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Elasticity of the hair cover in air-retaining Salvinia surfaces

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Abstract Immersed in water superhydrophobic surfaces (e.g., lotus) maintain thin temporary air films. In certain aquatic plants and animals, these films are thicker and more persistent. Floating ferns of the genus Salvinia show elaborated hierarchical superhydrophobic surface structures: a hairy cover of complex trichomes. In the case of *S. molesta*, they are eggbeater shaped and topped by hydrophilic tips, which pin the air–water interface and prevent rupture of contact. It has been proposed that these trichomes can oscillate with the air–water interface, when turbulences occur and thereby stabilize the air film. The deformability of such arrays of trichomes requires a certain elasticity of the structures. In this study, we determined the stiffness of the trichome coverage of *S. molesta* and three other *Salvinia* species. Our results confirm the elasticity of the trichome coverage in all investigated *Salvinia* species. We did not reveal a clear relationship between the time of air retention and stiffness of the trichome coverage, which means that the air retention function is additionally dependent on different parameters, e.g., the trichome shape and surface free energy. These data are not only interesting for *Salvinia* biology, but also important for the development of biomimetic air-retaining surfaces.

1 Introduction

Superhydrophobicity as a result of a hydrophobic surface chemistry combined with a hierarchically double-structured surface architecture is a feature of many biological surfaces [1, 2]. Immersed into water these surfaces maintain a thin air layer causing an easily visible silvery reflection. These double-structured surfaces exhibit also a highly reduced contact area for contamination particles resulting with the well-known self-cleaning properties of the lotus effect [2, 3]. These surfaces are of crucial interest for biomimetic technical applications (survey in [4]). However, double-structured lotus-effect surfaces maintain the thin air layer only temporarily, but there are several aquatic organisms like the back-swimmer *Notonecta* or the floating fern *Salvinia*, which maintain air layers under water for a longer time period [5]. The ability of superhydrophobic surfaces to retain an air film under water drew increased attention in the last years [6–11]. In contrast to surfaces with lotus effect, air-retaining surfaces are not self-cleaning and are characterized by a much more complex elaborate surface architecture. Air-retaining surfaces are of high technological interest as such an air film can lower the friction caused by water flowing over a solid surface [9, 12]. This drag-reducing property has high potential for technical applications. Applied to ship hulls such an air cover is able to save energy in shipping. Moreover, the transfer of air-retaining surfaces to the inside of pipe lines could reduce the energy

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needed to pump fluid through. Five criteria have been proposed to maintain an air layer under water [5] resulting in a phenomena we call Salvinia Effect [3]: (1) a hydrophobic chemistry which causes with (2) nanostructures superhydrophobicity, (3) hair-like structures (trichomes, microtrichia, setae) between some micrometers and several millimeters length which exhibit (4) undercuts and which are (5) elastic. Natural air-retaining surfaces occur in a variety of semi-aquatic and aquatic plants and animals. All of them show a combination of these specific surface structures and a hydrophobic surface chemistry. The air film kept under water on the majority of such surfaces lasts from a few hours up to several months [6]. One species with remarkable air-retaining properties is the floating fern *Salvinia molesta* [3, 8]. Its leaves normally float on the water surface. If submerged, the air film causes high buoyancy, so that the leaves after being submerged immediately buoy upwards to the surface again. *Salvinia* leaves are densely covered with hair-like structures called trichomes. Depending on the species, these trichomes can be either single and unicellular, or multicellular and grouped. A trichome group has a thick multicellular base, called "emergence," from where two or more unicellular trichomes arise [13]. A paradox additional feature occurs in three out of all 10 investigated *Salvinia* species [13]: hydrophilic tips of the eggbeater-shaped trichomes caused by a chemical heterogeneity of the otherwise superhydrophobic surface. This "Salvinia paradox" causes a refined physical mechanism to stabilize air layers in water under fluctuation conditions [8]. The same study suggests that the elastic properties of the trichomes are of advantage, because they allow adaptation to movements of the air–water interface in both directions and therefore prevent rupture of contact. In a recent study, the elastic properties of individual trichomes on the leaves of the floating water fern *Salvinia molesta* were measured quantitatively for the first time [11]. This was achieved by using a novel quantitative method, the capillary adhesion technique (CAT). These measurements yielded water adhesion forces of individual *Salvinia* trichomes of approximately 20 μN in the case of trichomes of *Salvinia molesta* and elastic force constants of individual trichomes of $D = 2.1$ N/m. The aim of the present study is to apply a different technique to measure the stiffness of the hairy cover (indumentums) as well as to compare the stiffness of different *Salvinia* species, an inevitable basis for the generation of biomimetic surfaces considering a wide range of applications.

