

Intra-seasonal variation and relationship among leaf traits of different forest herbs in a dry tropical environment

Prajjwal Dubey*, A. S. Raghubanshi and J. S. Singh

Ecosystems Analysis Laboratory, Department of Botany, Banaras Hindu University, Varanasi 221 005, India

A range of leaf traits was measured for 17 herbaceous species fortnightly from July to September for two years in a dry tropical forest. These species belonged to four life forms: annual grasses, perennial grasses, annual forbs and perennial forbs. The objectives of this study were: (i) to assess the effect of herb types on leaf traits in tropical dry deciduous forest of the Vindhyan highlands; (ii) to measure the growing season variation in the traits, and (iii) to investigate the relationships between the leaf traits.

Four sites, viz. Hathinala, Ranitali, Bokrakhari and Neuriuadamar were selected in the region. At each site, five quadrats, each 1 m × 1 m in size, were sampled randomly for herbaceous plants at 15-day interval in the wet season from July to September during 2006 and 2007. At each site soil samples and leaf samples were collected and analysed. Specific leaf area (SLA), leaf nitrogen (N), leaf phosphorus (P), leaf chlorophyll content, stomatal conductance (G_s) and photosynthetic rate (A_{mass}) were determined.

The seasonal pattern in leaf traits, in general, was an early season peak in SLA, leaf nitrogen and leaf phosphorus, and a midseason peak in G_s and A_{mass} . Annual forbs generally exhibited highest leaf trait values and the perennial grasses the lowest. Pearson correlation coefficients indicated significant positive relationship of SLA with all other leaf traits, between leaf N and leaf P and among chlorophyll, G_s and A_{mass} . The decline in leaf nitrogen and leaf phosphorus during the growing season occurred due to the retranslocation of these elements from the leaves into the storage or reproductive organs. The peaks in G_s and A_{mass} were associated with increase in soil moisture.

Keywords: Forest herbs, life forms, photosynthetic rate, specific leaf area, stomatal conductance.

LEAF traits are good predictors of plant performance as they are closely associated with growth and survival of the plant¹. These are often considered the principal traits with regard to plant resource use, biomass and ecosystem functioning, and the variations in them imply different strategies of plant species in some way^{2,3}. In addition,

these traits are easy to quantify and convenient to compare among large number of plant species⁴. Leaf structure, nutrient content and net photosynthetic capacity are key determinants of carbon dioxide and water vapour fluxes between the vegetation and the atmosphere, and of biogeochemical cycles that link soil, climate and the atmosphere⁵. These traits interact to determine plant behaviour and production, and provide a useful conceptual link between processes at short-term leaf scales and long-term whole plant and stand-level scales⁶⁻¹⁰. Fast-growing species tend to have short-lived leaves with a high specific leaf area (SLA). SLA is a potential predictive tool for the grazing responses; grazing-resistant species are reported to be shorter in height and have smaller, more tender leaves, with higher SLA than grazing-susceptible species¹¹. SLA represents the light-intercepting area of a leaf per unit dry mass, related to net assimilation rate^{8,12}, and besides being easy to measure, is a strong correlate of photosynthetic capacity and potential relative growth rate and is inversely related to the degree of physical defence of a leaf¹²⁻¹⁵. At the ecosystem level, SLA (or related leaf traits) and leaf nitrogen content (LNC) of component species may have a significant impact on primary productivity and nutrient cycling^{8,16,17}. SLA and LNC are important traits for plant growth and development because they provide information on main attributes such as relative growth rate and leaf gas exchange¹⁸. Leaf nitrogen (N) is integral to the proteins of photosynthetic machinery, especially Rubisco. Leaf phosphorus (P) is found in nucleic acids, lipid membranes and bioenergetics molecules such as ATP. Phosphorus is derived from weathering of soil minerals in a site, in contrast to nitrogen, much of which may be fixed from the atmosphere by the plants¹⁹. Above-ground productivity and quality of grasses is partly controlled by leaf traits and especially by the leaf lamina N content per unit fresh matter²⁰.

Leaf traits such as SLA and LNC could also be used as easily assessable predictive tools of litter decomposability, without requiring any detailed knowledge of individual species taxonomy and biology²¹. Photosynthetic characteristics and stomatal behaviour of plant species help predict carbon and water fluxes at the leaf, plant, ecosystem and biome levels⁹. Photosynthetic capacity is influenced both by stomatal conductance and the drawdown of

*For correspondence. (e-mail: prajjalg@gmail.com)

CO₂ concentration inside the leaf (carboxylation capacity)¹⁹.

Strong seasonal variations have been reported in several leaf traits such as SLA, LNC and leaf dry matter, particularly in herbaceous species²². These seasonal variations exhibit the response of plants to changing environmental conditions, such as light level and air temperature²³, water availability²⁴ and leaf ageing²⁵. Several studies have shown that dilution and retranslocation are the main factors related to seasonal fluctuations in leaf nutrient status²⁶. In order to understand the relationship between leaf traits and ecosystem processes, knowledge of the seasonal behaviour of the traits is required.

Forest herbs are usually characterized into life form classes, viz. annual grasses, annual forbs, perennial grasses and perennial forbs. These life form categories may show differential response to grazing and other environmental variables due to their differential growth characteristics.

The objectives of this study were: (i) to assess the effect of life form on leaf traits in the herbaceous flora of tropical dry deciduous forest of the Vindhyan highlands; (ii) to measure the growing season variation in the traits, and (iii) to investigate the relationships between the leaf traits.

