Research Article

Seed germination ecology of meadow knapweed (*Centaurea × moncktonii*) populations in New York State, USA

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Abstract

The introduced meadow knapweed (*Centaurea × moncktonii* C.E. Britton), a hybrid of black (*Centaurea nigra* L.) and brown (*Centaurea jacea* L.) knapweeds, is increasingly common in pastures, meadows, and waste areas across many U.S. states, including New York. We evaluated the effects of temperature, light, seed stratification, scarification, and population on percent germination in four experiments over 2 yr. Percent germination ranged from 3% to 100% across treatment combinations. Higher temperatures (30:20, 25:15, and sometimes 20:10 C day:night regimes compared with 15:5 C) promoted germination, especially when combined with the stimulatory effect of light (14:10 h L:D compared with continuous darkness). Under the three lowest temperature treatments, light increased percent germination by 15% to 86%. Cold-wet seed stratification also increased germination rates, especially at lower germination temperatures, but was not a prerequisite for germination. Scarification did not increase percent germination. Differences between *C. × moncktonii* populations were generally less significant than differences between temperature, light, and stratification treatments. Taken together, these results indicate that *C. × moncktonii* is capable of germinating under a broad range of environments, which may have facilitated this species' range expansion in recent decades. However, *C. × moncktonii* also shows evidence of germination polymorphism: some seeds will germinate under suboptimal conditions, while others may remain dormant until the abiotic environment improves. Subtle differences in dormancy mechanisms and their relative frequencies may affect phenological traits like the timing of seedling emergence and ultimately shape the sizes and ranges of *C. × moncktonii* populations.

Introduction

Mechanisms governing seed dormancy and germination exert powerful influences over the population dynamics of plant species. By limiting when and where seeds can germinate, dormancy helps regulate seed and seedling mortality rates, population densities, and species ranges (Baskin and Baskin 2014; Fenner and Thompson 2005). Dormancy status may be affected by environmental factors such as temperature, moisture, or light; the passage of time (e.g., afterripening); damage to the seed coat (e.g., scarification); and/or by plant hormones, including abscisic acid and gibberellins (Baskin and Baskin 2014; Finch-Savage and Leubner-Metzger 2006; Kucera et al. 2005). All dormancy mechanisms reflect selective pressure to discourage germination under (or immediately before) unfavorable conditions without preventing germination altogether. Optimal solutions to this trade-off depend on species and environment, which jointly dictate whether seeds germinate quasi-simultaneously or continuously across the growing season (Newman 1963; Salisbury 1929; Young et al. 1969). Species exhibiting simultaneous germination may achieve more emergence before competitors, which can provide a significant advantage to weeds and invasive herbaceous species (Radosevich et al. 2007; Zimdahl 2007). In contrast, species exhibiting continuous germination forfeit some favorable sites early in the growing season for the sake of insurance against catastrophic seedling losses due to weather or other unpredictable events (Newman 1963; Salisbury 1929). Some species combine simultaneous and continuous germination strategies (Clements et al. 2010; Newman 1963; Young et al. 1969), either by permitting simultaneous germination only in ideal conditions or by producing seeds with variable requirements for the release of primary dormancy. The latter tendency is known as germination polymorphism (Bewley and Black 1982; Clements et al. 2010).

Centaurea is a large genus of thistle-like plants in the family Asteraceae distinguished by its complicated internal taxonomy and the invasiveness of many representatives (DiTomaso 2000;

Garcia-Jacas et al. 2006; Keil and Ochsmann 2006). Most *Centaurea* species in the United States are introduced (USDA-NRCS 2020) and many arrived from Eurasia in the late 1800s or early 1900s (Roché and Roché 1988, 1991). There are now at least 32 species present in the United States, at least 11 of which are classified as noxious weeds in at least one state (USDA-NRCS 2020). Chemical and biological approaches to management have sometimes been successful (Harris and Cranston 1979; Miller and Lucero 2014; Sheley et al. 1998; Winston et al. 2012). However, management remains difficult because multiple traits may underlie invasiveness in these species, including allelopathy (Fletcher and Renney 1963), tolerance of diverse environmental conditions (Keil and Ochsmann 2006; Qaderi et al. 2013; USDA-NRCS 2020), and abundant seed production (Schirman 1981; Watson and Renney 1974).

