

**Growth Response of Three Native Shrubs
Planted as Un-rooted Cuttings
Across a Wetland Elevation Gradient:
Symphoricarpos albus
Rubus spectabilis, and
*Cornus sericea***

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Abstract

**Growth Response of Three Native Shrubs
Planted as Un-rooted Cuttings
Across a Wetland Elevation Gradient:
Symphoricarpos alba (snowberry)
Rubus spectabilis (salmonberry), and
Cornus sericea (red-twig dogwood)**

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Field propagation of unrooted cuttings (live-staking) provides functional advantages that make it a valuable revegetation tool. The factors affecting survival and growth of Puget Sound indigenous species planted as live-stakes under field conditions have not been systematically tested. Regression analyses were used to evaluate the relative effects of planting time, elevation, diameter, side cuts, number of nodes, and density on the survival and growth of *Symphoricarpos albus*, *Cornus sericea* and *Rubus spectabilis* planted as stakes across a wetland edge. All species were intolerant of spring inundation, and suffered 100% mortality where soils were super-saturated during bud flush. Survivorship increased to 68% for *Cornus sericea* and 100% for *Symphoricarpos albus* above the high water line. December, as compared to February planting, failed to improve survival or growth of all three species, and may reduce survival of *Cornus sericea* and *Rubus spectabilis*. Neither diameter, side cuts, nor density had any effect on survival and growth. For *Cornus sericea*, which can have widely spaced nodes, increasing nodal density increased odds for survival. For *Symphoricarpos albus* and *Cornus sericea*, Forty-centimeter cuttings of thin diameter produced substantial growth in one growing season without irrigation, indicating that field propagation using short cuttings may be a viable restoration practice. *Symphoricarpos albus* is currently not used for staking but has proven to be more reliable than *Cornus sericea*, which is in common use. Mortality appears to be primarily a threshold phenomenon caused by suppression of root initiation under hypoxic soil conditions.

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Dedication

For Ella Faith Cereghino

1. INTRODUCTION

The indigenous vegetation surrounding rivers, beaches, and wetlands in the lowland Puget Sound has been dramatically altered since European colonization resulting in the loss of historic habitat structure and function (Azous & Horner 1997; Collins & Sheikh 2003; Williams et al. 2001). Federal, state and local agencies, non-government organizations, private citizens, and land developers are planting native plants in an attempt to restore native vegetation. Not unlike the case of Humpty Dumpty, successful and efficient restoration of native vegetation depends on understanding: (1) the multiple factors that control spatial and temporal patterns of community composition, (2) the ecological tolerances and life histories of both native and invasive exotic species, and (3) the response of these species to the range of horticultural techniques used for propagation and control.

Live-staking is a horticultural technique commonly used to propagate the genus *Salix* (willow), where a live stem cutting is driven into moist soil, and due to the innate rooting ability of *Salix*, it roots and establishes (Carlson 1938; Chmelar 1974). Similar practices have been rigorously tested for the mass production of *Populus* hybrids for fiber production (Morgenson 1992). Many stream bank revegetation and biotechnical erosion control manuals imply the use of species other than *Salix* and *Populus* as un-rooted cuttings (Gray & Sotir 1996; Tuttle & Wenberg 1996; WADOE 1997). Nursery propagation of native species by cutting is common (Hambrick et al. 1991; Kruckeberg 1982), but few reports have discussed installation of Pacific Northwest species other than *Salix* and *Populus* in the field as un-rooted cuttings (Flessner 1997; Hennelly 1994; Stampfli 1992). Very few sources offer systematic experimentation that can be used to guide specific field practices (Darris 2002a, 2002b).

Cornus sericea (red-twig dogwood) is now commonly planted as un-rooted cuttings, however it is less reliable than *Salix* (Humphreys, pers. comm.; Myers, pers. comm.; Vanbianchi, pers. comm.), and the specific factors that limit survival in the field are not clear (Buis, pers. comm.). Restoration installation contractors often act in isolation of their competitors, on short timelines, with limited budgets and rigid specifications. Designers face similar limitations, and may have even less exposure to restoration practicum than contractors. Installation strategies are rarely designed to test hypotheses, different crews typically complete installation and monitoring, and monitoring is not funded to evaluate installation practices. The convergence of these factors creates little opportunity for precise assessment of revegetation methods. Despite the substantial cost savings offered by un-rooted cuttings, systematic experimentation with stakes in the field is rare and typically unpublished. Field practitioners face very basic decisions with little empirical information: what, when, and how should we harvest, how should we process, store and pre-treat cuttings, and when, where and how should we plant?

An extensive plant propagation literature offers a cacophony of overlapping, species-specific factors and methods that can potentially influence cutting success, including the growing conditions of stock plants (Andersen 1986; Moe & Andersen 1988), stock plant pruning (Cameron et al. 2001, 2003), wood age (Carlson 1950; Darris 2002b; Hartmann et al. 1990), location of cuttings within the stock plant (Howard et al. 1985; Howard & Ridout 1994),

geographic variation within a species (Puri & Swamy 1999), insufficient hormones or rooting cofactors (Edson et al. 1995; Haissig 1974b; St. Hilaire et al. 1996), timing of excision (Howard 1965; Landhausser & Lieffers 1997; Phipps & Netzer 1981; Zasada et al. 1994), pre-disposition of a species based on environmental adaptations (Ewing 1996; Gill 1975; Krasny et al. 1988), pretreatment and installation methods (Hansen & Phipps 1983; Hoag 1993; Hoag & Short 1992; Howard B.H. et al. 1985; Schaff et al. 2002), storage methods (Cram & Lindquist 1982; Phipps & Netzer 1981), disturbance on planting sites (Flessner 1997; Hoag & Short 1992), soil moisture and redox potential during root formation (Darris 2002a; Hansen & Phipps 1983; Rein et al. 1991; Stannard & Guenther 1999), and efficiency of carbohydrate metabolism (Tschaplinski & Blake 1989). Few of these studies provide evidence from which to infer the relative importance of these factors on the success of un-rooted cuttings planted for revegetation.

In this study three native shrubs, *Cornus sericea* (red-twig dogwood), *Rubus spectabilis* (salmonberry), and *Symphoricarpos albus* (snowberry), were planted as un-rooted cuttings across a wetland elevation/moisture gradient in the Union Bay Natural Area, Seattle, Washington. Their survival and growth was measured and evaluated as a function of the elevation, planting time, and the physical characteristics of each cutting (Table 4.1). These three species occur with varying frequency in Puget Sound lowland wetlands. *Cornus sericea* is primarily associated with wetland vegetation and is a likely indicator of hydric conditions. *Rubus spectabilis* occurs both in and out of wetlands, and is frequently associated with riparian forest. *Symphoricarpos albus* frequently occurs outside of wetlands and in quite dry locations (Reed 2003).

Table 1.1 – Habitat distribution of species studied

Nomenclature follows Kartesz (1994). Wetland identifier classification includes updates from the 1993 addendum to Reed (1988).

Species	Reed 1988	Hitchcock & Cronquist 1973	Klinka et al. 1989	Pojar & MacKinnon 1994
<i>Cornus sericea</i> <i>ssp. occidentalis</i> Red-twig dogwood	FACW	(no comment on habitat)	Very moist/wet soils, alluvial flooding, nitrogen-rich soils, moder/mull humus form	Moist soil, typically in swamps and streamside forest and scrub, but also in open upland forest and thickets.
<i>Rubus spectabilis</i> <i>var. spectabilis</i> Salmonberry	FAC+	moist woods to stream banks or swamps	Very moist/wet soils, fluctuating ground water table, nitrogen-rich soils, moder/mull humus form	Moist to wet places, often abundant along stream edges, avalanche tracks and in wet logged areas
<i>Symphoricarpos albus</i> var. <i>laevigatus</i> Snowberry	FACU	widespread species of thickets, woods, and open slopes	Broad moisture tolerance, nitrogen-rich soils, moder/mull humus form	Dry to moist, open forests, thickets, rocky slopes, river terraces, ravines

This study was designed to test several previously untested propositions that could be applied directly to field practice:

- (1) Soil saturation, as determined by elevation, affects live-stake survival and growth in palustrine wetlands.
- (2) Earlier planting time increases stakes survival and growth by increasing the time for root development before bud flush.
- (3) Species respond differently to soil saturation and inundation based on their adaptation to life in hypoxic conditions.
- (4) Among cuttings from equally aged stems, greater cutting diameter improves growth response.
- (5) Field planting of short cuttings (40 cm) is a viable practice for plant propagation in seasonally moist soils of the Pacific Northwest.

2. LITERATURE REVIEW

2.1 Scope and Definitions

Sticking cuttings in the field may be among the first practices of perennial agriculture. Aristotle (384-322 B.C.) reported the use of un-rooted cuttings in the field (Haissig & Davis 1994). Rooted poles from *Salix piperi* were used as self anchoring fish weir posts by local tribes (Gunther 1945). Living fences of woven rooted cuttings are still used to contain livestock in the remote undeveloped mountains of Southeast Asia (pers. obs.). Despite ancient roots and the utility of sticking cuttings in the field, very few authors describe systematic experimentation within the Pacific Northwest flora. The goal of this review is to collect information to support decision making during field propagation of un-rooted cuttings. Where possible, sources are selected that describe rooting of dormant cuttings from deciduous species in temperate climates.

There is a substantial plant propagation literature that evaluates a variety of procedures and isolates physiological processes related to adventitious rooting. The goal of this review is not to exhaustively cover the history and methodologies of plant propagation (see instead Davis et al. 1988; Hartmann et al. 1990; Macdonald 1986), nor scratch at the surface of the 125 year history of adventitious rooting research (see instead Haissig & Davis 1994; Haissig et al. 1988). The complexity of genetic factors, their nature and heritability, and their applicability to live-staking is also beyond the scope of this work (see instead Riemenschneider 1994, Haissig & Riemenschneider 1988; Ernst 1994). Four overlapping bodies of knowledge were integrated to complete this review:

- (1) Plant physiology studies that observe adventitious root formation under controlled conditions to determine factors controlling innate rooting potential of cuttings.
- (2) Plant propagation studies that demonstrate the effects of different propagation procedures.
- (3) Studies, reports, and manuals from land management professionals on the response of selected species to field propagation.
- (4) Unpublished information gained through conversations with landscape and nursery professionals based on their experimentation, field observations, and intuition.

Field propagation of un-rooted cuttings is a simple technology. During dormancy a stem from a parent plant is cut into sections. The stem sections (or cuttings) are then partially or completely buried on a field site – the simplest method being live-staking where a cutting is driven vertically into moist soil until only a portion remains above ground. If sufficient adventitious roots form in time to support shoot growth the plant may survive summer drought. The planting can persist if the species are sufficiently adapted to site conditions to persist and compete with co-occurring vegetation.

The ability of woody cuttings to self-anchor while dissipating energy in moving water, and to self-repair and increase their structural strength over time has led to the use of cuttings in a variety of erosion control systems collectively referred to as biotechnical erosion control, soil bioengineering, or ground bioengineering. Field propagation of the genus *Salix* (willow) is common and well documented throughout North America and Europe where sticking cuttings has evolved into a variety of techniques of increasing structural complexity: contour and down-slope fascine installation, live post installation, live fencing, brush layering, and brush mattresses. As described in a variety of readily available manuals and specifications these techniques can effectively stabilize steep slopes, slow and diffuse runoff, trap sediment, drain soil water, secure large woody debris, and diffuse or direct energy in stream and river channels (Bentrop & Hoag 1998; Gray & Sotir 1996; Morgan & Rickson 1995; Schiechl & Stern 1996; Tuttle & Wenberg 1996). Many of the shrub species that can be established by staking can form persistent shrub lands that are resistant to tree invasion. These shrublands may have value in power line rights-of-way or other controlled environments where low maintenance plant communities of limited height are desired (Meilleur et al. 1994).

Unrooted cuttings can be installed in environments where installation of potted material is not possible. Poles and stakes can be driven into riprap or cobbles where transplanting potted or bare root stock is not an option. Proboscis-like back-hoe attachments (Hoag & Ogle 1994), hand held water pressure drills (Hoag et al. 2001; Kaplan, pers. comm.) and impact drills (Kaplan, pers. comm.; Stamphli 1992) have been used to insert unrooted cutting deep into low water tables, through large riprap, and into roadside cut slopes and other compacted soils. Long pole cuttings can be inserted through gabions (rock filled wire baskets) during installation to reach soil and increase the ecological function of engineered structures (McCullah 1999).

This summary is divided into four sections that focus on the simple practice of staking, and moves from basic principles to field applications. Section 2.2 discusses the limitations of existing literature. Section 2.3 summarizes the advantages and risks that make staking unique and viable alternative to planting rooted or seed grown stock. Section 2.4 presents basic principles of adventitious rooting that are likely to affect rooting of stakes. Section 2.5 summarizes practical considerations including harvest, pre-treatment, storage and planting.

This review and the associated original experimental research is intended to assist practitioners in the lowland Pacific coastal rainforest ecosystem working with un-rooted cuttings, however the principles presented could be applied to work in many ecosystems. The term restoration is used occasionally, and should be understood in the context of the discussion provided by the Society for Ecological Restoration (2002). Native vegetation refers to the composition of species present in a defined region before the colonization of this region by European civilizations.

2.2 Limitations of Existing Information Sources

Publications that systematically evaluate rooting potential of Pacific Northwest native species under varied field conditions are practically non-existent. Many horticultural and physiological studies tend to focus on hard-to-root

species or clones which have value to the ornamental plant industry, use procedures not appropriate for economically viable field practice (Cameron et al. 2003; Cameron et al. 2001), focus on the isolation of biochemical processes that control rooting (Davis et al. 1988), or study the influence of stem characteristics that cannot be economically controlled during a harvest operation (Howard & Ridout 1991a, 1994). As a result there are severe disadvantages to using existing scientific literature as a foundation for determining field practices:

- (1) Given the limited number of native species studied, results must be extrapolated between cultivars and species, or even genera and families, while rooting potential clearly varies even within a species.
- (2) Statistically significant variation in average response under controlled conditions may not indicate economically or ecologically relevant differences in performance under field conditions.
- (3) Interaction between environmental factors and fluctuation of factors over time that influence rooting may overwhelm observed response to isolated factors under controlled experimental conditions.

Extrapolating greenhouse studies to the field may lead to unnecessarily increasing costs by trying to control all risks, even those that have no cost-benefit or bearing on field survival. To overcome these challenges physiological models must be tested under a range of realistic conditions that allows for better determination of how the complex interactive factors of the field environment influence cutting success.

Biotechnical manuals reviewed rarely cite sources of information or provide evidence supporting the specifications offered in the description of procedures (Gray & Sotir 1996; King County 1993, 2004; Schiechl & Stern 1996; WADOT 2000), or tend to avoid specific discussion of plant materials (BarkerBarker 1995; Morgan & Rickson 1995). Recommendations made in these manuals are sometimes adopted *verbatim* and used in contractual documents that limit the methods contractors are permitted to use in field practice (GCSWCD 2004). Field trials conducted by local or national resource agencies are sometimes implemented with limited resources to serve a specific resource protection needs (Hennelly 1994; Hoag & Short 1992; Stamphli 1992), and typically provide a limited description of methods, fail to control or discuss potentially confounding factors, or do not present data that can be independently evaluated.

A few recent studies specifically intend to inform field practices by systematically observing the response of cutting materials to field conditions (Darris 2002b; Hoag & Short 1992; Schaff et al. 2002). These studies have built on classic observational studies of *Salix* (willow) rooting (Carlson 1938, 1950) and commercially motivated research on the propagation of *Populus* (poplar and cottonwood) for which there is a sizable body of literature (Petersen & Phipps 1976; Phipps & Netzer 1981; Tschaplinski & Blake 1989). Even within the context of controlled experimentation, the mechanism through which rooting potential is controlled has not been clearly identified (Haissig & Davis 1994).

