

Female genital morphology and sperm priority patterns in spiders (Araneae)

GABRIELE UHL

University of Bonn, Institute of Zoology, Department of Ethology, Kirschallee 1, D- 53115 Bonn, Germany
(g.uhl@uni-bonn.de)

Abstract

For spiders, gross female spermathecal morphology has been widely used as the major predictor of sperm priority pattern depending either upon taxonomic classification or on the number of ducts that connect with the spermathecae. In order to establish whether, or to what degree, the female reproductive tract follows a cul-de-sac (one duct connects to the spermatheca) or a conduit design (two ducts connect to the spermatheca at opposite ends) I present information on genital morphology of two haplogyne species (*Pholcus phalangioides*, Pholcidae; *Dysdera erythrina*, Dysderidae) and two entelegyne species (*Nephila clavipes*, Tetragnathidae; *Pityohyphantes phrygianus*, Linyphiidae). Predictions based on female anatomy and copulatory mechanisms are compared to available data on sperm utilization patterns.

Female genital anatomy deviates markedly from the expected pattern in all cases. There are more than the two predicted types of sperm storage sites: sperm can either be stored in the bursa, or in spermathecae connected by two ducts that lie close together, or in multiple sperm stores of different morphology. If males are able to insert their genital structures as far as to the lumen of the female sperm storage organ, male manipulation of sperm masses stored from previous males are possible and changes in sperm priority patterns can be expected. Combined information on detailed female anatomy and copulatory mechanism do not suffice to make reliable predictions on the pattern of sperm priority. Possible reasons for this discrepancy are briefly outlined.

Key words: Sexual selection, sperm storage, female choice.

INTRODUCTION

Female spiders show a propensity to mate with more than one male (Austad 1984; Elgar 1998), and maintain sperm for long periods in their sperm-storage organs (e.g. Uhl 1993a). Moreover, male spiders are unable to monopolize access to a female for the duration of her reproductive life, mainly because life expectancy for males is usually shorter than for females (Elgar 1998). Sperm from one of several males may be utilized randomly as sperm mix in the spermathecae leading to similar fertilization success of successive males or to fertilization success

that depends on the relative number of sperm stored from each male. On the other hand, non-random utilization of sperm from a particular male is termed sperm precedence or priority, and is generally seen as a consequence of sperm stratification within the sperm storage organ of the female (Austad 1984; Elgar 1998; see Simmons & Siva-Jothy 1998 for definitions). Sperm from different males may remain stratified within the spermathecae either because sperm are non-motile, or are transferred or stored in distinctive portions, thus leading to a positional advantage. Whether the first or the

last male to mate sires most of the offspring may thus purely depend on the position of the sperm mass within the female sperm storage organ.

The commonly accepted hypothesis on sperm precedence patterns in spiders relies on the fact that the sperm masses are stratified within the female genital tract (Austad 1984). Austad proposed that spider spermathecal morphology may represent a phylogenetic constraint which would lead to a non-adaptive pattern of sperm priority (Fig. 1). The term 'non-adaptive' in this context probably meant that adaptation was not recent but related back to an ancestral stage. Spiders (Araneoclada) are classified into two groups, the Entelegynae and the Haplogynae (Coddington & Levi 1991). Formerly, spiders were said to exhibit a fundamental dichotomy in the female spermathecal morphology that divides along phylogenetic lines with the Haplogynae possessing one duct and the Entelegynae possessing two ducts that connect with the spermathecae. If this were consistently so, the hypothesis that the two groups have distinctly different sperm priority patterns would be based on a firm morphological basis. However, in a number of genera within the entelegyne families Uloboridae, Tetragnathidae, Anapidae and the superfamily Palpimanoidea reversal to the haplogyne condition with only a single spermathecal duct occurred (see Coddington & Levi 1991, and even Austad himself 1984). Likewise, within the haplogyne Pholcidae, there are at least two species in which two ducts have evolved independently (Huber 1996). The idea that Haplogynae und Entelegynae each have uniform female genital morphology is thus not supported. As a consequence, inferring distinctly different sperm precedence patterns for taxa of the two groups is untenable.

To examine the hypothesis that sperm priority patterns depend on the morphology of female genitalia independent of phylogenetic position seems to be a more rewarding task. In Austad's paper, this hypothesis is mixed with the previous one on phylogenetic constraints. If

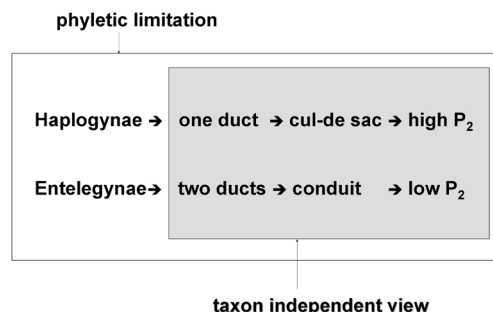


Fig. 1. The original hypothesis by Austad (1984) proposed a phyletic limitation to sperm precedence patterns in spiders. The 'taxon independent' hypothesis only assumes a connection between the number of ducts that connect to the spermathecae and P_2 values.

female genital morphology determines sperm priority patterns, and sperm stratification occurred, species with one or two ducts that connect with the spermathecae should exhibit distinctly different sperm priority patterns (Fig. 1). I will use the terms haplogyne genitalia or haplogyne condition for species with one duct and entelegyne genitalia or entelegyne condition for species with two ducts, irrespective of taxonomic classification. Thus a spider, classified as belonging to the Haplogynae can exhibit female genital morphology of the entelegyne condition as in the case of the two Pholcid species mentioned earlier.

