Preliminary Evaluation on Adventitious Rooting Hardwood Stem Cuttings of *Symphoricarpos oreophilus*, *Ribes cereum*, and *Cercocarpus montanus* from Throughout New Mexico[4]

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ABSTRACT

Current trends in restoration, reclamation and revegetation are focusing on using local sources of indigenous plants including woody shrubs. However, for many native shrubs propagation techniques are not well researched, resulting in increased production costs for those species. Further, propagation literature is often based on studies with a limited number of sources. This investigation was undertaken to evaluate the suitability of rooting of dormant hardwood cuttings of *Symphoricarpos oreophilus, Ribes cereum*, and *Cercocarpus montanus* as a means of plant propagation. Exogenous hormone application dosage and timing of collection were evaluated for seven sources of each species. Sources were selected to represent a range of latitudes from the southern part of the state to the northern part of the state. At the northern-most latitude, Molycorp, Inc. mine near Questa, NM, collections were made at three elevations ranging from 8,200 feet to 9,800 feet. Several rooting response variables were measured. *Ribes cereum* and *Cercocarpus montanus* had overall poor rooting in this investigation. Only the *Symphoricarpos oreophilus* had appreciable rooting. Exogenous hormone dosage, timing of collection and source of cuttings all influenced the rooting response. Late winter/early spring cuttings had the highest rooting percentages across all sources with the exception of the most northerly, highest elevation source. IBA/NAA applications from 250 to 1,000 ppm improved the percentage of cuttings rooting. There was considerable variation in rooting among the sources evaluated with the more northerly sources, from the Sandia Mountains northward, having overall the greatest percentage of cuttings rooting.

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tall (Harrington 1954, Vines 1960). Wax currant occurs on dry slopes, ridges, and plains at elevations of 1200-4000 meters from Montana west to British Columbia south to California and east to New Mexico (Vines 1960, Marshall and Winkler 1995). Wax currant is a potentially valuable species for the revegetation of disturbed sites. It occurs within numerous habitat types and plant communities- open forests, forest edges, and well as in shrub communities (Marshall and Winkler 1995). Wax currant grows on soil textures ranging from sandy to clayey, and has been observed to grow on rocky soils with little topsoil (Marshall and Winkler 1995). Wax currant occurs across a highly variable range of temperatures and precipitation amounts (Marshall and Winkler 1995). It has been observed to be an early colonizing species of disturbed sites within Douglas Fir communities, providing canopy favorable for the establishment of Douglas Fir seedlings (Marshall and Winkler 1995). This species can provide browse for deer when better browse is not available, and fruits are eaten by numerous bird species (Marshall and Winkler 1995).

Propagating native shrubs from vegetative material has potential advantages over propagation from seed. Seed propagation of native species can be made difficult by sporadic seed production in native stands, low seed viability necessitating seed refinement procedures, and seed dormancy, which must be overcome by scarification and stratification techniques (Dreesen and Harrington 1997). Some species may be propagated more easily, quickly, and economically by vegetative methods than by seed. The most widespread technique is rooting of stem cuttings.

Stem cuttings are able to develop into complete plants due to their ability to produce adventitious roots from within stem tissues. For most woody species propagation from stem cuttings requires exogenous auxin treatment (Kevers et al. 1997). Indole-3-acetic acid (IAA), a naturally occurring auxin, is known to promote adventitious root formation. Indole-3-butyric acid (IBA) and napthaleneacetic acid (NAA) are synthetic auxin found to be effective as root-promoting treatments for cuttings (De Klerk et al. 1999).

Little research has been conducted on the potential of vegetative propagation for large scale production of these species. Some research from the horticultural industry has illustrated that stem cuttings from other members of *Symphoricarpos* and *Ribes* can be induced to root readily (Anonymous 1948, Dirr and Heuser 1987, Plummer et al. 1969, Pfister 1974, Young and Young 1992). However, published literature on vegetative propagation of any species of *Cercocarpus* is lacking.

The objectives of this study were to examine the possibility of using vegetative propagation as means of propagating these three species. Additional objectives were to examine the influence of timing of collection and exogenous auxin application on adventitious root development. Lastly, the study was designed to examine the influence of provenance along a latitudinal gradient within New Mexico and an elevational gradient at the northern most sampling point on rooting response of these three species.

MATERIALS AND METHODS

For each species cuttings were taken at multiple locations (sources) throughout New Mexico in order to assess variability in response to various treatments. Sources were selected to encompass a range of latitudes across the state and a range of elevations at the Molycorp Mine in

Questa, New Mexico (Table 1). Cuttings were stuck on four dates approximately one month apart: January 17, February 14, March 14, and April 11, 1998. Within 48 hours prior to each date 110 cuttings from each source of each species were taken, sealed in polybags, placed into coolers and brought to Mora, NM. Only cuttings from the current season's growth were used. Eighteen centimeter cuttings were taken when possible. Many of the source plants at the Molycorp, Inc., site had insufficient growth the previous growing season to meet the eighteen centimeter cutting length target, however no cuttings less than 12.5 centimeters were used.

