

FOREST TREE BREEDING RESEARCH IN THREE  
REGIONS OF THE UNITED STATES AND ITS  
APPLICABILITY TO WEST PAKISTAN CONDITIONS

Thesis for the Degree of M. S.

MICHIGAN STATE UNIVERSITY

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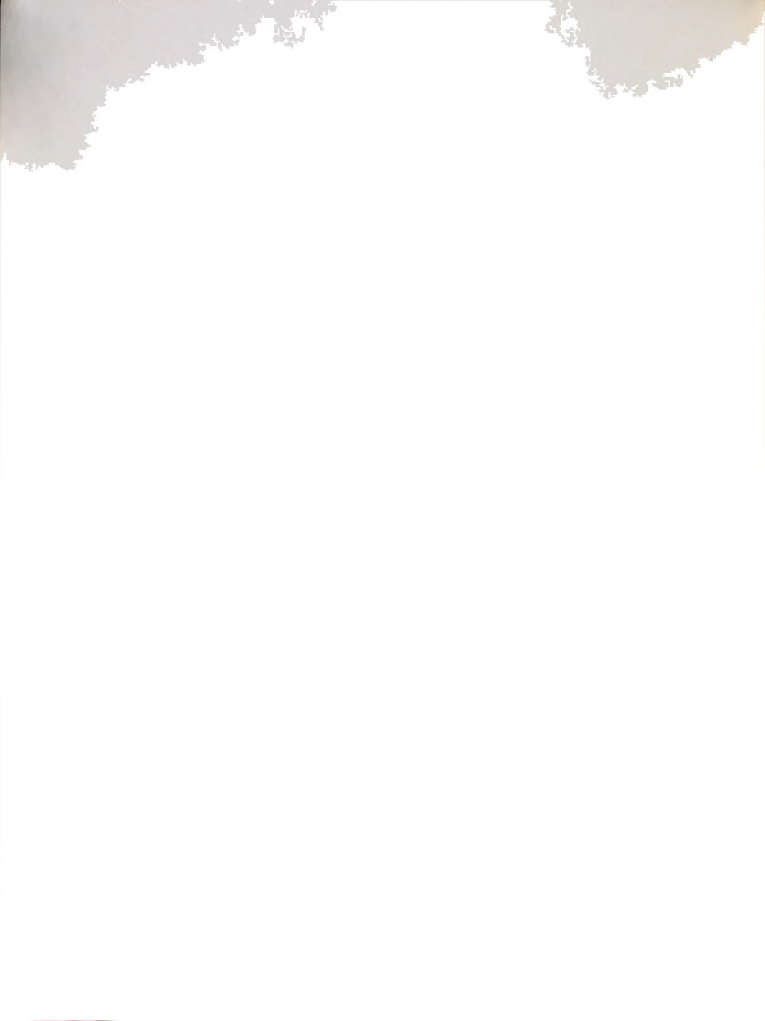
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## ABSTRACT

### FOREST TREE BREEDING RESEARCH IN THREE REGIONS OF THE UNITED STATES AND ITS APPLICABILITY TO WEST PAKISTAN CONDITIONS

by Javaid A. Ahsan

The United States is a country with diversified local features. The tree breeding methods adopted in the three regions represented by Michigan State University, East Lansing, Michigan; Institute of Forest Genetics, Placerville, California; and Institute of Forest Genetics, Gulfport, Mississippi, to a great extent, have been influenced by local factors.

Michigan State University has an active 10-year-old research and teaching program. There are three major objectives to its provenance testing work: determination of variation patterns in conifers and hardwoods, identification of desirable ecotypes for planting in specified areas, and at the same time providing material for further breeding operations. Another set of studies is designed to learn the inheritance of economic traits and select good parents for breeding, determine local variation patterns and establish seed orchards; the objective is being accomplished through 1-parent progeny tests which are converted to seed orchards by thinning poor individuals





Javaid A. Ahsan

and poor families. Allied problems, for example, determination of evolutionary and physiological factors responsible for variation, juvenile-mature relationships, are also studied. Much of the research is conducted in collaboration with other state experiment stations in the north central states, under the NC-51 project. A unique feature of the Michigan program is the emphasis on experimental methodology, aimed at making the work more efficient. The improvements obtained pertain to record systems, measurement philosophy, experimental layouts, planting methods, and nursery culture. The graduate teaching program utilizes segments of ongoing staff projects as dissertation topics, and involves genetics, taxonomy, and physiology.

At the Institute of Forest Genetics, Placerville, California, provenance and progeny tests were the first types of tree breeding studies. Fifteen years after its establishment, in 1940, the Institute's program switched to hybridization studies among the various pine species of the world. Reconnaissance of pine hybridization through crossability studies, has remained the major line of work. Economic potentialities of various hybrids and means to mass produce the desirable hybrids, are also studied. One of the major achievements of the Institute, during the past years, is the perfection of controlled pollination



techniques. In addition, some very useful hybrids have been made.

General objectives of tree breeding research at the Institute of Forest Genetics, Gulfport, have been laid down as "To determine the degree of genetic control of phenotypic characters of important southern pines and hardwoods and to develop methods of applying this control for the production of improved forest trees." Studies of variation through provenance tests and progeny tests, and also the intra- and interspecific hybridization, all contribute to the breeding program carried out at the Institute. A number of studies related to different aspects of tree breeding are thus underway. At Gulfport, generally more sophisticated types of experimental designs, that is, compact family block and lattice designs, are used.

West Pakistan consists mainly of arid alluvial plains with bushy vegetation, where irrigated plantations of hardwood species are raised artificially; and the mountaineous northern Himalayan region where coniferous species grow naturally. Important species of West Pakistan which need immediate improvement are Pinus griffithii, P. longifolia, Dalbergia sissoo, Acacia arabica and Populus euphratica. Cooperative provenance tests, such as those underway at the Michigan station

mechanisms. In addition, some very recent studies have  
been made on the effect of the environment on the  
development of the nervous system. It is now generally  
accepted that the environment has a profound influence  
on the development of the nervous system. The degree of  
genetic control of the development of the nervous system  
has been the subject of much discussion in the past.

For the purpose of this study, the following  
method was used. The subjects were divided into two  
groups. The first group consisted of 100 subjects  
who were given a standard test. The second group  
consisted of 100 subjects who were given a test  
which was similar to the first, but which was  
designed to measure a different aspect of the  
subject's ability. The results of the tests were  
compared and the differences between the two  
groups were noted. It was found that the subjects  
in the second group performed significantly better  
than those in the first group. This result  
suggests that the environment has a significant  
influence on the development of the nervous system.

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with the cooperation of NC-51 will deserve top priority, and should involve nursery experiments, permanent field plantations, and determination of juvenile-mature correlations. These should be followed, within a few years, by open-pollinated progeny test-seed orchards designed to determine the variability pattern within local populations and also obtain a modest amount of genetic improvement of actual planting stock. These will follow the Michigan pattern. At a much later date it will be desirable to follow the 2-parent progeny test designs used at Gulfport. The pine crossability patterns worked out at Placerville will be used to establish breeding arboreta consisting of the species most likely to cross with the two native pines, and pine hybridization will for the near future be limited to production and performance of hybrids involving those two species.





FOREST TREE BREEDING RESEARCH IN THREE REGIONS  
OF THE UNITED STATES AND ITS APPLICABILITY  
TO WEST PAKISTAN CONDITIONS

By

Javaid A. Ahsan, 1940-

A THESIS

Submitted to  
Michigan State University  
in partial fulfillment of the requirements  
for the degree of

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CHAPTER I  
INTRODUCTION





## CHAPTER I

### INTRODUCTION

This thesis is the outcome of the author's year long stay at Michigan State University as a student of Dr. Jonathan Wright, and a three month summer trip to the Institute of Forest Genetics at Placerville, California, and Gulfport, Mississippi, during 1966.

In a vast country like the United States there is a wide range of climates, geographical and botanical features, and public needs. Choice of forest tree breeding approach in a particular region is governed in large measure by local factors. The three breeding stations involved in this study, Michigan State University, Institute of Forest Genetics Placerville, and Institute of Forest Genetics Gulfport, thus have different approaches.

With the expanding field of forest genetics and the dynamic advancement in the related sciences, there is constant change in tree breeding methods. Therefore in the present study I tried to learn the best methods for present conditions as well as the processes by which these methods change.



The three research stations have engaged in breeding activities for many years. In order to determine how their experience can help implement tree breeding in Pakistan, an appraisal was made of the techniques employed at each center and then an attempt was made to evaluate the adaptability of each method to conditions prevailing in Pakistan.

Chapters II, III, and IV deal with the objectives and methods of tree breeding followed at each of the three research stations in the United States.

Chapter V embodies a description of the forest conditions in West Pakistan, necessity and objectives of tree improvement, and possible approaches to the problems in the light of breeding principles pursued at the three American centers of tree breeding.



CHAPTER II  
TREE BREEDING AT MICHIGAN  
STATE UNIVERSITY





## CHAPTER II

### TREE BREEDING AT MICHIGAN STATE UNIVERSITY

The history of forest genetics at Michigan State University is divisible into two phases, prior to 1957 and after 1957.

The earlier phase dates back to a time before forest tree breeding was a recognized branch of science. During the earlier years, the only aspect of forest genetics emphasized at Michigan State University was the introduction of exotic species. Professor W. J. Beal and, later, Professor E. E. Bogue, hoped to find species that would produce good wood quality and at the same time grow as fast or faster than the native species. Bogue started a forest nursery in 1904 from which western pines and firs as well as eastern white, jack and red pines and white spruce were sold for forest planting. He also started an arboretum for his first trials at East Lansing with the hope that progeny tests would follow. His experiments included many pine species: P. ponderosa, P. scopulorum, P. jeffreyi, P. flexilis, P. rigida, and P. strobus. Other conifers such as, Picea abies, P. engelmanni, and Larix decidua were also grown.



In 1917, Professor Chittenden established a trial plantation of Populus canadensis and P.deltoides. Introduction and trials of various varieties of poplars and hybrids continued through the next four decades. In the meantime, trials of exotic conifers were initiated at the W. K. Kellogg Forest, in the early 1930's. Ponderosa, Scotch,lodgepole, Austrian and Japanese red pine were established in forest plantations each of one acre or more. No tests on racial variation were established before 1957.

In 1957 Prof. Jonathan Wright joined the faculty of Michigan State University as a member of the forestry department. Systematic efforts were started to improve the trees of the region through the application of genetics. The tree improvement work was speeded up after participation of Michigan State University in the regional NC-51 project in 1960. NC-51 is a cooperative project, carried out by ten tree breeding agencies from different North Central states, with the financial assistance from the federal government. It is titled "Tree Improvement through Selection and Breeding of Forest Trees of Known Origin." An introduction of the project with special reference to Michigan State University is given in Appendix I.

IN THE DISTRICT COURT OF THE UNITED STATES FOR THE DISTRICT OF COLUMBIA

Case No. 100-100000-0000

JOHN DOE, Plaintiff,

vs.

JANE SMITH, Defendant.

JOHN DOE, Plaintiff, by and through the undersigned, do hereby certify that the within and foregoing is a true and correct copy of the original as the same appears in the files of the Court.

Witness my hand and seal of office this 1st day of January, 1900.

\_\_\_\_\_  
Clerk of the Court

Forests of Michigan.--Natural vegetation types in Michigan are spruce-fir, beech-maple-basswood-yellow birch-hemlock, jack pine-red pine-white pine, and aspen. Total annual precipitation averages about 30 inches of which 56 percent falls during the warm months, May through September. Irregular drought periods of 10 to 30 days, with little or no rain, are characteristic during the growing season. The mean annual temperature for the whole Lake States Region is about 44<sup>o</sup>F. Maxima of 90<sup>o</sup> to 100<sup>o</sup>F. are recorded throughout the region almost every summer. Minima below 0<sup>o</sup>F. are recorded every winter and have gone as low as -49<sup>o</sup>F. in the upper peninsula. Topography is typically level to gently rolling. Soils range from dry sandy soils occupied by jack pine or oaks, to loams with good conditions of moisture and fertility.

Unchecked forest fires in the wake of logging operations were an important factor responsible for the deterioration of growth and depletion of soil fertility. Aspen, jack pine and scrub-oak have characteristically invaded vast areas cut-over and burned lands. The forested area of the Lake States was 104 million acres at the time the first white men came. This area has been reduced to 56 million acres, of which one-fourth is barren or not restocking. A survey of the Michigan's forest resources reveals that natural mortality accounts



for more than one third of the total annual removal. Diseases are the greatest single cause of losses. Red pine, which is Michigan's most planted tree is attacked heavily by the European shoot moth.

The biggest job ahead is to improve the state's forests by reforesting the poor forest areas with the proper tree species which can exploit site quality to the maximum. In the following account, the role of Michigan State University will be evaluated in this task.

Major Objectives of Breeding and  
Lines of Work

The objectives of tree-breeding at Michigan State University may be summarized as follows:

- i. For important tree species to determine the pattern of variation and the best sources for planting in specified localities; to determine the evolutionary and physiological factors responsible for variation; and to furnish material for future breeding work (this objective is common with that of NC-51).
- ii. To learn the inheritance of economic traits, select good parents for breeding, determine local variation patterns and establish seed orchards.

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The first objective is being achieved by provenance studies. The preliminary results indicate the acceptable varieties. However, in order to obtain more concrete results, the studies are to be continued for the next several decades. At present most of the experiments are in various stages of establishment. They must be followed because every year the information becomes more valuable.

Very few hardwoods have been included in the studies of racial variation. A major hurdle has been the lack of satisfactory methodology which has so well been worked out for conifers. Difficulty of storing seeds from one year to the next (e.g. in red oak, cottonwood, and sugar maple); difficulty of planting and the need for intensive weed control, have been major problems.

Work on the physiological aspects of racial variation has started recently. It will involve chemical and other detailed analyses of material collected in well established outplantings. Such studies are inherently expensive and require specialized equipment and personnel. These conditions were met in the past three years.

Material for future breeding work will be furnished by selected races. By crossing selected races



within species and amongst different species, real improvement may be obtained. Nevertheless, selection of best races and subsequent improvement based upon them does not in any way exhaust all the possibilities of tree improvement. A second set of objectives therefore, envisages learning the local variation and inheritance of desirable characters, and selecting the best individuals for breeding and establishing seed orchards.

In order to aid the achievement of the major objectives, research is needed on many allied problems. One of the important problems is the long time required in breeding work. It can be attacked by determining the juvenile-mature correlations and by finding out the ways to induce early flowering. Research on juvenile-mature correlations has been started under NC-51 project in that all experiments have been measured when one and two year old and at intervals after the trees have been transferred to their permanent locations.

Relationship of the Breeding Approach  
to Local Conditions

Important local factors which have led to the choice of provenance studies as the main breeding method in Michigan State University, are discussed briefly.

SECRETARY OF THE INTERIOR

Washington, D. C.

Dear Sir:

I have the honor to acknowledge the receipt of your letter of the 10th inst. in relation to the above matter.

The Bureau is currently reviewing the same and will advise you as soon as a final decision has been reached.

Very respectfully,  
 [Signature]

Very truly yours,  
 [Signature]

Enclosed for you are the following documents:

- 1. Copy of the report of the field office.
- 2. Copy of the correspondence.
- 3. Copy of the Bureau's decision.

I am, Sir, very truly,  
 Yours,  
 [Signature]

Very truly yours,  
 [Signature]

Very truly yours,  
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Very truly yours,  
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Very truly yours,  
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Very truly yours,  
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Very truly yours,  
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Climate and edaphic factors.--For an efficient provenance or progeny test, climate and soil favorable for growth are necessary. Low mortality is of particular importance. Experience indicates that 85 percent survival is desirable in all tests. Another requirement is that the area should be uniform. Thus level or rolling topography is more desirable than a hilly site. Michigan and other areas of the North Central states, satisfy such requirements. In the prevailing climate, with adequate care, initial mortality can be kept below 5 percent.

