effects are roughly proportional to the size of the differences between error variances. The most important fact again is that the F test is so insensitive to heterogeneity of error variance that Box (1953) says it is neither necessary nor useful to test statistically for it. He explains that, when plot sizes are about equal, preliminary testing for heterogeneity may lead to more wrong conclusions than if homogeneity is assumed. The most serious distortions of significance levels due to heterogeneity come about throughout use of pooled error terms such as those often used for F tests (COCHRAN 1947). Use of pooled error terms requires extra caution.

In summary, SHIUE and PAULEY (1961) have overemphasized the efficacy of statistical tests for determining the validity of assumptions for the analysis of variance, and, while recognizing the dangers of heterogeneity, they have neglected to emphasize the general robustness of the frequently used F and two-tail t tests.

Summary

Presence of genetic diversity in a population is not sufficient basis for predicting, *a priori*, nonnormality or

heterogeneity in data from progeny and provenance tests with single-tree or multiple-tree plots. Rarely will the environmental component of error variation be so small that the genetic component could cause failure of the assumption of normality in the analysis of variance of such data. Data previously presented and discussed in the literature cannot be unambiguously interpreted to mean that single-tree plots are more likely to yield nonnormal distributions of error variation than are multiple-tree plots.

Confidence levels for the most frequently used tests of significance, F and two-tail t tests of means, are affected very little by deviations from normality and homogeneity of the magnitude usually encountered in experimental data. Therefore, it is neither necessary nor useful to routinely apply statistical procedures for testing normality and homogeneity.

When corrective procedures for nonnormality and heterogeneity of error variation are needed, they can be applied to data from single-tree plots as well as multiple-tree plots. Therefore, no special restrictions are encountered through the use of single-tree plots on this account.

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The Morphology and Behavior of Meiotic Chromosomes of Doriglas-Fir

(*Pseudotsuga menziesii*)

By GORDON K. LIVINGSTON¹

Introduction

Two recognized species of *Pseudotsuga* occur in North America (MUNZ and KECK, 1959). *Pseudotsuga menziesii* (MIRB.) FRANCO is the most widespread, and ranges 3000 miles from Canada to Mexico and extends eastward 1000 miles from the Pacific coast to the Rocky Mountains. *Pseudotsuga macrocarpa* MAYR., on the other hand, is restricted to southern California. Four other species occur in the eastern part of Asia and on the islands of Formosa and Japan. There are seven published accounts of the chromo-

somes of the American species, one of which (THOMAS and CHING, 1968) deals in part with an Asiatic species. With the exception of THOMAS and CHING's study on the somatic chromosomes of Douglas-fir in central Oregon, no studies have dealt with the chromosomal cytology of *Pseudotsuga* growing in its natural range. The author knows of no published report describing the meiotic chromosomes of Douglas-fir in its native habitat and of only one study in Europe, where this species was introduced in the 19th century.

SAX and SAX (1933) first reported the chromosome number of *Pseudotsuga menziesii* as $n = 13$. Their observations were based on haploid, female gametophyte tissue from trees planted at the Arnold Arboretum. However, both

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LANGLET (1943) and DURRIEU-VABRE (1958) reported $2n = 24$, the same as other genera of the *Pinaceae*, which has a basic number of $n = 12$. Their results were based on root-tip meristems of *P. menziesii* in Sweden and France, respectively. The only description of meiosis in Douglas-fir is that of ZENKE (1953), who reported that 13 bivalents appeared consistently at diakinesis and metaphase I. BARNER and CHRISTIANSEN (1962) found the number of somatic chromosomes in *P. menziesii* to be 26, which was in agreement with SAX and SAX, and with ZENKE. Their description of the karyotype, however, differs from that of SAX and SAX. The most recent chromosome count in this species is that of THOMAS and CHING (1968), who counted $2n = 26$ in root tips of *P. menziesii* in central Oregon. Thus, a survey of the literature shows there has not been universal agreement on the chromosome number or the karyotype in this commercially important lumber tree. This fact may reflect differences in cytological technique or natural variation within the species. A firm cytological basis for genetic studies needs to be established for this species in its natural range.

The main objective of this study was to describe microsporogenesis of *Pseudotsuga menziesii* in its natural range. All stages of meiosis from prophase I through microspore formation are illustrated with photomicrographs. The haploid chromosome complement is described, including positions of centromeres and secondary constrictions. A minor objective was to comparatively examine meiotic chromosomes of trees representing the Intermountain race of Douglas-fir.

Materials and Methods

Meiotic chromosomes were studied only in microsporocytes sampled from selected study trees. Male buds were first collected in January of 1963, from an open-grown Douglas-fir tree, *P. menziesii*, several miles north of Corvallis, Oregon. In 1964, three additional trees of this species were sampled on the campus of Oregon State University. In that same year, observations were also made of microsporocytes from *P. macrocarpa* growing on Mt. San Antonio, near Arcadia, California, at an elevation of 6,500 feet. From the Intermountain race of *P. menziesii*, microsporocytes were collected from four trees in the Wasatch Mountains near Provo, Utah, at an elevation of 6,300 feet in March of 1967.

