J Plant Res (2015) 128:573–584
DOI 10.1007/s10265-015-0721-z

REGULAR PAPER

Water relations and gas exchange of fan bryophytes and their adaptations to microhabitats in an Asian subtropical montane cloud forest

Liang Song[1] · Yong-Jiang Zhang[2] · Xi Chen[1,3] · Su Li[1] · Hua-Zheng Lu[1,3] · Chuan-Sheng Wu[1,3,4] · Zheng-Hong Tan[1] · Wen-Yao Liu[1] · Xian-Meng Shi[1,3]

Received: 9 July 2014 / Accepted: 3 February 2015 / Published online: 27 March 2015

Abstract Fan life forms are bryophytes with shoots rising from vertical substratum that branch repeatedly in the horizontal plane to form flattened photosynthetic surfaces, which are well suited for intercepting water from moving air. However, detailed water relations, gas exchange characteristics of fan bryophytes and their adaptations to particular microhabitats remain poorly understood. In this study, we measured and analyzed microclimatic data, as well as water release curves, pressure–volume relationships and photosynthetic water and light response curves for three common fan bryophytes in an Asian subtropical montane cloud forest (SMCF). Results demonstrate high relative humidity but low light levels and temperatures in the understory, and a strong effect of fog on water availability for bryophytes in the SMCF. The facts that fan bryophytes in dry air lose most of their free water within 1 h, and a strong dependence of net photosynthesis rates on water content, imply that the transition from a hydrated, photosynthetically active state to a dry, inactive state is rapid. In addition, fan bryophytes developed relatively high cell wall elasticity and the osmoregulatory capacity to tolerate desiccation. These fan bryophytes had low light saturation and compensation point of photosynthesis, indicating shade tolerance. It is likely that fan bryophytes can flourish on tree trunks in the SMCF because of substantial annual precipitation, average relative humidity, and frequent and persistent fog, which can provide continual water sources for them to intercept. Nevertheless, the low water retention capacity and strong dependence of net photosynthesis on water content of fan bryophytes indicate a high risk of unbalanced carbon budget if the frequency and severity of drought increase in the future as predicted.

Keywords Desiccation tolerance · Epiphyte · Fan life form · Photosynthesis · Poikilohydry · Water retention capacity

Introduction

Bryophytes are small plants with height ranged from one to a few millimeters up to a decimeter or more, with a few taxa of larger sizes. Most bryophytes have slender stems and unistratose leaves that lack a waxy cuticle (Miller 1982). As a successful group of nonvascular plants, bryophytes mostly avoid competition with tracheophytes by growing on hard, impenetrable substrata (e.g., tree bark and rocks), and tolerate frequent desiccation and nutrient-poor conditions (Bates 1998). Bryophytes are a typical group of poikilohydric plants; they usually dry out and suspend metabolism rapidly under dry conditions, but recover activity quickly when rehydrated (Proctor 2001; Green et al. 2011). Poikilohydric plants show rapid equilibration of tissue water content with the surroundings and adaptation to intermittent metabolism, which are generally optimal characteristics for diminutive

✉ Liang Song
songliang@xtbg.ac.cn

✉ Wen-Yao Liu
liuwy@xtbg.ac.cn

[1] Key Laboratory of Tropical Forest Ecology, Xishuangbanna Tropical Botanical Garden, Chinese Academy of Sciences, No. 88 Xuefu Road, Kunming 650223, Yunnan, China

[2] Department of Organismic and Evolutionary Biology, Harvard University, Cambridge, MA 02138, USA

[3] University of Chinese Academy of Sciences, Beijing 100049, China

[4] Ailaoshan Station for Subtropical Forest Ecosystem Studies, Jingdong 676209, China

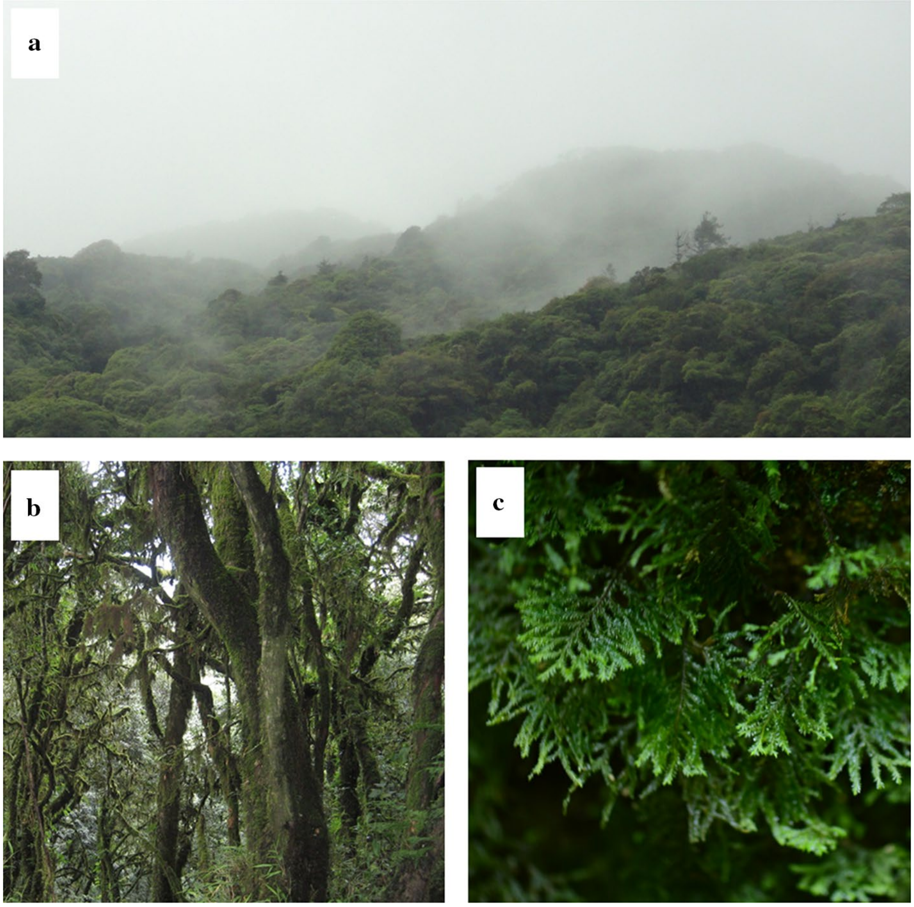

Fig. 1 Photos of study site and investigated species. **a** The subtropical montane cloud forest in the Ailao Mountains; **b** abundant epiphytic bryophytes in the understory; **c** fan bryophyte *Plagiochila assamica* Steph. (Photo credit: Liang Song)

plants such as bryophytes (Proctor 2001; Proctor et al. 2007; Green et al. 2011). In contrast, homoiohydric plants maintain high internal water potentials despite a low external water availability (Proctor and Tuba 2002).

Bryophyte life forms, which can be interpreted as recurring arrangements of photosynthetic tissues that minimize evaporative water loss and maximize primary production, are physiologically important for water storage and in determining boundary-layer diffusion resistance to water loss (Proctor 1990; Bates 1998; Kürschner et al. 1999). The life form is a useful concept for the ecophysiology of bryophyte, based on their poikilohydric nature and exceptionally strong dependence upon transient external water supplies (Bates 1998). There should be an ecophysiological basis for bryophyte life forms, but relevant data are very limited. Proctor (2002, 2004b) presented ecophysiological data on two tropical African pendulous epiphytic bryophytes and two Australasian pendulous species of *Weymouthia*. In another study, León-Vargas et al. (2006) addressed microclimate, light adaptation and desiccation tolerance of six bryophytes of pendulous growth form in two Venezuelan cloud forests. Zotz et al. (2000) found that water relations and carbon gain of moss *Grimmia pulvinata* (Hedw.) Sm. were related to cushion size. Fan life forms are bryophytes with shoots rising from a vertical substratum that branch repeatedly in the horizontal plane to form flattened photosynthetic surfaces, which are exposed on all sides to ambient air (Bates 1998; Ma et al. 2009). This type of bryophyte usually "stands" on tree trunk surfaces and has a very extensive surface area in contact with the air, and they are thus well suited for intercepting water from moving air (Bates 1998; Kürschner et al. 1999; Ma et al. 2009). However, to our knowledge, detailed water relations and gas exchange characteristics of fan bryophytes and their adaptations to those particular microhabitats are still poorly understood.

