Full Length Article

Variations in Leaf Morphological Traits of *Quercus guyavifolia* (Fagaceae) were Mainly Influenced by Water and Ultraviolet Irradiation at High Elevations on the Qinghai-Tibet Plateau, China

Mei Sun[1,2], Tao Su[1], Shi-Bao Zhang[3], Shu-Feng Li[1,2], Julie Anberree-Lebreton[1,2] and Zhe-Kun Zhou[1,3*]
[1]*Key Laboratory of Tropical Forest Ecology, Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences, Mengla, Yunnan 666303, China*
[2]*University of Chinese Academy of Sciences, Beijing 100049, China*
[3]*Kunming Institute of Botany, Chinese Academy of Sciences, Kunming, Yunnan 650204, China*
[*]*For correspondence: zhouzk@xtbg.ac.cn*

Abstract

Leaf morphological traits vary along elevational gradients so that plants can adapt to their surrounding habitat. However, the abiotic factors that shape those traits are still debatable, especially for plants at high elevations where the environment can be very harsh. *Quercus guyavifolia* H. Léveillé (Fagaceae) is a woody plant found along the southeastern boundary of the Qinghai-Tibet Plateau. Distribution elevations of the species range from 2000 m to 4500 m approximately, providing an excellent opportunity to explore correlations among leaf traits and environmental factors. We used multiple-regression models to investigate spatial trends in leaf morphology and their environmental determinants. As elevation increased, values for leaf area, length, width, the ratio of leaf length to width, shape factor, and petiole length and width decreased significantly and all were positively correlated with mean annual precipitation. Leaf length, the ratio of leaf length to width, and petiole length were negatively correlated with the daily mean maximum intensity of ultraviolet (UV)-B irradiation during the growing season. Our results indicated that the amounts of both precipitation and UV irradiation largely shape the leaf morphology in *Q. guyavifolia* along the elevation gradient. This study provides evidence for the adaptive plasticity of leaves in response to environments in regions at high elevations. © 2016 Friends Science Publishers

Keywords: Leaf morphology; Plasticity; Precipitation; Qinghai-Tibet Plateau; Quercus; UV-B

Introduction

The plasticity of leaf morphological traits across habitats has long been of interest to ecologists because these traits are considered good predictors of plant performance and adaptation (Díaz *et al*., 2004; Poorter and Bongers, 2006; Hernández-Calderón *et al*., 2014). Elevational clines represent a "natural laboratory" within which we can better understand how such traits fluctuate in response to growing conditions. This is because environmental factors change along an elevational gradient, even within a small geographical area in some cases (Hovenden and Brodribb, 2000; Thomas, 2011; Rezende *et al.*, 2015). Moreover, when one considers their close associations with environmental factors, these morphological traits can be used to predict the consequences of climate changes (Hudson *et al*., 2011; Tsonev *et al*., 2014).

The leaf morphological traits of many species vary with elevation (Cordell *et al*., 1998; Bresson *et al*., 2011; Guerin *et al*., 2012). Those variations are mainly shaped by fluctuations in environmental factors (Yin *et al*., 2004; Körner, 2007; Jump *et al*., 2009; Wang *et al*., 2014). For

example, the lower temperatures found at higher elevations can restrict the extension of leaves and reduce their size (Magnani and Borghetti, 1995). Stronger solar radiation at higher elevations may lead to lower leaf dry matter, smaller overall sizes, and shorter petioles (Niinemets *et al*., 2004; Pan *et al*., 2009). Greater radiation can also promote transpiration, although its effect may be buffered in smaller, shorter leaves (Givnish, 1987; Yang *et al*., 2014). Meanwhile, water availability is another important factor that influences leaf morphology (Givnish, 1987; Mahmood *et al.,* 2015). Under drought conditions, plants commonly produce smaller leaves and have a thinner vapor boundary layer so that evaporative water losses are decreased (Nicotra *et al*., 2008). Although the effects of environment and elevation on leaf traits have already been examined, most of the research has focused on regions at lower elevations, i.e. below 3000 m, and the abiotic factors that shape those traits are still debatable. However, since plants in higher elevations are more sensitive in the face of environmental changes, studies at these regions are crucial if we are to further investigate the adaptation of leaf morphology to harsh environments (Wang *et al*., 2014).
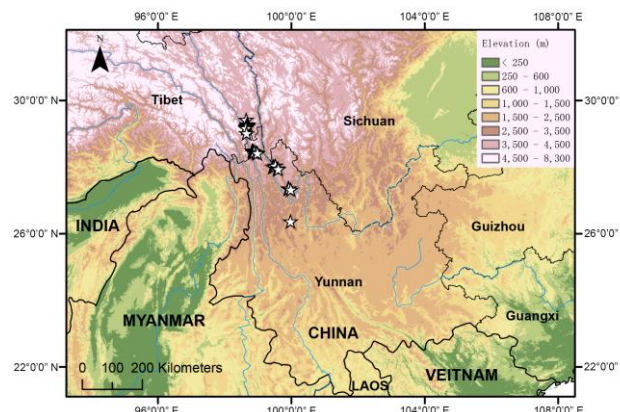
The southeastern boundary of the Qinghai-Tibet Plateau (QTP) refers to a mountainous region in East Tibet, West Sichuan, and Northwest Yunnan in China. This region is ideal for studying the plasticity of leaf structure and function because of its larger elevational gradients (average height >4000 m) and environmental variations. *Quercus guyavifolia* H. Léveillé is widely distributed along southesastern QTP. With a preferred elevation range between 2000 m and 4500 m, this species is distributed more widely than most other broad-leaved species (Zhou *et al.*, 2003, 2007; Su, 2010). It is also one of the dominant and established tree species in this region (Wu, 1980), existing mainly in evergreen broad-leaved forests. However, studies on morphology of this species are still rare. From these studies, leaf traits of this species (e.g., overall dimensions, physiological characters, and nutrient contents) vary extensively in alpine regions (Zhang *et al.*, 2005, 2007). Although previous findings have improved our understanding of how *Q. guyavifolia* adapt to environmental changes along elevations, no reports have described these trait variations along an entire elevation range of the distribution. Moreover, details about potential correlations between elevation and traits, such as leaf shape and petiole structure are still lacking, even though these data can reflect the light-interception and investments in leaf biomass (Takenaka, 1994; Niinemets *et al.*, 2004; Weijschedé *et al.*, 2006).

