Chapter 4
Parental Care

Lisa Filippi, Mantaro Hironaka, and Shintaro Nomakuchi

Abstract *Parastrachia japonensis* Scott (Heteroptera: Parastrachiidae) mothers guard eggs and nymphs and provision nymph-containing nests with drupes of the host tree, *Schoepfia jasminodora* (Schoepfiaceae). We describe our journey of discovery over the years from drawing conclusions about observations of natural behavior in the field to testing of hypotheses through manipulated field and laboratory studies. The journey has clarified the ecological constraints on the life history of *P. japonensis*: poor quality and abundance of drupes, predation, intraspecific competition, and severe weather. We explain how we identified patterns of nesting and the adaptiveness of the parental care behaviors that mitigate these constraints. Provisioning females choose only high-quality drupes to provision their nests because nymphs on their own cannot access sufficient high-quality drupes to sustain development. Female guarding of eggs and nymphs protects against predation by a ground beetle, but, importantly, provisioning also protects young because they remain in the safe confines of the nest longer if sufficient drupes are provided. Females also produce trophic eggs that enhance all measures of offspring success. They use risk-sensitive decision-making to choose when and where to nest and how many fertile and trophic eggs to produce. We explore how risk-sensitive decision-making allows mothers to best respond to the severe ecological conditions experienced during the nesting season that enhance their provisioning success and offspring success.

Keywords Cannibalism · Foraging · Intraspecific competition · Kin recognition · Maternal care · Resource constraints · Nymphal independence · Semelparity

L. Filippi (✉)
Department of Biology, Hofstra University, Hempstead, NY, USA
e-mail: Lisa.Filippi@Hofstra.edu

M. Hironaka
Department of Applied Entomology, Faculty of Bioproduction Science, Ishikawa Prefectural University, Nonoichi, Ishikawa, Japan
e-mail: hironaka@ishikawa-pu.ac.jp

S. Nomakuchi
Retired from the Faculty of Agriculture, Saga University, Saga, Japan

4.1 Introduction

The evolution of parental care represents an enormous leap in a species' life history. To make that leap, critical challenges had to have been met because, while providing parental care always enhances the success of offspring, it also always involves considerable trade-offs. The parent providing care typically experiences limits to foraging time, opportunities to mate and produce future offspring, ability to maintain its health, and even to its survivorship via increased susceptibility to predation and disease. Thus, when parental care emerges in a species, it can be assumed that the trade-offs always favor providing care as the best way to increase the provider's fitness (Clutton-Brock 1991). E.O. Wilson (1971, 1975) established four prime environmental movers that favor the evolution of parental care: unusually harsh and stressful environmental conditions, stable structured environments, predator pressure, and scarce, and specialized food resource. To be sure, these environmental states are commonly experienced by most insects, yet parental care in the insects is not the norm. In fact, outside the eusocial insects, relative to the number of insect species out there, parental care in the insects is pretty darn rare. Among an average estimated 5.5 million insect species (Stork 2018), subsociality, any care of offspring after oviposition (Michener 1969), has evolved in 50 families among more than 12 of the 25–30 insect orders (Costa 2006). Considering that there are an estimated 1000 insect families (Grimaldi and Engel 2005), only 5% of them have evolved any form of parental care, from the simplest behavior of guarding eggs to guarding immatures, provisioning immatures and the pinnacle of care, progressively provisioning immatures. Roughly 1% of all insect species progressively provision young, and that includes all Eusocial species (Royle et al. 2014). Rather, most insects have found the greatest success by evolving minimal investment in care beyond scattering their eggs over time and space in places where the offspring will have a fair chance of finding food and shelter after they hatch (Eickwort 1981a, b). Tallamy and Wood (1986) have categorized the parental care behaviors seen in insects that provide some form of care for their young as those that physically protect young from predation and other environmental dangers, provide and/or guard a resource vital to the offspring, or prepare a food resource for offspring feeding.

We will discuss the specific factors that have promoted the evolution of parental care in *Parastrachia. japonensis* Scott (Heteroptera: Parastrachiidae) from a theoretical framework as we move through the chapter. However, the field has advanced considerably in recent years, and for eloquent, thorough, and thoughtful coverage of these topics, we direct readers to Trumbo (1996), Costa (2006), Royle et al. (2012, 2014), Wong et al. (2013), and Gilbert and Manica (2015). Our goal is to take the reader on our specific journey, the one that revealed the marvelous and varied manifestations of parental care in *P. japonensis*. Prior to the start of our research at Mt. Hinokuma, it was known that female *P. japonensis* engaged in some parental care behaviors; the mother guarded a round egg mass (Miyamoto 1965) and young nymphs (Tachikawa and Schaefer 1985). The food resource had finally been identified as drupes of the gray twig tree, *Schoepfia jasminodora* (Schoepfiaceae)

(Gyôtoku and Tachikawa 1980; Tachikawa and Schaefer 1985; Tachikawa 1991). Moreover, Tachikawa and Schaefer (1985) suspected but never observed females provisioning drupes to the young. We recognized that if mothers were in fact repeatedly provisioning nests with drupes, it would represent an extremely unusual behavior in an insect, and it had never been reported for a phytophagous heteropteran. So, in 1989 we set out to find evidence of progressive provisioning behavior. Over the first 2 years of field work, we verified that females did in fact provision nests with drupes of the *Schoepfia jasminodora*.

Early studies from 1989 to 1991 focused on documenting and gathering evidence of the behaviors; later studies focused on the behaviors from a behavioral ecology perspective. Which of the ecological factors postulated by Wilson (1971, 1975) and Tallamy and Wood (1986) promoted their evolution? Because so little was known about this insect at the outset of our research, every behavior we documented (aggregating, mating, several parental care behaviors) was new and exciting. The early years in the field were nothing short of thrilling; there were so many avenues of research to pursue, and we felt like children in a candy store. We begin this chapter with a summary of the important findings of our first paper on parental care in *P. japonensis* (Tsukamoto and Tojo 1992).

4.2   Nesting and Maternal Care of Eggs

After feeding on the host tree for about 10 days, during which time egg development took place, inseminated females climbed down the host tree, their abdomens conspicuously laden with eggs, and roamed on the ground in and around the leaf litter searching for a site to nest. There was very little leaf litter under the host tree, and most females walked 5–10 m away from the host tree to nest. In fact, as described in Sect. 1.10, in 1990, only 4 of 24 nests were located within the area directly under the crown of the host tree, *S. jasminodora*, where drupes either landed, or where they had rolled to in the area outside the crown (foraging area). The remaining 20 (83%) nests were under the freshly fallen leaf litter of a large *Ilex chinensis* tree (Fig. 4.1; Tsukamoto and Tojo 1992). Females excavated a shallow burrow under the leaf litter and oviposited a round cream-colored egg mass, which gradually turned yellow, then pale pink, and finally red just prior to hatch. We verified that the mothers not only hovered over the egg mass, guarding it, but kept it suspended on the end of the stylets (Fig. 4.2a; Tsukamoto and Tojo 1992). Mothers remained in the nest until hatch about 10–14 days later, unless disturbed, in which case the female carried the eggs off to a new site.

Unlike other cydnid relatives that bring seeds into the nest and feed during the incubation period (Sites and McPherson 1982), females never brought drupes into the nest before the eggs hatched, and they never left to feed. Eggs that were attended by a female in the field always appeared healthy; however, six egg masses separated from the mother and placed on moist filter paper in individual lidded plastic cups in an incubator failed to hatch. Mites were eating some of the eggs in every mass, and
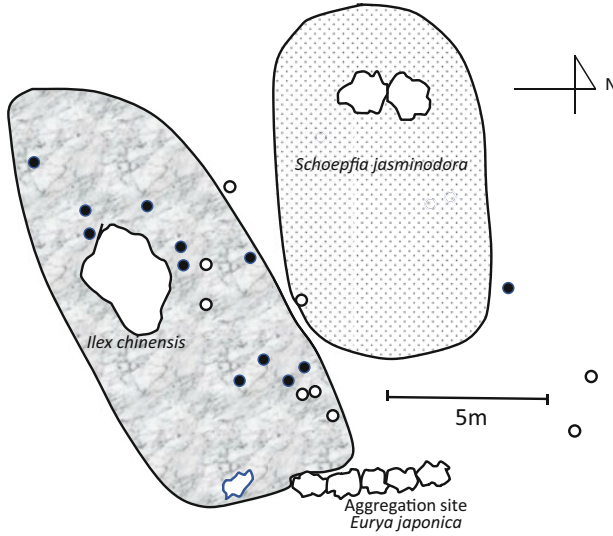
Fig. 4.1 Nesting site studied at Mt. Hinokuma in 1990. White patches represent the trees indicated. The site gradually sloped down from the host tree to the aggregation site. Dark circles, nests that contained drupes; white circles, nests that never contained drupes. Shaded area around *I. chinensis* indicates the area under the canopy of the tree where leaves fell; the dots around the host tree represent the area where drupes fell under the canopy and rolled downhill. (Adapted from Tsukamoto and Tojo 1992)

Fig. 4.2 Nesting behavior of *P. japonensis.* (**a**) Female guarding newly oviposited egg mass and (**b**) newly hatched nymphs in nest. Leaf litter covering was removed to take the photographs. Photo on left is credited to Hiromi Mukai. Adapted from Tsukamoto and Tojo (1992)

fungus was also growing as they gradually turned black and rotted (Tsukamoto and Tojo 1992). Maternal grooming of eggs that prevents fungal and other microbial infections has been reported for a number of insect species including earwigs (Meunier and Kölliker 2012; Boos et al. 2014), termites (Matsuura et al. 2007), and, a close relative of *P. japonensis*, the cydnid *Adomerus triguttulus* (Nakahira and

Kudo 2008), and we strongly suspect that in the field, *P. japonensis* mothers are grooming their eggs during incubation, removing fungus and mites.

We wondered whether females could distinguish their own eggs from those of their neighbors. While mothers of the reduviid bug *Pisilus tipuliformis* can distinguish their own eggs from those of other conspecific females (Parker 1965), *P. japonensis* mothers apparently lack that ability. We brought six females with their egg mass back to the laboratory and placed them in an incubator in individual cups. Each female was placed in the center of three egg masses, one of which was her own. The following morning two of the six females had claimed their own egg mass, three chose another female's eggs, and one female hovered over two of the three egg masses simultaneously, neither of which was her own (Tsukamoto and Tojo 1992)! In the field, females rarely leave their eggs unattended, so we expect that there really was no need for egg discrimination to evolve.

4.3 Provisioning Behavior

4.3.1 Method of Provisioning

In 1990, we observed the 24 nests several times a week from soon after oviposition until the third instar, when nymphs typically left the nest. We did not mark the females for this study, but at each observation, there was only ever one female in the nest, holding her eggs or guarding nymphs. We assumed it was the mother of the eggs in the nest. Females did not leave the nest until the eggs hatched. Within hours after hatch, females left the nest to forage for drupes. A female typically spent hours roaming about the foraging area, inspecting many drupes of different color and ripeness. When a suitable drupe was finally found, she snatched it up and quickly returned to the nest where she deposited it; the nymphs gathered on the drupe to feed (Fig. 4.3, Tsukamoto and Tojo 1992). Females transporting drupes moved with surprising speed and agility over the rough terrain in the field, despite the heavy and cumbersome burden. The average weight of a drupe was 265.5 ± 49.0 (S.D.) mg ($n = 10$). On the other hand, the average weight of a female soon after oviposition was 159.5 ± 28.6 (S.D.) mg ($n = 6$), but the weight of a provisioning female that hasn't fed for up to 2 weeks is likely to be considerably less. Moreover, females were frequently seen transporting two and occasionally three drupes together that were attached on the same bit of stem. If we do the math, females are dragging drupes that weigh an average of at least 1.7 to more than 5 times their weight at a formidable speed of 33.3 cm/min ($n = 5$; range = 17.5–61.4 cm/min) over an average distance of 254 cm (range 160–430 cm)! *Why all the haste?* Read on to Chap. 6 for the answer to that.

At the time of most observations (three to five times per week), the female was in the nest with the young. On two occasions, the female was away from the nest but was there at the next observation, along with several drupes. Occasionally a female that had been gone from the nest, presumably out foraging, was present in the nest at

Fig. 4.3 Nest provisioning behavior in *P. japonensis*. (**a** and **b**) Female transporting drupes to nest. Drupe is suspended on the end of the stylets. (**c**) Female in nest with young and drupe stockpile immediately after depositing a drupe in the nest. Two drupes attached by the same stem can be seen in the lower left of the photo. (**d**) Third instar nymphs gathering on drupe stockpile to feed. Adapted from Tsukamoto and Tojo (1992)

the next observation; however, there were no drupes in the nest. Apparently unable to find a suitable drupe, the female would return to the nest for a time before heading out to the foraging site again. Clearly finding a suitable drupe was no easy task.

4.3.2 Relationship Between Nest Distance and Provisioning Rate

We verified that females nesting both inside the foraging area and those nesting well outside the foraging area provisioned their nests with drupes (Fig. 4.1; Tsukamoto and Tojo 1992). Interestingly, but perhaps not surprisingly, in 1990, while 12/20 (60%) of the nests outside the foraging range eventually contained drupes, 3/4 (75%) nests within the foraging area eventually contained drupes. We did not want to disturb the nymphs in the nest, so rather than count the drupes at each observation, we counted them after the nests had been abandoned. However, because the entire provisioning period coincided with the rainy season, many of the nests were degraded at that point. Combining data from 1990 and 1991, we were able to
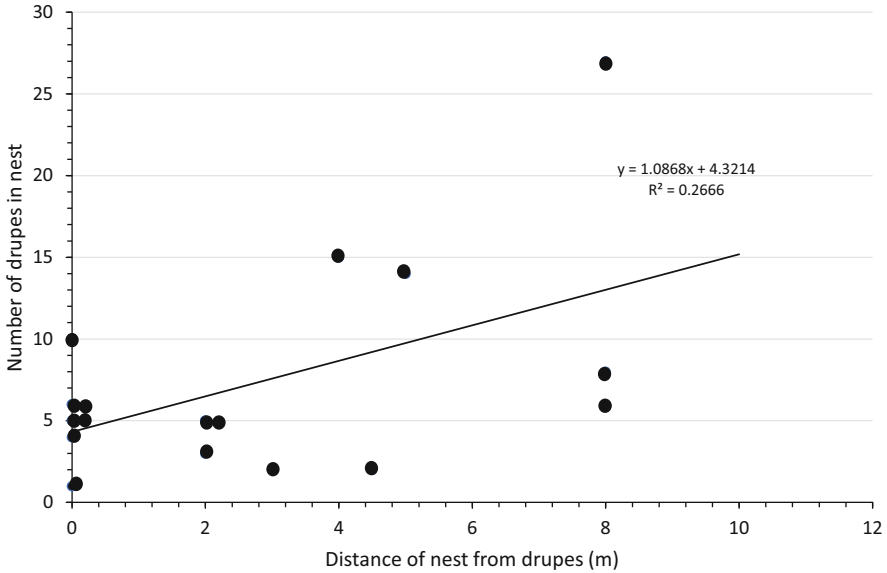
$$y = 1.0868x + 4.3214$$
$$R^2 = 0.2666$$

Fig. 4.4 Relationship between distance of nest from fallen drupe area and number of drupes provisioned. Only nests that had drupes provisioned are included. Data is from 1990 to 1991. $n = 17$. Mean number of drupes provisioned $\pm$ S.D. $= 7.2 \pm 6.2$. Mean distance from the fallen drupe zone $\pm$ S.D. $= 2.7 \pm 3.0$. Pearson correlation coefficient, $r = 0.5163$, $p = 0.034$. Data from 1990 and 1991 combined; adapted from Tsukamoto and Tojo (1992)

count the final number of drupes in 17 nests and plotted the number of drupes provisioned against the distance from the foraging area. Interestingly, there was a significant positive correlation between the distance of the nests from the fallen drupe area and the number of drupes that females provisioned (Fig. 4.4; modified from Tsukamoto and Tojo 1992). It seemed likely that the frail and vulnerable nymphs in nests that were farther away would have a difficult time accessing the foraging site, so females provisioned more drupes. The number of drupes provisioned ranged from 2 to 27, but 25% of all nests (1990 and 1991) never had any drupes. This percentage varies, though, because in 1991, 100% of 12 nests located and monitored from the egg stage eventually contained drupes (data for 1991 not specifically presented in Tsukamoto and Tojo 1992 but inferred).

