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The Effects of Fertilization and Herbivory on the Phenology of the Understory Vegetation of the Boreal Forest in Northwestern Canada

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Abstract

Environmental conditions associated with climate change, such as earlier snowmelt, warmer spring temperatures, and increased soil mineralization, have resulted in shifts in the timing of plant phenological events. We assessed the effects of fertilizer and herbivory on the phenology of 7 plant species from a boreal forest understory using long-term experimental plots in the southwest Yukon. Fertilizer and fencing treatments were initiated in 1990, and discontinued in half of each plot in 2000. There were few effects on phenology of either fertilizer or fencing. In some species, fertilizer affected the final phenological stage reached, but the presence and direction of the effect was species-dependent. *Epilobium angustifolium* was the only species where the timing of phenological stages responded to fertilization; early phenological stages were advanced with fertilizer. First leaf expansion for *Arctostaphylos uva-ursi* occurred earlier in fenced plots, although this effect disappeared in plots where the fencing treatment had been discontinued. We conclude that previously observed changes in species abundance with fertilizer treatments are likely not caused by changes in the phenology of these species, and are more likely due to fertilization imposed changes in vegetative growth.

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Introduction

As global temperatures continue to rise, considerable research effort has focused on how vegetation will respond to changing environmental conditions (Larigauderie and Kummerow, 1991; Wookey et al., 1993; Arft et al., 1999; Cleland et al., 2006). Environmental effects of warming in northern ecosystems may include changes in soil nutrient levels resulting from higher nutrient mineralization in soils (Aerts et al., 2006). Animal species are also expected to respond to climate change by shifting their ranges and distributions northward (Walther et al., 2002), potentially increasing herbivory on northern plant populations. Many plants have responded to climate change by emerging and flowering earlier than in the past; shifting phenological events have been documented across the globe (Walther et al., 2002; Parmesan, 2006; Miller-Rushing and Primack, 2008). It is predicted that the highest temperature increases will occur in northern regions (Wookey et al., 1993; Berteaux et al., 2004), which may influence the phenological development of northern plant populations. These predictions have caused northern plants to be labeled as an ''early warning system'' (Aerts et al., 2006).

Many phenological events, such as budding or flowering, are occurring earlier than in the past due to climate warming (Parmesan and Yohe, 2003). These changes may affect not only plants, but multiple trophic levels. For instance, plants that emerge earlier may be more susceptible to frost events, plants may begin to flower before the arrival or emergence of their pollinators, or plants may emerge earlier and provide more food resources to herbivores earlier in the growing season. As the timing of flowering and other correlated life history traits of plants change, interactions among species may also change either to the advantage or disadvantage of neighbors; for example, plants that begin to co-flower may either help to attract pollinators or compete for their services (Elzinga et al., 2007).

Monitoring phenological events, such as the timing of emergence or flowering, is a simple means to assess the effects of climate change on plant performance (Walther et al., 2002; Aerts et al., 2006; Zhang et al., 2010). Phenology is an adaptive trait that is able to evolve quickly in changing climatic conditions (Chuine, 2010) and may respond to treatments earlier than plant composition, especially in northern ecosystems composed of highly clonal species. Several species' distribution models show that phenology is a ''keystone trait'' in species distribution and fitness (Chuine, 2010). These models show that the northern limit of a species range is largely influenced by the inability to undergo full fruit maturation (Chuine, 2010), whereas the southern distribution is limited by the inability to flower due to a lack of cold temperatures necessary to disrupt bud dormancy (Chuine, 2010). Phenology also plays an especially critical role in the survival and adaptation of arctic populations, as phenological asynchrony between seasonal activities of species and environmental conditions may result in population crashes (Walther et al., 2002; Berteaux et al., 2004); therefore, it is crucial for species living in boreal environments to time their phenological activities accordingly (Chuine, 2010).

Most northern sites (areas of high latitude and altitude) have very low levels of available soil nutrients, particularly nitrogen, which can limit plant productivity and consequently reduce species abundance (Turkington et al., 2002; Aerts et al., 2006). These effects of nitrogen deposition on vegetation in the boreal forest often interact with biotic factors from multiple trophic levels, such as parasitic fungus, herbivores, and predators (Strengbom et al., 2005; Nordin et al., 2009). Herbivores may also directly have a substantial influence on the understory vegetation of the boreal

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TABLE 1

Summary of 2-way split-plot ANOVA testing the effects of fertilizer, fencing, and continuation of treatments on final phenological stage reached before plant senescence in seven boreal forest understory species. Only the effects of fertilizer and fencing are presented. Three species were not present in fertilized plots and only the effects of fencing are presented. Bold values are significant at $p < 0.05$.