Periodically, male buds were collected, the bud scales removed, and strobili fixed in Carnoy's solution, then stored at 3° C. Material treated in this manner was well preserved after three years. Microsporocytes were removed from the microsporangia and squashed in acetocarmine. The best preparations were made permanent by floating off the cover slip in a solution of one part ethanol and one part acetic acid and replacing it immediately over a mounting medium of Hoyer's solution. Photomicrographs were made with a phase-contrast, Zeiss photomicroscope, using Kodak high-contrast copy film.

Observations

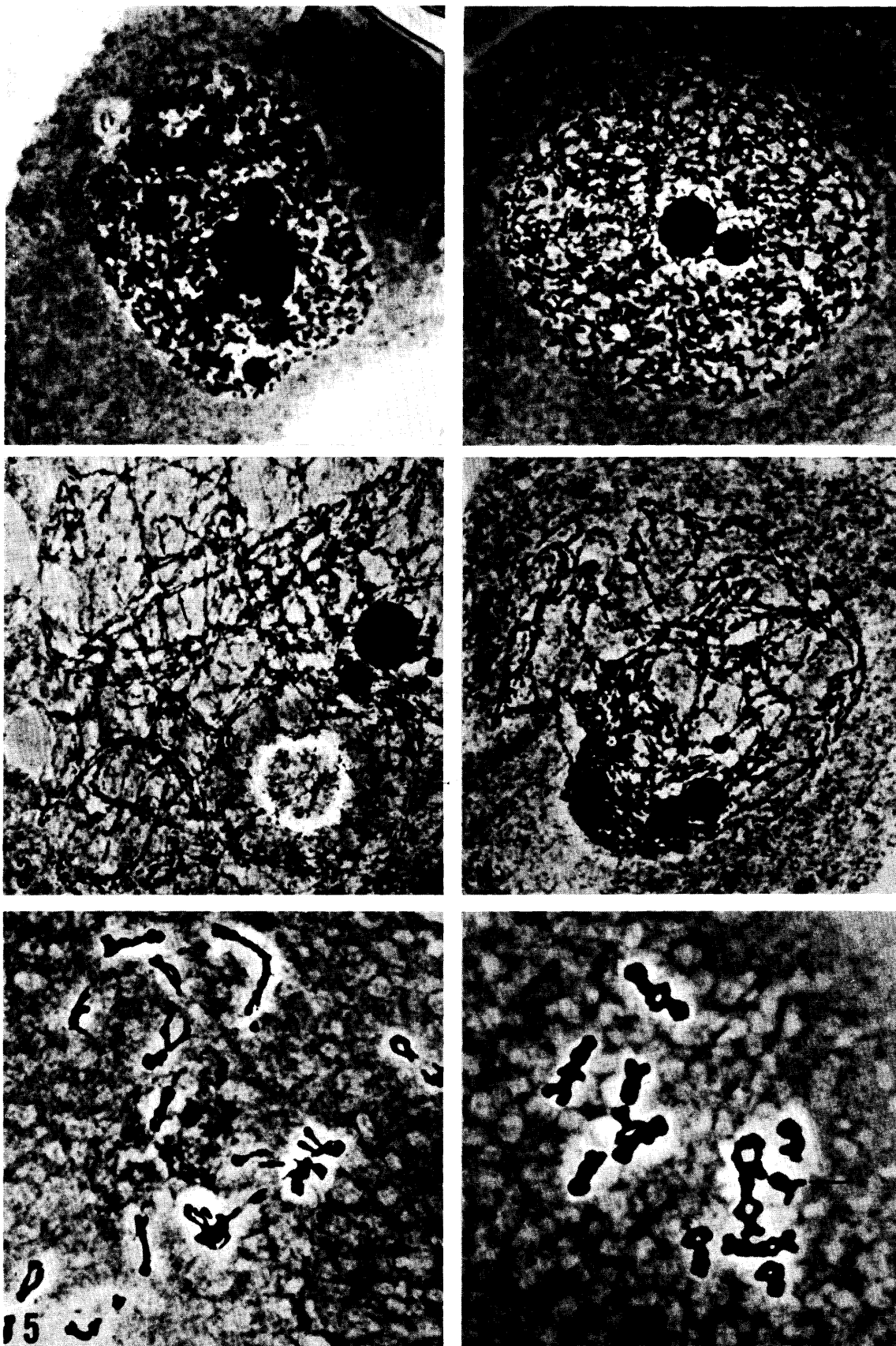
Male buds can be recognized on the terminal parts of branchlets in September or October of the year preceding pollen release. Archisporial cells differentiate in the fall of the year and overwinter as microsporocytes. Samples of microsporocytes were examined with the microscope at frequent intervals, often daily, starting in January of 1963. Meiosis was not visibly initiated until the latter part of

February in microsporocytes collected near Corvallis, Oregon. The division occurred late in January and March in material collected in southern California and Utah, respectively. The process appeared to be largely dependent on temperature, and once nuclear activity was fully initiated, both anaphase I and II were completed within a two- to three-day period on a tree. Following completion of meiosis, the microspores remained inactive for several days before the start of mitosis and the development of the male gametophyte.

