



Variability of water storage capacity in three lichen species

Anna Klamerus-Iwan¹ · Rafał Kozłowski² · Joanna Przybylska² · Wiktor Solarz¹ · Wojciech Sikora¹

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Abstract

As poikilohydric organisms, lichens are capable of storing significant amounts of atmospheric water. Epiphytes that intercept rainfall change the amount and chemical composition of throughfall water, affecting water balance and microclimate of forest ecosystems. The aim of the study was to investigate the differences in the process of changes/increase in the amount of water in three lichen species: *Evernia prunastrii*, *Hypogymnia tubulosa* and *Platismatia glauca*. In the experiment, conducted under laboratory conditions, samples of thalli were wetted with constant doses of water and weighed in order to determine the amount of water storage capacity from simulated rainfall. The studied lichen species differed in terms of process dynamics and values of water storage capacity, probably due to the morphological structure of thalli. Average water retention was the highest in *Platismatia glauca* (33.58 %), lower in *Evernia prunastrii* (19.77 %) and the lowest in *Hypogymnia tubulosa* (15.38 %). Analyzed taxa with larger water storage capacity are also known to be more sensitive to air pollution.

Keywords Ecohydrology · Epiphyte · Forest retention · Pollution · Rainfall · Water storage capacity

Introduction

As poikilohydric organisms with distinct morphological and physiological features, lichens, i.e. lichenised fungi, are able to absorb atmospheric water from the thallus surface. Apart from the growth form, determining surface area to volume ratio, some taxa have structures facilitating gas exchange, i.e. cyphellae and pseudocyphellae, enabling a higher water uptake and accumulation of contaminants (Józwiak 2014). Lichens are capable of storing even 3360 % of their dry weight in water; however, thallus supersaturation depresses photosynthesis (Lichner et al. 2012); and species from moist microhabitats have developed mechanisms to avoid excessive water

uptake (Hauck et al. 2008). Lichens can also absorb water from fog (Maphangwa et al. 2012). Due to their functional variability, lichens are present in nearly all climate regions. They dominate ca. 10 % of terrestrial ecosystems, covering areas with extreme conditions, inaccessible to most vascular plants, i.e. deserts, tundra and alpine habitats (Honegger 2007). Pypker et al. (2006) specifically mentions arboreal epiphytic lichens and their role in the canopy.

As lichens intercept and gradually evaporate rainwater, their presence modifies microclimate (Porada et al. 2014; Van Stan and Pypker 2015). Epiphytes significantly change the amount of water reaching the soil (Prada et al. 2009, 2012; Figueira et al. 2013) and reduce soil erosion (Lichner et al. 2012; Šurda et al. 2015; Yousefi et al. 2018). The global abundance of lichens, their high absorbency and efficient interception of atmospheric water suggest that they could play an important role in the forest water budget (Porada et al. 2018). Moreover, the accumulating capacity of epiphytic lichens can significantly decrease the concentration of trace elements in throughfall water (Cercasov et al. 2002). Phinney (2019) uses 3D modelling of topography of *Lobaria pulmonaria* thallus to facilitate the understanding of water storage pools.

Lichens also have the function of indicator species (bioindicators) (Hawksworth and Rose 1970). The lichen scale has been developed by Kiszka (1990) and modernised by Bielczyk (2001). On the lichen scale, 7 zones were

✉ Anna Klamerus-Iwan
a.klamerus-iwan@ur.krakow.pl

Rafał Kozłowski
rafal.kozlowski@ujk.edu.pl

Joanna Przybylska
joanna.przybylska@tbop.org.pl

¹ Faculty of Forestry, Department of Forest Engineering, University of Agriculture in Krakow, Al. 29 Listopada 46, 31-425 Krakow, Poland