Traits related to dormancy and germination vary among and within *Centaurea* species in North America. In yellow starthistle (*Centaurea solstitialis* L.), diffuse knapweed (*Centaurea diffusa* Lam.), and spotted knapweed [*Centaurea stoebe* L. ssp. *micranthos* (Gugler) Hayek, often reported as *Centaurea maculosa* Lam.], exposure to red light following darkness can stimulate germination, and far-red light can reverse the effect (Joley et al. 2003; Nolan and Upadhyaya 1988). White light was also shown to stimulate germination in *C. solstitialis* (Joley et al. 1997, 2003). However, Watson and Renney (1974) found that continuous light reduced germination rates in *C. diffusa* and *C. stoebe*, while a 12:12 L:D cycle was nearly indistinguishable from continuous darkness. The discrepancy could reflect the weakening of primary dormancy under warm, dry conditions (afterripening) in these species (Eddleman and Romo 1988; Nolan and Upadhyaya 1988). Cool-moist stratification further weakened dormancy in *C. stoebe* (Eddleman and Romo 1988), while cool temperatures followed by drying induced secondary dormancy in *C. solstitialis* (Joley et al. 2003).

Centaurea seeds are capable of germinating in a variety of environments. *Centaurea diffusa*, *C. solstitialis*, and purple starthistle (*Centaurea calcitrapa* L.) have germinated under temperatures ranging from 0 to 40 C, especially under alternating cold–warm temperature regimes (Clements et al. 2010; Pitcairn et al. 2002; Young et al. 2005). In these species, germination rates may be optimized under periods of 15 to 25 C alternating with 2 to 15 C (Clements et al. 2010; Joley et al. 2003; Pitcairn et al. 2002; Young et al. 2005). Moisture levels may also limit germination in *Centaurea* species (Eddleman and Romo 1988; Larson and Kiemnec 1997; Spears et al. 1980; Watson and Renney 1974). There are several reports of intraspecific variation in germination requirements, including variation in temperature optima across accessions of *C. solstitialis* (Young et al. 2005) and the coexistence of nondormant seeds, seeds with primary dormancy released by light, and dormant seeds insensitive to light on individual plants of *C. diffusa* and *C. stoebe* (Nolan and Upadhyaya 1988).

Meadow knapweed (*Centaurea × moncktonii* C.E. Britton = *Centaurea debeauxii* Godr. & Gren. ssp. *thuillieri* Dostál, *Centaurea jacea* L. var. *pratensis* W.D.J. Koch) is a hybrid of black (*Centaurea nigra* L.) and brown (*Centaurea jacea* L.) knapweeds, although Vochin knapweed (*Centaurea nigrescens* Willd.) may also contribute to the hybrid swarm in North America (Keil and Ochsmann 2006). It was likely introduced directly from Europe in the late 1800s (Roché and Roché 1991). This theory is consistent with the deep introgression reported in North American *C. × moncktonii* populations, although more recent hybridization in North America is also possible (Lachmuth et al. 2019). Like its parental

species, *C. × moncktonii* is a self-incompatible perennial that flowers between May and November (Hardy et al. 2001; Keil and Ochsmann 2006). It exhibits a preference for wetter environments (Roché and Roché 1991). Seed dispersal occurs by wind over short distances and primarily by humans over long distances (Roché and Roché 1991; Soons and Heil 2002).

Although *C. × moncktonii* was introduced to the United States more than a century ago, it has presented increasingly serious management problems in recent decades. It has now been reported in more than 20 states and is considered a noxious weed in the Pacific Northwest (Kartesz 2014; Keil and Ochsmann 2006; Winston et al. 2012). In New York, *C. × moncktonii* is becoming increasingly common in moist pastures, meadows, and waste areas (Eckel 2012; Weldy et al. 2020). The ongoing development of best management practices (Miller and Lucero 2014; Thorpe et al. 2009; Winston et al. 2012) may be aided by a better understanding of this invasive hybrid. Because *C. × moncktonii* is biologically and ecologically distinct from its parental species (Keil and Ochsmann 2006; Roché and Roché 1991; Winston et al. 2012), with evidence of transgressive segregation in some traits (Lachmuth et al. 2019), research efforts should be specific to *C. × moncktonii*. One important topic is germination, which is not well understood in either the hybrid or the parental species. *Centaurea × moncktonii* seeds usually germinate in the spring, but autumn germination can also occur (Milbrath and Biazzo 2020; Winston et al. 2012). Autumn germination also occurs in other *Centaurea* species (Lesica and Shelly 1996; Roché et al. 1997; Watson and Renney 1974), including *C. jacea* (Kahmen and Poschlod 2008). After observing high rates of autumn seedling emergence in field-placed seed pans (Milbrath and Biazzo 2020), we designed the present study to explore factors influencing seed germination in *C. × moncktonii*. We evaluated the effects of temperature, light, cold-wet stratification, scarification, and *C. × moncktonii* population on seed germinability. Based on the published germination requirements of other *Centaurea*, which often involve temperature and light, rarely include stratification or scarification, and can be similar between species, we developed five hypotheses:

1. *Centaurea × moncktonii* seed germination rates will be greatest at moderate temperatures.
2. More seeds will germinate in alternating light conditions than constant darkness.
3. Seed germination rates will not vary between populations.
4. Cold-wet stratification will not affect germination rates.
5. Scarification will not affect germination rates.

