Table 3. — The correlation coefficients comparing the effect of different number of seedlots, replications and plot size sampled.

No. of replications and plot size compared	No. of seedlots sampled				Average of 4 different seedlot sizes
	10	15	20	27	
II-1 vs X-4	0.658*	0.632*	0.625**	0.570**	0.621
IV-1 vs X-4	0.624	0.768**	0.708**	0.731**	0.708
VI-1 vs X-4	0.926**	0.925**	0.919**	0.900**	0.918
X-1 vs X-4	0.963**	0.950**	0.934**	0.914**	0.940

*Significant at the 5 percent level

**Significant at the 1 percent level

cations in the sample *(Figure 1)*. The amount of improvement in the sampling efficiency *(Table 2)* was 0.067 feet for plot size (average of 4 seedlot sizes × 9 different replications or 36 standard errors) and 0.279 feet for different replications sampled (average of 4 seedlot sizes × 4 plot sizes or 16 standard errors).

Regardless of the seedlot sizes *(Figure 1)*, measurement of the tallest per plot (top curves) was always less efficient than that of all 4 trees in the plot (bottom curves); similarly, sampling of just 2 replications was less efficient than that of 3 replications and as the number of replications increased, the standard error curves continued to drop and became leveled off when 6 or more replications were included in the sample. The results of the factual black pine data were comparable to those of the computer-simulated fictitious data reported by Lee (1981). Inclusion of 6 or more replications in the sample does not significantly improve the sampling efficiency.

The simple correlation analysis was conducted using seedlot means as items with (f - 2) degrees of freedom, where f is the number of seedlots sampled. The results are shown in *Table 3*. All but one correlation coefficient (= r) were statistically significant at the 5 or 1 percent level (number of replications in Roman numerals and plot size in Arabic figures). The strong correlation means superior seedlots or genetic material can be identified regardless of plot size or number of replications sampled in the present study.

Partial measurement is associated with a weak degree of loss (= 1 $-r^2$) in genetic information. When all four seedlot sizes were pooled together, it was possible to examine the combined plot size-replication effect. Sampling of 4 replications-tallest tree per plot combination (IV-1) resulted in a loss of 50 percent (r^2 = 0.50) in genetic information. This figure was reduced to 16 percent (r^2 = 0.84) when 6 replications-tallest per plot combination (VI-1) was compared with all 10 replications-all 4 trees per plot (X-4) combination. Sampling of 6 replications instead of all ten will save 40 percent in measurement effort.

When the effect of plot size alone is considered, there is a close similarity in statistical efficiency between black pine and ponderosa pine *(Pinus ponderosa* Dougl. ex Laws.) reported by Lee (1974). The loss in genetic information is under 12 percent (r^2 of X-1 vs. X-4 was 0.88 for black pine; r^2 of tallest vs. all 12 trees was 0.96 for ponderosa pine) in spite of the differences in tree age (14 years from seed for black pine and 2 years from seed for ponderosa pine) and plot size (4 trees were planted per plot for black pine, but 12 trees per plot for ponderosa pine) between the two pine species. Measurement of the tallest (or the best tree) per plot is efficient, time-saving, but accompanied by a small tolerable loss in genetic information.

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Genetic Variation in Needles of Pinus caribaea var. hondurensis Barr. et Golf. from natural Stands[1])

By R. Salazar

Tropical Agricultural Research and Training Center
(CATIE), Dept. of Renewable Natural Resources,
Turrialba, Costa Rica.

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Abstract

Analysis of the anatomy and morphology of mature needles of nine provenances of *Pinus caribaea* var. *hondurensis* Barr. and Golf. detected that most of the variation is present between and within trees.

Populations in Los Limones and Santa Clara from Honduras showed 80 and 50% respectively of the fascicles with four and five needles, which may be due to gene interchange with *Pinus oocarpa* Schiede. Further work will be necessary to determine if these two populations should be considered as hybrids.

Number of stomata, resin ducts and intrusion of hypoderm cells showed a slight clinal trend with respect to latitude but the provenance Alamicamba (lower value for stomata) and Melinda (higher value) were exceptions to the clinal trend. Also Melinda had shorter needles.

[1]) The paper is adapted from part of the author's Ph. D. Thesis, Forestry Department, Oxford University, England, 1981.

Key words: *Pinus varibea* var. *hondurensis* Barr. et Golf., Needles, Provenances, Anatomy, Morphology, Genetic variation.

Zusammenfassung

Eine Analyse der Anatomie und Morphologie der Alters-Nadeln von neun Herkünften von *Pinus caribea* var. *hondurensis* Barr. et Golf. zeigte, daß der größte Teil der Variation zwischen und innerhalb der Bäume vorhanden ist.

Bei Populationen in Los Limones und Santa Clara von Honduras zeigten 80 bzw. 50% der Kurztriebe vier oder fünf Nadeln, was auf eine Gen-Interaktion mit *Pinus oocarpa* Schiede zurückzuführen sein dürfte. Es werden weitere Untersuchungen nötig sein, um herauszufinden, ob diese beiden Populationen als Hybriden betrachtet werden müssen.

Die Anzahl der Stomata, der Harzkanäle und das Eindringen von Hypodermzellen zeigten einen leichten klinalen Trend in Bezug auf die geographische Breite. Die Provenienzen Alamicamba (geringerer Wert für die Stomata) und Melinda (höherer Wert) bildeten Ausnahmen beim klinalen Trend. Die Herkunft Melinda hatte außerdem kürzere Nadeln.