Planting practices.--Mention has already been made of the large scale reforestation needs in the region. A start toward this end has been made. By the end of 1948 the total area planted in the Lake States was about 1,500,000 acres. Considering all North Central states, at the end of 1962, 3,907,000 of forest and 951,000 acres of tree windbreak plantings had been established. During one year, 1962, 161,000 acres of forest land and 28,000 acres of wind breaks were planted within the region. The greatest challenge before the tree breeder of this area is to allow the planting of only superior varieties of both native and exotic trees, lest the mistake once committed in planting inferior varieties of Scotch pine is repeated. Until improved varieties are available for massive planting programs, it will be worthwhile to plant



the best natural races. Selection of races and individual trees constitute this preliminary phase of tree improvement in the region.

Christmas trees.--Michigan is second to Washington in Christmas tree production. Provenances which satisfy the requirements of good Christmas trees are therefore of much value. Growers are already making use of the 8-year data on Scotch pine and white spruce, buying seed from regions which yield trees with acceptable fall coloration and growth rate.

Participation of Michigan State University in NC-51 project.--Michigan State University is an active member of NC-51 project whose main part is provenance research, or testing of trees from different parts of species' natural range. Tree breeding research in the University should obviously be concentrated on this field.

Arboretum facilities.--Michigan State University campus is itself a large arboretum. The collections, while growing, are still deficient in the conifers. There are smaller arboreta in nearby Battle Creek and Ann Arbor. Interspecific hybridization will await the development of better collections.

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Salient Features of the Methodology  
at Michigan State University

In executing the tree breeding work, foremost considerations kept in view at Michigan are: reducing the amount of work without appreciably affecting the accuracy of results, lowering the cost of the experiments to the minimum, and simplification of methods. Consequently, allied research is carried out to find out ways and means to achieve these goals. Methods have been developed and certain rules laid down to be followed at every stage of the experiments. Distinctive features of the methodology are given below:

Nursery establishment and lifting.--In order to make full use of tractor driven mechanical equipment, trees, seedlings and transplants are grown in 43-inch-wide nursery beds which are prepared, sprayed and lifted by machine. All mapping is done in terms of distance rather than sequence. To re-establish the plot identities, if nursery stakes are removed, measurement with a tape is performed. A nursery map is always prepared before actual sowing.

A randomized complete block design is generally used. Seedlots are allotted to numbered packets which are randomized within replicates and the random order recorded on the nursery map. The randomization is done



by shuffling seed packets rather than by tables of random numbers.

Masking tape field labels, inscribed with a black China-marking pencil are used. Labels are attached close to the top of trees, with right side up, so as to be read standing up.

A mechanical lifter is used for loosening the trees. With the aid of a master list, markers are placed in the aisle along each seedlot to be included in a plantation, and seedlots are marked for pulling. Pulling starts at the corner diagonally opposite the plot stake which is not disturbed until the last tree is out. Each seedlot is tied separately. Each seedlot bundle contains some labelled trees.

Seedlings are transported to the planting shed. Then, in each seedlot labels are added to make the total number equal to the number of replicates. Each seedlot is then separated into 4-tree plot bundles each of which contains a labelled tree. Roots of seedlings in plot bundles are dipped in water to save them from drying out during handling. Each plot bundle is dropped into a separate pile representing a replicate. Plot bundles of each replicate are packed separately and labelled as 1, 2, 3 etc.

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random samples

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Weed control.--For planting, uniform sites of average to above-average quality, free from any shrub or tree growth, are chosen. They are prepared by plowing furrows to eradicate all the weeds well before the planting is carried out. Or weed-free strips are prepared by spraying dalapon or amino-triazole six months ahead of planting time. Simazine or simazine plus amino-triazole are used subsequent to planting. 2,4,5,-T is also used for killing the brush.

Planting.--Standard planting technique is used while planting, taking care to place root crown about 1 inch below the soil surface and to firm the soil about the roots. No planting is done within forty feet of existing tall timber.

Approximate replicate and plantation boundaries are marked by temporary stakes in a manner designed to make within replicate variation as small as possible. In aligning the plots, care is exercised to see that they follow obvious site gradients. All the plots are kept in the same direction, with straight rows. Square or approximately square replicates with regular outlines (except one or two on the border line of the plantation) are usually used.

A border row consisting of the same species is planted around the entire planting after all the



experimental trees have been planted. Identity of all the border rows is maintained.

Plot bundles are taken out from the numbered replicate bundles and planting is done in a random order within a replicate. The labelled tree is planted in the same relative position in each plot. In order to avoid confusion if a packer places less or more than the proper number of trees per bundle, a space is left unfilled or the excessive tree discarded. In case the labelled tree is not planted in its proper position it is replanted. If two labels are found in one plot bundle, one is torn off.

One of the outstanding developments made at Michigan State University in reducing the time and labor of planting, is the perfection of machine planting technique. It has been successfully applied in Scotch pine and red pine. In machine planting, it is essential that the replicates should have nearly regular boundaries, with their long direction in the direction of tractor travel. A base line is established with stakes at accurate distances along one edge of the plantation. Assume there are three replicates lying adjacent to one another in sequence 1, 2, 3, each having eight 4-tree plots, sixteen 4-tree plot bundles will be required to plant up one replicate. Sixteen plot bundles for each replicate will be placed on the tractor in the sequence

His is a very old and famous name in the history of the

country and he has been a member of the

House of Representatives for many years. He has been a member of the

Senate and has been a member of the Cabinet. He has been a member of the

Supreme Court and has been a member of the

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1, 2, 3. While the tractor travels, a walker will untie the 4-tree bundles and hand them to the tree planter. The planter will plant the 8 plots in replicate 1, then the 8 plots in replicate 2, etc. The walker will check the replicate boundaries to insure placement of the plots in correct order. At the end of the third replicate, the tractor will travel in the reverse direction, hence the replicates will be planted in the reverse sequence, that is, 3, 2, 1. Distances between adjacent plants are maintained uniform by planter, by observing the base line or the previously planted rows.

Mapping.--In the plantation maps, a row is always denoted by a number and a column by a letter. Only one entry per plot is made. A missing plot is indicated by a double dash (--) and plots of unknown identity by a question mark (?).

Field copies of the plantation map are prepared as the planting progresses or soon after it is completed. Then as soon as possible, 125 permanent copies are prepared by cutting mimeographed stencils. A copy of each map is distributed to all interested individuals.

Each map (on 8 x 10-inch paper) is a complete report, showing not only the location of the plantation and individual plots, but also giving all the details regarding establishment, weather at the time of planting;

While the tractor travels, a wheel will strike  
the ground which will cause the wheel to rise and  
fall. This will cause the tractor to vibrate and  
the engine to run unevenly. This will cause the  
engine to overheat and the tractor to stop.  
To prevent this, the tractor should be checked  
before it is used. The engine oil should be  
checked and the tractor should be serviced  
regularly. The tractor should be used on  
soft ground and the operator should wear  
seat belts. The tractor should be used  
carefully and the operator should be trained.

topography; soil type and soil condition; experimental design; plot size and direction; number of varieties; number of replicates and other pertinent information.

Measurement.--To ensure rapidity of work and accuracy, one or a few traits are measured at a time. Metric traits (height, diameter, etc.) are measured to an accuracy of 1/20th of the range between the extremes. Presence-absence traits are measured in terms of the number of trees per plot. Non-metrical traits such as color, are scored according to a system of numbered grades. In all the measurement work, only the plot means are recorded. This device saves considerable time.

Data recording.--There is a record folder maintained for every plantation. It is duplicated by carbon after the planting is over. One copy remains with the experimenter and the other with the ground man. Either can measure and record in his folder and transmit the carbon copy of the record to the other. Such an arrangement provides a complete and up-to-date record system in the two folders, all the time.

A record folder comprises five parts: plantation map, pages of accession record, a set of measurement instructions, field data sheets, and empty pages on which to record descriptions of measurements and appropriate notes regarding operations.



In addition to the data and other observations, results of various statistical analyses are placed in the folder under appropriate characters.

Experimental design.--Theory and practice of plot size to determine a suitable design for various experiments has been a subject of intensive investigation at Michigan station. It was found that for greatest efficiency 4-tree plots seem best for provenance studies and 8 to 10-tree linear plots, for 1-parent progeny tests.

A randomized complete block design has most commonly been used in various tests. The main advantage of this design is that pre-location of plots is not required. Moreover, it is far simpler to analyze statistically than more sophisticated designs.

Where the mortality is lower than five percent, instead of going into the lengthy, text-book procedure of calculating the values for missing plots, a short-cut is used. The mean value for the seedlot is entered and error degrees of freedom reduced by one correspondingly. The value of a missing plot can also be entered as the value of same plot in other replicates.



### Typical Studies

Being the main line of work, investigations on provenances, constitute the largest number of studies at the Michigan State University station. Important species involved are: Scotch pine, ponderosa pine, European black pine, eastern white pine, virginia pine, red pine, Japanese larch, white spruce, Douglas-fir, and white fir. Two typical studies, those of Scotch and eastern white pines, are described in detail.

Scotch pine.--The study consists of two phases. The nursery phase was accomplished wholly by Michigan State University. In the plantation phase, 31 permanent plantations were established in eight north Central states, including Michigan. The Michigan station pooled results and statistical analyses for measurements of all plantations through 7 years of age.

The two year nursery test was reported by Wright and Bull (1963). Seed was procured from 108 native and 13 planted stands of Scotch pine, from 21 countries of Europe and Eurasia. Each stand collection comprised seed from ten or more average trees from an area of several acres. Origin data and other pertinent information (about locality, altitude, date of collection, area, soil, age, height, diameter, etc.) was obtained with each collection.

INTERNAL SECURITY

Being the main line of work, investigation of  
provenance, ownership, etc. of the records of the  
the Michigan State University system. Important divisions  
involved are: General Plans, Research Plans, Economic  
Plans, Plans, extension work, etc., etc. etc. etc.  
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Sowing was done in sandy loam, highly fertile soil of Bogue Nursery in southern Michigan, in early May 1959. The nursery soil had already been treated with a fungicide-weed seed disinfectant (methyl bromide), the previous autumn. Sowing consisted of placement of row stakes and seed packets, checking packet identity against nursery map, hand planting the seeds 1/2 inch deep, in rows exactly six inches apart, sprinkling 1/2 inch of sand over the seeds, and covering the seedbeds with wire-screen and lath shades. A randomized complete block design, with four replicates, was used. Each plot representing one origin, consisted of one 46-inch row perpendicular to the long axis of the 500-foot seedbeds. Trees destined for outplanting were sown in a fifth, unmeasured replicate with each seedlot sown in a rectangular plot wherein the seeds were sown broadcast.

Germination was completed in three weeks after sowing. Watering was frequently done. A weed-free condition (with the applications of Stoddard solvent) was maintained. Mulching was done by a 1/2 inch saw dust layer applied before winter.

Measurements were made at intervals during the first three years. The measurements included all visible and differing traits. General rules for the measurement work have already been indicated.



Major geographical trends were found out in all the important characteristics. Trends for some of the traits are reproduced below:

Three-year height: Lowest, 12 cm. (northern Finland) to the highest, 71 cm. (Belgium). In central Europe, the trend was found reversed and southern origins were 50 cm. tall.

Summer foliage color: Medium green (central Europe) to dark green (northern Scandinavia and south-eastern Europe) to blue green (western Europe).

Autumn foliage color: Very yellow (Ural mountains) to yellow (Baltic countries) to dark green (south-eastern Europe) to blue green (western Europe).

Strength of root system expressed as ease of pulling at the age of six weeks: Easy (Germany, Belgium, Czechoslovakia) to difficult (northern Scandinavia, southern Europe).

Type of root system at age 3: Shallow and fibrous (northern Scandinavia, Siberia) to long and tap rooted (Spain).

Earliness of formation of secondary needles: Earliest (slow growing northern progenies), latest, early second year (progenies from Germany southwards).

Time of first year bud set: Mid-July (northern Finland) to early October (Spain).



April 1961 marked the beginning of the permanent phase of the Scotch pine study. Wright et. al. (1966) have given a full account of the commencement of this phase.

Trees from the unmeasured replicate at the Bogue Nursery, were lifted mechanically. After lifting, every fourth tree was labelled with masking tape and 4-tree bundles were tied, from which replicate bundles were made. Packing was done with tree roots in moist sphagnum moss and heavy water-proof paper wrapping. Planting material was shipped by surface carriers.

Experimental design of all the plantations was a randomized complete block design. In the Michigan plantations 10 replicates were made; in others the number of replicates varied from 4 to 10 per plantation. Except for Illinois, where two trees per plot were planted, 4-tree plots were used. Plots were in the form of rows. One or two rows border each plantation. Spacing was 7 x 7 or 8 x 8 feet, except in Nebraska, where 7 x 14 feet was maintained. From 60 to 108 different origins were represented in most plantations. Mortality ranged from 3 to 80 percent; it was lowest for varieties from Spain, southern France, and southern Scandinavia; highest for varieties from central Europe and the far north.

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After subjecting the data to the statistical analysis, the following conclusions, with regard to the intervarietal differences, could be drawn:

- Winter foliage color: Very yellow--Scandinavian and Siberian varieties; Green--varieties from Spain, southern France, the Balkans and Asia minor; Intermediate--central European varieties.
- Needle length: Longest--central European varieties; Shortest--south European varieties.
- Winter injury: Spanish variety (iberica) was prone to winter damage at seven out of sixteen plantations.
- Height at 5, 6 and 7 years (measured at 16 sites in 5 states): Fastest--var. haguenensis (Belgium, north France, Western Germany); var. east Anglia (England); var. hercynica (Germany, Czechoslovakia). Slowest--var. lapponica (northern Scandinavia); var. mongolica (Mongolia, eastern Siberia) and altaica (Altai Mountains of Siberia).
- Susceptibility to insect attack: Most resistant to European pine sawfly--var. uralensis (results from four different sites over three years) to white-pine weevil--var. iberica (two years infestation of one plantation).

1. The first part of the document is a list of names and addresses of the members of the committee. The names are listed in alphabetical order, and the addresses are given in full. The list includes names such as Mr. J. H. Smith, Mr. W. B. Jones, and Mr. C. D. Brown, among others.

2. The second part of the document is a report on the work of the committee during the past year. It describes the various projects and activities that have been undertaken, and the progress that has been made. The report is written in a clear and concise style, and is intended to provide a comprehensive overview of the committee's work.

3. The third part of the document is a list of recommendations for the future. These recommendations are based on the findings of the committee's work, and are intended to guide the organization in its future activities. The recommendations cover a wide range of areas, including financial matters, personnel, and general administration.

4. The fourth part of the document is a list of names and addresses of the members of the committee for the next year. This list is similar to the one in the first part of the document, and is intended to provide a clear record of the committee's membership for the coming year.

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Eastern white pine.--Six years results on the geographic variation of this species have been reported by Wright et. al. (1963). Collection of seed from 31 stands within the species' natural range was accomplished in the fall of 1956. Michigan State University received two-year-old seedlings of fifteen origins from the North-eastern Forest Experiment Station in the spring of 1959. They were grown in transplant beds for one year at East Lansing.

The 2-1 stock was planted in two permanent field tests in the spring of 1960, in a 10-replicated, four-tree-per-plot, randomized complete block design, with a spacing of 8 x 8 feet. The two outplantings are located at W. K. Kellogg Forest, Augusta, Michigan, and Fred Russ Forest in Cass County, Southwestern Michigan. The trees were slit planted. Cultural treatments in the Kellogg plantation consisted of spraying with simazine in 2-foot strips between the rows immediately after planting. Amitrol-T and simazine were applied during the second and third years. In the Russ plantation where the soil was a dense sod on a heavy clay, mulching was done around each tree in two foot square and the entire planting area was kept mowed.

