



## An intraspecific application of the leaf-height-seed ecology strategy scheme to forest herbs along a latitudinal gradient

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We applied the leaf-height-seed (LHS) ecology strategy scheme (a combination of three ecologically important traits: specific leaf area (SLA), seed mass and plant height) intraspecifically to two widespread European forest herbs along a latitudinal gradient. The aims of this study were to quantify LHS trait variation, disentangle the environmental factors affecting these traits and compare the within-species LHS trait relationships with latitude to previously established cross-species comparisons.

We measured LHS traits in 41 *Anemone nemorosa* and 44 *Milium effusum* populations along a 1900–2300 km latitudinal gradient from N France to N Sweden. We then applied multilevel models to identify the effects of regional (temperature, latitude) and local (soil fertility and acidity, overstorey canopy cover) environmental factors on LHS traits.

Both species displayed a significant 4% increase in plant height with every degree northward shift (almost a two-fold plant height difference between the southernmost and northernmost populations). Neither seed mass nor SLA showed a significant latitudinal cline. Temperature had a large effect on the three LHS traits of *Anemone*. Latitude, canopy cover and soil nutrients were related to the SLA and plant height of *Milium*. None of the investigated variables appeared to be related to the seed mass of *Milium*.

The variation in LHS traits indicates that the ecological strategy determined by the position of each population in this three-factor triangle is not constant along the latitudinal gradient. The significant increase in plant height suggests greater competitive abilities for both species in the northernmost populations. We also found that the studied environmental factors affected the LHS traits of the two species on various scales: spring-flowering *Anemone* was affected more by temperature, whereas early-summer flowering *Milium* was affected more by local and other latitude-related factors. Finally, previously reported cross-species correlations between LHS traits and latitude were generally unsupported by our within-species approach.

Functional life-history traits are increasingly used as the basis for a general ecological characterization and classification of a wide variety of organisms (Grime et al. 1988, Diaz et al. 2004, Freschet et al. 2010). Likewise, groupings of life-history traits that are consistent among different plant species have been widely used to predict

changes in the abundance and distribution of individual species and species groups in response to processes such as disturbance (Lavorel et al. 1997, Graae and Sunde 2000, Verheyen et al. 2003). The life-forms of Raunkiaer (1937) and the CSR-scheme of Grime (1974) have been applied and discussed for decades (see Craine 2005 for an

overview). More recently, Westoby (1998) proposed the leaf-height-seed (LHS) ecology strategy scheme as an alternative explanation for plant responses to environmental variation. The LHS scheme differs from other classifications in that it is based on three easily measured “soft” traits (sensu Diaz et al. 2004): 1) specific leaf area (SLA), 2) plant canopy height at maturity and 3) seed mass. SLA, for instance, contributes to the potential of a plant to respond to a change in stress exposure, whereas seed mass and plant height influence the ability of a plant to adjust to disturbance effects and other environmental changes (Westoby 1998, Poorter et al. 2009).

The LHS scheme has been used at the species level to compare species groups (Lavergne et al. 2003, Jardim and Batalha 2008) and at the community level to compare different forms of management or successional stages (Garnier et al. 2004, Moog et al. 2005, Golodets et al. 2009). To date, the LHS scheme has, to our knowledge, not been used to describe intraspecific trait variation among populations of a single species. Interspecific studies on how environmental factors influence a set of plant traits may be confounded by co-varying differences among taxa in, e.g. growth form, phenology, plant development time, predominant seed disperser assemblage or habitat type (Lavorel et al. 1997, Lord et al. 1997, Moles and Westoby 2003, Moles et al. 2007). For example, if seed mass or plant height across species increases towards the equator (Moles and Westoby 2003, Moles et al. 2007, 2009a, b), this pattern may be blurred by habitat variations, such as the decreasing proportion of trees towards the poles (Lord et al. 1997, Moles and Westoby 2003, Moles et al. 2007, 2009a, Tautenhahn et al. 2008). These drawbacks are largely avoided by using a within-species approach. Moreover, comparisons between the intraspecific and interspecific relationships for traits such as plant height are ecologically important issues that require further investigation (Moles et al. 2009a).