Introduction

The patchy distribution of the natural populations of *Pinus caribaea* var. *hondurensis* Barr. et Golf. covers several geoclimatic conditions and therefore genetic variation among provenances can be expected.

Considerable information is published about its natural distribution and morphological variation (Barrett and Golfari, 1962; Lückhoff, 1964; Abell, 1969; Kemp, 1973a, 1973b; Lamb 1973). There is, however, little available information with respect to the morphological and anatomical variation of the needles, within the natural distribution of *P. caribaea* var. *hondurensis*. Such information could be a valuable tool in the identification of provenances in juvenile stages.

Most of the information available is concerned with tree form and wood properties, where important differences between populations have been observed (Barrett and Golfari, 1962; Kemp, 1973a; Lamb, 1973; Greaves, 1978).

Variation in needle morphology has been found in several soft pines; it his been of great value in taxonomic studies, principally when no cones or flowers are available (Haller, 1965; Mergen, 1959; White and Beals, 1963). Mergen, Snyder and Burley (1966) found a North-South tendency in morphological traits of *P. elliottii* Engelm. Shorter needles, wider needles per unit lenght, more resin ducts, and smaller twigs were reported in provenances from xeric conditions. Harlow (1931) found that the number of resin canals in several pine species varied with the degree of exposure to sunlight. The shaded needles have less resin ducts. Nevertheless he stated that this variable could be important in pine identification, principally in species that normally have two or three needles per fascicle.

Mergen (1958a, 1958b) and Kriebel and Fowler (1965) found genetic variation in serration number, position and number of stomata per row, and position and number of resin ducts, in various species and hybrids of pines. Some of the traits also varied with the age of the tree and needle position. Geographic variation was found in *P. banksiana* Lamb. by Schoenike (1976) in terms of needle length, needle width, length per width, needle volume, curvature, number of stomata, serrations, resin ducts, and hypoderm layers. Fowler (1964) found in *P. resinosa* Ait. that the variation of some needle traits was random rather than systematic.

From the literature available on other species it is possible to see that needles are strongly affected by environmental conditions, variation that can be detected within the crown. Nevertheless the lack of information available for *P. caribaea* var. *hondurensis* makes it interesting to assess the morphology and anatomy of its needles in an attempt to determine geographic variation patterns.

Materials and Methods

To select the provenances, the area of natural distribution was divided in to four groups *(Figure 1)*, based on latitude and altitude.

The discontinuity in altitudinal stratification was due to the lack of samples in the Forest Herbarium, Oxford University, from where the material was taken. Two provenances were selected at random from each one of the four groups. The provenance Limones characterized as the driest area in the natural distribution of the variety was added. *Table 1* shows the nine populations selected and the respective geoclimatic information.

Needle samples have been collected since 1978 by the Forestry Department Oxford University. Trees were taken at intervals of approximately 60 km along the coastal stands while inland populations were selectively sampled as a result of their scattered distribution (Kemp, 1973a).

Ten or more well separated and mature trees per stands were selected. Two branch tips were taken from the centre of the crown and from the primary or secondary lateral branches in random compass directions.

Table 1. — Geoclimatic information of the provenances studied.

Nº	Country	Location	Latitude (ºN)	Longitude (ºW)	Altitude (masl)	Mean Annual Rainfall (mm)	Dry months Per year	Mean temperature (°C)
1	Nicaragua	Karawala (KAR)	12º 58'	83º 43'	10	3897	0	26.4
2	Nicaragua	Pantasma (PAN)	13º 20'	85º 57'	475	1400	5	20.7
3	Nicaragua	Alamicamba (ALA)	13º 34'	84º 17'	25	2610	3	27.3
4	Honduras	Santa Clara (STA)	13º 48'	86º 12'	700	1818	5	23.4
5	Honduras	Los Limones (LIM)	14º 03'	86º 42'	700	663	7	22.2
6	Guatemala	Poptun (POP)	16º 21'	89º 25'	500	1688	4	24.2
7	Belize	Las Lomitas (LOM)	16º 28'	88º 33'	30	2398	3	27.1
8	Belize	Mountain Pine Ridge (MPR)	16º 58'	89º 00'	487	1558	3	23.9
9	Belize	Melinda (MEL)	17º 01'	88º 20'	12	2137	2	26.9

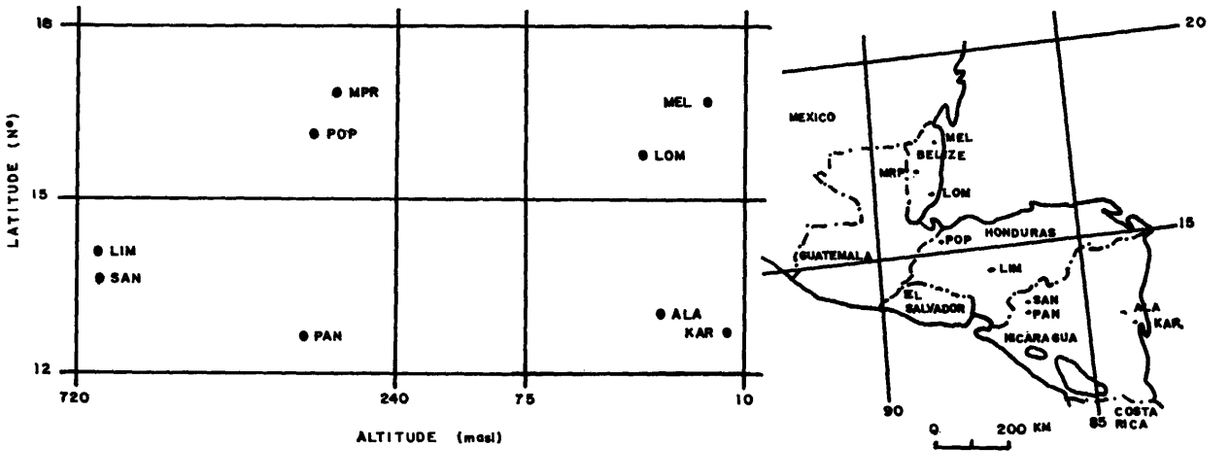

Figure 1. — Scheme of stratification used to select the provenances.