Given the absence of an organized academic interest in developing and testing live-stake or site-scale restoration technologies, and the unreliability of existing technical resources, the field practitioner has the potential to take a leadership role in advancing the science-based development of site restoration practice. To incorporate the communicability and repeatability of science into the art of revegetation would serve to increase interaction between practitioners, the rigor of methods, and out-planting success, for the benefit of our severely degraded ecosystems.

2.3 Advantages and Risks of Using Un-rooted Cuttings

The cost of ecological restoration is highly variable in part due to the variable costs of revegetation (Zentner et al. 2003). The full costs of successful revegetation includes not only the cost of installation but also the maintenance required to enable the planting to reach project objectives.

Given availability of cutting stock and volunteer labor, revegetation with live-stakes can be accomplished with no cash resources. Potential sources of cuttings include public and private land, harvest from suburban development sites, harvest of unwanted shrub regeneration on cleared forest land, utility rights-of-way, or stormwater retention sites, or through agricultural cutting block production. Local regulations may limit harvest of materials in sensitive areas (King County 1993).

Live-staking represents an opportunity to significantly reduce the per-plant costs of revegetation. Costs of plant installation are typically estimated based on the cost of materials and an estimate of hourly planting rate (Alexander, pers. comm.; Humphreys, pers. comm.; Partridge, pers. comm.). Availability of materials, logistics, hauling distances, and labor rates can cause actual revegetation costs to vary by an order of magnitude (Canty et al. 2003; Ogle et al. 2000). In the lowland Puget Sound, an 18" cutting may cost \$0.30-0.60 as compared to \$2.50-3.00 for a 1-gallon pot (2004 prices). According to one contact, one-gallon pots can be transported, placed, and planted at a rate of 5-10 per hour depending on site conditions, while stakes may be planted at a rate of 40-120 per hour (Humphreys, pers. comm.). Based on a billing rate of \$20 per hour, a stake can be installed for \$0.30-1.10 each whereas a one-gallon potted plant may cost \$4.50-7.00 installed. Thus 6 to 15 stakes can be installed for the price of each pot. Where staking materials are available for free from salvage locations, and where labor for stake harvest is subsidized through use of conservation corps programs, prisoner labor, or volunteers, the cost-advantage of staking increases even more dramatically.

The ease and cost effectiveness of staking can yield other benefits on a restoration site. The higher planting density afforded by stake planting is useful for rapid increase of cover and biomass on barren sites, and expansion of stake plantings by maintenance crews using on-site materials can offset losses in coverage due to failure of initial plantings (Kleindl, pers. comm.; Partridge, pers. comm.). Dense live-staking following mechanical or chemical treatment has been used to effectively control *Phalaris arundinacea* (reed canarygrass) (Brandvold, pers. comm.; Cantrell, pers. comm.; Vanbianchi, pers. comm.). Given predictable survivorship, reduced per-plant costs allow increased understory planting diversity or density. High-density plantings can more closely resemble natural

regeneration patterns, where high recruitment density results in rapid canopy closure and competitive thinning, which may decrease opportunities for non-native shade-intolerant species to invade.

Species that rapidly form adventitious roots are often species that are adapted to life in frequently flooded environments (Gill 1975; Krasny et al. 1988). The list of Pacific Northwest species with potential for staking is a role-call of common shrubs found in floodplains and wetlands (Appendix A; Kunze 1994). Staking as a propagation method may be particularly viable in these moist environments due to the potential for developing roots to reach moisture after more xeric soils begin to dry in late spring or early summer. Several shrub species that stake readily are also adapted to more mesic conditions (*Symphoricarpos albus*, *Philadelphus lewisii*, *Oemleria cerasiformis*, *Sambucus racemosa*). Supplemental irrigation may allow for staking under drier conditions. Agricultural-grade drip irrigation can increase survivorship and add as little as \$0.50 per plant (2004 prices), while saving water and reducing growth rates among weedy species by more precisely delivering water compared to overhead irrigation.

First-year survival and growth rates for stakes may be less than those for potted material for some species. Effective weed control over the establishment period may be critical for success of stake plantings (Vanbianchi, pers. comm.). The cost of increased planting density, site preparation, or maintenance to reach target planting coverage must be considered when comparing the cost-benefit of stakes to other stock. Survival and growth rate are not only controlled by stock type, but are also influenced by the adaptation of species to site conditions, the skill of the planter, the extent of aftercare, and the fertility of soils which can be partially mitigated by site preparation and management (Myers, pers. comm.; Partridge, pers. comm.).

There is a risk of catastrophic mortality of any planting where factors controlling survival are unknown, hydrology is not well surveyed, or where there is weak control over the quality of materials or planting technique. One informant even mentioned the importance of making sure that third party stake harvesters actually deliver living wood (Cooke, pers. comm.). Given the value of immediate success and short timelines for site development, potted material can offer an additional level of security on many projects (Myers, pers. comm.; Vanbianchi, pers. comm.) and increasing the predictability of stake survival may be necessary before stakes are widely used in some settings.

Field propagation does avoid some issues associated with nursery-grown potted stock. Potted stock may have difficulty adjusting to differences in soil characteristics between the potting medium and the native soil conditions or may be stunted due to sub-standard nursery practices (Harris et al. 2004; Hummel, pers. comm.). On potted stock plantings in poorly drained soils, runoff may collect in the plugs of more permeable potting medium resulting in localized hypoxia and high mortality (Harris et al. 2004). Addressing these issues during planting by removing pot-bound roots, removing potting medium or more elaborate construction of planting holes further increases the cost of planting potted stock and depends on well-trained labor. Root systems developing from stakes are formed under, and therefore are adapted to site conditions, and are free of defects caused by nursery or planting practices.

The initial growth rates of different species, planted as different stock types, under variable conditions, are poorly understood, and little empirical evidence is available to guide a practitioner. A window of opportunity follows site preparation when competition has been reduced. In highly degraded environments, where recruitment and competition of non-native species are strong, plantings must show adequate vigor and coverage within a brief 2-4 year window or site managers must choose between rapidly escalating maintenance costs or loss of plantings. Simultaneously, high-density plantings are avoided both due to higher cost and horticultural habits associated with managed landscapes. A more systematic analysis comparing the growth and survival of different stock types and species across soil, hydrologic, and light gradients would be a significant contribution to restoration field practice.

Genetic diversity may be compromised by stake propagation. A common objective of restoration is to mimic the structure and composition of reference vegetation (i.e. relatively un-degraded plant communities found in a similar location and under similar environmental conditions). Staking, as a form of vegetative reproduction, replicates the genetic identity of the parent plant, and may reduce the diversity of genetic material in a planting. Restoration planners have proposed to mimic not only composition and structure, but to the extent possible, the genetic character and variation of local or historic vegetation (Clewett et al. 2000; Jones 2003). Reduced genetic variation may reduce a population's ability to adapt to changing climatic or disturbance regimes. In application, genetic reference states are typically not available, or environmental conditions have changed such that a reference vegetation or genotype is not necessarily adapted to current site conditions (Jones 2003). The extent of genetic variability within Pacific Northwest native shrub populations has not been analyzed. Many species potentially propagated by staking also self-propagate vegetatively, both through rhizomatous expansion of clonal populations (*Symphoricarpos albus*, *Rubus spectabilis*, *Rubus parviflorus*, *Rosa nutkana*) or through dispersal of vegetative fragments with alluvial disturbance (*Salix spp.*, *Populus trichocarpa*, *Cornus sericea*). (Cooke 2004; Pojar & MacKinnon 1994). The relative contribution of sexual and vegetative reproduction on the genetic variation within native populations is undocumented. Washington State Department suggests using cuttings from 30-50 "unrelated" donor plants (WADOT 2000) a number identical to the policy established by Huber (1993). If this policy, or another more rigorous approach to maximize genetic variation were actually implemented the costs of harvesting staking materials would likely increase. Complicated harvest operations not only increase travel time and storage needs, but may increase management costs (King County 1993).

The U.S. Department of Agriculture's Natural Resource Conservation Service has run trials to select individual genotypes that root well when staked under agricultural field conditions. These individuals would be 'released' for mass-production and sale as 'performance-tested ecotypes' for stream bank erosion control (Zierke 1994). Agricultural mass production of genetic individuals for broad scale revegetation has been attempted in the forest nursery trade, and raises questions about the ability of genetically simplified populations to adapt to microclimatic variation, resist disease, and adapt to changing environmental conditions. Some NRCS staff have discussed selection of genetic stock based on adaptation to microclimate, but not in the context of population genetic diversity

(Crowder & Darris 1999). Using seed grown stock from a carefully selected provenance for cutting stock, or supplementing stake plantings with bareroot plantings of seed grown stock are two options for increasing genetic variation in staked out-plantings while taking advantage of the reduced costs offered by staking. Ironically staking has the potential to undermine the restoration of complex genetic structure, while simultaneously allowing cash-poor practitioners to mimic local genetic identity by cloning material from an adjacent plant community. The practical cost advantages of staking will remain in conflict with the ideal of precise restoration of diverse genetic identity, as long as restoration of native vegetation is limited by capital and time. Genetic management of revegetation will require changes in the legal structures that mandate revegetation, the timelines under which revegetation is implemented, and improved tracking of provenance in the native plant industry. On many projects, simple revegetation is still elusive without the addition of rigorous genetic objectives.

2.4 Theory – Variation in Rooting Potential

Hartmann (1990) divides the formation of adventitious roots into three phases: (1) the initiation of dedifferentiated root primordia, 2) the differentiation of root primordia into recognizable proto-roots, and 3) the development of vascular connections with the emergence of new roots through stem tissues. The development of roots is dependant on the formation of root primordia which as Cameroon et al. (2003) describe is dependant on cuttings being in a ‘favorable state’ at the time of excision.

Rooting potential is typically measured as the number, length or biomass of roots that form from a cutting. The concept of rooting potential is imprecise. The quantity of roots formed from a given cutting is controlled by factors both intrinsic and extrinsic to the cutting itself. Cameron et al. (2003) concludes that rooting is indeed dependent on environmental conditions that ‘support the expression of inherit rooting potential’. In short maximized rooting potential involves a series of potentially interactive conditions: the genetic predisposition of the genotype, the state and history of the stock plant tissue, pretreatment and storage conditions, and the dynamic environmental conditions in which the cutting is finally placed.

2.4.1 Predisposition and the Process of Adventitious Root Formation

A detailed review of early work on the origin, timing and development of adventitious roots has been completed by Haissig (1974a; 1974b; 1974c). In an intact plant, auxins are actively transported in the phloem, cell to cell, from the buds toward the roots (basipetally) at about 1 cm per hour (Salisbury & Ross 1985). When this flow is interrupted by excision or damage to the phloem, auxin begins to accumulate above the injury. The co-occurrence of sufficient auxin and rooting co-factors in predisposed cells may trigger the dedifferentiation that precedes formation of root primordia. Auxin may also trigger the growth of dormant root primordia that were pre-formed during shoot development (Haissig 1974b).

Species can be divided into those that pre-form root primordia during shoot development, and those where root primordia are induced only after cutting. The presence or absence of preformed root primordia in two *Populus* species has been associated with the rate of root development (Okoro & Grace 1976). Preformed root primordia have been identified in *Citrus medica* (citron) and some species of *Malus* (apple), *Cotoneaster*, *Lonicera* (honeysuckle), *Salix* (willow), *Populus* (cottonwood and poplar), *Ribes* (currant and gooseberry), and *Jasminum* (jasmine), and can be observed as protuberances after removing the bark (Carlson 1938; Haissig 1974a; Hartmann et al. 1990). Pre-formed root primordia may form in new shoots of *Salix* as early as June in Illinois (Carlson 1938) and may remain dormant for years until a change in the physical or biochemical environment stimulates their growth (Carlson 1950).

In woody plants, root primordia typically form in the young phloem, associated with parenchymous rays (Carlson 1938), or leaf traces adjacent to cambium, or even within cambium (Hartmann et al. 1990). The location of formation may vary depending on the species and the age of the stem. The primordia begin to develop the structures associated with a root tip as it pushes through the cortex dissolving thin walled cells and rupturing fibrous layers (Haissig 1974a). Over this period, carbohydrate levels in the cutting may drop as reserves are mobilized and transported to sites of root initiation (Okoro & Grace 1976). By the time roots have emerged they have completed their vascular connection to the xylem (Hartmann et al. 1990). As stems age, tissues capable of dedifferentiating to forming primordia may become less common, accounting for loss of rooting ability in older stems of some species (Haissig 1974a).

The delay between cutting excision and formation of root primordia, and again between formation of root primordia and emergence of roots, varies between species. *Salix* and *Populus* can produce roots in 7 days (Hoag 1994) (Tschaplinski & Blake 1989), or under ideal conditions in as few as two or three days (Alexander, pers. comm.). Shoots from buried *Populus* cuttings planted under field conditions typically emerge in 7-14 days (Morgenson 1992). Although there is considerable variation between species, several studies suggest that pre-formed root primordia most commonly form at nodes, or to a lesser degree between nodes, while induced primordia most commonly form near the base of cuttings (Carlson 1938; Haissig 1974a). *Salix cordata* produces up to five pre-formed primordia in a predictable arrangement around each bud in the first year of growth (Carlson 1938). A review of 107 species of *Salix* divided willow into two categories based on rooting habit. Vigorously rooting species root from throughout the stem, while the least vigorous species root primarily from the basal cut (Chmelar 1974). As wood grows radially pre-formed root initials grow with the phloem and continue to develop (Carlson 1950; Haissig 1974a). Additional root initials may form in the internodes as the wood ages. When older cuttings of *Salix cordata* were placed in water an increasing percentage of roots originated from internodes than from nodes (Carlson 1950). This clean division between basal rooting and nodal rooting was not clearly observed among 22 Pacific Northwest native shrubs tested by Darris (2002a). Many species appeared to root from nodes, the cutting base, and along internodes, and the distribution roots was not quantified.

Callus formation is frequently described as resulting in adventitious rooting (Dirr 1998), however Hartmann (1968) suggests that rooting is independent of callus formation, although both rise from similar internal and environmental conditions. Haissig (1974a) states that root primordia may form in undifferentiated callus tissue, however formation of callus does not assure root formation.

Varied rooting potential within a species is well represented by horticultural selections that are harder to root than their parent species (Cameron et al. 2003; Hambrick et al. 1991), varied rooting ability between selected hybrids of similar parentage (Morgenson 1992; Petersen & Phipps 1976), and varied rooting ability across the geographic range of a species (Puri & Swamy 1999). Prediction of rooting ability may require understanding patterns within a species as well as between species. Adventitious rooting has been clearly associated with species found in frequently flooded or inundated environments (Gill 1975; Krasny et al. 1988). Populations found where rapid root formation improves survival may develop a genetic predisposition to adventitious root formation. To what degree the rooting ability found in riparian species is an individual response to environmental conditions and/or a result of genetic selection has not been evaluated.

2.4.2 Timing of Excision and Planting

Stakes must be cut and planted so that sufficient roots form before the water demand from growing shoots exceeds supply, causing stomatal closure and inhibition of photosynthesis. Insufficient early root formation can increase mortality at bud flush (Okoro & Grace 1976). Inadequate root formation during early growth can have significant cumulative impact on growth rates. A seedling study of four *Populus* clones known to vary in their growth rates indicated that faster growing clones had greater rates of root elongation. A small difference in estimated net carbon assimilation rate (20%) resulted in double the biomass over the 5 week study as the impact of slightly higher growth rates is compounded by the accumulation of belowground structures over time (Pallardy & Kozlowski 1979). Loss of productivity due to water stress from slow root growth likely reduces the ability to grow roots to forage for water – a feedback that leads to dry season die-back or death.

