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(12) **United States Plant Patent**
Beineke(10) **Patent No.:** US PP17,125 P3
(45) **Date of Patent:** Oct. 3, 2006(54) **BLACK WALNUT TREE NAMED "BEINEKE 12"**

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PP9,925 P	6/1997	Jones
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PP14,839 P2	6/2004	Beineke
PP14,978 P2	7/2004	Beineke
PP15,079 P2	8/2004	Beineke

(50) Latin Name: *Juglans nigra L.*
Varietal Denomination: Beineke 12(75) Inventor: **Walter F. Beineke**, West LaFayette, IN
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(*) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 14 days.

(21) Appl. No.: **10/888,734**(22) Filed: **Jul. 9, 2004**(65) **Prior Publication Data**

US 2006/0010537 P1 Jan. 12, 2006

(51) **Int. Cl.**
A01H 5/00 (2006.01)(52) **U.S. Cl.** **Plt./154**(58) **Field of Classification Search** Plt./154
See application file for complete search history.(56) **References Cited**

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(57) **ABSTRACT**

A new and distinct cultivar of black walnut tree (*Juglans nigra* L.) which is distinctly characterized by extremely rapid growth rate, and excellent central stem tendency, thereby producing good timber qualities. This new variety of black walnut trees was discovered by the applicant near South Raub, Tippecanoe County, Ind. in a black walnut planting of seedling progeny from previously selected trees for outstanding timber production potential. This selection has been designated as BW509, a seedling progeny of BW 95 (unpatented) in records maintained by the applicant on the performance of this selection, and grafts made from the selection, and will be known henceforth as 'Beineke 12'.

3 Drawing Sheets**1**Latin name of the genus and species: *Juglans nigra* L.
Variety denomination: 'Beineke 12'.

BACKGROUND OF THE INVENTION

This new variety of black walnut tree (*Juglans nigra* L.) was discovered by the applicant near South Raub, Tippecanoe County, Ind. in a black walnut planting of seedling progeny from previously selected trees for outstanding timber producing potential. This selection has been designated as BW509, a seedling progeny of BW 95, unpatented, in records maintained by the applicant on the performance of this selection, and grafts made from the selection and will be known henceforth as 'Beineke 12'. The male parent is

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unknown, as is generally the case with black walnut trees. (Beineke, 1989)

SUMMARY OF THE INVENTION

A new and distinct cultivar of black walnut tree (*Juglans nigra* L.) which is distinctly characterized by extremely rapid growth rate, excellent resistance to anthracnose leaf disease, excellent central stem tendency, and excellent straightness, thereby producing excellent timber qualities, the trait of commercial interest. 'Beineke 12' was 9 years old when described at a location near South Raub, Ind.

After the original clone was selected, and assigned an identity number of BW509 the aforesaid tree was repro-

duced by collecting scions from it and grafting these onto common black walnut rootstocks at West Point, Ind. These asexual reproductions ran true to the originally discovered tree and to each other in all respects. A growth comparison between BW95 and 'Beineke 12' could not be made at this site because BW95 was not planted at South Raub or West Point and the original tree was cut down. However, since form is not site dependent, old form rating of BW95 on other sites can be used.

Color values used were from the Munsell Color Chart for Plant Tissues. However, color is too dependent on weather conditions and fertilization to be consistent or distinctive. For example, leaves can be made a deeper green by applying nitrogen. Walnut tree leaves turn yellow as the season progresses, especially if there is a lack of rainfall. As black walnut meats dry, they become darker. Simply being on the ground for a week causes the outer shell to darken. Bark color involves many shades of gray through brown and black.

'Beineke 12' is hardy un USDA zones 4,5,6,7, and 8.

DESCRIPTION OF THE DRAWINGS

FIG. 1 is a photograph showing the timber form of 'Beineke 12'.

FIG. 2 is a photograph showing the leaves of 'Beineke 12'.

FIG. 3 is a photograph showing the nuts of 'Beineke 12'.

BOTANICAL DESCRIPTION OF THE PLANTS

The botanical details of this new and distinct variety of walnut tree are as follows:

Tree:

Size.—Large, 36 ft. at 9 years; crown diameter of 15 ft.
Vigor.—Vigorous.

Growth rate.—Very rapid, 34% larger in diameter than the average of 'Purdue 1' (U.S. Plant Pat. No. 4,543) grafts, planted the same year on the same land. Diameter growth rate (at 4½ feet above the ground) averages 0.770 inches per year over 9 years, was 6.9 inches at 9 years.