Analysis of data at six years revealed that mortality had been fairly low, but had been the greatest for the very slow growing Minnesota and Nova Scotia



origins and the very fast growing southern Appalachian origins. The Tennessee origin attained the greatest height (84 cm.); the difference was significant from all but two origins. Next fastest growing origins were Georgia, Pennsylvania, southern Ontario, Massachusetts and the Catskill Mountains of New York. Foliage color differences were not significant from one another. Time of leaf fall was found significantly different among different sources.

Studies on physiological variation.--Ponderosa, virginia, eastern white, Austrian, jack, European black and Scotch pine provenances have been under investigation to determine the underlying physiological mechanism behind external growth characters. Leaves or twigs from existing provenance tests are subjected to chemical analysis. In this connection the gas chromatograph (for volatile terpenes) and mass spectrograph (for mineral elements in the foliage) are used. Also, growth-chambers are used to subject different races to known photoperiod, temperature, and light intensity regimes.

From what has been accomplished, it is evident that in nearly all of the 12 elements (N, K, P, Na, Ca, Mg, Mn, Fe, Cu, B, Zn and Al), there are strong genetic differences within Scotch and European black pines. It has been further indicated, for ponderosa pine, that coastal origins have much higher contents of N, P, K, Ca,



Mg, Mn, and B than do the hardier interior origins. In virginia pine, two southern origins which have suffered repeated winter-kill, differ from all others in a number of elements.

Scotch pine varieties with different degrees of winter yellowing were kept at different night and day temperatures during the spring. The results showed that the trees changed from winter-yellow to summer-green following a 3-day period during which the day-time temperatures rose to 60°F. Night temperatures seemed unimportant. Light was found necessary for the change. (White and Wright, 1966.)

Half-sib progeny tests.--Important species on which 1-parent progeny tests are being conducted are red pine, jack pine, eastern white pine and Scotch pine.

Work on the half-sib progeny tests was started in 1960 and a new species has been added about every two years. From 125 to 450 progenies are included in each experiment, which is first run through a replicated nursery test then planted at three to six locations in the state. The parents in some species (e.g. red pine) have been randomly selected and in others (e.g. jack pine) a very careful selection for the most important traits has been made. All except the jack pine experiment have been outplanted successfully with survival of



85 to 90 percent. Jack pine is still in the nursery stage. Early height measurements indicate that most of the variation is associated with stand rather than mother-tree within the stand. Differences in other traits have not been large when the seed was collected from a limited area such as southern Michigan (U.S.F.S. 1966).

Half-sib progeny tests are aimed at collecting heritability data regarding the important traits and subsequent development of seedling seed orchards, depending upon the magnitude of inheritance. Selective breeding over a number of generations leading to the formation of desired genotypes in the seed orchards, has been planned for some species (e.g., jack pine); in others, as in red pine, the seed orchards are intended to be from open pollinated progeny.

The jack pine progeny test and seed orchard project can be described in detail. The project was started in 1965 when parental selection of 450 superior trees was made from natural stands. Superiority of these trees was judged on branch fineness, stem straightness, and height growth. Special emphasis was placed, however, on favoring trees having non-serotinous cones. Selection intensity was such that each selected tree was the best of 100 to 500 dominant trees growing under more or less uniform conditions in an area of less than five acres.

85 on 90 percent. The fact that the total number  
 of eggs is large and that the eggs are laid  
 in a regular pattern is evidence that the  
 birds are nesting in the area. The birds  
 have been seen nesting in the area since  
 the first of the season. The birds are  
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Data on parental trees was recorded on specially prepared forms and included the following information:

Location of stand to nearest 40 acres.

Age, average height of dominant, stocking and soil type of parent stand.

Average stem straightness and branch size of parent stand, recorded as, below average, average, or above average for the county.

Superiority of selected over neighbouring comparable trees with respect to stem straightness, recorded as +50 if the 'plus tree' was best of 50, +70, if the tree was best of 70, and so on.

Superiority of selected to neighboring trees with respect to height growth and branch size, recorded in the same manner.

After the selection of individual trees about 40 cones per tree were collected in late September or early October of 1965 and 1966. Collection was done by felling the trees. A separate cloth bag was used for the cones from each tree. Each bag was numbered to correspond to the data sheet and dispatched to East Lansing, where cones were allowed to dry for three months before extraction and cleaning was done separately for each lot.

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Seed from the selected jack pines was sown in the spring of 1966, with a 4-replicated randomized complete block design, accompanied by a fifth block to be used for outplanting. Measurements of height and branch size will be made in the nursery and analyzed immediately. The measurable stock will remain in the nursery for five years. At a later date, the data will be used to establish juvenile-mature correlations.

Four permanent outplantings of the jack pine progenies will be established on available sites in lower Michigan in the spring of 1968. In later years, two additional outplantings will be made in the Upper Peninsula. Sites which are slightly above average in quality and adaptable to machine planting will be chosen. The following specifications have been outlined for the permanent phase of progeny tests:

Design: A randomized complete block design, each replicate to contain one 6-tree plot of each progeny.

Spacing: 6 x 6 feet; trees check-planted to keep the rows straight in both directions.

Number of replicates per planting: 10

Method of planting: Machine planting with a 5-man crew.

Rate of planting: 4000 to 5000 trees per acre.

Based upon the selected floor plan and how the  
 the spring of 1952, with a re-estimated amount of  
 plate block design, accompanied by a list of  
 used for original construction. The design and  
 also will be made in the future and exact measurements.  
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 years. At a later date, the work will be used to  
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 illustrate the design.

Care: Intensive, including brush control, possibly control of herbaceous weeds through chemicals, possible fertilization and pruning. No shearing to produce bush-type trees.

With the start of flowering the jack pine out-plantings will be measured. Measurement will include branch diameter, stem straightness, height, etc. Parent-progeny relationships will be determined by subjecting the data to analysis of variance and correlation analysis. Other sets of measurements will be made and statistically analyzed at five or ten year intervals.

Subsequent to each set of measurements, the plantings will be thinned so as to remove the poorest progenies (family selection). In the later thinnings, the poorest trees in the best progenies will be removed (combined family and mass selection). The genetic quality of the stand and of the seed produced will increase with each thinning. Theoretical aspect of the genetic gain is illustrated as follows:

- a. From the  $1/2$  sib open pollinated trees seed with  $1/2 K\Delta G$  will be had. The constant  $K$  refers to the heritability (portion of variance due to additive genetic factors). The genetic gain after mass selection is  $\Delta G$ .

Case: Invasive, including such as, possibly  
 control of herbaceous weeds through their  
 possible fertilization and growth. No  
 as production-type, possibly

When the first of flowering the first five

plants will be measured. The number of  
 plants at a given stage, possibly, and  
 growth rate, possibly, and the  
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- b. The expected genetic gain after family selection will be equal to  $1/2 K\Delta G + \Delta G_{hs}$ .  $\Delta G_{hs}$  denotes the gain from  $1/2$  sib family selection.
- c. The new expected gain as a result of the combined family and mass selection would be equal to  $1/2 K\Delta G + \Delta G_c$ .  $\Delta G_c$  represents the fraction of total gain due to combined family and mass selection.

Removal of the poorest progenies will mark the conversion of progeny tests to seed orchards. Other cultural treatments aiming at producing abundant cones and ease of collecting them, may include complete weed control, fertilization, and pruning of lower branches.

The above progeny test-seed orchards would be only a single step in a several generation improvement program, and because of this, they are to be replaced at the age of 20 by  $F_2$  orchards capable of producing even better seed.  $F_2$  orchards will involve two-parent (full sib) progenies. Theoretically, the expected gain from the initial full-sib seed orchard is  $K\Delta G$ , and after family selection, this gain will be raised to  $K\Delta G + \Delta G_{fs}$ . The later thinnings, which will be a combination of family and mass selection, will raise the expected genetic gain still further.

The expected genetic gain after selection  
 will be equal to the expected gain  
 from the selection of the best  
 individuals in the population.

The new expected gain as a result of the  
 combined family and mass selection would be  
 the sum of the expected gains from the  
 two selection methods.

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Selective breeding will be continued into the third and fourth generations if progress in obtaining better seed is shown.



CHAPTER III  
TREE BREEDING AT PLACERVILLE,  
CALIFORNIA



### CHAPTER III

#### TREE BREEDING AT PLACERVILLE, CALIFORNIA

Institute of Forest Genetics at Placerville, California, is probably the foremost institute of its kind in the world. It was established in 1925 as the Eddy Tree Breeding Station. The present name was adopted in 1932. In 1935, the United States Forest Service took over the administration and now the institute is a part of the Pacific Southwest Forest and Range Experiment Station in cooperation with the University of California.

The institute is located at an elevation of 2700 feet, near the lower edge of the main ponderosa pine zone in the central Sierras. Climate around the research station is comparatively mild; maximum temperatures rarely exceed 100°F. Winter minimums are usually near 20°F. Precipitation is about 40 inches, nearly all of which falls from October to May. The soil is a deep sandy loam of a remarkably uniform character.

It was Mr. Eddy's original intention to benefit timber-growing and lumber production throughout the United States. Hence softwoods, rather than hardwoods, were picked out for the principal attention. A decision



was also taken early in the life of the institute to confine most of the initial efforts to the species of one genus. Pines were chosen for intensive breeding research, for the following reasons. First, Pinus is the most widely distributed genus of conifers in the northern hemisphere and the United States. The genus is particularly well represented on the Pacific Coast, there being 20 different species and important varieties in California alone. Second, various species of Pinus have the most generally useful wood of any of the genera of conifers. Pine species supply a great part of the construction lumber for the United States and are notable timber producers throughout the world. Finally, Pinus is adaptable to a great variety of ecological conditions.

Tree-breeding at Placerville, is directed towards the following objectives:

A reconnaissance of pine hybridization.  
Evolving methods to mass produce the hybrids.  
Conducting investigations, involving short-term studies in other fields (physiology, taxonomy, morphology, etc.) to aid the exploratory work and facilitate the practical application of the results.

Exploratory investigations in pine hybridization.--

The investigations in hybridization are aimed directly

was also taken early in the life of the institute to con-  
line most of the initial efforts to the species of one  
genus. Plans were chosen for intensive breeding programs  
for the following reasons. First, it was felt that the most  
likely distribution of the genus in the northern  
hemisphere and the United States. The genus is particu-  
larly well represented on the Pacific Coast, there being  
20 different species and subspecies recorded in California  
alone. Second, a number of species of high economic value  
generally occur in the area of the Pacific Coast, and  
have species which are well adapted to the conditions  
of the Pacific Coast. Third, the Pacific Coast is a  
large area and the conditions are generally favorable  
for the breeding of the genus. Fourth, the Pacific Coast  
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at the improvement of pines. The objective is accomplished in the following way. First, production of a particular hybrid is attempted. Then, if the hybrid is obtained, its economic potentialities are studied through performance tests in the nursery and in field plantings. In the course of such testing, various treatments (inoculation with pathogens, forced attacks by insects, etc.) may be applied by cooperating scientists. Criteria used in judging the economic potentialities include rapidity of growth, adaptability to the locality where hybrid is to be used, cold hardiness and drought resistance, resistance to insects and diseases.

Pine hybridization reconnaissance has yielded data on the crossability of particular species and of hybrids, the ease with which hybrids and their progenies may be obtained, the inheritance of characters, and the economic potentialities of hybrids.

Initial crosses are attempted on a small scale without aiming at a specific location or use and without trying to select superior phenotypes as parents (avoiding, however, diseased and defective individuals). If sound seeds are produced from the crossings, they are kept separate by seed-tree and the seeds from each tree are tested against the wind pollinated seeds from the same tree. The results of such a procedure indicate what in general may be expected from other crossings between the species.

at the improvement of place. The objective is to be achieved

in the following way. First, production of a particular

type of bird is selected. Then, in the hybrid to be obtained

the genetic constitution is determined by means of a

series of tests in the laboratory and in the field.

The course of such tests, and the results obtained (including

the time between the start of the tests and the end of the

series of tests, and the results obtained, are recorded and

may be applied by comparing with the results obtained in

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As Critchfield (1963) puts it:

Once it is established that two species can be crossed, the ease with which they cross is of primary importance in determining how the hybrids can be used. If two species, cross with great difficulty, the hybrid is likely to find its chief use as a bridge for the transfer of characteristics between species. If the genetic barriers between species are weak, however, it may be possible to capitalize on the attributes of  $F_1$  hybrid itself.

The crossability patterns being evaluated at Placerville, when supplemented with biochemical, anatomical, and morphological studies, can ultimately provide a natural system for the taxonomy of genus Pinus. The earlier systems of inter-relationships of pines, proposed by Shaw (1914) and Pilger (Duffield, 1952), are based upon morphological evidence only. A classification based upon true phylogenetic values, would undoubtedly facilitate the job of pine tree-breeders all over the world.

Devising economical methods for the mass-production of desirable hybrids.--A first step was made and large trials of the most promising hybrids were started in 1946. In 1948, J. W. Duffield initiated a program for the quantity production of hybrid seeds. There were practical difficulties. In pines, four to ten years pass from the time of pollination to the first preliminary judgements on growth rate and general value under plantation conditions. Even more time is needed

in the field of genetics

It is well known that the genetic code is a set of rules by which the information encoded in DNA or mRNA is translated into proteins. The genetic code is nearly universal, with only a few minor variations in certain organisms. The code is read in groups of three nucleotides, called codons, which specify a particular amino acid. The start codon, AUG, codes for the amino acid methionine and also serves as the start signal for translation. The stop codons, UAG, UAA, and UGA, signal the end of translation. The genetic code is a key to understanding the relationship between genotype and phenotype.

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to decide if the hybrids will be satisfactory for timber production. Therefore, methods for reducing the time element have been evaluated in the institute. Is it possible to produce male and female flowers early in the life of a tree? This question led to grafting attempts with young seedlings on mature trees having abundant flowers, on the assumption that flowering of young seedlings might be promoted by a hormone crossing the graft. Mirov's work on hard and white pines (1949) proved such a possibility; male flowering was induced on 2-year-old seedlings.

In order to put the hybrid pines to practical use more quickly, F. I. Righter (1954), suggested that they be interplanted among regular planting stock or among the natural reproduction on cut over lands. If the hybrid proved unsuited to the situation, it will be crowded out; if it proves superior it will form the mature stand of trees. However, as pointed out by Righter, "The economic feasibility of such a course, depends on the total increase in yield from the hybrids in relation to the cost of seed-production carried forward at compound interest." The method has been put into practice, and interplanting tests of several species hybrids have been established on seven of the national forests in California (Righter and Duffield, 1950).

to decide if the machine will be satisfactory for the  
production. Therefore, methods for reducing the risk  
element have been evaluated in the literature.

It is possible to reduce the risk element in the  
life of a plant. This decision led to studying elements

with young seedlings or plants from having adequate  
fertilizer, the concentration and frequency of watering  
and the amount of light. The results showed that the  
young plants with adequate fertilizer, watering and light  
gave the best results in the field. The results also  
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light gave the best results in the field.

Supplementary studies.--While the production of hybrids has been the primary objective of work at Placerville, several supplementary investigations have been undertaken. A comprehensive 11-year study of the chemistry of pine oleoresins was completed by Mirov (1961). The investigation shed light on the biosystematic relationships of many of the pine species. A good example of how chemical composition of terpene may serve taxonomic purposes, is the relationship of Pinus helepensis and its supposed variety Pinus brutia. In the light of chemical differences of their terpenes, these two pines appear to be separate species. This conclusion has been supported by independent morphological studies and by entomological evidence.

Knowledge of terpene composition promises to be useful in the entomological studies concerned with insect-repellant or insect-attractive constituents of oleoresins. Recent investigations carried out by Smith (1966) indicate an association of resin vapor toxicity with the host specificity of the insects, Dendroctonus brevicomis, D. monticolae and D. jeffreyi.