Materials and methods

Study site

The study area lies in the Vindhyan plateau, Sonbhadra District (24°6'52"–24°26'16"N; 83°1'86"–83°9'60"E), Uttar Pradesh, India. The elevation above the mean sea level²⁷ ranges between 315 and 485 m. The climate is tropical with three seasons in a year, i.e. summer (March to mid June), rainy (mid June to September) and winter (October to February). October and March constitute the transition months between the rainy and winter seasons, and between winter and summer seasons respectively. The annual rainfall (1981–1988) varies between 926 mm at Obra meteorological station and 1145 mm at Renukoot meteorological station. About 85% of the annual rainfall occurs during the rainy season from the southwest monsoon²⁸. Red-coloured and fine-textured sandstone (Dhandraul orthoquartzite) is the most important rock of the area. Sandstone is generally underlain by shale and limestone. The soils derived from these rocks are residual ultisols and are sandy-loam in texture²⁹. These soils are part of the hyperthermic formation of typical plinthustults with ustorthents according to VII approximation of the USDA soil nomenclature³⁰. The potential natural vegetation of the region is tropical dry deciduous forest; the dominant tree species are *Anogeissus latifolia*, *Boswellia serrata*, *Buchanania lanzan*, *Diospyros melanoxylon*, *Hardwickia binata*, *Lagerstroemia parviflora*, *Lannea coromandelica*, *Madhuca longifolia*, *Shorea robusta* and

Terminalia tomentosa. Herbaceous vegetation is dominated by *Hyptis suaveolens*, *Sporobolus diander*, *Oplismenus compositus* and *Abutilon indicum*.

Method

Four sites, viz. Hathinala, Ranitali, Bokrakhari and Neuriadamar in the region were selected. These are considered replicate sites for the tropical dry deciduous forest of the Vindhyan hills. All experimental sites are situated within a radius of 50 km, between Obra and Renukoot meteorological stations. Hathinala and Neuriadamar sites belong to Renukoot forest division, while Ranitali and Bokrakhari sites belong to Obra forest division. At each site, five quadrats, each 1 m × 1 m in size, were sampled randomly for herbaceous plants at 15 days interval in the wet season from July to September during 2006 and 2007. On each sampling date new quadrats were laid. New leaves begin to expand at the start of the rainy season, which constitutes the peak growth period. Each year, the first sampling began on 3 July (Julian day 184) and was completed in two days at all sites. The last sampling of the season began on 18 September (Julian day 259) and was completed within two days at all sites. Soil moisture was measured by theta probe instrument (type ML1, Delta-T devices, Cambridge, England) at three locations on each site on each sampling date. Plants were classified into four life form classes: annual grasses, annual forbs, perennial grasses and perennial forbs. Light incident on the ground was measured by digital lux meter (type LX-101 Lutron). Data on climatic variables such as rainfall at the sampling dates were taken from the meteorological records of Obra and Renukoot forest divisions. From each site, five soil samples were collected from three random locations to a depth of 10 cm. Soil was analysed for pH (ref. 31) and texture³², organic carbon³³, nitrogen³⁴ and phosphorus³⁵. In all, 17 herbaceous species were recorded in the quadrats. For the analysis of leaf traits, 10 fully expanded leaves for each species were collected from as many individuals. Stomatal conductance (G_s) and photosynthetic rate (A_{mass}) were measured *in situ* by LCpro photosynthesis meter (model EN 11 ODB, ADC Bioscientific Ltd, England). Samples were transported to the laboratory in an ice-box, which provided a cool and wet environment. Leaf area was recorded using the leaf area meter (Systronics; Leaf area meter-211). Fresh leaves were dried at 80°C for 48 h to estimate their dry weight. SLA was determined as the ratio of leaf area to leaf dry weight. Leaf nitrogen was measured by Kjeldahl method³⁶ and phosphorus by phosphomolybdic blue colorimetric method³⁷. For measurement of leaf chlorophyll, fresh samples were crushed with 80% acetone solution and then filtered through a muslin cloth. The filtrate was collected and optical density was taken at 660 and 645 nm with a spectrophotometer³⁸. All analyses were

done separately for each species. Values were averaged across species within a life form class and across sites and years.

Statistical analyses

Data were analysed by ANOVA with leaf traits as dependent variables and life forms and sampling date as independent (fixed) variables. Relationships among leaf traits and between leaf traits and climatic variables were examined through two-tailed Pearson correlation coefficients. All the statistical analyses were conducted using SPSS³⁹.

Results

Soil is sandy loam in texture, slightly acidic and poor in nutrients (Table 1). On the average, about 75% of sunlight was incident on the ground as measured at the beginning of the rainy season. ANOVA indicated significant life form and sampling date effects on all the leaf traits (Table 2).

Specific leaf area

SLA generally exhibited an early season peak (Figure 1 a). SLA for annual grasses varied between 304 and 364 cm² g⁻¹ and averaged 332 cm² g⁻¹ across the season (Figure 1 a). Within this group, *Setaria glauca* showed the maximum peak SLA (Table 3). SLA for annual forbs varied between 341 and 398 cm² g⁻¹ and averaged 373 cm² g⁻¹ across the season (Figure 1 a). Within this group, *Alysicarpus vaginalis* showed the maximum peak SLA (Table 3). SLA for the perennial grass group varied between 262 and 300 cm² g⁻¹ and averaged 285 cm² g⁻¹ across the season (Figure 1 a). Within this group, *Heteropogon contortus* showed maximum peak SLA (Table 3). For perennial forbs, SLA ranged from 280 to 358 cm² g⁻¹ and averaged 312 cm² g⁻¹ across the season (Figure 1 a). Within this group, *Blepharis repens* showed the highest peak SLA (Table 3).

