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The Effects of Inbreeding in Black Walnut

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Introduction

Black walnut (*Juglans singra* L.) is not an abundant tree species in the eastern deciduous forest, but tends to occur in groups or as scattered individuals on good sites. Since colonial times, not only have black walnut trees been harvested for manufacturing purposes, but land clearing throughout the species' natural range has reduced its domain to small, isolated tracts, often less than 40 acres in size. Thus, the remaining black walnut population has suffered a reduction in effective breeding size by virtue of reduction in the number of breeding individuals within a stand and the likely inability of trees between isolated woodlots to interbreed.

Walnut trees probably occurred every few hundred feet in most deciduous forests of the Central States as it does today in the few remaining uncut woodlots. Therefore, it is probable that gene flow could readily occur among trees within stands and between nearby stands until recent times when cutting destroyed the pattern of continuous forest.

Is pollen dispersal distance great enough to overcome the wide spatial gaps in the breeding population? Black walnut is wind-pollinated and pollen is theoretically capable of traveling long distances; however, viable black walnut pollen grains are larger and heavier than most other wind pollinated hardwood or herbaceous species (1, 14). Thus, black walnut pollen may not travel as far as the pollen of some other anemophilous species. Persian walnut pollen may be carried up to one mile from its source, but its effective distance is only 200 to 300 feet (7). Wood (15) calculated that a stigma of Persian walnut in the lower branches of a tree would receive approximately 192 pollen grains while a stigma 500 feet from the pollen source under ideal conditions would receive only 24 grains, and a stigma at one-half mile would receive no pollen.

The viability of freshly dehisced pollen from 78 individual black walnut trees over a 3 year period averaged only 34 percent and ranged from 0 to 90 percent (8). Pollen grains that fail to germinate appear to be devoid of protoplasm, and, therefore, are probably lighter in weight. These dead pollen grains would tend to carry farther from their point of origin than viable grains.

Weather conditions cause dichogamy to vary from year to year; however, overlap of pollen maturation with female flower receptivity occurred in 41 percent of 34 black walnut trees during a spring of reasonably normal weather (8). As a result of these findings, selfing could occur in natural black walnut stands.

One factor that may reduce selfing is that excessive pollen on the stigmas inhibits normal fertilization. Beineke and Masters (2) found that only 8 percent of female flowers having excessive amounts of pollen applied developed viable seed, while 66 percent of wind-pollinated flowers produced seed. Thus, female flowers below ripening catkins on the same tree would be overwhelmed by copious amounts of pollen and abscise.

Considering all of these factors, if breeding among scattered stands is not occurring, is the forced breeding among closely related individuals within a stand detrimental? Inbreeding depression has been confirmed in a wide variety of other woody plant families. For example, in the Pinaceae, all genera show growth depression and reduced vigor of inbred seedlings (6, 12). Inbreeding depression has also been found in species related to black walnut. In pecan (*Carya illinoensis* [Wangenh.] K. Koch), inbred seedlings were 23 percent shorter than cross-pollinated seedlings produced by the same tree (10). Serr and Forde (11) found 3 of 45 selfed Payne cultivar Persian walnut (*Juglans regia* L.) seedlings were dwarfed, low in vigor, and susceptible to winter die-back. In general, the selfs had fewer desirable qualities and lower vigor than Payne crossed with other cultivars.

Selfing and related matings have been carried out in black walnut and Persian walnut (1, 15). In black walnut Beineke (1) found that forced selfing produced 22 percent seed set, while controlled-pollinated outcrosses produced 18 and wind-pollinated 49 percent seed set. In later studies, related matings (backcrosses) resulted in 62 percent seed set.

The objective of this long-term study has been to evaluate the effects of mating of closely related black walnut individuals.

Materials and Methods

In 1971, three black walnut trees in the vicinity West Lafayette, Indiana were selfed and outcrossed by controlled pollination techniques described by Beineke and Masters (2), Beineke and Masters (3), and Beineke, Masters and Pennington (4). Also, wind pollinated seed was collected from each tree. The resultant seedlings were outplanted in a progeny test in Park County, Indiana on land owned by Pierson-Hollowell Company. Survival, height, diameter, and stem form data was last obtained in 1986. Volume for each progeny was calculated according to Todhunter et al. (13).

