Cybaeus jinsekiensis n. sp., a spider species with protogynous maturation and mating plugs (Araneae: Cybaeidae)

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Abstract — A new medium-sized species of the genus Cybaeus (Araneae: Cybaeidae) from the Chugoku district, western Honshu, Japan, is described under the name C. jinsekiensis. This species shows close affinities to C. kuramotoi in external and genital morphology, and hence belongs to the kuramotoi-group. A periodical field survey on the phenotype and other biological features of C. jinsekiensis revealed that: 1) This species requires two years for the maturation; 2) The adults emerge in mid September in the area studied, and females reach adulthood about one or two weeks earlier than males (protogyny); 3) They copulate in autumn and produce eggs in late spring of the next year; 4) Female genitalia often bear one or two mating plugs, each of which originates from distal segment of embolus broken off from male palp after copulation; 5) The numbers of matured eggs stored in ovaries varied from 9 to 29; 6) C. jinsekiensis is often found in a silken tube-like retreat bearing two terminal openings with a pair of signal threads on each of both ends. Implications of both protogyny, which is reported in spiders for the first time, and mating plug in this species are briefly discussed.

Key words — Cybaeidae, Cybaeus jinsekiensis n. sp., geographic distribution, the kuramotoi-group, mating plug, phenology, protogyny

Species of the genus Cybaeus (Araneae: Cybaeidae) often have limited distributional areas, and exhibit remarkable geographic differentiations in Japan. A series of the medium-sized species, which show close affinities to Cybaeus kuramotoi Yaginuma 1963 in external and genital morphology, is the most diverse group in western Honshu. Of these, I described five new species as members of the kuramotoi-group (Ihara & Nojima 2004). However, their taxonomy is still far from complete, and there are many undescribed species which await formal description. To alleviate the poor taxonomical situation, I will describe here a new species of the group under the name of Cybaeus jinsekiensis.

Members of the kuramotoi-group, including the present new species, are commonly found in woodlands with moderate moisture. Adults are found from autumn to spring. They construct a silken tube-like retreat, which is characteristic of the genus Cybaeus in Japan. Collecting records revealed that the species of the group overwinters in both adults and juveniles, and two years are needed to mature after hatching (Ihara & Nojima 2004). These spiders seem to require a rather long period for the growth in spite of their small bodies (5−7 mm long). However, only fragmental knowledge has been accumulated for the biology and ecology of the spiders of the genus Cybaeus (Komatsu 1961; Ihara 2003a). In order to confirm the life cycle of Cybaeus jinsekiensis, I carried out a periodical field survey in an area in Jinseki-kôgen-chô, Hiroshima Prefecture, western Honshu. I will also present some biological data obtained on the new species.

Materials and Methods

Materials

Specimens of the present new species have been collected in an area with 50−1100 m in altitude in the Chugoku district, western Honshu. Collecting data of specimens in description will be given by the following order: locality, altitude of locality if available, number of individuals (juv. = juvenile), date collected, and name of the collector (KN = Koichi Nojima, YI = Yoh Ihara). The type specimens designated in this paper are deposited in the National Science Museum (Natural History), Tokyo. Other specimens are in my personal collection.

Morphological examination

All the measurements were made for the specimens immersed in 80% ethanol under a stereo dissecting microscope with an ocular micrometer. Body length of the specimens preserved in ethanol is liable to vary due to variable degrees of relaxation of the joint connecting cephalothorax and abdomen. Accordingly, I used carapace length as indicator of body size.

Female genitalia removed from the abdomen were cleared in hot 10% KOH and 3% H2O2 according to the
method described in Komatsu & Yaginuma (1968) to observe internal sclerotized structure.

Recognition of species
For closely related forms in the kuramotoi-group, I adopted partial distributional overlap of the two neighboring forms without any signs of hybridization as evidence for two distinct species which are reproductively isolated one another. On the other hand, two completely allopatric forms were considered as two independent species when morphological gap between the two are larger than the gap exhibited by the two forms with partial sympathy. Using these two criteria for the species recognition, I recognized a new species which belongs to the kuramotoi-group in the area studied. This species has a sufficient morphological gap when compared to the other species within the group.