Table 1. Physico-chemical characteristics of the soil

Parameter	Mean ± SE
Soil pH	6.56 ± 0.11
Clay content (%)	4 ± 1.1
Silt content (%)	28 ± 1.73
Sand content (%)	45 ± 1.64
Gravel content (%)	23 ± 2.03
Total soil carbon (%)	1.41 ± 0.07
Total soil nitrogen (%)	0.13 ± 0.01
Total soil phosphorus (%)	0.03 ± 0.01

Leaf nitrogen

In general, leaf N showed an early season peak (Figure 1 b). Leaf N for annual grasses varied between 0.01 and 0.03 g g⁻¹ and averaged 0.02 g g⁻¹ across the season (Figure 1 b). Within this group, *S. glauca* showed the maximum peak leaf N (Table 3). Leaf N for annual forbs ranged between 0.01 and 0.03 g g⁻¹ and averaged 0.02 g g⁻¹ across the season (Figure 1 b). Within this group, *A. vaginalis* showed the maximum peak leaf N (Table 3). For perennial grasses, leaf N varied between 0.01 and 0.02 g g⁻¹ and averaged 0.02 g g⁻¹ across the season (Figure 1 b). Within this group, *H. contortus* showed the maximum peak leaf N (Table 3). In perennial forbs, leaf N varied between 0.01 and 0.02 g g⁻¹ and averaged 0.02 g g⁻¹ across the season (Figure 1 b). Within this group, *B. repens* showed the greatest leaf N (Table 3).

Leaf phosphorus

Leaf P followed the seasonal pattern of leaf N, with peak values during the early part of growing season (Figure 1 c). Leaf P for annual grasses ranged from 0.001 to 0.002 g g⁻¹ and averaged 0.002 g g⁻¹ across the season (Figure 1 c). Within this group, *S. glauca* showed the maximum peak leaf P (Table 3). In the case of annual forbs, leaf P varied from 0.001 to 0.004 g g⁻¹ and averaged 0.002 g g⁻¹ (Figure 1 c). Within this group, *A. vaginalis* showed the highest value for leaf P (Table 3). For perennial grasses, leaf P varied between 0.001 and 0.002 g g⁻¹ and averaged 0.002 g g⁻¹ (Figure 1 c). Within this group *H. contortus* showed the maximum peak leaf P (Table 3). In perennial forbs, leaf P varied between 0.001 and 0.003 g g⁻¹ and averaged 0.002 g g⁻¹ (Figure 1 c). Among the species within this group, *B. repens* showed the maximum peak leaf P (Table 3).

Chlorophyll

Chlorophyll concentration generally increased with time and attained peak values in the later part of the growing season (Figure 2 a). Chlorophyll in annual grasses ranged between 1.3 and 1.80 mg g⁻¹ and averaged 1.58 mg g⁻¹ (Figure 2 a). Within this group, *S. glauca* showed the maximum peak chlorophyll (Table 3). For annual forbs, chlorophyll ranged from 1.48 to 1.94 mg g⁻¹ and averaged 1.66 mg g⁻¹ (Figure 2 a). Among the species within this group, *A. vaginalis* showed the maximum peak chlorophyll (Table 3). In the case of perennial grasses, chlorophyll varied between 1.04 and 1.62 mg g⁻¹ and averaged 1.38 mg g⁻¹ (Figure 2 a). Among the species within this group species, *H. contortus* showed the maximum peak chlorophyll (Table 3). For perennial forbs, chlorophyll varied from 1.47 to 1.81 mg g⁻¹ and averaged 1.63 mg g⁻¹ (Figure 2 a). Within this group *B. repens* showed the highest peak chlorophyll (Table 3).

Table 2. Summary of ANOVA on leaf traits

	df	Leaf chlorophyll (Chl)	Specific leaf area (SLA)	Leaf nitrogen (N)	Leaf phosphorus (P)	Photosynthetic rate (A_{mass})	Stomatal conductance (G_s)
Life form	3	18.9***	145.8***	52.4***	32.5***	62.39***	35.2***
Sampling date	5	8***	12.5***	119.8***	80.5***	9.05***	5.5***
Life form × sampling date	15	0.947 ^{ns}	0.676 ^{ns}	2.9**	1.9*	0.256 ^{ns}	0.733 ^{ns}
Error	62						