2 Materials and methods

2.1 Plant material

Four species of the floating fern genus Salvinia representing the whole morphological diversity of the 10 species

known were selected for the study. All plants were cultivated in the Botanical Gardens of the University of Bonn, Germany:

S. cucullata Roxb. Ex Boty (1833), accession no. 18268-7-2002, origin: South East Asia
S. minima Baker (1886), accession no. 8595-7-2002, origin: Central and South America;
S. molesta Mitchell (1972), accession no. 14459-1996, origin: Brazil
S. oblongifolia Mart. (1834), accession no. 14457-7-1996, origin: Brazil.

2.2 Microscopy

We investigated the surface morphology of fresh leaves using a digital microscope (VHX-1000 Keyence, Japan). The density of the microstructures was determined from top views of the leaves, while length, heights and diameter were determined from cross sections of fresh leaves. We measured the parameters of 10 samples for each species.

Additionally, we studied the plant surfaces using scanning electron microscopy (SEM). The plants were fixed in a formaldehyde solution (2 % formaldehyde, 65 % ethanol, 5 % acetic acid) for 2 h, dehydrated with ethanol, and dried in a critical point dryer (CPC020, Balzers Union, Liechtenstein). Specimens were coated with gold (25–30 nm) in a sputter coater (SCD-040, Balzers Union, Liechtenstein) and examined at 15 kV with a Stereoscan SEM (S200, Cambridge, UK), equipped with a lanthanum hexaboride filament.

2.3 Measurement of stiffness

Mechanical properties of the hairy coverage on leaves of four *Salvinia* species were measured in micro-indentation experiments by applying a micro-force tester Basalt-01 (Tetra GmbH, Ilmenau, Germany) according to [14] (see also [15, 16] for a more detailed description of the tester).

Fresh leaf blades were halved longitudinally with a razor blade. One half, attached to a metal holder by means of a double-sided adhesive tape with the adaxial surface facing up, was used as a lower sample (Fig. 1a). A sapphire sphere with a radius of 1.5 mm, fixed with a superglue to a glass spring having a spring constant of 137 N m^{-1}, served as an upper sample. The sphere was first brought close to the plant surface in the middle part of the sample, pressed against the sample with certain applied load and retracted from it. The motor drive speed for both loading and retracting was 70.3 μm s^{-1}, and the delay time between these processes was set for 3 s. The indentation depth ranged from 150 to 1500 μm depending on the plant species, because of different dimensions of trichomes: The leaf
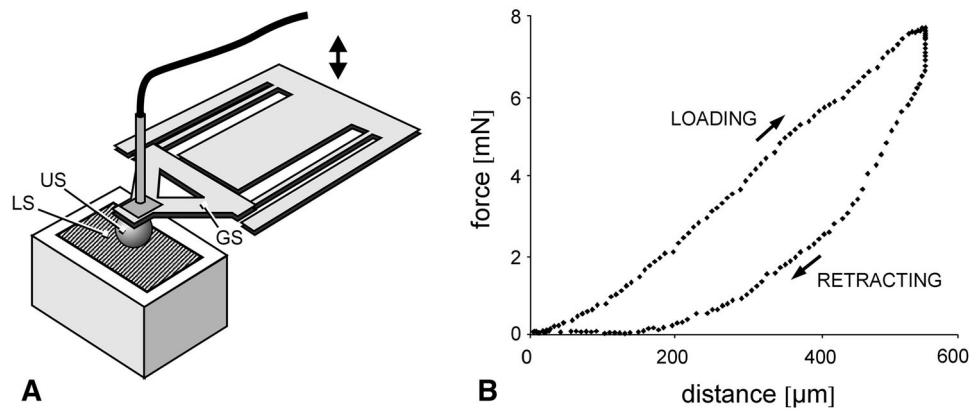
Fig. 1 Experimental setup (**a**) and typical force–distance curve (**b**) used for evaluation of mechanical properties (stiffness) of the trichome coverage of *Salvinia* leaves. LOADING, part of the curve corresponding to the loading process; RETRACTING, part of the curve corresponding to the retracting process; *GS* glass spring, *LS* lower sample (plant surface bearing a hairy coverage), *US* upper sample (*sapphire sphere*)