Sperm of haplogyne species passes along the single duct during copulation and again outward at oviposition (Fig. 2a). Austad (1984) termed this design, that is very similar to that in many insects (Walker 1980), 'cul-de-sac' (dead end). The entelegyne condition, on the other hand, was termed 'conduit' situation (one-way) and consists of a copulatory duct that leads to the spermatheca and a fertilization duct through which sperm reaches the eggs for fertilization (Fig. 2b). Predictions about sperm priority are that species with dead-end spermathecae should exhibit last male sperm priority as the last sperm to enter should lie closest to the single duct. This would represent a 'last in - first out' system. On the other hand, species

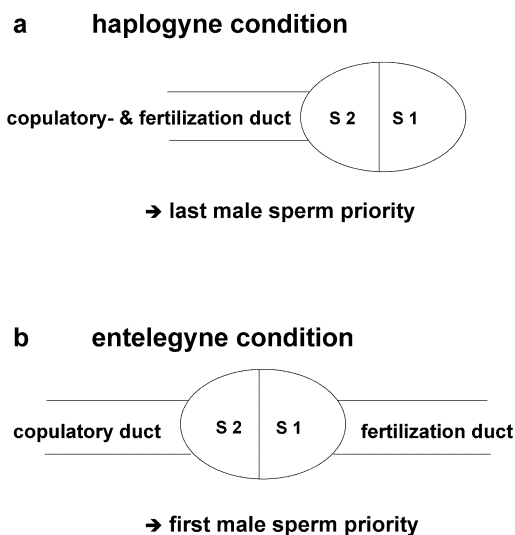


Fig. 2. Hypothetical sperm stratification inside the spermatheca of **(a)** spider with haplogyne genitalia and **(b)** spider with entelegyne genitalia and consequences of sperm priority patterns. S1: sperm from first male, S2: sperm from second male.

with one-way spermathecae are predicted to exhibit first male sperm priority, because the first sperm to enter should be closest to the fertilization duct and be the first to exit (first in - first out). The priority pattern should have implications for the mating strategy: species of the haplogyne condition should tend to guard mates just before egg laying, whereas species of the entelegyne condition should tend to guard penultimate females (see Elgar 1998).

Sperm utilization by females after multiple copulations is expressed as the proportion of offspring fathered by the last male to mate. In a typical experimental double-mating trial this is the proportion of eggs sired by the second male to mate, P_2 (Boormann & Parker 1976). Sperm mixing is expected to lead to P_2 values around 50% whereas sperm precedence of the first or second male leads to low or high P_2 values.

In order to test whether female genital morphology allows predictions on sperm priority patterns, the anatomy of the female reproductive tract needs to be examined in detail, thereby establishing whether, or to what de-

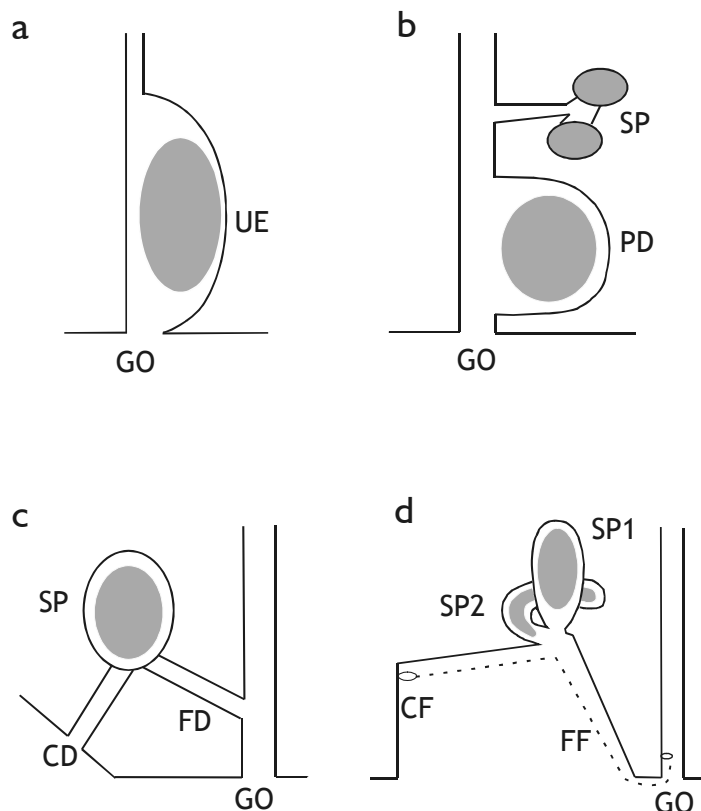
gree, it follows a cul-de-sac or conduit design. I therefore present detailed genital morphology for two haplogyne (*Pholcus phalangioides* (Fuesslin 1775) Pholcidae; *Dysdera erythrina* (Walckenaer, 1802), Dysderidae) and two entelegyne species (*Nephila edulis* (Labillardière, 1799), Tetragnathidae; *Pityohyphantes phrygianus* (C.L. Koch, 1836), Linyphiidae). For each species, I further present information on copulatory mechanisms. Possible access of male genitalia to the sperm storage site inside the female is crucial to the possibility that males physically manipulate sperm priority patterns. I also summarize published studies to check the predictions based on genital morphology and copulatory mechanism against paternity patterns.