The subtropical montane cloud forest (hereafter SMCF) located in the Ailao Mountain National Nature Reserve of Southwest China, also known as montane moist evergreen broadleaf forest, is generally characterized by persistent, frequent cloud cover at the canopy level (Fig. 1a). The forest harbors plentiful epiphytic bryophytes on tree trunks (Fig. 1b) (Shi and Zhu 2009; Chen et al. 2010; Song et al. 2012a; Zhang et al. 2013). The epiphytic bryoflora of the SMCF is not only remarkable in its abundance and richness of species, but also notable for the diversity of life forms represented (Ma et al. 2009; Song et al. 2011). Song et al. (2015) recorded eight life forms in the SMCF of the Ailao

Mountains and 36.2 % of the bryophytes growing on tree trunks at ~1.5 m above the forest floor showed fan forms (Fig. 1c). Considering that a life form spectrum allows one to obtain a realistic impression of the habitat (Frahm and Ohlemüller 2001), the predominance of fan life forms in the bryophyte cover on low tree trunks could indicate suitable conditions for the survival and growth of fans. This phenomenon poses the following two ecological and physiological questions. First, what are the microclimatic conditions on low tree trunks? Second, which water relations and gas exchange characteristics of fan bryophytes help them adapt to low tree trunk microhabitats?

In the present study, we quantified microclimatic characteristics of fan bryophyte microhabitats, and studied water relations (water release curves, pressure–volume relationships) and photosynthetic water and light responses of three representative fan bryophytes in a SMCF of southwest China. We address the following questions: (1) what are the water and light conditions at locations occupied by these fan bryophytes, and how do these environmental conditions influence their water and carbon balances? (2) What are the water retention and desiccation tolerance capacities of these fan bryophytes? (3) How does carbon assimilation of fan bryophytes respond to changes in shoot water content (WC) and photosynthetically active radiation (PAR)? Information regarding water retention, desiccation tolerance, and dependence of carbon assimilation on water content will further elucidate the potential performance of fan bryophytes under a predicted drier climate in the study region (Hijmans et al. 2005; Ramirez and Jarvis 2008; Fan and Thomas 2013).

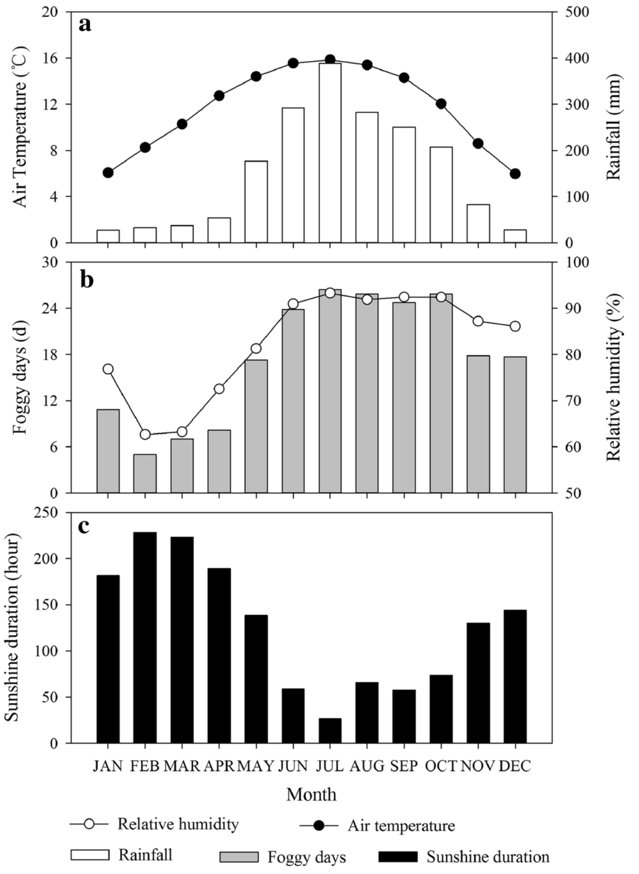

Fig. 2 **a** Mean monthly temperatures and rainfall, **b** foggy days and relative humidity, and **c** sunshine duration in the subtropical montane cloud forest during 2007–2013 (data from Ailaoshan Station for Subtropical Forest Ecosystem Studies, elevation 2480 m)

Materials and methods

Study site

We conducted the study in the Xujiaba region of Yunnan Province (24°32′N, 101°01′E), in a protected section of a pristine 5100-ha SMCF in the Ailao Mountain National Nature Reserve (Fig. 1a; 23°35′–24°44′N, 100°54′–101°01′E), with altitude between 2000 and 2600 m (Wu 1983). The study site is affected by both the southern subcurrent of the western airflow from India and Pakistan and the southwest monsoon, so it experiences a striking alternation of dry and wet conditions (Qiu and Xie 1998). Mean annual temperature of the study site is 11.6 °C, with minimum in December (6.0 °C) and maximum in July (15.8 °C) (Fig. 2a). Mean annual rainfall of the study site is 1859 mm. 86 % of the rainfall occur during the rainy season (May through October), resulting in a pronounced dry period from December through April (Fig. 2a). Mean relative humidity (RH) is 82.6 %, with the minimum in February (62.7 %) and maximum in July (93.3 %; Fig. 2b). Low-level cloud cover and fog are frequent (211 days per year), which compensates for a water shortage in dry months (Figs. 1a, 2b). Sunshine duration in the rainy season is much shorter than that in the dry season (Fig. 2c). The forest is primarily co-dominated by *Lithocarpus hancei* (Benth.) Rehder, *Castanopsis rufescens* (Hook.f.et Th.) Huang et Y.T.Chang, and *Lithocarpus xylocarpus* (Kurz) Markgr., among others (Wu 1983). The forest has four distinct strata, a canopy layer of trees (>20 m), a subcanopy layer of trees (<20 m), a shrub layer and an herb layer, interweaved with numerous lianas and epiphytes (Fig. 1b) (Qiu and Xie 1998). Detailed investigation of the vascular epiphytes and epiphytic bryophytes in this forest was carried out by Xu and Liu (2005) and Ma et al. (2009), and of the epiphytic lichens by Li et al. (2013).

Microclimatic measurements

On a triangular meteorological tower within the study area, two levels of instrument arms were mounted to measure microclimatic variables of the understory (1.5 m) and canopy (24 m). Instruments for measuring air temperature, RH (HMP45C; Vaisala Inc., Helsinki, Finland) and PAR (LQS70-10; Apogee Instruments, Inc., Logan, UT, USA) were installed at these heights, with a sampling interval of 30 min.

Measurements of water relations and gas exchange characteristics

One moss (*Homaliodendron flabellatum* (Sm.) Fleisch.) belonging to Neckeraceae and two liverworts [*Plagiochila arbuscula* (Brid. ex Lehm.) Lindenb. and *Plagiochila assamica* Steph.] belonging to Plagiochilaceae were selected for this study, because they are abundant and representative fan bryophytes and are easily sampled in the study area. All samples of *H. flabellatum*, *P. arbuscula*, and *P. assamica* were collected during the rainy season of 2012, from tree trunks at heights between 1.0 and 2.0 m above the forest floor. Samples were then transferred to the laboratory of the Xujiaba Ecological Field Station for subsequent measurements. After removing dirt and dead biomass, samples were stored outside in the shade for up to 1 week. One day prior to measurements, clean healthy shoots (terminal 1.5–2.5 cm of a stem, including the capitulum) were cut and wetted to ensure recovery of the metabolic processes. After being soaked in rainwater for 60 min so as to be fully hydrated, excess surface water was allowed to drip from the green healthy bryophyte shoots, and these were carefully blotted by placing the samples on blotting paper for 30 s.

