RESEARCH ARTICLE

# Wind induces variations in spider web geometry and sticky spiral droplet volume

Chao-Chia Wu[1], Sean J Blamires[2] Chung-Lin Wu[3], and I-Min Tso[1,2]*

[1]Department of Life Science, National Chung-Hsing University, Taichung 40227, Taiwan

[2]Department of Life Science, Tunghai University, Taichung 40704, Taiwan.

[3]Center for Measurement Standards, Industrial Technology Research Institute, Hsinchu

30011, Taiwan.

*Author for correspondence (spider@thu.edu.tw)

1

# SUMMARY

**Trap building by animals is rare because it comes at substantial costs. Using materials with properties that vary across environments maintains trap functionality. The sticky spiral silks of spider orb webs are used to catch flying prey. Web geometry, accompanied by compensatory changes in silk properties, may change across environments to sustain web functionality. We exposed the spider *Cyclosa mulmeinensis* to wind to test if wind-induced changes in web geometry are accompanied by changes in aggregate silk droplet morphology, axial thread width or spiral stickiness. We compared: (i) web catching area, (ii) length of total silks, (iii) mesh height, (iv) number of radii, (v) aggregate droplet morphology and (vi) spiral thread stickiness, between webs made by spiders exposed to wind with those not exposed to wind. We interpreted co-variation in droplet morphology or spiral stickiness with web capture area, mesh height or spiral length as the silk properties functionally compensating for changes in web geometry to reduce wind drag. Wind-exposed *C. mulmeinensis* built webs with smaller capture areas, shorter capture spiral lengths, and more widely spaced capture spirals, resulting in the expenditure of less silk. Individuals that were exposed to wind also deposited larger droplets of sticky silk but the stickiness of the spiral threads remained unchanged. The larger droplets may be a product of greater investment in water, or low molecular weight compounds facilitating atmospheric water uptake. Either way droplet dehydration in wind is likely to be minimized.**

## INTRODUCTION

The use of traps to capture prey has evolved independently in arachnids, larval dipterans, trichopterans and neuropterans (Viviani et al., 2002; Hansell, 2005; Scharf et al., 2011). The principal advantage of building a trap is that once the trap is built prey is captured at minimal expense of foraging time and energy (Lucas, 1985; Willis et al., 2011). The principal costs of trap building are the pre-investment in the raw materials and the time and energy required to build, repair and maintain it (Craig, 2003; Hansell, 2005; Hansell and Ruxton, 2013). Thus, for traps to be profitable they must be comprised of readily synthesizable materials that sustain their functionality for prolonged periods (Hansell, 2005). Additionally, as the traps may be exposed to spatially and temporally variable environments (Scharf et al., 2011) it may be imperative that the physical and chemical properties of the building materials are variable across different environments (Hansell, 2005; Liao et al., 2009; Blamires et al., 2012; Hansell and Ruxton, 2013).

Silk is a material consisting predominantly of protein that is secreted by some organisms. It has been a key innovation facilitating the building of aerial traps among spiders, larval dipterans and trichopterans (Craig, 1997, 2003). The type of silk used differs between spiders, dipterans and trichopterans but they both serve the function of absorbing the energy of moving prey (Craig, 2003; Hansell, 2005; Willis et al., 2011). The use of sticky silk by spiders is considered a key innovation that facilitated the radiation of >9,000 species of web builders (Craig, 2003; Blackledge et al., 2009). Two types of sticky silks are used by orb-web spiders: (1) Cribellar sticky silk, a "wooly" silk used by the Deinopoidea, who have retained these ancestral capture silks, and (2) the more derived

ecribellar sticky silk, which is used by most other orb-web spiders (Blackledge et al., 2009).

Ecribellar sticky silk, or spiral silk, is secreted from a single flagelliform spigot and two flanking

aggregate spigots to form a spigot "triad" (Opell et al., 2011a). The viscous aggregate silk and solid

flagelliform fibers are secreted simultaneously so the flagelliform fiber is spun doused with viscous

silk (Sahni et al., 2011). Upon spinning, surface tension forces result in coalescence of the viscous

aggregate silk into droplets along the flagelliform thread so as to superficially resemble beads along

a string (Kane et al., 2010; Opell and Hendricks, 2010; Sahni et al., 2011, Sahni et al., 2012).

Aggregate silk is a mixture of dissolved silk proteins, glycoprotein and low molecular weight

compounds (LMWCs) (Vollrath and Tillinghast, 1991; Vollrath and Knight, 2001). The stickiness of

spiral silk is conferred by the concentration of glycoprotiens within the aggregate silk and the

extensibility of the flagelliform threads (Bonthrone et al., 1992; Sahni et al., 2010; Opell et al.,

2011b). The well hydrated aggregate silk increases flagelliform thread extensibility by

supercontracting (plasticization of the thread accompanied by a loss of protein alignment) the spiral

thread (Guinea et al., 2010). The highly extensible flagelliform thread dissipates much of the kinetic

energy when prey strike the spirals (Opell and Hendricks, 2010; Sahni et al., 2010; Tarakanova and

Buehler, 2012), while the glycoproteins in the aggregate silk serves to retain the prey once it has

been captured (Sahni et al., 2011; Tarakanova and Buehler, 2012). The LMWCs facilitate water

uptake from the environment to the viscous droplets, thereby keeping the sticky spirals hydrated

(Vollrath et al., 1990; Townley et al., 1991; Edmonds and Vollrath, 1992; Sahni et al., 2010); the

greater the LMWC concentration the greater the uptake of water from the atmosphere (Sahni et al.,

2011). The LMWCs also influence spiral stickiness by keeping the glycoproteins soft and "tacky", a property implicit in the retention of prey (Sahni et al., 2011; Opell et al., 2011b).

Spider orb webs are placed into spatially and temporally unpredictable environments, which may induce physical and chemical property changes in the silks (Blamires et al., 2012). Orb-web spiders, accordingly, often build webs with varying geometries across environments (Sandoval, 1994; Vollrath et al., 1997; Liao et al., 2009; Blamires et al., 2011). Orb-web spiders may modulate the performance of their webs by modifying one or a combination of the following parameters: (i) size of the capture area, (ii) mean spacing between sticky spiral threads, or mesh height (although spider webs are not "meshed" we use this term to represent capture spiral spacing because it is the most commonly used term among comparable studies), or (iii) the number of spirals or radii used (Sandoval, 1994; Vollrath et al., 1997; Tso et al., 2007; Liao et al., 2009; Blamires, 2010). Since mesh height influences the size of the prey that can be retained by orb webs (Herberstein and Heiling, 1998; Blackledge and Zevenberg, 2006), mesh height may change in response to expected changes in the type or size of prey present (Sandoval, 1994; Herberstein and Heiling, 1998; Blamires, 2010; Blamires et al., 2011). Environmentally-induced changes in the stickiness of the spirals (Opell et al., 2011a; Sahni et al., 2011) may, however, also be associated with changes in mesh height as a means to maintain the ability to effectively absorb the kinetic energy of intercepted prey (Blackledge and Hayashi, 2006).