Species	d.f.	Fertilizer		Fence		Fertilizer × Fence	
		F	*P*	*F*	*P*	*F*	*P*
Achillea millefolium	1,12	0.09	0.766	0.26	0.621	1.25	0.286
Epilobium angustifolium	1,12	7.37	**0.019**	0.13	0.720	2.38	0.149
Festuca altaica	1,12	4.30	0.060	1.44	0.253	0.58	0.460
Mertensia paniculata	1,12	4.90	**0.047**	0.73	0.411	1.56	0.235
Arctostaphylos uva-ursi	1,6			1.71	0.239		
Linnaea borealis	1,6			1.69	0.241		
Lupinus arcticus	1,6			1.51	0.265		

forest. For example, approximately every 10 years, the snowshoe hare population can reach densities up to 300 km^{-2} in southwest Yukon (Krebs et al., 1986). The understory plants are the primary source of food for these herbivores during the snow-free season (Seccombe-Hett, 1999; Turkington et al., 2002) and during the hare population peaks plants are subject to increased levels of grazing. Both soil nutrient levels and herbivores have been previously shown to affect plant phenology. A study in an arctic tundra community showed few significant effects of fertilizer on phenological timing of tussock tundra species (sedges, deciduous shrubs, and evergreen shrubs) after 3 years, although it did show an advancement in the timing of leaf expansion in evergreen shrubs in fertilized plots (Larigauderie and Kummerow, 1991). Previous studies have also shown a significant effect of herbivory on phenology; Huhta et al. (2009) observed that injured or grazed plants flowered and set seed significantly later than uninjured plants.

In 1990 a long-term experiment was established in the understory of a boreal forest in southwest Yukon to determine how fertilization and the exclusion of major herbivores influenced individual plant species abundance and, as a result, community composition (Turkington et al., 2002). After 10 years of treatments, results indicated that the composition and abundance of the understory plant community are determined almost exclusively by nutrient availability (Turkington et al., 2002). Fertilization caused a decrease in plant species richness and evenness but the exclusion of herbivores had very little impact on the abundance of the vegetation or species diversity (Turkington et al., 2002). Other studies have also shown a similar decrease in plant species diversity with soil nutrient enrichment (Rajaniemi, 2002), especially if species were rare (Suding et al., 2005). The mechanism for this decline is not completely understood and still very much debated (Suding et al., 2005). Changes in plant phenology offer one potential mechanism for this decline; for example, decreases in a species' abundance may be due to decreases in seed output resulting from delayed flowering.

We used the long-term experimental plots of Turkington et al. (2002) to test whether plant phenology in the understory plant community is affected by nutrient availability or herbivory. In an earlier examination of these plots, species composition responded primarily to fertilizer, and not fencing (Turkington et al., 2002), and consequently we hypothesize that there will also be stronger effects of fertilizer than fencing on plant phenology. Despite the stronger effects of fertilizer on species composition, however, both treatments have the potential to affect plant phenology. First, changes in species composition resulting from changes in phenology may take multiple years to become apparent, especially in ecosystems dominated by perennial plants, and effects on phenology may occur in treatments where species composition effects are not yet apparent. Second, we examined plant phenology after an additional 6 years of treatments, compared with the Turkington et al. (2002) study, giving more time for effects of both treatments to become apparent. In the earlier study, fertilization significantly affected the cover of 7 species after 10 years of fertilization in these plots; four species increased (*Achillea millefolium*, *Epilobium angustifolium*, *Festuca altaica*, and *Mertensia paniculata*) and three decreased (*Linnaea borealis*, *Lupinus arcticus*, and *Arctostaphylos uva-ursi*), but fencing did not affect the cover of any species. We explored whether effects of fertilizer on species composition may be due, in part, to changes in plant phenology.

Materials and Methods

SITE DESCRIPTION

The study area is near Kluane Lake in the Shakwak Trench, a wide glacial valley in southwest Yukon in northern Canada, and is described in detail in Turkington et al. (2002). The area is in the rain shadow of the St. Elias Mountains and receives a mean annual precipitation of ca. 230 mm, mostly falling as rain during the summer months, but including an average annual snowfall of about 100 cm. The region is a closed to open spruce forest community and the dominant tree is white spruce (*Picea glauca* (Moench) Voss), interspersed with stands of trembling aspen (*Populus tremuloides* Michx.) and balsam poplar (*Populus balsamifera* L.). The understory is dominated by shrub willows (*Salix glauca* (L.) and other *Salix* spp.), dwarf birch (*Betula glandulosa* Michx.), soapberry (*Shepherdia canadensis* (L.) Nutt.), and a well-developed ground layer (see Table 1). Snowshoe hares (*Lepus americanus* Erxleben) are the primary herbivore and they undergo a regular 10- to 12-year population cycle. In 2006 the snowshoe hares were at the peak of this population cycle in the Kluane region (Sheriff et al., 2009). Other small mammals in this system, including the northern red backed vole (*Clethrionomys rutilus*) and the meadow vole (*Microtus pennsylvanicus*), include understory vegetation in their diet but these are not excluded with the mesh size used, and were not examined in this experiment. Nevertheless, although Treberg et al. (2010) showed that fertilizer may attract voles, there were few obvious signs of vole activity in our plots, regardless of fertilizer or fencing treatments.