Measurement of water release curves

As a first set, wet samples (one shoot each) were weighed using an electronic balance (AL204-IC; Mettler Toledo Instruments Ltd., Shanghai, China). Sample weights were recorded every 2 min during the first 60 min and at incremental intervals (5, 10, 30 min) over the next 260 min, until constant weights were obtained. After measurement, all samples were dried for 48 h at 70 °C to determine the dry weight (DW). Five replicates were measured for each species.

Measurement of pressure–volume curves

As a second set, wet samples (two shoots each) were placed in small polyethylene cups for ease of handling. Then, a succession of measurements of water potential at different WCs was made for each sample. Water potential was measured with the Dewpoint Water PotentiaMeter (WP4-T; Decagon Devices Inc., Pullman, WA, USA), and samples were weighed with an electronic balance (AL204-IC; Mettler Toledo Instruments Ltd., Shanghai, China). After measurement, all samples were dried for 48 h at 70 °C to determine the DW. Four replicates per species were measured.

Measurement of photosynthetic water and light response curves

As a third set, wet samples (three to four shoots each) were placed in a whole plant chamber (LI-6400-17; LI-COR, Lincoln, NE, USA) that connected to a portable infrared gas analyzer (LI-6400XT; LI-COR, Lincoln, NE, USA) to determine net photosynthetic rates (A_N). The chamber was illuminated by placing a full spectrum light source (LI-6400-18; LI-COR, Lincoln, NE, USA) at its top. For photosynthetic water response curves, a succession of measurements was made on each blotted sample until obtaining constant weight. PAR was set to 250 μmol m^{-2} s^{-1} (saturation for all species). After each weighing, the samples were allowed to lose up to a few milligrams of water before being resealed in the cuvette. Light response curves of bryophyte shoots were determined following 10 gradients of PAR: 200, 150, 125, 100, 80, 60, 40, 20, 10 and 0 μmol m^{-2} s^{-1}. To maintain relatively high and stable sample water content during the measurement, a piece of moist filter paper was placed in the bottom of the chamber. Empty chamber measurements with moist filter paper did not register significant CO_2 exchange signals, similar to other reports (Romero et al. 2006). The rate of CO_2 emission at zero PAR was assumed to be the respiration rate (R_d, μmol CO_2 m^{-2} s^{-1}). After all measurements were completed, samples were dried for 48 h at 70 °C to determine DW. All A_N is expressed relative to DW. Five replicates per species were measured.

Data analysis

Paired *t*-tests were conducted to compare microclimatic variables between the understory and forest canopy. Frequency distribution of RH and PAR during representative dry and rainy months (February and July, respectively) in 2013 were analyzed following León-Vargas et al. (2006). Here, we used microclimatic data from 2013 as a substitute for those in 2012 because the RH sensor at 1.5 m failed during the rainy season of the latter year, so RH data were not available then.

Evaporative water loss curves were fitted using exponential decay regressions:

$$\mathrm{WC} = y_0 + a \cdot e^{(-b \cdot t)}, \tag{1}$$

where t is evaporative time and a, b, y_0 are coefficients. Then, the initial ($t = 0$) WC of the samples can be calculated using ($y_0 + a$).

Relative water content (RWC, %) was calculated based on

$$\mathrm{RWC} = \frac{\mathrm{M} - \mathrm{DW}}{\mathrm{BW} - \mathrm{DW}}, \tag{2}$$

where M, BW, DW refer to real-time, blotted, and oven-dried weights, respectively.

Pressure–volume (P–V) curves were analyzed and the corresponding characteristics were calculated according to Beckett (1997). To estimate external capillary water (R_e) from a P–V curve, the water potential reciprocal ($-1/\Psi$, $-\mathrm{MPa}^{-1}$) was plotted as a function of 1 − RWC. The straight-line parts of the P–V curves below the turgor loss point (TLP) were fitted by linear regression:

$$-1/\psi = y_0 + \mathrm{a} \cdot (1 - \mathrm{RWC}). \tag{3}$$

Corresponding data points above the TLP were fitted by hyperbolic decay regression:

$$-1/\psi = y_0 + \frac{\mathrm{a} \cdot \mathrm{b}}{\mathrm{b} + 1 - \mathrm{RWC}}, \tag{4}$$

where Ψ is water potential and a, b, and y_0 are coefficients.

Turgor potential (Ψ_p, MPa) was calculated as the difference between the extrapolated linear portion of the curve and actual curve, and Ψ_p was then plotted as a function of RWC (Beckett 1997). In most of the cryptogams, Ψ_p did not decline with the initial loss of water. Water loss between 100 % RWC and RWC at which turgor began to decrease was assumed to be R_e (Beckett 1997). All RWC data were recalculated to exclude R_e. The P–V curves were then replotted and all parameters recalculated. RWC and osmotic potential at TLP ($\mathrm{RWC}_{\mathrm{TLP}}$, %; $\Psi_{\pi\mathrm{TLP}}$, MPa) were calculated according to the point of intersection of beeline and hyperbola (Fig. 3a). Water potential at saturated water content ($\Psi_{\pi s}$) was calculated by the negative reciprocal of intercept of the fitted linear model (Fig. 3a). Apoplastic water (R_a) was calculated from the x-intercept (Fig. 3a), and residual water is called symplast water (R_s). Ψ_p/RWC curves were fitted using exponential regression to calculate bulk elastic modulus (ε):

$$\psi_p = \mathrm{a} \cdot e^{\mathrm{b} \cdot \mathrm{RWC}}, \tag{5}$$

where RWC is the corrected relative water content and a and b are coefficients.

ε can generally increase continuously between the TLP and full turgor (Roberts et al. 1981; Tyree and Karamanos 1981), so it can only be defined precisely for a given Ψ_p. We took values of Ψ_p and ε from the fitted exponential models, calculating ε at an arbitrary Ψ_p of 1.0 MPa (Fig. 3b) following Beckett (1997).

The relationship between A_N and WC was fitted using logistic sigmoidal regression:

$$A_N = y_0 + \frac{\mathrm{a}}{1 + \left(\frac{\mathrm{WC}}{\mathrm{b}}\right)^{\mathrm{c}}}, \tag{6}$$

where A_N is net photosynthesis rate and a, b, c, y_0 are coefficients.

