PLSC 368 - Plant Propagation Name _____ Group_____
Lab Exercise 1
January 21, 2009

1. SEED STRATIFICATION AND SCARIFICATION

Note: You will learn procedures for removing seed dormancy in woody and herbaceous perennials by stratification and scarification.

1. Introduction

The seed has three basic parts: a) the embryo, b) the food storage tissue, and 3) the seed coat. The embryo and seed coat of woody seeds may require special preconditioning in order to overcome dormancy before germination will occur. Some seeds require a period of low temperatures for physiological changes or after-ripening to take place within the embryo before germination. The process of stratification will meet this requirement. The embryo dormancy can also be overcome by treating the seed with gibberellic acid (GA_3) in some woody and herbaceous perennial seeds.

The seed coat which protects the embryo from injury may be hard and impermeable to water. This will prevent the seed from imbibing the water necessary for germination to begin. In order to modify or alter this condition of the seed coat, the seeds should be scarified. Scarification can be accomplished by several methods: mechanical scarification, soaking the seed in hot water, or acid treatment.

For woody plant seeds which require special preconditioning for both the embryo and the seed coat (double dormancy), a combination of stratification and scarification may be necessary. Although these special dormancy requirements are met in nature, germination may occur at irregular intervals. Not only is more time required, but the resulting plants will be different ages and sizes. To the plant propagator, the uniformity of plant material is essential to a commercial production cycle. By using special preconditioning techniques, it is possible to control the germination process in many woody and herbaceous perennial species. The objectives of this lab are: 1) to learn procedures for cold-stratifying seeds to remove embryo dormancy, and 2) to practice various methods of removing seed coat dormancy in woody plants

2. Procedures

A. Seed Stratification

a. **Species used for class project**:

Species	Common Name	Group in Charge
1. *Acer ginela*	Amur Maple	
2. *Celtis occidentalis*	Northern Hackberry	
3. *Cornus stolonifera*	Red Osier Dogwood	
4. *Malus domestica*	Hardy Apple	
5. *Parthenocissus inserta*	Woodbine	
6. *Prunus americana*	American plum	
7. *Shepherdia argentea*	Buffaloberry	
8. *Syringa villosa*	Late Lilac	

b. **Treatments**

Stratification (weeks)	Date started	Date terminated	Number seeds/unit	No. of reps	Total no. of seeds
10 weeks	Jan 21	Apr 1	50	3	150
5 weeks	Feb 25	Apr 1	50	3	150
0 weeks	Apr 1	Apr 1	50	3	150

c. **Procedures**

Each group will obtain seeds of one or two species. Count seeds and use 50 or 100 seeds as a treatment unit. Mix seeds in a handful of moistened peat moss and place the mixture in a zip-lock bag. Keep one-tenth of the sealing of each zip-lock bag open to allow air entry. Store the seed samples in the refrigerator (4^{o}C) for a pre-determined time period (0, 5, 10 weeks) before germination experiment (Apr 1). Each treatment is replicated 3-4 times, depending on the availability of the seed. Seeds of each treatment are planted in a large cell pack or a tray filled with germination medium (Promix or Sunshine mix). Determine percent germination in two weeks and use the data for your report.

B. Seed Scarification

a. **Plants:** 1) Honeylocust (*Gleditsia triacanthos*)
2) Kentucky coffeetree (*Gymnocladus dioicus*)

b. **Treatments**:

	Number of seeds	
Treatment	Honeylocust	Kentucky coffeetree
1. Control	50	20
2. Mechanical Scarification*	50	20
3. Boiling Water	50	20
4. 4-Hour Acid Soak	50	20
5. 8-Hour Acid Soak	50	20
6. 12-Hour Acid Soak	50	20
7. 16-Hour Acid Soak	50	20

*All students are required to perform mechanical scarification treatment.

c. **Seed germination**:

Plant seeds in peat-lite mix (Promix or Sunshine mix) contained in large cell packs or open flats. Planting depth is about twice as deep as the thickness of the seed. Write the number of seeds planted per cell pack or tray on the label. Place the flats in the mist house until the seeds germinate and then move them to Range 8. Determine percent germination after seedling emergence is completed.

d. **Data:** Obtain percent germination for each treatment. Combine all data collected for the class and use them in your report.

3. Lab Report

Write a report summarizing the effect of seed treatment on percent germination and other observations and findings. **Your report should have: title, abstract, introduction, materials and methods, results, discussion, and several references. Lab report is due by April 22, 2009.**

Table 1. Seed treatment requirements for selected woody plants.