Wind is an example of an unpredictable environmental parameter that spiders may face and must be accounted for by an adjustment in web building and/or prey catching behaviors (Vollrath et

5

al., 1997; Liao et al., 2009; Turner et al., 2011; Cranford et al., 2012). Indeed, orb-web spiders that have been exposed to wind substantially alter the geometry of their webs upon exposure to strong wind. Such geometric alterations are characterized by a reduction in the number of radial threads in their webs, smaller web capture areas, greater spacing between spirals and a reduction in the length of the sticky spiral thread (Vollrath et al., 1997; Liao et al., 2009). In the orb-web spider, *Cyclosa mulmeinensis*, exposure to wind also induces the deposition of considerably stiffer radial threads, which, combined with a reduction in web capture area and spiral thread length, serves to reduce wind drag on the web and minimize tearing (Liao et al., 2009). The likely cost of such changes in web geometry and silk properties is a reduction in the number of prey that can be effectively caught (Blackledge and Zevenberg, 2006; Blamires et al., 2011; Tarakanova and Buehler, 2012).

As evaporative water loss across organic membranes substantially increases with the rate of air flow (Mellanby, 1934; Willmer et al., 2000), water is lost from the sticky spiral silks more readily when they are in wind than when they are in still air. It thus seems that an additional cost of placing an orb web into wind is a high risk of silk property variation through dehydration. Accordingly, in order to maintain the efficiency at which the web absorbs and dissipates the kinetic energy of intercepted prey and retains any intercepted prey, orb-web spiders exposed to windy conditions should use silk with some degree of resistance to dehydration. For instance, wind exposed spiders might reduce dehydration from the spiral threads by secreting aggregate silk with specific properties. This may be accomplished by either secreting aggregate silks that form larger droplets so the proportion of droplet surface area that is exposed to wind is reduced, or aggregate silks with

droplets of greater LMWC concentration; thus shifting the droplet-atmosphere water exchange

equilibrium (Vollrath et al., 1990; Townley et al., 1991; Edmonds and Vollrath, 1992; Opell et al.,

2011a).

Spiders make decisions about the geometry and properties of a web they will build at a

particular location based on their previous experiences at the location (Venner et al., 2000; Blamires,

2010). These experiences are likely to be assessed by their prey capture experiences (Heiling and

Herberstein, 1999; Venner et al., 2000; Mestre and Lubin, 2011), or by the quality and quantity of

specific stimuli (Blamires et al., 2011). The stimuli received may include a combination of

web-borne tactile stimuli detected by the strain sensitive slit sensilla, or changes in air pressure

detected by the pressure sensitive trichobothriae (Barth, 2002; French et al., 2002). Exposure to

strong wind is likely to influence the type and frequency of prey caught and the tactile and airborne

stimuli received by spiders. Accordingly, spiders may use a combination of these cues to make

decisions about the geometric features of its web when building at a location where it has

experienced strong wind.

Here we tested the role of wind in inducing concurrent web geometric and spiral thread

property variations using the dust spider, *Cyclosa mulmeinensis*, an orb-web spider that regularly

builds its webs in strong winds (Liao et al., 2009; Blamires et al., 2010). We measured and

compared the geometries of *C. mulmeinensis* webs with and without prior wind exposure and

measured and compared the viscous silk droplet morphology and spiral thread stickiness. We

interpreted across treatment changes in droplet volume, surface area to volume ratio, or thread

7

stickiness, congruent with variations in web capture area, mesh height, or sticky spiral length, as *C. mulmeinensis* functionally varying their silk properties to account for changes in web geometry. Conversely, we interpreted a lack of congruency between changes in droplet morphology, thread stickiness and changes in web capture area, mesh height, or spiral length, as implying that when *C. mulmeinensis* builds webs in locations exposed to wind they compromise between prey capture efficiency, web dehydration, and protection from wind drag.

## METHODS

We collected adult female *Cyclosa mulmeinensis* from Huwei, Yunlin County, Taiwan (120°22'31.47" E, 23°38'57.54" N) all year round during 2011. Spiders and webs were collected by placing two circular wooden frames (diameter = 200 mm) with superglue around their rims on either side of a web and moving them toward each other carefully until they touched. The frames were pressed firmly together in order to stick them to each other. Any web components lying outside the frames were burnt away using a stick of incense. We temporarily removed each spider from the web to measured its mass (± 0.1mg) using an electronic balance (PJ300; Mettler Toledo, Greifensee, Switzerland). The spiders were returned to their webs and taken back to the laboratory within the frames and acclimated in the laboratory on their frame-mounted webs under a 12: 12 h light-dark cycle for three days. We fed them one *Drosophila melanogaster* each day and lightly sprayed the webs with tap water before destroying the webs.

## Experiments

*Cyclosa mulmeinensis* is a small orb-web spider (adult body length < 6 mm), so making repeated

measurements on the same individual can be stressful and this may confound the experiments. We thus randomly divided 120 spiders into two groups; a pretreatment and a treatment group ($n$ = 60 in each). The individuals in the pretreatment group were further randomly divided into two subgroups ($n$ = 30 in each subgroup); designated P1 and P2. These spiders were given three days to build a web on their circular frames, after which web geometry (see 'Web geometry measurements') and spiral properties (see 'Droplet morphology measurements' and 'Thread stickiness measurements') were measured and compared. These web and silk measurements served as the pre-treatment standards for the following experiments.

Spiders in the treatment group were divided into a wind exposed subgroup (W subgroup) and an unexposed subgroup (N subgroup) ($n$ = 30 in each subgroup). We subjected individuals in the W subgroup to wind of constant speed (1.1 ms$^{-1}$) over seven days while on their webs, while individuals on webs in the N subgroup were placed in the same laboratory as the W subgroup for the same seven days but not subjected to wind. The wind was generated by 120 x 120 mm electric fans (Cooler Master; AREC Peripherals, Inc., Taipei, Taiwan) placed 400 mm from the spiders' dorsum. Relative humidity and temperature data loggers (Hobo U23, Pro v2, Onset, USA) were set up in the laboratory beside six representative webs from each treatment to make sure that relative humidity (W = 62.81 ± 1.06 %, N = 62.57 ± 0.94%, ANOVA: $F$ = 0.03, d.f. = 1,12, $P$ = 0.86) and temperature (W = 18.81°C ± 0.10, N = 18.75 ± 0.11°C, ANOVA: $F$ = 0.16, d.f. = 1,12, $P$ = 0.69) did not differ significantly between the locations where spiders in the W and N subgroups were placed. After seven days the experiment was terminated and we destroyed the webs of all individuals.