samples were indented up to the half height of their trichomes. The precise positioning between the counterparts and driven distances was monitored by the stereomicroscope Leica MZ 12.5 with a built-in video camera Leica IC A coupled with a video recorder. After each measurement, the sapphire sphere was cleaned with acetone. Experiments were performed at room temperature (22–25 °C) at a relative humidity of 47–56 %. For each plant species, five leaves with one test per each half were examined. In total, 40 individual tests with plant surfaces were carried out.

The recorded force–distance curves (Fig. 1b) were used to calculate the stiffness of the hairy coverage of the leaves. The trichomes deformed when the sapphire sphere pressed against the sample. The indentation depth into the array of trichomes was obtained by subtracting spring deflections in two separate experiments: on the plant sample and on a hard sample (glass plate) under the same applied force [15, 16].

To estimate the stiffness of the hairy leaf coverage, the loading part of the curve showing more linear character was used. The stiffness k was calculated according to Hooke's law of elasticity, which states that the change in length of a spring is in direct proportion with the load applied to it $k = \frac{F}{\delta}$, where F is the applied force and δ is the displacement (indentation depth) produced by the applied force along the same degree of freedom (in our case, indentation into the plant material).

3 Results

3.1 Surface architecture of Salvinia

The trichome morphology considerably varies between the four investigated *Salvinia* species (Fig. 2; Table 1). The "hairs" are a combination of a multicellular base

(emergence) topped by 1, 2 or 4 solitary multicellular trichomes which may remain connected on their tips like a basket resulting in the eggbeater-shaped trichomes like in *S. molesta*. *Salvinia cucullata* has one multicellular trichome per emergence, *S. oblongifolia* two and *S. minima* four free trichomes. In *S. molestea*, these four trichomes remain connected in the eggbeater-shaped structure. This combination between emergence and trichomes (shortly called "trichome") can considerably vary shape, height (about 180–2900 um) and diameter (270–600 um). Density of the emergence and trichomes varies from 2 to 26 structures per square millimeter. Due to these structural differences, the overall height of the structures differs considerably (310–2900 μm). This must lead to a varying height of the retained air film.

3.2 *Stiffness of* Salvinia *leaves*

The total stiffness of the trichome array showed significant differences in the investigated *Salvinia* species (Fig. 4) (Kruskal–Wallis test $H_{3.36} = 26.3$, $P < 0.001$; Dunn's method for all pair wise multiple comparison procedures $P < 0.05$). Two groups can be distinguished showing significant differences between each other (Dunn's method $P < 0.05$). Total stiffness was considerably lower in the first group (*S. cucullata* 4.3 ± 2.4 N/m; *S. molesta* 4.6 ± 2.2 N/m), if compared to that in the second one (*S. minima* 15.7 ± 4.8 N/m, *S. oblongifolia* 20.7 ± 8.1 N/m) (Fig. 3a).