Females, constrained by obligate semelparity, began dying off when their nymphs were in the third instar, though occasionally they lived and provisioned through the fifth; none ever had a second egg mass, and we have never seen a female lay a second egg mass no matter how much food we provided (unpublished observation, LF). As the mothers died off, nymphs gradually moved to the foraging area where they aggregated as they fed on the drupes.

4.3.3 Conclusions of Our Earliest Study on Parental Care in **P. japonensis**

Most females nested well outside the foraging area, where the leaf litter provides better cover and likely more sanitary conditions (Fig. 4.1). We verified that females do not leave their eggs or the nest unattended until hatch (Fig. 4.2); if disturbed, they move to a new spot with the eggs. They cannot distinguish their eggs from other conspecific eggs, likely because over the course of their natural evolutionary history the need rarely arose. We verified that most females do, indeed, progressively provision their nests with drupes (Figs. 4.1 and 4.3), and this was the first report of a female phytophagous heteropteran progressively provisioning a nest; however, this report came out simultaneously with that of a related cydnid, *Adomerus triguttulus*, that also progressively provisions its nest (Nakahira 1992). While more females nesting within the foraging area provisioned their nests, those nesting farther away that did provision the nest stockpiled more drupes (Fig. 4.4). Nymphs typically remain feeding in the nest until the third instar, the time when females typically begin to die off, and nymphs gradually relocate to the foraging area.

So why the enormous effort of progressive provisioning? Because the females seemed to reject most of the drupes they inspected, we suspected that the resource, while seemingly abundant, was generally of poor quality, and screening by females would provide quicker access to higher quality food. We also concluded that the distance between most nests was prohibitive to young and vulnerable nymphs. However, these and other ecological factors were not clarified in this study. The findings of subsequent studies gradually revealed some fascinating answers!

4.4 Thinking About the Big "Whys"

Now that the parental care behaviors, guarding of eggs and nymphs and progressive provisioning, were confirmed, the next series of studies set out to gather information that could help us answer some of the big *why* questions of behavioral ecology (Tinbergen 1963; Davies et al. 2012): the proximate developmental and mechanistic causes and the ultimate adaptive and evolutionary causes. Our first goal was to explore two of the ecological factors proposed by Wilson (1971, 1975) and Tallamy and Wood (1986) that might reveal an adaptive value of the behaviors, predation pressure, and resource availability. Because parental care will always reduce future reproductive potential, when it evolves evolutionary forces will have arrived at a pattern that results in optimizing the trade-offs in favor of care over no care. When predation risk is low and resources are abundant and available for a prolonged period, iteroparity should be favored, with females producing offspring either singly or in batches repeatedly over space and time, and providing no care, for as long as the resource lasts. However, if eggs and/or early-stage immatures suffer from heavy predation, it would pay for females to reduce predation risk by investing in guarding

for a time before producing additional offspring (Tallamy and Denno 1982). On the other hand, if the resource is ephemeral or scarce, rendering production of additional eggs unprofitable, *and* predation pressure is high, evolution should favor producing a limited number of offspring and investing in extensive parental care (Kudo 1990). This seems to be what is going on with *P. japonensis*.

4.5   Nesting Pattern and Longevity of Nests in 1994

This was year five of our research (1994), and we came to recognize that, although the location of the aggregations in the trees around the host trees was fairly fluid from year to year, and the numbers of nesting females varied considerably, the general pattern of the nests at this site, where females preferred to make them, remained largely the same (Fig. 4.5; Filippi-Tsukamoto et al. 1995).

Because we recognized that collecting large numbers of bugs for experiments in the laboratory and manipulating them in the field like we did for the mating experiments was sure to affect the outcome of any collection site's reproductive success, we had one area at Mt. Hinokuma that we left completely intact, never collecting or manipulating bugs in the subpopulation in any way. We limited our
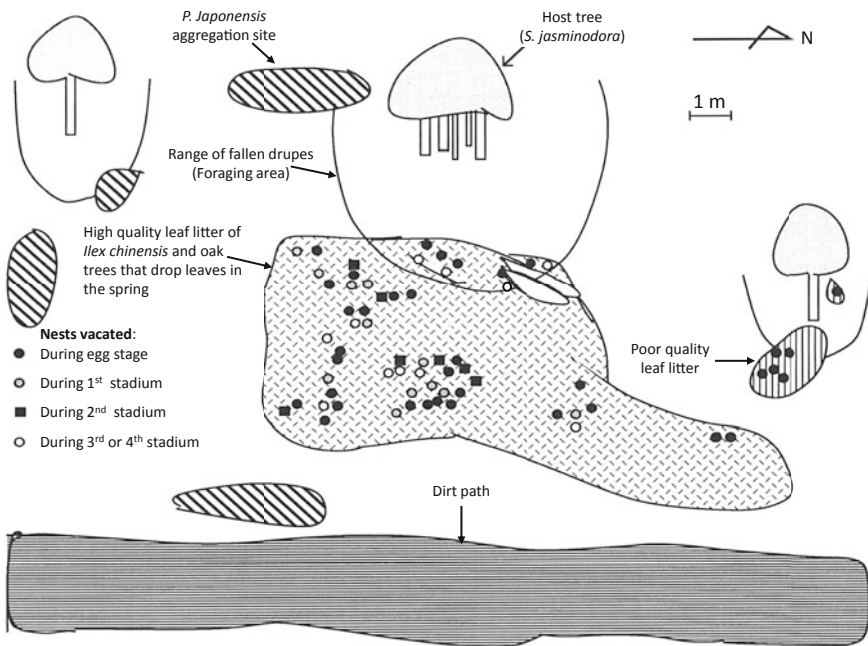

Fig. 4.5 Pattern of nesting and nest persistence at an unmanipulated field site at Mt. Hinokuma in 1994. Adapted from Filippi-Tsukamoto et al. (1995)

work at that site to observing the natural condition, monitoring the natural pattern of nesting each year, and the changes in the size and movement of the subpopulation. Although lifting the leaf litter to observe might have had some effect on the success of the nest, we considered that the disturbance was minimal.

As before, in 1994 females tended to nest at some distance from the host tree and clearly preferred to nest under the leaf litter of trees that dropped their leaves in the spring (*Ilex chinensis* and some oaks). In 1994, we found 66 nests soon after females had oviposited and monitored them regularly throughout the season to gain a better understanding of nesting success in the field. In 1994, the distance of nests from the foraging area ranged from 0 to 5.2 m, mean $\pm$ SD $= 2.3 \pm 1.5$ m (Fig. 4.5; Filippi-Tsukamoto et al. 1995). Of course, it is possible we missed some nests, but these distances were considerably shorter than those from the previous study at the same site. 16.7% of the nests were located within the foraging area, and the remaining 83.3% were outside the foraging area. Only 45% (30) of the nests persisted to the first stadium. It is possible that some of the females relocated their nests, but we searched for new nests daily and did not find any. We had come to realize that predation was a big selective pressure for nesting *P. japonensis*. 16 (24%) of the original 66 nests (53% of the nests that survived to the first stadium) persisted to second stadium, and only 11 (<17%) of the 66 nests (69% of the nests that persisted to the second stadium) persisted to the third or fourth stadium, which is when we typically see nymphs moving over to the foraging area. We were fairly certain these numbers varied from year to year, but the great variation stressed the fact that successful nesting is clearly a challenge for *P. japonensis* mothers. We suspected that the high rate of nest failure was due to predation (see the next section) and to the stressful environment experienced during the heavy rains and flooding of the rainy season, which coincides with the nesting season in Kyushu, Japan.

Second instars rarely relocate to the foraging area, and most nymphs seem to relocate to the foraging area at the third stadium or fourth stadium. Thus, we considered that the 17% of nests that persisted until that point were successful nests and those that vacated before that likely failed. We played with the data presented from the 1994 nesting site in Filippi-Tsukamoto et al. (1995) to see if there was a correlation between distance from the foraging area and success of the nests. Surprisingly, no significant difference between the distance to the foraging area of nests that succeeded to the third instar, and those that failed before the third stadium was found (distance for successful nests, 1.9 ± 1.68 m, and for failed nests, 2.38 ± 1.49 m <mean $\pm$ S.D.>). However, interestingly, in 1994, 36% of nests within the foraging area were successful, while only 17% of those outside the foraging area were. Later studies show that these percentages vary dramatically with the conditions prevailing each year.

4.6 Maternal Egg Guarding Behavior

Protection of eggs from parasitism (Odhiambo 1959), predators (Nafus and Schreiner 1988), and desiccation (Nalepa and Bell 1997; Nalepa et al. 2008) is the simplest form of parental care in the insects. The eggs are usually located in a stationary site that can be defended by covering them with the body (Kudo et al. 1989) or in a nest and may be carried off to safety. The next level would be guarding of young larvae or nymphs from predators, either passively or with aggressive displays (Tallamy and Denno 1981, 1982; Tallamy and Wood 1986; Nafus and Schreiner 1988; Choe 1989) or sometimes by following them around, as with the extended care seen in *Elasmucha dorsalis* (Kudo et al. 1989). By 1991, we had identified the first known predators of *P. japonensis*. On 13 occasions, adults and larvae of the carabid ground beetle (*Apotomopterus dehaanii*, ~30 cm length) were observed feeding on eggs, nymphs, and adults of *P. japonensis*. They appeared to be attracted to the high density of *P. japonensis* on the ground during the nesting season (*P. japonensis* are typically aggregated on shrubs and trees over a meter above the ground), as they were commonly observed roaming specifically throughout the nesting and foraging area during that time. We also frequently observed ants carrying off eggs and nymphs one by one from their nests after immobilizing the mother by feeding on her legs, antennae, and wings but leaving the toxic body (Sect. 1.14) behind. This relentless predation interfered with many an experiment!

We assessed whether females guarding eggs ($n = 17$) and nymphs ($n = 8$) displayed a defensive response against a simulated predator. A female was exposed to one of three levels of stimulation that simulated the disturbance of a predator: the mildest stimulation was rustling the litter close to the nest (approach of a predator) with a disposable chopstick (one of our favorite field tools!); the next intense level of stimulation was gentle probing of the female (predator in the nest) with the chopstick; and the most intense level of stimulation was more forceful probing (attack by the predator). We used a different female for each trial, and the subjects were naturally nesting females. Females guarding an egg mass displayed four behaviors in response to the threat, or no reaction at all. From mildest response to most aggressive, females would flagellate the probe with the antennae; feign death (thanatosis); turn about on the egg mass, presenting the dorsum to the disturbance; and finally lift the egg mass and carry it off in escape. Interestingly, several females did not respond at all to any level of stimulation, but the most frequent behavior was to flee with the egg mass (33% of females with the two mildest stimulations, and over 70% of females with the most intense stimulation; Table 4.1; Filippi-Tsukamoto et al. 1995). Only 2/28 (7%) females displayed a milder response before finally escaping. In other words, most females did not seem to display progressively intense responses but had a particular response that they displayed. Notably, no female ever left her egg mass behind. However, with females guarding nymphs, none failed to respond, and at the level of probing, most females escaped the nest leaving the nymphs behind. While none of the females guarding eggs made the typical chattering sound heard when aggregations are disturbed, two females

Table 4.1 Defensive behaviors of egg and nymph guarding females

| Behavior mild ↓ Intense | Stimulation mild → intense | | | | |
| | Eggs | | | Nymphs | |
	Rustling litter	Gentle probing	Strong probing	Rustling litter	Gentle probing
No reaction	2	2	1	2	0
Flagellate antenna	4	1	0	0	0
Feign death	4	2	1	0	0
Chatter	0	0	0	2	0
Present dorsum	2	3	0	2	1
Escape	6	4	5	4	4
Total responses	18	12	7	8	4
Total females	17	11	7	8	4

Adapted from Filippi-Tsukamoto et al. (1995)

guarding nymphs did. We suspect this alarm sound evolved for an aggregation to deter vertebrate predators, and one female chattering would not be a very effective deterrent. There is no way for a female to guard nymphs, which scattered into the crevices of the nest upon disturbance, so we concluded that by running away from the nest, the female was making her larger body the target of the threat.

We repeated the trials on females with eggs, but this time stimulated them with live ground beetle larvae ($n = 11$) or adults ($n = 13$) tethered to a string. The responses were similar, but the females tended to present their dorsum to the intruder more often, keeping the eggs under the abdomen, suspended on the stylets. When the disturbance became too intense, they fled with the eggs. The broad and hard dorsum of the female's body is apparently a good defense against predation, and when that fails, escape with the eggs, or running off to distract and attract the predator when nymphs are in the nest were the most prevalent responses.

4.7 Kin Recognition and Nest Fidelity

We verified the earlier finding that females cannot distinguish their own eggs (Table 4.2; Filippi-Tsukamoto et al. 1995). Females exposed to three egg masses, one of which was her own, chose the one that was nearest. We also removed the egg

Table 4.2 Female discrimination of eggs

| | Number of times that female chose own egg mass | | | |
	0	1	2	3
Observed	2	4	3	1
Expected	2.96	4.44	2.22	0.37

$n = 10$; $\chi^2 = 1702$, $p > 0.05$
Adapted from Filippi-Tsukamoto et al. (1995)

masses from three females in the field, and each of the three tried to steal the egg masses from other nesting females, clearly not caring whether they were her own, just that she knew she was supposed to be guarding an egg mass. Again, it is reasonable that the ability to distinguish their own eggs from those of conspecifics has likely not evolved because females rarely leave the egg mass unattended.

We know that females cannot distinguish their own eggs, but they progressively provision nests, which means they must repeatedly leave and return to the nest. Therefore, we needed to verify that females were not simply entering any nest but that they could distinguish their own nymphs or nest. We observed the behavior of 17 provisioning females to verify whether they displayed nest fidelity; in other words, were females only provisioning their own offspring? Thus, in this study, nests were labeled, and the females in each nest were individually marked with liquid paint markers. Because we wanted to manipulate some nests to ascertain whether females possibly use chemical cues to find their nest, ten gravid females were allowed to nest on black mesh plastic gardening trays (56 cm L × 36 cm W × 8 cm H) that had been piled with soil and leaf litter similar to the conditions of the surrounding area. The trays were set in the field about one meter distant from each other. We also labeled nests and females in seven naturally occurring nests in the field. We observed the nests nearly daily throughout the reproductive season and documented the presence or absence of the female in the nest and, when a female was there, whether she was in her own nest. Females were sometimes absent from the nest, probably out foraging, and then were back in the nest at a later observation (Fig. 4.6; Filippi-Tsukamoto et al. 1995). In this study, we never observed a female in a nest that was not her own; thus, females have at least nest fidelity. However, in 1993, on a check of 18 natural nests being observed, 2 females were in a nest. The owner of the nest was one of the two. The other female appeared to be feeding on a drupe in the nest. See Sect. 6.4 for more about what we learned regarding this behavior.