EXPERIMENTAL DESIGN

To assess the effects of various treatments on the phenology of plant species we used experimental plots established in 1990.

The experiment site is in moderately open spruce forest (45–60% canopy cover, typically 160–220 stems ha^{-1}) with a well-developed (>90% cover) understory and was approximately 2 km to the south of Boutellier summit on the Alaska highway (61°02′N, 138°22′W). There are sixteen 5 m × 5 m plots placed in small forest openings that were dominated by understory species and did not have any rooted trees. Only a few had scattered shrubs, most of which were less than 1m tall. The experiment was a 2 × 2 factorial design, with fertilization and herbivore reduction (fencing) as the main treatments, with plots divided randomly among the four treatments. Fences were 1 m high and made of galvanized chicken wire with 2.5 cm mesh. Fertilizer (N:P:K 35:10:5) was applied as slow release pellets in each year after snowmelt, between mid-May and early June, at a rate of 1.25 kg per 5 m × 5 m plot per year, resulting in an addition of 17.5 g N m^{-2} yr^{-1}, 5 g P m^{-2} yr^{-1} and 2.5 g K m^{-2} yr^{-1}. After 10 years of study, at the end of the 1999 growing season, the plots were divided in half (bisecting each subplot) and beginning in 2000 one-half continued to receive the original treatment, while the other half no longer received any treatment. More details of the study site and experimental treatments are described in Krebs et al. (2001) and Turkington et al. (2002).

PHENOLOGICAL ASSESSMENT

Phenological stages were assessed on the 7 most abundant species in the experimental plots: arctic lupine (*Lupinus arcticus* S. Wats), northern rough fescue (*Festuca altaica* Trin.), twin flower (*Linnaea borealis* L.), bearberry (*Arctostaphylos uva-ursi* (L.) Spreng.), bluebells (*Mertensia paniculata* (Aiton) G. Don), yarrow (*Achillea millefolium* L. var. *borealis* (Bong.) Breitung), and fireweed (*Epilobium angustifolium* L. s.l.). Their phenology was recorded throughout the 2006 growing season for each treatment, in both treated plots and those in which the treatments had been discontinued in 2000.

As all the plants we examined are perennials, individuals were marked in late August 2005. Three individuals of each of the 7 species were marked in both the treated and untreated halves of each plot using plastic rings secured to the soil surface. For species in which discrete individuals are difficult to determine (*F. altaica*, *A. uva-ursi*, *L. borealis*), 3 transects of 1 m × 0.1 m containing numerous stems of each species were marked in each plot to indicate which individuals would be examined. Throughout the 2006 growing season, beginning immediately following snowmelt in early May, marked plants were visited every third day and the phenological stage recorded as follows: shoot emergence, first true leaf expanded, presence of buds, presence of first fully developed flower, presence of fruits, presence of ripened fruits/seeds, and senescence. Shoot emergence for most species was defined as their first emergence from the soil surface, but for the two prostrate shrubs, *A. uva-ursi* and *L. borealis*, emergence was defined as the first appearance of new growth. The appearance of buds was obvious for all species except *A. uva-ursi* in which buds were formed in the previous season. Ripened fruits and seeds were recorded when the first fruits had dropped, withered, or dehisced. Senescence was recorded when the majority of the leaves had yellowed, leaves had abscised, or the plant was substantially wilted. Evidence of herbivory was observed in some of the plots over the course of the season; fewer than 5% of the marked individuals, however, showed signs of herbivory. No direct damage of fertilizer (e.g. fertilizer burns on leaves) was observed on any of the marked plants.