Beside the total stiffness of the entire trichome array, we calculated the stiffness per trichome group from the data in Table 1 (Fig. 4b). Calculated stiffness of per trichome group differs significantly between species (Kruskal–Wallis test $H_{3.36} = 34.86$, $P < 0.001$). The stiffness related to the trichome group is significantly the highest in *S. minima*

Fig. 2 Surface structures of the leaves of four different *Salvinia* species: **a** *S. molesta*, **b** *S. minima*, **c** *S. cucullata*, **d** *S. oblongifolia*. Images are all taken with the digital microscope and shown in the same scale [17]
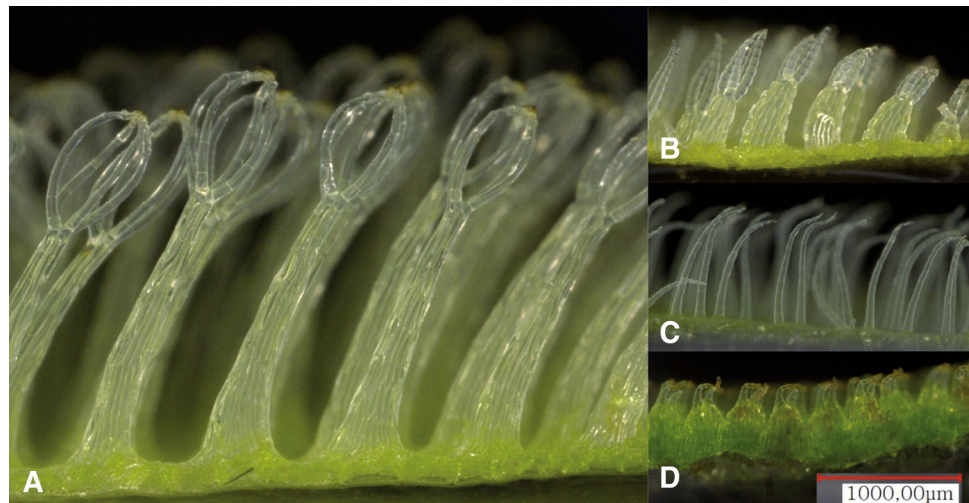
Table 1 Structural morphological data of the four investigated *Salvinia* species (m ± SD) [17]

		S. oblongifolia	*S. cucullata*	*S. minima*	*S. molesta*
Density (trichome groups)	1/mm²	25.8 ± 3.3	13.0 ± 2.6	2.3 ± 0.2	1.6 ± 0.2
Overall height of structures	µm	309.7 ± 41.0	558.3 ± 143.2	919.4 ± 106.8	2628.9 ± 285.1
Length of structure	µm	369.6 ± 46.4	609.5 ± 151.7	995.0 ± 108.0	2921.6 ± 264.1
Length of emergence	µm	177.2 ± 30.1		443.3 ± 93.3	1954.9 ± 301.8
Length of trichomes	µm	222.1 ± 27.6	609.5 ± 151.7	562.4 ± 45.9	1160.0 ± 88
Diameter of emergence (at base)	µm	269.1 ± 8.6		287.4 ± 18.1	589.7 ± 45.0
Diameter of emergence (at tip)	µm	168.2 ± 13.3		139.5 ± 11.3	177.0 ± 13.3
Diameter of whole eggbeater structure	µm				612.6 ± 73.5
Diameter of trichome (at base)	µm	68.5 ± 6.8	40.5 ± 6.3	71.2 ± 6.9	71.0 ± 6.7
Diameter of trichome (at tip)	µm	26.5 ± 4.8	9.6 ± 1.3	20.9 ± 3.0	
Trichome length/total length		0.56	1.00	0.56	0.33
Length–width quotient trichome group		1.37	15.05	13.97	4.95

(6.96 ± 2.15 N/m) compared to those in all other *Salvinia* species (Dunn's method $P < 0.05$). *S. molesta* has an average stiffness of 2.82 ± 1.32 N/m per trichome group, which is considerably higher than those in *S. cucullata* (0.28 ± 0.12 N/m) and *S. oblongifolia* (0.80 ± 0.31 N/m) (Dunn's method $P > 0.05$). The latter two *Salvinia* species show no significant differences (Dunn's method $P > 0.05$).