Cuttings from each source of each species were trimmed to 15 centimeters (or slightly less in the case of shorter cuttings) in length with a diagonal cut at the basal end. Terminal buds were removed, and cuttings were randomly divided into 5 groups of 20 cuttings each, by source. Each of the 5 groups underwent a different hormone treatment. The hormones used were equal parts IBA and NAA dissolved in 50% isopropyl alcohol. Hormone concentrations of 4000, 2000, 1000, 500, and 0 parts per million (ppm) were used for mountain mahogany and wax currant. Hormone concentrations were 2000, 1000, 500, 250, and 0 ppm were used for mountain snowberry. Cuttings were given a five-second quick-dip in their respective hormone treatments to a depth of 2 centimeters, allowed to air dry, and stuck to a depth of at least 7.5 cm into cells containing a mixture of 2 parts peat and 1 part each of perlite and vermiculite. Copper-coated, 77-cell styroblocks were used for wax currant and mountain snowberry, and copper-coated, 112-cell styroblocks were used for mountain mahogany. Cuttings were watered in and kept moist throughout the experiment.

Due to other activities in the greenhouse, three types of greenhouse benches were used to conduct this study. Bench one was a bottom heated bench with a traveling boom mist system and a wet canvas canopy suspended 1.2 meters above the cuttings. The second bench used was also a bottom heat bench, however, humidity was enhanced by using a small greenhouse fogger with a clear polyethylene covering suspended 1 meter above the seedlings. The third bench was a conventional greenhouse bench with no special features to elevate humidity levels above greenhouse levels. The bottom heat temperatures were set at 20°C for the first two benches. Cuttings stuck on the first date were placed on a bench for 30 days then moved to the second bench for the remainder of the study. Cuttings stuck on the second date were placed on the first bench for 30 days at which they were moved to the third or conventional bench for the remainder of the study. Cuttings stuck on the third and fourth collection dates were placed on the first bench and remained there for the remainder of the study. Cuttings stuck on the third and fourth dates were placed on the boom bench and not moved. Thirty days after the cuttings were stuck the wet canvas covering was removed from the bench, in effect mimicking a typical greenhouse bench.

Cuttings were monitored for leaf burst at weekly intervals throughout the study. After 22 weeks (150 days) cuttings were removed from the media, and their roots were measured. Stem caliper, number of root loci and length of the longest primary root were measured, and a subjective rating from 0-4 for the amount of root branching was assigned. A rating of 0 represented no root branch and a rating of 4 represented heavy root branching.

Categorical analysis of variance was used to determine treatment (collection date, hormone concentration, and cutting source and their interactions) differences in the number rooted cuttings. A generalized-least-squares approach was used in order to address the high frequency

Table 1. Cutting Source Information

Species		Lot Name	Latitude (approx.)	Location	Elevation
mountain snowberry		Capulin	36 degrees 42'	Questa, NM	9,800 ft
mountain snowberry		Mill	36 degrees 42'	Questa, NM	8,200 ft
mountain snowberry		Spring Gulch	36 degrees 42'	Questa, NM	8,700 ft
mountain snowberry		Mora	36 degrees 00'	Mora, NM	7,200 ft
mountain snowberry	(a)	Sandia	35 degrees 10'	Albuquerque, NM	7,700 ft
mountain snowberry	(a)	Sacramento	32 degrees 58'	Cloudcroft, NM	8,600 ft
wax currant		Capulin	36 degrees 42'	Questa, NM	9,800 ft
wax currant		Mill	36 degrees 42'	Questa, NM	8,200 ft
wax currant		Mahogany Point	36 degrees 42'	Questa, NM	8,800 ft
wax currant		Mora	36 degrees 00'	Mora, NM	7,200 ft
mountain mahogany		Capulin	36 degrees 42'	Questa, NM	9,800 ft
mountain mahogany		Mill	36 degrees 42'	Questa, NM	8,200 ft
mountain mahogany		Mohagany Point	36 degrees 42'	Questa, NM	8,800 ft
mountain mahogany		Mora	36 degrees 00'	Mora, NM	7,200 ft
mountain mahogany		Sandia	35 degrees 10'	Albuquerque, NM	9,300 ft
mountain mahogany		Sacramento	32 degrees 58'	Cloudcroft, NM	8,600 ft

(a) Snowberry shrubs at these locations were not identified to species with certainty, and may be another snowberry species or mixture of snowberry species.

of low cell counts. Pairwise comparisons were made using an alpha value of .05 divided by the number of comparisons.

Analysis of root branching of rooted cuttings was only undertaken for mountain snowberry. The low rooting success of wax currant and mountain mahogany precluded any analysis of root branching in these species. To facilitate analysis of root branching, root branching ratings were divided into 2 categories. Ratings of 0, 1, and 2 were combined to form a low root-branching category, and ratings of 3 and 4 were combined to form a high root-branching category. Categorical analysis of variance was then used to determine how cutting source, collection date, and hormone concentration affected root branching. Pairwise comparisons were carried out using an alpha value of .05 divided by the number of comparisons.

