Results of nine year trials on vegetative propagation of Bombacopsis quinata (Jacq.) Dugant by branch setts.[1]

By G. H. MELCHIOR[2] and M. QUIJADA R.[3]

(Received February 1972)

Because of the importance of Bombacopsis quinata (saquisaqui) in Venezuela and the difficulties of obtaining seed of this tropical tree species, investigations on methods for its vegetative propagation began in 1961. The main practical purpose was to afforest exploited areas using rooted cuttings. By 1963 a successful method had been devised namely to use lengths of branch or "setts" 5 to 25 cms in diameter and 80 cms long as cuttings and plant them four weeks to a fortnight before the trees in natural stands were flushing, that is before the rainy season begins in April (MELCHIOR 1965). Between 1965 and 1967, 11 clones were rooted and various ramets of these flowered abundantly during the 1968 dry season. Following this observation the goals of the work were changed to a long term programme for producing improved seed in a clonal seed orchard (MELCHIOR 1970). Today the clonal gardens, containing some 50 clones from Venezuelaan provenances of saquisaqui, are used as an experimental site to obtain more detailed information on flowering and its relation to the breeding system. These clonal gardens are also being used as seed producing area, even though they still are of unknown genetic quality (MELCHIOR and QUIJADA, in preparation).

Because vegetative propagation of saquisaqui by branch setts has become routine in tree breeding in Venezuela, the most important results from nine years of experiments are presented here. A more detailed report will be published in the near future in the Venezuelaan Forestry Review (MELCHIOR and QUIJADA, in preparation).

Results *and* Discussion: The first test revealed a significant dependence of rooting ability on the date when the cuttings were taken. However, setts planted at different dates originated from different trees, because the individuals which were felled on each occasion had to be used immediately. These first tests made in October, January, April, May involved few clones and setts less than 5 cms in diameter. Because of the relatively small diameter of the cuttings and the small number of clones these first trials cannot be compared with those which followed.

The next set of results were also obtained with material of different clones: 30 were used in March (501 setts), 5 in April (167 setts), 4 in May (108 setts) and 2 in June (25 setts). The dimensions of the setts were those described in the introduction to this paper. The R X 2 test again showed a highly significant dependence of rooting on the date when the cuttings were taken (Tab. 1.1). The months of March and April gave a rooting percentage of about 50, whereas May and June showed values of 2 and 28 respectively.

It will be easy in future to confirm the assumed dependence of the rooting ability on the date when cuttings are taken because sufficient secondary material of the same clones is now available for planting at different times.

A trial in 1968 enabled us to test by analysis of variance differences in rooting ability between provenances, clones within provenances and diameters of cuttings. Setts of different diameters from four trees in each of three provenances were taken as cuttings; these were planted at three locations each with 6 ramets in such a way that each ramet was surrounded by others of a different clone.

One of the three test areas was permanently submerged in water during the rainy season and all the cuttings planted there died. Therefore, this site was not included in the calculations. The two remaining sites showed no significant differences, probably because of the single number of degrees of freedom.

Table 1. — Rooting ability of Bombacopsis quinata cuttings. Comparison of different factors by various χ^2-tests (SACHS 1968, pp. 341—345; 448—453). The numbers 1 to 13 indicate the numbers in the text and the asterisks indicate the level of significance (***: a = 0.001; **: a = 0.01; n.s. = not significant).

No.	The Dependence of Rooting ability on Time of planting	N	K	d.f.	χ^2
1	Time of planting	801	4	3	88,480***
2	Primary (P)J Secondary (S) setts	1122	—	1	510,599***
3	PIS setts, clone 1.63	124	—	1	14,770***
4	PIS setts, clone 1.65	135	—	1	10,584***
5	PIS setts, clone 3.65	161	—	1	52,699***
6	P/S setts, clone 4.65	115	—	1	2,821 n.s.
7	PIS setts, clone 5.65	150	—	1	61,167***
8	PIS setts, clone 6.65	39	—	1	3,687 n.s.
9	PIS setts, clone 8.65	169	—	1	62,247***
10	P/S setts, clone 1.66	169	—	1	9,202**
11	P/S setts, clone 1.67	60	—	1	3,076 n.s.
12	Primary clones	845	9	8	217,110***
13	Secondary clones	277	9	8	22,946**

---

[1] In honour of the 65th birthday of Professor WOLFGANG LANGNER. This work was made possible only by the consistent and interested help of our technical cooperators: Srs. RAPAEL CORROZ, VALMORE GUTIERREZ U., GERMÁN TORRES y AMADEO ZAPATA. We gratefully acknowledge their cooperation.