ns, Not significant, * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

Table 3. Peak values (\pm SE) of leaf traits of different herbaceous species

Species	SLA	N	P	Chl	A_{mass}	G_s
Annual grass						
<i>Digitaria adscendens</i>	305 \pm 0.33 (184)	0.02 \pm 0.00 (184)	0.002 \pm 0.00 (184)	1.75 \pm 0.06 (229)	64.05 \pm 0.52 (229)	0.24 \pm 0.01 (229)
<i>Setaria glauca</i>	428 \pm 1.45 (199)	0.03 \pm 0.00 (199)	0.004 \pm 0.00 (199)	1.96 \pm 0.01 (229)	162.64 \pm 0.55 (229)	0.40 \pm 0.01 (229)
<i>Eragrostis tenella</i>	295 \pm 1.42 (214)	0.02 \pm 0.00 (214)	0.002 \pm 0.00 (214)	1.69 \pm 0.1 (229)	76.70 \pm 0.63 (229)	0.31 \pm 0.01 (229)
Annual forb						
<i>Abutilon indicum</i>	391 \pm 0.38 (184)	0.03 \pm 0.00 (184)	0.003 \pm 0.00 (184)	1.64 \pm 0.01 (214)	121.21 \pm 0.75 (214)	0.35 \pm 0.003 (214)
<i>Phyllanthus virgatus</i>	326 \pm 0.83 (214)	0.02 \pm 0.00 (214)	0.003 \pm 0.00 (214)	1.64 \pm 0.01 (244)	94.54 \pm 3.34 (244)	0.44 \pm 0.02 (244)
<i>Tridax procumbens</i>	399 \pm 1.76 (214)	0.03 \pm 0.00 (214)	0.003 \pm 0.00 (214)	1.98 \pm 0.05 (244)	151.62 \pm 0.54 (244)	0.46 \pm 0.01 (244)
<i>Alysicarpus vaginalis</i>	405 \pm 3.7 (184)	0.03 \pm 0.00 (184)	0.004 \pm 0.00 (184)	2.33 \pm 0.01 (244)	149.85 \pm 1.08 (244)	0.46 \pm 0.01 (244)
<i>Hyptis suaveolens</i>	403 \pm 1.17 (214)	0.03 \pm 0.00 (214)	0.003 \pm 0.00 (214)	1.64 \pm 0.01 (244)	141.05 \pm 0.05 (244)	0.4 \pm 0.01 (244)
Perennial grass						
<i>Dichanthium annulatum</i>	292 \pm 1.67 (184)	0.02 \pm 0.00 (184)	0.002 \pm 0.00 (184)	1.56 \pm 0.21 (229)	87.60 \pm 1 (229)	0.31 \pm 0.01 (229)
<i>Sporobolus diander</i>	262.84 \pm 0.84 (199)	0.02 \pm 0.00 (199)	0.002 \pm 0.00 (199)	1.84 \pm 0.08 (229)	78.9 \pm 0.93 (229)	0.32 \pm 0.01 (229)
<i>Oplismenus compositus</i>	274 \pm 1.23 (184)	0.02 \pm 0.00 (184)	0.002 \pm 0.00 (184)	1.32 \pm 0.00 (214)	63.02 \pm 0.41 (214)	0.23 \pm 0.01 (214)
<i>Heteropogon contortus</i>	388 \pm 3.21 (199)	0.03 \pm 0.00 (199)	0.003 \pm 0.00 (199)	2 \pm 0.05 (229)	124.16 \pm 0.69 (229)	0.40 \pm 0.01 (229)
Perennial forb						
<i>Alternanthera sessilis</i>	290 \pm 1.13 (199)	0.02 \pm 0.00 (199)	0.002 \pm 0.00 (199)	1.79 \pm 0.05 (214)	81.20 \pm 1.82 (214)	0.31 \pm 0.01 (214)
<i>Blepharis repens</i>	419 \pm 2.31 (184)	0.03 \pm 0.00 (184)	0.004 \pm 0.00 (184)	2.64 \pm 0.01 (214)	159.22 \pm 1.27 (214)	0.39 \pm 0.01 (214)
<i>Caesulia axillaries</i>	296 \pm 0.5 (184)	0.02 \pm 0.00 (184)	0.002 \pm 0.00 (184)	1.72 \pm 0.04 (214)	85.84 \pm 0.77 (214)	0.29 \pm 0.01 (214)
<i>Sida acuta</i>	286 \pm 0.97 (199)	0.02 \pm 0.00 (199)	0.002 \pm 0.00 (199)	2.47 \pm 0.02 (244)	105.82 \pm 0.65 (244)	0.38 \pm 0.003 (244)
<i>Evolvulus numularius</i>	394 \pm 1.15 (214)	0.02 \pm 0.00 (214)	0.003 \pm 0.00 (214)	2.25 \pm 0.01 (244)	133.96 \pm 0.91 (244)	0.38 \pm 0.01 (244)

Numbers in parentheses are Julian days when peak values were recorded.

Stomatal conductance

G_s of annual grasses varied between 0.24 and 0.35 mol m⁻² s⁻¹ and averaged 0.27 mol m⁻² s⁻¹ across the season (Figure 2 b). Within this group, *S. glauca* showed the maximum

peak G_s (Table 3). For the annual forbs, G_s varied between 0.33 and 0.40 mol m⁻² s⁻¹ and averaged 0.36 mol m⁻² s⁻¹ (Figure 2 b). Within this group, *A. vaginalis* showed the maximum peak G_s (Table 3). For perennial grasses, G_s varied between 0.17 and

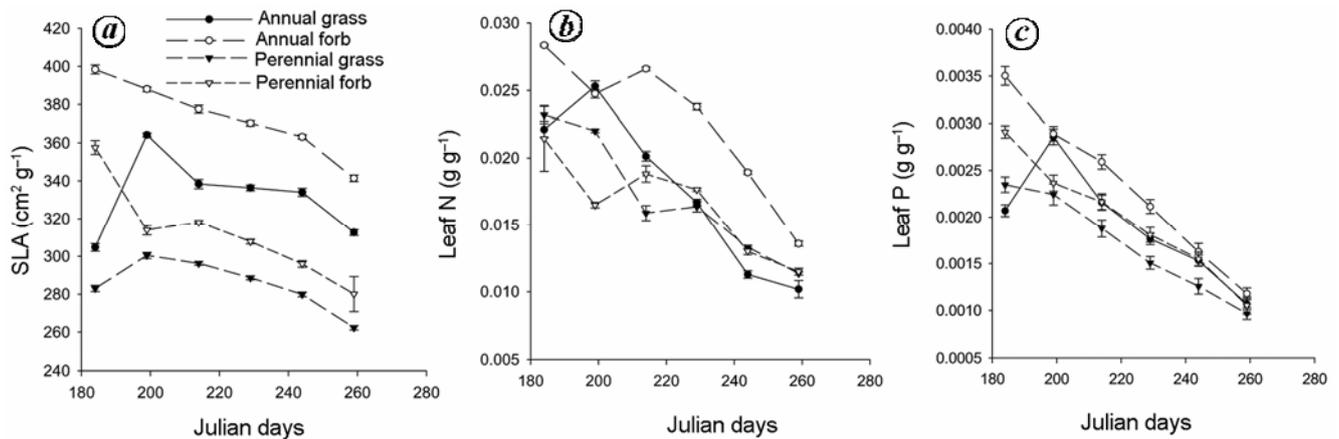


Figure 1. Intra-seasonal variations in (a) specific leaf area (SLA), (b) leaf nitrogen (N) and (c) leaf phosphorus (P) of annual grass, annual forb, perennial grass and perennial forb.