Quantitative knowledge about trait responses across various spatial scales is still largely lacking (Ordoñez et al. 2009). Such knowledge, however, is urgently needed to better understand ecosystem functioning (McGill et al. 2006) and predict responses to future climate change (De Frenne et al. 2009). The macroclimatic variation along latitudinal gradients offers an opportunity to elucidate variation in plant traits and to unravel the relative importance of a set of environmental factors (temperature, soil fertility, etc.; Austin 1999, McGill et al. 2006, De Frenne et al. 2009, 2010, Graae et al. 2009; see Fukami and Wardle 2005 for a recent review). Both observational studies and common-garden experiments along latitudinal transects at the inter- and intraspecific level show that seed mass and plant height generally decrease with latitude (Moles and Westoby 2003, Murray et al. 2004, Pakeman et al. 2008, Moles et al. 2009a, b), whereas SLA increases with latitude within species (Clevering et al. 2001, Etterson 2004, Miyazawa and Lechowicz 2004). Interspecific comparisons of SLA with latitude on a global scale are still lacking, and the three LHS traits have rarely been investigated intraspecifically from a combined perspective. Furthermore, little information is available about the scale on which the environmental factors affect the LHS traits,

such as soil fertility on a local scale and climate on a regional scale (already stressed by Westoby 1998).

In the present study, we investigated intraspecific variation in the LHS traits for multiple populations of two common European herbaceous forest plant species along a latitudinal gradient. In an effort to make our results more widely applicable, the study species belong to two different phenological groups (Kudo et al. 2008, De Frenne et al. 2009) and life-forms: *Anemone nemorosa* is a spring flowering geophyte, and *Milium effusum* is an early-summer flowering hemicryptophytic grass. We specifically wanted to gain insight into 1) the extent of variation in the three LHS traits within these species along a latitudinal gradient, 2) which environmental factors are most related to the observed variation in LHS traits and 3) whether the within-species relationships differ from previously established cross-species comparisons.

## Methods

### Study species

We selected two forest herbs for this study: *Anemone nemorosa* (hereafter referred to as *Anemone*; Ranunculaceae) and *Milium effusum* (referred to as *Milium*; Poaceae). Both species are widespread forest herbs within Europe (Hultén and Fries 1986). The northern edge of the distribution of *Anemone* in Sweden is situated around the city Umeå (63.8°N) whereas *Milium* is distributed as far as the North Cape (71.2°N; Hultén and Fries 1986). Close to their northern range edge, both species occur only in small deciduous forest patches within a largely boreal forest (*Anemone*) or sub-arctic tundra (*Milium*) matrix (De Frenne unpubl.). Although *Anemone* usually produces 10–30 achenes (hereafter referred to as “seeds”) per year per flowering ramet (referred to as “individual”) for sexual reproduction (De Frenne et al. 2010), vegetative propagation through rhizomes is common. *Milium* usually produces 100–300 caryopses (referred to as “seeds”) per individual per year and develops short stolons for vegetative resprouting. *Anemone* typically has very slow colonization rates whereas *Milium* is somewhat faster (Verheyen et al. 2003, Brunet 2007), probably due to epizoochorous dispersal by large mammals (Heinken and Raudnitschka 2002).

### Study regions and populations

We studied the species in seven (*Anemone*) and eight (*Milium*) regions located along an approximately 1900 km (*Anemone*) to 2300 km (*Milium*) latitudinal gradient from N France via Belgium and Germany to N Sweden. The latitudinal gradient exhibits a mean annual temperature difference of 6.5°C and 8.1°C between the southernmost (Amiens) and northernmost (Umeå and Abisko; for *Anemone* and *Milium*, respectively) populations (Table 1; FAO 2005). In 2008, we selected six populations of each species in each region (except five *Anemone* populations in Umeå and two *Milium* populations in Stockholm), thus a total of 41 *Anemone* and 44 *Milium* populations. Within each region, the populations were randomly selected within an area of ca 20 × 20 km<sup>2</sup>. All populations

Table 1. Location and climatic characterization of the study regions and collection dates (Julian days) along the latitudinal gradient (parentheses indicate the number of populations sampled in each region).

Region	City	Lat. (°N)	Long. (°E)	MAT <sup>1</sup> (°C)	MAP <sup>2</sup> (mm)	PET <sup>3</sup> (mm)	PET: MAP <sup>4</sup>	Collection dates	
								<i>Anemone</i>	<i>Milium</i>
France	Amiens	49.8	2.1	9.9	671	571	0.85	140 (6)	157 (6)
Belgium	Brakel	50.9	3.8	9.7	821	580	0.71	130 and 131 (6)	157 (6)
NE Germany	Potsdam	52.6	13.0	8.7	585	642	1.1	136 and 137 (6)	162–164 and 167 (6)
NW Germany	Bremen	53.3	9.2	8.3	761	557	0.73	142 (6)	165 (6)
S Sweden	Lund	55.6	13.3	7.9	732	545	0.74	141 and 143 (6)	170 and 171 (6)
C Sweden	Stockholm	59.0	17.5	6.6	535	606	1.13	157 (6)	198 (2)
C-N Sweden	Umeå	63.8	20.0	3.4	603	474	0.79	176 and 177 (5)	187 and 196–198 (6)
N Sweden	Abisko	68.4	18.8	1.8	305	318	1.03	– (0)	235, 239 and 242 (6)

<sup>1</sup>MAT: mean (1961–1990) annual temperature; <sup>2</sup>MAP: mean annual precipitation; <sup>3</sup>PET: potential evapotranspiration; <sup>4</sup>PET:MAP ratio (FAO 2005).

occurred in deciduous forests that were unaffected by major recent disturbances (management, grazing, etc.).

