

Ecology of a wood-feeding cockroach, *Salganea taiwanensis ryukyuanus*, focusing on mating behavior and interspecific interactions

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**Ecology of a wood-feeding cockroach, *Salganea taiwanensis ryukyuanus*,
focusing on mating behavior and interspecific interactions**

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General introduction

To date, only one species has been known that female and male within a mating pair eat the mate one another: in the mating pairs of the wood-feeding cockroach, *Salganea taiwanensis*, females and males each eat their mate's wings. This wood-feeding cockroach is subsocial species and the parents care for their offspring biparentally (Maekawa et al. 2008). New adults have long wings, but over 99% of parents observed in the field showed only small parts remaining around the base of their wings. Nymphs of this species are raised in a rotten log by their parents until they emerge into new adults. New adults fly away to search for a mate. After mating, a mating pair makes a gallery in a rotten log and is likely to be strict monogamy (Bell et al. 2007). Both female and male of *S. taiwanensis* eat the wings of mate which should be the cooperator for taking care of the offspring.

This mutual wing-eating behavior has something in common at a glance with two behaviors that have been studied: sexual cannibalism and nuptial feeding. In the sexual cannibalism, only one sex in a mating pair consumes one's mate, which occurs in only a limited number of species in arthropods (Prenter et al. 2006). The cannibalistic sex, either of males and females, kills the mate and obtains nutrition for reproduction. In mutual wing-eating, both female and male eat only wings which is non-fleshy and may have low or no nutritional value. Furthermore, the female and male mate with the individual whose wings were eaten by them in *S. taiwanensis*. Therefore, *S. taiwanensis* cannot obtain benefits in the same way as the sexual cannibalism.

In the other behavior, the nuptial feeding, males usually feed females material other than sperm. The males that feed females obtain a chance of increasing the number of offspring (Lewis et al. 2014). The reversed type, where females feed males, is reported in only one example. In Zeus bugs, females feed males with their gland secretion to reduce kleptoparasitism by males (Arnqvist et al. 2006). The wings of *S. taiwanensis* can be interpreted as nuptial food gifts in mutual wing-eating. Male kleptoparasitism, however, has not been reported in *S. taiwanensis*. Moreover, the pairs of *S. taiwanensis* have sufficient food readily available, the rotten wood, around their galleries. Therefore, the reported reason why females feed males cannot explain mutual wing-eating. The remaining question is why females also give males wings.

For either behavior, sexual cannibalism or nuptial feeding, mutual wing-eating is the first example in which both sexes consume and are eaten. Thus, the significance of mutual wing-eating is probably different from unilateral sexual cannibalism and nuptial feeding. Mutual wing-eating may shed new light on the relationship between males and females in mating pairs.

In this study, as a step to clarify the significance of mutual wing-eating behavior, I conducted a field survey and laboratory experiments. Because there was little knowledge on mutual wing-eating behavior, I recorded the behavior of mating pairs to understand the adaptive effect of the mutual wing-eating behavior. To observe this mating behavior, the rearing and observation system of *S. taiwanensis* was needed. However, no method had been established. I established the rearing system that enables to observe the behavior of wood-feeding cockroaches. *Salganea taiwanensis* has not been studied enough in the field, and little is known about its ecology except the biparental care in rotten logs. Interspecific relationships of *S. taiwanensis* are important for understanding the benefits and costs of its mating behavior. I conducted a field survey and discovered an associate insect. This also helped to understand their ecological roles.

References

- Arnqvist G, Jones TM, Elgar MA. 2006. Sex-role reversed nuptial feeding reduces male kleptoparasitism of females in Zeus bugs (Heteroptera; Veliidae). *Biology Letters* 2: 491–493.
- Bell WJ, Roth LM, Nalepa CA. 2007. *Cockroaches: ecology, behavior, and natural history*. JHU Press, Baltimore.
- Lewis SM, Vahed K, Koene JM, Engqvist L, Bussière LF, Perry JC, Gwynne D, Lehmann GUC. 2014. Emerging issues in the evolution of animal nuptial gifts. *Biology Letters* 10: 20140336.
- Maekawa K, Matsumoto T, Nalepa CA. 2008. Social biology of the wood-feeding cockroach genus *Salganea* (Dictyoptera, Blaberidae, Panesthiinae): Did ovoviviparity prevent the evolution of eusociality in the lineage? *Insectes Sociaux* 55: 107–114.
- Prenter J, MacNeil C, Elwood RW. 2006. Sexual cannibalism and mate choice. *Animal Behaviour* 71: 481–490.

Chapter 1

Mutual eating between female and male within mating pairs in a wood-feeding cockroach, *Salganea taiwanensis ryukyuanus*

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Abstract

The behavior of eating their mates or a part of their mate's body has been studied in sexual cannibalism or nuptial feeding. In these behaviors, only one sex eats the other unilaterally. Within mating pairs of a wood-feeding cockroach (*Salganea taiwanensis*), males and females eat the mate's wings each other, which is the first "mutual" case in these behaviors. Because the evolution of sexual cannibalism and nuptial feeding has been explained based on unilaterality, this mutual eating should have a new significance of reproduction. I described this behavior quantitatively and suggest a new hypothesis based on true monogamy, the mating system of *S. taiwanensis*, copulating with only one mate throughout life. Under true monogamy, the fitness of the mate is the same as own fitness and the pairs are free from sexual conflict. If the wing eating increases the ability of the mate to raise the offspring, this behavior is adaptive for the eater as well as its mate.

Introduction

A variety of insects and spiders eat their mates or what their mate provides. These feeding behaviors have long been studied in two contexts: sexual cannibalism and nuptial feeding. In sexual cannibalism, females usually consume males while the reverse type is rare. In nuptial feeding, males usually feed females material other than sperm while the reverse type is reported in only one example. To date, observation of these behaviors has identified only unilateral eating one sex (Prenter et al. 2006; Lewis et al. 2014). The only exception is within mating pairs of the wood-feeding cockroach, *Salganea taiwanensis*, males and females each eat their mate's wings. In this species, new adults fly away from their natal colony, which is in a rotten log, to search for a mate. After mating, a mating pair makes a gallery in a rotten log, takes care of their offspring biparentally (Maekawa et al. 2008), and is likely to mate for life (Bell et al. 2007). New adults have long wings, but parents do not (Fig. 1). Over 99% (330/331 individuals) of parents observed in the field showed only small parts remaining around the base of their wings.