Analysis of number of root loci and length of longest root for rooted cuttings was also undertaken for mountain snowberry only, because too few cuttings rooted successfully for wax currant or mountain mahogany. Analysis of variance (ANOVA) was used to determine differences in the number of root loci and length of longest root as effected by hormone concentration, collection date, and cutting source. Least square means were used for pairwise comparisons.

RESULTS

The overall low rooting percentages of the wax currant (< 5%) and the mountain mahogany (< 1%) precluded any analysis of the data. Therefore the results and discussion will only relate to the responses found in the mountain snowberry portion of the study. Treatment effects were considered meaningful at the alpha < 0.05 level.

Cutting source, collection date, and hormone treatment all affected rooting response (Table 2), as did the interactions of cutting source with collection date (Figure 1) and collection date with hormone concentration (Figure 2). Cuttings from the Mill source at Molycorp Mine had the highest overall rooting percentage, averaging 50%, approximately double the lowest rooting percentage, 25%, for cuttings from the Sacramento site (Table 2). All other sources had overall rooting percentages ranging from 35% to 43%.

The greatest percentage of cuttings rooting successfully were collected in March followed by April, February, and lastly January (Table 2). Using rooting of cuttings collected in January as a baseline, March cuttings showed a 140% improvement, April cuttings showed an 87% improvement, and February cuttings showed a 41% improvement. Rooting percentage improved for most sources when collection date was delayed, reaching a maximum in March (Figure 1). However rooting percentage of Sandia cuttings fell when collection was delayed from January to February, but then rose to a maximum in March. Other exceptions to the overall trend were the February peak in rooting of cuttings from Capulin and the continued high rooting, relative to March, of Spring Gulch cuttings taken at the April sampling date.

The percentage of cuttings rooted was improved by treatment with 250, 500, or 1000 parts per million IBA/NAA solution (Table 2). Treatment with 2000 ppm IBA/NAA resulted in rooting percentages no different from that of the control.

Cuttings collected at different dates responded differently to hormone concentration (Figure 2). While overall cuttings taken in March had the best rooting, cuttings taken at this

Table 2. Mountain Snowberry Cutting Study- Main Effects of Source, Collection Date, and IBA/NAA Concentration on Percentage of Cuttings Rooted after 150 Days

Cutting Source	Mean Percent Rooted [1]	Standard Error
Capulin	39.0 (b)	2.44
Spring Gulch	43.0 (ab)	2.48
Mill	50.3 (a)	2.50
Sandia	36.5 (b)	2.41
Mora	35.0 (b)	2.38
Sacramento	24.8 (c)	2.16

Collection Date	Mean Percent Rooted [1]	Standard Error
January 17	22.8 (d)	1.71
February 14	32.3 (c)	1.91
March 14	54.8 (a)	2.03
April 14	42.3 (b)	2.02

IBA/NAA Concentration (parts per million)	Mean Percent Rooted [1]	Standard Error
0	32.1 (b)	2.13
250	45.2 (a)	2.27
500	42.5 (a)	2.26
1000	40.8 (a)	2.24
2000	29.8 (b)	2.09

[1] Mean rooting percents followed by the same letter are not significantly different.