To determine whether females were using chemical cues to locate their nest, rather than only other cues, such as celestial and magnetic, we also removed four females from their nests and set them on the ground about a meter away from the nest. While they were out of their nests, we switched the nests around, each one about one meter away from the original site, by moving the entire tray that the nests were in. By the next morning, three of the four females were in the correct nest at the new location, and the fourth one was never seen again (Filippi-Tsukamoto et al. 1995). It seems there is a chemical cue that the females use to recognize their own nest or nymphs that is effective from at least one meter away. See Chap. 6 for exciting information on how females successfully navigate the very long and rambling routes they take while provisioning.
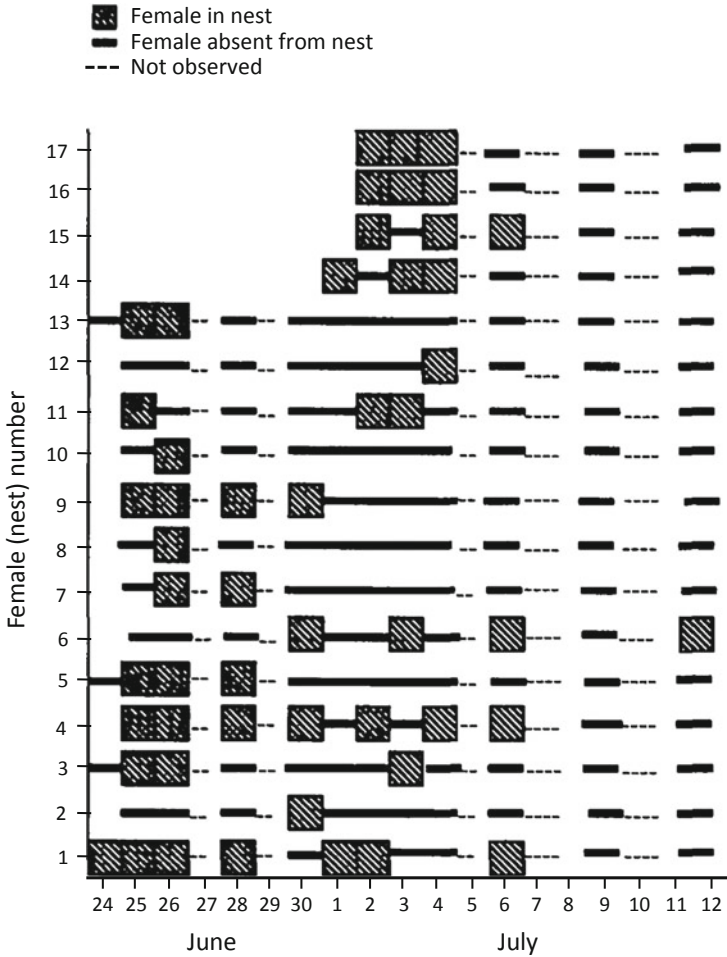
Fig. 4.6 Female nest fidelity. Females frequently left the nests to forage for drupes. Nests were observed for the presence (shaded squares) or absence (bold lines) of a female, and verification that when a female was present, it was the owner of the nest. Observations of a nest began on the date that the first icon is seen. Dotted lines indicate days of no observation. Adapted from Filippi-Tsukamoto et al. (1995)

4.8 Provisioning Behavior

Provisioning a food resource that the offspring could access soon after hatch (Wood 1976), either by preparing a substrate for feeding, such as mass provisioning a dung ball and laying eggs in it for the hatchlings to feed on in a single effort is the next level of care beyond guarding (Brussard and Visser 1987; Hunt and Simmons 1998).

A parent might also excrete substances as food (Becqueart 1935) or regurgitate substances that facilitate feeding for altricial young.

Progressive provisioning is a much rarer behavior in the insects. Burying beetle parents prepare carrion as a single mass for hatchlings to feed on, but they also repeatedly express partially digested carrion into the mouths of offspring (Eggert and Sakaluk 1995; Trumbo 1996, 2012). In the Reduviidae, males repeatedly transport prey items to their young (Ralston 1977; Machado and Trumbo 2018). Among phytophagous heteropterans, progressive provisioning occurs in *P. japonensis* and members of a related cluster of genera within the Sehirinae, a subfamily of the Cydnidae. Seeds or fruits are transported repeatedly to the nest either during the egg guarding phase and after hatch (*Adomerus triguttulus*, Nakahira 1992, 1994; *Sehirus cinctus*, Sites and McPherson 1982; Kight 1997) or after hatch only (*Adomerus rotundus*, Inadomi et al. 2014; *Canthophorus niveimarginatus*, Filippi et al. 2009; *Adomerus variegatus*, Mukai et al. 2010; and *Parastrachia japonensis*).

Of 43 nests with females tending an egg mass that were initially found and marked in 1993, only 18 could be observed through the early nymphal stage. Three of the females that had disappeared were later seen foraging for drupes in the foraging area, so we assumed they had relocated their nests during the egg stage. We never saw any of the other 22 females again. Provisioning females set out on foraging forays soon after hatch. It appeared that first instars were feeding on the eggshells, but, *more on that* in Sect. 4.17. Females left the nest and roamed for hours at a time inspecting drupes, and rejecting most, before returning to the nest. Frequently a female would return without a drupe, stay in the nest for a short time, and then head out again in search of a drupe. Nymphs often climbed onto the mother's dorsum and probed it with their proboscis, apparently in an attempt to feed on her. Females responded by rocking their bodies from side to side to shake them off. As we will see later in this section, at some point the females lost the ability to shake them off!

One foraging female was tracked for 2 h, during which time she covered 30 meters while inspecting ten drupes before finally accepting one for transport back to the nest. Another female that we observed for 2 h on each of 3 consecutive days spent between 15 min and 2 h before finding a drupe that was acceptable. Like all other females, once the drupe was taken up on the stylets, she zipped in a straight beeline directly back to her nest. As soon as the female deposited the drupe in the nest, the young gathered on it and fed, while the mother stood guard. She invariably stayed only about 5 min before taking off on another foraging foray. Often, nymphs would climb on her dorsum and attempt to feed on her. What was particularly interesting was that, while we observed her behavior for 3 days, on days 2 and 3 she continued to provision the nest, although the nymphs had already departed! In fact, in 5 of the 18 nests that had been observed up to the third stadium, drupes continued to increase in number for 1–3 days after the nymphs had abandoned the nest (Filippi-Tsukamoto et al. 1995)! In this obligate semelparous species, it appears that a switch is turned on at hatch taking the mother out of egg guarding mode and bringing her into provisioning mode. There apparently is no turn off switch to the provisioning mode. We frequently also saw mothers that had died either near their nest or somewhere along

the path of their arduous provisioning task being fed on by aggregates of nymphs. We surmised that females just keep provisioning until they die because there is no question of conserving resources for future reproductive efforts. If they die near their nest, their body will continue to provide sustenance to their offspring, but we expect that many females die far from the nest, after giving their all to the provisioning effort, and are fed on by the offspring of other females.

4.9   Variation in Number of Provisioned Drupes and Nymphal Nesting Stage

One other interesting finding that surely could have some evolutionary import is that some females are better provisioners than others. There was considerable variation in the final number of drupes that females provisioned and this was positively correlated with the duration of the nymphal nesting stage (Fig. 4.7; Filippi-Tsukamoto et al. 1995). In 1993, among the 18 nests monitored until the nymphs left, the final number of drupes provisioned to nests ranged from 0 to 40 (mean ± S.D.,
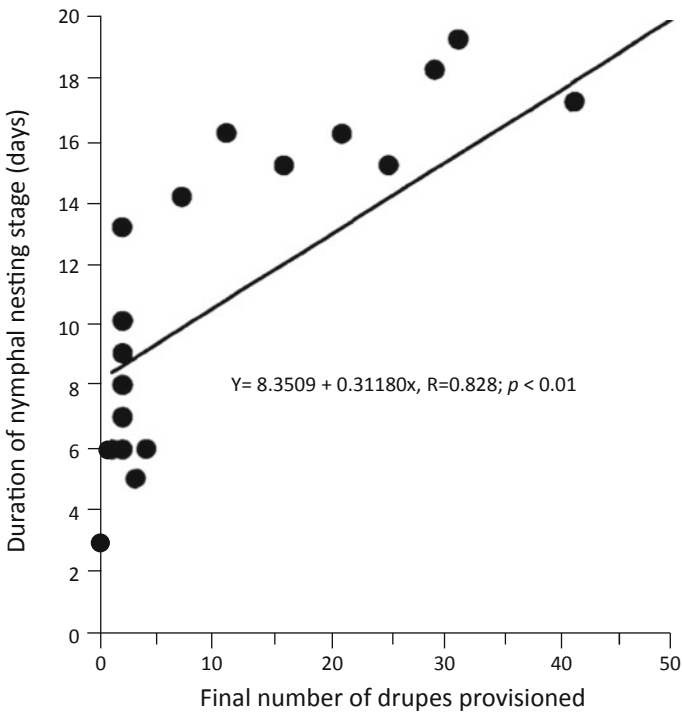

Fig. 4.7 Impact of the number of drupes provisioned on the duration of the nymphal nesting stage. Adapted from Filippi-Tsukamoto et al. (1995)

Y= 8.3509 + 0.31180x, R=0.828; $p < 0.01$

10.3 ± 12.7). Good drupes seem to fall in patches under the host tree, and finding a good patch was like striking gold. The female would return to the site repeatedly and bring lots of good drupes back to the nest. Some females apparently did not come across a good patch or might just have been bad foragers. We witnessed just such a situation. As diligent at searching as the poor provider was, she was only able to secure a few drupes. On the other hand, the female in a nest about 30 cm away (most nests are not this close together) was a much better provider. The poor provider returned to her nest one day to find that her offspring had moved over to the neighbor's nest. The neighbor ended up feeding twice as many young, and the original mother kept provisioning, albeit poorly, a nest with no young!

The variation in the number of drupes provided is even more stark among years. In 1994, the year represented in Fig. 4.5, only 8 of 11 nests contained any drupes at all; 7 of the 8 nests contained only 1 or 2 drupes, while the remaining nest contained 28! On the other hand, in 1992, many nests contained nearly 100 drupes, and 1 held the record thus far at a remarkable 155 drupes! As shown in Fig. 1.10, there is considerable variation in the abundance of drupes overall from year to year and, importantly, in the abundance of drupes that are acceptable to provisioning females. As will be demonstrated in Sect. 4.19, there is also considerable variation in the number of nesting females from year to year, and the intensity of inclement weather, and these factors also contribute to the success of provisioning females.

The duration of the nymphal nesting stage of the 18 nests monitored in 1993 also showed considerable variation, ranging from 5 to 19 days (mean ± S.D., 11.6 ± 4.8), and there was a positive correlation between the number of drupes provisioned and the duration that nymphs remained in the nest (Fig. 4.7; Filippi-Tsukamoto et al. 1995). Like the variation seen for provisioned drupe numbers over the years, variation in the average duration of the nesting stage was also great, and with the third stadium, which occurs at about 10 days after hatch, being the one that is correlated with young safely reaching the foraging area and feeding with nymphs from other nests, we considered nesting durations shorter than 10 days as being unsuccessful. In 1993, 10 out of the original 43 (23%) nests contained nymphs up to that point. In 1994, only 11/66 (16.7%) nests did so. On the other hand, in 1992, when so many drupes were provisioned, there were nymphs in nests that had reached the fifth instar. Within years and between years, variations seen in provisioning abundance and nesting duration are stark and suggest that several factors, including resource abundance, predator pressure, intraspecific competition, and weather extremes, were at play. We explore these possibilities in Sect. 4.19.

4.10 Effects of Drupes and Female Presence

The females were clearly providing food for the young, but we were interested in seeing separately how the presence of drupes and a tending female affected the behavior and survivorship of nymphs and also how the female's tending behavior and survivorship might change with the presence or absence of drupes. We could not
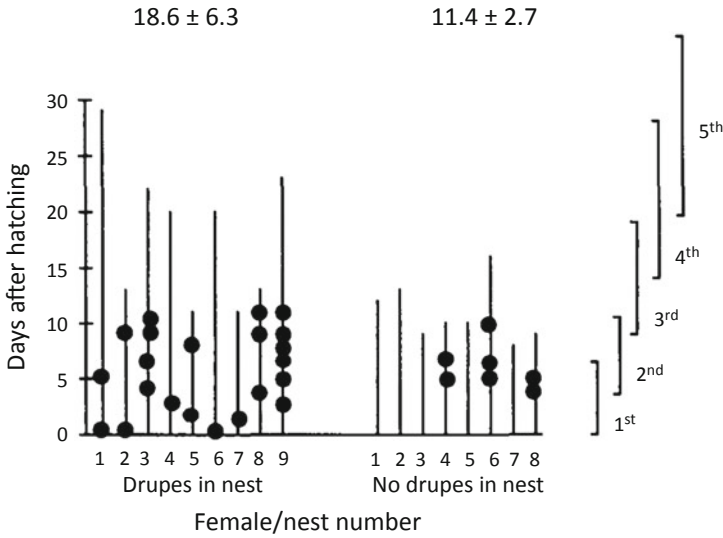
Fig. 4.8 The effect of drupe presence or absence in the nest on female behavior and life span. Life span of each female is represented by the vertical lines; black dots indicate female was in the nest at the time of observation. For comparison of female attendance in nest, Mann-Whitney U test, $p < 0.01$; for life span, Student t-test, $p < 0.05$. Adapted from Filippi-Tsukamoto et al. (1995)

control for predation effect with natural nests, so we carried out a semi-manipulated study by enclosing 40 gravid females in cylindrical chicken wire cages (40 h × 30 D cm) that were then covered with black netting and fastened at the top. The enclosures were set 8 cm into the soil at Mt. Hinokuma, which effectively excluded the worst predators, and were layered with the soil and litter that the females typically nest in. Females oviposited in the cages (no food provided, as they do not feed at this stage in the field), and the nest/cages were divided into four conditions upon hatch: female + nymphs + drupes in the nest, nymphs + drupes in the nest, female + nymphs, no drupes in the nest, and nymphs without a female or drupes. We observed the nests daily to see if the female was guarding her eggs/ nymphs and monitored survivorship of the females and the nymphs. Some of the nests failed very early on and were not used in the study. In the two groups where females were present, females were observed in the nest 20.3% of the times checked in the group that had drupes ($n = 9$), but only 7.3% of the times checked when no drupes were in the nest ($n = 8$) (Fig. 4.8; Filippi-Tsukamoto et al. 1995). This suggests that females are more inclined to invest time guarding their nymphs when ample drupes are available but likely spend more time out searching for drupes when they are scarce, which would leave the nymphs vulnerable to predators for longer periods of time. Extrapolating this finding to the natural field, the data also suggest that nymphs would be more vulnerable to predation in years when drupes are scarce because females would be away from the nest for longer periods of time as they searched for drupes. Female life span was also significantly longer when drupes were
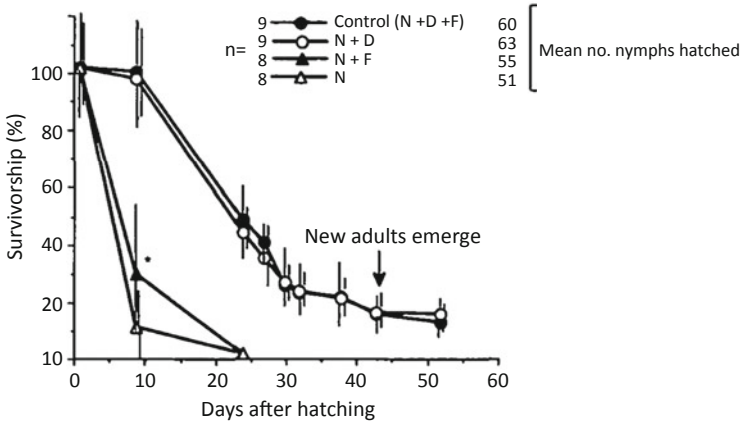
Fig. 4.9 Survival rate of nymphs under different nesting conditions. *N* nymphs; *D* drupes present; *F* female present. *Significant difference at day 10 between nymphs with and without the presence of mother, in the absence of drupes; Student's t-test, $p < 0.05$. Adapted from Filippi-Tsukamoto et al. (1995)

present in the nest, indicating that provisioning has a cost in reduced survivorship (Fig. 4.8; mean days ± S.D., 18.6 ± 6.3 with drupes, 11.4 ± 2.7 without drupes; Filippi-Tsukamoto et al. 1995). Though we have never seen a female feeding in the nest, the shorter female life span in the no-drupe group suggests that they do feed while out foraging for drupes.

