EXPLORING FUNCTIONAL ASSOCIATIONS BETWEEN SPIDER CRIBELLA AND CALAMISTRA

Brent D. Opell, **Jamel S. Sandidge**[1] and **Jason E. Bond**[2]:   Department of Biology, Virginia Polytechnic Institute and State University, Blacksburg, Virginia 24061 USA

ABSTRACT.   A spider's calamistrum draws silk fibrils from its cribellum and helps combine them with supporting strands to form a cribellar prey capture thread. Despite the close functional association of these two features, this study shows that there is a great deal of variability in the ratio of cribellum width to calamistrum length. When the independent contrast method was used to examine these two features in 11 species representing seven families, no relationship was found. Likewise, no relationship was found among nine species representing seven genera of the family Uloboridae. Only among the 14 species of *Mallos* (Dictynidae) was calamistrum length directly related to cribellum width. This suggests that, above the genus level, differences in spinning behavior and morphological features such as leg length and abdomen size and shape influence the relationship of these two features.

Keywords: cribellar thread, cribellate spiders, independent contrast method, functional linkage

The outer surfaces of a cribellar capture thread are formed of thousands of fine, looped fibrils that are produced by spinning spigots on the cribellum (Eberhard & Pereira 1993; Opell 1994a, 1995, 1996; Peters 1983, 1984, 1986, 1992). These fibrils are drawn from the cribellum and manipulated by a setal comb on the fourth walking leg, termed the calamistrum, as they are combined with axial and, in some cases, paracribellar fibers to form a completed capture thread. This functional linkage between the calamistrum and the cribellum suggests that their features should also be closely related. The most obvious features to exhibit this relationship should be calamistrum length and cribellum width. We predict that the calamistrum must be long enough to fully span the cribellum as it sweeps over it in a combing motion. However, cribellum width may not be the only factor that influences calamistrum length. The effective length of a calamistrum is probably determined by such factors as the angle at which the calamistrum passes over the cribellum and the lateral movement of the calamistrum during a combing stroke. Although these features

and their relationships are poorly studied, they are likely to be affected by the length and width of a spider's abdomen, the length of a spider's fourth legs, by the manner in which the combing leg is supported (Eberhard 1988), and probably by other details of the combing behavior such as the length of each combing stroke.

The diversity in cribellar thread-combing behavior documented by Eberhard (1988) suggests that the ratio of calamistrum length to cribellum width may differ considerably among cribellate taxa. The null hypothesis of this study is that this ratio is uniform for all cribellate taxa. Using the comparative method of phylogenetic systematics (Harvey & Pagel 1991), we test this hypothesis at three hierarchical levels: the interfamilial level, the intrafamilial level, and the intrageneric level. The degree to which differences in behavior and other aspects of anatomy influence the ratio of calamistrum length to cribellum width will affect the level at which the null hypothesis will be rejected. As behavioral and morphological features should be most similar within members of the same genus, it should be more difficult to reject the null hypothesis at this level than at more inclusive levels.

METHODS

Measurements.—The fourth legs and cribella of spiders were removed and mounted in   water-soluble   medium   on   microscope

[1] *Current address:*   Dept. of Entomology, University of Kansas, Lawrence, Kansas 66045 USA
[2] *Current address:*   Dept. of Zoology, Insect Division, Field Museum of Natural History, Roosevelt Road at Lake Shore Drive, Chicago, Illinois 60605 USA

Table 1.—Means and standard deviations of the ratio calamistrum length to cribellum width of representative species.

Family Species	n	$\bar{x}$	SD
Uloboridae			
Miagrammopes animotus	31	1.35	0.10
Uloborus glomosus	21	1.10	0.13
Octonoba sinensis	24	1.27	0.11
Dictynidae			
Mexitilia trivittata	6	1.67	0.24
Mallos bryantii	5	1.55	0.25
Mallo niveus	9	1.58	0.27
Mallos mians	8	1.49	0.19

slides. Calamistrum length and cribellum width were measured to at least the nearest 20μm under a compound or dissecting microscope. Two indices can be used for calamistrum length: the distance separating the tips of the proximal and distal-most setae of the calamistrum and the distance separating the proximal and distal-most setal bases. We chose the second index for two reasons. First, it can be more consistently measured and is not affected by missing setae. Second, it does not make any assumptions about the deflection of calamistrum setae during cribellar fibril combing. In the case of those species with divided cribella, cribellum width included the central region that separated the two halves of the cribellum. We measured a single mature female per species. We reasoned that, as the cribellum and calamistrum must be functionally linked throughout an individual's development, these measurements would provide a more rigorous test of the hypothesis than would the use of mean values derived from several individuals of a species. Table 1 gives the variance of the ratio of calamistrum length to cribellum width for seven species included in this study.

