

HEAT AVOIDANCE LIFE HISTORY STRATEGY CONTROLS THE DISTRIBUTION OF GEOTHERMAL *AGROSTIS* IN YELLOWSTONE

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Abstract. Two *Agrostis* taxa occur in geothermal habitats of Yellowstone National Park: *Agrostis rossiae* is reported to be endemic to the thermal areas and *A. scabra* occurs both in thermal and nonthermal habitats. Thermal populations of both taxa are always surrounded by a nonthermal population of *A. scabra* that is reproductively isolated from the thermal plants by its later flowering time (summer rather than spring). Since *Agrostis* species are well known for their ability to form edaphic ecotypes, we used common garden and greenhouse experiments to test the hypothesis that “soil type” (a complex variable including factors such as soil chemistry and soil biotic community) was responsible for the separation of the thermal and nonthermal populations. In addition, field monitoring and growth chamber experiments were used to determine whether soil temperature affected the local distribution of these taxa. The results indicate that the thermal and nonthermal taxa are affected similarly by both soil temperature and soil type. The separate distribution of the thermal and nonthermal taxa is not due to the greater tolerance of one of these taxa for a particular factor, but instead to the interaction between seasonal fluctuations in soil temperature and the life history of the thermal and nonthermal plants. Nonthermal *Agrostis* plants, which are perennial, are excluded from the thermal habitats by lethal summer soil temperatures that limit the duration of the growing season, effectively selecting for precocious flowering and an annual habit. The thermal plants, in contrast, do not grow in the nonthermal matrix in prevailing cool field temperatures, but do grow in these habitats under artificially elevated temperatures. The thermal taxa may have reduced competitive ability at cooler temperatures. Thermal *Agrostis* have adopted a “stress avoidance” strategy, unlike previously studied thermal plants in Yellowstone, e.g., *Dicanthelium lanuginosum*, which are stress tolerant.

Key words: *Agrostis*; bentgrass; geothermal; Gramineae; heat tolerance; life history; Poaceae; soil temperature; stress avoidance; stress tolerance; thermal; Yellowstone.

INTRODUCTION

Stressful habitats often support plants that have sacrificed competitive ability or fecundity in favor of the ability to tolerate a narrow range of extreme conditions (Grime 1977), but a single habitat may pose different adaptive challenges for different species (Stanton et al. 2000, Wilson and Lee 2000). Serpentine endemics, for example, have developed diverse ways of coping with the soils on which they grow, such as preferentially excluding chromium from roots or accumulating calcium (Kruckeberg 1985). Moreover, different species may evolve alternative adaptations to the same stressor. Desert plants, for example, have evolved a variety of drought-tolerance mechanisms, including CAM photosynthesis, succulent stems, or ephemeral life histories (Ehleringer 1985).

The geothermally influenced habitats of Yellowstone National Park, Wyoming, USA contain a unique assemblage of plant species that differs markedly from the surrounding nonthermal vegetation, including a

number of species that occur only in thermal areas (Despain 1990, Whipple 2001). Elevated soil temperatures are the most apparent environmental factor distinguishing the thermal habitats from their nonthermal counterparts, and a number of authors have concluded that heat stress is the most important factor controlling the spatial distribution and zonation of plants in and around geothermally influenced habitats of North America (Sheppard 1971, Pavlik and Enberg 2001, Stout and Al-Niemi 2002), New Zealand (Given 1980, Burns 1997) and Japan (Glime and Iwatsuki 1990, 1997). But thermal habitats also contain unique soils that consist primarily of hydrothermally altered rhyolite that is permeated by steam, H₂S, and other gasses and they are sometimes very acidic (White et al. 1971, Fournier 1989). It is therefore possible that some plant species in thermal habitats may have been more influenced by edaphic factors other than soil temperature during their evolution (e.g., soil chemistry or biotic community), or that, as has been documented for desert species, different species may have evolved alternative adaptations to heat stress.

Agrostis taxa in geothermally influenced habitats of Yellowstone National Park provide a unique opportunity to separate soil temperature from other edaphic

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factors that may be influencing plant distribution. *Agrostis* species are well known for their ability to form ecotypes in response to localized variations in soil chemistry (Jowett 1964, Jain and Bradshaw 1966, Archambault and Winterhalder 1995), and therefore seem likely to have evolved specialized adaptations to differences between thermal and nonthermal soils. Two thermal *Agrostis* species are currently recognized: *Agrostis rossiae* Vasey is endemic to Yellowstone thermal areas (Swallen 1948), while *Agrostis scabra* Willdenow is thought to occur both in the thermal areas and in a variety of nonthermal habitats (Hitchcock 1950, Hitchcock and Cronquist 1973). In 1999, one of the present authors (M. Tercek) noticed that every thermal *Agrostis* population (whether *A. rossiae* or *A. scabra*) is surrounded by a nonthermal population of *A. scabra* that is reproductively isolated from the thermal plants due to its later flowering time. Seeds of thermal *Agrostis* populations germinate in December through January, when all nonthermal habitats are covered by snow; and the thermal plants (all annuals) are killed by rising soil temperatures in mid-June. Nonthermal *Agrostis scabra* populations, in contrast, do not initiate new growth from their perennial root stocks until late June, when the snow has completely melted; and they flower in mid-July to mid-August. All three taxa (*Agrostis rossiae*, thermal *A. scabra*, and nonthermal *A. scabra*) maintain distinctive morphologies when grown under uniform conditions (Tercek et al. 2003). In populations growing near a localized heat source, the nonthermal population is distributed concentrically around the thermal population.

This arrangement of the thermal and nonthermal *Agrostis* populations initially suggested that the thermal *Agrostis* populations might be independently evolved edaphic ecotypes of nonthermal *A. scabra*, but Tercek et al. (2003), using RAPD genetic markers, found that nonthermal *A. scabra* plants are no more closely related to the thermal populations than a number of other nonthermal species that occur in Wyoming. Thermal *A. scabra* was found to be most closely related to the endemic *A. rossiae*, and all the Yellowstone thermal populations were most closely related to other thermal *Agrostis* populations located in Lassen Volcanic National Park, California, and the Valley of the Geysers, Kamchatka, Russian Federation. These findings demonstrate the existence of a previously unrecognized, thermally adapted taxon that is reproductively isolated from nonthermal *A. scabra*, which always occurs near it.