² Institute of Geography and Environmental Sciences, Jan Kochanowski University, ul. Uniwersytecka 7, 25-406 Kielce, Poland

identified depending on the SO_2 concentration [$\mu\text{g}/\text{m}^3$]. Zone I: above $170 \mu\text{g}/\text{m}^3$, is the so-called lichen desert; zone II ranges from 100 to $170 \mu\text{g}/\text{m}^3$; zone III has $70\text{--}100 \mu\text{g}/\text{m}^3$; zone IV: $50\text{--}70 \mu\text{g}/\text{m}^3$; zone V: $40\text{--}50 \mu\text{g}/\text{m}^3$; zone VI has $30\text{--}40 \mu\text{g}/\text{m}^3$; and zone VII is below $30 \mu\text{g}/\text{m}^3$. The higher the zone, the lower the contamination tolerance range. The lichen structure is also associated with tolerance to pollution. In zone II, lichens usually have a crusty thallus. In zone III lichens are fine-leaved whereas in zone IV they are leafy, and this is where *Hypogymnia psysodes* belongs. In zone V, next to leafy lichens, bushy species such as *Evernia prunastri* appear. Zones VI and VII include all previous lichens types as well as one of the more demanding species in terms of air purity: *Platismatia glauca*. Poleotolerance as well as structure types has also been included in the Italian classification:

(<http://dryades.units.it/italic/?procedure=base&t=59&c=60#growth:%20growth%20forms%20of%20lichens>).

Total rainfall interception by forests includes both water storage and evaporation processes. The dynamic interchange between this state (storage) and flux (evaporation) can be a complex, because it depends on the spatial heterogeneity of canopy surfaces, their surface characteristics, and rainfall conditions (Dohnal et al. 2014). A key trait for integration of canopy elements into rainfall interception models is the water storage capacity under given conditions (often determined under simulated rainfall conditions). Thus, the aim of the study was to investigate the differences in the total water storage capacity and temporal variability for three lichen species common to Northeastern Poland, under simulated rainfall conditions. Moreover, the results obtained were compared with the resilience of various lichens to air pollution.

Material and methods

Analysed species of lichens

The samples were collected in the Puszcza Borecka Forest (NE Poland). The area of ca. 18 thousand hectares is dominated by deciduous and mixed forests with a significant share of spruce, some parts are also covered by coniferous mixed-species stands, bog woodlands and alluvial forests (Kondracki 1972). It is one of the areas of the best air quality in Poland (GIOŚ 2018).

In terms of climate regionalisation, the research area is located in the Węgorzewo Region, where the mean annual temperature is $6.1\text{--}6.5^\circ\text{C}$, the coldest month is February (the mean ca. -4°C), the warmest month is July (the mean ca. $+16.8^\circ\text{C}$) and the growing season lasts ca. 200 days (Stopa-Boryczka 1986). Rainfall frequency in the Węgorzewo Region (170–180 days of $>0.1 \text{ mm}$ precipitation) is higher than in many parts of Polish lowlands (Woś 1999). Its annual precipitation has been given as

ranging from 620 to 660 mm (Stopa-Boryczka 1986) and up to 700 mm (Woś 1999), as in the other postglacial lakelands of northern Poland with elevations of 200 m a. s. l. or more, which trap humid polar maritime air masses moving from the north-west. More recent measurements from the Puszcza Borecka Forest show annual precipitation ranging from 494.5 to 855.7 mm in 2003 and 2007 (Skotak et al. 2018). The climate of the examined area and especially of the Puszcza Borecka Forest is influenced by the landforms and landscape (related to the presence of high morainal hills; as mentioned above) as well as the microclimate resulting from the local relief and plant cover types (Zalewska 2012). All major young glacial landforms can be found here, with clear domination of hilly ground moraine areas showing highly diversified relief, with elevations of 133–223 m a.s.l. and many hills above 200 m a.s.l. Local height differences are considerable (on average 20–50 m, max. 90 m). The largest area is covered by subcontinental *Tilio-Carpinetum* forest showing considerable local habitat variation related primarily to soil moisture and nutrient content (Polakowski 1961; Polakowski 1963; Endler et al. 1991; Matuszkiewicz 2005). *Tilio-Carpinetum typicum* phytocoenoses are most common, occurring on slopes and flat-topped hills. Multilayered stands are typically composed of hornbeam *Carpinus betulus*, common oak *Quercus robur*, small-leaved linden (lime) *Tilia cordata*, and Norway spruce *Picea abies* as a naturally accompanying species (Czarnota and Kukwa 2008).

