Emergence Timing and Morphological Characteristics of *Galium* Species in Western Canada

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Abstract

Catchweed bedstraw (*Galium aparine* L.) and false cleavers (*Galium spurium* L.), collectively known as cleavers, have become an increasing problem in grain fields across the Canadian prairies. The increasing presence of cleavers on the prairies has resulted in canola infested with cleavers seeds, and its tangling behaviour can cause difficulties during harvest. Although there are many herbicide options available, the development of resistance in cleavers to Group 2 and Group 4 (quinclorac) herbicides has complicated their management. Understanding germination and emergence characteristics can provide a better understanding of how populations of *Galium* species are behaving in western Canada, and whether emergence timing of these populations are preventing adequate weed control. The objectives of this research are to 1) To determine the emergence timing of several cleavers populations 2) To characterize the morphological characteristics and differences between the populations 3) To determine the speciation and relatedness of the populations. Germination, emergence, and morphological characteristics were examined in 6 representative samples of cleavers obtained from various locations across the prairies. To determine if populations are acting as winter or summer annuals, two sowing dates (early May and early September) were utilized. Throughout the experiment, measurements on morphological characteristics such as leaf area, leaf weight, branch number, shoot biomass, plant height, flowering, seed production and thousand seed weight were taken. After the first year of data collection, differences between populations with regard to some morphological characteristics was observed. Emergence patterns suggest that the populations start to emerge at the same time, but the time to reach 50% and 100% emergence is different between populations. Among populations, fall emergence appeared to be very low in comparison to spring emergence.

Introduction

Galium aparine is often referred to as ‘catchweed bedstraw’. This species prefers to grow in shady, moist areas (Moore 1975). Chromosome number and ploidy level are the only true way of distinguishing cleaver species from one another. *G, aparine* is normally a hexaploid (Podlech and Dieterle 1969) with a chromosome number 2n=66 (Malik and Vanden Born 1988). The common name for *Galium spurium* is ‘false cleavers’. This species was introduced into Eastern Canada as a contaminant of shipped grain, which then moved west due to its preference for drier, sunnier habitats (Malik and Vanden Born 1987a). *G. spurium* is thought to be a more competitive species than *G. aparine* in crops since it is better adapted to the prairie climate and nutrient-rich soils. Minor morphological details such as smaller cotyledons, lighter color, stiffer, rougher stems, more branched and ‘stickier’ leaves may differentiate *G. spurium* from *G. aparine* (Malik and Vanden Born 1988). *G. spurium* has a chromosome number 2n=20 and no recorded polyploidy (Malik and Vanden Born 1988; Moore 1975).

Galium aparine and *Galium spurium,* commonly referred to as cleavers in Western Canada, have become, prominent weed species on the Canadian Prairies. In the latest prairie weed survey of canola, cleavers were ranked 9th in the 2000's, up from 30th in the 1980's (Leeson et al. 2005). There is a similar trend in Saskatchewan canola fields where cleavers are #6 in relative abundance, up from #31 in the 1970's (Leeson 2012). Both species share similar morphological characteristics, which makes them difficult to distinguish from one another. Both species are annuals, but can act as a biennial if the plant enters winter in a vegetative state. They have similar germination behaviour, short seed viability and prefer to grow in cool, moist, nutrient-rich soils (Malik and Vanden Born 1988; Moore 1975). Leaves and seeds are covered with stiff, hooked bristles that allow the species to attach to the fur, feathers or clothing of passing animals (Defelice 2002; Moore 1975). Seeds are also light enough to be spread by wind, and a hollow space between the two carpels helps seeds float on water. More importantly, cleavers are the same size and shape of canola that is grown across Canada. Due to their similarities, cleavers cannot be easily separated from canola.

