# Propagation of Ornamental Plants for Oklahoma



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# Propagation of Ornamental Plants for Oklahoma

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Most ornamental plants in Oklahoma can be propagated locally by nursery personnel, which allows them to increase the number of plants with specific desirable characteristics. Plants can be propagated by asexual or sexual means. Asexual propagation refers to the multiplications of plants from vegetative plant parts such as shoots, roots, and leaves, while sexual propagation involves the growing of plants from seed.

# **Asexual Propagation**

Asexual propagation is the only practical means of reproduction when plants do not produce viable seed, or seeds are difficult to germinate. The most important reason for using asexual reproduction is to grow plants with the same characteristics as the parent plant. This method can produce marketable plants in less growing time and increase profits.

# **Rooting Media**

Many people have tried to define or describe the ideal rooting medium for asexual propagation. In general, it has been found that one ideal rooting medium does not exist but several combinations of materials are best. A mixture of equal volumes of peat moss and coarse perlite is a suitable rooting mixture for most plants. Combinations of materials such as rockwool, shredded sphagnum, vermiculite, and sand have also proven satisfactory.

Peat and perlite or peat and ground pine bark in a 1:1 ratio works well in containers approximately 2 ½ inches deep. If the pot is less than 2 ½ inches deep, use more perlite or bark to slightly increase aeration, and if it is deeper, a slightly larger proportion of peat could be used. Drainage and aeration are functions of the depth of the mix; therefore, finer particles can be used for deeper containers, which drain better than smaller ones, with satisfactory results. A detailed list of growing media is given in Table 1.

The medium should drain freely and be free of disease organisms and weed seeds. Pasteurization of media components reduces diseases and other pests. Some commercial growers mix and pasteurize their own media. Perlite and vermiculite are sterile when purchased. Good quality peat moss, although not sterile, is clean and ready for use directly from the bale. In contrast, ground pine bark or sand should be pasteurized prior to use. Small quantities can be pasteurized by placing a two-inch layer of moist medium on a tray in an oven at 170° F (76° C) for one hour. Packaged media can also be purchased.

The open wound at the base of the cutting combined with the warm, moist environment is an ideal entrance site for diseases. In most cases, if a disease is present, cuttings will die while still in the propagation area. In other instances, the young plants may appear healthy and succumb only after an environmental stress predisposes them to the disease.

## **Containers Versus Beds**

Individual containers allow rooting medium to be transferred along with the rooted cutting to the field or to a larger container for growing. The root system is undisturbed and establishment is very rapid. Generally, when cuttings are rooted in small containers, a smaller volume of rooting medium is provided for each cutting as compared to bed-grown liners; however, there is little difference in the volume of rooting medium used and, likewise, in the cost of rooting medium. Bedding plant trays and other planting containers can be used once or, if labor and facilities are available, can be dipped in a disinfectant solution and used again. The advantage of trays is that workers handle many plants at one time rather than individual pots; therefore, labor and space are used efficiently in the propagation house at a very low cost.

Rooting cuttings in beds can be accomplished for a lower initial investment with several times more plants for each square foot of propagation bed. However, cost for each liner is similar for containers and beds because crowding, stunting, and disease problems are associated with propagation beds after several cycles of cuttings. Isolation and removal of diseased plants is easier in containers. In addition, slow release fertilizer incorporated into the rooting medium greatly increases plant quality when liners are propagated in containers, but is only effective during the initial rooting when beds are used.

When cuttings are crowded together in bulk beds during propagation, it is impossible to remove the plant liners without greatly disturbing and damaging the root system. Liners in containers are undisturbed and growth is continuous with no transplant shock as long as they do not remain in the containers too long.

Medium	Origin	Weight <sup>a</sup>	Water Holding Capacity	Nutients <sup>b</sup> Present	pH and Buffering <sup>c</sup> Capacity	Drainage	Need for Sterilization
Soil	Organic & inorganic components, rock plus subsoil, variable composition-solid, liquid & gas	Usually heavy	Variable	Some- many	Variable	Variable	Yes
Sand	Small rock particles usually silicaceous	Very heavy	Little or none	None	Neutral & none	Very good	Yes
Peat	Remains of bog vegetation-partly decomposed	Medium to light	High	1% N	Very acidic & high	Poor (slow)	Usually recommended
Spagnum moss	Dehydrated remains of acid bog plants, genus <i>Sphagnum</i>	Very light	Very high	Similar to peat	Very acidic & high	Slow	Not recommended (fungistatic)
Vermiculite	Mica type material expanded by extreme heat	Very light	High	Mg & K	Neutral & high	Poor (compacts with use)	No
Perlite	Volcanic ore processed and subjected to very high temperatures	Very light	Good	None	Neutral & none	Very good	No
Leaf molds- composts	Leaves, soil, and added N	Moderate to heavy	Good to high	N & some others	Acid & good	Poor to good	Yes, but this causes problems
Pine bark	Lumber industry by-product	Light	Good	Some, slowly available need N added	Acid & fair to good	Very good	Recommended
Hardwood bark	Lumber industry by-product	Light	Good	Some, slowly available, need N added	Initially acid by pH rises as decomposition proceeds, fair to good	Very good	Recommended, pasteurization achieved by composting
Sawdust	Lumber industry by-product	Light	Good	Some, slowly available, needs N added	Initially acid but pH rises as decomposition proceeds, fair to good	Fair	Recommended pasteurization achieved by composting

#### Table 1. Characteristics of Commonly Used Propagation Media.

a Relative dry weights based on equal volumes of the various propagation media presented in this table.

b N = nitrogen, K = potassium, Mg = magnesium.

c Buffering capacity refers to the capacity for resisting changes in pH.

# Water Quality

Water quality is important in mist propagation. Since nearly all of the mist applied to leaves of cuttings evaporates, any salts or debris in the water is left behind to accumulate on the leaf surface. In some instances, salts may shade the leaf enough to reduce photosynthesis and subsequent rooting and growth. Salts may also accumulate in the rooting medium and reduce rooting. Approximate guidelines for interpreting water sample results are given in Table 2. Chlorinated city water is preferred over lake or stream water, since it contains few, if any, living disease organisms and algae. Water from deep wells is generally satisfactory unless boron content or total salts are excessive.

Water pressure of 45 to 60 psi is needed to provide a mist with small droplets and good coverage. Low water pressure is a frequent cause of poor moisture distribution in the propagation greenhouse.

Solu Bridge Reading Quality	Total Salts in Parts per Million (PPM)	Water
0.00 - 0.25 0.25 - 0.75 0.75 - 1.50 1.50 - 2.00 2.00 or above	0 - 150 150 - 500 500 - 1,500 1,500 or above	Very Good Good Fair Questionable Unsuitable

#### Table 2. General Interpretation of Water Quality Using a Solu Bridge.

#### Cuttings

The most common method of propagating plants asexually is from cuttings, which can be made from stems, roots, or leaves. Cuttings should be taken from healthy plants with desirable characteristics, and placed in a warm, humid environment to prevent drying and hasten root development. After they have been transplanted into the rooting media, heat applied at the bottom of the containers will enhance rooting of many species. Refer to Figure 1 for a review of plant parts and terminology.

Polarity is important when planting cuttings. The piece of cutting that was originally at the top should be oriented upwards when inserted in the medium. Roots will eventually form from the lower portion of the cutting. If polarity is reversed, the cutting may not root well enough to produce a plant. See Photo 1.

#### **Selection of Cuttings**

In general, the more vigorous and healthy the parent plant, the better the cuttings root and grow. One should also check carefully to be sure it is the desired species or cultivar.

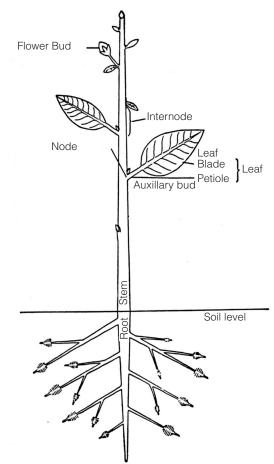
The size and type of cuttings are sometimes dictated by the size and vigor of the branches on the parent plant. In general, however, cuttings should be 5 to 7 inches long and  $\frac{1}{4}$  to  $\frac{1}{2}$  inch in diameter. Slender branches and all branches from shaded portions of



Photo 1. Note *Sansevieria trifasciata* must be positioned correctly, as should all plant cutting, for successful root initiation to occur.

the parent plant should be avoided since they are lower in carbohydrates and less likely to root and become vigorous plants. Give each cutting enough space to avoid overcrowding of the foliage and excessive shading of some leaves. Many species will root poorly, or not at all, when part or all of the leaves are removed from the cutting. The practice of removing one-fourth to one-half of each leaf was developed prior to intermittent mist systems and should not be used for most species in modern propagation facilities.

Cuttings can be classified as herbaceous or woody cuttings. Herbaceous cuttings are





Scientific Name	Common Name	Kind of Cutting <sup>a</sup>
A <i>belia</i> spp.	Abelia	SH, HW
A <i>cer</i> spp.	Maple	SW, SH
Amelanchier alnifolia	Serviceberry	SW
Berberis julianae	Wintergreen barberry	SH
Berberis x mentorensis	Mentor barberry	SH
Berberis thunbergii	Japanese barbery	SH, HW
Betula spp.	Birch	ŚŴ
Buxus microphylla	Littleleaf boxwood	SH, HW
Buxus sempervirens	Common boxwood	SH, HW
Campsis spp.	Trumpet creeper	SW, SH, HW
Catalpa spp.	Catalpa	SW
Ceanothus spp.	Ceanothus	SW, SH, HW
Cedrus spp.	Cedar	SH, HW
Celastrus spp.	Bittersweet	SW, SH, HW
Cercis spp.	Redbud	SW SH
Chaenomeles spp.	Flowering quince	
Chameacyparis spp.	Chamaecyparis, False cypress	SH, HW
Chionanthus spp.	Fringe tree	SW
<i>Clematis</i> spp.	Clematis	SW, SH
Cornus spp	Dogwood	SW, SH
Cotinus coggygria	Smoke tree	SW
Cotoneaster spp.	Cotoneaster	SW, SH
<i>Cytisus</i> spp.	Broom	SW, HW
Cupressocyparis leylandii	Leyland cypress	SH, HW
<i>Deutzia</i> spp.	Deutzia	SW, HW
Elaeagnus angustifolia	Russian olive	HW
<i>Euonymus</i> spp.	Euonymus	SH, HW
Forsythia spp.	Forsythia	SW, SH, HW
Ginkgo biloba	Ginkgo, Maidenhair tree	SW
Gleditsia triacanthos	Honeylocust	HW
ledera helix	English ivy	SH
Hibiscus syriacus	Shrub-althea, Rose of Sharon	SW, HW
<i>lydrangea</i> spp.	Hydrangea	SW, HW
<i>Typericum</i> spp.	St. Johnswort	SW
lex aquifolium	English holly	SH
lex x attenuata 'Fosteri'	Foster's holly	SH
lex cornuta	Chinese holly	SH, HW
lex crenata	Japanese holly	SH, HW
		011
lex opaca	American holly	SH
lex vomitoria	Yaupon holly	SH
lasminum spp.	Jasmine Chinese iuniner	SH
luniperus chinesis	Chinese juniper	SH, HW
luniperus conferta	Shore juniper	SH. HW
luniperus horizontalis	Creeping juniper	SH, HW
<i>Koelreuteria</i> spp.	Goldenrain tree	SW
agerstroemia indica	Crapemyrtle	SH
<i>igustrum</i> spp.	Privet	SW, SH, HW
iquidambar styraciflua	Sweet gum	SW
<i>onicera</i> spp.	Honeysuckle	SW, HW
<i>lagnolia</i> spp.	Magnolia	SW, SH
Nahonia spp.	Mahonia	SH
<i>falus</i> spp.	Crabapple	SW, SH
<i>letasequoia glyptostroboides</i>	Dawn redwood	SW, HW
Aorus alba	Mulberry	SW
Parthenocissus quinquefolia	Virginia creeper	SW, HW
Parthenocissus tricuspidata	Boston ivy	SW, HW
Philadelphus spp.	Mockorange	SW, HW