This mutual wing-eating behavior is sexual cannibalism or nuptial feeding. Sexual cannibalism is the behavior of consuming one's mate, which occurs in only a few species, such as spiders and mantises (Prenter et al. 2006). The cannibalistic sex, either of males and females, kills and consumes the mate and consequently, obtains benefits, which may include obtaining nutrition for reproduction. In the sexual cannibalism by males, males may have the benefit of avoiding mating with inferior females (Prenter et al. 2006). In mutual wing-eating, female and male eat only non-fleshy wings, which may have low or no nutritional value, and they mate with the individual whose wings were eaten by them. Therefore, *S. taiwanensis* cannot obtain benefits in the same way as reported cases of sexual cannibalism. In nuptial feeding, males usually feed females with food gifts and obtain benefits, which is to increase the number of offspring (Lewis et al. 2014). Another benefit suggested in Zeus bugs is that females feed males to reduce kleptoparasitism by males (Arnqvist et al. 2006). In *S. taiwanensis*, wings are nuptial food gifts in nuptial feeding. Male kleptoparasitism, however, has not been reported in *S. taiwanensis*, and the pairs of *S. taiwanensis* have sufficient food readily available, the rotten wood, around their galleries. Therefore, the remaining question is why females also give males wings.

In either case, cannibalism or feeding, mutual wing-eating is the first example in

which both sexes consume and are eaten. Thus, the significance of mutual wing-eating is probably different from unilateral sexual cannibalism and nuptial feeding. Mutual wing-eating may shed new light on the relationship between males and females in mating pairs.

Obata (1998) reported mutual wing-eating takes at least three days in his master thesis. He observed pairs only for the initial 3 hours of mating and checked the condition of the wings visually only once a day. Since this thesis is the only report on mutual wing-eating, no detailed observations on behavior and relationships between mates are available. Mutual wing-eating behavior is reported here quantitatively to describe the entire sequence of mutual wing-eating and discuss how this behavior evolves.

Materials and Methods

Field colonies of *S. taiwanensis ryukyuanus*, including last instar nymphs, were collected from Yona Field, Okinawa, Japan (26°45'18"N, 128°13'09"E). New adults that emerged from last instar nymphs were used for observation.

I created 24 new adult pairs using males and females from different colonies. Pairs were recorded with video cameras for three days. The end of mutual wing-eating was defined as the time when wings of both sexes became as short as those of wild parents. Mutual wing-eating terminated in 12 pairs. Since most wild parents lose their wings, these 12 pairs were considered to be in the same condition as wild parents and were used for analysis. Of the remaining pairs, nine pairs ate wings partially and three pairs did not eat wings.

Results

The area of the wings eaten was $1.54 \pm 0.16 \text{ cm}^2$ (66.7% of the entire wing area) in males and $1.58 \pm 0.46 \text{ cm}^2$ (68.3%) in females (mean \pm SD, male: $n = 10$, female: $n = 10$; four individuals that escaped after wing-eating were not measured). There was no significant difference between sexes (Welch's t-test, $t = -0.27$, $df = 11.2$, $p = 0.79$).

S. taiwanensis ate its mate's wings in the following typical sequence. The "actor" is the individual that is actively eating the wings. The "recipient" is the mate of the actor.

- (1) The actor approached its mate, exhibiting antennating (12/12 pairs).
- (2) The actor licked its mate's body parts (grooming, 12/12 pairs). The body parts groomed were the head, pronotum, abdominal tergum, wings, and legs. This step was occasionally skipped.
- (3) The actor climbed onto the recipient's back (Fig. 2) and ate the recipient's wings (wing-eating, 12/12 pairs, Movie S2).
- (4) During (2) and (3), the recipient did not walk (12/12 pairs). The recipient did lean either toward the actor (dorsal-exposure, 12/12 pairs, Movie S2) or toward the opposite side of the actor (ventral-exposure, 10/12 pairs, Movie). Dorsal-exposure was more frequently observed when wings were being eaten rather than groomed (Mantel-Haenszel test, common odds ratio = 2.51, $df = 1$, $p < 0.001$). Ventral-exposure was observed only during grooming (Mantel-Haenszel test, common odds ratio = 0.026, $df = 1$, $p < 0.001$).
- (5) The actor stopped eating although its mate's wings were partially intact. In 61.7% of times of stopping, the recipient remained motionless. In the other 38.3% of times, the recipient violently shook its body left and right (wagging, 12/12 pairs, Movie S3). After the recipients wagged, the actor stopped eating more often (Mantel-Haenszel test, common odds ratio = 5.71, $df = 1$, $p < 0.001$).

A sequence from step (1) to (5) was defined as one bout (Fig. 3). The actor in each bout was either the same or the opposite sex as the actor in the preceding bout. The female and male ate in several bouts until the end of mutual wing-eating. Once a bout was stopped, the pair usually did not groom and eat wings until the next bout. During the intervals between bouts, both sexes were either motionless, walking, or digging. Digging is scratching the ground using the forelegs (9/12 pairs, Movie S4). In the first bout for each pair, the actor was almost always male (11/12 pairs).

Discussion

Mutual wing-eating, found in most wild pairs of *S. taiwanensis*, is considered sexual cannibalism or nuptial feeding. In all the known examples of these behaviors, one sex consumed the mate or gift unilaterally (Prenter et al. 2006; Lewis et al. 2014). Mutual wing-eating in *S. taiwanensis* is unique and may offer a challenge to the present understanding of the adaptive significance of sexual cannibalism or nuptial feeding.

Sexual cannibalism usually involves the female consuming the male. The reverse

type is rare. The benefits of sexual cannibalism may include obtaining nourishment for reproduction or encouraging males to avoid mating with inferior females (Prenter et al. 2006). Mutual wing-eating is like sexual cannibalism, but it differs in that one mate is not killed. Since wings include no flesh, the value of wings as a food resource appears to be negligible.

In nuptial feeding, males usually give materials other than sperm to improve their number of offspring (Lewis et al. 2014). The reverse type only occurs among Zeus bugs, where females feed males to reduce the rate of male kleptoparasitism (Arnqvist et al. 2006). Although sufficient food was provided, *S. taiwanensis* still engaged in wing-eating. *Salganea taiwanensis* did not eat wings to compensate for a lack of food. There exists only one example where one individual gives a body part to its mate, i.e., sagebrush crickets (Morris 1979). The sagebrush cricket male allows the female to feed on his fleshy hind wings during copulation to increase the chances of the transfer of a spermatophore (Sakaluk et al. 2019). *S. taiwanensis*, however, cannot eat its mate's wings during copulation because of the end-to-end position. Also, the wings are unlikely to be of nutritional value. Therefore, mutual wing-eating cannot be explained by the previous interpretations of nuptial feeding (Lewis et al. 2014).