4 Discussion

4.1 Influence of surface structure on stiffness

In the investigated *Salvinia* species, the morphology of the surface microstructures strongly differs in height, shape and diameter of emergences and number of trichomes. Moreover, the shape of the surface structures was very different, which is reflected in varying relationships between emergence and trichome lengths, as well as in different numbers of trichomes originating from a single emergence.

Due to the different overall height of the structures, we used different indentation depths in different species. The estimated stiffness of the trichome arrays shows a dependence on the aspect ratio of the overall structure (Fig. 5). The higher the aspect ratio of the structure is, the more deformable the array of the structures is. The softest microstructure arrays were found in *S. cucullata*, the only species with solitary scattered trichomes without emergences. These single trichomes are very flexible and, therefore, do not provide much resistance against external load. Also the microstructures of *S. molesta* are very pliant. The eggbeater-shaped trichome groups have a very long emergence which most likely contributes to the high deformability of the total structure. We found the lowest aspect ratio in *S. oblongifolia*, the species with the stiffest
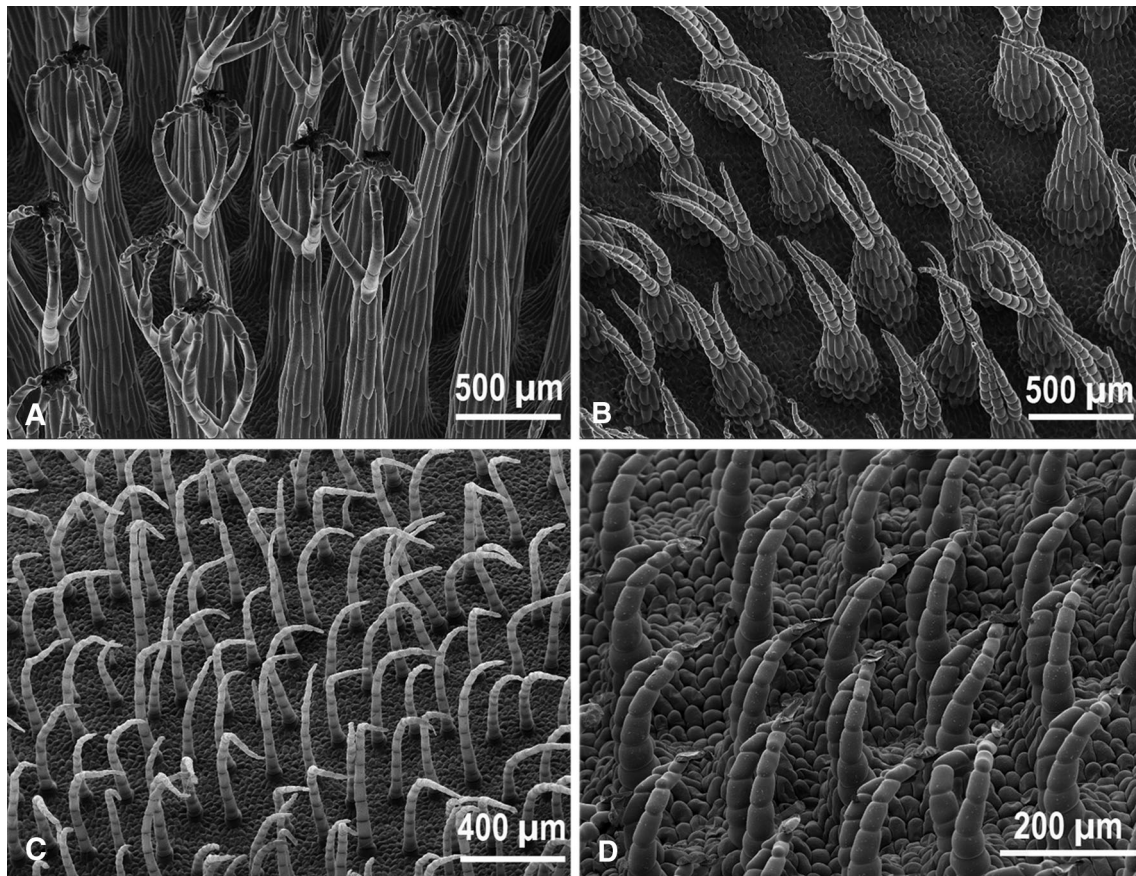
Fig. 3 SEM images of the same four *Salvinia* species: **a** *S. molesta*, **b** *S. minima*, **c** *S. cucullata*, **d** *S. oblongifolia*
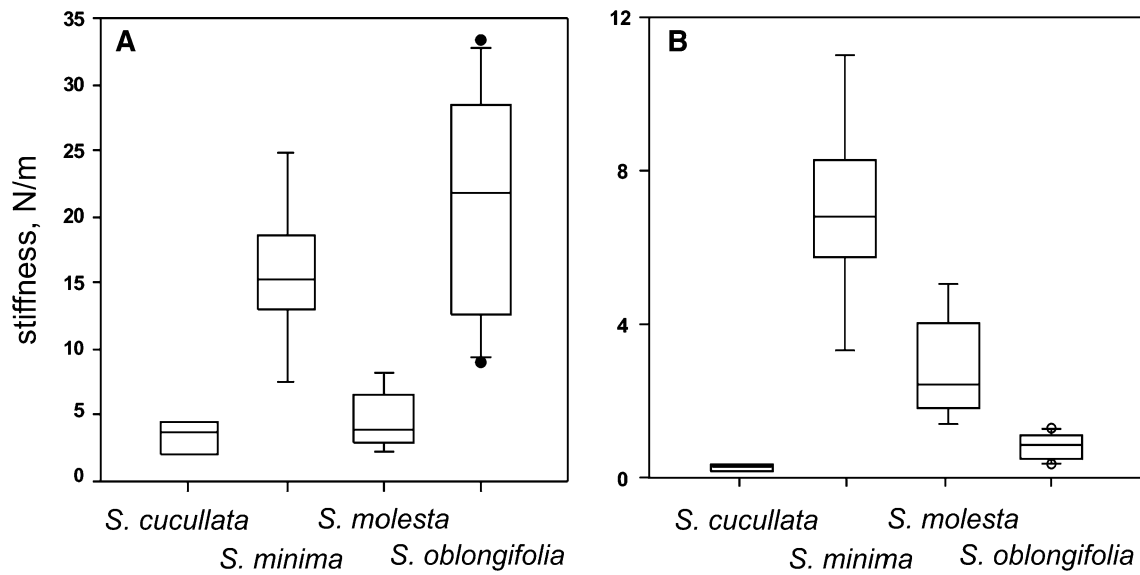