Nymphs also had a greater tendency to remain aggregated when drupes or a female were present (70% of observations), and this tendency was enhanced when both drupes and a female were present (82% of observations) (Filippi-Tsukamoto et al. 1995). Feeding is more efficient in aggregations, desiccation is less of a problem, and certainly aggregations keep one safer from predators (dilution effect). Not surprisingly, survivorship of nymphs was also affected by the presence or absence of drupes in the nest (Fig. 4.9).

There was no difference in the survival rate of nymphs in the presence and absence of a tending female as long as food was provided (Fig. 4.9), and the body length of both males and females was the same in both groups (Filippi-Tsukamoto et al. 1995), suggesting that the female is not treating the drupes to facilitate feeding by the nymphs. However, these nymphs were protected from predators. This finding indicated that when nymphs had ready access to food and predators were excluded, they could manage to feed and survive. Interestingly, at day 10, nymphs with a tending female had significantly greater survivorship than those without a tending female (Fig. 4.9; Filippi-Tsukamoto et al. 1995). In the foraging area, cannibalism is rampant, and we suspect that females were mitigating cannibalism among the hungry nymphs.

4.11 Conclusions from This Study

So, what did we glean from these studies? We confirmed that females cannot distinguish their eggs from those of conspecific females (Table 4.2) but distinguish their nymph-containing nest from other nests (Fig. 4.6). We confirmed that females do not feed while incubating their eggs and found that they display several defensive behaviors to would-be predators (carabid ground beetle) (Table 4.1). Once nymphs hatch, females trade-off between guarding and provisioning behaviors. They repeatedly leave the nest to search for drupes but cannot always find an acceptable one, verifying that the resource is constrained. Provisioning provides nymphs with access to high-quality drupes that would be difficult to access (Figs. 4.8 and 4.9), particularly because most nests are far from the foraging area. Females never provision other nests but sometimes unwittingly provision nymphs that intrude into their nest. There is great variation in the number of drupes provisioned among females and among years, likely due to resource availability, competition with other females, and inclement weather. Duration of the nymphal nesting period is positively correlated with the number of drupes a female provides (Fig. 4.7). In this way, progressive provisioning is likely to protect nymphs from predation. Nymphs will abandon the nest if provisioning is too low (Fig. 4.8). Females continue to provision until they die, even after the nymphs have left the nest. Females invest more in guarding at the nest when drupes are abundant, and nymphs remain aggregated more when a female and drupes are present in the nest. Finally, females apparently mitigate cannibalism in the nest. The next set of studies sought to test some of these conclusions.

4.12 Testing the Speculated Functions of Progressive Provisioning

We will take a detour from the chronological presentation of our journey of discovery to maintain the continuity of this part of the story: the adaptive value of progressive provisioning. In Chap. 6, when we get into answering the proximal mechanistic and developmental "whys," we present a detailed description of how we came to understand why females spend so much time inspecting a variety of drupes before selecting one (Nomakuchi et al. 1998), but in order to maintain the flow of our story, suffice it to say here that we discovered most drupes on the ground, regardless of how enticing they appear (bright red, purple) do not contain much endosperm at all, which is the part of the drupe that *P. japonensis* feeds on, and provisioning females were choosing those drupes that contained a hefty amount of endosperm. However, knowing this does not necessarily mean that nymphs really require females to bring them good food, or even that it enhances nymphal success. In fact, as demonstrated in the last series of studies, a number of females, albeit a small percentage, actually nest where the drupes are scattered on the ground and are presumably accessible to nymphs after a bit of hunting. Yet, females in those nests

also provisioned them. So, we tested the effect of provisioning from the resource constraint hypothesis. We also knew that ground beetles were predators and concluded that the mother's defensive behaviors and provisioning behavior, which kept the vulnerable nymphs in the safety of the nest longer, were adaptive but had not tested it. In the reproductive seasons of 1995 and 1996, we set out to test these hypotheses.

4.12.1 Testing the Resource Constraint Hypothesis

We tested the hypothesis that, because the resource is of such poor quality, progressive provisioning is essential for nymphs to acquire sufficient nutrients, even when the nest is situated among the scattered drupes of the average (typical) quality in the field. In 1995, there were very few nesting females to be found, but we managed to collect 13 females with an egg mass. To exclude the potential effect of predators, we prepared square cages made of a PVC pipe frame ($50 \, \text{cm}^2 \times 20 \, \text{cm h}$), covered on all sides except the bottom with black mesh. A square opening ($10 \, \text{cm}^2$) at the top, fastened with Velcro, allowed access to the inside of the cage. The cages were set ~7 cm into the ground in a gently sloped area in the forest at Mt. Hinokuma that had been cleared of groundcover (~10 m^2) (Filippi et al. 2000). We monitored the number and quality of drupes that were falling from the host tree that year using a seed trap made by forming a 1 m^2 section of black mesh into a triangle and suspending it on three 1.2 m garden poles. The seed trap was set under the crown of the main host tree for that area, and drupes that fell into it were counted and their quality assessed on a weekly basis. We used this information to determine the number and type of drupes to seed the cages with. We divided the cages into two groups, one that was provided with typical drupes that were falling in the area and one that was provided with only high-quality drupes. Neither of the groups had the drupes placed inside the nest. Both had them scattered about outside the nest, as if the nest were situated under the host tree, in the same density that was occurring under the host tree. To eliminate the possibility that the female is facilitating feeding or providing some other assistance in the nest, females were removed at hatch. Based on the data from the seed traps, we gradually increased the number of drupes in each cage to 25 by day 15 after hatch. We monitored the development rate and the survivorship of offspring in the two groups.

Drupe Availability

In 1995, the date that good drupes began falling was much later than the date that females were seen out in the foraging area searching for drupes to provision their nests with (Fig. 4.10; extrapolated from Filippi et al. 2002). There were large fluctuations in the numbers of drupes collected, likely due to heavy rains that occurred during the rainy season, which typically lasts throughout most of the
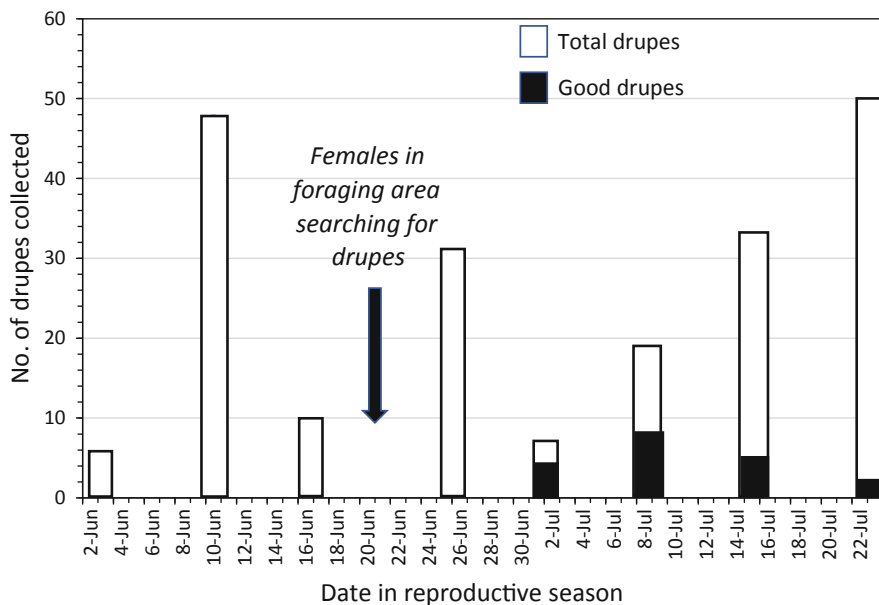
Fig. 4.10 Drupe abundance at the experimental site in 1995. Based on data presented in Filippi et al. (2002)

reproductive season, and a very low percentage of those drupes were good, ranging from 0 to 57%. Clearly in 1995, resource was poor (see Fig. 1.10 for comparison of drupe quality over 4 years).

Development Rate

The behavior of nymphs in the two groups differed considerably. Nymphs in the good drupe groups were seen outside the nest feeding on drupes in aggregations from day 2 after hatch, while those in the typical drupe group were scattered about the cage on day 2, with a few individuals on each of several different drupes. Successful feeding by nymphs requires that they aggregate in large numbers on a drupe and simultaneously inject the enzyme that liquefies the stony endosperm (Tachikawa and Schaefer 1985; Tsukamoto and Tojo 1992), so it is unlikely that feeding in that dispersed way would be successful. There was a significant difference in the development rate between the two groups. The duration of the first and second stadia was shorter for nymphs in the good drupe group ($n = 7$), and they molted to the second stadium significantly earlier than those in the typical drupe group ($n = 6$) (Fig. 4.11a; Mann-Whitney U test, $p = 0.001$; Filippi et al. 2000).
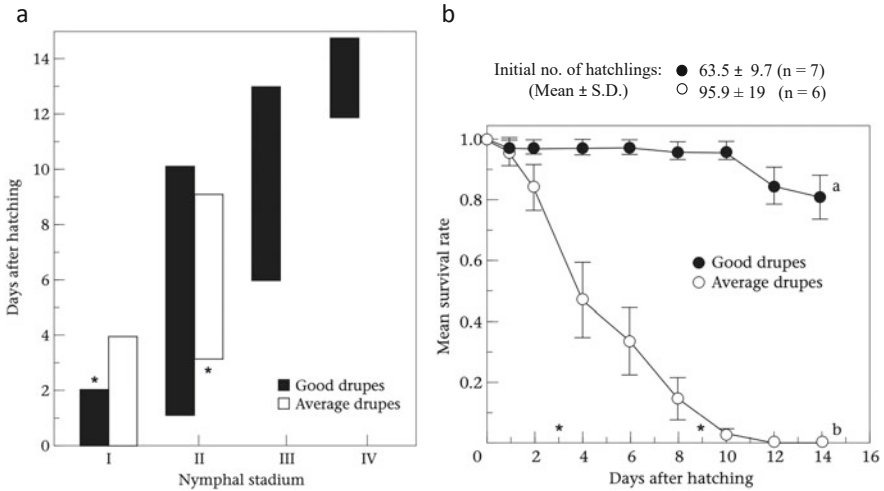
Fig. 4.11 Impact of drupe quality on success of *P. japonensis* offspring. (**a**) Development rate. *, Mann-Whitney U test, $p = 0.001$; (**b**) survivorship. *, Significant difference from days 2–4 and 8–10, $p = 0.012$ and 0.047, respectively. Different letters indicate significant difference, Mann-Whitney U test, $p = 0.003$. Adapted from Filippi et al. (2000)

Survivorship

There was also a significant difference in survivorship between the two groups between days 2–4 and 8–10 (Mann-Whitney U test, $p = 0.012$ and 0.047, respectively), and at day 14, by which time all nymphs in the average drupe group had died ($p = 0.003$; Fig. 4.11b; Filippi et al. 2000). Clearly, the data support the resource constraint hypothesis. The quality of most drupes is typically so poor that provisioning not only enhances offspring success but also is essential, *at least* when drupe quality and abundance are similar to that encountered in 1995. We predict that the data would look quite different in years when the proportion of good drupes is very high and their availability coincides with the presence of foraging females.

4.12.2   Testing the Predation Hypothesis

Using cages like the ones used in the previous experiments, in 1996 we tested the hypothesis that provisioning mitigates predation risk to nymphs. We set up 25 cages at the same site that was used in 1995. The cages were divided into five groups ($n = 5$ per group) based on whether a female was present, whether a predator (ground beetle, *Apotomopterus dehaanii*) was present, and whether the drupes, *all good drupes*, were placed inside the nest or scattered around the outside of the nest, as done for the previous experiment (Table 4.3; Filippi et al. 2000). One cohort from each group except Group D failed very early on from death of the female or loss to

Table 4.3 Experimental design for test of Predation Hypothesis

Group (n)	No. of nymphs Mean ± S.D.	Female	Predator	Drupes
A (4)	63.3 ± 3.3	Present	Absent	Outside nest
B (4)	33.0 ± 10.5	Present	Present	Outside nest
C (4)	55.4 ± 15.3	Absent	Present	Inside nest
D (5)	45.0 ± 17.1	Absent	Present	Outside nest
E (4)	45.3 ± 7.0	Absent	Absent	Outside nest

Adapted from Filippi et al. (2000)

flooding, so all groups but D had four cohorts; D had five cohorts. Survivorship of nymphs was assessed over the first 10 days of the nesting period, after which young are typically in the much less vulnerable third stadium and begin moving on to the foraging area.

The survival curves of Groups A, B, and C were similar until day 6 with a survival rate of 70% (Fig. 4.12a; Filippi et al. 2000). Groups A and B had a female present, but group B also had a predator. Good drupes were scattered outside the nest in both groups. There was no difference in survivorship between these two groups throughout, indicating that female presence effectively mitigates predator threat beyond the provisioning of drupes, which keeps the young in the nest. Group C was provisioned by the researchers, which kept the nymphs safely aggregated in the nest, but in the absence of a tending female, the predator, enclosed in the cage, was eventually able to find the nymphs. By day 10, the survivorship in Group C was significantly lower than that of Groups A and B, reduced to 20%, which was about 30% lower than Groups A and B (comparison between Groups A and C, ANOVA, $p = 0.0005$, Bonferroni $p = 0.01$, and between Groups B and C, ANOVA, $p = 0.0005$, Bonferroni $p = 0.006$).

We examined the effect on survivorship of the mother's presence (by excluding her) in terms of food access and protection from predators. We compared Groups D and E, neither of which had a mother present, both of which had drupes outside the nest, but Group D had a predator, and Group E did not. By day 1, the survivorship of Group D was significantly lower (20%) than that of Group E (ANOVA, $p = 0.013$, Bonferroni, $p = 0.009$; Fig. 4.12b). By day 10, the difference in survivorship between the two groups was 42% (ANOVA, $p = 0.0005$, Bonferroni, $p = 0.0001$).

Finally, to confirm the mother's role in protecting young from predation, we compared the effect on survivorship of young in nests without a female but with a predator present when drupes were inside the nest (provisioned, Group C) or scattered outside (Group D). By day 2, the group with drupes outside the nest had significantly (33%) lower survivorship than the group that had been artificially provisioned. However, by day 6, the ground beetle had clearly found the nest, and the survivorship began to decrease markedly, with no difference in survivorship between the two groups by day 10. Clearly, the role of the female in protecting young from predators is significant, even when they do not have to leave the nest to feed.