ECOSYSTEM PROPERTIES

Ecosystem variables were examined in the 2005 growing season, the year before the phenological assessment. Both percentage light interception and soil moisture were determined in late July at peak flowering. Percentage light interception of the understory canopy was determined at approximately solar noon using a quantum meter with six evenly spaced sensors on a 50-cm wand (Apogee Instruments Inc., Logan, Utah, U.S.A.) at 2 m above the soil surface (above all understory vegetation but below the tree canopy), and also at the soil surface. Soil moisture (%) was measured using a water content sensor (Hydrosense Water content measurement system; Campbell Scientific, Thuringowa Central, Queensland, Australia) at a depth of 10 cm. For both light and soil moisture, a single measurement was taken in the middle of each subplot and the average of these 5 measurements was used in analysis.

Nutrient supply rates were estimated using ion exchange membranes (Plant Root Simulator (PRS)^{TM} probes; Western Ag Innovations Inc., Saskatoon, Saskatchewan, Canada) using separate cation- and anion-exchange resin membranes. A single probe each for cations and anions was placed in each subplot and subplot probes were pooled during analysis to account for soil heterogeneity. The PRS probes were inserted into the soil in mid-June and left in place until the end of the growing season (mid-August) to measure *in situ* nutrient supply rates. Probes were analyzed by Western Ag Innovations Inc. for NO_3^+, NH_4^-, Total N, P, K, S, Ca, Mg, Mn, Fe, Cu, Zn, B, Al, and Pb.

DATA ANALYSIS

Data were analyzed as a two-way factorial split-plot ANOVA, with fertilization and fencing as the main effects, and treatment continued (whether the treatment had been stopped in 2000) as the split-plot effect. When there was a significant interaction between treatments, analyses were run separately for different treatment levels. Soil nutrients were log transformed as required to conform to assumptions of normality. Final phenological stage reached before senescence was assessed by assigning phenological stages consecutive numbers from 1 to 7; these values were analyzed independently for each species. For timing of the different phenological stages, analyses were done on the Julian date for each phenological stage separately for each species. Because not all individuals reached all stages, analyses were only conducted on stages where a minimum of 3 replicates reached that phenological stage. Three of the species (*A. uva-ursi*, *L. borealis*, *and L. arcticus*) disappeared from fertilized plots early in the experiment (Turkington et al., 2002) and for these species data were analyzed as a split-plot ANOVA, with fencing as the only main factor, and treatment continued as the split-plot effect. Analyses were conducted using JMP statistical software (2003, SAS Institute, Cary, North Carolina, U.S.A.).

Results

PHENOLOGICAL ASSESSMENT

The final phenological stage reached before senescence was affected by fertilization, with the presence and direction of the effect depending on species (Table 1, Fig. 1). Fencing, in contrast, had no effect on the final phenological stage reached nor was there an interaction between fertilization and fencing treatment. Responses to neither fertilizer nor fencing were affected by the length of time the plots had been treated (i.e., no interaction between Treatments Continued and either Fencing or Fertilizer; data not shown).

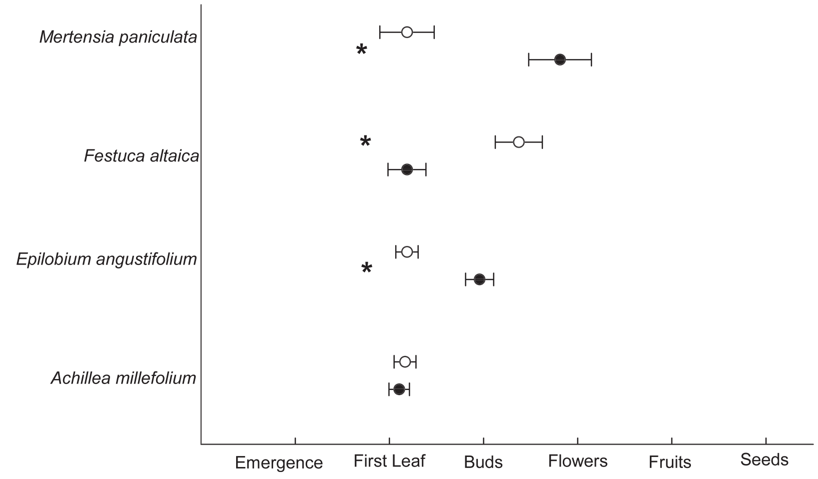

FIGURE 1. Mean (±SE) final phenological stage reached before senescence for four boreal forest understory species in different fertilizer treatments. * indicates a significant ($p < 0.05$) difference between fertilizer treatments within a species. Filled symbols represent fertilized plots, and open symbols represent unfertilized plots.