To allow time for root development, stakes are typically harvested and planted as hardwood cuttings (cuttings that are taken during shoot dormancy). Little information on the best time during dormancy for cutting excision and planting. The biochemical condition of cuttings and, in particular, carbohydrate and nitrogen concentrations can vary over the course of a year, and more specifically, over the course of the dormant season. Hambrick et al (1991) found that rooting ability of a *Rosa multiflora* hybrid was greatest when total (soluble and insoluble) carbohydrate/nitrogen ratios were lowest, during a period from November 15th to December 15th (Texas, USA) suggesting that biochemical cycles associated with dormancy may also influence rooting ability of excised cuttings.

Biotechnical manuals and specifications provide a wide range of rationales for timing of staking. Early spring is prescribed by some (WADOT 2000), others suggest fall and spring (Myers Biodynamics 1993), others suggest the entire period from leaf fall to bud break (Schiechl & Stern 1996). The only rationales found for not planting during

winter was the risk of soil compaction and erosion during winter construction (King County 1993), the potential for loss of material during flood events in riparian environments, and the potential for frost heave in cold winter climates (Crowder & Edelen 1996).

The extent to which roots develop in cuttings stuck in winter during bud dormancy is undocumented. Neither temperature nor moisture are limiting for photosynthesis in conifers during our mild maritime winters, however, deciduous species may not be able to take advantage of the extended growing season when buds are dormant (Waring & Franklin 1979). The presence of leaves is essential for rooting olive cuttings (Hartmann & Loreti 1965). Limitation of rooting by shoot dormancy may relate to associated changes in the concentration and distribution of growth regulating compounds and resources required for root initiation. In a rooting trial of 107 species of the genus *Salix*, 50% of species grew roots and shoots simultaneously, 10% grew roots slightly later than shoots, and 40% formed roots only after a delay of up to 15 days (Chmelar 1974). Slower rooting was correlated with lower levels of root production. This ability to form roots rapidly following bud break may also correlate with the presence of preformed root primordia, although this was not explicitly studied.

Changes in rooting potential have been documented to occur before bud break. In hardwood cuttings of apple, Howard (1965) observed that rate of root formation increased as cuttings were taken later in the dormant season and suggested that decreasing bud dormancy over winter led to increasing production of basipetally transported rooting co-factors. This effect of increased rooting ability during late dormancy was also found in *Actinidia* by Ercisli (2003) but not observed in *Salix* with pre-formed root primordia (Swingle (1929) cited in Hartmann (1968) and Haissling (1974a)). If cuttings are able to develop root primordia during dormancy there may be some benefit to taking and planting cuttings in early winter. This may be counterbalanced by the risks of desiccation faced by cuttings placed in the field before root formation. Moist cold storage of cuttings between early excision and out planting may be a beneficial combination of treatments and will be discussed in section 2.5.5.

The use of leafy cuttings in the field is not common practice. Although vigorously growing shoots (called softwood cuttings) may root well in controlled nursery environments (Hartmann et al. 1990) and may be necessary for propagation of some difficult-to-root species (Howard 1996), no author suggests field staking during bud break or early shoot extension. Easy rooting taxa like *Salix* have been successfully rooted when planted as semi-hardwood cuttings (taken after cessation of shoot extension but before leaf senescence when the stems are well buried and sufficient soil moisture is present (Myers, pers. comm.)).

Several studies have examined the ability of root or shoot fragments to establish when cut and buried during the growing season, with later season wood showing increasing ability to survive excision, which was correlated with increasing levels of carbohydrate accumulation (Landhausser & Lieffers 1997). Given adequate moisture, field staking may begin well before dormancy for some species, although some practitioners are inclined to strip leaves to reduce transpiration that may desiccate cuttings (Darris, pers. comm.). Others indicate that if leave senescence has

begun that the shock of excision will cause rapid leaf drop making stripping unnecessary (Cullum, pers. comm.). Given the potential benefits to rooting from compounds produced by shoot metabolism, and the ability of cuttings to maintain water status given sufficient soil moisture, excision and planting just prior to dormancy deserves further exploration.

2.4.3 Carbohydrate and Nitrogen Status of Stock Plants

The growing conditions of stock plants will influence the nutrient status of cuttings. The risks of poor rooting ability in cuttings from plants growing under environmental stress or competition has been mentioned (Hoag 1993), but clear connection between growing conditions of stock plants, variation in stem nutrient content, and rooting ability of stakes has not been tested for native species.

A detailed summary of metabolism during adventitious rooting can be found in Haissig (1974c). Varied results in published studies provide a complex picture of the relationship between plant nutrient status and rooting ability. A single measure of total non-structural carbohydrates does not correlate readily with rooting ability in all species. Carbohydrates do accumulate at sites of root initiation (Haissig 1974c), and carbohydrate levels have been observed to fall rapidly during root formation in *Populus* cuttings (Okoro & Grace 1976). The ability of *Rubus spectabilis* to re-grow shoots and roots from rhizome fragments has been correlated to seasonal changes in non-structural carbohydrate levels, with rhizomes harvested in early summer having the lowest carbohydrate content and the least ability to form shoots or roots (Zasada et al. 1994). The ability of stem fragments of *Cornus sericea* to root also increased over late summer as carbohydrate content increased (Landhausser & Lieffers 1997). This pattern of increasing rooting ability approaching dormancy was also observed in root fragments of *Cornus sericea*, *Viburnum edule*, and *Corylus cornuta*. Interpreting carbohydrate concentrations as limiting rooting potential may be inaccurate. The influence of carbohydrates may simply parallel the effects of other environmental and developmental fluxes (Veierskov 1988). In some cases, variation in rooting potential may be controlled not by carbohydrate concentration, but by mechanisms of carbohydrate metabolism, with individuals that are able to biochemically mobilize carbohydrate reserves more rapidly showing increased early root growth (Tschaplinski & Blake 1989). Where plant material is not grown under deficiency there is no clear evidence that inadequate carbohydrate levels would be a limiting factor in root development in hardwood cuttings. There are many other factors related to timing of excision and planting (sections 2.5.1 and 2.4.2) that must be considered in conjunction with carbohydrate status.

The effect of nitrogen levels in cuttings on rooting ability is also complex. Nitrogen (N) will move to the base of a cutting after excision, similar to carbohydrates. However, this movement of N has been observed whether or not rooting is successful (Haissig 1974c). Some studies suggest that a high carbon/nitrogen ratio favors primordia development, however, total carbon (C) or N levels may independently influence rooting regardless of their ratio, with excessively low N levels reducing rooting ability (Haissig 1974c). The balance between different nitrogen-

containing compounds may further affect rooting ability (Veierskov 1988). A species may be able to generate high nitrogen levels through effective mobilization, despite low total nitrogen levels within the cutting. It is likely that there is an optimum window of available nitrogen, above or below which rooting ability may be compromised (Haissig 1974c). This being said, sub-optimal nitrogen fertilization has been associated with increased rooting potential in some herbaceous species (Andersen 1986). At this point insufficient evidence exists to inform a practical strategy for manipulation of nitrogen levels in cuttings, and nutrient levels must be considered in conjunction with many other factors that may effect rooting ability.

2.5 Practicum – Maximizing Rooting Potential

The plant nursery industry uses a variety of treatments to increase rooting potential. For revegetation the cost per cutting must be kept low to maintain the economic advantages of planting un-rooted cuttings. This limits the scope of procedures that can be economically integrated into a revegetation project to increase rooting potential. For these reasons techniques that involve labor-intensive treatment of individual cuttings like girdling, or that involve significant control of growing climate, like increasing day length or carbon dioxide concentration, will not be considered.

2.5.1 Harvesting and Producing Cuttings

Several sources recommend transporting branches whole, to be cut into stakes just before installation (King County 2004; Schiechl & Stern 1996). Others process stakes at the harvest site for easy transport.

For optimal re-growth of shrubby species, Gray and Sotir (1996) suggest that leaving a minimum of 8 inches of stump after cutting, and that selective rather than clear cutting may be better for healthy regeneration. The traditional horticultural practice of cutting stems at an angle to prevent water from collecting on cut surfaces may further prevent damage of stock plants (King County 1993). Hoag (1992) advises taking no more than 1/3 of stems from a shrub or tree, cutting from within the crown, making clean cuts with sharp tools, and spreading the harvest throughout the stand. Re-growth of *Salix* following severe stake harvest may be reduced when it is under strong competition from *Phalaris arundinacea* (reed canarygrass) (Whitacre, pers. comm.). Salvage sites where all vegetation will soon be destroyed offer a labor-efficient/low impact alternative to cutting intact vegetation.

Harvest stands may be infected with disease or insects that compromise stake survival. The willow bore (*Cryptorhynchus lapathi*) is endemic in the Pacific Northwest and can kill individual plants (King County 1993). One local practitioner has discontinued wild harvest of *Cornus sericea* after observation that cutting led to extensive fungal infection that subsequently reduced the vigor of the stand (Whitacre, pers. comm.). Sterilization of cutting tools to prevent the transfer of wood-borne disease is common throughout the nursery trade (Harris et al. 2004).

Agricultural production of cuttings for restoration plantings has been conducted by the United States Department of Agriculture, Natural Resource Conservation Service (Crowder & Darris 1999). These installations alternately called

cutting blocks, stooling beds, or coppice plantations, have been observed to vigorously produce yearly poplar cuttings for 10-15 years following planting (Morgenson 1992). The productivity of other species under coppice is not common. Meilleur et al. (1997) experimented with severe pruning to encourage shrubland expansion in power line rights-of-way. *C. sericea* significantly increased the length of new shoots following severe cutting, and shrub height rebounded more quickly following cutting than *Salix petiolaris*. Productivity following severe pruning, or under a continuous pruning regime will likely vary between species. Faced with high demand for revegetation products some authors have begun investigating micropropagation techniques that dramatically increase the number of clones that can be produced from limited parent materials (Edson et al. 1995).

Agricultural production offers many advantages. Wood age can be controlled and harvest labor can be reduced. Marginal agricultural land can be converted to lightly managed cutting blocks that increase wetland buffer function and/or wildlife habitat while producing a resource for restoration. The growing conditions of cutting stock can be manipulated to maximize rooting potential. If the harvest cycle is reduced to every two years, seed can be produced as a secondary by product. A number of manuals are available that explore the design and management of agroforestry systems that can incorporate coppice as part of a more complex production system that maximize landscape yield by increasing yield complexity and diversity (Mollison 1988).

2.5.2 Water – Moisture Content, Soaking, and Site Hydrology

There is a clear relationship between plants adapted to life in the floodplain environment and the ability to form adventitious roots. The development of stem-borne adventitious roots in naturally occurring stands of *Alnus glutinosa*, *Salix spp*, and *Populus spp*. is stimulated by periodic soil inundation (Gill 1975; Krasny et al. 1988). The native deciduous shrubs with potential for staking are commonly adapted to life in hydric soils (Kunze 1994; Reed 2003). Salisbury and Ross (1985) suggest that accumulation of ethylene may cause adventitious root initiation, however the role of ethylene in inducing rooting is not clear (Jarvis 1986; Mudge 1988). The degree to which adventitious rooting is controlled genetically or by physiological response to the environment has not been adequately investigated. No experimental evidence indicates that cuttings from parent stock growing in hypoxic soil have increased adventitious rooting as compared to stock growing in aerobic soils - a proposition that could be easily tested.

High soil moisture appears to benefit rooting given sufficient diffusion of oxygen. Rein (1991) rooted woody cuttings of *Juniperus* (juniper), *Rhododendron* and *Ilex* (holly) in a well drained rooting medium at varied moisture levels. Survival was highest in the wettest material where water weight was at 625% of dry media weight. Cuttings were better able to uptake water through cut stems in the wettest media (Rein et al. 1991), a phenomena that has also be observed where cuttings soaking in water grew and remained turgid in the absence of roots (Stannard & Guenther 1999). After planting in dry soil, water content of small *Populus* cuttings decreased before rooting, while cuttings in moist soil retained their water content with a corresponding increase in survivorship and growth (Hansen & Phipps

1983). Low soil moisture has also negatively affected growth and survivorship of large *Salix nigra* pole cuttings (Pezeshki et al. 1998).

Where soil pores are filled with water, oxygen cannot diffuse at a rate sufficient to replace oxygen consumed by root and microbial metabolism resulting in a hypoxic (low-oxygen) environment (Mitsch & Gosselink 2000; Salisbury & Ross 1985). Given sufficient aeration, roots will develop in standing water (hence the success of hydroponic gardening). Species vary in their ability to form new roots in a hypoxic environment. Where *Hedera helix* (English Ivy) requires dissolved oxygen levels of 10 ppm, *Salix* will initiate roots at oxygen levels as low as 1 ppm, albeit at a reduced rate (Zimmerman 1930). In a potted trial, *Populus trichocarpa* was unable to extend roots into the hypoxic zone, while *Salix lucida* both extended roots into hypoxic substrates and formed adventitious roots on stems submerged in hypoxic media (Stannard & Guenther 1999). Wetland plants adapt to life in hypoxic soil conditions through a variety of mechanisms, some that are intrinsic to the metabolism of the species, and others that develop over time (Mitsch & Gosselink 2000). A species that can tolerate significant saturation in the early growing season when established may not have the physiological adaptations to initiate adventitious roots under saturated soil conditions. Even where adaptation to saturated soils exist, low redox potential may limit growth (Pezeshki et al. 1998). Hoag (2001) agrees that a frequent cause of staking failure is planting in soils that are too wet. Where soil pore space has been destroyed by compaction, common on extensively graded restoration sites, hypoxic conditions may occur with typical rainfall limiting root formation.

Water stress may cause cutting failure over summer months. Mulch reduces evaporation of soil water and increases percolation of rainfall, extending the season where soils have moisture levels hospitable to plant growth (Harris et al. 2004). The cost of soil de-compaction, amendment, and/or mulching may discourage their large-scale application, however the benefits to plant survival and growth may be substantial (Harris et al. 2004; Myers, pers. comm.). Fabric weed barriers significantly increase soil moisture, increase cutting survival, and can be installed mechanically for under \$0.40 per square foot (Hoag & Sellers 1996; Simonson & Cornforth 2001). As a secondary benefit these fabrics can effectively control *Rubus discolor* (Himalayan blackberry) within 1 to 3 years (Carrell 2004; Starflower Foundation 1997).

Soaking cuttings before planting is not uniformly practiced. Hoag states that soaking stimulates root production in *Salix* (Hoag 1994; Hoag et al. 1995), which is consistent with Carlson's investigation of *Salix cordata* (1938). When soaking is considered at all it is usually recommended, often without regard to climate, species, or the oxygen content of water. Hoag (1992; 1993; 1994) has successively recommended soaking cuttings from 1 to 14 days, a minimum of 24 hours, or 2-5 days for poles. In Idaho, Zierke (1994) soaked cuttings for 12 days, while in Western Washington, Alexander (2004) has observed soaked cuttings of willow that have begun rooting in a few days. Several Western Washington sources made no mention of soaking (Brandvold, pers. comm.; Myers Biodynamics 1993; Tuttle & Wenberg 1996; WADOE 1997; WADOT 2001).

