

Do really all wolf spiders carry spiderlings on their opisthosomas? The case of *Hygrolycosa rubrofasciata* (Araneae: Lycosidae)

Petr Dolejš

doi: 10.5431/aramit4507

Abstract. Wolf spider females are characterised by carrying cocoons attached to their spinnerets. Emerged spiderlings are carried on the females' opisthosomas, with the exception of three Japanese lycosid species who carry spiderlings on empty cocoons. Here, the same behaviour is recorded in a European spider: the drumming wolf spider *Hygrolycosa rubrofasciata*. Spiderlings of this species do not try to climb on the female's opisthosoma, even when they are adopted by a female of a species with a normal pulli-carrying behaviour. This behaviour occurs in Trechaleidae and four unrelated species of Lycosidae inhabiting wet habitats and is therefore regarded as an adaptation to the unsuitable environment.

Keywords: Cocoons, female abdominal knobbed hairs, humid habitats, pulli-carrying behaviour, spiderling clusters

Female wolf spiders are known for their care of both cocoons and spiderlings (Foelix 2011). They carry their cocoons attached onto the spinnerets (cocoon-carrying behaviour) and their spiderlings on the opisthosoma (pulli-carrying behaviour) (Fujii 1976). All lycosids show cocoon-carrying behaviour, there are, however, three exceptions concerning pulli-carrying. Fujii (1976) found that females of *Arctosa ebicha* Yaginuma, 1960 and *Arctosa fujiii* Tanaka, 1985 (sub *Lycosa* sp.) do not carry their spiderlings on the opisthosoma. Juveniles of these species stay for several hours on the cocoon surface and then disperse within one to four days (Fujii 1976). This behaviour is also typical for members of the putative sister family Trechaleidae, which also carry cocoons attached to the spinnerets like lycosids (Carico 1993). The same behaviour was also recorded for females of *Hygrolycosa umidicola* Tanaka, 1978 (Suwa in litt. 1977, Yaginuma 1991). Females of both *A. ebicha* and *A. fujiii* lack the abdominal knobbed hairs (Fujii 1983) that enable the spiderlings to climb onto the mother's opisthosoma (Rover et al. 1973). Kronstedt (1984, 1996) recorded that these knobbed hairs are also absent in females of *Hygrolycosa rubrofasciata* (Ohlert, 1865) and suggested that this may be associated with the pulli-carrying behaviour. Ahtiainen et al. (2002) noted that "the offspring (of *H. rubrofasciata*) usually remain on the female's abdomen or on top of the empty egg sac for a day to chitinise their exoskeleton, after which they disperse". Thus, this species was chosen for the present study to clarify its pulli-carrying behaviour.

Hygrolycosa Dahl, 1908 is still of uncertain subfamilial affinities. It belongs either to Piratinae (Zyuzin 1993) or Venoniinae (Murphy et al. 2006), and currently contains four species (Platnick 2013). The drumming wolf spider, *Hygrolycosa rubrofasciata* is a rare, Palearctic, Eurosiberian wolf spider inhabiting reed swamps, alder forests and marshy pine forests (Buchar & Růžicka 2002). It is a diurnal species (Kronstedt 1984, 1996), well known for its audible acoustic courtship behaviour (Helversen in: Rovner 1975, Kronstedt 1984, 1996, Köhler & Tembrock 1987) and for the lowest male diploid chromosome number ($2n\delta = 20$) among European wolf spiders (Gorlov et al. 1995). Both males and females mature in autumn (Kronstedt 1984) and it takes two or three years to complete their life cycle (Kotiaho et al. 1996, Vertainen et al. 2000). The life cycle is a transition between diplochronous – with mating periods in autumn and spring – and stenochronous with a mating period occurring only in spring (Braun 1976 and references therein). Males die during or immediately after the mating season (in spring); however, females can survive until the next mating season (Kotiaho et al. 1999). In northern Europe, females produce more than one cocoon (Vertainen et al. 2000), and in central Europe, females can make up to three cocoons per year (Dolejš pers. obs.). The cocoons contain about 60 eggs (Wiebes 1959). The pulli-carrying behaviour of this species has, however, not been thor-

Petr DOLEJŠ, Department of Zoology, National Museum – Natural History Museum, Cirkusová 1740, CZ – 192 00 Praha 9 – Horní Počernice, Czech Republic; e-mail: petr_dolejs@nm.cz. Department of Zoology, Charles University in Prague, Faculty of Science, Viničná 7, CZ – 128 44 Praha 2, Czech Republic; e-mail: petr.dolejs@natur.cuni.cz

oughly investigated previously. Therefore, this study aims to investigate (1) the pulli-carrying behaviour of *H. rubrofasciata*, (2) the behaviour of *H. ubrofasciata* spiderlings on “substitute” mothers and “adoptive” spiderlings on *H. rubrofasciata* mothers after exchange of cocoons, and (3) the surface of the *H. rubrofasciata* cocoon in an attempt to determine the possible responsible structures for its pulli-carrying behaviour.