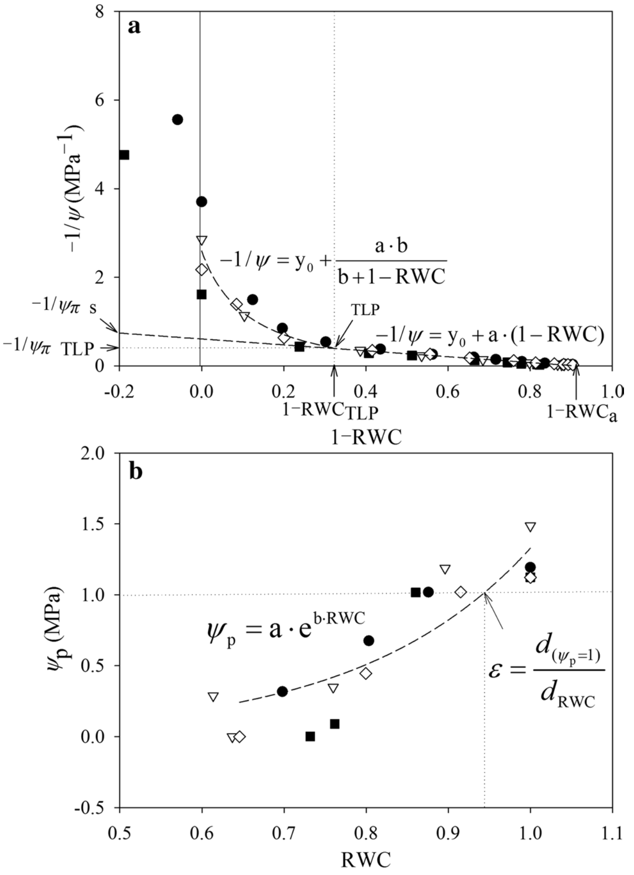

Fig. 3 Schematic diagrams showing how parameters of water relations were calculated, based on **a** pressure–volume curves and **b** graphs of turgor pressure (Ψ_p) against relative water content (RWC) (data for *Plagiochila arbuscula*). *TLP* turgor loss point. Different symbols indicate replicates of specific species. *Short dashed lines* in upper graph show hyperbolic decay regression and linear regressions for curved and straight-line part of the pressure–volume curves. *Short dashed curves* in the lower graph show exponential regressions for Ψ_p/RWC curves. Relative water content and osmotic water potential at turgor loss ($\mathrm{RWC}_{\mathrm{TLP}}$, $\Psi_{\pi\mathrm{TLP}}$) were calculated based on point of intersection of beeline and hyperbola. Osmotic potential at full turgor ($\Psi_{\pi s}$) was calculated by negative reciprocal of intercept of the fitted linear model. Apoplastic water (R_a) was calculated according to the x-intercept of the fitted linear model; residual water content can be used to calculate symplast water (R_s). Bulk elastic modulus (ε) was calculated from the fitted exponential models at an arbitrary Ψ_P of 1.0 MPa

Relationships between A_N and PAR were fitted by a modified rectangular hyperbola, which showed advantages over the non-rectangular hyperbola, rectangular hyperbola, and Prado–Moraes models in C_3 plants (Ye 2007; Ye and Yu 2008).

$$A_N = \mathrm{a} \cdot \frac{1 - \mathrm{b} \cdot \mathrm{I}}{1 + \mathrm{c} \cdot \mathrm{I}} \cdot \mathrm{I} - R_d, \tag{7}$$

where I is light intensity; R_d is the dark respiration rate; a, b, c are coefficients.

The main photosynthetic characteristics were calculated according to Ye (2007); Ye and Yu (2008). These included the light-saturated net photosynthetic rate (A_{nmax}; μmol CO_2 m^{-2} s^{-1}), light saturation point (I_{sat}: light intensity on the light curve beyond, which further increases will not augment the rate of photosynthesis; μmol m^{-2} s^{-1}), and light compensation point (I_c: light intensity on the light curve where the rate of photosynthesis exactly matches the rate of respiration; μmol m^{-2} s^{-1}).

All data were submitted to normality and homoscedasticity tests before applying further statistical methods. Differences in water relations and gas exchange characteristics were compared using one-way ANOVA with Tukey's post hoc tests via SPSS 16.0 software (SPSS Inc., Chicago, IL, USA). All regressions and figures were made using SigmaPlot 12.5 (Systat Software Inc., San Jose, CA, USA).

Results

Microclimatic conditions

Compared with the forest canopy, significantly lower air temperatures (paired *t*-test, $t = 4.21$, $P = 0.001$) and PAR ($t = 2.177$, $P = 0.052$), but higher RH ($t = 9.027$, $P < 0.001$) were detected in the understory (Fig. 4). The detailed frequency distribution of RH indicated that the understory was a very moist microhabitat, especially during the rainy season (with 93.7 % of RH data recorded in July at more than 90 %; Table 1). In addition, the frequency distribution of PAR further confirmed very low light conditions in the understory (with 56.3 % of the PAR data recorded in February less than 1 μmol m^{-2} s^{-1}; Table 2).

Water relations and gas exchange of fan bryophytes

There was a reasonably good exponential decay regression fit between the WC of shoots and time for all three experimental species, with r^2 values 0.98–0.99 (Fig. 5). Average initial water contents of blotted samples of *H. flabellatum*, *P. arbuscula*, and *P. assamica* were 318.8, 402.6, and 451.7 %, respectively. All bryophyte samples lost water very quickly, and all dried up within an hour after being exposed to air with no water supply (Fig. 5). The measured final WCs were very low, with averages 11.6, 12.3, and 11.8 % for *H. flabellatum*, *P. arbuscula*, and *P. assamica*, respectively (model-fit values were much smaller). Further ANOVA results indicated no difference in average final WC (after air drying for 320 min) among different bryophyte species ($F_{2,14} = 0.055$, $P = 0.946$).

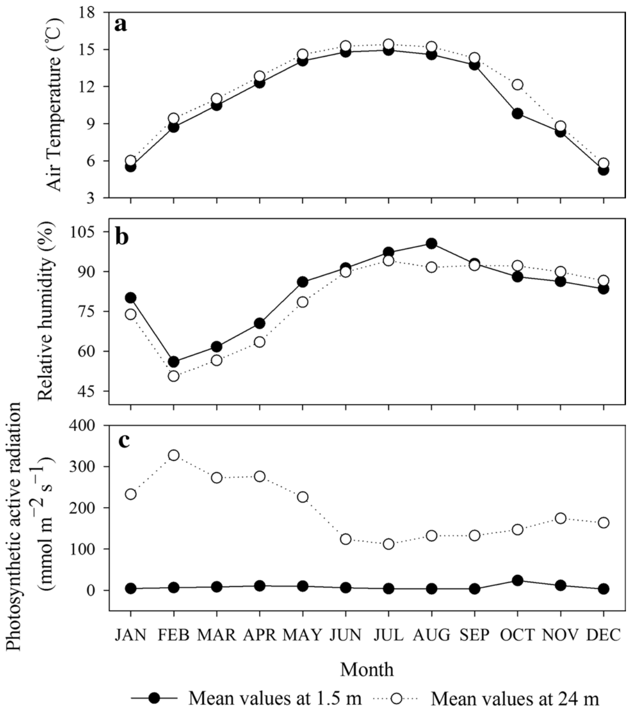

Fig. 4 Comparison of mean **a** air temperature, **b** relative humidity, and **c** photosynthetic active radiation between understory at ~1.5 m and forest canopy at ~24 m in the subtropical montane cloud forest during 2007–2013 (data from Ailaoshan Station for Subtropical Forest Ecosystem Studies, elevation 2480 m)

P–V curves are shown in Fig. 6. According to ANOVA results, significant differences were detected in R_s ($F_{2,11} = 16.330$, $P = 0.001$), $\Psi_{\pi TLP}$ ($F_{2,11} = 17.992$, $P = 0.001$), and $\Psi_{\pi s}$ ($F_{2,11} = 7.839$, $P = 0.011$), but no significant differences in R_a ($F_{2,11} = 1.791$, $P = 0.221$), RWC_{TLP} ($F_{2,11} = 0.334$, $P = 0.724$), and ε ($F_{2,11} = 0.167$, $P = 0.849$) were detected among the three studied bryophytes (Table 3). $\Psi_{\pi TLP}$ ranged from −4.2 MPa in *H. flabellatum* to −2.6 MPa in *P. assamica*, and $\Psi_{\pi s}$ from −2.5 to −1.6 MPa. The moss *H. flabellatum* had lower R_s, $\Psi_{\pi TLP}$, and $\Psi_{\pi s}$ (Table 3) compared with the two liverworts, and ε of the bryophytes was 4.2–4.5 (Table 3).

The response of A_N to WC in the three fan bryophyte species showed similar trends, rising rapidly with increasing WC (Fig. 7). The fitted sigmoidal models (logistic, four parameters) for A_N had r^2 from 0.85 to 0.95 (Fig. 7). A_N increased with WC, reaching 0-nmol CO_2 g^{-1} DW s^{-1} (respiration rates equal to photosynthetic rate) as WC approached 80 % DW (82.7, 104.1, and 81.7 % for *H. flabellatum*, *P. arbuscula*, and *P. assamica*, respectively).