The goal of the present study was to determine whether the concentric zonation of *Agrostis* in and around the thermal areas is indeed controlled by soil temperature, as has been found for other thermal taxa, or whether some other aspect of the soils (e.g., soil chemistry, biotic interactions) is instead maintaining distinct thermal and nonthermal populations. To test the hypothesis that soil temperature is the important

factor, data loggers were used to measure temperature at 2 cm root depth for a period of 3 yr, and the results were used to design a series of laboratory growth experiments. This approach allowed us to measure both the heat tolerance of the study taxa as well as the interaction between their life histories and the soil temperature regime found in the field. Previously studied thermal plants, e.g., *Dicanthelium lanuginosum*, have adapted to their habitat primarily through heat tolerance (Stout et al. 1997, Redman et al. 2002), but the annual life history of the thermal *Agrostis* suggested that they may have evolved a stress-avoidance strategy similar to that found in desert ephemerals. The nontemperature edaphic factors were dealt with simultaneously under the heading of "soil type" and their hypothesized control of plant zonation was tested in a series of common-garden and greenhouse experiments.

By comparing thermal and nonthermal congeners, we were able to determine the ecological significance of our results, rather than merely correlating the maximum growth of the thermal taxa with particular soil types or temperature regimes. Adaptations that are shared by both thermal and nonthermal taxa clearly do not control the zonation of plants in the thermal habitats.

METHODS

Habitat description

Thermal *Agrostis* populations range in size from a solitary plant to ~100 m in diameter (~10 000 plants). In Yellowstone, they usually occur in "vapor-dominated" thermal areas, which are partially isolated from the water table of nearby hot springs either by their slightly higher elevation or by nonporous rock layers (White et al. 1971). Vapor-dominated thermal soils are easily distinguished from the surrounding nonthermal soils by their brown-white, bleached color and are composed mainly of a thermally degraded, siliceous residue containing pockets of bleached clays. They are permeated by a number of gasses, including steam and H₂S, which, along with soil-borne Fe₂S, is converted to sulfuric acid by endemic bacteria, significantly lowering the soil pH (White et al. 1971, Fliermans and Brock 1972, Mosser et al. 1973, Fournier 1989). Unlike some vapor-dominated thermal areas, however, the habitats in which thermal *Agrostis* occur usually contain a layer of the moss *Racomitrium canescens* (Shepard 1971; M. Tercek, *personal observation*). They also support several thermophilic soil fungi (Redman et al. 1999), a heat-tolerant grass, *Dichanthelium lanuginosum*, which depends on a mycorrhizal partner for its survival on thermal soils (Stout et al. 1997, Redman et al. 2002), and a small number of annual forbs.

Field monitoring of soil temperature and moisture

For the purposes of this study, the "thermal" habitat is defined as the area occupied by either of the endemic

thermal taxa *Agrostis rossiae* (AR) or thermal *A. scabra* (TAS). *Agrostis* populations in most thermal habitats exhibit complex patterns of spatial distribution. However, a number of habitats were identified that exhibit an “ideal” zonation of plant taxa, in which a thermal *Agrostis* population (either AR or TAS) surrounds a localized heat source, e.g., a single fumarole (steam vent), and is itself concentrically surrounded by a nonthermal *A. scabra* (NAS) population. Soil temperature data loggers were placed in the hottest thermal location (nearest the steam vent, referred to as “hot”), the coolest thermal location (outer edge of thermal population, referred to as “cool”) and at the coolest edge of the nonthermal NAS zone in each study site. All temperature readings were taken among the roots of a living plant at 2 cm standard depth. Measurements were collected hourly during the period February 2000 to February 2001, and again from May 2001 to August 2002. At least three populations were monitored during each season, however, the choice of populations monitored during each season varied (data not presented).

Soil moisture was measured twice each week during May through August of 2000, 2001, and 2002 with gypsum block sensors placed next to each temperature data logger. The plants next to each sensor were closely observed, and their dates of flowering, seed set, and death were recorded.

Growth experiments in laboratory and greenhouse

The field monitoring data were used to design temperature experiments. All the other temperature-independent edaphic factors were investigated simultaneously in common-garden and greenhouse experiments that used “soil types” as fixed factors. Seed samples used in every experiment were collected from across the entire geographic range of each taxon (eight populations each of AR and TAS, 14 populations of NAS) and mixed before they were assigned to experimental treatments. Soil samples were assigned randomly to treatments instead of being mixed, and each soil treatment included replicates from at least three different populations. The results of all experiments were tested with SPSS 8.0 (SPSS, Chicago, Illinois, USA) standard or factorial ANOVA. In order to correct for their heteroscedascity, all germination rates were arcsine transformed prior to ANOVA (Snedecor 1956); however, since this did not change the significance of any of the tests, the untransformed data are presented below.

Effect of soil type on seed germination.—To test whether differences between thermal and nonthermal soil types have an effect on germination, soil was collected to 10 cm depth from AR, TAS, and NAS populations and immediately shipped to the laboratory in ventilated containers. Transport time varied from 2 to 6 d and soil remained moist during transit. The top 2.5 cm of soil was removed from the samples, since a preliminary experiment had shown that all native seeds

are thus eliminated, and the remaining soil was spread into 10 × 30 cm germination trays to 7 cm depth. Twenty seeds of AR, TAS, or NAS were sown into each tray in a full-factorial soil × seed design. Twelve replicate trays were placed in a chamber maintained at a constant 10°C and exposed to 15 h light:9 h dark by fluorescent full-spectrum lamps. The other replicates ($n = 30$ for NAS and TAS, $n = 32$ for AR) were grown in a greenhouse with fluorescent 15 h light:9 h dark cycles and an average temperature of 20.26°C (1 SD = 5.9). The percentage of seeds germinated after one month was recorded. Approximately 98% of the seedlings survived to flowering in every treatment.

Effect of temperature on seed germination.—To test whether temperature had an effect on germination success, Petri dishes (9.5 cm diameter) containing moistened filter paper were sown with 20 seeds of AR ($n = 37$ sets of 20 seeds), TAS ($n = 34$ sets), or NAS ($n = 35$ sets) and heated in a water bath for 2 wk. Treatment temperatures were 5, 10, 30, 35, 40, and 45°C, and an unregulated “room temperature” (15–25°C) control. The percentage of seeds germinated was recorded after the 2-wk heat treatment and again after the treated seeds had been maintained for an additional 2 wk at unregulated room temperature and 15 h light:9 h dark under full-spectrum lamps. Water bath treatments did not vary more than $\pm 2^\circ\text{C}$. No new germination was discovered when seeds were counted again 8 wk after heat treatment.

Effect of soil temperature on plant growth.—Plastic pots containing a 6 cm depth of potting soil were sown with AR, TAS, and NAS seeds spaced 1.5 cm apart in rows 6 cm apart and placed in growth chambers in which the soil temperature had been adjusted to 10, 30, or 40°C. In addition, a set of unregulated control replicates was maintained at room (air) temperature, which averaged 21.4°C (1 SD = 2.8) during the experiment. Sample sizes are reported in *Results*. Temperatures did not deviate more than $\pm 2^\circ\text{C}$ in the regulated treatments. All treatments were exposed to 15 h light:9 h dark with fluorescent full-spectrum lamps. Adult plants were collected 50 d after germination and fresh root and shoot maximum lengths were measured.

pH tolerance.—Since vapor-dominated thermal soils are acidic (Fournier 1989), the effect of pH on growth was investigated. Thirty percent Hoagland’s solution with a pH of 2.0, 2.5, 3.0, 4.0, 5.0, or 6.0 (adjusted with NaOH or H₂SO₄) was allowed to diffuse through holes in the bottom of plastic tubes containing 10 cm depth of sand that had been sown with AR, TAS, and NAS seeds spaced 1.5 cm apart. The lengths of fresh roots and shoots were measured 50 d after germination. Sample sizes are reported in *Results*.