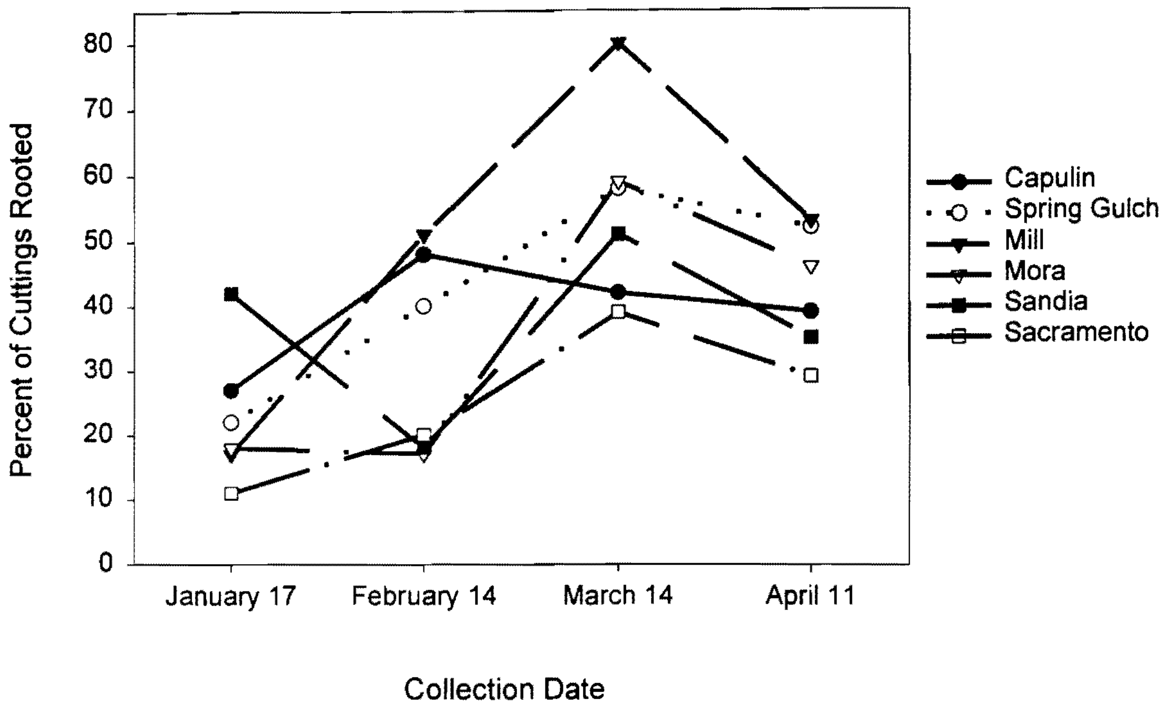

Figure 1. Mountain Snowberry Cutting Study- Effect of the Interaction between Cutting Source and Collection Date on Percent of Cuttings Rooted 150 Days after Sticking

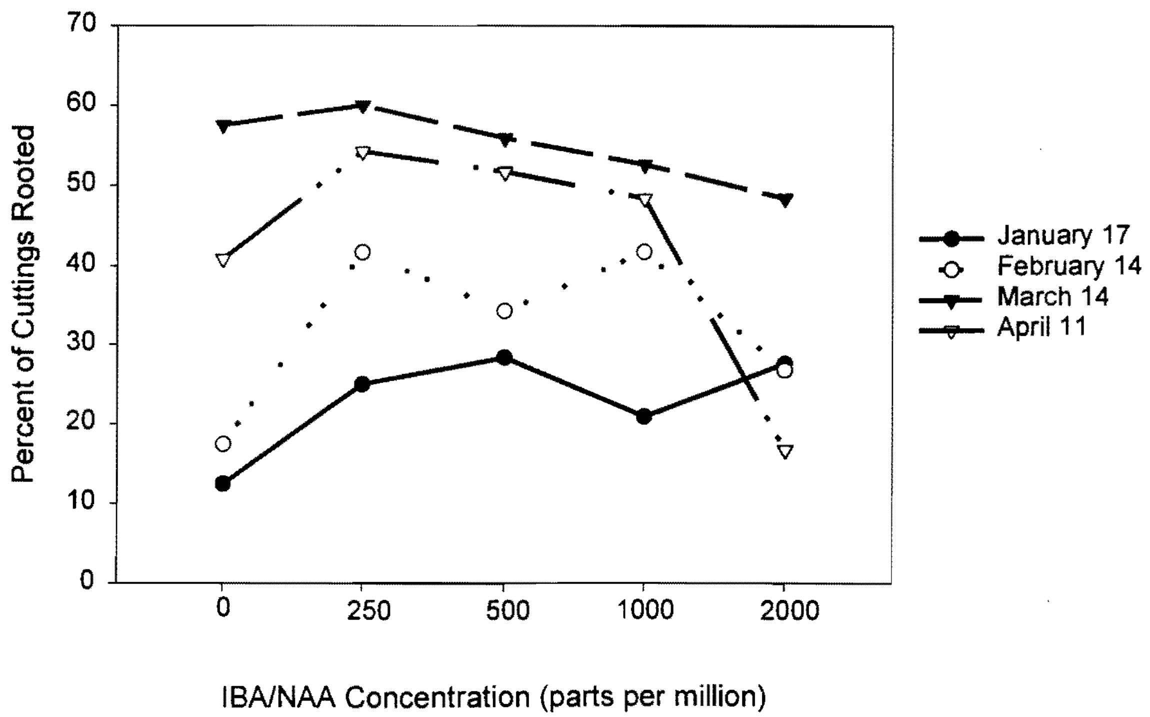

Figure 2. Mountain Snowberry Cutting Study- Effect of Interaction between Hormone Concentration and Collection Date on Percent of Cuttings Rooted 150 Days after Sticking.

time did not benefit from any hormone treatment. Cuttings collected on the other 3 dates all benefited from the intermediate concentrations hormone concentrations of hormone application, but optimal concentration varied by source and collection date.

Collection date was the only factor affecting the root branching of cuttings. Pairwise comparisons of collection dates indicated cuttings collected in January and February had a higher percentage with high root-branching ratings than did those collected in March or April (Table 3). This result contrasts sharply with the effect date had on rooting response, in which later collections, March and April, had greater frequencies of cuttings rooting (Table 2).

Cutting source, collection date, hormone concentration, and all interactions of these factors affected the number of root loci per cutting (Table 4, Figure 3, Figure 4). Low numbers of root loci for Sacramento correlates with this sources low rooting percentage overall, but did not for the other sources (Tables 2, 4). January and March cuttings had the highest number of loci, followed by April, and lastly by February (Table 4). This result varies from the rooting response analysis only in the relative position of January, which had the highest number of loci per cutting, but the lowest rooting percentage (Tables 2, 4).