Propagation of pines by cutting and grafting is desirable in some research work. N. T. Mirov, assisted by E. F. Kimbrough, demonstrated that not only branchlets but even needle bundles could be used for cuttings and for scions in grafting. By use of these methods, portions





of valuable trees from other localities may be propagated, or as many genetically identical offspring as may be needed can be had from any particular tree.

An investigation on seed size and seedling size relationships sought ways to judge inherent vigor of young trees. It was found that several factors other than inherent capacity to grow fast affect the size of seedling trees for several years. Differences in time of germination and in seed size are most important. There was often a difference of a month or two between the germination of the earliest and the latest seeds. When seeds are late to germinate, the young trees have a shorter growing season the first year and are, therefore smaller. It may take several years for the tree to overcome this initial handicap. Schedules have been developed for chilling pine seeds that make it possible to germinate all seeds within three days. The stimulus given by the food supply in large seeds may mark the seedlings' true growth rate for two or three years. To overcome this difficulty, all seeds for experimental use are weighed and segregated into classes so that the growth of trees from seed of like weight may be compared.

One of the most critical steps in the pine hybridization program is the provision needed for long-term storage without loss of viability. Investigations relating to long storage of pollen have been carried out

of various types from other localities are reported.

Some very generally identical specimens are reported

from the same locality and are reported as being

identical to those from other localities.

In some cases the specimens are reported as being

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Some specimens are reported as being identical to

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at various humidities in refrigerators and deep-freeze chambers (Stanley et. al., 1960; Duffield and Callaham, 1959). The results indicate that pollen should be stored at 0°C. or below and 10 percent relative humidity. Also pine pollen kept at -23°C. and -50°C. has germinated well. The studies remain underway and each year pollen samples are removed from a deep freezer and tested for viability and fertility.

Local Conditions Affecting the  
Placerville Program

To select suitable strains of Pinus ponderosa, a provenance study was initiated by Lloyd Austin and A. R. Liddicoet in 1929. The seed was collected from South Dakota to British Columbia and southward to Arizona and California. Other smaller tests were started from time to time. In 1937, an experiment was established to test 729 individual seed trees of ponderosa pine.

In 1940, the emphasis of breeding work swung to hybridization because (U.S.D.A., 1948): "(i) Maintaining experimental control is difficult, and (ii) the costs of conducting such tests may be excessive when compared with the benefits to be expected from this method of pine breeding."

Choice of hybridization as the dominant breeding



method at the Institute during the last quarter of century can be traced back to other factors.

Pinus is the largest coniferous genus and owing to the great number and diversity of the species and varied places in which they grow naturally throughout the northern hemisphere, there are almost endless possibilities for producing new combinations of desired characters.

An arboretum is a pre-requisite for carrying out an efficient hybridization program. By the time hybridization was adopted as the chief breeding method at Placerville, in 1940, an arboretum was ready to facilitate the job. The Eddy arboretum, named after the Institute's founder, contains the most complete collection of pines (75 species) in the world. In addition to this, a large number of trees in the arboretum are raised from the nursery stock which provide testing material. Four specific purposes are achieved with this material. They are: (1) to study early growth of hybrids and other pedigreed seedlings, (2) to grow pedigreed planting stock for the Institute's plantation tests here and elsewhere, (3) to facilitate insect and disease studies, and (4) to prove and demonstrate techniques and results.

The climate at Placerville is sufficiently mild to permit assembling large collections of the different species of Pinus. In this climate, all but a few of the most tropical of the world's species of pines can readily be grown to seed bearing age. The average growing season



at the research station is approximately 225 to 250 days.

There is abundant opportunity to obtain nearby test areas in the foothills and higher reaches of the Sierra where the minimum temperatures range from  $-30^{\circ}$  to  $+16^{\circ}$ F. Austin stated in 1937

. . . On the Atlantic Seaboard one would have to travel about a thousand miles, from Florida to New England, to secure as great a range in minimum temperatures. Thus the Institute is exceptionally well situated to assemble for hybridization purposes pines of nearly all degrees of cold-hardiness, and to develop and test strains that will combine various desirable characteristics with considerable cold resistance. . . .

Before establishing the Institute, its founder James Eddy consulted Luther Burbank who had been practicing hybridization in horticulture. Burbank believed that pines could be bred as successfully as giant daisies or potatoes and therefore he encouraged Eddy to develop such plans for the forest trees. It can thus be assumed that from the very beginning, Eddy was prejudiced in favor of hybridization.

Although as early as 1927, attempts were made to hybridize different species of pines, yet the proper techniques had not been developed at that time and with one exception, those early crossings failed (U.S.D.A., 1948). With the arrival of F. I. Righter in 1940 as the head of the Institute, a new impetus was given to the practice of hybridization. Righter was particularly

at the research station in approximately 1953 or 1954.  
 There is abundant opportunity to obtain certain  
 data from the bookkeeping and other records of the  
 Bureau which are available to the public. It is  
 possible that certain data are available in 1953.

On the Atlantic seaboard one would have  
 to travel about a thousand miles from Florida to  
 New England to obtain a record of a single individual.  
 The fact that the Bureau has a record of a  
 well established individual in Florida is  
 not surprising. It is not surprising that the  
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interested in this approach and assisted by W. C. Cumming and A. R. Liddicoet, he perfected techniques of controlled pollination in pines suited to Sierra Nevada conditions. Methods to isolate ovulate flowers, extract, test and store pollen, to pollinate the ovulate flowers, and to test hybrids were developed during his tenure. Through 1964, when he retired from the Institute, some ninety hybrid combinations, including first generation ( $F_1$ ), second generation ( $F_2$ ), back-crosses and multi-species hybrids, had been reported.

Interest in hybridization has not dwindled with the retirement of Righter. His absence has been compensated by William Critchfield on the staff. Critchfield has continued interest in such hybrids as Pinus attenuata X radiata var. binata; P. muricata X attenuata; P. sabiniana X coulteri; P. sabiniana X torreyana; and P. jeffreyi X engelmannii.

The Sierra Nevada mountains range from 200 to 10,000 feet above sea level. Such topography offers extreme variation in edaphic and climatic factors associated with altitude and is therefore hardly suitable for large racial selection studies and progeny tests which demand high degree of uniformity in the environment for proper evaluation.

introduced in this approach and resulted in a  
 Cushman and A. E. Robinson, in 1934, published  
 controlled pollination in which pollen was  
 collected from flowers to isolate overwintering  
 and store pollen, to pollinate the flowers  
 and to test hybrid vigor was reported in 1934.  
 and a method for the isolation of pollen from  
 this case, the pollen was collected from the  
 flowers and stored in a dry place for use  
 in the following year.

The method described in this paper is a  
 simple and practical method for the isolation  
 of pollen from flowers and its storage for  
 use in the following year. It is suitable  
 for use in the laboratory and in the field.  
 The method is described in detail in the  
 following pages.

Techniques of Controlled Pollination

Techniques of controlled pollination have been developed at Placerville over a period of twenty years. Improvements have resulted in (i) isolation of ovulate flowers, (ii) collection and preparation of pollen for use, (iii) pollination of female flowers, and (iv) protection of cones. When properly modified, the Placerville methods are applicable to other cone bearing trees in other regions.

For the sake of convenience, seven distinct stages of the development of ovulate strobili are arbitrarily recognized at the Institute. They are : (1) buds small, (2) buds large, (3) buds opening, (4) flowers partly open, (5) flowers maximum, (6) flowers closed, and (7) cones enlarging.

The buds are enclosed in pollination bags as soon as they are recognizable, commonly at the "bud small" stage, but occasionally at the "bud large" stage.

The Institute has perfected its own pollen-proof bag. It was found that bags of materials lacking requisite toughness were useless for Sierra Nevada conditions. Moisture-tight bags were rejected as the vapor from transpiration would condense in them and obstruct the vision through transparent substances. Furthermore, the weight of water that accumulates in such bags would cause the



limbs of some species to bend down and sometimes break off. For quite a few years, Institute's standard bag has been a 14.5 x 8-inch bag, made up of a piece of finely woven Army duck, with a 3 x 4-inch window made of cellulose acetate (.01 inches thick). The bag, although tough, proved too heavy for certain species such as Pinus glabra and P. helepensis which have relatively slender shoots. In recent years, a "kraft paper bag" has been used. The bag has the same dimensions as above and is similarly provided with a 6 x 3-inch cellulose acetate window. Kraft paper has proved satisfactory because of its light weight and toughness. Initial cost of Kraft bags is higher than sausage casings, however, they can be re-used.

Before re-use, pollination bags are cleaned with a vacuum cleaner, re-lined at the neck with a 1/2 x 6-inch cotton wand, and sealed with gummed paper. They are then subjected to heat (80°F.) for 15 hours to kill any pollen. Immediately prior to use, the bags are inspected for any cracks in the paper or window.

While bagging, if there is a choice between two or three flowers at the "bud small" stage and six or eight flowers at the "bud large" stage, the former is bagged. That the latter may pass the receptive stage before pollination is possible. To pull in long branches, a four-inch hook attached to an eight foot length of stiff No. 6 wire is used.



The number of flowers enclosed in each bag, after tagging the branches, is recorded on a prescribed form which is reproduced as Appendix II. The flowers that are used as controls are also tagged. This allows a check on the effectiveness of pollination.

Technicians at Placerville collect male catkins when a clear liquid oozes out when pressed, that is before perfect maturity is attained (yellowish juicy liquid is the indication of complete ripeness of the male catkins). At this stage a few male catkins are mature on the tree.

In the laboratory, male catkins are washed and dried before the pollen is extracted. Extractors consist of sterilized 12-inch x 30-inch canvas bags and funnels which empty through a rubber tube into glass receptacles. A four inch circle of 60 mesh screen is soldered within each funnel just above its spout to catch coarse materials, such as catkin particles, dried pitch, etc., which otherwise would mix with the pollen. A sieve of the same diameter as the funnel is held about two inches above the funnel by metal legs that are soldered to the bottom of the hoop. The bags have two windows. The receptacles are glass bottles equipped with a rubber cap in which a cross (+) is cut to permit entry of the rubber tube into the receptacle. When the tube is forced through the cut in the cap, the cap makes a tight fit around the tube. When the tube is withdrawn, the cap closes automatically.

The number of flowers which are produced by the plant during the period of its life is recorded in a special table which is reproduced in Appendix II. The flowers which are used for analysis are those which are collected on the 15th day of the flowering period. The flowers which are collected on the 15th day of the flowering period are those which are collected on the 15th day of the flowering period.

When the first flower buds appear, the plants are divided into two groups: one group is used for the study of the effect of the hormone on the growth of the plant, and the other group is used for the study of the effect of the hormone on the flowering of the plant.

The results of the experiment show that the hormone has a significant effect on the growth and flowering of the plant. The plants which were treated with the hormone showed a significant increase in the number of flowers produced during the period of their life compared with the control plants.

The results of the experiment also show that the hormone has a significant effect on the flowering of the plant. The plants which were treated with the hormone showed a significant increase in the number of flowers which were collected on the 15th day of the flowering period compared with the control plants.

The results of the experiment are summarized in the following table:

Group	Number of flowers produced during the period of life	Number of flowers collected on the 15th day of the flowering period
Control	100	50
Hormone treated	150	75

The results of the experiment show that the hormone has a significant effect on the growth and flowering of the plant. The plants which were treated with the hormone showed a significant increase in the number of flowers produced during the period of their life compared with the control plants.



The catkin-washing operation is very carefully done. The catkins are loaded into a funnel and washed with a stream of lukewarm water, then the funnel is immersed in a warm water bath. While still immersed the catkin-containing funnel is fitted into the lower end of the canvas extractor. Then extractor and funnel are removed from the water and the lower end of the funnel plugged with cotton. Then the extractor is placed to dry in a heated room with circulating air. Pollen is ready in 1 to 3 days and is drawn off through a small rubber tube, the end of which is sterilized (with alcohol) after each use.

Testing the pollen.--Pollen is tested for viability before use, using the 'hanging drop' method described by Righter (1939). Holes are punched in a 2-millimeter layer of paraffin placed in the top of a Petri dish, and in these holes the liquid culture media is placed. A few drops of distilled water are placed in the Petri dish to maintain humidity. The inside periphery of the top section is coated with vaseline to provide a seal. For preparing the culture, a sterile hypodermic syringe equipped with a rubber bulb and 15 gauge needle is used. A portion of each lot of pollen is withdrawn from its container into a separate syringe and distilled water is next drawn into the syringe and mixed with the



pollen by shaking. To place the culture in the Petri dish, a drop of the pollen water mixture is squeezed into a hole. The dish is then placed in an electric oven, adjusted to maintain the temperature at about 26°C.

Pollinating the flowers.--When the flowers begin to emerge from the buds they are watched closely so that they can be pollinated between the fourth to sixth (preferably fifth) stages. The materials include labelled vials of pollen, labelled hypodermic syringes of 10 cc. capacity equipped with rubber bulbs and 16 gauge needles whose points are protected with corks, aluminum data tags, pertinent data, record sheets in a loose-leaf note book, plungers for clearing needles that become plugged, wires, a rope, pencils and a knapsack.

When flowers are ready for pollination, the technician forces the pollen into the bag through the needle of hypodermic syringe. The puncture is closed by an adhesive tape. The bag is then shaken vigorously to distribute the pollen thoroughly around the flowers.

A properly labelled 8-inch aluminum tag is fastened to the branch a few inches below the bag. Stapled to the tag is a strip of colored cloth to serve as a flag. A tag applies to all cones maturing on the stem above it. Pollinations on the same tree are often made over a several-day period. Pollination bags are removed after the ovulate flowers reach stage 7.

police by showing. To place the camera on the level  
 dish, a drop of the police water mixture is applied  
 into a hole. The dish is then placed in an upright  
 over, adjusted to camera. The camera is then

Pollinating the Flowers—When the flowers begin

to emerge from the cross they are worked closely so that  
 they can be pollinated between the fourth to sixth

(previously mentioned) days. The pollination should be done  
 with a pollen brush. The pollen is taken from the  
 anthers of a flower and rubbed on the stigma of another

flower. It is important that the pollen be fresh and  
 that the stigma be receptive. The pollen should be  
 applied to the stigma of the flower to be pollinated

as soon as possible after the pollen is taken from the  
 donor flower. The pollen should be applied to the  
 stigma of the flower to be pollinated as soon as

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 as soon as possible after the pollen is taken from the

Protection of seed and cone.--Cloth bags are applied to the conelets pollinated during the previous seasons to guard against attack by squirrels and insects. These bags also catch seed from cones opening prematurely. Cone bagging is generally done in the spring and before the attacking insects become active.

#### Hybrids Made at Placerville

Since the inception of the Institute of Forest Genetics, about ninety different hybrids ( $F_1$  inter-specific crosses, back-crosses, 3-way hybrids) have been produced. Some hybrids were easy to make. In contrast, Pinus nigra X resinosa yielded only 7 seeds after several years' effort. Reports by Liddicoet and Righter (1960) and Critchfield (1963; 1966) cover most of the hybrids produced. Although most of the hybrids are too young to show their true capabilities, as Duffield and Snyder (1958) point out "Evidence from natural hybrids found in the wild as well as from artificial hybrids, gives a large measure of confidence in the long term performance of hybrids." Some of the outstanding hybrids growing at the Eddy Arboretum are mentioned as follows.

Pinus ponderosa X latifolia shows early growth advantages over P. ponderosa for reforestation in the California region (Righter and Duffield, 1951). This



combination has a long, stout tap root which can be of great value in a region of high summer temperature and little or no summer rain fall. It outgrows P. ponderosa in height and diameter.

Hybrids between jack pine of the Lake States and lodgepole pine of the Rockies and Sierra Nevada, have the good form of lodgepole pine and a growth rate almost equal to jack pine. It should be of value in the Lake States and in the Rocky Mountains.