Phylogenetic analysis.—This study includes representatives of the infraorder Araneomorphae, the family Uloboridae, and the dictynid genus *Mallos* O. Pickard-Cambridge 1902 (Figs. 1–3) and uses the phylogenies of Griswold et al. (in press), Coddington (1990), and Bond & Opell (1997), respectively. To analyze the relationships of calamistrum length and cribellum width in a phylogenetic context we used the independent contrasts method of

Felsenstein (1985), as implemented by the Comparative Analysis of Independent Contrasts program of Purvis & Rambaut (1995). All branch lengths were treated as equal. This method minimizes the influence of non-independence of the data due to phylogenetic relationship by analyzing directional changes in continuous characters. It does so by computing differences between the features of sister taxa (both extant taxa and their inferred ancestors). These differences are then normalized and relationships among the resulting independent contrast values are examined using regression statistics (see Harvey & Pagel 1991 for a review of this approach).

All known species of the genus *Mallos* were included in the analysis of the relationship between the calamistrum length and cribellum width. In contrast, analyses of the other two clades included only some of the known members. We examined the consequences of partial sampling by analyzing the relationship between calamistrum length and cribellum width within subsets of the genus *Mallos*. We used a random number generator to select seven of the 14 species of *Mallos*. After constructing a pruned phylogeny that included these seven species and *Mexitilia trivittata* (Banks 1901) as an outgroup, we ran an independent contrast analysis for calamistrum length and cribellum width. This procedure was repeated until a total of ten analyses had been run. We then repeated the entire procedure a second time with nine species of *Mallos* being selected each time.

RESULTS

Values for calamistrum length and cribellum width are given in Figs. 1–3. Within the Araneomorphae, the ratio of calamistrum length to cribellum width ranged from 0.99–2.57; and an independent contrast analysis showed that there was no relationship between the dimensions of these two features ($F = 0.09$, $R^2 = 0.01$, $P = 0.77$). Within the Uloboridae the ratio of calamistrum length to cribellum width ranged from 1.07–2.06 and an independent contrast analysis showed that there was no relationship between the dimensions of these two features ($F = 0.63$, $R^2 = 0.10$, $P = 0.46$). When this analysis is restricted to orb-weaving uloborids of the genera *Waitkera* Opell 1979, *Siratoba* Opell 1979, *Uloborus* Latreille 1806, *Octonoba* Opell

Figure 1.

Filistatidae
Kukulcania hibernalis 479, 472
(Hentz 1842) (0.99)

Oecobiidae
Platoecobius floridanus 218, 392
(Banks 1896) (1.80)

Uloboridae
Waitkera waitakerensis 381, 780
(Chamberlain 1946) (2.04)

Uloborus glomosus 574, 672
(Walckenaer 1837) (1.17)

Dictynidae
Mexitilia trivittata 448, 816
(Banks 1901) (1.82)

Mallos bryanti 480, 760
Gertsch 1946 (1.58)

Amaurobiidae
Callobius bennetti 686, 1102
(Blackwall 1846) (1.61)

Neolanidae
Neolana pallida 444, 1142
Forster & Wilton 1973 (2.57)

Desidae
Matachia livor 840, 959
(Urquhart 1892) (1.14)

Badumna insignis 697, 1499
(L. Koch 1872) (2.15)

Badumna longinqua 664, 1204
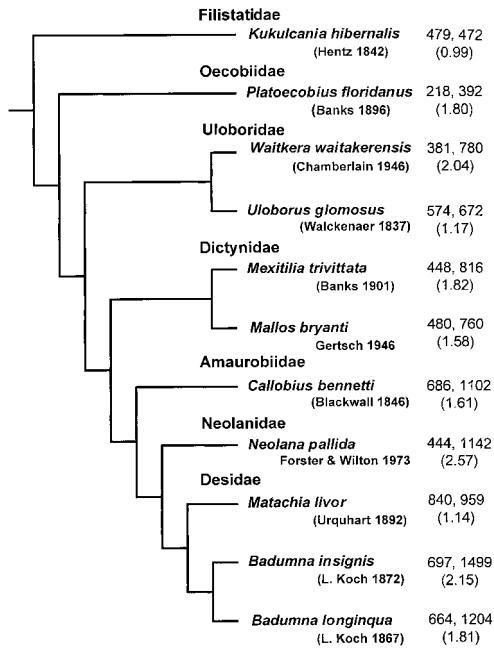(L. Koch 1867) (1.81)

Figure 1.—Phylogeny of species representing seven families (from Griswold et al. 1999). Following each species is the width of its cribellum and the length of its calamistrum, both in μm. Ratios of calamistrum length to cribellum width are in parentheses.