There were few effects of either fertilization or fencing on the timing of any of the 7 phenological stages for any of the species (Table 2, Fig. 2b). *Epilobium angustifolium* was the only species for which fertilization affected the timing of phenological events (Table 2) and it advanced the timing of the early phenological stages, first leaf expansion (4.5 days earlier), budding (7.2 days earlier), and had marginal effects on emergence (3.5 days earlier) (Table 2, Fig. 2). Fencing, again, had no effect on the timing of any phenological stage, nor did fertilization or fencing treatments interact for any of the species (Table 2). Finally, the effect of fertilization or fencing was also not affected by the length of time the plots had been treated (i.e., no interaction between Treatments Continued and either Fencing or Fertilizer; data not shown).

For the three species that were absent from the fertilized plots, only the effects of fencing were examined. There were few effects of fencing (Table 1). Leaf expansion for *A. uva-ursi* occurred earlier in fenced plots, and this only occurred in plots where the fencing treatments had been maintained (Fence × Treatments Continued: $F_{1,29} = 5.15$, $P = 0.031$; Fig 3).

ECOSYSTEM PROPERTIES

Maximum daily temperatures at a nearby weather station (Environment Canada, Haines Junction, Yukon Territory, weather station) ranged between 16 °C and 26 °C during the study period (Fig. 2a); 22 March was the first day of 2006 where the average temperature was above 0 °C (Environment Canada, 2010), although snowmelt did not occur until early May. Light interception was not significantly affected by either fertilizer or fencing treatments (Table 3). Soil moisture showed a significant interaction between fertilizer and fencing treatments (Table 3), because fertilized plots had lower soil moisture only when plots were also fenced (Fig. 4). There was a significant interaction between fertilizer and continuation of treatments for Total N (Table 3, Fig. 4), P (Table 3, Fig. 4), NO_3^- ($F_{1,12} = 153.51$, $p < 0.001$), and Mn ($F_{1,12} = 70.02$, $p < 0.001$) because there was a larger difference between fertilized and unfertilized plots when treatments were continued, although both treated and untreated plots showed a significant effect of fertilizer. There was also a significant interaction between fertilizer and continuation of treatments for K (Table 3, Fig. 4), Fe ($F_{1,12} = 68.06$, $p < 0.001$), Zn ($F_{1,12} = 22.29$, $p < 0.001$), and Al ($F_{1,12} = 25.52$, $p < 0.001$), but for these nutrients there was only a significant effect of fertilizer in treated plots. There were no significant treatment effects on NH_4^+, Ca, Mg, B, or S.

Discussion

This study explored the influences of fertilization and herbivory on the timing of phenological stages of 7 plant species in the

TABLE 2

Summary of 2-way split-plot ANOVA testing the effects of fertilizer, fencing, and continuation of treatments on timing of phenological stages in 4 boreal forest understory species. Only effects of fertilizer and fencing are presented. Values are presented only for those stages with adequate replication. Bold values are significant at $p < 0.05$.

	Achillea millefolium							*Epilobium angustifolium*						
		Fertilizer		Fence		Fertilizer × Fence			Fertilizer		Fence		Fertilizer × Fence	
	d.f.	*F*	*P*	*F*	*P*	*F*	*P*	d.f.	*F*	*P*	*F*	*P*	*F*	*P*
Emergence	1,12	0.31	0.588	0.44	0.521	0.62	0.446	1,12	4.31	0.060	<0.001	0.996	4.61	0.053
Full Leaf	1,12	0.44	0.520	0.19	0.673	1.35	0.267	1,12	4.76	**0.050**	0.18	0.680	0.25	0.628
Buds								1,9	5.15	**0.049**	0.02	0.902	3.79	0.083
	Festuca altaica							*Mertensia paniculata*						
		Fertilizer		Fence		Fertilizer × Fence			Fertilizer		Fence		Fertilizer × Fence	
	d.f.	*F*	*P*	*F*	*P*	*F*	*P*	d.f.	*F*	*P*	*F*	*P*	*F*	*P*
Emergence	1,12	1.55	0.236	0.01	0.940	1.55	0.236	1,10	0.12	0.738	0.10	0.754	0.40	0.540
Full Leaf	1,12	0.52	0.483	0.37	0.553	1.93	0.190	1,10	0.17	0.689	0.05	0.820	0.09	0.775
Buds														