The pH of the soil was measured in the “cool thermal,” “hot thermal,” and NAS habitats of four AR populations and four TAS populations in the field. For each sample, ~10 g of soil was removed, immediately mixed 1:1 with distilled H₂O, and tested in the field

with a portable pH meter. The pH of duplicate samples collected from the same collection sites was determined after two months of air drying, using the same methods and a laboratory pH meter.

Competition experiment.—To determine whether temperature affects the success of thermal *Agrostis* growing within an intact, nonthermal, plant community, eight 1.5 × 0.5 m blocks of undisturbed soil were field collected to 0.5 m depth in the NAS zone of four thermal habitats. This was the maximum soil collection allowed under Park Service regulations. The blocks were collected in January, before any seeds or perennial plants had initiated growth, and flown intact to the laboratory in ventilated containers that remained moist. Fifty seeds of AR were sown directly onto the vegetated surface of each block. Half of the blocks were maintained at a constant 10°C, while the other half were placed in a greenhouse that had an average temperature (measured hourly) of 20.26°C (1 SD = 5.9). Both treatments were exposed to 15 h light:9 h dark under fluorescent full-spectrum lamps. Native seeds and perennial plants in the soil blocks were allowed to grow, thus providing the competition for the experimentally introduced seeds. Germination and survival rates were recorded after 4 wk and again after 12 wk.

Common-garden experiments

These experiments were designed to separate the effects of soil type and temperature regime on the germination and growth of AR and NAS under field conditions. Since it had been shown in a preliminary experiment that the top 2.5 cm of soil contains all of the native seeds (M. Tercek, *unpublished data*), this layer was removed from all of the 60 × 60 cm experimental plots prior to the planting of experimental seeds. Two experiments were conducted.

The first experiment began in late January, when AR seeds were germinating but adjacent nonthermal areas were still covered with snow. Forty experimental plots were established in the AR and NAS zones of five populations (eight plots in each population, four in each planting zone). Native soil was left in half of the plots, and half of the plots were “swapped” to the opposing planting zone as intact, 30 cm deep, soil blocks (AR soil blocks moved to NAS zone and vice-versa). Fifty seeds of either AR or NAS were sown in each plot in a full factorial design: seed × soil type (swapped or unswapped) × planting zone. The number of plants present in each plot was recorded on 15 May, and the phenology of the plants was followed until they senesced (15 June through 1 July).

Since no seeds of either AR or NAS germinated in the nonthermal habitat during the first experiment, a second experiment designed to test the effect of field soils under a warmer temperature regime was initiated 15 May. Six 60 × 60 cm plots were prepared in the NAS zones of three populations as described above, and 50 seeds of either AR or NAS were sown in each.

No soil blocks were swapped in this experiment; all contained native soil. Percent germination and plant phenology were recorded until 1 July.

RESULTS

Field monitoring of soil temperature and moisture

Soil temperature was strongly influenced by both daily and seasonal fluctuations in air temperature. Temperatures in any one location varied by as much as 60°C during a year, with the lowest average temperatures (calculated for each hour of the year) occurring in December through January and the highest in July through August. Temperature readings taken from the same habitat (i.e., hottest, coolest, or nonthermal) in different populations were treated as replicates, and all of the data were time synchronized and averaged over the three-year collection period. For example, the temperature readings taken at 0000 hours on 15 May 2000 in all of the “hot” habitats (locations nearest the heat source) in every study population were averaged with the corresponding “hot” readings taken at 0000 hours on 15 May 2001 and 0000 hours on 15 May 2002. The same was done with the “cool” (thermal locations furthest from heat source) and nonthermal temperature readings. This procedure produced three sawtooth-shaped “average temperature” time series that retained the diurnal fluctuations characteristic of the original data (Fig. 1). The single coolest and hottest temperatures measured in each 24-h period during the three years of monitoring were also plotted as separate graph traces. In order to calculate the length of the potential growing season, Fig. 1 has been annotated with the results of the laboratory growth experiments.

During the summer months, thermal soil temperatures were greatly affected by moisture. Soils in the thermal habitats remained moist throughout the year, but soils in the NAS zone desiccated quickly when the snow melted and rainfall ceased in May and June (Fig. 2). This drop in nonthermal soil moisture was consistently accompanied by a sharp increase in thermal soil temperatures. Thermal *Agrostis* plants were always killed when soil temperatures reached 40–45°C (Fig. 2). Plants in “hot thermal” habitats were always killed earlier in the season than plants in the “cool thermal” habitats, and in some cases the return of rainfall delayed plant death in the “cool thermal” habitats by as much as one month. These results suggest that two different water sources are affecting thermal soil temperature. Steam keeps the “hot habitats” moist all year, but plant life in these habitats relies on the regulating effect of rainfall and nonthermal zone soil water.

Effect of soil type on seed germination

The germination success of all three study taxa was affected similarly by soil type (Fig. 3A, Table 1; soil × taxon interaction not significant). The germination ratio of AR:NAS:TAS was approximately 1.30:1.36:1