Apart from physical male manipulation of stored sperm masses there are numerous other, more cryptic possible mechanisms, e.g. chemical manipulation. Products of the male reproductive organs that are transferred during copulation often induce female resistance to further mating, earlier oviposition, and even sperm transport (Eberhard 1997). On the other hand, female behavioural, morphological or physiological mechanisms that occur during or after copulation were shown to impose a bias on male reproductive success (Eberhard 1996). Although cryptic processes can be expected to play an important role, I will restrict this paper mainly to the question of whether female genital morphology and copulatory mechanism allow predictions on the pattern of sperm priority.

Pholcus phalangioides

The cellar spider *P. phalangioides* has only one genital opening through which copulation and egg laying are achieved as in typical haplogyne spiders. However, there are no spatially separated sperm storage organs of the cul-de-sac type (Fig. 3a). Sperm is stored in the bursa itself, and is embedded in a secretion produced by the female before copulation (Uhl 1994a). Complicated glands produce this matrix, and possibly also cause sperm activation (Uhl 1994b). What was formerly described as sper-

Fig. 3. Schematic presentation of female genital anatomy and place of sperm storage in (a) *Pholcus phalangioides* (Pholcidae), (b) *Dysdera erythrina* (Dysderidae) (c) *Nephila edulis* (Tetragnathidae) and (d) *Pityohyphantes phrygianus* (Linyphiidae). CD: copulatory duct, CF: copulatory fold, FD: fertilization duct, FF: fertilization fold, GO: genital opening, PD: posterior diverticulum, SP: spermatheca, UE: uterus eternus (= bursa). Dotted lines indicate the existence of folds instead of ducts. Shaded areas indicate places of sperm storage.



mathecae by Wiehle (1933) only rarely contains a few sperm. This structure was shown to have a different function: it is a fold connected to muscles that serves to open the genital valve before egg laying (Uhl 1994a). Overall, genital morphology in *P. phalangioides* deviates markedly from the expected pattern.

During copulation, several parts of the male pedipalp are inserted into the female: the procurus, the embolus, the appendix (a coupling structure) and the uncus (Uhl et al. 1995). The embolus is inserted directly into the female secretion where the sperm are stored and semi-thin sections from twice mated females showed no obvious stratification of sperm masses (Uhl 1994a). The male performs rhythmic twisting movements with the simultaneously inserted pedipalps during copulation, which result in extrusion of sperm near the centre of the female genital opening (Uhl et al. 1995).

Two different predictions arise from these

findings: if female genital morphology played the most important role, sperm mixing should occur and P_2 should vary around 50%, whereas sperm displacement via pedipalp movement would predict a last male advantage. Investigations on sperm priority in the cellar spider showed a mean P_2 value of 66% for first broods of seven females (Yoward 1998, Tab. 1), despite a much shorter copulation duration in second matings compared to first ones (Uhl 1993b; Yoward 1998). The analysis of a larger sample size of 47 first broods showed high mean paternity value of 78% for second males (Schäfer & Uhl 2002). However, both investigations demonstrate highly variable paternity values (Tab. 1). The number of pedipalp movements a male performs during copulation is a good predictor of his fertilization success (Schäfer & Uhl 2002). Thus, the number of movements very likely influences the degree of displacement of previous male's sperm in second matings.

Table 1. Compilation of data available both on sperm priority and genital mechanisms in spiders.^a: calculated from Yoward's data for first broods to allow comparison between species^b: note that mean P₂ values did not meet assumptions of normality.

Species (n)	Mean	P ₂ in %		Genital mechanism known?	Male manipulation possible?	Predictions possible ?	Source
		Range (sd)(se)	median				
Haplogyne genital structure							
Bursal storage							
<i>Pholcus phalangioides</i> (7)	0.66 ^a	0.39-1.00 (0.22)	0.66	yes	yes	yes	Yoward 1996; Uhl et al. 1995
(47)	0.78	0.00-1.00 (0.25)	0.89				Schäfer & Uhl 2002.
<i>Holocnemus pluchei</i> (40)	0.74 ^b	0.09-1.00 (0.04)	?	yes	yes	yes	Kaster & Jakob 1997; Huber 1995
<i>Physocyclus globosus</i> (12)	0.38	0.00-1.00 (0.30)	0.44	yes	yes	no	Eberhard et al. 1993; Huber & Eberhard 1997
Cul-de-sac							
<i>Tetragnatha extensa</i> (7)	~ 0.70	(0.36)	?	~ yes	no access	yes	West & Toft 1999
Entelegyne genital structure							
„Cul-de-sac“ type							
<i>Nephila clavipes</i> (63)	0.18	0.00-1.00 (0.32)	0.02	~ yes	yes	no	Christenson & Cohn 1988
<i>Nephila edulis</i> (8)	0.66	(0.56)	0.83	yes	yes	yes	Schneider et al. 2000; Uhl & Vollrath 1998
<i>Nephila plumipes</i> (33)	0.46	(0.05)	0.42	~ yes	yes	no	Schneider & Elgar 2001
<i>Latrodectus hasselti</i> (11)	0.56	0.00-1.00		~ yes	yes	~yes	Andrade 1996; see text for morphological data on various species