The sequence of microsporogenesis in *Pseudotsuga* is exemplary of the regular stages of meiosis. The chromosomes are large and easily stained, and permit close examination of each stage in considerable detail. The premeiotic cell persists in the metabolic condition (Fig. 1) for many weeks prior to the onset of meiosis. At this stage, the carmine stain clearly differentiates the nucleus from the cytoplasm and also reveals a large nucleolus along with one or two smaller nucleolar bodies. The chromatic material in the preleptotene stage appears as granules (Fig. 1) which may be fine, coiled threads uniformly dispersed throughout the nucleus. The initiation of meiosis is marked by an increase in nuclear volume at the same time the leptotene strands begin to straighten and become thicker (Fig. 2). Due to differential uncoiling of the chromonemata, the chromomeres begin to show a linear pattern at leptotene, revealing individual chromosomal strands. The strands are highly attenuated, and at this stage synaptic forces begin to bring homologous chromosomes together, initiating zygotene (Fig. 3). The chromosome strands are now more conspicuous and in several areas the duality (note arrows) of the strands and chromomeres can be seen, showing intimate pairing. When zygotene synapsis is completed the nucleus is at pachytene (Fig. 4). The pachytene strands appear thicker due to contraction and close apposition of homologues; when examined closely at higher magnification, the strand is visibly double and homologous chromomeres are resolved. At this stage, the nucleoli are highly variable in number; usually both large and small ones are in close association with the chromatin. The stages of leptotene through pachytene are illustrated with microsporocytes of big-cone Douglas-fir (*P. macrocarpa*) but corresponding stages of *P. menziesii* are the same.

The chromosomes continue to contract and thicken as the bivalents enter the diplotene stage, which is characterized by the repulsion of homologues. Complete separation does not take place, however, because each bivalent is held together by several chiasmata (Fig. 5). Bivalents at this stage could not be counted, due to the presence of large achromatic regions which caused bivalents to appear diffuse. These nonstaining regions, presumably heterochromatin, are found in nearly all bivalents. Many hundreds of cells were examined for the diplotene stage, which was the most difficult to find, perhaps indicating its short duration.

As further contraction proceeds, the bivalents enter diakinesis (Fig. 6) and the heterochromatic regions are no longer visible. At this stage in meiosis, Douglas-fir exhibits a definitive haploid number of 13. The bivalents show characteristic configurations due to centric loops, which are formed by repulsion of homologous centromeres, while chiasmata continue to hold the homologues together. At diakinesis, three classes of chromosomes can easily be distinguished according to the position of their centromeres. The centromere is located in the median section of five bivalents (metacentric), six bivalents show subterminal



Figs. 1—6. — Meiosis in the microsporocytes of *Pseudotsuga*. — Fig. 1: *P. menziesii*, premeiotic nucleus, $\times 850$. Figs. 2—4: *P. macrocarpa*. Fig. 2: Leptotene, $\times 680$. Fig. 3: Zygotene showing synapsis of homologous strands (note arrows), $\times 530$. Fig. 4: Pachytene, $\times 530$. Figs. 5—6: *P. menziesii*. Fig. 5: Late diplotene, $\times 730$. Fig. 6: Diakinesis (arrow denotes one telocentric), $\times 960$.

centromeres, and two bivalents behave as if their centromeres were terminal (telocentric). One of the telocentric bivalents showing centromere repulsion can be seen in Fig. 6 (arrow) but the other is obscured by another bivalent. Diakinesis and subsequent stages of meiosis confirm the morphology of these three classes of chromosomes as they were first described by BARNER and CHRISTIANSEN (1962).

Once the nuclear membrane and the nucleolus have disappeared, the bivalents move into the equatorial plate region at metaphase I (Fig. 7). The chromosomes are fully contracted, and the centromeres, relative arm lengths, and chiasmata determine the appearance of metaphase I bivalents. Again, three types of chromosomes are clearly distinguished as centromeres begin to lead homologous chromosomes to opposite poles. The arrows in Fig. 7 indicate the two bivalents interpreted as telocentrics, because cytologically they appear to have terminal spindle fiber attachments. As the bivalents disjoin in anaphase I (Fig. 8), the chromatids become visible and the metacentric and subterminal chromosomes are readily seen as double V-shaped structures. The two telocentric chromosomes, however, can be recognized as single V-shaped (see arrows) elements as the chromosomes continue poleward in anaphase I. Infrequently, chromosomes have difficulty in disjoining, for unknown reasons, and a chromosome bridge is formed (Fig. 8). Anaphase bridges appear to be rare, and in those few observed, fragments were not found nor were telocentric chromosomes involved. The chromosomes in late anaphase I (Fig. 9) congregate near the poles and show sister chromatids held in association by their centromeres. Chromosomes elongate as they advance in telophase, presumably by a loosening of the coils; they become more diffuse, then pass into an interphase stage (Fig. 10). The interphase nuclei show chromatin similar in appearance to the premeiotic nuclei, and each contains one or more nucleoli.

Following a brief interphase, the chromatin again contracts in preparation for the second division. Individual chromosomes cannot be followed in early prophase II (Fig. 11), because achromatic regions impart a highly segmented appearance to each chromosome. This effect is typical in both diplotene and early prophase II and may result from the differential staining of heterochromatin and euchromatin. As contraction continues in prophase II, the chromosomes exhibit continuity in stain affinity except for secondary constrictions (Fig. 12). The double nature of prophase II chromosomes is exceptionally clear in Fig. 12, the two chromatids of each chromosome are widely separated except at the centromere. The arrows in Fig. 12 denote areas which are interpreted as three secondary constrictions. These features are not always observed. The secondary constrictions which have been found in various preparations, however, were always located on metacentric chromosomes.

