ROOTING STEM CUTTINGS OF SHANTUNG MAPLE (*ACER TRUNCATUM*), MOUND LAYERING SHANTUNG AND CADDO SUGAR MAPLES (*ACER SACCHARUM*), AND USING EASTERN REDCEDAR (*JUNIPERUS VIRGINIANA*) AS A SUBSTRATE COMPONENT IN STEM CUTTING PROPAGATION

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Abstract

Heat and drought tolerance make shantung maple (*Acer truncatum*) and caddo sugar maple (*A. saccharum*) good candidates for midwestern landscapes. Improving cutting propagation or mound layering techniques could increase the availability of these species.

The influence of time of year, cutting position, and auxin concentration, formulation, and solvent on rooting of stem cuttings of shantung maple was investigated. Semi-hardwood cuttings rooted best (55%). Generally, rooting percentage decreased as indole-3-butyric acid (IBA) concentration increased. Cutting position, auxin formulation, and solvent did not affect rooting. Mean root number and mean root length were unaffected by treatments. Results suggest semi-hardwood cuttings and low IBA concentrations [< 2500 ppm (0.25%)] promote rooting.

Auxin concentration influenced rooting of caddo and shantung maple mound layered shoots. Rooting peaked at 15,000 ppm (1.5%) IBA for both caddo (71%) and shantung maples (34%). Mean root number for caddo, but not shantung, increased as IBA concentration increased. Differences in mean root length were not significant. Growers may now propagate caddo maple by mound layering. For shantung maple propagation, stem cuttings are recommended.

Propagation substrates can strongly influence rooting success of stem cuttings. Eastern redcedar (*Juniperus virginiana*) chips (ERC) have been suggested as a propagation substrate component. This report investigated ERC as a perlite substitute in a 3 perlite: 1 sphagnum peat moss (v/v) rooting substrate. Stem cuttings of spreading euonymus (*Euonymus kiautschovicus*), forsythia (*Forsythia* x *intermedia*), English ivy (*Hedera helix*), lantana (*Lantana camara*), and coleus (*Solenostemon scutellarioides*) were rooted in substrates containing increasing concentrations of ERC hammer milled to pass a 4.8 mm (0.19 in) screen. All species rooted well (≥95%) in all substrates except forsythia which rooted poorly in all substrates (8% to 36%). ERC did not affect mean root number or mean root length in any species except spreading euonymus where mean root number peaked at 0% and 100% ERC content and mean root length decreased with increasing ERC content. Bulk density, container capacity, and total porosity increased as

ERC replaced perlite. Physical properties of all substrates were suitable for cutting propagation. ERC can effectively replace perlite in rooting substrates for many ornamental species.

Table of Contents

List of Tables

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Chapter 1 - Literature Review

Part I - Propagation of Caddo Sugar Maple and Shantung Maple

Introduction

Recent studies have confirmed the value of healthy landscapes in urban centers. In these often small spaces, trees play a large role in framing microclimates and offering long-lasting benefits. Besides the psychological value of gardens demonstrated by Freeman et al. (2012), trees also provide economic and environmental benefits to the landscape. A study by Sander et al. (2010) demonstrated that having tree cover within 250 m (820 ft) of a house significantly increased its market value. Nowak et al. (2006) showed the ability of trees to reduce air pollution. Jensen et al. (2003) showed that homes in locations with a higher leaf area index tend to use less energy during the summer than homes in locations with a low leaf area index. This demonstrates the benefits of shade provided by trees. Xiao and McPherson (2003) even suggested that rain interception by trees plays a role in reducing the rate of stormwater runoff. Clearly, establishing and maintaining healthy trees is a worthwhile investment in urban locations.

Trees in midwestern landscapes face a host of physical and physiological challenges. The use and maintenance of outdoor spaces often leads to tree wounding. Furthermore, the space available for tree growth is often limited by the size of a yard, surrounding buildings, or overhead power lines. Trees must be carefully selected for their location based on their mature size to avoid costly maintenance needs in the future. Besides these physical challenges, trees in midwestern landscapes are exposed to a host of physiological stresses ranging from bitter cold in the winter to the prolonged drought and heat of summer.

With these challenges, selecting appropriate trees for midwestern landscapes is a difficult task. Horticulturists can use two strategies to find plants suited to these locations. First, they can search the region for native plants with suitable characteristics for urban landscapes. Second, they can select landscape plants that are native to other regions of the world with similar climates. The two maples described in this thesis represent both of these strategies. Caddo sugar maple (*Acer saccharcum* Marsh. subsp. *saccharum*) (caddo maple) is native to central Oklahoma. Shantung maple (*A. truncatum* Bunge) is native to northern China, which has a climate similar to the midwestern region of the United States. The tolerance for drought and heat which allows caddo and shantung maples to thrive in their native ranges also suits them well for service in midwestern landscapes.

Caddo Maple

Origin

The term “caddo maple” refers to a disjunct population of sugar maples native to central Oklahoma (Dent and Adams, 1983; Dirr, 2009). These maples get their name from the Caddo Native Americans and are considered to be near the western extreme of the natural sugar maple range. Sugar maples are native to the northeastern states. Preston and Braham (2002) describe the native sugar maple range as reaching as far west as eastern Minnesota and eastern Kansas. Dirr (2009) notes that sugar maples can be found as far south as Georgia and Texas. Simpson and Hipp (1993) specifically define caddo maples as the population of sugar maples occurring in the calcareous canyon soils of Caddo and Canadian County, Oklahoma [about 80 km (50 mi) west of Oklahoma City]. This distinguishes them from the maples occurring in the acidic soils of the Wichita Mountains in Comanche County, south of Caddo County. Caddo maples are specifically associated with the Red Rock Canyon Area near Hinton, Oklahoma (Pair, 1994).

Visual Characteristics

Caddo maples grown in managed landscapes stand 9 to 15 m (30 to 50 ft) tall at maturity (Dirr, 2009). Open-grown specimens develop rounded heads with diameters of about 12 m (40 ft) (K-State Res. and Ext., 2008). Preston and Braham (2002) classify sugar maple as a slow growing tree. However, a trial of caddo maples grew 5.6 m (18.5 ft) in 8 years at the John C. Pair Horticulture Center near Haysville, Kans. (Dirr, 2009). Sugar maples may live for 300 to 400 yr (Preston and Braham, 2002), but the stress of urban settings likely reduces their life span.

Like other maples, sugar maples develop extensive root systems, especially near the soil surface. In their natural forest setting, these roots absorb nutrients from the layer of decomposing leaf litter (Preston and Braham, 2002). In the landscape, these surface roots compete with turfgrass.

Sugar maple trunks can reach 60 to 90 cm (2 to 3 ft) in diameter. Their dark grey bark is smooth during juvenility, but develops long shallow fissures at maturity. Twigs are red-brown and glabrous. Buds are red-brown, about 6 mm (0.25 in) in length with acute tips (Preston and Braham, 2002).

Leaves of caddo maples are simple, arranged oppositely, and smaller than other sugar maples (Pair, 1994). Though leaves are five lobed, the two basal lobes are suppressed. Margins are entire. The U-shaped sinuses extend about half way to where the petiole attaches to the leaf blade (Preston and Braham, 2002). Dark green, leathery foliage during the summer turns shades of yellow, orange, or red during autumn (Dirr, 2009). Pair (1994) notes that caddo maples develop fall color late. Selections such as the 'John Pair' caddo maple have been made for early fall color (Dirr, 2009).

Flowers appear in spring as leaves emerge. Trees are polygamous, bearing perfect, pistillate, and staminate flowers on the same plant. Flowers are yellow-green, apetalous, and borne in clusters on long pedicels (Hardin et al., 2001; Preston and Braham, 2002).

Fruits are red-brown, double samaras with parallel to slightly divergent wings ≈2.5 cm (1 in) in length, and ripen in the fall. Sugar maples produce good seed crops every 1 to 4 yr (Hardin et al., 2001; Preston and Braham, 2002).

Landscape Use

Simpson and Hipp (1993) recommend caddo maples for sites with alkaline soils (≈7.5 pH). Studies by Pair (1994) and Griffin (2005) showed that caddo maples have better drought tolerance than several common sugar maples. Their leaves are also thicker, making them more resistant to wind tatter, a common problem of maples in the Great Plains region of the United States. Pair concluded that caddo maples are ideal for the drought, wind, and shady conditions that occur in urban landscapes.

Cultivars

There are primarily three caddo maple cultivars on the market today.

‘John Pair’

The ‘John Pair’ caddo maple (*A. saccharum* Marsh. subsp. *saccharum* ‘John Pair’) is a compact cultivar named posthumously in honor of Dr. John Pair. It matures to 9 m tall x 9 m wide (30 ft tall x 30 ft wide) and is hardy to zone 5. Its dark green, glossy leaves are tatter resistant and are often held into winter. Pair selected this cultivar for its early, brilliant red fall color (Dirr, 2009).

‘Autumn Splendor’

Another caddo maple selected by Dr. John Pair, ‘Autumn Splendor’ caddo maple (*A. saccharum* Marsh. subsp. *saccharum* ‘Autumn Splendor’) was released by J. Frank Schmidt and Son Co. in 2006. Dirr (2009) notes that ‘Autumn Splendor’ caddo maple showed faster growth than ‘John Pair’ caddo maple in one trial. The tree matures to 14 m tall x 12 m wide (45 ft tall x 40 ft wide) with orange-red fall color.

Flash Fire™

Released in 2010 by J. Frank Schmidt and Son Co., Flash Fire™ caddo maple (*A. saccharum* Marsh. subsp. *saccharum* ‘JFS-Caddo2’) was selected for its bright red fall color and mildew resistance. The tree has a broad oval form, is hardy to zone 5, and matures to 14 m tall x 12 m wide (45 ft tall x 40 ft wide) (J. Frank Schmidt & Son Co., 2010).

Summary

As a near native to the midwest, caddo maples hold good potential as urban trees. Their tolerance for alkaline soils and their excellent summer and fall colors position them as both healthy and attractive trees for urban sites. Large enough to shade a yard and tough enough to withstand scorching winds, caddo maples deserve a place in midwestern landscapes.

Shantung Maple

Origin

Shantung maple is native to northern China, Russia, Korea, and Japan (Dirr, 2009; Pair, 1986). Its common name is likely derived from the Shandong province in northeastern China. It is also known as purpleblow maple – a name which describes the rich color of its tender shoots and leaves. Its specific epithet refers to the shape of its leaf bases, though this varies. Although currently considered to be a unique species, shantung maple has been classified and re-classified many times due to its close relationship to painted maple [*A. pictum* subsp. *mono* (Maxim.) H. Ohashi] (also known as mono maple) and coliseum maple (*A. cappadocicum* Gled.) (Browse, 1990).

Visual Characteristics

Mature shantung maples reach heights of 6 to 8 m (20 to 25 ft) and canopy spreads of about the same or slightly less. The tree is round-headed and slow growing with reports of specimens in the midwest that averaged 43 cm (1.4 ft) of growth per year during a 9-yr observation (Dirr, 2009).

Trunks display rough fissured grey-brown bark and branching is often dense. Twigs are glabrous, green becoming dark brown as they lignify. Buds are four-sided, imbricate, red-brown, about 6 mm (0.25 in) in length, and glabrous except for a tuft of hair at their tips (Dirr, 2009).

Emerging leaves are purplish-red, becoming dark, glossy green above and pale green below. Leaves tend to have 5 (sometimes 7) lobes. Lobes are ovate with acuminate tips. Margins are usually entire, though lobes of some specimens are incised. Leaf bases are truncate to hastate. Leaves are glabrous except for a tuft of hair where petiole attaches to blade. This distinguishes shantung maple from painted maple, which has pubescent petioles. Fall leaf color ranges from yellow to reddish-purple (Dirr, 2009; Toy, 1966).

Flowers are green-yellow, 1.3 cm (0.5 in) wide, and develop in spring, held upright on stalks 1.3 cm (0.5 in) in length. Fruit is a double samara with wings 3 to 4 cm (1.25 to 1.5 in) in length (Dirr, 2009). Wings are right to obtusely angled (90 to 160°).

Besides shantung maple, the similar painted maple also may be encountered in the landscape. Painted maple grows twice as large as shantung maple, reaching heights of 15 to 18 m (50 to 60 ft) with a vase shaped crown. Painted maple also has smoother bark and larger leaves than shantung maple. Leaves of painted maple have pubescent petioles in contrast to the glabrous petioles of shantung maples (Dirr, 2009).

Landscape Use

Shantung maples were first introduced to the U.S. in 1881 (Pair, 1986) and are hardy to zone 3, though this likely depends on provenance (Dirr, 2009; Pair, 1986). At least three characteristics of shantung maples suit them well for midwestern states. First, their drought tolerance prepares them to survive the dry summers common in the Midwest. In a reforestation study in the loess plateau of China, Cao et al. (2008) found that improved soil moisture only slightly improved growth of shantung maples. This demonstrates the excellent drought tolerance of this species. In most cases, merely surviving is insufficient performance for landscape plants. They must also look attractive. Shantung maples easily meet this criterion with their purple tinged new growth, glossy summer foliage, and fall colors ranging from yellow to red. Lastly, the size of shantung maples makes them ideal for many landscaping situations. Maturing at 6 to 8 m (20 to 25 ft) (Dirr, 2009), they make excellent specimen trees and can fit easily in areas where power lines are a concern. As an added advantage to the great landscaping qualities this tree offers, Pair (1986) noted that shantung maples growing in Kansas trials were generally disease-free except for a few cases of tar spot at a trial in Colby, Kans. Shantung maples seem excellent candidates for many landscape situations in midwestern states and would likely be used more frequently if they were more available.

Cultivars

Despite the diversity of leaf shapes and fall colors available among shantung maple specimens, relatively few cultivars are commercially available.

'Fire Dragon'

'Fire Dragon' shantung maple (*A. truncatum* 'Fire Dragon') was selected in Fort Worth, Texas in 1999 and introduced by Keith Johansson (Metro Maples, Fort Worth, Texas). This cultivar grows well on high pH soil and displays brilliant red fall color. Johansson noted its tolerance for drought, wind, and ice (Johansson, 2006).

Main Street™

Main Street™ shantung maple (*A. truncatum* 'AT-WF1') was introduced by Mike Worthington (Worthington Farms, Inc., Greenville, NC). The original specimen reached

6 m tall x 4 m wide (20 ft tall x 14 ft wide) in 10 yr without irrigation. The dense foliage on its tight crown turns orange-red during autumn. (Dirr, 2009).

Sunset Series

Since 1989, J. Frank Schmidt and Son, Co. have released several hybrid crosses of shantung maple and Norway maple (*A. platanoides* L.), all with “sunset” in their trade name. Norwegian Sunset™ maple (*A. truncatum* x *A. platanoides* ‘Keithsform’) and Pacific Sunset™ maple (*A. truncatum* x *A. platanoides* ‘Warrenred’) are similar with green foliage and orange-red fall color, though Pacific Sunset maple has a finer branch structure and displays its fall color earlier than Norwegian Sunset maple (Dirr, 2009). Crimson Sunset™ maple (*A. truncatum* x *A. platanoides* ‘JFS-KW202’) is a hybrid of shantung maple and ‘Crimson King’ Norway maple (*A. platanoides* ‘Crimson King’), a popular cultivar with purple foliage. It displays the purple foliage associated with ‘Crimson King’ Norway maple, but offers better heat tolerance. (J. Frank Schmidt & Son Co., 2011).