Lichen samples were collected in the area of Podleśne village in the Puszcza Borecka Forest on 31.03.2019. Based on meteorological data obtained from the Mikołajki station approx. 40 km from the sampling site, it was noted that the average daily temperature amounted to 6.9°C , while the monthly average was 4.4°C ; and the relative air humidity was 66%, while the monthly average was 75 % (<https://en.tutiempo.net/climate/2019/ws-122800.html>). On that day, no precipitation was recorded and the monthly precipitation average amounted to 38.9 mm. In the period preceding the sampling (from 20th March), the average daily temperature ranged from 1.2 to 10.6°C , whereas the average daily humidity ranged from 61 to 85 %. In that period, the total precipitation amounted to 5.9 mm as recorded on 27th and 28th March, i.e. 3 and 4 days before the sampling.

Tree lichen species were analysed in the experiment. *Hypogymnia tubulosa* (Fig. 1) has a foliose, rosette-like thallus with tubular lobes adhering to the substrate. Soredia are grouped into convex knots, located at the end of lobes. *Platismatia glauca* (Fig. 2) forms large, foliose thalli with broad, upcurved lobes, corrugated margins and marginal isidia and soredia. *Evernia prunastri* (Fig. 3) has a fruticose or subfruticose, branched thallus, drooping or erect, with flat, pitted and rugose lobes. Soredia are numerous, often on ridges and margins (Fałtynowicz 2012). Lichen samples located on dry branches were collected at the height of 1–3 m above ground from living spruce tree stems.



Fig. 1 *Hypogymnia tubulosa*

As in other areas dominated by forest communities, the lichen biota of the analysed area consists mainly of epiphytes (Cieśliński and Tobolewski 1988; Cieśliński 1995, 1997; Cieśliński et al. 1995; Bystrek and Kolanko 2000). Sixty-eight percent of all identified species were found on the bark of trees, 38 % on dead wood, 35% on rocky substrates and 11% on the ground (Zalewska 2012). The main morphological forms of the recorded lichens show a percentage structure typical of woodland, with predominance of species having crustose thalli (71 %) and low shares of foliose (16 %) and fruticose (13 %) lichens. These relations are shaped by the abundance of epiphytic lichens, which have an almost identical percentage structure of forms. The percentages of foliose and/or fruticose forms are higher in the other habitat groups, probably due to the higher frequency of their substrates in well-insolated places.

Among the ecological groups, the epiphytes had the most diverse taxonomic composition (206 species) and the largest share of species with a high number of records (Zalewska 2012).

Measurements of water storage capacity

The samples were transported on spruce branches. For each species, 15 fragments of thallus of different sizes were

selected. The experiment was conducted under laboratory conditions (temperature 21 °C, air humidity 52 %). All samples were stored under the same conditions and wetting was performed on a single day so as to ensure result comparability. The measurements were divided into 21 runs, during which the samples were wetted with constant doses of water ($P = 10$ g). Each run lasted for 15 s and a subsequent dose of water was sprayed immediately after the sample was weighed. The average intensity of simulated rainfall amounted to $0.68 \text{ g} \cdot \text{s}^{-1}$. The wetting was performed using a garden sprinkler. The sprinkler nozzle was supported on a stand, on a constant level above the lichen sample. Water doses were measured from a calibrated container. Lichen samples were arranged on a mesh base above the container (Fig. 4). The lichen thallus was separated by means of tweezers from the tree bark on which it had been transported. The purpose was to eliminate water interception by the bark. The difference in the weight of a sample before and after wetting was equal to the amount of water (W) stored by lichen thalli in each run. The results of subsequent measurements reflected the increase in the weight of water intercepted by the lichens. The actual water storage capacity (S), i.e. water gain after each rainfall dose, was calculated per unit of wet biomass (bm). Relating water storage capacity to biomass and the relevant calculations are based on other studies (Garcia-Estringana et al. 2010). Water storage capacity in



Fig. 2 *Platismatia glauca*



Fig. 3 *Evernia prunastri*

grams of water per gram of thalli [$\text{g} \cdot \text{g}^{-1}$] was converted to the percentage gain of stored water, where each dose was treated as 100 % of rainfall. Water storage capacity expressed as a proportion of rainfall provides a better understanding of the amount of water absorbed by lichens. Water storage capacity is presented as a process of filling a container, which, in this experiment, was the sample of lichen thalli.

Statistical analysis

One-way analysis of variance was performed in order to verify the differences between the taxa. Multiple comparisons were conducted with the Sidak post-hoc test, using an index derived from the mean results of 21 water storage capacity measurements.