Fig. 4.12 Survival curves of provisioned and unprovisioned nymphs in the presence or absence of a predator and/or a female. (**a**) Comparison of groups A, B, and C. Different letters indicate significant differences in final survival rate at $P < 0.01$ in pairwise comparisons by a one-way ANOVA with a post hoc Bonferroni adjustment for multiple comparisons. (**b**) Comparison of groups D and E. *Mortality rate for the interval showed significant difference at $P = 0.008$; †marginally significant difference at $P = 0.019$ by pairwise comparisons using a one-way ANOVA with a post hoc Bonferroni adjustment for multiple comparisons. Different letters indicate significant difference in final survival rate at $P = 0.0001$. (**c**) Comparison of groups C. *Significant difference in mortality rate for the interval at $P = 0.002$ by a one-way ANOVA with a post hoc Bonferroni adjustment for multiple comparisons. Error bars indicate SE. Adapted from Filippi et al. (2000)
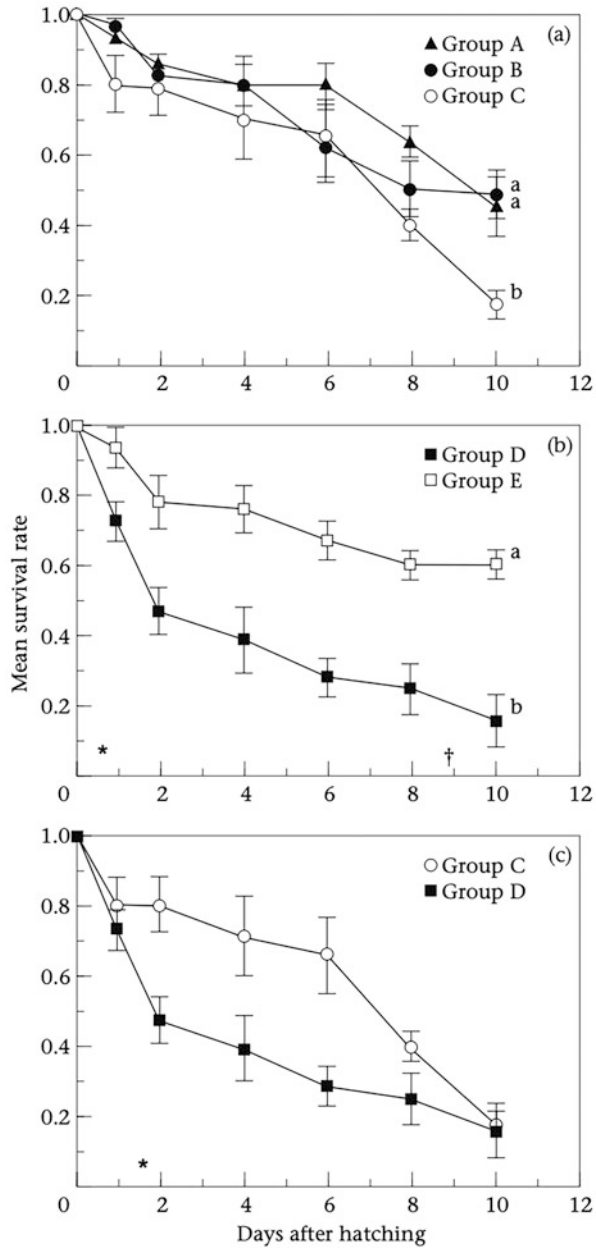
4.13 Conclusions from These Studies

Two assumptions about the adaptive significance of progressive provisioning were tested and resolved. The hypothesis that females provision because the resource is constrained was supported. Drupe availability is ephemeral, and, when available, it is scarce (Fig. 4.10). Moreover, as was the situation in 1995, the availability of resource does not always coincide with the timing of need. These resource constraints should strongly select for subsocial behavior, such as progressive provisioning, so that young might receive better access to the poor food resource (Tallamy and Wood 1986; Eggert et al. 1998). In the condition simulating a nest situated right in the middle of the foraging area, development and survivorship of the nymphs were high when only good drupes were scattered outside the nest but decreased significantly when nymphs were forced to find good drupes on their own from among the many drupes scattered on the ground (Fig. 4.11), verifying the essential role of a progressively provisioning mother to give young access to the scarce good drupes.

The predation hypothesis was also supported. Even with good drupes in the nest, in the absence of a tending female, young eventually succumbed to predation (Fig. 4.12). As long as a provisioning female was present, survivorship in the presence or absence of a predator was the same but decreased significantly when the female was absent (Fig. 4.12a). Further, as long as no predator was present, survivorship was high even if the *good* drupes were only scattered about outside the nest (Fig. 4.12b), but with a predator present and no female to protect the young even when drupes were placed inside the nest, the predator eventually found the young (Fig. 4.12c). The findings that parental care mitigates predator pressure in *P. japonensis* are consistent with findings in numerous other species (Wilson 1971; Tallamy and Wood 1986; Kudo et al. 1989; Wyatt and Foster 1989; Diesel 1992; Royle et al. 2012). These studies demonstrated that progressive provisioning provides both essential access to the food resource and vital protection from predation.

4.14 Predation Risk to Nymphs of Different Stages

Knowing that *P. japonensis* is preyed upon by the ground beetle and that there is variation in the stadium that nymphs vacate the nest left us with another question that begged to be answered. The assumption was that younger nymphs would be more vulnerable to predation, and we know that staying in the nest with mom keeps them safe, yet they do emerge at all different stages and head to the foraging area. In 1998 we tested whether nymphs of different stages had different levels of vulnerability to predators. To understand the natural pattern of when nymphs relocate to the foraging area, we counted the numbers of nymphs in each stadium in a 42 m^2 quadrat of 1 m^2 plots in the foraging area daily from June 15 to July 9. Although, as previously noted (Filippi-Tsukamoto et al. 1995), the third stadium seemed to be the time that nymphs
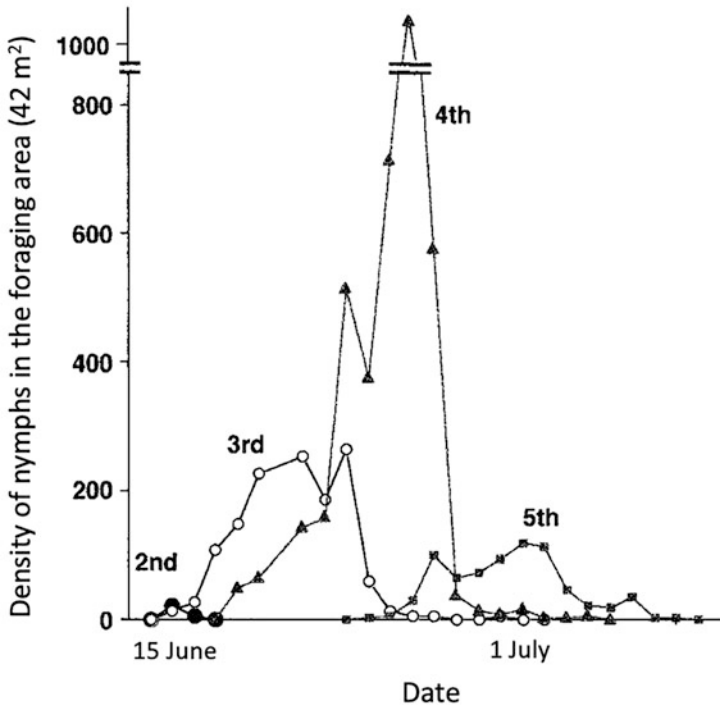
Fig. 4.13 Nymphal occurrence pattern in the foraging area. Adapted from Nomakuchi et al. (2001)

started leaving the nest, 3.6 times as many of the nymphs on the ground were fourth stadium compared to third (Fig. 4.13; Nomakuchi et al. 2001) indicating that most stay in the nest until the end of third stadium. We were surprised to note that even second instars were seen in the foraging area, though in very small numbers. We suspect these are the offspring of poor provisioners and that the variation in stadia that relocate to the foraging area is also related to the provisioning success of their mothers.

The very low number of fifth instars represented in the graph is likely related to the fact that we only counted individuals in the quadrat, and fifth instars tended to spend a considerable amount of time (~ 10 days or so) in aggregations on the lower trunks/branches of trees adjacent to the foraging area and were not counted.

We anticipated that offspring would leave the nest and relocate to the foraging area at a stadium that is less vulnerable to predation. Therefore, we tested the hypothesis that stadia occurring in high abundance in the foraging area would be less vulnerable to predation. We set up 12 square cages ($50 \times 50 \times 25$ cm^2) covered in fine black mesh in the field as described for the previous studies. To simulate the conditions of the foraging area where leaf litter and ground cover is sparse, a small pile of leaves was set in one corner of the cage where nymphs could hide, and the rest of the cage was left largely bare. Drupes were scattered on the bottom of the cage in the density that occurred under the host tree; however, to minimize the confounding
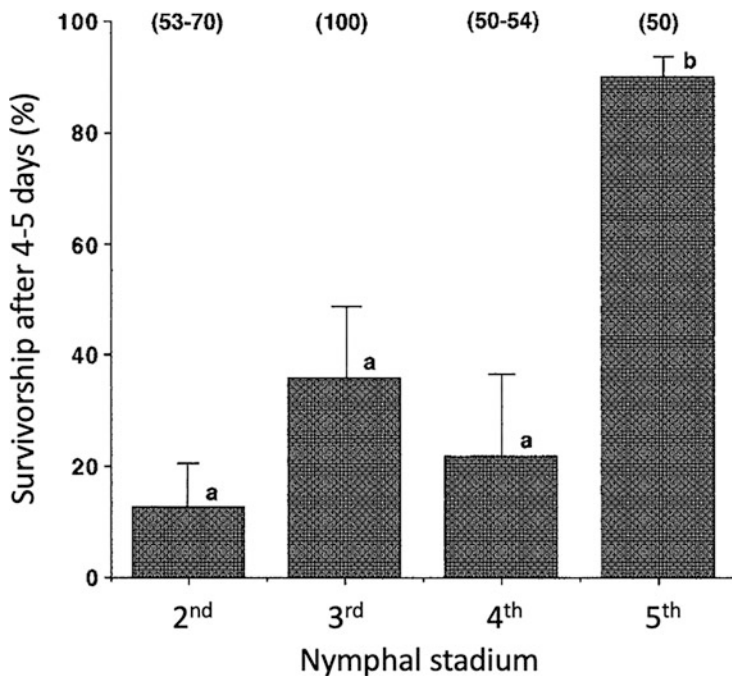
Fig. 4.14 Survivorship of *P. japonensis* nymphs at different nymphal stadia in the presence of a predator, the carabid ground beetle, *Apotomopterus dehaanii*. Adapted from Nomakuchi et al. (2001)

variable that typical (poor) drupes would have on survivorship, only good drupes were used. A nest containing cohorts of second, third, or fourth stadium nymphs was placed under the leaf litter in the corner of each cage, and one ground beetle (*Apotomopterus dehaanii*) was released into each cage. Three replicates were done for each nymphal stadium, and survivorship was assessed after 4–5 days. Although fourth stadium nymphs were most abundant on the ground in the foraging area, there was no difference in survivorship between second, third, and fourth stadium nymphs; only fifth stadium nymphs had significantly higher survivorship (Fig. 4.14; ANOVA, $p = 0.004$, Bonferroni, $p < 0.05$; Nomakuchi et al. 2001).

The optimal duration of the nesting stage of immature insects with regard to predation will vary with the degree of mobility of the young at different stages, in other words, their ability to evade predators. The young of several subsocial holometabolous species with parental care, such as the burying beetle, *Necrophorus* (Eggert and Müller 1997), and the dung beetles, *Copris*, *Synapsis*, and *Heliocopris* (Halffter 1997), often leave the nest just before pupation or after eclosing as adults because the larval stage has far less mobility than the adult stage, and so is much more vulnerable to predation.

The pattern of nymphal occurrence in the foraging area is a manifestation of both direct and indirect effects of predation (Nomakuchi et al. 2001). In other words, there

is predation pressure acting directly on the independent nymphs which results in the observed distribution pattern of nymphs in the different nymphal stadia. Because the data showed that second–fourth stadium nymphs have equal predation pressure, the earlier a nymph leaves the nest, the longer it is exposed to the predation pressure and the less likely it is to survive. In fact, the actual number of nymphs that we were able to count might be far less than the numbers that emerged, because many more of the youngest, most vulnerable ones could have been eaten.

The indirect predation effect would occur over evolutionary time and would form the distribution of nymphal stadia at the time of independence. This effect would work primarily on female fitness traits, specifically provisioning behavior in this case, because if predation pressure on younger nymphs outside of the nest is higher than that inside the nest, selection should favor a pattern of parental care that results in nymphs becoming independent at a later stadium or even at the adult stage. In fact, the survival rate of *P. japonensis* nymphs *is* higher inside a nest than outside (Filippi et al. 2000), and there is a positive correlation between the number of drupes provisioned and the duration of the nymphal nesting stage (Filippi-Tsukamoto et al. 1995), so we would expect selection to favor prolonged provisioning in this semelparous species. However, the intense constraints on resource availability (Fig. 1.10; Filippi et al. 2002) confound female efforts and success at provisioning.

4.15 Impact of Female Provisioning Capacity on Duration of Nymphal Stage

While the interactions between parents and offspring have long been a focus of parental care studies (Clutton-Brock 1991; Mock and Parker 1997), interest in the evolutionary basis of parental care has shifted from a static concept focusing on optimal behaviors from the parent's perspective (Lack 1947) to a more dynamic understanding that a conflict must exist between parents and offspring and that conflict will determine the manner and duration of parental care (Godfray 1995). Subsocial behavior in invertebrates has been the focus of numerous studies (Choe and Crespi 1997), but most often from the perspective of parental optimality. Few studies have examined the conflict from the perspective of offspring response to the parental care they receive (Wyatt and Foster 1989; Agrawal et al. 2001).

Typically, when a parent provides care, in particular, supplies food, to offspring, a conflict arises between parent and offspring because for the parent the optimal timing for offspring transition to feeding independence comes earlier than that of offspring, who want to stay in the nest and receive food longer (Trivers 1974; Leigh and Smiseth 2012). However, in the case of *P. japonensis*, one can almost consider that the conflict runs in the reverse direction. Because early departure from the nest leads to dramatically increased predation risk (Filippi et al. 2000; Nomakuchi et al. 2001), and, importantly, females have no option of future reproductive success, female fitness would be optimized by provisioning enough food to keep the young in the

nest until the adult stage as the nymphs should leave the nest when food becomes insufficient. Some hungry young insects like larvae of the burying beetle (Eggert and Müller 1997) and dung beetles (Halffter 1997; Sato 1997) remain in the "nest" until the adult stage, completely dependent on food being provisioned by the parent, and larval begging results in delivery of more food (Smiseth and Moore 2002). The behavior described earlier of *P. japonensis* offspring attempting to feed on the mother in the nest (Filippi et al. 2000) effectively acts as begging behavior because the female aggressively rocks her body to shake the young off and makes a chattering alarm call and then quickly leaves the nest to forage once again. However, as we have discussed (and will explain in detail in Chap. 6), good drupes are scarce and hard to find, and foraging mothers, notwithstanding their diligent efforts, aren't always successful. So how does provisioning capacity by *P. japonensis* mothers relate to duration of the nymphal nesting period?

Although we observed in an earlier study that there was a positive correlation between the number of drupes provisioned and the duration of the nymphal nesting period, a correlation, of course, is not cause and effect. Other factors, for example, safety against predators, could be keeping the nymphs in the nest. To sort this out, we decided to test the hypothesis that the provisioning capacity of females was directly responsible for the longer nymphal nesting period. Between 1996 and 2000, we carried out three field studies from mid-June through late July to (1) determine the beginning of nest departure (1996); (2) examine the relationship between the duration of the nesting period, i.e., from hatching to departure from the nest and the provisioning effort of the mother (1998); and (3) examine the rate of nymphal accession to the foraging site after departing from the nest (2000). All studies were carried out at our original field site in Mt. Hinokuma Prefectural Park in Kanzaki town, Saga, Japan.