The present study assessed nine provenances with ten trees per provenance, and five fascicles per tree.

The following 17 morphological and anatomical traits were assessed:

1.	Number of needles per fascicle	nf
2.	Needle length (cm)	nl
3.	Serration number per 5 mm segment	sn
4.	Breadth cross-section (mm)	bcs
5.	Height cross-section (mm)	hcs
6.	Rows of stomata on abaxial surface	rsb
7.	Stomata number/row on abaxial surface	sb
8.	Rows of stomata on adaxial surface	rsa
9.	Stomata number/row on adaxial surface	sa
10.	Total number of stomatal rows per 5 mm segment	tsr
11.	Total number of stomata per 5 mm segment	tns
12.	Number of resin ducts in internal position	rip
13.	Number of resin ducts in medial position	rmp
14.	Total number of resin ducts	trd
15.	Diameter of the resin ducts (μ)	did
16.	Number of cells around resin ducts	crd
17.	Intrusion of hypoderm cells into the chlorenchyma	ihc

Needle length was measured to the nearest millimetre (mm) on the longest needle in the fascicle. A section 4 cm long was taken from the centre of the needle; for the remaining traits it was boiled and stored in 30% alcohol. From this material, traits three to eleven (except four and five) were assessed in a 5 mm section under a light microscope.

The last six variables were examined from hand-cut transverese sections, mounted on slides with a drop of hot lactic acid, and measured with a Projectina projector at × 100 magnification. Traits four and five were measured from the same section but with × 10 magnification.

Serration number was counted along one edge of the needle. Breath of cross-section is the distance of the straight line between the two edges of the abaxial surface. Height of cross-section is the distance of the straight line between the centre of the abaxial surface and the point where the two abaxial surfaces are joined. Stomatal number per row was recorded in a complete row. Resin ducts in internal and medial position were recorded agreeing with Lückhoff (1964). The number of cells around the largest resin duct was counted; the resin duct internal diameter was recorded in microns (μ). When more than one layer of hypoderm cells was present, it was taken as an intrusion.

Each of the 17 traits was assessed for between and within population variability using the following fully random model:

$$Yijk = \mu + Pi + Tj/i + Eijk$$

Where:

Yijk = mean value of the trait Y in the k^{th} fascicle of the j^{th} tree in the i^{th} provenance.

μ = true mean effect;

Pi = effect of the i^{th} provenance; i = 1,2 . . . . . 9;

Tj/i = effect of the j^{th} tree in the i^{th} provenance; j = 1, 2 10

Eijk = error specified by the effect of k^{th} fascicle of the j^{th} tree in the i^{th} provenance; k = 1, 2 . . . . . 5.

The analysis of variance and the expectation mean squares, are shown in *Table 2.*

Table 2. — Analysis of variance and expected mean squares.

Entry number	Sources of variation	d. f.	Test against entry	Expected mean squares
1	Provenances (P)	(P-1)	2	$\sigma^2_F + f\sigma^2_{T/P} + ft\sigma^2_P$
2	Trees (T) in P	P(T-1)	3	$\sigma^2_F + f\sigma^2_{T/P}$
3	Fascicles (F) in T in P	PT(F-1)		σ^2_F

The following basic model of multilinear regression analysis was used, to achieve a better understanding of the relationship of each dependent variable with the independents:

$$Y = a + b_1 x_1 + b_2 x^2_1 + b_3 x_2 + b_4 x^2_2 + b_5 x_3 + b_6 x^2_3 \ldots$$

where Y is the dependent variable; a the regression constant; $b_1, b_2, b_3 \ldots\ldots b_6$ regression coefficients; and $x_1, x_2, x_3 \ldots\ldots$ independent variables.

Principal component and cluster analysis were used to detect any specific provenance grouping, in terms of the provenance means of all the traits studied.

Results and Discussion

The results of the analysis of variance *(Table 3)* show that all the traits except breadth of the cross-section, and the number of stomata in abaxial and adaxial surfaces, are significantly different between the nine populations.

Table 3. — Summary of analyses of variance for morphological and anatomical traits of the needles.