# Table 3. Stage of Maturity Necessary for Rooting Stem Cuttings of Woody Orna-mentals.

Scientific Name	Common Name	Kind of Cutting <sup>a</sup>
Photinia spp.	Photinia	SH, HW
Pinus mugo	Mugo pine	SH
Pinus strobus	Eastern white pine	HW
<i>Populus</i> spp.	Poplar, aspen, Cottonwood	SW, HW
Prunus spp.	Fowering cherry	SW, SH
Pyracantha spp.	Pyracantha, Firethorn	SH
Pyrus calleryana	Callery pear	SH
Rhododendron spp.	Azalea (deciduous)	SW
Rhododendron spp.	Azalea (evergreen & semi-evergreen)	SH
Rhododendron spp.	Rhododendron	SH, HW
Rhus spp.	Sumac	SW
Rosa spp.	Rose	SW, SH, HW
Salix spp.	Willow	SW, SH, HW
Sambucus spp.	Elderberry	SW
Spiraea spp.	Spirea	SW
Syringa spp.	Lilac	SW
Taxus spp.	Yew	SH, HW
Thuja occindentalis	American arborvitae	SH, HW
Thuja orientalis	Oriental arborvitae	SW
Tilia americana	American linden, Basswood	SW
<i>Ulmus</i> spp.	Elm	SW
Virburnum spp.	Viburnum	SW, HW
Weigela spp.	Weigela	SW, HW
Wisteria spp.	Wisteria	SW

\* SW = softwood, SH = semi-hardwood, HW = hardwood. Rooting success (percentage) will vary with species/cultivar and environmental conditions.

referred to as stem, leaf, leaf-bud, and root cuttings. Woody cuttings come from either deciduous plants that lose leaves and go through a dormancy period or narrow-leaved evergreen plants which do not lose their leaves and do not go through a noticeable dormancy. These woody cuttings include softwood, semihardwood, and hardwood cuttings. Reference to various species and the appropriate type(s) of cuttings is listed in Table 3.

#### Herbaceous Cuttings

Herbaceous plants are non-woody species. A few examples of these are begonias (*Begonia* spp.) and geraniums (*Pelargonium* spp.). The sections removed for cuttings may or may not contain leaves, depending upon the species' requirements and type of cutting. Most plants can be propagated any time of the year since many of these are grown in greenhouses.

#### Woody Cuttings

**Softwood.** These cuttings usually form roots the quickest (two to five weeks) of the three woody cutting techniques. Plant examples include forsythia (*Forsythia* spp.), lilacs (*Syringa* spp.), and magnolias (*Magnolia* 

spp.). Leafy shoots are removed from the new, soft growth of the season. Cuttings should be harvested in the morning hours and rooted in a moist, humid environment at a media temperature of 75 to 80° F (23 to 27° C) from bottom heat and 70 F (21° C) for aboveground leaf portions.

Each cutting should have two or more nodes and range from 3 to 5 inches long. They should be removed from lateral branches and be of average diameter in relation to other branches on the plant. The bottom cut should be made below a node. Any large leaves, flowers, or flower buds must be removed, since they compete with newly forming roots for essential nutrients.

**Semihardwood.** The cuttings from broadleaved evergreens, such as hollies (*llex* spp.), euonymus (*Euonymus* spp.), and azaleas (*Rhododendron* spp.), are removed during the summer months after new growth has slightly matured and is semihard. Deciduous plants, including camellias (*Camellia* spp.) and pittosporums (*Pittosporum* spp.), have cuttings removed during middle to late summer before dormancy begins.

The cuttings should be taken in the morning hours. All large leaves, flowers, and flower buds must be removed as with the softwood cuttings, The average length ranges from 3 to 6 inches with the bottom cut below a node. Terminal cuttings are the most common; however, more than one section may be used from the same branch if there is enough length. Proper care must be maintained to prevent moisture loss until the cutting has rooted.

**Hardwood.** These cuttings are removed from narrow-leaved evergreens and deciduous ornamentals, such as junipers (*Juniperus* spp.), pines (*Pinus* spp.), spruces (*Picea* spp.), honeysuckle (*Lonicera* spp.), spirea (*Spiraea* spp.), plums (*Prunus* spp.), and willows (*Salix* spp.). Hardwood cuttings are taken from dormant wood during the late fall, winter, or early spring. The central and bottom sections of the plants are better sources for cutting material. There must be at least two nodes on each piece. The lengths range from 4 to 30 inches and the diameter may be ¼ to 2 inches. Hardwood cuttings must be stored correctly to prevent moisture loss until needed.

There are three types of hardwood cuttings—straight, heel, and mallet. The straight is the most common, which is a piece of the branch. The heel has a piece of older wood from the main stem and the mallet has a larger section of the stem. These forms enable the cuttings to root more efficiently, depending upon the species.

#### Juvenility

For difficult-to-root plants, the age of the parent plant may be a critical factor in propagation success. In most cases, cuttings obtained from seedling plants root easier, quicker and in greater percentages than cuttings from older plants in an adult growth phase. This has been shown to be true for a number of species, including Chinese pistache (*Pistacia chinensis*), which is a highly valued ornamental tree for Oklahoma landscapes. The greatest success for rooting stem cuttings to date has been from seedlings.

The lower portion of many trees, such as oaks (*Quercus* spp.), retain their leaves throughout the winter months (this is the juvenile growth stage). When taking cuttings, obtain sticks from this juvenile portion of the stock plant.

Some propagators deliberately keep plants in a juvenile growth stage. Hedging and shearing have worked for several species when cuttings were taken from the forced succulent growth. Since the juvenility factors vary so widely among woody species, it is best to refer to a propagation book which lists specific plants and methods discovered to overcome the juvenility factor; however, for some species, trial and error will be required.

#### Stem Cuttings

Stem cuttings, the most commonly used cuttings, are used for numerous rooting, grafting, and budding purposes. Stem cuttings can be taken at different stages of vegetative maturity and may consist of just the growing tip of a plant or subterminal stem sections. As mentioned, the species of the pant and use of the cutting will determine whether herbaceous, softwood, semihardwood, or hardwood cuttings should be removed.

Stem cuttings are removed using a clean, sharp knife or pruner. Cuttings 4 to 6 inches in length are appropriate for most plants. Leaves are removed from the bottom inch of stem cuttings, which are stuck upright in a propagation medium—usually ½ to 1 inch below the soil surface. The position of the cutting on the plant affects desired growth. Cuttings removed form lateral sections of some plants, such as a juniper (*Juniperus* spp.), may grow in a spreading manner rather than upright. Be sure to remove cuttings from the appropriate areas on the plant material for the growth habit desired.

#### Leaf Cuttings

Leaf cuttings consist of only the leaf blade or the leaf blade and petiole (the leaf stem). African violets (Saintpaulia ionantha), begonias (Begonia spp.), and sansevierias (Sansevieria trifasciata) are commonly propagated by leaf cuttings directly inserted into rooting medium. Leaf cuttings of some plants, such as the Rex begonia (Begonia rex), are wounded by cutting the underside of the main veins before placing the leaf surface flat and in form contact with the propagation medium. Sometimes these leaves should be pinned to moist media with toothpicks. Leaf cuttings of many plants can be stuck upright in the propagation medium. Roots and new shoots will start at the base of the leaf or at points where the veins are cut.

#### **Leaf-bud Cuttings**

Leaf-bud cuttings include the leaf blade, petiole, and  $\frac{1}{2}$  to 1 inch segment of the stem.

Auxiliary buds located at the union of the petiole and stem produce new shoots under warm, humid conditions. This method is often used for plants in short supply that have long internodes, such as the philodendron (*Philodendron* spp.). Every node (joint) on the stem can be a cutting. Other common examples are rhododendrons (*Rhododendron* spp.) and many of the bramble crops such as blackberry (*Rubus allegheniensis*).

#### **Root Cuttings**

Root cuttings are usually taken from young plants in early spring or late winter before they start growing. Healthy roots have ample food (carbohydrates) stored to support shoot development during this time of year. Root cuttings are typically 2 to 7 inches in length, depending upon root diameter. Large roots can be cut shorter than small roots and still have an adequate food supply for root and shoot initiation. Small, delicate root cuttings (1/8 to 1/4 inch in diameter) should be positioned horizontally, 1/2 inch deep in the propagation medium. Larger root cuttings (1/4 to 1/2 inch in diameter) can be planted vertically with the end of the cutting originally nearest the plant

crown (the transition from roots to shoots) positioned upward. Optimum temperatures for most root cuttings range from 55 to 65° F (13 to 18° C). Root cuttings may be transplanted after shoots have emerged and sufficient new secondary roots have developed. The principle disadvantage of this method is the amount of work involved in obtaining the root cuttings. Many species such as the Kentucky coffee tree (*Gymnocladus dioicus*) are successfully propagated by this method. See Table 4 for detailed examples.

#### Wounding

In a number of woody species, wounding the basal portion of stem cuttings promotes rooting. The two basic types of wounding are light and heavy.

A light wound could be described as vertical cuts on the basal stem penetrating the bark to the wood. These cuts, usually numbering two or four, would be equidistant and oriented parallel to the long axis of the cutting. The cuts would not be made deep enough to split the stem. Junipers (*Juniperus* spp.), for example, respond to light wounding. Strip-

Scientific Name	Common Name
Ailanthus altissima Albizia julibrissin	Tree-of-Heaven Silk Tree
Broussonetia papyrifera	Paper Mulberry
Campsis radicans	Trumpet Vine
Celastrus scandens	American Bittersweet
Chaenomeles japonica	Japanese Flowering Quince
Forsythia x intermedia	Forsythia
Hypericum calycimun	St. Johnswort
Koelreuteria paniculata	Goldenrain Tree
Malus spp.	Apple, Flowering Crabapple
Myrica pennsylvanica	Bayberry
<i>Populus</i> spp.	Poplars
Prunus spp.	Plums
Pyrus calleryana cvs.	Callery Pear cvs.
Rhus copallina	Shining Sumac
Rhus glabra	Smoth Sumac
Rhus typhina	Staghorn Sumac
Robinia pseudoacacia	Balck Locust
Rosa spp.	Roses
Rubus spp.	Blackberries, Raspberries
Sassafras albidum	Sassafras
Styphnolobium japonicum	Japanese Pagodatree
Symphoricarpos hancockii	Coralberry
Syringa vulgaris	Lilac

Table 4. Oklahoma Species Propagated by Root Cuttings.

ping leaves and lower side branches from the basal portion of cuttings can also be regarded as a light wound. Although wounding stimulates rooting, the greatest rooting response is achieved when cuttings are treated with a root-promoting compound following wounding.

A heavy wound would consist of the removal of a thin strip of bark on opposite sides of the basal stem exposing the cambium. Magnolias (*Magnolia* spp.) and rhododendrons (*Rhododendron* spp.) often respond to heavy wounding.

Girdling is another form of wounding that promotes rooting on plants such as hibiscus (*Hibiscus* spp.) and water oak (*Quercus nigra*). This process prevents hormones, carbohydrates, and other rooting factors from moving to the root system. These are contained in a concentrated area so new roots form.