In this study, we collected leaves of *Q. guyavifolia* at 29 high-elevation sites, and applied multiple-regression models to explore: (1) how leaf morphological traits change with increasing elevation; (2) the key environmental factor(s) that contribute to those variations; and (3) the ecological implications of altered leaf morphology for plant adaptability.

Materials and Methods

Leaves from *Q. guyavifolia* trees (3–6 m tall) were collected in June 2012 from 29 sampling sites with minimal human activity along the southeastern boundary of the QTP in southwestern China (Fig. 1). Elevations at these sites range from 2343 m to 4242 m. At each site, four trees were randomly selected, and a single branch with more than 50 sun leaves was collected from each tree. Because *Q. guyavifolia* is an evergreen species, we chose only mature leaves and excluded any young shoots. Branches were labeled and sealed separately in plastic bags prior to measurements.

For each site, the mean annual temperature (MAT) and mean annual precipitation (MAP) were extracted from a global gridded climate dataset (precision: 0.16º × 0.16º; http://www.paleo.bris.ac.uk/). The daily maximum ultraviolet-B irradiation (280–315 nm) in 2011 was obtained from a TUV (Tropospheric Ultraviolet and Visible) radiation model (http://cprm.acd.ucar.edu/Models/TUV/Interactive_TUV/).
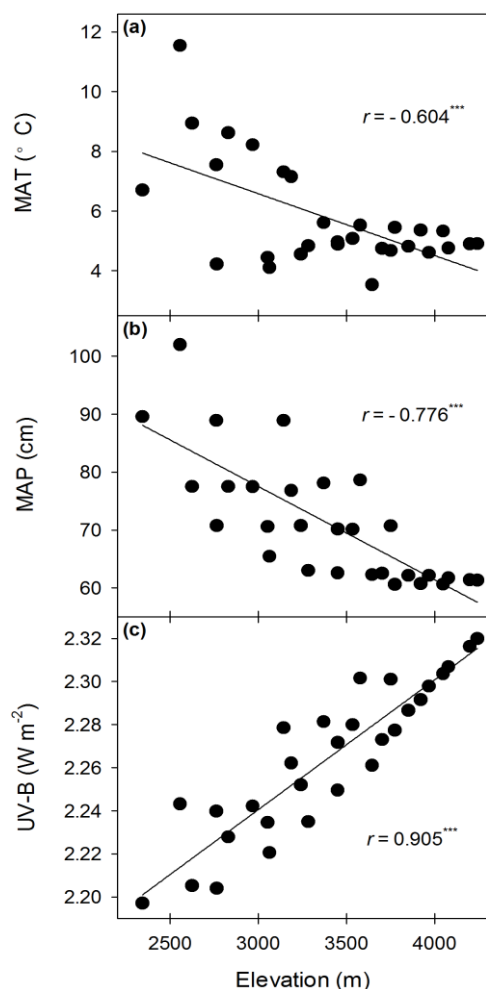

Fig. 1: Sampling sites (white stars) for *Quercus guyavifolia* along southeastern boundary of the Qinghai-Tibet Plateau

UV-B was calculated as the daily mean maximum ultraviolet-B intensity during the growing season from March to October. Across our sampling sites, the elevational changes of MAT and MAP were significantly decreased, while UV-B increased remarkably with increasing elevations (Fig. 2).

Leaf traits were scored in the Paleoecology Laboratory of Xishuangbanna Tropical Botanical Garden, the Chinese Academy of Sciences. Thirty mature, undamaged leaves were randomly collected from each sampled branch. These leaves were conserved in FAA (formalin, glacial acetic acid, ethanol, and distilled water; 10:5:50:35; v:v:v:v) for 48 h, then photographed by an HP Scanjet G4050 digital scanner (Chongqing, China). For each leaf, six leaf morphological characters were measured with ImageJ v. 1.48 (Wayne Rasband, National Institutes of Health, USA; available in the website: http://rsb.info.nih.gov/ij/), including leaf area (LA; mm^2), leaf perimeter (LP), leaf length (LL; mm), leaf width (LW; mm), petiole length (PL; mm), and petiole width (PW; mm). To quantify leaf shape, we determined the ratio of leaf length to width (LL/LW) and shape factor (SF, leaf perimeter^2/leaf area), where the latter represents the relative amount of leaf edge independent of size (Sack *et al.*, 2003).

