Protocol: p8 Setup & Measure qGen2 Experiment in 2016

Background

In the summer of 2013 we initiated an experiment, called qGen2, to assess adaptive potential in *Echinacea angustifolia*. In summer 2013, we crossed maternal plants in p1 with pollen from Landfill and Staffanson. That October, in a newly cleared plot (p8), we sowed the resulting achenes. We scattered achenes within 1-meter long segments (between 0.05 to 0.95m) marked with nails at 0cm and 100cm along a measuring tape. In the spring of 2014 we located, measured, and marked each seedling with a plastic toothpick placed 2cm north of the plant. We used colored toothpicks (blu, org, wht, grn, red, yel) in rotation so duplicate colors are not next to each other.

Goals

1. To assess survival and measure leaf lengths in 2 year old progeny of qGen2 crosses.
2. To mark and measure newly emerged seedlings or juvenile plants that were not found last year.

Equipment Needed

Visor with assigned rows & segments, meter stick, toothpicks, rulers, 50-meter, 30-meter, and 5-meter tapes, *Echinacea* seedling search image in your head

Plot setup / Flag plan

The outline of p8 is not a rectangle. In general, **697 is the lowest Position** (on the South end closest to HWY 27) and **755 is the highest Position** (on the North end closest to the Hjelm House). Planted **Rows** range from **104** on the West edge (closest to Tower Rd.) to **181** on the East edge (closest to the electric lines). Note that not all rows are planted. See figure 1 for map of flags left over the winter.

Lay Measuring Tapes

Use meter sticks to measure from 697 to 700. On shorter rows (< 120), set 30-meter tapes at 700. On all other rows (shorter rows >160 included), Set 50-meter tapes with 0 at 700 (or with 10m at pos 710, etc) so the tape can be used to check the position of the segment being measured. Use meter sticks or 5-meter tapes for positions 750-754. See table in Figure 2 to assist with setting up measuring tapes.

Search & Measure:
Most often we will be measuring in blocks of 10 (1-meter) segments. Choose a visor and open the form "2016MeasureQ2" and locate the starting row and nail. **Double check that you are in the correct location!** Enter initials for you and your partner in the "**measuredBy**" field for your first and last plant measured. There will be a record for each plant that was found in 2014 & 2015. Each record will list the **row, nail, seedlingCt2015** (for the segment), **tPickColor, northDist, eastDir, and eastDist** for the plant. Records will be in order of northDist (South to North). Use the arrow buttons to scroll through the records for plants in the segment while you and your partner search the segment for the plants & toothpicks listed.

Visualize the entire segment and the records for that segment. Scan for Echinacea plants with no toothpick and toothpicks with no Echinacea plants. If there are any issues about the segment—i.e. if any toothpicks don't match the records or if there are missing records—then confer with Gretel or Stuart. Please do not move any toothpicks or edit any records. If everything looks as expected, then verify the plant identity (based on toothpick color) and location as follows:

1. Double check that the **row** and **nail** being searched match the position in the visor! If you are working on nail 700, you should be between nails at positions 700 and 701.
2. Double check the **tPickColor** (blu, org, wht, grn, red, yel).
3. Verify **northDist**: the distance of your plant along the tape in centimeters. Distances should range from 5 to 95 centimeters.
4. Verify **eastDir**: the direction of your plant in relation to the tape (West, On, or East of the tape). Remember that West is toward Tower Rd.
5. Verify **eastDist**: the east/west distance of the plant from the tape in centimeters. If your plant is west of the tape (toward Tower Rd), it will be recorded in negative centimeters. If your plant is East of the tape (toward Electric lines), it will be recorded in positive centimeters. If your plant is on the tape, it will be recorded as 0 cm. Example: plant 10 cm west of tape: "-10."

Measure each plant as follows:

6. **plantStatus:** Record the status of the plant and the toothpick in this multiselection box: Basal, Crisp, Can't Find Plant, tPick Present, tPick Absent
7. **1LeafCm**: Record the length of first leaf in cm. Record additional leaves in **2LeafCm**, **3LeafCm**, **4LeafCm** , and **5LeafCm** fields. Measure from the ground to the top of the leaf using a ruler. Orient the ruler straight up.
8. **plantNotes**: Record anything unusual or noteworthy about the plant in this field. Use a semicolon to separate notes – *no commas!*
9. **segmentNotes**: Record anything unusual or noteworthy about the segment in this field. Use a semicolon to separate notes – *no commas!*
10. **cotyledonStatus**: Ignore unless you are measuring a plant with cotyledons. Using the multi-selection box, select the status of the cotyledons. You can make multiple selections. Options include: present, not present, green, yellow, brown, shriveled, 1, 3. Most EA seedlings have 2 cotyledons, but some may have 1 or 3.
11. **Completed**: Click the completed checkbox & proceed to the next record.
12. Repeat for each plant found in the segment.