Entry No.	Sources of variation	d.f.	Test against entry	nf	nl	an	bcs	ncs	rsb	sb	rsa	sa	trs	tns	rip	rmp	tnr	drd	crd	ihc
											MEAN SQUARES									
1	Provenances (P)	8	2	2.55	83.51	181.60	2.00	0.17	39.03	106.80	54.79	123.70	143.70	408800	2.92	3.99	0.91	51.26	85.81	6.91
2	Trees (T) in P	81	3	0.37	34.90	73.83	2.18	0.03	10.79	97.35	16.66	95.27	33.70	7623	0.66	0.74	0.26	696	1.31	0.85
3	Fascicles in T in P	36		0.06	11.50	9.09	2.07	0.00	2.88	19.83	8.13	18.64	6.50	11160	0.15	0.24	0.04	132	1.44	0.35
											F TEST									
1	P			***	*	*	NS	***	**	NS	**	NS	***	***	***	***	*	***	***	***
2	T in P			***	***	***	NS	***	***	***	***	***	***	***	***	***	***	***	NS	**
											VARIANCE COMPONENTS (%)									
1	P			25.91	8.73	8.91	0.00	15.04	11.23	0.53	7.19	1.76	14.97	21.37	15.04	16.18	15.65	26.56	9.05	16.54
2	T in P			36.44	52.45	53.51	1.05	34.24	31.49	43.65	16.11	44.33	40.11	42.21	34.24	25.32	44.40	33.79	0.00	7.98
3	F in T in P			37.61	38.81	37.58	98.95	50.73	57.27	55.82	76.70	53.91	44.93	36.22	50.73	58.49	41.95	39.65	90.95	75.48

* significant at P <0.05 of probability
** significant at P <0.01 of probability
*** significant at P <0.001 of probability
NS not significant at P <0.05 of probability

number of needles per fascicle, total number of stomata, and diameter of the resin ducts showed 25.91, 21.23, and 26.56% respectively of genetic variation as the highest. In the other 14 traits, although most of them showed significant differences between provenances, the majority of the variation was concentrated between trees and between fascicles in trees. Hence the taxonomic value of needle traits in this variety could be low, apart from the first traits which were shown to have high genetic variation.

The genetic variation detected in the number of needles per fascicle is due to the provenances Santa Clara and Los Limones that had 80 and 50% respectively of the fascicles with four and five needles. The Studientized range test *(Figure 2)* separated these two populations completely from the rest. Normally the number of needles per fascicle is quite constant within a species or decreses slightly in response to dry conditions (Haller, 1965; Burley, Burrows and Waters, 1967; Burley and Burrows, 1972). It is possible

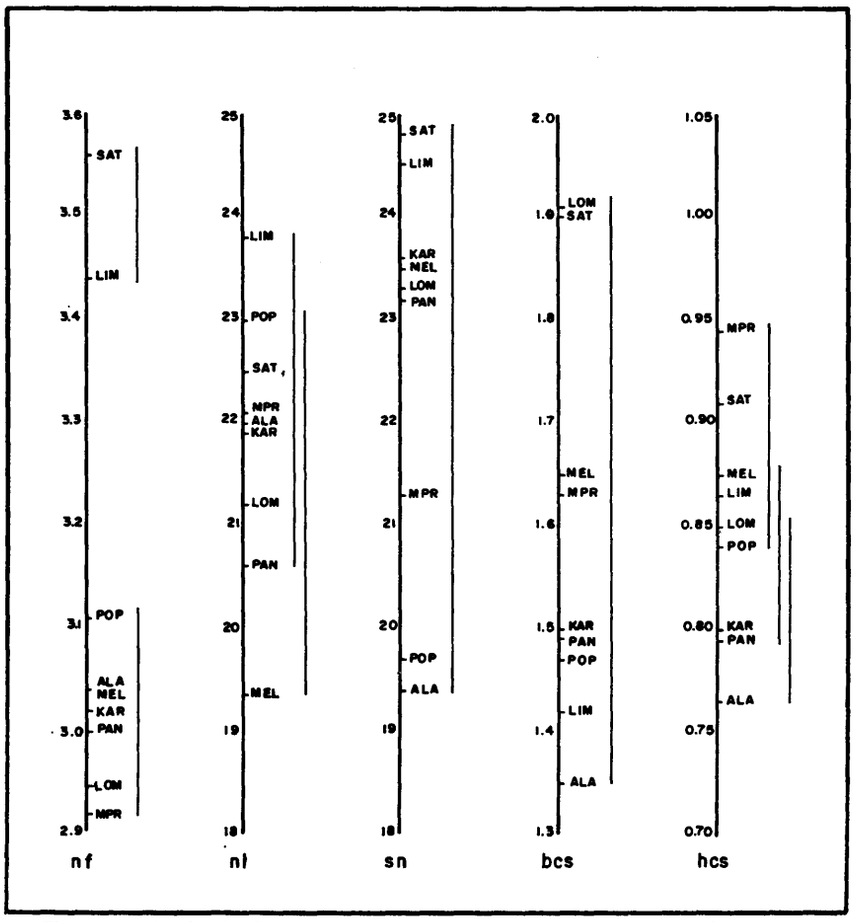

Figure 2. — Studentized range test for morphological traits of the needles ($P < 0.05$).