Prior to our data analyses, we checked the assumption of normality and homoscedasticity of the observed morphological traits. Elevational variations in studied leaf morphological traits were then explored with Pearson-bivariate correlations. Principle Component Analysis (PCA) was also performed to further explore the variations of leaf morphological traits among the sample sites. Because it was possible for environmental factors to be correlated with each other, we used multiple-regression models with the 'stepwise' option to minimize the co-correlation impact. All analyses were performed with R statistical program v. 3.01 (R Development Core Team, Vienna, Austria; available in the website: http://ftp.ctex.org/mirrors/CRAN/).
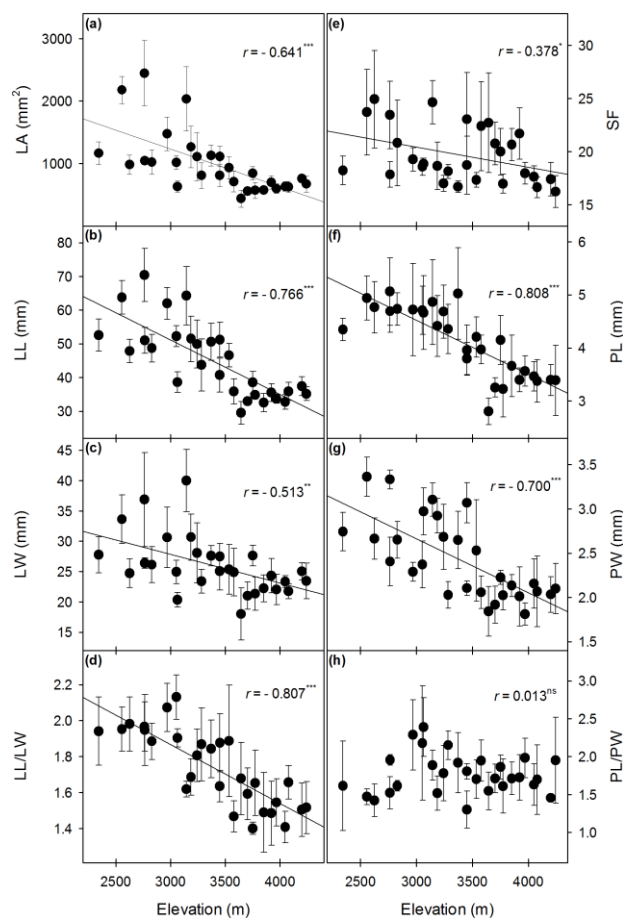
Fig. 2: Variations of mean annual temperature (MAT), mean annual precipitation (MAP), and the daily mean maximum ultraviolet-B intensity during the growing season (UV-B) along elevations in southeastern boundary of the Qinghai-Tibet Plateau. ***, *p*<0.001

Fig. 3: Elevation-associated variations in leaf morphological traits (mean values) for *Quercus guyavifolia* across 29 collection sites. LA, leaf area; LL, leaf length; LW, leaf width; LL/LW, the ratio of leaf length to width; SF, shape factor; PL, petiole length; PW, petiole width; PL/PW, the ratio of petiole length to width. Vertical error bars denote standard deviation. *, *p*<0.05; **, *p*<0.01; ***, *p*<0.001

Results

All studied leaf morphological traits varied noticeably among 29 sampling sites (Table 1). Among the eight traits investigated here, leaf area (LA) showed the widest variability, ranging from 438 to 2446 mm^2, and having the maximum coefficient of variation (CV=49.6%). By contrast, the ratio of leaf length to width (LL/LW) varied the least (CV=12.2%), with a range of 1.40 to 2.13 (Table 1).

With increasing elevation, LA, LL, LW, the LL/LW ratio, shape factor (SF), petiole length (PL), and petiole width (PW) were all decreased significantly; however, the PL/PW ratio was generally stable with elevation (Fig. 3).

In the Principle Component Analysis (PCA) for leaf traits values, the first two PCA axes explained for 60.68% and 18.65% of the total variation (Table 2; Fig. 4).

LA, LL, LW, LL/LW, SF, PL, and PW were significantly correlated with the first axis. Meanwhile, LL/LW, SF and PL/PW were significantly correlated with the second axis (Table 2; Fig. 4a). Sample sites were mainly loaded along the first PCA axis (Fig. 4b). Sites with low elevation were loaded on the positive side of the first axis, while the high elevation sites were loaded on the negative side (Fig. 4b). This implied that plants from lower altitude have higher leaf dimensions.

The results from stepwise-regression models showed that LA, SF, and PW were significantly correlated with MAP (respective *p*-values of 10^{-9}, 10^{-2} and 10^{-7}), whereas those models excluded MAT and UV-B as independents (Table 3). Both MAP and UV-B affected LL, LW, LL/LW, and PL (Table 3). LA, LL, LW, LL/LW, SF, PL, and PW were positively correlated with MAP (Fig. 5).

Table 1: Examined leaf morphological traits of *Quercus guyavifolia*

Variable	Abbreviation	Unit	Mean (Min.-Max.)	Coefficient of variation (CV, %)
Leaf area	LA	mm^2	993 (438-2446)	49.6
Leaf length	LL	mm	44.8 (29.5-70.4)	24.6
Leaf width	LW	mm	26.0 (18.0-40.0)	18.5
The ratio of leaf length to width	LL/LW	--	1.74 (1.40-2.13)	12.2
Shape factor	SF	--	19.7 (16.2-24.9)	13.3
Petiole length	PL	mm	4.12 (2.81-5.06)	15.9
Petiole width	PW	mm	2.42 (1.81-3.36)	18.8
The ratio of petiole length to width	PL/PW	--	1.77 (1.30-2.39)	15.0

Table 2: Correlations (*r*) of eight leaf morphological traits with the first two axes of principle component analysis (PCA) in 29 collecting sites

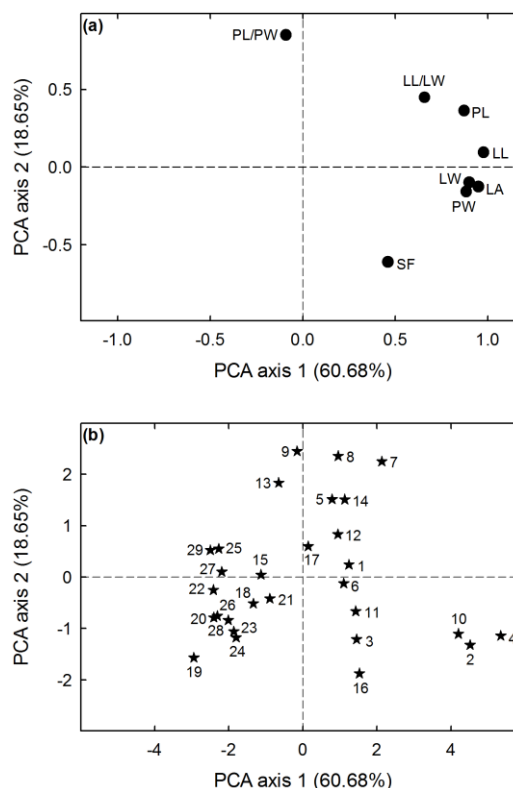| Leaf morphological traits	PCA axis 1		PCA axis 2	
	r	*p*	*r*	*p*
Leaf area	0.949***	0.000	-0.126	0.516
Leaf length	0.976***	0.000	0.096	0.621
Leaf width	0.883***	0.000	-0.158	0.414
The ratio of leaf length to width	0.658***	0.000	0.450*	0.014
Shape factor	0.460*	0.012	-0.611***	0.000
Petiole length	0.871***	0.000	0.364	0.053
Petiole width	0.899***	0.000	-0.098	0.612
The ratio of petiole length to width	-0.092	0.636	0.852***	0.000

Significant correlations are shown in boldface. *, *p*<0.05; ***, *p*<0.001

By contrast, LL, LL/LW and PL were negatively correlated with UV-B (Fig. 5).

