Propagation of Narrowleaf Cottonwood by Stem Cuttings II: Media Density and Indole-3-butyric Acid Concentration.

by:

John T. Harrington, Stephen Hine, and David R. Dreesen

Introduction

Narrowleaf cottonwood (*Populus angustifolia* James), occurs throughout the Rocky Mountains of North America. Its natural range is from southern Canada to northern Mexico (Little 1968). Typically, this species is located in riparian areas, however, it has been found in drastically disturbed upland sites (Harrington and Dreesen personal observations). No work has been done on the physiology, specifically water relations, of these upland sources. However, the ability to naturally colonize such sites indicates members of this species may be suitable for high elevation revegetation projects.

Published protocols for producing narrowleaf cottonwood in either nursery beds or containers is lacking. Scant information exists on species sensitivity to media density (Harrington et al. earlier in this proceeding). Rooting hormones are not commonly used in *Populus* propagation and in some cases have been inhibitory to root production ((Phipps et al 1977; Philippis 1966 as cited by Phipps et al. 1977). Indole-3-butyric acid is frequently used to promote root initiation with difficult to root species. This is due, in part, to its beneficial effects and its low toxicity even at high concentrations. In *Populus alba*, high levels of exogenous auxin (4,000 ppm IBA and 2,000 ppm IBA) improved rooting while lower concentrations did not improve the overall response (Al-Kinany 1981). When developing a propagation protocol for a new species it is still necessary to examine the effect of hormone level on the rooting response. In container nurseries, small increases in overall rooting can result in cost effective production of the crop.

The objectives of this study were to examine the effects of media density and IBA level on the rooting response and shoot growth of narrowleaf cottonwood cuttings.

A secondary objective was to examine the influence of stock plant location (vigor) on these responses.

Methods and Materials

Stem (branch) cuttings used in this study originated from five distinct stands (ecotypes) of narrowleaf cottonwood growing at the Molycorp Inc. molybdenum mine in Questa, New Mexico. This area is located in the Red River canyon approximately five miles east of the town of Questa, New Mexico. Ecotype locations are described in Table 1. Ecotypes were separated by no less than 1,000 meters. Stem cuttings originally taken from these ecotypes in 1992 were used to establish stooling blocks at the Los Lunas Plant Materials Center in Los Lunas, NM. The stooling blocks were established in the spring of 1993. Source identification of the stooling block material was maintained to the ecotype level but not to the individual tree.

Table 1. Ecotype locations and elevations:

Ecotype	Elevation
Capulin	9,800
Raspberry Ridge	9,850
Pinon Knob	9,300
Neutral	8,600
River	8,100

The stem cuttings used in this study were harvested from both the original stands at the mine as well as from the 3-year-old stooling block material growing in Los Lunas, NM. The source plants at the mine site ranged in age from 3 years to 15 years. When possible branches were harvested from young trees or younger materials from older trees. Branches were harvested in the last week of February 1996 for the Los Lunas source material and the first week of March 1996 for the mine source material. Branches were then transported to the Mora Research Center and stored in coolers (2 - 4°C) until used in the study (less than 2 weeks). Individual branches were subdivided into stem cuttings immediately prior to treatment imposition. Stem cutting length ranged from 10 cm to 15 cm in length and contained a minimum of 3 buds. Subdivision of the stem cuttings was done using a hand pruner.

The rooting hormone used in this study was indole-3-butyric acid (IBA). A stock solution of 1000 ppm IBA was prepared. Test concentrations of 125 ppm, 250 ppm, and 500 ppm were prepared by dilution from the stock solution. Distilled, de-ionized water was used as a control. Rooting hormone application was a 5 second dip into the appropriate hormone treatment immediately followed by sticking the cutting into the container.

The rooting media evaluated in this study were a 2:1:1, a 1:1:1, a 1:2:1, and a 1:3:1 ratio of peat:perlite:vermiculite (v:v:v). Media components were mixed using a large paddle mortar mixer. Slow release fertilizer (Osmocote 14-14-14; 3 month) was incorporated into the media at the rate of 4kg/m^3 during the mixing process.