Summary

Although not a native to midwestern states, shantung maples are well suited to the conditions present in urban landscapes. With their compact size, drought tolerance, and brilliant fall colors, shantung maples have advantages over several species of popular specimen trees used today. As more cultivars are developed, shantung maple will likely gain popularity in the future.

Propagation Methods

Perhaps the greatest obstacle to caddo and shantung maple’s success in commercial markets has been the challenges associated with clonally propagating these species. Although seedling production and grafting both yield usable specimens, these methods are accompanied with certain disadvantages. An efficient method of propagating caddo and shantung maples by vegetative cuttings or layering could increase the availability of these maples and eliminate many of the complications associated with other methods of propagation.

Seed

Seed propagation is one of the simplest methods of propagating maples. However, this process does not yield genetically identical specimens. Both caddo and shantung maple seeds ripen in the fall. (Ackerman, 1957; Preston and Braham, 2002). Browse (1990) recommends harvesting shantung maple seeds while they are still green, suggesting that dry seed coats may hinder successful germination.

Seeds of both species require a period of moist stratification at 1 C (34 F) (Pair, 1986; Preston and Braham, 2002). Caddo maple seeds need 60 to 90 d of stratification according to Dirr (2009). On the other hand, Pair (1986) reports excellent germination of shantung maples after only 45 d of stratification. Dirr (2009) also notes that sugar maple seeds are often non-viable. Furthermore, seeds may not be widely available every year, as experienced by Simpson and Hipp (1993) who were unable to obtain caddo maple seeds in Fall 1977. In contrast to caddo maples, shantung maples produce abundant seed which germinates readily. Ackerman (1957), Browse (1990), and Pair (1986) all report about 90% germination of shantung maple seeds. Good seed production requires pollination between trees because, although shantung maple is monoecious, the male and female flowers open at different times (Dirr, 2009).

Seed propagation offers the advantages of being cheap and simple, but it is only useful when genetic variation is acceptable. Therefore, although it is useful for developing new cultivars, it is not an effective method of propagating specific clones. One application of seed propagation is rootstock production for the grafting techniques that will be discussed next.

Grafting

Many of the maple cultivars available today are propagated by grafting. Unfortunately, very little specific information is available regarding caddo or shantung maple grafting. This portion of the literature review relies heavily on the work conducted with Japanese maple (*A. palmatum* Thunb.) and Norway maple.

Due to the historically low efficiency of cutting propagation, both caddo and shantung maples are usually propagated by grafting onto seedling rootstocks. Pair (1994) briefly mentions that caddo maples can be successfully grafted onto caddo seedlings in

July and August using T-budding. Alternatively, Vertrees (1978) states that many nurserymen graft Asiatic maples in late winter using side veneer grafting. This would likely work for shantung maples as well.

Howard (1993) extensively studied grafting of Norway maple, a species related to shantung maple. (Shantung and Norway maples can hybridize as in Crimson Sunset™ maple.) He showed that chip budding in early August onto rootstocks planted the previous winter gave satisfactory results in certain cases. Of the factors affecting successful bud take, he suggested that rootstock growth played a stronger roll than weather conditions. Howard and Oakley (1997) continued the work and found that vigorous rootstock growth greatly improved bud-take. In a deep sand bed with trickle fertigation, they achieved grafting efficiencies as high as 100%. Although Howard (1993) suggests chip-budding offers advantages over the older T-budding technique, Pair et al. (1996) note successful shantung maple grafting using the T-budding approach.

The main advantage of grafting, when compared to seed propagation, is that the shoot systems of new plants are genetically identical to the scion-donor plant. Grafting has long been used as a method of propagating plants whose cuttings are difficult to root. When bud-grafting is used, a large number of plants can be obtained from a relatively small amount of scion wood. Furthermore, some nurserymen prefer grafted trees over cuttings, because grafted trees can be placed on a vigorous seedling rootstock with a natural rooting pattern, while cuttings form modified rooting patterns.

On the other hand, several disadvantages also come with grafting. In some cases, rootstock material is prone to develop suckers which compete with the desired scion. Homeowners may have difficulty selecting the appropriate shoots to remove. Care should also be taken to select rootstocks with an appropriate provenance. Although caddo maples are likely compatible with other sugar maple rootstocks, their tolerance for alkaline soil and drought may be lost when less tolerant rootstocks are chosen (Le Duc and Pair, 2000). Lastly, grafting is a labor intensive technique. Some nurseries may lack the technical expertise or the experience necessary to manage their own grafting program.

Despite these challenges, grafting has historically been the most effective method for propagating many maple cultivars. With proper materials and management, it is a commercially feasible method of maple production.

Cuttings

Maples can be propagated by cuttings, but reported rooting responses vary widely among researchers. Many factors must be considered. Successful maple propagation relies on selecting good cuttings. Most maple studies use 8 to 15 cm (3 to 6 in) terminal cuttings (Chapman, 1979; Dunn and Townsend, 1954; Enright, 1958; Koelling, 1968; Pair, 1986; Snow, 1941). Anstey (1969) stated that 20 to 23 cm (8 to 9 in) cuttings of Japanese maples rooted better than 7 to 10 cm (3 to 4 in) cuttings. However, large numbers of long terminal cuttings are not always available when few stock plants exist (Enright, 1958). In some cases, cuttings as short as 5 cm (2 in) have rooted well. (Gabriel et al., 1961; Tousignant et al., 2003).

Growth stage of stock plants influences rooting success more than cutting length. Propagators categorize woody cuttings into one of three growth stages: softwood, semi-hardwood, and hardwood. Softwood refers to tender, actively growing tissue and is characterized by expanding leaves, unlignified stems, absence of a terminal bud, and often coloration that differs from mature tissue, such as the yellow new growth of ‘Hearts of Gold’ eastern redbud (*Cercis canadensis* L. ‘Hearts of Gold’). Maples can produce multiple flushes of softwood during a single growing season. Plants raised from softwood cuttings tend to grow vigorously, even during their first season. However, softwood cuttings are prone to rot during mist propagation (Chapman, 1979). Generally, propagators prefer softwood cuttings (if they root successfully) over other stages of growth because they are available earlier in the season which allows rooted cuttings more time to develop before winter.

Propagators refer to cuttings taken during summer as “semi-hardwood”. Semi-hardwood cuttings are taken after current year shoots finish elongating, lignify, and terminal buds develop, but before leaves senesce and plants go dormant for winter. For species with especially tender new growth, semi-hardwood cuttings may root better than softwood cuttings because they are less prone to rot.

Hardwood refers to dormant plant material. Hardwood cuttings are collected during the winter, normally from the recent season’s growth. Until leaves develop, hardwood cuttings are incapable of manufacturing photosynthates. Thus, they must rely on non-structural carbohydrate reserves during rooting and leafing-out. Some propagators harvest dormant large-diameter limbs during winter and place them in greenhouses, where they produce softwood cuttings earlier than stock plants grown outdoors. Preece et al. (2002) describe a method of forcing 3 to 25 cm (1 to 10 in) diameter hardwood stem sections to produce softwood cuttings which can be collected for tissue culture work or cutting propagation.

For maple propagation, many propagators recommend softwood cuttings (Chapman, 1979; Coggeshall, 1957; Snow, 1941). Proponents for softwood cuttings emphasize the advantages of juvenile tissue (Chapman, 1979; Coggeshall, 1957) and extensive cutting growth before winter (Anstey, 1969). For some maples, partially lignified softwood seems superior to tender new growth. Studying amur maple (*A. ginnala*), hedge maple (*A. campestre*), Norway maple, and red maple (*A. rubrum*), Chapman (1979) reported that softwood cuttings taken in May sometimes rotted before rooting occurred, but that most softwood cuttings taken in June rooted successfully. Vertrees (1978) noted that Japanese maple can be rooted by semi-hardwood or hardwood cuttings. This demonstrates that other stages of growth besides softwood can also root successfully.

As cuttings are taken later in the year concerns about overwintering arise. Dixon (1980) noted that cuttings of Japanese maple did not survive winter unless they had broken bud before winter. Pre-winter growth replenishes carbohydrate reserves of newly rooted cuttings (Pair, 1986; Smalley and Dirr, 1987). The work of Smalley and Dirr (1987) emphasizes that winter survival depends more on nonstructural carbohydrate reserves than on pre-winter growth. Cuttings of red maple ‘October Glory’ were harvested in June and August. If cuttings did not break bud before winter, cuttings from the later collection date survived winter better than cuttings harvested in June. Measuring nonstructural carbohydrate levels of cuttings mid-winter, Smalley and Dirr found that August cuttings contained carbohydrate levels comparable to cuttings taken in June which had broken bud and grown. The cuttings least likely to overwinter successfully were those collected in June which had not broken bud and whose reserves could not be replenished until the following spring. All cuttings that broke bud after rooting overwintered successfully.

The work of Smalley and Dirr (1987) suggests the possibility of successful late season maple propagation. Pair’s work (1986) with shantung maple supports this conclusion. He showed that although cuttings rooted poorly in May (due partly to adverse conditions in outdoor mist beds), 15 cm (6 in) semi-hardwood cuttings collected in early August rooted satisfactorily. Successful overwintering was achieved by extending the photoperiod to encourage shoot growth of rooted cuttings.

Cutting Propagation of Caddo Maple

Researchers have studied cutting propagation of sugar maples extensively, hoping to propagate clones with superior sap for use in the maple syrup industry. Several papers demonstrate that cuttings possess maximum rooting ability as shoots finish elongating and terminal buds develop (Dunn and Townsend, 1954; Enright, 1958; Koelling, 1968; Tousignant et al., 2003; Yawney and Donnelly, 1981). At this stage leaves are full size, petioles are reddish-purple at their base, stems are lignifying, and terminal buds have about two visible scales (Yawney and Donnelly, 1981). This correlates to stem water potentials between 55% and 75% and occurs after about 270 degree-days above 5 C (41 F) in Quebec, Canada (Tousignant et al., 2003). The number of visible terminal bud scales correlates to stem water potential and might indicate optimal cutting harvest times (Tousignant et al., 2003). However, optimal harvest time will vary from year to year with weather conditions and from plant to plant due to genetic differences (Alsup, 2001; Donnelly and Yawney, 1972).

Many scientists have investigated which auxin types and rates stimulate optimal rooting in sugar maples. However, results are non-conclusive. Donnelly and Yawney (1972) tested several concentrations of indole-3-butyric acid (IBA), 1-naphthalene acetic acid (NAA), and combinations of the two, but observed no significant differences in rooting (25% to 50%) among auxin treatments. Enright (1958) used rates as high as 20,000 ppm (2.0%) IBA to stimulate rooting. Working specifically with caddo maples, Alsup (2001) observed only 30% rooting of stem cuttings treated with IBA ranging from 5000 to 15,000 ppm (0.5% to 1.5%). Many of the differences in results among these studies can be attributed to genetic differences among stock plants. Yawney and

Donnelly (1972) and Alsup (2001) demonstrated that genetically unique trees respond differently to auxin.

In early work on sugar maple cutting propagation, Snow (1941) observed 65% rooting after a 50 ppm (0.05%) IBA soak for 3 h. Then in 1958, Enright reported 90% rooting success with 50 sugar maple cuttings collected in June in Maryland and rooted in a greenhouse in coarse sand under intermittent mist. His highest auxin rate, a 5 sec dip in 20,000 ppm (2.0%) IBA solution, caused the best rooting. Other scientists have observed similar results. While studying timing of cutting harvest, Donnelly and Yawney (1972) observed up to 85% rooting of cuttings taken in early June in Vermont. Working with 24 clones harvested in early June in southeast Canada, Tousignant et al. (2003), averaged ≈75% rooting from most trees after a 4000 ppm (0.4%) IBA talc treatment. These examples demonstrate that sugar maples can be efficiently propagated from cuttings in ideal circumstances. However, due to genetic differences among stock plants, experimentation will be needed to determine ideal IBA treatments for individual specimens.

Cutting Propagation of Shantung Maple

Compared to the extensive work devoted to improving sugar maple propagation by cuttings, relatively few studies have dealt with shantung maple. Vertrees (1978) investigated the rooting ability of semi-hardwood stem cuttings from several Asiatic maples, including shantung maple. Cuttings were treated with 8000 ppm (0.8%) IBA talc and although most species rooted well, cuttings of shantung maple failed to root.

Pair (1986) propagated shantung maple stem cuttings with better success. Results from his work suggest that semi-hardwood cuttings root better than softwood cuttings. Treating both terminal and subterminal 15 cm (6 in) cuttings with a 5 s dip in solutions of up to 5000 ppm (0.5%) IBA (solvent not specified), he observed 62% rooting overall from semi-hardwood cuttings. Unfortunately, this study lacked sufficient repetition to support strong conclusions. Further research is needed to verify these results.

Podaras and Bassuk (1996) investigated shantung maple as part of a broad study on maple species propagation. Shantung maple stem cuttings were dipped in solutions of IBA [0 to 10,000 ppm (0% to 1.0%)] dissolved in 1 ethanol: 1 water (v/v). A four-year-old greenhouse-grown specimen rooted best (51% to 56%) when treated with 0 and 1000 ppm (0% and 0.1%) IBA. When shoots of the greenhouse-grown stock plant were banded and etiolated, rooting increased to 88% at the only IBA concentration evaluated [5000 ppm (0.5%)]. Cuttings from a mature (about sixteen-year-old) field grown specimen rooted poorly (≤ 21%) (Podaras and Bassuk, 1996). This suggests that loss of stock plant juvenility reduces the rooting success of stem cuttings.

Besides the ontogenic aging of stock plants, work by Chapman (1979) suggests that stem cuttings from many maple species only root successfully during short periods of the year. In that study, cuttings of amur maple, hedge maple, Norway maple, and red maple were collected every two weeks from late May to early July in Midland, Mich. Chapman reported that rooting peaked for cuttings of amur maple (86%), hedge maple (75%), and Norway maple (85%) collected on 26 June, 4 June, and 18 June, respectively. Rooting from each of the other cutting dates was less than ≤ 45% for these species. Red maple cuttings rooted well (60% to 90%) during a two week period (16 June to 26 June). Results from this study indicate that collecting cuttings at the appropriate developmental stage is critical. Variations in stock plant development could lead to starkly different results, not only between studies, but even on specific years of a single study.

Layering

For species which fail to root well from stem cuttings, many growers have found layering a suitable method of propagation. Many types of layering exist. Some, such as serpentine layering, require that stock plants have very flexible stems. Others, such as air layering, are well suited to stock plants with rigid stems but produce few daughter plants and are not practical for large scale propagation of most species. In the nursery industry, mound layering is one of the most popular layering techniques for propagating woody species.