Discussion

We found that leaf morphological traits of *Q. guyavifolia* varied significantly across our 29 study sites (Table 1). This indicated that those traits had significantly adaptive plasticity under different environmental conditions (Givnish *et al*., 2004; Dunbar-Co *et al*., 2009; Sun *et al*., 2014), which could potentially facilitate the high adaptive ability of *Q. guyavifolia* along high elevations.

In mountainous regions, leaf traits within a single species usually exhibit morphological variations along an elevational gradient (Bresson *et al*., 2011). *Q. guyavifolia* is distributed across a wide elevation range, and leaves in higher elevations exhibited lower leaf area, length, and width; lower shape factor and ratio of leaf length to width; and lower petiole length and width (Figs. 3–4; Table 2). Moreover, this elevational range from 2343 to 4242 m represents much higher altitudes than most previous studies of this kind (e.g., Cordell *et al*., 1998; Bresson *et al*., 2011; Guerin *et al*., 2012; Li and Bao, 2014). Those earlier investigations tended to focus primarily on the leaf size, which (consistent with our findings) is reduced at higher elevations. For example, the average leaf area from samples of *Metrosideros polymorpha* Gaudichaud (Myrtaceae), a dominant Hawaiian tree, decreases significantly with


Fig. 4: Principle component analysis (PCA) for eight leaf morphological traits of *Q. guyavifolia* in 29 collecting sites. Loading of sampling sites with elevations from low to high (numbers from 1 to 29) along PCA axis are presented in Panel (b). LA, leaf area (mm^2); LL, leaf length (mm); LW, leaf width (mm); LL/LW, the ratio of leaf length to width; SF, shape factor; PL, petiole length (mm); PW, petiole width (mm); PL/PW, the ratio of petiole length to width

elevations from 107 to 2469 m (Cordell *et al*., 1998); leaf size for both *Fagus sylvatica* Linnaeus and *Q. petraea* (Mattuschka) Lieblein are greatly reduced between 100 and 1600 m (Bresson *et al*., 2011); and the leaf area of *Dodonaea viscosa* Jacquin decreases as elevation increases from 300 to 800 m (Guerin *et al*., 2012). However, the opposite trend has also been reported by Li and Bao (2014) in a study with *Campylotropis polyantha* (Franchet) Schindler in southwest China. These observations demonstrate that patterns of leaf morphological plasticity can differ among species under various environmental backgrounds.

Abiotic conditions are the major factors that shape plant traits along elevational gradients (Hovenden and Brodribb, 2000; Körner, 2007; Thomas, 2011; Guerin *et al*., 2012; Rezende *et al*., 2015). Our results demonstrate that leaf area, shape factor, and petiole width are mainly influenced by MAP, whereas variations in leaf length, width, the ratio of leaf length to width, and petiole length are due to elevation-dependent fluctuations in both MAP and UV-B (Table 3). Among our collection sites,
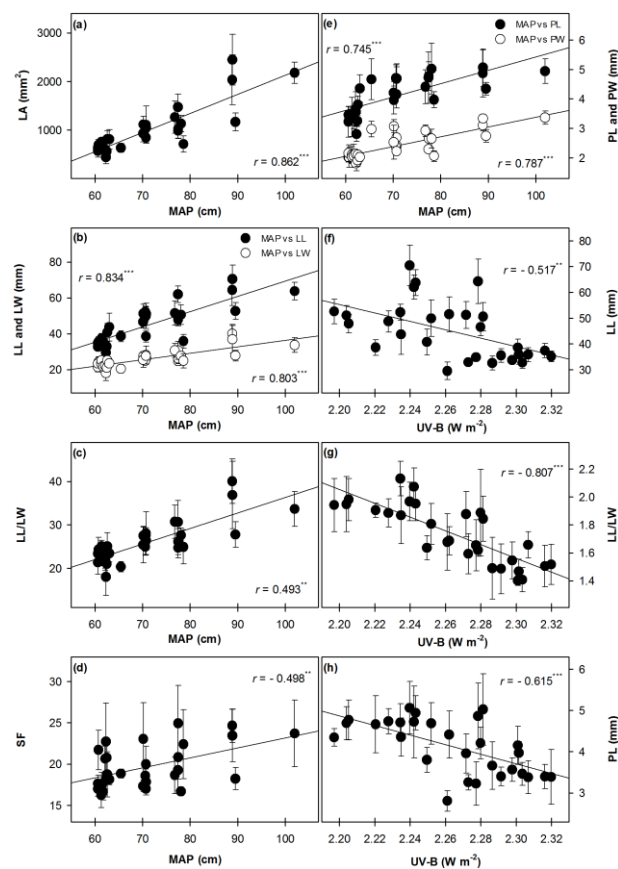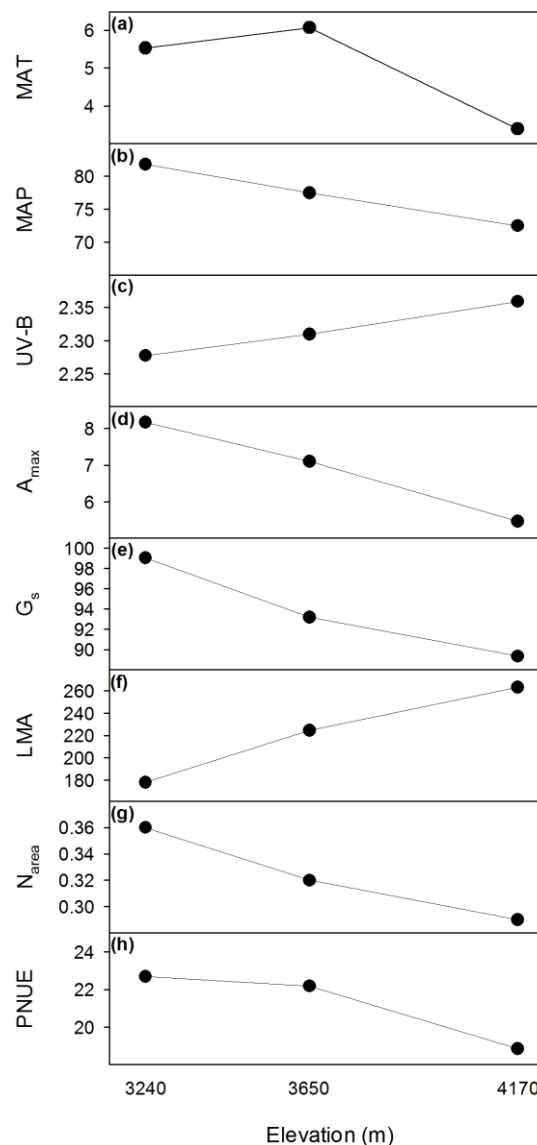
Fig. 5: Significant correlations among leaf morphological traits (site mean values) and environmental factors. LA, leaf area; LL, leaf length; LW, leaf width; LL/LW, the ratio of leaf length to width; SF, shape factor; PL, petiole length; PW, petiole width; PL/PW, the ratio of petiole length to width; MAP, mean annual precipitation; UV-B, the daily mean maximum ultraviolet-B intensity during the growing season. Vertical error bars denote standard deviation. **, $p<0.01$; ***, $p<0.001$