Form.—Excellent timber form, 59.1% better than average of the entire planting and the same as 'Purdue 1'. Stem form was obtained by subjectively rating the straightness of the main stem on a scale of 1 to 5 with 1 representing a perfectly straight stem; 2, a slight crook or deviation of the central stem; 3, about average straightness; 4, several severe crooks or a single fork; and, 5, a very crooked, forked and/or leaning central stem. 'Beineke 12' averages 1 on the 1 to 5 scale, while parent BW95 averaged 3 on another site.

The trees of the present invention are grown in plantations, not in open fields (not natural stands). In plantations, trees are upright and have no distinctive or characteristic crown shape because all branches are seeking to grow upwards.

Branches: Diameter depends on age and size of tree, varies from ½" to 12", bark color varies from grays to browns.

Leaves:

Compound leaves.—Size — Large; average length — 17.55"; width 8.73". Compared to 'Purdue 1' (Plant Pat. No. 4,543), the leaves of 'Beineke 12' are

longer. 'Beineke 12' averages 3.05 inches longer than 'Purdue 1'.

Leaflets.—Size — Average; average length — 4.43"; average width 1.45", average number of leaflets — 18.7 — lanceolate; acutely pointed, rounded base. The leaflets of 'Beineke 12' are 0.06 inches longer and 0.05 inches narrower than 'Purdue 1'. 'Purdue 1' has an unusually long, narrow leaflet compared to most other black walnuts as does 'Beineke 12.' 'Beineke 12' averages 1.5 fewer leaflets than 'Purdue 1'. Leaflet number appears to be a consistent trait within tree and year to year.

Thickness.—Thin.

Texture.—Smooth.

Margin.—Serrated.

Petioles.—Short.

Color.—Topside — dark green (5GY3/4 by the Munsell Color Chart for Plant Tissues). Underside — light green (5GY5/4 on the Munsell Color Chart for Plant Tissues).

Anthracnose resistance.—Excellent.

Nut:

Size.—Large; average length — 1.4"; average diameter in suture plane — 1.18"; average diameter cheek to cheek — 1.46."

Uniformly of size.—Not much variation.

Form.—Rounded; flattened in suture plane. See FIG. 3.

Blossom end.—Pointed, acute.

Basal end.—Flat.

Thickness of shell.—Thick.

Ridges.—Sharp.

Color.—Mottled, 5YR3/2 and 2.5YR3/4 by the Munsell Color Chart for Plant Tissues. The nut of 'Beineke 12' averages 0.35 inches shorter than 'Purdue 1' (Plant Pat. No. 4,543). 'Beineke 12' averages 0.06 inches wider in the suture plane and 0.21 inches wider cheek to cheek than 'Purdue 1'.

Nut with husk:

Size.—Medium; average length — 2.46". Average suture plane width — 2.16"; average cheek to cheek width — 2.32".

Husk thickness.—0.92 inches.

Form.—Almost round except basal end protruding.

Blossom end.—Rounded, slightly indented.

Basal end.—Protruding.

Surface.—Smooth, not waxy, slight warts.

Color.—Greenish-yellow, 2.5 GY 7/10 by the Munsell Color Chart for Plant Tissue. The nut in the husk of 'Beineke 12' is about the same length as 'Purdue 1' (Plant Pat. No. 4,543). 'Beineke 12' averages 0.29 inches wider in the suture plane and 0.26 inches wider cheek to cheek than 'Purdue 1.' The husk of 'Beineke 12' averages 0.17 inches thicker than 'Purdue 1'.

Flowering habit:

Age at which trees start producing catkins.—Early, 5 years.

Number of catkins produced.—Few.

Age at which trees start producing pistillate flowers.—Early, about 6 years.

Number of pistillate flowers produced by young trees.—Few.

Lateral shoots producing pistillate flowers.—None.

Number of pistillate flowers per inflorescence.—2.

Flower season: Flowers typically in May in Indiana. There are probably 1-million pollen per catkin. Female flowers

are about $\frac{1}{16}$ " long and grow to two "pollen pick up points" which subsequently break apart. Pollen exists as "dust" which is not feasible to quantitate.

Nut crop:

Bearing.—Biennial.

Productivity.—Low.

Ripening period.—Middle October.

Evenness of maturity (period between first and last nuts are ready for harvest).—Even.

Quality.—Good.

Distribution of nuts on tree.—Throughout.