Sperm extrusion was also found for another pholcid spider, *Physocyclus globosus* (Taczanowski, 1874) (Huber & Eberhard 1997). Sperm masses only emerged in copulations with non-virgin females, they appeared gradually rather than in step with male pedipalp movements and continued to emerge after copulation ended. Double mating experiments revealed a mean P₂ of 38% (Eberhard et al. 1993; Tab.1). Copulation durations in virgin versus mated females are not nearly as different as in *P. phalangioides* (Huber & Eberhard 1997; Uhl 1993b). In *P. phalangioides*, sperm extrusion can also occur in copulations with virgin females, extrusion seems to appear in step with male movements and does not continue after copulation. Apparently, closely related species with similar genital morphology have evolved very differ-

ent male and female mechanisms that may bias paternity. Further investigations are needed that examine whether and to what extent a male removes his own sperm relative to his rival's sperm. Also, sperm transfer may not be a time dependent process as is often assumed: it may either relate to pedipalp movements or to copulation duration. For *P. phalangioides* preliminary results suggest the former mechanism (Uhl, unpublished). In fact, time independent sperm transfer was found in *Frontinella communis* (Hentz, 1850) (Austad 1982, syn. *F. pyramitela*) and *Micrathena gracilis* (Walckenaer, 1805) (Bukowski & Christenson 1997). Both studies investigated sperm release from the male palp by interrupting copulations and counting the remaining number of sperm in the palp. Other studies that suggest that sperm

transfer were time related actually investigated sperm uptake and storage by the female, not sperm release. However, these may be distinctly separate processes that are often confounded (Bukowski & Christenson 1997).

Data on sperm priority and copulatory mechanism exists for yet another Pholcid spider, *Holocnemus pluchei* (Scopoli, 1763). High last male sperm precedence in this species (Kaster & Jakob 1997) may also be a result of male manipulation, as male genitalia reach the place of sperm storage (Huber 1995).

Dysdera erythrina

Semi-thin sections show that two structures function as sperm storage organs in *D. erythrina*: the so-called posterior diverticulum (PD), a large dilatation of the genital cavity similar to the structure found in *Pholcus phalangoides*, and a bilobed anterior spermatheca (Fig. 3b; Cooke 1966; Schult 1980; Uhl 2000). Thus, genital morphology deviates strongly from the presumed haplogyne pattern. Both structures are equipped with glandular tissue but the glandular tissue of the two is markedly different. The glandular tissue of the spermatheca is composed of complicated glandular units around cuticular ductules, whereas the glandular tissue of the posterior diverticulum is composed of rather simple gland cells (Uhl 2000). The products presumably differ, leading to possibly different storage conditions for the spermatozoa. Encapsulated spermatozoa are found in each lobe of the spermatheca. Sperm seems to be packed much tighter in the spermathecae than in the posterior diverticulum.

The male pedipalp in *D. erythrina* is a simple, blunt tipped structure, and thus probably reaches only as far as the posterior diverticulum (PD). Due to its size, it is highly unlikely that it enters the duct to the bilobed spermatheca. Thus, male manipulation is only possible for the PD, not for the spermatheca and it is tempting to suggest that the two types of sperm storage organs have evolved to allow (in the case of the PD) or prevent (in the case of the spermatheca) males from accessing previously

stored sperm. Multiple organs may further facilitate specialization in short-term and long-term sperm storage as occurs in *Drosophila* (Pitnick et al. 1999). To date, there is no information on sperm priority pattern in *D. erythrina*.

Nephila edulis

Because *N. edulis* is an entelegyne spider, we would expect to find two ducts that connect on opposite sides of the spermathecae to form a one-way system. Female genital morphology in *N. edulis* does possess two ducts (in and out) connected to each of the two spermatheca, but the details of the connections to the spermathecae differ from the assumptions made by the Austad hypothesis. The ducts are close together and the reproductive tract looks more like a cul-de-sac than a conduit (Fig. 3c). The spermatheca lacks a septum that would create the equivalent of a one-way system inside of the spermathecal lumen. Based on female morphology alone, the sperm from the last male should be closer to the fertilisation duct, and last male sperm priority should prevail. The male pedipalp in *N. edulis* consists of a compact genital bulb provided with a long conductor that supports the sperm transferring structure, the embolus (Uhl & Vollrath 1998). The embolus is rolled up inside the bulb and can be pushed out of the tip of the conductor. During copulation, the embolus reaches the lumen of the spermathecae (Uhl & Vollrath 1998) which makes stratification unlikely and speaks in favour of sperm mixing. Male manipulation of previous males' sperm, also seems possible.

An investigation on sperm priority in *N. edulis* shows a mean P_2 value of 66% (median 83%) based on 8 matings (Schneider et al. 2000) (Tab. 1), which tentatively suggests that sperm manipulation by subsequent males is possible. On the other hand, in *N. edulis* duration and frequency of copulation is a very good predictor of paternity independent of mating order, which suggests that sperm are utilized according to relative numbers. As mentioned above, caution has to be applied when assuming grad-

ual sperm transfer on the basis of a correlation between copulation duration and sperm utilization. In *N. clavipes* (Linnaeus, 1767) this correlation was found although sperm transfer occurs during the first of many insertion bouts (T. E. Christenson pers. comm.). In *N. clavipes* clear first male advantage was demonstrated (mean P_2 value: 18%) (Christenson & Cohn 1988). Besides possible male manipulation an alternative explanation for the priority pattern would be that significantly different sperm numbers are taken up by the female depending on the mating order (T.E. Christenson pers. comm.). Investigations on sperm release and uptake by Cohn (1988) point in this direction. In *N. plumipes* (Latreille, 1804) equal sperm numbers seem to mix in the female spermatheca (mean P_2 : 46%, Schneider & Elgar 2001, Tab. 1). Although female and male morphology appears to be very similar in the three *Nephila* species, transfer, storage and utilization mechanisms seem to be quite different. Clearly, further investigations are needed.