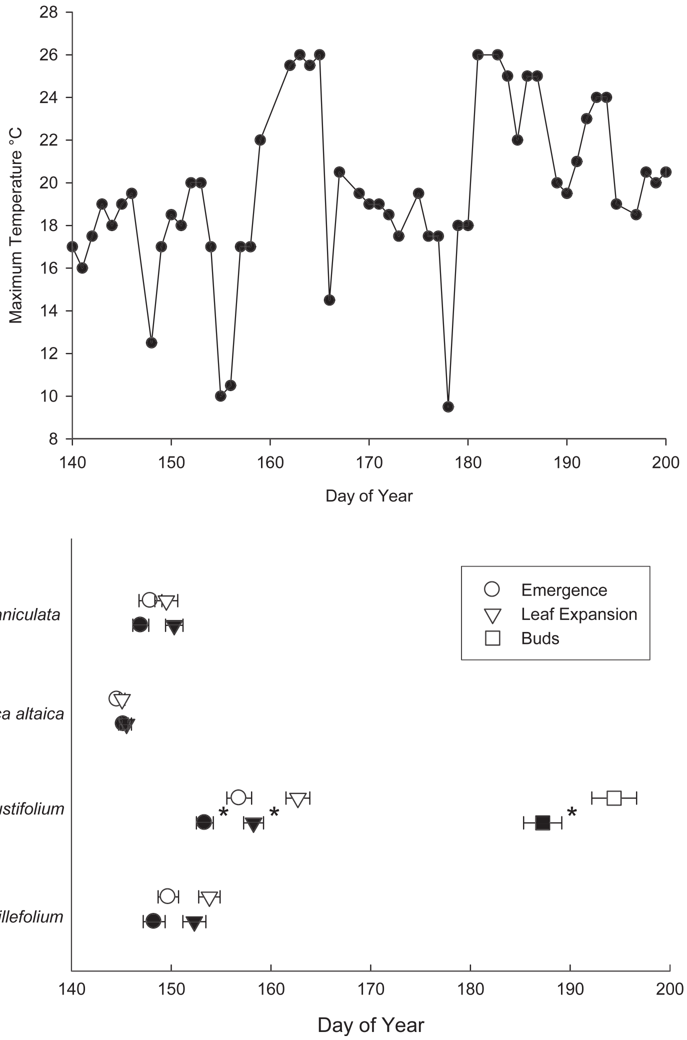

FIGURE 2. (a) Maximum daily (day 140 is 20 May 2006) temperature at a nearby Environment Canada weather station (Haines Junction). (b) Mean day of year (±SE) of major phenological stages for four boreal forest understory species in different fertilizer treatments. * indicates a significant ($p < 0.05$) difference between fertilizer treatments within both stage and species. Filled symbols represent fertilized plots, and open symbols represent unfertilized plots.

understory of the boreal forest. In an earlier study, fertilization significantly affected the cover of these 7 species after 10 years of fertilization in these plots; four species increased (*A. millefolium*, *E. angustifolium*, *F. altaica*, and *M. paniculata*) and three decreased (*L. borealis*, *L. arcticus*, and *A. uva-ursi*) (Turkington et al., 2002). After 17 years of fertilization, however, we found that fertilizer had little to no effect on the phenological timing, nor the final phenological stage reached, of these species. In sites with closed vegetation cover dominated by clonally proliferating species, such as this boreal forest, competition for light, nutrients, and/or water tends to be severe (Wookey et al., 1993). As a result, plants may strategically invest greater resources in vegetative growth rather than reproductive growth, because the opportunity for successful germination or seedling development tends to decrease as competition increases (Wookey et al., 1993; Arft et al., 1999). All 7 species we examined are clonal, and we hypothesize that the increased abundance observed by Turkington et al. (2002), is likely due to effects of fertilizer on vegetative, rather than sexual reproduction.

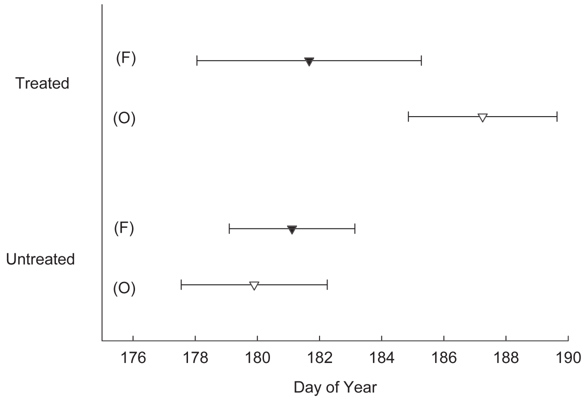

FIGURE 3. Timing (±SE) of leaf expansion for *Arctostaphylos uva-ursi* in fenced and open plots for plots where the treatments had been maintained for the duration of the experiment (Treated) and plots where the treatments were ceased after 10 years (Untreated). Filled symbols represent fenced plots and open symbols represent open plots (Day 176 is 25 June 2006).