## LHS traits

At seed maturity, we measured plant height and collected all seeds and leaves of 15 randomly chosen individuals per population and species. Individuals that appeared heavily attacked by insects, pathogens or other organisms were avoided. Plants were considered mature when natural seed dispersal began, i.e. when seeds fell easily from the plant (Baskin and Baskin 1998). Collection dates are shown in Table 1. The following data collection protocol was adapted from Cornelissen et al. (2003). Plant canopy height at maturity was determined by measuring the total height from the ground level to the top for all 15 individuals per population. In *Anemone*, we calculated mean air-dry seed mass as the total seed mass per individual (weighed to the nearest 0.1 mg) divided by the total number of seeds of each individual, whereas for *Milium*, we determined mean seed mass from a randomly selected 50-seed subsample per individual. Leaves of each individual were air-dried flat in dry paper for 3 d and shipped to Belgium. Subsequently, leaves were oven-dried (50°C) to constant mass for 3 d. Leaf area (LA; mm<sup>2</sup>) was measured with a Li-Cor Portable Area Meter Li-3000 (Li-Cor Biosciences, NE, USA). Hence, LA was measured on dry leaves and not on water-saturated leaves (Garnier et al. 2001) due to logistic constraints along the large geographic gradient, shipping restrictions and to standardize the LA measurements in a single lab with one LA meter. We believe that the introduced “shrinkage” error by drying leaves of these particular species is negligible in an intraspecific relative comparison. Leaf dry matter (LDM; mg) was weighed to the nearest 0.1 mg. Specific leaf area (SLA; mm<sup>2</sup> mg<sup>−1</sup>) was calculated by dividing LA by LDM.

## Environmental variables

We used the number of growing degree hours (GDH) above 5°C from 1 January until the population- and species-specific seed collection dates as a measure of temperature (Lindsey and Newman 1956, De Frenne et al. 2009). The calculations of GDH followed those of Lindsey and Newman (1956) using daily minimum and maximum temperatures from weather stations as nearby as possible to the sampled populations (mostly < 40 km). We included the latitude of each region in the analysis to account for regional environmental variables not measured directly along the gradient but expected to differ among sampling regions, such as precipitation, photoperiod and semi-darkness (Mills 2008), and land-use and climate history shaping distributions of species and populations (Jansson and Dynesius 2002, Moles et al. 2007, De Frenne et al. 2009).

Within each population, we visually estimated the percentage of overstorey canopy cover as a measure of light availability. In addition, we collected five 4-cm deep soil cores from below the litter layer. The soil samples from each population were combined, oven-dried (40°C) to constant mass and passed through a 2 mm-sieve. All samples were analyzed for pH (determined from a solution of 10 g of

soil and 25 ml of 0.01 M  $\text{CaCl}_2$  with a standard glass electrode), plant available phosphorus (P), calcium (Ca), magnesium (Mg), potassium (K) (extraction with ammonium lactate and photometric determination by flow injection analysis for P and flame atomic absorption spectrophotometry, SpectrAA-220, Varian, for cations; all in mg per 100 g soil), carbon (C) and nitrogen (N) (elemental analyzer; %).

## Data analysis

We used population-level means for all analyses. Moreover, data were not transformed as assumptions of statistical tests in terms of normality and homogeneity of variances were fulfilled. To account for multicollinearity among soil variables, we performed a principal component analysis (PCA) with VARIMAX-rotation to reduce the set of soil variables to two components for both species. The first two PCA axes explained 67.4 and 71.2% of the variation in soil variables for *Anemone* and *Milium*, respectively. For both species, the first PCA axis was a soil nutrient axis (Pearson correlations between axis 1 and soil N, Mg, C and K concentration:  $r > 0.550$ ;  $p < 0.001$ )

whereas the second PCA axis was a soil acidity axis (Pearson correlations between axis 2 and soil pH and Ca concentration:  $r > 0.859$ ;  $p < 0.001$ ). Sample scores for the first two principal components were used in the multilevel models (see below).