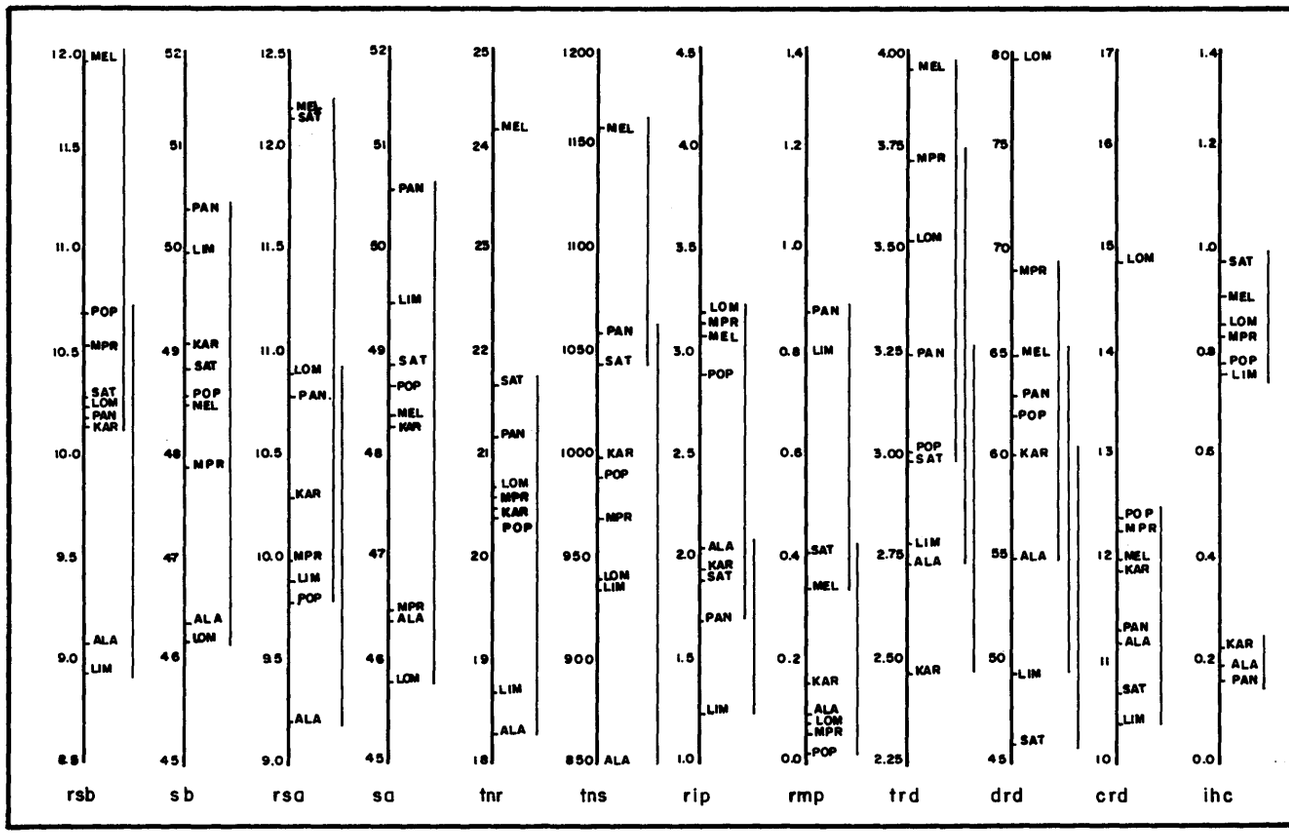

Figure 3. — Studentized range test for anatomical traits of the needles ($P < 0.05$).

that the high transpiration area exhibited by these two populations that have a long dry season is balanced by having few stomata and thick hypoderm, as can be observed in *Figure 3.* The high frequency of fascicles with more than three needles could be due to hybridisation with *P. oocarpa* SCHIEDE, a species which is in contact with *P. caribaea* in Limones and relatively near it in Santa Clara. *Figure 3* also shows that there are significant differences in terms of stomatal rows and total number of stomata, where Melinda provenance can be characterized by having the higher values and Alamicamba the lowest values. The average total number of rows was 20.7 and Melinda has 24.2, and Alamicamba 18.3. The average total number of stomata in 5 mm section was 994.4, and Melinda shows 1160 and Alamicamba 850 *(Table 4).* This source of variation could be the result of small gene modifications in the Southward migration of the variety.

MERGEN (1958a) found that, in *P. elliottii* var. *elliottii* LITTLE and DORMAN, the number of stomata per row can be used as a test to evaluate putative hybrids. He suggested that this trait was relatively independent of the environment.

Diameter of the resin ducts was the other trait with high genetic variation. The Lomitas population was isolated from the rest with resin ducts of 80.0 μ diameter compared with 60.9 μ as average for the nine populations

Table 4. — Means of morphological and anatomical characteristics of the needles.

variables / Provenances	nf	nl	sn	bcs	hcs	rsb	sb	rsa	sa	tsr	tns	rip	rmp	trd	did	crd	ihc
POP	3.06	23.01	19.68	1.74	0.84	10.64	48.60	9.78	48.72	20.40	989.04	2.92	0.02	3.09	62.20	12.40	0.78
MPR	2.92	22.21	22.62	1.63	0.94	10.54	47.92	10.04	46.50	20.60	969.12	3.12	0.07	3.70	68.78	12.28	0.84
LOM	2.94	21.21	23.34	1.90	0.85	10.24	46.20	10.90	45.80	20.74	938.38	3.18	0.08	3.56	79.76	14.88	0.84
ALA	3.04	22.03	19.48	1.35	0.77	9.10	46.36	9.20	46.44	18.30	845.54	2.09	0.08	2.73	54.90	11.24	0.18
KAR	3.02	21.96	23.64	1.50	0.80	10.14	49.06	10.34	48.32	20.48	1000.94	1.97	0.16	2.48	59.58	11.96	0.24
PAN	3.00	20.63	23.20	1.49	0.79	10.18	50.54	10.84	50.62	21.22	1063.06	1.73	0.87	3.25	63.02	14.30	0.14
MEL	3.04	19.35	23.48	1.65	0.88	11.96	48.48	12.22	48.12	24.16	1163.40	3.11	0.34	3.93	64.58	12.02	0.90
LIM	3.44	23.75	24.46	1.42	0.89	8.94	50.02	9.86	49.52	18.74	935.10	1.25	0.81	2.80	49.50	10.49	0.76
SAT	3.56	22.54	24.78	1.90	0.91	10.26	48.84	12.16	48.88	21.70	1045.42	1.90	0.41	2.99	46.24	10.72	0.98
$\bar{X}$	3.11	21.84	22.74	1.59	0.85	10.23	48.45	10.59	48.14	20.70	994.44	2.31	0.25	3.15	60.95	11.91	0.66
$S\bar{X}$	0.09	0.83	1.21	0.21	0.02	0.46	1.39	0.58	1.38	0.84	39.05	0.12	0.12	0.07	3.73	0.53	0.13
$S\bar{e}$	0.12	1.18	1.72	0.29	0.17	0.66	1.97	0.82	1.95	1.19	3.93	0.16	0.17	0.10	5.27	0.75	0.18
CV (%)	2.76	3.83	5.34	13.11	7.60	4.34	2.88	5.45	2.87	4.07	55.22	7.58	24.59	4.02	6.12	4.47	19.70