Field survey
A first periodical field survey was carried out from August to October in 2003 and April 2004 in an area with elevations of 300–500 m in Jinseki-kōgen-chô, Hiroshima Prefecture. I also collected specimens periodically from June to September in 2005 in the same area. To pursue adult emergence, specimens were collected three times (15–16, 23 and 29) in September 2005. On the other hand, as the spider matures in autumn, it is likely that drop of the air temperature may be responsible for the onset of the final molting. Therefore, altitudinal variation in adult emergence was also surveyed in 14–16 September 2005. The annual mean temperature of Jinseki-kōgen-chô is 11.0°C, and normal monthly mean temperature of August, September and October are 22.9°C, 18.7°C, and 12.2°C, respectively (the meteorological data at the Yuki Observation Site at elevation of 510 m altitudes near the study area, by Japan Meteorological Agency). The mean temperatures of September in 2003 and 2005 were 19.9°C and 20.6°C respectively, and they were higher than the normal year.

Description

Cybaeus jinsekiensis n. sp.

Japanese name: Jinseki-namihiagumo

(Figs. 1A–D, 2, 3A–D, 4A–B, 10, 11A–D)

Diagnosis. Due to the similarity in external morphology, diagnosis of each species relies primarily upon genital morphology of both sexes within the kuramotoi-group. This species can be distinguished from other members of the group by the following points: the angular patellar apophysis and arc shaped large apical element of embolus in male palp, the circular opening of epigynum in female.

Description. Male (holotype). Measurements (in mm). Body length 5.50; carapace length 2.91, width 1.94, head region width 1.12; abdomen length 2.63, width 1.90; sternum length 1.35, width 1.22; labium length 0.31, width 0.40. Length of legs (femur/ patella/ tibia/ metatarsus/ tarsus; total): Leg I: 2.30/ 0.88/ 2.07/ 2.00/ 1.39; 8.64. Leg II: 2.22/ 0.85/ 1.88/ 1.91/ 1.45; 8.31. Leg III: 2.00/ 0.81/ 1.47/ 1.81/ 1.05; 7.14. Leg IV: 2.44/ 0.80/ 2.12/ 2.52/ 1.32; 9.20.

Head region narrow, ratio of width to thoracic region 0.58 (Fig. 1A). Thoracic region as high as head region. Anterior eye row slightly procured as seen from front, posterior eye row slightly recurved as seen from above. Diameter of eyes: anterior median eyes the smallest, about half to other eyes. Ocular area twice as wide as long. Clypeus shorter than length of median ocular area, 0.84 in ratio. Chelicera geniculate in front, promargin of fung furrow with 3 teeth, retromargin with 4 (or 3) teeth and 4 (or 5) denticles, and basally with lateral condyle. Length of legs: 4 > 1 > 2 > 3. Tibia I with 2–2–2–2 ventral spines and 2 prolateral spines and 1 (left) or 2 (right) retrolateral spines; metatarsus I with 2–2–2 ventral spines, 2 prolateral spines and 1 retrolateral spine; tibia II with 2–2–1 (retromargin)-2 ventral spines and 3 prolateral spines and 1 retrolateral spine; metatarsus II 2–2–3 ventral spines, 4 prolateral spines and 1 retrolateral spine.

Palp (Figs. 2, 3A–D). Relatively thick and short in proportion. Cymbium relatively short, broad in prolateral side. Tibia slightly longer than patella. Patella retrolaterally with a hooked apophysis furnished with conical teeth. Genital bulb circle, and relatively large. Conductor with a large

**Fig. 1.** Shape of carapace and coloration pattern of abdomen of Cybaeus jinsekiensis n. sp.: A, C male (holotype); B, D female (paratype) —— A-B carapace, dorsal view; C-D abdomen, dorsal view. (Scale: 1.0 mm).
apical element of embolus.