Once cuttings were treated they were stuck in 164 cm^3 Ray-Leach Super Cells. Stem cuttings were placed in a greenhouse on a propagation bench with bottom heat which kept root zone temperature at 24°C. Greenhouse temperatures were 20 - 22°C days and 16 - 18°C nights. Photoperiod was a 10-hr light/14-hr dark with the dark cycle being interrupted twice at 5 hours and 10 hours with 30 minute light periods. Artificial light used to extend the ambient light period and light interruptions was provided by 1000-watt high pressure sodium vapor lamps suspended 3 meters above the stem cuttings.

Cuttings were misted 4 times daily until the majority of cuttings had significant bud break (approximately 3 weeks). Following budbreak, cuttings were irrigated as needed, increasing from once every three days at the beginning of the study to once every day at week 20. Fertilizer applications consisted of foliar feed applications of a 25 ppm N solution of Peter's Foliar Feed following every second irrigation from week 4 through week 12. At week 13, this fertilization was increased to applications of 100 ppm N of Peter's Conifer Grower (20-7-19) every other irrigation . This fertilization rate was retained until the end of the study. At week 20, cuttings were non-destructively analyzed for rooting response, seedling height and number of leaves.

The treatment structure for this study was a factorial design with two source plant locations, five ecotypes, four media combinations, and four levels of IBA application. The planting design was a randomized complete block design. Due to limited plant material availability, a variable number of trees (2 - 8) were used to represent each ecotype by site combination. Each tree/ecotype/site was represented in each of the 16 media by hormone treatments by 14 stem cuttings. Data was analyzed using analysis of variance procedures.

Results

Overall the rooting response was quite high in this study, averaging over 80%. As a single factor, stock plant location accounted for the greatest variability observed in the rooting response (Table 2). Overall cuttings collected from the younger stooling blocks in Los Lunas had a higher rooting response (Figure 1). Regardless of ecotype, cuttings from the stooling blocks in Los Lunas rooted between 94% and 96%. As elevation of mine collected ecotype decreased rooting percent increased from 62% to 85%. IBA level also influenced rooting response. Study wide concentrations of 250 ppm IBA and 500 ppm IBA improved rooting percentages when compared to untreated cuttings. The 125 ppm IBA treatment appeared to have an inhibitory effect on rooting, lowering rooting from 85% for control treated cuttings to 80% (Figure 2).

Stock plant location (site) had the greatest impact on shoot growth of the variables monitored (Table 3). The material from the younger Los Lunas stooling blocks showed greater growth than materials originating directly from the Questa mine (Figure 3). Growth was up to 100% greater when cuttings originated from Los Lunas. In four of the five ecotypes evaluated, stem cuttings from Los Lunas had greater than 50% more growth by the end of the 20 weeks (Figure 3). Only in the case of cuttings derived from the river ecotype was there no difference in growth. This ecotype had the greatest shoot growth of all the ecotypes originating from the mine and was comparable to two of the ecotypes from Los Lunas.

The influence of media composition on shoot growth was strongly influenced by ecotype. The four upper elevation ecotypes all had greater shoot growth on the lighter, more porous media (Figure 4). The ecotype from the base of the valley near the river had the greatest shoot growth on the two intermediate density media. Shoot growth was lowest on the least porous media (2:1:1; peat:perlite:vermiculite) across all

Table 2. Analysis of variance table for the effects of rooting media, IBA (auxin) level, stock plant location (site) and ecotype on the rooting percentage of narrowleaf cottonwood cuttings.