A series of regression analyses was conducted to find the model describing the relationship between time (subsequent doses of water) and value of water retained on a lichen sample. Linear, logarithmic, inverse, quadratic and cubic functions were tested. The coefficient of determination (R^2) was used to evaluate the goodness-of-fit of the models. A higher value of R^2 indicates a higher proportion of total variation of the

observed phenomenon explained by the model. In the last step, a cubic function was adapted and regression analysis was performed for each lichen species separately.

Results

Differences in the water storage capacity of lichen species

There was a statistically significant difference between the taxa in terms of the mean value of water storage capacity, as determined by one-way ANOVA ($F(2.942) = 930.49$, $p < 0.001$, $\eta^2 = 0.66$). The Sidak post-hoc test revealed that the water storage capacity value was the highest in *Platismatia glauca*, lower in *Evernia prunastri* and the lowest in *Hypogymnia tubulosa*: 33.58 % vs. 19.77 % vs. 15.38 %, respectively (Fig. 5).

Relationship between time and water storage capacity

The process of water storage capacity in 21 runs of the experiment was best described by a cubic function. With the first doses of water water storage capacity was growing dynamically, then it decreased and slightly increased again in the last measurements. The test results for each model are presented in Table 1. Parameters of the cubic function relating time (subsequent doses of water) and value of water storage capacity are presented in Table 2 and the curve is given in Fig. 6.

The cubic model for the water storage capacity of three lichen species

A regression analysis indicated that in all samples of lichen taxa each subsequent dose of simulated rainfall significantly affected the value of water storage capacity; *Evernia prunastri* $F(3.311) = 78.33$, $p < 0.001$; *Hypogymnia tubulosa*



Fig. 4 Lichen thallus sample on a mesh before simulated rainfall

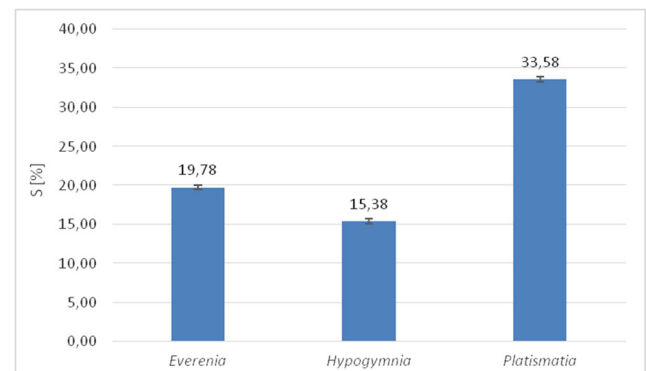


Fig. 5 Differences in water storage capacity of three lichen species. Error bars indicate the standard error of the mean. S [%] - water storage capacity as the portion of simulated rainfall stored by lichens

Table 1 Test results for analysed models

Function	<i>R</i>	<i>R</i> ²	<i>R</i> ² _{adj}	<i>SE</i>
Linear	0.35	0.12	0.12	8.91
Logarithmic	0.40	0.16	0.16	8.73
Inverse	0.37	0.14	0.14	8.85
Quadratic	0.40	0.16	0.16	8.73
Cubic	0.41	0.17	0.16	8.71

SE standard error

R correlation coefficient

*R*² coefficient of determination

*R*²_{adj} adjusted *R*²

$F(3.311) = 152.31$, $p < 0.001$; *Platismatia glauca* $F(3.311) = 126.913$, $p < 0.001$ (Table 3). The coefficient of determination revealed that subsequent doses of water affected water storage capacity mainly in *Hypogymnia tubulosa*, to a lesser extent in *Platismatia glauca*, while in *Evernia prunastri* the correlation between the two variables was the weakest (Table 4). The dynamics of the process was similar in all analysed taxa. The first doses of water increased water storage capacity dynamically; afterwards it decreased and then slightly increased again. The cubic model for each species is presented in Fig. 7.