The number of root loci per cutting did not consistently increase with increasing hormone concentration (Table 4). Although all dosages except 1000 ppm resulted in a higher number of root loci per cutting than the control, the best treatment increased the number of root loci by only 16% of the control. The effect of hormone concentration on mean number of loci varied highly among sources (Figure 3). The shape of the response pattern as well as optimal hormone concentration varied among sources.

Collection date also influenced the effect of hormone concentration on number of root loci (Figure 4). January and February cuttings were negatively impacted by increasing hormone concentration, while April cuttings had increased numbers of root loci with increasing concentration. March cuttings, on the other hand, showed a positive response to moderate concentrations of IBA/NAA, with a drop-off as concentration was increased.

Cutting source, collection date, and hormone concentration affected the length of the longest root per cutting, as did the interaction between cutting source and collection date (alpha<.05).

Average length of longest root increased from the southerly sources, (Sacramento) to the northerly sources, (Molycorp sources; Table 5). This trend strongly resembles the source trend found in rooting response (Table 2).

As collection date was delayed the mean length of longest root decreased (Table 5). This response contrasts with the results of the rooting response analysis (Table2), but is more or less consistent with the root branching and root loci analysis (Tables 3, 4).

Pairwise comparisons of hormone concentrations shows that the highest concentration (2000 ppm) resulted in the highest mean length of the longest root, and all other concentrations were equally worse (Table 5). This result contrasts with the result from the analysis of rooting response, in which 2000 ppm and the control treatment resulted in the fewest cuttings rooted (Table 2), and the root loci analysis in which there was no trend in response to increasing hormonal concentration (Table 4).

Table 3. Mountain Snowberry Cutting Study- Pairwise Date Comparisons for Root Branching Rating

Collection Date	Percent of Cuttings with High Root Branching	Standard Error
January 17	83.9 a[1]	3.1
February 14	85.6 a	2.5
March 14	60.6 b	2.7
April 11	54.8 b	3.1

[1] Mean cutting percents followed by same letter are not significantly different.

Table 4. Mountain Snowberrry Cutting Study- Main Effects of Source, Collection Date, and Hormone Concentration on Number of Root Loci per Rooted Cutting

Cutting Source	Mean Number of Root Loci [1]	Standard Error
Capulin	10.9 de	0.37
Spring Gulch	11.1 d	0.40
Mill	12.8 c	0.39
Mora	14.0 b	0.52
Sandia	16.6 a	0.58
Sacramento	9.6 e	0.58

Collection Date	Mean Number of Root Loci [1]	Standard Error
January 17	14.0 a	0.64
February 14	10.3 b	0.34
March 14	13.6 a	0.34
April 11	12.3 b	0.36

IBA/NAA Concentration (parts per million)	Mean Number of Root Loci [1]	Standard Error
0	11.5 c	0.43
250	12.7 ab	0.43
500	13.4 a	0.42
1000	12.3 bc	0.46
2000	12.9 ab	0.53

[1] Root loci means followed by same letter are not significantly different.

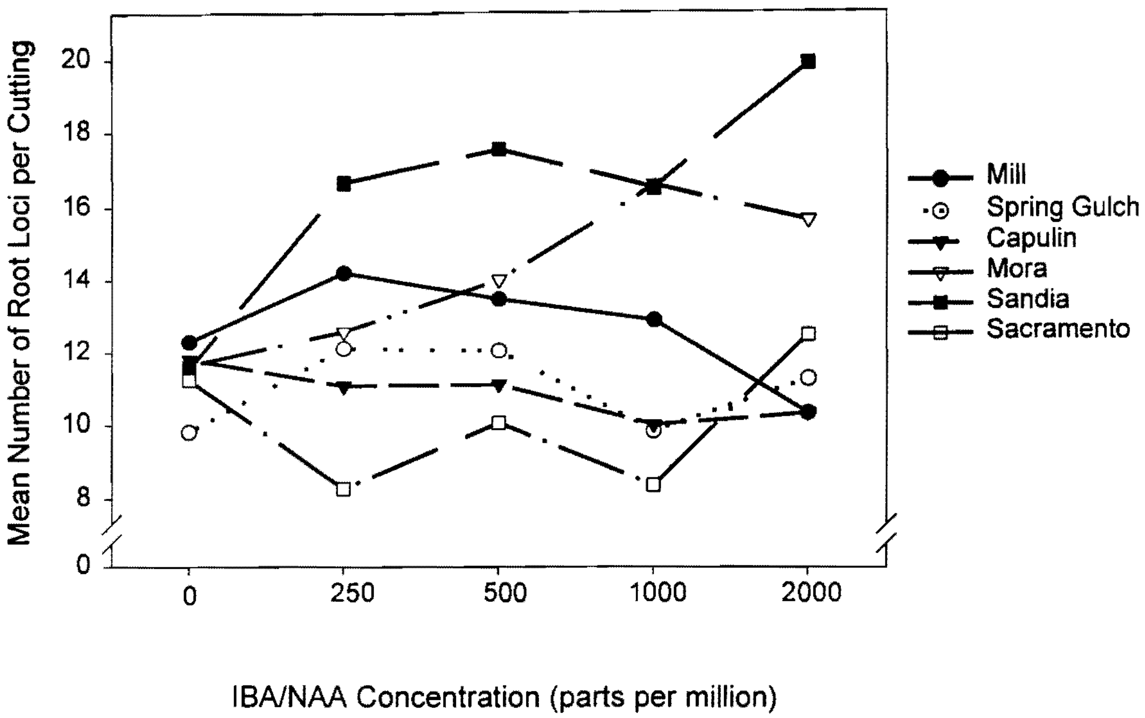

Figure 3. Mountain Snowberry Cutting Study- Effect of Hormone Concentration on Mean Number of Root Loci per Rooted Cutting 150 Days after Sticking by Source