The increase in the frequency of cleavers in the fields of western Canada has made it vital to understand why this has occurred as well as how to better utilize control strategies. Malik and Vanden Born (1988) speculate that this increase may be due to the similarity in seed size with canola. False cleavers have been shown to reduce canola yields by 18% when present at 100 plants per square meter. Furthermore, its tangling behaviour makes harvesting operations difficult (Malik and Vanden Born 1987b). Current broadleaf herbicide options are unable to effectively control cleavers. This is partially due to the development of resistance to Group 2 herbicides in cleavers, but also to poor spray timing (Hall et al. 1998). Imazamox, imazapyr, and imazethapyr are Group 2 herbicide options in Group 2 resistant canola. Imazamox alone and in combination with clopyralid will only suppress cleavers, while combinations of imazapyr and imazethapyr with imazamox can provide great control if the cleavers are not resistant. Glyphosate provides excellent cleavers control, but can only be used on glyphosate resistant canola. Glufosinate, known as Liberty, can control cleavers in glufosinate resistant canola, but some producers find that it is often not successful on cleavers past the 2 whorl stage (Saskatchewan Ministry of Agriculture 2012). The evolution of resistance to pesticides has led producers to find other means by which to control pests. To successfully accomplish this, an understanding of pest biology is necessary. Morphological characteristics can be used to measure the competitive ability of weed populations and their relatedness (Hübner et al. 2003). In Europe, small differences in morphological characteristics were found between populations of *G. aparine.* Hübner et al. (2003) determined that leaf size, shoot length, and plant weight were dependent on environmental conditions during growth, while length-width ratio of the cotyledons, number of leaves per whorl, length of internodes, and 1000-seed weight are determined genetically. Differences in competitiveness will likely stem from genetic characteristics, but how plants respond to stresses in their environment will also impact competition. Studying emergence and germination timing will allow for the ability to predict weed emergence and improve control strategies. The objectives of this research are to 1) To determine the emergence timing of several cleavers populations 2) To characterize the morphological characteristics and differences between the populations 3) To determine the speciation and relatedness of the populations.

Materials and Method

This experiment was a common garden with a randomized complete block design containing 4 replications at two locations (Kernen and Goodale Research Farm) in 2013. Two treatment factors were used, including seeding date (spring and fall) and populations (Trawin, Heavin, Clancy, Lacombe, Vegreville, SPG, Germany, and UK). A total of 64 microplots, 1 m wide x 2 m long, were present at each location. Cleavers samples were collected from six locations across western Canada and 2 from Europe. Land locations are in Table 1.

Table 1: Sources of cleavers seed.

Name	Land Location	Name	Land Location
Lacombe	NW 35-40-26 W3	Melfort - Heavin	NW 25-45-17 W2
Vegreville	SW 25-52-15 W4	Melfort - Trawin	NW 11-45-18 W2
Carrot River - Clancy	SW 23-51-11 W2	Germany	N/A
Saskatoon - SPG	E1/2 1-36-4 W3	United Kingdom	N/A

Samples of each population were blended with sand and then broadcasted by hand onto fallow at a rate of 400 seeds per m^2. Plots were raked individually to cover seeds with soil. Planting took place in early May and early September to determine competitive ability during crop growth and the potential for over-wintering plants. Over-wintering plants could have a large competitive advantage the next year due to their advanced stage. Soil preparation included soil samples to determine nutrient requirements, tillage to eliminate residue and a pre-burn herbicide application of glyphosate (900 g ae/ha) before seeding to control weeds. Group 1 herbicides were used to control grassy weeds as needed, while broadleaf weeds were hand weeded throughout the season. Newly emerged cleavers plants within three randomly placed 0.15 m^2 quadrats were recorded daily and marked with a rubber band to ensure they are not counted twice. Mean values of the four reps for each population was used for the creation of graphs in excel. Differences in morphological traits between populations were analyzed using PROC GLM in SAS 9.3 (SAS 2003). Morphological characters were taken as described in Table 2.

Table 2: Morphological characteristics to be measured.