Benefits of soaking are not always consistent. Hoag (1993) also reported no conclusive results from an experiment using pre-soaking as an experimental treatment. Where soaking has any reported effect, it appears to be beneficial, however not all cuttings benefit equally. Soaked and warmed *Populus* cuttings of diverse parentage rooted and grew more vigorously than un-soaked cuttings however this effect was most substantial when cuttings were planted in a low-moisture rooting medium. Significant improvement in *Populus* rooting was also seen by increasing soaking time from 3 days to 10 days (Hansen & Phipps 1983). Soaking until just before bark rupture (7 days) improved survival in hybrid *Populus* cuttings in field trials as well, however not all hybrids responded to the soaking treatment (Petersen & Phipps 1976). A recent study by Schaff et al (2002) measured rooting in pole cuttings (5 cm diameter, 1.5 m length) of *Salix nigra*. Ten days of soaking were required for significant effects, and only cuttings planted in dry environments consistently increased root or shoot dry weight. Strangely, in this study, short duration soaking reduced growth. Cuttings that were soaked for three days had lower measured mean growth than both cuttings that were soaked for ten days and cuttings that were un-soaked, with significant differences between 3 and 10 day treatments in several growth measures. The ability of soaked cuttings to partially overcome reduced adventitious root development dry soils was further confirmed with *Populus x euramericana* stem cuttings by Puri & Thompson (2003b) and further explains the importance of soaking to practitioners in arid climates. Interaction between soaking treatments and soil moisture regime may account for cases where soaking provides no measurable benefits. Given that most soils in the Pacific Northwest are likely to be quite moist during late winter and spring, soaking may have limited benefits. However, soaking may not just improve water status of cuttings and may increase rooting ability of genera other than *Salix* by stimulating the pre-formation of root primordia, which could improve survivorship and growth after planting.

2.5.3 Growth-Regulating Compounds

Auxins are a group of growth-regulating compounds that cause a variety of effects in plant cells. Indole-3-acetic acid (IAA) is an auxin produced endogenously in buds and leaves and is critical for root formation. Indole-B-butyric acid (IBA) and naphthalene-acetic acid (NAA) are synthetic auxins commonly used in the nursery industry. The hard-to-root species *Mussaenda erythrophylla* was tested for auxin response and observed for production of root primordia. Increased rooting in response to hormone treatment was correlated with increased development of root primordia (St. Hilaire et al. 1996). The synthetic auxins tend to be more effective at stimulating rooting than naturally occurring compounds because they are resistant to the enzyme systems that neutralize the naturally occurring IAA (Salisbury & Ross 1985). Plants may be divided into three classes based on rooting response to exogenously applied auxin: 1) Plants that root well regardless of supplemental auxin, suggesting that all factors necessary for rooting are present, 2) Plants for which rooting increases with supplemental auxins such that rooting is auxin limited, and 3) Plants for which rooting is limited by some factor other than auxins, such that exogenous application of auxins fails to stimulate rooting. Rooting co-factors (which Hartmann (1968) identifies as bud-formed phenolic compounds) must be present in addition to auxin for successful rooting. A wide variety of rooting

co-factors have been identified, and attempts to isolate specific compounds and their sources suggest that necessary co-factors for adventitious root production are species specific (Haissig 1974b). Even where application of auxin does increase rooting, Leibig's law of the minimum suggests that another factor may prevent continued improvement of rooting response once auxin is no longer deficient (Salisbury & Ross 1985). The factors limiting rooting can vary between ecotypes within a species. Puri and Swamy (1999) discovered that variation in rooting ability among cuttings from 12 provenances was amplified by the application of auxin suggesting that some variation in rooting ability among ecotypes was masked by auxin limitation. A similar pattern of increased variation under auxin treatment has been observed in comparisons of rooting ability between cuttings from young and old trees (Swamy et al. 2002).

A large number of studies have been conducted solely to identify optimal hormone application levels to maximize rooting of hardwood cuttings. Concentrations of 2000-5000 ppm of IBA or IBA/NAA solutions are typical for the propagation industry (Dirr 1998). Based on preliminary work by Darris (2002) the majority of 22 native species tested showed no significant response to treatment with several dilutions of Woods Rooting Compound (1.03% indole-B-butyric acid and 0.519% naphthaleneacetic acid) suggesting that these species are not auxin limited in their ability to form root primordia. Application of growth regulators to willow have been observed to reduce root formation or stake survival (Darris 2002a; Hoag & Short 1992), while dilute applications of growth regulators (0.3% indole-B-butyric acid) improved root formation of the harder-to-root *Salix scouleriana* more than concentrated applications (Edson et al. 1995). Hoag (1992) suggested that treatment with growth regulating compounds offered no economic advantage on large field installations of *Salix* and *Populus*.

2.5.4 Wounding Bark and Counting Nodes

All cuttings are wounded when they are excised from their parent stem. As mentioned in section 2.4.1, carbohydrates and growth regulators travel basipetally in phloem and accumulate at the basal end of a cutting. Superficial lateral cuts that sever the active transport of compounds in the phloem would likely cause accumulation of those substances associated with formation of root primordia near the wounds. Hartmann (1990) suggests that wounding bark may assist root emergence through woody sclerenchyma, increase absorption of water and externally applied rooting hormones, and stimulate production of root primordia due to the natural accumulation of hormones and carbohydrates near the wound. The effects of wounding on root production in native species have not been documented.

Nodes are a common site for the formation of root primordia particularly in young stems (Carlson 1938; Haissig 1974c; Hartmann & Loreti 1965). When propagating *Populus* hybrids, Edson (1995) was careful to insure that a node was located just above the basal cut. A node buried underground is recommended for propagation of *Sambucus racemosa* (Vanbianchi, pers. comm.). The presence of nodes in the rooting zone may be important to take advantage of pre-formed root primordia associated with nodes. However, some species including some species

of *Salix*, appear to form roots primarily from induced primordia near the cutting base (Chmelar 1974; Haissig 1974c; Hartmann & Loreti 1965) or all along the stem (Darris 2002a; Stannard & Guenther 1999) making node position less critical.

Darris (2002) characterized the general location of root initiation among Pacific Northwest native plants. All species that rooted adequately for consideration as stakes rooted from both the base and nodes, further obscuring the potential importance of nodes as rooting sites. The Washington Department of Transportation (2001) specifies that two undamaged buds should be left above ground in stake installations. Hoag (1993) warns against damaging buds during installation while encouraging removal of apical buds. However, mass-produced cuttings are typically cut without regard to node location with acceptable results (Hartmann et al. 1990; Morgenson 1992).

Careful management of node location on cuttings may ultimately be determined by project scale and labor costs. Based on the two to three foot length recommended in many cutting specifications, and the depth of recommended planting, insuring the presence of adequate nodes in ideal locations may not be necessary. More systematic species-specific investigation of the effect of cutting node location and subsequent rooting will be needed to determine the degree to which this factor affects survival and growth of cuttings.

2.5.5 Storing Cuttings

Several authors suggest that immediate planting with a minimum of storage increases success of staking (Gray & Sotir 1996; King County 2004) and some contract specification require planting within 8 hours of cutting, or moist storage for no more than two days (GCSWCD 2004). Some practitioners will frequently cut and plant on-site during maintenance (Partridge, pers. comm.), while others will keep cuttings for a week under careful storage (Alexander, pers. comm.). Other specifications make no mention of storage or time allowed between cutting and planting (WADOT 2001) or provide recommendations for long term storage that may involve storage in snow, cold water, or under refrigeration (Bentrup & Hoag 1998; Fischenich 2001; Schiechl & Stern 1996). Long-term storage can allow practitioners to harvest during shoot dormancy, but then store stakes through spring to avoid damage from flooding (Hoag & Short 1992).

Propagation manuals may recommend storage of hardwood cuttings before sticking to improve rooting (Dirr 1998; Hartmann et al. 1990). Careful studies of cuttings indicate that cuttings can absorb water through cut stems. Cuttings may continue to absorb water for at least a few days after excision, although rates of water absorption may subsequently drop (Rein et al. 1991). For cuttings where root primordia form before shoot development, soaking or storage in a moist medium allows for development of primordia while protecting cuttings from desiccation that might occur if planted in mid-winter.

Few studies have explicitly studied the effects of storage. Cram and Lindquist (1982) tested November-harvested *Populus* and *Salix* cuttings buried in sawdust outdoors compared to cuttings stored in polyethylene bags at 0, 24 and

35° Fahrenheit (-18, -4, and 2° Celsius). Fresh cuttings were harvested in early April when the heeled-in cuttings were lifted, and both the fresh and the heeled-in cuttings were stored in polyethylene bags at 24° F until all treatments were simultaneously planted in spring. Although there was some variation among species and varieties, cuttings that were heeled-in before bag storage had the highest moisture content and rooting ability, even better than cuttings harvested later in winter and stored for less time under refrigeration. Hoag (1993) found no significant difference in cutting survival between cuttings planted fresh and those stored at 28-34° F for several months. Storage of *Salix* in an icebox for 10 months produced no change in rooting ability (Chmelar 1974). Storage of November-harvested cuttings at temperatures below freezing may cause delayed shoot development compared to cuttings harvested and stored later in winter (Phipps & Netzer 1981), with some practitioners recommending two or three weeks at 41° F to provide the time required for biochemical processes to occur during dormancy prior to shoot expansion (King County 1993). Storage under moist burlap, in plastic, or wrapped in PVC film is necessary to prevent dehydration (Cram & Lindquist 1982; Schiechl & Stern 1996).

Careful storage can preserve stake moisture content for planting, extend the planting season, and improve rooting ability of some species. Experimental results suggest that careful storage of cuttings even for long periods may be possible without reduction in rooting potential.

2.5.6 Cutting Thickness, Planting Depth, and Sources of Field Mortality

Poor or unexpected soil conditions at planting frequently cause revegetation failure (Myers, pers. comm.; Vanbianchi, pers. comm.). Site assessment and preparation are topics of critical importance that will not be discussed at length. Morgan and Rickson's (1995) suggestion that 'conditions be suitable for plant growth' will require repetition and refinement for years to come.

Several sources recommend using cuttings of a certain minimum diameter (Bentrup & Hoag 1998; GCSWCD 2004; Gray & Sotir 1996; Hoag 1993; King County 2004; Myers Biodynamics 1993). *Salix* (willow) are the most commonly used taxa for field propagation (Bentrup & Hoag 1998; Gray & Sotir 1996). *Salix* cuttings as large as 8 inches in diameter will root well after planting (Hoag & Short 1992). Enormous *Salix fragilis* logs 70 cm in diameter have been stuck and rooted (Chmelar 1974), and massive *Populus trichocarpa* logs will root when partially buried in alluvial sediments (personal observation). Large cutting diameter is frequently cited as increasing planting success of *Salix* and *Populus* by increasing available energy reserves (Bentrup & Hoag 1998; Hoag 1993). Insufficient cutting diameter has been cited as a cause of failure for willow-stake plantings (Hoag 2001).

The recommendation of using thick willow cuttings may stem in part from differences in rooting ability among *Salix* species. A study of rooting in 107 species of the genus *Salix* indicated that thicker cuttings were only advantageous for the less vigorously rooting species, while aggressively rooting species grew well regardless of diameter (Chmelar 1974). Hoag, who works in arid climates, has described increased success with thicker cuttings (Hoag 2001), and uses a wider variety of willows than used in the PNW (Hoag & Sellers 1996). *Salix geyeriana* and *S.*

scouleriana are less vigorously rooting than *S. sitchensis* and *S. lucida*, which are more commonly used in the Puget Trough. Thick cuttings may be a practical way of ensuring success when planting multiple difficult-to-differentiate species with varied rooting abilities, although thick cuttings may not improve rooting of all species equally.

Other guidelines suggest using young shoots (WADOT 2000). *Populus* hybrids achieve high growth rates under agricultural field management and irrigation using 7- to 12-inch cuttings of one-year-old wood sometimes as little as 3/8 inch in diameter (Cram & Lindquist 1982; Morgenson 1992; Petersen & Phipps 1976; Phipps & Netzer 1981; Tschaplinski & Blake 1989). Long slender branches are recommended for construction of fascines of *Cornus sericea* (Gray & Sotir 1996) and *Rubus spectabilis* (Darris 2002b). Young stems are preferred for rooting of *Populus trichocarpa* and *Salix lucida* (Stannard & Guenther 1999). Many hard-to-root species will only root from first-year wood (Haissig 1974c; Hartmann et al. 1990). Among one-year-old stems, larger *Populus* cuttings showed no greater ability to root in dry medium or after pre-soaking (Hansen & Phipps 1983).

Howard and Ridout (1991b) completed a series of experiments to examine rooting ability of *Prunus insitita* cv. 'Pixy' in relation to cutting diameter, and the location of the cut on severely pruned stock plants. Within a wood age class they identified that thin shoots that often emerged from more basal locations had a higher rooting potential. Where shoots of varied diameter emerged from more distal origins, thinner shoots tended to have a higher rooting ability, however this increased rooting ability was compromised by an increase in basal rot, which had to be counteracted with better drained rooting medium (Howard & Ridout 1991a). Further experiments to partition sources of variation in rooting potential suggested that potential was independently developed in each shoot, that the actual height above the ground where the cut occurs did not affect rooting ability, and confirmed that in severely pruned parent stock, the thinner shoots appeared to have a higher rooting potential (Howard & Ridout 1994). Rooting ability within a plant appears to be controlled by a diversity of interrelated factors that are species specific.

Larger cutting diameter from older wood may increase resistance to desiccation due to a higher surface-to-volume ratio and greater bark thickness, although no studies have tested this proposition. In more temperate climates Sciechtl and Stern (1996) agree that 'more substantial' cuttings ensure successful sprouting and rooting because of their ability to resist desiccation. Approaches that rely on large-diameter cuttings are unlikely to work for species where rooting ability declines with wood age.

A significant portion of the stem of easy rooting species like *Rubus spectabilis* or *Sambucus racemosa* contains spongy pith that could make these species particularly vulnerable to desiccation (King County 2004). Capping the distal end of cuttings with latex paint or a 50% paint/50% water mixture immediately after harvest is a common practice in arid regions (Bentrup & Hoag 1998; Hoag 1993; Hoag & Short 1992). However, it is rarely recommended by practitioners in the Pacific Northwest (Alexander, pers. comm.; Buis, pers. comm.; King County 2004). Painting cutting ends or other preventative measures should not be categorically excluded where desiccation limits survival of certain species, or in specific environments. Painting cuttings may also increase visibility, identify

of correct planting orientation, and color code stakes for variable planting locations or treatments (King County 2004).

Specification of large cutting diameters may be preferred for reasons other than rooting ability. Large-diameter cuttings possess the mechanical strength to withstand deep installation, which in turn may increase resistance to disturbance, or better position new roots in moist soil strata. Deep planting has been advised by several practitioners working in arid environments. If cuttings are not planted in the capillary fringe of the low water table there may be no reliable source of water for rooting (Bentrup & Hoag 1998; Hoag 1993; Kaplan, pers. comm.). In a study by Hoag & Short (1992), short willow cuttings of narrow diameter planted along a reservoir shoreline showed excellent rooting success, but were subsequently damaged by wave action, and submerged under fluctuating water levels. Increased diameter and length were used in subsequent installations to increase survivorship in the reservoir environment. In this case deep planting stabilized cuttings exposed to wave action, and long stick lengths allowed the tops of cuttings to remain above high water.

A variety of methods have been developed for planting un-rooted cuttings. Due to the risk of splitting the stakes before reaching the desired depth, the preferred tool for hand installation is the rubber mallet or dead-blow hammer (a plastic head filled with sand). Specifications may direct installers to remove and replace split stakes (GCSWCD 2004). Some field practitioners will cut the shattered top, leaving the shortened stake in place given that it may well root and support the objective of revegetation even though not counting toward installation specifications (Humphreys, pers. comm.). Other specifications indicate that cuttings should have no more than 6 inches of exposed top with any excess to be cut (Fischenich 2001).

Specifications suggest stakes lengths of 18 to 36 inches, with burial of 2/3 to 4/5 of the stake below ground. This may require contractors to install stakes up to 28 inches below the ground surface. While some suggest “the deeper the better” (King County 2004), several practitioners question the need for deep installation in our climate (Alexander, pers. comm.; Buis, pers. comm.; Moore, pers. comm.). Where soils are moist and root development is limited by hypoxic conditions, deep planting may not increase either the length of stake in a position to form roots, or the rate of water absorption by un-rooted tissues. No published study has documented the role of installation depth among species, which is exceptional considering the cost of labor and materials required to install stakes 2 foot depth in our stony and frequently compacted soils.