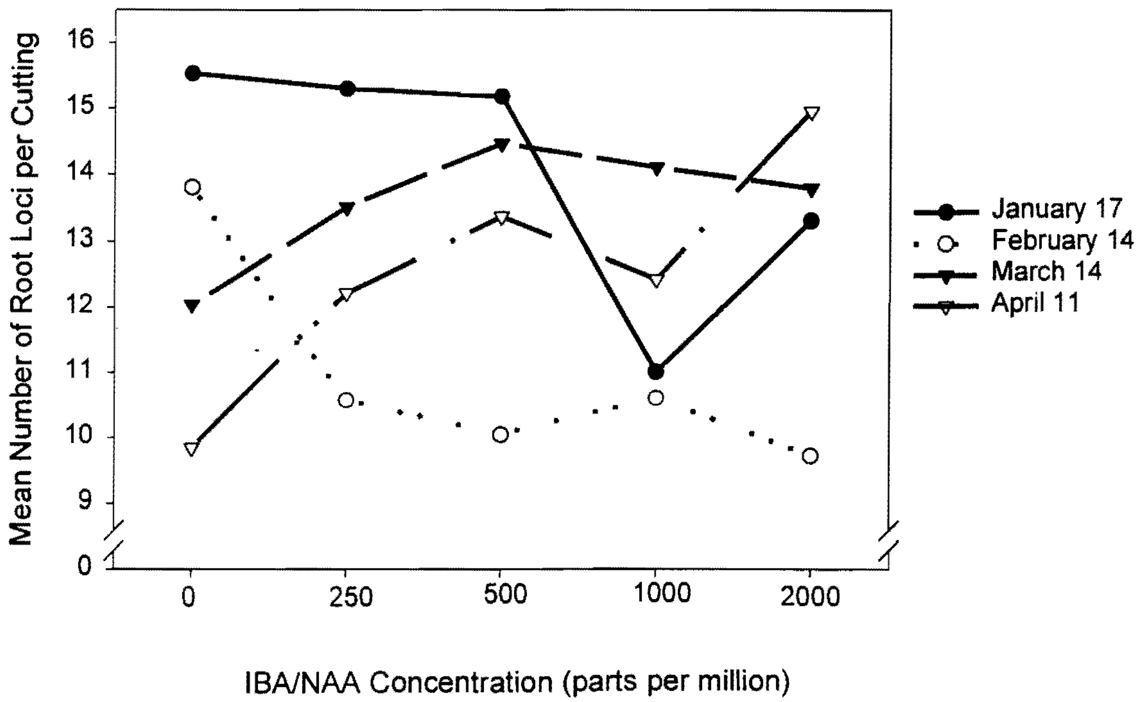

Figure 4. Mountain Snowberry Cutting Study- Effect of Hormone Concentration on Mean Number of Root Loci per Rooted Cutting 150 Days after Sticking by Collection Date

Table 5. Mountain Snowberry Cutting Study- Main Effects of Source, Collection Date, and Hormone Concentration on Length of Longest Root 150 Days after Sticking

Cutting Source	Mean Length of Longest Root (mm)[1]	Standard Error
Capulin	94.6 a	2.68
Spring Gulch	98.0 a	2.93
Mill	91.7 a b	2.41
Mora	85.6 b c	3.66
Sandia	78.7 c	2.39
Sacramento	63.4 d	3.27

Collection Date	Mean Length of Longest Root (mm)[1]	Standard Error
January 17	106.2 a	3.94
February 14	95.5 b	2.40
March 14	85.8 c	2.06
April 11	72.9 d	1.61

IBA/NAA Concentration (parts per million)	Mean Length of Longest Root (mm)[1]	Standard Error
0	84.1 b	2.84
250	88.3 b	2.48
500	86.6 b	2.13
1000	83.6 b	3.92
2000	95.4 a	3.66

[1] Mean longest root lengths followed by same letter are not significantly different.

Source response to collection date measured as length of longest root varied (Figure 5). Although most sources show a decline in the mean length of the longest root as collection date was delayed, Spring Gulch shows an increase through March, followed by a decline from March to April. Also the mean longest root length for Mora cuttings taken in January is much higher than all other source by date combinations, but drops in February to a more mid-range level. There was also much variability in the source by date interaction for rooting response (Figure 1) and number of root loci (Figure 4).