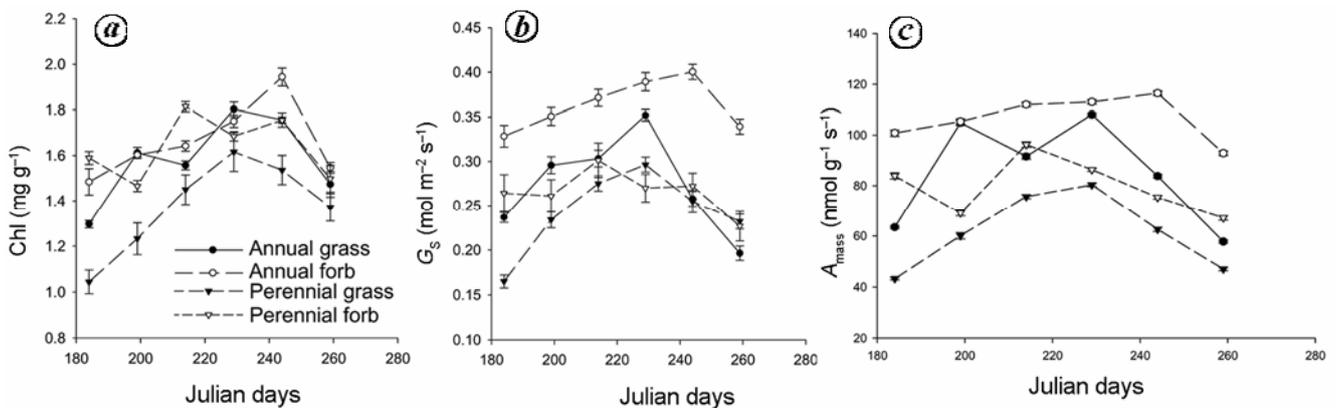


Figure 2. Intra-seasonal variations in (a) leaf chlorophyll (Chl), (b) stomatal conductance (G_s) and (c) photosynthetic rate (A_{mass}) of annual grass, annual forb, perennial grass and perennial forb.

0.29 mol m⁻² s⁻¹ and averaged 0.24 mol m⁻² s⁻¹ (Figure 2b). Within this group, *H. contortus* showed the maximum peak G_s (Table 3). For perennial forbs, G_s varied between 0.26 and 0.30 mol m⁻² s⁻¹ and averaged 0.27 mol m⁻² s⁻¹ (Figure 2b). Within this group, *B. repens* showed the maximum peak G_s (Table 3). In general, G_s exhibited a mid-season peak (Figure 2b).

Photosynthetic rate

For annual grasses A_{mass} varied between 57.76 and 108.12 nmol g⁻¹ s⁻¹ and averaged 84.93 nmol g⁻¹ s⁻¹ (Figure 2c). Within this group, *S. glauca* showed the maximum peak A_{mass} (Table 3). For annual forbs, A_{mass} varied between 92.88 and 116.42 nmol g⁻¹ s⁻¹ and averaged 106.74 nmol g⁻¹ s⁻¹ across the season (Figure 2c). Within this group, *A. vaginalis* showed the maximum peak A_{mass} (Table 3). In the case of perennial grasses, A_{mass} varied between 43.24 and 80.33 nmol g⁻¹ s⁻¹ and averaged 61.48 nmol g⁻¹ s⁻¹ (Figure 2c). Within this group, *H. contortus* showed the maximum peak A_{mass} (Table 3). For

perennial forbs, A_{mass} varied between 67.05 and 96.27 nmol g⁻¹ s⁻¹ and averaged 79.67 nmol g⁻¹ s⁻¹ (Figure 2c). Within this group, *B. repens* showed the maximum peak A_{mass} (Table 3). In general, A_{mass} showed a mid-season peak (Figure 2c).

Among the life form classes, annual forbs generally exhibited the highest values of leaf traits and perennial grasses the lowest. Species in the same life form attained peak values on different Julian days for different leaf traits (Table 3). For instance, the annual forb *Abutilon indicum* showed peak value for leaf chlorophyll at the third sampling date (Julian day 214), whereas another annual forb, *Phyllanthus virgatus* showed peak value for leaf chlorophyll at the fifth sampling date (Julian day 244).

Pearson correlation coefficients indicated significant positive relationship of SLA with all other leaf traits, between leaf N and leaf P and among chlorophyll, G_s and A_{mass} (Table 4).

Rainfall in the preceding 15 Julian days before each sampling event, and soil moisture on each sampling date are shown in Table 5. Rainfall was maximum during the

Table 4. Pearson correlation between leaf traits

	Chl	SLA	N	P	A_{mass}
SLA	0.282**				
N	-0.155 ^{ns}	0.703**			
P	-0.047 ^{ns}	0.675**	0.916**		
A_{mass}	0.706**	0.788**	0.393**	0.409**	
G_s	0.714**	0.633**	0.268*	0.295**	0.907**

*Correlation is significant at the 0.05 level (two-tailed). **Correlation is significant at the 0.01 level (two-tailed). ns, Not significant.