To explore a new hypothesis for the significance of mutual wing-eating, the life-history of *S. taiwanensis* is helpful. The mating pair of *S. taiwanensis* begin to care for the offspring biparentally after making a gallery in a rotten log. Females give birth to their offspring in multiple cycles for several years (Maekawa et al. 2008). For this reason, the adult life span is expected to be several years, and the parents are expected to live in the gallery together for their lifespan. This mating system of *S. taiwanensis* is considered true monogamy, where mates copulate only with each other for life (Bell et al. 2007). There are three reasons for true monogamy. First, since *S. taiwanensis* is a wood-feeding cockroach, outside of the rotten log, it loses the food. Second, because the density of reproductive individuals seems not to be high (observation by H.O.), an adult may seldom encounter another mate. Third, particularly after losing the wings, the adults cannot fly ever, the risk of predation outside the gallery may be high. In light of these, it is probably not adaptive to go outside the gallery. Further, no colony with three or more wingless adults has been observed in the field (observation by H.O., in my study area). This shows that other adults hardly ever enter galleries with pairs. Therefore, after mating, adults would not leave the mate and encounter another adult

except for the mate ever.

I propose a hypothesis where mate's fitness is mutually increased by mutual wing-eating in *S. taiwanensis*. In true monogamy, a female's fitness is equal to that of the male (i.e., the number of offspring produced by the pair). Thus, any behavior that benefits one member of the pair also benefits the other. Further, because *S. taiwanensis* cares for offspring biparentally, increasing its mate's survival or care ability leads to an increase in its fitness. Under this hypothesis, if *S. taiwanensis* benefits by losing its wings, recipients are not expected to resist the eating. Recipients are stationary while their wings are being eaten, which is consistent with this expectation. Three factors may make losing of wings beneficial: (i) wing management, in which adults can reduce maintenance costs of wings and flight muscles and invest further in reproduction or care (histolysis in flight muscles was observed in several wild parents); (ii) hygiene management against mites or mold, in which the absence of mites and mold would improve colony health (aggregations of mites and mold were observed on the wings of newly emerged adults in the laboratory); and (iii) adaptation for the insect gallery, in which wings are more physically inconvenient for adults in the gallery.

Actors in the first bouts were almost always males (11/12 pairs). Although both females and males benefit from mutual wing-eating, benefits may not be incurred equally. Some adaptive effects could differentially favor males.

This hypothesis is based on true monogamy, which is complete and lifelong. This is the only condition in which mating is free of sexual conflict (Lessells 2006), however, it has not been demonstrated in the field. Cooperation between males and females may be compromised by betrayal behavior. Generally, individuals that exploit benefits gained by mates' investment with less investment of their own may improve their fitness (e.g., Kölliker 2012). However, by assuming that mating is conflict-free, cooperation without betrayal should exist. Such cooperation can be called true cooperation. Mutual wing-eating may be an example of true cooperation and may help explain some interactions between females and males from a new viewpoint.

References

Arnqvist G, Jones TM, Elgar MA (2006) Sex-role reversed nuptial feeding reduces male kleptoparasitism of females in Zeus bugs (Heteroptera; Veliidae). *Biology Letters* 2, 491–493.

- Bell WJ, Roth LM, Nalepa CA (2007) Cockroaches: ecology, behavior, and natural history. JHU Press, Baltimore, MD.
- Fox LR (1975) Cannibalism in natural populations. *Annual review of ecology and systematics* 6, 87–106.
- Kölliker M (2012) The evolution of parental care. Royle NJ, Smiseth PT, Kölliker M [eds.]. OUP Oxford, New York.
- Lessells CM (2006) The evolutionary outcome of sexual conflict. *Philosophical Transactions of the Royal Society B: Biological Sciences* 361, 301–317.
- Lewis SM, Vahed K, Koene JM, Engqvist L, Bussière LF, Perry JC, Gwynne D, Lehmann GUC (2014) Emerging issues in the evolution of animal nuptial gifts. *Biology Letters* 10, 20140336.
- Maekawa K, Matsumoto T, Nalepa CA (2008) Social biology of the wood-feeding cockroach genus *Salganea* (Dictyoptera, Blaberidae, Panesthiinae): Did ovoviviparity prevent the evolution of eusociality in the lineage? *Insectes Sociaux* 55, 107–114.
- Morris GK (1979) Mating Systems, Paternal Investment and Aggressive Behavior of Acoustic Orthoptera. *The Florida Entomologist* 62, 9–17.
- Obata Y (1988) Master of Science Thesis, Department of Biology, University of Tokyo, Tokyo Behavioral and ecological studies on life history traits and familial relationship in the wood-feeding cockroaches *Panesthia angustipennis spadica* Shiraki, *Salganea esakii* and *S. taiwanensis* Roth (Blattaria: Blaberidae, Panesthiinae). University of Tokyo.
- Prenter J, MacNeil C, Elwood RW (2006) Sexual cannibalism and mate choice. *Animal Behaviour* 71, 481–490.
- Sakaluk SK, Duffield KR, Rapkin J, Sadd BM, Hunt J (2019) *Advances in the Study of Behavior* The troublesome gift: The spermatophylax as a purveyor of sexual conflict and coercion in crickets. 1st ed. Elsevier Inc.
- Shimada K, Maekawa K (2011) Description of the basic features of parent-offspring stomodeal trophallaxis in the subsocial wood-feeding cockroach *Salganea esakii* (Dictyoptera, Blaberidae, Panesthiinae). *Entomological Science* 14, 9–12.

Figures



Fig. 1

Adults of *Salganea taiwanensis* before (right) and after (left) mutual wing-eating behavior. The right individual has long intact wings. The left individual has short wings. Most part of its wings has been eaten by its mate.

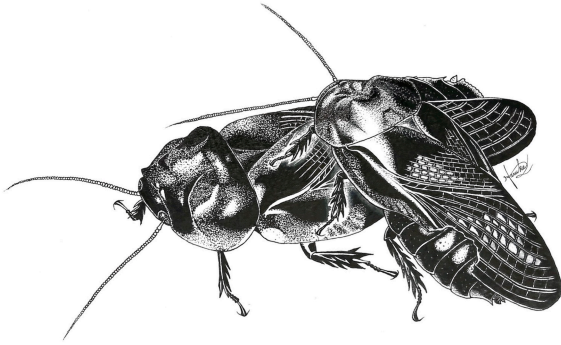


Fig. 2

A drawing of a mating pair during mutual wing-eating behavior by the author (H.O.).
The individual in the front has wings chewed by the mate and is eating the wings of the mate.