The process of etiolation is used on plants that do not root well with auxin. Plants are grown in heavy shade to produce shoots which are removed from the stock plants and readily form roots. A few species include clematis (*Clematis* spp.) and lilacs (*Syringa vulgaris* cvs). A lighter shade condition is used for species that do not require the extreme method of etiolation. A few of these plants are roses (*Rosa* spp.) and the Japanese euonymus (*Euonymus japonica*).

# Hormonal Treatment of Cuttings

A widely used propagation practice is the treatment of cuttings with root-promoting compounds. However, these materials are usually limited to cuttings from difficult-to-root plants. The treatment of cuttings, which normally root easily without treatment with root-promoting compounds, may only result in additional expense without any benefits to the propagator. Cuttings are treated with root-promoting compounds to:

- increase the percentage of cuttings which form roots,
- quicken root initiation,
- increase the number and quality of roots produced per cutting, and
- increase the uniformity of rooting for crop scheduling.

The chemical compounds most often used to treat cuttings for root promotion are known as auxins. A landmark in the history of plant propagation was the discovery in 1934-35 of the role of auxin [indoleacetic acid (IAA)] in promoting adventitious root initiation. This advancement led to auxin treatment of cuttings to stimulate rooting and made it possible to root large quantities of cuttings consistently from many difficult-to-root plants.

IAA is the only naturally occurring auxin found in plants; however, the synthetic auxins, indolebutyric acid (IBA), and naphthaleneacetic acid (NAA), have strong root-promoting properties. One other compound, 2,4-dichlorophenoxyacetic acid (2,4-D), is used for rooting plants as well as a herbicide in large doses. These materials may be used alone or in combination to treat cuttings. As expected, when they are combined, their effectiveness is increased. IBA and NAA are most often used because they are more effective in promoting root formation in cuttings from a wide range of plants. The common auxin-type rooting preparations are: (a) application of auxintalcum powder mixtures, (b) the concentrated solution-dip method (quick-dip method), and (c) the dilute solution soaking method.

Commercial auxin formulations consist of: (1) an auxin or several auxins dispersed in talcum powder or gel form, or (2) one or more auxins dissolved in a solvent. The solutions are usually concentrated and must be diluted by the propagator before use.

Of the two commercial rooting formulations, the auxin-talcum powder mixtures have remained popular. These formulations are used widely and sold under numerous trade names. They are available in varying strengths and combinations of active ingredients.

Despite availability of commercial formulations, many propagators prefer to purchase the reagent grade of a particular auxin or auxins and prepare their own rooting formulations. Propagator prepared rooting formulations can be just as effective as commercial preparations if correct procedures are followed. These can be more economical than purchasing commercial products, and allow propagators greater flexibility in terms of the concentrations of formulations they could prepare. The reagent grade IBA is more effective than IAA or NAA; however, propagators who desire to prepare their own concentrated solutions may experience difficulty purchasing a reagent grade of various auxins, particularly IBA.

Whether commercial or propagator prepared, rooting formulations are used to treat cuttings. The various treatment techniques utilized have distinct advantages and disadvantages. Keep in mind that each procedure can yield satisfactory results when used properly. Choice of a particular method will depend on many factors and should be suited to a propagator's particular operation.

#### **Auxin-Powder/Gel Mixtures**

This method consists of dipping the bottom 1/2 to 2 inches of a cutting into an auxintalcum powder mixture. The talcum powder merely serves as a carrier for the auxin. This type of preparation can be purchased commercially from various nursery supply houses or prepared by a propagator if reagent grade auxin is available. The strength of the solution can vary from 1,000 ppm to 30,000 ppm depending on the species or cultivars to be rooted and product used. The mixture can contain more than one auxin and may also contain a fungicide. When formulating auxin-talcum powder mixtures, a very important step is thorough blending of the auxin(s) and talcum powder.

A knife or trowel should be used to make a trench in the rooting medium into which the base of the cuttings are placed. The base of the cutting should be moist so that the powder will stick. After dipping, the cutting is lightly tapped to remove any excess powder and immediately inserted into the rooting medium. The rooting medium should be gently pressed around each cutting. It is important to place the cutting in a prepared hole or trench, otherwise the rooting powder may be wiped off with friction.

Cuttings should never be dipped into the entire stock of powder because this can result in microbial contamination of the stock and lead to early deterioration. When treating cuttings, remove a small quantity of the rooting powder and place it into another container for treatment. Remove only material needed for a particular length of time. Any excess left after treatment should be used quickly or discarded.

The advantages of using auxin-talcum powder formulations are their availability and ease of application. The disadvantages include the lack of uniformity to potentially varying amounts of powder applied to each cutting, and the cost of commercial powder preparations can be high when large quantities of cuttings are to be treated.

### Concentrated-Solution-Dip Method

This procedure involves dipping the bottom  $\frac{1}{4}$  to  $\frac{1}{2}$  inch of stem cuttings into a concentrated auxin solution very briefly (1 to 5 seconds) followed by insertion of the cuttings into the rooting medium. The strength of the solution can vary from 500 to 30,0000 ppm (0.05 to 3.0 percent), depending on the species or cultivars to be rooted and the particular chemical used.

There are several advantages associated with using this quick-dip technique: (a) if reagent grade auxin is used to prepare the solution, the solution will be cheaper than the equivalent amount of a commercial rooting preparation (more cuttings treated at a lower cost); (b) the procedure is fast and easy to use; and (c) uniform results are obtained particularly when treating cuttings uniform in length and tied in bundles.

The disadvantages associated with this techniques are: (a) if the acid formulation of an auxin is used, the propagator may have difficulty getting the chemical into solution, and (b) although application is simple, slight miscalculations can cause problems.

As mentioned above, the propagator may have difficulty getting the chemical into solution. The reagent grade of most auxins is usually synthesized in acid formulation. In this form, the chemicals are generally insoluble in water and must be dissolved in alcohol. Ethyl, methyl, or isopropyl alcohol are all satisfactory solvents. Full-strength (100%) alcohol should be used when preparing concentrated auxin solutions and, prior to use, should be reduced to half strength (50%) using distilled water.

Although the acid formulation of an auxin is alcohol soluble, it is often necessary to use a mechanical stirring device to get the chemical completely into solution. The higher the concentration of auxin desired in solution, the more stirring is required.

Labels of bottles containing the acid formulation of an auxin will simply indicate the chemical name of the contents. Thus; remember that alcohol must be used to dissolve the material; however, if the label indicates the chemical is the potassium salt (K-salt) formulation, then the compound is water-soluble. Distilled water should be used rather than tap water. Potassium salt formulations of IBA and NAA are available but are more expensive than the acid formulations.

When using the guick-dip method, a few precautions must be taken. If care is not exercised, the concentration of the solution will vary and become too concentrated or too diluted. For example, if the acid formulation is being used (the auxin dissolved in alcohol), evaporation can rapidly cause the solution to become more concentrated. The increasing concentration can cause injury or death to the cuttings; however, if the cuttings are wet, the concentrated solution can be diluted, thus reducing the efficacy. To avoid either, an increase or decrease in concentration, it is advisable to use only a small portion of the stock solution at any time. Any solution remaining after use should always be discarded and not poured back into the stock solution.

On a large production scale, the best way to avoid an increase or decrease in concentration is to periodically mix fresh solutions. Stock solutions should be kept in tightly capped bottles and stored in darkened containers in a refrigerator. Use solutions within three to four months.

#### Dilute Solution Soaking Method

This method consists of soaking the bottom 1 to 2 inches of stem cuttings in the dilute solution of a rooting compound for several hours (2 to 24) prior to inserting the cuttings into rooting medium. The solution concentration generally varies from 20 to 500 ppm of active ingredient, depending upon the active ingredient(s), soaking time, and the species or cultivars treated. The solutions can contain more than one auxin.

This dilute solution soaking method is not currently popular, since it has no real advantages and several disadvantages. The procedure is slow, special equipment is needed for soaking the cuttings, and the amount of solution absorbed can vary depending upon environmental conditions.

# Hardening Rooted Cuttings

There must be a transitional period to allow new roots and leaves formed in a humid environment to adjust gradually to hot, dry conditions in full sun. The first step in hardening is to decrease the humidity by increasing the interval between misting and/or increasing the ventilation if in an enclosed rooting structure (humidity chamber). After the gradual decrease in moisture, the light intensity can be increased gradually by moving the plants into areas where they will receive increasing amounts of direct sunlight. Plants that have been adequately hardened are more likely to survive when transplanted into larger containers or in the landscape.

### Layering

Layering is a relatively easy method of propagation in which new plants are formed while attached to the parent plant. The new plant receives nutrients and water from the parent plant until roots develop. This method of asexual propagation yields a large plant in a relatively short time, and is an excellent way to produce a small number of plants or to propagate plants that are difficult to increase by other methods. Layering outdoors is best performed during spring and summer months, although it can be done during any season of the year.

Healthy, maturing branches that are growing vigorously and have been exposed to light should be chosen for layering, since they usually have more food reserve (carbohydrates) and, therefore root faster. Branches from <sup>1</sup>/<sub>4</sub> to about <sup>3</sup>/<sub>4</sub> inch in diameter are best for layering. If possible, select wood for layering that would normally be pruned when shaping the plant. The various types of layering are air, tip, trench, serpentine, and mound. Air and tip layering are the most popular methods.

## **Air Layering**

Air layering is commonly used for fiddleleaf figs (Ficus lyrata), rubber plants (Ficus elastica), crotons (Codiaeum variegatum), hibiscus (Hibiscus spp.), azaleas (Rhododendron spp.), and magnolias (Magnolia spp.). The first step in air layering is to remove leaves and twigs on the selected limb 3 to 4 inches above and below the point where the air layer is made. The air layer is usually made at least 12 to 15 inches below the tip of the branch. The branch is wounded to induce rooting. One method consists of removing a 1/2 to 1 inch ring of bark and, with a knife, scraping clean the wood underneath. This ensures complete removal of the cambium layer-the layer of cells between the bark and the wood. If the cambium layer is not removed completely, new bark may develop instead of roots. A second method of wounding involves making either a long cut slanting upward about 1/4

to ½ the way through the twig, or two small cuts on opposite sides of large branches or on branches having brittle wood. One cut should be slightly higher on the branch than the other, and the cuts should not be too deep or the branch may break. The incision is kept open by inserting a small chip of wood or a toothpick to prevent the cut from closing.

A rooting hormone can be applied around and just above the wound on difficult-to-root plants to hasten rooting, but hormones are unnecessary for most air layering. The wounded area should be bound with a handful of moist sphagnum moss. Squeeze excessive moisture from the moss before placing it completely around the stem at the wound. Tie the moss firmly in place with strong twine or fabric. Wrap the sphagnum ball with clear polyethylene film and tie securely with plastic covered wire or strong rubber bands above and below the ball to prevent the moss from drying. The ball should then be covered with aluminum foil of freezer paper to prevent excessive heat buildup under the plastic.

When a mass of roots has developed in the sphagnum ball (one to six months, depending upon plant species and time of year), the layered branch can be removed from the parent plant (see Photo 2). It is best to allow the new plant to develop a larger root system in a container or protected holding area before planting it in open areas where high light



Photo 2. Air layering of Fiscus spp. soil Soil Propagation of Ornamental Plants for Oklahoma

intensities and dry conditions usually prevail. Layers removed during the growing season should be potted in containers and hardened like the rooted cuttings discussed previously.