Source	df	MS	Pr > F
Model	159	551.45	0.0001
Media (M)	3	45.25	0.7195
IBA Dosage (D)	3	643.50	0.0003
M x D	9	175.03	0.0813
Site (S)	1	41955.74	0.0001
M x S	3	342.19	0.0185
D x S	3	384.88	0.0105
M x D x S	9	115.97	0.3297
Ecotype (E)	4	1026.86	0.0001
M x E	12	97.81	0.4807
D x E	12	170.56	0.0686
M x D x E	36	130.98	0.1272
S x E	4	1290.08	0.0001
M x S x E	12	129.48	0.2290
D x S x E	12	305.22	0.0005
M x D x S x E	36	125.33	0.1709
Error	329	101.20	
Total	488		

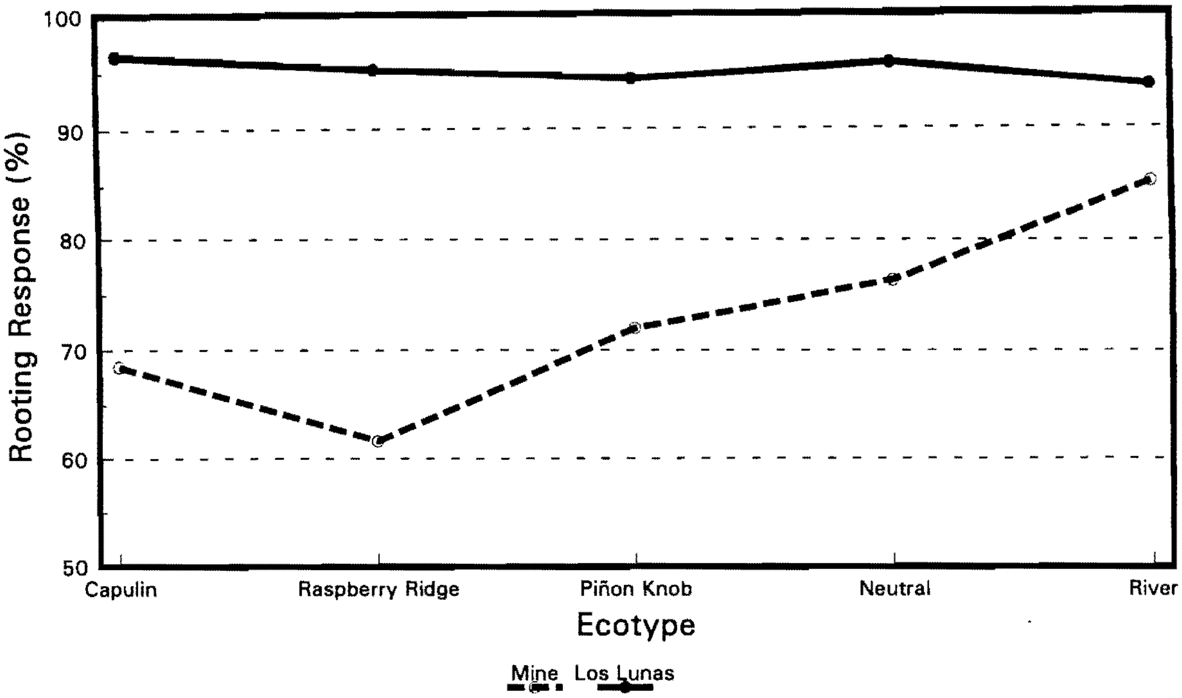

Figure 1. Influence of stock plant location and ecotype on the percentage of narrowleaf cottonwood cuttings rooting in this study.