Discussion

The values of water storage capacity in the experiment ranged from 15 % to 33 % and were slightly lower than the results based on mathematical models, calculated for bryophytes and lichens together in the temperate climate zone (Porada et al. 2018). Epiphytes in rainforests, i.e. lichens, bryophytes and ferns, are capable of storing as much as 80% of rainwater in the initial phase of precipitation (Veneklaas et al. 1990; Fačková et al. 2019). However, our results indicate that, after 21 runs of the experiment, lichen thalli did not achieve the maximum potential water storage capacity. The intensity

Table 2 Parameters of the cubic function relating time and water storage capacity of lichens

	<i>B</i>	<i>SE</i>	β	<i>t</i>	<i>p</i>
Iteration	2.90	0.53	1.85	5.51	<0.01
Iteration ** 2	−0.18	0.05	−2.61	−3.30	<0.01
Iteration ** 3	0.00	0.00	1.13	2.33	0.02
(α)	9.86	1.37		7.19	<0.01

t *t*-value

p probability value

β standardised coefficient

B unstandardised coefficient (results in measurement scale)

SE standard error

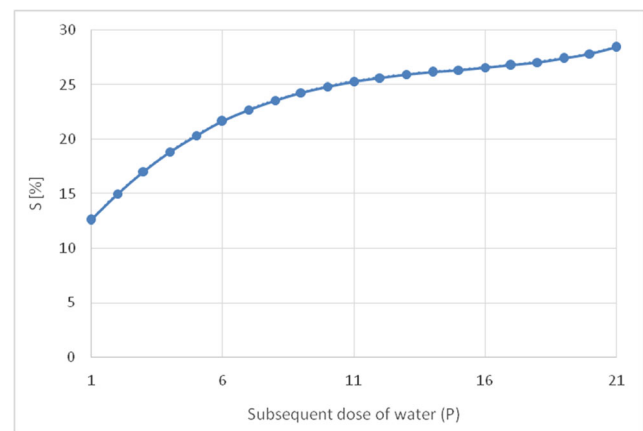


Fig. 6 Cubic function relating time (subsequent doses of water) and water storage capacity of water by all three lichen species

of simulated rainfall was not high, which proves that precipitation of low intensity adversely affects the time required for gaining the maximum water storage capacity (Klamerus-Iwan 2014). Obtaining the results that are lower than in other studies may also result from obtaining the values of water storage capacity that were close to those under field conditions.

The differences in the amount of the water retained could be related to morphological features of the analysed taxa. *Platismatia glauca* has a foliose, flat thallus with corrugated margins, which increases the capability of rainwater storage. *Evernia prunastri* with branched, fruticose thallus has a medium water storing capacity. However, the model characterising the relationship between time and changes in the water retained suggests that, in the case of this species, prolonged exposure to rainwater could increase its overall storage capacity. The amounts of stored water were the lowest in *Hypogymnia tubulosa* with a foliose thallus and convex lobes. The curves of all three cubic models were similar, yet in *Platismatia glauca* the initial storage capacity was the most dynamic and the curve was the most concave. Additionally, nonvascular epiphytes are able to significantly influence the atmospheric moisture dynamics even during non-rain periods (Pypker et al. 2017).

The anatomical structure of the thallus, including the presence of structures increasing its gas exchange, can be another factor affecting the water storage capacity of lichens (Ahmadjian and Hale 1973). Research on the absorption of contaminants from a water solution suggests that the presence of pores facilitates water absorption. Out of the three analysed species, pseudocyphellae are present in the thalli of *Hypogymnia tubulosa* and *Evernia prunastri*, i.e. taxa with lower water storage capacity values, rather than in *Platismatia glauca*, which does not have such structures. Therefore, the anatomical structure seems to be an important factor, next to the morphology of a thallus and its chemical properties, regulating the amounts of absorbed water.

Table 3 Relationship between water storage capacity and subsequent doses of simulated rainfall in three analysed lichen species

		<i>B</i>	<i>SE</i>	β	<i>t</i>	<i>p</i>
<i>Everenia prunastri</i> F(3,311) = 78,33; <i>p</i> < 0,001	Iteration	2.779	0.48	2.79	5.79	<0.01
	Iteration ** 2	−0.171	0.05	−3.89	−3.41	<0.01
	Iteration ** 3	0.004	0.00	1.70	2.43	0.02
	(Constant)	6.878	1.25		5.51	<0.01
<i>Hypogymnia tubulosa</i> F(3,311) = 152,31; <i>p</i> < 0,001	Iteration	2.231	0.25	3.56	8.78	<0.01
	Iteration ** 2	−0.138	0.03	−5.00	−5.21	<0.01
	Iteration ** 3	0.003	0.00	2.12	3.60	<0.01
	(Constant)	5.386	0.66		8.15	<0.01
<i>Platismatia glauca</i> F(3,311) = 126,913; <i>p</i> < 0,001	Iteration	3.693	0.45	3.52	8.21	<0.01
	Iteration ** 2	−0.235	0.05	−5.07	−5.01	<0.01
	Iteration ** 3	0.005	0.00	2.21	3.56	<0.01
	(Constant)	17.302	1.17		14.80	<0.01

t *t*-value*p* probability value β standardised coefficient*B* unstandardised coefficient (results in measurement scale)*SE* standard error