[2] Institut für Forstgenetik und Forstpflanzenzüchtung, D-207 Schmalenbeck, Siekerlandstr. 2, Bundesforschungsanstalt für Forst- und Holzwirtschaft, B.R.D.

[3] Sección de Genética Forestal, Instituto de Silvicultura, Facultad de Ciencias Forestales (Apartado 305), Universidad de los Andes, Mérida/Venezuela.

The other results revealed significant differences between provenances, that is the rooting ability of cuttings from the drier climate being less than that from the more humid one. The intraclass-correlation (I. C.) for differences between provenances was 21.2 percent.

Thicker primary cuttings (> 9.9 cms) rooted better than thinner ones (< 9.9 cms) and had an I. C. of 24.9 percent. For clones within provenances the intraclass-correlation was 22.9 percent. For the experimental error, which was pooled over non-significant interactions (provenances × treatment and clone × treatment) the result was 31 percent.

Since the diameter of a tree — for the lack of other criteria in the tropics — is often used as an indicator of the age of an individual, a correlation analysis between the stem diameter of the parent trees (DBH) and rooting ability of the cuttings taken from them was made and this showed a non-significant coefficient ($r = —0.075$, 40 d.f.). Because older trees of saquisaqui yielded cuttings showing an inferior rooting ability than those from younger trees (MELCHIOR, in preparation), this result clearly contradicts the stem diameter-age relationship.

Primary and secondary setts also showed significant differences in rooting ability *(Tab. 1.2);* secondary cuttings were more easily rooted (92.8%) than primary ones (17.8%) and this was the case in 6 out of 9 clones from which both the two groups of setts could be compared *(Tab. 1.3—1.11).*

Differences were found between clones for primary and secondary cuttings *(Tab. 1.12, 1.13),* but there seems to be a higher heterogeneity in the primary cuttings. When primary and secondary cuttings of five comparable clones were classified according to their diameter, we also found, that primary cuttings showed a lower rooting ability than secondary ones, and additionally a higher variation for the two different diameter groups of primary cuttings than for the two groups of secondary cuttings *(Tab. 2).* Comparing the smaller diameter groups of both types of cuttings with the groups of thicker cuttings significant differences between them were found, and a lower, but still highly significant variation was observed for cuttings thicker than 9.9 cms than for the corresponding groups which were thinner than 9.9 cms.

Thus the rooting percentage did not increase to the same extent in thick and thin cuttings according whether they were primary or secondary but it is stronger in the smaller diameters. Cuttings with diameter less than 9.9 cms increased in rooting ability from primary to secondary cuttings from 14.4 to 93.5 percent whereas those with a diameter of more than 9.9 cms increased in rooting ability from 64.2 to 95 percent.

Table 2. — Comparison of primary cuttings (n_1, n_2 (I)) of 4 (+1) clones with the secondary cuttings of the same clones (n_3, n_4 (I)); and cuttings of diameters thicker than 9.9 cms (n_1, n_3 (II)) with those thinner than 9.9 cms (n_2, n_4 (II)) using the BRANDT SNEDECOR formula (SACHS 1968, pp. 453—460). Level of significance: see *table 1.*

Components of variation	$\hat{\chi}^2$	d.f.
Differences between $\bar{p}$'s of the sample groups		
$n_1 + n_2$ and $n_3 + n_4$ (I)	312,3846***	1
$n_1 + n_3$ and $n_2 + n_4$ (II)	80,7666***	1
Variation between p's within		
$n_1 + n_2$ (I)	115,6584***	7
$n_3 + n_4$ (I)	2,3629 n.s.	7
$n_1 + n_3$ (II)	37,2725***	7
$n_2 + n_4$ (II)	312,3668***	7
Total deviations of the p's from $\hat{p}$ within $n = n_1 + n_2 + n_3 + n_4$ (I, II)	430,4059***	15