Fig. 6: Elevational variations of some environmental factors and physiological characters based on our data collection in conjunction with data from Zhang et al. (2005). We collected the environmental factors (MAT, MAP, UV-B) using longitude, latitude, and elevation. The light-saturated photosynthetic rate (A_{max}, µmol m^{-2} s^{-1}), stomatal conductance (G_s, mmol m^{-2} s^{-1}), leaf mass per area (LMA, g m^{-2}), and leaf nitrogen concentration (N_{area}, g m^{-2}) were extracted from Zhang et al. (2005). Based on these sets of measurements, leaf nitrogen use efficiency (PNUE, µmol g^{-1} s^{-1}) was calculated as A_{max} divided by N_{area}

MAP decreases as elevation increases (Fig. 2), suggesting that plants at higher locations may endure more serious water limitations because of reduced precipitation. Moreover, solar irradiation (usually proportional to UV-B irradiation) is more intense at higher elevations (Körner, 2007; Jump *et al.*, 2009; Thomas, 2011). This trend promotes transpiration losses, further inducing a water deficit (Hovenden and Brodribb, 2000; Guo *et al.*, 2013). Because MAP and UV-B proved to be the major factors determining the performance of leaf traits in our study (Table 3; Fig. 5), their fluctuations may reflect the enhanced capacity by plants to adapt to water deficits at higher elevations.

Consistent with our results, Givnish (1987) showed that water stress is a consequence of lower precipitation inputs and stronger solar radiation at high elevations, and that it is the main factor regulating leaf morphology.

Peppe *et al.* (2011) also implied that water is a key determinant of leaf size, whereas temperature is relatively less critical. However, because temperature declines significantly with increasing elevation (Fig. 2; Körner, 2007; Thomas, 2011), such chilling conditions can increase water viscosity and reduce enzymatic activity, thereby inducing

Table 3: Correlations among leaf morphological traits and environmental factors based on bivariate correlations and stepwise-regression models

Leaf traits	Bivariate correlations		Stepwise-regression models				Leaf traits	Bivariate correlations		Stepwise-regression models			
	Environment	r	Variable	Coefficient	R^2	p		Environment	r	Variable	Coefficient	R^2	p
LA	MAT	0.711***	MAP	39.3***	0.744	10^{-9}	SF	MAT	0.438*	MAP	0.121**	0.248	10^{-2}
	MAP	0.862***	Intercept	-1807***				MAP	0.498**	Intercept	11.1***		
	UV-B	-0.339						UV-B	-0.807***				
LL	MAT	0.695***	MAP	0.768***	0.723	10^{-8}	PL	MAT	0.598***	MAP	-1.32*	0.742	10^{-7}
	MAP	0.834***	UV-B	-58.1				MAP	0.745***	UV-B	-49.1**		
	UV-B	-0.517**	Intercept	121				UV-B	-0.615***	MAP:UV-B	0.601**		
										Intercept	112**		
LW	MAT	0.630***	MAP	0.401***	0.683	10^{-7}	PW	MAT	0.598***	MAP	-0.0331***	0.620	10^{-7}
	MAP	0.803***	UV-B	30.3				MAP	0.787***	Intercept	0.0668		
	UV-B	-0.180	Intercept	-71.1				UV-B	-0.462*				
LL/LW	MAT	0.453*	MAP	0.00352	0.672	10^{-7}	PL/PW	MAT	0.271	None			
	MAP	0.493**	UV-B	-0.0181***				MAP	0.169				
	UV-B	-0.807***	Intercept	11.5***				UV-B	-0.104				

LA, leaf area (mm²); LL, leaf length (mm); LW, leaf width (mm); LL/LW, the ratio of leaf length to width; SF, shape factor; PL, petiole length (mm); PW, petiole width (mm); PL/PW, the ratio of petiole length to width; MAT, mean annual temperature (°C); MAP, mean annual precipitation (cm); UV-B, the daily mean maximum ultraviolet-B intensity during the growing season (W m⁻²). *, $p<0.05$; **, $p<0.01$; ***, $p<0.001$

physiological water stress (Magnani and Borghetti, 1995; López-Bernal *et al.,* 2015). We also noted significant bivariate correlations between MAT and leaf morphological traits (Table 3). However, no such statistical correlations were found after stepwise-regression models were applied, suggesting that the effect of temperature on leaf morphology may not be as obvious as the influences of precipitation and UV-B irradiation.