Mound layering, also known as stooling, allows growers to propagate stock plants without using a greenhouse or intermittent mist system. In this technique, stock plants are initially planted in beds and allowed to become established for at least one year. At the beginning of the first production season, the grower cuts stock plants back to near ground level, leaving only the stumps. These stumps, or stools, produce a flush of shoots during the growing season. Depending on the grower's technique, a suitable rooting substrate such as soil, sawdust, or compost is piled over the stools before, as, or after the shoots emerge (Hartmann et al., 2011b). This substrate is kept moist during the remainder of the growing season as the shoots produce root systems at their bases. When the shoots go dormant in the fall, the rooting substrate is removed and rooted shoots are severed from the stock plants. Growers can handle these shoots just as they would handle bare-root liners (Hartmann et al., 2011b). A well-managed stool bed can remain productive for 15- to 20-yr (Hartmann et al., 2011b).

In many cases, wounding or girdling can improve the rooting success of mound layering. When the base of a shoot is wounded, the flow of solutes through the phloem is interrupted and photosynthates accumulate above the wound (Hartmann et al., 2011b). Girdling causes a similar response when the girth of shoots is restricted by wire, cable ties, or similar materials (Richards and Rupp, 2011; Rupp et al., 2013). Besides causing an accumulation of photosynthates, wounding and girdling also increase the concentration of hormones in the shoot (Hartmann et al., 2011b). These, combined with exogenously applied auxins, can improve the rooting response.

Mound layering has been used to propagate many hard to root species, including apple rootstocks (*Malus* spp. Mill.) (Howard, 1977), oaks (*Quercus bicolor* Willd. and *Q. macrocarpa* Michx.) (Amissah and Bassuk, 2005), and Chinese pistache (*Pistacia chinensis* Bunge) (Dunn and Cole, 1995). However, very few projects have investigated the response of maple (*Acer* L.) to mound layering.

Work by Rupp et al. (2013) dealt with rooting wild accessions of bigtooth maple [*A. saccharum* subsp. *grandidentatum* (Nutt.) Desmarais] in a three-year-old stooling bed. Girdling and auxin application [4000 ppm (0.4%) IBA + 2000 ppm (0.2%) NAA dissolved in 25% aqueous ethanol] both improved rooting when applied separately. However, best rooting (87%) occurred when stems were both girdled and treated with auxin. This work with a species closely related to caddo maple demonstrates the benefits of girdling and auxin application when rooting maples by mound layering.

Alsup (2001) conducted research dealing specifically with caddo maples. In that study, two-year-old seedlings were cut back to establish a stool bed. Treatments were applied when shoots were 12 cm (4.7 in) in height. Soil was immediately mounded to ≈9 cm (3.5 in) around shoots and more soil was added as the shoots elongated. Rooting only improved significantly when stems received both treatments: a longitudinal cut at the shoot base [8 mm (0.31 in) in length x 1 mm (0.04 in) deep] and 5000 ppm (0.5%) IBA in 70% isopropyl alcohol applied to wound. Stock plants in this study were small and a limited number of shoots were available. Alsup recommends that future mound layering studies of caddo maple use stock plant beds established from larger specimens.

The results from work by Rupp et al. (2013) and Alsup (2001) agree. Sugar maples (both bigtooth and caddo maples) root better when their phloem is interrupted at the shoot base, whether by girdling or wounding. Auxin application to the base of shoots further stimulates rooting. However, neither study sought to determine an optimal auxin concentration for rooting stimulation. Future work could likely improve rooting of both species by investigating their rooting responses to auxin concentration and determining the ideal severity of wounding or girdling.

In summary, preliminary work suggests mound layering holds great potential as an effective method of propagating maples. However, few maple species have been tested using this technique and detailed procedures have not been developed. A vegetative propagation alternative to stem cuttings and grafting would give growers more options for propagating maples and more opportunity to offer maple cultivars to the nursery industry.

Part II - Alternatives to Perlite in Vegetative Propagation Substrates

In recent years, economic and environmental concerns have spurred interest in alternative horticulture substrates. Researchers have identified several promising and practical alternative substrate components for nursery and greenhouse crop production. Among these, eastern redcedar [*Juniperus virginiana* (L.)] chips (ERC) deserve further investigation as a replacement for perlite in propagation substrates.

The Diversity of Alternative Substrates

Considering the large number of published studies related to this topic, it is no surprise that multiple alternatives have been recommended for each of the major components in nursery and greenhouse substrates. Alternatives to pine bark include clean chip residual from pine plantations (Boyer et al., 2008) and ERC hammer milled to pass a 9.5 mm (0.38 in) screen (Carmichael, 2013) or 20 mm (0.79 in) screen (Griffin, 2009). Peat moss alternatives investigated for containerized production include coconut coir (Lanzi et al., 2009; Noguera et al., 2000), cotton gin compost (Cole et al., 2005), and ERC hammer milled to pass a 4.8 mm (0.19 in) screen (Starr, 2011). Starr (2011) compared five substrates containing 0%, 25%, 50%, 75% and 100% ERC and 25% perlite (except the 100% ERC substrate). The remaining volume was filled with sphagnum peat moss. In that study, growth of petunia (*Petunia ×hybrida* Juss.), New Guinea impatiens (*Impatiens hawkeri* W. Bull.), and vinca [*Catharanthus roseus* (L.) G. Don] decreased as ERC content increased. Starr notes that adding ERC to substrates tends to increase their air space while reducing their container capacity and suggests that ERC may be better suited as a replacement for perlite.

Extensive work has investigated possible substitutes for perlite. The candidate materials can be generally classified into two groups: synthetic and organic. Most options are organic. In the synthetic category, ground automobile tires were evaluated as a perlite substitute, but they decreased growth of chrysanthemum (*Chrysanthemum morifolium* Ramat.) and produced high concentrations of zinc in the substrate (Bowman et al., 1994). Evans (2011) reports favorable results using ground waste glass treated at high temperatures to form growstones [Growstones, Earthstone Corp., Santa Fe, NM]. When growstones comprised more than 25% of substrates by volume, they provided greater air space and water holding capacity than equal volumes of perlite.

Organic alternatives to perlite include harvested wood based products and industrial byproducts. Murphy et al. (2011), evaluated hammer milled chips of sweet-gum (*Liquidambar styraciflua* L.), hickory (*Carya* sp. Nutt.), and eastern redcedar. They report that ERC concentrations as high as 25% for petunia (*Petunia ×hybrida* Juss. ‘Dreams Sky Blue’) and 50% for impatiens (*Impatiens walleriana* Hook. f. ‘Super Elfin Salmon’) and vinca [*Catharanthus roseus* (L.) G. Don ‘Cooler Peppermint’] did not reduce plant growth. ERC performed better than similar substrates containing sweet-gum or hickory. Similar work by Owen et al. (2013), demonstrated that pine wood chips are also a feasible alternative to perlite.

Several industrial byproducts have also been tested. Evans (2004) evaluated ground bovine bone as a perlite substitute. When blended with peat moss based substrates, ground bovine bone increased airspace, similarly to perlite, though the magnitude of the increase depended on how finely the bones had been ground. Evans does not recommend bovine bone as a perlite substitute because it leads to elevated pH, electrical conductivity, and ammonium in substrates. Parboiled fresh rice hulls, a byproduct of the rice industry, have become a popular substitute for perlite. Evans and Gachukia (2007) reported that parboiled fresh rice hulls increase air space and decrease container capacity more than equivalent amounts of perlite when incorporated as more than 20% (by volume) of a substrate. Another agricultural byproduct, processed corncob, has been successfully substituted for perlite. Weldon et al. (2011) reported that impatiens and petunias grew as well or better than controls in pine bark-based substrates containing 10%, 20%, or 30% processed corncob. Lastly, Vandiver et al. (2013) compared distilled ERC to rice hulls as a substrate component. Distilled ERC are a byproduct of cedar oil production (Vandiver et al., 2013). Both materials gave satisfactory results. Compared to rice hulls, distilled ERC provided more water holding capacity but less air space in substrates.

Alternative Substrates for Propagation

Despite the wide array of alternative substrate components available, most propagation studies have focused on evaluating substitutes for peat moss. Seed germination work and cutting propagation work have both been investigated.

Rose and Haase (2000) germinated seeds of douglas-fir [*Pseudotsuga menziesii* (Mirb.) Franco] in a substrate containing coconut coir. Seedlings which developed were nitrogen deficient, and the authors attribute this to the high C:N ratio of coconut coir. Bustamante et al. (2008) germinated seeds of four species of leafy vegetables or herbs in substrates containing distillery waste (exhausted grape marc) and poultry or cattle manure. Seeds germinated well and without signs of nutrient deficiency.

Several studies have evaluated stem cutting propagation in substrates containing peat moss alternatives. Chong (1999) investigated composted municipal solid waste (leaf and yard waste) as a substitute for perlite and peat moss. The author reports that municipal solid waste functions best as a peat moss substitute in rooting substrates. In another study, stem cuttings of foliage plants were rooted in substrates containing cowpeat (composted dairy manure) (Li et al., 2009) All cuttings produced similar growth in all treatments except heartleaf philodendron [*Philodendron scandens* subsp. *oxycardium* (Schott) G.S. Bunting] which developed a smaller root system in substrates containing high proportions of cowpeat. The high nutrient level of cowpeat substrates likely explains this observation (Li et al., 2009). Buck and Evans (2010) analyzed substrates containing hammer milled parboiled rice hulls. Though no plants were grown, the authors report that physical properties measured from substrates were all within recommended ranges.

Starr (2011) studied the potential of ERC as a substitute for peat moss. ERC were hammer milled to pass a 4.8 mm (0.19 in) screen. Herbaceous cuttings were rooted in substrates containing 0%, 25%, 50%, 75%, and 100% ERC. The remaining volume of each substrate was perlite. ERC content did not affect rooting of any of the species in the study. However, cuttings in all substrates, including the 100% perlite, rooted poorly. The author attributed this poor rooting to a suboptimal propagation environment.

Working with woody ornamentals, Brock et al. (2012) also investigated ERC as a substitute for peat moss in propagation substrates. Beginning with a 100% perlite substrate, four additional substrates were prepared by replacing 25%, 50%, 75%, and 100% of the perlite with ERC hammer milled to pass a 4.8 mm (0.19 in) screen. A standard propagation substrate of 75% perlite and 25% peat moss was also included for comparison. Stem cuttings of ‘Green Giant’ arborvitae (*Thuja* L. ×‘Green Giant’) rooted as well (91%) as controls at 25% ERC, but rooting was reduced at greater ERC contents. Stem cuttings from lacebark elm (*Ulmus parvifolia* Jacq. ‘Emerald Prairie’) showed a strong negative relationship between rooting percent and increasing ERC content. However, ERC content did not affect mean root number or root dry weights of either species. The results of this experiment support the suggestion made by Starr (2011). ERC hammer milled to pass a 4.8 mm (0.19 in) screen may be better suited as a replacement for perlite than as a replacement for peat moss.

Although not directly studying perlite substitutes, Bilderback and Lorscheider (1995) successfully propagated anise (*Illicium parvifloram* Michx. ex Vent.) in a non-perlite substrate of 100% double processed pine bark. Cuttings rooted as well in the double processed pine bark as they did in other commercial rooting substrates, including a perlite-based substrate.

Another alternative to perlite-based rooting substrates is sand. In the past sand was used frequently as in propagation. However, sand is not well suited to many of the needs of commercial propagators today. If stem cuttings were rooted in cells of liner trays filled with sand, their root systems would not maintain integrity during transplant. Furthermore, the high bulk density of sand would increase shipping costs. With these considerations, perlite alternatives from renewable or byproduct sources are preferred.

The Need for Perlite Alternatives

The search for perlite substitutes is not driven by poor performance of perlite in rooting substrates. Growers have learned to depend on the excellent aerating properties of perlite and commonly include it as 30% to 100% of their rooting substrates (Moore, 1987). The motivation to find perlite substitutes is based on concerns about limited resources, rising energy costs, and the dusty nature of perlite in its dry state.

The precursor to perlite is an amorphous silicate obtained by mining (Moore, 1987). Perlite mines are a non-renewable resource (Owen et al., 2013). Finding alternatives to perlite would reduce the rate at which these resources are consumed.

As energy costs continue to rise, perlite costs have also risen. Producing perlite is an energy intensive process. The raw amorphous silicate particles are heated at 593 to 871 C (1100 to 1600 F), which both sterilizes the particles and causes the small amount of entrapped water within them to become steam, forcing the particles to expand (Hartmann et al., 2011a; Moore, 1987). Rising production costs and shipping costs are increasing perlite prices for growers (Evans, 2011; Owen et al., 2013; Vandiver et al., 2013).

Lastly, growers would like to find an alternative to perlite which is cleaner and safer. When handled in its dry state, perlite releases large amounts of dust that is an eye and lung irritant (Evans, 2011; Murphy et al., 2011; Weldon et al., 2011). The recommended exposure limit to perlite dust is $5\ \mathrm{mg{\cdot}m^{-3}}$ (5 ppb) (OSHA, 2014). Although perlite is not frequently associated with serious health issues, some studies have reported perlite related cases of reactive airway dysfunction (Du et al., 2010) and a reduced transfer factor of carbon monoxide in the lung (Polatli et al., 2001).

The concerns discussed above are important to keep in mind as potential perlite substitutes are considered. Ideally, new substrate components should resolve one or more of the concerns associated with perlite without causing other major concerns.

Eastern Redcedar as a Substitute for Perlite

Eastern redcedar resolves most of the concerns associated with perlite. Native to the Great Plains and the eastern half of the U.S. (Hardin et al., 2001), eastern redcedar is both a renewable resource and locally available to many growers. Although ornamental cultivars of eastern redcedar are commonly used in landscapes, trees growing in grasslands are considered weeds. Because birds spread the seeds of eastern redcedar, the trees quickly spread to new areas where they rapidly convert grasslands to forest in as little as forty years (Briggs et al., 2002). Historically, fire has held back the encroachment of eastern redcedar into grasslands. However, increased grazing intensity in recent years has hindered thorough grassland burning (Hoch, 2000). As the number of trees per acre increases, biomass productivity and species diversity decrease. Briggs et al. (2002) reported a 99% loss in biomass productivity and a severe loss of species diversity in closed canopy eastern redcedar forests. Hoch (2000) found that the eastern redcedar population in counties of Kansas correlated to the human population in those counties. As human population increased, fire frequency decreased, and eastern redcedar population increased. In one study in the Flint Hills region of Kansas, closed canopy forest cover increased 120% between 1986 and 2000 (Hoch, 2000).