Plant Characteristic	Methodology
Leaf Area	Randomly select 5 plants per plot, remove leaves and determine leaf area with leaf area meter.
Leaf Weight	Determine weight of leaves from 5 randomly selected plants per plot plants in grams.
Flowering Notes	Determine the number of days to flowering for each plot and time between 5%, 50%, and 95% flowering.
Total Biomass	Cut 5 randomly selected plants per plot at soil surface, dry and determine weight in grams.
Plant Height	Determine height of 5 randomly selected plants per plot during seed maturation.
Leaf Number	Prior to harvest, 5 plants will be randomly selected from each plot and the number of leaflets per whorl will be count at the 5th and 10th whorl.
Number of Branches	Prior to harvest, 5 plants will be randomly selected per plot and the number of branches per plant will be counted.
Seed Production	At maturity, each plot will be cut, dried and then threshed through a combine to determine seed production.
Thousand Seed Weight	Seed from each plot will be run through a seed counter for a 1000 seeds and then weighed.

Results and Discussion

Due to a high background population of cleavers at the Kernen site that was seen in unseeded plots, graphs only represent results from the Goodale location. All populations started to emergence around the same time, but some finished germinating earlier than others (Figure 1).

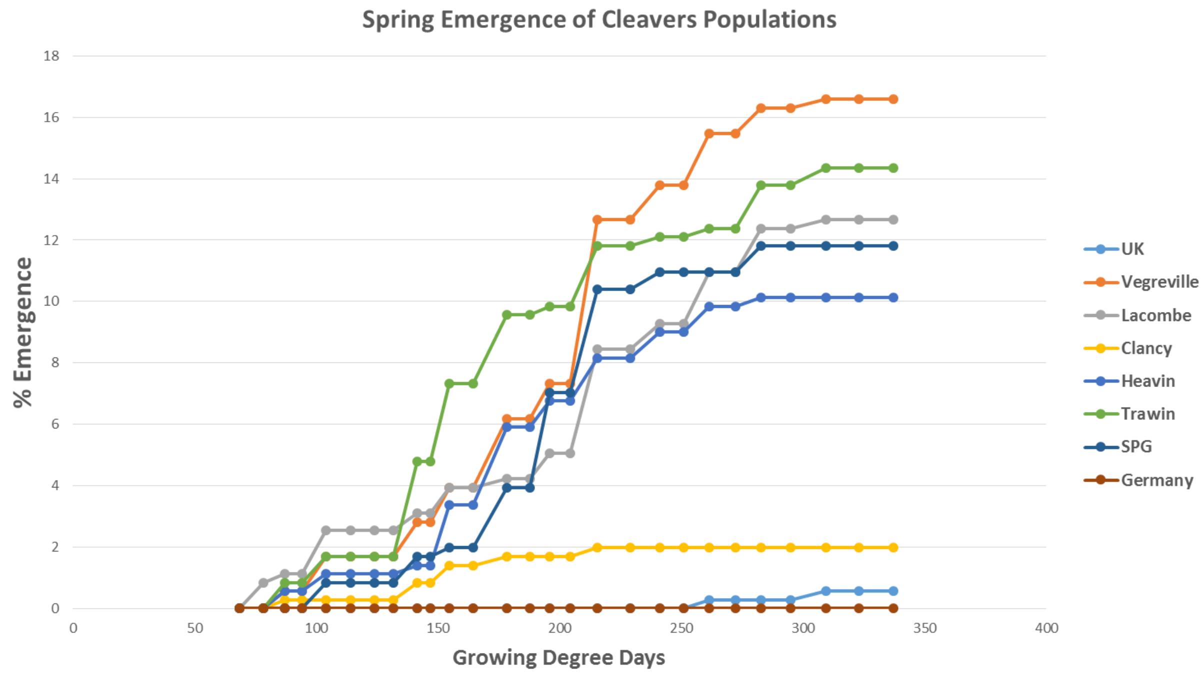

Figure 1: Spring emergence of the six Canadian populations at Goodale in 2013.