#### **Tip Layering**

Tip layering is proven means of propagating climbing roses (*Rosa* spp.), jasmine (*Jasminum* spp.), abelia (*Abelia* spp.), and pyracantha (*Pyracantha* spp.). Most plants with a trailing or vining growth habit can be propagated by this method. A low branch, or one that can be bent easily to the ground, is chosen. The bark is injured (in the manner previously described for air layering) about  $\frac{1}{2}$  to 1 inch along the stem and 4 to 5 inches back from the tip, and the injured area is anchored 2 to 3 inches in the soil. Be sure to keep the soil moist.

Spring is the best time to tip layer because the injured portion will develop roots during warm summer months. Spring layers can be cut from the parent and planted in late fall or left until the following spring. The layered portion should be checked for roots before removal from the parent plant.

# Trench and Serpentine Layering

These methods are similar to tip layering, except that a longer branch is placed in a trench and covered with soil. These methods produce several new plants from each layered branch. Trench layering is useful on plants whose buds will break and start to grow under the soil surface. Willows (Salix spp.), viburnum (*Viburnum* spp.), and dogwood (*Cornus* spp.) can be trench layered. The entire branch, except the tip, is placed in a trench and covered with soil. Serpentine layering involves burying every other but, leaving the alternate bud above ground. This method requires plants with pliable or vining stems, such as grapes (Vitis spp.) and trumpet creeper (Campsis radicans).

#### **Mound Layering**

This can be used to propagate many of the heavy stemmed or closely branched plants such as flowering quince (*Chaenomeles speciosa*). Mound layering is started in spring. The plant is cut back severely prior to spring growth; new shoots that emerge are wounded (as described for air layering), and soil is mounded around the base of the plant. Soil should be mounded up in several stages throughout the first half of the growing season to a maximum of about 1 ½ feet. Adding peat or sphagnum moss to the mounded soil helps when removing the rooted branches. It takes about one growing season to produce shoots that have rooted sufficiently for transplants.

### **Division**

Plants with a multi-stem or clumping growth habit, offshoots, or with underground storage structures such as rhizomes or tubers can be propagated by division. Division involves cutting large clumps into smaller sections, making sure that each smaller clump has an adequate amount of stems, leaves, roots, and buds to survive transplanting. House plants, ferns, daylilies (Hemerocallis spp.), bulbous plants, nandina (Nandina spp.), and liriope (Liriope spp.) are commonly propagated by division. Division is an excellent way to increase the area in the landscape covered with ground covers such as liriope. Each season, dig the plants from a portion or all of the ground cover area, divide the clumps, and replant them into a larger area. Some plants can be pulled apart, but many must be cut. Transplant the separated clumps at the same depth they were growing originally. Do not divide plants when they are flowering because they are low in nutrients during this period. Flowering and seed production compete with the regeneration of roots or other vegetative parts for energy and nutrients. Plants can be divided any other time during the growing season.

#### Grafting

The goal of grafting is to join two plants or plant parts together in such a manner that they will unite and grow as one. Many plants do not readily form root systems from other techniques described. Many ornamentals are grafted to create a more desirable specimen that is pest resistant, more environmentally adaptable, either smaller or larger in size, and of higher quality. Grafting can also be used to repair plants damaged by mechanical or environmental means. New cultivars are created by grafting and budding. Many broadleaf evergreens and deciduous ornamentals, such as azaleas (Rhododendron spp.), junipers (Juniperus spp.), magnolias (Magnolia spp.), rhododendrons (Rhododendron spp.), roses (Rosa spp.), and fruit and nut trees, are propagated with this method of grafting or budding. Recently, grafting of vegetables in the Solanaceae and *Cucurbitaceae* families has gained interest for improved yields and increased disease resistance.

A few precautions should be followed when grafting. A sharpened, fixed straight edged grafting knife is required. The plant material should be free from diseases and insects. Small nails, twine, and rubber band strips may be necessary to support the grafts as they heal. Moisture loss must be prevented, so adhesive tape, parafilm, plastic, polyethylene tape, raffia, or grafting wax are commonly used.

Two pieces of plant material are essential for grafting. The scion is the upper portion of the graft. It is a short piece of a stem with two or more buds and is usually dormant. To prevent the scion from breaking dormancy too early, it should be wrapped in moist peat moss and covered with paper towels or plastic to avoid moisture loss, and kept in cool storage. The stock (rootstock or understock) becomes the lower portion which forms the root system. Rootsocks usually must have a very healthy root system. Often, stocks are grown from seed and are one to two years old. Either dormant or actively growing wood may be used. Early spring is the ideal time for most species. However, more accurate information may be furnished by an Extension educator from your local county office.

A few rules should be followed to ensure a successful graft union. The stock and scion must be compatible and polarity maintained. Plants that are botanically similar are best. Exceptions do exist, but normally plants are only compatible within a genus whose species are genetically similar. The cambial section of the scion should be in direct contact with the cambium of the stock to allow the graft union to heal. Also, the graft should be wrapped securely to avoid dehydration. The temperature may range from 55 to 90° F. The graft must be monitored after completion to ensure proper growth. Often, the stock develops shoots that compete with the scion and must be pruned. Callus (undifferentiated cells) formed around the scion and stock also requires monitoring. Occasionally, an interstock (inserted between the stock and scion) is used to aid in compatibility and control growth.

Symptoms of incompatibility may not appear for several years and may be confused with pest or environmental problems. A few indicators are:

• failure to form a successful union,

- yellow foliage late in season, followed by early defoliation, shoot dieback, and the general unhealthy appearance in the tree due to the blockage of nutrients and water from stock to scion,
- premature death after one to two years in nursery,
- marked differences in growth rate and vigor between stock and scion,
- difference between stock and scion in time of year when vegetation occurs,
- overgrowth at, above, or below the graft union, and
- the plant breaks cleanly at the graft union.

## Whip and Tongue Graft

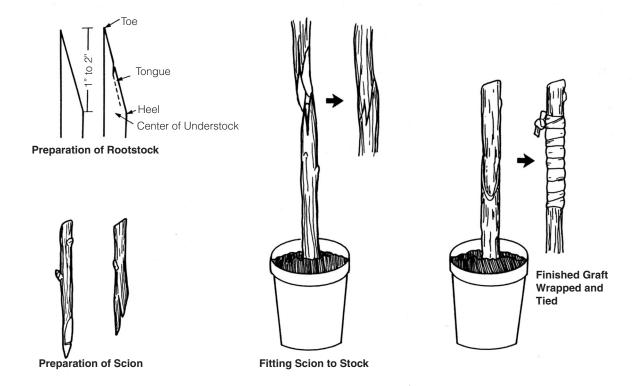
The scions and stocks from either dormant or active wood are cut to form an interlocking segment. Once proper contact has been made, the graft heals quickly and forms a strong union. Both the stock and scion should be 1/4 to 1/2 inch in diameter. Each piece is cut the same to lock together. First, one slanting cut is made about 1 to 2 1/2 inches long. A second cut is made 1/3 from the tip (Figure 2). Each piece is then inserted, tied, and covered with parafilm or wax.

# **Splice Graft**

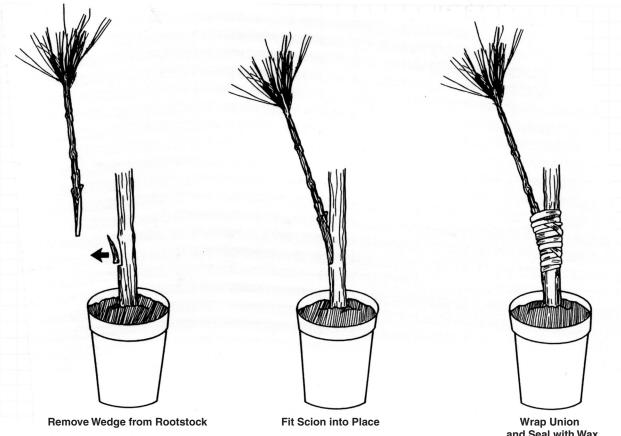
This is similar to the whip and tongue technique but has only one slanting cut; therefore, precise contact with the cambium layers is essential for success. This is usually conducted on plant material that has a pithy stem or inflexible wood to create a tongue.

## Side (Stub) Graft

This method permits the use of a larger stock than scion. Dormant or active wood may be used. The scion is inserted into the side of the stock to create a new branch. The stock may be up to 1 inch in diameter and the scion 3 inches long with up to three buds. The cut in the stock is about 1 inch long and at an angle so when pressure is applied to the stock, the cut opens, then closes when pressure is removed. The scion is cut to form a V-shaped end approximately 1 inch long on each side. The scion is then inserted into the stock. If necessary, two small flat-headed nails (20 gauge, 5/8 inch long) may be used. Then the graft should be wrapped. The stock can be removed above the union when grafting procedures are completed.



#### Figure 2. Whip and tongue graft.



Scion

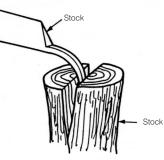
Cambium

Bark

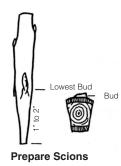
Scions Properly Aligned in Cleft

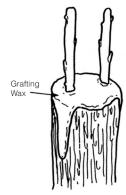
Figure 3. Side veneer graft.

Wrap Union and Seal with Wax



Saw Off Stock and Make Cleft







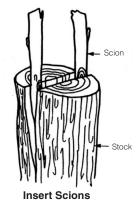


Figure 4. Cleft graft.

#### Side Veneer Graft

The side veneer graft uses dormant or active wood and is similar to the stub graft, but the stock is planted in a container. Also, no flaps are needed. The scion only needs to be 1 inch long with one slanted cut. A nail or tie may be required to hold the scion in place. Wrap is also used for this procedure. Conifers and deciduous trees and shrubs may be grafted with this technique (see Figure 3).

#### **Cleft Graft**

Cleft grafting is used in a variety of species for topworking small trees (Figure 4), branches of large trees, and crown grafting camellias. This can be done on dormant wood in the early spring as buds swell. The stock can range from 1 to 4 inches in diameter. The scion must have two to three buds, be 3/8 to ½ inch in diameter, and 3 to 4 inches long. Two scions are normally used to obtain two separate plants from the same technique, or to have an alternate plant in reserve should one die.

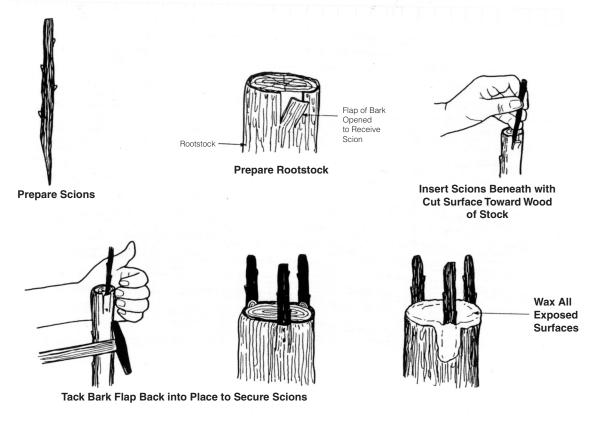
A large knife (butcher knife or cleft grafting tool) is used to make one single split, 2 to 3 inches down the center of the stock trunk. The split must be straight to allow for proper placement of scion(s). A tool may be used to separate the split for scion insertion. The basal (bottom) end of the scion(s) should form a wedge approximately 2 inches long. The scion(s) is then placed on the outer edge of the stock next to the cambium layer. If the scion is place incorrectly in the stock, or is too small, contact between the two cambial layers will not be accomplished.

