**Kennedy Lab**

Bioassay Procedure for *Pseudotsuga*

1. Prepare seeds (30 days)

-Get seeds, cheesecloth, a stapler, a ziplock bag, and some straws.

-Cut a piece of cheesecloth to around 1-1.5ft long, and fold it so that it is doubled over at least 4 times (this is going to hold the small seeds, so the mesh has to be thick enough so that they will not slip through even over a long period of time soaking in water).

-Measure out the amount of seeds needed, and pile them on the cut cheesecloth.

-Fold the cheesecloth around the seed pile to make a bag, and staple the bag shut (make sure the bag is very secure, since the seeds have a tendency to fall out).

-Place the pouch with the seeds into a container of tap water so that the pouch is completely submerged/floating, and cover the container.

-Let the seeds soak for 24 hours.

-Take the pouch out, and outside or in the sink shake the pouch around for a bit until it is fairy dry (kind of subjective, but we don’t want the seeds to be too wet or they will mold. Mostly try to get as much water out as you can without messing up the pouch or letting seeds escape).

-Put the pouch in a ziplock bag, and put the straws in the bag, and zip the bag so that the straws poke out the top (the straws provide ventilation for the seeds).

-Refrigerate for 4 weeks, every couple of days turning over the pouch to keep standing water from accumulating around any portion of the seeds.

-Plant seeds directly on soil surface, with a very light dusting of soil to hold them in place (just a few mm of soil at most).

-Should see germinate within 7-10 days of planting.