

It is apparent that the nutrient medium and controlled environment of this experiment did not necessarily enhance bud formation or rapid development of apical-type meristematic centers on other parts of the embryo (i.e., the cotyledons). However, formation of a cotyledonary vascular system was rapid as well as initial leaf primordia evidence about the apical dome.

Prospects for further tissue culture work on the excised embryos of ponderosa pine look promising. Embryos are relatively easy to work with as compared to other gymnosperm material and they react rapidly to culture media. Further work is planned to determine possibilities and factors involved with plantlet formation from excised embryos cultured in vitro. To be able to develop many plantlets of ponderosa pine from a single embryo would allow rapid clonal propagation of genetically identical individual-for genetic studies and tree improvement work.

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#### Abstract

Early stages of growth were studied on excised embryos of ponderosa pine (*Pinus ponderosa* LAWS.) grown on a chemically defined nutrient medium. Morphological and histological studies were made throughout a five week

study period. Rapid differentiation of a primary vascular system and primary leaves were noted.

Key words: Tissue culture, *Pinus ponderosa* LAWS.

#### Zusammenfassung

Bei der Kultur von Embryonen von *Pinus ponderosa* LAWS. konnte Kallusbildung beobachtet werden, wobei solche undifferenzierten Gewebepartien blattprimordiale Auswölbungen hervorbrachten. Die zum Teil erfolgte Weiterentwicklung solcher Auswölbungen gibt Anlaß, darin eine Knospenbildung zu vermuten, die evtl. zur Regeneration ganzer Pflanzen geeignet erscheint.

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## Viable Seed from a Shortleaf Pine 13 Months Old

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Scientists studying tree improvement would like to shorten the maturation period of southern pine cones, since the lengthy ripening (normally about 21 months) causes major delays in obtaining seed from the offspring of controlled crosses. Cones have reportedly matured during their first year (KATSUTA 1970, McLEMORE, in press), but there is no record that such cones have produced viable seed. This note describes a technique to induce shortleaf pine (*Pinus echinata* MILL.) cones to mature and yield viable seed in 13 months.

#### Procedures

Pine trees usually produce no cones before the age of 10 to 15 years, when tree heights of 30 to 40 feet make potting and indoor testing impractical. Therefore, it was necessary to obtain samples by grafting. Fifteen soft-tissue branches of shortleaf pine were collected and placed in a greenhouse in early April 1974. Attached were a total of 45 conelets, which had been wind pollinated immediately before collection. On April 18, the branches were cleft-grafted onto 2-year-old potted slash pine seedlings (*P. elliottii* ENGELM.) about 50 cm tall. By May 13, all but one of the scions were dead, and all but three conelets on the successful graft had aborted.

From early June through mid-July (45 days) the surviving ramet with its three conelets was subjected to treatments simulating abbreviated natural seasons. At the beginning and end of the treatment period, the graft was placed in a growth chamber for 7 days and subjected to alternating 8-hour photoperiods (15,000 lux) at 13° C and darkness at 7° C. During the 31 intervening days, the ramet was moved to a cold room (1° C), where fluorescent lights (1,000 lux) were turned on for 8 hours on weekdays but were omitted on weekends. On July 18, the graft was placed in an air conditioned greenhouse (23° C), where the cones remained until harvesting.

The conelets remained bright green throughout the summer, when they normally turn brown. In late September, they had begun to enlarge, and by mid-November, they appeared fully grown and measured approximately 3.5 cm long and 1.5 cm in diameter, slightly smaller than normal for mature shortleaf pine cones. The diminished size was probably caused by insufficient nutrition due to grafting and to the fact that the seedlings did not have enough rootstock to maintain full-sized cones.

One cone was removed from the ramet on December 31, 1974, one on February 28, 1975, and one on May 1, 1975. Immediately after harvesting, each cone was kiln-dried for 48 hours at 38° C and opened mechanically for seed removal. The seeds were left unstratified and were sown on a moist sand-peat medium for germination tests.