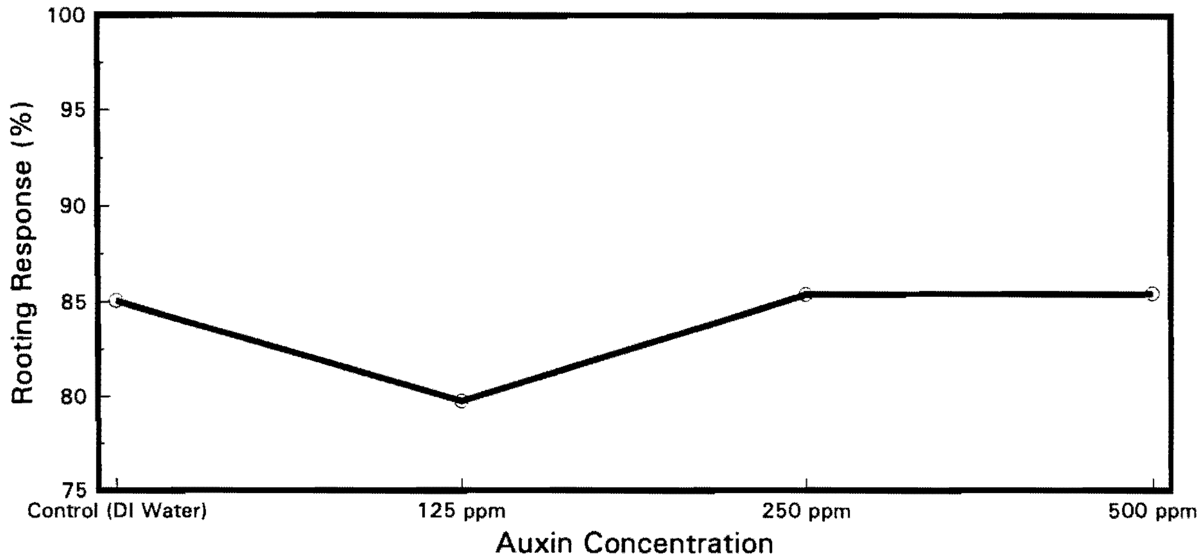

Figure 2. Influence of auxin (IBA) concentration on the percentage of narrowleaf cottonwood cuttings rooting in this study.

Table 3. Analysis of variance table for the effects of rooting media, IBA (auxin) level, stock plant location (site) and ecotype on the shoot growth of rooted narrowleaf cottonwood cuttings.

Source	df	MS	Pr > F
Model	159	2200.24	0.0001
Media (M)	3	11423.24	0.0001
IBA Dosage (D)	3	1734.52	0.0002
M x D	9	822.82	0.0007
Site (S)	1	100145.58	0.0001
M x S	3	1039.58	0.0070
D x S	3	1727.02	0.0002
M x D x S	9	120.21	0.8972
Ecotype (E)	4	1706.32	0.0001
M x E	12	938.25	0.0001
D x E	12	396.14	0.1020
M x D x E	36	467.76	0.0020
S x E	4	6506.36	0.0001
M x S x E	12	735.48	0.0006
D x S x E	12	697.89	0.0012
M x D x S x E	36	445.01	0.0043
Error	6703	257.15	
Total	6862		

