

Structure of Female Genitalia in the Korean Wood-Feeding Cockroach, *Cryptocercus kyebangensis*

Yung Chul Park* and Jae Chun Choe

Behavioral Ecology Laboratory, School of Biological Sciences, Seoul National University, Seoul 151-742, Korea

Key Words:

Cryptocercus kyebangensis
Female genitalia
Ovipositor
Spermatheca
Wood-feeding cockroach

Female genital morphology of *Cryptocercus kyebangensis* is described, focusing on the structural features of ovipositors at each developmental stage. Ovipositor valves were the first genital structures to appear in female nymphs. The caudal margin of the 9th sternum was nearly straight in the 1st instars of both sexes, with a slight median notch, but was deeply concave in the female nymphs of the 2nd instar. The 1st valve of ovipositor budded off from membranous fold between the 8th and 9th sternum and a pair of the 2nd valve came from the 9th sternum. Separation of styli occurred in the female nymphs of the 8th or 9th instar. Some parts of the female genitalia were sclerotized after the final molt. Spermathecae were the fork type and leaned a little to the right in the middle line of the genital chamber from ventro-posterior view. The trace sealed with sclerite materials existed on the spermathecal opening. The genital segments (segments VIII, IX, and X) were concealed completely by the 7th tergum and sternum posteriorly expanded. Terminal lobes which exist in the Blattidae were not observed in *C. kyebangensis*. Instead, the apicolateral emargination existed on the terminal abdominal segment of *C. kyebangensis* females.

Woodroaches of the genus *Cryptocercus* live in montane forest areas in temperate regions. There are currently nine species recognized in *Cryptocercus*. Five of them occur in the Nearctic (Cleveland et al., 1934; Bey-Bienko, 1950; Nalepa et al., 1997; Burnside et al., 1999). The other four species of *Cryptocercus* occur in the Palearctic (Bey-Bienko, 1950; Grandcolas, 2000; Nalepa et al., 2001; Grandcolas et al., 2001). Despite the extensive studies on the phylogenetic position of *Cryptocercus* (Cleveland et al., 1934; McKittrick, 1964, 1965; Grandcolas and Deleporte, 1992; Bandi et al., 1995; Grandcolas, 1994, 1997, 1999; Gäde et al., 1997; Nalepa and Bandi, 1999; Maekawa and Matsu-moto, 2000), however, the phylogenetic relationship among *Cryptocercus* species has still been controversial. Kirby (1904) and Bey-Bienko (1950) placed the genus *Cryptocercus* in the Panesthiidae. Since some Panesthiines harbor symbiotic bacteria and amoebae rather than flagellates to digest their woody diet, the inclusion of *Cryptocercus* in this group, however, has been questioned by Cleveland et al. (1934) and Marks and Lawson (1962). *Cryptocercus* harbors similar gut symbionts to those of the lower termite, especially *Mastotermes darwiniensis* (Bobyleva, 1975), and female genitalia and proventriculus characteristics of *Cryptocercus* are also similar to those of lower termite

(Cleveland et al., 1934; McKittrick, 1964, 1965). Based on these similarities between the lower termite and *Cryptocercus*, Cleveland et al. (1934) and McKittrick (1964) suggested that *Cryptocercus* is more closely related to termites than to cockroaches. A different view on the phylogenetic position of *Cryptocercus* was proposed recently by Grandcolas (1994, 1997, 1999). He assigned *Cryptocercus* to the subfamily Polyphaginae on the basis of newly derived character states including the morphology of genital sclerites.

Despite the controversies on the phylogenetic position of *Cryptocercus*, most of what is known about the morphology of *Cryptocercus* have been primarily based on North American species, especially on *Cryptocercus punctulatus*. In *Cryptocercus*, morphological characteristics of the female genitalia including spermatheca were often used to determine its phylogenetic position (McKittrick, 1964, 1965; Grandcolas, 1994). Especially, McKittrick (1964) used the female genitalia to reveal the phylogenetic relationship between *Cryptocercus* and termites. However, the study of McKittrick (1964) was based on *C. cryptocercus*, only a North American *Cryptocercus*.

The objective of the present study is to provide comparative information on East Asian *Cryptocercus* using Korean *Cryptocercus*. In the present study, we describe morphological characteristics of its female genitalia, especially focusing on its ovipositor features at each developmental stage, in *Cryptocercus kyebangensis* recently described by Grandcolas et al. (2001).