Materials and Methods

Seed Collection

Mature capitula were collected in August 2015 for Experiments 1–3 from four sites in central and eastern New York State and in August 2016 for Experiment 4 from one of the previous sites in central New York State (Table 1). The Jacobson site (private property; 42.4981°N, 76.2475°W) in Cortland County is a little-used hayfield on Erie silt loam soil (fine-loamy, mixed, active, mesic Aeric Fragiaquepts). McLean Meadow (Cornell Botanic Gardens; 42.5453°N, 76.2681°W) in Tompkins County is a formerly abandoned pasture on Howard (loamy-skeletal, mixed, active, mesic Glossic Hapludalfs) and Palmyra (fine-loamy over sandy or sandy-skeletal, mixed, active, mesic Glossic Hapludalfs) soils that is mowed every few years to maintain it as a meadow.

Table 1. Seed handling and treatments for four germination experiments on *Centaurea × moncktonii* seeds collected in New York State (*n* = 5 per treatment combination).

Experiment	Scarified	Cold-dry storage	Cold-wet stratified	Temperature	Light	Population[a]
		wk		C, L:D	h, L:D	
1	Yes	4	No	15:5, 20:10, 25:15, 30:20	14:10 or 0:24	Jacobson, McLean
2	Yes	4	Yes	15:5, 20:10, 25:15, 30:20	14:10 or 0:24	Jacobson, McLean
3	Yes	24	Yes	15:5, 20:10, 25:15, 30:20	14:10 or 0:24	Jacobson, McLean, FLNF, Fort Plain
4	Yes, No	13	No	15:5, 20:10, 25:15, 30:20	14:10 or 0:24	FLNF

[a]FLNF, Finger Lakes National Forest.

The Finger Lakes National Forest site (FLNF, USDA Forest Service; 42.5283°N, 76.7764°W) in Schuyler County is a periodically mowed pasture on Erie silt loam soil. The Fort Plain site (private property; 42.9003°N, 74.7458°W) in Montgomery County in eastern New York, on Hornell silt loam soil (fine, illitic, acid, mesic Aeric Endoaquepts), has been in hay production or grass fallow for many years. All sites had extensive populations of *C. × moncktonii* that have been present for at least 10 yr (Milbrath and Biazzo 2020).

Capitula were stored in paper bags at room temperature for 4 to 6 wk before cleaning. A rubbing board was used to separate the seeds from the dried capitula, potentially scarifying the seeds, and chaff was removed by sieving followed by a seed blower (General Seed Blower, New Brunswick General Sheet Metal Works, New Brunswick, NJ). For the non-scarified treatment in Experiment 4 (Table 1), seeds were gently separated from the capitula by hand. All seeds were then stored dry at 4 C for 4 wk (Experiments 1, 2), 13 wk (Experiment 4), or 24 wk (Experiment 3) before experiments were initiated (Table 1). Seeds were counted into lots of 50 using a seed counter (Seedburo Seed Counter, Seedburo Equipment, Des Plaines, IL) and placed on moist blotter paper in petri dishes, which were sealed with Parafilm® (Bemis Company, Neenah, WI).

Experimental Design and Treatments

Experiment 1. Four separate experiments were performed. Factors included temperature and light for all experiments and seed population, stratification, and scarification treatments for select experiments (Table 1). Experiment 1 followed a three-way factorial design involving four temperatures, two light conditions, and two populations. Temperature treatments consisted of day and night temperatures on a 14:10 h cycle: 15:5 C (day:night), 20:10 C, 25:15 C, and 30:20 C. These temperature regimes are reasonable for New York State and the *Centaurea* genus (Clements et al. 2010; Joley et al. 2003; Pitcairn et al. 2002; Young et al. 2005). One incubator (model I36VL, Percival Scientific, Perry, IA) was used for each temperature, with petri dishes distributed randomly within incubators. For the light treatment, uncovered petri dishes of seeds were exposed to a 14:10 h L:D photoperiod with light levels of approximately 80 µmol m^{-2} sec^{-1} at a distance of 15 cm from the light fixtures. Petri dishes of seeds in the dark treatment were placed in the same growth chambers but were wrapped in aluminum foil. All petri dishes were placed in sealed plastic bags to further reduce drying. Non-stratified, scarified seeds from the Jacobson and McLean populations were tested. Each treatment combination was replicated five times for a total of 80 experimental units (each experimental unit was a dish of 50 seeds).