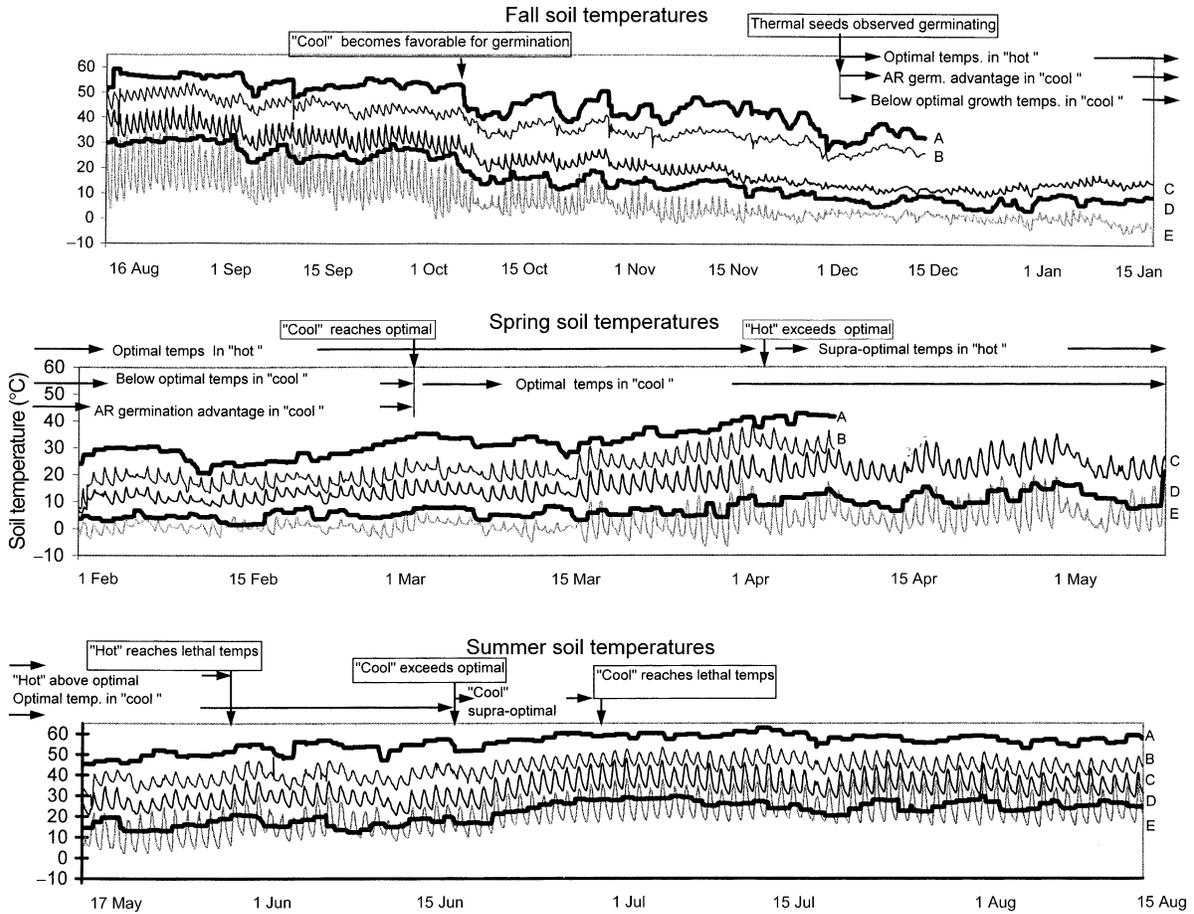


FIG. 1. Soil temperatures at 2 cm root depth in *Agrostis* populations of Yellowstone (three years of data are averaged into a single annual cycle): (A) maximum in all thermal habitats for each 24-h period, (B) 3-yr average of all “hot thermal” habitats (calculated hourly), (C) average of “cool thermal” habitats, (D) minimum in all thermal habitats for each 24-h period, and (E) 3-yr average of nonthermal zone.

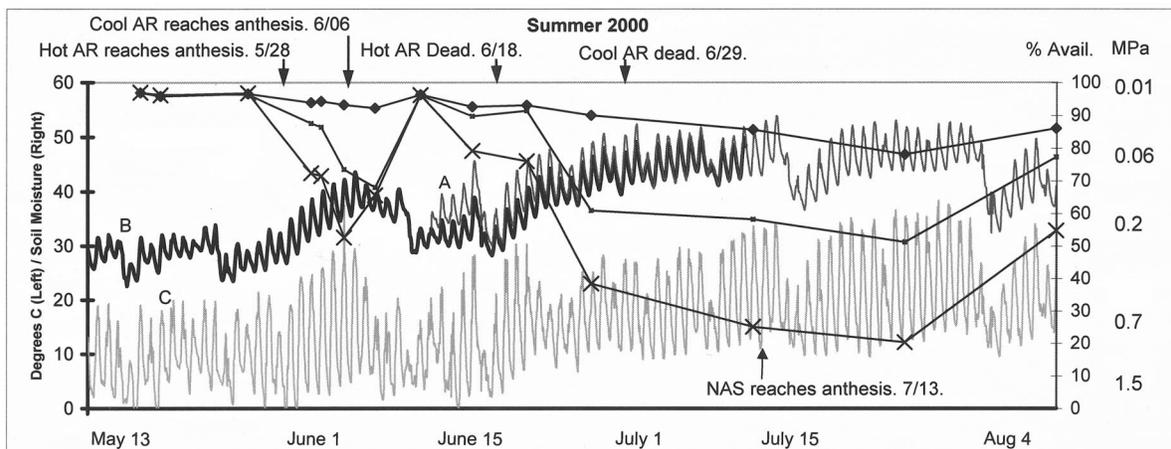


FIG. 2. The relationship among soil temperature, soil moisture, and survival of thermal *Agrostis* populations in Yellowstone National Park. This pattern was observed 12 times during the 3-yr study in both *Agrostis rossiae* and thermal *Agrostis scabra* populations. Symbols for soil moisture (right axis, straight lines): diamonds = “hot thermal” habitat; squares = “cool thermal” habitat; × = nonthermal habitat. Temperature readings (left axis, sawtooth lines) are not average values but are taken directly from data-loggers at single locations. Trace A (thin line) is the “hot thermal” soil temperature, trace B (heavy black line) is the “cool thermal” location, and trace C (gray line) is the nonthermal *Agrostis scabra* soil temperature.