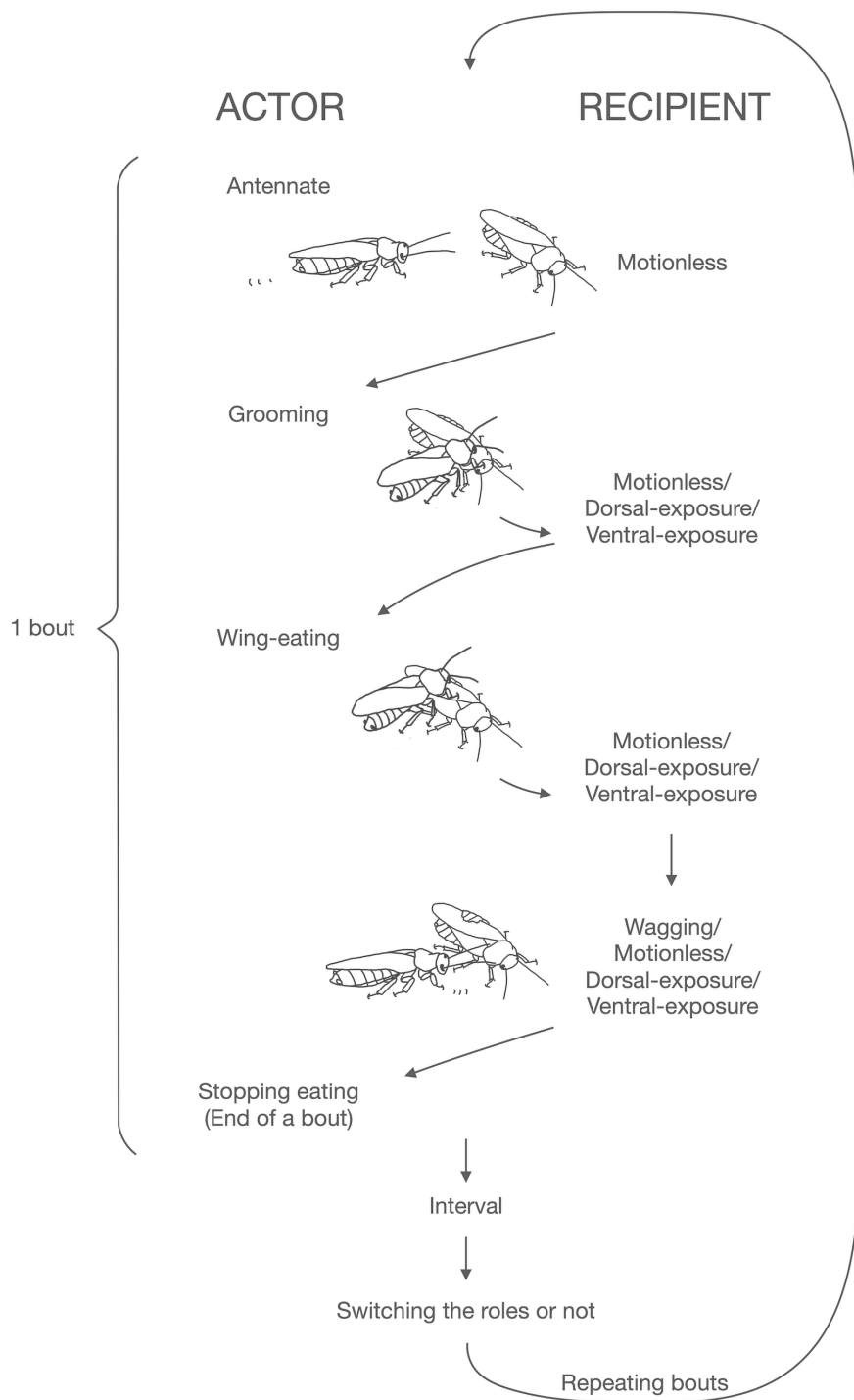


Fig. 3

A typical sequence of mutual wing-eating behavior. Both male and female can be an actor in each bout.

Chapter 2

An observable and efficient rearing system for wood-feeding cockroaches

Abstract

Plant decomposition is an essential component of forest ecosystems. Wood-feeding cockroaches are responsible for the decomposition of rotten logs. However, the ecology of wood-feeding cockroaches is not well understood. To understand how they decompose rotten logs in the gallery inside the logs, it is necessary to observe them in laboratories. Here, I present an observable and cost-effective for rearing these log decomposers using a culture dish and an artificial fungal meal. The culture dish is traditionally used to cultivate fungi or bacteria with the lid on. The artificial fungal meal is a commercial product for stage beetle larvae, which are a kind of log decomposers. These let researchers easy to deal with wood-feeding cockroaches. In the course of keeping over 1,000 individuals of the wood-feeding cockroaches, I have developed effective techniques to rear them under the observable system for several years. I present these techniques along with several tips to keep the wood-feeding cockroaches healthy and observe them using video cameras. These techniques remove many of the limitations for study on the plant decomposers. When researchers use this observable rearing system, I can conduct rearing experiments easily and well understand the ecology of wood-feeding cockroaches and insects associating with them.

Introduction

The essentiality of plant decomposers in forest ecosystems is unquestioned (Odum & Biever 1984; Whitford & Wade 2002). Soils provide ecosystem services that are fundamental to life (Daily 2013). Among plant decomposers, *Salganea* and other wood-feeding cockroaches in general (Bell *et al.* 2007), can have a substantial ecological impact on the decomposition of logs because of their large body size and social behavior (Maekawa *et al.* 2008). *Salganea raggei* and *S. matsumotoi*, for example, were some of the largest wood-feeding insects at the study site at Mt. Victoria in Myanmar (Kon *et al.* 2004). *Salganea* is a subsocial wood-feeding cockroach. In each one colony, the parents and offspring live in galleries inside a rotten log. They not only ingest wood but also shred it without consumption when excavating galleries (Bell *et al.* 2007). It is necessary to observe their behavior within the log to understand how *Salganea* decomposes rotten logs. There has been no method for rearing *Salganea* colonies under observable conditions. When a rearing system is established, it should be applicable not only to wood-feeding cockroaches but also to other log decomposers whose ecology is still unknown.