Table 5. Soil moisture on sampling dates and rainfall during the preceding 15 days of sampling

Sampling date (Julian days)	Soil moisture (mean % \pm SE)	Rainfall in the preceding 15 days (mm)
3 July (184)	22.7 (\pm 1.8)	292
18 July (199)	22.3 (\pm 1.2)	294
4 August (214)	24.2 (\pm 2.4)	364
19 August (229)	25 (\pm 1.7)	365
3 September (244)	19.2 (\pm 2.2)	222
18 September (259)	17.3 (\pm 2.4)	220

fourth sampling interval (between 214 and 229 Julian day), and soil moisture was maximum on the fourth sampling date (229 Julian day). As expected, rainfall and soil moisture were interrelated ($r = 0.89$, $P < 0.01$). The mid-season peaks in G_s and A_{mass} generally coincided with high soil moisture condition. Rainfall was positively related to G_s ($r = 0.75$, $P < 0.01$) and A_{mass} ($r = 0.73$, $P < 0.05$). Both G_s ($r = 0.74$, $P < 0.01$) and A_{mass} ($r = 0.69$, $P < 0.05$) were also positively related with soil moisture.

Discussion

SLA, leaf N and leaf P were the highest in young leaves and declined through the season as the plants accumulated leaf biomass. Apart from the dilution in leaf N and leaf P due to increasing leaf biomass, organic N and P fractions were probably hydrolysed and inorganic P and amino acid N were translocated out of leaves, as reported by Chapin and Kedrowski⁴⁰. Garnier *et al.*²² argued that dilution by stored carbohydrates and/or nutrient deficiencies induced by low water availability may trigger retranslocation of nutrients towards storage organs. Many herbaceous plants die-off completely above-ground and store reserves in root and rhizomes during hostile period^{41,42}. During the course of leaf senescence, protein, nucleic acid and other nitrogenous compounds are degraded, and their nitrogenous contents are remobilized and retranslocated into other actively growing organs^{43,44}. In nutrient-poor environment, nutrients are conserved mainly by extending the lifespan of the plants or by retranslocation⁴⁵. The retranslocation of nitrogen and phosphorus to different sink organs (e.g. flowers, fruits and rhizomes) could contribute to this decrease²⁷. As

indicated by ANOVA, there was a significant effect of life form on N and P; thus the magnitude of decline in leaf N and P varied across life forms. Annual grasses showed 59.8% decline in leaf N, 62.7% in leaf P, and annual forbs 52.1% in leaf N, 66.4% in leaf P. Perennial grasses showed 51% decline in leaf N, 58.9% in leaf P, and perennial forbs 46.4% in leaf N, 63.8% in leaf P. Annual grasses and forbs showed a greater decline in leaf N and leaf P presumably because of massive redistribution to reproductive organs. Several other studies on herbaceous species have also reported that the retranslocation process occurs during leaf senescence⁴⁶⁻⁴⁸.

Mid-season peak in G_s and A_{mass} in our study could be attributed to better environmental conditions caused by increased soil moisture. Subsequent fall in these attributes was due to senescence caused by soil moisture depletion and other environmental factors⁴⁹.

A number of studies have reported a positive association between SLA and leaf nitrogen concentration⁵⁰⁻⁵². Garnier *et al.*²² have also reported that positive associations are usually found between SLA and nitrogen concentration at the leaf level. The high correlation between leaf N and leaf P as found in our study reflects their close association in plant biochemistry, particularly protein synthesis²⁷. The combination of a high N investment in photosynthetic enzymes and pigments, and a high SLA can provide high metabolism (gas exchange rate) and greater light harvesting per unit tissue^{8,53}. Positive correlation among SLA, A_{mass} and G_s was also reported⁵⁴⁻⁵⁷. Wright *et al.*⁵⁸ reported a number of broad relationships between leaf traits and climate. Ordoñez *et al.*⁵⁹ found that precipitation affected the trade-off between traits associated with growth and resource conservation strategies.

Our study showed that the initial part of the growing season is the best for sampling in the case of SLA, leaf N and leaf P, while the mid-season period is better for quantifying G_s and A_{mass} . Further, since SLA had significant correlation with all other leaf traits and its seasonal pattern was consistent across all life forms, it can be considered the most important trait for the study of forest herbs, and can predict photosynthetic rate (hence productivity) and decomposability of the herbaceous biomass.