Waitkera waitakerensis 381, 780
(Chamberlain 1946) (2.04)

Siratoba referena 223, 460
(Muma & Gertsch 1964) (2.06)

Hyptiotes cavatus 390, 740
(Hentz 1847) (1.90)

Hyptiotes gertschi 465, 820
Chamb. & Ivie 1935 (1.76)

Miagrammopes animotus 540, 700
Chickering 1968 (1.30)

Miagrammopes species 426, 720
(1.69)

Uloborus glomosus 574, 672
(Walckenaer 1837) (1.17)

Octonoba sinensis 560, 600
(Simon 1880) (1.07)

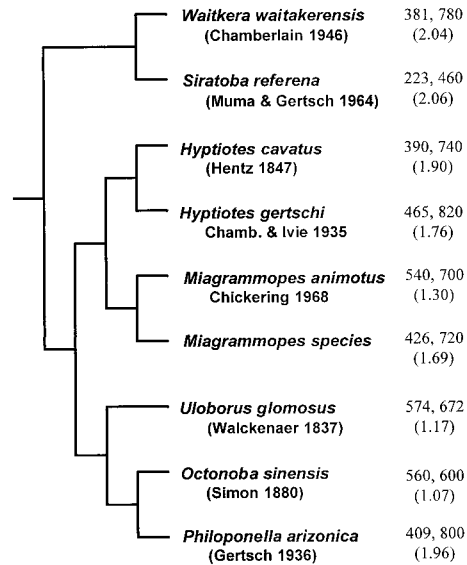*Philoponella arizonica* 409, 800
(Gertsch 1936) (1.96)

Figure 2.—Phylogeny of species belonging to the family Uloboridae (from Coddington 1990). Following each species is the width of its cribellum and the length of its calamistrum, both in μm. Ratios of calamistrum length to cribellum width are in parentheses.

1979, and *Philoponella* Mello-Leitão 1917 an independent contrast analysis still fails to show a relationship between calamistrum length and cribellum width ($F = 0.11$, $R^2 = 0.05$, $P = 0.77$).

Within the genus *Mallos*, the calamistrum length to cribellum width ratio ranged only from 1.26–1.82 and an independent contrast analysis showed that there was a relationship ($F = 8.40$, $R^2 = 0.41$, $P = 0.013$) between the dimensions of these two features (Fig. 4). However, in only three of the ten subsets that included seven *Mallos* species plus *Mexitilia trivittata* was there a significant relationship between calamistrum length and cribellum width ($F = 8.91–22.75$, $R^2 = 0.64–0.82$, $P = 0.005–0.031$). When the sample size was increased to include nine *Mallos* species, seven of the ten samples showed a relationship between these features ($F = 5.61–19.95$, $R^2 = 0.45–0.74$, $P = 0.050–0.003$).

DISCUSSION

The size of a spider's cribellum and the number of spigots that it bears are the main factors that correlate with the stickiness of the cribellar thread that it produces (Opell 1994a, 1995, in press). However, differences in the way cribellar fibrils are combined with supporting fibers can alter thread stickiness (Opell 1994b), as can the deposition of linear cribellar threads in a looped manner when they are placed in the web (Opell, unpub. data). Although cribellum shape differs among taxa, spigot number is generally related to cribellum width. This evolutionary plasticity in cribellum width is reflected by differences in calamistrum length.