In addition, *S. taiwanensis* is the only species reported that females and males in a mating pair eat one another's wings (Osaki & Kasuya 2021). This mating behavior is the first mutual sexual cannibalism and important to understand the strategy of females and males. To study this behavior, I was required to establish the observable and long-term rearing system because mutual wing-eating and all behavior following it was considered occurred inside rotten logs. I improved the observation method in Osaki and Kasuya (2021) and have made long-term rearing possible.

To establish the rearing system, there were three problems that I had to solve to facilitate rearing *S. taiwanensis* in the laboratory. The first was that the wood-feeding cockroach would soon die if the environment of the rotten logs in the field was not stably maintained. At first, I had brought pieces of the rotten logs (maximum about 50×150×30 mm) from the field where the wood-feeding cockroaches were collected and used them for rearing the cockroaches in the laboratory. However, about a month later, the wood-feeding cockroaches stopped eating the scraps and died. One reason for their death could be that the rotten logs contained microorganisms that were harmful to the wood-feeding cockroach and these microorganisms had multiplied under the rearing conditions. The second problem is that, I have to maintain a comfortable environment

for a long time because of the longevity of *S. taiwanensis* whose expected lifespan is several years (Osaki & Kasuya 2021). The last problem is that, in a rearing environment, *S. taiwanensis* must still be able to observe their behavior within a rotten wood. Many studies have been conducted on *Cryptocercus punctulatus*, which is also a subsocial wood-feeding cockroach. In these studies, natural rotten logs were placed in the field and *C. punctulatus* was reared in it (Nalepa 1984). However, in this system, the logs must be destroyed to observe the wood-feeding cockroach inside. Therefore, it was impossible to observe their behavior inside the gallery. In addition, the availability of food is an important factor in long-term rearing. *S. taiwanensis* feeds on a large amount of rotten wood. Although it is necessary to change their food frequently, it is a burden for breeders to go and collect rotten logs in good condition every time. Therefore, I present an observable and efficient rearing system. This methodology may be used to solve these problems.

Materials & Methods

The rearing system is shown in Figure 1A. Powdered cellulose moistened with water was spread in culture dishes (the white part), and an artificial fungal meal was crushed and spread on top of the powdered cellulose (the brown part) (Fig. 1B). Powdered cellulose is used as a moisturizer because it has excellent moisture retention properties and is harmless to cockroaches, even if it is consumed.

I used transparent culture dishes (Thermo Fisher Scientific K.K., Waltham, MA, US), powdered cellulose (KC Flock; Nippon Paper Industries Co., Ltd., Tokyo, JP), water, and artificial fungal meals made with white-rot fungus (DEBURO; Fujikon Co., Ltd., Osaka, JP). Other necessary equipment included Bondic (Witmer Industrial, Niagara Falls, NY, US), a 1.5 L container, a hand mixer, and rubber gloves (Fig. 1C). The cost for a full setup is approximately 2,000 JPY.

The advantages of using culture dishes include the following three points. First, high transparency allows for observation of the inside. Second, a slight gap between the dish and the lid allows for ventilation while maintaining high humidity levels. Finally, you can wash the dishes and use them repeatedly. The artificial fungal meal made with white-rot fungus is a Japanese product called “kinshi-bin” used in rearing stag beetle larvae. It is a hard-cupped sawdust infested with only one species of white-rot fungi, such as *Pleurotus ostreatus*. This meal is suitable as food for wood-feeding cockroaches

because they are often collected from rotten logs with stag beetle (*Dorcus titanus*) larvae in the field. *Salganea* can be rear for several days with only the powdered cellulose or the artificial fungal meal, however, it would be impossible to rear the cockroaches for several years. The powdered cellulose has no nutrition from fungus. The artificial fungal meal may be easy to be too dried for the cockroaches in the culture dishes in the incubator.

The size of the dish was determined by the colony size. If only young nymphs comprise the colony, it is better to use a medium-sized dish (outer dimensions 140 × 100 × 20 mm). However, because the lids of the medium-sized dishes are too light, possibly facilitating the escape of the cockroaches, it is necessary to fix the lid with tape. For the other types of colonies, which are composed of mature nymphs or include at least one adult, a large dish should be used (outer dimensions 245 × 245 × 25 mm).

To create a small space between the lid and the container, protrusions were made in the four corners of the container with Bondic (Witmer Industrial, Niagara Falls, NY, US) (Fig. 2). If the space between the lid and the container is too tight, the rearing system will completely seal with the water inside, and the cockroaches will suffocate.

I will describe the process of setting up the rearing system when using the large dish. The powdered cellulose was mixed in a 1.5 L container at a ratio of 300 mL of powder to 500 mL of water. To mix evenly, the container was shaken 10 times. When completely mixed, the mixture was poured into the dish. The dishes were then dried at 65°C for 24 h to allow the extra water to evaporate. If water comes out when you press the mixture with your finger, allow it to dry for several hours because that indicates that there is too much water within it.

The artificial fungal meal made with white-rot fungi was at 5°C until using it in this rearing system. A measure of 120 mL of the artificial fungal meal was crushed in a 1.5 L container with hands that were covered with rubber gloves. The glove is necessary to prevent the contamination of *Penicillium* in this rearing system. If the artificial fungal meal is too hard to crush by hand, use a hand mixer. When the artificial fungal meal is crushed to the point where it resembles sawdust powder, the mixture is spread on top of the cellulose in the dish. At this time, the space above the food was adjusted to leave approximately 7 mm of space within (the height of the adult). This is because *S. taiwanensis* behaves calmly when its tergum is in contact with the lid (Observation by the author). It is consistent that the galleries of wild colonies are approximately 7–10

mm high. The dishes with colonies are kept in the incubator at 25°C under constant darkness.

When the collection of feces becomes noticeable, it is time to change the moisturizer and food. In addition, if *Penicillium* appears, the moisturizer and food should be changed as soon as possible.

Results & Discussion

I established this rearing system in 2019. In 2018, I collected 32 wild colonies of *S. taiwanensis* and attempted to rear them, but all colonies died within a year. After establishment, I have reared 68 colonies (22 colonies were collected in Apr.–Oct. 2019, seven colonies in Feb. 2020, seven colonies in Apr. 2021, and 32 colonies were established by the new adults reared with this rearing system in Jun.–Aug. 2020) of 72 colonies. Only four colonies have died. In the 68 colonies, the colony size has been 14.5 ± 5.8 (mean \pm SD). I made the protrusions with Bondic from Jan. 2021. Table 1 shows the mortality rate before and after making protrusions. The dead adult/nymph ratio was 0.18. Of the four colonies that died, two died from suffocation, one from mold, and the other from anything not identified. With this rearing system, individuals of *S. taiwanensis* have been able to produce young and the nymphs have become adults. In Jun.–Aug. 2020, I made the 32 pairs with these new adults and all pairs reproduced. Consequently, important events in growth and reproduction, such as multiple molts, emerging, and hatch, of *S. taiwanensis* were observed with this rearing system in culture. The closest species, *S. esakii*, could also be reared in this way. Now, I succeeded in rearing three colonies of six colonies of *S. esakii*.