Dishes were checked for germination daily for 7 d, every other day for the next 10 d, and then twice more before the experiment ended after 28 d. More than 90% of germination occurred within the first 7 d (data not shown). Percent germination was calculated for each dish, with germination defined as the emergence of the radicle from the seed coat. Seeds for the dark treatment were checked in a dark room under a dim green light (flashlight wrapped in multiple layers of green plastic film; Bemiss-Jason Cellophane, Neenah, WI) (Nolan and Upadhyaya 1988; Oliveira and Garcia 2019). Aluminum foil was replaced each time the seeds were checked to prevent seeds from being exposed to light from pinholes resulting from the folding and unfolding of the foil.

Viability of seeds for each dish was estimated immediately following the experiment by holding nongerminated seeds at ca. 28 C for an additional 2 wk of germination followed by a 0.1% tetrazolium chloride test of remaining firm seeds. Seeds that were covered with fungi or that collapsed when pressed gently were considered dead. Total viability averaged 93% to 95% across experiments.

Experiment 2. The experimental design and data collection were the same as for Experiment 1, except that dishes of seeds were cold-wet stratified for 28 d at 4 C before testing the seeds for germination (Table 1). Dishes for the dark treatment were wrapped in aluminum foil before stratification. A total of 80 experimental units were used.

Experiment 3. The experimental design and data collection were similar to Experiment 2, including cold-wet stratification of seeds, except that four populations were tested (Jacobson, McLean, FLNF, Fort Plain; Table 1) for a total of 160 experimental units. Light-proof bags (Orca Grow Film, Urban Sunshine, Orlando, FL) were used instead of aluminum foil for the dark treatment.

Experiment 4. The experimental design and data collection were similar to Experiment 1, except that two scarification treatments—scarified (rubbing board removal of seeds) and non-scarified (hand removal of seeds)—were tested with a single population (FLNF; Table 1). Light-proof bags were used for the dark treatment. A total of 80 experimental units were used.

Statistical Analysis

For each experiment, percent germination data were transformed using the arcsine square-root transformation and analyzed with ANOVA (PROC MIXED, SAS Institute, Cary, NC). Standard errors for back-transformed data were approximated using the methods of Deming (1964). Stepwise removal of nonsignificant interaction terms determined the best model. Means for significant factors were separated using Tukey's honest significant difference (HSD) test.

Results and Discussion

We conducted four factorial experiments to test the effects of temperature, light, stratification, scarification, and population on *C. × moncktonii* seed germinability, which enabled us to evaluate our five hypotheses.

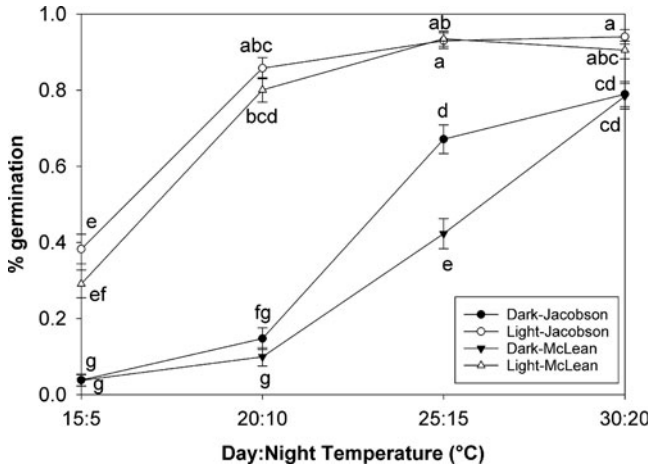*Hypothesis 1: Centaurea × moncktonii Seed Germination Rates Will Be Greatest at Moderate Temperatures*

Figure 1. Percent germination in Experiment 1 (non-stratified and scarified seeds) from two *Centaurea × moncktonii* populations from New York State under different temperature regimes and light environments. Points not labeled with the same lowercase letter are significantly different according to Tukey's HSD test.