The ratio of calamistrum length to cribellum width differs among taxa; but, with one exception, it always exceeds one. In *Kululcania hibernalis* (Hentz 1842) calamistrum length and cribellum width are essentially the same. This suggests that the production of a cribellar thread requires the calamistrum to span the complete width of the cribellum during a combing stroke. It is possible that a calamistrum could comb fibrils from only part of the cribellum spigots, but this seems unlikely for two reasons. First, as the spigots of the cribellum are probably not regionally controlled, non-calamistrum setae on other parts
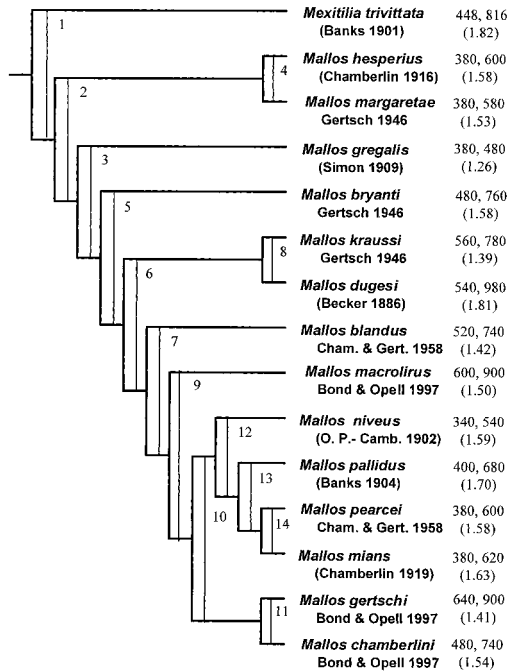
Mexitilia trivittata (Banks 1901)	448, 816	(1.82)
Mallos hesperius (Chamberlin 1916)	380, 600	(1.58)
Mallos margaretae Gertsch 1946	380, 580	(1.53)
Mallos gregalis (Simon 1909)	380, 480	(1.26)
Mallos bryanti Gertsch 1946	480, 760	(1.58)
Mallos kraussi Gertsch 1946	560, 780	(1.39)
Mallos dugesi (Becker 1886)	540, 980	(1.81)
Mallos blandus Cham. & Gert. 1958	520, 740	(1.42)
Mallos macrolirus Bond & Opell 1997	600, 900	(1.50)
Mallos niveus (O. P.- Camb. 1902)	340, 540	(1.59)
Mallos pallidus (Banks 1904)	400, 680	(1.70)
Mallos pearcei Cham. & Gert. 1958	380, 600	(1.58)
Mallos mians (Chamberlin 1919)	380, 620	(1.63)
Mallos gertschi Bond & Opell 1997	640, 900	(1.41)
Mallos chamberlini Bond & Opell 1997	480, 740	(1.54)

Figure 3.—Phylogeny of the 14 known species of *Mallos* and a representative of its sister group *Mexitilia* (from Bond & Opell 1997). Following each species is the width of its cribellum and the length of its calamistrum, both in μm. Ratios of calamistrum length to cribellum width are in parentheses. Numbers near vertical lines denote the sister groups whose independent contrasts are given in Figure 4.
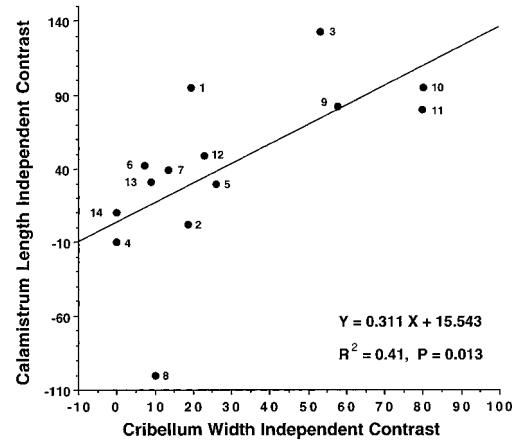

Figure 4.—Regression of independent contrast values for cribellum width and calamistrum length for 14 *Mallos* species and *Mexitilia trivittata*. Numbers identify the sister groups in Figure 3 from which these values were computed.

of the combing leg that contacted cribellum spigots would tend to draw fibrils from them and these would become stuck to the leg or catch on the forming cribellar thread, thereby interfering with cribellar thread production. Second, cribellar thread is materially costly to produce (Opell 1997, 1998) and it seems unlikely that a cribellum with an unused lateral region would be retained. The apparent ease with which the cribellum itself is lost is documented by a number of families, genera, and even species pairs (putative sister species) that have both cribellate and ecribellate members (Forster 1970; Forster & Wilton 1973).

This study shows that at higher taxonomic levels, there is no uniform relationship between cribellum width and calamistrum length. This suggests that the angle at which a calamisturm passes over a cribellum or the amount of lateral movement of the calamistrum during a combing stroke differs greatly

among spiders. As noted in the introduction, a variety of morphological and behavioral factors may influence the position and path of the calamistrum.