Prior to metaphase II, the nuclear membrane disappears and the chromosomes become oriented with their centric regions lying in the equatorial plate. At metaphase II (Fig. 13) the two chromatids of each chromosome are no longer widely separated but generally appear to lie parallel to each other. It is at this stage, early anaphase II, that the centromere of each chromosome becomes functionally double and the two chromatids disjoin (Fig. 14). Figure 15 is an enlarged view of one of the divisions in an advance stage of anaphase II. At this stage the 13 chromosomes are easily counted, and readily characterized according to the location of their centromeres. A subterminal centromere is observed in six chromosomes, which appear J-shaped.

Median centromeres are seen in five V-shaped chromosomes. Anaphase II, like several previous stages, demonstrates the pair of telocentric chromosomes. At this stage, however, the telocentrics appear as rods (note arrows, Fig. 15), in contrast to their appearance as single V's in anaphase I. As the four groups of chromosomes reach the poles in late anaphase II (Fig. 16) and complete telophase, they become enclosed by a nuclear membrane. They then become attenuated and appear diffuse upon entering the metabolic stage (Fig. 17) as microspore nuclei. Cytokinesis soon follows, separating the four microspores (Fig. 18), each of which then develops into a mature pollen grain.

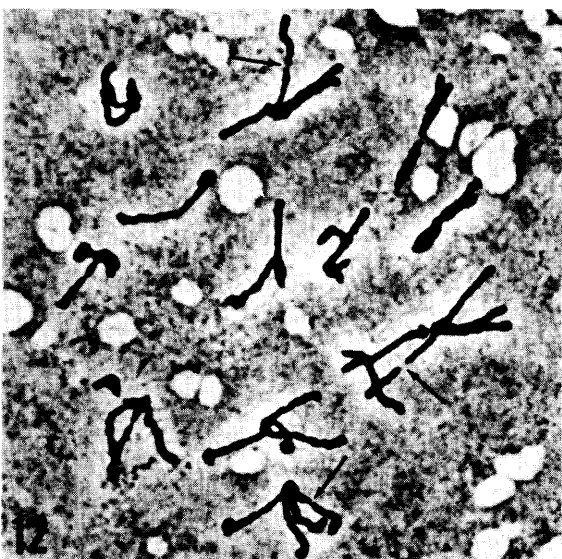
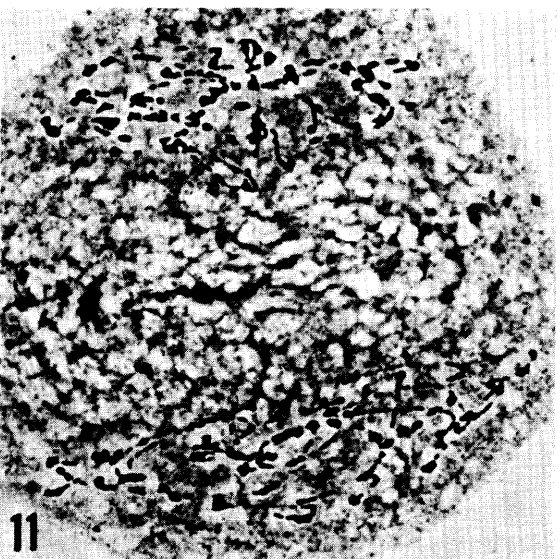
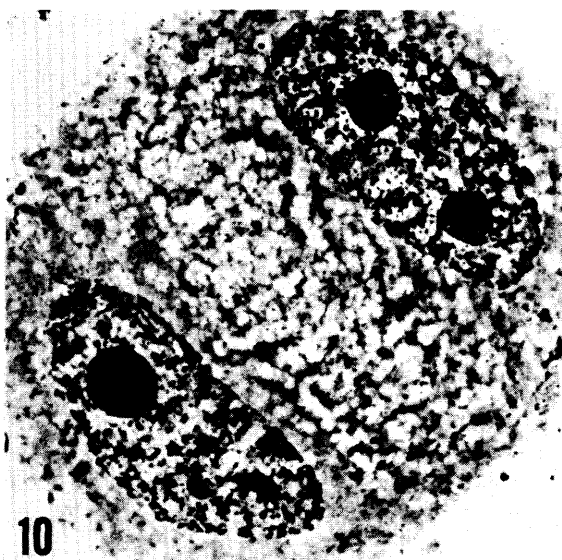
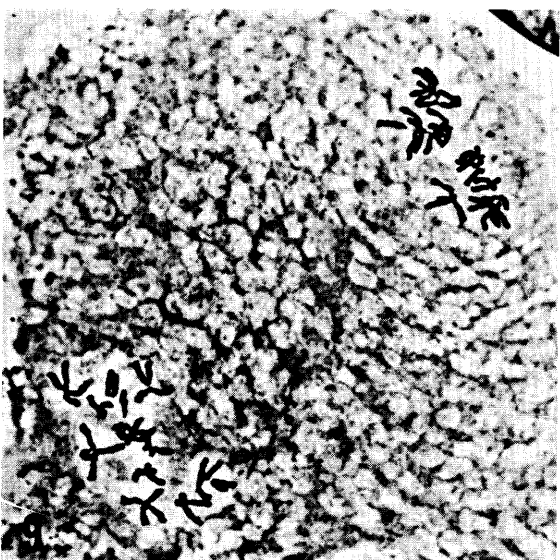
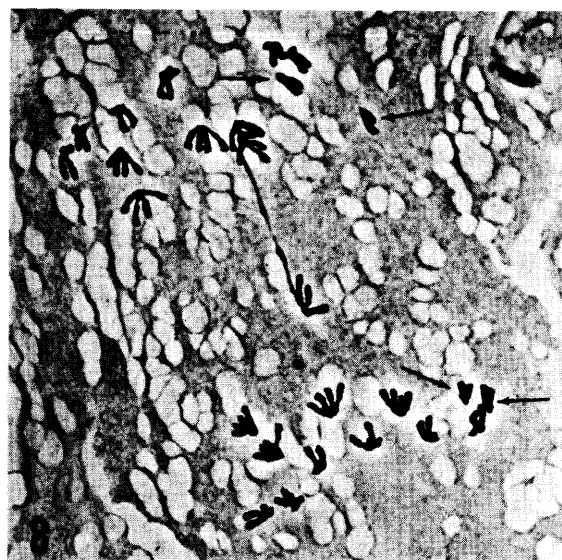
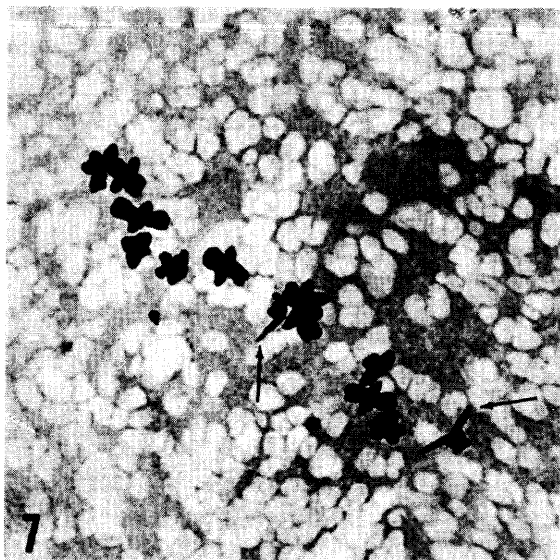
Observations of microsporocytes in four trees from the Intermountain race showed the same karyotype as the coastal race. Chromosome counts at metaphase I clearly revealed 13 bivalents. Examination at anaphase I and II confirmed these counts. As with the coastal race, three classes of chromosomes were readily distinguished according to the position of the centromere. Five chromosomes were metacentric, six were subterminal with respect to the centromere, and two were telocentric.