Figure 3. Percent germination in Experiment 3 (cold-wet stratified and scarified seeds) from four *Centaurea × moncktonii* populations from New York State under different temperature regimes and light environments. FLNF, Finger Lakes National Forest. Points not labeled with the same lowercase letter are significantly different according to Tukey's HSD test.

Figure 2. Percent germination in Experiment 2 (cold-wet stratified and scarified seeds) averaged over two *Centaurea × moncktonii* populations from New York State under different temperature regimes and light environments. Points not labeled with the same lowercase letter are significantly different according to Tukey's HSD test.
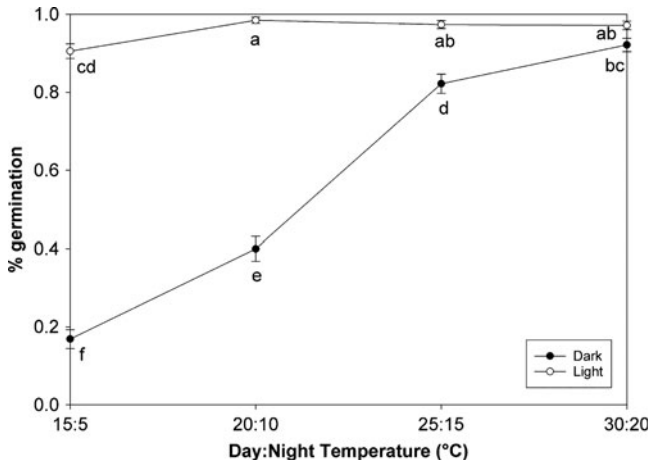

Figure 4. Percent germination in Experiment 4 (non-stratified and scarified or non-scarified seeds) from the Finger Lakes National Forest (FLNF) population in New York State under different temperature regimes and light environments. Points not labeled with the same lowercase letter are significantly different according to Tukey's HSD test.

Across all experiments and populations, percent germination was typically highest for the three warmest temperature regimes under light. Differences between these three regimes were usually not significant. For example, germination under light in Experiment 1 was higher (80% to 94%) under the three warmest temperature regimes than in the coldest treatment (29% to 38%) (Figure 1). Experiment 2 was different in that germination under light was highest (97% to 99% averaged over both populations) under the three warmest temperature regimes but also high (91%) at 15:5 C (Figure 2). The temperature responses observed under light in Experiments 3 and 4 were qualitatively similar to the patterns observed in Experiments 2 and 1, respectively (Figures 3 and 4), with all populations in Experiment 3 achieving more than 80% germination, even in the coldest treatment. Without light, germination was highest in the warmest and, in some cases, the second-warmest temperature regimes (Figures 1–4). In Experiment 1, dark germination rates
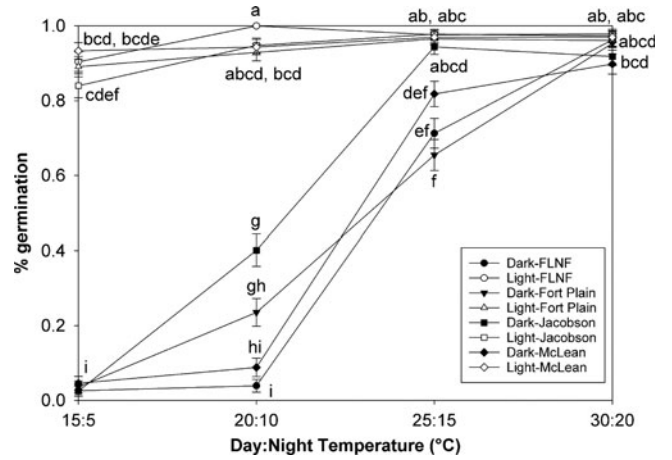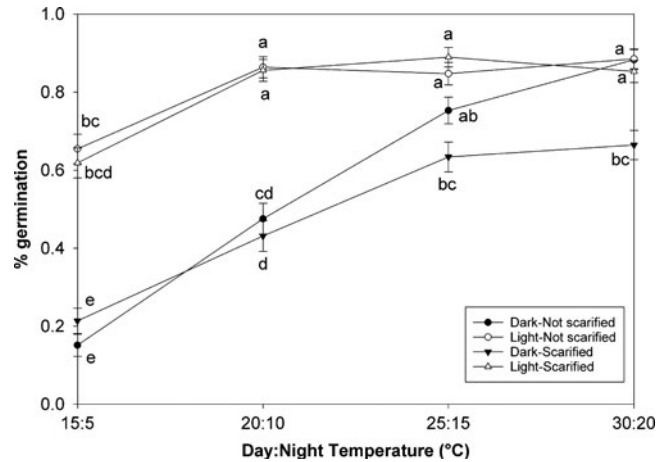
were higher in the warmest two treatments (42% to 78%) than in the coldest two treatments (4% to 15%) (Figure 1). In Experiments 2, 3, and 4, dark germination often increased as temperatures increased (Figures 2–4). In all cases, germination without light was higher under 25:15 C regimes than under 20:10 C regimes (Figures 1–4). Because percent germination generally increased (and did not decrease) with increasing temperatures when other factors were held constant (Table 2), our hypothesis is refuted.