Even among orb-weaving species of the family Uloboridae that support the combing leg in the same manner (Eberhard 1988; Opell unpub. obs. for *Waitkera waitakerensis* (Chamberlain 1946)*, Siratoba referena* (Muma & Gertsch 1946), *Uloborus glomosus* (Walckenaer 1837), *Octonoba sinensis* (Simon 1880)) and share more similar body plans (abdomen dimensions, leg lengths, and ratios of leg articles; Opell 1979), the ratio of calamistrum length to cribellum width differs considerably. It is only within the genus *Mallos* that a clade-specific correlation between calamistrum length and cribellum width can be demonstrated. Even here this relationship is not exceedingly strong, as it begins to decay when sample size decreases.

As comparisons of calamistrum length and cribellum width within the family Uloboridae and among families are based on small samples, it is possible that an increased sample size would establish a significant relationship between these features. However, in comparisons of other spider features similar phylogenetic representation has been sufficient to demonstrate significant relationships (Opell 1994a, 1996, 1997, 1998, 1999, in press). Therefore, if there is a general relationship be-

tween calamistrum and cribellum features, it is weaker than those of other aspects of the phenotype.

ACKNOWLEDGMENTS

LITERATURE CITED

Banks, N. 1896. New North American spiders and mites. Trans. American Entomol. Soc., 23:57–77.

Banks, N. 1901. Some Arachnida from New Mexico. Proc. Acad. Nat. Sci., Philadelphia, 53:581–590.

Banks, N. 1904. Some Arachnida from California. Proc. California Acad. Sci., 3:331–376.

Becker, L. 1886. Diagnoses de quelques Arachnides nouveaux. Ann. Soc. Entomol. Belgique, 30:XXIII–XXVII.

Blackwall, J. 1846. Notice of spiders captured by Professor Potter in Canada, with descriptions of such spiders as appear to be new to science. Ann. Mag. Nat. Hist., 17:30–44, 76–82.

Bond, J.E. & B.D. Opell. 1997. Systematics of the spider genera *Mallos* and *Mexitilia* (Araneae, Dictynidae). Zool. J. Linn. Soc., 119:389–445.

Chamberlain, G. 1946. Revision of the Araneae of New Zealand (Part 2). Rec. Auckland Inst. Mus., 3:85–97.

Chamberlin, R.V. 1916. Results of the Yale Peruvian Expedition of 1911: The Arachnida. Bull. Mus. Comp. Zool., 60:177–299.

Chamberlin, R.V. 1919. New Californian spiders. J. Entomol. Zool., Claremont, 12:1–17.

Chamberlin, R.V. & W.J. Gertsch. 1958. The spider family Dictynidae in America north of Mexico. Bull. American Mus. Nat. Hist., 116:1–152.

Chamberlin, R.V. & W. Ivie. 1935. Miscellaneous new American spiders. Bull. Univ. Utah, 26:1–79.

Chickering, A.M. 1968. The genus *Miagrammopes* (Araneae, Uloboridae) in Panama and the West Indies, Brev. Mus. Comp. Zool., 289:1–28.

Coddington, J.A. 1990. Ontogeny and homology in the male palpus of orb-weaving spiders and their relatives, with comments on phylogeny (Araneoclada: Araneoidea, Deinopoidea). Smith. Contrib. Zool., 496:1–52.

Eberhard, W.G. 1988. Combing and sticky silk attachment behaviour by cribellate spiders and its taxonomic implications. Bull. British Arachnol. Soc., 7:247–251.

Eberhard, W.G. & F. Pereira. 1993. Ultrastructure of cribellate silk of nine species in eight families and possible taxonomic implications. (Araneae: Amaurobiidae, Deinopidae, Desidae, Dictynidae, Filistatidae, Hypochilidae, Stiphidiidae, Tengellidae). J. Arachnol., 21:161–174.

Felsenstein, J. 1985. Phylogenies and the comparative method. American Nat., 125:1–15.

Forster, R.R. 1970. The spiders of New Zealand, part III: Desidae, Dictynidae, Hahniidae, Amaurobioididae, Nicodamidae. Otago Mus. Bull., 3:1–184.

Forster, R.R. & C.L. Wilton. 1973. The spiders of New Zealand, part IV: Agelenidae, Stiphiidae, Amphinectidae, Amaurobiidae, Neolanidae, Ctenidae, Psechridae. Otago Mus. Bull., 4:1–309.

Gertsch, W.J. 1936. Further diagnosis of new American spiders. American Mus. Nov., 852:1–27.

Gertsch, W.J. 1946. Notes on American spiders of the family Dictynidae. Amererican Mus. Nov., 1319:1–21.