Deeply planting stakes may be unnecessary on many sites in the PNW, and Hoag (1993) identifies that correct planting depth depends on water table depth and the expected level of erosive disturbance. Planting stakes deeply into anoxic soils where rooting is suppressed is unlikely to improve survivorship, and may consequently waste materials and labor. Moore (2004) does suggest that deep planting can reduce a cuttings susceptibility to disturbance, and that reducing height above the ground surface may reduce risk of flood damage on flooded sites where a taller stake may collect flotsam.

Deep planting is universally practiced in arid climates where the summer capillary fringe may be 2 to 6 feet below the ground surface even on a river bank (Bentrup & Hoag 1998; Hoag 1993; Kaplan, pers. comm.). On some sites ground-water monitoring and observation of soil texture are used to determine the appropriate depth for planting (Kaplan, pers. comm.). *Salix* and *Populus*, which retain their rooting ability in older wood (Carlson 1950; Chmelar 1974) such that large poles can be successfully rooted in the field (Hoag 1993; Kaplan, pers. comm.; Moore, pers. comm.; Schaff et al. 2002), are commonly used in these environments. Hoag (1999) has successfully bundled cuttings for deep installation where large diameter poles were not available, and found that bundles are less likely to die from beaver damage.

A variety of tools have been developed to facilitate deep stake planting including custom built back-hoe attachments for creating pilot holes and driving live poles (Hoag 2001), hand-held pressurized water drills to create deep pilot holes (Hoag et al. 2001), and roto-hammers, commonly used for drilling concrete (Hennelly 1994; Stamphli 1992). Hand held jack hammers with the chisel replaced with a welded pipe section have been used to insert substantial cuttings in Europe (Schiechl & Stern 1996). Good contact between cuttings and soil was commonly cited as critical for success (Hoag 2001; Myers Biodynamics 1993). Stamphli (1991) commented on the difficulty of backfilling drill holes around stakes and found free-flowing dry sand useful for the task. Deep planting also has had value where there was a risk of physical disturbance, either due to alluvial flooding, wave action, or human and animal traffic (Hoag 1993; Hoag & Short 1992; Moore, pers. comm.). Even with shallow planting, creating a pilot hole with a iron bar may be necessary (Brandvold, pers. comm.; Humphreys, pers. comm.; Myers Biodynamics 1993), although Myers (pers. comm.) suggested that pilot holes increase the possibility of poor soil-stake contact.

Recommended stake planting density and pattern on revegetation sites varies and is largely based on the performance of *Salix*. Schiechl & Stern (1996) recommend 2-5 cuttings per square meter and King County (1993) suggest a similar range of 2-4 cuttings per square yard. Extremely dense plantings (1 foot centers) have been installed but may interfere with weed control activity if stake growth is not adequate (Brandvold, pers. comm.). A specification provided by Myers (pers. comm.) suggests planting clusters of 5 cuttings at 12 foot spacing to insure even cover.

Although some sources prescribe planting at right angles to the soils surface (King County 1993) there is no consistent rationale for why cuttings must be planted vertically. Planting poles at an angle can increase immediate shading of stream channels (Hoag 1998) and may increase the length of the stake located in soil conditions conducive to rooting (Myers Biodynamics 1993). Horizontal or near horizontal burial is practiced using fascines (Bentrup & Hoag 1998; Gray & Sotir 1996; Tuttle & Wenberg 1996) and horizontal burial is more common than vertical burial in alluvial environments (pers. obs.). Vanbianchi (pers. comm.) reported rooting native cuttings in moist sawdust, suggesting that encasement in mineral soil is not necessary for root formation. Unrooted cuttings could be scattered and buried under mulch.

Several studies have explored the rooting ability of rhizome fragments and found that *Viburnum edule*, *Corylus cornuta*, and *Rosa* all rooted effectively from even 10-cm fragments (Landhausser & Lieffers 1997). *Rubus spectabilis* also roots readily from rhizomes (Zasada et al. 1994). A large number of Pacific Northwest native shrubs are adapted to vegetative self-propagation using rhizomes, and could be considered for vegetative propagation, particularly where rhizome harvest from a salvage site is feasible.

After 12 interviews and numerous informal conversations opinions outnumbered and frequently contradicted empirical observation suggesting that the potential of vegetative propagation has been poorly explored and is primarily limited by the assumptions of practitioners, lack of familiarity with the natural capacities of the species planted, and a shortage of resources that can be allocated to experimental work.

Even where stakes survive, herbivory may limit the success of stake plantings on some sites. For example, deer or rodent browse has limited growth of stakes in Oregon (Flessner 1997). Spring shoots of *Rubus spectabilis* composes 25% of the diet of Columbian black-tailed in Olympic National Park (Leslie et al. 1984), and may completely alter riparian vegetation composition and structure across whole watersheds (Schreiner et al. 1996; Woodward et al. 1994). *Cornus sericea* is heavily and selectively browsed in some locations (pers. obs.). On the Oregon coast, *Castor canadensis* (beaver) depend heavily on *Salix* as a food source (Maser et al. 1981). Selective browsing of new growth on woody deciduous shrubs may substantially impact a stake planting during and after establishment. Hoag (1993) suggests that *Spirea douglasii* and *Symphoricarpos albus* may be less palatable to livestock. Diversifying staked species may increase success under browsing pressure.

3. METHODS

3.1 Experimental Design

This experiment aimed to identify factors controlling survival and short-term growth in three indigenous Puget Sound shrubs with potential for live-staking. *Symphoricarpos albus* and *Rubus spectabilis* show the ability to rapidly form adventitious roots (Darris 2002a, 2002b), are frequently found in wetland and riparian systems, and are commonly included in revegetation designs. They are currently planted as bare root seedlings or potted stock. Their performance as live-stakes has not been tested under diverse field conditions. *Cornus sericea* is frequently staked and so could serve as a benchmark for evaluating the relative performance of *Symphoricarpos* and *Rubus*. *Cornus* is considered less reliable than *Salix*, and so there is some interest in determining what factors control its success.

Soil moisture and reduction-oxidation potential strongly influence adventitious root formation (Hansen & Phipps 1983; Hoag 2001; Pezeshki et al. 1998; Rein 1991; Stannard & Guenther 1999; Zimmerman 1930). Although previous studies have compared adventitious root development among a few static moisture regimes (Darris 2002a; Pezeshki et al 1998; Schaff et al. 2002), moisture variation in the field exists as a continuous gradient. Restoration practitioners need to make decisions about where along this gradient to plant. In this experiment, stakes were planted across a wetland edge to describe survival and growth response under a range of field conditions. Elevation relative to water level was used as a surrogate for soil moisture regime as it is easily measured in the field and thus is available to restoration practitioners. An arbitrary elevation datum was established on a staff gauge in the pond below the elevation of the planting. All elevation data are measured in centimeters above this arbitrary datum.

Planting in late fall is typical for lowland Puget Sound. It is hypothesized that plants grow roots during winter shoot dormancy that increase survival over the dry season. Planting time has not been explicitly tested in the field as a predictor of first-year survival and growth. A December planting and a February planting were interspersed across the elevation gradient to test for increased survival from early planting.

Numerous biotechnical erosion control manuals instruct that thick cuttings be deeply inserted into the soil (Bentrup & Hoag 1998; GCSWCD 2004; King County 2004; Myers Biodynamic 1993). However, agricultural production of *Populus* is successful with thin, short cuttings (Morgenson 1992). Diameter has been cited as important for stake survival with *Salix* and *Populus* (Bentrup & Hoag 1998; Hoag 2001; Schiechl & Stern 1996). Diameter is correlated with stem age, which can independently affect adventitious rooting ability (Darris 2002a; Hartmann et al. 1990). For this study, stem age and stake length were kept constant while diameter, weight, side branch cuts, and number of nodes were measured and entered as factors into regression analyses. Stakes were kept short (40 cm) as short stakes save materials and labor, while the advantages of long stakes for revegetation are untested.

To minimize variation between experimental units, stems were harvested randomly from stands growing under reasonably homogeneous light, soil, and moisture conditions. All stakes within a planting were harvested over a 1-3

hour period, and processed, stored, measured, and installed in as short a period as possible. No soaking, bark laceration, or hormone treatment was used, although systematic experimentation with these treatments is warranted.

An outdoor site with smooth topography, homogeneous soils, and stable hydrology was selected to make elevation a meaningful surrogate for moisture regime. To facilitate installation and maintenance, the planting site was divided into nine blocks. Each block contained 60 planting locations at 20 cm spacing in a 5 x 12 grid (100 x 240 cm). Each block contained 10 stakes of each species and planting time (3 species x 2 planting times). At each of the two planting times, individual stakes were randomly assigned to blocks, and planting locations within each block. Thus, planting times and species were evenly distributed throughout the planting belt, while variations in stake characteristics were randomly distributed throughout the planting.

The nine planting blocks were oriented perpendicular to the slope and distributed from 47 cm below high water to 36 cm above high water. To more evenly distribute cuttings across elevation, the distance between blocks was increased at higher elevations to compensate for reduced slope (Figure 3.1).

Two measurements of growth were selected to quantify the competitive ability of survived stakes. Total shoot length was served as an easy-to-measure surrogate for biomass. Vertical height was measured as an indicator of the stakes ability to successfully compete for light. Spring bud flush can be delayed by soil moisture conditions (pers. obs.), so the rate of spring shoot emergence was monitored to determine if delayed development resulted in reduced growth and competitive ability.

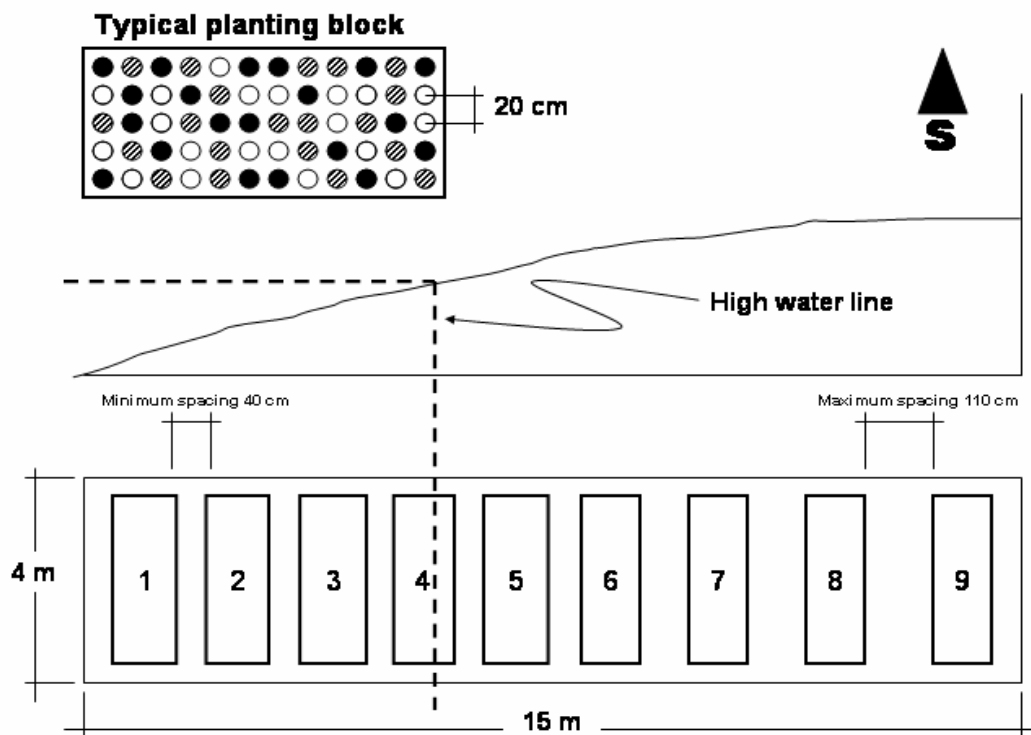


Figure 3.1 – Planting schematic

Dimensions are not to scale. Belt transect of planting blocks is arranged perpendicular to the contour. Blocks east of block 4 were inundated during the early growing season. Typical planting block represents the random distribution of 3 species and two planting times throughout each block. Spacing between each cutting is 20 cm. Planting blocks are oriented North to South. Spacing between blocks was increased at higher elevations to distribute stakes evenly across elevation, despite decreasing slope.

3.1 Cutting Harvest and Processing

Hardwood stem cuttings of *Rubus spectabilis*, *Symphoricarpos albus*, and *Cornus sericea* were harvested, measured, and planted at two times in the winter of 2002-2003 (Table 3.1). At each planting time all three species were harvested within a three-day period and planted within a week of harvest. At both harvest times stems were bare of leaves. All procedures were designed to mimic typical restoration field practices and were kept consistent between planting times.

Table 3.1 - Project timeline

Date	Event
5-7 December 2002	First stake harvest
12 December 2002	First planting
14-16 February 2003	Second stake harvest
20 February 2003	Second planting
29 March 2003	First shoot observation
20 April 2003	Second shoot observation
6 May 2003	Third shoot observation
28 May 2003	Fourth shoot observation
November 2003	Growth data collection

Even-aged stems were selectively cut from harvest sites at low elevations in King County, Washington (Table 3.2). A standard wood age was chosen based on work by Darris (2002b) and wood age was estimated by counting apical bud scars backwards from distal buds as corroborated by observation of shoot bifurcation. Excised stems were cut into 40 cm segments in the field regardless of node location. Thin distal stem segments with numerous side branches were discarded. Cuttings were bundled and buried in moist sawdust. Time between harvest and storage was between two and six hours for each species and planting time.

Table 3.2 - Cutting harvest locations.

Stem age was estimated by observation of yearly growth increments and side branch bifurcation. Coordinates are based on North American Datum 1983. Elevation is approximate and based on USGS quarter quad interpolation. Nomenclature follows Hitchcock and Cronquist (1973)

Species	Stem Age	Location	Latitude/Longitude	Elevation
<i>Cornus sericea</i>	2-3 years	University of Washington	47.6579°N, 122.2914°W	~ 10 m
<i>Rubus spectabilis</i>	2 years	Sammamish Plateau	47.6798°N, 122.0274°W	~ 170 m
<i>Symphoricarpos albus</i>	3 years	Snoqualmie Floodplain	47.6875°N, 121.9475°W	~ 40 m

Thin cuttings are more likely to break during installation so the cutting harvest was 'high-graded' by discarding 10-20% of the smallest diameter cuttings. Both two- and three-year-old wood from *Cornus sericea* was used and was distributed evenly among planting blocks and planting times. This variation in wood age was recorded as an indicator variable for later analysis. Cuttings were randomly assigned to treatment groups, individually labeled, and measured for length, midpoint diameter, and damp weight. The total number of buds and any cut lateral branches were counted. The occasional presence of a black fungal stain on some *Rubus spectabilis* cuttings was also noted. Except during initial sorting and briefly during measurement, cuttings were kept moist and covered until planting. Based on Daris (2002a) no auxin treatment was used. Stakes were not painted, soaked or given any other pretreatment.

3.2 Experimental Site Preparation, Planting, and Maintenance

The planting site was located on the shore of Central Pond in the Union Bay Natural Area, Seattle, Washington (Latitude 47.6552°N, Longitude -122.2937°W, ~ 10 m elevation) - the site of the City of Seattle's Montlake Landfill decommissioned between 1966 and 1971. After closure and capping, landfill materials and the underlying peat subsided, creating an undulating landscape cradling several ponds and vernal pools. High water elevation is stable and limited by overflow to Lake Washington, and is maintained throughout the early growing season. Soils are very compact and poorly developed. Dunn (1966) describes landfill reclamation procedures. The site is currently managed for wildlife habitat (UBPC, 1995). The poor soil conditions and exposure to wind and sun are typical of urban or sub-urban restoration sites degraded by historic development. Field observations of soils were made using Washington State wetland determination protocols (WADOE, 1994) and USDA-NRCS definitions (Schoeneberger et al. 2002).