1. Poorter, L. and Bongers, F., Leaf traits are good predictor of plant performance across 53 rain forest species. *Ecology*, 2006, **87**, 1733–1743.
2. Westoby, M. A., Leaf–height–seed (LHS) plant ecology strategy scheme. *Plant Soil*, 1998, **199**, 213–227.
3. Vendramini, F., Diaz, S., Gurvich, D. E., Wilson, P. J., Thompson, K. and Hodgson, J. G., Leaf traits as indicators of resource-use strategy in floras with succulent species. *New Phytol.*, 2002, **154**, 147–157.
4. Liu, J., Zeng, D., Lee, D. K., Fan, Z. and Zhong, L., Leaf traits and their interrelationship of 23 plant species in southeast of Keerqin Sandy Lands, China. *Front. Biol. China.*, 2008, **3**, 332–337.
5. Reich, P. B., Wright, I. J. and Lusk, C. H., Predicting leaf physiology from simple plant and climate attributes: a global GLOPNET analysis. *Ecol. Appl.*, 2007, **17**, 1982–1988.
6. Chabot, B. F. and Hicks, D. J., The ecology of leaf life spans. *Annu. Rev. Ecol. Syst.*, 1982, **13**, 229–259.
7. Field, C., Allocating leaf nitrogen for the maximization of carbon gain: leaf age as a control on the allocation program. *Oecologia*, 1983, **56**, 341–347.
8. Reich, P. B., Walters, M. B. and Ellsworth, D. S., Leaf life-span in relation to leaf, plant, and stand characteristics among diverse ecosystems. *Ecol. Monogr.*, 1992, **62**, 365–392.
9. Schulze, E. D., Kelliher, F. M., Korner, C., Lloyd, J. and Leuning, R., Relationships among maximum stomatal conductance, ecosystem surface conductance, carbon assimilation rate, and plant nitrogen nutrition: a global ecology scaling exercise. *Annu. Rev. Ecol. Syst.*, 1994, **25**, 629–660.
10. Meir, P. *et al.*, Acclimation of photosynthetic capacity to irradiance in tree canopies in relation to leaf nitrogen concentration and leaf mass per unit area. *Plant Cell Environ.*, 2002, **25**, 343–357.
11. Díaz, S., Noy-Meir, I. and Cabido, M., Can grazing response of herbaceous plants be predicted from simple vegetative traits? *J. Appl. Ecol.* 2001, **38**, 497–508.
12. Reich, P. B., Uhl, C., Walters, M. B. and Ellsworth, D. S., Leaf lifespan as a determinant of leaf structure and function among 23 tree species in Amazonian forest communities. *Oecologia*, 1991, **86**, 16–24.
13. Reich, P. B., Walters, M. B. and Ellsworth, D. S., From tropics to tundra: global convergence in plant functioning. *Proc. Natl. Acad. Sci. USA*, 1997, **94**, 13730–13734.
14. Wright, I. J. and Westoby, M., Leaves at low versus high rainfall: coordination of structure, lifespan and physiology. *New Phytol.*, 2002, **155**, 403–404.
15. Cornelissen, J. H. C. *et al.*, Handbook of protocols for standardised and easy measurements of plant functional traits worldwide. *Aust. J. Bot.*, 2003, **51**, 335–380.
16. Cornelissen, J. H. C. *et al.*, Leaf structure and defense control litter decomposition rate across species and life forms in regional flora on two continents. *New Phytol.*, 1999, **143**, 191–200.
17. Aerts, R. and Chapin, F. S., The mineral nutrition of wild plants revisited: a re-evaluation of processes and patterns. *Adv. Ecol. Res.*, 2000, **30**, 1–67.
18. Garnier, E., Cordonnier, P., Guillermin, J. L. and Sonié, L., Specific leaf area and leaf nitrogen concentration in annual and perennial grass species growing in Mediterranean old-fields. *Oecologia*, 1997, **111**, 490–498.
19. Wright, I. J. *et al.*, The worldwide leaf economics spectrum. *Nature*, 2004, **428**, 821–827.
20. Pontes, L., Das, J., Soussana, F., Louault, F., Andueza, D. and Carrere, P., Leaf traits affect the above-ground productivity and quality of pasture grasses. *Funct. Ecol.*, 2007, **21**, 844–853.
21. Fortunel, C. *et al.*, Leaf traits capture the effects of land use changes and climate on litter decomposability of grasslands across Europe. *Ecology*, 2009, **90**, 598–611.
22. Garnier, E. *et al.*, Consistency of species ranking based on functional leaf traits. *New Phytol.*, 2001, **152**, 69–83.
23. Björkman, O., Responses to different quantum flux densities. In *Physiological Plant Ecology I Responses to the Physical Environment* (eds Lange, O. L. *et al.*), Springer-Verlag, Berlin, 1981, pp. 57–107.
24. Ehleringer, J. R. and Cook, C. S., Photosynthesis in *Enceliafarinosa* Gray in response to decreasing leaf water potential. *Plant Physiol.*, 1984, **75**, 688–693.
25. Wilson, J. R., Variation of leaf characteristics with level of insertion on a grass tiller. I. Development rate, chemical composition and dry matter digestibility. *Aust. J. Agric. Res.*, 1976, **27**, 343–354.
26. Roca-Pérez, L., Boluda, R. and Pérez-Bermúdez, P., Seasonal variation in nutrient status of foxglove leaves. *J. Plant Nutr.*, 2006, **29**, 1077–1084.
27. Singh, J. S. and Singh, V. K., Phenology of seasonally dry tropical forest. *Curr. Sci.*, 1992, **63**, 684–688.
28. Pandey, C. B. and Singh, J. S., Rainfall and grazing effects on net primary productivity in a tropical savanna. *India. Ecology*, 1992, **73**, 2007–2021.
29. Raghubanshi, A. S., Effect of topography on selected soil properties and nitrogen mineralization in a dry tropical forest. *Soil Biol. Biochem.*, 1992, **24**, 145–150.
30. Singh, A. K., Raghubanshi, A. S. and Singh, J. S., Medical ethnobotany of the tribals of Sonaghati of Sonebhadra District, Uttar Pradesh, India. *J. Ethnopharmacol.*, 2002, **81**, 31–41.
31. U.S. Salinity Laboratory Staff, Handbook 60, U.S. Government Printing Office, Washington, DC, 1954.
32. Sheldrick, B. H. and Wang, C., Particle-size analysis. In *Soil Sampling and Methods of Analysis* (ed. Carter, M. R.), Lewis Publ., Boca Raton, FL, 1993, pp. 499–517.
33. Walkley, A. and Black, J. A., An examination of the Degtjareff method for determining soil organic matter and a proposed modification of the chromic titration method. *Soil Sci.*, 1934, **37**, 29–38.
34. Bremner, J. M. and Mulvaney, C. S., Nitrogen – Total. In *Methods of Soil Analysis: Part 2. Chemical and Microbiological Properties* (eds Page, A. L. *et al.*), Agronomy Monograph 9, ASA-SSSA Madison, WI, 1982, 2nd edn, pp. 595–624.
35. Olsen, S. R. and Sommers, L. E., Phosphorus. In *Methods of Soil Analysis: Part 2. Chemical and Microbiological Properties* (eds Page, A. L. *et al.*), Agronomy Monograph 9, ASA-SSSA Madison, WI, 1982, 2nd edn, pp. 403–430.
36. Bradstreet, R. B., *The Kjeldahl Method for Organic Nitrogen*, Academic Press, New York, 1965, p. 239.
37. Anderson, J. M. and Ingram, J. S. I., In *Tropical Soil Biology and Fertility: A Handbook of Methods of Analysis*, CAB International, Wallingford, UK, 1989, 1st edn.
38. Arnon, D. I., Copper enzymes in isolated chloroplasts: polyphenol oxidases in *Beta vulgaris*. *Plant Physiol.*, 1949, **24**, 1–15.
39. SPSS/PC, SPSS/PC for the IBM PC/XT/AT, Chicago, IL, 2004.
40. Chapin III, F. S. and Kedrowski, R. A., Seasonal changes in nitrogen and phosphorus fractions in autumn retranslocation in evergreen and deciduous taiga trees. *Ecology*, 1983, **64**, 376–391.