Griswold, C.E., J.A. Coddington, N.I. Platnick, & R.R. Forster. 1999. Towards a phylogeny of entelegyne spiders (Araneae, Entelegynae). J. Arachnol.,27(1):53–63.

Harvey, P.H. & M.D. Pagel. 1991. The Comparative Method in Evolutionary Biology. Oxford Univ. Press, New York.

Hentz, N.M. 1842. Descriptions and figures of Araneides of the United States. Boston J. Nat. Hist., 4:54–57.

Hentz, N.M. 1847. Descriptions and figures of Araneides of the United States. Boston J. Nat. Hist., 5:443–478.

Koch, L. 1867. Beschreibungen neuer Arachniden und Myriapoden. Verh. Zool. Bot. Ges. Wien, 17:173–250.

Koch, L. 1872. Die Arachniden Australiens, nach der Natur beschrieben und abgebildet, Part 2, Pp. 105–368. Nürnberg.

Latreille, P.A. 1806. Genera Crustaceorum et Insectorum, Aranéides, Part 1: Aranéides, pp. 82–127. Paris.

Mello-Leitão, C.F. 1917. Generos e species novas de Araneidos. Arch. Esc. Sup. Agr. Med. Veter., 1:3–19.

Muma, M.M. & W.J. Gertsch. 1964. The spider

family Uloboridae in North America north of Mexico. American Mus. Nov., 2196:1–43.

Opell, B.D. 1979. Revision of the genera and tropical American species of the spider family Uloboridae. Bull. Mus. Comp. Zool., 148:433–549.

Opell, B.D. 1994a. Factors governing the stickiness of cribellar prey capture threads in the spider family Uloboridae. J. Morph., 221:111–119.

Opell, B.D. 1994b. Increased stickiness of prey capture threads accompanying web reduction in the spider family Uloboridae. Funct. Ecol., 8:85–90.

Opell, B.D. 1995. Ontogenetic changes in cribellum spigot number and cribellar prey capture thread stickiness in the spider family Uloboridae. J. Morph., 224:47–56.

Opell, B.D. 1996. Functional similarities of spider webs with diverse architectures. American Nat., 148:630–648.

Opell, B.D. 1997. The material cost and stickiness of capture threads and the evolution of orb-weaving spiders. Biol. J. Linnean Soc., 62:443–458.

Opell, B.D. 1998. Economics of spider orb-webs: the benefits of producing adhesive capture thread and of recycling silk. Funct. Ecol., 12:613–624.

Opell, B.D. 1999. Redesigning spider webs: stickiness, capture area, and the evolution of modern orb-webs. Evol. Ecol. Res. In press.

Opell, B.D. In press. Changes in spinning anatomy and thread stickiness associated with the origin of orb-weaving spiders. Biol. J. Linnean Soc.

Peters, H.M. 1983. Struktur und Herstellung der Fangfäden cribellater Spinnen (Arachnida: Araneae). Verhand. Naturwissen. Ver., Hamburg, 26: 241–253.

Peters, H.M. 1984. The spinning apparatus of Uloboridae in relation to the structure and construction of capture threads (Arachnida, Araneida). Zoomorph., 104:96–104.

Peters, H.M. 1986. Fine structure and function of capture threads. Pp. 187–202, *In* Ecophysiology of Spiders (W. Nentwig, ed.). Springer Verlag, New York.

Peters, H.M. 1992. On the spinning apparatus and structure of the capture threads of *Deinopis subrufus* (Araneae, Deinopidae). Zoomorph., 112: 27–37.

Pickard-Cambridge, O. 1902. Arachnida. Araneida. Pp. 305–316, *In* Biologia Centrali-Americana.

Purvis, A. & A. Rambaut. 1995. Comparative analysis by independent contrasts (CAIC): an Apple Macintosh application for analysing comparative data. Comp. Appl. Biosci., 11:247–251.

Simon, E. 1880. Etudes arachnologiques. 11e Mémoire. XVII. Arachnides recueillis aux environs de Pékin par M.V. Collin de Plancy. Ann. Soc. Entomol. France, 10:97–128.

Simon, E. 1909. Sur l'Araignèe Mosquero. Paris Academy of Sciences, 148:736–737.

Urquhart, A.T. 1892. Descriptions of new species of Araneae. Trans. New Zealand Inst., 25:165–190.

Walckenaer, C.A. 1837. Histoire naturelle des Insectes. Aptères. Tome I, Pp. 1–682. Paris.