In another experiment, an outstanding seedling was selected and replicated by grafting. When the grafts reached flowering age, pollen was collected from its maternal parent and applied to the stigmas of female flowers on the grafted seedling to produce a backcross. Seedlings were outplanted in a progeny test at Purdue University's Martell Forest near West Lafayette, Indiana.

Results

Remeasurement of the 14-year study confirms earlier speculation that inbreeding and closely related matings in black walnut reduce growth rate and vigor (1). At age 14, inbreeding depression occurred in seed germination (11 percent), height (9 percent), diameter (18 percent), volume (36 percent), and form (16 percent) as compared to outcrossed progeny; however, seedling survival of selfs after outplanting was as good as the outcrossed seedlings (Table 1).

Table 1. Inbreeding depression in selfed and backcrossed black walnut seedlings.

	Number of Seedlings	Seed Germination (Percent)	Seedling Survival (Percent)	Height (m)	DBH (cm)	Volume[1] (cu.m)	Form[2]
14-year-old self test							
Self	32	72.7	88.9	4.42	5.97	.0081	3.73
Outcross	61	81.4	88.4	4.83	7.32	.0127	3.22
Wind	98	97.9	97.0	4.80	6.93	.0115	3.57
Inbreeding depression (%)		10.7[3]	—	8.5	18.4	36.2	15.8
7-year-old backcross test							
Backcross	8	—	87.5	3.58	2.90	.0019	3.71
Female parental	10	—	100	4.46	4.45	.0048	3.60
Male parental	4	—	100	4.49	4.39	.0047	3.75
Inbreeding depression (%) (male, female mean)		—	12.5	20.1	34.5	60.3	1.0

[1] From "A Volume Equation for Seedling and Sapling Black Walnut," 1979. M. N. Todhunter, S. E. McKeand, and W. F. Beineke. Purdue Agr. Expt. Sta. Bul. No. 197. 3 p.

[2] From 1 = excellent to 5 = very poor stem form.

[3] Calculated using outcross as the basis.

In the seven-year-old backcross study, inbreeding depression was greater than was observed from selfs with the exception of form. At age seven, survival, height, diameter, and volume were depressed by 12.5, 20.1, 34.5, and 60.3 percent, respectively, as compared to wind-pollinated progeny (Table 1). Stem form was nearly the same as the male and female parental mean.

Discussion

Evidence from these experiments suggests that selfs and closely related matings produce black walnut trees of less than average growth rate and vigor. If this is occurring in our natural stands, the effect on naturally regenerated populations of black walnut could be a reduction in growth and form. As the best trees are cut leaving the worst to reproduce, and as distances between trees and woodlots increases, the detrimental effects of related matings may increase.

In forest openings on good sites, vigorous competition develops among trees, shrub, and herbaceous species. Since black walnut is intolerant, seedlings that are the product of selfing or related matings usually would be eliminated. Hence, few walnut seedlings may become established in the next generation of the forest. This could account for the paucity of walnut seedlings in forest openings with nearby seed sources. However, if inbred or related seedlings survive to maturity, further degradation of the forest could occur.

In addition, seed collected or seedling production by the state nursery systems contains a percentage of selfed seed. Since selfed seed germinates and survives nearly as well as outcrossed seed (Table 1), seedlings grown in the pampered nursery environment are eventually outplanted in plantations where every tree is expected to grow and survive to merchantable size and quality. This may be one reason significant numbers of seedlings perform poorly in most black walnut plantations.

Assuming that outcrossing occurs more frequently than inbreeding in black walnut, what is the fate of the outcrossed individual in these small isolated woodlands? The few poor-quality individuals left for breeding purposes probably perpetuate their kind; but, in addition, lacking pollination from sources outside the small, isolated woodland, the progenies will tend to become homozygous at additional loci in future generations, and undesirable genes could become fixed due to genetic drift (16).

The current interest by various agencies to bring black walnut seed orchards into production has values for reasons other than the production of genetically improved seedlings (5, 9). Black walnut seed orchards serve the vital function of providing genetic diversity that can be returned to plantations and forest openings. Contrary to popular opinion concerning the perceived threat of monoculture posed by the genetic improvement of trees, seed orchards bring together diverse genetic combinations not possible in nature. Since selections are scattered throughout a region and only one tree is selected from the same stand, related matings are unlikely events. Related matings are still possible, but careful observation and avoidance of clones that consistently overlap in flowering times should insure a minimum of selfing.

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