The last important step is the waxing of the graft. The entire section must be covered, including the split in the stock. The wax should be reapplied as necessary to cover any openings until the graft has healed.

#### **Bark Graft**

The bark graft is performed on thick barked trees, such as pecan or walnut. This method is easy and successful when correctly performed. While it is conducted on actively growing stock wood, the scions must be dormant. Any shoots that grow from the scion should be staked to prevent wind breakage.

Two parallel cuts that are approximately 2 inches long are made through the bark and one at the top to connect them. The scion should be the same width as the bark cutting,



#### Figure 5. Bark graft.

and the basal end of the scion should be cut in a wedge shape for proper insertion into the stock. The cut is opened quickly and the scion inserted to prevent any moisture loss. Then, two small nails are used to hold the scion and stock in place (Figure 5).

#### **Approach Graft**

There are three ways to perform approach grafts. The approach graft is performed on two plants in separate containers. It is done while the bark is slipping (plants are actively growing). The upper portion of the union and the bottom of the scion below the union should be removed carefully to avoid breakage. Occasionally, the graft may be removed in section, if the plant specimen is top heavy. Wax should thoroughly cover the entire graft to prevent moisture loss.

## **Splice Approach Graft**

Camellias are most often propagated with this technique. This method uses two separately grown plants with equal stem sizes. On each stem a 1- to 2-inch piece is removed through the bark to the cambium. The cambium layers must be matched for contact. The grafted area is then tied with twine or nursery tape. Finally, wax is used to cover the entire area. Each plant is determined to be either the stock or scion. After the union is formed, the root system of the scion is cut and the upper portion of the original stock is removed.

#### **Tongued Approach Graft**

The tongued approach graft is similar to the splice approach graft, but a second downward cut on the stock and upward cut on the scion are made to form a tongue. The cambium layers are matched, and then the graft is covered with wax or parafilm. This may eliminate the need for twine or nursery tape.

#### Inlay Approach Graft

This is applied to plant material in which the bark of the stock is thicker than that of the scion. The same sized piece of bark is removed from the stock and scion. The stems are nailed to prevent cambium layers of each piece from separating. The graft is sealed with wax or parafilm to minimize moisture loss.

# **Wound Repairs**

Many older plants that have been severely damaged either mechanically, environmentally, or pathogenically can be repaired with two grafting techniques.

#### **Inarch Graft**

This is performed on ornamentals in which the root system or basal portion of the tree trunk has been damaged. The size of the plant will determine the number of seedlings required. Seedlings are planted next to the tree to be grafted at a spacing of 5 to 6 inches, if severely damaged. They are normally transplanted while dormant and the grafting procedure can be performed in early spring on active wood. The damaged tree will receive nutrients and water from the seedlings.

The tree is cut at the base about 6 inches long through the bark to the wood. The piece is removed except for a small flap on top. The seedlings should be 1/4 to 1/2 inch in diameter toward the upper half. The sides of the seedlings closest to the tree trunk are cut 6 inches down. A second piece is cut on the opposite side to form a wedge. The seedling is placed next to the trunk with the top covered by the lip of the tree trunk. Nails are used to hold the seedlings in place with wax, sealing the entire graft (Figure 6).

## **Bridge Graft**

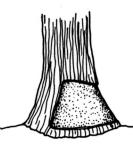
This procedure is performed on trees in which the trunk has been damaged, but the rood system is intact. It is performed when the bark is slipping in early spring. The scions are dormant with one year growth and ¼ to ½ inch in diameter. The damaged area on the trunk is removed. One scion is spaced every 2 to 3 inches around the trunk. Polarity is extremely important in this technique. Nails may be used if needed, and wax should cover the section, preventing exposure to pathogens (Figure 7).

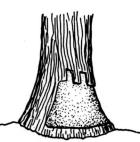
# **Budding**

Budding is similar to grafting, but the scions consist of small pieces of bark and wood with one bud removed from a budstick (a large scion). Often, budding is more successful due to the formation of a stronger union, in which shoots are not broken by the wind as in grafting. Plants which can be budded include roses (*Rosa* spp.), maples (*Acer* spp.), crabapples (*Malus* spp.), honeylocust (*Gleditsia triacanthos*), and many fruit trees. Special budding knives are available, depending on the type of technique used. Pathogen free scions and stocks should be used. The rooted

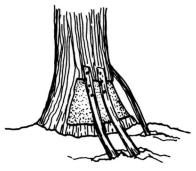


Damaged Basal Portion of Tree Trunk





**Injured Area Prepared to Receive Scions** 



Scions Planted Next to Injured Tree

Figure 6. Inarch graft.



**Finished Inarch** 

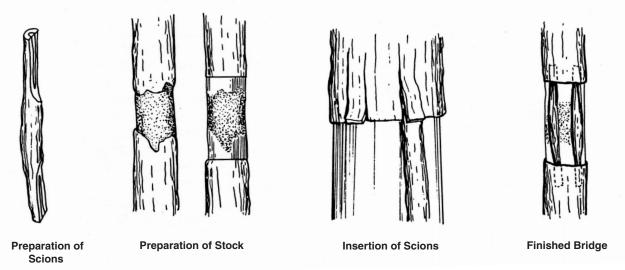


Figure 7. Bridge graft.

stock may range in age from 1 to 3 years. Budding is performed while the bark is slipping, except for chip budding, and polarity should be maintained.

#### Fall Budding

This is done in late summer or early fall when the bark on the stock is in active growth. Vegetative buds should be used (they are **Propagation of Ornamental Plants for Oklahoma** 

pointed and slender as compared to the round and plump flower buds). This technique has advantages over the spring budding:

- budsticks can be readily used and do not have to be stored,
- this is the longest budding period,
- the warmer temperatures enable the union to heal more rapidly, and
- the buds begin to grow early in spring.

The budsticks should be the current season's growth from healthy specimens. The leaves should be removed leaving petiols, which serve as indicators for a successful union. Higher quality buds are used from the middle of the bottom portion of the budstick. Within two to three weeks, unions should be completely healed. Suckers should be removed at the time of budding.

The top of the rootstock should be cut in spring before the new growth appears. This will allow the grated buds to grow by eliminating apical dominance of the stock. The stock should not be cut earlier or the buds will break dormancy too early. The cut should slant away from the bud. As the bud forms a shoot, the rest of the stock should be cut. If any shoots (suckers) appear from the stock, they should be removed. If necessary, stakes may be used to support the plant during winds.

#### **Spring Budding**

Often, spring budding is used to replace failed fall buddings. Spring budding is performed in early spring as the bark is slipping. The budsticks must be dormant and taken before buds begin to swell; therefore, they must be stored in a chilled environment—29 to 32° F (-2 to 0 C). They should be wrapped in dampened peat moss to avoid moisture loss. As with fall budding, the top of the rootstock must be cut about two weeks later. This allows the new bud to grow. Any shoots from the rootstock should be removed.

#### **June Budding**

This method of budding is performed in regions with long growing seasons, such as the southern U.S. and California. It is too hot to be used in parts of Texas; not all areas in Oklahoma may be suitable for this method due to excessive heat. June budding is used on stone fruits, like peach, apricot, and plum, and done before mid-June. Spring and fall budded trees are larger than June budded trees at the end of the growing season because they had a longer growth period.

The buds are from the same season's growth. The bud is placed so that three to four leaves remain below the new bud on the stock. After four days, the stock can be cut back approximately 3 ½ inches above the bud. After 10 to 14 days, it may be cut again. As with fall and spring budding, any shoots from the rootstock should be removed.

#### **T-budding**

T-budding is commonly used for fall, spring, and June budding (Figure 8). Polarity is very important with this budding. The stock trees are <sup>1</sup>/<sub>4</sub> to 1 inch in diameter and actively growing. A T-shaped cut is placed in the stock plant. Normally, the bark is cut 2 to 10 inches above the soil surface. The horizontal cut is made after a vertical 1 inch long cut, so a twist may be given to open the flaps.

The removal of a bud from the budstick is more difficult. A curved cut is made from approximately ½ inch below the bud to 1 inch above the bud so a small part of the wood will be retrieved. A second cut is made across the top to release the bud section. This is called a shield and should not dry. The shield is then inserted into the vertical cut. It is wrapped with tape (do not cover the bud), which must be removed after the bud had formed a union to avoid girdling. Parafilm may also be used for wrapping and can temporarily cover the bud.

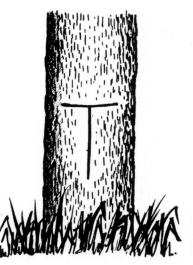
#### **Inverted T-Budding**

This is the same at T-budding, except the cuts made in the scion are inverted. This is used in wet areas, or if the stock plant bleeds (loses sap) excessively. Water and sap are able to drain to avoid disease and rot.

#### **Patch Budding**

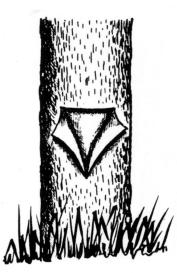
While this technique is more successful than T-budding, it is more difficult and requires a special tool to properly cut a uniform scion and stock section. Patch budding is normally used on thick barked trees—pecans and walnuts—and performed on actively growing shoots.

The scion must fit exactly into the stock opening. The scion should be ½ to 1 inch in diameter and the stock can be up to 4 inches. The stock is cut on smooth bark about 1/3 around. Two horizontal cuts are made to form a rectangle. The bark is then removed. The same is performed with the budstick; however, the bud piece must not be pulled from the stock, but rather gently slid to the side to avoid leaving the bud behind (Figure 9). This section is then inserted firmly into the stock, and parafilm is used to hold it in place. If other wrapping material is used, do not cover the bud, and remove wrapping within 10 to 14 days to avoid girdling.



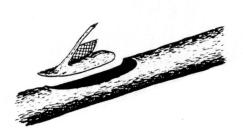
#### 1. Make first cut.

The first cut is made lengthwise of the stem near the ground line, preferable on the north side of the stock. Next, make the cross cut by a rolling movement of the knife, which lifts the corners of the bark where the two cuts cross each other.



#### 2. Open the matrix.

The same area with the matrix opened out to receive the bud.



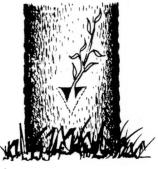


#### 3. Remove the bud.

Remove the bud from the bud stick by making a cut across the top through the bark and peel the bark and bud from the stick, leaving the wood attached to the stock. The bud may be held on the blade of the knife with the thumb on the leaf stub while it is being inserted in the stock. If the bark does not slip, leave the wood.

#### 4. Wrap the bud.

Secure the bud to the stock by wrapping with 1/8 inch rubber budding strips, making at least two wraps below the bud and two above. The band maintains constant pressure, but expands with the growth of the tree.



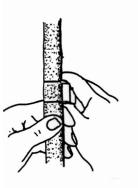
5. New growth starts. Shows the new growth from the inserted bud.



#### 6. Remove the top.

When the forced bud starts to grow, remove the top of the seedling just above the new shoot.

#### Figure 8. T-budding.



#### 1. Double cut the stock.

Select a smooth convenient location on the stock. Make a double cut approximately 1 1/2 inches long by rotating a double-bladed budding knife around the stock.

5. Transfer the patch from budstick to

the patch containing the bud between the

knife and thumb or finger and thumb and

quickly place in the matrix on the stock

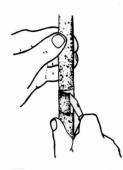
flush with the right side of the connecting (perpendicular) cut made on the stock. The bark flap on the stock is then creased and torn or cut to fit the patch A slight overlap of the bark flap over the patch can

When completely loosened, pick up



#### 2. Double cut the budwood.

Center a plump healthy looking bud between the budding knife blades. Begin on the left side of the bud and, with firm pressure, rotate the knife to the right. Make the double cut through the bark and approximately 1 1/2 inches long.