Spiders built new webs on their frames within 24 h, upon which we immediately measured and compared between subgoups: (i) web geometric parameters: catching area, total (sticky and non-sticky) silk length, total spiral thread length, mesh height and the number of radii, (ii) droplet morphology and (iii) spiral stickiness, from samples of spiral threads from each web as follow.

## Web geometry measurements

We counted the number of radii and sticky spirals in each web along four cardinal directions (up, down, left and right). We then measured the hub and total radius of the upper and lower portions of each web. These variables were used to calculate: (1) catching area, (2) total silk length, (3) total spiral length and (4) mesh height, using the formulae (Herberstein and Tso, 2000):

$$\left[ \frac{\pi(r_{au})^2}{2} - \frac{\pi(Hr_u)^2}{2} \right] + \left[ \frac{\pi(r_{al})^2}{2} - \frac{\pi(Hr_l)^2}{2} \right]$$

where

$$r_{au} = \frac{r_u + (d_h / 2)}{2}$$

and

$$r_{al} = \frac{r_l + (d_h / 2)}{2}$$

where $r_u$ is the radius of the upper portion of the web, $r_l$ is the radius of lower portion of web, $d_h$ is the width of web, $Hr_u$ is the radius of upper portion of hub and $Hr_l$ is the radius of lower portion of hub.

Total silk length was estimated from the formula (Tso et al., 2007):

Total silk length $= \pi \left( \overline{X}_{Rweb} + \overline{X}_{Rhub} \right) \overline{X}_{spiral} + \left( \overline{X}_{Rweb} + \overline{X}_{Rhub} \right) \overline{X}_{radii}$

10

where $\overline{X}_{Rweb}$ is the average radius of web, $\overline{X}_{Rhub}$ is the average radius of hub, $\overline{X}_{spiral}$ is the average

number of sticky spirals and $\overline{X}_{radii}$ is the average number of radii. Because the total silk length was

the sum of radii and sticky spiral length in a web, we used the following formula for length of sticky

spiral to calculate the total spiral length:

$$\text{Total spiral length} = \pi\left(\overline{X}_{Rweb} + \overline{X}_{Rhub}\right)\overline{X}_{spiral}$$

The mesh height was calculated by the formula (Tso et al., 2007):

$$\text{Mesh height} = \frac{(r_u + r_l) - (Hr_u + Hr_l)}{S_u + S_l - 2}$$

where $S_u$ is the number of sticky spirals in upper half of web and $S_l$ is the number of sticky

spirals in the lower half of the web.

## Droplet morphology measurements

We collected three spirals from each web between two 25 mm x 35 mm plastic frames with

double-sided adhesive tape around their 5 mm wide border. The frames were placed in front of and

behind a selected set of spirals and moved together until they touched securing the spirals within.

The spirals mounted within the frames were freed from the surrounding web using a hot soldering

iron. The frames always contained at least five spirals rows so the ensuing measurements were

replicated five times per sample. Spirals were collected from the lower portion of the web on all

occasions.

The spiral-containing frames were gently placed onto parallel matchsticks placed 20 mm apart

on a microscope slide so that the threads and their droplets had no contact with any surface that

could distort their shape. We viewed and photographed the spirals under 100x and 1000x

magnification using a polarized light microscope (Olympus BX-50, Tokyo, Japan) connected to a

digital camera (UC-Series, Nikon, Tokyo, Japan). From the photographs we calculated the number

of gluey silk droplets per 0.5 mm length of flagelliform thread (DV/0.5mm), the width of the

flagelliform thread (SD), and measured the length and width of nine randomly selected droplets

using VIS Plus (Liion Opto-Electronics Technology, Taichung, Taiwan) imaging software. From the

latter measurements, we determined the mean droplet volume (DV) using the formulae (Opell and

Hendricks, 2007):

$$DV = \frac{2\pi(h)^2 b}{15}$$

where $h$ is the width of the droplet and $b$ is the length of the droplet. We then calculated the average

spacing between droplets (DS) and DV/0.5mm according to Opell and Hendricks (Opell and

Hendricks, 2007). Since it is an indicator of water loss potential from sticky silk, we calculated the

surface area to volume ratio of the droplets by first calculating droplet surface area using the

formula:

$$\text{Droplets surface area} = \frac{4\pi hb}{3}$$

The droplets surface area to volume ratio (DSAVR) was then calculated as the droplet surface area

divided by droplet volume. All measurements were done as soon as possible after collection and the

treatments were sampled in random order so that the time taken after web building to view the

droplets had minimal affect on the measurements made.

## Thread stickiness measurements

We used 11 x 11 mm U-shaped cardboard frames (Agnarsson and Blackledge, 2009) to collect three

spiral threads from the lower portion of each web. We lightly touched the tips of the U frame to a

length of spiral thread. Threads within U frames were freed from the rest of the web using a hot iron

and reinforced onto the frame border attachment sites using a drop of Elmer's glue. We placed the

top (i.e. so the open end of the U faced downward) of the frame within the uppermost grips of a

Nano Bionix tensile tester (MTS Systems Corporation, Oakridge TN, USA) and a 6 x 2 mm

stainless steel stage was mounted securely onto a pin using super glue and placed in the lowermost

grips. We then lowered the card at 0.01 mms$^{-1}$ until the thread touched the stage. The specimen was

held in position for 60 seconds, allowing the thread to adhere to the stage, before the thread was

pulled up at 1 mms$^{-1}$ until the thread detached from the stage. The force ($\mu$N) required to pull the

thread off the stage was measured and indicated the stickiness of the thread (Opell, 1989). We

repeated this procedure 10 times, using a different part of the stage each time, for each of the three

threads from each web to obtain an average per thread. The stage was cleaned with alcohol before

testing each thread. As with the measurements of droplet morphology, all measurements were done

as soon as possible after collection and the treatments were sampled in random order. All

measurements of droplet morphology and thread stickiness were made at room temperature (~20°C)

and relative humidity (~60%).

**Analyses**

We assessed whether the web and spiral properties differed between the P1 and P2 subgroups using

analyses of variance (ANOVA). We assessed whether spider weight and spiral features differed

between the pretreatment subgroups, and the W and N treatment subgroups using ANOVA. We used

13

multivariate analyses of variance (MANOVA) to determine whether web geometric parameters, thread stickiness and/or droplet morphology differed between the P1 and P2 pre-treatment subgroups and between the W and N treatment subgroups. When a MANOVA showed a significant difference we performed individual ANOVAs on each of the variables to ascertain the significantly differing variable across the treatments. We performed Kolmogorov-Smirnov tests to assess the normality of the data prior to all analyses. $Log_{10}$ transformations were done on data that failed the test ($P < 0.05$), which normalized it ($P > 0.05$) on all occasions. Variations in thread droplet morphology, spacing or stickiness with web capture area, mesh height or spiral length across treatments were used to interpret whether, and to what extent, silk properties compensated for changes in web geometry.