Light response of the photosynthetic rate in the three bryophyte species was well described by a modified rectangular hyperbolic function (r^2 between 0.93 and 0.98; Fig. 7). Significant differences were detected in A_N ($F_{2,14} = 6.734$, $P = 0.011$) and R_d ($F_{2,11} = 5.964$,

Table 1 Percentage frequency distribution of relative humidity (RH) during February and July, 2013 at different heights in the subtropical montane cloud forest

RH interval (%)	February		July	
	1.5 m	24 m	1.5 m	24 m
90–100	0.7	4.3	93.7	84.9
80–90	8.9	5.0	5.8	9.8
70–80	14.4	8.3	0.3	3.5
60–70	18.1	13.3	0.1	0.9
50–60	15.8	19.9	0.2	0.2
<50	42.3	49.3	0.0	0.8

Table 2 Percentage frequency distribution of photosynthetically active radiation (PAR) during February and July 2013, at different heights in the subtropical montane cloud forest

PAR interval	February		July	
	1.5 m	24 m	1.5 m	24 m
>100	1.0	33.6	0.0	15.2
50–100	2.2	4.5	0.3	14.4
10–50	14.1	7.8	12.3	18.0
1–10	26.3	3.9	34.2	7.0
<1	56.3	50.3	53.1	45.4

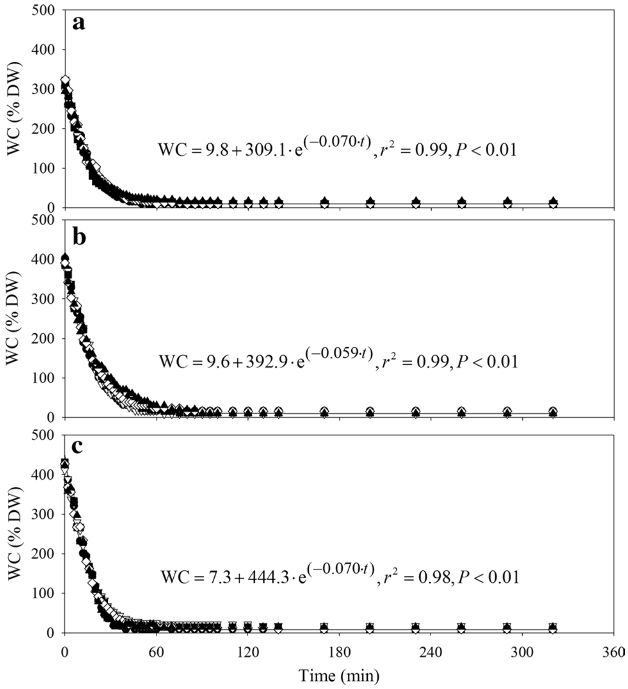

Fig. 5 Water release curves of **a** *Homaliodendron flabellatum*, **b** *Plagiochila arbuscula*, and **c** *P. assamica* (N = 5). *WC* water content

$P = 0.016$), but not in I_c ($F_{2,\,14} = 2.912$, $P = 0.093$) and I_s ($F_{2,\,14} = 0.038$, $P = 0.963$). Light compensation points (all less than 5 μmol m^{-2} s^{-1}; Table 4) and light saturation points (all less than 150 μmol m^{-2} s^{-1}; Table 4) were low for all three species. The moss *H. flabellatum* showed lower maximum A_N and R_d relative to the two liverworts (Table 4).

Discussion

Microclimatic conditions of the subtropical forest

Microclimate describes well the habitat under which epiphytic bryophytes on trees survive and grow in a SMCF. Similar to León-Vargas et al. (2006), measurements at 1.5 m above the soil surface showed quite high RH but very low temperature and light levels (Fig. 4). These results suggest that epiphytic bryophytes in the understory may remain continuously moist for days during the rainy season. RH was greater than 80 % for most of the time during that season. Even during the driest month, RH was near 60 % (Fig. 4). Frequent and persistent fog provides an important water source, which can alleviate water shortages for epiphytic bryophytes, especially during the dry season (Fig. 2b). However, bryophytes experience dry periods during the dry season and extreme drought in dry years (Song et al. 2012b). In addition, light is limiting for most plants, including bryophytes, in the deep shade of the understory (mean PAR at ~1.5 m above the soil surface was 7.8 μmol m^{-2} s^{-1}; Fig. 4). However, the deep shade minimizes evaporation of poikilohydric plants such as filmy ferns and bryophytes, and nullifies the advantages of typical vascular plants with ventilated mesophyll and stomatal control over water loss (Proctor 2012).

Water relations of fan bryophytes

Because fan bryophytes usually stand on tree trunks, they have very extensive surface areas in contact with air. However, their structure is like a double-edged sword. On one hand, they are better suited for intercepting water from moving air than many other bryophytes (Bates 1998; Kürschner et al. 1999). On the other hand, with their loose and open structures, fan bryophytes inevitably have weak resistance to water loss, owing to limited external capillary space (Bates 1998). Our results show that the three fan bryophytes lost most of their free water within 1 h when exposed to air without moisture supply, suggesting weak water retention capacity of these

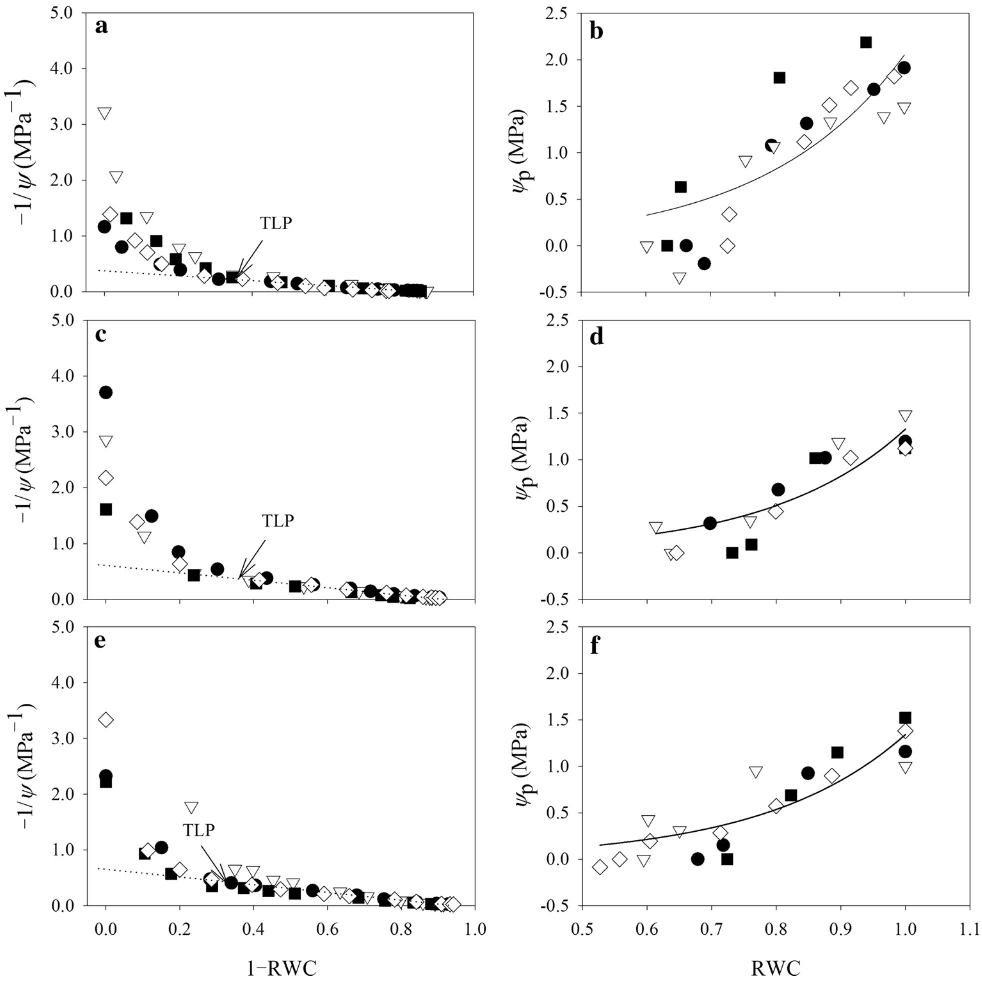

Fig. 6 Pressure–volume curves (*left panel*) and graphs of turgor pressure against relative water content (*right panel*) of **a**, **b** *H. flabellatum*, **c**, **d** *P. arbuscula*, and **e**, **f** *P. assamica* (N = 4). Abbreviations and symbols are in Fig. 3