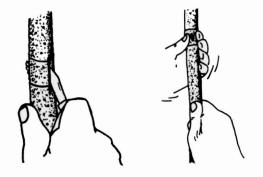


# 3. Complete preparation of stock.

Connect the double cut on the right side with one perpendicular cut. This can be done with one of the budding knife blades or a single-bladed knife.



**4. Raise the bark** on the stock with a flip of the knife point or the tongue attached to the butt end of some budding knives.



# 6. Complete preparation of the patch for removal from the budwood.

Connect the double cut on both sides of the bud with two perpendicular cuts. Loosen the patch containing the bud at all four corners with the knife point. Hold the patch with the bud located between the left thumb and index finger. Twist the left hand toward either direction; with the right hand, rotate the budstick in the opposite direction.



#### 7. Seal the patch.

Wrap with polyethylene plastic budding tape, rubber budding strips, or masking tape. Several other materials, such as waxed cloth and grafting tape, may be used successfully. Pull the wraps firmly and allow them to overlap slightly to seal out excessive air and water and to prevent drying. Cover all exposed areas well. Allow the bud to show through between the wraps.

Figure 9. Patch budding.

stock.

be left

#### **Chip Budding**

This method is used on bark that is not slipping. It can be accomplished in early spring before active growth or indoors for bench grafting. The first cut in the budstick is made 1/4 inch below the bud at a 45-degree angle. The second is made 1/2 inch above the bud and connects with the first. The stock is cut in the same manner. The bud is placed in the stock and wrapped with tape or parafilm. It is important to wrap well to prevent moisture loss or displacement of the chip. Wrapping material must be removed after a union has been made to avoid girdling. If chip budded in the spring, the stock must also be cut after 12 days.

#### Topworking—Topgrafting or Topbudding

Topworking is used to change the cultivar of an existing tree, shrub, or vine. Any budding or grafting technique described previously may be used. For topgrafting, the upper portion of the plant is removed. Grafts are then placed on the lower base of the trunk(s). For topbudding, buds may be placed in healthy branches of the tree. Shoots should be removed as necessary from the stock. The stock should also be cut as described, depending upon the budding technique used.

For an overview of budding, grafting, and related topics for fruit and nut trees refer to OSU Extension Fact Sheets HLA-6204, HLA-6205, HLA-6206, HLA-6227, and HLA-6230.

# **Sexual Propagation**

Seed propagation is the least expensive way to produce large numbers of new plants, but seedling characteristics are usually quite variable which may be a disadvantage. Genetic variability, however, offers an opportunity to select seedlings with new or different features. Seed propagation is a means of reproducing plants that are extremely difficult or impossible to propagate vegetatively.

# Seed Collection and Storage

#### Provenance

Provenance denoted the original geographic source of seed used for propagation. The provenance of a species can be critical for plants that have many races or ecotypes. A species can differ drastically in habit and cold hardiness if grown in the northern versus the southern U.S. It has long been recognized that trees may be inferior if grown far from seed sources; therefore, choose seed sources close to Oklahoma or from areas which mimic Oklahoma's climate. The United States Department of Agriculture has implemented a policy that seed should come from a region with similar:

- climatic influences,
- length of growing season,
- mean temperature during the growing season,
- frequency of summer droughts, and
- latitude.

#### **Seed Maturity and Collection**

There are no set rules for determining when seeds of plants are mature and ready for collection. Changes in size, shape, weight, and color of fruit serve as visual guides to seed maturation. Mature fruit should be collected because viability may decrease after they fall to the ground. The period of seed viability for some plants is short, sometimes only a few months.

Evergreens or conifers, such as some pines (*Pinus* spp.), require special care for seed collections. Many cones will open faster when artificially dried. Most will open at 115 to 140° F (46 to 60° C) after four to six hours, depending on the species. Experiment with lower temperatures and times first, since overexposure to heat may kill the seeds. After the cones have dried, the scales will open and expose the seeds. The seeds should be immediately removed by shaking the cones.

#### **Seed Storage**

Some seeds need not be planted immediately, but can be stored under controlled environmental conditions. Although optimum seed storage conditions differ with plant species, seeds should be separated from fleshy pulp as soon as possible after collection. Southern magnolias (*Magnolia grandiflora*) and yews (*Taxus* spp.) are examples of plants with fleshy fruit. The flesh or pulp should be removed to aid drying, and because the pulp may contain chemicals that inhibit germination. Removal of the pulp by hand is possible for a small number of fruit, but alternative methods can be used for greater quantities of fruit. The pulp can be removed by allowing the fruit to soften in water and then scraping them over a wire screen. A blender with rubber tubing on the blades can also be used. Another method of pulp removal involves placing the fruit in a container with water and a small amount of coarse sand. Use a wire brush on an electric drill to stir the mixture and remove the pulp. Spread the clean seeds in thin layers in the sun or a warm room to dry.

Optimum storage temperature and seed moisture content varies with species, but generally seeds should be stored at 40° F (5° C) in an environment with 30 to 35 percent relative humidity. Household refrigerators usually maintain temperatures suitable for seed stoage, but the relative humidity may exceed the optimum for some seeds. Seeds can be stored in metal cans, plastic bags, or paper or aluminum foil lined envelopes. A protective fungicide treatment is advised for seed known to be susceptible to fugal diseases. Common seed fungicides for vegetable, fruit, and ornamental crops include Captan, Thiram, and Mancozeb. Read the label listed plants and seed as well as instruction for use.

#### **Seed Germination**

Proper moisture, oxygen, temperature, and sometimes light must be provided for germination. Although optimum conditions differ with plant species, general recommendations can be made. Optimum temperatures for germination of most ornamental plant seeds are 75 to 80° F (24 to 27° C). A variation of 9° F (5° C) between day and night temperatures stimulates the germination of some species. The lower temperature should be during the dark period.

The germination media must hold adequate water, yet drain freely. A mixture of equal volumes of peat moss and builder's sand is suitable, but other materials, such as shredded sphagnum, vermiculite, and perlite used alone or in combinations are satisfactory. The particle size of germination media in relation to the seed size should be considered. A small seed planted in a media with large particle may dry rapidly, even though the media particles are moist, because there is inadequate surface contact between the seed and the germination media. Also, small seeds may move too deep in the container of media that has too large particles. The media should be pasteurized to prevent diseases. Dampening-off, a common disease of seedlings, is caused primarily by the fungi *Pythium* spp. And *Rhizoctonia* spp. As described previously, sterile propagation media can be purchased, or a small quantity can be pasteurized in an oven. Heating a 2-inch layer of moist medium at 170° F (76.7° C) for 1 hour will kill pathogenic bacteria and fungi. The media should be moistened before the seeds are planted, and kept moist, but not too wet, for optimum germination. A fungicide treatment may be justified when specific seedlings are known to be susceptible to soilborne fungi.

Seed should not be planted deeper than one to two time their diameter. Small seed should be scattered over the media surface or planted in rows. Keep in mind some seed like lettuce needs light to germinate and should not be covered up. Medium-sized seeds sown on the surface should be covered with a thin layer of shredded sphagnum or peat moss. Larger seeds should be planted at a depth less than their diameter, since a 2- to 3-inch plating depth is maximum for any species.

#### Seed Dormancy

Although seeds of many ornamental plants in Oklahoma are ready to germinate as soon as the fruit matures, some seeds will not germinate until certain internal or external conditions are overcome. Such seed dormancy can be caused by an impermeable or hard seed coat which inhibits water movement into the seed or physically restricts embryo expansion. Seeds may also contain chemicals that inhibit germination. Some chemical inhibitors are water soluble and can be leached from the seeds by soaking them in water. Other inhibitors must be degraded or modified by exposure to certain environmental conditions such as cold temperatures. Seeds can exhibit dormancy due to an immature embryo, in which case proper storage allows further embryo development. Seeds can also be dormant due to more than one factor.

## **Double Dormancy**

Some seeds require periods of warm and moist conditions followed by cool, moist stratification for germination to occur. The earlier warm period allows for chemical and microbial weathering, which begins to break down the seed coat. The cool, moist conditions allow for

embryo dormancy to be completed. Many species respond to acid scarification followed immediately by cold stratification. In some cases, several months time has been gained.

# **Water Soaking Treatment**

Seeds soaked in water may have their seed coat dormancy overcome without further special treatment. The seeds should be planted promptly after the treatment is completed. Many seed species can be placed in room temperature water overnight and will become sufficiently softened. However, others require a hot water treatment. Water may be heated to 212° F (100° C), and the seeds then placed in the container. At this point, the container should be removed from the source of heat to allow the water to cool down slowly. Some seeds have responded favorably to being placed in boiling water for a few minutes; however, this is risky and has a high potential for destroying the seeds' embryos. Soaking times vary greatly with species, and unless reported in the literature, the grower must experiment for the ideal treatment.

# **Scarification**

Seed dormancy allows seeds to germinate only when conditions are optimal for germination and seedling growth. Dormancy caused by a hard seed coat can be overcome by penetrating or breaking the seed coat, known as scarification.

## **Acid Scarification**

Acid treatments are often needed for species that have an impermeable or hard seed coat. Although concentrated sulfuric acid is highly effective, precautions must be taken. The acid is corrosive and reacts violently with water. NEVER ADD CONCENTRATED ACID TO WATER. ALWAYS ADD WATER TO THE ACID TO AVOID EXPLOSION. Protective measures must be taken to safeguard the skin and eyes. Always wear safety goggles.

Place dry seeds into glass containers and immerse them in the concentrated acid. The solution may need to be stirred occasionally, with caution, to avoid splattering of the acid. The exposure time of the seeds to the acid varies greatly with the species. Depending upon the thickness and resistance of the seed coat, a few minutes to a few hours may be necessary. These time intervals have been established for many commonly grown ornamentals. When the seed coat has been significantly eroded, all seeds should be removed from the solution. Seeds must be washed with running water for several minutes and planted. If the propagator is working with a species not reported in the literature, more information may be obtained from a commercial nursery or an Extension office.

#### **Mechanical Scarification**

Although acids and hot water treatments are sometimes used in commercial nurseries to break or soften the seed coat, mechanical scarification is most suited for the landscape gardener. Small numbers of seeds can be scarified by rolling them on a cement floor using a brick or board, by rubbing the seeds with sandpaper, or by cutting the seed coat with a knife. Mechanical devices may be purchased or constructed to scarify larger numbers of seeds. The seed coat should be dull in appearance after scarification, but not deeply pitted or cracked enough to expose or injure the embryo. Scarified seeds will not store after scarification and should be planted as soon as possible after treatment.

# **Stratification**

Seeds of many Oklahoma plants require a cold period before they will germinate. This requirement is satisfied by cold stratification storing the seeds in a cold, moist environment. Seeds are mixed with moist sphagnum peat or vermiculite after a 12- to 24-hour soak in wa-

Table 5. List of plants that need scarification or stratification for germination.

Scarification	Stratification
Cotoneaster Hawthorns Goldenrain tree Wisteria Lupine Baptisia Dogwood Acacia Black locust Camellia Kentucky coffee tree Persimmon Redbud	Veronica Lavender Phlox Rudbeckia Delphinium Ascelpias Penstemon Gaura Silene Phlox Sedum Hellebore

ter at room temperature. It is also advisable to spray the seeds with a protective fungicide treatment before putting them in refrigerated storage. The seeds should be stored for two to six months at 37 to 40° F (3 to 5° C). Temperatures in household refrigerators are usually adequate. Suitable containers for stratification are flats, trays, boxes, or cans that provide aeration, prevent drying, and allow drainage, but do not retain excessive moisture that would cause rot. Polyethylene bags nor more than 0.004 inch (4 mil) thick may also be used. Seeds should be planted immediately after removal from refrigeration.