In situations where grassfires are not a desirable option or trees are too large to remove by burning, eastern redcedar trees are often cut down, piled, and occasionally burned. These “trash trees” can also be chipped for use as landscape mulch (Carmichael, 2013). Processing this mulch through a hammer mill yields ERC, a potential component of horticulture substrates (Carmichael, 2013; Murphy et al., 2011; Starr, 2011). The size of the openings in the hammer mill screen should be selected based on the intended use of the substrate. To prepare a pine bark substitute, Carmichael (2013) hammer milled eastern redcedar to pass a 9.5 mm (0.38 in) screen. When preparing a perlite substitute for container production, Murphy et al. (2011) hammer milled eastern redcedar to pass a 6.4 mm (0.25 in) screen. Adjusting the screen size influences physical properties of the substrate.

Starr (2011) and Vandiver et al. (2013) both suggest that ERC may be suitable as an alternative to perlite. Murphy et al. (2011) demonstrated ERC can indeed replace perlite without reducing growth of bedding plant species in a substrate used for greenhouse production. Because rooting substrates often contain a large proportion of perlite, further research should be conducted to investigate the suitability of ERC as a perlite alternative in cutting propagation. This research could allow growers to obtain more of their substrate materials from local, sustainable sources.

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Chapter 2 - Rooting Response of Stem Cuttings of Shantung Maple *(Acer truncatum*) to Time of Year, Cutting Position, and Auxin Concentration, Formulation, and Solvent

Introduction

Several characteristics of shantung maple (*Acer truncatum* Bunge) suit it for landscapes in midwestern states. This drought tolerant species fits well into urban landscapes reaching 6 to 8 m (20 to 25 ft) in height (Dirr, 2009). Trees have new growth tinged purple and fall colors ranging from yellow to red to purple. Although the species sprouts easily from seed (Ackerman, 1957; Browse, 1990; and Pair, 1986), stem cuttings fail to consistently root in high numbers. Currently, growers propagate the few existing cultivars by more labor intensive grafting techniques. An efficient method of propagating shantung maples by stem cuttings could improve the availability and popularity of this promising species.

Previous attempts to propagate shantung maple by stem cuttings have produced varied results. In a casual study of maple propagation, Vertrees (1978) recorded that cultivars of *A. truncatum* collected during late June in Oregon failed to root when treated with 8000 ppm (0.8%) indole-3-butyric acid (IBA) in talc powder (IBA/talc). He suggests that a 1000 ppm (0.1%) IBA quick-dip be used and notes that collecting cuttings at a proper developmental stage is critical. Podaras and Bassuk (1996) treated softwood stem cuttings from a 4-year-old greenhouse-grown shantung maple with a 20 s dip in solutions of 0; 1,000; 5,000; and 10,000 ppm (0, 0.1, 0.5, and 1.0%) IBA dissolved in 1 ethanol: 1 water (v/v). They obtained best rooting (51% to 56%) at 0 & 1000 ppm (0% & 0.1%) IBA. Etiolating and banding cuttings before harvest increased rooting to 88% with 5000 ppm (0.5%) IBA treatment. Softwood cuttings collected from a mature field specimen rooted poorly (≤ 21%).

Pair (1986) reported good rooting of semi-hardwood shoot tip cuttings from 3-year-old shantung maple seedlings [75% rooting after a quick (5 s) dip in 1000 ppm (0.1%) IBA; solvent not specified]. Rooting of subterminal cuttings (the cutting immediately proximal to the shoot tip cutting) reached 85% at 5000 ppm (0.5%). This data suggests that stock plant maturity plays an important role in rooting of stem cuttings. Unfortunately, this experiment lacked sufficient repetition to provide great confidence in the results.

The objective of this study was to build upon Pair's 1986 work using a statistically stronger experimental design to investigate the role of stock plant growth stage, cutting position, and IBA concentration, formulation, and solvent on rooting of stem cuttings of shantung maple.

Materials and Methods

Procedures were similar each year. Therefore, they will be discussed together.

Stock plants

Stock plants were 19 mature shantung maple specimens of seedling origin (non-clonal) ≈20 years old, growing at the Kansas State University John C. Pair Horticulture Center near Haysville, Kans. Ten of the trees were topped at 1.5 m (4.9 ft) to encourage epicormic shoot production in Spring 2010. During 2011 and 2012, the trees received supplemental water due to unusually severe drought. In Spring 2013, nine more trees were topped at 1.5 m (4.9 ft) to increase available cutting material. All cuttings in this study were collected from the original ten stock plants except the second-flush softwood cuttings of 2013, which were collected from the nine new stock plants of 2013. All trees received 14 g (0.5 oz) of nitrogen (N) from urea (46N-0P-0K) applied in April the year cuttings were to be taken.

Cuttings

For each experiment in this study, cuttings were categorized as softwood, semi-hardwood, second-flush softwood, or hardwood. Softwood cuttings were collected from actively growing shoots before leaves at the shoot apex fully expanded and stems lignified. At this stage, leaves and shoot tips were colored red to purple. Semi-hardwood cuttings were collected after shoot elongation ceased, leaves near the shoot apex reached their full size, and stems were lignified. At this stage, stems turned from green to brown and little of the red to purple color remained in the leaves. Second-flush softwood cuttings were identical to softwood cuttings except they were collected from the flush of growth which occurred in late summer. Hardwood cuttings where collected in late winter from dormant shoots. All cuttings were collected from the most recent season growth.

During mornings when plants were well hydrated, terminal stem cuttings ≈20 cm (8 in) in length were harvested except for the second-flush cuttings of 2013 when ≈30 cm (12 in) stem cuttings were collected. Cuttings were placed in plastic bags and stored in a cooling room at 10 C (50 F) if not immediately processed. Each cutting was trimmed to 15 cm (6 in) from the terminal bud. In addition to the second-flush softwood terminal cuttings of 2013, subterminal cuttings were also collected. These 15 cm (6 in) subterminal cuttings were taken directly proximal to the terminal cutting beginning with the next node. Leaves were stripped from the basal half of each cutting before treatments were applied. Cuttings were lightly misted with water occasionally during processing to prevent desiccation.

Cuttings were thoroughly mixed before applying treatments to blend any genetic variation of stock plant propensity for rooting. However, in the second-flush softwood cuttings of 2013, terminal stem cuttings from each stock plant were assigned to a specific block to ensure genetic uniformity within each block.

Cuttings for all experiments were inserted to a depth of 5 cm (2.0 in) in 38-cell trays with individual cells [6.4 cm dia. x 12.7 cm deep (2.5 in dia. x 5 in deep)] (X-38ST, Landmark Plastic, Akron, Ohio) filled with moist 3 perlite (Sun Gro Horticulture, Agawam, Mass.): 1 sphagnum peat moss (Ferti-lome, Bonham, Texas) (v/v) substrate. Trays of cuttings received intermittent mist during natural daylight hours in a polycarbonate greenhouse covered with 63% shadecloth (WS63, DeWitt Co., Sikeston, Mo.). Mist on softwood and semi-hardwood cuttings operated for 6 s every 7 min, whereas mist on hardwood cuttings operated for 10 s every 30 min. No supplemental light was provided. Temperature in the greenhouse was set to 21/16 C (70/60 F) (day/night) and controlled using an evaporative cooling system.

Treatments

Treatments in this study consisted of IBA concentrations of 0; 2,500; 5,000; 10,000; and 15,000 ppm (0%, 0.25%, 0.5%, 1.0%, and 1.5%), IBA formulation [liquid or talc powder (Rhizopon AA #1, #2, and #3; Rhizopon B.V.; Hazerswoude-Rijndijk, Netherlands], IBA solvent [water or 1 ethanol: 1 water (v/v)], and cutting position (terminal or subterminal). The potassium (K) salt of indole-3-butyric acid (K-IBA) (Sigma-Aldrich, St. Louis, Mo.) was dissolved in reverse osmosis water (treatment K-IBA/H_2O) whereas the free acid of IBA (≥ 99.0%,Sigma-Aldrich, St. Louis, Mo.) was dissolved in aqueous ethanol (treatment IBA/EtOH).

Liquid IBA treatments were applied by dipping the basal 1 cm (0.4 in) of cuttings in the treatment solution for 5 s. Treated cuttings were allowed to air dry for 5 min to allow the auxin to adhere to the stem tissue. Powder formulations were applied by dipping basal 1 cm (0.4 in) of cutting in powder and then gently tapping cutting to remove excess talc.

Experiment 1-3: To determine the influence of stock plant growth stage, softwood (16 June 2011), semi-hardwood (5 Aug. 2011), and hardwood (20 Feb. 2012) cuttings were treated with K-IBA at 0; 2,500; 5,000; or 10,000 ppm (0%, 0.25%, 0.5%, or 1.0%) in water. The experimental design was a randomized complete block design with 5 cuttings (subsamples) per K-IBA treatment and 8 replications. Cuttings were harvested and data collected after 18 weeks (softwood), 11 weeks (semi-hardwood), and 12 weeks (hardwood). Data included percent rooting, root number and root length. For all experiments a cutting was considered rooted if it had one primary root greater than 0.2 cm (0.08 in) in length (to distinguish roots from callus).

Experiment 4 & 5: To determine the influence of liquid or talc-powder IBA application method, softwood (7 June 2012) and semi-hardwood (27 July 2012) cuttings were treated with 0; 2,500; 5,000; or 10,000 ppm (0%, 0.25%, 0.5%, or 1.0%) K-IBA/H_2O or 1000, 3000, or 8000 ppm (0.1%, 0.3%, or 0.8%) IBA/talc. The experimental design was a randomized complete block design with 5 cuttings (subsamples) per treatment and 7 replications. Cuttings were harvested and data collected after 20 weeks (softwood) and 16 weeks (semi-hardwood). Data included percent rooting, root number and root length.

Experiment 6: To investigate the influence of water or ethanol as a solvent, semi-hardwood cuttings (26 June 2013) were treated with 0; 2,500; 5,000; 10,000; or 15,000 ppm (0%, 0.25%, 0.5%, 1.0%, or 1.5%) K-IBA/H_2O or IBA/EtOH. The experimental design was a randomized complete block design with a factorial arrangement of treatments. The factorial arrangement consisted of 5 auxin concentrations and 2 solvents. Each treatment contained 5 cuttings (subsamples) and was replicated 8 times except 15,000 ppm (1.5%) IBA/EtOH which had only 5 replications in a single flat due to a shortage of stock material. Cuttings received 4 preventative fungicide soil drench treatments at 1 week intervals from 27 June to 1 Aug. using a rotation of mefenoxam [active ingredient at 2.6 $mL\cdot L^{-1}$ (0.33 fl $oz\cdot gal^{-1}$) (Mefenoxam 2AQ, Quali-Pro, Pasadena, Texas)], thiophanate methyl [active ingredient at 304 $mg\cdot L^{-1}$ (0.04 $oz\cdot gal^{-1}$) (3336WP, Cleary Chemicals LLC, Dayton, N.J.)], and azoxystrobin [active ingridient at 22.5 $mg\cdot L^{-1}$ (0.003 $oz\cdot gal^{-1}$) (Heritage Fungicide, Syngenta Group Company, Greensboro, N.C.)]. Cuttings were harvested and data collected after 16 weeks. Data included percent rooting, root number and root length.

Experiment 7: To determine the effect of cutting position on rooting, experiment 6 was repeated with second-flush softwood (14 Aug. 2013) shoot tip cuttings and subterminal cuttings. The experimental design was a randomized complete block design with a three-way factorial arrangement of treatments (5 IBA concentration x 2 solvent x 2 cutting position). There were 5 cuttings (subsamples) per treatment and 8 replications. In this experiment each replication represented a different stock plant (genotype) except for the subterminal cuttings, which were combined among genotypes to create 4 replications due to a lack of stock plant material. Cuttings were harvested and data collected after 11 weeks. Data included percent rooting, root number and root length.

Statistical Analysis

Data were subjected to analysis of variance using the general linear models (GLM) procedure of SAS (Statistical Analysis System, Version 9.2, SAS Institute Inc., Cary, N.C.). Data for one block of the 2013 semi-hardwood cuttings was removed from the dataset because it was located on the sunniest, driest corner of the mist bench and was an outlier. Where appropriate, data were also subjected to regression analysis.

Results

Experiments 1-3

K-IBA/H_2O did not affect rooting percent, root number, or root length of stem cuttings at any growth stage (Table 2.1). Rooting averaged 8.8% in softwood cuttings and 44.4% in semi-hardwood cuttings. Hardwood cuttings did not root (0%). Average root number per rooted cutting was 1.5 and 2.4 in softwood and semi-hardwood cuttings, respectively. Root length averaged 13.0 cm (5.1 in) in softwood cuttings and 8.0 cm (3.1 in) in semi-hardwood cuttings.

Experiments 4 & 5

Neither K-IBA/H_2O nor IBA/talc influenced rooting percent, root number, or average root length of softwood or semi-hardwood stem cuttings (Table 2.2). Softwood cuttings did not root (0%). Mean rooting of semi-hardwood cuttings was 14.3% and 12.4% for K-IBA/H_2O and IBA/talc, respectively. Root number averaged 1.4 and 1.8 for K-IBA/H_2O and IBA/talc, respectively. Root length averaged 11.1 cm (4.4 in) and 10.3 cm (4.1 in) for K-IBA and IBA/talc, respectively. IBA formulation did not cause any significant differences in rooting, root number, or root length.

Experiment 6

Semi-hardwood cuttings had a clear rooting response to IBA concentration (Table 2.3). Rooting responded in a negative linear relationship to increasing IBA concentration regardless of the solvent (K-IBA/H_2O or IBA/EtOH). Mean root number and average length were unaffected by IBA concentration. Furthermore, solvent type (H_2O or EtOH) did not influence rooting, root number, or average root length. There was no significant interaction between IBA concentration and solvent.

Experiment 7

The main effects of IBA concentration, IBA solvent, and cutting position did not influence rooting percent, root number, or average root length from second-flush softwood cuttings (Table 2.4). Rooting of K-IBA/H_2O treatments averaged 5% and 1.6% for terminal and subterminal cuttings, respectively. Rooting of IBA/EtOH treatments averaged 10% and 7.2% for terminal and subterminal cuttings, respectively. Overall, shoot tip cuttings rooted at 7.5% whereas subterminal cuttings rooted at 4.8%. No interaction was observed between IBA concentration, solvent, or cutting position. Overall root number per rooted cutting was 2.0 with an average root length of 6.6 cm (2.6 in).