Table 3 Parameters of water relations in (a, b) *Homaliodendron flabellatum*, (c, d) *Plagiochila arbuscula*, and (e, f) *P. assamica* (N = 4)

Species	R_s (% DW)	R_a (% DW)	RWC_{TLP} (%)	$\Psi_{\pi TLP}$ (MPa)	$\Psi_{\pi s}$ (MPa)	ε
Homaliodendron flabellatum	112.0 ± 9.1a	19.6 ± 3.3a	64.3 ± 3.7a	−4.2 ± 0.3a	−2.5 ± 0.2a	4.2 ± 0.3a
Plagiochila arbuscula	194.5 ± 13.1b	19.5 ± 5.8a	67.4 ± 2.2a	−2.6 ± 0.2b	−1.7 ± 0.1b	4.5 ± 0.5a
P. assamica	173.4 ± 9.2b	10.2 ± 2.0a	63.9 ± 3.8a	−2.6 ± 0.2b	−1.6 ± 0.2b	4.5 ± 0.6a

Values are mean ± SE. Different lowercase letters indicate significant difference for α = 0.05

bryophytes. The high abundance of fan bryophytes with low water retention capacity in the SMCF (Ma et al. 2009) is likely facilitated by the heavy annual precipitation (1859 mm), high average RH (82.6 %), and frequent and long duration of fog (211 foggy days annually; Fig. 2), which may provide a continuous water source for the bryophytes to intercept. The fog could be a very important water source for fans, especially in the dry season (Fig. 2b). The present study provides some of the scarce empirical evidence of the low water retention capacity of fan bryophytes and implies that these organisms are often associated with continuous water supply from frequent rain, clouds or fogs.

The water relations characteristics of fan bryophytes in this study are generally consistent with those of Santarius (1994), Beckett (1997), Proctor et al. (1998), and Proctor (1999). Therefore, our results add evidence for the generally mesic pattern of physiological adaption of bryophytes. Nevertheless, fan bryophytes show tolerance to desiccation, which may occur at any time of year (Proctor 2004a; León-Vargas et al. 2006). ε is of ecophysiological interest in plants subject to periodic water stress. In this study, generally low ε (4.2–4.5) and thus high cell elasticity were detected for the fan bryophytes relative to other studies (9–34) (Beckett 1997). This could be because fan bryophytes need high cell

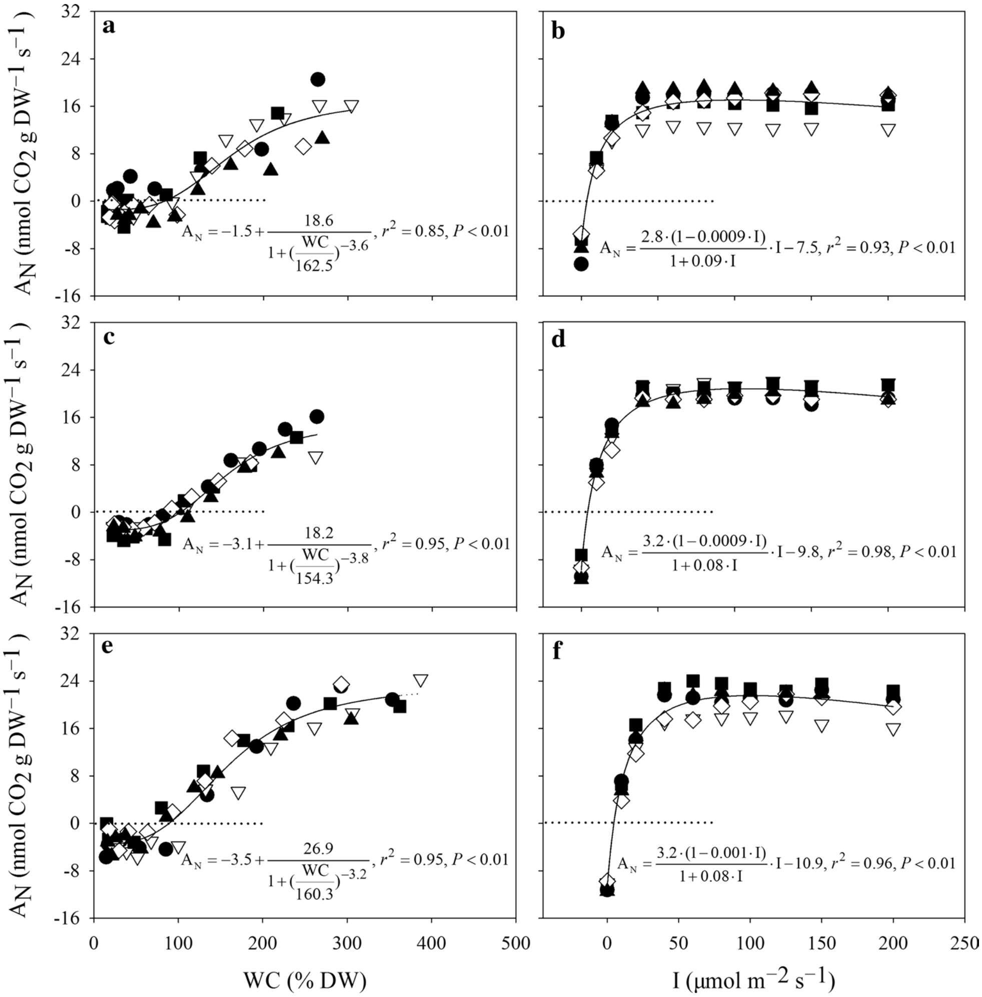

Fig. 7 Photosynthetic water and light response curves of **a**, **b** *H. flabellatum*, **c**, **d** *P. arbuscula*, and **e**, **f** *P. assamica* (N = 5). Symbol shapes refer to different samples. *Dotted lines* in figures indicate zero net photosynthetic rates (A_N). WC and I in figures refer to water content and light intensity, respectively

Table 4 Light saturation point (I_s), light compensation point (I_c), maximum net photosynthetic rate (A_{Nmax}), and dark respiration (R_d) derived from photosynthetic light response curves of (a) *H. flabellatum*, (b) *P. arbuscula*, and (c) *P. assamica* (N = 5)

Species	I_s (μmol m^{-2} s^{-1})	I_c (μmol m^{-2} s^{-1})	A_{Nmax} (nmol CO_2 g^{-1} DW s^{-1})	R_d (nmol CO_2 g^{-1} DW s^{-1})
Homaliodendron flabellatum	101.9 ± 8.5a	3.6 ± 0.3a	17.1 ± 1.2a	7.5 ± 0.9a
Plagiochila arbuscula	104.5 ± 4.9a	4.2 ± 0.3a	20.8 ± 0.5b	9.8 ± 0.7ab
P. assamica	103.3 ± 6.9a	4.7 ± 0.3a	21.7 ± 1.0b	10.9 ± 0.4b

Values are mean ± SE. Different lowercase letters indicate significant difference for α = 0.05

wall elasticity to tolerate desiccation during dry periods, owing to their weaker water retention capacity. Additionally, the osmotic potentials at turgor loss point of the fan bryophytes were lower than co-occurring angiosperm species (Zhang YJ, unpublished data), suggesting greater drought tolerance of fan bryophytes compared with angiosperms. Thus, fan bryophytes have weak water retention abilities but high desiccation tolerance, for surviving dry periods. Further experiments should be done to explore the desiccation tolerance of different life form bryophyte species in various microhabitats.