Ponderosa pine of the Sierra Nevadas P. ponderosa X var. scopulorum pine in the Rockies, equals ponderosa pine in growth rate at four years and exceeds it after the fifth year. This hybrid presumably has some of the hardiness of the scopulorum parent. It is a prospect for trial in the Great Basin area and eastwards.

Jeffrey X (Jeffrey X Coulter) pine is a vigorous hybrid which is resistant to resin midge and pine reproduction weevil, both serious pine plantation pests. It is cold-hardy throughout much of the ponderosa pine belt.

Hybrids between eastern white and Himalayan white pines grow faster and are more resistant to white pine blister rust than eastern white pine.

History of the knobcone X Monterey pine hybrids can be given in detail. Knobcone pine is a small, thin-foliaged frost resistant tree of the Coast ranges and Sierra Nevada foot-hills of California. Monterey pine

combination of wind, snow and fog which was of  
great value in a region of high ocean temperature  
little or no surface wind. It was observed that  
in height and direction.

Hydrate between each line of the Lake Basin and  
Kokopole line of the Rockies and Alaskan ranges,  
the good part of the Kokopole line and a small part of the  
equal to that of the Alaskan range. It is observed that  
Kokopole and Alaskan ranges are of the same height.

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is a rapidly growing, heavy-foliaged, frost-tender species of the central California coast. The hybrids grow almost as fast as Monterey pine but the important characteristic is their ability to withstand temperatures as low as 16°F. They should be useful as a fast-growing ornamental tree or for windbreaks and erosion control in localities too cold for Monterey pine. This combination was the first pine hybrid produced at the Institute of Forest Genetics. It has been described by Stockwell and Righter (1946) and Little and Righter (1965).

Fresh pollen was collected from native Monterey pine and placed on receptive conelets of a native knobcone pine near Placerville in April, 1927. Hand-pollinated cones were harvested in the autumn of 1928 and the hybrid seeds from these cones were sown in a seedbed the following spring. Twenty-eight 2-year old seedlings were planted in the Eddy Arboretum for testing. Planting was done at 15 x 15 feet. A few seedlings of both parental species were planted adjacent to the hybrids.

The hybrids withstood cold which killed 3 to 5-year old Monterey pine seedlings. At age 16 the hybrids were 42 to 62 feet (average 52) tall, and 9 to 19 (average 13) inches in diameter. Adjacent knobcone pines were 40 feet in height and 10 inches in diameter. Twenty F<sub>1</sub> trees measured at age 30 (1959) averaged 73 feet high

is a rapidly growing, heavy-billed, black-headed  
 species of the Central California coast. The birds  
 grow almost as fast as honeycreepers but the honeycreepers  
 characteristic of their ability to withstand temperatures  
 as low as 10° F. They should be useful as a fast-growing  
 species for windbreaks and ornamental plantings in  
 localities too cold for honeycreepers. This conifer  
 was first introduced to California in 1858.

It is a small, bushy tree, 10 to 15 feet high,  
 with a dense, rounded top. The leaves are dark green,  
 and the flowers are small and white. The fruit is a  
 small, round, red berry. It is a very hardy  
 plant, and is well adapted to the climate of  
 California. It is a very useful plant for  
 windbreaks and ornamental plantings. It is  
 a very hardy plant, and is well adapted to  
 the climate of California. It is a very useful  
 plant for windbreaks and ornamental plantings.

and 17 inches in diameter.

Cones and catkins were seen on some hybrids at age 4; at age 10 all bore cones.

In 1940, open pollinated seeds from  $F_1$  hybrids (actually  $F_2$  seed) and knobcone seeds were planted in a seedbed. At age 5  $F_2$  hybrids were taller than knobcone pine seedlings. The growth rate of some of the  $F_2$  population compared favorably with the best Monterey pine seedlings.

The hybrid test started in 1927 was repeated in 1947. Results were the same as before.

Field trials of  $F_2$  Monterey X knobcone hybrids and seven other hybrids were initiated in 1950 at three test sites (elevation 5,200-6,400 feet) in the Stanislaus-Tuolumne Experimental Forest of California. Air temperature at the test site could vary between  $-10^{\circ}$  and  $96^{\circ}$ F. The seedlings were planted as 1-1 stock, 25 trees per plot. At age 10, the Monterey-knobcone hybrids had failed at all three sites.

$F_1$  Monterey-knobcone hybrids are being mass produced by the U. S. Forest Service for field trials in several places in California.

and 17 inches in diameter.

Cones and needles were seen in some specimens.

Age 4; at age 10 all cones were seen.

At age 10, the hybridized seeds from 1 hybrid

(generally F<sub>2</sub> seeds) and inbred seeds were planted in a

bed. At age 2 F<sub>2</sub> hybrids were better than inbred ones

in seed yield. The growth rate of the F<sub>2</sub> hybrids

was not much different from that of the inbred ones.

Conclusions.

1. The growth rate of the F<sub>2</sub> hybrids was not much

different from that of the inbred ones.

2. The seed yield of the F<sub>2</sub> hybrids was better than

that of the inbred ones.

3. The growth rate of the F<sub>2</sub> hybrids was not much

different from that of the inbred ones.

4. The seed yield of the F<sub>2</sub> hybrids was better than

that of the inbred ones.

5. The growth rate of the F<sub>2</sub> hybrids was not much

different from that of the inbred ones.

6. The seed yield of the F<sub>2</sub> hybrids was better than

that of the inbred ones.

7. The growth rate of the F<sub>2</sub> hybrids was not much

different from that of the inbred ones.

8. The seed yield of the F<sub>2</sub> hybrids was better than

that of the inbred ones.

9. The growth rate of the F<sub>2</sub> hybrids was not much

different from that of the inbred ones.

10. The seed yield of the F<sub>2</sub> hybrids was better than

that of the inbred ones.

CHAPTER IV  
TREE BREEDING AT GULFPORT,  
MISSISSIPPI



## CHAPTER IV

### TREE BREEDING AT GULFPORT, MISSISSIPPI

The (Southern) Institute of Forest Genetics was established at Gulfport, Mississippi, in 1954, by the United States Forest Service. It was preceded by the Gulfcoast Research Center of the Southern Forest Experiment Station.

The Institute is advantageously located for research on the southern pines on which it has concentrated all of its early efforts. Longleaf (Pinus palustris) and loblolly (P. taeda) pines, their natural hybrid (X P. sonderoggeri), slash (P. elliotii) and shortleaf (P. echinata) pines, and one minor species, spruce pine (P. glabra) occur naturally in the immediate vicinity. Sand (P. clausa), pond (P. rigida var. serotina) and virginia (P. virginiana) pines, occur naturally within 200 miles. Baldcypress (Taxodium distichum) and red cedar (Juniperus virginiana), are other conifers of the area.

There are also excellent hardwoods. Yellow-poplar (Liriodendron tulipifera), red gum (Liquidambar styraciflua), cottonwood (Populus deltoides), black





tupelo (Nyssa sylvatica), water tupelo (N. aquatica), black willow (Salix nigra), black walnut (Juglans nigra), Sycamore (Platanus occidentalis), and several species of oak (Quercus sp.), are some of the valuable hardwood species of the area.

Climate of the region is characterized by long hot humid summers and short mild winters. July average temperature is 85°F., January average is 55°F. Moderate temperatures, the long growing season (280 days), a well distributed rainfall (annually 55 inches), all contribute to a luxuriant vegetation.

Soils of the Gulf Coast are red and yellow podzolic; they have undergone severe laterization. They range from sandy to clay. Generally, these soils are poor for agricultural crops and good to excellent tree growth.

The Institute administers the 4,000-acre Harrison Experimental Forest at Saucier, and the 1000-acre McNeill Experimental Forest at McNeill, Mississippi. A 10-acre arboretum is being established at the Harrison Experimental Forest.

In 1961, the Institute was assigned the specific responsibility for the Forest Service's research on the breeding techniques of southern hardwoods. Initial studies are devoted to cottonwood, sweet-gum, sycamore,



and five of the economically important oaks--cherrybark, Nuttall, swamp white, Shumard, and willow. The pine work continues.

#### Major Objectives of Breeding Work

The general objective of the Institute's breeding program is to determine the degree of genetic control of phenotypic characters of the important southern pines and hardwoods and to develop methods of applying this control for the production of improved forest trees. The work falls into four phases: the study of variation, the utilization of this variation in inheritance and hybridization studies, development of most promising strains of trees, and silvicultural relations of genetically improved varieties.

Improvement of characters such as vigor, form, wood quality, and resistance to diseases, insects, and adverse environmental factors are sought through selection, hybridization and modification, with special emphasis on the mode of inheritance. Economically important characters receive major emphasis. Progeny tests, provenance tests, inter- and intra-specific hybridization, are being employed.

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The general objective of this...

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Relationship of Breeding Work  
to Local Conditions

The South is a major producer of wood for lumber and pulp. The population of the United States and the world is increasing and this means a greater demand for southern wood fibre. Of the United State's annual production of 13 billion cubic-feet of wood, the South grows nearly half. Of the estimated goal of nearly 20 billion cubic feet within the next fifteen to twenty years, the Southern states are assigned the task of growing 10 billion. Tree breeders must explore all the methods to increase wood production. There is also a demand for better quality.

Southern forest plantations are managed more intensively than in other parts of the United States. Artificial planting is common and there is a great opportunity to plant new varieties. Test planting in the South is greatly facilitated by large private tree planting programs.

The Institute is fortunate in having a well qualified crew of tree breeders who specialise in different aspects of forest genetics. This coupled with the existence of most modern equipment and other facilities, does not warrant limiting the breeding activities to the minimum number of approaches.

Historical Statistics of the United States  
to 1890

The following table shows the population of the United States and the number of persons engaged in agriculture, stock raising, and fishing, and the number of persons engaged in manufacturing and commerce, and the number of persons engaged in other occupations, from 1790 to 1890. The population of the United States in 1790 was 3,929,214, and in 1890 it was 62,946,582. The number of persons engaged in agriculture, stock raising, and fishing in 1790 was 2,000,000, and in 1890 it was 12,000,000. The number of persons engaged in manufacturing and commerce in 1790 was 1,000,000, and in 1890 it was 30,000,000. The number of persons engaged in other occupations in 1790 was 929,214, and in 1890 it was 30,946,582.

Year	Population	Agriculture, stock raising, and fishing	Manufacturing and commerce	Other occupations
1790	3,929,214	2,000,000	1,000,000	929,214
1800	3,929,214	2,000,000	1,000,000	929,214
1810	3,929,214	2,000,000	1,000,000	929,214
1820	3,929,214	2,000,000	1,000,000	929,214
1830	3,929,214	2,000,000	1,000,000	929,214
1840	3,929,214	2,000,000	1,000,000	929,214
1850	3,929,214	2,000,000	1,000,000	929,214
1860	3,929,214	2,000,000	1,000,000	929,214
1870	3,929,214	2,000,000	1,000,000	929,214
1880	3,929,214	2,000,000	1,000,000	929,214
1890	62,946,582	12,000,000	30,000,000	30,946,582

Pines have been the most commercially important trees in the South and are grown mainly for sawlogs and pulpwood. In recent years there has been a phenomenal increase in the use of hardwoods for the pulp. Considering only one state, Mississippi, in the late 1940's, round hardwood pulpwood averaged less than 200,000 cords annually, or 15 percent of the total pulpwood cordage. Production of hardwoods since 1955 has averaged about 880,000 cords annually, or 44 percent of the round pulpwood output. These considerations have led the tree breeders of the South to place more emphasis on hardwood improvement.

#### Work in Progress

Variation among southern pine races.--In the South local seed is used for reforestation. The economic returns from following this principle, which was derived from early research (Wakeley, 1944, 1953, 1959, 1961; Wakeley, et. al., 1951) has probably more than paid for all subsequent tree improvement research in the several times over. However, practical problems still exist. In case of a crop failure, how far away is it safe to acquire seed? Are there sources better than the local ones?

These have been the most common types of  
cases in the field and are usually  
found in the following order:  
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At present, the Institute is carrying out several studies relating to the seed sources of southern pines, independently or in cooperation with the other agencies. One such study was designed to probe desirability of using non-local seed of slash pine. The study was started by E. B. Snyder in 1955 when seeds collected from sixty-four 20- to 30-year old plantations were outplanted in the Biloxi Ranger District and the MacNeill Experimental Forest. The parent trees were of south Georgia or north-east Florida origin. Seeds from eight local (south Mississippi) slash pines were also sown in the experiment. To date no significant differences have been found among the sources in cone-maturity, cone-length, number of seeds per cone, percent of full seeds, seed weight, wing weight, wing color, and seed-coat appearance. However, significant differences in survival were found in dry years, Florida source being poorest. There were no differences in fusiform rust resistance.

Another seed source study on loblolly pine was started in 1958. Seed was collected from throughout the range of the loblolly pine (Texas, Arkansas, Louisiana, Mississippi, Alabama, Georgia, South Carolina, North Carolina, Virginia and Maryland).

Single-tree Inheritance.--Individual tree variation exists in various species and races (Dorman, 1961).



A wide tree-to-tree variation in economically important characters means considerable progress by selecting trees with valuable traits.

Variation and heritability studies present a broad field of investigations to the Institute of Forest Genetics. These investigations relate to genetic diversity in loblolly pine and sweetgum, within Mississippi. The objectives are: to describe and compare the genetic variation associated with individual trees and stands, and to select outstanding genotypes, on the basis of progeny performance.

The approach to the first generation of testing consists of two steps: first, a screening of many randomly selected families (half-sib progenies) in a few extreme environments within the region considered, and second, the selection and testing of outstanding families in several varied environments. A secondary objective of the study is the comparison of the pattern of genetic variation in two genetically unrelated but ecologically associated species.

Seed collection from 678 sweetgum in 139 stands and 567 loblolly trees in 118 stands was done in 1962-63-64. Stands were selected along 20 latitudinal transects chosen at intervals of 15 minutes. Seedlings for all permanent outplantings were grown in Harrison Experimental Forest nursery in 1965. Three replications of a



compact family block design were used. Progenies were scored in the nursery for germination rate, height and other distinguishing traits. Three outplantings of all families of each species were established spring 1966. Compact family block design with 20 replications of the 678 gum families and the 567 pine families were used at each planting. One-tree plots at a spacing of 8 x 8 feet were employed.

Evaluation will be done for the following characteristics: survival at age 1, height and diameter at ages 3, 5, and later years, phenological differences whenever they appear, fusiform rust infection, and crown characteristics.

Polygenic inheritance in diallel crosses of longleaf pine is another important study. It utilizes the method of diallel cross for heterozygous organisms. The information to be derived includes the type and relative effects of gene and environmental action, combining ability, effects of selfing, and differences among reciprocals. Thirteen longleaf pine trees, average to superior were intercrossed in 1956-57. Experimental design consisted of 4-tree subplots of the compact family block type; subplots were paired to contain the cross and its reciprocal. Whole plots were arranged in a 9 x 10 rectangular lattice of eight replications. In the



compact family block design were used. Progenies were scored in the nursery for germination rate, height and other distinguishing traits. Three outplantings of all families of each species were established spring 1966. Compact family block design with 20 replications of the 678 gum families and the 567 pine families were used at each planting. One-tree plots at a spacing of 8 x 8 feet were employed.

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Company family blood being used. Provisions were  
made in the company for continuation after death and  
other distinguishing factors. These provisions of all  
families of each generation were established in 1900.  
Company family blood being used with no restriction of the  
any families and the 50% plan families were used in  
each planting. One-time blood was used in 1908 and  
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families of each generation were established in 1900.  
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families of each generation were established in 1900.  
Company family blood being used with no restriction of the  
any families and the 50% plan families were used in  
each planting. One-time blood was used in 1908 and  
was corrected.



spring of 1960, planting was made on 24 acres at the Harrison Experimental Forest, with a stand density of 349 trees per acre.