4.15.1 Experiment 1: Determining the Timing of Nymphal Independence

To determine the point at which departure from the nest ensues, in 1996 we constructed a 50 cm^2, open-bottom mesh-covered nest cage as described in the earlier studies and set it into the ground. For each nest cage, we constructed an identical cage and set it into the ground 50 cm away from the nest cage. This was the foraging cage, meant to simulate the foraging site, where drupes were supplied. We connected the two cages with a mesh-covered passageway so that nymphs could leave their nest, which had been set in the nest cage, and freely move to the foraging cage (Fig. 4.15; Nomakuchi et al. 2005). The bottom of the nesting cage was layered firmly with soil and then with leaf litter, similar to the nest sites in the field. The base of the passageway was a wooden board, and the bottom of the foraging cage was a white Styrofoam board to facilitate observation of the drupes. A female and her egg mass were placed into each of ten nesting cages. Inspection of the cages was made

Fig. 4.15 Experimental field cage to assess timing of nymphal departure. Adapted from Nomakuchi et al. (2005)
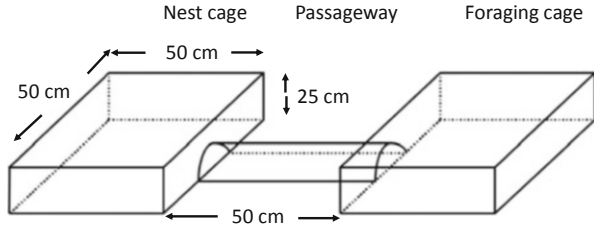
daily. After hatch, the female could leave her nest and enter the foraging cage to provision her nest. Throughout the second stadium, nine drupes were evenly placed in the foraging cage. Each drupe that a female removed from the foraging cage was replaced at the time of daily inspection. Of note, larger nymphs within a cohort seemed to access the drupes females provisioned more easily, excluding the smaller nymphs. This apparently led to the large range in the duration of a given stadium. At the end of the second stadium, the number of drupes in the foraging cage was increased to 18. The first time a nymph was observed in the foraging cage was considered the start of nymphal departure from the nest. Nymphal hatch was synchronized within a clutch. However, even under the minimally constrained conditions of this field experiment (no predation and guaranteed access to food), the average number of offspring $\pm$ S.D. decreased steadily from hatch ($n = 10$, 68.8 ± 27.7) to first sign of independence ($n = 10$, 45.6 ± 28.6) to emergence of first adults ($n = 8$, 27.9 ± 24.1) for a final average decrease of over 60%. The duration of first, second, third, fourth, and fifth nymphal stadia, determined by >50% of individuals molting to a given stage, was 2, 2, 4, 4, and 14 days, respectively (Nomakuchi et al. 2005). Nymphs remained in the nest feeding on provisioned drupes for several days after hatch and began to walk around the outside of, but remained near, the nest apparently when the drupe level became insufficient. Eventually they left the nest and made their way over to the foraging cage. Once in the foraging cage, they apparently did not return to the nest, though some apparently returned to the nest cage, and aggregated in the corner. Those in the foraging cage aggregated on drupes and fed.

Most cohorts began departing from the nest cage and relocating to the foraging cage at the fourth nymphal stadium (Fig. 4.16; Nomakuchi et al. 2005). However, in a cage where the mother had died during the late second/early third stadium, the nymphs left the nest during the third stadium. On the other hand, in another cage where the mother had died early, nymphs remained until the adult stage, probably because the mother had already provisioned many drupes. All females in this experiment had equal access to drupes, so clearly there is intra individual variation in the provisioning capacity that has nothing to do with resource availability. Once independence began, the average percentage $\pm$ S.E. of nymphs that had relocated to the foraging cage was $12.8 \pm 9.4\%$ on the first day and increased to $36.6 \pm 27.9\%$ on the second day after initiation of independence. As reported in Filippi et al. (2000), nymphs had relocated to the provisioning area, and, in fact, provisioning always continued until the death of the female (Nomakuchi et al. 2005); however, there was
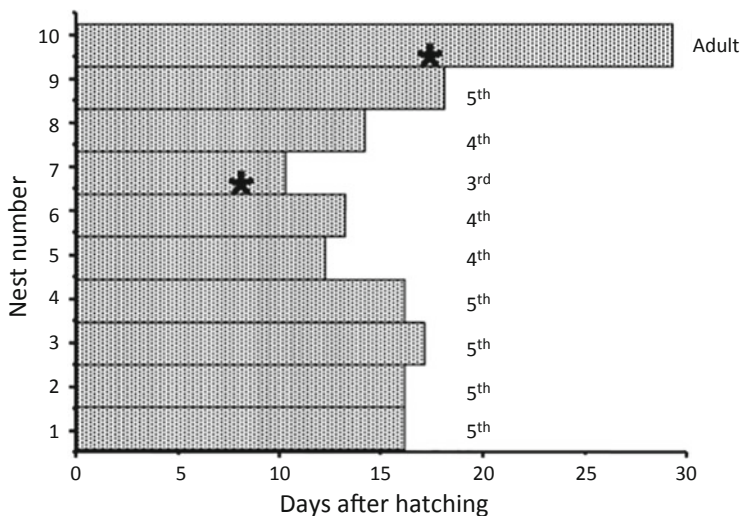
Fig. 4.16 Duration of nymphal nesting period. Stadium at the start of relocation from the nest to the foraging area is indicated on the right of each bar. Asterisks indicate death of the females that died before the start of nymphal independence. All other females died after nymphal independence. Adapted from Nomakuchi et al. (2005)

no significant difference between the duration of the nymphal nesting period and the duration of female provisioning (Wilcoxon test, $T = 9$, $n = 9$, $P > 0.05$).

To test our prediction that a progressively increasing number of drupes per day would be required to sustain nymphal development and that once the amount provisioned failed to be sufficient, the nymphs would depart from the nest an index of average provisioning rate (IAPR) was calculated. IAPR is a sequential daily calculation of the total number of drupes provided per cumulative number of nymphs in the nest until the day the first nymphs departed the nest for the foraging cage. The index values were subjected to a randomization test for evaluation. The IAPR value was positively correlated with the duration of the nymphal nesting period (Fig. 4.17; Pearson correlation coefficient, $r = 0.89$, $n = 9$, $p < 0.01$). However, it is possible that there was a spurious correlation between duration of the nymphal nesting period and IAPR because IAPR values tended to increase over days as a result of increasing provisioning frequency and decreasing number of nymphs. Therefore, an exact randomization test was performed to confirm the validity of the correlation. We were able to verify that departure from the nest cages began significantly earlier in nests that had fewer drupes provisioned ($p = 0.0421$; Nomakuchi et al. 2005).
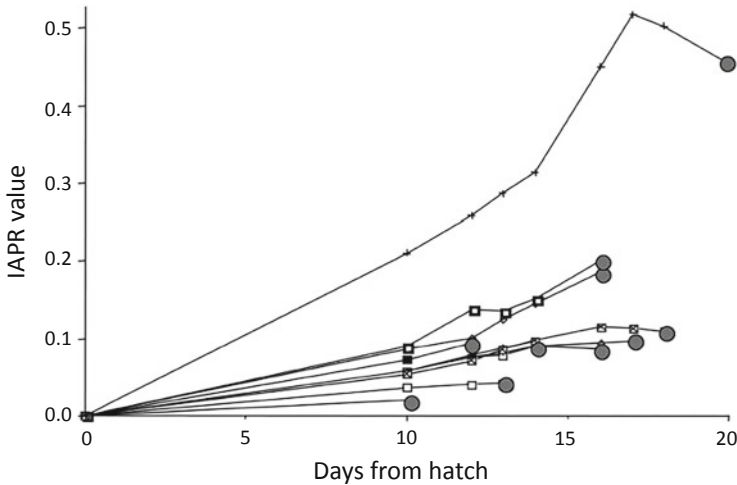
Fig. 4.17 Impact of maternal provisioning of drupes on duration of nymphal nesting stage. Each line represents the pattern of a different nest. Terminal point of line indicates beginning of nymphal independence. See text for explanation of IAPR value. Pearson correlation coefficient done for terminal points only. $N = 9$, $r = 0.89$, $p < 0.01$. Adapted from Nomakuchi et al. (2005)

4.15.2   Experiment 2: Impact of Provisioning Capacity on Timing of Nymphal Independence

To confirm the hypothesis that female provisioning capacity is positively correlated with the duration of the period prior to nymphal departure from the nest, we put females with an egg mass into a nest cage as in Experiment 1. We removed the mothers at the second stadium to avoid the possibility that another aspect of female care was affecting the duration of the nesting period prior to independence and provided the nymphs with poor or rich resource abundance in the nest. The number of drupes placed in the nests daily was regulated according to the number of nymphs in the nest each day using the IAPR values calculated in the previous Experiment. The number of drupes in the rich group was maintained at about four times the number in the poor resource group. The average number of days until departure from the nest in the poor resource group $\pm$ S.D. was 12.2 ± 2.7 days ($n = 6$), while the corresponding number of days in the rich resource group was 18.0 ± 3.1 days, $n = 5$. The duration of the nymphal stage prior to departure from the nest was significantly longer in the rich resource group (Fig. 4.18; Mann-Whitney U test, $p = 0.026$; Nomakuchi et al. 2005). Clearly, nymphs in nests with females that provision more drupes will depart for the foraging site later than those that are provisioned with fewer drupes.
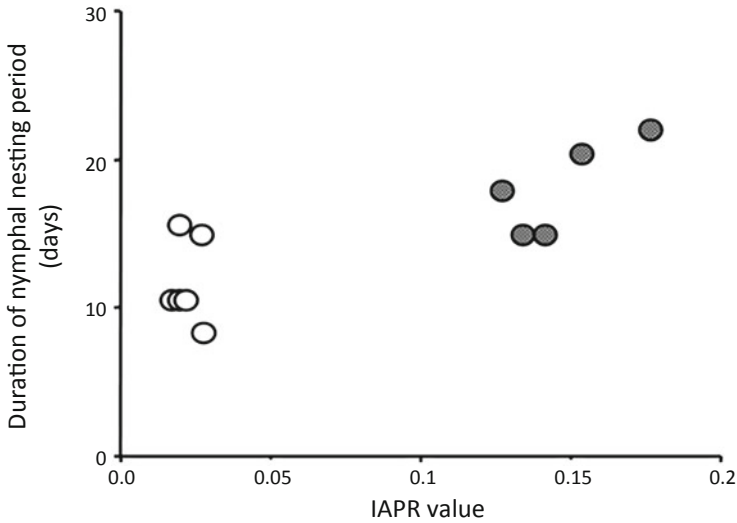
Fig. 4.18 Impact of provisioning level on duration of nymphal nesting period. Nymphs received either poor (open circles) or rich (shaded circles) provisioning. Females were removed at the second stadium for both conditions. Adapted from Nomakuchi et al. (2005)

4.15.3 Experiment 3: Distance and Success Rate of Nymphal Instars Reaching the Foraging Area

Finally, to determine how nymphal stadium and distance from the foraging area impact success of nymphs accessing the foraging area, in 2000 we reared nymphs with drupes in the laboratory at 25 °C from the egg stage to the second, third, and fourth nymphal stadia. The young nymphs were too soft to mark, so to ensure that any nymphs we encountered in this study were from among the ones released, we established an artificial foraging area more than 10 m from any host tree or subpopulation of *P. japonensis*. We placed 50 drupes in a 1 m^2 plot and then released 40–50 second, third, and fourth stadium nymphs either 1 m or 5 m from the "foraging site." We checked the number of nymphs that had reached the "foraging area" daily and removed them to avoid double counting. We repeated the trials five times for the second stadium nymphs and ten times for third and fourth stadium nymphs. We continued each trial until no more nymphs entered the "foraging area." There was a significant increase in success rate of nymphs as the stadia progressed (Fig. 4.19; Two-way ANOVA, $p < 0.001$; Nomakuchi et al. 2005). The success rate was also lower for nymphs released from further away; however, the combined effects of age and distance were not significant. Younger nymphs clearly have a much lower likelihood of successfully reaching the foraging area, particularly because many nests are located more than 5 m from the foraging area.
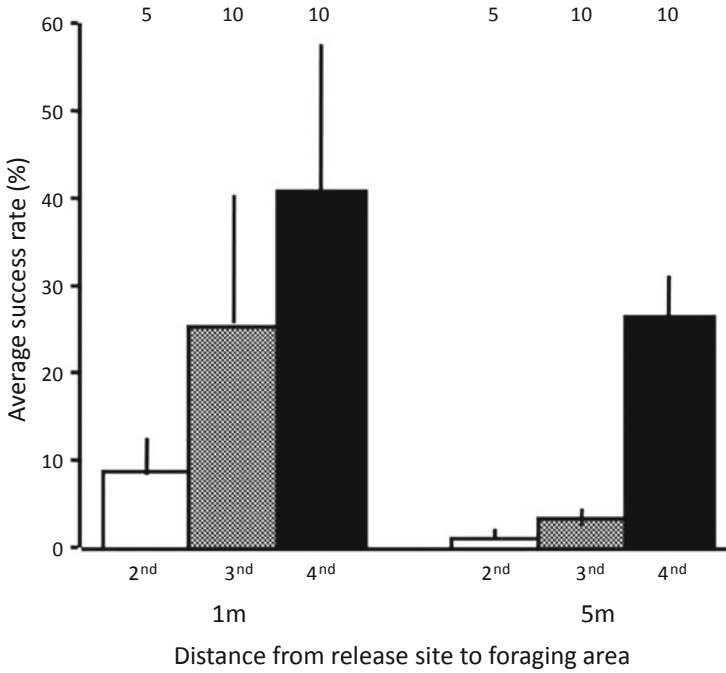
Fig. 4.19 Impact of distance from the foraging area on the success rate of different instars reaching it. Numbers above bars represent number of trials. Error bars represent S.D. Adapted from Nomakuchi et al. (2005)

4.16 Conclusions About the Impact of Provisioning on the Nymphal Nesting Period

The first series of experiments showed that, when excluding fifth instars who tend to aggregate on tree trunks, by far the greatest number of nymphs in the foraging area was in the fourth stadium (Figs. 4.13 and 4.16), suggesting that nymphs at that stage can succeed outside the nest much better than younger nymphs. On the other hand, the very low number of second and third instars in the foraging area suggests that when food becomes insufficient in the nest, they will be forced to leave. However, while only fifth instars seemed to be considerably less vulnerable to predation and fourth instars suffered equal predation rates to second and third instars (Fig. 4.14), the longer nymphs are exposed to predators during the vulnerable stages (second–fourth stadia), the more likely it is that they will succumb to predation before reaching the fifth stadium. Thus, nymphs leaving at the fourth stadium stand a far greater chance of reaching the fifth stadium and the adult stage than those leaving at the second or third stadium.

We have also demonstrated that the duration of the nesting period is positively correlated with the number of drupes that a female is able to provide (Figs. 4.17 and

4.18); however, because good drupes are scarce, the mother is not always able to provision enough to meet their needs so they will be forced to leave at a vulnerable stage. Moreover, the distance to the foraging area is negatively correlated with successful ability to reach it for all stadia, but the impact of distance is far greater for second and third instars than fourth instars (Fig. 4.19) who are much more mobile and better able to navigate the rough terrain as they make their way to the foraging area which can be up to 12 m away from the nest. It is a game of deadly trade-offs for nymphs who must balance the risk of starving in the nest, or being cannibalized by hungry siblings there, against the threat of predation and other physical hazards awaiting them outside the nest when they leave for the foraging area!