Due to extreme intraspecific male size variability in *Nephila* (Vollrath 1980), one might expect small males to be at a disadvantage if it comes to copulatory mechanisms and sperm transfer. Size variability could lead to different degrees of mating efficiency and fertilization success for males of different sizes. However, in *N. edulis* genital characters show negative allometric values when plotted against somatic characters, which means that small males have relatively large genitalia and large males relatively small genitalia (Uhl & Vollrath 2000). Although male somatic size has a coefficient of variation of about 45%, genital variability is only 20% ($p < 0.01$, Lewontin's method 1966). Mean values of male and female genitalic characters match surprisingly well: embolus length minus conductor length in the male has a mean of 1.3 mm while mean copulatory duct length in the female is 1.28 mm (Uhl & Vollrath 2000). There seem to be strong selective advantages leading towards intermediate, standardized sizes of male genitalia as in many other species of insects and spiders (Eberhard et al. 1998). It

seems that males adapt their genital size to that appropriate to the most common female size.

As a consequence of this finding, it might be expected that copulatory mechanisms for males of different sizes should be similar. However, the study by Schneider et al. (2000) showed that small males had a mating advantage, they mated for longer and fertilized more eggs than large males.

Pityohyphantes phrygianus

We might expect linyphiids to represent 'proper' conduit type spiders, as the linyphiid *Frontinella communis* exhibits clear first male sperm priority with little variation in P_2 (Austad 1982).

The epigynum of *P. phrygianus* has a scape, with an atrium on both sides. There are two spermathecae on each side, one is straight and thumb-like (spI) and the other is twisted (spII) and surrounds the straight one half way (Fig. 3d). Both spermathecae extend from a massive U-shaped structure. As in *D. erythrina*, the composition of the associated glands differs between spermathecae, as only the twisted spermathecae exhibit a strip of glandular ductules. The internal characteristics are quite complicated: a fold rather than a tube functions as a copulatory 'duct', leading to the spermathecae (Uhl & Gunnarsson 2001). This copulatory fold is 'sealed' after copulation with a homogeneous secretion, probably to impair copulatory success of subsequent males. At the base of the spermathecae there is a valve-like structure that makes intromission of male genital structure unlikely. Surprisingly, a fertilisation duct does not exist in *P. phrygianus* (Uhl & Gunnarsson 2001). A fold extends from the base of the spermathecae laterally along the ventral wall towards the opening of the atrium. From there, it turns into the epigastric fold leading towards the gonoduct. This fold is the only connection between the spermathecae and the oviduct. Such folds have also been found in other Linyphiids, including *Lepthyphantes* (Saaristo & Tanasevitch 1996) and *Batyphantes gracilis* (Blackwall, 1841) (M. Saaristo pers. comm.).

Histological sections depicted by Engelhardt (1910) on *Linyphia triangularis* (Clerck, 1757) also point in this direction (but see van Helsdingen 1969).

In *P. phrygianus*, various apical parts of the male palp (the 'embolic division' sensu Merrett 1963) probably couple to the knob of the u-shaped base, an interpretation derived from the position of the mating plug. If this is the case, males are not expected to be able to physically manipulate stored sperm masses directly. The predicted sperm priority pattern based on female genital morphology is last male priority against which the production of a mating plug evolved.

Other species

Investigations on sperm priority patterns are generally rare for spiders, and information on genital morphology and copulatory mechanism for these species is often unavailable. However, there is some information on both aspects for Tetragnathidae and Theridiidae. Published data on sperm priority are summarized in Elgar (1998) and Uhl & Vollrath (1998).

The Tetragnathinae is an entelegyne spider subfamily, in which female genitalia of the haplogyne condition occur. West & Toft (1999) found that the last male fertilized about 70% of the eggs in first egg sacs of *Tetragnatha extensa* (Linnaeus, 1758) (Tab.1). From what is known about male and female genital morphology of *Tetragnatha* species it is questionable whether the male pedipalp has access to the spermathecae (e.g. Wiehle 1963; Uhl et al. 1992), which would lead to the prediction of last male sperm priority as was found for *T. extensa*. However, in a drawing of genitalia of *T. montana* Simon, 1900 in functional contact, it seems as if the emboli were inserted into the spermathecae (Huber & Senglet 1997). Unfortunately, female genitalia are not fully depicted which leaves copulatory mechanism obscure. The tetragnathid *Leucauge mariana* (Keyserling, 1881), on the other hand, possesses a conduit design with three successive sperm storage chambers (Eberhard & Huber 1998). Moreover, the male embolus may reach as far as to the lumen of the

first chamber and may thus be able to manipulate stored sperm.