The longleaf diallel progenies will be evaluated for survival, height, branch angle, needle characteristics, and oleoresin content.

Interspecific hybridization.--One problem tackled by the Institute of Forest Genetics at Gulfport is determination of the value of interspecific hybridization as a method of improving southern pines. Crossability results with the four major southern pine species have confirmed those of Placerville. Crosses involving loblolly or shortleaf are relatively easy to make. Longleaf X slash pine hybrids are also obtained easily.

Attempts to synthesize sonderegger pine (longleaf X loblolly) have been unsuccessful at Gulfport, as in Placerville (Critchfield, 1963). An investigation into the crossability and incompatibility of this cross is now underway at Gulfport.

Many valuable characteristics have been noticed in southern pine interspecific hybrids, for example, rust resistance in shortleaf X loblolly and tip-moth resistance in shortleaf X slash hybrids. Longleaf X slash pines survived well, bypassed the grass stage of longleaf, were taller than longleaf pine at age 8 and were moderately

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susceptible to fusiform rust. Shortleaf hybrids were moderately resistant to fusiform rust and superior in height growth to pure shortleaf pine. They were inferior in growth and survival to longleaf, loblolly, and slash pines (Schmitt, 1966).

An important phase of the hybrid evaluation program at Gulfport, is testing a series of selected individuals and races as parents and observing the progeny at different sites. Screening tests of most possible interspecific southern pines were undertaken in 1957 and 1959. Analysis of four plantings ranging in age from 6 to 8 years has revealed that site, cultural conditions and individual parents had a marked effect upon hybrid performance. These investigations also indicate that hybrid survival is adequate and fusiform rust resistance of three combinations (longleaf X slash, shortleaf X loblolly, and shortleaf X slash) is exceptional (Schmitt, 1966).

A study of shortleaf geographic races X slash pine was installed in the spring of 1964. Controlled crosses were made between one race of slash pine and eight races of shortleaf pine. The main object was to learn whether fast growing shortleaf X slash pine hybrids can be planted north of the slash pine range.

Shortleaf X slash may prove to be of practical value where slash pine is an accepted species for planting

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and in parts of Arkansas and Oklahoma and the unglaciated parts of central states where shortleaf is the preferred planting species. In the former it is thought that the greater rust resistance of the hybrids gives them an advantage over slash pine, despite their somewhat slower growth. In the latter, it is hoped that faster growth and better recovery from tip-moth attack will make the hybrids superior to shortleaf (Wells, 1963). Also, parent-progeny correlations will be calculated.

Techniques.--One of the greatest problems in southern test plantations is the presence of a number of obnoxious weeds. They compete with the trees and present difficulty at time of measurements. One reason for not employing machine planting is that it is difficult to follow a straight line due to the dense cover of weeds on the ground (personal discussions with O. O. Wells). Simazine, Amino triazole, Dalapon and 2, 4, 5-T have been only partially effective. Research is needed to keep the experimental areas weed-free.

At Gulfport, there is a trend toward the use of more sophisticated types of experimental designs, that is, compact family block design and lattice type design. Compact family block design differs from a randomized complete block design in that the individual families from a single stand are in consecutive plots (rows)

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pairs of central colors which are the  
primary colors. In the center of the  
circle are the colors of the rainbow  
which are the colors of the spectrum  
and the colors of the rainbow are the  
primary colors. In the center of the  
circle are the colors of the rainbow  
which are the colors of the spectrum  
and the colors of the rainbow are the  
primary colors.

rather than scattered throughout a replication. This arrangement results in maximum precision for detection of differences between families within stands and slightly underestimates the differences between stands (Wright, 1963). Another advantage is the relative ease with which the data can be recorded and tabulated (Wells, 1964).

Compact family block design is appropriate where genotypes divide themselves naturally into groups and where the within-group differences are smaller or of more interest than those between groups. For genotypes whose differences are unknown or of equal size or interest (for example, estimation of genetic gains, combining abilities, and components of variance), Dr. Bayne Snyder at Gulfport, prefers a lattice design. Although it is complicated he asserts that newly developed computer programs will ease statistical analyses and minimize time of operations at all stages (Snyder, 1966).

A modified form of Michigan's folder system has been in partial use at Gulfport by O. O. Wells. A photocopy of the pertinent data remains with the cooperator and the original with the experimenter. Plantation maps do not contain information on soil, weather, spacing, experimental design as do Michigan maps. Standard accession record forms have recently been introduced (Appendix III).





To keep the identity of planting material in the nursery, paper field labels are used by Bayne Snyder. Inscription is done with any water-proof ink. One advantage of such paper made field labels as compared to the masking tape labels of Michigan is that inscriptions can be made in the office rather than in the field. However, this advantage is outbalanced by the longer life of masking tape as a field label.

At Gulfport transparent sausage casings are used for bagging female cones. Tests are underway to evaluate the performance of painted sausage casings to obtain cooler temperatures.

The warm humid climate of southern Mississippi is conducive to rapid deterioration of pollen. The pollen storage problem is acute. Presence of 10 to 14 percent moisture in the "dry" extracted pollen is actually the cause of rapid spoilage of pollen in the South. Snyder (1961) has found a correlation between the abundance of pollen saw fly larvae (Xylea sp.) and the relative humidity of the extractor. He has recommended that the relative humidity should be 35 percent or less.

Southern pine pollen is stored in sealed or unsealed containers, and wet or dry, with variable results. Deep-freezing pine storage method at Placerville indicates the advantage over ordinary storage in the refrigerator. This method, however, promises to be effective

The first step in the identification of plastic material is to  
 determine its general type. This is done by using a series of  
 tests which are described in the literature. The first of these  
 tests is the burning test. This test is used to determine the  
 general type of plastic. The second test is the solubility test.  
 This test is used to determine the specific type of plastic.  
 The third test is the density test. This test is used to  
 determine the specific type of plastic. The fourth test is the  
 infrared spectrum test. This test is used to determine the  
 specific type of plastic. The fifth test is the mass spectrum  
 test. This test is used to determine the specific type of plastic.

The next step in the identification of plastic material is to  
 determine its composition. This is done by using a series of  
 tests which are described in the literature. The first of these  
 tests is the elemental analysis test. This test is used to  
 determine the elemental composition of the plastic. The second  
 test is the infrared spectrum test. This test is used to  
 determine the infrared spectrum of the plastic. The third test  
 is the mass spectrum test. This test is used to determine the  
 mass spectrum of the plastic. The fourth test is the x-ray  
 fluorescence test. This test is used to determine the x-ray  
 fluorescence spectrum of the plastic. The fifth test is the  
 thermogravimetric analysis test. This test is used to determine  
 the thermogravimetric analysis spectrum of the plastic.

The final step in the identification of plastic material is to  
 determine its source. This is done by using a series of tests  
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 infrared spectrum test. This test is used to determine the  
 specific type of plastic. The fifth test is the mass spectrum  
 test. This test is used to determine the specific type of plastic.

in Gulfport only after the pollen is brought down to a certain minimum moisture content (personal discussions with E. B. Snyder). Unsuccessful attempts to freeze-dry pollen of longleaf pine under lyophilized conditions (i.e., suspending the pollen in Seitz-filtered bovine serum, freeze drying and sealing in vacuum) have also been made (Hasseltine and Snyder, 1958).

To test the pollen germination, a solution of 10 percent sugar and 75 percent agar is made. It is cooled to 50°C. and a small drop placed on a microscope slide. Pollen is dropped by tapping it off a small loop of wire. Slides are placed vertically in a slide rack over water in a sealed container and examined after 60-72 hours.

This method is fast, about 60 samples per hour can be prepared. It does not require delicate handling, and the large drop buffers against drying during handling. It is easy to handle microscopically. Contamination is minimized compared to systems having many samples close together within one container.

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 with E. S. Fisher). (Enclosed is a copy of the  
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CHAPTER V  
TREE BREEDING IN WEST PAKISTAN



## CHAPTER V

### TREE BREEDING IN WEST PAKISTAN

West Pakistan is situated between  $24^{\circ}$  and  $37^{\circ}$  N. latitudes and  $61^{\circ}$  and  $75^{\circ}$  W. longitudes in the west of the Indian sub-continent of Asia. Its total land area is 310,236 square miles. Two thirds of the total area is covered by two mountain ranges, one in the north, and the other in the west. The northern range is the western extremity of the great Himalayas whereas the western range is a part of Suleman series. The rest of the country is deep alluvial plain of the Indus river system.

West Pakistan experiences a rigorous continental climate. It is very hot in summer and very cold in winter. Most of the country is arid to semi-arid. The northern mountains receive 30 to 50 inches of precipitation while the plains and the western mountains get only 2 to 10 inches per year. Snow fall constitutes a very significant part of the precipitation in the mountaineous region. The higher mountain peaks (above 15,000 feet) are under perpetual snow. The lower peaks and ridges, that is, up to 2000 feet also carry thick layers of snow 6 to 9 months in the year. Frost is experienced throughout the province to a varying degree in the winter season.

WESTERN MINING IN WEST VIRGINIA

1877

West Virginia is situated between 37° and 41° N. latitude and 77° and 82° W. longitude. It is bounded on the north by the Potomac River, on the east by the Maryland and Pennsylvania lines, on the south by the Kentucky line, and on the west by the Ohio River. The area of the State is 62,000 square miles. The population in 1870 was 400,000. The State is rich in coal, iron, and other minerals. The coal fields are extensive and produce a large amount of fuel for the West. The iron mines are also numerous and produce a large amount of pig iron and steel. The State is also rich in other minerals, such as zinc, lead, and copper. The mining industry is the backbone of the State's economy. The coal fields are situated in the western part of the State, and the iron mines are situated in the eastern part. The mining industry has been the main source of wealth for the State since its settlement. The coal fields were discovered in the 18th century, and the iron mines were discovered in the 19th century. The mining industry has grown steadily since that time, and it is now one of the most important industries in the State. The coal fields produce a large amount of fuel for the West, and the iron mines produce a large amount of pig iron and steel. The State is also rich in other minerals, such as zinc, lead, and copper. The mining industry is the backbone of the State's economy. The coal fields are situated in the western part of the State, and the iron mines are situated in the eastern part. The mining industry has been the main source of wealth for the State since its settlement. The coal fields were discovered in the 18th century, and the iron mines were discovered in the 19th century. The mining industry has grown steadily since that time, and it is now one of the most important industries in the State. The coal fields produce a large amount of fuel for the West, and the iron mines produce a large amount of pig iron and steel. The State is also rich in other minerals, such as zinc, lead, and copper. The mining industry is the backbone of the State's economy.



Due to rigorous climate and extreme aridity, the natural vegetation of West Pakistan is mostly xerophytic and sparse. According to Champion's classification (Champion, 1936), the plains' vegetation is tropical thorn forest which preponderates in thorny leguminous species like Acacia spp. and Prosopis spp. Along the rivers the natural vegetation is composed of Tamarix articulata, T. dioica and Populus euphratica, which are the pioneer species on the silt deposits left annually by the rivers after the flood recedes. Considerable parts of these riverine areas have been converted to timber forests, mainly of Acacia arabica, through direct seeding. Irrigated plantations have also been established artificially in the plains, where sub-marginal land and irrigation water are made available. The primary species used is sissoo (Dalbergia sissoo) with understory of Morus. Subtropical dry evergreen forest in which Olea cuspidata and Acacia modesta are the dominant species occurs on the low hills and the outer Himalayas, from 2,000 to 4,000 feet elevation. Subtropical pine (Pinus longifolia) forest lies between 3,000 to 6,000 feet in the north-western Himalayas. Moist and dry temperate forests consist of coniferous species--Pinus griffithii (Himalayan white pine), Cedrus deodara (deodar), Abies pindrow (fir), Picea morinda (spruce), Juniperus macropoda (cedar) and

Due to rigorous climate and extreme aridity, the natural vegetation of West Pakistan is mostly xerophytic and sparse. According to Choudhury's classification (Choudhury, 1952), the climate of West Pakistan is semi-arid to arid. The natural vegetation of West Pakistan is mostly xerophytic and sparse. According to Choudhury's classification (Choudhury, 1952), the climate of West Pakistan is semi-arid to arid. The natural vegetation of West Pakistan is mostly xerophytic and sparse. According to Choudhury's classification (Choudhury, 1952), the climate of West Pakistan is semi-arid to arid.

with some scattered hardwoods--Quercus incana (oak) and Acer pictum (maple). They occur between 5,000 and 10,000 feet in the Himalayas. Alpine forests are primarily located from 10,000 feet to timber line, with Abies pindrow as the important timber species.

### Necessity for Tree Improvement in West Pakistan

West Pakistan is an extreme wood-deficit area. Its land area classed as forests is less than 4 percent. Even with the present low standard of living, it imports 5 million cubic feet of timber per year. Its population increases at the rate of 2 percent per year (F.A.O., 1961). Its economy is expanding fast and industries are growing rapidly.

To solve the problem of wood shortage either additional area should be brought under forests or the productivity of existing forests must be enhanced. The second approach is most feasible. The forests can be made more productive by silvicultural treatments in combination with genetically better planting stock

The importance of tree breeding in West Pakistan is obvious, in view of the growing demands of new industries. Total dependance on natural regeneration is giving way to large-scale artificial plantations. In

with some scattered individuals (mostly small) and

also along (small). They were between 2.00 and 10.00

feet in the distance, some of them were

located near the edge of the field, with

some of the larger trees.

THE UNIVERSITY OF CALIFORNIA  
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the plains vigorous efforts are being made to raise more irrigated plantations.

Another factor calling for the attention of tree breeders is the gradual degeneration of the gene pool in the natural forests of West Pakistan. The present genetically poor stock is the result of past selective logging of economically superior trees, leaving the inferior ones to breed. Such a situation is most common in the community owned coniferous forests. In establishing artificial plantations a frequent practice has been to collect seed from the inferior or isolated trees which are prolific seed producers (Champion, 1933). This malpractice continues today.

#### Previous Work and Achievements

Foresters in Indo-Pakistan subcontinent have been slow to appreciate the importance of tree breeding. H. G. Champion was probably the first man who drew attention to the subject. In 1925 he gave an account of the twisted fibre in the wood of chir pine (Pinus longifolia) and its possible inheritance. In 1930 he established the first provenance test at Dehra Dun to support his observations regarding racial differences in the wood of this native pine.

The plain vigorous efforts are being made to make more irrigated plantations.

Another factor which has the effect of increasing the production of the goods pool is the general degeneration of the goods pool in the present.

The general increase of West African The present generationally poor stock is the result of that selection.

logged of economically important trees, leaving the ground with no more than a few scattered trees.

As the number of trees per acre is reduced, the amount of fuel wood available is also reduced.

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The amount of fuel wood available is also reduced.

In 1929, thirty deodar (Cedrus deodara) and two chir pine trees of exceptional superiority in height, age, form and girth, were selected. A project was initiated in 1935 in the Punjab area to select a good race of sissoo (Dalbergia sissoo) which should combine a good stem form, shape, frost hardiness and a high timber quality. In 1941 (Anonymous, 1941), progeny from selected parent trees from the best forests of Hala Range in Hyderabad was raised in the Kathar Barrage area. Possibilities of improving teak (Tectona grandis) through ecotypic selection were also explored.

In 1947 the Pakistan Forest Research Institute came into being. Research in forest tree breeding received little emphasis, however. Cytogenetical aspects were studied briefly by M. I. Khan, who determined somatic chromosome number in Acacia spp. and hybrids (Khan, 1951). In 1954 the same author emphasized the urgent need of research in tree breeding. But it was not until 1961 that concerted efforts were made by the Silvicultural Branch of the Forest Research Institute to systematize the tree improvement work. A thorough search for superior germplasm was made in irrigated plantations. As a result, 140 sissoo and 27 mulberry trees were selected as 'plus trees.' Progenies raised from thirty of the selected sissoo trees are under study. In 1964, phenotypic

... in 1933, thirty books (English language) and two  
... of exceptional quality in English  
... and their work, which was published in 1933

In each book a series of notes (English language) which should contain a good  
... (English language) which should contain a good

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... (English language) and a high quality

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selection of 26 trees of Populus euphratica was made after a careful survey of the riverine forests in the southeastern region of West Pakistan. Similar work was initiated in 1965 in the northern coniferous forests. So far 65 chir pine and 26 Himalayan white pine plus trees have been selected (Khan, 1966).

