# REMEDIATION OF DEICER SALT CONTAMINATED SOILS USING NATIVE MONTANA PLANTS

Task 3 – Greenhouse Testing Plan

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#### Materials

Materials and supplies to be utilized for the Greenhouse Testing Plan include:

- Greenhouse
- Greenhouse Bench Space
- Supplemental Fluorescent Lighting
- 30 Greenhouse Megastore Grower Select Plug Trays, 72 square plugs
- 30 Greenhouse Megastore Germination Trays, no holes
- Colored Plastic Labels
- Qilebi 2 Gallon Watering Can, removable head
- Sodium Chloride (NaCl, rock salt, Whitefish, MT MDT Storage Facility)
- Hanna Instruments 98130 pH/Conductivity/TDS tester
- 100 ml plastic beaker
- Bamboo Stirring Stick
- Small Catch Trays
- 50/50 topsoil/sand mix
- Seeds
- Tally Counter

### **Methods**

## **Salt Concentration Watering Selection**

A salt (NaCl) concentration of 5-10 dS/m (2,500 - 5,000 ppm) in water was selected based on preliminary greenhouse testing of watering methods (top, bottom, and a combination of both), measurements of average soil salt concentrations from pour-through tests, the range of tolerances within our plant candidates, and the difficulties maintaining soil electrical conductivity (EC) over time. Half of the trays will have salt dosing of soil EC 5-10 dS/m (2,500 - 5,000 ppm), while the other half will be the control, using the standard greenhouse water (0.267  $\pm 0.01$  dS/m, 134  $\pm 5$  ppm).

## Sample Tray Design

The greenhouse experiment will use a randomized block design. Thirty (30) plug trays will be placed on the greenhouse bench, with fifteen (15) trays having a water regime treated with salt and fifteen (15) control trays (greenhouse water, here in referred to as clean water). There will be five replicates for each treatment and plant type within the fifteen (15) trays. The fourteen (14) plant candidates will be seeded for an entire row once per replicate treatment (Figure 1). Within each treatment, watered with salty or clean water, there will be four extra rows in the plug trays that will be filled with randomly selected plant species, the purple-to-blue

gradient shown in Figure 1, and will not be measured. The plug trays will be distributed systematically across the bench space to ensure variability in placement and environmental exposure, which are accounted for in the randomized block design.

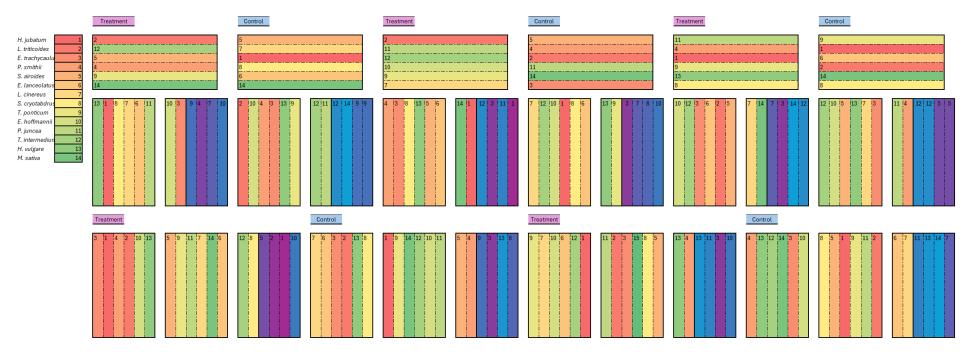


Figure 1: Greenhouse randomized block design for planting.

Labels displaying information on replicate, treatment (salt and control watering), and plant variety will be used.

## **Plug Tray Method**

The greenhouse experiment will use the fourteen (14) salt tolerant plant varieties previously selected (Table 1). A 50/50 topsoil/sand mix will be made in the plant growth center on MSU's campus using their materials; the topsoil will not be autoclaved. The soil mix will then be pre-wet with clean water and used to fill the thirty (30) plug trays. Premade labels will be placed into the filled soil trays, and three seeds will be planted in each plug according to the variety listed for that row. After planting, trays will be moved into the greenhouse space, placed according to Sample Tray Design (Figure 1). All trays will be initially watered once with clean water.

Table 1. Plant species identified for use in Task 3 Greenhouse testing.