These data suggest that the optimal temperature regime for germination of New York populations of *C. × moncktonii* may be 20 to 30 C alternating with 10 to 20 C. These ranges are slightly warmer than those measured in other *Centaurea* species, in which germination may be maximized under periods of 15 to 25 C alternating with 2 to 15 C (Clements et al. 2010; Joley et al. 2003; Pitcairn et al. 2002; Young et al. 2005). The difference could reflect either

Table 2. ANOVA results for four experiments examining the effects of temperature, light, seed population and/or scarification on non-stratified or stratified seeds of *Centaurea × moncktonii* from New York State.

Factor	num. df, den. df[a]	*F*-value	P-value
Experiment 1			
Temperature (T)	3, 64	308.46	<0.001
Light (L)	1, 64	554.35	<0.001
Population (P)	1, 64	12.36	<0.001
T*L	3, 64	36.72	<0.001
T*P	3, 64	0.86	0.465
L*P	1, 64	0.42	0.517
T*L*P	3, 64	4.08	0.010
Experiment 2			
Temperature	3, 71	99.1	<0.001
Light	1, 71	475.96	<0.001
Population	1, 71	29.18	<0.001
T*L	3, 71	61.31	<0.001
Experiment 3			
Temperature	3, 128	377.32	<0.001
Light	1, 128	1510.68	<0.001
Population	3, 128	3.37	0.0206
T*L	3, 128	241.27	<0.001
T*P	9, 128	5.12	<0.001
L*P	3, 128	12.74	<0.001
T*L*P	9, 128	7.06	<0.001
Experiment 4			
Temperature (T)	3, 64	109.09	<0.001
Light (L)	1, 64	252.81	<0.001
Scarification (S)	1, 64	6.23	0.015
T*L	3, 64	20.5	<0.001
T*S	3, 64	3.75	0.015
L*S	1, 64	4.27	0.043
T*L*S	3, 64	3.91	0.012

[a]Abbreviations: num. df, numerator degrees of freedom; den. df, denominator degrees of freedom.

interspecific variation or a regional effect; previous studies were carried out in the western United States. Environmental requirements for germination may be relatively evolvable (Hierro et al. 2009; Pitcairn et al. 2002; Young et al. 2005).

Hypothesis 2: More Seeds Will Germinate in Alternating Lighted Conditions Than Constant Darkness

The lower germination rates and increased temperature responses observed in darkness (Figures 1–4) indicate that light and a light–temperature interaction, respectively, affect percent germination. Both factors were highly significant in all experiments (Table 2). The 14:10 h L:D cycle always increased germination relative to continuous darkness under the 15:5 and 20:10 C temperature regimes and usually also under the 25:15 C regime (Figures 1–4). Because alternating lighted conditions stimulated germination in all experiments, our hypothesis is supported.

Most previous studies have found that light can stimulate germination in *Centaurea* species (Joley et al. 1997, 2003; Nolan and Upadhyaya 1988), with occasional exceptions potentially linked to storage treatments that weakened primary dormancy (Nolan and Upadhyaya 1988; Watson and Renney 1974). The significant effects reported here agree with this consensus. Our findings suggest that *C. × moncktonii* germination may be inhibited by low-light environments or deep burial in soil or litter. However, light limitation will not completely prevent germination. Under warm temperature regimes, we observed high germination rates in the dark relative to some previous *Centaurea* measurements (Joley et al. 1997, 2003; Nolan and Upadhyaya 1988). High rates of germination in the dark have

been reported before (Maguire and Overland [1959] cited in Young et al. 2005; Clements et al. 2010; Davis 1990; Eddleman and Romo 1988; Pitcairn et al. 2002; Watson and Renney 1974; Young et al. 2005), including in the parental species *C. nigra* (Silvertown 1980). Our findings might represent a conservative estimate of the stimulatory effect of light: there is some evidence that the dim green light used to check dark treatments can affect germination rate (Baskin and Baskin 2014). This possibility should be accounted for in future investigations of dark germination and the consistently significant interactions between temperature and light environments (Table 2).