Discussion

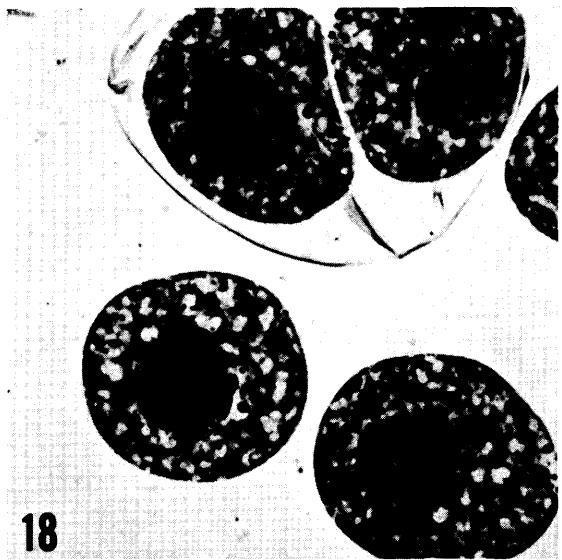
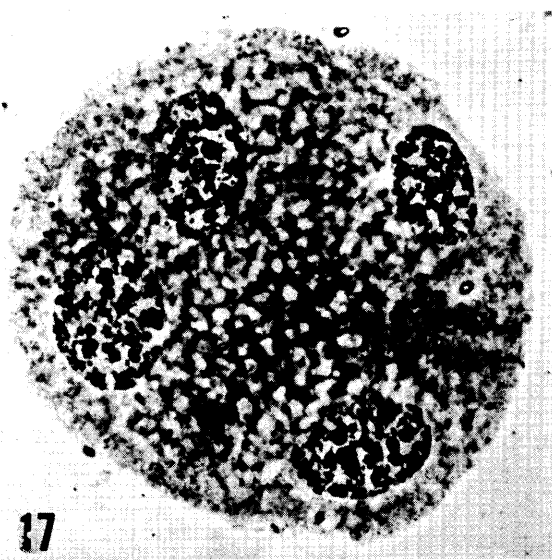
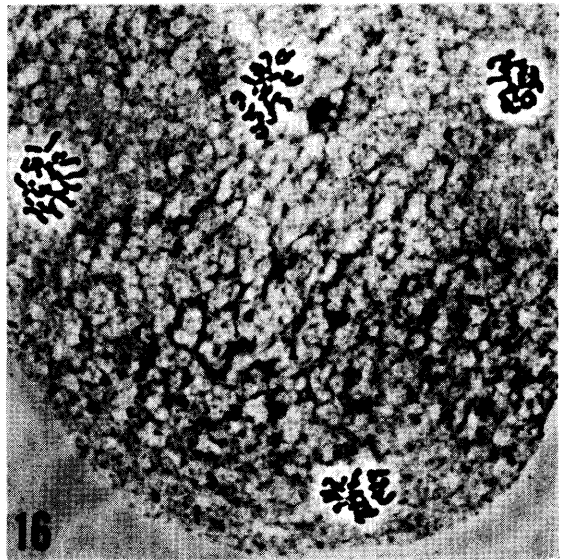
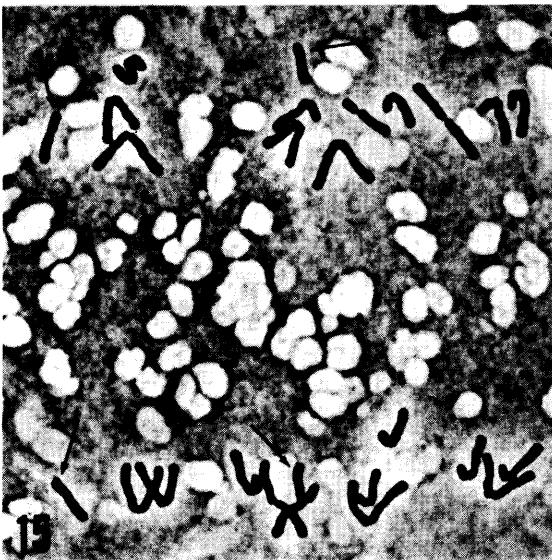
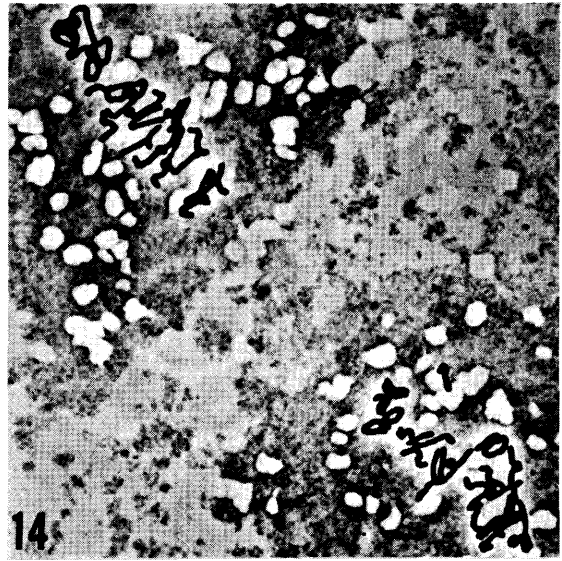
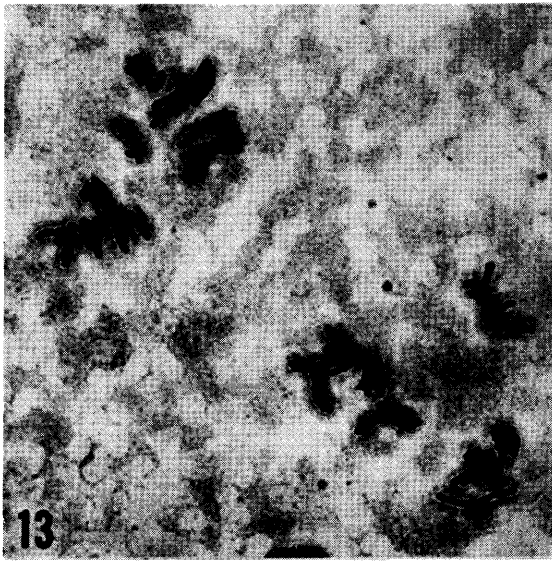
The foregoing observations on meiotic divisions in four trees at Corvallis, Oregon, indicate that the haploid chromosome number of coastal Douglas-fir (*Pseudotsuga menziesii*) is 13. Since LANGLET (1943) and DURRIEU-VABRE (1958) reported 24 chromosomes for this species, it was suspected that Douglas-fir might have different chromosomal races over its wide distribution in western North America. Accordingly, limited observations were made of meiotic chromosomes of Intermountain Douglas-fir growing in the Wasatch Mountains in central Utah. This brief survey clearly showed no karyotypic differences between the two races.

LANGLET's report dealt only briefly with cytology, and no details were given regarding the observations. DURRIEU-VABRE also examined chromosomes from root-tip meristems and found a diploid condition of 24 chromosomes, the same as other genera of *Pinaceae*. She suggested that the 13th chromosome, first described by SAX and SAX (1933), was due to breakage of a fragile centromere during preparation. This separated the arms of a single chromosome, resulting in a miscount. Her explanation, however, is contradicted by the 13 bivalents regularly found in meiosis by ZENKE (1953), and confirmed in this study. Unfortunately, DURRIEU-VABRE did not report the source of her material. Her observations, however, still suggest the possible existence of different chromosomal races in this species.