Coloration. Carapace bright brown with reticulate brownish black markings on lateral sides of the head and radical bands on the thorax. Chelicerae, maxillae, labium and sternum yellowish brown; chelicerae darker than the others. Legs yellowish brown with dark grayish annulations. Dorsum of abdomen olive black with dull yellow chevron pattern as shown Fig. 1C.

Female (one of paratypes). Measurements (in mm). Body length 6.00; carapace length 2.83, width 1.92, head region width 1.29; abdomen length 3.25, width 2.45. Length of legs (femur/ patella/ tibia/ metatarsus/ tarsus; total) as follows. Leg I: 2.05/ 0.85/ 1.78/ 1.60/ 1.06; 7.34. Leg II: 1.87/ 0.81/ 1.57/ 1.50/ 1.00; 6.75. Leg III: 1.74/ 0.75/ 1.24/ 1.50/ 0.87; 6.10. Leg IV: 2.14/ 0.80/ 1.83/ 2.20/ 1.06; 8.03. Tibia I with 2/2–2/0 ventral spines and 1 (left) or 2 (right) prolateral spines; metatarsus I with 2/2–2 ventral spines, 2 prolateral spines and 1 retrolateral spine; tibia II with 2/2–1(retromargin)-2 ventral spines and 3 prolateral spines; metatarsus II 2/2–3 ventral spines, 4 prolateral spines and 1 retrolateral spine.

Similar to male in coloration. Carapace longer than male. Head region large, ratio of width to thoracic region 0.67 (cf. Fig. 1B with 1A). Abdomen larger and more rounded (cf. Fig. 1D with 1C), legs shorter than those of male.

Genitalia (Fig. 4A–B). Epigynum simple, with a large and circular opening; posteriorly with two copulatory pores. End of spermatheca extended and bent, connecting ducts long.

Specimens examined. Type series. Tantō, Yuki, Jinsekikōgen-chō, Jinsekigun, Hiroshima Prefecture, Japan: holotype (♂) and paratype (1♀), 17-X-2003, Yoh Ihara leg.


Phenology and Biology

Phenology

Figure 6 shows seasonal change of the number of individuals collected through the periodic survey in 2003 in an area with elevations of 300–500 m in Jinseki-kōgen-chō. Specimens collected in mid September consisted of both adults and penultimate juveniles, while samples collected in August comprised of penultimate juveniles alone in both sexes. In the sample collected in mid September, the majority of the females were adults, whereas most of the males were still penultimate. On the other hand, all the specimens collected in October had already become adults. The earliest data of adult emergence in the present survey was of female from an area in 520 m in altitude in Jinseki-kōgen-chō, and it was 9 September 2004. Discrepancy in adult emergence between females and males can be seen also in the samples collected during September in 2005 from areas ranging from 470 to 580 m in altitude (Fig. 7), where females reached adulthood about a week earlier than males. That is, in 15–16 September, most of the females of the adults, while all the males were still penultimate juveniles. Additional samples collected on 23 and 29 September showed that the time lag in the adult emergence between females and males is about a week.

Figure 8 shows altitudinal variation in adult emergence surveyed in 14–16 September 2005. In those samples, percentages of adults increased with altitude of the localities. It is considered that the earlier temperature drop in localities with higher altitudes leads the penultimate juveniles of the species in those areas to molt for adulthood earlier. The time lag in final molting between the two localities that differ about 500 m in altitude seems to be equivalent to about two weeks (compare Figs. 8 and 7). The data depicted in Fig. 8 also showed that the ratios of adults to juveniles were higher in females than males again.