4.17   Provisioning of Nutrition *Before* Provisioning Drupes: Production of Trophic Eggs

We have mentioned the observation of newly hatched nymphs feeding on the egg cases (Filippi-Tsukamoto et al. 1995). Another observation that warranted explanation was that some eggs in *P. japonensis* egg masses invariably failed to turn pink, develop eyespots, and hatch. We decided to investigate whether females were in fact producing trophic eggs to feed their newly hatched offspring. Trophic eggs are eggs that are produced as a food for young, and young feeding on them is not considered oophagy or cannibalism because the mother has deliberately produced them for the express purpose of enhancing offspring development. Trophic egg production represents an extreme version of food caching that allows mothers to progressively provision young as a supplement to other food sources (Alexander 1974; Polis 1981, 1984; Mock and Parker 1997). Prior to our first study on trophic eggs in *P. japonensis*, production of trophic eggs had been documented in a variety of insect species but most commonly, by far, in the Hymenoptera (Wilson 1975; Polis 1981; Kukuk 1992; Choe and Crespi 1997; Iwanishi et al. 2003). Trophic eggs had also been reported in a few subsocial species, including a burrower cricket (*Anurogryllus muticus* DeGeer; West and Alexander 1963) and a cydnid (*Adomerus triguttulus* Motchulsky), a relative of *P. japonensis* with similar types of extensive parental care behaviors (Nakahira 1994). We now know that most of the parental cydnids do in fact also produce trophic eggs, and we will discuss those in Sect. 4.18. For foraging *P. japonensis* females, food availability is scarce, and provisioning efforts are not always successful. In fact, in some years the timing of drupe availability is totally out of synch with the timing of nymphal need (see Sect. 4.19). Cannibalism is also rampant in this species, and it would behoove *P. japonensis* mothers to evolve some mechanism to keep the young ones from eating each other while they are out trying to sequester a meal (Trivers 1985; Crespi 1992; Kukuk 1992; Mock and Parker 1997; Kim and Roland 2000)! The duration of the developmental period and the ultimate size of offspring can be greatly enhanced by trophic eggs (Baur 1992; Kam et al. 2000; Kim and Roland 2000; Kudo and Nakahira 2004). However, feeding on eggs does not necessarily mean that the female has produced trophic eggs; it could simply be cannibalism.

Perry and Roitberg ([2006](#)) proposed criteria that should be met prior to designating eggs that offspring feed on as trophic. In order to establish a cohesive and consistent definition of trophic eggs from an evolutionary standpoint, they proposed two hypotheses that could be tested to determine whether the putative trophic eggs had evolved. First, the eggs should differ phenotypically from fertile eggs, such that their production involves lower cost because females should invest less in them than in viable eggs destined to become offspring. Most often trophic eggs are smaller than viable eggs, rounder (Koedam et al. [2001](#)) and have a softer shell, making them easier for young nymphs to penetrate. They might also have other structural differences, such as fewer or no micropyles, where the sperm penetrate the egg, as in the trophic eggs of *Adomerus triguttulus* (Kudo and Nakahira [2004](#)). Second, the functional value of the trophic eggs should be determined. What is the adaptive value of the putative trophic eggs? We carried out a series of experiments to test these two hypotheses and to demonstrate that female *P. japonensis* produce trophic eggs to enhance offspring success.

4.17.1   Are Eggs Fed on by New Hatchlings Morphologically Different from Viable Eggs?

Female *P. japonensis* lay all their trophic eggs at the time of initial oviposition. Fertile and unfertilized eggs can be easily distinguished because the fertile eggs gradually turn from cream to yellow to pink and finally deep pink with red eyespots (Fig. [4.20](#)). To eliminate the possibility that eggs being fed on by newly hatched

Fig. 4.20 Mother *P. japonensis* guarding her egg mass near hatch. Cream-colored eggs are unfertilized eggs; pink eggs are fertilized eggs; red dots are eyespots. Photo credit, Mantaro Hironaka

Table 4.4 Dimensions and weight of fertilized and unfertilized eggs

	Length (mm) Mean $\pm$ S.D.	Width (mm) Mean $\pm$ S.D	Weight (mg) Mean $\pm$ S.D
Fertilized eggs	1.51 ± 0.04	0.95 ± 0.03	$0.80 \pm 7.5 \times 10^{-2}$
Unfertilized eggs	1.47 ± 0.06	0.96 ± 0.04	$0.78 \pm 8.1 \times 10^{-2}$
P value (student's t-test	0.0001	0.22	0.0035

10 fertilized and 10 unfertilized eggs were measured from each of 5 egg masses for a total of 50 eggs of each type. Adapted from Hironaka et al. (2005)

nymphs were not simply eggs that had failed to get fertilized, we assessed whether fertilized and unfertilized eggs in an egg mass differ morphologically, other than color. We collected eight females with their egg masses from the field site at Mt. Hinokuma in Kanzaki Town, Saga, Japan, and brought them into the lab for incubation in individual clear plastic, lidded cups at 25 °C. Just prior to hatch, as evidenced by the clear presence of eyespots, mothers were removed. We then gently dissociated five egg masses and measured length and width of ten fertile and ten unfertilized eggs from each mass (total 50 eggs of each type). Individual egg weight was estimated by taking the average weight of the ten eggs that had been measured. While the average length of fertilized eggs (range 1.42–1.59 mm) was significantly longer than that of unfertilized eggs (range 1.33–1.6 mm), there was no difference in width between the two egg types, resulting in the fertilized eggs having an oval shape and the unfertilized eggs having a rounder shape (Table 4.4; Hironaka et al. 2005). Fertilized eggs were also significantly heavier than unfertilized eggs. Moreover, the unfertilized eggs were softer when pressure was applied with a tweezer (unpublished observation) and had fewer and flattened micropyles (S. Munshi and L. Filippi, unpublished data) compared to fertilized eggs. These phenotypic differences meet the first criterion proposed by Perry and Roitberg (2006) as necessary to conclude that females are producing trophic eggs.

4.17.2 Numbers/Proportions of Fertile and Unfertilized Eggs

In 2000, there was considerable variation in the average total number of eggs in an egg mass; among 51 dissociated egg masses, the average number of eggs was 130.5 ± 36.2. There was also considerable variation in the number of fertilized eggs in a mass (88.8 ± 34.2). There was less variation in the number of unfertilized eggs in a mass (41.7 ± 13.0). The average percentage of trophic eggs in an egg mass was 33.5 ± 11.9, showing the least amount of variation. The frequency distributions of these egg composition parameters are shown in Fig. 4.21.
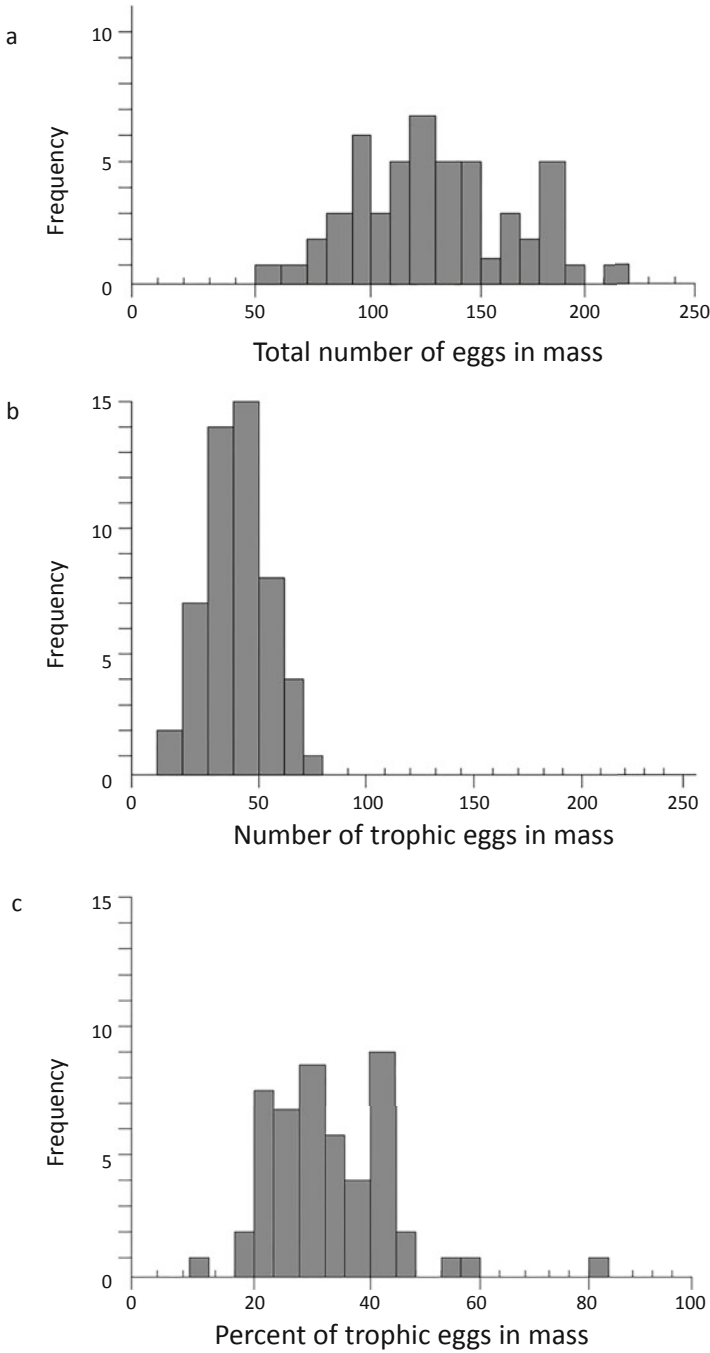
Fig. 4.21 Frequency distributions showing composition of egg masses. (**a**) Total number of eggs; (**b**) number of trophic eggs; (**c**) percent of trophic eggs in mass. $N = 51$ dissociated egg masses. Adapted from Hironaka et al. (2005)

4.17.3 Impact of Feeding on Unfertilized Eggs on Nymphal Weight at 24 h

Because newly hatched nymphs were seen probing egg cases immediately after hatch in the earlier studies (Filippi-Tsukamoto et al. 1995), we were interested in examining how that presumptive feeding impacted nymphal growth. A preliminary finding that 89.1% of trophic eggs in a mass are located on the outside of the mass (Hironaka et al. 2005) allowed us to perform a trophic egg removal experiment to assess this. We knew that by removing surface unfertilized eggs, most would be removed. For this study, we chose 36 females with egg masses that contained between 25 and 45% unfertilized eggs in their mass (see Hironaka et al. 2005 for details on how we calculated this). We separated the mother from all egg masses and, using a fine paintbrush (one of our favorite tools!), gently removed all trophic eggs from the surface of 18 of the egg masses (experimental group). We then allowed all females to retrieve their egg masses. The 18 egg masses that were left intact comprised the control group. When the eggs hatched, nymphs were clearly seen probing their egg masses. After 24 h we weighed the nymphs. Control nymphs that had access to the unfertilized eggs were significantly heavier by 16% at 24 h (range 0.94–1.7 mg) than the unfertilized egg removal nymphs (range 0.8–1.3 mg) (Fig. 4.22; Hironaka et al. 2005). Clearly, the control nymphs fed on those unfertilized eggs.
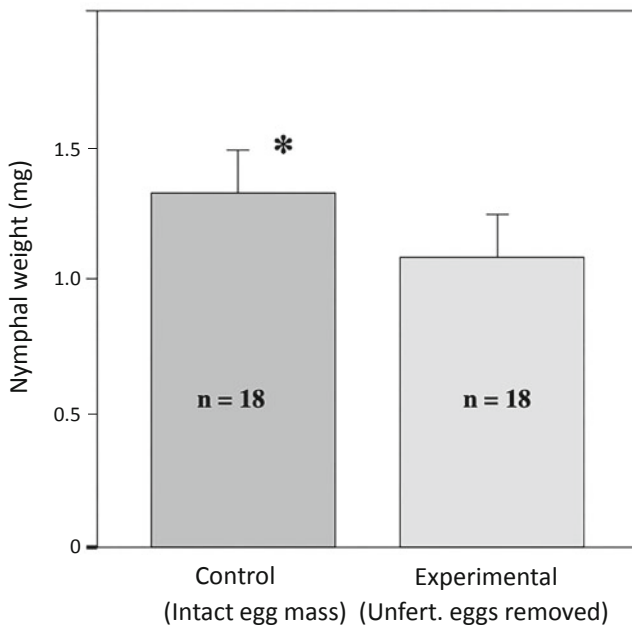

Fig. 4.22 Impact of removal of surface unfertilized eggs on average nymphal weight. *Mann-Whitney U test, $p < 0.002$. Adapted from Hironaka et al. (2005)

4.17.4   Impact of Access to Unfertilized Eggs on Nymphal Development Rate and Survivorship

There were other significant differences between nymphs with and without access to the unfertilized eggs. Twice as many first stadium nymphs had molted to the second stadium by 48 h (Mann-Whitney U test, $p < 0.005$; Fig. 4.23a; Hironaka et al. 2005), and survivorship at day 10 of nymphs with access to unfertilized eggs was over three times greater than that of nymphs deprived of access to unfertilized eggs (Mann-Whitney U test, $p < 0.003$; Fig. 4.23b; Hironaka et al. 2005). Nymphs clearly

Fig. 4.23 Impact of access to unfertilized eggs on nymphal success. (**a**) Percent of nymphs that had molted to the second stadium by 48 h; Mann-Whitney U test, $p < 0.005$; (**b**) percent of nymphal survivorship at day 10; Mann-Whitney U test, $p < 0.003$. Adapted from Hironaka et al. (2005)
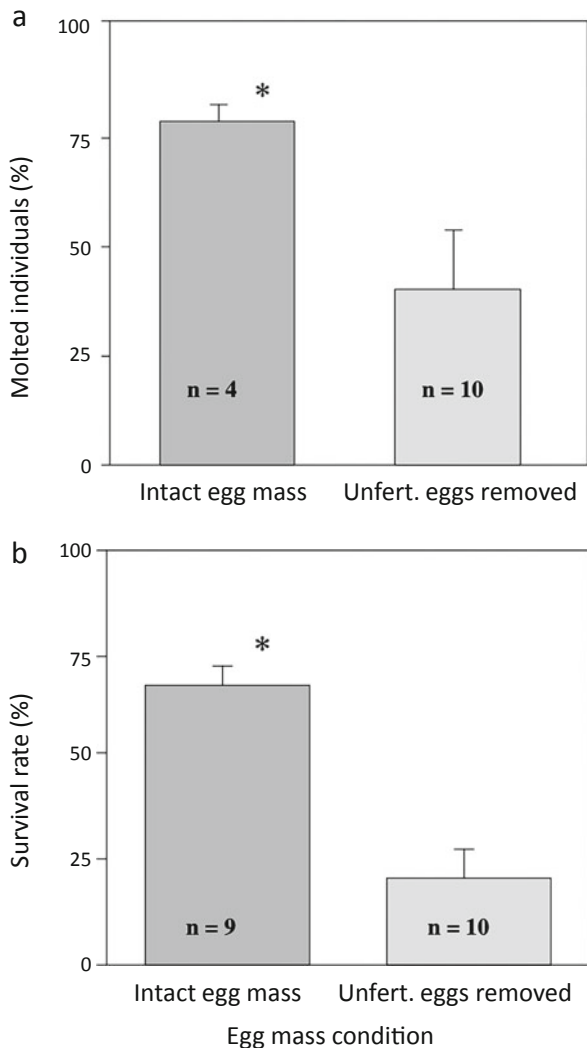
benefitted from access to unfertilized eggs because they were significantly heavier, molted to the second stadium significantly faster, and had significantly higher survivorship.

The findings of this study demonstrate that females are producing unfertilized eggs for nymphs to feed on; these eggs meet the criteria proposed by Perry and Roitberg (2006) for trophic eggs. They are phenotypically distinct from fertile eggs (Table 4.4) and, being smaller and more fragile, are undoubtedly less costly to make, and their adaptive value to offspring success has been demonstrated (Figs. 4.21, 4.22, and 4.23). Because of the scarcity and unpredictability of drupe availability, females often fail to find a drupe for a day or two after hatch. Trophic egg production as a way to ameliorate the poor resource condition (Kudo and Nakahira 2004) would fulfill the premise of the "ice-box" or "food-cache" hypothesis (Alexander 1974; Polis 1981, 1984). Trophic eggs can sustain young nymphs until drupes can be found. It is unlikely that nymphs feed on fertile eggs because of synchronized hatch (see Chap. 5), but hungry nymphs could feed on each other. Having access to trophic eggs should also mitigate a parent-offspring conflict by reducing cannibalism in the nest (Crespi 1992; Kukuk 1992; Mock and Parker 1997).