When primary and secondary setts were planted during the optimal time for rooting in March and April, secondary setts of small diameter attained the rooting ability of those thicker than 9.9 cms (more than 90 percent). So, the low water and nutrient content of thin primary cuttings cannot be of great importance in their poor rooting ability. It is suggested that the physiological age of the ortet (that is, cyclophysis) and/or the effect of topophysis has influenced the rooting success of primary cuttings to a considerable degree. These influences can apparently be completely eliminated by one more step of homeoplastic propagation and the propagated secondary plant material is rejuvenated in its rooting ability. But the same secondary "rejuvenated" cuttings also flowered within one year after vegetative propagation in the same manner as primary ones. Thus, their behaviour in flowering and fruiting is similar to that of mature trees. Therefore, if high rooting ability is a criterion of the juvenile phase, then flowering ability must be independent of other physiological stages in saquisaqui.

Summary

1) *Bombacopsis* setts 5—25 cms in diameter rooted best between March and April, some time before the species flushes in stands.
2) Primary setts from a dry region and with small diameter did not root so well as those from a more humid region and with bigger diameter. There were differences between clones within provenances.
3) There is no relation between the diameter of the parent tree — as indication to its age — and the ability of cuttings to root.
4) Secondary cuttings rooted better than primary cuttings; thin secondary cuttings (thinner than 9.9 cms) reach the same rooting percentage as thick primary cuttings (thicker than 9.9 cms).
5) Because thin secondary cuttings and primary cuttings flowered abundantly one year after rooting and the rooting ability increases from primary cutting to secondary cutting, it is concluded that flowering ability is independent of physiological age in *Bombacopsis.*

Key words: Bombacopsis quinata, rooting ability, primary cuttings, secondary cuttings, differences between provenances, clonal differences, influence of diameter, flowering ability.

Zusammenfassung

1) *Bombacopsis*-Rollen von 5—25 cm Durchmesser bewurzeln sich optimal in der Zeit zwischen März und April, einige Zeit bevor die Art im Bestand austreibt.
2) Primär-Rollen aus einer trockenen Region und geringen Durchmessers bewurzelten sich weniger gut als solche aus einem mehr feuchten Gebiet und größeren Durchmessers. Zwischen Klonen in Provenienzen bestehen Unterschiede.
3) Zwischen dem Durchmesser des Ausgangsbaumes — als Hinweis auf sein Alter — und Bewurzelungsfähigkeit bestehen keine Beziehungen.
4) Sekundärstecklinge bewurzeln sich besser als Primärstecklinge; Sekundärstecklinge, die dünner als 9,9 cm waren, erreichen das gleiche Bewurzelungsprozent wie Primär- und Sekundärstecklinge mit einem Durchmesser von mehr als 9,9 cm.

5) Da auch dünne Sekundärstecklinge wie Primärstecklinge ein Jahr nach dem Steckzeitpunkt und sehr zahlreich blühen und ihre Bewurzelungsfähigkeit vom Primärsteckling zum Sekundärsteckling steigt, wird bei *Bombacopsis* auf Unabhängigkeit der Blühfähigkeit vom physiologischen Alter geschlossen.

Schlagworte: Bombacopsis quinata, Bewurzelungsfähigkeit, Primärstecklinge, Sekundärstecklinge, Provenienzunterschiede, Klonunterschiede, Einfluß des Durchmessers, Blühfähigkeit.

Literature Cited

Melchior, G. H.: Über die Vegetativvermehrung von *Bombacopsis quinata* (Jacq.) Dugant. Silvae Genetica **14**, 148—154 (1965). — Melchior, G. H.: Die Vegetativvermehrung von *Bombacopsis quinata* (Jacq.) Dugant als Grundlage für seine Saatguterzeugung in Venezuela. Beitrag, IUFRO Sektion 23, Arbeitsgruppe 2 / Tropischer Waldbau. 30. 9. — 7. 10. 1970, Ljubljana/Jugoslawien. — Sachs, L. Statistische Auswertungsmethoden. Springer Verlag Berlin, Heidelberg, New York, 1968.