Gas exchange of fan bryophytes

Net photosynthetic rates of fan bryophytes are strongly dependent on WC, which increases sigmoidaly with increasing WC. A decline in photosynthetic rate at high WC has been detected in some bryophyte species (Dilks and Proctor 1979), suggesting depression of carbon gain at such WC. However, this was not observed for any of the three bryophyte species in the present study. These fan bryophytes may have structural or physiological adaptations to facilitate gas exchange at high WC during the rainy season. Nevertheless, the WC of the blotted bryophyte materials in this study (with maximum <500 %; see Fig. 7) did not reach the maximum WC of the saturated bryophyte materials, which was between 700 and 2000 % (Dilks and Proctor 1979).

It has been reported that the light compensation points of 18 tropical bryophyte species were from 22 μmol m^{-2} s^{-1} in *Octoblepharum pulvinatum* (Dozy & Molk.) Mitt. to 159 μmol m^{-2} s^{-1} in *Frullania mirabilis* J.B. Jack & Stephani (Wagner et al. 2013). In addition, Marschall and Proctor (2004) found that the light saturation points of 39 mosses and 16 liverworts varied from 79 μmol m^{-2} s^{-1} in *Jubula hutchinsiae* (Hook.) Dumort. to 2549 μmol m^{-2} s^{-1} in *Polytrichum piliferum* Hedw., with median 583 μmol m^{-2} s^{-1} for mosses and 214 μmol m^{-2} s^{-1} for liverworts. The latter may be overestimated because the light saturation points were mainly taken from light curves of relative electron transport rate (not CO_2 exchange), based on measurements of (modulated) chlorophyll florescence (Marschall and Proctor 2004). In our study, we measured light-response curves of gas exchange for three fan bryophyte species and found relatively small values of light compensation points (<5 μmol m^{-2} s^{-1}; Table 4) and light saturation points (<105 μmol m^{-2} s^{-1}; Table 4). Similarly, some filmy fern species from comparable habitats and of comparable life forms show comparable photosynthetic light saturation points (Proctor 2012). Low light compensation and saturation points are adaptations to low light levels.

Interspecific comparisons

It had been repeatedly confirmed that the ratio of species richness of liverworts to mosses correlate well with water availability, because liverworts usually favor wet habitats (Wolf 1993; Frahm and Ohlemüller 2001). For example, there was a distinct increase of liverworts with increasing water input from eastern to western Europe (Duell 1984). In addition, liverworts accounted for 80 % of bryophytes in the Chocó region, one of the heaviest precipitation regions with nearly 15,000 mm $year^{-1}$ (Frahm 1994). Our results provide a physiological explanation for these patterns. According to P–V relationships, the moss (*H. flabellatum*) in our study had higher osmoregulation capacity (more negative $\Psi_{\pi TLP}$ and $\Psi_{\pi s}$), and therefore greater desiccation tolerance than the liverworts (*P. arbuscula* and *P. assamica*). This suggests that liverworts may have less tolerance to drought compared with mosses. Further study is needed to confirm this hypothesis, considering that very few species have been included in the present study.

Ecological implications

Our results suggest that net photosynthesis rates of fan bryophytes are greatly dependent on WC. Because fan bryophytes lost most of their free water within 1 h in the dry air without water supply (Fig. 2), the implication is that the transition from a hydrated, photosynthetically active state to a dry, inactive state is rapid. In other words, fan bryophytes dry up and suspend photosynthesis quickly when water is unavailable. These fan bryophytes developed desiccation tolerance, which can be considered a special adaptive strategy to unpredictable dry periods or extreme drought in moist cloud forests (Proctor 2000, 2001). Considering abundant desiccation-tolerant epiphytic bryophytes investigated in SMCFs (Ma 2009; Song et al. 2011), we suggest that desiccation-tolerant bryophytes are among the most conspicuously successful plants in habitats with reasonably regular moisture supply.

Considering that carbon gain and growth are restricted to periods of sufficient hydration, capturing and preserving water are thus suggested to be crucial adaptations for bryophytes (Norris 1990; Wagner et al. 2013). The low water retention capacity and strong dependence of net photosynthesis on the WC of fan bryophytes thus imply a high risk of carbon imbalance under dry conditions. This situation would be exacerbated by the fact that faster drying could lead to more frequent rewetting, causing carbon loss through carbohydrate leaching (Coxson et al. 1992). The study area of Yunnan is currently experiencing drought (Fan and Thomas 2013). There was a severe drought from the end of 2009 to early 2010, which can be considered the worst over the past century (Qiu 2010; Wang 2010). This drought has been proven to result in remarkably reduced rates of growth and detrimental effects on the health of several epiphytic bryophytes (Song et al. 2012b). Since climate conditions in the SMCF are predicted to become drier by the 2050s (Hijmans et al. 2005; Ramirez and Jarvis 2008), the results of this study, together with those of a previous transplantation experiment (Song et al. 2012b), suggest a dim future for epiphytic bryophytes.

Conclusions

Our results suggest that fog helps maintain relatively high water availability during most of the year for tree trunk fan bryophytes in an Asian subtropical cloud forest. The fan bryophyte species studied have limited water retention capacities, but have developed relatively high cell wall elasticity and osmoregulation capacity so as to tolerate desiccation. In addition, relatively low light saturation and compensation points may help them tolerate shaded tree trunk microhabitats. The carbon gain of these fan bryophytes is highly water-dependent. Therefore, fan bryophytes can flourish on tree trunks in the subtropical cloud forest because of heavy annual precipitation, high average relative humidity, and frequent and long duration of fog, which provide continuous water supplies for them to intercept. However, the low water retention capacity and strong dependence of net photosynthesis on the water content of fan bryophytes suggest a high risk of carbon imbalance for them under increasing severity and frequency of drought. Therefore, this study points to an uncertain future for climate-sensitive epiphytic bryophytes under ongoing climatic change.

Acknowledgments This work was supported by the National Natural Science Foundation of China (31300382, U1133605, U1202234), CAS135 Program (XTBG-F01), West Light Foundation and the Research Foundation for members of the Youth Innovation Promotion Association of the Chinese Academy of Sciences to Liang Song. We thank Ailaoshan Station for Subtropical Forest Ecosystem Studies and Central Laboratory of Xishuangbanna Tropical Botanical Garden for Granting permission and facilitating this research. We are also grateful to Dr. Kouki Hikosaka, Mr. Steven Hunter, and the two anonymous reviewers for their constructive comments and corrections to the manuscript.

References

Bates JW (1998) Is' life-form'a useful concept in bryophyte ecology? Oikos 82:223–237

Beckett RP (1997) Pressure–volume analysis of a range of poikilohydric plants implies the existence of negative turgor in vegetative cells. Ann Bot 79:145–152

Chen L, Liu W-Y, Wang G-S (2010) Estimation of epiphytic biomass and nutrient pools in the subtropical montane cloud forest in the Ailao Mountains, south-western China. Ecol Res 25:315–325

Coxson DS, McIntyre DD, Vogel HJ (1992) Pulse release of sugars and polyols from canopy bryophytes in tropical montane rain forest (Guadeloupe, French West Indies). Biotropica 24:121–133

Dilks TJK, Proctor MCF (1979) Photosynthesis, respiration and water content in bryophytes. New Phytol 82:97–114

Duell R (1984) Computerized evaluation of the distribution of European liverworts. J Hattori Bot Lab 56:1–5

Fan Z-X, Thomas A (2013) Spatiotemporal variability of reference evapotranspiration and its contributing climatic factors in Yunnan Province, SW China, 1961–2004. Clim Change 116:309–325

Frahm J-P (1994) A contribution to the bryoflora of the Chocó region, Colombia. I. Mosses. Trop Bryol 9:89–110