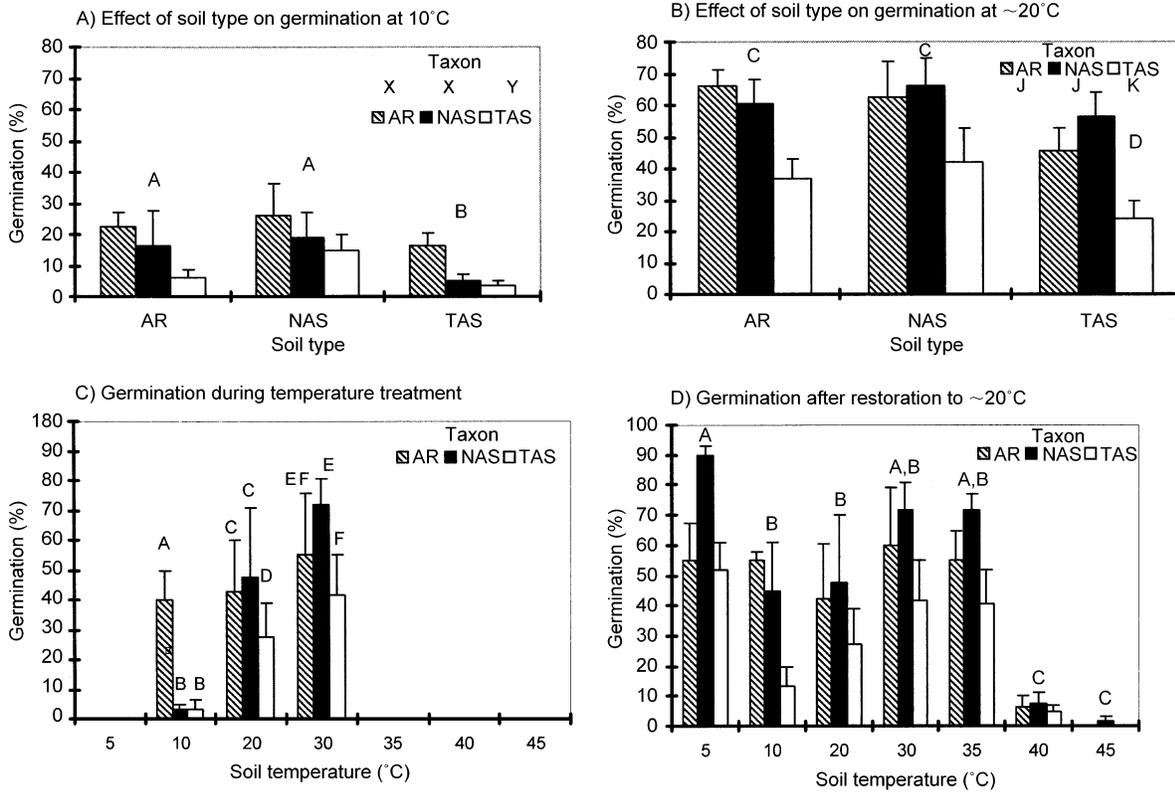


FIG. 3. Germination of *A. rossiae* (AR), thermal *A. scabra* (TAS), and nonthermal *A. scabra* (NAS). (A) Germination at 10°C in all three soils. Different letters over the bars (A, B) indicate significant differences among three-taxon soil-type means; different letters under key (X, Y) indicate significant differences among three-soil-type taxon means ($P < 0.05$). (B) Germination in the three soils at 20°C. Different letters over the bars (C, D) indicate significant differences among three-taxon soil-type means; different letters under key (J, K) indicate significant differences among three-soil-type taxon means ($P < 0.05$). (C) Germination during two-week heat treatments. Different letters over the bars (A–F) indicate significant differences among taxa within treatments ($P < 0.05$). (D) Germination two weeks after restoration to 20°C. Different letters over bars (A–C) indicate differences among treatment means ($P < 0.05$). Error bars show 1 SE.

in every soil at 20°C and 2.59:1.60:1.00 in every soil at 10°C, which suggested that AR had a germination advantage over the other taxa at 10°C. Germination rates were lower at 10°C for all taxa, and there were significant differences among taxa in all soils (Fig. 3A). Tukey's post-hoc tests showed that TAS had lower germination rates than the other two taxa in every soil and that TAS soil produced significantly lower germination rates for all taxa (Fig. 3A and B, Table 1).

TABLE 1. Comparison of germination rate of *A. rossiae*, thermal *A. scabra*, and nonthermal *A. scabra* at 10°C and 20°C in all three soils.

Factor	df	F	P
Soil	2	7.28	<0.001
Taxon	2	3.22	0.044
Temperature	1	72.29	<0.001
Soil × taxon	4	0.17	0.952
Soil × temperature	2	1.58	0.210
Taxon × temperature	2	0.65	0.524
Soil × taxon × temperature	4	0.12	0.976
Error	110		

Effect of temperature on seed germination

The germination of the seeds in the Petri dishes during the temperature treatments was significantly affected by both temperature and taxon (Fig. 3C, Table 2). AR had a significant (12:1:1) germination advantage over the other taxa at 10°C (simple contrasts, Fig. 3C),

TABLE 2. Comparison of germination rate of *A. rossiae*, thermal *A. scabra*, and nonthermal *A. scabra* during temperature treatment and after restoration to 20°C.

Factor	df	F	P
Germination during temperature treatment			
Taxon	2	4.16	0.019
Temperature	6	39.38	<0.001
Taxon × temperature	12	2.31	0.013
Error	85		
Germination after restoration to 20°C			
Taxon	2	11.57	<0.001
Temperature	6	36.29	<0.001
Taxon × temperature	12	1.52	0.132
Error	850		