## RESULTS

Web geometric parameters did not differ significantly between P1 and P2 pretreatment subgroups (MANOVA: $\lambda = 0.92$, $F = 0.95$, d.f. = 5,55, $P = 0.45$; Table 1), so we were confident that web geometric parameters were homogeneous among pre-treated spiders. Web geometry differed significantly between W and N subgroups (MANOVA: $\lambda = 0.79$, $F = 5.07$, d.f. = 5,96, $P < 0.001$). We therefore used ANOVAs to examine the individual variables and found that capture area ($F = 4.35$, d.f. = 1,100, $P = <0.001$), total silk length ($F = 10.31$, d.f. = 1,100, $P = 0.004$) and total spiral length ($F = 12.55$, d.f. = 1,100, $P = 0.001$) of the webs of spiders in the W subgroup were significantly smaller than those of spiders in the N subgroup (Fig. 1A-C). Moreover, the mesh height in the webs of spiders in the W subgroup were significantly larger ($F = 11.40$, d.f. = 1,100, $P$

= 0.001) than those in the webs of spiders in the N subgroup (Fig. 1D).

No features of the sticky spirals differed between the P1 and P2 pre-treatment subgroups (MANOVA: $\lambda = 0.82$, $F = 1.07$, d.f. = 7,35, $P = 0.40$; Table 2), so we were confident that droplet morphology and spiral stickiness were homogeneous among pre-treated spiders. Among the treatment subgroups, we found that the DV ($F = 5.43$, d.f. = 1,100, $P = 0.02$), DVP/0.5mm ($F = 11.79$, d.f. = 1,100, $P <0.001$) and droplet surface area ($F = 5.21$, d.f. = 1,100, $P = 0.02$) in the W subgroup were significantly greater than those of the N subgroup (Fig. 2C-E) and the DSAVR in the W subgroup was significantly smaller ($F = 3.94$, d.f. = 1,100, $P = 0.05$) than that of N subgroup (Fig. 2F). DN, DS and SD, however, did not significantly differ ($P > 0.05$) between the subgroups (Fig. 2A,B,G). Despite these morphological changes to the droplets, the stickiness of the capture spirals did not differ significantly between any of the pre-treatment ($F = 1.22$, d.f. = 1,35, $P = 0.31$; Fig. 3A) or treatment ($F = 1.64$, d.f. = 1,36, $P = 0.21$; Fig. 3B) subgroups.

**DISCUSSION**

We found that recent exposure to wind induces the orb-web spider *Cyclosa mulmeinensis* to build webs with smaller capture areas and spiral thread length, larger mesh heights, and use less silk, than they would in the absence of wind exposure. These changes in web geometry are consistent with findings for these and other orb-web spiders when exposed to wind (Vollrath et al., 1997; Liao et al., 2009), which are predicted to constitute a response to reduce wind-induced damage to webs (Liao et al., 2009; Cranford et al., 2012). *Cyclosa mulmeinensis* increases radial thread stiffness upon exposure to wind (see Liao et al., 2009), and this may enable the radii to take more of the burden of

prey impact absorption away from the spiral threads (Lin et al., 1995; Cranford et al., 2012; Sensenig et al., 2012), thus, partially offsetting the effects on prey capture performance induced by the shorter spiral thread.

We found that the spiral threads of spiders exposed to wind had a similar number of, albeit larger, aggregate (viscous) droplets compared to those of spiders that had not been exposed to wind. However, the larger droplets did not correspond to the spiral threads being any stickier. The lack of change in thread stickiness despite an increase in droplet size, a reduction in spiral length, and an increase in mesh height, across treatments is inconsistent with our *a priori* prediction of aggregate silk properties functionally compensating for any necessary changes in web geometry. A likely consequence of the larger glue droplets was a reduction in droplet surface area to volume ratio, which may serve to reduce evaporative water loss from the droplets. There seems to be two possible explanations for the larger droplets in wind-exposed spiders. One is that there is a trade-off between the investment in glycoprotein and LMWCs with wind exposure favouring LMWCs, perhaps for greater desiccation resistance. The other possibility is that the larger droplets of wind exposed spiders have the same amount of glycoprotein as the control group, but greater amounts of the LMWCs. The consequence of this would be that at a given humidity the wind exposed droplets would be more fully hydrated and more prone to loosing adhesion due to the over lubrication phenomena documented by Sahin et al. (Sahni et al., 2011). Thus, it seems that enhanced droplet hydration or water retention due to larger surface to volume ratio would be at odds with optimal glycoprotein hydration and adhesive optimization.

Empirical studies (Herberstein and Heiling, 1998; Blackledge and Zevenberg, 2006; Blamires et al., 2011; Sensenig et al., 2012) and computer simulations (Lin et al., 1995; Cranford et al., 2012; Tarakanova and Beuhler, 2012) demonstrate that many aspects of web geometry are important predictors of prey capture performance in orb webs. Indeed, the combined influence of geometry and silk properties on prey capture performance of spider webs (Lin et al., 1995; Sensenig et al., 2012; Cranford et al., 2012) explains why spiders that switch diets vary both the geometry of their webs and the properties of their silks (Tso et al., 2005, 2007; Blamires et al., 2011; Blamires and Tso, 2013). These variations include changes to mesh height, number of radials and spiral length, which affect the number, size, shape, mass and kinetic energy of the prey that can be effectively caught (Blackledge and Zevenberg, 2006; Sensenig et al., 2010, 2012; Blamires et al., 2011; Cranford et al., 2012). The geometric differences that we found between webs built by spiders that had been exposed to wind compared to those that had not been exposed to wind are likely to correspond with variation in prey capture performance of the respective webs (Cranford et al., 2012). We predict that the geometry of the webs built by spiders that had been exposed to wind are likely to ultimately accommodate fewer prey than would the geometry of the webs built by the spiders that had not been exposed to wind (Herberstein and Heiling, 1998; Blackledge and Zevenberg, 2006; Blamires et al., 2011).