The traits of leaf area, length, width, the ratio of leaf length to width, shape factor, petiole length, and petiole width were all positively correlated with MAP (Fig. 5). Because precipitation influences water availability, one might expect that plants at higher elevations (i.e., where MAP values are lower; Fig. 2) would experience more drought stress. To counteract that phenomenon, smaller leaves and reduced values of the ratio of leaf length to width, shape factor, petiole length, and petiole width can work to optimize the water balance between supply and loss within a plant.

Smaller leaves have thinner vapor boundary layers, which then support more rapid cooling and restrict evaporation and corresponding water losses (Nicotra *et al.*, 2008). Although leaf length, width, ratio of leaf length to width, shape factor, petiole length, and petiole width may change allometrically with leaf area, their decreases in parallel with lower MAP may also alleviate the effects of a water deficit (Dunbar-Co *et al.*, 2009; Tošković and Veličković, 2013). Water is transported from branches through conduits in the petiole and middle veins to the leaf lamina (Sack and Holbrook, 2006). Therefore, plants with shorter leaves (lower leaf length and ratio of leaf length to width) and petioles (smaller petiole length) will have less resistance to water flow and enhanced water supplies to the lamina (Noda *et al.*, 2004; Guo *et al.*, 2013). These types of adaptations may enable plants to reduce the damage of low

leaf hydraulic conductance, photosynthesis, and nutrient availability in high-elevation water-stressed regions, as shown in Zhang *et al.* (2005) where leaf assimilation rate, gas conductance, nitrogen content per leaf area, and nitrogen use efficiency all decreased with increasing elevation (Fig. 6). Additionally, a reduced petiole width may also lead to smaller vessel diameters in those petioles. In fact, larger vessels tend to have higher water conductance and transport rates, but are also more vulnerable to cavitation and deformation of leaf tissues (Sack and Frole, 2006; Zhang *et al*., 2014b). Here, we suspect that a trade-off exists between increased water conductance (shorter petiole length) and lower vessel vulnerability (thinner petiole width) at higher elevations. The consistence of the ratio of petiole length to width across elevations supports this trade-off (Fig. 3h; Fig. 4).

Leaf length, the ratio of leaf length to width, and petiole length were negatively correlated with UV-B (Fig. 5). These traits are thought to be associated with investments on leaf biomass (Niinemets *et al*., 2004; Weijschedé *et al*., 2006). Smaller leaf length, the ratio of leaf length to width, and petiole length values suggest either lower "construction costs" for petioles or the formation of more robust midribs that will support shorter petioles and leaf blades (Niinemets *et al*., 2004). Strong UV-B irradiation impedes the leaf biomass investment at high elevations, resulting in shorter petioles and leaf blades (Pan *et al*., 2009). Both a broad blade (low leaf length and ratio of leaf length to width) and a short petiole (small petiole length) contribute to reduced light capture per unit leaf area, because of the increase in aggregate leaf area (more self-shading) around the stem (Takenaka, 1994). Intense UV-B irradiation impedes CO_2 uptake and can signal a light-responsive microenvironment within leaves, resulting in greater suppression of photosynthesis or photoinhibition

(Teramura and Sullivan, 1994; Rozema *et al.*, 1997; Jansen *et al.*, 1998; Tang *et al.*, 2015). Therefore, at high elevations, plants with shorter petioles and leaves may incur less damage from intense UV-B radiation if self-shading increases and the efficiency of light capture is diminished (Shigesada and Okubo, 1981; Takenaka, 1994; Ackerly, 1999; Pearcy *et al.*, 2005).

The elevations at which we collected leaves of *Q. guyavifolia* were selected to represent the entire range of distribution for this species, which is larger than most for other evergreen broadleaf trees (Zhou *et al.*, 2007; Su, 2010). The significant variability in leaf traits reflects their plasticity, and it demonstrates their important roles in the adaptability of this species to alpine environments. During the past five decades, both temperature and precipitation have increased along the southeastern boundary of the Qinghai-Tibet Plateau (Fan *et al.*, 2008; Qin *et al.*, 2010; Zhang *et al.*, 2014a). Along with these climatic changes, the alpine timberline has also risen in this region (Moseley, 2006). As one of the dominant tree species there (Wu, 1980), *Q. guyavifolia* may continue to spread upward on those mountain slopes. Given their significant plasticity, the morphological traits examined here should contribute to the adaptations by these plants to much higher elevations. However, UV-B irradiation also becomes more intense with increasing elevation, such that a trade-off must exist between the benefit of increased temperature/precipitation and the negative influence of intensive UV-B irradiation.

Conclusion

Leaf area, length, width, the ratio of leaf length to width, shape factor, petiole length, and petiole width decreased significantly for *Q. guyavifolia* as sampling moved upward along our elevational gradient. All these seven traits were positively correlated with mean annual precipitation, but leaf length, the ratio of leaf length to width, and petiole length were also negatively correlated with the daily mean maximum UV-B intensity during the growing season. These significant elevation-related variations may have contributed to the successful adaptation of *Q. guyavifolia* on the boundary of the Qinghai-Tibet Plateau.