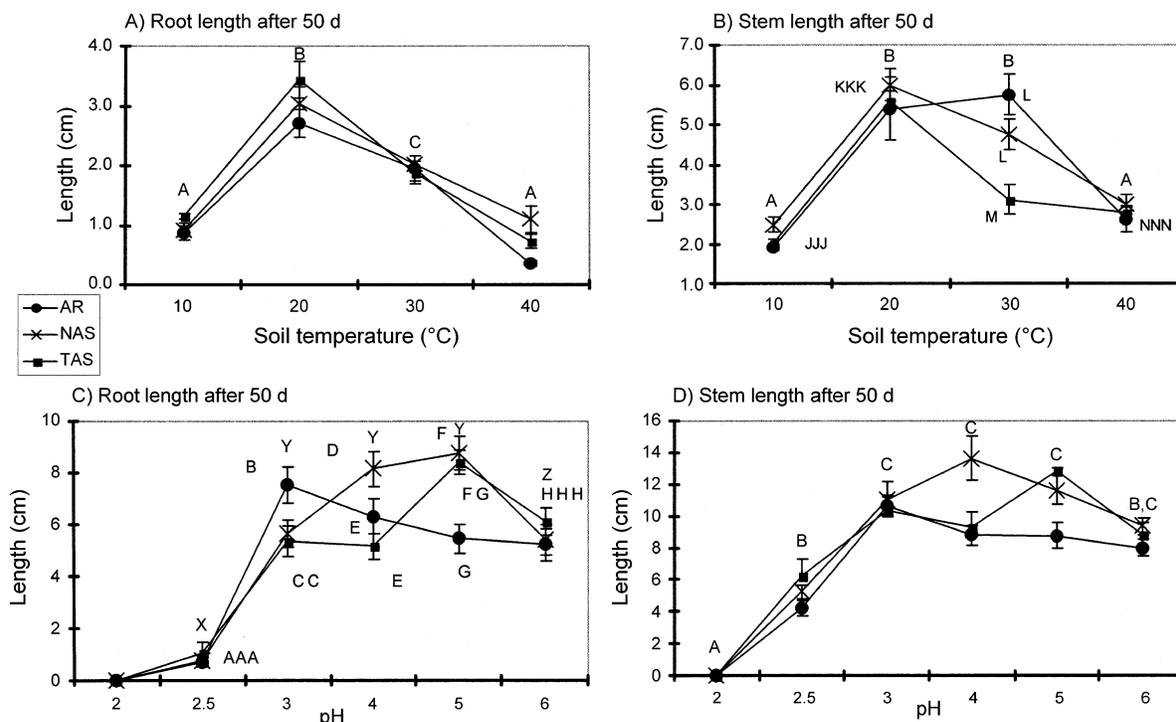


FIG. 4. Growth responses of *A. rossiae* (AR), thermal *A. scabra* (TAS), and nonthermal *A. scabra* (NAS). (A) Root response to temperature. Different letters over data points indicate significant differences among three-taxa means at each temperature ($P < 0.05$). Sample sizes for AT, NAS, and TAS, respectively, were 9, 10, and 5 at 10°C; 12, 18, and 9 at 20°C; 19, 20, and 16 at 30°C; and 5, 8, and 16 at 40°C. (B) Stem response to temperature. Different letters (A and B) indicate significant differences among three-taxa means at each temperature; different letters (J–N) indicate significant differences among taxa within temperatures ($P < 0.05$). (C) Root response to soil pH. Different letters (A–H) indicate significant differences among taxa within pH ($P < 0.05$); different letters (X–Z) indicate significantly different three-taxa means. Sample sizes for AR, NAS, and TAS, respectively, were 5, 8, and 3 at pH 2.5; 16, 18, and 13 at pH 3.0; 14, 14, and 17 at pH 4.0; 5, 11, and 5 at pH 5.0; and 6, 11, and 8 at pH 6.0. (D) Stem response to pH. Different letters indicate significant differences among three-taxa means at each pH ($P < 0.05$). Error bars show 1 SE. At 10°C, $n = 9$ for AR, 10 for NAS, and 5 for TAS. At 20°C, $n = 12$ for AR, 18 for NAS, and 9 for TAS. At 30°C, $n = 19$ for AR, 20 for NAS, and 16 for TAS. At 40°C, $n = 5$ for AR, 8 for NAS, and 16 for TAS.

which confirms the cold-temperature advantage of AR observed in the soil experiment (above). TAS germination rates were lower than AR/NAS at 20°C (contrasts, Fig. 3C, $P = 0.047$) and lower than NAS at 30°C ($P = 0.036$).

Once the seeds had been removed from the water baths and restored to 20°C, there were no significant

differences among taxa within temperature treatments (Fig. 3D, Table 2, taxon \times temperature interaction not significant). Seeds that previously had been exposed to 40°C and 45°C had significantly reduced germination (Tukey's tests, Fig. 3D). AR seeds from the 10°C treatment lost their germination advantage when returned to room temperature, and the 5 and 35°C treatments were able to germinate normally, despite their inhibition during the temperature treatments (Fig. 3C and D).

Effect of temperature on plant growth

Temperature had a significant effect on root length after 50 d, but there were no differences among taxa (Fig. 4A, Table 3). Tukey's tests showed that roots were significantly longer at 20°C than at all other temperatures (Fig. 4A). The 10°C and 40°C root lengths did not differ significantly, even though the plants in the two treatments looked strikingly different. The 10°C plants were green and healthy, while the 40°C plants had shriveled, discolored leaves by the end of the experiment. Stems of all taxa were significantly longer

TABLE 3. Growth responses of *A. rossiae*, thermal *A. scabra*, and nonthermal *A. scabra* to temperature regime.

Factor	df	F	P
Root length after 50 d			
Taxon	2	2.06	0.131
Temperature	3	55.90	<0.001
Taxon \times temperature	6	0.98	0.440
Error	135		
Stem length after 50 d			
Taxon	2	2.09	0.127
Temperature	3	30.16	<0.001
Taxon \times temperature	6	2.86	0.012
Error	135		

TABLE 4. Growth responses of *A. rossiae*, thermal *A. scabra*, and nonthermal *A. scabra* to different pH.

Factor	df	F	P
Root length after 50 d			
pH	5	19.27	<0.001
Taxon	2	0.49	0.617
pH × Taxon	10	2.80	0.004
Error	136		
Stem length after 50 d			
pH	5	11.98	<0.001
Taxon	2	1.72	0.184
pH × Taxon	10	1.35	0.213
Error	136		

at 20–30°C than at 10°C or 40°C. TAS stems were significantly shorter than AR/NAS at 30°C (Fig. 4B, Table 3). There were no other differences among taxa. Interestingly, AR and TAS in the 30°C treatment were the only plants to flower by the end of the 50-d experiment.

pH tolerance

The pH at which maximum growth occurred differed among taxa (Fig. 4C). AR had its maximum root length at pH 3, where it was significantly greater than TAS/NAS, and declined at higher pH levels (Fig. 4C, Table 4). TAS and NAS had maximum root lengths at pH 5. Stem length showed the same pattern; however there were no significant differences among taxa within pH treatments (pH × taxon interaction not significant; Fig. 4D, Table 4). Tukey’s tests showed that mean stem lengths calculated for all taxa did not differ at pH 3–6 (Fig. 4D).

The range of soil pH measured in thermal soils overlapped greatly with corresponding values from nonthermal soils. Soil pH was 3.9–5.6 in the AR habitats (mean ± 1 SD = 4.7 ± 0.5), 4.4–5.6 in TAS habitats (5.1 ± 0.63), and 4.3–6.4 in the NAS habitats (5.5 ± 0.65). The pH in “cool” thermal habitats was not consistently higher or lower than that in “hot” habitats. Measurements made in the field with a portable pH meter differed from duplicate measurements made in the laboratory by less than 0.3 pH units.

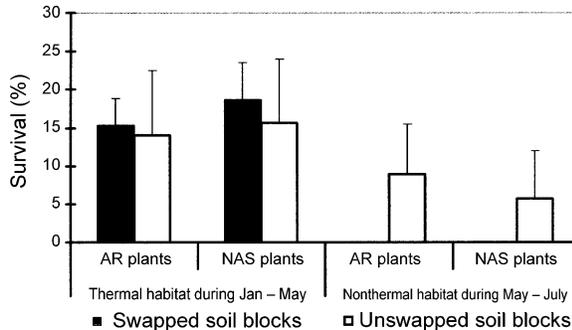


FIG. 5. Survival of *A. rossiae* (AR) and nonthermal *A. scabra* (NAS) in field common-garden experiments. In thermal habitats, seeds were planted in January, and survival was measured in May. In nonthermal habitats, seeds were planted in May, and survival was measured in July. In the thermal common gardens (January–May experiment), soil in half of the plots was replaced (“swapped”) with intact soil blocks from adjacent nonthermal habitat.

Common-garden experiments

During the first common-garden experiment, in which seeds were planted in field populations during January, survival was not affected by either seed type (AR or TAS) or by soil-block swapping in the thermal habitats (Fig. 5, Table 5). However none of plants, neither AR nor NAS, survived in the nonthermal habitat during this period. Interestingly, all of the plants present in the thermal experimental plots on 15 May (whether AR or NAS) survived until soil temperatures reached 40–45°C on 15 June through 1 July (Figs. 1 and 2). At this time, the AR plants had already set seed, while the NAS plants, though healthy, had not.