The planting location had even topography and no observable source of concentrated upland runoff. Soil texture was uniform except for a shallow 1-2 cm layer of fine sediment deposited below the high water line. A layer of hypoxic soils was identified by reduced chroma, the presence of iron concentrations and depletions (mottling), and reduced rooting density of the dominant upland grass, *Agrostis stolonifera* (Vepraskas, 1999). The depth to hypoxic soils increased evenly with distance from the water line and elevation.



Figure 3.2 – Planting site before installation

Photo taken during low water in fall 2002. The top of the dark vegetation band indicates winter/spring high water elevation. Planting was distributed from the bottom of existing vegetation to above the high water line.

Above-ground vegetation was cut and removed, *Rubus discolor* (Himalayan blackberry) root crowns were grubbed, and the site was tilled by three passes of a 15-HP rear-tine roto-tiller. The site was raked smooth and mulched with

4 inches of un-composted arborist chips. Site preparation was consistent with intensive regional revegetation practices in the absence of herbicide.

Well-defined bands of existing vegetation indicate the elevation of high water in Central Pond (Figure 3.1). The lowest band is inundated for most of spring and is dominated by *Epilobium spp.* with unvegetated mud providing >25% cover. A middle band is only inundated for the beginning of the growing season, and is dominated by *Solanum dulcamara*, *Epilobium spp.* and *Lythrum salicaria*, with *Lycopus uniflorus* becoming dominant at higher elevations. The highest band is rarely inundated and dominated by *Rubus discolor* and *Agrostis stolonifera*, with *Cirsium vulgare* common. Upland vegetation was mowed several times a year to control *Rubus discolor*. The transition between the middle and highest band is located at approximately 70 cm elevation.

Out-plantings of cuttings occurred over single afternoons within one week of harvest dates. Cuttings were driven 30 cm into the substrate using a rubber mallet. Approximately 10 cm of the distal end was left above the mulch level. Cuttings that broke during installation were removed and replaced.

Branta Canadensis (Canadian geese) were observed on the site, particularly near the water line. Guano was removed, and no new deposits were discovered after installation of a three-foot fence for goose exclusion. By the time of the second planting, rising water levels inundated the lowest three planting blocks. Due to the impracticality of planting cuttings underwater the second planting was not installed in blocks 1 and 2. *Rubus discolor* regrowth was not evenly distributed across the planting belt and was concentrated in blocks 6, 7 and 8. Because of the fear that competing vegetation would increase transpiration, reduce soil moisture, and thereby differentially affect stake growth in the immediate vicinity, *R. discolor* shoots growing within the planting were cut at mulch level throughout the growing season to reduce their influence on soil moisture. Further removal of *R. discolor* was not attempted due to the risks of physically injuring adjacent cutting roots. Access to the planting area was limited to pathways between planting blocks for weeding and data collection.

3.3 Elevation, Survival and Growth Data Collection

The corner elevations of each planting block were surveyed with a theodolite, using a staff gauge in the pond as an arbitrary datum. The elevation of each planting location was interpolated based on the surveyed corner points, assuming straight-line slopes between measured points. All elevations measurements were recorded and analyzed relative to a arbitrary zero point located 23cm below the lowest planting.

Shoot development was characterized at four observation times during the early growing season to measure bud flush (Table 3.1). Four shoot development classes were designed to capture the range of variation observed among the cuttings (Table 3.3).

Table 3.3 - Four level phenologic scale

Observed phenologic level over four observations was summed to construct a flushing index to describe the rate of spring shoot growth.

Bud Flushing Level	Criteria
0	No evidence of shoot emergence
1	Shoot emerged, less than 3 cm length, no leaves formed
2	Leaves formed, shoot less than 3 cm length
3	Shoot greater than 3 cm length

At the end of the growing season height, number of shoots, and individual shoot lengths were measured for each cutting. Shoot length was measured as the maximum distance along the stem from shoot origin to the most distant apical bud of either the main shoot, or a side branch if the side branch was longer than the main shoot. Height was measured from the mulch surface to the highest shoot perpendicular to the ground surface. The presence of shoots emerging from below the mulch surface, and a three-level variable describing the degree of apical control among all shoots was also recorded. Survivorship after one growing season was measured as the presence of live buds above ground after leaf senescence.

Weight data from ten *Symphoricarpos albus* stakes were accidentally not collected prior to planting, and so these ten individuals were omitted from analyses involving weight or density, accounting for the discrepancy in degrees of freedom between table 4.7 and table 4.8.

4. RESULTS

4.1 Data Manipulation

The shoot development score of individual cuttings across four observation times were summed to create a ‘flushing index’ that describes the earliness and rate of shoot emergence. A score of 0 indicated no evidence of growth at each of the four observations. A score of 12 indicated shoots in excess of 3 cm were at each observation. Total shoot length and average shoot length were calculated. All measurements were taken during dormancy, one growing season after planting.

For each species, separate regression analyses were used to evaluate survivorship, flushing index, and total shoot length in relation to elevation, planting time, and the physical characteristics of the cuttings (Table 4.1). Binomial logistic regression was used to evaluate factors potentially affecting stake survival, while linear multiple regression was used to evaluate factors potentially affecting total shoot length and the flushing index score. Errors in data collection resulted in 11 *Symphoricarpos albus* stakes being excluded from regression analyses. For all analyses the null hypothesis was rejected where statistical tests indicated that the probability of the data resulting from a random distribution were less than 0.05. Data were organized using Microsoft Excel™ 2002, and exported to SPSS™ 11.5 for statistical analyses.

Table 4.1 - Variables used in regression analysis

Units of measure and notes on variable characteristics follow the variable’s name.

Type	Cutting characteristics
Indicator	Diameter (mm, continuous)
Indicator	Weight (g, continuous)
Indicator	Number of buds (count)
Indicator	Number of side branch cuts (count)
Indicator	Density (g cm ⁻³ , continuous, derived estimate)
Indicator	Wood age (binomial: 2-year/3-year, <i>Cornus sericea</i> only)
Indicator	Fungal stain (binomial: presence/absence, <i>Rubus spectabilis</i> only)
Type	Experimental Treatments
Indicator	Elevation (cm, continuous)
Indicator	Planting time (binomial: early planting as the reference state)
Type	Growth Response
Response	Number of shoots (count)
Response	Total shoot length (cm; continuous sum of individual shoot length)
Response	Flushing Index (ordinal; 12 point index of 4 observations)
Response	Survival (binomial; live/dead)

4.2 Stake Characteristics

Descriptive statistics of cutting parameters are listed by species in table 4.2. *Cornus sericea* cuttings were greatest in mean weight and diameter, followed by *Rubus spectabilis* and *Symphoricarpos albus*. Despite small physical dimensions *Symphoricarpos albus* cuttings had the highest density, followed by *Cornus sericea* and *Rubus spectabilis*. No *Symphoricarpos albus* cuttings broke during installation as compared to six *Rubus spectabilis* cuttings and two *Cornus sericea* cuttings. *Cornus sericea* cuttings had the greatest range in diameter, weight, density, number of buds, and number of cuts.

Table 4.2 - Descriptive statistics of cutting parameters

'Diameter' is maximum median diameter. 'Weight' is based on damp weight. 'Number of buds' indicates the number of locations where buds would typically form for the species. *Cornus sericea* and *Symphoricarpos albus* have two buds per node. Number of cuts indicates the number of side-branch stubs left exposed after removal of side branches during processing.

***Cornus sericea* (Red-twig dogwood)**

	Min	Max	Mean	SD
Diameter (mm)	8.0	24.9	13.53	3.43
Weight (g)	17.7	175.7	53.85	27.92
Density (g cm ⁻¹)	0.3	1.2	0.91	0.10
Number of Buds	2	12	5.88	2.11
Number of Cuts	0	11	1.70	2.48

***Rubus spectabilis* (Salmonberry)**

	Min	Max	Mean	SD
Diameter (mm)	6.3	15.8	10.26	1.86
Weight (g)	11.4	74.3	28.68	12.09
Density (g cm ⁻¹)	0.6	1.2	0.83	0.10
Number of Buds	3	8	4.73	0.98
Number of Cuts	0	3	0.16	0.46

***Symphoricarpos albus* (Snowberry)**

	Min	Max	Mean	SD
Diameter (mm)	5.7	13.2	8.31	1.48
Weight (g)	9.2	53.6	22.47	8.58
Density (g cm ⁻¹)	0.8	1.2	1.04	0.06
Number of Buds	4	12	7.37	2.40
Number of Cuts	0	2	0.25	0.57

No systematic relationship was detected between planting elevation and either stake diameter, weight, or number of buds, indicating effective randomization of cutting distribution. There was a significant but very weak negative relationship between diameter and number of buds for all species ($r^2 < 0.05$). There was also a very weak relationship between diameter and density that was not consistent among species. *Cornus sericea* density decreased with diameter ($p = 0.003$, $r^2 = 0.056$), while *Rubus spectabilis* density increased with diameter ($p < .001$, $r^2 = 0.11$). No relationship between diameter and density was observed in *Symphoricarpos albus*. There was a predictably strong correlation between diameter and weight, however neither of these two parameters were retained as significant in final regression models. Only weight and diameter displayed substantial collinearity based on observation of variance inflation factor (VIF) statistics.

Unexpectedly there was no systematic relationship between the number of buds and the number of shoots for any species. Neither consideration of planting time nor elevation improved prediction of the number of shoots (results not shown). *Symphoricarpos albus* was extremely prolific in producing adventitious shoots resulting in both the highest number of shoots observed (7 shoots) and the highest mean number of shoots ($3.38 \text{ shoots} \pm 1.58 \text{ SD}$, among plants growing above high water). As many as six shoots originated from a single node with two buds.

4.3 Factors Affecting Survival

For each species a binary logistic regression model was fitted to predict likelihood of survival using all indicator variables (Table 4.1). An interaction factor was added combining planting time and elevation. For each species, a likelihood-ratio test of model fit indicated that estimated coefficients of indicator variables significantly improved model fit ($p < 0.001$). Due to the risk of type II error in using the Wald statistic (Menard, 2002), the significance of individual parameters was evaluated using a likelihood-ratio test, comparing nested models using the chi-squared difference between the full model and a model with the tested parameter removed. A final reduced model was fitted using only those parameters determined to be significant by this method. The difference in model deviance ($\Delta - 2LL$) between the full model and the reduced model was also tested for significance, with lack of significance confirming that the removed variables did not contribute significantly to model fit. Parameter coefficients presented in Table 4.3 are taken from the final reduced models. The Nagelkerke's R-Square statistic was used in conjunction with the overall percent of correctly predicted observations (% Correct) from a cross-tabulated classification table, to evaluate the effectiveness of the model in explaining variation in the data (Table 4.3). Survival models only explained a modest amount of variation in the data for *Cornus sericea* ($r^2 = 0.42$) and *Rubus spectabilis* ($r^2 = 0.41$). *Symphoricarpos albus*, which had 100% survival above the high water line, very low survival below the high water line, and was only influenced by elevation had a predictably powerful survival model ($r^2 = 0.88$) (Table 4.3).

For all species addition of elevation caused the greatest improvement in model fit with survivorship increasing with elevation ($p < 0.001$). *Symphoricarpos albus* demonstrated the most sudden response in the logit function to change in elevation ($b_1 = 0.260$) and only elevation significantly improved prediction of survival. Planting time and number of buds also significantly improved model fit for *Cornus sericea*. Late planting time and number of buds correlated with an increased probability of survival ($p = 0.05$; $p = 0.005$). In addition to the ubiquitous influence of elevation, model prediction of *Rubus spectabilis* survival was improved by inclusion of an interaction between elevation and planting time ($p = 0.037$). Twelve of 160 *Rubus spectabilis* stakes survived, ten of these were from the late planting, and eight of those 10 were in blocks 8 and 9 at high elevations, suggesting that high elevation and late planting in conjunction may be required for survival. Unexpectedly, the presence of a seemingly superficial fungal stain on *Rubus spectabilis* cuttings significantly improved survivorship ($p = 0.005$).

Table 4.3 - Regression of survival for each species

A binary logistic regression model was fitted to data for each species using least likelihood. Column two indicates the significance of the change in model log-likelihood between the full model containing all parameters as compared to the reduced model with nonsignificant parameters removed, and justifies the simplification of the model. Nagelkerke's R-square is a measure of model effectiveness in explaining data variability, standardized to a scale from 0 to 1. The significance of individual parameters in the reduced model is described using a chi-square nested model test of likelihood-ratio.

Species	$\Delta -2LL$ p-value	R-square	% Correct	Parameters	Estimate	$\Delta -2LL$	p-value
<i>C. sericea</i>	0.40	0.417	73.3%	elevation	0.055	-35.717	< 0.001
				early planting	-1.066	-7.801	0.005
				number of buds	0.256	-7.745	0.005
<i>R. spectabilis</i>	0.14	0.414	94.6 %	elevation	0.082	-17.090	< 0.001
				fungal stain	-1.932	-7.719	0.005
				elevation*planting	-0.017	-4.360	0.037
<i>S. albus</i>	0.99	0.881	96.2 %	elevation	0.260	-143.436	< 0.001

Although the models identified elevation as a significant factor influencing survival, examination of mean survival suggests that position in relation to the high water line may be the critical factor. Mean survivorship was very different between cuttings planted above the high water compared to cuttings planted below high water (Table 4.4). When a logistic regression model is fitted using only those *Cornus sericea* cuttings planted above the high water mark, elevation becomes insignificant in predicting survivorship ($p = 0.97$), although a high number of buds and low density were weakly correlated with higher survival of cuttings (Table 4.5).

Table 4.4 - Summary of mean response for cuttings in inundated and drained locations

Mean survivorship, flushing index, and total shoot length is compared between cuttings planted above and below the high water mark (70 cm). Mean flushing index, mean total shoot length, and mean number of shoots were calculated only from the measurement of survived cuttings (n).

Species	Moisture	Mean statistic (Standard Deviation)					
	Level	n	Survivorship	n	Flushing Index	Shoot Length (cm)	# of Shoots
<i>C. sericea</i>	above HW	106	68%	72	6.0 (± 2.5)	106.8 (± 53.0)	2.07 (± 1.0)
	below HW	55	13%	7	3.9 (± 2.5)	63.1 (± 25.2)	2.43 (± 1.0)
<i>R. spectabilis</i>	above HW	101	12%	12	5.6 (± 3.3)	13.0 (± 10.0)	1.58 (± 0.7)
	below HW	57	0%	0	0	0	0
<i>S. albus</i>	above HW	103	100%	103	8.0 (± 2.3)	144.0 (± 74.1)	3.38 (± 1.6)
	below HW	55	27%	15	4.20 (± 3.2)	46.33 (± 47.5)	3.20 (± 1.7)

Table 4.5 – Logistic regression of survival for *Cornus sericea* planted above high water.

Binary logistic regression using least likelihood was used to fit all parameters. Column two indicates the significance of the change in model log-likelihood between the full model containing all parameters as compared to the reduced model with insignificant parameters removed. Nagelkerke's R-square is a measure of model effectiveness standardized to a scale from 0 to 1. The significance of individual parameters in the reduced model is described using a chi-square nested model likelihood-ratio test.

Species	$\Delta -2LL$	R-square	% Correct	Parameters	coefficient (b ₁)	$\Delta -2LL$	p-value
<i>C. sericea</i>	p = 0.44	0.14	69.8%	number of buds	0.395	-11.11	0.001
				density	-5.578	-4.60	0.032

Table 4.6 - Multiple linear regression of *Cornus sericea*

ANOVA tables for multiple linear regression models fitted to predict flushing rate and total shoot length using all measured parameters as predictor variables. Residuals followed a linear distribution with a normally distributed variance. Individual parameters determined as significant by t-test are listed below the appropriate model.