Fig. 4 a Total stiffness [N/m] of the trichome array on the leaves *S. cucullata*, *S. minima*, *S. molesta* and *S. oblongifolia*. **b** Stiffness [N/m] normalized per trichome group for the same *Salvinia* species
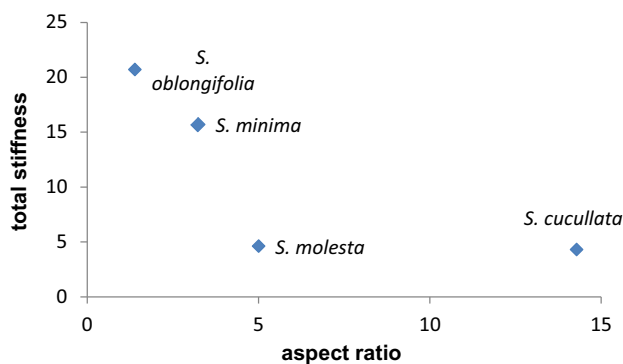
Fig. 5 Total stiffness in relation to the aspect ratio (length–width quotient) of the trichome groups in the four investigated *Salvinia* species. The quotient was calculated from the total length of the structures and the diameter of the base (emergence)

microstructures. *S. minima* show a relatively high stiffness for its length–width quotient. In *S. minima* and *S. oblongifolia*, single trichomes and their emergences are almost of the same length. The measured stiffness is likely a result of the complex morphological structure, but may also depend on the indentation depth. These stiffness data of trichome groups of *S. molesta* are in good agreement with the stiffness measured for the case of individual trichomes of *Salvinia molesta* [11], measuring elastic force constants of the order of 2.1 ± 0.2 N/m per trichome by performing direct measurements on individual trichomes. In comparison, measurements of human hairs showed a much lower stiffness of 0.2 ± 0.001 mN/m [11].

4.2 Influence of structure and stiffness on air retention capability

The average duration of air retention shows considerable differences between the investigated *Salvinia* species,
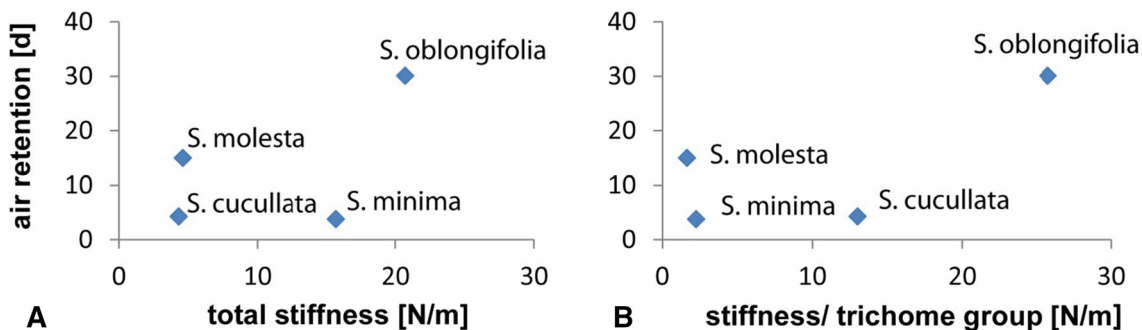
which varied between 4 and 30 days [18]. Despite their different shape, the contact angle of the investigated *Salvinia* species (160.5°–162.3°) is very similar [19] and can therefore be excluded as an important impact factor for the duration time of air retention. Stiffness has been assumed to have an impact on the duration time of air retention, but our results suggest that this is not a primary factor as well. The species with the stiffest microstructures (*S. oblongifolia*) was the species with the longest duration of air retention. However, there was no general direct trend in the relationship between the time of air retention and the stiffness of the trichome arrays (Fig. 6a) and the stiffness per trichome group (Fig. 6b). This result can be explained by the stronger contribution of the particular shape, dimension and distribution of individual trichomes rather than the stiffness of the trichome arrays to the air-retaining function of the trichome coverage.

It seems that in spite of the strong difference in stiffness of trichome arrays in different *Salvinia* species, other parameters such as morphology and density of individual trichomes are more important for the duration of air retention. In the case of *S. molesta*, the species with the hydrophilic tips on the otherwise superhydrophobic microstructures (Salvinia Paradox), our results confirm and quantify the previously presumed soft and deformable properties of the arrays [8]. Our measurements show stiffness between 4 and 21 N/m of the *Salvinia* species. This should support the pronounced ability to pin the air–water interface to the hydrophilic tips of the eggbeater shape. When the tips of the microstructures are able to deform and adapt to the oscillating air–water interface while turbulences occur, such adaptability with some damping should reduce the probability of interface separation from the tips of the trichomes and therefore lead to the air film stabilization.

Fig. 6 Air retention time in relation to (**a**) total stiffness and (**b**) stiffness per trichome group

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