Table 5. — Multiple regression analysis.

Dependent variables	Constant	Independent variables regression coefficient			Multiple correlation coefficient (R)	Determination coefficient (R^2%)	Variance ratio (F)
		Latitude	Altitude	Rainfall			
nf	3.58	-0.042	0.0005	-	0.47	27	***
nl	18.89	-	0.0045	0.0008	0.32	10	*
sn	21.43	-	0.0025	0.0003	0.13	2	NS
bcs	- 0.91	0.192	0.0007	-	0.20	4	NS
hcs	0.28	0.029	0.0002	0.0001	0.32	27	***
rsb	4.11	0.371	-	0.0003	0.37	13	**
sb	51.49	-0.063	0.0026	-	0.20	4	NS
rsa	7.59	0.154	0.0007	0.0003	0.11	1	NS
sa	37.23	-0.494	0.1530	-0.0009	0.21	5	NS
tnr	13.15	0.456	-	0.0004	0.24	6	NS
tns	803.80	12.900	-	-	0.15	2	***
rip	- 1.67	0.701	0.0007	-	0.50	24	***
rmp	3.53	-0.127	-0.0008	-0.0005	0.42	18	***
tnr	1.33	0.187	-0.0008	-0.0003	0.41	17	**
drd	17.04	3.285	-0.0150	-	0.50	25	***
crd	6.98	0.362	-0.0020	-	0.60	36	***
ihc	- 1.78	0.154	0.0004	-	0.66	43	***

* significant at $P < 0.05$ of probability
** significant at $P < 0.01$ of probability
***significant at $P < 0.001$ of probability
NS not significant at $P < 0.05$ of probability

assessed *(Figure 3* and *Table 4).* In the present analysis it was found that the northern provenances plus Santa Clara and Limones were characterized by having a high foliar area *(Figure 3);* also they had 80% of their needles with intrusion of hypoderm cells into the chorenchyma as a way to control the transpiration rate. The three more tropical provenances from Nicaragua (Alamicamba, Karawala, and Patasma) were grouped together by the absence of intrusions.

The multiple regression analysis *(Table 5)* suggested that the populations from northern latitudes and high elevations had greater numbers of needles per fascicle, bigger diameters of the resin ducts, more cells around the ducts, and more intrusion of the hypoderm cells; the respective determination coefficients are 27, 25, 36 and 43%. The stomatal traits were not associated with the environment variables assessed as reported by MERGEN (1958a) in other species. In the case of the increase in needles per fascicle it is feasible to assume a gene flow from *P. oocarpa* forest.

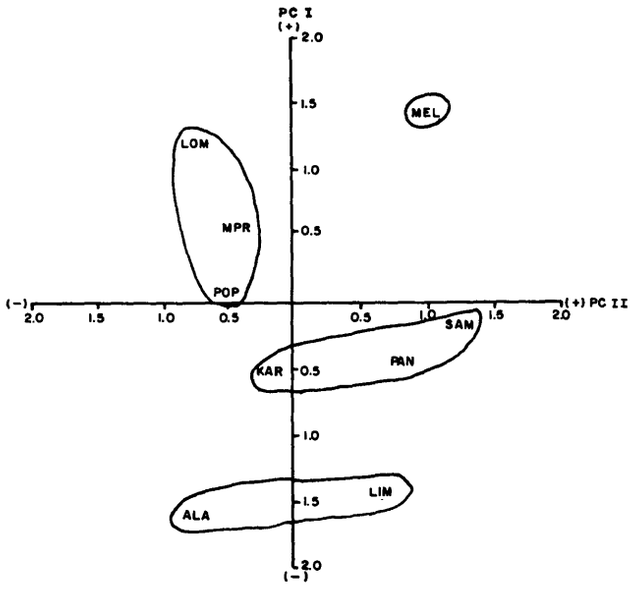

Figure 4. — Projection of the components I and II.

The principal component analysis detected that 83.2% of the total variation in the 17 traits was contained in the first three components. The first one accounted for 38.5% of the total variation and it was largely composed by number of resin ducts, resin ducts in internal position, diameter of the resin ducts, and stomatal rows in abaxial surface

Table 6. — Latent roots and percentage of variation in each component.