I used this system for recording the behavior of the mating pair of *S. taiwanensis* inside a small dish ($\phi 90 \times 15$ mm) with the lid on (Movie S1). If the lid had sawdust on it, it was possible to observe the inside after replacing the lid with a new one and leaving the pair for maximum 30 min. To record their behavior using video cameras while reproducing the darkness of their gallery in the wild, I turned on a red light, which the cockroach cannot sense, 30 cm above the dishes. The light and dishes were placed so that the angle between them was approximately 30° to ensure that the light did not reflect off the video image. Because the transparency of the lid is high and the light was able to reach the inside even with the lid on, I was able to observe their behavior.

The described methodology for rearing plant decomposers is efficient and generally

does not require extra equipment or permits beyond the usual rearing of insects. Needless to say, researchers not only can rear the wood-feeding cockroaches also study their emerging, reproduction, and parental care with this methodology. This observable system can also be applied in ecological studies of other wood-feeding insects whose ecology are unknown. For example, the insects associating with *Salganea*, such as *Coilodera penicillata* and *C. miksici* of the cetoniine beetles (Kon *et al.* 2004) and *Daulocoris formosanus* of the flat bug (Osaki *et al.* 2018). This observable rearing system already has successful cases to rear four individuals of *Cylindrocaulus patalis* (Murakami, pers. comm.), which is a log decomposer and has unique ecology to raise the offspring. This methodology will facilitate a breakthrough in understanding the ecology of log decomposers and wood-feeding insects.

References

- Bell WJ, Roth LM, Nalepa CA (2007) Cockroaches: ecology, behavior, and natural history. JHU Press, Baltimore, MD.
- Daily GC (2013) Nature's Services: Societal Dependence on Natural Ecosystems (1997). In: Robin L, Sörlin S, Warde P (eds) The Future of Nature, pp 454–464. Yale University Press, London.
- Kon M, Maekawa K, Sakai K, Kusakabe Y, Araya K (2004) The cetoniine beetles, *Coilodera penicillata* and *C. miksici* (Coleoptera, Scarabaeidae, Cetoniinae), collected from the galleries of wood-feeding cockroaches (Blattaria, Blaberidae, *Salganea*) in Myanmar. *Kogane* (5), 16–20.
- Maekawa K, Matsumoto T, Nalepa CA (2008) Social biology of the wood-feeding cockroach genus *Salganea* (Dictyoptera, Blaberidae, Panesthiinae): Did ovoviviparity prevent the evolution of eusociality in the lineage? *Insectes Sociaux* 55, 107–114.
- Nalepa CA (1984) Colony composition, protozoan transfer and some life history characteristics of the woodroach *Cryptocercus punctulatus* Scudder (Dictyoptera: Cryptocercidae). *Behavioral Ecology and Sociobiology* 14, 273–279.
- Odum EP, Biever LJ (1984) Resource Quality, Mutualism, and Energy Partitioning in Food Chains. *American Naturalist* 124, 360–376.
- Osaki H, Kasuya E (2021) Mutual wing-eating between female and male within mating pairs in wood-feeding cockroach. *Ethology* 127, 433–437.

- Osaki H, Shimamoto S, Kasuya E (2018) First record of habitat of *Daulocoris formosanus* (Hemiptera: Aradidae): Colonies discovered in nests of *Salganea taiwanensis ryukyuanus* (Dictyoptera: Blaberidae). *Entomological Science* 21, 340–342.
- Whitford W, Wade EL (2002) Decomposition and Nutrient Cycling. In: Whitford W, Wade EL (eds) *Ecology of Desert Systems*, pp 235–274. Academic Press, London.

Table 1

The mortality rate of colony members before and after making protrusions in Jan. 2021.

	rearing months	<i>n</i>	mortality rate	
			mean	SD
before (~Dec. 2020)	28	9	0.212	0.273
	25	4	0.444	0.096
	22	4	0.304	0.342
	18	10	0.248	0.273
	16	8	0.196	0.176
	14	8	0.000	0.000
	13	9	0.116	0.114
	12	6	0.261	0.175
	10	7	0.200	0.157
after (Jan. 2021~)	8	68	0.000	0.000

The colonies in “before“ period are all colonies excepted for seven colonies collected in Apr. 2021. In the “before“ period, four colonies were dead. The colonies in “after“ period are all colonies excepted for these dead four colonies. The rearing months shows the time length of rearing from collecting or establishment to now (Aug. 2021).