Other studies have investigated the effects of fertilization on plant phenology (Larigauderie and Kummerow, 1991; Wookey et al., 1993; Erhardt et al., 2005; Cleland et al., 2006), but none have imposed fertilization treatments for this extended length of time. In our study, *E. angustifolium* was the only species in which the phenological timing was affected by fertilizer, with early phenological stages (emergence, first full leaf, and budding) occurring approximately 3–7 days earlier in fertilized plots. In contrast, Erhardt et al. (2005) found no direct effect of fertilizer on *E. angustifolium*, although they found that only with fertilization did elevated CO_2 marginally advance flowering date in a growth chamber experiment.

TABLE 3

Summary of 2-way split-plot ANOVA testing the effects of fertilizer, fencing, and continuation of treatments on light interception, soil moisture and soil total nitrogen (N), phosphorus (P), and potassium (K). Bold values are significant at $p < 0.05$.

		Light		Soil moisture		Total N		P		K	
Source	df	F	P	F	P	F	P	F	P	F	P
Fertilizer	1,12	2.24	0.160	1.82	0.202	**232.86**	**<0.001**	**50.12**	**<0.001**	**26.10**	**<0.001**
Fence	1,12	1.42	0.267	0.17	0.686	0.18	0.68	0.38	0.549	0.26	0.62
Fertilizer × Fence	1,12	0.01	0.916	**5.16**	**0.042**	2.94	0.112	0.46	0.512	2.00	0.183
Treated	1,12	0.67	0.429	0.57	0.464	**221.49**	**<0.001**	**17.26**	**0.001**	**30.83**	**<0.001**
Fertilizer × Treated	1,12	4.01	0.058	0.00	0.923	**193.21**	**<0.001**	**15.16**	**0.002**	**37.15**	**<0.001**
Fence × Treated	1,12	2.32	0.154	0.29	0.602	0.49	0.496	0.01	0.937	0.06	0.812
Fertilizer × Fence × Treated	1,12	0.44	0.521	0.14	0.712	0.36	0.557	0.12	0.733	1.64	0.224

We did find, however, that both *E. angustifolium* and *M. paniculata* were more likely, and *F. altaica* less likely, to reach flowering with higher nutrient levels. All three of these species increased in abundance with fertilization (Turkington et al., 2002). We propose that the increase in *E. angustifolium* and *M. paniculata* abundance could be partially due to an increased potential to produce seed, resulting from a greater likelihood of reaching flowering. Conversely, *F. altaica* may have fewer resources going to reproductive growth (resulting in less flowering) and more to vegetative growth in fertilized plots. This warrants further examination as we cannot examine these mechanisms directly with the data collected in this study. A study in temperate grasslands also showed that fertilizer, in combination with elevated CO_2, significantly accelerated flowering in forbs but delayed flowering in grasses, which decreased phenological complementarity and potentially increased competition between these two functional groups (Cleland et al., 2006). The balance between sexual reproduction and clonal growth has been predicted to shift towards more sexual reproduction as temperature increases (Aerts et al., 2006), given that many studies have observed high arctic plants producing greater seed counts when subjected to higher temperatures (Arft et al., 1999). However, it is difficult to predict how the later phenological stages, such as fruiting or seeding, will respond to higher temperatures, because sexual reproduction also requires greater nutrient investments than clonal growth (Aerts and Chapin, 2000) and temperature increases may lead to greater soil nutrient availability (Jonasson et al., 1993; Aerts et al., 2006).

The three species that declined in abundance with fertilization (*Linnaea borealis*, *Lupinus arcticus*, and *Arctostaphylos uva-ursi*) in the earlier experiment (Turkington et al., 2002) were nearly completely eliminated from fertilized plots by 2006, and thus we could only examine the effects of fencing, and not fertilizer, on these species. Fencing had little effect on the phenology of any of the 7 species we examined, except for a delay in leaf expansion in *A. uva-ursi* without fences. In our boreal system, Turkington et al. (2002) detected few effects of fencing on species diversity and abundance, and concluded that snowshoe hares, the dominant herbivore in the system, have only marginal impacts on plant cover even during peak hare densities. Herbivory may have little impact on the understory plants for a number of reasons; first, snowshoe hares may rely more on woody shrubs than other understory species (Turkington et al., 2002); second, much of the understory vegetation may be unavailable due to chemical defenses (Seccombe-Hett, 1999); and third, snowshoe hare populations are regulated both by predators and winter food