To explore whether and to what extent the three LHS traits varied along the latitudinal gradient, we calculated the variation in LHS trait values using latitude as a predictor variable in multilevel models (SPSS MIXED procedure with maximum likelihood estimation). A random effect term “region” was added to the multilevel models to account for the hierarchical nature of the data and the spatial autocorrelation between populations from the same region (Hox 2002, De Frenne et al. 2009). Secondly, the regional (temperature and latitude) and local (soil nutrients axis, soil acidity axis and canopy cover) predictor variables were related to the variation in LHS trait values using multilevel models. Again, the multilevel model acknowledges that regional environmental variables only account for regional level variance, omitting pseudo-replication of the predictors at the population level. To detect possible multicollinearity between the predictor variables, variance inflation factors (VIF) were calculated according to Quinn and Keough (2002). The calculated

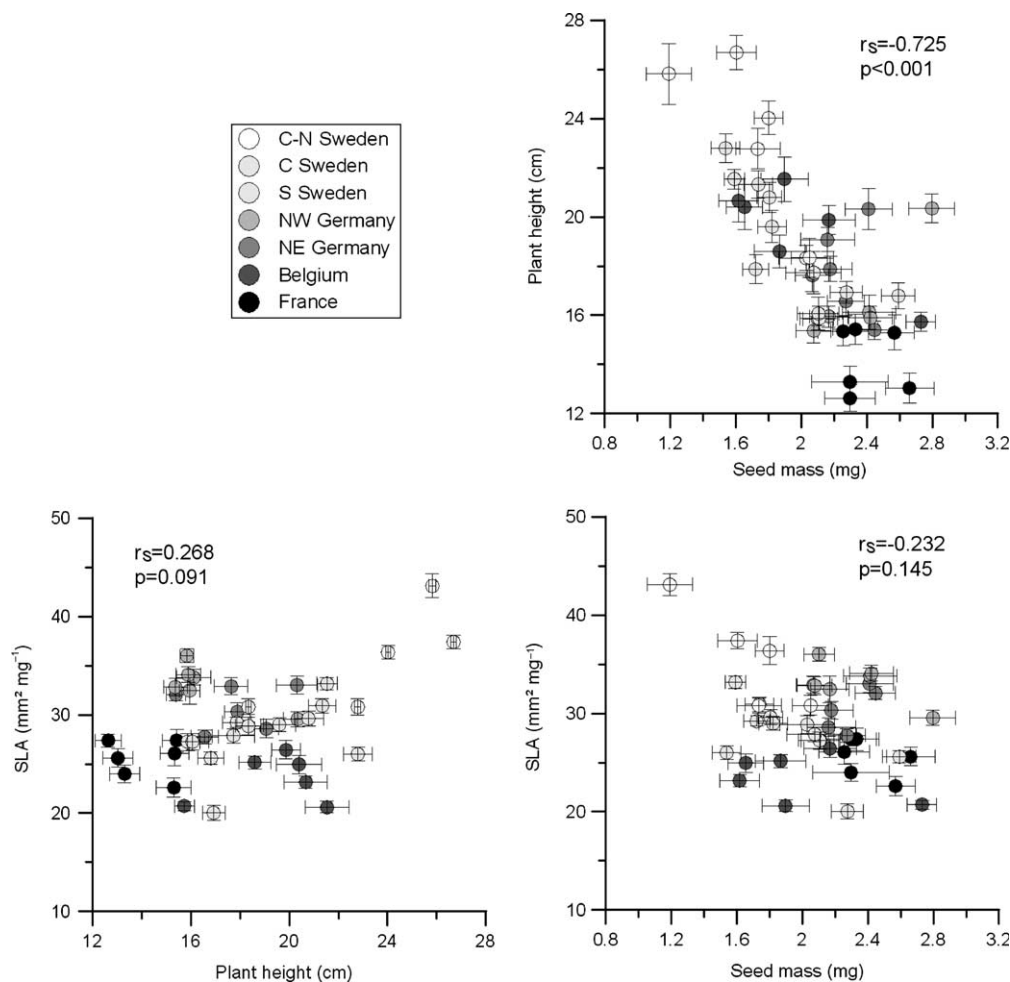


Figure 1. Leaf-height-seed relationships for *Anemone nemorosa* populations along the latitudinal gradient. Every dot represents a mean per population  $\pm$  SE with the grey scaling indicating southerly (black) to northerly (white) regions. Spearman rank correlations between LHS traits ( $r_s$ ) and p-value are also represented ( $n = 41$ ).