* To whom correspondence should be addressed.
Tel: +82-2-880-8157, Fax: +82-2-882-7195
E-mail: parkyungchul@hotmail.com

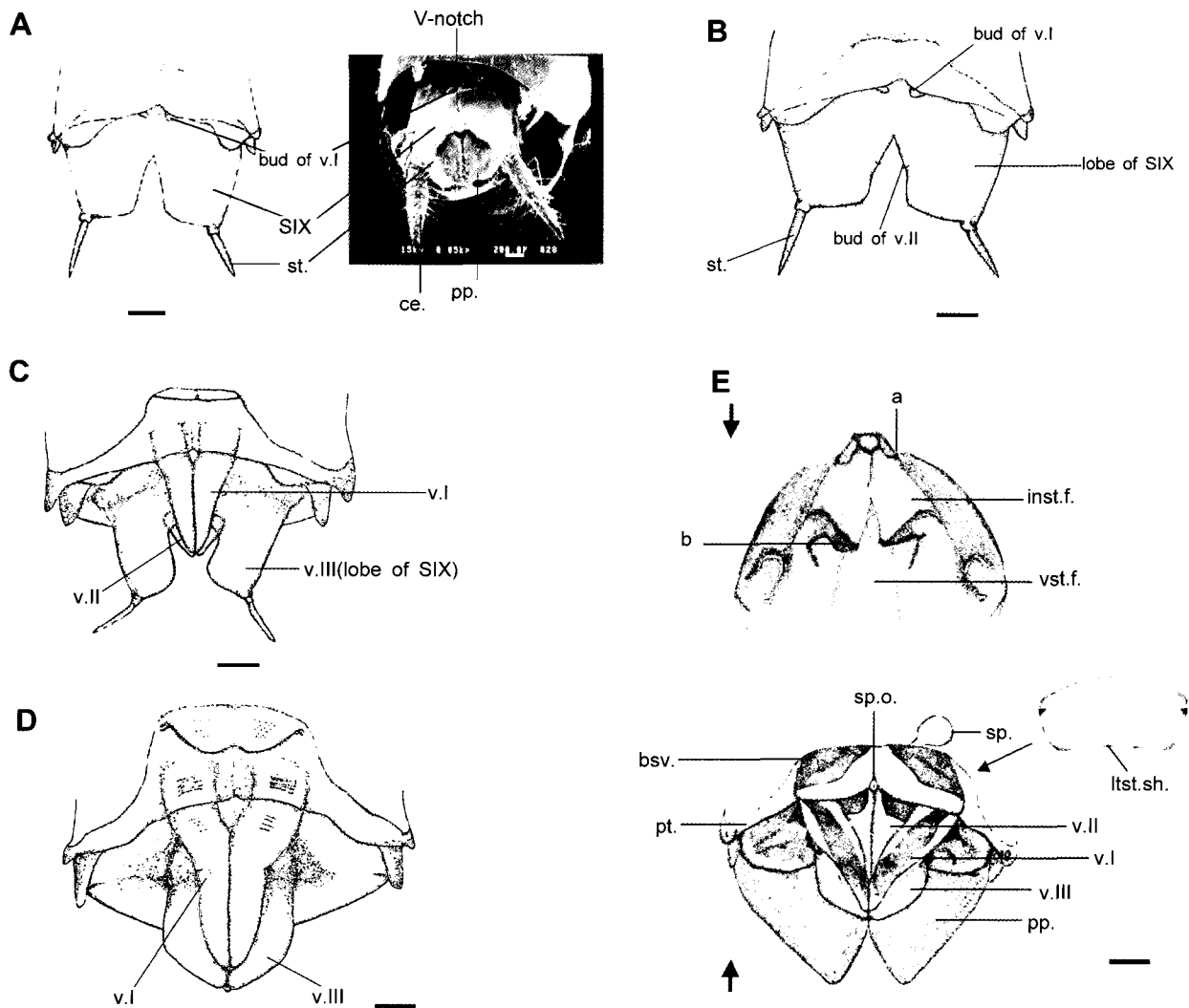


Fig. 1. Postero-ventral views of female genitalia in *C. kyebangensis*. A, fourth instar (head capsule width: 1.67 mm); B, fifth instar (head capsule width: 1.90 mm); C, seventh instar (head capsule width: 2.67 mm); D, last instar (head capsule width: 4.05 mm); E, adult female (head capsule width: 4.5 mm), apicolateral emargination (a), and a sclerotized-tip of intersternal folds (b). Scale bars = 0.13 mm (A), 0.15 mm (B), 0.3 mm (C), 0.34 mm (D), and 0.65 mm (E).