The radii and spiral threads are the web components that bear much of the burden of absorbing the kinetic energy of prey on impact (Cranford et al., 2012; Sensenig et al., 2012; Tarakanova and Beuhler, 2012). Accordingly, they need to be incorporated into webs under specific tensions (Craig,

17

2003; Sensenig et al., 2012). Too much tension will mean that prey, depending on its size and flight velocity, will either fly through the web or bounce off the web, a phenomena known as the 'trampoline effect' (Craig, 2003; Blackledge and Hayashi, 2006; Kelly et al., 2011; Sensenig et al., 2012). Moreover, localized tearing becomes increasingly likely in strong wind if webs are under excessive tension. Accordingly, radii tension might be loosened or fewer radial threads used when webs are in wind (Lin et al., 1995; Aoyanagi and Okumura, 2010; Cranford et al., 2012). We found that *C. mulmeinensis* used fewer radii, although we did not directly measure radii tension herein. The longer the spiral, the more a web is likely to experience sagging under wind drag (Lin et al. 1995; Sensenig et al., 2010). This explains why *C. mulmeinensis* uses shorter spiral threads with wider spaces between spiral turns when they build their webs upon exposure to wind (Liao et al., 2009).

We found, in addition to variations in web geometry, that webs built by *C. mulmeinensis* that had been exposed to wind have larger aggregate silk droplets. Nevertheless, the larger droplets did not affect the stickiness of the spiral thread. The extensibility of the flagelliform thread principally influences the stickiness of the spiral threads (Opell and Hendricks, 2010; Sahni et al., 2010; Tarakanova and Buehler, 2012). It seems that the larger droplets had no affect on flagelliform thread extensibility via supercontraction of the flagelliform thread; if it had we would have found significant differences in thread stickiness between treatment subgroups. The production of larger droplets could have been a result of the secretion of aggregate silk with greater water content forming larger droplets. This would suggest that there were reductions in the glycoprotein and

LMWC concentrations. We consider this scenario unlikely as the droplets in the W and N treatment group would have had different surface tensions which would have been identifiable by the droplets of the W and N group differing in their flatness under magnification (Opell and Schwend, 2007); which we never observed. In addition, windy conditions are typically drying conditions (Willmer et al., 2011), so it seems counterintuitive that under these conditions a relatively small spider would expend more water on silk, particularly when the threads produced under these conditions are no stickier and no more likely to capture enough prey to recover their water investment. A more plausible explanation, but one requiring confirmation, is that the aggregate silks of the spiders exposed to wind had greater concentrations of LMWCs, so took in water more from the atmosphere post-deposition (Townley et al., 1991; Opell et al., 2011a).

Despite the production of larger aggregate silk droplets and the maintenance of consistent spiral stickiness across treatments, functional augmentation of spiral threads in *C. mulmeinensis* webs was to be expected because of the probable dehydrating influence of persistent strong winds on the silk. Spiral silks may lose their stickiness over time due to water loss to the environment (Edmonds and Vollrath, 1992; Opell et al., 2011a). The production of larger aggregate silk droplets by *C. mulmeinensis*, regardless of whether they were deposited with a greater water content or gain water from the atmosphere, probably serve to reduce the droplet surface area to volume ratio and curtail evaporative water loss to some extent. Furthermore, if higher concentrations of LMWCs were added to the viscous silks of wind-exposed spiders it would potentially facilitate silk rehydration when the wind subsides (Edmonds and Vollrath, 1992; Sahni et al., 2010).

19

Although variation in the geometry of *C. mulmeinensis* webs has the benefit of reducing wind drag and web damage when in strong wind (Liao et al., 2009), we interpreted the lack of congruent changes in droplet morphology, thread stickiness and web geometry as negatively affecting the prey capture performance of webs of spiders exposed to wind. Furthermore, in strong wind prey fly in multiple directions and at a multitude of speeds and debris may regularly strike and deposit onto webs, so the responsiveness of orb-web spiders to prey capture is severely hampered (Turner et al., 2011). Despite these austere consequences, *C. mulmeinensis* consistently builds webs in windy locations (Liao et al., 2009; Blamires et al., 2010). We thus expect that they use means other than varying their silk properties to offset the constraints placed on prey capture performance.

In certain circumstances, aggregating webs may enhance the prey capture efficiency of spider webs compared to webs in isolation. This is because prey bounce off successive webs before eventually being captured by a centrally placed web. This phenomenon has been called the 'ricochet effect' (Uetz et al., 1989) and competition for the central position has been called 'shadow competition' (Rao, 2009). Shadow competition may provide a runaway selective mechanism on the size of spider web aggregations (Uetz et al., 1989; Rao, 2009; Blamires et al., 2010; Mestre and Lubin 2011). It might, accordingly, be desirable for *C. mulmeinensis* to aggregate their webs when strong winds are considered likely. Indeed, aggregations of *C. mulmeinensis* webs are often found at locations where the wind is consistently strong, e.g. when webs are placed along the foreshore (Blamires et al., 2010). Aggregating by *C. mulmeinensis* thus could be to take advantage of 'ricochet effects' under these circumstances.

To summarize, variations in orb web geometry and spiral silk properties provide a means to maintain web function in variable environments. We found that *C. mulmeinensis* exposed to wind built webs with smaller capture areas and spiral thread lengths, but wider mesh heights, and used less silk, than those that had not been exposed to wind. We, however, found no change in the stickiness of spiral threads, despite significant differences in the size of the aggregate silk droplets. The larger droplets seem to be a consequence of either a greater water investment in aggregate silk or a greater investment in LMWCs facilitating greater water uptake into the droplets from the atmosphere. In either event dehydration of the silks when in wind is at least partially mitigated. There is likely to be a compromise between prey capture efficiency and a reduction of wind drag and dehydration in the webs of wind exposed spiders. It seems plausible that *C. mulmeinensis* aggregate their webs to take advantage of 'ricochet effects', thereby enhancing their prey capture efficiently when building webs at windy locations.

## ACKNOWLEDGEMENTS

## REFERENCES

**Agnarsson, 1. and Blackledge, T. A.** (2009). Can a spider web be too sticky? Tensile mechanics constrain the evolution of capture spiral stickiness in orb-web spiders. *J. Zool.* **278,** 134-140.

**Aoyanagi, Y. and Okumura, K.** (2010). Simple model for the mechanics of spider webs. *Phys.*

*Rev. Lett*. **104,** 38102-1-38102-4.

**Barth, F. G.** (2002). Spider senses— technical perfection and biology. *Zoology* **105,** 271-285.

**Blackledge, T. A. and Hayashi, C. Y.** (2006). Silken toolkits: biomechanics of silk fibers spun by

the orb web spider *Argiope argentata* (Fabricius 1775). *J. Exp. Biol*. **209,** 2452-2461.

**Blackledge, T. A. and Zevenbergen, J. M.** (2006). Mesh width influences prey retention in spider

orb webs. *Ethology* **112,** 1194-1201.

**Blackledge, T. A., Scharff, N., Coddington, J. A., Szuts, T., Wenzel, J. W., Hayashi, C. Y. and**

**Agnarsson, I.** (2009). Reconstructing web evolution and spider diversification in the

molecular era. *Proc. Nat. Acad. Sci. USA* **106,** 5229-5234.