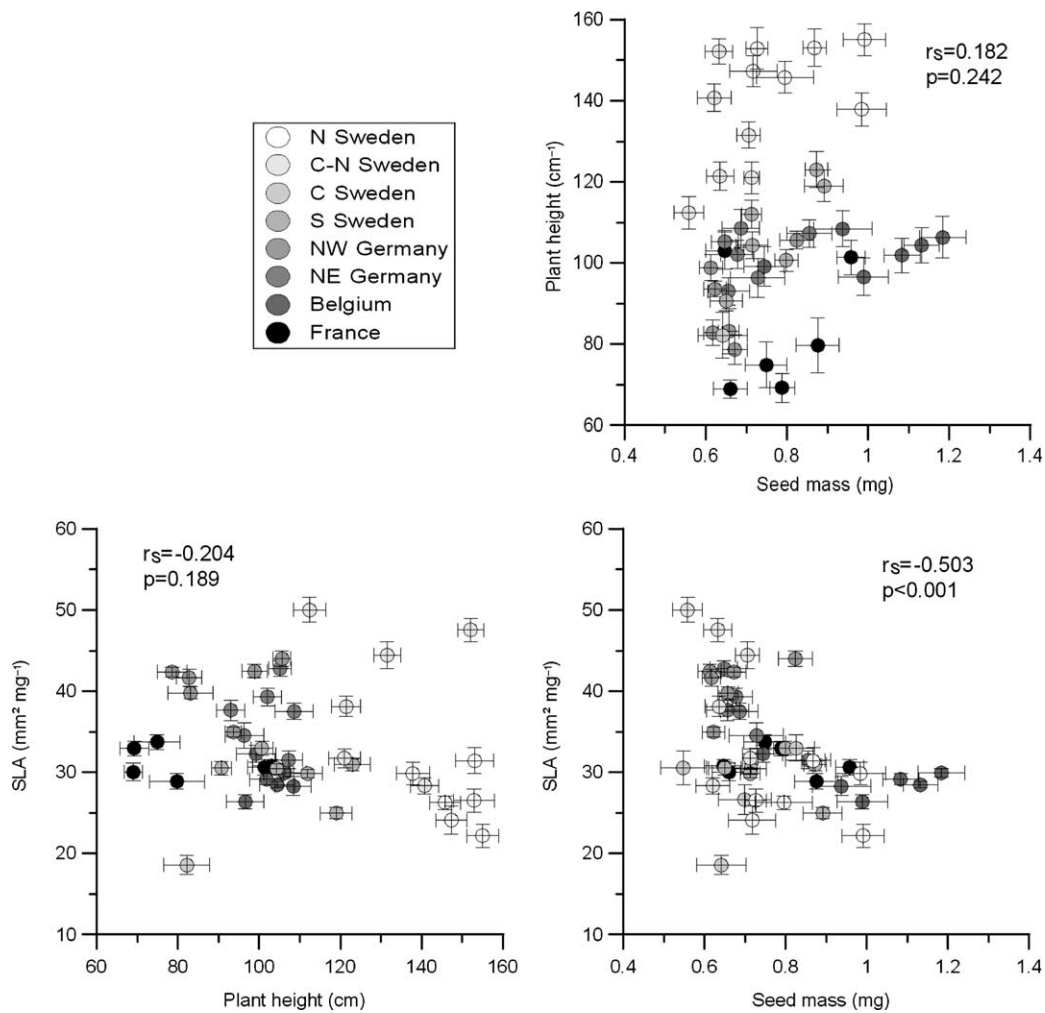


Figure 2. Leaf-height-seed relationships for *Milium effusum* populations along the latitudinal gradient. Every dot represents a mean per population  $\pm$  SE with the grey scaling indicating southerly (black) to northerly (white) regions. Spearman rank correlations between LHS traits ( $r_s$ ) and p-value are also represented ( $n = 44$ ).

VIF were lower than 10 ( $<4.85$  for *Anemone* and  $<4.32$  for *Milium*), indicating low collinearity (Quinn and Keough 2002). To avoid overfitting, the five predictors were first evaluated based on the  $-2$  Log Likelihood information criterion (deviance; Hox 2002) as stand-alone predictors. Subsequently, extra predictors were added one-by-one to the first model with the lowest deviance. If the deviance decreased significantly (Chi-square tested; Hox 2002), this procedure was repeated. Only the final models are presented in the results. All analyses were performed with SPSS 15.0.