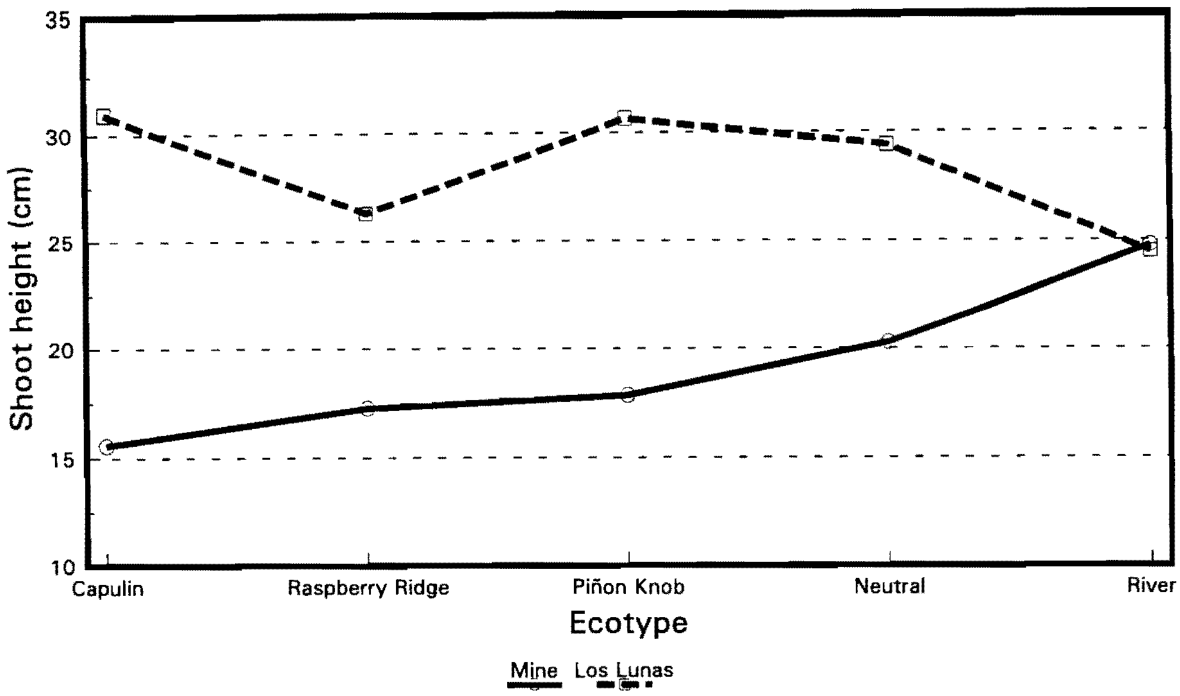

Figure 3. Influence of ecotype on shoot growth of rooted narrowleaf cottonwood cuttings collected from the mine and from stooling blocks growing in Los Lunas, NM.

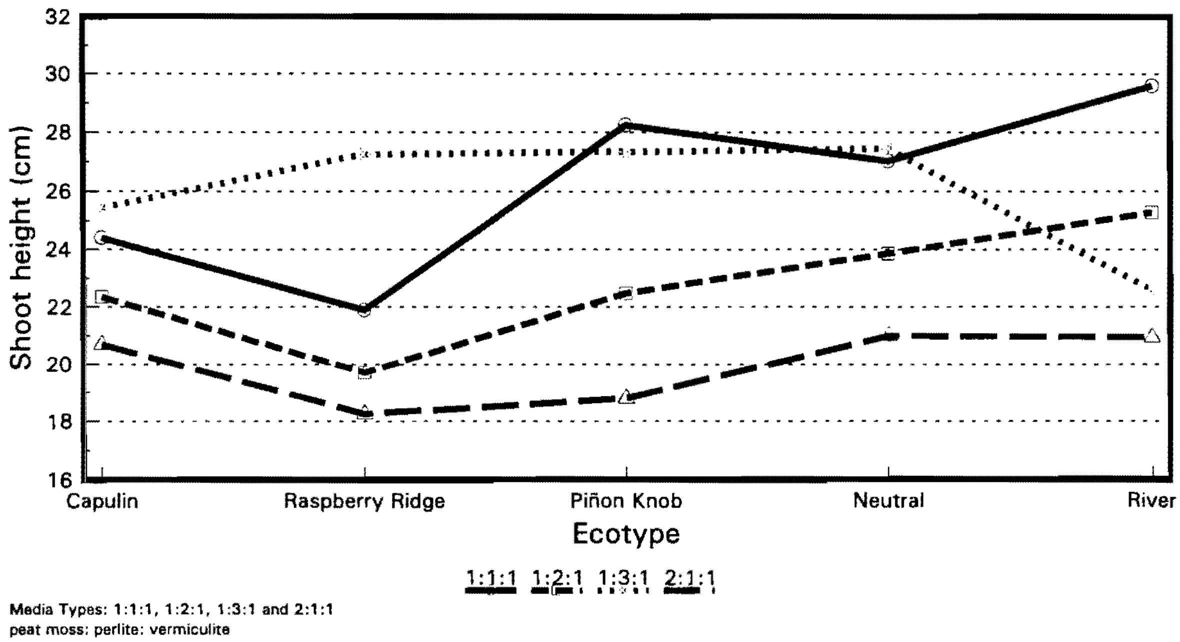

Figure 4. Influence of media density on the shoot growth of rooted narrowleaf cottonwood cuttings from five ecotypes examined in this study.

ecotypes (Figure 4). As was the case with rooting response, the two upper concentrations of IBA (250 ppm & 500 ppm) did comparable to the control treatment in shoot growth (Figure 5). Only at the lowest IBA level, 125 ppm IBA, was shoot growth negatively impacted by the treatment.