Life cycle

Figure 9 shows seasonal transition of the body size of individuals collected through the periodic survey in 2003–2005 in areas with elevations of 300–500 m in Jinseki-

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kōgen-chō. Specimens collected during the survey can be classified into six classes (five juvenile and an adult stages, see Fig. 9–10 I–VI) based on the carapace length. The smallest juveniles in the field (it is possible that they are not of the first instar) were found in July and August. They were collected with the other class of juveniles that differ enormously in size, suggesting an overlap of the two different generations. They probably overwinter as juveniles and adults, respectively. The body size of juveniles showed no considerable increments from October to April. Individuals overwintered as juveniles molt to adults in the

**Fig. 5.** Geographic distributions of *Cybaeus jinsekiensis* n. sp. and other species of the *kuramotoi*-group in the Chugoku district, western Honshu, Japan.

**Fig. 6.** Seasonal change of relative frequencies of penultimate juveniles and adults of *Cybaeus jinsekiensis* n. sp. in Jinseki-kōgen-chō in 2003.

**Fig. 7.** Sexual differentiation in adult emergence of *Cybaeus jinsekiensis* n. sp. in Jinseki-kōgen-chō and adjacent areas around ca. 500 m in altitude in 2005. Ōhagi, Nara, Jōchi and Takekawauchi are located in Jinseki-kōgen-cho, and Kusshiro in Shōbara-sh. Tōjō-cho is adjacent to Jinseki-kōgen-chō.
next September. It is considered that oviposition takes place in spring by overwintering adult female. Thus, it explicitly showed that two years are needed for the maturation of *Cybaeus jinsekiensis*.

**Sex ratio**

A total of 524 individuals of adults comprising 213 males and 311 females, were collected in this study. Female-biased sex ratio was observed over adult period (deviation from 1:1 is significant: $\chi^2 = 18.3$, $P < 0.001$). However, actual sex ratio show approximately 1:1 when it was determined by recruited adults and penultimate juveniles in the periodical surveys. In the surveys, male to female ratios were 35:33 in August and September in 2003 (see Fig. 6), and 48:48 in 14–16 September 2005 (see Fig. 8).

**Mating plug**

Each of the females often had one or two mating plugs on her epigynal opening. The mating plug is an apical element of embolus of male palp (Fig. 11, cf. A with B). After the copulation, it is detached from male palp and is inlaid to acrevice of epigynal plate to block a copulatory pore (Fig. 11, cf. C with D). The apical element of embolus of left palp is attached to epigynal opening on female’s left side of abdomen. Females bearing the mating plugs usually emerge after mid October, and increase in number in later season of the adult stage. For example, in an area with 460–570 m in altitude in Jinsenki-kōgen-cho, 66% of females (40 of 61 females) bore the mating plugs on their epigyna on 5 November 2005, though in all of 25 females collected on...
17–18 October 2005 the mating plugs were absent. Of the 40 females bearing plugs on 5 November, 21 females had a plug on her left side (on the right side of the ventral view of the abdomen) alone and 11 females right side alone, whereas 8 females had two plugs on both lateral sides. Similarly, males lacking the apical element of embolus of its palp increase in number in late autumn. Therefore, it is concluded that _Cybaeus jinsekiensis_ copulate in late autumn.

**Eggs**

Egg sac of _Cybaeus jinsekiensis_ has not been found in the field. However, it is likely that females of this species produce egg sacs in late spring, because females collected from March to May bear mature eggs in their ovaries (in females collected in January, ovaries were still immature, though they accumulated some yolk). The number of matured eggs stored in ovaries varied from 9 to 29 (n = 6) and was correlated with the body size of females (Fig. 12).

**Habitat and Retreat**

The spiders of _Cybaeus jinsekiensis_ were usually found inside and around the silken tube-like retreats in the earlier season of the adult stage. The retreats were found under logs or rocks on humid forest floor and on the soiled wall of the overhang rocks protruded on the roadside slope along the forest road. However, they seemed to wander in leaf litter layer on forest floor in later season, because retreats without resident spiders increased in the season.