Methods

Females were collected at their first cocoon-carrying stage in the Kokořínsko Protected Landscape Area during a field course “Fauna of the Czech Republic”. There were six females collected at the locality Černý důl Natural Monument (50°29'N, 14°37'E; grid mapping square 5553; 310 m a.s.l.) on 10.5.2011, six females were collected at the locality Mokřady Horní Liběchovky Nature Reserve (50°31'N, 14°32'E; grid mapping square 5453; 260 m a.s.l.) on 11.5.2011, and two females were collected at the locality Jestřebské slatiny (50°36'N, 14°37'E; grid mapping square 5453; 250 m a.s.l.) on 23.5.2011 (lgt. Antonín Kůrka). Voucher specimens are deposited in the National Museum in Prague (N^os P6A 5070–5072).

Pulli-carrying behaviour was studied in the laboratory. In order to study differences in behaviour in test tubes and in simulated natural conditions, seven females were held in plastic test tubes (length 10 cm, diameter 15 mm) with cotton wool as a source of water and seven females were kept in four diagonally divided glass terraria (14 × 11 × 8 cm) with 3 cm of leaves. The programme NCSS 2007 (Hintze 2006) was used to test normality, to calculate descriptive statistics on the length of the pulli-carrying period, and to compare (Two-Sample T-Test) the length of the pulli-carrying period of females kept in test tubes with those kept in terraria.

After the females of *H. rubrofasciata* made the second cocoon, the behaviour of females with exchanged cocoons were tested. Cocoons of three females (in terraria) were replaced by cocoons of *Pardosa amen-tata* (Clerck, 1757), a species that displays a normal pulli-carrying behaviour (Vlijm et al. 1963). Three females of the latter species were collected at their second cocoon-carrying stage from Žleby (49°53'N, 15°29'E; grid mapping square 6158; 240 m a.s.l.). They were kept in terraria as described above for *H. rubrofasciata*. After the spiderlings of both species emerged from the cocoons, the behaviour of spider-

lings on their “adoptive” mothers was examined and documented (digital camera Olympus C-7070 WZ). In order to compare possible structural differences in cocoon surface, empty cocoons of both species were inspected under a scanning electron microscope JEOL 6380 LV.

Results

Pulli-carrying behaviour of *Hygrolycosa rubrofasciata*

After emergence from the cocoon, spiderlings of *H. rubrofasciata* did not climb onto the opisthosoma of their mother. Instead, they stayed on the surface of the cocoon (Fig. 1). Spiderlings of females kept both in terraria and test tubes behaved in the same way. However, time spent on the cocoon surface differed significantly in both groups ($p = 0.0031\%$). Spiderlings of females kept in terraria dispersed within 1–3 days ($\bar{X} = 2.29$, $SD = 0.756$, $n = 7$), but those of females kept in test tubes within 4–6 days ($\bar{X} = 5.00$, $SD = 0.816$, $n = 7$). In cases of larger clutch size, a few spiderlings stepped on the mother's opisthosoma using their forelegs, however they always remained in contact with the cocoon or with lower layer of spiderlings on the cocoon surface. Females detached their empty cocoons after the spiderlings had dispersed.

Behaviour of spiderlings on substitute females

All females (of both *H. rubrofasciata* and *P. amentata*) accepted, sooner or later, cocoons of the other species. After emergence, spiderlings of *H. rubrofasciata* did not try to climb on the opisthosomas of *P. amentata* females and remained on the cocoon surface (Fig. 2). By contrast, spiderlings of *P. amentata* tried to climb



Fig. 1: Pulli-carrying behaviour of *Hygrolycosa rubrofasciata*. Newly emerged spiderlings occupy the surface of the cocoon instead of climbing onto the opisthosoma of their mother.

onto the opisthosoma of *H. rubrofasciata* females, yet only few spiderlings were successful in settling on a substitute mother. The majority of spiderlings formed a cluster either beside the empty cocoon or on the ventral surface of the female's opisthosoma – in the area where the cocoon was touching the opisthosoma. Therefore, *H. rubrofasciata* females were carrying two 'pellets': the empty cocoon and the cluster of *P. amentata* spiderlings (Fig. 3). However, the females lost the clusters within the same day as the spiderlings emerged and the spiderlings dispersed the following day. Examination of the cocoon surfaces revealed no differences between the cocoon structures of both species (Figs 4, 5).