**Blamires, S. J.** (2010). Plasticity in extended phenotypes: orb web architectural responses to

variations in prey parameters. *J. Exp. Biol*. **213,** 3207-3212.

**Blamires, S. J. and Tso, I. M.** (2013). Nutrient-mediated architectural plasticity of a predatory trap.

*PLoS ONE* **8,** 54558.

**Blamires, S. J., Chao, I. C., Liao, C. P. and Tso, I. M.** (2011). Multiple prey cues induce foraging

flexibility in a trap-building predator. *Anim. Behav*. **81,** 955-961.

**Blamires, S. J., Lee, Y. H., Chang, C. M., Lin, I. T., Chen, J. A., Lin, T. Y. and Tso, I. M.** (2010).

Multiple structures interactively influence prey capture efficiency in spider orb webs. *Anim.*

*Behav*. **80,** 947-953.

**Blamires, S. J., Wu, C. L., Blackledge, T. A. and Tso, I. M.** (2012). Environmentally-induced post

spin property changes in spider silks: influences of web types, spidroin composition and

ecology. *Biol. J. Linn. Soc*. **106,** 580-588.

**Bonthrone, K. M., Vollrath, F., Hunter, B. K. and Sanders, J. K. M.** (1992). The elasticity of spider's webs is due to water-induced mobility at a molecular level. *Proc. Roy. Soc. B* **248,** 141-144.

**Craig, C. L.** (1997). Evolution of arthropod silks. *Ann. Rev. Entomol*. **42,** 231-267.

**Craig, C. L.** (2003). S*piderwebs and silks: tracing evolution from molecules to genes to phenotypes*. Oxford, U.K.: Oxford University Press.

**Cranford, S. W., Tarakanova, A., Pugno, N. M. and Buehler, M. J.** (2012). Nonlinear material behaviour of spider silk yields robust webs. *Nature* **482,** 72-76.

**Edmonds, D. and Vollrath, F.** (1992). The contribution of atmospheric water vapour to the formation and efficiency of a spider's web. *Proc. Roy. Soc. B*. **248,** 145-148.

**French, A. S., Torkkeli, P. H. and Seyfarth, E. A.** (2002). From stress and strain to spikes: mechanotransduction in spider slit sensilla. *J. Comp. Physiol. A* **188**, 739-752.

**Guinea, G. V., Cerdeira, M., Plaza, G. R., Elices, M. And Perez-Reiguero, J.** (2010). Recovery in viscid line fibers. *Biomacromolecules* **11,** 1174-1179.

**Hansell, M. H.** (2005). *Animal architectures*. Oxford, U.K.: Oxford University Press.

**Hansell, M. H. and Ruxton, G. D.** (2013). Exploring the dichotomy between animals building using self-secreted materials and using materials collected from the environment. *Biol. J. Linn. Soc*. **108,** 688-701.

**Heiling, A. M. and Herberstein, M. E.** (1999). The role of experience in web building spiders

(Araneidae). *Anim. Cogn*. **2,** 171-177.

**Herberstein, M. E. and Heiling, A. M.** (1998). Does mesh height influence prey length in orb-web
spiders. *Eur. J. Entomol*. **95,** 367-371.

**Herberstein, M. E. and Tso, I. M.** (2000). Evaluation of formulae to estimate the capture area and
mesh height of orb webs (Araneoidea, Araneae). *J. Arachnol*. **28,** 180-184.

**Kane, D. M., Naidoo, N. and Staib, G. R.** (2010). Atomic force microscopy of orb-spider web
silks to measure surface nanostructuring and evaluate silk fibers per strand. *J. Appl. Phys*. **108,**
073509.

**Kelly, S. P., Sensenig, A. Lorentz, K. A. and Blackledge, T. A.** (2011). Damping capacity is
evolutionarily conserved in the radial silk of orb-weaving spiders. *Zoology* **114,** 233-238.

**Liao, C. P., Chi, K. J. and Tso, I. M.** (2009). The effects of wind on trap structural and material
properties of a sit-and-wait predator. *Behav. Ecol*. **20,** 1194-1203.

**Lin, L. H., Edmonds, D. T. and Vollrath, F.** (1995). Structural engineering of an orb spiders web.
*Nature* **373,** 146-148.

**Lucas, J. R.** (1985). Metabolic rates and pit-construction costs of two antlion species. *J. Anim. Ecol*.
**54,** 295-309.

**Mellanby, K.** (1934). The evaporation of water from insects. *J. Exp. Biol*. **10,** 317–333.

**Mestre, L. and Lubin, Y.** (2011). Settling where the food is: prey abundance promotes colony
formation and increases group size in a web-building spider. *Anim. Behav*. **81,** 741-748.

**Opell, D. B.** (1989). Measuring the stickiness of spider prey capture threads. *J. Arachnol*. **17,**

112-114.

**Opell, B. D. and Hendricks, M. L.** (2007). Adhesive recruitment by the viscous capture threads of araneoid orb-weaving spiders. *J. Exp. Biol*. **210,** 553-560.

**Opell, B. D. and Hendricks, M. L.** (2010). The role of granules within viscous capture threads of orb-weaving spiders. *J. Exp. Biol*. **213,** 339-346.

**Opell, B. D. and Schwend, H. S.** (2007). The effect of insect surface features on the adhesion of viscous capture threads spun by orb-weaving spiders. *J. Exp. Biol*. **210,** 2352-2360.

**Opell, D. B., Karinshak, S. E. and Siger, M. A.** (2011a). Humidity affects the extensibility of an orb-weaving spider's viscous thread droplets. *J. Exp. Biol*. **214,** 2988-2993.

**Opell, B. D., Schwend, H. S. and Vito, S. T.** (2011b). Constraints on the adhesion of viscous threads spun by orb-weaving spiders: the tensile strength of glycoprotein glue exceeds its adhesion. *J. Exp. Biol*. **214,** 2237-2241.

**Roa, D.** (2009). Experimental evidence for the amelioration of shadow competition in an orb-web spider through the 'ricochet' effect. *Ethology* **115,** 691-697.

**Sahni, V., Blackledge, T. A. & Dhinojwala, A.** (2010). Viscoelastic solids explain spider web stickiness. *Nat. Comm*. **1,** 19.

**Sahni, V., Blackledge, T. A. and Dhinojwala, A.** (2011). Changes in the adhesive properties of spider aggregate glue during the evolution of cobwebs. *Sci. Rep*. **1,** 41.

**Sahni, V., Labhasetwar, D. V. & Dhinojwala, A.** (2012). Spider silk inspired microthreads. *Langmuir* **28,** 2206-2210.

**Sandoval, C. P.** (1994). Plasticity in web design in the spider *Parawixia bistriata*: a response to variable prey type. *Funct. Ecol*. **8,** 701–707.