Flushing Rate ($r^2 = 0.24$)

Source	df	Mean Square	F	Sig.
Regression	8	15.089	2.813	0.009
Residual	70	5.364		
Total	78			

Parameter	estimate	t	sig
Elevation	0.056	2.793	0.007

Total Shoot Length (not significant)

Source	df	Mean Square	F	Sig.
Regression	8	4,131.457	1.584	0.145
Residual	70	2,608.431		
Total	78			

elevation and growth may be present.

Symphoricarpos albus showed little clear growth response to measured variables (Table 4.7). No parameter was significantly correlated to total shoot length. Only twelve *Rubus spectabilis* cuttings survived, and the fitted regression model did not show any significant ability to predict either flushing index or total shoot length although greater growth appeared to occur on cuttings at higher elevations (results not shown).

Simple linear regression models were used to determine whether flushing index was a good predictor of total shoot length for each species. For *Cornus sericea* and *Symphoricarpos albus* there was a significant

4.4 Factors Affecting Flushing Rate and Growth

For each species, multiple linear regression models fitted with all independent variables were used to evaluate the factors potentially affecting flushing index or total shoot length among cuttings surviving the growing season. Few models showed any significant predictive value, although there was a weak relationship between independent variables and flush rate of *Cornus sericea* (Table 4.6; $r^2 = 0.24$, $p = 0.009$). Flushing rate correlated with increase in elevation such that for every 20 cm increase in elevation the flushing index increased by an average of one point (Table 4.6; $p = 0.007$). No relationship between any independent variable and total shoot length was observed, however examination of residual plots of *Cornus sericea* suggested that a nonlinear relationship between

Table 4.7 - Multiple linear regression of *Symphoricarpos albus*

Models were fitted using OLS methods to predict either flushing rate or total shoot length using all measured parameters as predictor variables. Models were simplified using a backwards elimination procedure (Zar 1999). Individual parameter coefficients were tested using a *t* statistic ($H_0: b_i = 0$). Parameters whose coefficients are significantly different from zero with $\alpha = 0.05$ were retained in the final model. Residuals followed a linear distribution with a normally distributed variance.

Flushing Rate ($r^2 = 0.06$)

Source	df	Mean Square	F	Sig.
Regression	7	35.199	6.181	< 0.001
Residual	100	5.695		
Total	107			

Parameter	estimate	t	sig
Early Planting	-1.346	-2.510	0.014
Elevation	0.092	5.547	0.000

Total Shoot Length (not significant)

Source	df	Mean Square	F	Sig.
Regression	5	12,939.907	2.171	0.063
Residual	102	5,961.509		
Total	107			

Table 4.8 - Prediction of total shoot length using flushing index

Simple linear regression models were fitted using OLS methods to predict either flushing rate or total shoot length using all measured parameters as predictor variables. Model significance was tested using an F statistic ($H_0: b_i = 0$). Residuals followed a linear distribution with a normally distributed variance. *Rubus spectabilis* was not significant and is not shown.

***Cornus sericea* ($r^2 = 0.06$)**

Source	df	Mean Square	F	Sig.
Regression	1	13,701.748	5.224	0.025
Residual	77	2,622.598		
Total	78			

***Symphoricarpos albus* ($r^2 = 0.15$)**

Source	df	Mean Square	F	Sig.
Regression	1	113,846.073	21.902	< 0.001
Residual	116	5,198.025		
Total	117			

relationship, however the ability of these models to account for variation in total shoot length was extremely poor ($r^2 = 0.06$; 0.15; Table 4.8), suggesting that initial rate of shoot development was a poor predictor of productivity over the growing season in these plantings.

For *Cornus sericea* and *Symphoricarpos albus*, comparison of mean total shoot length between planting blocks suggested that although there was no linear influence of elevation on growth, there appeared to be a substantial decline in growth at the highest elevations with a peak in blocks five and six (Table 4.9). Examination of standardized residual plots revealed systematic variation that suggests that a non-linear model may be more appropriate for these data. Quadratic regression models were fitted including a squared elevation parameter in addition to elevation as a predictor of *Cornus sericea* and *Symphoricarpos albus* total shoot length. Where the linear model failed the quadratic model showed a weak but significant relationship between the elevation parameter and total shoot length. Similar analyses were conducted for height and average shoot length and the addition of a quadratic component significantly improved prediction of mean height and mean average shoot length and reduced unexplained variability in the data (Figure 4.3; Tables 4.10, 4.11 and 4.12). However, this pattern of declining growth at high elevations was not strong for *Symphoricarpos albus*. A strong threshold in growth is evident between stakes planted above and below high water (Table 4.10), and neither quadratic nor linear models detect a relationship between elevation and growth when stakes planted above high water are evaluated in isolation.

Table 4.9 –Survivorship and mean total shoot length by planting block

Percent survived is in bold. Mean total shoot length and standard deviation is measured in centimeters and only considers survived cuttings. Elevation for each block is described as centimeters above or below the winter-early spring high water level (located 61cm above the summer low water level)

Species	PLANTING BLOCK (with elevation range)								
	1 -47cm to -42 cm	2 -37 cm to -27 cm	3 -21 cm to -12 cm	4 -7 cm to 1 cm	5 +4 cm to +9cm	6 +10 cm to +17 cm	7 +18 cm to +22 cm	8 +25 cm to +32 cm	9 +33 cm to +37 cm
<i>C. sericea</i>	0%	0%	10%	45%	67%	75%	60%	60%	75%
	0	0	63.5 ± 10.6	82.0 ± 34.7	138.1 ± 57.8	123.73 ± 50.9	103.5 ± 55.9	107.1 ± 48.4	63.3 ± 28.5
<i>R. spectabilis</i>	0%	0%	0%	0%	10%	0%	0%	20%	32%
	0	0	0	0	10.0 ± 8.5	0	0	19.0 ± 13.0	10.0 ± 7.7
<i>S. albus</i>	0%	0%	15%	85%	100%	100%	100%	100%	100%
	0	0	60.7 ± 20.6	58.2 ± 58.7	168.2 ± 86.7	152.3 ± 84.6	126.3 ± 49.1	158.5 ± 80.7	127.2 ± 59.0

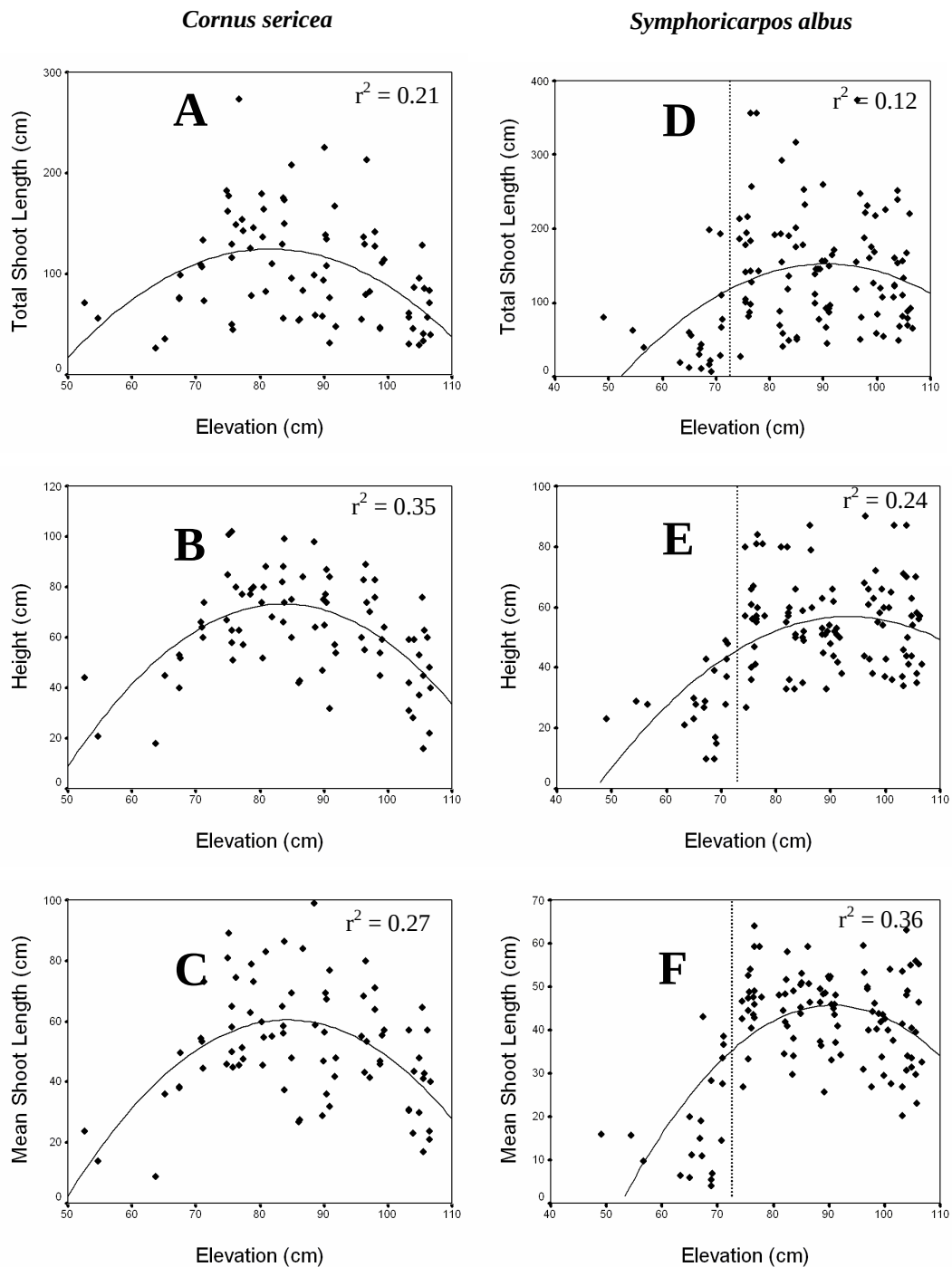


Figure 4.1 – Non-linear growth response of *C. sericea* and *S. albus* across elevation.

Ordinary least squares method was used to fit a quadratic model of elevation as a predictor of growth. There was a significant relationship between elevation and all three growth measures that weakly explained observed variation in growth (Tables 4.11 and 4.12). When compared to plots of *Cornus sericea*, *Symphoricarpos albus* appears to be more of a threshold response with a distinct increase in growth performance above ~73 cm elevation as indicated by the dashed line in figures D-F. The decline in growth at higher elevation is more pronounced with *Cornus sericea*.

Table 4.11 - Non-linear growth response to elevation of *Symphoricarpos albus*

Quadratic linear regression models were fitted with elevation and elevation² as predictors of total shoot length, height, and mean shoot length. Both parameters were found to be significantly different than zero for all growth measures.

Total Shoot Length ($r^2 = 0.119$)				
Source	df	Mean Square	F	Sig.
Regression	2	42,657.329	7.768	0.001
Residual	115	5,491.324		
Total	117			

Parameter	Estimate	Std Error	t	sig
Elevation	18.82	5.788	3.252	0.002
Elevation ²	-0.104	0.034	-3.045	0.003

Height ($r^2 = 0.239$)				
Source	df	Mean Square	F	Sig.
Regression	2	4,058.012	18.062	<0.001
Residual	115	224.670		
Total	117			

Parameter	Estimate	Std Error	t	sig
Elevation	4.979	1.171	4.253	<0.001
Elevation ²	-0.027	0.007	-3.865	<0.001

Average Shoot Length ($r^2 = 0.364$)				
Source	df	Mean Square	F	Sig.
Regression	2	3,992.971	32.959	<0.001
Residual	115	121.150		
Total	117			

Parameter	Estimate	Std Error	t	sig
Elevation	5.697	0.860	6.628	<0.001
Elevation ²	-0.031	0.005	-6.192	<0.001

Table 4.12 - Non-linear growth response to elevation in *Cornus sericea*

Quadratic linear regression models were fitted with elevation and elevation² as predictors of total shoot length, height, and mean shoot length. Both parameters were found to be significantly different than zero for all growth measures.

Total Shoot Length ($r^2 = 0.207$)				
Source	df	Mean Square	F	Sig.
Regression	2	22,284.84	9.90	<0.001
Residual	76	2,250.94		
Total	78			

Parameter	Estimate	Std Error	t	sig
Elevation	17.58	4.568	3.850	<0.001
Elevation ²	-0.108	0.027	-4.033	<0.001

Height ($r^2 = 0.35$)				
Source	df	Mean Square	F	Sig.
Regression	2	5,304.583	20.49	<0.001
Residual	76	258.813		
Total	78			

Parameter	Estimate	Std Error	t	sig
Elevation	9.523	1.549	6.149	<0.001
Elevation ²	-0.057	0.009	-6.283	<0.001

Average Shoot Length ($r^2 = 0.285$)				
Source	df	Mean Square	F	Sig.
Regression	2	3,895.992	15.13	<0.001
Residual	76	257.496		
Total	78			

Parameter	Estimate	Std Error	t	sig
Elevation	8.330	1.545	5.392	<0.001
Elevation ²	-0.049	0.009	-5.467	<0.001

5. DISCUSSION

Some clear results can be applied directly to field practice, while other less explicit patterns suggest directions for further experimentation. *Symphoricarpos albus* performs well as a live-stake under a range of conditions. The 100% survivorship of *S. albus* stakes planted above the high water line indicates that the tolerance to drought during establishment was not challenged, despite lack of irrigation and compacted soils. *Rubus spectabilis* appears to have little value as a live-stake using traditional planting methods, which is consistent with the experience of other practitioners (Alexander, pers. comm.; Darris 2002a).

Elevation was consistently the strongest factor affecting survival for all species, survival was markedly different between plantings above and below the high water mark, and no stakes survived continuous inundation during winter and the early growing season as found in blocks one and two. For *Cornus sericea* and *Symphoricarpos albus* survivorship increased rapidly across blocks three and four, which straddle the high water line. Elevation was not significant for predicting *Cornus sericea* or *Symphoricarpos albus* survival above the high water mark, indicating that survivorship was a threshold event pivoting on the elevation of the spring high water line. This is consistent with observations that adventitious root production is suppressed under hypoxic conditions (Pezeshki et al. 1998; Stannard & Guenther 1999; Zimmerman 1930).

Ability to form new roots under hypoxic conditions has been observed to vary among species (Stannard & Guenther 1999; Zimmerman 1930) reflecting varied physiological strategies for avoiding or tolerating the stress of hypoxia (Mitsch & Gosselink 2000). In contrast to *Salix*, which can metabolize and form roots at very low soil oxygen levels, *Rubus spectabilis* cannot support root formation from un-rooted cuttings in hypoxic soils. Although total survival was limited, *Rubus spectabilis* survival improved with elevation even among stakes planted above the high water mark, suggesting that rooting ability of *Rubus spectabilis* cuttings may be more sensitive to hypoxic conditions than *Cornus sericea* or *Symphoricarpos albus*. However, extremely low survivorship even above the high water line (12%) and minimal growth (13.0 cm mean total shoot length), suggest that *Rubus spectabilis* survival and root production were also strongly limited by other factors.

Spring water level was relatively stable in central pond over the early growing season, and only began to drop significantly during the early summer. Many wetland systems, particularly in the urbanizing landscape, are less stable with dramatic fluctuation of water elevation following rainfall (Azous & Horner 1997). The response of stakes to periodic inundation may be different from response to continuous inundation, rendering the location of the high water line less important to stake survival, and perhaps distributing the variation in survival over a wider elevational range. Redox potential decreases with duration of saturation and periodically inundated soils should experience more variation in redox potential over time as well as higher minimum redox levels (Richardson & Vapraskas 2001). The response of photosynthetic productivity to periodic inundation varies among species (Ewing

1996) and low but fluctuating soil redox potential caused by periodic inundation may require different adaptive strategies than those necessary under more persistent inundation.