Acknowledgements

References

Ackerly, D., 1999. Self-shading, carbon gain and leaf dynamics: a test of alternative optimality models. *Oecologia*, 119: 300–310

Bresson, C.C., Y. Vitasse, A. Kremer and S. Delzon, 2011. To what extent is altitudinal variation of functional traits driven by genetic adaptation in European oak and beech? *Tree Physiol.*, 31: 1164–1174

Cordell, S., G. Goldstein, D. Mueller-Dombois, D. Webb and P.M. Vitousek, 1998. Physiological and morphological variation in *Metrosideros polymorpha*, a dominant Hawaiian tree species, along an altitudinal gradient: the role of phenotypic plasticity. *Oecologia*, 113: 188–196

Díaz, S., J.G. Hodgson, K. Thompson, M. Cabido, J.H.C. Cornelissen, A. Jalili and M.R. Zak, 2004. The plant traits that drive ecosystems: evidence from three continents. *J. Veg. Sci.*, 15: 295–304

Dunbar-Co, S., M.J. Sporck and L. Sack, 2009. Leaf trait diversification and design in seven rare taxa of the Hawaiian *Plantago* radiation. *Int. J. Plant Sci.*, 170: 61–75

Fan, Z.X., A. Bräuning and K.F. Cao, 2008. Annual temperature reconstruction in the central Hengduan Mountains, China, as deduced from tree rings. *Dendrochronologia*, 26: 97–107

Givnish, T.J., 1987. Comparative studies of leaf form: assessing the relative roles of selective pressures and phylogenetic constraints. *New Phytol.*, 106: 131–160

Givnish, T.J., K.C. Millam, T.M. Evans, J.C. Hall, J.C. Pires, P.E. Berry and K.J. Sytsma, 2004. Ancient vicariance or recent long-distance dispersal? Inferences about phylogeny and South American-African disjunctions in Rapateaceae and Bromeliaceae based on *ndh*F sequence data. *Int. J. Plant Sci.*, 165: S35–S54

Guerin, G.R., H. Wen and A.J. Lowe, 2012. Leaf morphology shift linked to climate change. *Biol. Lett.*, 8: 882–886

Guo, X., W.H. Guo, Y.J. Luo, X.F. Tan, N. Du and R.Q. Wang, 2013. Morphological and biomass characteristic acclimation of trident maple (*Acer buergerianum* Miq.) in response to light and water stress. *Acta Physiol. Plant.*, 35: 1149–1159

Hernández-Calderón, E., R. Méndez-Alonzo, J. Martínez-Cruz, A. González-Rodríguez and K. Oyama, 2014. Altitudinal changes in tree leaf and stem functional diversity in a semi-tropical mountain. *J. Veg. Sci.*, 25: 955–966

Hovenden, M.J. and T. Brodribb, 2000. Altitude of origin influences stomatal conductance and therefore maximum assimilation rate in Southern Beech, *Nothofagus cunninghamii*. *Funct. Plant Biol.*, 27: 451–456

Hudson, J.M.G., G.H.R. Henry and W.K. Cornwell, 2011. Taller and larger: shifts in Arctic tundra leaf traits after 16 years of experimental warming. *Global Change Biol.*, 17: 1013–1021

Jansen, M.A., V. Gaba and B.M. Greenberg, 1998. Higher plants and UV-B radiation: balancing damage, repair and acclimation. *Trends Plant Sci.*, 3: 131–135

Jump, A.S., C. Mátyás and J. Peñuelas, 2009. The altitude-for-latitude disparity in the range retractions of woody species. *Trends Ecol. Evol.*, 24: 694–701

Körner, C., 2007. The use of 'altitude' in ecological research. *Trends Ecol. Evol.*, 22: 569–574

Li, F.L. and W.K. Bao, 2014. Elevational trends in leaf size of *Campylotropis polyantha* in the arid Minjiang River valley, SW China. *J Arid Environ.*, 108: 1–9

López-Bernal, Á., O. García-Tejera, L. Testi, F. Orgaz and F.J. Villalobos, 2015. Low winter temperatures induce a disturbance of water relations in field olive trees. *Trees-Struct. Funct.*, 29: 1247–1257

Magnani, F. and M. Borghetti, 1995. Interpretation of seasonal changes of xylem embolism and plant hydraulic resistance in *Fagus sylvatica*. *Plant Cell Environ.*, 18: 689–696

Mahmood, T., A. Saeed, A.R. Saleem, H. Rasool, M. Haworth and M. Centritto, 2015. Divergent gas-exchange, physiological, isotopic and compositional responses of two wood-crop species to water deficit: *Ziziphus nummularia* and *Corymbia citriodora*. *Int. J. Agric. Biol.*, 17: 681–690

Moseley, R.K., 2006. Historical landscape change in northwestern Yunnan, China. *Mountain Res. Dev.*, 26: 214–219

Nicotra, A.B., M.J. Cosgrove, A. Cowling, C.D. Schlichting and C.S. Jones, 2008. Leaf shape linked to photosynthetic rates and temperature optima in South African *Pelargonium* species. *Oecologia*, 154: 625–635

Niinemets, Ü., N. Al Afas, A. Cescatti, A. Pellis and R. Ceulemans, 2004. Petiole length and biomass investment in support modify light interception efficiency in dense poplar plantations. *Tree Physiol.*, 24: 141–154

Noda, H., H. Muraoka and I. Washitani, 2004. Morphological and physiological acclimation responses to contrasting light and water regimes in *Primula sieboldii*. *Ecol. Res.*, 19: 331–340

Pan, H.L., M.H. Li, X.H. Cai, J. Wu, Z. Du and X.L. Liu, 2009. Responses of growth and ecophysiology of plants to altitude. *Ecol. Environ. Sci.*, 18: 722–730

Pearcy, R.W., H. Muraoka and F. Valladares, 2005. Crown architecture in sun and shade environments: assessing function and trade-offs with a three-dimensional simulation model. *New Phytol.*, 166: 791–800

Peppe, D.J., D.L. Royer, B. Cariglino, S.Y. Oliver, S. Newman, E. Leight and I.J. Wright, 2011. Sensitivity of leaf size and shape to climate: global patterns and paleoclimatic applications. *New Phytol.*, 190: 724–739

Poorter, L. and F. Bongers, 2006. Leaf traits are good predictors of plant performance across 53 rain forest species. *Ecology*, 87: 1733–1743