**Scharf, I., Lubin, Y. & Ovadia, O.** (2011). Foraging decisions and behavioural flexibility in trap-building predators: a review. *Biol. Rev*. **86,** 626-639.

**Senenig, A, Agnarsson, I. and Blackledge, T. A.** (2010). Behavioural and biomaterial coevolution in spider orb webs. *J. Evol. Biol*. **23,** 1839-1856.

**Senenig, A, Lorentz, K. A, Kelly, S. P. and Blackledge, T. A.** (2012). Spider orb webs rely on radial threads to absorb prey kinetic energy. *J. Roy. Soc. Interf*. **9,** 1880-1891.

**Tarakanova, A. and Buehler M. J.** (2012). The role of capture spiral silk properties in the diversification of orb webs. *J. Roy. Soc. Interf*. **9,** 3240-3248.

**Townley, M. A., Bernstein, D. T., Gallagher, K. S. and Tillinghast, E. K.** (1991). Comparative study of orb-web hygroscopicity and adhesive spiral composition in three araneid spiders. *J. Exp. Zool.* **259,** 154-165.

**Tso, I. M., Chiang, S. Y. and Blackledge, T. A**. (2007). Does the giant wood spider *Nephila pilipes* respond to prey variation by altering web or silk properties? *Ethology* **113,** 324-333.

**Tso, I. M., Wu, H. C. and Hwang, I. R.** (2005). Giant wood spider *Nephila pilipes* alters silk protein in response to prey variation. *J. Exp. Biol*. **208,** 1053-1061.

**Turner, J., Vollrath, F. & Hesselberg, T.** (2011). Wind speed affects prey-catching behaviour in an orb web spider. *Naturwissenschaften* **98,** 1063-1067.

**Uetz, G. W.** (1989). The 'ricochet effect' and prey capture in colonial spiders. *Oecologia* **81,**

154-159.

**Venner, S., Pasquet, A. and Leborgne, R.** (2000). Web-building behaviour in the orb-weaving

spider *Zygiella x-notata*: influence of experience. *Anim. Behav*. **59,** 603-611.

**Viviani, V. R., Hastings, J. W. and Wilson, T.** (2002). Two bioluminescent diptera: the North

American *Orfelia fultoni* and the Australian *Arachnocampa flava*. Similar niche, different

bioluminescence. *Photochem. Photobiol*. **75,** 22-27.

**Vollrath, F. and Knight, D. P.** (2001). Liquid crystalline spinning of spider silk. *Nature* **410,**

541-548.

**Vollrath, F. and Tillinghast, E. K.** (1991). Glycoprotein glue beneath a spider web's aqueous coat.

*Naturwissenschaften* **78,** 557-559.

**Vollrath, F., Downes, M. and Krackow, S.** (1997). Design variability in web geometry of an

orb-weaving spider. *Physiol. Behav*. **62,** 735-743.

**Vollrath, F., Fairbrother, W. J., Williams, R. J. P., Tillinghast, E. K., Bernstein, D. T.,**

**Gallagher, K. S. and Townley, M. A.** (1990). Compounds in the droplets of the orb spider's

viscid spiral. *Nature* **345,** 526-528.

**Willis, R. E., White, C. R. and Merritt, D. J.** (2011). Using light as a lure is an efficient predatory

strategy in *Arachnocampa flava*, an Australian glowworm. *J. Comp. Physiol. B* **181,** 477-486.

**Willmer, P. Stone, G. and Johnston, I. A.** (2000). *Environmental Physiology of Animals*. New York:

Blackwell Publishing.

**Figure legends**

Fig. 1. Comparisons of the web geometric features: (A) web catching area , (B) total length of silk used per web , (C) total length of spiral thread , (D) mesh height and (E) number of radii, between the spiders exposed to wind (W) and spiders not exposed to wind (N) treatment subgroups. Error bars represent s.e.m.

Fig. 2. Comparisons of the droplet morphometric features: (A) droplet number (DN) per 0.5 mm of thread, (B) droplet spacing (DS) , (C) droplet volume (DV), (D) droplet volume per 0.5 mm of thread, (E) droplet surface area (DSA), (F) droplet surface area to volume ratio (DSAVR) and (G) flagelliform spiral thread diameter (SD), between the spiders exposed to wind (W) and spiders not exposed to wind (N) treatment subgroups. Error bars represent s.e.m.