On the Development of Pollen and the Fertilization Mechanisms of Larix and Pseudotsuga menziesii

By H. Christiansen

The Laboratory of Genetics of the Royal
Veterinary and Agricultural University
Copenhagen and The Danish State Forestry's
Tree Improvement Station, Humlebaek, Denmark

Introduction

Since Geleznov (8) and (5) in 1849 demonstrated the unique pollen-catching device in *Larix,* and Strasburger (18) and Beljaeff (2) in 1884—93 discussed the character of the generative pollen cell in the species, the different stages of the development of the pollen and the pollination and fertilization mechanisms of *Larix* have been the subject of several investigations, but, nevertheless, important details in these processes are still more or less in the dark.

This may seem strange, but it is a natural consequence of the absence of sufficiently detailed, continuous investigations of complicated processes like the pollination and fertilization mechanisms in conifers. The main causes are: undependable flowering, adverse climatic conditions, attacks by insects, great variations in structure even in closely related species or within the same genus (cf. Ferguson (7), p. 217, point 14) and technical difficulties.

The knowledge in this field is therefore based on many descriptions of limited details (Sterling (17)) and sometimes, as it would seem, on homologies and analogies not too well founded.

Sterling ((17) p. 188) states:

> "Among the gymnosperms the greatest amount of variation in sexual development occurs in the Coniferales and Taxales. It would appear that a closer investigation of gametophyte development in this group could provide further clues to their phylogenetic development and their interrelationships. Nevertheless there are many gaps in the knowledge of the different genera".

It also appears from Sterling's review that the interpretations of different investigators are by no means always unanimous. He says f. inst., (p. 168):

> "However, resemblances among lower and higher plants in the organization of the male gametophyte (and its gametangium) are less readily perceived. Here only two homologues have been universally admitted: the first cell of the gametophyte (the spore) and the last one (the male gamete or sperm)".

and in the next paragraph:

> "Because of the reduced structure of the gymnospermous male gametophyte, homologies of its cells have not been unambiguously interpreted. Differences in concepts of morphology have been reflected in differences of terminology."

It is evident that in the circumstances the study of the history of the pollen development in gymnosperms is extremely difficult although Sterling's excellent review of the situation, particularly as regards the Coniferales and Taxales, is a great help. Particularly as regards the pollen cells, different interpretations and varying results of different investigations have, of course, numerous causes, most of them understandable. Remarkable are the uncertainties connected with the determination of the exact number of prothallial cells in several groups. When, f. inst. in respect of the *Araucariaceae* ((17) p. 192), it is documented that 13 to 40 secondary prothallial cells are produced, the suspicion involuntarily arises, that such — and perhaps other similar, although less spectacular — variations may be due to the recently discovered ability of pollen cells to divide (and produce haploid plants) when subjected to abnormal growth conditions. If this is possible, it involves a new complication of an already complicated situation, and new investigations may be required to find out the actual state of affairs.

The present investigation aims to fill in and elucidate some of the above mentioned gaps, which are of taxonomic as well as of practical significance. The subjects of the investigation are particularly the events occurring during the development of the pollen from the tetrad stage till the male gametes arrive at the archegones.

According to Sterling ((17) p. 189) the male gametophyte in *Larix, Picea, Abies* and *Pseudotsuga* before pollination consists of a five-celled row i. e.: 2 prothallial cells, 1 stalk (or sterile) cell, 1 spermatogenous cell and 1 tube cell. We found in *Larix* as a maximum: 2 prothallial cells, a dubious stalk cell, a spermatogenous cell, but no ordinary tube cell and no generative cell.

According to Chamberlain ((3), p. 227) *Larix* is taxonomically classified under the *Abietaceae,* the pollen of which produce a pollen tube and readily germinate on artificial substrate. According to our findings pollen of *Larix* (and *Pseudotsuga)* produce no ordinary pollen tube, and it has hitherto not been possible with pollen of the two species to make it discharge male cells on artificial substrate (4c).

The unusual interphase nucleus *(fig. 10),* the unique internal structure of the mature pollen grain *(figs. 15—18),* the absence of a pollen tube and the impossibility of artificially producing discharge of the male cells seem to indicate that *Larix,* as regards the male gametophyte, must be placed somewhere between the *Ginkgoales* and pollen-tube-producing *Coniferales.*