In the entelegyne genus *Latrodectus* (Theridiidae), genital morphology of both sexes does not vary considerably between species. The two spermathecae are heavily sclerotized, dumb-bell shaped structures with anterior and posterior lobes. These lobes are connected by an intermediate, more slender part. In some species like in *L. hesperus* Chamberlin & Ivie, 1935, the spermathecae are of the functional cul-de-sac type with copulatory and fertilization duct close together (Bhatnagar & Rempel 1962, misidentified as *L. curacaviensis*), whereas in other species (e.g. *L. hystrix* Simon, 1890; *L. geometricus* C.L. Koch, 1841; *L. cinctus* Blackwall, 1865; *L. renivulvatus* Dahl, 1902; Knoflach & van Harten 2002; *L. revivensis*: Berendonck & Greven 2002) the ducts are further apart. At the end of copulation, the tip of the embolus typically breaks off and remains either in the narrow entrance of the spermatheca, as for example in *L. dahli* Levi, 1959, or is deeply inserted in the lumen of the spermatheca (*L. geometricus*) (Knoflach & Van Harten 2002). The broken embolus tips do not necessarily prevent the female from remating, but in the case of *L. renivulvatus*, in which whole emboli may break and plug the copulatory ducts (Knoflach & van Harten 2002) successful remating seems impossible. In *L. dahli* and *L. geometricus* Knoflach & van Harten report cases in which two tips were found in one spermathecal entrance. For the latter species Müller (1985) detected four tips in the spermatheca and one in the bursa. In *L. hesperus* and *L. revivensis*, however, only a single tip was found in each spermathecal entrance, whereas several more could be found in the bursa (Bhatnagar & Rempel 1962; Berendonck & Greven 2002). The data from *L. hesperus*, *L. revivensis* and *L. renivulvatus* suggest that successful intromission of palps is only possible once for each spermatheca. If a male is allowed to inseminate only one spermatheca but not the other, an additional copulation with another male may occur, which will lead to sperm of different males being stored in different storage

organs. The only information on sperm priority we have to date is from a study on yet another species, *L. hasselti* Thorell, 1870, in which sperm mixing with considerable variation occurs (Andrade 1996). It remains to be clarified whether mixed paternity in *L. hasselti* results from sperm mixing in a given spermathecae or from activation of sperm stored in different spermathecae. The latter situation may explain why some males have no paternity success whereas others fertilize 100% of the eggs.

Is sperm stratification a plausible assumption?

In the testis, male spiders produce encapsulated spermatozoa. These remain encapsulated during sperm induction into the male pedipalp and are transferred in an encapsulated state to the female (Alberti 1990). Whereas in *Leucauge mariana* (Eberhard & Huber 1998) sperm activation (better: decapsulation) within the female occurs soon after insemination, sperm are stored in an inactive state in *Pholcus phalangioides*, probably until shortly before egg laying (Uhl 1994a). Both studies investigated the conditions of sperm in situ, in the female sperm storage organ. A study on *Nephila clavipes* (Brown 1985) demonstrated that decapsulation took 7 to 18 days from mating, depending on whether the female had moulted the same day or mated later in adulthood. Brown squeezed the spermathecal content onto a slide containing physiological saline, which may have influenced the results as sperm become active when transferred to physiological saline. It should be noted that decapsulated sperm do not necessarily move in the female genital tract as it may require additional stimuli for sperm to become mobile. These findings show that although sperm stratification may occur in some taxa it is not a general characteristic of spiders. Female glandular secretion seems to trigger the process of activation both in *L. mariana* and *P. phalangioides* (Eberhard & Huber 1998; Uhl 1994b), which suggests that females have the potential of biasing the fertilization success of rival males by selectively activating stored sperm.

CONCLUSION

Each of the four spiders investigated deviates considerably from the assumptions underlying the modified Austad's hypothesis. Obviously, female genital morphology is extremely variable even within families. Data on morphology and sperm usage show that sperm precedence patterns cannot be predicted by the number of ducts connected to a spermatheca. Even if species are classified according to their specific design of female genital morphology, independent of their phylogenetic position, and even if knowledge on copulatory mechanisms is included, predictions on sperm precedence patterns are difficult to make. Unfortunately, there is only little information on detailed genital morphology, copulatory mechanisms and P_2 values for single spider species which would help to clarify the matter. Beyond morphology, manifold processes of male and female manipulation, male and female age, remating intervals and body size may influence sperm transfer, storage and usage. Adaptation, not constraints seems to play the major role in shaping sperm priority patterns. Thus, researchers should refrain from assuming particular precedence patterns solely on the basis of either taxonomic classification or the number of ducts present.

ACKNOWLEDGEMENTS

I cordially thank Søren Toft and Nikolaj Scharff for inviting me to give this talk at the 19th European Colloquium of Arachnology in Århus. William Eberhard, Jutta Schneider, Tina Berendonck, Barbara Knoflach-Thaler, Michael Schmitt and an anonymous referee gave valuable comments on a previous draft of the manuscript. I am especially grateful to Bill Eberhard, whose thorough comments helped to substantially improve this paper.

REFERENCES

- Alberti, G. 1990. Comparative spermatology of Araneae. *Acta Zoologica Fennica* 190, 17-34.
 Andrade, M.C.B. 1996. Sexual selection for male sacrifice in the Australian redback spider. *Science* 271, 70-72.