Fig. 3. Mean (± s.e.m.) stickiness values for spirals in webs of the (A) P1 and P2 pretreatment groups and (B) W (spiders exposed to wind) and N (spiders not exposed to wind) treatment groups. Error bars represent s.e.m.
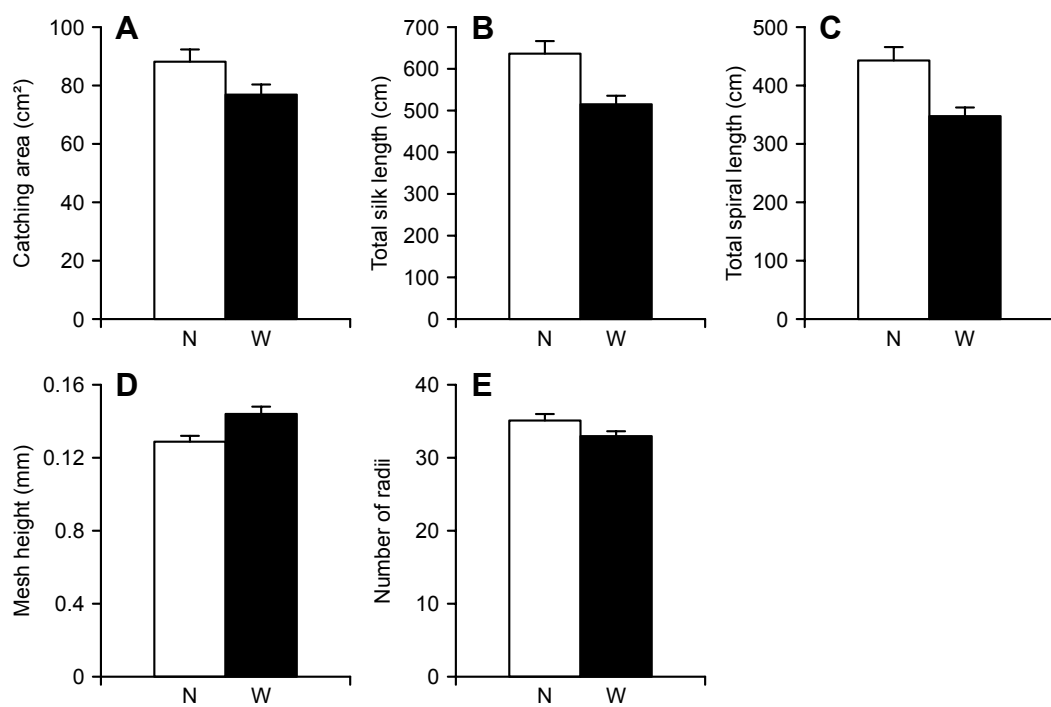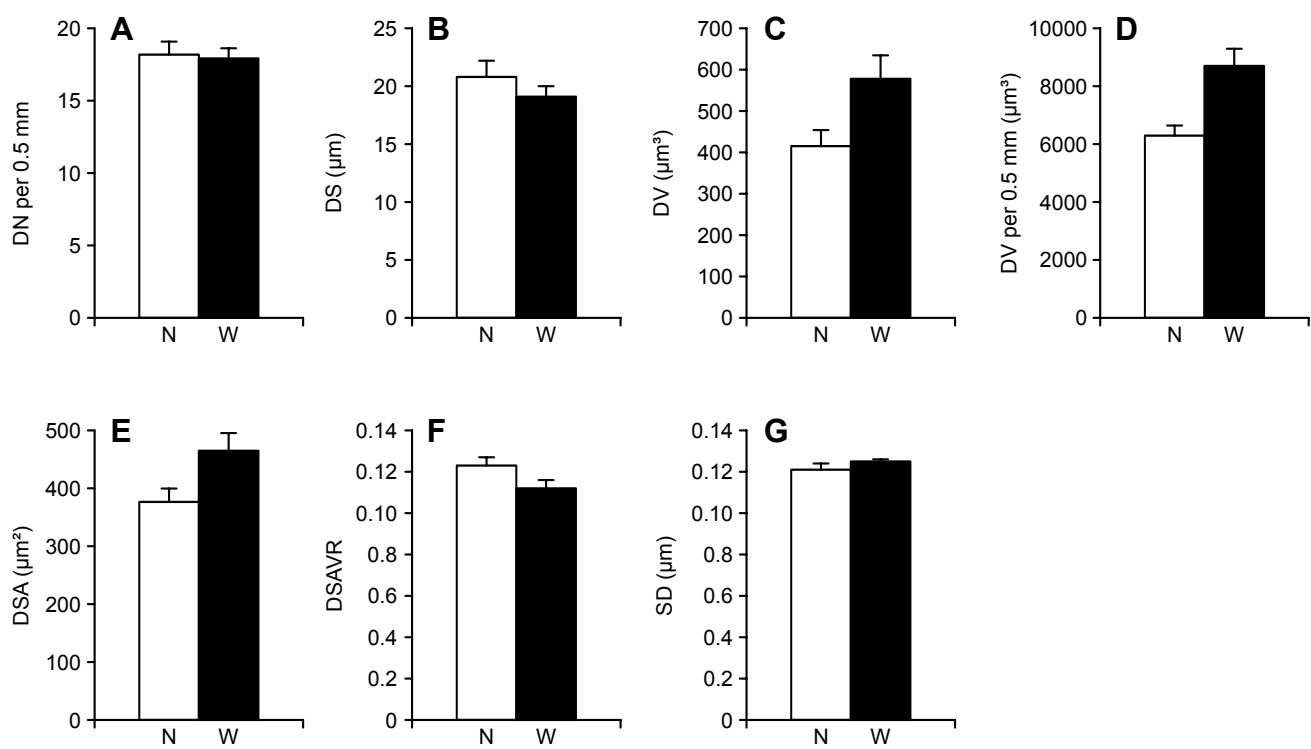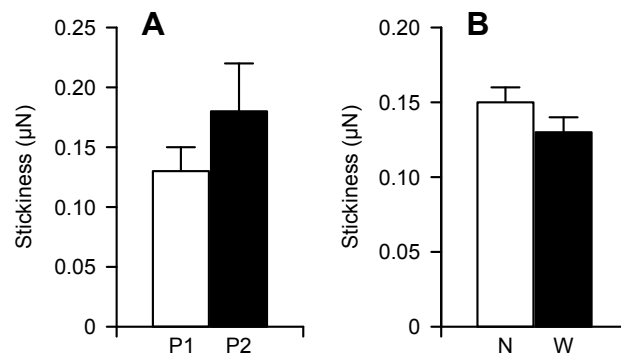
Figure 1

Figure 2

Figure 3

Table 1. Mean (± s.e.m) web geometric parameters of *Cyclosa mulmeinensis* webs from the P1 and P2 pre-treatment subgroups.

| Treatment | Catching area (cm$^2$) | Total silk length (cm) | Total spiral length (cm) | Mesh height (mm) | Number of radii |
|---|---|---|---|---|---|
| P1 | 84.96 ± 4.22 | 617.00 ± 28.26 | 419.40 ± 20.59 | 1.55 ± 0.06 | 36.61 ± 0.85 |
| P2 | 79.97 ± 4.57 | 597.10 ± 32.89 | 413.36 ± 24.42 | 1.48 ± 0.06 | 35.10 ± 0.94 |
| $F_{1, 59}$ | 0.65 | 0.76 | 0.04 | 0.21 | 1.44 |
| *P* | 0.42 | 0.39 | 0.85 | 0.65 | 0.24 |

Results of ANOVA tests comparing the subgroups for each parameter are shown.

Table 2. Mean (± s.e.m) features of the sticky spirals from webs built by *Cyclosa mulmeinensis* in the P1 and P2 pre-treatment subgroups.

| Treatment | DN per 0.5 mm | DS (μm) | DV (μm$^3$) | DV per 0.5 mm (μm$^3$) | DSA (μm$^2$) | DSAVR | SD (μm) |
|---|---|---|---|---|---|---|---|
| P1 | 18.43 ± 0.8 | 19.06 ± 1.1 | 386.67 ± 39.2 | 6326.39 ± 448.4 | 363.49 ± 24.7 | 1.23 ± 0.04 | 1.20 ± 0.01 |
| P2 | 17.62 ± 0.8 | 20.11 ± 1.4 | 434.44 ± 42.6 | 6837.94 ± 431.3 | 394.90 ± 25.7 | 1.17 ± 0.04 | 1.21 ± 0.02 |
| $F_{1, 59}$ | 0.44 | 0.35 | 0.68 | 0.67 | 0.77 | 1.05 | 0.19 |
| $P$ | 0.51 | 0.55 | 0.41 | 0.41 | 0.38 | 0.31 | 0.66 |

Results of ANOVA tests comparing the subgroups for each parameter are shown.

DN = droplet number; DS = droplet spacing; DV = droplet volume; SD = spiral diameter; DSA = droplet surface area; DSAVR = droplet surface area

to volume ratio; DN = droplet number; DS = droplet spacing; DV = droplet volume; SD = spiral diameter; DSA = droplet surface area; DSAVR =

droplet surface area to volume ratio; SD = spiral thread diameter.