Fig. 2: Behaviour of *Hygrolycosa rubrofasciata* spiderlings on a substitute *Pardosa amentata* mother. The spiderlings remained on the cocoon surface and did not try to climb onto the female's opisthosoma.

Discussion

A generally accepted statement that females of all wolf spiders carry spiderlings on their opisthosomas was contested. Fujii (1976) was the first author to report "abnormal" behaviour in two *Arctosa* species which carried spiderlings on their cocoons instead of on their opisthosomas. Suwa (in litt. 1977) and Yaginuma (1991) reported similar behaviour for *Hygrolycosa umidicola*, and Kronestedt (1984) and Ahtiainen et al. (2002) suggested such a possibility in *Hygrolycosa rubrofasciata* too. In the present study, it was confirmed unequivocally that females of *H. rubrofasciata* do not carry spiderlings on their opisthosomas, but instead on their empty cocoons. Until now, this

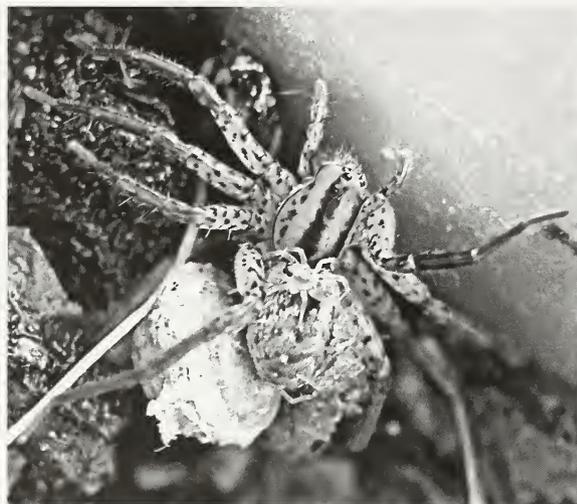
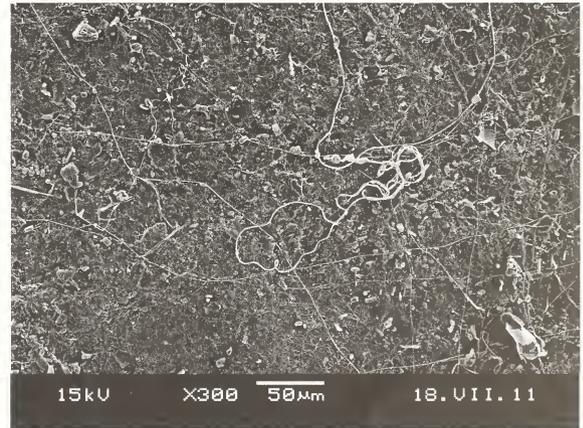
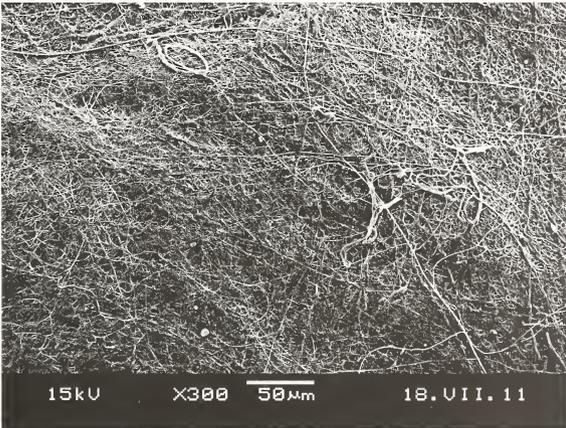
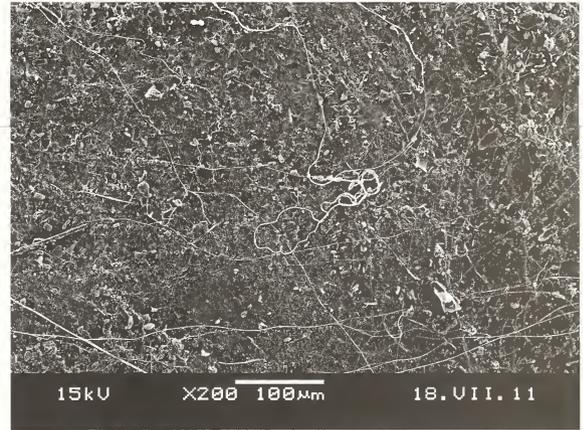
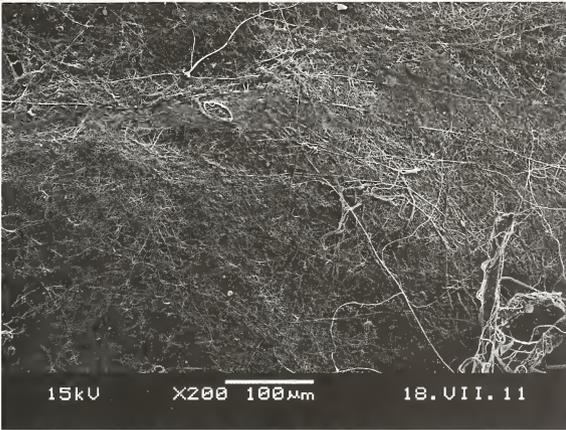
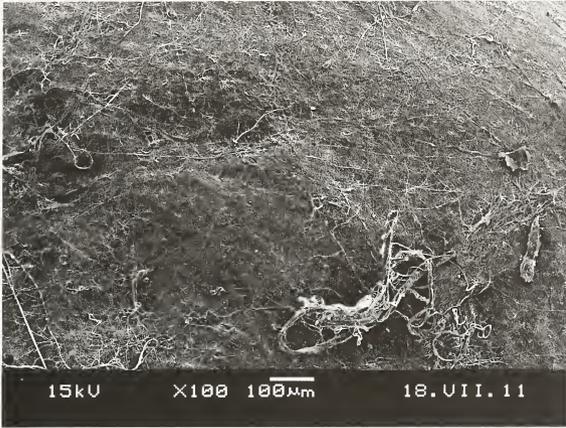