Material and Methods

Woodroaches of *Cryptocercus kyebangensis* were collected from rotting logs at Gyebang-san in 1997. Neonates were obtained by incubating oothecae at $25 \pm 2^\circ\text{C}$. Ages of nymphs were assigned using the criterion of Park and Choe (in prep.). The genital morphology was examined by removing the 7th sternum under a binocular microscope (Olympus Model SZ-STU1) with a graduated eyepiece and scanning electron microscope. For the SEM analyses, the structure was fixed for 12 h in 2.5% glutaraldehyde in 0.1 M phosphate buffer, washed in buffer, postfixed for 12 h in 2% osmium tetroxide in the same buffer, dehydrated in a graded series of ethanol solutions. Specimens were mounted on stubs, coated with gold

using an ion coater (Eiko Model IB-3), and examined with an SEM (Akashi Model SR 50A) operated at an accelerating voltage of 15 kV.

Abbreviations used in the description were taken from the modification of McKittrick (1964) as follows; bsv. = basivalvula, inst.f. = intersternal fold, ltst.sh. = laterosternal shelf, pp. = paraprocts, pt. = paratergites, SIX = sternum IX, sp. = spermatheca, sp.o. = spermathecal opening, st. = stylus, ce. = cercus, vst.f. = vestibular floor, v.I = first valve, v.II = second valve, and v.III = third valve.

Results and Discussion

Development of female reproductive organs

In the earliest instars, the 9th sternum of female

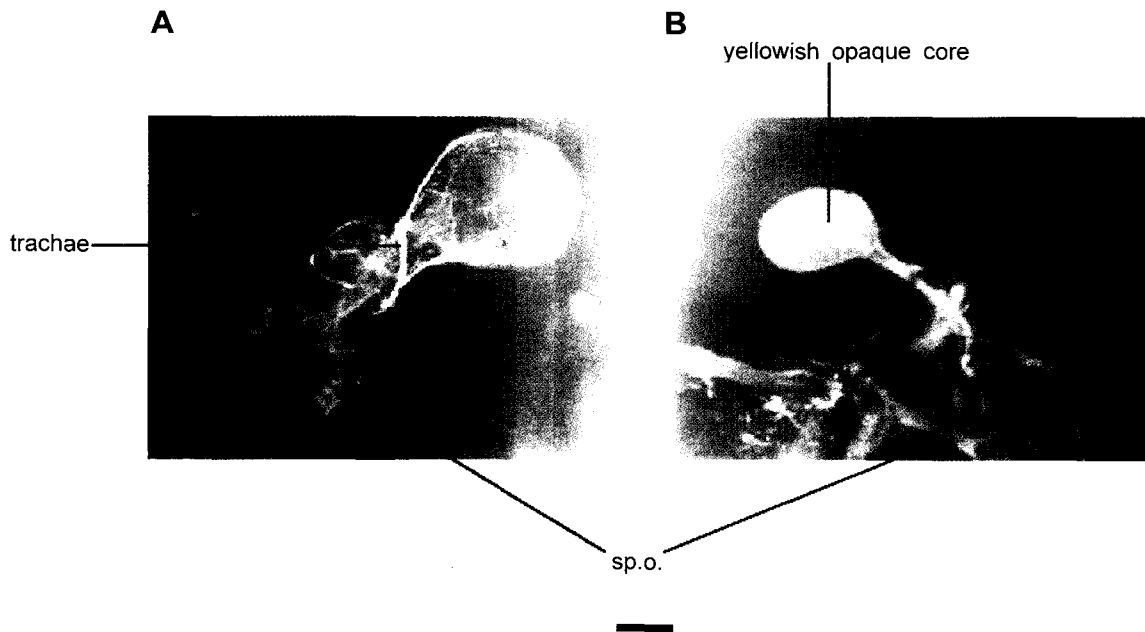


Fig. 2. Spermatheca and surrounding sclerites of *Cryptocercus kyebangensis*. Spermatheca covered by whitish material and trachea (A), and observed after the removal of the exterior materials (B). Scale bar = 0.3 mm.

nymphs bears styli laterally and the caudal margin of the sternum is nearly straight. From the 2nd instar, a slight median notch (V-notch) appears obviously in the caudal margin of the 9th sternum, and it broadens and deepens in the successive instars. The lateral sternal lobes support the styli and later differentiate into the 3rd pair of valves. The incipient 1st pair of valves becomes apparent in the middle of the posterior edge of the 8th sternum in the 4th instar (Fig. 1A). As the 1st valves enlarge, the posterolateral corners of the 8th sternum lengthen narrowly. The median notch deepens broadly in the successive instars, giving rising to anteriorly to a pair of small buds that later become the 2nd pair of valves (Fig. 1B). During the successive instars, the buds of the 1st valves expand and overlap with those of the 2nd valves (Fig. 1C). A pair of the 3rd valves migrate medially in the successive instars, with the progressive diminution of the lobes lateral to the 2nd valves until the last instar stage. One or both of the styli are lost at the 8th or 9th instar. Melanization traces are observed to the points on the lateral lobes from which the styli are detached. In the last instar, the 1st valves overlap with the 3rd valves, covering the 2nd valves (Fig. 1D).