Hypothesis 3: Seed Germination Rates Will Not Vary between Populations

Germination rates were similar across populations under all temperature regimes with light and the lowest and highest temperature treatments in the dark (Figures 1 and 3), although there was some variability between populations under midrange temperature treatments in the dark (Figures 1 and 3). In Experiment 2, the Jacobson population had a higher overall germination rate (86%) than the McLean population (77%) (data not shown). In one case, population affected responses to temperature. In Experiment 3, the lowest germination (3% to 9%) occurred at 15:5 C in the dark for all four populations and also at 20:10 C in the dark for the FLNF and McLean populations, while the Fort Plain and Jacobson populations had higher germination at 20:10 C (Figure 3). However, dark germination rates differed between the 15:5 C and 20:10 C treatments for the FLNF population in Experiment 4 (Figure 4). Apart from the above exceptions, differences between populations were usually insignificant (Figures 1 and 3). Our hypothesis is generally supported.

Germination requirements and rates can vary substantially within *Centaurea* species (Clements et al. 2010; Pitcairn et al. 2002; Young et al. 2005). However, the four sites in this study were distinguished by soil type and land-use characteristics rather than by geographic distance or climate. Optimal dormancy mechanisms and germination behavior may not differ strongly between sites. Emerging differences between population gene pools could also be blurred by migration or breeding with individuals elsewhere on the admixture continuum between *C. nigra* and *C. jacea* (Lachmuth et al. 2019). A prior field-based study reported larger differences in *C. × moncktonii* germination across the same four populations tested here (Milbrath and Biazzo 2020). Our results are not incompatible with this report, as observed variation between field sites may have a strong environmental component. It is also worth noting that, while differences between populations typically fell below the significance threshold in pairwise comparisons within treatments (Figures 1 and 3), they did add explanatory power at the experimental level (Table 2).

Hypothesis 4: Cold-Wet Stratification Will Not Affect Germination Rates

Cold-wet stratification appeared to increase germination rates relative to non-stratified seeds (compare Figures 1 and 2), particularly at lower temperatures in either light treatment. The hypothesis is refuted, although future work should confirm this result by testing stratified against non-stratified seeds in a single experiment. Cold-wet stratification has been shown to break dormancy in *C. stoebe* (Eddleman and Romo 1988), whereas other North American and European *Centaurea* species may not germinate at higher rates following stratification (Luna et al. 2008; Milberg and Andersson 1998; White et al. 2009). The presence or absence

of stratification effects may reflect native habitat: cold and wet conditions are more likely to alleviate dormancy in temperate species that typically germinate after cold, wet winters (Baskin and Baskin 2014). Other aspects of handling, such as seed storage time before stratification, can also enhance or diminish the effects of stratification (Eddleman and Romo 1988). In this study, the duration of cold-dry storage (4 wk, Experiment 2; or 24 wk, Experiment 3) before stratification did not have an obvious impact on germination rates (compare Figures 2 and 3). However, it is possible that testing short and long storage periods within the same experiment, including a no-storage treatment, or increasing storage temperature would reveal subtle effects attributable to afterripening.

Hypothesis 5: Scarification Will Not Affect Germination Rates

For non-stratified seeds of the FLNF population, scarification did not significantly increase germination relative to non-scarified seeds under any combination of temperature and light treatments (Figure 4). A significant interaction did occur among temperature, light, and scarification treatments (Table 2), which is partially attributable to the fact that non-scarified seeds had a higher germination rate than scarified seeds under the 30:20 C temperature regime in darkness (Figure 4). Because other differences between scarified and non-scarified seed germination rates were not significant (Figure 4), our hypothesis is mostly supported. There has been little work on the effects of scarification in *Centaurea* species, although it seems to be unnecessary in Californian *C. solstitialis* (Benefield et al. 2001). In European *Centaurea* taxa, elaiosome removal may either promote or inhibit germination (Imbert 2006; Viegi et al. 2003), and scarification by fire does not promote germination (Riba et al. 2002). It is not yet clear how these results relate to mechanical scarification by rubbing board (present study) or to events that increase seed permeability in nature (Baskin and Baskin 2014).