Common / Scientific / Family / Variety	Native Status	Salt Tolerance
Foxtail Barley / Hordeum jubatum / Poaceae	Native	High
Beardless Wildrye / Leymus triticoides / Poaceae /	Native	12,000 ppm
Slender Wheatgrass / Elymus trachycaulus / Poaceae / Pryor	Native	10,000 ppm
Western Wheatgrass / Pascopyrum smithii / Poaceae	Native	7,000 ppm
Alkali Sacaton / Sporobolus airoides / Poaceae	Native	7,000 ppm
Thickspike Wheatgrass / Elymus lanceolatus / Poaceae	Native	5,500 ppm
Basin Wildrye / Leymus cinereus / Poaceae	Native	5,000 ppm
Sand Dropseed / Sporobolus cryptandrus / Poaceae	Native	2.000 ppm
Tall Wheatgrass / Thinopyrum ponticum / Poaceae / Alkar	Non-native	11,000 ppm
<b>Green Wheatgrass</b> / Elymus hoffmannii / Poaceae / AC Saltlander	Non-native	10,000 ppm
Russian Wildrye / Psathyrostachys juncea / Poaceae / Bozoisky II	Non-native	7,000 ppm
Pubescent Wheatgrass / Thinopyrum intermedium / Poaceae / Luna	Non-native	6,000 ppm
Barley / Hordeum vulgare / Poaceae / Haymaker	Non-native	6,000 ppm
Alfalfa / Medicago sativa / Fabaceae / FSG 423ST	Non-native	3,000 ppm

#### Watering

Watering will be done every three days, split between top and bottom watering. The treatment group will be top-watered with 750 mL of clean water and bottom-watered with 750 mL of 5 dS/m (2,500 ppm) salt solution. The control group will only receive clean water but will follow the same top and bottom watering regimes. Following bottom watering, the water will be left in the tray for one (1) hour before being removed.

#### **Soil Water Monitoring**

Pour-through tests according to methods developed by Purdue University Extension (Nemali, 2018) will be performed weekly for top and bottom watering to monitor soil EC levels and provide additional information during data analysis, and to inform potential shifts in our watering regime if EC levels exceed our set range of 5-10 dS/m (2,500-5,000 ppm). A handheld EC meter (Hanna Instruments 98130 pH / Conductivity / TDS tester) will be used to measure soil water EC and pH and will be calibrated every use to ensure consistent results. If the soil EC is too high, treatment water salt concentration will be reduced to 2.5 dS/m (1250 ppm) and monitored to slow the buildup of salt in the soil.

#### **Plant Germination Data Collection**

Plant germination will be recorded using a Tally Counter every three days to compare plant variety performance. After germination, plugs will be thinned to one seedling per plug to reduce competition and increase visibility of individuals for visual observations. Plants will be grown for three to four months.

## **Plant Harvest and Prep**

After three to four months, the plants will be removed from the soil and the roots cleaned of debris. The soil from the trays will be collected and tested for final EC measurements, along with pH and other nutrient concentrations. Plant material will first be rinsed with distilled water, then dried for 2 days at 70°C in a drying oven. After drying is complete, plant material will be separated into above- and below-ground tissue. For testing, plant tissue from all fourteen (14) replicates making up a row in each treatment will be combined to create one larger sample. This will create five samples for each plant type and treatment.

To prepare a sample to determine plant tissue chloride concentration, 0.10-0.15 grams (g) of dried tissue of plant material will be added to three separate test tubes for milling to create triplicate samples for each plant type and treatment. After milling, the resulting powder will be put into 100 mL of deionized water and placed in a 90°C water bath for 60 minutes (Asch et al., 2022).

#### **Plant Tissue Testing**

Plant tissue extracts will be tested for EC levels following the same procedures as soil

extracts.

## **Soil Testing**

Following the conclusion of our plant growth tests and plant harvesting, soil will be collected and composited based on treatment. An extract will be produced for each of the fourteen (14) plant types and treatment following the previously used Florida Method of Test for Chloride in Soil and Water (FDOT State Materials Office, 2023). These soil extracts will then have their EC measured using a handheld EC meter.

Throughout the greenhouse experiment and sampling process, pictures will be taken to record the effort. Germination, soil EC, growth statistics, plant chloride concentrations and other data will be recorded in an excel sheet and stored on the Montana State OneDrive for safety.

#### References

- Asch, Julia, et al. "Comprehensive Assessment of Extraction Methods for Plant Tissue Samples for Determining Sodium and Potassium via Flame Photometer and Chloride via Automated Flow Analysis \*." *Journal of Plant Nutrition and Soil Science*, vol. 185, no. 2, Feb. 2022, pp. 308–16, https://doi.org/10.1002/jpln.202100344.
- FDOT State Materials Office. Florida Method of Test for Chloride in Soil and Water. No. FM5-552, 24 Aug. 2023. Florida Sampling and Testing Methods, <a href="www.fdot.gov/docs/default-source/materials/administration/resources/library/publications/fstm/methods/fm5-552.pdf">www.fdot.gov/docs/default-source/materials/administration/resources/library/publications/fstm/methods/fm5-552.pdf</a>. Accessed 7 Aug. 2025.
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