Structures of female genitalia

The genital structures of *C. kyebangensis* females are shown in Fig. 1E. They are similar to those of *C. punctulatus* described by McKittrick (1964). The genital segments (VIII, IX, and X) of *C. kyebangensis* are concealed completely by the 7th sternum and tergum

expanded posteriorly. They are very small and lightly sclerotized. Terminal lobes which exist in the Blattidae were not observed in *C. kyebangensis*. Instead, the apicolateral emargination existed on the terminal abdominal segment of *C. kyebangensis* females. On the terminal sternum, the membranes of the vestibular floor (vst.f.) are expanded into a conspicuous pair of intersternal folds (inst.f.), with their tips sclerotized lightly. The bases of the 1st valves are posterior to the basivalvulae (bsv.). A pair of the 2nd valves (v.II) are concealed by the 1st (v.I) and 3rd valves (v.III). The basivalvulae are well developed in the roof of the genital chamber and lie adjacent to the spermathecal area. Basivalvulae and spermathecal opening (sp.o.) are covered with laterosternite shelf (ltst.sh.). Spermathecae (sp.) lean a little to the right from the middle line of the genital chamber. The trace sealed with sclerite materials exists on the spermathecal opening. In *C. kyebangensis*, spermathecae are the fork type as those of *C. punctulatus* (McKittrick, 1964) and is wrapped in whitish materials with tracheal ducts entangled (Fig. 2A). A pale yellowish opaque core is present inside the spermathecae observed after the removal of exterior materials (Fig. 2B).

Taxonomic significance

This study includes the first description on female genitalia of the Korean wood-feeding cockroach, *C. kyebangensis*. The ovipositor valves were the first genital structures to appear in female nymphs. In *Cryptocercus kyebangensis*, the differentiation of

ovipositor showed a similar pattern to that of *Eurycotis floridana* (Walker) and *Leucophaea maderae* (Fabricius) described by McKittrick (1964). Since the positions and configurations of the 1st and 2nd pairs of valves are very similar during development, it is possible that these structures are homologous as suggested by McKittrick (1964). Unlike the cockroaches of the Blattidae, the terminal lobes did not exist in *C. kyebangensis*. It is possible that the apicolateral emargination existed on the terminal sternum (Fig. 5a) was the precursor to the completely divided terminal lobes of cockroaches belonging to the Blattidae, as suggested by Appel (1989). According to Nalepa and Bell (1997), cockroaches show the entire range of reproductive modes including oviparous, ovoviviparous and viviparous. In cockroaches, especially, the reproductive mode of oviparous is observed in nearly all families except Blaberidae and some Blattellidae (Nalepa and Bell, 1997). The oviparous is also shown in the woodroaches of the genus *Cryptocercus* (Nalepa, 1988; Park et al., in prep.). The genital structures of *Cryptocercus* females need to be studied further in relation to the evolution of their reproductive mode.

Acknowledgements

We are grateful to Dr. Hyun-Woo Lee and Prof. Chong-Wook Park in Plant Taxonomic Lab., Seoul National University for their assistance and help for the SEM photos. We wish to thank Yeon-Suk Lee for her assistance for the drawings. This work was supported by grants from the BK21 Research Fellowship from the Korean Ministry of Education and Human Resources Development and Korea Science and Engineering Foundation (KOSEF 985-0500-007-2).