Figures

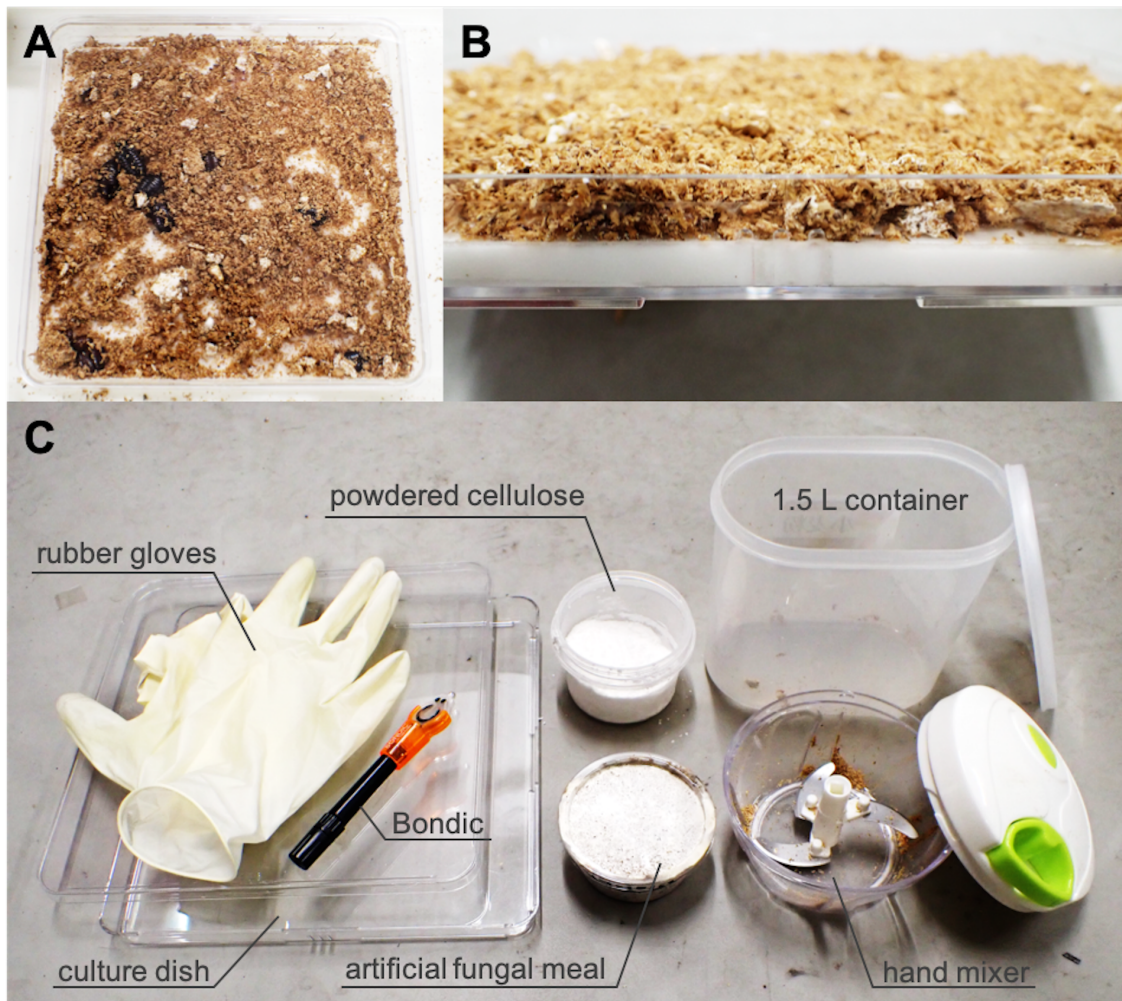


Fig. 1

(A) The observable rearing system with a colony of *S. taiwanensis*. (B) The side view of the rearing system. (C) The materials and tools used to set up the rearing system.



Fig. 2

The protrusion made by Bondic on a corner of the dish (inside the circle).

Chapter 3

Ecology of *Daulocoris formosanus*, an associate insect of *Salganea taiwanensis ryukyuanus*

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Abstract

I collected colonies of *Daulocoris formosanus* from nests of the subsocial wood-feeding cockroach *Salganea taiwanensis ryukyuanus* for the first time. To understand the ecology of *D. formosanus*, I observed and characterized their behavior in a laboratory setting. I provide the first detailed record of the habitat, colony structure, and feeding behavior of *D. formosanus*.

Introduction

Daulocoris formosanus Kormilev, 1971 is a large flat bug belonging to the family Aradidae. Some genera of Aradidae live in colonies (Usinger and Matsuda 1959); however, very little is known regarding the ecology of *D. formosanus*, including their habitat, colony structure, and feeding behavior. In Japan, they were discovered when researchers accidentally collected not more than three individuals in one instance. Although there are several specimens in total (unpublished data, specimens preserved in Tokyo University of Agriculture), none of their colonies were found and no information on their ecology was acquired. In Taiwan, there are a series of specimens caught at the same time; however, no ecological information has been reported (Kormilev 1971).

Therefore, to understand the ecology of this species, I collected and characterized *D. formosanus* colonies in April 2016 and May 2017. This is the first report detailing the habitat, colony structure, and feeding behavior of this species. I describe the collecting circumstances and the behaviors that I observed.

Field survey and observation in culture

Field research was conducted twice, during April 2016 and May 2017, at the Yona Field in the experimental forest of the University of the Ryukyus, which is located on Okinawa Island (26°45'17.80"N, 128°13'9.46"E). Upon collection, individuals within the same tunnel of cockroaches were considered a single colony. I investigated 57 cockroach nests and the collection comprised two colonies and one individual. The colony collected in 2016 was designated as colony A and the one collected in 2017 was designated as colony B. The individual was also collected in 2017. For collection, rotten logs along the trail were split with a hatchet, and insects inside were collected and placed in plastic containers (206×104×41 mm) for transport, with each colony housed in the same container with particles of the rotten log. I kept colony B without the cockroaches for 5 months in an incubator maintained at 25°C with 24 D and observed it every day by my eyes. I kept the cockroaches in the plastic container filled with pieces of nest logs.

Colony structure

Colony A was comprised of 5 adult males, 1 adult female, and 1 nymph collected on April 18, 2016. There were more individuals in colony A at the collection site, but I was

unable to collect all the individuals. Colony B was comprised of 3 adult males, 3 adult females, and 4 nymphs collected on May 13, 2017. The individual was an adult female collected on May 15, 2017. In colony B, 12 nymphs hatched in the incubator on the following day; however, I could not locate the remains of eggshells.

Collection conditions and habitat

D. formosanus lived in the nests of wood-feeding cockroaches, *Salganea taiwanensis ryukyuanus* Asahina, 1988 (Fig. 1a). This is the first record of aradids found in the nests of wood-feeding cockroaches. All rotten logs where *D. formosanus* were found had extensive white-rot fungus. Each log was 20–30 cm in diameter. I could not identify the plant species from which the wood was derived due to extensive rot damage (Fig. 1b).

Frass and scraps of wood made by cockroaches had accumulated on tunnel walls. *D. formosanus* remained stationary on the walls. For this reason, they were surrounded with fine frass and scraps, which adhered to their body surface (Fig. 1c).

Relationship to *S. taiwanensis ryukyuanus*

Several aradid species including a species of *Brachyrhynchus*, which is a closely related genus of *Daulocoris*, have been observed to be living with termites (Kormilev 1955a, b; Usinger & Matsuda 1959; Kormilev 1967, 1976, 1980, 1982). These authors wrote that the relations between aradids and termites are symbiotic.

However, in my field observations, *S. taiwanensis ryukyuanus* behaved independently of *D. formosanus*. This conclusion was based on the lack of contacts, such as antennating or biting, between *D. formosanus* and *S. taiwanensis ryukyuanus*. Based on these field observations, I suggest that *D. formosanus* and *S. taiwanensis ryukyuanus* cohabitate but do not engage in the special relationship with direct touch.