Ecological Implications

Centaurea × moncktonii seed germination occurred across a wide range of abiotic environments, although rates were highest under warm temperatures and light following wet-cold stratification. Under these optimal conditions, germination rate approached 100% for all populations (Figures 2 and 3). These values are consistent with the maximum germination rates of other North American *Centaurea* species (Joley et al. 2003; Nolan and Upadhyaya 1988; Pitcairn et al. 2002; Young et al. 2005). They are also consistent with estimates of initial seed viability in *C. × moncktonii*, which are higher than the actual germination rates achieved in field-placed trays (Milbrath and Biazzo 2020). The potential for high germination rates may be adaptive if the multiyear survival of dormant seeds is low (Milbrath and Biazzo 2020).

For all experiments and populations, minimum germination occurred under the coldest temperature regime(s) in the dark. Germination rates were always below 21% under these conditions, regardless of stratification, storage time, and scarification treatment (Figures 1–4). Although we did not test stratified against non-stratified seeds within a single experiment, our data suggest that cold-wet stratification increased germination rates under cold and/or dark conditions (compare Figure 1 with Figures 2 and 3). Most dramatically, stratification increased germination under the coldest light treatment from under 40% to more than 80% for all populations (compare Figure 1 with Figures 2 and 3). This finding may help explain why the primary seedling flush occurs in spring

(Winston et al. 2012; LRM, personal observation): after dormancy-weakening winter exposure to wet and cold conditions, a broad range of abiotic environments will support germination. In autumn, seeds have not recently experienced winter conditions, so they may be more dormant and may require more specific temperature and light environments to germinate.

A small percentage of seeds in every population germinated even under unfavorable conditions (Figures 1–4). After observing a similar trend in *C. diffusa* and *C. stoebe*, Nolan and Upadhyaya (1988) concluded that seeds fell into three categories: seeds without primary dormancy, seeds with primary dormancy released by light, and seeds with primary dormancy not released by light. All three categories were represented in each species, site, and individual plant. The authors viewed this germination polymorphism as an adaption to promote distribution in time (Bewley and Black 1982; Nolan and Upadhyaya 1988). In *C. diffusa*, variation in germination time not only provides population-level protection against disaster (Berube and Myers 1982) but also decreases intraspecific competition (Sheley and Larson 1996). Our findings suggest that *C. × moncktonii* may likewise rely on germination polymorphism, perhaps for similar reasons. For *C. × moncktonii* seeds, variation in light requirements is only one of several sources of variation in germinability (Table 2). Nolan and Upadhyaya (1988) also reported population-level and plant-level differences in average germinability, which reflected differences in the proportions of seeds in the three dormancy categories. It could be useful to view the subtle population-level differences observed in *C. × moncktonii* (Figures 1–3) through a similar lens. If selection and/or genetic drift can modify the relative frequencies of traits related to germination, these changes may lead to measurable changes in the distribution of germination requirements within the population and ultimately to visible changes in outcomes like the frequency of autumn germination.

Several authors have suggested that the invasiveness and competitiveness of *C. × moncktonii* (Lachmuth et al. 2019; Vilà et al. 2000) and other *Centaurea* hybrids (Blair et al. 2012; Blair and Hufbauer 2010) may reflect increased fitness due to hybridization. The classic hypothesis of increased hybrid fitness (Ellstrand and Schierenbeck 2000; Hovick and Whitney 2014) is theoretically consistent with the fact that *C. × moncktonii* may occupy larger or different ranges than the parental species *C. nigra* and *C. jacea* in North America (Roché and Roché 1991). However, this hypothesis cannot be tested using only the data presented here, given the scarcity of information about germination requirements for the parental species. The relationships between dormancy traits, germination rates, and recruitment rates are also complex and highly dependent on environmental conditions (Milbrath and Biazzo 2020). Ongoing research efforts seek to characterize seedling emergence success and establishment in different habitats.

Conclusions

Centaurea × moncktonii, a hybrid of *C. nigra* and *C. jacea*, is an invasive weed found in many parts of North America (Keil and Ochsmann 2006). Seeds from four New York populations of *C. × moncktonii* germinated under a broad range of temperature, light, seed stratification, and scarification treatments. However, the treatments had significant impacts on germination rates, which were maximized under warm temperatures and light following wet-cold stratification. These findings suggest that *C. × moncktonii* uses germination polymorphism (Bewley and Black 1982; Nolan

and Upadhyaya 1988) to combine simultaneous and continuous germination strategies (Clements et al. 2010; Salisbury 1929). Nearly all seeds can germinate under optimal conditions, whereas suboptimal conditions allow only weakly dormant and non-dormant seeds to germinate. This approach allows for adaptation to diverse environments and may facilitate future population increases and range expansion.

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