Fig. 3: Behaviour of *Pardosa amentata* spiderlings on substitute *Hygrolycosa rubrofasciata* mother. Most of the spiderlings were not successful in settling on the opisthosoma of a substitute female. Instead, they formed a cluster beside the empty cocoon. The female was carrying both the spiderling cluster and the empty cocoon for a while, until she lost the cluster.

unusual behaviour was only known from four lycosid species: *A. ebicha*, *A. fujiii*, *H. rubrofasciata* and *H. umidicola*.

The time which the spiderlings spent on the cocoon surface differed between females kept in terraria and those in the test tubes. In natural conditions, the spiderlings probably disperse within two days – this is maybe the reason why pulli-carrying behaviour has not yet been observed in the field. In the test tubes, spiderlings probably did not have enough space to disperse and therefore remained longer on the cocoon surface. A similarly short pulli-carrying period was reported for *A. fujiii*, but three times longer for *A. ebicha* (Fujii 1976). However, the females of these species were kept in small glass vials and therefore the length of the period could be affected. There are no data about the length of pulli-carrying period in *H. umidicola*.

After cocoon exchange, spiderlings behaved on the substitute mother as they would on their own mother: *H. rubrofasciata* spiderlings remained on the cocoon surface, although they were carried by a *Pardosa* female. Similarly, *Pardosa* spiderling tried to climb on the *H. rubrofasciata* female, but only few of them were successful – presumably due to the absence of abdominal knobbed hairs. The majority of spiderlings tended to aggregate elsewhere. It seems therefore that the primary behaviour of the



Figs 4a-c: Cocoon surface of *Hygrolycosa rubrofasciata*. Scale bars: a) and b) 100 μm , c) 50 μm

Figs 5a-c: Cocoon surface of *Pardosa amentata*. Scale bars: a) and b) 100 μm , c) 50 μm

spiderlings is species-specific and does not depend on the mother. The same results were also obtained by Fujii (1980). He tested behaviour of *Pardosa astrigera* L. Koch, 1878 which carry spiderlings normally on the opisthosoma, and *A. fujiii* (sub *Arctosa* sp.), which carry spiderlings on the cocoon (see Fu-

jii 1980: figs. 1G and 1H). Both *Pardosa* spiderlings failed to settle on the substitute mother (*A. fujiii*, *H. rubrofasciata*) and formed a cluster. This behaviour disagrees with observations in *Schizocosa crassipes* (Walckenaer, 1837) and *Rabidoso rabida* (Walckenaer, 1837) in which the spiderlings never clus-

tered when separated from their mothers (Higashi & Rovner 1975).