I did not observe any attacks toward *D. formosanus* by *S. taiwanensis ryukyuanus*. This species of cockroach is generally considered subsocial and lives in tunnels inside rotten logs (Costa 2006). Subsocal cockroaches defend their nests from invaders without attacking or causing injury (Costa 2006), which is consistent with my observation that *S. taiwanensis ryukyuanus* did not attack *D. formosanus*. Some species of Aradidae have been found in close proximity to termites (no specific name was mentioned) or ants (*Pachycondyla chinensis* and *Paratrechina amia*) and even when ants are aggressive against invaders, they behave independently of aradids (Kormilev

1976; S. Nagashima 2017, personal communication). Thus, the behavior of *S. taiwanensis ryukyuanus* toward *D. formosanus* is similar to that of these hosts.

Although there is no direct touch between the two species, *D. formosanus* and *S. taiwanensis ryukyuanus* possibly engage in a symbiotic relationship. It is based on the fact that it was only in nests of *S. taiwanensis ryukyuanus* that researchers collected colonies of *D. formosanus* having their nymphs. In colony B that I kept in culture, all nymphs died in their fourth instar. There is a possibility that nymphs need *S. taiwanensis ryukyuanus* to become adults.

Feeding behavior in culture

I observed two times that *D. formosanus* individual from colony B in culture pushed their stylet into the rotten log. Some fungus-feeding aradids have been reported to feed in a similar manner (Usinger and Matsuda 1959). I speculate that *D. formosanus* feeds on tunnel walls containing *S. taiwanensis ryukyuanus* rather than on the surface of logs under the bark because the tunnel walls are softer, moister, and closer to the fresh fungus than the surface is. In the case of Mezirinae, flat bugs found in close proximity to termites, it has been suggested that they live in this location because of a food source (Kormilev and Froeschner 1987).

References

- Costa JT (2006) *The Other Insect Societies*. Harvard University Press, Cambridge, MA.
- Kormilev NA (1955a) Notes on Aradidae from Eastern Hemisphere III (Hemiptera): On some Aradidae from Japan, Russian Far East and Formosa (Taiwan). *Mushi*, 28, 31–41.
- Kormilev NA (1955b) Notes on Aradidae from the Eastern Hemisphere (Hemiptera). VII. Aradidae from Fukien, South China. *Quarterly Journal of the Taiwan Museum*, 8, 177–191.
- Kormilev NA (1967) Aradidae in the South Australian Museum, Adelaide II. (Hemiptera-Heteroptera). *Records of the South Australian Museum*, 15, 513–550.
- Kormilev NA (1971) Mezirinae of the Oriental region and South Pacific (Hemiptera-Heteroptera: Aradidae). *Pacific Insects Monograph*, 26, 1–165.
- Kormilev NA (1976) Notes on Aradidae from China and India. *Pacific Insects*, 17, 87–90.

- Kormilev NA (1980) A new species of the genus *Daulocoris* Usinger and Matsuda, 1959, from Sumatra (Hemiptera: Aradidae). *Sociobiology*, 5, 21–24.
- Kormilev NA (1982) A new species of *Neuroctenus* from nests of termites (Hemiptera: Aradidae). *Sociobiology* 7, 25–28.
- Kormilev NA, Froeschner RC (1987) Flat bugs of the world : a synonymic list (Heteroptera: Aradidae). *Entomography*, 5, 1–245.
- Usinger RL, Matsuda R (1959) Classification of the Aradidae (Hemiptera-Heteroptera). British Museum, London.

Figures

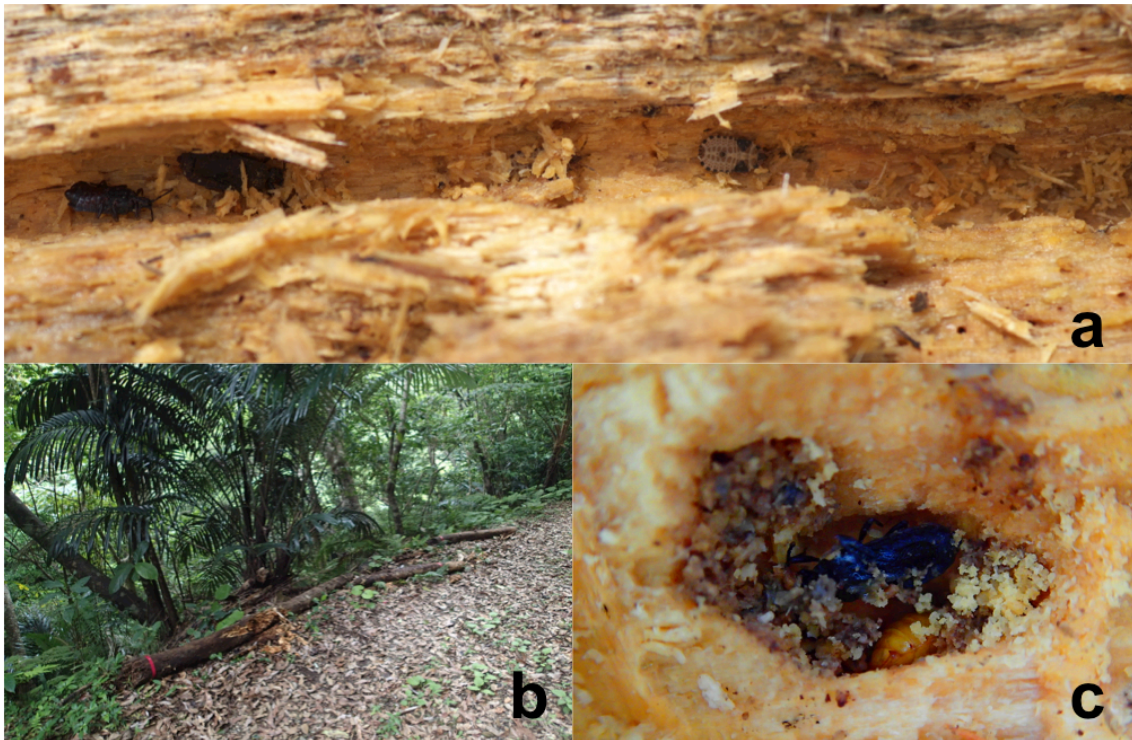


Fig. 1

(a) Colony B contains 2 adults and 2 nymphs of *Daulocoris formosanus* in the nest of *Salganea taiwanensis ryukyuanus*; (b) the location where *D. formosanus* colonies were found; (c) an adult member of colony A with a nymph of *S. taiwanensis ryukyuanus*.



Fig. 2

An adult female from colony B feeding on the rotten log in culture.

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