Seed from Germany did not emerge at all and emergence from the United Kingdom population was very late (250 GDD) and exhibited low numbers (<1%). Clancy behaved similar to the European populations and finished emerging early in the season, with very low overall emergence (2%). Emergence timing of the other Canadian populations were similar to each other throughout the spring. Time to reach 50% emergence was between 210-268 GDD. Trawin was more robust earlier in the season and was 50% emerged at 210 GDD, while Lacombe's median emergence time was later at 268 GDD. Variance in the emergence timing likely contributes to poor control. If cleavers emerge after a pre-seed burn off or in-crop herbicide application, the cleavers will compete and contaminate the crop. Even if plants emerge before an in-crop spray, for most herbicides, they must not be past a certain stage for good control. The percentage of total emergence varied between the populations. Clancy and the European population appear significantly lower than the remaining Canadian populations. While Clancy has the lowest total emergence, the few plants produced significantly more seed per plant than any of the other Canadian populations.

Fall emerging cleavers has also contributed to poor control over time. Fall emergence timing shows great differences between the populations as well (Figure 2).

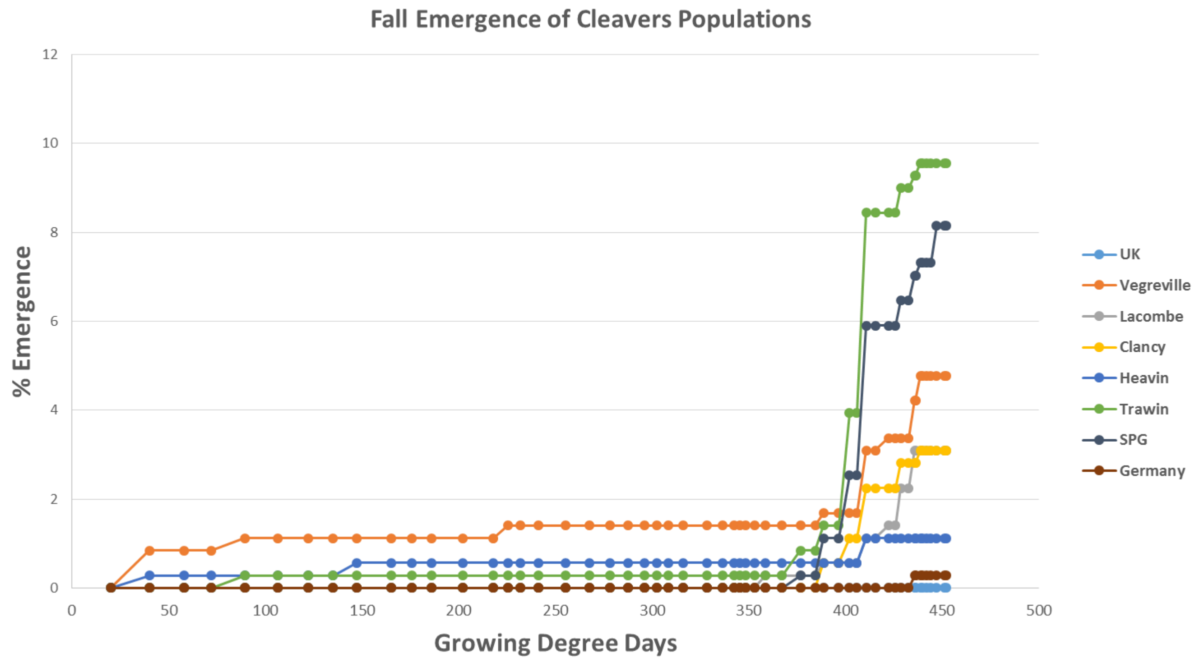

Figure 2: Fall emergence curves of the six Canadian populations at Goodale in 2013.