This study confirms the findings of SAX and SAX of a chromosome number of $n = 13$ for Douglas-fir. However, in contrast with their results, meiotic figures observed in all trees of this study clearly showed the presence of two chromosomes with terminal centromeres, rather than one. The ratio of the three classes of chromosomes was 5:6:2 (metacentric:subterminal:telocentric), instead of 6:6:1 reported by SAX and SAX. Their findings could be accounted for more easily on the basis of isochromosome formation, involving one of the telocentrics, than on the basis of a fragile centromere as suggested by DURRIEU-VABRE. Following centromere misdivision, separated arms, each bearing a part of the centromere, may show the union of sister chromatids within the centromere and open out to form an isochromosome (LEWIS and JOHN, 1963). Such an occurrence in only one of the two telocentrics could result in a karyo-



Figs. 7–12. — Meiosis in the microsporocytes of *P. menziesii*. — Fig. 7: Metaphase I (arrows denote telocentrics), $\times 630$. Fig. 8: Anaphase I (note telocentrics), $\times 500$. Fig. 9: Late anaphase I, $\times 500$. Fig. 10: Interphase, $\times 500$. Fig. 11: Prophase II, $\times 510$. Fig. 12: Late prophase II (arrows denote secondary constrictions on metacentric chromosomes), $\times 800$.



Figs. 13—18. — Meiosis in the microsporocytes of *P. menziesii*. — Fig. 13: Metaphase II, $\times 660$. Fig. 14: Early anaphase II, $\times 510$. Fig. 15: Anaphase II (arrows denote telocentrics), $\times 810$. Fig. 16: Late anaphase II, $\times 520$. Fig. 17: Microspore nuclei, $\times 510$. Fig. 18: Microspores, $\times 520$.

type of 6:6:1 as reported by SAX and SAX. This study however, always showed the presence of two telocentrics rather than one.

The cytological evidence on meiotic cells in this paper is in full agreement with BARNER and CHRISTIANSEN's (1962) observations of the basic karyotype in somatic cells of Douglas-fir. They observed two telocentric chromosomes in root-tip meristems and suggested that the extra pair of chromosomes may be a result of fragmentation of a chromosome at the centromere, rather than arising by duplication as proposed by SAX and SAX. BARNER and CHRISTIANSEN first advanced the suggestion that centromere breakage may account for the extra chromosome found in *Pseudotsuga menziesii*.

Similar breakage also occurs in chromosomes of other plants, as reported by DARLINGTON (1956), LEWIS and JOHN (1963), MARKS (1957), and KHOSHOO (1960). According to DARLINGTON, centromere misdivision can often give rise to plants with new chromosome numbers, without altering the genetic content of the chromosome complement. He stated, "The breakage of a chromosome is often the first visible step in the breakage of a species". Often such changes in basic numbers come to distinguish species, genera, and even larger groups in the course of evolution. Many cytotoxic studies indicate that chromosome breakage at the centromere is a common means by which chromosome numbers are increased. MCCLINTOCK (1938) provided genetic evidence in maize that both parts of a centromere, transversely broken by X-ray treatment, could successfully function in the mitotic process.

If centromere misdivision has yielded the extra chromosome of Douglas-fir, it is surprising that the two resulting telocentric chromosomes have shown such stability in division and have been perpetuated in natural populations. Regarding the position of the centromere in a chromosome, a number of investigators, including LEWITSKY (1931) and RHOADES (1940), hold that no chromosome has a true terminal centromere but that all are interstitial. They maintain that those chromosomes which appear telocentric possess a minute second arm so small as to escape observation, unless special techniques are used at critical stages. KAUFMANN (1934) showed that the rod-shaped X chromosome of *Drosophila melanogaster* has such a subterminal centromere. Both genetic and cytological evidence has since confirmed his observation.

According to RHOADES, "There is apparently no certain case of a telocentric chromosome in the regular chromosomal complement of any plant, and it is possible, though not as thoroughly established, that a similar condition is true among animals." He showed, in studies of a telocentric chromosome in hyperploid maize, that such a chromosome was involved in producing isochromosomes at meiosis, and its subsequent loss and modification in somatic tissue indicated that a terminal centromere is unstable. RHOADES stated that such "... instability may apply to all telocentric chromosomes and account for the fact that telocentric chromosomes are rarely, if ever, found in the normal chromosome complement of any organism." DARLINGTON (1939) also suggested that the absence of a terminal centromere may be due to their instability and account for their disappearance through natural selection. MARKS (1957), however, concluded that, "There is no evidence that a telocentric chromosome is unstable because its centromere is terminal." He suggests that their rarity is due, not to inefficient centromeres, but rather to low survival of cells in which they occur, because of genetic imbalance brought about by chromosome deficiencies and duplications.

It is problematical whether the two rod-shaped chromosomes in Douglas-fir are true telocentrics. A genetic analysis would provide the most critical test, but forest trees do not lend themselves to standard genetic methods. From a cytological standpoint, however, the centromeres appear terminally located, even when viewed at maximum resolution and magnification. Careful observation of meiosis disclosed that behavior of the telocentrics was as regular during pairing and segregation as for the other 11 chromosomes in the complement. This indicates their stability in the genome.