- Austad, S.N. 1982. First male sperm priority in the bowl and doily spider, *Frontinella pyramitela* (Walckenaer). *Evolution* 36, 777-785.
- Austad, S.N. 1984. Evolution of sperm priority patterns in spiders. In: *Sperm competition and the evolution of mating systems*. (R.L. Smith ed.), pp. 223-249. Harvard University Press, Cambridge Mass.
- Berendonck, B. & Greven, H. 2002. Morphology of female and male genitalia in *Latrodectus revivensis* Shulov, 1948 (Araneae, Theridiidae) with regards to sperm priority patterns. In: *European Arachnology 2000* (S. Toft & N. Scharff eds.), pp. 157-167. Aarhus University Press, Aarhus.
- Bhatnagar, R.D.S. & Rempel, J.G. 1962. The structure, function, and postembryonic development of the male and female copulatory organs of the black widow spider *Latrodectus curacaviensis* (Müller). *Canadian Journal of Zoology* 40, 465-510.
- Boorman, E. & Parker, G.A. 1976. Sperm (ejaculate) competition in *Drosophila melanogaster*, and the reproductive value of females to male in relation to female age and mating status. *Ecological Entomology* 1, 145-155.
- Brown, S.G. 1985. Mating behavior of the golden-orb-weaving spider, *Nephila clavipes*: II. Sperm capacitation, sperm competition, and fecundity. *Journal of Comparative Psychology* 99, 167-175.
- Bukowski, T.C. & Christenson, T.E. 1997. Determinants of sperm release and storage in a spiny orbweaving spider. *Animal Behaviour* 53, 381-395.
- Christenson, T.E. & Cohn, J. 1988. Male advantage for egg fertilization in the golden orb-weaving spider (*Nephila clavipes*). *Journal of Comparative Psychology* 102, 312-318.
- Coddington, J.A. & Levi, H.W. 1991. Systematics and evolution of spiders (Araneae). *Annual Review of Ecology and Systematics* 22, 565-92.
- Cohn, J. 1990. Is it the size that counts? Palp morphology, sperm storage, and egg hatching frequency in *Nephila clavipes* (Araneae, Araneidae). *Journal of Arachnology* 18, 59-71.
- Cooke, J.A.L. 1966. Synopsis of the structure and function of the genitalia in *Dysdera crocata*. *Senckenbergiana Biologica* 47, 35-43.
- Eberhard, W.G. 1996. *Female Control: Sexual Selection by Cryptic Female Choice*. Monographs in Behavior and Ecology, Princeton University Press, New Jersey.
- Eberhard, W.G. & Huber, B.A. 1998. Courtship, copulation, and sperm transfer in *Leucauge mariana* (Araneae, Tetragnathidae) with implications for higher classification. *Journal of Arachnology* 26, 342-368.
- Eberhard, W.G., Guzman-Gomez, S. & Catley, K.M. 1993. Correlation between spermathecal morphology and mating systems in spiders. *Biological Journal of the Linnean Society* 50, 197-209.
- Eberhard, W.G., Huber, B.A., Rodriguez, S.R.L., Briceño, R.D., Salas, I. & Rodriguez, V. 1998. One size fits all? Relationships between the size and degree of variation in genitalia and other body parts in twenty species of insects and spiders. *Evolution* 52, 415-431.
- Elgar, M.A. 1998. Sperm competition and sexual selection in spiders and other arachnids. In: *Sperm competition and sexual selection*. (T. R. Birkhead & A.P. Møller eds.), pp. 307-339. Academic Press, San Diego.
- Engelhardt, V. 1910. Beiträge zur Kenntnis der weiblichen Copulationsorgane einiger Spinnen. *Zeitschrift für Wissenschaftliche Zoologie* 96, 32-117.
- Huber, B.A. 1995. Copulatory mechanism in *Holocnemus pluchei* and *Pholcus opilionoides*, with notes on male cheliceral apophyses and stridulatory organs in Pholcidae (Araneae). *Acta Zoologica* 76, 291-300.
- Huber, B.A. 1996. On American 'Micromerys' and *Metagonia* (Araneae, Pholcidae), with notes on natural history and genital mechanics. *Zoologica Scripta* 25, 341-362.
- Huber, B.A. & Eberhard, W.G. 1997. Courtship, copulation, and genital mechanics in *Physocyclus globosus* (Araneae, Pholcidae). *Canadian Journal of Zoology* 74, 905-918.