RESEARCH ARTICLES

41. Chapin III, F. S., The mineral nutrition of wild plants. *Annu. Rev. Ecol. Syst.*, 1982, **11**, 233–260.
42. Aerts, R., Aboveground biomass and nutrient dynamics of *Calluna vulgaris* and *Molinia caerulea* in a dry heathland. *Oikos*, 1989, **6**, 31–38.
43. Peoples, M. B. and Dalling, M. J., The interplay between proteolysis and amino acid metabolism during senescence and nitrogen reallocation. In *Senescence and Aging in Plants* (eds Nooden, L. D. and Leopold, A. C.), Academic Press, California, USA, 1988, pp. 181–217.
44. Mae, T., Leaf senescence and nitrogen metabolism. In *Plant Cell Death Processes* (ed. Nooden, L. D.), Elsevier Academic Press, CA, USA, 2004, pp 157–168.
45. Eckstein, R. L., Karlsson, P. S. and Weih, M., Leaf lifespan and nutrient resorption as determinants of plant nutrient conservation in temperate–arctic regions. *New Phytol.*, 1999, **143**, 177–189.
46. Boerner, R. E. J., Seasonal nutrient dynamics, nutrient resorption and mycorrhizal infection intensity of two perennial forest herbs. *Am. J. Bot.*, 1986, **73**, 1249–1257.
47. Ono, K., Terashima, I. and Watanabe, A., Interaction between nitrogen deficit of a plant and nitrogen content in the old leaves. *Plant Cell Physiol.*, 1996, **37**, 1083–1089.
48. De Mars, B. G. and Baerner, R. E. J., Foliar phosphorus and nitrogen resorption in three woodland herbs of contrasting phenology. *Castanea*, 1997, **62**, 43–54.
49. Mishra, M. K. and Mishra, B. N., Seasonal changes in leaf area index and chlorophyll in an Indian grassland. *J. Ecol.*, 1981, **69**, 797–805.
50. Sobrado, M. A. and Medina, E., General morphology, anatomical structure, and nutrient content of sclerophyllous leaves of the 'Bana' vegetation of Amazonas. *Oecologia*, 1980, **45**, 341–345.
51. Brown, R. and Wilson, H. J. R., Nitrogen response of *Panicum* species differing in CO₂ fixation pathways. II. CO₂ exchange characteristics. *Crop Sci.*, 1983, **23**, 1154–1159.
52. Kull, O. and Niinemets, Ü, Variations in leaf morphometry and nitrogen concentration in *Betula pendula* Roth., *Corylus avellana* L. and *Lonicera xylosteum* L., *Tree Physiol.*, 1993, **12**, 311–318.
53. Field, C. and Mooney, H. A., *On the Economy of Plant Form and Function* (ed. Givnish, T.), Cambridge University Press, Cambridge, UK, 1986, pp. 25–55.
54. Santiago, L. S. and Wright, S. J., Leaf functional traits of tropical forest plants in relation to growth form. *Funct. Ecol.*, 2007, **21**, 19–27.
55. Ojima, M., Improvement of photosynthetic capacity in soybean variety. *Jpn. Agric. Res. Q.*, 1974, **8**, 6–12.
56. Buttery, B. R. and Buzzell, R. I., The relationship between chlorophyll content and rate of photosynthesis in soybeans. *Can. J. Plant Sci.*, 1977, **57**, 1–5.
57. Schreiber, U., Bilger, W. and Neubauer, C., Chlorophyll fluorescence as a noninvasive indicator of rapid assessment of *in vivo* photosynthesis. In *Ecophysiology of Photosynthesis* (eds Schultze, E. O. and Caldwell, M. M.), Springer Verlag, New York, 1995, pp. 48–70.
58. Wright, I. J. *et al.*, Modulation of leaf economic traits and trait relationships by climate. *Global Ecol. Biogeogr.*, 2005, **14**, 411–421.
59. Ordoñez, J. C., Bodegom, P. M. V., Witte, J. P. M., Wright, I. J., Reich, P. B. and Aerts, R., A global study of relationships between leaf traits, climate and soil measures of nutrient fertility. *Global Ecol. Biogeogr.*, 2009, **18**, 137–149.

ACKNOWLEDGEMENTS. P.D. thanks the University Grant Commission, New Delhi, for providing financial assistance. J.S.S. is supported under the NASI Senior Scientist Scheme.

Received 26 February 2010; revised accepted 11 November 2010