Site and ecotype factors had the greatest impact on the number of leaves produced (Table 4). Source plant location (site) accounted for majority of variation found. The material from the Los Lunas stooling blocks had over 50% more leaves than seedlings originating from stem cuttings obtained from the mine (Figure 6). As was the case with shoot growth, rooted stem cuttings from the river ecotype collected at the mine had similar leaf numbers as those produced from Los Lunas derived materials.

Applications

Again, stock plant vigor was an overriding factor on rooting success and subsequent cutting growth. Overall, rooting percentage for stooling block derived material averaged 95% whereas cuttings collected from natural growing stands at the mine ranged from 62% to 85%. Exogenous hormone application (IBA) did not significantly improve rooting at any level. Media density influenced shoot growth to a greater extent than it did root initiation. There was considerable variability across ecotypes in shoot growth sensitivity to media composition. The highest elevation, upland sources grew best on the lightest, most porous media (1:3:1). In contrast the two lowest elevation sources grew better on a moderately porous media (1:1:1). Our standard production mix, 2:1:1, was worst for shoot growth.

Table 4. Analysis of variance table for the effects of rooting media, IBA (auxin) level, stock plant location (site) and ecotype on the number of leaves produced on rooted narrowleaf cottonwood cuttings.

Source	df	MS	Pr > F
Model	159	256.49	0.0001
Media (M)	3	153.14	0.0006
IBA Dosage (D)	3	357.59	0.0001
M x D	9	76.28	0.0023
Site (S)	1	15084.27	0.0001
M x S	3	174.08	0.0002
D x S	3	193.51	0.0001
M x D x S	9	32.60	0.2764
Ecotype (E)	4	803.56	0.0001
M x E	12	79.30	0.0004
D x E	12	60.22	0.0076
M x D x E	36	57.71	0.0001
S x E	4	635.49	0.0001
M x S x E	12	113.89	0.0001
D x S x E	12	96.17	0.0001
M x D x S x E	36	55.90	0.0001
Error	6703	26.69	
Total	6862		