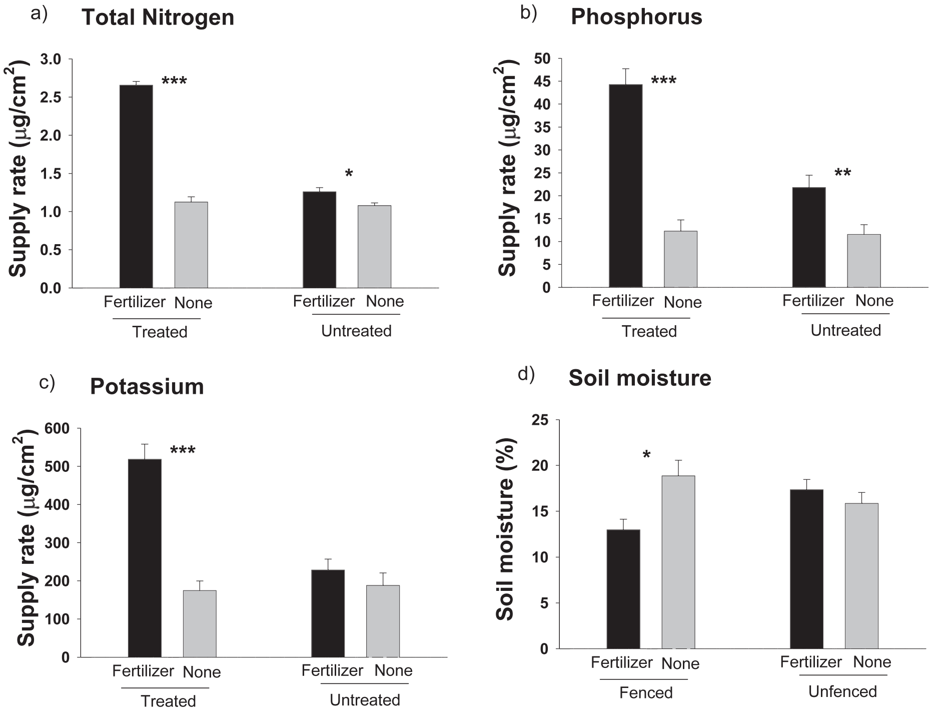

FIGURE 4. Mean nutrient supply rate (+SE) for (a) total nitrogen, (b) phosphorus, and (c) potassium. Treated plots received fertilizer, or not, annually for 16 years; untreated plots received fertilizer, or not, annually for 10 years, and then the fertilizer treatment was discontinued for years 11–16. Data are averaged across fencing treatments. (d) Soil moisture in fertilized and unfertilized plots, separated by fencing treatments. Data are averaged across treated and untreated plots. * $p < 0.05$, ** $p < 0.01$, and *** $p < 0.001$.

shortages so their population numbers are rarely high enough to have an impact on the understory plants (Krebs et al., 1995).

Effects of fencing on the phenology of *A. uva-ursi* disappeared when fences were removed after 10 years of treatment, whereas the effects of fertilizer on *E. angustifolium*, *M. paniculata*, and *F. altaica* were still detected 7 years after fertilizer treatments had stopped. This may be a legacy effect of fertilizer, but not fencing, on soil properties; although there was no difference between formerly fertilized and never fertilized plots for some nutrients (including K), a number of soil nutrients (including N and P) in these plots still showed effects of fertilization 6 years after the treatments were stopped. Alternatively, as these plants are perennial, the plants themselves may still have legacy effects of fertilizer in their storage organs, enabling them to respond to these treatments after nutrient levels have declined in the soils. Although soil moisture was affected by a combination of fertilizer and fencing, these effects did not correlate to any described phenological effects.

Environmental factors other than nutrient availability and herbivory may be determining phenological timing. A number of studies have investigated how biotic factors, such as pollinators and pre-dispersal seed predators (Elzinga et al., 2007), and abiotic factors, such as temperature, elevated CO_2, light availability, snow cover period, and precipitation, influence phenological timing (Cleland et al., 2006; Erhardt et al., 2005; Wookey et al., 1993). Many of these factors, however, have only significantly affected phenology when in combination. For example, Erhardt et al. (2005) stated that the response of plants to elevated CO_2 may depend significantly on nutrient availability.

In conclusion, changes in species abundance with fertilizer in this system (Turkington et al., 2002) have likely not been caused by changes in the phenology of these species. Rather, these changes in abundance are more likely due to changes in clonal growth caused by fertilization. Alternatively, our conclusions may be based on the timing of this experiment as 17 years of fertilization may not have been long enough to affect phenology in such highly clonal species; many ecological processes occur slowly and initial measurements seldom reflect more permanent long-term responses (Turkington et al., 2002). Predicted environmental changes in northern ecosystems, however, include more than just changes in soil conditions (Erhardt et al., 2005; Cleland et al., 2006), and further studies are required to understand how soil nutrients or herbivory may interact with other environmental changes to influence phenology in this northern ecosystem.

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