For most sources cuttings from all collection dates leafed out rapidly following sticking and nearly all cuttings were leafed out after 3 weeks with the exception of the Mora cuttings taken in January and February which were poorly leafed out after 3 weeks (Figure 6).

DISCUSSION

Species of snowberry (*Symphoricarpos*) have been successfully propagated by stem cuttings (Anonymous 1948, Dirr and Heuser 1987). Wildings of mountain snowberry, consisting of a small piece of stem having a short length of root, were found to propagate easily (Plummer et al. 1969). Softwood and semi-hardwood cuttings of common snowberry (*S. Albus*) taken June through August rooted from 90%-100%, when treated with IBA-talc solutions of 1000-3000 ppm (Dirr and Heuser 1987). Hardwood cuttings of common snowberry taken in December and January and treated with 3000 ppm IBA-talc rooted 90-100% in 4-6 weeks (Dirr and Heuser 1987).

This study employed a quick dip in a solution of equal parts IBA and NAA to improve the potential for increased root initiation. Concentrations of 250-1000 parts per million IBA/NAA increased the rooting percentage in mountain snowberry cuttings compared to either the 2000 parts per million or no hormone treatments. However, this improvement varied by source and by collection date, and rooting did not improve with any hormone treatment for some source and collection date combinations. Variation in source response to hormone treatment may be ecotypic and/or related to differing environmental conditions at each collection site. Both of these factors could influence the stem morphology, affecting absorption of the hormone solution as well as the duration of time adequate levels of hormone were maintained within the stem.

The growing conditions of the parent plant strongly influence cutting rooting ability (Moe and Anderson 1988). Climate and microclimate of native stands influences the duration of bud dormancy in winter as well as the accumulation of carbohydrates within stem tissue in summer, and these factors will vary from year to year. Hardwood cuttings taken before adequate chilling has taken place root poorly

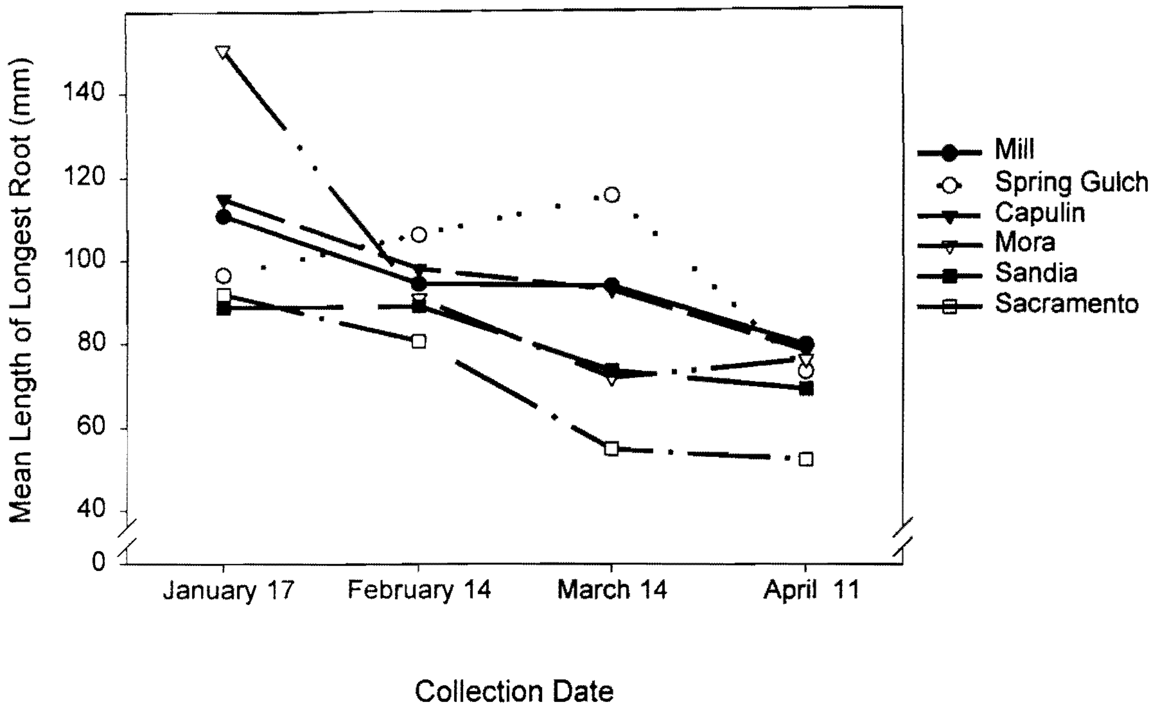

Figure 5. Mountain Snowberry Cutting Study- Effect of Collection Date on Mean Length of Longest Root by Source 150 Days after Sticking