Discussion

Timing

Results from experiments 1 to 5 suggest that maximum rooting potential for terminal stem cuttings occurred during the semi-hardwood stage of shoot development (Tables 2.1 and 2.2). The results also suggest considerable variability from year to year with semi-hardwood cuttings rooting 44% in 2011, but only 14% in 2012. Similar to the current study, Pair (1986) found that semi-hardwood stem cuttings of shantung maple rooted better than softwood cuttings. Vertrees (1978) attributes failed rooting of shantung maple cultivars to poor timing of cutting harvest. He notes that tender cuttings and excessively mature cuttings tend to root poorly. Chapman and Hoover (1981) reaffirm this conclusion with their study of rooting response in hedge maple (*A. campestre* L.), Norway maple (*A. platanoides* L.), and red maple (*A. rubrum* L.). They state that stem cuttings of these species are prone to rot when collected before rapid shoot elongation has finished. Collecting cuttings too late is also a possibility. Chapman (1979) collected stem cuttings of hedge maple and Norway maple at 2-week intervals from late spring to early summer. He found that rooting percentage briefly peaked on 4 June and 18 June for hedge maple (75%) and Norway maple (85%), respectively. Stem cuttings rooted poorly (≤40%) before and after peak dates. This emphasizes the importance of propagating maples during their ideal developmental stage. Working with sugar maples (*A. saccharum* Marsh.), Tousignant et al. (2003) found that water content measurements and terminal bud scale counts accurately predicted optimum periods for stem cutting propagation. They determined that peak rooting generally correlated to 270 growing degree days above 5 C (41 F). Future studies may improve rooting success of shantung stem cuttings by relating specific shoot developmental stages or growing degree days to rooting responses. Results from the current study suggest that optimal rooting occurs near the time when shoots have finished elongating, leaves near the apex have fully expanded, and stem tissue has lignified.

IBA Concentration

Of the seven sets of cuttings observed in this study, only the semi-hardwood cuttings of 2013 (Table 2.3) showed a significant response to IBA concentration. After poor rooting in experiments 4 and 5, a high IBA concentration treatment [15,000 ppm (1.5%)] was included in experiments 6 and 7 to eliminate any question of underdosing. Rooting of semi-hardwood cuttings in 2013 displayed a strong negative response to increasing IBA concentration. This suggests that rooting was not hindered by a lack of IBA. Second-flush softwood cuttings of 2013 (Table 2.4) do not show any significant relationship between rooting, root number, and root length, but overall results are low, likely due to collecting cuttings when they were too tender.

Condition of stock plants in this study best explains the varied responses of stem cuttings to IBA concentration. In 2011, when semi-hardwood cuttings rooted uniformly at all IBA concentrations stock plants had recently been pruned back to 1.5 m (4.9 ft) to encourage epicormic shoot formation. In 2012, differences in semi-hardwood rooting were nonsignificant, but rooting percentages appeared to favor low IBA concentrations. In 2013, low IBA concentrations caused significantly better rooting than high IBA concentrations. Thus, during the three seasons of this study, semi-hardwood stem cuttings increased in sensitivity to IBA concentration. During this time, stock plants were annually hedged (pruned to previous year pruning wound) to supply cutting material, which may have rejuvenated stock plants. If sensitivity to high IBA concentration is a trait of juvenile plants, previous studies with juvenile shantung stock plants should show a similar trend. Although Pair (1986) did not observe this trend, his study contained few repetitions and statistical analysis is not provided. Better support for this hypothesis is provided in work by Podaras and Bassuk (1996) where a 4-year-old greenhouse-grown shantung maple shows higher sensitivity to IBA concentration than a 16-year-old field grown specimen. In that study, the 4-year-old plant rooted best at low IBA concentration [56% rooting at 1000 ppm (0.1%) IBA dissolved in 1 ethanol: 1 water (v/v)], while the 16-yr-old tree showed a weak preference for higher IBA concentration [21% rooting at

5000 ppm (0.5%) IBA]. This supports the hypothesis that stem cuttings from younger shantung maples respond better to low IBA concentrations than stem cuttings of mature trees. Further research is needed to clearly determine the influence of ontogenic aging on the response of shantung maple stem cuttings to IBA concentration. The study by Podaras and Bassuk (1996) also suggests that low concentrations of IBA may encourage rooting of shantung maple cuttings. Because the lowest IBA concentration applied in the current study was 2500 ppm (0.25%), any stimulatory effect of lower IBA concentrations would not have been observed. A study with smaller differences in treatment IBA concentration in the 0 to 2500 ppm (0% to 0.25%) range may produce better rooting results.

Interestingly, root number and average root length of semi-hardwood cuttings of 2013 show no response to IBA concentration. Davies and Haissig (1990) note that roots arise from either preformed or induced root primordia. Fink (1982) states that species of maple (*Acer* L.) do have preformed root primordia. In the current study, IBA neither hindered preformed primordia development nor induced additional primordia development as demonstrated by the consistent root number. Uniform root length indicates treatments rooted simultaneously. Similar to work by Chong (1981), high IBA concentrations did not hinder root elongation.

Formulation

Similar rooting between cuttings treated with liquid IBA formulations and those treated with powder IBA formulations was unexpected. Normally, liquid IBA formulations provide better rooting than powder formulations (Hartmann et al., 2011b). Cuttings likely absorb less IBA from powder formulations than from liquid formulations, thus leading to the frequent difference in rooting success. However, because IBA concentration did not affect rooting in semi-hardwood stem cuttings of 2012, formulation differences did not affect rooting either.

Solvent

Solutions containing ethanol can influence rooting by improving uptake of IBA in stem cuttings (Heung and McGuire, 1973), or by damaging tender tissues (Hartmann et al., 2011b). In this study, differences in rooting, root number, and root length were not related to solvent. Further work could investigate an increase in the concentration of ethanol or include other solvents such as dimethyl sulfoxide (DMSO) and polyethylene glycol (PEG) which improved root number and length in work by Dirr (1989)

Cutting Position

Subterminal cuttings were included in this study because results from Pair's study (1986) suggested that subterminal cuttings root slightly better (69%) than terminal cuttings (55%). This observation is supported by Hartmann et al. (2011a) who stated that cuttings taken from lower sections of shoots root better in some species. Haissig (1972) noted that in brittle willow (*Salix fragilis* L.) root primordia develop in subterminal nodes as shoots elongate. This should cause better rooting in subterminal cuttings. In the current study, terminal and subterminal cuttings showed no differences in rooting. Either subterminal cuttings of shantung maple have no rooting advantage over terminal cuttings, or the maximum rooting potential of subterminal cuttings was reduced in this study by the tender condition of subterminal cuttings which were only investigated as second-flush softwood cuttings. Either way, subterminal cuttings root at least as well as terminal cuttings. Using both terminal and subterminal cutting material for propagation could speed the establishment of clonal populations of future shantung maple selections.

Conclusion

Rooting efficiency from mature shantung maples remains low. Successful propagation depends primarily on collecting stem cuttings at their optimum developmental stage, which occurs near the time when shoots lignify and leaves at the shoot apex reach their mature size. This study suggests that treating cuttings with IBA concentrations ≥ 2500 ppm (0.25%) will not improve rooting and may decrease rooting.

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Table 2.1

Percent rooting, mean root number, and mean root length of softwood (16 June 2011), semi-hardwood (5 Aug. 2011), and hardwood (20 Feb. 2012) stem cuttings of *Acer truncatum* treated with the potassium (K) salt of indole-3-butyric acid (IBA) dissolved in water (H_2O).

	Softwood			Semi-hardwood			Hardwood		
K-IBA/H_2O (ppm)	Rooting (%)	Root No.	Root Length (cm)	Rooting (%)	Root No.	Root Length (cm)	Rooting (%)	Root No.	Root Length (cm)
0	12.5	2.1	14.3	42.5	2.4	7.0	0.0	-	-
2,500	0.0	-	-	30.0	2.0	7.9	0.0	-	-
5,000	10.0	1.3	9.7	55.0	3.0	8.3	0.0	-	-
10,000	12.5	1.2	14.9	50.0	2.3	8.6	0.0	-	-
Significance	NS[y]	NS	NS	NS	NS	NS	NS	NS	NS
Column Mean[z]	8.8	1.5	13.0	44.4	2.4	8.0	0.0	-	-

[z] Mean across all K-IBA concentrations

[y] NS = nonsignificant at $P \leq 0.05$

n=40 stem cuttings per treatment

Table 2.2

Percent rooting, mean root number, and mean root length of softwood (7 June 2012), and semi-hardwood (27 July 2012) stem cuttings of *Acer truncatum* treated with either the potassium (K) salt of indole-3-butyric acid (IBA) dissolved in water (H_2O) or IBA suspended in talc.

	Softwood			Semi-hardwood		
K-IBA/H_2O (ppm)	Rooting (%)	Root No.	Root Length (cm)	Rooting (%)	Root No.	Root Length (cm)
0	0.0	-	-	22.9	1.9	8.9
2,500	0.0	-	-	22.9	1.7	10.1
5,000	0.0	-	-	8.6	1.0	13.7
10,000	0.0	-	-	2.9	1.0	11.5
Significance	NS[y]	-	-	NS	NS	NS
Column Mean[z]	0.0	-	-	14.3	1.4	11.1
IBA/talc (ppm)						
1000	0.0	-	-	17.1	2.1	10.6
3000	0.0	-	-	8.6	1.8	8.8
8000	0.0	-	-	11.4	1.5	11.6
Significance	NS	-	-	NS	NS	NS
Column Mean	0.0	-	-	12.4	1.8	10.3

[z] Mean across all K-IBA concentrations

[y] NS = nonsignificant at $P \leq 0.05$

n=35 stem cuttings per treatment

Table 2.3

Percent rooting, mean root number, and mean root length of semi-hardwood (26 June 2013) stem cuttings of *Acer truncatum* treated with either the potassium (K) salt of indole-3-butyric acid (IBA) dissolved in water (H_2O) or the free acid of IBA dissolved in 1 ethanol (EtOH): 1 water (v/v).

	Rooting (%)		Root No.		Root Length (cm)	
IBA (ppm)	K-IBA/H_2O	IBA/EtOH	K-IBA/H_2O	IBA/EtOH	K-IBA/H_2O	IBA/EtOH
0	37.1	48.6	1.4	3.1	7.3	7.6
2,500	25.7	25.7	2.2	3.7	7.3	7.6
5,000	20.0	28.6	1.3	1.6	7.0	7.2
10,000	14.3	14.3	1.1	1.9	8.5	5.7
15,000	2.9	10.0	2.0	1.0	3.0	11.5
Linear	**[z]	*	NS	NS	NS	NS

[z] Nonsignificant (NS) at $P \leq 0.05$, (*) Significant at $P \leq 0.05$, or (**) Significant at $P \leq 0.01$

n=35 stem cuttings per treatment except 15,000 ppm (1.5%) IBA/EtOH where n=25 stem cuttings.

Table 2.4

Percent rooting, mean root number, and mean root length of second-flush softwood (14 Aug. 2013) terminal and subterminal stem cuttings of *Acer truncatum* treated with either the potassium (K) salt of indole-3-butyric acid (IBA) dissolved in water (H_2O) or the free acid of IBA dissolved in 1 ethanol (EtOH): 1 water (v/v).

K-IBA/H_2O (ppm)	Rooting (%)		Root No.		Root Length (cm)	
	Terminal	Subterminal	Terminal	Subterminal	Terminal	Subterminal
0	7.5	4.0	1.0	2.0	9.5	7.6
2,500	5.0	0.0	1.5	-	3.4	-
5,000	5.0	4.0	4.0	1.0	5.1	4.0
10,000	2.5	4.0	1.0	3.0	6.9	4.3
15,000	5.0	0.0	2.5	-	7.6	-
Significance	NS[z]	NS	NS	NS	NS	NS
IBA/EtOH (ppm)	Terminal	Subterminal	Terminal	Subterminal	Terminal	Subterminal
0	12.5	12.0	1.7	2.8	5.4	7.9
2,500	15.0	8.0	2.4	1.5	8.3	12.1
5,000	15.0	4.0	2.3	1.0	3.2	5.5
10,000	5.0	8.0	2.0	1.5	3.3	13.8
15,000	2.5	4.0	3.0	1.0	6.7	3.3
Significance	NS	NS	NS	NS	NS	NS

[z] NS = nonsignificant at $P \leq 0.05$

n= 40 stem cuttings per terminal treatment; n=20 stem cuttings per subterminal treatment

Chapter 3 - Auxin Concentration Affects Adventitious Rooting of Mound Layered Caddo Sugar Maple (*Acer saccharum*) and Shantung Maple (*Acer truncatum*)

Introduction

The ornamental landscape industry seeks plants with attractive, dependable growth characteristics. Caddo sugar maple (*Acer saccharum* Marsh. subsp. *saccharum*) (caddo maple) and shantung maple (*A. truncatum* Bunge.) both offer desirable traits. Caddo maple originated from a disjunct population of sugar maples native to central Oklahoma. Trees are known for their tolerance to drought, heat, wind, and alkaline soil (Griffin, 2014; Pair, 1994b; Simpson and Hipp, 1993). Shantung maples are native to northern China, Russia, Japan, and Korea (Dirr, 2009; Pair, 1986). They also perform well in hot, dry environments and possess excellent disease resistance (Griffin, 2014; Pair, 1986; Pair et al., 1996). Maturing at 6 to 8 m (20 to 25 ft) (Dirr, 2009), shantung maples fit nicely into urban landscapes.

Both species are relatively difficult to propagate asexually, which hinders the introduction and availability of improved cultivars. Seed propagation is possible (Ackerman, 1957; Hartmann et al., 2011; Pair, 1986), but not useful for maintaining unique genotypes. Growers may propagate cultivars by winter side-veneer grafting or summer T-budding (Le Duc and Pair, 2000; Pair, 1994a; Pair et al., 1996; Vertrees, 1978), but these methods are labor intensive. Furthermore, nurseries sometimes graft caddo maples onto other more readily available sugar maple rootstocks. This may compromise the drought tolerant characteristics of this species (Le Duc and Pair, 2000).

Alternatives to grafting include propagation by vegetative cuttings and layering of stock plants. Vegetative cuttings produce mixed results. For shantung maples, Podaras and Bassuk (1996) report rooting percentages as high as 88% from softwood stem cuttings that were banded, etiolated, and treated with 5000 ppm (0.5%) indole-3-butyric acid (IBA). Without banding or etiolating, Pair (1986) achieved 62% overall rooting of semi-hardwood stem cuttings treated with 0 to 5000 ppm (0% to 0.5%) IBA. In recent work using a range of cutting dates, IBA concentrations, and solvents, stem cuttings of shantung failed to root greater than 55% (Brock, 2014).

Caddo maple also roots poorly from cuttings. Working with sugar maples, Morsink (1971) reports 80% to 89% rooting of softwood stump sprouts collected as 35 to 65 cm (14 to 26 in) cuttings without IBA application. However, when Alsup (2001) worked specifically with caddo maples, only 30% of softwood stem cuttings rooted in treatments ranging from 5000 to 15,000 ppm (0.5% to 1.5%) IBA.

When a species roots poorly from cuttings, growers may choose to propagate stock plants by layering. Certain species of Apples (*Malus* Mill.) (Howard, 1977), oaks [*Quercus bicolor* Willd. (swamp white oak) and *Q. macrocarpa* Michx. (bur oak)] (Amissah and Bassuk, 2005), and Chinese pistache (*Pistacia chinensis* Bunge) (Dunn and Cole, 1995) respond favorably to mound layering. Rupp et al. (2013) successfully used mound layering to root shoots of bigtooth maple [*A. saccharum* subsp. *grandidentatum* (Nutt.) Desmarais]. Applying 4000 ppm (0.4%) IBA with 2000 ppm (0.2%) 1-naphthalene acetic acid (NAA) dissolved in 1 ethanol (EtOH): 4 water, they observed 16% rooting. Girdling the base of stems combined with the IBA and NAA treatment increased rooting to 87%.

