Germination of Doubly Dormant Woody Ornamental Seeds

Steven E. Newman, Ph.D.
Department of Horticulture, Mississippi Agricultural and Forestry Experiment Station, Mississippi State University, P O Drawer T, Mississippi 39762-5519

INTRODUCTION

Many temperate zone plants have developed highly specialized organs for protection from would-be foragers, drought, and temperature extremes. For example, *Opuntia* spines protect it from animals; barley awns help dissipate heat; corms, bulbs and tubers serve as reservoirs of food and water; winter bud scales protect shoot apices form cold winter winds; and seed serve to transmit genetic information accumulated in response to the environment to new generations (Janick, 1986).

Probably the most important adaptation and the most intriguing mechanism temperate plants have developed is the ability to survive the long winter months in a state of rest Herbaceous perennials survive by underground storage structures, woody plants by winter buds and massive root systems, and annual plants by seeds (Khan, et al. 1977, Oosting, 1956).

Seeds are the primary means of surviving and increasing the population. Seeds rest through conditions unfavorable for growth by mechanisms best described in two categories, quiescence and dormancy "A quiescent seed is readily germinable with non-specific trigger agents, such as sufficient moisture and optimum temperature" (Jann and Amen, 1977) Annual plants, for the most part, fall within this category in that they germinate rapidly, grow, flower, set seed, and die in one season These plants produce many seeds so that at least a few will germinate and mature to produce more seed

True seed dormancy is the inability of a seed to germinate, even under conditions that are normally considered favorable for germination (Jann and Amen, 1977). Dormant seed germinates in response to specific triggering agents, which are environmental factors correlated to their respective inhibition mechanisms.

After the dormancy requirements have been satisfied, the seed is capable of germination Physiologically, germination is the sequential process including resumption of previously depressed metabolic pathways and the differentiation of oxidative and synthetic pathways This ultimately brings the embryonic axis into a state of active growth, which was temporarily suspended during quiescence or dormancy Morphologically, germination is the transformation of an embryo into an actively growing seedling (Jann and Amen, 1977).

SEED QUIESCENCE AND DORMANCY

The common inhibitor mechanisms include the following:

Seed Coat Impermeability.This mechanism involves the inability of water or gasses to enter the seed either by an impermeable seed coat or by seed coat resistance to swelling (Khan, et al., 1977)

Most species with an impermeable seed coat will germinate rapidly after the seed

coat is made permeable (Hartmann, et al , 1990; Krugman, et al., 1974; Macdonald, 1986). Seed coat impermeability is reversed in nature by various environmental factors including mechanical abrasion, alternate freezing and thawing, attack by soil microorganisms, passage through the digestive tract of animals and fire (Hartmann, et al., 1990). Most seed of the Fabaceae (Leguminoseae) family have an impermeable covering, as do many species of Sapotaceae, Ericaceae, Rhamnaceae, Anacardiaceae, and Sapindaceae families (Krugman, et al., 1974) Seed coat impermeability is due to presence of a layer of palisade-like macrosclerid cells (Hartmann, et al., 1990). They are especially thick-walled with a waxy external cuticle.

Any process that alters the seed covering to make it permeable to water and gases is called scarification. Seeds can be mechanically scarified by rubbing on sandpaper, cutting with a file, or cracking in a vise (Hartmann, et al., 1990; Krugman, et al , 1974; Macdonald, 1986). Large lots of seeds can be scarified in large motor-driven tumblers with an abrasive lining (Macdonald, 1986). After the scarification process is complete, the seed coat should be dull and not deeply pitted or cracked (Hartmann, et al., 1990) Scarified seeds are more susceptible to pathogens and do not store as well as non-scarified seed (Krugman, et al., 1974; Macdonald, 1986).

Water soaks will soften some hard seed coats (Macdonald, 1986) Hot water (170-212°F) can sometimes be used but care must be taken to avoid high-temperature injury to the embryo. Seed that has been treated with a hot water soak is alluring to rodents so must be protected from them.

Acid scarification is a common method of modifying seed coats and is easy to perform. Seeds are soaked in full strength sulfuric acid (H_2SO_4, specific gravity 1.84) for different lengths of time based upon the seed coat thickness (Hartmann, et al., 1990; Krugman, et al., 1974; Macdonald, 1986) Acid scarification is a convenient method that leaves the seed clean, firm and unswollen (Krugman, et al , 1974). The drawbacks of acid are the safety hazards to personnel and the need to determine accurately the length of treatment time (Krugman, et al., 1974). As with hot water treatments, seeds that have been treated with acid may attract rodents.