Frahm J-P, Ohlemüller R (2001) Ecology of bryophytes along altitudinal and latitudinal gradients in New Zealand. Studies in austral temperate rain forest bryophytes 15. Trop Bryol 20:117–137

Green TGA, Sancho LG, Pintado A (2011) Ecophysiology of desiccation/rehydration cycles in mosses and lichens. In: Lüttge U, Beck E, Bartels D (eds) Plant desiccation tolerance, vol 215. Springer, Heidelberg, pp 89–120

Hijmans RJ, Cameron SE, Parra JL, Jones PG, Jarvis A (2005) Very high resolution interpolated climate surfaces for global land areas. Int J Climatol 25:1965–1978

Kürschner H, Frey W, Parolly G (1999) Patterns and adaptive trends of life forms, life strategies and ecomorphological structures in tropical epiphytic bryophytes—a pantropical synopsis. Nova Hedwig 69:73–100

León-Vargas Y, Engwald S, Proctor MC (2006) Microclimate, light adaptation and desiccation tolerance of epiphytic bryophytes in two Venezuelan cloud forests. J Biogeogr 33:901–913

Li S, Liu W-Y, Li D-W (2013) Epiphytic lichens in subtropical forest ecosystems in southwest China: species diversity and implications for conservation. Biol Conserv 159:88–95

Ma W (2009) The composition and biomass of epiphytic materials and their relationships with ecological factors in Xujiaba region from Ailao Mountain, Yunnan. Unpublished Ph.D. Thesis, Graduate School of the Chinese Academy of Sciences, Beijing

Ma W-Z, Liu W-Y, Li X-J (2009) Species composition and life forms of epiphytic bryophytes in old-growth and secondary forests in Mt. Ailao, SW China. Cryptogam Bryol 30:477–500

Marschall M, Proctor MCF (2004) Are bryophytes shade plants? Photosynthetic light responses and proportions of chlorophyll a, chlorophyll b and total carotenoids. Ann Bot 94:593–603

Miller HA (1982) Bryophyte evolution and geography. Biol J Linn Soc 18:145–196

Norris D (1990) Bryophytes in perennially moist forests of Papua New Guinea: ecological orientation and predictions of disturbance effects. Bot J Linn Soc 104:281–291

Proctor MCF (1990) The physiological basis of bryophyte production. Bot J Linn Soc 104:61–77

Proctor MCF (1999) Water-relations parameters of some bryophytes evaluated by thermocouple psychrometry. J Bryol 21:263–270

Proctor MCF (2000) The bryophyte paradox: tolerance of desiccation, evasion of drought. Plant Ecol 151:41–49

Proctor MCF (2001) Patterns of desiccation tolerance and recovery in bryophytes. Plant Growth Regul 35:147–156

Proctor MCF (2002) Ecophysiological measurements on two pendulous forest mosses from Uganda, *Pilotrichella ampullacea* and *Floribundaria floribunda*. J Bryol 24:223–232

Proctor MCF (2004a) How long must a desiccation-tolerant moss tolerate desiccation? Some results of 2 years' data logging on Grimmia pulvinata. Physiol Plant 122:21–27

Proctor MCF (2004b) Light and desiccation responses of *Weymouthia mollis* and *W. cochlearifolia*, two pendulous rainforest epiphytes from Australia and New Zealand. J Bryol 26:167–173

Proctor MCF (2012) Light and desiccation responses of some Hymenophyllaceae (filmy ferns) from Trinidad, Venezuela and New Zealand: poikilohydry in a light-limited but low evaporation ecological niche. Ann Bot 109:1019–1026

Proctor MCF, Tuba Z (2002) Poikilohydry and homoihydry: antithesis or spectrum of possibilities? New Phytol 156:327–349

Proctor MCF, Nagy Z, Csintalan Z, Takács Z (1998) Water-content components in bryophytes: analysis of pressure-volume relationships. J Exp Bot 49:1845–1854

Proctor MCF, Oliver MJ, Wood AJ, Alpert P, Stark LR, Cleavitt NL, Mishler BD (2007) Desiccation-tolerance in bryophytes: a review. Bryologist 110:595–621

Qiu J (2010) China drought highlights future climate threats. Nature 465:142–143

Qiu X-Z, Xie S-C (1998) Studies on the forest ecosystem in Ailao Mountains, Yunnan, China. Yunnan Sciences and Technology Press, Kunming

Ramirez J, Jarvis A (2008) High resolution statistically downscaled future climate surfaces. International Center for Tropical Agriculture (CIAT); CGIAR Research Program on Climate Change, Agriculture and Food Security (CCAFS) Cali, Colombia. http://wwwccafs-climateorg/

Roberts SW, Strain BR, Knoerr KR (1981) Seasonal variation of leaf tissue elasticity in four forest tree species. Physiol Plant 52:245–250

Romero C, Putz FE, Kitajima K (2006) Ecophysiology in relation to exposure of pendant epiphytic bryophytes in the canopy of a tropical montane oak forest. Biotropica 38:35–41

Santarius KA (1994) Apoplasmic water fractions and osmotic potentials at full turgidity of some Bryidae. Planta 193:32–37

Shi J-P, Zhu H (2009) Tree species composition and diversity of tropical mountain cloud forest in the Yunnan, southwestern China. Ecol Res 24:83–92

Song L, Liu W-Y, Ma W-Z, Tan Z-H (2011) Bole epiphytic bryophytes on *Lithocarpus xylocarpus* (Kurz) Markgr. in the Ailao Mountains, SW China. Ecol Res 26:351–363

Song L, Liu W-Y, Ma W-Z, Qi J-H (2012a) Response of epiphytic bryophytes to simulated N deposition in a subtropical montane cloud forest in southwestern China. Oecologia 170:847–856

Song L, Liu W-Y, Nadkarni NM (2012b) Response of non-vascular epiphytes to simulated climate change in a montane moist evergreen broad-leaved forest in southwest China. Biol Conserv 152:127–135

Song L et al (2015) Bole bryophyte diversity and distribution patterns along three altitudinal gradients in Yunnan, China. J Veg Sci (published online). http://onlinelibrary.wiley.com/doi/10.1111/jvs.12263/abstract

Tyree M, Karamanos A (1981) Water stress as an ecological factor. In: Grace J, Ford E, Jarvis PG (eds) 21st symposium of British Ecological Society. Blackwell Scientific Publications, Oxford

Wagner S, Zotz G, Allen NS, Bader MY (2013) Altitudinal changes in temperature responses of net photosynthesis and dark respiration in tropical bryophytes. Ann Bot 111:455–465

Wang X-J (2010) System thinking for drought management in Southwest China. China Water Resour 7:11–13

Wolf JH (1993) Diversity patterns and biomass of epiphytic bryophytes and lichens along an altitudinal gradient in the northern Andes. Ann Mo Bot Gard 928–960

Wu Z-Y (ed) (1983) Research of forest ecosystem on Ailao Mountains, Yunnan. Yunnan Science and Technology Press, Kunming

Xu H-Q, Liu W-Y (2005) Species diversity and distribution of epiphytes in the montane moist evergreen broad-leaved forest in Ailao Mountain, Yunnan. Biodivers Sci 13:137–147

Ye Z-P (2007) A new model for relationship between irradiance and the rate of photosynthesis in *Oryza sativa*. Photosynthetica 45:637–640

Ye Z-P, Yu Q (2008) Comparison of new and several classical models of photosynthesis in response to irradiance. J Plant Ecol (Chinese Version) 32:1356–1361

Zhang Y-J, Yang Q-Y, Lee DW, Goldstein G, Cao K-F (2013) Extended leaf senescence promotes carbon gain and nutrient resorption: importance of maintaining winter photosynthesis in subtropical forests. Oecologia 173:721–730

Zotz G, Schweikert A, Jetz W, Westerman H (2000) Water relations and carbon gain are closely related to cushion size in the moss *Grimmia pulvinata*. New Phytol 148:59–67