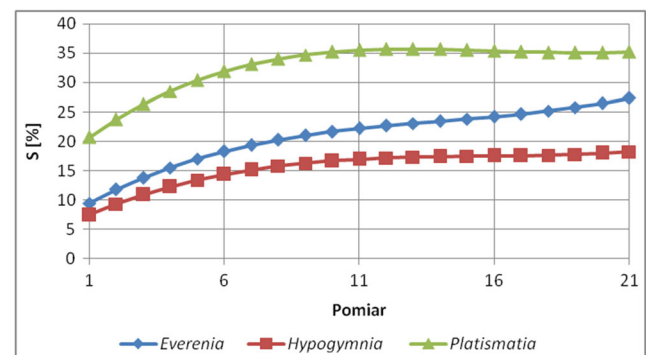
Hydrophobicity, characteristic of taxa inhabiting places exposed to rainwater, protects lichens from supersaturation, depressing photosynthesis (Honegger 2007). Additionally, it prevents dissolved pollutants from penetrating the thalli (Hauck et al. 2008). Research on wettability of the thallus surface indicates that *Platismatia glauca* and *Hypogymnia tubulosa* are moderately hydrophobic, while *Everenia prunastri* has a hydrophilic thallus; however, the differences between the analysed taxa were small: the contact angles ranged from 89° in *E. prunastri* to 95° in *P. glauca* and 96° in *H. tubulosa* (Hauck et al. 2008).

Without specialised dermal tissue, lichens are prone to absorb contaminants from the atmospheric air (Tretiach et al. 2007). The use of lichens in biomonitoring leads to delimitation of zones with different air quality (Svoboda 2007; Kubiak 2005), as selected species are more or less vulnerable to air pollution (Kiszka 1990; Bielczyk 2001; Conti and Cecchetti

2001). *Hypogymnia tubulosa*, which in our experiment intercepted the smallest amount of water, is found in areas with moderate air pollution (zone IV) and it is the least sensitive of the three studied species. *Everenia prunastri* grows under conditions of low pollution (zone V), while *Platismatia glauca*, the species with the highest interception rate, is the most sensitive one and can be found only in unpolluted areas (zone VI). Therefore, our results concerning water storage capacity indicate future research is merited to assess potential linkages between rainfall interception and absorption of contaminants into thalli. Atmospheric pollution can lead to a decline in species with a higher water storage capacity and, as a consequence, affect the forest water budget. However, in terms of hydrology, nonvascular epiphytes are a major canopy structural factor in the increased rainfall interception observed in old growth forests (Pypker et al. 2005).

Table 4 Test results for the analysed models

Species	<i>R</i>	<i>R</i> ²	<i>R</i> ² _{adj}	<i>SE</i>
<i>Everenia prunastri</i>	0.66	0.43	0.42	4.58
<i>Hypogymnia tubulosa</i>	0.77	0.60	0.59	2.43
<i>Platismatia glauca</i>	0.74	0.55	0.55	4.29

SE standard error*R* correlation coefficient*R*² coefficient of determination*R*²_{adj} adjusted *R*²**Fig. 7** Cubic functions relating time (subsequent doses of water) and water storage capacity of water by studied lichen species

Conclusions

Results on the water storage temporal dynamics and water storage capacity of lichen thalli for three study lichen species differed with regard to morphology and anatomical structure. Water retention was highest in *Platismatia glauca*, lower in *Evernia prunastrii* and lowest in *Hypogymnia tubulosa*. The morphological structure of lichen thalli may be the most important factor explaining differences in water storage temporal dynamics and capacity between analyzed species. The study lichen species with the highest water storage capacity is a species known to be more sensitive to air pollution. Thus, we recommend future research examine (a) how thallus morphology drives rainwater storage capacity and retention, as well as (b) how these rainwater storage and retention dynamics influence thalli's air pollution sensitivity.

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Compliance with ethical standards

Conflict of interest The authors declare no conflict of interest.

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