After-Ripening Requirement. This type indicates the presence of an undeveloped or rudimentary embryo, the presence of chemical germination inhibitors, or the lack of chemical stimulus Any one or combination of these will maintain the seed in dormancy until a cold temperature stratification (moist-chilling) period occurs. This results in a maturation of the embryo, removal of inhibitors, or the production of chemical promoters (Khan, et al., 1977).

After-ripening requirements are usually satisfied by stratification (moist-chilling). Temperatures from 33 to 41°F allow certain physiological changes within the embryo to occur (Hartmann, et al., 1990). Most of the changes are biochemical, but seeds of many species have immature embryos that must grow and develop before germination is possible (Krugman, et al., 1974; Macdonald, 1986). Rudolf (1961) reported that 60% of over 400 woody plant species tested required some after-ripening to prompt germination

Many different techniques have been developed and many are specific to the species involved, the equipment to be used, or personal preference. The critical

factors require the use of a high-moisture-holding medium, adequate aeration, and proper temperatures (Hartmann, et al., 1990; Krugman, et al., 1974).

Many chemicals, such as gibberellin (GA), cytokinin, and ethylene, have been used to promote seed germination Of all these, GA has the most pronounced effect (Hartmann, et al., 1990). ". . . GA will stimulate germination in seeds where dormancy or quiescence is imposed by a wide variety of mechanisms, eg. incomplete embryo development, mechanically resistant seed coats, presence of germination inhibitors, and factors relating to the physiological competence of the embryo axis (Jones and Stoddart, 1977). GA has been used to replace cold stratification of *Corylus avellana* seed (Brinkman, 1974; Frankland and Wareing, 1966; Jarvis and Wilson, 1977). Exogenously applied GA activity in *Corylus* appears to affect germination at two sites, the embryonic axis, stimulating growth, and the cotyledon, stimulating carbohydrate metabolism (Jarvis, et al., 1978). Other woody species where GA soaks of 200-500 ppm for 24 hours have been shown to replace moist chilling include *Carpinus caroliniana* (Bretzloff and Pellett, 1979), *Vaccinium ashei* (Ballington, 1976), *Prunus persica* (Hundal and Khajuria, 1979), *V. macrocarpon* (Devlin and Karczmarczyk, 1975), *Fagus sylvatica* (Franklan and Wareing, 1966), *Liquidambar styraciflua* (Burns, 1967), *Ulmus* sp., *Pinus sylvestris, Picea glauca,* and *Picea pungens* (Grover, 1962).

Exogenous GA has been demonstrated to improve seed germination and shorten the moisture-chilling period of several woody ornamental plants, and GA combined with moist-chilling may be used to germinate seed of species from GA that applications has been demonstrated in many woody species, including *Quercus rubra* (Vogt, 1970), *Pyrus pashia* (Dhillon and Sharma, 1987), *C. caroliniana* (Bretzloff and Pellett, 1979), *Acer tataricum* (Nikolaeva, et al., 1973), and *Ostrya virginiana* (Newman, 1981).

Seeds of many woody species have several forms of dormancy and are called doubly dormant (Bonner, et al. 1974; Dirr and Heuser, 1987; Hartmann, et al., 1990; Macdonald, 1986). More than 14% of the species and cultivars listed by Dirr and Heuser (1987) require combinations of dormancy treatments to induce seed germination. The most common form is an impermeable seed coat along with an internal dormancy requiring after-ripening. Any of the seed coat scarification treatments followed by moist chilling will satisfactorily remove both forms of dormancy (Bonner, et al. 1974; Dirr and Heuser, 1987; Hartmann, et al., 1990; Macdonald, 1986).

Some species have a hard seed coat that is not completely impermeable and a warm stratification period of several months may degrade the coat enough for organic acids and microorganisms to act on the seed coat or "bony" endocarp and allow imbibition (Hartmann, et al., 1990; Khan, et al., 1977; Krugman, et al., 1974; Macdonald, 1986).

Warm followed by cold stratification is a treatment used on woody species with under-developed embryos The warm temperature promotes embryo maturation. The epicotyl is dormant in many species, such as *Davidia involucrata, Paeonia suffruticosa, Chionathus retusus,* and *Aesculus parviflora* (Macdonald, 1986), but the radicle is not (Dirr and Heuser, 1987, Krugman, et al., 1974; Macdonald, 1986). A warm temperature period (constant 50-77°F or 68 and 86°F alternating diurnally) allows the radicle to emerge and become established. The following cold temperature period removes epicotyl dormancy (Krugman, et al., 1974; Macdonald,

1986). Seeds naturally receive this warm-cold treatment during the summer-winter seasons (Macdonald, 1986). Germination generally then occurs 18 months after fertilization.