#### **Seedling Establishment**

Seed germination and early seedling development are best accomplished in a moist environment with moderate temperatures (75 to 80° F or 24 to 27° C). Although light is not required for germination of many seeds, high intensity light is necessary to produce stocky, strong seedlings. Low intensity light will result in weak and spindly, pale green seedlings.

Seedlings planted close together quickly become crowded, resulting in slow growth and weak, spindly stems. Crowded seedlings must be transplanted with wider spacing into flats or individual or multi-celled containers. Seedlings can be grown in these containers until they are mature enough to transplant into larger containers or the landscape.

Tender seedlings transplanted without a transition period into a hot dry environment have a poor survival rates. The environment in which seedlings are grown should be modified, and gradually hardened until it is similar to the environment into which they will be transplanted. Watering frequency should be decreased gradually followed by a gradual increase in light intensity.

# Small-Scale Propagation Units

The key to successfully rooting cuttings and seed germination is a moist environment maintained at a favorable temperature. Environmental control is less important for other propagation methods, such as layering, because the mother plant provides some degree of support to the developing new plant. However, most cuttings and young seedlings are susceptible to environmental stress and will survive only if an appropriate environment is provided. An environment with a relative humidity near 100 percent will minimize water loss from cuttings and developing seedlings, although water loss is less critical for seedlings than cuttings. Cuttings do not have roots; therefore, they cannot take water from the media to replace that which is lost through the leaves. If high rates of water loss occur, cuttings will dry out.

Temperature influences the physiological activity of plants. Excessively high or low temperatures injure plants or slow their growth and development, but temperatures in the range of 70 to 80° F (21 to 27° C) stimulate optimum growth and development for most plants.

The home gardener can provide a warm, humid environment for seed germination and rooting of cuttings by constructing or purchasing small scale propagation units. These units are inexpensive, require little attention, and are convenient to use in the home landscape or indoors.

A propagation unit can be made from a terrarium or aquarium. These structures are usually constructed of glass or Plexiglass, but a suitable structure could be constructed of wood and glass or plastic. Approximately 2 to 4 inches of propagation media can be placed in the bottom of the tank, and cuttings may be stuck or seed sown directly in the media. Alternatively, propagation media can be placed on top of 2 inches of gravel in the tank. A glass or plastic cover should be put on the container after adequate moisture has been added.

Large plastic pots and a plastic bag can be used to create a suitable propagation environment. Stick the cutting in a moist propagation medium and the container in a large, clear plastic bag. Wire hoops or stakes can be used to prevent the plastic bag from laying on the cuttings or seedlings.



Photo 3. Propagation unit.

A plastic bag alone can serve as a propagation environment. Simply place moist propagation media in the bottom of the bag, insert the cuttings, and tie the top of the bag closed.

Put the structure in diffused light and NEV-ER in full sun. The temperature in these sealed units will rapidly become too high in full sun, and cutting or seedling injury or death will result. Units kept indoors should be placed near a north window or under fluorescent lights for 12 to 16 hours per day. Temperatures of 65 to 80° F (18 to 27° C) should be maintained. Although these units are designed to prevent moisture loss, routine examination of the moisture level is suggested. Add moisture if no water had condensed on the inside of these units overnight, or if the propagation media appears dry.

For the advanced hobbyist and commercial grower, refer to OSU Extension Fact Sheets HLA-6708 for an overview of humidity chambers and automated mist systems.

## **Field Seeding**

Many evergreen and deciduous plants are sown by seed in outdoor field beds. Expenses are usually reduced, since less labor is required for transplanting and greenhouse space can be used for other more tender plants.

The soil should be fertile, well-drained, and of a good quality for seed germination and growth. Many propagators use raised beds for better drainage. The common size of these beds is 3 ½ to 4 feet wide, which allows easy access. For maximum sun exposure, north-south orientation is recommended.

There are many herbicides available for weed control in both pre-emergent and postemergent forms. Their labels specify the proper dosage and rate of control for the various growth stages of the plants.

The outdoor environment can provide the conditions necessary to meet dormancy

Spores

Ferns reproduce with spores. A group of spores is called sori. The reproduction process of ferns is both asexual and sexual. Sporophytes are the conspicuous part of the plant which produce spores. The sporophytic generation is the first part and is called the asexual stage. Later, gametophytes form which cannot be seen and are referred to as the sexual stage of ferns. requirements depending upon the seed or cutting, season, and area. Thus, labor can be minimized.

The rate for sowing seeds will depend upon the growers situation and the seeds' requirements. The following formula may be used to determine a ratio for seeds rates:

Weight of seeds to sow per area = density desired [plants per unit area]/[purity percent x germination percent x field factor x number of seeds per unity weight (seed count)].

Percent purity and germination are expressed as a decimal in this equation and may be obtained from the seed producer. The seed count and density desired will be determined by the grower. The field factor is formed by the experience of lost ratios from previous crops.

Some seeds may require a fungicide treatment to ensure seedling survival. Various techniques may be used for planting the seeds—hand broadcast, seeders, hand drilling, or tractor precision drills. The general rule for seed depth is applied—twice the seed diameter. Hand or tractor rollers may be used after sowing to ensure soil contact with the seed.

Continued surveillance of the bed is necessary to monitor for fertilizer, herbicidal, and insecticidal requirements. The plants will remain in these beds for one to thee years and will be replanted to another location for continued growth or sale. They may be packaged as bare root, balled and burlaped, or containerized plants.

Ornamental, fruit, and nut trees used as rootstock liners are planted in rows rather than beds. The seeds or cuttings are placed 3 to 6 inches apart in row 4 feet apart. They can be grafted and budded directly in these rows once they have grown to the desired size.

Spores appear throughout the year, depending upon the temperature, humidity, and maturity of the spores. They may be found on the underside of fronds and are tan, orange or brown in color.

Water is required for fertilization. The spores, or sori, may be removed from the frond and mixed with tap or distilled water. The solution is distributed onto the growing medium

with a pipet or syringe. The growing medium must remain moist and a covering should be used to maintain constant humidity. The ferns may be thinned with tweezers as they mature and transplanted accordingly.

# Tissue Culture – Micropropagation

Tissue culture differs from traditional types of propagation by the high degree of control that is required for a successful outcome. Each step of the process is manipulated to maximize the size, quantity and quality of the new plants.

Tissue culture is based on totipotency. Totipotency is the concept that every living cell has the capacity to reproduce the entire plant, regardless of whether the cell is harvested from the flower, stem, root, etc. although this is true in theory, cells are difficult to manipulate into regenerating a new plant, at least in an economical fashion. Therefore, because of the technical difficulty in tissue regeneration and the often higher expense compared to traditional propagation methods, tissue culture or micropropagation is still limited in use in ornamental horticulture. Most businesses that have tried tissue culture in Oklahoma have since discontinued this form of propagation. Consequently, it is not advisable to attempt tissue culture on a large basis without considerable research into its necessity.

Micropropagation is the correct term for most types of ornamental plant propagation in test tubes (in vitro). This refers to the application of tissue culture technique to the perpetuation of plants beginning with very small plant sections (pieces of stem, leaf, etc.) grown aseptically in a test tube or similar vessel (Photo 4).

# Advantages to Tissue Culture

Mass propagation of specific clones. Commercial laboratories have produced millions of micropropagated plants per year, which is particularly important when:

- the natural rate of increase or tradition methods of propagation are slow, i.e. palms, ferns, orchids, etc.,
- a new cultivar needs to be produced quickly in sufficient numbers to market, and
- a cultivar cannot be easily or economically propagated by traditional methods.



#### Photo 4.

Production of disease-free plants. This permits:

- eliminating disease from the cultivar initially,
- preventing reinfection,
- allowing plants to go through quarantine barriers.

Clonal propagation of parental stocks for hybrid seed production, which is done for many of the commercially important vegetable crops.

Provide year-round nursery production. Production can be more easily scheduled to accommodate market demands.

# Disadvantages of Tissue Culture

- Expensive facilities with highly trained workers are required.
- Production costs may be very high due to much hand labor needed for specific procedures.
- Contamination is a constant threat and when left unchecked, could destroy large numbers of plants in a very short time.
- The variability in individual plants occurs.

# The Four Stages of Micropropagation Establishment

The explant (piece of shoot, stem, leaf, etc.) is placed in sterile culture (Photo 5). Initiation of callus, shoots, roots, etc. must occur for successful establishment. This can often be manipulated by altering the ingredients and their concentrations within the growing media. The complex media includes the following ingredients:

- mineral nutrients—basically the same ingredients used in traditional fertilizers such as nitrogen, phosphorus, potassium, etc.,
- carbohydrates,
- vitamins, and
- hormones and growth regulators.

You can mix your own ingredients or buy premixed ingredients. Common mixes include Murashige and Skoog (MS), Gamborg B-5, and Woody Plant Material (WPM). In addition to the above mentioned ingredients, agar is added to solidify the media, auxins and cytokinins are added to induce roots or shoots, respectively, deionized water free of impurities is used, and the pH is adjusted to a range of 5.3 to 5.8. The media must be sterilized with an autoclave. However, some ingredients require cold sterilization (by chemicals or filters) to remain stable.

Explant selection is also made during this stage. Pieces of leaves with cotyledons, flower scapes, shoot tips, lateral buds, embryos, and other plant organs or tissues can be divided into several explants. For woody plants, the actively growing shoot tip is often the best explant source. Explant preparation must also occur under sterile conditions. Special transfer hoods minimize the chance of pathogens settling on carefully disinfected explants.



Photo 5.

#### **Shoot Multiplication**

As this stage, the original explant gives rise to a cluster of microshoots that must be divided. Upon division, each microshoot is transferred into new media which is often of a different composition than the original media. Media is generally higher in cytokinins than auxins.

#### Pretansplant, In Vitro Rooting

During this stage, the very tender microplants are hardened off to be prepared for the transfer from test tubes to much harsher conditions outside of the culture environment. Normally, the third step requires conditions quite different from the previous two steps. The media's ingredients are again adjusted with higher auxins than cytokinin concentrations for the transfer process, and the light intensity is often increased.

### Transplant, Acclimation, and Ex Vitro Rooting

Rooted or sometimes unrooted microshoots are removed from their growth vessels and transplanted into a conventional pasteurized growing media. This is a critical stage and the shoots must be allowed to slowly adjust to the new harsher conditions. Direct light should be avoided, and the shoots often benefit from mist or a humidity chamber for a few days. Once properly acclimated (adapted to the new environment), the young plants should be treated like any other seedling, plug, or newly rooted cutting. Some authorities place this final stage in with the third stage described earlier.

# Major Types of Tissue Regeneration Shoot Apex Culture

This procedure is widely used for producing disease-free stock of geraniums (*Pelargonium x hortorum*), chrysanthemums (*Chrysanthemums* spp.) and other ornamentals. Normally, the shoot apex is trimmed to 0.5 to 1.0 mm under a dissecting microscope. Sections of the shoot apex serve as explants.

#### **Axillary Shoot Proliferation**

Lateral meristems are stimulated to break dormancy and develop new shoots. Because many more plants can be harvested by this means, it is very common for commercial propagation of ornamentals and certain food crops.