Do a double-check search for *Echinacea* plants with no toothpicks. We are really keen to find some new plants and see if they are seedlings or juveniles.

13. Create a new record and enter all data for any plants or seedlings that have not previously been found. Use new, uniquely colored (or shaped) toothpicks.
14. Double check that you have entered data for all of the plants in the segment.
15. Go on to the next segment.

If you have *any* questions, please ask Gretel or Stuart.

Figure 1. Winter Flag Plan. Black dots are nails, all other dots are flags.

Flag placement

1. Every row (100 – 189) Min & Max Positions
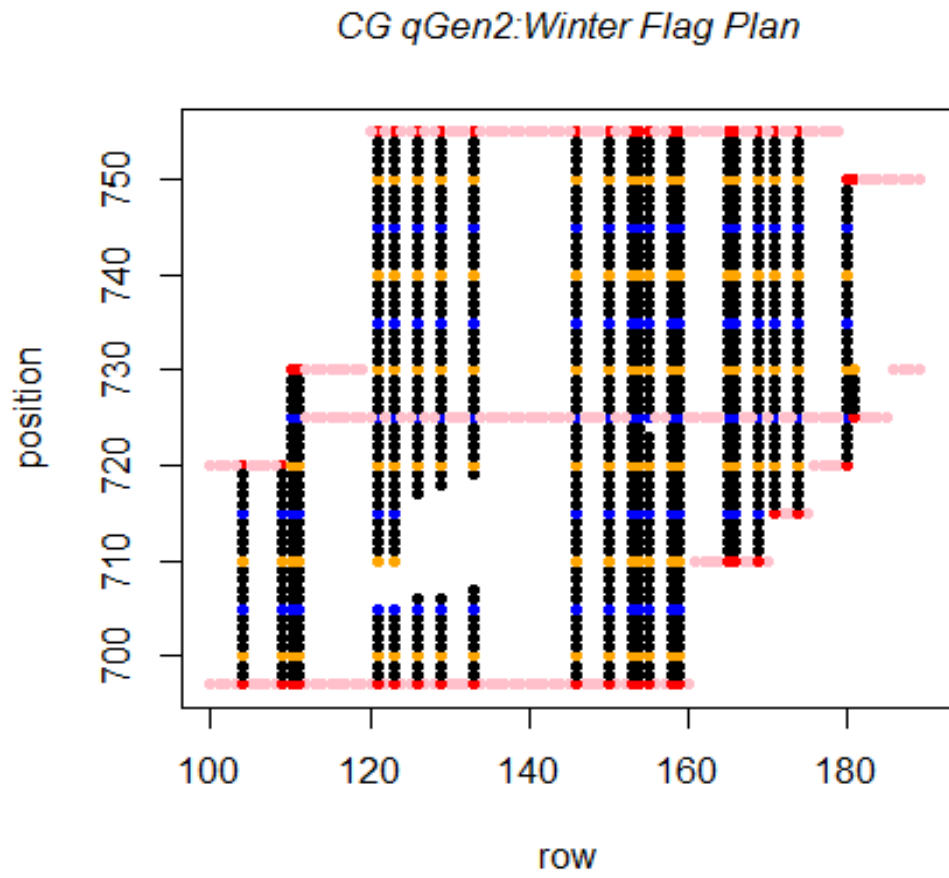2. Rows divisible by 5 at position 725

CG qGen2:Winter Flag Plan

Figure 2. Rows & Position to be searched w/ count of positions

	qGen2MeasureStartingPositions					
row	seg1min	seg1max	seg2min	seg2max	notes	count
104	697	719				23
109	697	719				23
110	697	729				33
111	697	729				33
121	697	705	710	754	tpplot caused gap	54
123	697	705	710	754	tpplot caused gap	54
126	697	706	717	754	tpplot caused gap	48
129	697	706	718	754	tpplot caused gap	47
133	697	707	719	754	tpplot caused gap	47
146	697	754				58
150	697	754				58
153	697	754				58
154	697	754				58
155	697	723	725	754	rock @724 caused gap	57
158	697	754				58
159	697	754				58
165	710	754				45
166	710	754				45
169	710	754				45
171	715	754				40
174	715	754				40
180	720	749				30
181	725	730				6