Nandina seeds, which have an under-developed or rudimentary embryo (Dirr and Heuser, 1987), are best handled by collecting the berries during late fall to midwinter, and storing them in a cool and dry place until the following spring. Then soak the berries for 24 hours in water, macerate the flesh from the seed, sow the seed in flats or pots of pine bark, and place the containers under shade with irrigation The seed will then germinate in late October. During the warm summer months, the rudimentary embryo matures and germinates during the cooler months

Doubly dormant seeds collected during the late summer and planted immediately, taking advantage of the warm soils, will often germinate the following spring. However, seed collected, stored and planted in the fall will usually germinate the second spring (Dirr and Heuser, 1987). Early collection may avoid the hard seed coat condition (Macdonald, 1986).

Seed collected while still green, prior to the development of thick seed coats or prior to the accumulation of inhibitors in the seed coat, can be sown immediately for germination during the following spring. Unchilled mature *Malus* will germinate if the seed coat is removed (Powell, 1987) Many species collected green will germinate immediately upon sowing (Sandahl, 1941).

In seeds of species requiring a warm temperature stratification period followed by moist-chilling, exogenous GA has been demonstrated to replace the warm phase and shorten the cold phase (Macdonald, 1986; Newman, 1981; Simancik, 1970; Wicislinska, 1977) GA does not seem to be the only factor in removing dormancy but possibly induces embryo maturation. Moist-chilling then removes any other form of dormancy. GA effects are also enhanced with scarification (Newman, 1981; Simancik, 1970).

Photoblasticity. Photoblasticity is the requirement of exposure to light, which releases or initiates hormonal control of germination (Khan, et al., 1977).

TECHNIQUES FOR BREAKING SEED DORMANCY.

Approximately two-thirds of the tree species native to North America yielding sound seed will not germinate under favorable conditions (Krugman, et al., 1977). Under natural conditions, morphological and physiological changes necessary for the removal of dormancy take place gradually in response to varying environmental factors. "By duplicating key conditions of the natural environment in the laboratory or nursery, dormant seeds can be induced to germinate" (Krugman, et al., 1977).

Delayed germination due to dormancy is a serious problem to the nurseryman (Krugman, et al., 1977). Natural irregular germination occurring over a two or three year period leads to irregularly aged stock, ties up bed space, hampers direct-seeding operations, and increases the time sown seed is exposed to predators, disease, and adverse weather conditions (Krugman, et al., 1977; Macdonald, 1986).

FINAL POINTERS

Study the literature first when attempting to germinate woody plant seeds that are doubly dormant or of an unfamiliar species. The Proceedings of the International Plant Propagators' Society is an indispensable reference for the propagator, and their indices are a good place to start. Other good references include: The reference manual of woody plant propagation: From seed to tissue culture by M.A. Dirr and C.W. Heuser, Jr (1987); Plant propagation: Principles and practices, 5th ed., by H.T. Hartmann, D E. Kester, and F T. Davies, Jr. (1990); Practical woody plant propagation for nursery growers, vol. 1., by B. Macdonald (1986); and Seeds of woody plants in the United States, published by the USDA Forest Service (1974) All of these books should be on the ready-reference shelf of any serious propagator.

Controlled temperature germination chambers are a must for seed treatment. Many growers simply sow the seed and place the containers under a greenhouse bench or under shade for warm stratification. This is not satisfactory due to poor sanitation and little temperature control Expensive seed-treatment chambers can be purchased or chambers can be constructed from discarded refrigerators or incubators. Refrigerated truck boxes that are no longer road worthy can easily be converted into large germination chambers (Macdonald, 1986).

The germination environment is critical. The seed must be sound and clean. The pre-germination medium must be pathogen-free, have a high moisture-holding capacity, and be well aerated (Hartmann, et al., 1990). Finally, the germination chamber must have precise temperature control Any temperature extreme during pre-germination treatment may induce secondary dormancy, which is usually very difficult to break (Dirr and Heuser, 1987; Hartmann, et al , 1990, Macdonald, 1986).

LITERATURE CITED

Abdalla, S.T. and **A.D. McKelvie.** 1980 The interaction of chilling and gibberellic acid on the germination of seed of ornamental plants. Seed Sci Technol 8(2) 139-144

Ballington, J.R., G. J. Galletta, and **D.M. Pharr.** 1976 Gibberellin effects on rabbit-eye blueberry seed germination HortScience 11(4) 410-411

Bonner, F. T., B.F. McLemore, and **J.P. Barnett.** 1974 Presowing treatment of seed to speed germination, p 126-135 in Seeds of woody plants in the United States USDA Forest Service Agriculture Handbook no 450

Bretzloff, L.V. and **N.E. Pellett.** 1979 Effect of stratification and gibberellic acid on the germination of *Carpinus caroliniana* Walt HortScience 14(5) 621-622