During the second experiment, in which AR and NAS seeds were planted in nonthermal plots during May, AR and NAS survived at the same rate (Fig. 5, Table 5). Soil moisture in the NAS zone dropped quickly in early June (Fig. 2), making it necessary to water the experimental seedlings daily. By 1 July, when the experimental plants were killed by rising soil temperatures, all AR plants had flowered, while the NAS seedlings had not. Survival rate in the NAS zone was not significantly less than in the AR zone (Fig. 5).

TABLE 5. Survival of *A. rossiae*, thermal *A. scabra*, and nonthermal *A. scabra* in field common-garden experiments.

Factor	Both experiments			First experiment (Jan–May)		
	df	F	P	df	F	P
Swapping	1	0.36	0.557	1	0.32	0.578
Taxon	1	0.01	0.920	1	0.40	0.534
Taxon × swapping	1	0.06	0.808	1	0.06	0.818
Habitat type†	1	2.78	0.110			
Habitat type × taxon	1	0.30	0.588			
Error	21			17		

Note: See Fig. 5 legend for details.

† Thermal vs. nonthermal.

Competition experiment

No AR plants grew in the NAS zone soil blocks that were maintained at 10°C, despite the AR germination advantage in the absence of competition at this temperature during the soil- and seed-heating experiments (Figs. 3 and 4). In the ~20°C treatment, the four soil blocks had 65, 71, 3, and 2 AR plants, respectively, out of 50 seeds sown. The fact that more than 50 seeds germinated in some of the blocks indicates that an AR seed bank was present in at least some of the non-thermal soil blocks, even though AR did not occur normally in the area from which the soil blocks were collected. All AR plants in the ~20°C soil blocks flowered within 4 wk. Many nonthermal species grew successfully in both temperature treatments.

Effect of temperature on time to flowering

During the growth experiments, a relationship between soil temperature and the time from germination until flowering became apparent. AR and TAS, which are annuals, flowered after ~60 d at 10°C and in only 36 d at 30°C. NAS took ~160 d to reach flowering at its optimum growing temperature (Fig. 3A and B) of 20–30°C. NAS plants at 10°C had greatly reduced growth, confirming the findings presented in Fig. 3A and B, and did not appear to be near anthesis after 180 d.

DISCUSSION

Why is nonthermal *A. scabra* excluded from the thermal habitat?

The thermal habitats have lethal summer soil temperatures that prevent the growth of perennial roots (Fig. 1), and the thermal growing season is not long enough to allow the slow-growing NAS to produce seeds within one year. Precocious flowering, therefore, appears to be the most important difference between the thermal taxa and NAS. AR and TAS, which are annuals, require 30–70 d to reach flowering, depending on the temperature to which they are exposed, while NAS requires ~160 d to reach flowering at 20–30°C (the temperature range producing maximum growth), and it does not reach anthesis after 180 d at 10°C (data not shown). Comparing the results of our growth experiments with the temperature regime observed in the field (Fig. 1) reveals that the growing season in the “hot thermal” habitat is only ~120 d (1 December to 1 April)—not sufficient for flowering of NAS. After 1 April, temperatures in the “hot” habitats are consistently near sublethal 40°C; the plants typically have discolored leaves, produce no new growth (M. Tercek, *personal observation*), and are finally killed when temperatures reach ~45°C near the beginning of June. Similarly, the growing season in the “cool” thermal habitats is approximately 1 December, when earliest germination has been observed (M. Tercek, *personal observation*; J. Whipple, *personal communication*), until

1 July or ~210 d, with favorable growth temperatures (Fig. 4A and B; 20–40°C) occurring for only ~105 d (Fig. 1; 1 March to 15 June). To grow in this zone, NAS plants must pass through a long period of sub-optimal ~10°C soil temperatures during December to March (Fig. 1). In the laboratory experiments, NAS plants exposed to 10°C had significantly reduced growth (did not differ in length from plants in the sublethal 40°C treatments after 50 d; Fig. 4A and B) and did not appear to be near anthesis after 180 d. Furthermore, air temperatures in the thermal areas are consistently 10–30°C less than soil temperatures in the thermal habitats (data not shown), which could further slow the growth rate of NAS.

This interpretation is supported by the results of the first common-garden experiment, in which AR and NAS seeds planted in the thermal areas in January survived at the same rate (Fig. 5). Both AR and NAS appeared healthy on 15 May and both were killed when soil temperature reached 40–45°C (15 June to 1 July) in every study population. The only difference between the thermal and nonthermal taxa in this experiment was AR’s ability to set seed before soil temperatures reached lethal levels. Despite their ability to grow in the thermal common gardens, NAS seedlings seldom occur in the thermal habitats at distances >1–2 m from the main body of the nonthermal population. This is likely due to the fact that the study taxa have very poor dispersal abilities. Tercek et al. (2003) found almost no gene flow between thermal populations that are sometimes only meters apart, and they also found evidence for poor dispersability in several nonthermal *Agrostis* species.

Poor dispersability would seem to expose the seeds of AR and TAS to lethal summer soil temperatures, and thus heat would exclude both the thermal and nonthermal taxa from the thermal habitats. However, some seeds may remain in the glumes until later in the season, when cooler temperatures prevail, and seeds that dehiscence earlier often land on the moss *Racomitrium canescens*, which blankets most of the thermal *Agrostis* habitats. This moss seems to shield the dormant seeds from the temperature regime experienced by plants that have actively growing roots (M. Tercek, *personal observation*).

Why are *Agrostis rossiae* and thermal *Agrostis scabra* excluded from nonthermal habitat?

The combined results of the competition, common-garden, and soil experiments suggest that competition from the nonthermal plants, exacerbated by cooler temperatures, is responsible for the restricted thermal distribution of AR and TAS. In the absence of competition (when the top 2.5 cm of soil had been removed), AR had a germination advantage at 10°C in both thermal and nonthermal soil (Figs. 3 and 4); and the thermal taxa had survival rates equivalent to NAS in nonthermal soils under warmer conditions (second common-

garden experiment; Fig. 5). Indeed, the growth of AR in nonthermal soil was prevented only once: in the competition experiment, under cool (10°C) conditions, in the presence of competition from intact, nonthermal perennials. Nonthermal competition did not inhibit growth of AR under warmer (20°C) conditions.

Admittedly, competition may not be the only factor affecting AR/TAS success in the nonthermal habitat at lower temperatures. Simon (1974), for example, found that seeds exposed to suboptimal temperatures are vulnerable to attack by fungi. Nevertheless, the fact that more seeds germinated than were planted in the 20°C treatments of the competition experiment suggests that a viable AR seed bank persists in NAS soil during the winter; and that suppression of the growth of annual thermal taxa occurs after germination.