THOMAS and CHING (1968) suggested a mechanism involving chromosome translocation to explain the origin of the two telocentrics in *P. menziesii*. They ruled out the alternate hypothesis of centromere misdivision on the basis of results of earlier workers (MCCLINTOCK, 1932 and RHOADES, 1940), who held that telocentrics are unstable, and that such behavior explains why they are rarely, if ever, found in nature. However, in the latest review on the chromosome complement, JOHN and LEWIS (1968) cite a case where an increase in chromosome number has actually taken place by "centric fission" or misdivision of the centromere (DARLINGTON and LACOUR, 1950). Not only do telocentrics exist in natural populations, but the chromosome complement of *Tradescantia micrantha* was recently discovered to consist entirely of telocentrics (JONES and COLDEN, 1968). Also, in a survey of 52 species covering all genera of the *Podocarpaceae*, HAIR and BEUZENBERG (1958) found a regular numerical relationship between metacentrics and telocentrics. They reported that with successive change in basic number, a metacentric replaces or is replaced by two telocentrics. Thus, in the light of recent investigations, it is known that, contrary to earlier generalization, telocentric chromosomes do exist; they are stable in some species at least; and they can arise by centromere misdivision. The question of whether chromosome translocation or centromere misdivision has altered the karyotype of Douglas-fir therefore remains purely speculative.

There is substantial reason to believe that *P. menziesii* arose from a progenitor with 24 chromosomes, but we have no experimental proof of how this increase in chromosome number came about. The evidence favoring this view is that the *Pinaceae*, to which Douglas-fir belongs, has a basic chromosome number of 12. The only genera showing exception to this basic number are *Pseudolarix*, a monotype with $n = 22$, and *Pseudotsuga menziesii* with $n = 13$. Conforming with the rest of the family, however, *P. macrocarpa* and *P. wilsoniana* both have a diploid number of $2n = 24$ [CHRISTIANSEN (1963), THOMAS and CHING (1968)]. Neither of these two species has any rod-shaped chromosomes like *P. menziesii*, and both have one more metacentric chromosome. Also, both species are geographically separated from Douglas-fir. Whatever the mechanism for repatterning the chromosome complement has been, it seems clear that the species with the modified karyotype is now dominant, occurring over wide ranges of latitudes, altitudes, and habitats. Such vast environmental distribution could differentiate chromosomal races, but preliminary observations on the Intermountain race indicate similarity in karyotype with the coastal race.

Acknowledgements

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Summary

Microsporogenesis is described and illustrated in the male strobili of *Pseudotsuga menziesii* (MIRB.) FRANCO from the premeiotic mother cell through microspore formation. A total of eight trees were examined cytologically, of which four represented the coastal and four represented the In-termountain race of Douglas-fir. Trees from both geographic regions showed a haploid chromosome number of 13. Similarly, both races exhibit three types of chromosomes according to the position of the centromere. Among the 13 chromosomes, five had median centromeres, six had sub-terminal centromeres, and two behaved as though the centromeres were terminal. In addition to primary constrictions, sometimes up to three secondary constrictions were observed which were always in association with meta-centric chromosomes. The possible origin of the two telocentric chromosomes, unique to *P. menziesii*, is discussed.

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Induction florale précoce chez *Cupressus arizonica* et *Chamaecyparis lawsoniana*

Action de l'acide gibbéréllique et d'autres substances de croissance

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Introduction

La mise au point de méthodes permettant de provoquer une floraison précoce représenterait un outil de travail de première importance pour l'amélioration des arbres forestiers.

Depuis longtemps, les forestiers se sont attachés à résoudre ce problème, sans que pour l'instant une solution véritable ait pu être trouvée. La première solution a consisté à expérimenter un certain nombre de techniques déjà utilisées pour les plantes horticoles et les arbres fruitiers. MATTHEWS (1963), dans une étude sur les différents facteurs susceptibles d'intervenir dans la floraison des arbres forestiers, signale les techniques de taille des racines, d'annellation ou d'étranglement des tiges d'arcure des branches. Ces techniques permettent effectivement d'augmenter souvent l'intensité de la floraison, parfois aussi de gagner quelques années par rapport à l'âge normal d'apparition des premières fleurs. Ce gain de temps n'est cependant pas assez important pour que puissent être appliquées aux arbres forestiers les techniques classiques d'amélioration qui demandent un travail sur plusieurs générations. Aussi

MATTHEWS pense-t-il que le véritable progrès passe par une meilleure connaissance de la physiologie de la floraison, et, en particulier, par l'étude du rôle des substances de croissance susceptibles de pouvoir intervenir dans ce phénomène.

Or, dès 1958, des chercheurs japonais (KATO *et al.* 1958, 1960; HASHIZUME 1959—1963) signalaient la possibilité de provoquer la floraison de jeunes plants (2 ans) de divers Taxodiacees et Cupressacées par application d'acide gibbéréllique. En 1965 PHARIS et ses collaborateurs réussissaient à provoquer, par application d'acide gibbéréllique, la floraison de plants de *Cupressus arizonica* âgés seulement de 88 jours. Par la suite, PHARIS *et al.* (1967, 1969) ont pu préciser chez *Cupressus arizonica* et *Thuja plicata* dans quelles conditions climatiques pouvait s'effectuer cette induction. Ils ont notamment montré de quelle façon il était possible d'agir sur la proportion d'inflorescences mâles ou femelles en faisant varier les conditions photopériodiques.

Au cours du présent travail, nous avons essayé non seulement d'intensifier l'induction florale précoce, mais encore d'augmenter la production d'inflorescences femelles. Nous avons aussi suivi, notamment par une étude histologique, l'effet de la photopériode sur la maturation des inflorescences produites.

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