Few populations emerged soon after seeding, but the majority of the fall emergence occurred right before the first snowfall in late October. Once again the European populations did poorly in comparison to Canadian populations. Seed from Heavin had the lowest emergence when compared to the other Canadian populations, while Trawin had the highest. This observation suggests that there can be high variation in emergence timing from cleavers in close proximity. Other factors also may have contributed to differences. For example, the maternal environment can have a strong influence on some characteristics of the seed. Farming practices such as tillage can also recruit cleavers from seed burial as seen in results by Reid and Van Acker (2005). Total emergence also varied between populations. Clancy was similar to Lacombe and Vegreville seed, while Trawin and SPG appear to have a greater amount of total germination in the fall. If any of the plants survive the winter in a vegetative state, these plants will have a significant advantage over field crops. Growers that have fall emerging cleavers are going to have to implement some form of control in the fall. For the plants to survive the winter, they have to be in an older vegetative state. Since majority of the seeds emerged before the first snowfall, it is unlikely that any will survive the winter. While none of the populations reached over 10% total emergence in the fall, these plants can produce a large amount of seed to contribute to spring and fall emergence next year.

Analysis of morphological data using PROC GLM in SAS 9.3 showed that the only characteristics to exhibited significant differences between populations were height, yield per plant, and thousand seed weight as seen in Table 2.

Table 2: Effects of population on plant height, yield, and thousand seed weight in *Galium*.

	Morphological Characteristics		
Population	Height	Yield	Thousand Seed Weight
Clancy	100.00 AB	12.51 B	1.93 B
Heavin	102.50 AB	7.92 BC	2.44 A
Lacombe	96.25 AB	7.64 C	2.35 AB
SPG	121.25 A	7.37 C	2.43 A
Trawin	116.25 A	8.75 BC	2.14 AB
Vegreville	105.00 AB	6.97 C	2.31 AB
United Kingdom	67.50 B	17.16 A	2.44 A
SEM	9.473	0.994	0.105
Statistical Analysis		P-Value	
Population Effect	0.0213	<.0001	0.0206

Note: Means with the same letter in the same row are not significantly different (P>0.05). The multi-treatment comparisons using Tukey method. SEM = standard error of mean.

The differences between the yields of each population was strongly significant (P<.0001) and the differences between height and thousand seed weight were significant (P=0.0213 and P= 0.0206 respectively). The SPG and Trawin populations were significantly taller than the United Kingdom population, but not significantly different from the other Canadian populations. Hübner et al. (2003) concluded with their experiment that height was not a genetically controlled trait. The environment has a strong influence on height and therefore, we would expect plants at the same site to show no significant difference. From this, height is not a contributing factor to the different competitive abilities of populations in western Canada. The highest yielding population was from the United Kingdom. Clancy was the second highest yielding, but was not significantly different from the Heavin and Trawin populations. The similarities in yield between Clancy, Trawin and Heavin was likely not significant because the seed sources were in close proximity to one another. As the location of sourced seed moved west to Saskatoon and Alberta, the overall seed yields decreased; therefore cleavers plants in the Melfort area could have a competitive advantage as demonstrated by their higher yield. Thousand seed weight of Heavin, SPG, and United Kingdom seed was significantly higher than that of Clancy. Remaining populations were not significantly different from any of the other populations. Previous research by Hübner et al. (2003) also saw that the highest yielding population produced the lightest seed. Although they do not make any comment on why this may be, they did conclude that the highest yielding population was clearly distinguishable from all other populations. No significant differences were seen in the morphological data at the Kernen site. This can be attributed to the high amount of background cleavers that interfered with data collection.

Conclusion

Emergence timing shows that there are differences in populations with regard to the time they take to achieve 50% and 100% emergence. The proportion of emergence also varied among the populations. These differences in emergence timing likely impact the efficacy of weed control when spraying. The stage of the cleavers when spraying is very important to their control. They need to be small to have accurate control, and early emerging populations can escape this. Later emerging plants can also escape control, but may not be able to compete with established crops. Producers need to scout their field regularly before they make spraying decisions to ensure appropriate control levels are met. Effective product selection for the control of cleavers is very important as some populations have shown a slight competitive advantage over other populations.

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