- Kaster, J.L. & Jakob, E.M. 1997. Last-male sperm priority in a haplogyne spider (Araneae: Pholcidae): correlations between female morphology and patterns of sperm usage. *Annals of the Entomological Society of America* 90, 254-259.
- Knoflach, B. & van Harten, A. 2002. The genus *Latrodectus* (Araneae: Theridiidae) from mainland Yemen, the Socotra Archipelago and adjacent countries. *Fauna of Arabia* 19, 321-361.
- Lewontin, R.C. 1966. On the measurement of relative variability. *Systematic Zoology* 15, 141-142.
- Merrett, P. 1963. The palps of male spiders of the family Linyphiidae. *Proceedings of the Zoological Society* 140, 347-467.
- Müller, G.H. 1985. Abgebrochene Emboli in der Vulva der 'Schwarzen Witwe' *Latrodectus geometricus* C.L. Koch 1841 (Arachnida: Araneae: Theridiidae). *Entomologische Zeitschrift* 95, 27-30.
- Pitnick, S. Markow, T. & Spicer G.S. 1999. Evolution of multiple sperm storage organs in *Drosophila*. *Evolution* 53, 1804-1822.
- Saaristo, M.I. & Tanasevitch, A.V. 1996. Redelimitation of the subfamily Micronetinae Hull, 1920 and the genus *Lepthyphantes* Menge, 1866 with descriptions of some new genera (Aranei, Linyphiidae). *Berichte des nat.-med. Vereins Innsbruck* 83, 163-186.
- Schäfer, M.A. & Uhl, G. 2002. Determinants of paternity success in the spider *Pholcus phalangioides* (Pholcidae: Araneae): the role of male and female mating behaviour. *Behavioural Ecology and Sociobiology* 51, 368-377.
- Schneider, J.M. & Elgar, M.A. 2001. Sexual cannibalism and sperm competition in the golden orb-spider *Nephila plumipes* (Araneoida): female and male perspective. *Behavioural Ecology* 12, 547-552.
- Schneider, J.M., Herberstein, M.E., de Creispigny, F.C., Ramamurthy, S. & Elgar, M.A. 2000. Sperm competition and small size advantage for males of the golden orb-web spider *Nephila edulis*. *Journal of Evolutionary Biology* 13, 939-946.
- Schult, J. 1980. Die Genitalstrukturen haplogyner Araneae unter phylogenetischem Aspekt (Arachnida). PhD thesis University Hamburg.
- Simmons, L.W. & Siva-Jothy, M.T. 1998. Sperm competition in insects: mechanisms and the potential for selection. In: *Sperm competition and sexual selection*. (T.E. Birkhead & A.P. Møller eds.), pp. 341-434. Academic Press, San Diego.
- Uhl, G. 1993a. Sperm storage and repeated egg production in female *Pholcus phalangioides* Fuesslin (Araneae). *Bulletin de la Société Neuchâteloise des Sciences Naturelles* 116 (1), 245-252.
- Uhl, G. 1993b. Mating behaviour and female sperm storage in *Pholcus phalangioides* (Fuesslin) (Araneae). *Memoirs of the Queensland Museum* 33, 667-674.
- Uhl, G. 1994a. Genital morphology and sperm storage in *Pholcus phalangioides* (Fuesslin, 1775) (Pholcidae; Araneae). *Acta Zoologica* 75, 1-12.
- Uhl, G. 1994b. Ultrastructure of the accessory glands in female genitalia of *Pholcus phalangioides* (Fuesslin, 1775) (Pholcidae; Araneae). *Acta Zoologica* 75, 13-25.
- Uhl, G. 2000. Two distinctly different sperm storage organs in female *Dysdera erythrina* (Araneae: Dysderidae). *Arthropod Structure and Development* 29, 163-169.
- Uhl, G. & Gunnarsson B. 2001. Female genitalia in *Pityohyphantes phrygianus*: a spider with skewed sex ratio. *Journal of Zoology* 255, 367-376.
- Uhl, G. & Vollrath, F. 1998. Genital morphology of *Nephila edulis*: implications for sperm competition in spiders. *Canadian Journal of Zoology* 76, 39-47.
- Uhl, G. & Vollrath, F. 2000. Extreme body size variability in the golden silk spider (*Nephila edulis*) does not extend to genitalia. *Journal of Zoology* 251, 7-14.
- Uhl, G.; Huber, B.A. & Rose, W. 1995. Male pedipalp morphology and copulatory mechanism in *Pholcus phalangioides* (Fuesslin, 1775) (Araneae, Pholcidae). *Bulletin of the British Arachnological Society* 10, 1-9.

- Uhl, G., Sacher, P., Weiss, I. & Kraus, O. 1992. Europäische Vorkommen von *Tetragnatha shoshone* (Arachnida, Araneae, Tetragnathidae). *Verhandlungen des naturwissenschaftlichen Vereins Hamburg N.F.* 33, 247-261.
- Van Helsdingen, P. J. 1969. A reclassification of the species of *Linyphia* based on the functioning of the genitalia (Araneae, Linyphiidae), I. *Zoologische Verhandlungen* 105, 1-303.
- Vollrath, F. 1980. Male body size and fitness in the web-building spider *Nephila calvipes*. *Zeitschrift für Tierpsychologie* 53, 61-78.
- Walker, W.F. 1980. Sperm utilization strategies in nonsocial insects. *American Naturalist* 115, 780-799.
- West, H.P. & Toft, S. 1999. Last-male sperm priority and the mating system of the haplogyne spider *Tetragnatha extensa* (Araneae: Tetragnathidae). *Journal of Insect Behavior* 12, 433-450.
- Wiehle, H. 1933. *Holocnemus hispanicus* sp.n. und die Gattungen *Holocnemus* Simon und *Crossoprisa* Simon. *Zoologischer Anzeiger* 104, 241-252.
- Wiehle, H. 1963. Spinnentiere oder Arachnoidea (Araneae). XII. Tetragnathidae—Streckspinnen und Dickkiefer. In: *Die Tierwelt Deutschlands und der angrenzenden Meeressteile* (F. Dahl ed.), pp. 1-76. Gustav Fischer, Jena.
- Yoward, P.J. 1998. Sperm competition in *Pholcus phalangioides* (Fuesslin, 1775) (Araneae, Pholcidae) - shorter second copulations gain a higher paternity reward than first copulations. In: *Proceedings of the 17th European Colloquium of Arachnology, Edinburgh 1997* (P.A. Selden ed.), pp. 167-170. British Arachnological Society, Burnham Beeches, Bucks.