## Results

There was clear variation in the three LHS traits along the latitudinal gradient for both species (Fig. 1, 2). *Anemone* and *Milium* individuals growing in northern populations were significantly taller at maturity compared to individuals in more southerly distributed populations (Table 2). For instance, French *Anemone* and *Milium* were on average ( $\pm$  SE) 14.2 ( $\pm 0.3$ ) cm and 82.9 ( $\pm 2.4$ ) cm tall, whereas the northernmost Swedish individuals measured 23.7 ( $\pm 0.5$ ) cm in Umeå and 148.7 ( $\pm 1.8$ ) cm in Abisko,

Table 2. Effects of latitude on leaf-height-seed traits of *Anemone nemorosa* and *Milium effusum* along the latitudinal gradient. Results from multilevel model analyses.

	LHS trait	Par. est. <sup>1</sup>	DF <sup>2</sup>	t-value	p-value
<i>Anemone</i>	Plant height	0.427	7.19	2.51	0.039 *
	SLA	0.526	7.17	1.93	0.094 (*)
	Seed mass	-0.037	7.32	-2.08	0.074 (*)
<i>Milium</i>	Plant height	3.00	3.94	6.84	0.003 **
	SLA	-0.163	7.41	-0.49	0.639 (ns)
	Seed mass	-0.003	7.83	-0.51	0.610 (ns)

(ns):  $p > 0.1$ ; (\*):  $p < 0.1$ ; \*:  $p < 0.05$ ; \*\*:  $p < 0.01$ . <sup>1</sup>Parameter estimate; <sup>2</sup>degrees of freedom.

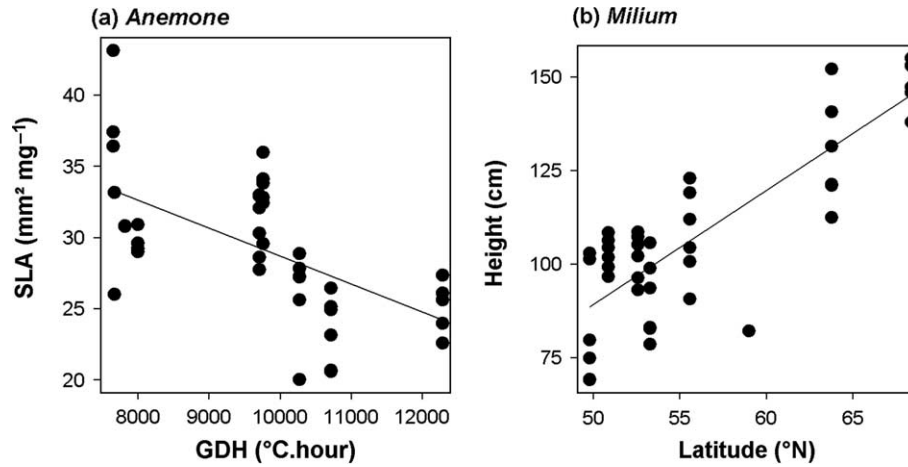


Figure 3. The effect of (a) temperature (growing degree hours, GDH) on specific leaf area (SLA) of *Anemone nemorosa* (n = 41) and (b) latitude on plant height of *Milium effusum* (n = 44) along the latitudinal gradient. See Table 2 and 3 for statistical significances.

respectively (Fig. 3b). This means that the plant height of *Anemone* and *Milium* increased significantly by 4.8% (0.43 cm) and 4.3% (3.0 cm) respectively, per degree latitude poleward shift. In the case of *Anemone*, seed mass and plant height were negatively correlated (Fig. 1), as were seed mass and SLA in *Milium* (Fig. 2). The remaining LHS traits of *Anemone* and *Milium* were uncorrelated. Seed mass and SLA were unrelated to latitude in *Milium*, whereas in *Anemone* the relation was only marginally significant (Table 2).

The height of *Anemone* and *Milium* increased significantly with increasing latitude (Table 2; Fig. 3b), but height differences in *Anemone* were mainly related to accumulated temperature (Table 3). Hence, plant height of both species was determined by regional factors (temperature and latitude), although canopy cover also affected plant height of the summer-flowering *Milium* (Table 3), i.e. plants were taller under an open canopy. SLA of both species decreased with increasing soil nutrients and, for *Anemone*, with increasing temperature (Fig. 3a). Finally, temperature (GDH) had a significant, positive effect on the seed mass of *Anemone*, whereas none of the studied variables significantly affected the seed mass of *Milium* (Table 3).