Brinkman, K.A. 1974 *Corylus* L Hazel, p 343-345 in Seeds of woody plants in the United States USDA Forest Service Agriculture Handbook no 450

Burns, R.B. 1967 Gibberellic acid stimulates germination of sweetgum seed. Forest Sci 13(4) 438-439

Devlin, R.M. and **J. Karczmarczyk.** 1975 Effect of light and gibberellic acid on the germination of 'Early Black' cranberry seeds Hort Res 5(1) 19-22

Dhillon, B.S. and **M.R. Sharma.** 1978 Note on the germination of wild pear seeds Indian J Agric Sci 48(6) 370-372

Dirr, Michael A. and **Charles W. Heuser, Jr.** 1978 The reference manual of woody plant propagation From seed to tissue culture Varsity Press, Inc , Athens, Georgia

Frankland, B. and **P.F. Wareing.** 1966 Hormonal regulation of elm, scotch pine, Colorado and white spruce, Forest Sci 8(2) 187-190

Hartmann, H.T., D.E. Kester, and **F.T. Davies, Jr.** 1990. Plant propagation: Principles and practices, 5th ed. Prentice Hall, Englewood Cliffs, New Jersey

Hundal, P.S. and **H.N. Khajuria.** 1979 Effect of GA, and thiourea on seed germination of different varieties of peach Indian J Agric Sci 49(6) 417-419.

Janick, J. 1986. Horticultural science, 4th ed. W H. Freeman, San Francisco, California.

Jann, R.C. and **R.D. Amen.** 1977 What is germination?, p. 77-109 in. A Khan (ed) The physiology and biochemistry of seed dormancy and germination North -Holland Publishing Co

Jarvis, B.C. and **D. Wilson.** 1977 Gibberellin effects within hazel (*Corylus avellana* L) seed during breaking of dormancy. I A direct effect of gibberellin on the embryonic axis New Phytol 78(2) 397-401

Jarvis, B.C., D. Wilson and **M.W. Fowler.** 1978 Growth of isolated embryonic axes from dormant seeds of hazel (*Corylus avellana* L) New Phytol 80(1)·117-123

Jones, R.L. and **J.L. Stoddart.** 1977 Gibberellin and seed germination, p 77-109 in A Khan (ed) The physiology and biochemistry of seed dormancy and germination North-Holland Publishing Co

Khan, A.A., C.E. Hert, E.C. Waters, C.C. Anojulie, and **L. Anderson.** 1977 Seed dormancy Changing concepts and theories, p 29-50 in A Khan (ed) The physiology and biochemistry of seed dormancy and germination. North-Holland Publishing Co

Krugman, S.L., W.I. Stein, and **D.M. Schmitt.** 1974 Seed biology. Seeds of woody plants in the United States. USDA Forest Service, Agriculture Handbook no 450

Macdonald, B. 1986 Practical woody plant propagation for nursery growers, vol I Timber Press, Portland, Oregon

Newman, S.E. 1981 Seed germination of *Ostrya virginiana* (Mill) K Koch MS Thesis University of Nebraska, Lincoln, Nebraska.

Nikolaeva, M.G., T.V. Daletskaya, M.V. Razumova, and **N.N. Kofanova.** 1973 Effects of gibberellin and kinetin on embryo growth and seed germination in spindle tree an tatar maple Sov Plant Physiol 20(4) 600-605.

Oosting, H.J. 1956 The study of plant communities W H. Freeman, San Francisco, California.

Pollock, B.M. and **V.K. Toole.** 1961 After ripening, rest period and dormancy, p. 101-112 in. Seeds Yearbook of agriculture USDA, Washington, D C

Powell, L.E. 1987 Hormonal aspects of bud and seed dormancy in temperate-zone woody plants HortScience 22(5) 845-850

Rudolf, P.O. 1961 Collecting and handling seeds of forestry trees, p 221-226. in· Seeds. Yearbook of agriculture USDA, Washington, D C

Sandahl. P.L. 1941 Seed germination Parks and Recreation 24(2) 508

Simancik, F. 1970 Germination of seeds of *Prunus laurocerasus* L after gibberellic acid treatment at warm, cold and warm-followed-by-cold stratifications Proc Int Seed Testing Assn 35(2) 393-403.

Toole, E.H., V.K. Toole, and **E.A. Gorman.** 1955 Photocontrol of *Lepidium* seed germination Plant Physiol. 30(1) 5-21

Vogt, A.R. 1970 Effect of gibberellic acid on germination and initial seedling growth of northern red oak. Forest Sci 16(4) 453-459.

Wcislinska, B. 1977 The role of gibberellic acid (GA3 in the removal of dormancy in *Fraxinus excelsior* L seeds Biol Plant 19(5) 370-376