Components	Roots	Variation in percentage	
		simple	cumulative
1	6.54	38.45	38.45
2	5.03	29.58	68.03
3	2.58	15.19	83.22
4	1.05	6.17	89.40
5	0.88	5.18	94.58

(Tables 6 and *7);* for simple descriptive purposes this was called "resin ducts". The second one account for 29.6% of the variation and was called "stomatal density" because it was composed of most of the stomata traits. The combination of the two components *(Figure 4)* separated the nine populations into four groups mainly due to the component II, following a North-South tendency as emphasized previously. Again the Melinda population was separated by its high stomatal density.

Some 44.7% of the variation was represented by the combination of components I and III, the last one formed largely by the traits concerned with needle volume. Its graphic representation *(Figure 5)* confirmed the previous assessments, where Santa Clara and Limones are grouped together by having more needle volume, and Melinda is separated by having low needle volume.

The cluster analysis shows through the dendrogram *(Figure 6)* that the populations Poptun, Karawala, Mountain Pine Ridge, Lomitas, Pantasma and Santa Clara were similar with respect to the needle traits assessed. Only Melinda and Alamicamba were distinguished as distinct

Table 7. — Proportional weightings for each component.

Variables	Vector for components				
	1	2	3	4	5
nf	-0.524	0.763	0.820	0.091	-0.607
nl	-0.738	-0.195	0.876	0.460	0.381
sn	0.749	0.353	0.711	-0.734	-0.487
bcs	0.418	0.483	0.945	0.714	0.813
hcs	0.115	-0.957	0.402	-1.000	0.368
rsb	0.930	0.414	-0.383	0.681	-0.290
sb	-0.462	0.972	-0.383	0.251	0.893
rsa	0.625	0.955	0.072	-0.372	-0.837
sa	-0.484	0.976	-0.479	0.247	0.404
tnr	0.833	0.762	-0.330	0.200	-0.485
tns	0.544	1.000	-0.479	0.326	-0.209
rip	1.000	-0.475	0.016	0.547	0.096
rmp	-0.481	0.914	-0.263	-0.600	0.909
trd	0.964	0.181	-0.067	0.251	0.828
drd	0.835	-0.552	-0.300	-0.535	1.000
crd	0.818	-0.639	0.049	-0.651	0.397
ihc	0.672	0.233	1.000	0.318	0.386

populations as suggested by the univariate analysis where clear differences were observed in the variables related to stomata. However, they were not associated with the climatic variables *(Table 5)*; this must be considered ecotipic variation as a result of the natural selection.

Conclusions

The assessment of the needle anatomy and morphology in *P. caribaea* var. *hondurensis* showed that differences between provenances accounted for very little of the total variation. Most of the variation was concentrated in trees in provenances and in fascicles in trees.

It is feasible to assume that hybridisation with *P. oocarpa* could have taken place in the populations Limones and Santa Clara, because they presented a high frequency of fascicles with four and five needles. The Limones population showed a low number of stomata, a feature that could play an important role in reducing transpiration rate.

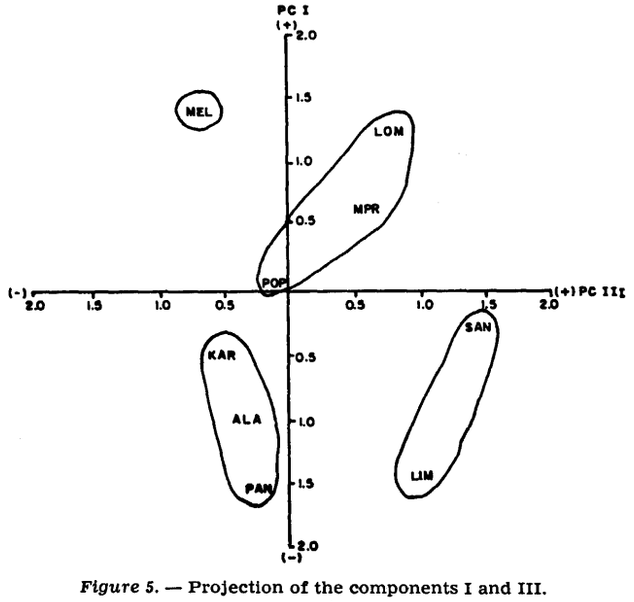

Figure 5. — Projection of the components I and III.

The Melinda population from the northern distribution had the highest values in terms of stomatal traits; it showed shorter needles maybe as a way to control the transpiration rate. The Alamicamba source had the lowest values with respect to stomata.

Most of the variables showed and ecotypic variation, with the exception of height of cross section and intrusion of hypoderm cells that showed a latitudinal clinal trend.

In this preliminary assessment most of the variation was found to be between fascicles; this suggests that the traits studied are highly sensitive to the environmental conditions and emphasizes the necessity of analysing comparable samples within the crown of the tree. Great attention should therefore be given to the sampling methods in future studies.

In general terms, this preliminary assessment of the needles has drawn attention to the variation showed by the population at Limones, Santa Clara, Melinda, and Alamicamba. More detailed study would be required to confirm these differences, and perhaps split these four populations from the rest.