Alsup (2001) attempted to propagate caddo maple by mound layering shoots from 2-year-old seedlings. In that study the author attempted to improve root production using wounding and IBA application. Rooting was improved when shoots received both treatments: longitudinal cut at the shoot base [8 mm (0.31 in) in length by 1 (0.04 in) mm deep] and 5000 ppm (0.5%) IBA in 70% isopropyl alcohol applied to wound. Alsup reported that wounding and IBA application increased rooting and suggested that future studies establish stool beds from larger stock plants.

The objective of the current study was to determine the optimal IBA concentrations for rooting mound layered caddo maple and shantung maple shoots.

Materials and Methods

Mound layering procedures were similar in 2012 and 2013. They are described together here, except where substantial differences require specific explanation.

Stock Plants

Stock plants for this project were 15 to 20-year-old field grown caddo and shantung maples of seedling origin (non-clonal) growing in a Canadian-Waldeck fine sandy loam soil at the Kansas State University John C. Pair Horticulture Center near Haysville, Kans. Stock plants were cut off at 10 to 15 cm (4 to 6 in) above the soil surface in Spring 2010. Stump diameters were 7 to 13 cm (3 to 5 in). All trees received 14 g (0.5 oz) of nitrogen (N) from urea (46N-0P-0K) in April of years mound layering was to occur. Shoots remaining from previous years were pruned away at their base each spring.

When shoots reached 50 to 100 cm (20 to 39 in) in height and were actively growing, stock plants were thinned in preparation for mound layering. Leaves were stripped from basal 35 cm (14 in) of shoots and stems of less than 0.5 cm (0.2 in) basal diameter were removed. In 2013, leaves of shantung shoots were stripped from basal 20 cm (8 in) due to a shortage of tall shoots. Most stock plants produced 15 to 20 shoots. Treatments were applied within 13 d of stock plant preparation.

Treatments

In 2012, treatments were three rates of the potassium (K) salt of IBA (K-IBA) (Sigma-Aldrich, St. Louis, Mo.) dissolved in reverse osmosis water at 0; 5,000; and 10,000 ppm (0%, 0.5%, and 1.0%). In 2013, the K-IBA solution was replaced with four rates of the free acid of IBA (≥ 99.0%,Sigma-Aldrich, St. Louis, Mo.) dissolved in 1 EtOH: 1 water (v/v) at 0; 10,000; 15,000; and 20,000 ppm (0%, 1.0%, 1.5%, and 2.0%). Treatments were randomly assigned to shoots and denoted by colored tags fastened to each shoot. Each treatment was assigned to five shoots (five subsamples) on each stump (block) when possible. In 2013, if 20 shoots were available on a stump, then all four IBA treatments were applied to that stump. If less than 20 shoots were available, then the number of subsamples of the 15,000 ppm (1.5%) IBA treatment was reduced for that stump. Treatments were applied to caddo maple on 27 June 2012 and 25 June 2013 and to shantung maple on 8 June 2012 and 10 July 2013.

Auxin solution was applied after stems were wounded. In 2012, stems were wounded between the second and third basal nodes with two 5 cm (2 in) longitudinal cuts through the phloem, on opposite sides of the stem. At harvest, it was observed that these lightly wounded shoots had completely sealed over with little sign of callus development. It was therefore hypothesized that a heavy wound may be needed. In 2013, heavy wounding was achieved by scraping one side of each stem with a grafting knife to expose its xylem at an internode 5 to 10 cm (2 to 4 in) from its point of shoot attachment to the stump, leaving a wound 3 to 4 cm (1.2 to 1.6 in) in length and 0.25 to 0.5 cm (0.1 to 0.2 in) wide. Regardless of the wounding method, the assigned auxin solution was applied immediately to the circumference of each stem at the wound site using dedicated foam brushes.

After allowing the auxin to adhere to the shoot surface (5 min), substrate retention rings were placed around treated stock plants. Inverted 46 L (12 gal), 38 cm (15 in) deep nursery pots (15S, Poly-Tainer Inc., Simi Valley, Calif.) with their bottoms cut out were used to retain the substrate for the shantung maples of 2012. For the caddo maples of 2012 and both species in 2013, substrate retention rings were made from 160 cm x 38 cm (60 in x 15 in) sections of perforated root wrap (RootBuilder II Expandable Container, RootMaker Products Co., LLC, Huntsville, Ala.) secured with cable ties to form rings with 50 cm (20 in) diameters and 75 L (20 gal) capacity. The outer side of all rings was painted white to reduce heating due to light absorption.

Once the substrate retention rings were placed around the auxin treated shoots, they were backfilled with a commercial container production substrate (Metro-Mix 900, Sun Gro Horticulture, Agawam, Mass.) to cover the portion of the shoots which had been stripped of leaves. After watering, additional substrate was added if settling had occurred. All wound sites were covered by at least 10 cm (4 in) of substrate.

Maintenance

Supplemental water was applied as needed by overhead irrigation and hand watering to maintain moist substrate conditions in the rooting substrate and in the soil of the stock plant root zones. Five thermometers were randomly assigned to five mound layered plants during 2013. They were placed vertically in the top 20 cm (8 in) of the growing media, 8 cm (3 in) from the south facing (warmest) side of each ring.

Harvest

Dormant shoots of both species were harvested from the first experiment on 19 Nov. 2012; 21 and 24 weeks after treating the caddo and shantung maples, respectively. Dormant shoots of both species were harvested from the second experiment on 12 Mar. 2014; 37 and 35 weeks after treating the caddo and shantung maples in 2013, respectively. Substrate retention rings were lifted or unwrapped and growing substrate was gently removed. Rooted shoots were severed from the stock plants at their point of origin. Root number and length of primary roots was measured for each shoot.

Statistical Analysis

When rooting data was collected from the shantung maple shoots of 2013, substrate surrounding 6 of the 20 stock plants had settled or eroded during the winter – exposing the wound and treatment sites on 1 to 3 of the shoots of those stock plants. Roots developed from the exposed wound on only one shoot, likely before the substrate settled. Data was analyzed with and without those six plants to compare their effect. Conclusions were not altered by the omission or inclusion of the stock plants in question. Therefore, all stock plant data was included in the final analysis.

The experimental design was a randomized complete block design with 3 (2012) or 4 (2013) auxin concentrations. On stock plants with sufficient numbers of shoots, there were five shoots (subsamples) per auxin concentration. There were 19 replications (blocks) of caddo maple both years. Of the shantung maples, there were 19 replications (blocks) in 2012, but 20 replications in 2013 because treatments were applied to an extra stock plant from the same population as the other stock plants. Data were subjected to analysis of variance using the general linear models (GLM) procedure of SAS (Statistical Analysis System, Version 9.2, SAS Institute Inc., Cary, N.C.). Where appropriate, data were also subjected to regression analysis.

Results

Caddo maples (2012 and 2013) and shantung maples (2013) rooted successfully by mound layering. Stock plants remained healthy throughout both seasons. Substrate temperatures reached 29 C (85 F) during afternoons on days when air temperatures reached 38 C (100 F).

Caddo Maples

K-IBA and IBA concentration affected rooting of caddo maple shoots. Rooting responded in a positive linear relationship to increasing concentrations of K-IBA and IBA in 2012 and 2013, respectively (Tables 3.1 and 3.2). In 2012, rooting peaked at 37% with the highest K-IBA rate [10,000 ppm (1.0%)], while only 4.3% of shoots which received no K-IBA rooted. In 2013, rooting percentage increased from 10.0% at 0 ppm (0%) IBA to 71.1% at 15,000 ppm (1.5%). Rooting dropped to 54% when IBA was increased to 20,000 ppm (2.0%).

Mean root number of caddo maple shoots was significantly influenced by IBA concentration only in 2013 (Table 3.2). Root number responded in a positive linear relationship to increasing IBA concentration reaching a maximum of 29.5 roots per rooted shoot at 20,000 ppm (2.0%) IBA. Differences in root number for 2012 were unaffected by IBA concentration.

Mean root length of caddo maple shoots was not significantly influenced by auxin concentration in either experiment (Table 3.1 and 3.2). Interestingly, mean root length 21 weeks after treatment averaged 16.2 cm (6.4 in) in 2012, but only 12.2 cm (4.8 in) after 37 weeks in 2013.

Shantung Maples

All shoots of shantung maple failed to root in 2012. Shoots appeared healthy throughout growing season. When the growing substrate was removed at the end of the experiment, shoots had sealed over wound sites without producing roots.

There was a strong quadratic rooting response to increasing IBA concentration in shantung maple shoots in 2013 (Table 3.3). Rooting peaked at 32% with 15,000 ppm (1.5%) IBA before declining to 15.9% rooting at 20,000 ppm (2.0%). Neither mean root number (5.9) nor mean root length [11.4 cm (4.5 in)] responded to IBA concentration.

Discussion

Results from these two experiments followed expected patterns and agree well with related literature.

Caddo Maples

Establishing the response of rooting to IBA concentration was one of the key purposes of this experiment. Work by Rupp et al. (2013) and Alsup (2001) had demonstrated that IBA treatments could improve rooting of layered sugar maple subspecies, but optimal rates had not been established. Based on results from the current study, 15,000 ppm (1.5%) IBA promotes best rooting of mound layered caddo maple shoots. Further work may improve rooting of specific cultivars by investigating finer rate increments within the 10,000 to 20,000 ppm (1.0 to 2.0%) range.

This current work reports 71.1% rooting at 15,000 ppm (1.5%) IBA for mound layered caddo maple shoots. Of the stem cutting and mound layering propagation methods investigated by Alsup (2001), caddo maple rooted best (52.9%) from shoots which were wounded and treated with IBA before mound layering. The current study has improved the efficiency of caddo maple propagation by mound layering, making it now one of the most productive methods of obtaining caddo maple clones on their own root systems.

The positive relationship between IBA concentration and root length agrees with the current scientific understanding that IBA induces root initiation (Taiz and Zeiger, 2006). Although IBA concentrations in this study failed to show the upper limit of the positive response of mean root number to IBA concentration, they do suggest an upper limit to rooting percentage. Additionally, 29.5 roots per rooted shoot is more than sufficient to support the shoots after harvest.

The similarity of mean root length between the two experiments suggests that most first season root elongation from layered shoots occurs by late autumn. Although shoots treated in 2013 were left on stock plants 4 months longer than those treated in 2012, their mean root length was no longer than the mean root length of shoots of 2012. This may be partially due to the unusually severe winter of 2013/2014 which kept the substrate around stock plants frozen until March 2014. When shoots were harvested in 2014, root tips were white and beginning to grow.

Shantung Maples

The effect of IBA concentration upon rooting of shantung maple parallels the response observed in caddo maple. Based on this study, mound layered shantung maple shoots root best when treated with 15,000 ppm (1.5%) IBA. As with the caddo maples, further research may optimize this IBA concentration for specific cultivars by experimenting with smaller increment rates from 10,000 to 20,000 ppm (1.0 to 2.0%). This relatively high IBA rate is not surprising for a species notoriously difficult to root. However, the high IBA rate optimal for mound layering does seem odd when compared to a recent study which showed that rooting of shantung maple stem cuttings has a negative linear relationship to increasing concentrations of IBA (Brock, 2014). The difference in size and developmental state of propagation material in the two studies likely accounts for the different responses. Semi-hardwood stem cuttings have a smaller diameter and are more tender than the lignified bases of shoots used for mound layering.

Although the current study has demonstrated the feasibility of rooting shantung maples by mound layering, the efficiency of this method remains low. Currently, stem cuttings are the most efficient method of propagating shantung maple cultivars on their own root systems. Future work may improve the efficiency of both cutting propagation and mound layering. However, given that stem tip cuttings can already provide nearly 50% rooting and use stock plants more efficiently, mound layering of shantung maples does not seem to hold great commercial potential, though it may be useful for rooting particularly difficult cultivars or for the amateur lacking greenhouse facilities.

Differences Between Years

The improved rooting of shoots in 2013 compared to 2012 cannot be attributed to any single factor because adjustments were made to the experimental design in 2013 and other changes occurred. These differences included a new style of substrate retention rings for the shantung maples, heavier wounding of shoots, and replacing water with 1 EtOH: 1 water as an auxin solvent. Other differences include yearly variation in the developmental stage at which treatments were applied and the age of the stock plants which were potentially rejuvenated by successive seasons of topping. Of these factors, heavier wounding and the IBA solvent probably played the biggest role. Heavy wounding likely interfered more with carbohydrate translocation out of the shoot than the light wounding did in 2012. This principle is often applied to air layering. Using 1 EtOH: 1 water should have improved penetration of IBA into shoot tissue. When applied to stem cuttings of shantung maple, recent work (Brock, 2014) found that 1 EtOH: 1 water slightly improved rooting (48.6%) compared to treating cuttings with only water (37.1%).

Conclusion

Caddo and shantung maples both root best in mound layering situations when treated with 15,000 ppm (1.5%) IBA. Using 1 EtOH: 1 water as an IBA solvent and heavily wounding the base of stems likely enhance rooting. Growers may now propagate caddo maple on its own root system by mound layering. For shantung maple propagation, stem cuttings remain the best alternative to grafting.

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Table 3.1

Percent rooting, mean root number, and mean length of primary roots of layered caddo maple (*Acer saccharum* subsp. *saccharum*) shoots treated 27 June 2012 with potassium (K) salt of indole-3-butyric acid (K-IBA) dissolved in water.

K-IBA (ppm)	Rooting^z (%)	Root No.	Root Length (cm)
0	4.3	1.5	15.0
5,000	12.6	2.8	16.1
10,000	37.0	7.8	17.6
Column Mean^y	18.0	4.0	16.2
Linear^x	**	NS	NS

^{z} n = 89, 89, and 90 shoots for 0; 5,000; and 10,000 ppm, respectively

^{y}Mean across all K-IBA treatments

^{x}Nonsignificant (NS) at $P \leq 0.05$ or (**) Significant at $P \leq 0.01$

Table 3.2

Percent rooting, mean root number, and mean length of primary roots of layered caddo maple (*Acer saccharum* subsp. *saccharum*) shoots treated 25 June 2013 with the free acid of indole-3-butyric acid (IBA) dissolved in 1 ethanol (EtOH): 1 water (v/v).

IBA (ppm)	Rooting^z (%)	Root No.	Root Length (cm)
0	10.0	4.3	10.8
10,000	44.4	10.2	13.1
15,000	71.1	19.2	11.9
20,000	54.1	29.5	12.9
Column Mean^y	44.9	15.8	12.2
Linear^x	**	**	NS
Quadratic	*	NS	NS

^{z}n = 89, 91, 67, and 90 shoots for 0; 10,000; 15,000; and 20,000 ppm, respectively

^{y}Mean across all IBA treatments

^{x}Nonsignificant (NS) at $P \leq 0.05$, (*) Significant at $P \leq 0.05$, or (**) Significant at $P \leq 0.01$

Table 3.3

Percent rooting, mean root number, and mean length of primary roots of layered shantung maple (*Acer truncatum*) shoots treated 10 July 2013 with the free acid of indole-3-butyric acid (IBA) dissolved in 1 ethanol (EtOH): 1 water (v/v).