Each retreat had two terminal openings (“V letter-shaped type” in Komatsu 1961) with a pair of signal threads on each of the both ends (Fig. 13). It was coated with particles of detritus of plants or sand grains. Juveniles, at least in the second and later stages (see Fig. 9), also build a retreat of the same structure as that of adults, though it is smaller than those constructed by adults. Komatsu (1961, 1968) classified the retreats of the Japanese species of the genus _Cybaeus_ into four types: 1) V letter-shaped with two entrances; 2) V letter-shaped with three entrances; 3) Y letter-shaped with three entrances; and 4) hexagonal in shape with three entrances. Most of the Japanese species of _Cybaeus_ build a retreat of V letter-shaped with two entrances (C. _s Sanctus_, Komatsu 1961; _miyosii_-group, Ibara 2003b; _kuramotoi_-group, Ibara & Nojima 2004). In addition, I have observed this type of retreat in the following species: _C. ashikitaensis_ (Komatsu 1968); _C. jaanaensis_ Komatsu 1968; _C. kirigaminensis_ Komatsu 1963; _C. kiuchi Komatsu 1965; C. _mellotei_ (Simon 1886); _C. shinkaii_ (Komatsu 1970); _C. totoriensis_ Ibara 1994. A retreat similar to this
has been known in *Cicurina bryantae* Exline 1936 (Dictynidae) from U. S. A. (Bennet 1985). However, retreat of *Cicurina bryantae* lacks signal threads.

**Discussion**

Among several biological features of *Cybaeus jinsekiensis* revealed in the present study, most notable one is protogynous maturation of the species. Protogyny, which refers to the trend that the females emerge somewhat before the males, probably has not been reported in spiders, though it seems to be dominant in harvestmen of the superfamily Phalangioidea (Tsurusaki 2003). In spiders, especially in “entelegyne” which usually bear spermathecae of the conduit type, it is expected that the first male sperm priority and occurrence of mating plugs are widespread (Austad 1984). Several empirical studies that have so far been accumulated (Jackson 1980; Martyniuk & Jaenike 1982; Watson 1991; Masumoto 1991, 1993, 1994; Uhl & Vollrath 1998) seem to generally support the Austad’s prediction, though some studies showed commitment of other factors such as male body size (Watson 1990) or duration of copulation in the sperm competition (Schneider 1997; Schneider & Elgar 1991; Schneider et al. 2000, 2005; Uhl 1998; Drengsgaard & Toft 1999). On the other hand, occurrence of mating plugs is often associated with precopulatory mate guarding and protandry in insects (Thornhill & Alcock 1983). The reason for the association is easy to understand because mating plug is primarily a device to secure first male sperm precedence. Spiders are also outstanding for its propensity to evolve precopulatory mate guarding and protandrous maturation (Riley 1983; Toft 1989; Watson, 1990; Miyashita 1994; Prenter et al. 1994; Holdsworth & Morse 2000) and according to the Riley’s estimation, precopulatory mate guarding has been evolved at least four times in Araneae.

In this context, occurrence of both the protogyny and mating plug in *Cybaeus jinsekiensis* is extraordinary and puzzling. One possibility that might explain the association is that males which mate with fresh adult females bearing less sclerotized genitalia may be maladaptive because such less sclerotized female genitalia are flexible and mating plugs attached to the female genitalia would be liable to drop off. In *C. jinsekiensis*, copulation takes place in autumn and oviposition in spring of the next year, and hence time lag between the two events is rather long. On the other hand, males that lost their intromittent organs to make mating plugs loose further chances of copulation with other females. Thus they should not be indifferent to optimal timing of copulation.

If this reasoning is correct, we would expect that association between mating plug and precopulatory mate guarding or protandry can be found in species such as *Frontinella pyramitela* (Linyphiidae, Austad 1984), *Phidippus johnsoni* (Salticidae, Jackson 1980), where time lag between copulation and oviposition is rather short and the other type of association between mating plug and protogyny in species, such as *C. jinsekiensis*, with longer time lag due to adult overwintering which intervenes between copulation and oviposition. In the latter case, postcopulatory mate guarding might also be expected, though it is not yet confirmed, in the species.

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