## Discussion

The LHS scheme (Westoby 1998) is used to classify species or communities according to three easily measured “soft” (sensu Diaz et al. 2004) traits: SLA, canopy height and seed mass. This study is the first to apply the LHS scheme intraspecifically in order to assess life-history trait variation of a single species along a (complex) environmental gradient. Using two forest herbs as a case study, the LHS scheme enabled us to clearly distinguish between different populations along the gradient. Our results demonstrate that LHS traits are useful for discriminating fundamental characteristics of growth and reproduction within a single species. Unlike cross-species observational quantifications, where a large part of the variability in plant traits occurs between coexisting species within a single site (Moles et al. 2007, 2009a), the within-species approach more clearly identifies the environmental variables shaping specific plant traits.

The variation in LHS traits for both species indicates that the ecological strategy determined by the position of each population in the LHS volume is not constant along the latitudinal gradient. Because taller plants possess

Table 3. Effects of regional and local environmental factors on leaf-height-seed traits of *Anemone nemorosa* and *Milium effusum* along the latitudinal gradient. Results from multilevel models with predictor variables: 1) temperature [GDH], 2) latitude, 3) soil nutrients axis [first PCA axis], 4) soil acidity axis [second PCA axis] and 5) overstorey canopy cover.

	LHS trait	Environmental factor	−2LL <sup>1</sup>	Par. est. <sup>2</sup>	DF <sup>3</sup>	t-value	p-value
<i>Anemone</i>	Plant height	Temperature	178.3	−1.67E-3	7.05	−4.33	0.003 **
	SLA	Temperature	211.7	−2.15E-3	7.12	−3.04	0.019 *
		Soil nutrients axis		−1.10	40.6	−1.80	0.079 (*)
<i>Milium</i>	Seed mass	Temperature	9.7	1.54E-4	7.23	3.61	0.008 **
	Plant height	Latitude	329.0	2.15	9.47	5.07	<0.001 ***
		Canopy cover		−0.40	34.2	−3.29	0.002 **
	SLA	Soil nutrients axis	267.4	−1.98	44.0	−2.37	0.022 *
	Seed mass	(ns) <sup>4</sup>					

(ns): p > 0.1; (\*): p < 0.1; \*: p < 0.05; \*\*: p < 0.01; \*\*\*: p < 0.001. <sup>1</sup> −2LL = −2 Log Likelihood (deviance) model fit information criterion. <sup>2</sup>Parameter estimate; <sup>3</sup>degrees of freedom. <sup>4</sup>None of the studied factors was significant.

greater competitive abilities than smaller individuals (Grime et al. 1988, Westoby 1998, Lavergne et al. 2003, Moog et al. 2005) and LHS traits clearly correspond with CSR strategies (Moog et al. 2005), the significant latitudinal increase in plant height suggests that *Anemone* and *Milium* individuals in the northern populations are more competitive. Of course, more research is needed to confirm this hypothesis in relation to coexisting species and to identify the factors driving this pattern experimentally. Furthermore, our analyses could be extended to a larger set of plant species to allow for generalization among functional types or growth forms. Finally, the criterion of LHS trait independence (Westoby 1998, Golodets et al. 2009) was mostly upheld, as most correlations between LHS traits were not significant.

### LHS trait variation and environmental factors

Firstly, plant height of both *Anemone* and *Milium* increased by >4% for every degree northward shift. Northern populations experience a longer photoperiod in the growing season when the temperature is above the threshold for plant development, and the studied forest type in the north is more open with lower canopy cover (from ~90% in France to ~40% in N Sweden), which leads to a clear northward increase in potentially available radiation during the growing season. This increase in aboveground resource availability may result in a higher possibility of increased growth and a taller plant stature in northern populations, but may also influence interspecific competition and height growth of other species (see above). Secondly, increasing temperature (GDH) had a positive effect on the seed mass of *Anemone*, but not on the seed mass of *Milium*. This confirms previous work along this latitudinal gradient, which showed that temperature had significant, positive effects on seed mass of *Anemone* (De Frenne et al. 2009, 2010), but that the effect of temperature was minor on summer-flowering forest herbs such as *Milium* (De Frenne et al. 2009, Graae et al. 2009). And thirdly, we found that both species displayed lower SLA values when they grew in a nutrient-rich environment. Two recent meta-analyses (Ordoñez et al. 2009, Poorter et al. 2009) demonstrated that irradiance, temperature, submergence and precipitation are the primary determinants of SLA, but that plants also tend to increase SLA with increasing soil fertility. The latter effect, however, is mostly observed in extremely nutrient-poor or nutrient-rich habitats, which differs from the growing situations of our study species. The comparison of the two species reveals that *Anemone* (spring-flowering) is more temperature sensitive than the early-summer flowering *Milium*, which confirms previous divergent responses to temperature between species belonging to different phenological groups (Sherry et al. 2007, Kudo et al. 2008, De Frenne et al. 2009).