IBA (ppm)	Rooting[z] (%)	Root No.	Root Length (cm)
0	0.0	-	-
10,000	19.0	6.7	9.5
15,000	32.4	4.3	11.7
20,000	15.9	6.7	13.0
Column Mean[y]	16.8	5.9	11.4
Linear[x]	**	NS	NS
Quadratic	**	NS	NS

[z] n = 100, 100, 94, and 100 shoots for 0; 10,000; 15,000; and 20,000 ppm, respectively
[y] Mean across all IBA treatments
[x] Nonsignificant (NS) at $P \leq 0.05$ or (**) Significant at $P \leq 0.01$

Chapter 4 - Rooting Stem Cuttings of Herbaceous and Woody Ornamentals in Substrates Containing Eastern Redcedar (*Juniperus virginiana*)

Introduction

Perlite is a key component of most commercial substrates used for cutting propagation. Because of its aerating characteristics, light weight (80 to 128 kg·m^{-3}) (5 to 8 lb·ft^{-3}), and water holding capacity, growers commonly incorporate it as 30% to 100% by volume of their rooting substrates (Moore, 1987). Perlite is mined as an amorphous silicate and heated at 593 to 871 C (1100 to 1600 F), changing the water inside the silicate to steam and expanding the particle (Moore, 1987). Characteristics of the final product are determined by the initial particle size and the heating process (Moore, 1987). Hartmann et al. (2011) notes the sterile nature of perlite as an advantage, but recognizes its dusty nature as a disadvantage. Perlite dust is an eye and lung irritant. The recommended exposure limit to perlite dust is a time weighted average of 5 mg·m^{-3} (5 ppb) (OSHA, 2014). In one case of acute exposure to perlite dust, three out of twenty-four workers developed long term respiratory health problems (Du et al., 2010). However, severe health issues are not commonly associated with perlite.

Although perlite remains standard in much of the horticulture industry, researchers have investigated alternative substrate components that could replace or reduce perlite both in propagation and production substrates. Recent work by Starr (2011) demonstrates that hammer milled eastern redcedar (*Juniperus virginiana* L.) chips (ERC), can replace perlite without reducing propagation success with stem cuttings of chrysanthemum (*Chrysanthemum morifolium* Ramat. ‘Abelle’), ivy geranium [*Pelargonium peltatum* (L.) L’Her. ‘Colorcade Cherry Red’], hibiscus (*Hibiscus rosa-sinensis* L.; cultivar unknown), privet (*Ligustrum* ×*vicaryi* Rehder ‘Golden Vicary’), and ‘Green Giant’ arborvitae (*Thuja* L. ×‘Green Giant’).

Eastern redcedar is native to the Great Plains and the eastern half of the U.S. (Hardin et al., 2001). Although commonly grown in landscapes, eastern redcedar becomes a nuisance when it invades grasslands. In one study in the Flint Hills region of Kansas, closed canopy eastern redcedar forest cover increased by 120% between 1986 and 2000 (Hoch, 2000). Eastern redcedar encroachment causes both economic and ecological consequences with a 99% reduction in herbaceous biomass productivity and severe loss of plant species diversity (Briggs et al., 2002). Increased grazing pressure and decreased fire frequency and intensity are the major factors in the conversion of grasslands to woodlands (Briggs et al., 2002; Hoch, 2000). In some cases, eastern redcedar trees are cut and cleared from grasslands. Although this woody material is often burned, it can also be chipped and used for other purposes. Processing coarsely chipped eastern redcedar through a hammer mill yields material suitable as an ornamental crop substrate component.

The purpose of this study was to investigate the potential of ERC as a substitute for perlite in a general purpose, 3 perlite: 1 sphagnum peat moss (v/v) rooting substrate used for propagation of spreading euonymus (*Euonymus kiautschovicus* Loes.; cultivar unknown), forsythia (*Forsythia ×intermedia* Zab.; cultivar unknown), English ivy (*Hedera helix* L. ‘Anne Marie’), lantana (*Lantana camara* L. ‘Irene’), and coleus [*Solenostemon scutellarioides* (L.) Codd ‘Defiance’)].

Materials and Methods

Experimental Design

This study compared rooting of six plant species in five substrates. Each of 6 blocks contained 1 flat of each substrate and each flat contained 6 randomly assigned subsamples of each species, arranged identically in each flat. The flats were randomly arranged under intermittent mist within each block.

Substrate

On 6 Dec. 2013 coarsely chipped eastern redcedar (Queal Enterprises, Pratt, Kans.) which had been hammer milled to pass a 9.5 mm (0.38 in) screen was further processed through a hammer mill (Model 30HMBL, C.S. Bell Co., Tiffin, Ohio) to pass a 4.8 mm (0.19 in) screen. These processed chips were used to prepare five substrates of increasing ERC content (0%, 25%, 50%, 75%, and 100% by vol.). All substrates contained 25% sphagnum peat moss by volume (Ferti-lome, Bonham, Texas) except the 100% ERC substrate. The remaining volume of each substrate was coarse perlite (Sun Gro Horticulture, Agawam, Mass.). Fresh samples of each substrate were collected for physical property analysis.

Clean flats [40 cm x 40 cm x 12.7 cm with 5 mm screen bottom (15.75 in x 15.75 in x 5 in with 0.20 in screen bottom] (AFlat5, Anderson Die and Manufacturing Inc., Portland, Ore.) were dipped in a sterilizing solution [3.96 $ml \cdot L^{-1}$ (0.5 $fl. oz \cdot gal^{-1}$) Green-Shield, Whitmire Micro-Gen Research Laboratories, Inc., St. Louis, Mo.] before each was filled with one of the five substrates. Three days before the first cuttings were inserted, all flats were placed under intermittent mist (8 s every 4 min during natural daylight hours) [Flora-Mist #300A, 0.25 $L \cdot min^{-1}$ (0.066 $gal \cdot min^{-1}$), Hummert International, St. Louis, Mo.] in a glass greenhouse with natural photoperiod and constant temperature set at 28.5 C (84 F).

Cutting Material and Setup

Woody cuttings of spreading euonymus and forsythia were harvested 16 Dec. 2013 from the Kansas State University campus, Manhattan, Kans. Stem tissue from the most recent year was selected and cuts were made above nodes to form 10 to 15 cm (4 to 6 in) shoot tip stem cuttings. Cuttings were kept moist during harvesting and processing. Any remaining leaves were stripped from dormant forsythia cuttings and from the basal half of euonymus cuttings. The bottom 1 cm (0.4 in) of each cutting was dipped 5 s in a 1000 ppm (0.1%) solution of potassium (K) salt of indole-3-butyric acid (K-IBA) (7012I, Research Organics, Inc., Cleveland, Ohio) dissolved in distilled water. After 5 min, cuttings were inserted 5 cm (2 in) deep into each flat, and substrate was firmed around each cutting. Intermittent mist was initially set at 6 s every 8 min, 24 $h \cdot d^{-1}$, but was increased to 6 s every 4 min after 1 week.

Herbaceous stem cuttings from a commercial unrooted cutting supplier (North Carolina Farms Inc., Indian Trail, N.C.) arrived 3 Jan. 2014 and were stored in moist newspaper at 6.7 C (44 F) until treated and inserted into substrates on 4 Jan. 2014.

Single node stem cuttings of English ivy were 2 to 4 cm (0.8 to 1.6 in) in length and had been trimmed just above a node at both ends by the supplier. A small number of the ivy cuttings, had one or two roots measuring 1 to 2 cm (0.4 to 0.8 in) in length at nodes when cuttings arrived. These roots were removed to ensure that roots measured at harvest had developed in the assigned substrate. Lantana and coleus both arrived as 2 to 4 cm (0.8 to 1.6 in) stem tip cuttings without visible roots.

The basal 1 cm (0.4 in) of English ivy and lantana cuttings was dipped in a 1000 ppm (0.1%) K-IBA solution for 5 s and allowed to rest 5 min. Coleus cuttings were not treated with K-IBA. Six cuttings of each species were individually inserted 1 to 2 cm (0.4 to 0.8 in) deep into dibbled holes in each substrate which was gently firmed around cuttings to ensure good stem to substrate contact. Cuttings were kept moist during entire setup procedure.

Maintenance

Seventeen days after the herbaceous cuttings were inserted, intermittent mist was reduced to 6 s every 8 min to begin hardening off the rooted cuttings. At 21 d, mist was further reduced to 6 s every 16 min to prevent rot of herbaceous cuttings. At the conclusion of the experiment, pH and electrical conductivity (EC) of water samples from the mist system were measured.

Harvest

Cuttings were destructively harvested to measure rooting, mean root number and mean length of primary roots. Coleus cuttings were harvested 25 d after insertion in the propagation substrates. English ivy and lantana were harvested at 32 d after insertion with spreading euonymus cuttings harvested at 51 d. Lastly, forsythia cuttings were harvested after 59 d in the substrates. When roots could not be measured immediately, cuttings were wrapped in moist paper towels, placed inside re-sealable plastic bags, and stored in a walk in cooler set to 12 C (53 F) [Cooler was adjusted to 3 C (38 F) for the last 4 d the English ivy cuttings were stored]. Roots were defined as any linear root growth longer than 0.2 mm (0.01 in) to distinguish roots from callus tissue.

Substrate Physical Properties

Particle size distribution was determined from 100 g (3.53 oz) samples of oven dried [24 h at 105 C (221 F)] substrate separated on a sieve shaker delivering 278 oscillations·min^{-1} and 159 tap·min^{-1} (Ro Tap RX-29, W.S. Tyler, Mentor, Ohio) with 12 sieve opening sizes of 12.7, 9.5, 6.3, 3.35, 2.36, 2.00, 1.40, 1.00, 0.50, 0.25, 0.106, and 0.053 mm (0.5, 0.375, 0.248, 0.132, 0.093, 0.079, 0.055, 0.039, 0.020, 0.010, 0.004, and 0.002 in) plus a catch pan. Sieves were divided into two sets – the largest six and the smallest six, and samples were run for 3 min on each. Mass of material caught in each sieve was measured and the value was divided by 100 g (3.53 oz) to determine its proportion of the original substrate.

Four replications of each substrate were measured according to the procedure described by North Carolina State University's Horticultural Substrates Laboratory (Fonteno and Harden, 2003) to determine air space, container capacity, total porosity, and bulk density. Before physical properties were measured, samples were adjusted to 35% volumetric water content using distilled water. Moistened samples were placed inside closed plastic bags for 11 h to equilibrate. Aluminum cores measuring 7.6 cm dia. x 7.6 cm tall (3 in dia. x 3 in tall) were filled with substrate as described by Fonteno and Harden (2003) and dropped 6 times from a height of 5 cm (1.97 in) to establish substrate structure. During the saturation procedure, the 3 perlite: 1 sphagnum peat moss (v/v) substrate was prone to float out of the core. To prevent this, the top of the core was covered with a perforated plastic petri dish, weighted and arranged upright to minimize interference with the measurements. Bulk density was determined from 347.5 cm^3 (21.21 in^3) samples dried at 105 C (221 F) in a forced air oven (13-247-725F, Thermo Fisher Scientific Inc., Pittsburgh, Pa.) for at least 24 h.

Statistical Analysis

The propagation experimental design was a randomized complete block design with five substrate treatments and 6 single cutting subsamples per replication. Each substrate was replicated 6 times. Data were analyzed with the general linear models (GLM) procedure of SAS (Statistical Analysis System, Version 9.2, SAS Institute Inc., Cary, N.C.). Because coleus cuttings in one block were clearly damaged by an edge effect, coleus data from this block was omitted from the final analysis. Sieve shaker results showed that particle size varied among substrates in three distinct ranges. These ranges were based on particle diameter and defined as coarse [>2.36 mm (>0.093 in)], medium [2.36 to >0.5 mm (0.093 to >0.020 in)], and fine [≤0.5 mm (≤0.020 in)]. Substrates were analyzed a second time using these three categories. Differences in physical properties were tested for significance using a protected means separation [Waller-Duncan K ratio t-test (α=0.05)].

Results and Discussion

In general, percent rooting was high (≥ 95%) except for forsythia (25.6%) (Tables 4.1 - 4.5), and unaffected by substrate ERC content. Mean root number per rooted cutting and mean root length were also unaffected by substrate ERC content, with the exception of spreading euonymus (Table 4.1). Physical properties of the five substrates were generally within recommended ranges (Table 4.7) with total porosity slightly above and bulk density below recommendations.

Rooting

Shoot tip cuttings of spreading euonymus rooted 96.1% and rooting was unaffected by ERC substrate content (Table 4.1). This agrees with Dirr (2009) who stated that cuttings of spreading euonymus root easily. Cuttings in the standard substrate [3 perlite: 1 sphagnum peat moss (v/v)] rooted well (97.2%) with a mean of 36.3 roots per rooted cutting and a mean root length of 7.1 cm (2.8 in). This confirms that euonymus cuttings used in this experiment were capable of rooting successfully and that including ERC in growing substrates does not negatively influence percent rooting.

For spreading euonymus, mean root number per rooted cutting and mean root length did respond to ERC substrate content. The response of mean root number to increasing ERC substrate content was quadratic in nature. Greatest root number peaked at 0% and 100% ERC with means of 36.3 and 25.6 roots, respectively. Intermediate ERC substrate contents produced the lowest mean root number (≈20). No single substrate component or physical property entirely explains the trend observed in root number. However, root length also had a general decline with increasing ERC content. The decline was linear with a maximum mean root length of 7.1 cm (2.8 in) with 0% ERC substrate content. This response could be explained by the increasing substrate bulk density with increasing ERC content. Kirkham (2005) demonstrates that increasing bulk density increases the work roots must do to elongate. Thus, increasing ERC content increases mechanical resistance to root elongation. Chong (1999) observed similar results with burning bush [*Euonymus alatus* (Thunb.) Seibold] when rooting cuttings in six substrates containing 0% to 75% by volume composted municipal solid waste. In that study, root length was inversely related to increasing substrate bulk density.

Explaining the difference in euonymus root length by differences in bulk density faces the dilemma that low substrate bulk densities, such as those observed in the current study [0.17 $g \cdot cm^{-3}$ (10.6 $lb \cdot ft^{-3}$) at the highest ERC content], are much lower than typical landscape soil conditions. This does not negate the current results, however. There was still a range of bulk densities among the substrates (although all were quite low) and the length of euonymus roots corresponded closely to the differences in bulk density. Furthermore, comparing the difference in mean root length to the difference in bulk density at increasing levels of ERC reveals that the magnitude of the change at each level is similar. This strongly suggests that bulk density was the main factor influencing mean root length of euonymus cuttings in this study.