Competitive interactions are mediated by one or more limiting resources (Chapin et al. 1987). If competition is indeed restricting the distribution of AR and TAS, water may be the limiting resource. Our field data indicate that nonthermal soils are quite dry by the time they reach the 20–30°C optimum for AR or TAS in June and July (Fig. 2). Under these conditions, the longer, perennial roots of the nonthermal community would be better adapted for obtaining water. In addition, by the time the optimal 20–30°C has been reached, the perennial plants have grown to an extent that would allow them to shade or crowd AR and TAS (M. Tercek, *personal observation*). In contrast, the competition experiment showed that nonthermal soil blocks that were immediately transferred from a dormant winter state to 20–30°C did not suppress AR, presumably because the slower-growing nonthermal community was not yet large enough to affect the growth of AR.

Study taxa respond similarly to heat stress and soil type

The thermal taxa do not differ from NAS in their response to either heat or soil type. Instead, the thermal habitats are distinguished by a short growing season that requires precocious flowering and an annual habit, while the nonthermal habitats are dominated by slow-growing, competitive perennials. The growth experiments support this conclusion. In the absence of competition, thermal and nonthermal taxa did not differ in their growth response to soil temperature, except in the high temperature 30°C treatment, where TAS actually had shorter stems than NAS (Fig. 4B). Similarly, all three study taxa were affected equally by soil type. Thermal and nonthermal taxa survived at the same rate in both soil types during the common-garden experiments (Fig. 5), and TAS soils induced the lowest germination for all three taxa, with TAS seeds having the lowest germination rate in every soil and temperature treatment (Fig. 3). These findings are somewhat unexpected, given the tendency of *Agrostis* taxa to form distinct races in response to soil chemistry (Jowett 1964, Archambault and Winterhalder 1995). None of

the study taxa are as heat resistant as *Dichanthelium lanuginosum*, which occurs in Yellowstone thermal soils with temperatures as high as 50°C (Redman et al. 2002), even though all three (AR, TAS, and NAS) are more heat resistant than *Agrostis palustris*, a cool season turf grass, which fares poorly after 50 d of exposure to 35°C:30°C day:night temperatures (Huang et al. 2001, Xu and Huang 2001).

Other factors

There may be many factors which affect each taxon differently and yet are nevertheless not responsible for the spatial separation of thermal and nonthermal *Agrostis*. The pH experiment, for example, demonstrated that AR had peak growth in more acidic soil, at pH = 3, than either TAS or NAS did, which both peaked at pH = 5 (Fig. 4C and D). However, these optima did not directly correspond to the ranges of soil pH in either the thermal (pH = 3.9–5.6) or nonthermal (pH = 4.3–6.4) habitats, which overlapped greatly. Similarly, Stout et al. (1997) found *Dichanthelium lanuginosum*, which occurs in the same habitats as AR and TAS, to be more acid tolerant than is perhaps required by field conditions, capable of growing in experimental cultures maintained at pH = 2.0, even though few plants occur in soils with pH < 3.5. Furthermore, it seems likely that there are other factors not considered in this study that affect plant growth in thermal areas. Redman et al. (2002), for example, have shown that *Dichanthelium lanuginosum* benefits from a mycorrhizal association at high (~50°C) soil temperatures, even though this species often occurs in cooler areas.

Broader significance

Like many earlier studies, it was shown that soil temperature was the most important factor controlling the zonation of plant taxa in geothermal habitats. However, the fortuitous occurrence of a closely related congener in the adjacent nonthermal habitat has revealed that, unlike species examined previously, thermal *Agrostis* are not unusually heat tolerant, nor do they appear to be specifically adapted to any other edaphic factors, e.g., soil chemistry, but merely reproduce more quickly and escape the heat. These findings suggest that the short growing season is the primary factor preventing more competitive species from colonizing the thermal areas, and they highlight the thermal community's vulnerability to invasion by weedy annuals. Yellowstone's thermal areas already have been colonized by a number of weeds that are usually found in disturbed habitats, including *Bromus tectorum* (Whipple 2001).

Implications for theories of life history evolution.—Many other studies have documented species that, like AR and TAS, use annual or ephemeral habits to avoid, rather than tolerate, stress; and this feature of plant life histories is one of the concepts underlying the CSR plant classification scheme of Grime (1977). This

scheme has been one of the most useful generalizations in community ecology; however, as originally formulated, it was intended to characterize particular habitats as well as plant life histories (Wilson and Lee 2000). Hodgson et al. (1999) explicitly continue this line of thinking with their development of quantitative CSR plant attributes, such as specific leaf area, flowering period, and canopy height that can be entered into spreadsheets and used to categorize habitats as, e.g., eutrophied or disturbed.

The results of the present study suggest that classifying habitats in this way may not be useful, since different species may respond to the same stressor in different ways. *Dichanthelium lanuginosum*, a perennial, has evolved elaborate heat tolerance adaptations to the thermal habitats (Redman et al. 2002); while AR and TAS have instead evolved heat avoidance mechanisms. Both the stress tolerators and stress avoiders are reacting to the same environmental factor (heat) in different ways. Similarly, Stanton et al. (2000) have argued that the CSR status of a particular species may be as much due to preexisting phylogenetic and physiological constraints as to the prevailing selection pressures, and that perennial species are generally more likely to evolve stress tolerance adaptations when they colonize a new habitat, whereas annuals tend to evolve toward stress avoidance. Indeed, the present study supports Stanton et al. (2000), but leaves unclear whether or not the thermal taxa have evolved an annual habit specifically as an adaptation to thermal habitats, or whether they are merely descended from an annual ancestor that opportunistically colonized thermal habitats.

The results of the present study indicate that the thermal habitats are vulnerable to colonization by ruderal exotics, but the thermal *Agrostis* themselves are not completely ruderal. They possess many of the traits that would be expected of ruderal species (annual habit, rapid growth, plastic response of flowering time in response to a stressor like heat), yet according to the genetic work of Tercek et al. (2003), they are poor dispersers. This trait makes adaptive sense, in view of the fact that thermal habitats are small and scattered, making a seed bank a better investment than dispersing seeds into habitats in which they have little chance of surviving. A similar trend was documented by Carlquist (1974), who showed that island species often lose the dispersal ability of their mainland ancestors. Some species in vernal pools of California are thought to have poor dispersal for the same reason (Baskin 1994). The predictable, seasonally favorable nature of the thermal habitats (Figs. 1 and 2) might also select for poor dispersal: the thermal taxa do not need to compensate for years of erratically harsh conditions by seeking new habitats. Similarly, Volis et al. (2002) found that desert ecotypes of barley (*Hordeum spontaneum*) have evolved better dispersal mechanisms than ecotypes growing in seasonal Mediterranean climates. Many of

these questions regarding the life history evolution of the thermal *Agrostis* could be elucidated with a comparative study of other *Agrostis* species. In particular, it would be useful to know the life history of the non-thermal progenitor of AR and TAS, which may occur near other thermally active areas in Alaska or the Aleutian Islands (Juday 1998, Tercek et al. 2003).

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