In order to learn the techniques involved in hybridization, attempts have been made at the following crosses:

Pinus longifolia X P. canariensis (New Zealand)

Pinus griffithii X P. strobus (U.S.A.)

Pinus griffithii X P. flexilis (U.S.A.)

Pinus griffithii X P. monticola (U.S.A.)

Cedrus deodara X C. libanotica (Turkey)

An arboretum at Batrassi (approx. 3500 ft. elevation) was started in 1962. It contains a number of pine species from all over the world, besides other conifers and hardwood species.

#### Tree Breeding Goals

General goals of tree breeding in West Pakistan should be to investigate the genetics of all major species of economic importance and to increase their genetic potential for superior tree quality, growth and other important traits. Research on the reproductive

collected at 30 sites in the northern region of the study area. The results of the analysis are presented in Table 1. The results show that the mean values of the variables studied are significantly different from those reported in the literature for the same region. In particular, the mean values of the variables studied are significantly higher than those reported in the literature. This may be due to the fact that the study area is a high-altitude region with a high degree of isolation. The results also show that the mean values of the variables studied are significantly lower than those reported in the literature. This may be due to the fact that the study area is a high-altitude region with a high degree of isolation.

In order to test the hypothesis that the mean values of the variables studied are significantly different from those reported in the literature, a t-test was performed. The results of the t-test are presented in Table 2. The results show that the mean values of the variables studied are significantly different from those reported in the literature. This supports the hypothesis that the mean values of the variables studied are significantly different from those reported in the literature.

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processes and breeding techniques should supplement any tree improvement program.

There are a number of species in West Pakistan, both coniferous and hardwood, which are of commercial importance. Only a few species can be chosen initially. Choice should be based upon their economic importance and improvement potential. The following five species should be given first priority: Pinus griffithii, Pinus longifolia, Dalbergia sissoo, Acacia arabica, and Populus euphratica.

Pines are given top priority because of their great commercial importance in West Pakistan. Also Pinus is the chief coniferous genus of the northern hemisphere. Much genetic research work has been accomplished, particularly in the United States and Europe. Many of the methods and principles perfected in other countries can be used in Pakistan.

Pine breeding goals should be a high rate of growth in Pinus griffithii, improvement in oleoresin production and wood quality, and growth rate in P. longifolia.

Dalbergia sissoo is the major species of irrigated plantations and of the riverine forests of the former Punjab area of West Pakistan and is by far the most commonly used species all over the plains. Its major use lies in furniture and construction. Growth rate

process and feeding techniques should be reviewed and improved. There are a number of factors which are important in the design of a feeding program.

Both conditions and materials, which are of considerable importance. Only a few factors can be shown initially.

Choice should be based upon their economic importance and improvement program. The amount of feed eaten should be determined.

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and stem form are the main characters needing improvement. Stem crookedness is common in irrigated plantations. Also, sissou is highly susceptible to defoliator (Plecoptera reflexa) which sometimes kills trees outright. Varieties resistant to this insect are needed. Improvement may also be required in the wood quality of sissou.

Acacia arabica covers extensive areas in southern West Pakistan and is used for tannin, lumber, mine props and fuel. It is very frost tender. As one proceeds north, frost casualties increase; even large poles and young trees are killed back in cold years. Increased hardiness should be the primary improvement objective.

Populus euphratica should be included in the breeding program on account of its prospective value for pulp, plywood and matches. It has an extensive ecological range, from the hottest parts of West Pakistan plains to the upper limits of tree growth (Troup, 1921). Other species can be hybridized with Populus euphratica to produce a hybrid which would embody several desirable characteristics. Improvement should stress better growth rate, disease resistance and stem form.

Being an underdeveloped country, still building up its economy, Pakistan cannot afford to embark upon gigantic and money consuming projects at the very beginning of tree improvement program. Economical devices in breeding should be preferred at all the stages of the work.

and often form the main structure of the  
 stem or backbone of the plant.  
 Also, there is a large amount of  
 (the so-called) which sometimes kills the  
 plants resistant to this insect are needed. Improvement  
 may also be required in the wood quality of the  
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Pakistan is very short of trained and skilled personnel to carry out breeding work. Introducing a complex breeding technique would therefore not solve the problems. Lack of proper laboratory and equipment in the initial stages also call for simplicity of the approach.

The necessity for large quantities of superior wood has been emphasized. Along with long-term breeding methods, workers in the country must apply short cuts and depend much upon the methods which give early results.

Some Local Considerations Involved in the  
Implementation of a Tree Breeding  
Program in West Pakistan

Staff and facilities.--At present there is one U.N.D.P. (United Nations Development Project) expert, Dr. Mirko Vidakovic of Yugoslavia, working as an expert in genetics in the Pakistan Forest Research Institute at Peshawar. He will remain there for the next two years. I will join the Institute as a geneticist in April, 1967. It is expected that the number of permanent geneticists in the Institute will increase within the next ten years to four. Presently there is one technician with cross pollination experience. For immediate needs, four forest





rangers will be trained in breeding work. The training will consist of imparting philosophy of the work involved, and laboratory and field studies, for a period of nine months. Ten additional persons will be trained during the next years.

Approximately 30,000 man-hours of common labor per year will be available for the execution of tree improvement work.

Nursery and planting sites will be available on state-owned land. Approximately three man-months will be required for site preparation for each large experiment. This will include clearing of trees and shrubs, terracing (in the hills), furrowing, and levelling.

Existing laboratory equipment consists of controlled environment chambers, refrigerators, incubators, and electric computers. A modern green-house will be constructed in the near future with the help of U.N.D.P. funds.

Regional cooperative project.--Before the initiation of breeding studies, arrangements will be made to have a joint, cooperative program of research, particularly on provenances, with the tree breeders of Indian Forest Research Institute, Dehra Dun, for species of common interest to Pakistan and India. The cooperation may be on a non-governmental basis. It will involve free

workers will be trained in pressing work. The program will consist of imparting knowledge of the work process and laboratory and field practice. The duration of nine months. The additional periods will be trained during the work hours.

Approximately 10,000 man-hours of training

per year will be available for the expansion of work

improvement work.

Training of workers in the field

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exchange of material and data between the Pakistani tree breeders and their counterparts in India. Annual meetings will be held to discuss the past achievements and plans for the future. General pattern of this cooperative project will be similar to that of NC-51. If there would be a fair chance that such cooperation could be obtained after a few years, provenance studies in Pakistan will be delayed until the cooperation becomes effective.

A 10-Year Breeding Program  
for West Pakistan

The projects and experiments listed below are in the order of priority. A time schedule has been given only for the first experiment of this program. Experience and extent of success gained with every experiment will govern the time for the subsequent experiment. Again, local circumstances (availability of funds, labor and other facilities, unprecedented occurrences) would influence the times set for the experiments.

Project 1: Provenance studies of the three important species of West Pakistan.--All the three species picked for provenance studies, Pinus griffithii, P. longifolia and Dalbergia sissoo, occupy extensive ranges in the



Indo-Pakistan subcontinent. Although there is every reason to suspect the occurrence of racial variation in these species, little work has been done to explore such a possibility. The primary goal will be to determine the best ecotypes for planting in varied localities. Also, the experiments will be designed in such a manner as to provide breeding material in future hybridization studies.

Experiment 1A. To determine the extent and pattern of variation in *Pinus griffithii* and to select the most promising geographic and altitudinal ecotypes.--There are indications of interest on the part of NC-51 members in provenance tests of *Pinus griffithii*. J. J. Jokela, the chairman of that project will be approached to work out an arrangement utilizing PL-480 funds for execution of the experiment.

In general the study will follow the Michigan pattern (in this regard, Scotch pine study will serve as a model). In 1968 request will be made to district forest officers in West Pakistan, Afghanistan, India, Sikkim, Nepal and Bhutan, to provide seed from 10 or more average trees per stand. Hopefully it will be possible to obtain 100 such stand collections. Pertinent information on locality, altitude, date of collection, area, soil, age, height, diameter of trees will also be obtained. The seed will be stored dry at 40° F.

Indo-European environment. Although there is every reason to suspect the occurrence of racial variation in these species, little work has been done to explore this possibility. The present study will be to determine the best approach for planning a varied localities. Also, the experiments will be designed to show a method to provide genetic material to future population studies.

The present study is a preliminary investigation of the genetic variation in the population of the species. The results of this study will be used to determine the best approach for planning a varied localities. Also, the experiments will be designed to show a method to provide genetic material to future population studies.

The present study is a preliminary investigation of the genetic variation in the population of the species. The results of this study will be used to determine the best approach for planning a varied localities. Also, the experiments will be designed to show a method to provide genetic material to future population studies.

It is intended to have one centrally located nursery site and three sites for installing the plantation tests. The nursery will be established near Abbottabad. Planting sites will be located in Kaghan, Gallis, and Murree areas. The exact location of the sites will be decided after a thorough survey of the areas. It is desirable that the sites be uniform, above average in quality, and near the main road connecting the nursery and planting areas. All will be within 250 miles of Peshawar.

In November 1969, sowing will be done. Two weeks prior to sowing each seed lot will be given a "cold water stratification treatment" (Rudolph, 1950). Three weeks prior to sowing the seedbeds will be treated with the combination fungicide-weed seed disinfectant 'Mylone'. Fertilizer, if recommended by the soil chemist, will be applied. Supplementary experiments may be needed to determine chemicals more suited than 'Milon.'

Local practice of sowing three-foot-wide terraced hill nurseries, will be adopted. Each plot will consist of a single row running across the seed beds. Distance between the successive rows will be at least 12 inches. A randomized complete block design similar to that used in Michigan will be adopted. That will be easier to analyze than the lattice designs used at Gulfport.





In addition to the four replicates for nursery measurement, a fifth will be established to provide material for outplantings; this will consist of 8 x 3-foot plots sown at a density of 25 per square foot. Due to the abundance of cheap labor, these plots will be sown in rows, as at Placerville. Seeds will be sown half inches deep and covered with one third of an inch of sand. Mulching will be done by covering the beds with polyethylene film and then burlap until germination starts in the spring. About 50 percent high shade will be preferable until August of the first year. Seed beds will be covered with wire screen which will be removed after germination. Damping off, if it appears, will be controlled by "Captan." Control of white grubs will be exercised by the application of Dieldrin (at the rate of 1.5 pounds per acre); of cut worms by poison bait or D.D.T.

Adequate watering is extremely important. Frequency of watering will depend upon the site. Frequent shallower watering will be substituted by less frequent, deeper watering as the plants grow older.

Preparation of sites for outplanting will be completed by 1971. All the trees, scrubs and grasses will be cleared. Before planting a low volatile ester of 2,4,5-T, 3 parts concentrate in 100 parts of diesel



oil will be applied as a stump spray (White, 1965). In March of that year seedlings will be lifted for out-planting. The outplanting procedure and design will in general follow those used in Michigan for hand-established plantations. Planting stock will be shipped in a state truck driven by one of the professional geneticists, who can then stay at the planting site to supervise.

Planting will be carried out with open rooted stock, after pruning the roots and dipping them in liquid mud. After planting, Simazine (4 lbs. per acre in 100 gallons of water) will be sprayed over the newly planted trees. Application will be in coarse, low pressure (20-30 psi) water spray in 24-inch bands (White, 1965).

Due to uneven topography, it may not be possible to locate all replicates at the same place. They can be separated if the site conditions within a replicate remain as uniform as possible. The trees will be spaced 8 x 8 feet. Differences in visible traits will be measured in the nursery. Particular notice will be taken of mortality, growth, height, stem form, disease and frost resistance.

In order to determine whether the visible differences between the extreme varieties are due to chance or otherwise, the data will be subjected to analysis of



variance. Plot means will be used as items. Statisticians will be consulted to carry out detailed analyses.

All failures in the test plantings will be replaced in November 1972 with the onset of winter rains. No further replacement will be carried out.

The nursery experiment will probably be abandoned after 5 years, perhaps earlier. The outplantings will be maintained indefinitely although recent experience at the Michigan station indicates that they may yield 90 percent of their useful information by age 10. Probably detailed measurement will be made in the permanent outplantings at ages 5 and 10. Thereafter just enough measurements will be made to determine whether earlier conclusions remain true.

Experiment 1 B. Provenance test of *Pinus longifolia*.--The extent and pattern of variation in *Pinus longifolia* will be studied in a provenance test similar to that already outlined for *Pinus griffithii*. The nursery study will be located near Abbottabad. Permanent plantings will be established in Murree, Haripur and Siran forest divisions.

Experiment 1 C. Provenance test of *Dalbergia sissoo*.--The extent and pattern of variation in *Dalbergia sissoo* will be studied in a similar provenance

variance. Hot spots will be used as areas. Stations  
plans will be considered to carry out local projects.  
All facilities in the west will be used to  
expand in November 1972 with the onset of winter season.

No further expansion will be carried out.  
The primary expansion will be in the  
after 3 years, perhaps earlier. The total of expansion

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test. An existing nursery maintained by the Silviculture Research Division, Northern Zone, near Lahore, will be utilized for the nursery test. The planting sites, all located within 250 miles of the nursery site, are: Machu (Leiah), Shorkot (Multan), Chhanga Manga (Lahore), Peshawar.

Sowing will take place in March. Planting will be carried out with one year old sissoo stumps consisting of 3 inches of stem and 9 inches of root portion.

Project 2: Study of local variation in Pinus griffithii, P. longifolia, Dalbergia sissoo and Acacia arabica; establishment of seed orchards.--Experience indicates a good deal of local variation in these species. Phenotypic selection of superior trees in the local populations is being done intensively. Investigations to know the extent of local variation and heritability of useful characters, are necessary to find out the true value of phenotypic selections. Seed orchards are needed to produce improved seed for implementing the large scale planting programs in West Pakistan. The goals are to determine the best genotypes and the extent of heritability; to produce seed of a high genetic quality in the seed orchards.

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Experiment 2A. Open-pollinated, progeny test-seed orchards of Pinus griffithii.--The jack pine progeny test-seed-orchard project at Michigan station, will serve as a model.

Seed will be collected from about 400 selected trees by felling. Details of nursery treatment and planting given for experiment 1A will be followed. A 4-replicated randomized complete block design will be used in the Abbottabad nursery; a fifth replicate will be used for outplanting. Measurements will be made in the nursery intensively for five years and later, data will be used to establish juvenile-mature correlations.

Sites for the three permanent outplantings will be located in Murree, Kaghan and Gallies. Design of the plantings will be a randomized complete block; with 10 replicates, and 6-tree plots. Plantations will be measured at five-year intervals and parent-progeny relationships will be worked out. After each measurement, the plantings will be thinned to the best individuals in the best families.

Experiment 2B. Open pollinated, progeny test seed orchards of Pinus longifolia.--Details will be the same as for experiment 2A. A nursery will be located at Abbottabad. Plantings will be established in Murree, Haripur and Siran forest divisions..



Experiment 2C. Open pollinated progeny test-seed orchard of *Acacia arabica*.---Major details will be the same as for experiment 2A. The number of selected parents will be 60. A nursery maintained by the Silviculture Research Division, Southern Zone, at Miani research station, will be utilized for the nursery test and raising the planting stock. Sowing will be done in early February. A pretreatment consisting of soaking the seed in boiling water for 20-30 seconds and then allowing the water to cool for 24 hours, will be given. Sowing will be done from the seed of the same year.