Qin, N.X., X. Chen, G.B. Fu, J.Q. Zhai and X.W. Xue, 2010. Precipitation and temperature trends for the Southwest China: 1960–2007. *Hydrol. Process.*, 24: 3733–3744

Rezende, V.L., P.V. Eisenlohr, A.C. Vibrans and A.T. de Oliveira-Filho, 2015. Humidity, low temperature extremes, and space influence floristic variation across an insightful gradient in the Subtropical Atlantic Forest. *Plant Ecol.*, 216: 759–774

Rozema, J., J. van de Staaij, L.O. Björn and M. Caldwell, 1997. UV-B as an environmental factor in plant life: stress and regulation. *Trends Ecol. Evol.*, 12: 22–28

Sack, L. and K. Frole, 2006. Leaf structural diversity is related to hydraulic capacity in tropical rain forest trees. *Ecology*, 87: 483–491

Sack, L. and N.M. Holbrook, 2006. Leaf hydraulics. *Annu. Rev. Plant Biol.*, 57: 361–381

Sack, L., P.D. Cowan, N. Jaikumar and N.M. Holbrook, 2003. The 'hydrology' of leaves: co-ordination of structure and function in temperate woody species. *Plant Cell Environ.*, 26: 1343–1356

Shigesada, N. and A. Okubo, 1981. Analysis of the self-shading effect on algal vertical distribution in natural waters. *J. Math. Biol.*, 12: 311–326

Su, T., 2010. On the establishment of the leaf physiognomy-climate model and a study of the late Pliocene Yangjie Flora, Southwest China. *Ph.D. Dissertation*, pp: 26–28. Kunming Institute of Botany, Chinese Academy of Sciences

Sun, M., S.J. Yang, J.L. Zhang, M. Bartlett and S.B. Zhang, 2014. Correlated evolution in traits influencing leaf water balance in *Dendrobium* (Orchidaceae). *Plant Ecol.*, 215: 1255–1267

Takenaka, A., 1994. Effects of leaf blade narrowness and petiole length on the light capture efficiency of a shoot. *Ecol. Res.*, 9: 109–114

Tang, H., Y.Y. Hu, W.W. Yu, L.L. Song and J.S. Wu, 2015. Growth, photosynthetic and physiological responses of *Torreya grandis* seedlings to varied light environments. *Trees Str. Funct.*, 29: 1011–1022

Teramura, A.H. and J.H. Sullivan, 1994. Effects of UV-B radiation on photosynthesis and growth of terrestrial plants. *Photosynth. Res.*, 39: 463–473

Thomas, S.C., 2011. Genetic vs. phenotypic responses of trees to altitude. *Tree Physiol.*, 31: 1161–1163

Tošković, D. and M. Veličković, 2013. Comparison of allometric relationships and morphological differences in *Tilia cordata* Mill. outer leaves exposed to different environmental conditions. *Plant Biosyst.*, 147: 611–619

Tsonev, T., S. Wahbi, P. Sun, G. Sorrentino and M. Centritto, 2014. Gas exchange, water relations and their relationships with photochemical reflectance index in *Quercus ilex* plants during water stress and recovery. *Int. J. Agric. Biol.*, 16: 335–341

Wang, X.F., R.Y. Li, X.Z. Li, F.J. Ma, B.N. Sun, J.Y. Wu and Y.K. Wang, 2014. Variations in leaf characteristics of three species of angiosperms with changing of altitude in Qilian Mountains and their inland high-altitude pattern. *Sci. Chin. Earth Sci.*, 57: 662–670

Weijschedé, J., J. Martínková, H. De Kroon and H. Huber, 2006. Shade avoidance in *Trifolium repens*: costs and benefits of plasticity in petiole length and leaf size. *New Phytol.*, 172: 655–666

Wu, Z.Y., 1980. *Vegetation in China*. Science Press, Beijing, China

Yang, S.J., M. Sun, Y.J. Zhang, H. Cochard and K.F. Cao, 2014. Strong leaf morphological, anatomical, and physiological responses of a subtropical woody bamboo (*Sinarundinaria nitida*) to contrasting light environments. *Plant Ecol.*, 215: 97–109

Yin, C.Y., B.L. Duan, X. Wang and C.Y. Li, 2004. Morphological and physiological responses of two contrasting poplar species to drought stress and exogenous abscisic acid application. *Plant Sci.*, 167: 1091–1097

Zhang, K.X., S.M. Pan, L.G. Cao, Y. Wang, Y.F. Zhao and W. Zhang, 2014a. Spatial distribution and temporal trends in precipitation extremes over the Hengduan Mountains region, China, from 1961 to 2012. *Quatern. Int.*, 349: 346–356

Zhang, S.B., Z.K. Zhou, H. Hu, K. Xu, N. Yan and S.Y. Li, 2005. Photosynthetic performances of *Quercus pannosa* vary with altitude in the Hengduan Mountains, southwest China. *For. Ecol. Manage.*, 212: 291–301

Zhang, S.B., Z.K. Zhou, H. Hu and K. Xu, 2007. Gas exchange and resource utilization in two alpine oaks at different altitudes in the Hengduan Mountains. *Can. J. For. Res.*, 37: 1184–1193

Zhang, Y.J., F.E. Rockwell, J.K. Wheeler and N.M. Holbrook, 2014b. Reversible deformation of transfusion tracheids in *Taxus baccata* is associated with a reversible decrease in leaf hydraulic conductance. *Plant Physiol.*, 165: 1557–1565

Zhou, Z.K., C.X. Pu and W.Y. Chen, 2003. Relationships of the distributions of *Quercus* sect. Heterobalanus (Fagaceae) and uplift of Himalayas. *Adv. Earth Sci.*, 18: 884–890

Zhou, Z.K., Q.S. Yang and K. Xia, 2007. Fossils of *Quercus* sect. *Heterobalanus* can help explain the uplift of the Himalayas. *Chin. Sci. Bull.*, 52: 238–247