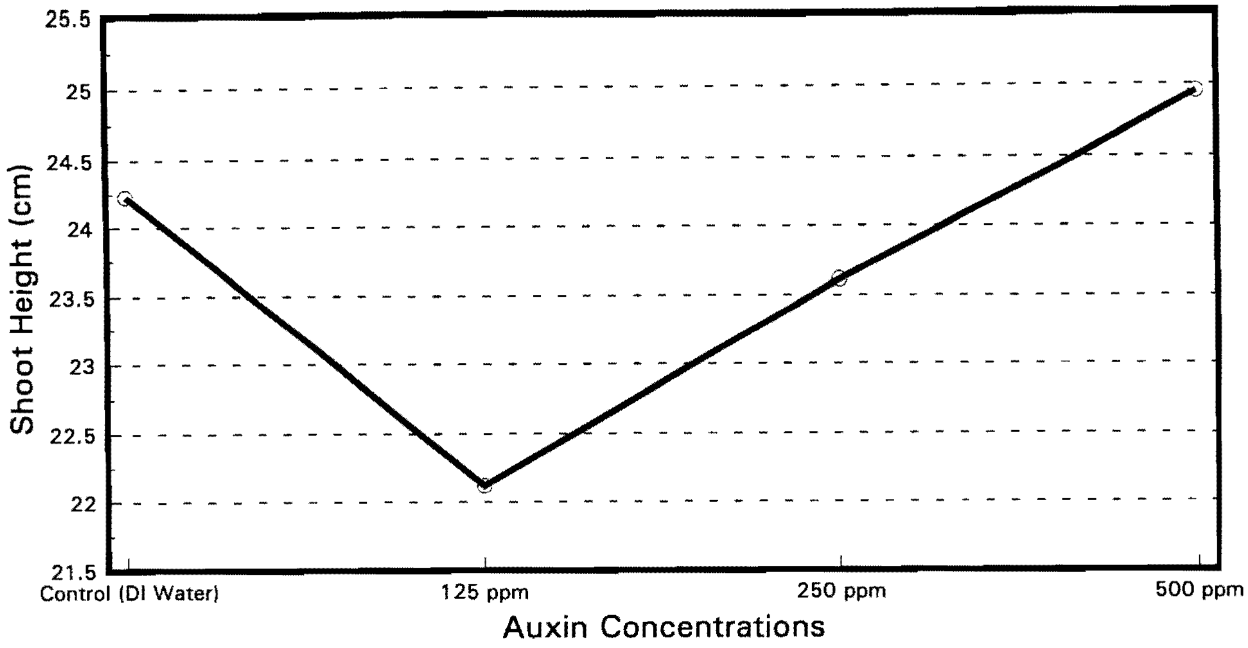

Figure 5. Influence of auxin (IBA) concentration on the shoot growth of narrowleaf cottonwood cuttings.

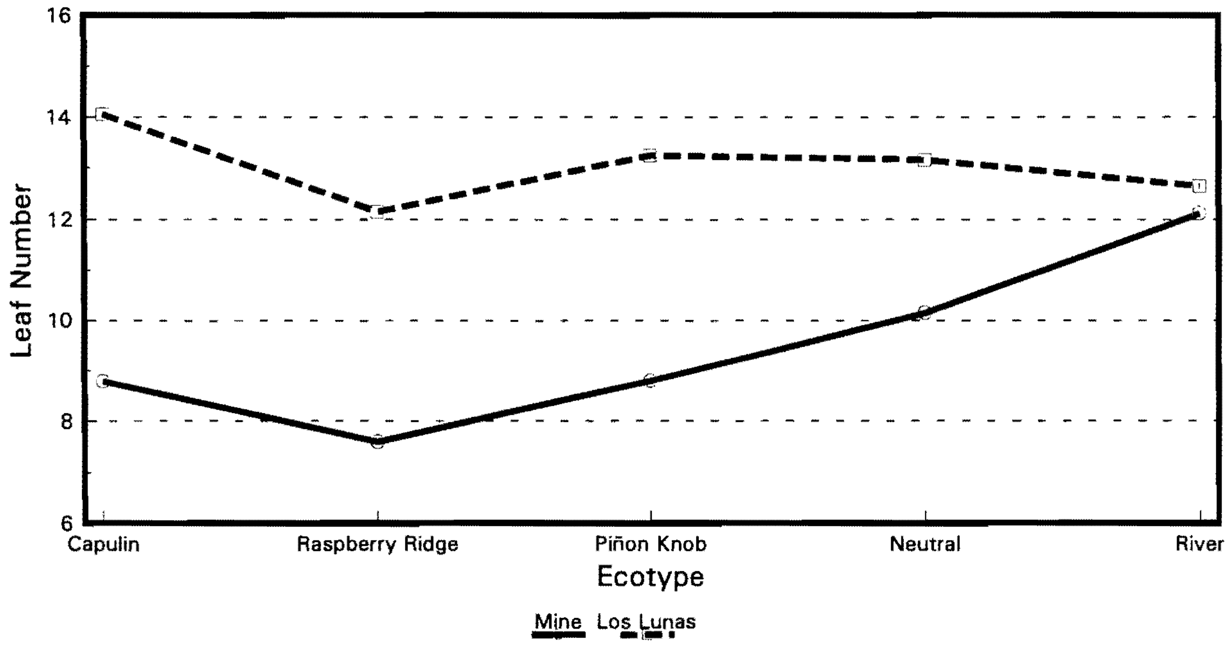

Figure 6. Influence of stock plant location (vigor) on the number of leaves produced on rooted narrowleaf cottonwood cuttings for five ecotypes of the species.

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