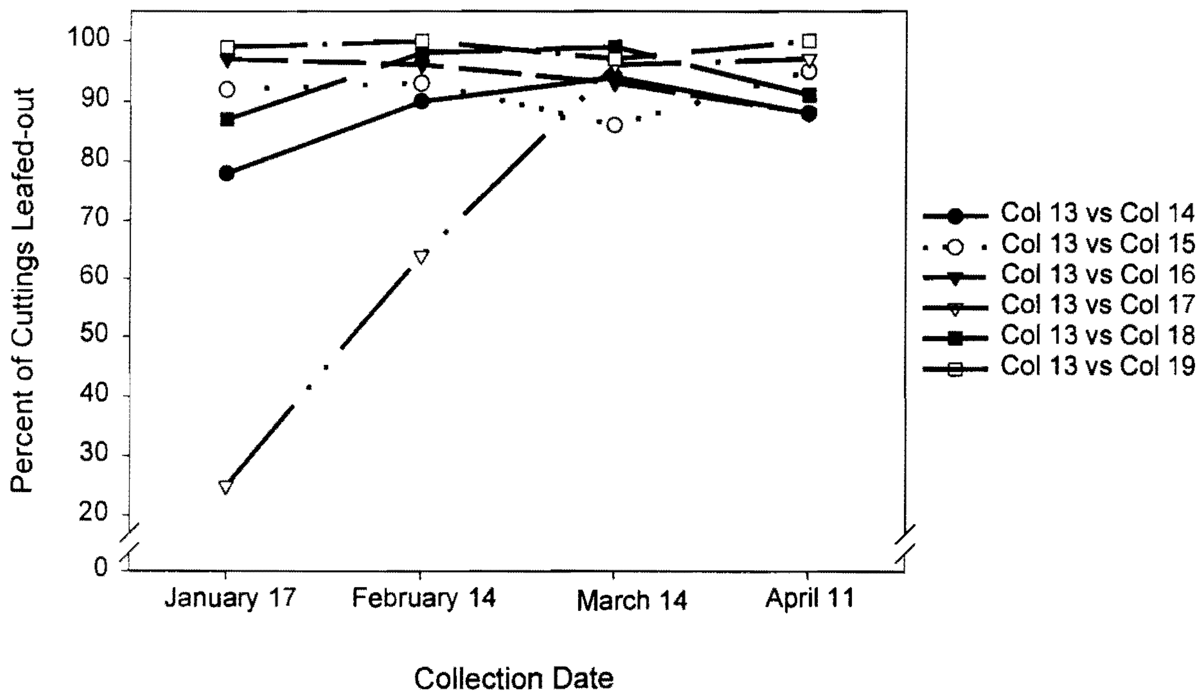

Figure 6. Effect of Collection Date on Number of Cuttings Leafed-out by Source 3 Weeks after Sticking

unless chilled further, and cuttings taken too late in the season may root poorly if the parent plant is in a high state of water stress prior to cutting (Loach 1988). Carbohydrate content within stem tissue may also vary during the dormant season influencing the optimal date for cuttings to be collected (George and McKell 1978).

Rooting percent varied among the sources with the three sources from the more northerly latitudes having the highest percentage of cuttings rooting. Within, the most northerly sources, the three Molycorp mine sources, rooting percentage was inversely related elevation. The shorter growing season at higher elevations may result in less stored carbohydrates in stems. Cuttings from Sandia and Mora had less cuttings root when compared to the Molycorp sources, but a greater number when compared to the most southerly source, Sacramento. The lack of any climatic data from several of these collection sites precludes the assignment of these trends to the stock plant environment or stock plant genotype.

Timing of cutting also influences the response to cultural treatments. Typically, non-dormant hardwood cuttings should be stuck in advance of warm spring temperatures, which can induce rapid shoot growth before adequate root development has taken place resulting in tissue desiccation (Hartmann and Kester 1990). Reduction of the leaf-to-air vapor pressure gradient, which is the driving force behind leaf water loss in cuttings, can be accomplished by maintaining low leaf temperatures by cooling, shading, or misting and by maximizing the ambient humidity in the rooting environment (Loach 1988). These needs must be balanced by the need for adequate irradiance, and the need to maintain the base of cuttings at an optimal level of warmth (generally 23°C - 27°C) for root initiation and development (Hartmann and Kester 1990). In this study, efforts were made to create similar greenhouse conditions and regimes for the various collection dates being examined, a span of 12 weeks. In this study, optimal rooting response occurred in late winter/early spring for all but the highest elevation source from the Molycorp site which had optimal rooting at the preceding collection date. This may be related to the carbohydrate supplies of the cuttings.

Levels of non-structural carbohydrates in mature stems of mountain snowberry reach a yearly high just before the onset of dormancy, and this pool is partially depleted during dormancy (George and McKell 1978). These reserves provide resources to basal root sprouts, which rapidly elongate in the early spring prior to the resumption of growth along older stems (George and McKell 1978). Levels of carbohydrate reserves in stems may also influence the rooting response in stem cuttings as the new root tissues have to compete with other strong sinks, such as expanding leaves, for these reserves.

There were no discernable elevational or latitudinal patterns in meeting chilling requirement (timing of collection) based on initial leafing-out. Nor were there any discernable elevational or latitudinal patterns in rooting percent with the exception of the highest elevation source from the Molycorp site, Capulin.

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