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## Results

The first cone yielded 44 immature seeds, and the second produced 66. X-rays taken before sowing revealed fully developed embryos. The last cone yielded 52 seeds of which 10 germinated within 10 days; no other seeds germinated. The seedlings all had four cotyledons rather than the five to seven normal for shortleaf pine, a possible result of the small seed size.

The total time from pollination to maturity was slightly over 1 year, about 7 months less than the time required in nature. The early development was probably stimulated by the changes in temperature and the shortened day lengths.

The maturation period might possibly be reduced by another 2 to 3 months. If a winter grafting could be accomplished while the strobili are still in the bud stage (stage 1), the development of the female strobili to the receptive stage (stage 5) might be accelerated in the greenhouse, and pollination might then be accomplished in January or February instead of in April. Grafting and massive conelet abortions might be avoided by developing stock that can produce cones at age 4 or 5, when the trees are small enough for potting and indoor handling.

## Abstract

A wind-pollinated shortleaf pine conelet was subjected to varying temperatures and shortened day length to simulate abbreviated seasons and induce early development. The conelet produced viable seed in slightly less than 13 months.

*Key words:* *Pinus echinata*, conelet, development, maturation, germination.

## Zusammenfassung

Durch Pfropfung von frei abgeblühten Reisern von *Pinus echinata* MILL. auf *Pinus elliottii* ENGELM. kurz nach der Bestäubung im April 1974 und anschließende Behandlung durch Veränderung der Tageslänge und Temperatur sowie mit fluoreszierendem Licht konnten in einem Fall innerhalb von 13 Monaten reife Zapfen mit keimfähigen Samen erzielt werden.

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# Some results on changes of leaf characters and readiness to flower in *Chamaecyparis pisifera*

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By its polymorphism of the foliage *Chamaecyparis pisifera* is a well known ornamental garden species in many parts of the world. Especially gardeners defined two different forms: The "juvenile" squarrosa type and the "adult" squamosa type with scale formed foliage (BEISSNER 1879, DALLIMORE and JACKSON 1966). Normally the type of foliage changes from squarrosa to squamosa type after a few years after sowing. The retention of the juvenile squarrosa type in older plants is genetically fixed, which is demonstrated by LANGNER (1964). He analysed a selfed progeny showing a segregation in squarrosa and squamosa types. Both types can be distributed widely commercially by their vegetative propagation.

In a progeny of a *Chamaecyparis pisifera* Plumosa Aurea which originated with a high probability from selfing we observed four types of seedlings 6 years after seed-

the squarrosa type (sr)

a mixed squarrosa-plumosa type in which branches or branch parts showed the needle and the plumosa character (sp),

a plumosa type with scaly leaves grown together with the branch in their basal parts and in the apical part

tapering in a point (p) and the squamosa type with shapes with rounded off apical parts and laid on the branches in its whole length (sm); (LANGNER and MELCHIOR 1968).

The broad variation of morphological and physiological characters between and within leaf types were described by the same authors.

In the following some results on (1) the changes of these leaf characters and (2) the influence of gibberellic acid ( $GA_3$ ) on the strobili production of some selected clones of this progeny are presented and more detailed elsewhere (MELCHIOR, in print).

Considering the topic (1) the seedlings from seed to an age of some 80 months have been assessed concerning their leaf character: As already mentioned by LANGNER (1964) *Chamaecyparis pisifera* within the relatively short time of two years changes the needle type from the sr to the sm type. This happened in the observed progeny (86 clones) of the genetically fixed sm-types during the 2nd growing season within some 3 to 5 months. In the same period two more clones changed into the p- and the sp-type. Most of the clones changed within an age of 2 to 4 years into the p-type, 8 changed at an age of 4 to 6 years into the mixed sp-type and more than a quarter of the clones left un-

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