GENETIC METHOD OF IDENTIFICATION

DNA "fingerprint" for identification of 'Beineke 12':

DNA was isolated from the leaves of 'Beineke 12'. For purposes of DNA fingerprinting, eleven highly polymorphic loci from a suite of microsatellites developed by Woeste et al. (2002) were chosen. Microsatellites sizes were checked against previously published standards and verified by a second independent analysis. The "fingerprint" is the collection of microsatellite allele sizes at each locus for 'Beineke 12'.

DNA was isolated from the leaves of 4 black walnut trees obtained from Walter Beineke using CTAB extraction buffer (50 mM TRIS-HCL, pH 8.0, 20 mM EDTA, pH 8.0, 0.7 M NaCl, 0.4 M LiCl, 2% SDS, 2% CTAB, nd 1% PVP). After isolation the DNA from each tree was quantified and diluted with manopure distilled water to a final concentration of 5 ng/microliter. The samples were stored in 96-well plates at -20 degrees C.

For purposes of DNA fingerprinting, eleven highly polymorphic loci from a suite of microsatellites developed by Woeste et al. (2002) were chosen. Amplification of each locus was performed with an MJ Research Tetrad Thermocycler (Waltham, Mass.) using 10 microliter reactions in 96-well plates. The PCR reaction mix contained 2 microliters of the aforementioned black walnut DNA, 5 microliter Sigma Taq ReadyMix (Sigma Aldrich, St. Louis, Mo.), 0.4 microliter of a 20 pmol mixture of forward and reverse fluorescence labeled primer, and 3 microliter PCR grade water supplied with the Sigma ReadyMix. PCR amplification was for 30 cycles of 94 degrees C. for 20 sec, 55 degrees C. for 30 sec, and 72 degrees C. for 1 min. All primers were annealed at 55 degrees C. The products were then held at 4 degrees C. until aliquots could be loaded into 6% Long Ranger (polyacrylamide) denaturing gels (BMA, Rockland, Me.). For each individual 0.5 microliter PCR product was added to 0.75 microliter blue dextran and 0.25 microliter of CXR 350 bp Ladder Standard (Applied Biosystems, Inc., Foster City, Calif.) in a new 96-well plate. The samples were denatured for 2 min at 95 degrees C. and loaded onto a CAL96 96-well laminated membrane comb (The Gel Company, San Francisco, Calif.). Electrophoresis was at 3,000 V, 60 mA, 200 Watts, 50 degrees C. for 2 hours using an ABI 377 (Perkin Elmer) with 36 cm plates and 0.2 mm spacers. The resulting data was analyzed using ABI's GeneScan 3.1.2 and Genotyper 2.5 (Perkin Elmer). Microsatellite sizes were checked against previously published standards and verified by a second independent analysis. The "fingerprint" is the collection of microsatellite allele sizes at each locus for each tree.

Locus	Forward (SEQ ID NOS: 1–10)
WGA6	CCATGAAACTTCATGCGTTG
WGA24	TCCCCCTGAAATCTTCTCCT
WGA27	AACCCCTACAACGCCTGATG
WGA32	CTCGGTAAGGCCACACCAATT
WGA72	AAACCCACCTAAACCCCTGCA
WGA89	ACCCATCTTCACGTGTGTG
WGA90	CTTGTAAATGCCCTCTGCTC
WGA97	GGAGAGGAAAGGAATCCAAA
WGA69	TTAGTTAGCAAACCCACCCG
WGA76	AGGGCACTCCCTATGAGGT
WGA82	TGCCGACACTCCTCACTTC
Locus	Reverse (SEQ ID NOS: 11–22)
WGA6	CATCCCAGCGAAGGTTG
WGA24	TTCTCGTGGTGCTTGTGAG
WGA27	TGCTCAGGCTCCACTTCC
WGA32	ACGGGCAGTGTATGCATGTA
WGA72	ACCCATCCATGATCTTCAA
WGA89	TGCCTAATTAGCAATTCCA
WGA90	TACCTGCAACCGTTACACA
WGA97	TTGAACAAAAGGCCGTTTC
WGA69	AGATGCACAGACCAACCTC
WGA76	CAGTCTCATCCCTTTTCC
WGA82	CGTGATGTCGACGGCTG

The best interpretation of the current data indicates that the probability that any other black walnut tree would have the collection of microsatellite allele sizes listed is estimated to be less than 3×10^{-14} .

Sizes (bp) of microsatellites at 11 loci used to fingerprint 'Beineke 12' (2 alleles at each locus).

Microsatellites used to fingerprint 'Beineke 12':

WGA6	WGA24	WGA27	WGA32	WGA72
142	142	238	242	229 233 175 183 149 149
WGA89		WGA90		WGA97
199	199	152	158	155 155
WGA69		WGA76		WGA82
172	176	232	236	180 196

DOCUMENTS CITED

Beineke, Walter F. (1989) Twenty years of black walnut genetic improvement at Purdue University *North. J. Appl. For.* 6:68–71.