Plant	Species	Requiremen
American Linden	*Tilia americana*	Seeds exhibit great variation from year to year for no explainable reason. Seeds usually germinate well if they are picked before the leafy bract and seed coat turn from gray to brown.
American Sweetgum	*Liquidambar styraciflua*	Seeds have a very shallow dormancy which can be overcome by 1/2 to 2 months stratification.
Amur Maple	*Acer ginnala*	Fresh seed can be directly sown and require no pretreatment.
Birch	*Betula* spp.	Seeds require no pretreatment as long as light is provided.
Black Locust	*Robinia pseudoacacia*	Mechanical, acid, or hot water scarification is required, as with most species in the Pea (Fabaceae).
Black Walnut	*Juglans nigra*	Needs 3-4 months of stratification.
Catalpa	*Catalpa* spp.	Seed require no pretreatment.
Cherry, Plum	*Prunus* spp.	Germination requirements vary widely. Some seed sources require no stratification while others may require a 3-month cold period.
Common Lilac	*Syringa vulgaris*	Variable dormancy requirement; 2 month cold stratification results in 93% germination.
Eastern Redcedar	*Juniperus virginiana*	Requires 2-3 months cold stratification.
European Mountain-Ash	*Sorbus aucuparia*	Requires a three month cold stratification as do most other *Sorbus* species.
Fragrant Sumac	*Rhus aromatica*	Requires 30-40 min scarification in sulfuric acid followed by 1-2 months cold stratification.
Green Ash	*Fraxinus pennsylvanica*	Seeds may require 2-3 months cold stratification.
Hackberry	*Celtis occidentalis*	Requires 2-3 months stratification.
Honeylocust	*Gleditsia triacanthos*	Needs scarification: mechanical, or 1-2 hours of scarification in sulfuric acid.
Ironwood	*Ostrya virginiana*	Needs cold stratification for 3-4 months.
Kentucky Coffeetree	*Gymnocladus dioicus*	Requires 2-4 hours of acid treatment, or mechanical scarification.

Plant	Species	Requirement
Larch	*Larix* spp.	Seeds require no treatment; however, one to two months cold stratification may hasten and unify germination.
Pine	*Pinus* spp.	Species vary widely in germination requirements. Most do not require pretreatment. Some species may require artificial light or cold treatment.
Redbud	*Cercis canadensis*	Requires 30 minutes scarification in sulfuric acid followed by 3 months stratification. Hot (190^{O}F) water soak is also used in place of acid treatment.
Rocky Mountain Juniper	*Juniperus scopulorum*	Requires the equivalent of two winter cold periods: 3 months warm followed by 3 months cold will substitute for this process.
Russian Olive	*Elaeagnus angustifolia*	Requires 2-3 months of cold stratification. Seed coat may be exceptionally hard and scarification may increase germination percentage.
Silver Buffaloberry	*Shepherdia argentea*	Requires 2-3 months of cold stratification (40% germination). 97% germination after cold stratification for 3.5 years.
Spruce	*Picea* spp.	No pretreatment is required, although a 24-hour water soak or a 3-week cold stratification may unify germination.
Ussurian Pear	*Pyrus ussuriensis*	Needs 30-60 days of cold stratification.
Willow	*Salix* spp.	Seeds require no pretreatment.

List of Seeds Used for Stratification Studies in 2008 and 2009

Provided by Greg Morganson, Lincoln Oaks Nursery, 3310 University Drive, PO Box 1601, Bismarck, ND 58520, phone 701-223-8575, fax 701-223-1291, e-mail <*lincoln@tic.bisman.com*>

Year collected	Species	Common name	Comments
2005	*Ginkgo biloba*	Ginkgo	Warm and cold stratification for 1-2 months enhances seed germination
2005	*Pyrus ussuriensis*	Ussurian Pear	Requires 90 days cold stratification at 34-38 oF
2005	*Rhus glabra*	Smooth Sumac	Hot water treatment followed by 3-month cold stratification for good germination
2005	*Sorbus aucuparia*	European Mountain-ash	Requires 2-3 mopnths days of cold stratification
2006	*Celtis occidentalis*	Northern Hackberry	Requires 90 days cold stratification at 34-38oF
2005	*Cornus stolonifera*	Red Osier Dogwood	Requires 30 days warm and 90 days cold stratification at 41 oF
2006	*Caragana arborescens*	Caragana or Siberian Peashrub	Germination of untreated seeds is erratic. Light acid scarification for 15 minutes, 2-4 weeks of stratification desirable
2006	*Malus baccata*	Siberian crabapple	Requires 1-4 month cold stratificaion
2006	*Prunus americana*	American plum	For best germination, 5 weeks of warm stratification (60-70°F) followed by 3-6 months cold stratification at 41 °F needed
2006	*Shepherdia argentea*	Buffaloberry	Requires 60-90 days of cold stratification at 41°F