#### **Adventitious Shoot Initiation**

Explants of various tissues can be stimulated to develop callus, which then give rise to adventitious shoots. An example is African Violet (*Saintpaulia ionantha*) leaves, which can be divided into many explants. Each explant will develop microshoots with the proper media and environmental conditions.

#### Organogenesis

Adventitious shoots or sometimes roots are formed from callus cells. Plant parts are first placed in culture and callus cells proliferate. From these callus cells, adventitious shoots or roots are formed depending on the hormonal balance in the growing media.

#### **Embryogenesis**

Embryos can be formed outside of the normal fertilization process. Somatic embryos can be harvested from reproductive tissues, vegetative callus of mature plants, and seedling tissues.

# Basic Laboratory Requirements

- 1. Laboratory working areas.
- 2. Transfer hoods and designated areas for aseptic (sterile) explant transfers.
- 3. Autoclave for sterilizing media, instruments, etc.
- 4. Distilled and deionized water, various chemicals, etc.
- 5. Various instruments, depending on which procedures used.
- 6. Culture rooms that can be carefully monitored for temperature, light, and humidity.

Most commercial tissue culture laboratories have invested several thousands of dollars in basic beginning costs. The high initial and ongoing costs, coupled with the high risk of contamination, prevent most growers from ever practicing large scale tissue culture. Only species that can ensure the grower a profit from the investment can be considered candidates for these special propagative procedures.

It is advisable to seek counsel from propagators currently using tissue culture procedures. Also, refer to publications listed at the back of this chapter for further information on applications and procedures for micropropagation.

# Importance of Sanitation

Sanitation is an important part of any successful propagation program, whether it be traditional or tissue cultivated. Four considerations are needed to achieve and maintain such sanitary conditions.

First, when propagating a particular plant, either by seed or cuttings, the propagator should begin with clean stock plants or seed. Cuttings or seed taken from these plants should be free from disease organisms and insects. Various treatments can be used to eliminate such troublesome pests. Even though the plant material may appear to be free of noxious organisms, fungicidal treatment prior to, or after placement in the propagating medium is a suggested precautionary measure.

Second, cuttings should be inserted, or seeds sown, in a propagating media that is free from weeds or weed seeds, nematodes, and various disease organisms (such as bacteria, fungi, or viruses). Pasteurized media can be prepared by heat (steam or electric) treatment or chemical fumigation.

Third, one should inspect propagation containers, propagation benches, media bins, and tools prior to use for sterility. After a crop has been raised or cuttings have been rooted and removed, propagation benches should be disinfected prior to reuse. All propagation media kept in bins in a greenhouse should be pasteurized prior to storage, and periodic sterilization of such bins is recommended. Tools should be disinfected regularly, especially when they are used on sensitive species. Many propagators dip their tools in disinfectant after each cut and between every plant. Numerous disinfectants are available on the market depending upon the situation, however, a 10 percent household bleach concentration works well for most applications. Plants or cuttings of an unknown origin should not be brought into the propagation house because they might be infected with various disease organisms and insects, which could infect healthy cuttings or plants.

Finally, general cleanliness must be maintained to avoid recontamination or introduction of noxious organisms in the propagation area. As soon as a disease problem is observed, diseased plants or cuttings should be immediately removed. Identify the disease organism(s) and begin proper treatment of cuttings or plants as a preventative measure. Weeds should not be permitted in the propagation house or area as they can serve as a source of weed seed, disease, and insect infestation. Plant debris can harbor insect and disease organisms and should always be removed. Water hose nozzles should not come in contact with the floor because they could pick up and later distribute harmful pathogens. If the nozzles do become contaminated, they can be disinfected with the same solution as the tools. Hoses should be placed on a wall bracket when not in use.

In addition, try to limit or discourage unauthorized individuals from visiting the propagation house. The constant movement of people through a propagation house can be a potential source of disease or insect introduction, particularly if these people have had prior contact with disease or insect infected material. Lastly, do not allow workers to smoke around the plants because tobacco may spread various disease organisms.

# **Plant Patents**

The Plant Patent Law was enacted in 1930 to protects the originator of new plant materials against competition. This 20-year protection has proved to be an incentive for a considerable amount of work into new plant materials for everyone to enjoy.

If a grower finds a mutation on any plant, the discovery is protected under a plant patent. Any distinct and new variety of plant (including cultivated mutants, hybrids, and newly found seedlings), other than a tuber, propagated plant, or a plant found in an uncultivated state (the wild), is qualified under this law. Plants which can be propagated asexually, and in some cases sexually, are afforded protection under the law. The U.S. Plant Variety Protection Act, in addition to the original plant patent law, affords protection to certain seed propagated cultivars.

Virtually any new characteristic in a plant enables it to be patented. Greater resistance to environmental conditions, immunity from disease, and any change in size or color of any plant part(s) are just a few of the possibilities.

The applicant for a plant patent must be the individual who identified and then propagated the unique plant. Application procedures and fees can be found by contacting the patent offices or by visiting their web sites. Patent offices are listed in the back of this publication.

# Plant Propagation Literature

- Bailey, L.H. *The Nursery Manual: A Complete Guide to the Multiplication of Plants.* New York: MacMillan Co. 1967.
- Ball, V. (ed.) *The Ball Red Book* (13<sup>th</sup> Ed.).
  West Chicago, IL: George J. Ball, Inc., 1975. 502 pp.
- Browse, Philip M. *Plant Propagation*. New York: Simon & Schuster. 1979. 96 pp.
- Chen, S., D.A. Evans, W.R. Sharp, P.V. Ammirato, and M.R. Sondahl (eds.). *Handbook of Plant Cell Culture*—Volume 6. American Nurseryman Publishing Company.
- Dirr, Michael A. and Charles W. Heuser, Jr. *The Reference Manual of Woody Plant Propagation* (2<sup>nd</sup> ed.). Athens, Georgia: Varsity Press, 2006. 239 pp.
- "Forest tree seed orchards," Directory of Industry, State and Federal Forest Tree Seed Orchards in the U.S. Washington, D.C.: USDA. (26).
- Free, M. *Plant Propagation in Pictures*. (Rev. and ed. By M.J. Dietz). Garden City, New York: Doubleday and Co., Inc., 1979.
- Hartmann, H.T., D.E. Kester, F.T. Davies, and R. L. Geneve. *Plant Propagation Principles and Practices*. New Jersey: Prentice – Hall Inc., 7<sup>th</sup> ed., 2002.
- Lipp, L.F., P.K. Nelson and C.S. Umbreit (ed.). *Handbook on Propagation*. New York: 1958.
- Mahlstede, J.P. and E.S. Haber. Plant Propagation. Chicago: American Nurseryman, 1957. 412 pp. (10).
- Mayer, A.M. and A. Polijakoff-Mayber. *The Germination of Seeds.* New York: Pergamon Press, 1975.
- Wells, James S. *Plant Propagation Practices.* Chicago, IL: American Nurseryman Publishing Co., 1985. 367 pp.

## Oklahoma Propagation Supply Companies

American Plant Products 9200 Northwest 10<sup>th</sup> Street Oklahoma City, OK 73127 (405) 787-4833 www.americanplant.com

#### Other Propagation Supply Companies

A.H. Hummert Seed Co. 2746 Chouteau Avenue St. Louis, MO 73103 (800) 325-3055 www.hummert.com

Herbst Brother Seedsmen, Inc. 1000 N. Main Street Brewster, NY 10509 (914) 279-2971

A.M. Leonard, Inc. P.O. Box 816 Piqua, OH 45356 (800) 543-8955 http://amieo.com

Hortus USA 245 W. 24<sup>th</sup> Street New York, NY 10011-1717 (212) 929-0927 www.hortus.com

Agri-Products, Inc. 1805 Easy Street Wenatchee, WA 98801 (509) 663-5096 www.agriproductsinc.com

Smithers-Oasis U.S. Grower Product Division P.O. Box 118 Kent, OH 44240 (800) 321-8286 www.smithersoasis.com/us

Ben Meadows Company 3589 Broad Street Atlanta, GA 30366 (800) 241-6401 www.benmeadows.com

Stuewe & Sons, Inc. 2290 S.E. Kiger Island Drive Corvallis OR 97333-9461 www.stuewe.com

Capital Agricultural Service & Supply Co. (CASSCO) 611 Sutton Bridge Road Gadsden AL 35901 (800) 633-5888 http://www.cassco.cc/

Walter E. Clark & Son P.O. Box 756 Orange, CT 06477 (203) 795-1235 www.waltereclark.com

E.C. Geiger Box 285 Harlesville, PA 19438 (215) 256-6511 www.hortnet.com/ecgeiger

#### **Tissue Culture Supply Houses**

Fisher Scientific 2000 Park Lane Dr. Pittsburg, PA 15275 (800) 766-7000 www.fishersci.com

Labconco Corporation 8811 Prospect Kansas City, MO 64132 www.labconco.com

Magenta Corp. 4149 W. Montrose Ave. Chicago, IL 60641 www.magentacorp.com

Sigma Chemical Co. P.O. Box 14508 St. Louis, MO 63178 www.sigmaaldrich.com

United States Plastic Corp. 1390 Neubrecht Road Lima, OH 45811 www.usplastic.com

#### Sources of Reagent Grade Auxins

Fisher Scientific Company 3315 Winton Road Raleigh, NC 27604 www.fishersci.com

Sigma Chemical Co. P.O. Box 14508 St. Louis, MO 63178 www.sigmaaldrich.com

United States Biochemical Corp. P.O. Box 22400 Cleveland, OH 44122 (800) 321-9322 www.Usbweb.com

# Seed Companies (shrubs and trees)

F.W. Schumacher Co., Inc. 36 Spring Hill Road Sandwich, MA 02563-1023 (508) 888-0659 www.treeshrubseeds.com

Lawyer Nursery 950 Highway 200 W. Plains, MT 59859 (406) 826-3881 www.lawyernursery.com

Plants of the Southwest 1570 Pacheso Street Santa Fe, NM 87501 www.plantsofthesouthwest.com

Sheffield's Seed Company, Inc. 273 Rt. 34 Locke, NY 13092 (315) 497-1058 www.sheffields.com

Silvaseed Co. P.O. Box 118 Roy, WA 98580 www.silvaseed.com

Swift Seed Company Box B Jaroso, CO 81138 (719) 672-3739 www.deanswiftseed.com

Wild Seed 2021 South Forest Ave. Tempe, AZ 8522

#### **Seeders**

Berry Seeder Company Rt. 4 Box 230 Elizabeth City, NC 27909 (800) 327-3239 http://www.berryseederco.com/

B.F.G. Supply Company 14500 Kinsman Road Burton, OH 44021 (800) 321-0608 www.bfgsupply.com

#### Associations

Oklahoma Greenhouse Growers Association 400 N. Portland OKC, OK 73107 (405) 942-5276 www.ogga.org

Oklahoma State Nurserymen's Association 400 N. Portland OKC, OK 73107 (405) 942-5276 www.oknla.org

National Association of Plant Patent Owners 1250 I Street, NW, Suite 500 Washington, DC 20055 www.anla.org/industry/patents/index.htm

International Plant Propagators Society Center for Urban Horticulture University of Washington, GF-15 Seattle, WA 98195 www.ipps.org

#### **Patent Offices**

Plant Variety Protection Office Room 500 National Agricultural Library Building Beltsville, MD 20705 (310) 344-2518

U.S. Patent and trademark Office Washington, DC 29231 (703)-557-4636

Plant Variety Protection Office http://www.ams.usda.gov/AMSv1.0/pvpo

U.S. Patent and Trademark Office http://www.uspto.gov/

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