Permanent plantings will be located at three above-average sites in the Hyderabad region. Planting will be done with balled two-year old stock.

Project 3: Hybridization of *Populus euphratica* with other species.---Hybridization in genus *Populus* has been an effective method of improvement. There are good chances that *Populus euphratica* will combine with many exotic poplars to produce progeny with hybrid vigor and other desirable traits. The primary goals are to produce hybrids combining ecological adaptability of the native *Populus euphratica* with good stem form of many of the exotic species; to produce the effects of hybrid vigor.



Experiment 3A. Populus euphratica X P. deltoides hybridization.--U.S.F.S. will be requested to send pollen from four selected trees of Populus deltoides; it will be stored in the refrigerator upon its arrival. In late December, about two weeks before the anthesis occurs in P. euphratica, 2 to 3 feet branches bearing female flowers will be shot down from the twenty selected individuals, in the Hyberabad region. They will be brought to moist atmosphere of the green house and grafted onto rooted stocks. Pollinations will be done in late January.

From four male parents and twenty female parents, eighty combinations will be obtained. Seed will be kept separate for each combination. Progenies kept separate by families will be raised in the experimental nursery at Peshawar. Individuals from different progenies will be compared for height growth. In order to evaluate the advisability of juvenile selection of hybrid progenies, 25 largest and 25 smallest trees from every progeny will be planted in a randomized block-split plot design.

The project will be continued after the first phase and systematic hybridization experiments with the important species will be carried out. Tests for the effectiveness of clonal selection will be a major part of the project.



Developing Project 4, Determining effective pollination techniques in Acacia arabica and possibilities of inter-specific hybridization.--Some of the exotic Acacias are known for their hardiness and high yield of tannin. By hybridization it is possible to combine these traits with the site adaptability of the native Acacia.

Experiment 4A, Pollination techniques for Acacia arabica.--In Acacia emasculation damages the tender buds so much so that they soon drop. Hot weather coupled with wind and storms also take heavy toll of the flowers. Emasculation and pollination under varied conditions of green house will be made to find a solution to these problems.

Future hybridization experiments will depend upon the perfection of pollination techniques in Acacia. If successful techniques can be evolved, the following crosses will be made during the next phase of the breeding program:

For frost resistance: A. Arabica X A. modesta,  
A. jacquemontii and A. Catechu.

For drought resistance: A. Arabica X A. modesta,  
A. farnesiana, A. senegal, and A. jacquemontii.

For higher yield of tannin: A. arabica X A. modesta.

Project 5, Pollination techniques in Dalbergia sissoo and intra- and interspecific hybridization.--

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Development of pollination techniques is necessary for testing 2-parent progenies and combining desirable traits in intra- and interspecific hybrids. No hybridization has been attempted on sissoo. Due to self pollination, sissoo trees occur in 'pure lines.' There is thus a chance of producing hybrid vigor in the progenies by effecting cross pollination.

Experiment 5 A. Pollination techniques for Dalbergia sissoo.--During early April before natural pollination occurs, 30 flowers will be opened and emasculated. There will be no need of isolation by pollination bags because of the self-pollinating habit. There will be three pollination treatments: (1) rubbing split anthers directly over the stigma (10 flowers), (2) brushing preserved pollen over the stigma (10 flowers), and (3) none.

In developing a pollen forcing procedure, branches bearing flowers will be collected two weeks before anthesis occurs. They will be brought to the green house and pistils will be removed. Two treatments will be attempted: (1) grafting onto the rootstocks and (2) keeping the lower ends in the water.

Future hybridization experiments will depend upon perfection of pollination techniques in sissoo. If successful techniques can be evolved, the following



interspecific crosses will be made later: Dalbergia  
sissoo X D. latifolia, and D. sissoo X D. cochinchinensis.

Future breeding work.--The above program will be only a first step for the tree improvement in West Pakistan. After this preliminary stage is successfully completed, it can be expected that information regarding the provenances, individual tree variation, heritability and cross pollination in respect to particular species will enable us to commence advanced breeding work. During the next phase, 2-parent progeny tests designed on the Gulfport pattern will be used for acquiring more knowledge of inheritance. Inter-racial crosses will be attempted to combine different desirable traits of ecotypes. Intra-specific crosses will also be made.

Interspecific hybridization of pines will be confined to the production and performance of hybrids involving the two native species only. The pine crossability pattern worked out at Placerville will be used to establish breeding arboreta consisting of the species most likely to cross with the two native pines. Pronounced hybrid vigor has been demonstrated in the following crosses in the United States (Wright, 1962): Pinus griffithii X P. strobus, Pinus griffithii X P. monticola, and Pinus flexilis X P. griffithii. A successful cross between Pinus longifolia and P. canariensis has also been reported (Liddicoet

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and Richter, 1960). Efforts will be made to obtain the  
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the project.

APPENDIX





## APPENDIX I

### NC-51 PROJECT

NC-51 project was started in 1960 with the support of federal funds allocated under the Hatch Act, 1886. There are 10 state agricultural experiment stations of the north central states as well as the Rocky Mountain and North Central Forest Experiment Stations, of the United States Forest Service, involved.

The project is an excellent example of cooperation amongst various agencies in the field of forest genetics. The representatives from the above-mentioned agencies meet once a year, discuss the progress of work and decide about the future goals and plans. The rigidity of rules is observed in the matters of technical cooperation. Nevertheless, various members are free to publish the findings independently.

In its first phase, that is 1960-1965, NC-51 project consisted of a series of provenance studies which aimed at providing information on the broad genetic variation patterns in the major forest tree species. Range wide tests were initiated on 19 species; tests organised by other agencies on 6 additional species were also

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WORLD-PI PROGRESS

WORLD-PI PROGRESS was started in 1950 and has since

been a steady growth in the number of members and in the

amount of money raised for the benefit of the poor and

the needy in all parts of the world.

The organization has a wide membership and is active in

many countries and is a member of the United Nations.

It is a non-profit organization and its funds are used

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participated by several of the NC-51 states. Important species under investigations have been red pine, eastern white pine, Scotch pine, ponderosa pine, Japanese larch, balsam fir, white fir, Douglas-fir, eastern cottonwood and red oak. The experiments are in various stages of establishment and are continued in the next phase. The overall objectives have been extended from provenance tests to other aspects of forest genetics. A summary of these objectives is reproduced below:

- (a) Determine the range and pattern of genetic diversity within selected forest tree species
- (b) Determine the physiological and evolutionary mechanisms responsible for growth and morphological differences
- (c) Develop useful hybrids within and between species by crossing selected races, utilizing material in existing provenance tests as parents. Where possible develop methods of stimulating early fruit production to facilitate the breeding
- (d) Establish the usefulness of juvenile performance data and shorten testing periods by means of juvenile-mature correlations and theoretical analysis.

Regional cooperation in the field of forest genetics as envisaged by NC-51 project, is necessitated due to several factors; listed by Wright (1965), they are:

participated by several of the 10-11 classes. Important  
specimens under investigation were from the pine, eastern  
white pine, Scotch pine, Douglas fir, Japanese larch,  
Douglas fir, white fir, Douglas-fir, eastern white pine  
and balsam. The experiments are in various stages of  
completion and are continuing in the laboratory. The  
overall objectives have been achieved and the results  
tests to date indicate that the objectives were met.  
There are no further specimens to be collected.  
The specimens are being prepared for the  
division and will be available for the  
division.

First, all species designated for improvement are being planted in several states. Second, the amount of improvement needed is too great to be handled by a single agency. Third, little of this type of research has yet developed to the point where the most important answers are applicable to specific localities. Most overlap state lines. Fourth, a coordinated approach has proved fruitful in this and other regions.

#### Value of NC-51 to Michigan State University

Michigan State University Forest-Genetics branch is a small organization with too limited resources to carry out large-scale experiments. Its participation in NC-51 project has made it possible to embark upon gigantic studies, a classical example of which is Scotch pine provenance test. Without the cooperation of other states, perhaps, it would not have been possible to test the species over a varied number of sites and gather a wealth of extremely useful information with regard to the geographic pattern and performance of different seed origins.

Besides the material benefits, several useful technical lessons have been learned as a result of a joint approach. For example, desirability of having a rangewide coverage, relatively intense sampling and field tests over a wide variety of sites, has been realized.

The following information was obtained from the files of the  
 Bureau of the Federal Bureau of Investigation, Department of Justice,  
 and the Bureau of the Federal Reserve System, Department of the Treasury,  
 in connection with the investigation of the activities of the  
 Communist Party, United States of America, and its various  
 branches and chapters in the United States and its possessions.  
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THE FEDERAL BUREAU OF INVESTIGATION OF THE DEPARTMENT OF JUSTICE

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 Bureau of the Federal Reserve System, Department of the Treasury,  
 in connection with the investigation of the activities of the  
 Communist Party, United States of America, and its various  
 branches and chapters in the United States and its possessions.

A previous practice in NC-51 experiments had been to raise the stock to plantable size and then distribute it for immediate planting. There were timing difficulties and seedlings which did well under some conditions did not perform well under others. Lining out of the stock for one year prior to outplanting has lead to far better results.

It has been further learned that, to find out the details of variability pattern, 90 to 95 percent survival and a start of all the species under weed free and uniform conditions is a must.

Small plots, many replicated designs have proved to be more than satisfactory in a project like the NC-51, where too many origins and too many localities are involved.

#### Contribution of Michigan State University to NC-51

Michigan State University has all along been a very active member of the NC-51 project. Besides the fact that measurements and analyses for many of the species have been accomplished at this station, Michigan State University has distributed planting stock of a number of species to other NC-51 agencies for the purpose of testing and pooling down the results. Michigan has been the initiating agency for the provenance studies of the following species: red pine, eastern white pine, Scotch pine, European black pine, ponderosa pine, limber pine, Japanese larch. In

A previous practice in NO-2 experiments had been to raise the stock to planktonic size and then transfer it to immediate plankton. This was done in the offices and the media were held under aseptic conditions for not greater than 24 hours. It has been found that the best results are obtained when the media are held for not more than 24 hours prior to introduction to the culture.

It has been found that the best results are obtained when the media are held for not more than 24 hours prior to introduction to the culture. The media are held in a sterile container and are transferred to the culture medium as soon as possible. The media are held in a sterile container and are transferred to the culture medium as soon as possible. The media are held in a sterile container and are transferred to the culture medium as soon as possible.

The media are held in a sterile container and are transferred to the culture medium as soon as possible. The media are held in a sterile container and are transferred to the culture medium as soon as possible. The media are held in a sterile container and are transferred to the culture medium as soon as possible.



improving the methodology of experiments, Michigan has formulated some useful rules for the measurement and record keeping which have benefitted other members of the project as well. Another useful device perfected at Michigan has been that of machine planting.

improving the technology of equipment, including the  
development of some new types for the measurement of  
to measure the rate of change of the  
to measure the rate of change of the  
to measure the rate of change of the

APPENDIX II

Pollination Record Form Used at the  
Institute of Forest Genetics,  
Placerville

19\_\_\_\_

Experiment number \_\_\_\_\_ Seed parent \_\_\_\_\_

Flowers bagged \_\_\_\_\_ Maturity \_\_\_\_\_ Disbagged \_\_\_\_\_

Cones bagged \_\_\_\_\_ Collected \_\_\_\_\_

Tag number	Number of flowers	Pollination data				Crop	
		Pollen parent, with year of pollen collection	Pollinations			Cones bagged	Cones collected
			Date	Maturity	By		

Experiment 11  
Elimination Reactions  
Acetate of Isopropyl Alcohol  
Alcoholic

Experiment number: 11  
Name: \_\_\_\_\_  
Date: \_\_\_\_\_  
Section: \_\_\_\_\_

Time	Observations
0 min	Reaction mixture is colorless.
5 min	Reaction mixture becomes cloudy.
10 min	Reaction mixture becomes more cloudy.
15 min	Reaction mixture becomes very cloudy.
20 min	Reaction mixture becomes very cloudy.
25 min	Reaction mixture becomes very cloudy.
30 min	Reaction mixture becomes very cloudy.
35 min	Reaction mixture becomes very cloudy.
40 min	Reaction mixture becomes very cloudy.
45 min	Reaction mixture becomes very cloudy.
50 min	Reaction mixture becomes very cloudy.
55 min	Reaction mixture becomes very cloudy.
60 min	Reaction mixture becomes very cloudy.

Accession number

////////////////////

APPENDIX III

SOUTHERN FOREST EXPERIMENT STATION

LIVING PLANT MATERIAL ACCESSION RECORD

Type of material (seed, pollen, ramet, if other, specify)

Species or hybrid \_\_\_\_\_

Pedigree (if applicable) \_\_\_\_\_

Supplier's name and address (if plant material has been sent to Southern Station by outside address) \_\_\_\_\_

Supplier's identification of plant material \_\_\_\_\_

Amount of material collected \_\_\_\_\_

Source: Lat. \_\_\_\_\_ Long. \_\_\_\_\_ State \_\_\_\_\_ County \_\_\_\_\_ Range \_\_\_\_\_

Township \_\_\_\_\_ Section \_\_\_\_\_ Elevation \_\_\_\_\_ Land grant \_\_\_\_\_

Detailed directions: \_\_\_\_\_

Aspect \_\_\_\_\_ Slope \_\_\_\_\_ Soil \_\_\_\_\_

Physiography (floodplain, upland, ridge, etc.) \_\_\_\_\_

Planted or natural stand (seed origin, if planted) \_\_\_\_\_

Stand history \_\_\_\_\_

Size of collection area \_\_\_\_\_

Number of plants collected from \_\_\_\_\_

Estimate of average age \_\_\_\_\_

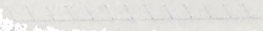
Associated species \_\_\_\_\_

Comments \_\_\_\_\_

Collected by \_\_\_\_\_ Collection date \_\_\_\_\_

Intended use (study no. or other) \_\_\_\_\_

Accession number



III

NOITAKI PULMINEKZ TEREKOT BENTIVOR

NOITAKI PULMINEKZ TEREKOT BENTIVOR

Type of material

Species of plant

Location

Collector

Date

Number

Notes

Remarks

Other

Disposition

Remarks

Other

Disposition

Remarks

Other

Disposition

Remarks

Other

Disposition

Remarks

This form should be filled out whenever plant material of any type is collected or received. If the material is to be used in a specific study original and 1 copy are required. If a specific study is not involved the original only will suffice. A Southern Station Accession Number should be assigned at this time. This number consists of 2 letters and 10 numerals as follows:

1. A 2-letter, 4-digit designation of the Southern Station Project that is collecting or receiving the plant material (S01801, S01401, etc.). This part of the number is already on the form.
2. A 2-digit designation year of collection or receipt (66, 67, etc.).
3. A 4-digit number within a group of number assigned to the collector by the project leader. Within this group, the numbers may be consecutive or they may conform to some special scheme called for by a particular study.

Example: /S/O/1/4/0/1/6/6/2/0/2/1

This was the twenty-first collection made in 1966 by the man assigned accession numbers 2000-2999; Institute of Forest Genetics, Southern Forest Experiment Station.  
ALWAYS FILL IN ALL 12 SPACES.

All different collections from a given parent or ortet should be assigned separate lot numbers.

Outgoing shipments of the plant material recorded on this form should be accompanied by a copy of this form.

Filing.--If a collection is made as part of a study, the original should be kept in a central project file and the copy with the other records for that particular study. If the collection is made not as part of a study the original should be kept in the central project file (no copy is necessary in this case). The central project file should categorize the accession forms by:

- I. Type of material (seed, pollen, ramet, or other)
  - A. Year of collection
    1. Species
      - a. Increasing order of accession number





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EXHIBIT 100

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Report of the Selective Service System, 1964-1965

Austin, Texas  
1965

Report of the Selective Service System, 1964-1965

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UNITED STATES AND ITS APPLICABILITY TO WEST PAKISTAN  
CONDITIONS

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