### Intra- vs interspecific relationships

Some of our results support previously reported interspecific relationships between seed mass, SLA or plant height

(Westoby et al. 1996, Falster and Westoby 2003). For example, we found negative correlations between seed mass and height in *Anemone* and between SLA and seed mass in *Milium*. These correlations may reflect a fundamental trade-off among plants, e.g. between investment in height growth (access to light) and reproduction (heavier seeds) (Falster and Westoby 2003, Diaz et al. 2004, Freschet et al. 2010). But, as most of the inter-trait correlations were not significant, the criterion of LHS trait independence (Westoby 1998, Golodets et al. 2009) was mostly (but not always) upheld. In contrast, the known interspecific trends with latitude are generally not confirmed by our intraspecific comparison. Both *Anemone* and *Milium* individuals were significantly taller in the north, despite previous evidence that plant height across species and within herbs tends to decrease from the equator to the poles (Moles et al. 2009a). Moreover, species at high latitudes in general have a lower seed mass than species at low latitudes (Moles and Westoby 2003, Murray et al. 2004, Moles et al. 2007, 2009b, Pakeman et al. 2008), although it should be noted that this pattern is very weak within herbs (Moles et al. 2007). This trend is also absent in our data (see also De Frenne et al. 2009, 2010). Finally, our study shows no relationship between SLA and latitude, but a global cross-species quantification of the relationship between latitude and SLA is still lacking. Hence, the existing cross-species LHS correlations with latitude were generally unsupported within our study species.

As our analyses are based on only two European temperate forest herbs, our findings do not cast doubt on the global cross-species patterns in plant height and seed mass. Yet, the results of our study do demonstrate that on a smaller scale (one continent) and within one functional plant type (forest herbs), the intraspecific variation of these important life-history traits with latitude may differ strongly from the overall interspecific patterns. This is probably due to a combination of several factors, including the exclusion of confounding cross-species effects through growth form, seed disperser assemblage (e.g. tropical species that are more adapted to dispersal by vertebrates) or habitat type (Lavorel et al. 1997, Lord et al. 1997, Moles and Westoby 2003, Moles et al. 2007, 2009a, Tautenhahn et al. 2008), as well as the latitudinal range studied.

All of our study populations were situated within deciduous forest, whereas large-scale cross-species patterns in plant height were derived by combining different ecosystems (tropical forest, desert, tundra, etc.; Moles et al. 2009a) along a much larger latitudinal range. The 29-fold decrease in mean height between plant species growing within 15° of the equator compared to those growing between 60 and 75°N is mostly driven by differences in growth form and precipitation. For instance, there is a lack of small species in sites with high temperature, precipitation and/or productivity and there are large treeless areas in the north (Moles et al. 2009a). In contrast, growth form and precipitation are of minor importance in our study because we investigated intraspecific patterns and there are no extreme differences in precipitation among the study regions (Table 1). Our approach probably increases the significance of the effect of light availability (see above) on LHS traits, as compared to growth form and precipitation.

Moles and Westoby (2003) found a significant within-species relationship between seed mass and latitude for 78 species, but the intraspecific slope was only two-sevenths of the interspecific slope. They also found that species with a wider latitudinal range were more likely to show a decline in seed mass with latitude. However, the sampled latitudinal ranges for *Anemone* and *Milium* in our study were 14° and 18.6°, respectively, which is a wide range when compared to the other studies compiled in Moles and Westoby (2003). Hence, the within-species decline in seed mass towards the poles is rather idiosyncratic (see also Murray et al. 2004, De Frenne et al. 2009) compared to the decline found across species.

## Conclusions

The LHS traits of multiple populations of a single species varied clearly along the latitudinal gradient. We found an almost two-fold increase in plant height between the southernmost and northernmost *Anemone* (14° latitudinal range) and *Milium* (18.6° latitudinal range) populations, which suggests increased competitive abilities in the north for both species. Furthermore, we found evidence that environmental factors on different scales (local vs regional) influence the two study species differently: *Anemone*, a spring-flowering herb, is influenced most by temperature, whereas *Milium*, an early-summer flowering grass, is affected more by factors operating on the local scale (canopy cover, soil nutrients) and by other latitude-related variables. Finally, most previously established cross-species correlations are unsupported within our study species, which suggests that the global patterns in LHS traits found across species are not as widespread within species as previously thought. Further research into the causes (phenovs genotypic) and relativity (in relationship to coexisting species) of the within-species variation in LHS traits and ecological strategy of these forest herbs and other plant species with latitude is required.

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