Woeste, K., Burns, R., Rhodes, O., and Michler, C. (2002) Thirty polymorphic nuclear microsatellite loci from black walnut. *Journal of Heredity.* 93:58–60.

SEQUENCE LISTING

<160> NUMBER OF SEQ ID NOS: 22

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<212> TYPE: DNA

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<210> SEQ ID NO 3

<211> LENGTH: 20

<212> TYPE: DNA

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<400> SEQUENCE: 3

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20

<210> SEQ ID NO 4

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<212> TYPE: DNA

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<400> SEQUENCE: 4

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<212> TYPE: DNA

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<212> TYPE: DNA

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20

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<210> SEQ ID NO 8

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<212> TYPE: DNA

-continued

-continued

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<211> LENGTH: 20
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<210> SEQ ID NO 20
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<212> TYPE: DNA
<213> ORGANISM: Juglans nigra

<400> SEQUENCE: 22

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I claim:

1. A new and distinct variety of black walnut tree named 'Beineke 12' substantially as illustrated and described,

which has excellent timber quality, extremely rapid growth rate, and excellent central stem tendency.

* * * * *



FIG. 1

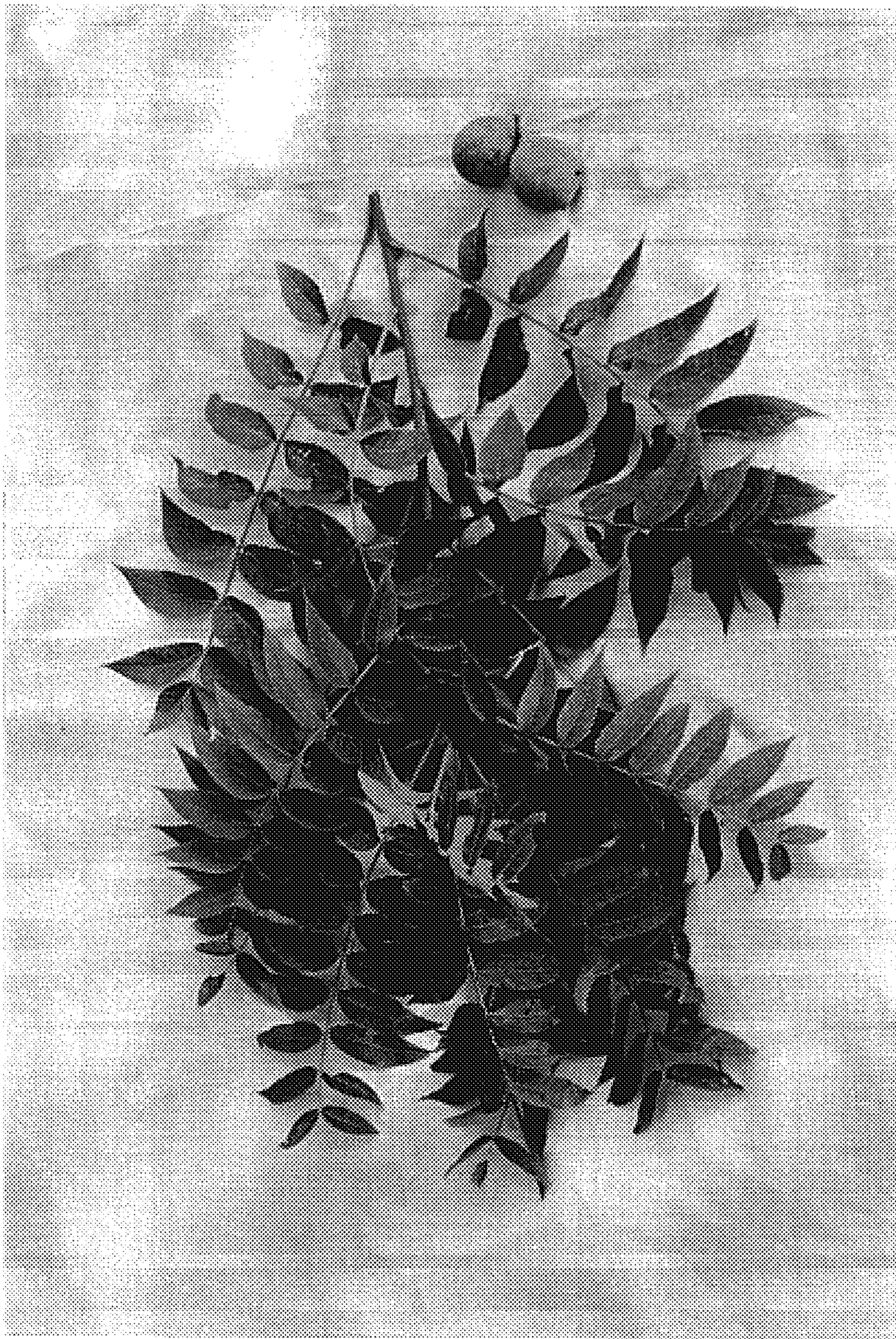


FIG. 2

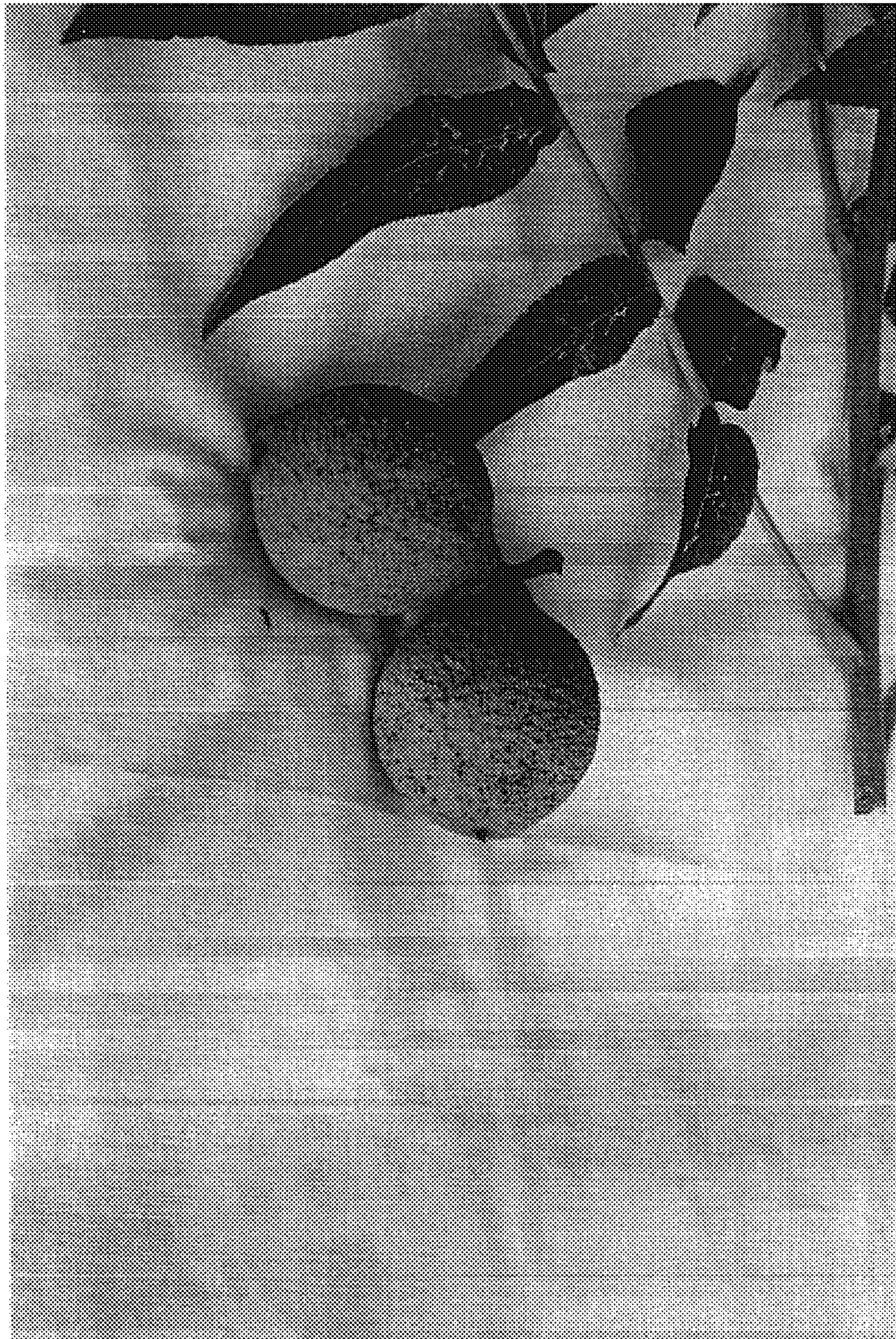


FIG. 3