Low soil porosity is common on re-graded restoration sites due to heavy equipment compaction and limited conservation of topsoil. Runoff can create a mosaic of hypoxic soils at low points during the early spring growing season. Given the apparent sensitivity of tested species to early growing season inundation, runoff accumulation at low points in areas of subtle topography could create patchy mortality that corresponds with regions of low soil redox potential resulting from persistent saturation.

Several bundles of *Rubus spectabilis* stakes were left stored in sawdust over the same early growing season. Rooting and growth were vigorous and far greater than with the field planted stakes. These results suggest that *Rubus spectabilis* may improve survivorship and growth where cuttings are completely buried. Over half the diameter of salmonberry stems typically contains soft pith that may increase the rate of water loss from cuttings where the pith is exposed. The potential for *Rubus spectabilis* stakes to dry out quickly has been mentioned by King County (2004). Burial of cuttings horizontally under mulch, or sealing the apical cut with latex paint, as commonly done with *Salix* in arid climates (Hoag 1993), may improve survival. Rhizome cuttings, buried under mulch or planted, is another approach that should be considered, as rhizomes readily sprout (Zasada et al. 1994), and *Rubus spectabilis* is commonly available for indiscriminant harvest from salvage sites.

By contrast, *Symphoricarpos albus*, with no current regional use as staked cuttings, suffered no mortality above the high water line, at any planting time. Growth was significant, with an average of over three shoots forming over 140 cm of new growth with no supplemental irrigation. By every measure, *Symphoricarpos albus* growth exceeded *Cornus sericea* planted in the same environment. Survival for *Cornus sericea* planted above the high water mark was typical for restoration sites (Humphreys, pers. comm.; Myers 2004a) suggesting that conditions at the test site were comparable to typical restoration sites with similar preparation. Although the full tolerances of *Symphoricarpos albus* is unknown, it can be planted with confidence above the level of persistent standing water, and it may prove useful where dry summer conditions limit the competitiveness of more wetland dependant species.

The significant non-linear relationship between elevation and all growth measures of *Cornus sericea* and *Symphoricarpos albus* suggests that growth declines under increasing moisture stress, and that these species may show optimal growth on the wetland edge. However, an edge effect resulting from the planting design should not be ruled out. The highest elevation block had an exposed western aspect and may have received higher levels of direct radiation than the lower elevation blocks that were more completely shaded by the new growth (Figure 4.1).

Previous studies provide some support for the hypothesis that given sufficient gas diffusion, increased soil moisture increases the rooting of cuttings (Rein et al. 1991). The growth response of species to different moisture regimes has not been well tested and more carefully designed tests across a broader range of conditions may reveal that

Cornus sericea is most competitive under a narrow range of conditions. Future studies should include characterization of soil water regime to provide a mechanistic basis for reduced performance.

Early planting time did not improve survival of *Symphoricarpos albus* and was significantly correlated with reduced survival in *Cornus sericea* when stakes were planted below high water. These results do not support the hypothesis that stakes planted in December would have more opportunity to form roots before bud flush. Metabolism is partially controlled by temperature (Salisbury & Ross 1985), and soil temperatures may have decreased by the December planting such that stakes were not able to make substantial root growth. Alternately, if soils were sufficiently compact to remain hypoxic under frequent winter rainfall, root formation could have been further suppressed. Cuttings of *Populus balsamifera* have failed to produce roots under hypoxic conditions (Stannard & Guenther 1999), and root growth under low pore water oxygen saturation (1.0 ppm) reduced rooting even in *Salix* (Zimmerman 1930). Early planting may reduce survival where stakes are exposed to prolonged low-oxygen conditions over winter. Planting following the beginning of heavy rains in fall, in areas where soils are not inundated over winter, may be a more appropriate setting to test the value of the early root growth hypothesis (Darris, pers. comm.).

Stakes planted in December, without the appropriate soil conditions for early root growth, may lose water as they sit exposed to sun and wind. Cuttings that lose water content have reduced rooting ability in the greenhouse (Hansen & Phipps 1983). Cuttings lose their ability to absorb water through basal cuts within a week limiting the ability of cuttings to absorb water before a root system forms (Rein et al. 1991). In a field setting it is likely that cutting moisture content would decline until roots form, particularly in exposed sections of stem containing buds necessary for shoot development making a benefit from early planting contingent on root formation.

At 40 cm length, stakes used in this study were about half the length of the industry standard and frequently less than 1 cm in diameter. Survival of stakes planted above the high water mark was 68% for *Cornus sericea* and 100% for *Symphoricarpos albus*, well within or exceeding industry expectations for a stake planting. *Cornus sericea* had the greatest variation in stake diameter, with stakes ranging from 8 mm to almost 25 mm, however increasing diameter did not improve survival or any growth measure in any of the analyses attempted. This weakens the hypothesis that larger cuttings have greater energy reserves that increase rooting ability (Hoag & Short 1992), and indicates that some species such as *Symphoricarpos albus* perform well from small cuttings. The greater availability of carbohydrates hypothesized for larger volume cuttings has not been tested and a number of factors would need to be controlled. Hybrid *Populus* cultivars have shown varied ability to mobilize carbohydrate reserves, independent of actual mobile carbohydrate levels (Tschaplinski & Blake 1989) suggesting that presence of carbohydrates through tissue does not necessarily translate to increased root formation. Change in rooting ability due to seasonal fluxes of non-structural carbohydrates may be more important than stem size (Meilleur et al. 1994; Zasada et al. 1994).

Cornus sericea survival consistently decreased among stakes with fewer nodes. It is unclear whether reduced survival resulted from an inadequate number of nodes located in the rooting zone and/or above ground. Sixteen percent of *Cornus sericea* stakes initiated shoots from below the mulch surface indicating that below ground buds are able to contribute shoots. Either cutting longer stakes or selecting branches with higher nodal density would both increase the average number of nodes on a cutting, and might improve survival. Given that rooting may be limited by the presence of oxygen in the soil profile, higher nodal density would increase the likelihood of a node being present in an advantageous position in the soil profile where stakes are cut with out regard to node location. The relative value of nodes as sites for rooting and the most advantageous position for them to occur in the soil profile has not been thoroughly described and varies significantly among species (Chmelar 1974; Darris 2002a).

For all three species there was a very weak negative correlation between the number of nodes and diameter. Howard and Ridout (1991) observed that dominant shoots have both higher stem elongation rates and larger diameters, and that cuttings from subordinate shoots (with narrower diameter) had a better rooting ability. Higher growth rates due to shoot dominance could increase the distance between nodes, causing a correlation between larger diameter and fewer nodes. Many plants will increase inter-nodal length in response the increased far-red/infrared fluorescence found under a leafy canopy (Salisbury & Ross 1985). New shoots in *Cornus sericea* often emerge from within the crown, and elongate rapidly with large inter-nodes. These shoots frequently lack side shoots making them desirable for stake production. Agricultural production of even-aged stems under full sunlight might increase nodal density, increasing the survival of *Cornus sericea* stakes.

Presence of a black fungal stain on the surface of *Rubus spectabilis* stakes appeared to increase survival. Salmonberry survival was so low that these observations are taken as cautionary but not conclusive. No other stake characteristics were correlated with the presence of the fungal stain, and no staining of pith or wood was observed during processing.

Models used to predict survival of *Cornus sericea* were generally weak, indicating that unmeasured factors contribute to survival factors or that random variation prevent precise prediction of survival. Increasing control of experimental conditions is not unconditionally desirable. Results of experiments under increasingly artificial environments may increasingly deviate from field outcomes, reducing the immediate value of findings for field practitioners. Increased control of cutting variation requires precision that cannot be duplicated in commercial practice. However, controlled experimentation may reveal specific mechanisms that control rooting within and between the species of our native flora and additional labor could then lead to more accurate prediction of whole plant response in a complex natural environment. Given the absence of economic interest in this line of inquiry, carefully designed field trials offer a return on investment best suited for improving field practices to increase the effectiveness of revegetation efforts.

A number of topics for further study were suggested by the literature review, conversations with informants, or field observations. Response patterns are likely to differ among species, and so experimentation with a diversity of species is advantageous. Observation of variation among species will simultaneously provide specific information for field revegetation practice, while outlining functional groupings of species that could lead to isolation of the physiological mechanism(s) in a more controlled setting. Developing a more precise picture of the horticultural response of a range of species within our lowland wetland vegetation will clarify the factors that affect competitive ability, leading to a more precise understanding of the factors affecting community composition and succession. The following propositions should be explored for the immediate benefit of restoration practice, and trials could be incorporated into existing restoration plantings:

- (1) Shoots emerged from buried buds for all species planted (data not shown). Partially processed cuttings of shoots or roots scattered on a soil surface and covered with mulch may root readily, allowing low cost plantings to be installed as part of site preparation with a savings of labor. This may be particularly valuable for species that root well from cuttings but are easily desiccated (*Rubus spectabilis* and *Sambucus racemosa*).
- (2) For species without pre-formed root primordia, storage in moist sawdust may allow for development of root initials in a protected environment, priming cuttings for out-planting, and increasing rooting (Vanbianchi, pers. comm.). This approach may be more advantageous than early planting due to loss of moisture from exposure in the field.
- (3) Soaking cuttings before planting can stimulate pre-formed primordia (Carlson 1938) and improve performance in dry soils (Hansen & Phipps 1983), but duration of soaking (Schaff et al. 2002) may affect success.
- (4) Root development in fall, before soil temperatures drop may increase the ability of some species to support spring growth (Darris, pers. comm.). *Oemleria cerasiformis* and *Sambucus racemosa* break buds very early in spring and may benefit from fall planting or 'priming' that allows for early root development.
- (5) The growth environment of stock plants can be manipulated under agricultural conditions to affect the biochemical state of cuttings at the time of excision. Limited information is available on shoot production rates or response of native shrubs to heavy pruning (Tappeiner et al. 2001). Stakes grown in full sun under fertile conditions may root more effectively than wildcrafted stakes due to greater nodal density or improved nutrient status in excised stems.
- (6) Genotypes within a species growing under conditions of periodic flooding and sediment deposition may have increased rooting ability, and may be better adapted to life in hypoxic soils.

- (7) Survival and growth of different stock types should be tested with side-by-side comparison of cuttings, bareroot stock, and different sized potted stock of native plant species under varied growing conditions. Increasing stock weight and size dramatically increases per-stem installation costs, and requires adequate and verified benefits. Based on observations of *Symphoricarpos albus*, cuttings may perform as well as potted stock.

6. CONCLUSION

Symphoricarpos albus, *Cornus sericea* and *Rubus spectabilis* are intolerant of spring inundation when planted as unrooted cuttings, and suffered 100% mortality where cuttings were submerged during bud flush. Survivorship rapidly increased to 68% for *Cornus sericea* and 100% for *Symphoricarpos albus* above the high water line. Response to wetland elevation in these two species appears to be a threshold phenomenon related to soil saturation during bud flush. *Symphoricarpos albus* is currently not used for staking but has proven to be more reliable than *Cornus sericea*, which is commonly used.

December planting failed to improve survival or growth, and may reduce survival of *Cornus sericea* and *Rubus spectabilis*. Diameter, which has been credited with improving stake survival, had no effect on any growth measure for any of the species tested. For *Cornus sericea*, which has the potential for widely spaced nodes, increasing nodal density was correlated with greater survival. Forty-centimeter cuttings of thin diameter produced substantial growth in one growing season without irrigation, indicating that broadly distributed specifications for 60-100 cm stakes may be unnecessary where disturbance is not a factor.

Although I strongly encourage experimentation, the following suggestions are made to maximize successful staking of *Cornus sericea* and *Symphoricarpos albus*:

- ◆ Harvest and plant stakes in late winter before buds begin to swell to minimize moisture loss between planting and root development.
- ◆ If planting cannot occur immediately after harvest, store stakes in a cool, moist, well oxygenated environment, and plant before buds begin to swell.
- ◆ For *Cornus sericea*, select stakes with dense internodes, or if internodes are long, cut stakes to a length such that there are at least 3 nodes per stake.
- ◆ Avoid planting in anaerobic soil conditions, as indicated by redoximorphic indicators like low soil chroma (grayish soils), rotten egg smells, or standing water.
- ◆ It is unnecessary to select stakes based on large diameter.
- ◆ It is unnecessary to plant stakes deeply where rooting is shallow, except where physical disturbance may dislodge stakes.

A large number of potentially effective staking methods are precluded by rigid installation specifications and should be considered by practitioners:

- ◆ Planting depth may be less important than the length of root forming stem located in soil depths where moisture and oxygen levels are suitable for adventitious root production. For this reason, vertical planting may not be the most effective installation method.
- ◆ Depending on the species, stem age may significantly control rooting ability more than stake diameter.
- ◆ Rooting may occur in soil or mulch allowing for planting through mulch, and burial under much as installation options.
- ◆ Root cuttings under mulch may be an effective field propagation method for rhizomatous species, or species that form shoots from damaged roots.
- ◆ Staking may be an effective propagation method in more upland areas, where species like *Symphoricarpos albus* are well adapted.

The potential for vegetative reproduction in many native species appears to be unexplored and under-utilized. Increasing the number of species used as un-rooted cuttings is limited primarily by lack of experimental evidence, and the inability of practitioners to experiment within the confines of the marketplace. A broad range of topics related to revegetation practice using un-rooted cuttings are largely unexplored.

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Appendix A – List of Species With Potential for Field Propagation

SPECIES	Status
<i>Corunus sericea</i>	commonly used
<i>Corylus cornuta</i>	potential for root cuttings
<i>Crataegus douglasii</i>	potential for root cuttings
<i>Lonicera involucrata</i>	uncommonly used
<i>Malus fusca</i>	rare use reported
<i>Oemleria cerasiformis</i>	rare use reported
<i>Oplopanax horridus</i>	unknown
<i>Philadelphus lewesii</i>	rare use reported
<i>Physocarpus capitatus</i>	uncommonly used
<i>Populus tremuloides</i>	potential for root cuttings
<i>Populus trichocarpa</i>	commonly used
<i>Ribes bracteosum</i>	unknown
<i>Ribes divaricatum</i>	unknown
<i>Ribes lacustre</i>	unknown
<i>Ribes laxiflorum</i>	unknown
<i>Ribes sanguineum</i>	rare use reported
<i>Rosa gymnocarpa</i>	rare use reported
<i>Rosa nutkana</i>	rare use reported
<i>Rosa pisocarpa</i>	unknown
<i>Rubus leucodermis</i>	unknown
<i>Rubus parviflorus</i>	unknown
<i>Rubus spectabilis</i>	rare use reported
<i>Salix hookeriana</i>	commonly used
<i>Salix lucida</i>	commonly used
<i>Salix scouleriana</i>	commonly used
<i>Salix sitchensis</i>	commonly used
<i>Sambucus racemosa</i>	rare use reported
<i>Spiraea douglasii</i>	uncommonly used
<i>Symphoricarpos albus</i>	excellent potential (see study)
<i>Viburnum edule</i>	unknown

Appendix B – Photographic Figures



Figure B.1 – Collection site for *Rubus spectabilis*, Sammamish Plateau



Figure B.2 – Collection site for *Symphoricarpos albus*, Snoqualmie Floodplain



Figure B.3 – 40-cm *Rubus spectabilis* stakes ready for planting



Figure B.4 – Experimental site, Union Bay Natural Area, Seattle, WA



Figure B.5 – Planting site after December installation



Figure B.6 – Planting frame with initial planting of marked stakes



Figure B.7 – High water inundating planting block 3



Figure B.8 – Bud flush of *Symphoricarpos albus*, level 2 phenology