References

- Appel AG (1989) Rapid and non-destructive gender determination of nymphal and adult *Cryptocercus punctulatus* Scudder (Dictyoptera: Cryptocercidae). *Proc Entomol Soc Wash* 91: 286-287.
- Bandi C, Sironi M, Damiani G, Magrassi L, Nalepa C, Laudani U, and Sacchi L (1995) The establishment of intracellular symbiosis in an ancestor of cockroaches and termites. *Proc R Soc Lond B* 259: 293-299.
- Bey-Bienko GY (1950) Fauna of the USSR. Insects. Blattodea. Institute of Zoology, Academy of Sciences, URSS, Moscow. pp 1-342 [in Russian].
- Bobyleva NN (1975) Morphology and evolution of intestinal parasitic flagellates of the far-eastern roach *Cryptocercus relictus*. *Acta Protozool* 14: 109-169.
- Burnside CA, Smith PT, and Kambhampati S (1999) Three new species of the woodroach, *Cryptocercus* (Blattodea: Cryptocercidae), from the eastern United States. *J Kans Entomol Soc* 72: 361-378.
- Cleveland LR, Hall SR, Sanders EP, and Collier J (1934) The wood-feeding roach *Cryptocercus*, its protozoa, and the symbiosis between protozoa and roach. *Mem Am Acad Arts Sci* 17: 85-342.
- Gäde G, Grandcolas P, and Kellner R (1997) Structural data on hypertrehalosaemic neuropeptides from *Cryptocercus punctulatus* and *Therea petiveriana*: how do they fit into the phylogeny of cockroaches? *Proc R Soc London B* 264: 763-768.
- Grandcolas P (1994) Phylogenetic systematics of the subfamily Polyphaginae, with the assignment of *Cryptocercus* Scudder, 1862 to this taxon (Blattaria, Blaberoidea, Polyphagidae). *Syst Entomol* 19: 145-158.
- Grandcolas P (1997) What did the ancestors of the woodroach *Cryptocercus* look like? A phylogenetic study of the origin of subsociality in the subfamily Polyphaginae (Dictyoptera, Blattaria). *Mem Mus Natn Hist Nat* 173: 231-252.
- Grandcolas P (1999) Systematics, endosymbiosis, and biogeography of *Cryptocercus clevelandi* and *C. punctulatus* (Blattaria: Polyphagidae) from North America: a phylogenetic perspective. *Ann Entomol Soc Am* 92: 285-291.
- Grandcolas P (2000) *Cryptocercus matilei* n. sp., Sichuan de Chine [Dictyoptera, Blattaria, Polyphaginae]. *Rev Entomol* 22: 223-226.
- Grandcolas P and Deleporte P (1992) La position systématique de *Cryptocercus* Scudder, 1862 et ses implications évolutives. *Comp Rend Acad Sci Paris* 315: 317-322.
- Grandcolas P, Park YC, Choe JC, Piulachs MD, Belles X, D'Haese C, Farine JP, and Brossut R (2001) What does reveal *Cryptocercus kyebangensis*, n. sp. from South Korea about *Cryptocercus* evolution? A study in morphology, molecular phylogeny and chemistry of tergal glands (Dictyoptera, Blattaria, Polyphagidae). *Proc Acad Nat Sci Phila* 151: 61-79.
- Kirby WF (1904) A Synonymic Catalogue of the Orthoptera. British Museum of Natural History, London. Vol 1. pp 1-501.
- Maekawa K and Matsumoto T (2000) Molecular phylogeny of cockroaches (Blattaria) based on mitochondrial COII gene sequences. *Syst Entomol* 25: 511-519.
- Marks E and Lawson FA (1962) A comparative study of the dictyopteran ovipositor. *J Morphol* 111: 139-172.
- McKittrick FA (1964) Evolutionary studies of cockroaches. Cornell University Agricultural Experimental Station, Ithaca. pp 1-389.
- McKittrick FA (1965) A contribution to the understanding of cockroach-termite affinities. *Ann Entomol Soc Am* 58: 18-22.
- Nalepa CA (1988) Reproduction in the woodroach *Cryptocercus punctulatus* Scudder (Dictyoptera: Cryptocercidae): mating, oviposition, and hatch. *Ann Entomol Soc Am* 81: 637-641.
- Nalepa CA and Bell WJ (1997) Postovulation parental investment and parental care in cockroaches. In: Choe JC and Crespi BJ (eds), *The Evolution of Social Behavior in Insects and Arachnids*, Cambridge University Press, Cambridge, pp 26-51.
- Nalepa CA, Byers CW, Bandi C, and Sironi M (1997) Description of *Cryptocercus clevelandi* (Dictyoptera: Cryptocercidae) from the northwestern United States: molecular analysis of bacterial symbionts in its fat body, and notes on biology, distribution, and biogeography. *Ann Entomol Soc Am* 90: 416-424.
- Nalepa CA and Bandi C (1999) Phylogenetic status, distribution, and biogeography of *Cryptocercus* (Dictyoptera: Cryptocercidae). *Ann Entomol Soc Am* 92: 292-302.
- Nalepa CA, Li L, Wen-Hua L, and Lazell J (2001) Rediscovery of the wood-eating cockroach *Cryptocercus primarius* (Dictyoptera: Cryptocercidae) in China, with notes on ecology and distribution. *Acta Zootaxonom Sinica* 26: 184-190.

[Received November 22, 2001; accepted December 5, 2001]