A difference in pH of the substrates is not likely to have influenced root number or length. Irrigation water in this experiment had a pH of 7.4 and an EC of 220 $mS \cdot cm^{-1}$. Starr (2011) did not observe any clear relationship between ERC content and the pH of propagation substrate leachates from ERC substrates under intermittent mist.

Allelopathic chemicals found in ERC are not likely responsible for the decrease in root length. Work by Smith (1986) shows that most of the allelopathic effects of eastern redcedar influence seedling germination and not general plant growth. Because other species in the current study did not show such a response, an allelopathic effect is either unlikely or species specific.

Overall, forsythia cuttings rooted poorly (Table 4.2). Most of the cuttings bloomed and developed leaves while under mist, and many desiccated when the intermittent mist was reduced (to 6 s every 16 min) 40 d after the experiment began. Warm greenhouse conditions likely forced forsythia cuttings to initiate shoot growth before adventitious root initiation. Cooler air temperature and bottom heat may have induced rooting prior to bud expansion and growth. Cuttings rooted less than expected with 25.6% rooting, a mean of 3.6 roots per rooted cuttings, and a mean root length of 1.5 cm (0.59 in) (Table 4.2).Rooting percentage dropped sharply to 8.3% rooting in 100%

ERC, though the difference is not significant. This suggests that 100% ERC is a poor substrate for rooting forsythia, though further research would be needed to confirm this conclusion.

Rooting of herbaceous cuttings was more successful. Stem cuttings of English ivy rooted at 98.9% with a mean of 11.8 roots per cutting and mean root length of 4.5 cm (1.8 in) (Table 4.3). Cuttings of lantana rooted at 97.2% with a mean root number of 7.8 and mean root length of 3.5 cm (1.4 in) (Table 4.4). Similarly, cuttings of coleus rooted at 94.7%, with a mean root number of 12.3, and mean root length was 5.5 cm (2.2 in) (Table 4.5). These results demonstrate that cuttings of certain species can root successfully in ERC substrates.

The species rooted in this project are known for their high rooting potential and success. They were chosen as ideal candidates for demonstrating the feasibility of rooting cuttings in ERC substrates. Now that the suitability of ERC as a propagation substrate has been established, future work ought to focus on identifying the rooting substrate preferences of individual species, especially those that are difficult to root.

Physical Properties

During particle size analysis, no particles were caught on the two largest sieves [12.7 and 9.5 mm (0.5 and 0.375 in) openings]. Although a fine dust accumulated in the pan below the smallest sieve for each substrate, the quantity was insufficient to register on a scale accurate to 0.1 g (0.004 oz). Analysis of particle size distribution showed that the proportion of particles of a given size differed greatly among substrates in three distinct ranges, but converged to similar proportions of particles at two sizes: >2.00 mm but <2.36 mm and >0.25 mm but <0.50 mm (>0.079 in but <0.093 in and >0.010 in but <0.020 in) (Table 4.6). Dividing particle size distribution into categories of coarse [>2.36 mm (>0.079 in)], medium 2.36 mm to >0.5 mm (0.093 to >0.020 in)], and fine [≤0.5 mm (≤0.020 in)] clarified the results. Replacing perlite, but not peat, with ERC reduced the amount of coarse particles in the substrate. Increasing the ERC content increased the amount of medium sized particles in all cases. Replacing perlite or peat moss with ERC decreased the amount of fine particles in the substrate except from 50% to 75% ERC, where fine particle content was unchanged. Considering substrate components based on their particle size distributions helps to explain their interaction with other substrate components. This leads to a greater understanding of how each component influences substrate physical properties such as container capacity.

Significant differences (α=0.05) among substrates were observed for all physical properties (Table 4.7) except air space, which averaged 30.5% of substrate volume. Container capacity generally increased as ERC content increased, reaching 57.6% of volume at 75% ERC before dropping to 50.8% when ERC replaced peat moss in the 100% ERC substrate. The increase in container capacity as ERC rose from 25% to 75% seems largely due to the uniform particle size of ERC which were 70.7% medium sized particles. These particles nested closely together, increasing the water holding capacity of the substrate. Bilderback and Lorscheider (1995) observed a similar result in a substrate containing double processed pine bark with a high proportion of uniformly sized particles. The decrease in container capacity between 75% and 100% ERC substrates is clearly due to the decrease in sphagnum peat moss, which has a high water holding capacity. The corresponding increase in air space demonstrates that peat moss, which was contributing mostly fine particles, had been nesting between the ERC particles in other substrates. When the peat moss was removed, these pores had an increased volume. Thus, air space increased and container capacity decreased when peat moss was replaced by ERC.

Total porosity rose steadily from 79.2% of volume at 0% ERC to 88.9% of volume at 75% ERC. However, when ERC content increased to 100%, porosity decreased slightly. This trend is a direct result of the changes seen in air space and container capacity. Both air space and container capacity generally increase from 0% to 75% ERC content. Although air space increases again when ERC reaches 100%, container capacity drops sharply with the loss of peat moss. Combined, these factors caused the slight decrease in total porosity at 100% ERC.

Bulk density rose steadily from 0.09 $g \cdot cm^{-3}$ (5.6 $lb \cdot ft^{-3}$) at 0% ERC to 0.17 $g \cdot cm^{-3}$ (10.6 $lb \cdot ft^{-3}$) at 100% ERC. Clearly, ERC is a denser substrate component than either perlite or sphagnum peat moss.

Maronek et al. (1985) provide recommended ranges for physical properties of propagation substrates (Table 4.7). In the current experiment, air space and container capacity were within the recommended ranges for all substrates. Total porosity was above the recommended range of 40% to 60% porosity in all cases, but was closest (79.2%) in the control substrate. High porosity may lead to poor contact between substrate and cutting tissue (Maronek et al., 1985), but in this experiment, high porosity did not seem to be a problem as most species rooted well. Bulk density was also outside the recommended range of 0.3 to 0.8 $g{\cdot}cm^{-3}$ (18.7 to 49.9 $lb{\cdot}ft^{-3}$) and only reached 0.17 $g{\cdot}cm^{-3}$ (10.6 $lb{\cdot}ft^{-3}$) at the highest ERC content. Similarly, Starr (2011) determined the bulk density of his 100% ERC substrate to be 0.18 $g{\cdot}cm^{-3}$ (11.2 $lb{\cdot}ft^{-3}$). The range for bulk density recommended by Maronek et al. is influenced by the ballast needed in substrates used for liner production in 5 to 10 cm (2 to 4 in) pots. Though substrates from this experiment may not be dense enough for container production applications, they appear suitable as propagation substrates.

Conclusion

ERC substrates have excellent potential for cutting propagation. Although roots of certain species such as spreading euonymus and possibly forsythia may develop poorly in ERC substrates, other species including English ivy, lantana, and coleus root well in substrates containing up to 100% ERC. Propagators seeking alternatives to perlite should seriously consider ERC as a component of their propagation substrate.

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Table 4.1

Percent rooting, mean root number, and mean root length of stem cuttings of spreading euonymus (*Euonymus kiatschovicus*; cultivar unknown) inserted into substrates containing eastern redcedar (*Juniperus virginiana*) chips (ERC).

ERC[z] (% vol.)	Rooting[y] (%)	Root No.	Root Length (cm)
0	97.2	36.3	7.1
25	100.0	20.1	5.9
50	97.2	20.7	4.1
75	88.9	20.2	2.6
100	97.2	25.6	2.3
Column mean	96.1	24.6	4.4
Linear	NS[x]	NS	**
Quadratic	NS	**	NS

[z] Hammer milled to pass a 4.8 mm (0.19 in) screen. All substrates contained 25% peat by volume except the 100% ERC treatment. Perlite filled the remaining volume.

[y] n=36 stem cuttings per treatment

[x] Not significant (NS) at *P*≤0.05, (*) Significant at *P*≤0.05, or (**) Significant at *P*≤0.01

Table 4.2

Percent rooting, mean root number, and mean root length of stem cuttings of forsythia (*Forsythia ×intermedia*; cultivar unknown) inserted into substrates containing eastern redcedar (*Juniperus virginiana*) chips (ERC).

ERC[z] (% vol.)	Rooting[y] (%)	Root No.	Root Length (cm)
0	27.8	4.2	1.6
25	30.6	3.9	1.3
50	36.1	3.9	1.2
75	25.0	2.9	2.1
100	8.3	2.5	1.0
Column mean	25.6	3.6	1.5
Significance	NS[x]	NS	NS

[z] Hammer milled to pass a 4.8 mm (0.19 in) screen. All substrates contained 25% peat by volume except the 100% ERC treatment. Perlite filled the remaining volume.

[y] n=36 stem cuttings per treatment

[x] Not significant (NS) at $P \leq 0.05$

Table 4.3

Percent rooting, mean root number, and mean root length of stem cuttings of English ivy (*Hedera helix* ‘Anne Marie’) inserted into substrates containing eastern redcedar (*Juniperus virginiana*) chips (ERC).

ERC[z] (% vol.)	Rooting[y] (%)	Root No.	Root Length (cm)
0	94.4	11.5	4.8
25	100.0	13.0	4.3
50	100.0	11.5	4.7
75	100.0	12.8	4.1
100	100.0	10.0	4.6
Column mean	98.9	11.75	4.5
Significance	NS[x]	NS	NS

[z] Hammer milled to pass a 4.8 mm (0.19 in) screen. All substrates contained 25% peat by volume except the 100% ERC treatment. Perlite filled the remaining volume.
[y] n=36 stem cuttings per treatment
[x] Not significant (NS) at $P \leq 0.05$

Table 4.4

Percent rooting, mean root number, and mean root length of stem cuttings of lantana (*Lantana camara* 'Irene') inserted into substrates containing eastern redcedar (*Juniperus virginiana*) chips (ERC).

ERC[z] (% vol.)	Rooting[y] (%)	Root No.	Root Length (cm)
0	94.4	6.7	3.0
25	100.0	7.9	3.8
50	94.4	8.3	3.6
75	97.2	8.0	3.5
100	100.0	8.1	3.4
Column mean	97.2	7.8	3.5
Significance	NS[x]	NS	NS

[z] Hammer milled to pass a 4.8 mm (0.19 in) screen. All substrates contained 25% peat by volume except the 100% ERC treatment. Perlite filled the remaining volume.

[y] n=36 stem cuttings per treatment

[x] Not significant (NS) at $P \leq 0.05$

Table 4.5

Percent rooting, mean root number, and mean root length of stem cuttings of coleus (*Solenostemon scutellarioides* 'Defiance') inserted into substrates containing eastern redcedar (*Juniperus virginiana*) chips (ERC).

ERC[z] (% vol.)	Rooting[y] (%)	Root No.	Root Length (cm)
0	100.0	11.6	5.4
25	86.7	14.3	5.5
50	90.0	12.4	5.9
75	96.7	10.8	5.3
100	100.0	12.3	5.3
Column mean	94.7	12.3	5.5
Significance	NS[x]	NS	NS

[z] Hammer milled to pass a 4.8 mm (0.19 in) screen. All substrates contained 25% peat by volume except the 100% ERC treatment. Perlite filled the remaining volume.

[y] n=30 stem cuttings per treatment

[x] Not significant (NS) at $P \leq 0.05$

Table 4.6

Particle size distribution[z] of substrates containing eastern redcedar (*Juniperus virginiana*) chips (ERC).

U.S.A. Standard Test Sieve No.	Sieve Opening (mm)	Substrate									
		0% ERC[y]		25% ERC		50% ERC		75% ERC		100% ERC	
0.25 in	6.30	0.7[x]	a[w]	0.4	ab	0.4	b	0.3	bc	0.0	c
6	3.35	20.60	a	10.2	b	4.6	c	1.7	d	1.2	d
8	2.36	16.4	a	12.2	b	9.7	c	8.0	d	8.3	d
10	2.00	5.5	d	7.1	c	7.3	c	7.8	b	8.3	a
14	1.40	7.8	e	15.2	d	18.5	c	21.7	b	23.0	a
18	1.00	6.4	e	12.2	d	15.4	c	18.2	b	19.8	a
35	0.50	12.8	d	16.5	c	18.3	b	19.7	a	19.6	a
60	0.25	13.0	b	13.2	b	14.3	a	14.3	a	12.9	b
140	0.106	11.2	a	9.7	b	9.2	b	7.0	c	5.5	d
270	0.053	3.9	a	3.0	b	1.9	c	1.0	d	0.8	d
Pan	-	0	NS[v]	0		0		0		0	
Coarse	>2.36	37.7	a	22.8	b	14.7	c	10.1	d	9.6	d
Medium	2.36 to >0.5	32.5	e	50.9	d	59.5	c	67.3	b	70.7	a
Fine	≤0.5	28.1	a	25.9	b	25.4	b	22.3	c	19.2	d

[z] From 100.0 g (3.53 oz) samples dried at 105 C (221 F) and separated on sieve shaker for 3 minutes (278 oscillations·min^{-1}, 159 taps·min^{-1}) (Ro Tap RX-29, W.S. Tyler, Mentor, Ohio).

[y] Percent volume of total substrate. ERC were hammer milled to pass a 4.8 mm (0.19 in) screen. All substrates contained 25% peat by volume except the 100% ERC treatment. Perlite filled the remaining volume.

[x] Percent of total sample weight collected from sieve. Column totals do not equal 100% because of particle losses due to static electricity.

[w] Means separated within row using Waller-Duncan K-ratio t test (n=3, α=0.05).

[v] Means not significantly different at α=0.05.

Table 4.7

Physical properties[z] of substrates containing eastern redcedar (*Juniperus virginiana*) chips (ERC).

ERC[y] (% vol)	Air Space[x] (% vol.)		Container Capacity[w] (% vol.)		Total Porosity[v] (% vol.)		Bulk Density[u] g·cm^{-3}	
0	26.0	NS[t]	53.2	bc[s]	79.2	c	0.09	e
25	29.7		51.1	c	81.3	bc	0.11	d
50	30.1		55.1	ab	85.2	ab	0.13	c
75	31.4		57.6	a	88.9	a	0.16	b
100	35.4		50.8	c	86.2	ab	0.17	a
Recommended Range[r]	15-40		20-60		40-60		0.3-0.8	

[z] Measured according to procedures described by the Horticultural Substrates Laboratory at N. C. State Univ. (Fonteno and Harden, 2003)

[y] Hammer milled to pass a 4.8 mm (0.19 in) screen. All substrates contained 25% peat by volume except the 100% ERC treatment. Perlite filled the remaining volume.

[x] Air Space = (volume of water drained) ÷ (initial volume of sample)

[w] Container Capacity = [(wet weight) - (oven dry weight)] ÷ (initial volume of sample)

[v] Total Porosity = (air space) + (container capacity)

[u] Bulk Density = (oven dry weight) ÷ (initial volume of sample)

[t] Differences not significant (NS) at α=0.05

[s] Mean separation within columns using Waller-Duncan K-ratio t test (n=4, α=0.05).

[r] From Maronek et al., 1985.

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