Methods experiment vegetation cutting in Remmene 2016

Question: Do plants in plots where the vegetation is cut show higher predation and lower fitness compared to plants with similar traits in uncut plots?

How does removing the vegetation affect ant abundance?

* 1st VISIT (2-3 July):

MARKING OF PLOTS AND CUTTING: Mark 20 plots and cut vegetation in 10 of them. Try to set the plots in the same area where we marked plants last year, mostly along or close to the ditch, and approximately in a line along it, so they are easy to find. Avoid setting them in the area to the right of the ditch, which was cut last year. Decide on site the size of the plots in order to get a reasonable number of plants into each of them (20+), a starting size could be 2 m x 1 m. Mark plots with 4 small wooden poles labeled with tape and a number from 1 to 20. Try to orient all of them in the same direction and put for example red tape on the upper left corner and white tape in the lower right corner. Note X and Y coordinates of one of the corners of the plot (specify), using phone GPS. If some plots are difficult to find, put tape marks on bushes, etc. Try to find plots where plants have no eggs. Randomize the treatments “Cut” and “Uncut” (Control). In the “Cut” plots, cut all the vegetation except the *Gentiana* plants using garden scissors.

MARKING OF PLANTS: In both treatments, if there are eggs on any of the plants, mark these plants and count the eggs on them. Mark with wool and tape label with plot number + plant number (e.g. 1.1) around the most advanced shoot (this will be the shoot selected for measurements). Count eggs on this shoot and note them on the protocol. Do not mark any other plants in this visit (i.e. do not mark plants without eggs).

If there are extra iButtons after accounting for those to be used in the grid design, consider putting some in the plots, for example 4 iButtons per plot at 50, 25 cm from corners (mark location with flag + label).

* 2nd VISIT (9-17 July):

MARKING OF PLANTS: In each of the plots, mark as many plants as possible, starting with 20 per plot. In the “Cut” plots, avoid if possible marking plants very close to the borders of the plot, where the vegetation will not be cut all around the plant individual. Mark with wool and tape label with plot number + plant number (e.g. 1.1) around the most advanced shoot. For each marked plant (considering also the ones marked in the first visit, if any), count and note in the protocol the number of shoots.

MEASUREMENT OF INTERACTION INTENSITY: Count the number of eggs per marked shoot.

1st ANT COLLECTION: Set 4 baits per plot at 50, 25 cm from corners (mark location with flag + label). At each bait location, put an open plastic vial containing a sugar cube on the soil. Set them in the morning and check them 1-2 times during the day and a last time before leaving. If any ants are found, put the lid and remove the bait (replace it with another if that is before the end of the day?). Take home, freeze and then store the dead ants into 70% alcohol.

Label with adhesive label + pencil with Location, Date, Plot number, Bait number.

* 3rd VISIT (29 July-5 August):

Consider marking more plants in the plots if needed / possible (e.g. newly emerged plants, or plants that did not have buds last time).

MEASUREMENTS OF TRAITS, INTERACTION INTENSITY, CONTEXT, (FITNESS): For the selected shoot in each marked plant, count and note shoot height (cm), number of flowers (i.e. count of all buds + open and dry flowers), developmental stage of the most advanced bud (stages a-f) and number of eggs. Count and note number of intact / predated / aborted buds / flowers / fruits (if present). Center a 30 cm2 square (use carpenter rule) on the plant and measure vegetation height (cm, in “Uncut” plots, take two measures if uneven) and count number of reproductive *Gentiana* shoots (i.e. shoots with open or closed buds) into this square (count also eggs on these plants if there is time left).

2nd ANT COLLECTION

* 4th VISIT (September):

MEASUREMENTS OF FITNESS: For the selected shoot in each marked plant, count and note number of of intact / predated / aborted buds / flowers / fruits (if present).

FRUIT COLLECTION: Collect one fruit of each type (predated / unpredated) from the selected shoot in each marked plant when available.

3rd ANT COLLECTION (if needed, depending on the amount of samples already collected).