Spiderlings settle only on suitable surfaces (Engelhardt 1964, Rovner et al. 1973, Fujii 1983). Therefore, it is curious that there are no 'proper structures' on the cocoon surface of *H. rubrofasciata*. Unfortunately, only a few workers examined cocoon microstructure (e.g. De Bakker et al. 2006), but none of them considered lycosid and trechaleid cocoons. For this reason, it would be of value to study the mechanics underlying lycosid and trechaleid spiderling behaviour.

The clustering of spiderlings on an empty cocoon for a short time – instead of on their mother's opisthosoma – in *A. ebicha* and *A. fujiii* is peculiar because at least *A. ebicha* is related neither to the genus *Hygrolycosa* nor to other *Arctosa* species (Murphy et al. 2006), and probably belongs (together with the related *Arctosa kwangreungensis* Paik & Tanaka, 1986) to a separate, undescribed genus (Framenau in litt. 5. xii. 2007). Interestingly, members of *Hygrolycosa*, both *A. ebicha* and *A. fujiii*, and the most members of the family Trechaleidae live in very humid or even wet habitats (Tanaka 1978, 1991, Carico 2005). Little drops of water remaining on the opisthosomal surface and hairs probably create unsuitable conditions for clustering the spiderlings. The dragline threads eventually forming a layer over the surface and providing a good means of attachment for the spiderlings (Rovner et al. 1973) probably cannot be attached to the moist surface or hairs either. Moisture can even paste the knobbed hairs together and disable the spiderlings' ability to cling onto their mother's opisthosoma.

For this reason, the cocoon surface seems to be more water-repellent (Hieber 1984 in: Hieber 1992) and presents better place to cluster, despite the absence of any auxiliary structures. Fujii (1983) reported the absence of knobbed hairs, but normal pulli-carrying behaviour, in *Arctosa subamylacea* (Bösenberg & Strand, 1906) and *Arctosa depectinata* (Bösenberg & Strand, 1906). These two species live mainly in fields (Tanaka 1991) so clustering on the cocoon has not developed. On the other hand, Fujii (1980), who also tested spiderling behaviour after cocoon exchange in *Pardosa astrigera* and *Pirata subpiraticus* (Bösenberg & Strand, 1906), showed that spiderlings of *P. astrigera* climbed indeed onto the opisthosoma of wet-habitat-dwelling *P. subpiraticus* but all of them dropped from her till the next day and clustered elsewhere. Correspondingly, spiderlings of most of *Pirata* (and

Piratula) species do not mount the opisthosoma of the female, but remain in the silken retreat (Nielsen 1932). Therefore, living in wet habitats rather than lacking knobbed hairs seems to be the main reason for modifying pulli-carrying behaviour.

This contribution shows how little is known about basic biological data for wolf spiders. It is necessary to study the pulli-carrying behaviour and female abdominal knobbed hairs of other wet-habitat lycosids (*Hygrolycosa*, *Arctosa*, *Pirata*, *Piratula*) and trechaleids. As Fujii (1976) already stated, more data must be obtained to clarify the evolution of pulli-carrying behaviour in lycosids and related families.

Acknowledgements

My great thanks belong to Hozumi Tanaka who informed me about the behaviour of *H. umidicola*, provided me with unpublished information from Masayoshi Suwa, and encouraged me to study *H. rubrofasciata*. I would like to thank the participants of the field course "Fauna of the Czech Republic", namely Lucie Jarošová, who collected females of *H. rubrofasciata* in Kokořínsko PLA, and to Antonín Kúrka who collected them at Jestřebské slatiny. I wish to thank Volker W. Framenau for providing me with his unpublished data. I am indebted to Jan Buchar and Hozumi Tanaka for valuable discussion and critical reading of the manuscript. I also thank Torbjörn Kronstedt and an anonymous reviewer for important comments and valuable suggestions that improved the earlier version of the manuscript. This work was financially supported by Ministry of Culture of the Czech Republic (DKRVO 2013/13, National Museum, 00023272).

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Jahr/Year: 2013

Band/Volume: [45](#)

Autor(en)/Author(s): Dolejs Petr

Artikel/Article: [Do really all wolf spiders carry spiderlings on their opisthosomas? The case of *Hygrolycosa rubrofasciata* \(Araneae: Lycosidae\) 30-35](#)