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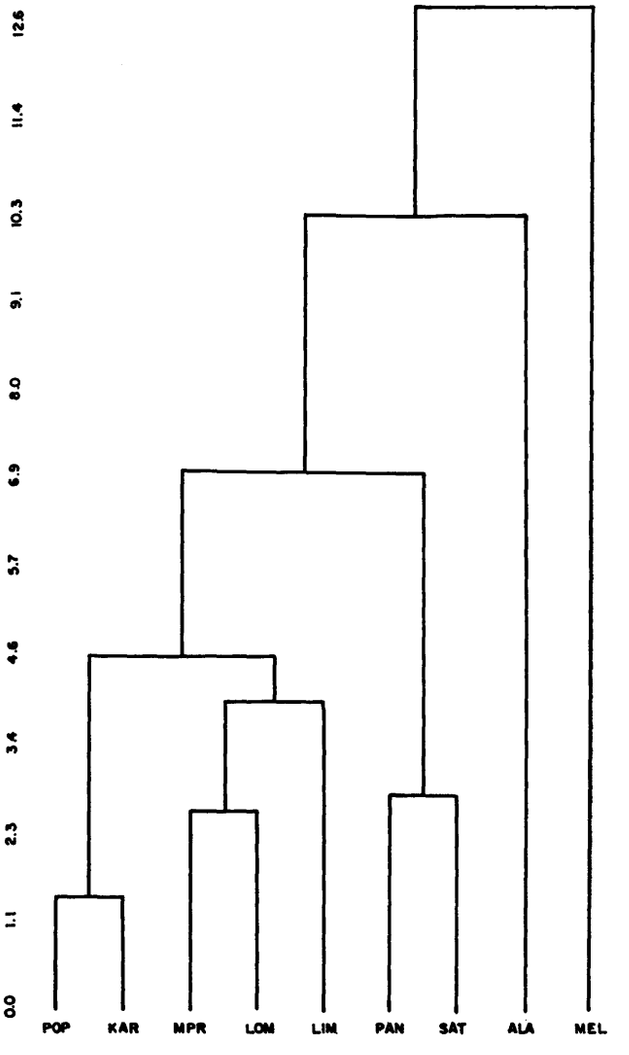

Figure 6. — Dendrogram showing the grouping pattern of provenances in terms of needle traits.

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Stand and Seed Source Variation in Peroxidase Isozymes of Quercus rubra L.[1])

By D. B. Houston[2])

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Summary

Variability in *Quercus rubra* foliar peroxidase isozymes was studied in 11 seed sources and 3 local stand collections. Analysis of isozyme frequency distributions by Berry's measure of distinctiveness, D^2, revealed significant differences for all but one (Michigan vs Pennsylvania) of the 55 pair-wise seed source comparisons. All sources could be tentatively differentiated from one another based on the presence or absence of one or more isozyme bands, or by large frequency differences between one or more isozymes.

Berry's D^2 also indicated the existence of significant differences in isozyme frequency distributions between trees (i. e. between four half-sib families) sampled in each of 3 Ohio red oak stands, as well as between stands.

Key words: northern red oak, isozymes, peroxidase, geographic variation.

Zusammenfassung

Bei 11 Provenienzen und 3 örtlichen Herkünften von *Quercus rubra* wurde die Variabilität der Blatt-Peroxydase-Isoenzyme untersucht. Eine Analyse der Häufigkeitsverteilung der Isoenzyme mit der Charakterisierungsmethode von Berry, D^2, zeigte signifikante Unterschiede für alle der 55 paarweisen Herkunftsvergleiche bis auf einen (Michigan gegenüber Pennsylvania). Alle Herkünfte konnten im Versuch voneinander durch das Vorhanden- oder Nichtvorhandensein eines oder mehrerer Enzymbänder oder die sehr unterschiedlichen Häufigkeiten bei einem oder mehreren Isoenzymen unterschieden werden.

Berry's D^2 zeigte außerdem die Existenz signifikanter Unterschiede in der Isoenzymhäufigkeitsverteilung sowohl zwischen Bäumen (z. B. zwischen 4 Halbgeschwisterfamilien), als Stichprobe aus je 3 *Quercus rubra* Beständen in Ohio, als auch zwischen Beständen, an.

Introduction

The analysis of genetic variation among and within forest tree populations traditionally has been approached by studying quantitative aspects of inheritance and natural variation for a wide variety of physiological and morphological characters (Libby *et al.* 1969). The application of electrophoretic techniques to such analyses overcomes many of the inherent disadvantages in traditional methods of characterizing genetic variability within and among tree populations by permitting precise determinations of allelic variation.

Many recent isozyme studies have focused on gymnosperms (e. g. Yeh and El-Kassaby 1980, Feret 1974, Mitton *et al.* 1977); considerably less work has been performed with hardwoods. Peroxidase inheritance patterns have been determined in *Ulmus* (Feret and Stairs 1971), *Populus* (Guzina 1978, Mitton and Grant 1980) and *Liriodendron* (Houston and Hood 1982). Kim (1979) has identified the mode of inheritance for leucine aminopeptidase and acid phosphatase isozymes in *Fagus*. Mitton and Grant (1980) also studied phosphohexose isomerase and glutamate dehydrogenase inheritance in *Populus*.

Developmental and inter-specific variability in several isozymes has also been reported for *Quercus* (Mayberry and Feret 1977, Tobolski 1978, Olsson 1975). Studies of intraspecific variation in enzyme systems of *Robinia* (Huang *et al.* 1977), *Betula* (Payne and Fairbrothers 1973), *Prunus* (Lewis and Cech 1969) and *Juglans* (Clarkson *et al.* 1974)

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[2]) Associate Professor, Dept. of Forestry, OARDC and OSU, Wooster, OH 44691, USA.