The pattern of deposition of trophic eggs is also adaptive. Because 89% of the trophic eggs are located on the surface of the egg mass (Hironaka et al. 2005), predation and physical damage will occur more on the less costly trophic eggs, and loss of fertile eggs is likely reduced (Nakahira 1994; Kudo and Nakahira 2004).

4.17.5 Risk-Sensitive Decision-Making as the Source of Variation in Trophic Egg Production

The large degree of individual variation in the number of trophic eggs produced and the percentage of the egg mass that is trophic eggs led us to the next study. We suspected that females were adjusting these numbers based on the resource availability and the condition of the female. Even within a given reproductive season, depending on whether a female oviposits early or late in the season, resource availability can fluctuate dramatically. Females that oviposit early have fewer females to compete with when their eggs hatch, and their young have no older nymphs to compete with for food once they depart for the foraging area. However, the female risks not having any drupes at all if they are not yet ripe when her eggs hatch. See Sect. 4.19 for more on this topic. On the other hand, laying ones' eggs later in the season might coincide with more mature drupes falling from the host tree for provisioning; however, there will also be more females to compete with for access to those drupes and, importantly, plenty of older nymphs for their offspring to compete with when they get to the foraging area. Phenotypic plasticity in deposition of fertile and trophic eggs would allow females to respond to the resource availability and their physical condition when developing and laying eggs (Bradshaw 1965; Fordyce 2006; Whitman and Agrawal 2009). In addition to assessing the food availability for provisioning, mothers should be able to gather

information that would allow them to assess the level of resource constraint that offspring will encounter from competition with siblings and offspring from other cohorts when they become independent and relocate to the foraging area and adjust number, size, and types of eggs they produce accordingly (Mock and Parker 1997; Creighton 2005). We tested the hypothesis that timing of oviposition (early or late) and maternal phenotype (relative weight) were factors being used by females to make risk-sensitive decisions about the number, size, and types of eggs they produce. We predicted that females ovipositing later in the season would make more trophic eggs than those ovipositing earlier in response to the higher level of competition they and their offspring would encounter. We also predicted that relatively heavier females just prior to oviposition would produce more of both types of eggs as they would have more energetic resources to produce the eggs and to provision them (Filippi et al. 2012).

4.17.6 Temporal Variation in Female and Egg Parameters During the Nesting Season

During the nesting season of 2005, we mass marked pregnant females, easily distinguished by their distended abdomen, roaming about the nesting area searching for a nest with one of three different colors based on the phase of the nesting season (early, middle, late). The females had mated in the field and fed on the host trees there, presumably gathering information about resource availability and competition level while doing so. The first pregnant females were seen on May 29. Between May 29 and May 31, we marked 63 females and designated them the early group. Numbers of pregnant females dropped after that, and we marked only five females from June 1 to June 3. These females were designated as the middle group and were marked for purposes of exclusion from the early and late groups. We marked 46 females from June 4 to June 7 that were designated the late group. All females were released where we had found them as soon as they were marked and allowed to nest. After about a week, we began searching for the nests of the females we had labeled. To minimize confounding variables, we concentrated our search efforts on a 15 m^2 area that we recognized as being suitable for nesting and having consistent features throughout. We were able to recover 13 early females and 12 late females and their egg masses. We recorded the distance of each nest to the nearest appropriate-sized host tree and carefully transferred them with their nests to plastic cups containing leaf litter and soil and brought them back to the lab for incubation and then analysis. We incubated them at 25 °C, under a photoregime of 16L:8D (Filippi et al. 2012).

One day prior to the anticipated hatch date, based on the deep pink color of the fertile eggs and the dark red eyespots, the egg mass was separated from the female, and the female's weight and pronotum width were measured. We also weighed the egg mass and, after dissociating the egg mass with a fine paintbrush, counted the numbers of fertile and trophic eggs in each mass. In 2005, there were significant differences between early and late nesting females for all parameters measured

except two (Table 4.5; Filippi et al. 2012). The nests of late nesting females were, on average, 2.5 m closer to the foraging site, but the range of distances was similar in both groups (2.5–12.1 m for early nesters and 3.1–12.7 m for late nesters). Nesting closer to the foraging area is considered a riskier behavior because of the lack of protective leaf litter and the greater density of foraging females and, later in the season, foraging nymphs; females would be expected to nest there when drupes or nests are particularly scarce (Filippi et al. 2002, 2005). The best nesting sites were probably saturated by the time late nesters were ovipositing.

The body weight of late nesting females was 6% heavier than that of early nesters (Table 4.5; Filippi et al. 2012). Because there was no difference in body size as measured by pronotum width, this difference in weight indicates that late nesting females had more fat body reserves after producing their eggs which could be used for provisioning. It is unclear whether these females started out with more fat body reserves after breaking reproductive diapause or if they actively fed on the host tree longer, accruing enough energy to produce eggs and build up their own reserves. The data from our earlier study on mating behavior indicate that fat body reserves after breaking diapause decrease for a time during the mating season before they build up again during feeding on the host tree (Fig. 3.5), which would support the latter scenario. Nesting further from the host tree would require more work getting to and from the foraging site and transporting drupes back to the nest. The extra body weight would support this additional effort.

The egg masses of late nesting females were nearly 20% heavier than those of early nesters. This extra weight was clearly relegated to trophic eggs, because although late nesters produced 10% more total eggs, they produced 16% fewer fertile eggs and 40% more trophic eggs. On average, a late nester's egg mass was comprised of 44% more trophic eggs than an early nester's (Table 4.5). Late nesting females were apparently responding to the likely shortage of drupes that would occur later in the season because provisioning females had been removing drupes from the foraging area for days before late nesters' eggs hatched and their third and fourth stadium offspring would enter a foraging stage overrun with clusters of older nymphs feeding on drupes.

4.17.7 Impact of Body Size on the Number of Eggs Produced

There was no correlation between body size (PNW) and the number of eggs produced for either group (not shown); however, there was a significant positive correlation between body weight and the total number of eggs produced for both early and late nesters (Fig. 4.24a; Filippi et al. 2012). Interestingly, the body weight of only the late nesters showed a significant positive correlation with the number of fertile eggs produced (Fig. 4.24b; Filippi et al. 2012). It is difficult to draw conclusions here because the outlier late female in the figure clearly impacted the significance, and when that female was removed from the analysis, the finding was no longer significant.

Table 4.5 Female and egg parameters of early and late nesting females

Timing of oviposition	Distance from host tree (m)	Female PNW (mm)	Female Bwt (g)	Egg mass wt (g)	Individ. egg wt (g)	Total no. eggs	No. fert. Eggs	No. trophic eggs	% trophic eggs
Early $n = 13$	8.70 ± 2.87	7.43 ± 0.19	0.18 ± 0.02	0.09 ± 0.01	$8.4 \times 10^{-4} \pm 5.59 \times 10^{-5}$	113.62 ± 12.37	74.54 ± 12.14	39.08 ± 13.38	34.08 ± 10.43
Late $n = 12$	6.35 ± 2.73	7.36 ± 0.29	0.19 ± 0.02	0.11 ± 0.02	$8.8 \times 10^{-4} \pm 9.75 \times 10^{-5}$	126.92 ± 16.51	62.75 ± 13.09	64.17 ± 8.75	50.79 ± 5.89
$P =$	0.04	0.45	0.04	0.02	0.22	0.03	0.03	**0.0001**	**0.0001**

Values are means $\pm$ S.D. PNW = pronotum width. Female BWT = body weight just prior to hatch. Adapted from Filippi et al. (2012)
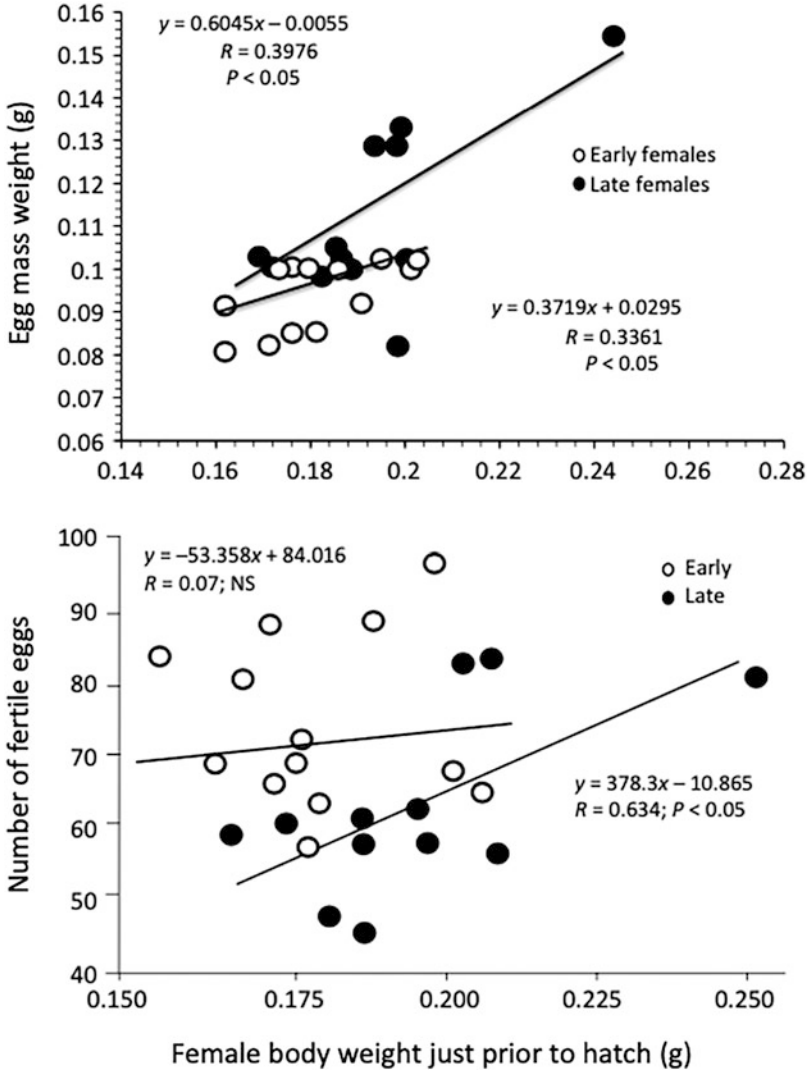
Fig. 4.24 Relationship between female body weight just prior to hatch and egg abundance. (**a**) Total egg mass; (**b**) fertile eggs. Adapted from Filippi et al. (2012)

The degree of variation in a data set is always an intriguing source of more questions when the variation is real, and not a source of error. The tendency for the earlier females to have a much greater degree of variation in the number of trophic eggs produced than late females (33% of the average for early females and 10% of the average for late females; Table 4.5) was curious, particularly because that difference in variation was not present for fertile eggs (~16% and ~21% of the average for early and late females, respectively), but the difference was not significant. LF recalls being
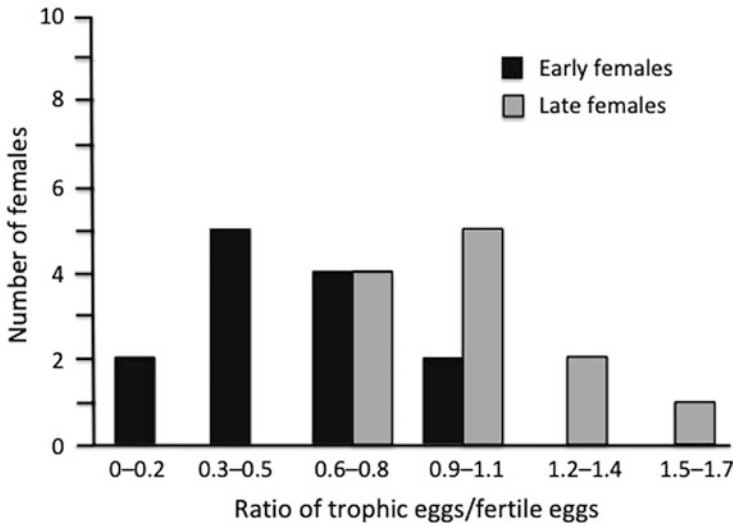
Fig. 4.25 Frequency distribution of the rate of trophic eggs produced per fertile egg. Adapted from Filippi et al. (2012)

frustrated when analyzing the data, because although we detected something intriguing was afoot, nothing stood out until we arranged the data as number of trophic eggs per fertile egg in each group. And there it was! Most late females produced at least one trophic egg per fertile egg (1.06 ± 0.27, mean $\pm$ S.D.), while very few early females did so and more than half of the early females produced 0.5 or less trophic eggs per fertile egg (0.55 ± 0.24, mean $\pm$ S.D.; Fig. 4.25).

There is a physiological limitation to the amount of provisioning a female can manage. Females should be designed to produce an appropriate number of eggs based on their ability to provision those nymphs (Lack 1947; Monaghan and Nager 1997). The data suggest that the number of fertile eggs that early females produced might be the maximum number that a female could provision and any extra body fat reserves that early females possessed were relegated to trophic eggs. On the other hand, while both groups showed a positive correlation between body weight and total egg number, only late females showed that same correlation between body weight and the number of fertile eggs. The data suggest that to mitigate an anticipated scarcity of drupes, late females obligatively produce eggs such that the number of fertile eggs matches trophic and, when resources allow, facultatively produce additional fertile eggs.

We carried out a binomial generalized linear model (GLM) analysis to determine the factors that had the greatest impact on the percentage of trophic eggs that a female incorporated into her egg mass. Timing of oviposition and distance of the nest from the foraging area had the greatest impact (Table 4.6). Late females produced a higher percentage of trophic eggs, as did females nesting closer to the foraging area. This suggests that late females, who likely nested closer to the

Table 4.6 Coefficient table of binomial GLM for analysis of percentage of trophic eggs

Coefficients	Estimate	SE	X value	P-value
Intercept	−0.37288	0.38439	−0.970	0.332
Early/late oviposition	0.57169	0.08702	6.570	0.0000
Distance from host tree	−0.05775	0.01403	−4.116	0.0000
Female BWT Just prior to hatch	−1.95321	1.95897	−0.997	0.319

GLM generalized linear model. Adopted from Filippi et al. (2012)

foraging site because more distant, protected nest sites were saturated with early nesting females, were responding to the disturbance to the nest and competition that their nymphs would experience from the many provisioning females and foraging older nymphs.

The hypothesis that the resource exposure as a function of timing of oviposition was determinant in the number of trophic eggs produced was supported (Fig. 4.25; Tables 4.5 and 4.6). Moreover, the hypothesis that maternal phenotype (body weight) was also a determinant was also supported (Fig. 4.24; Table 4.5). The data clearly indicated that females have the capacity to adjust the number and types of eggs they produce to best respond to the availability of the resource, which is based on numbers of drupes produced by the host tree and competition with foraging females and older nymphs at the time their nymphs will be foraging. The findings that late females produce on average about one trophic egg for each of the hatchlings to feed on while early females produce about one half of a trophic egg per nymph (Fig. 4.26) suggests that, because of less resource availability to later nymphs, trophic eggs are much more important to the success of late offspring than early ones. It will be of great interest to explore whether there is a genetic predisposition to when and how females oviposit, or whether it is simply a plastic response to the resources each female was able to accrue prior to diapause. Also, clarifying the cues females use to assess resource level would be a fascinating undertaking. Do they "count" the number of females they are exposed to while they are moving about the host tree feeding to produce eggs? There are no drupes on the ground when females descend the host tree to oviposit, so they cannot be assessing resource level by the number of drupes that they encounter on the ground while looking for a nest site. It could also be that they are "counting" the absolute number of drupes on the host tree that they encounter versus the number that have been fed on by other females. All intriguing questions, so much left to discover!

4.18   Timing and Manner of Trophic Egg Deposition in the Cydnid Relatives of *P. japonensis*

We have stated that 89% of trophic eggs produced by *P. japonensis* females are deposited on the outside of the egg mass, that they are all deposited at the time of initial oviposition, and that survivorship was critical to having access to trophic eggs
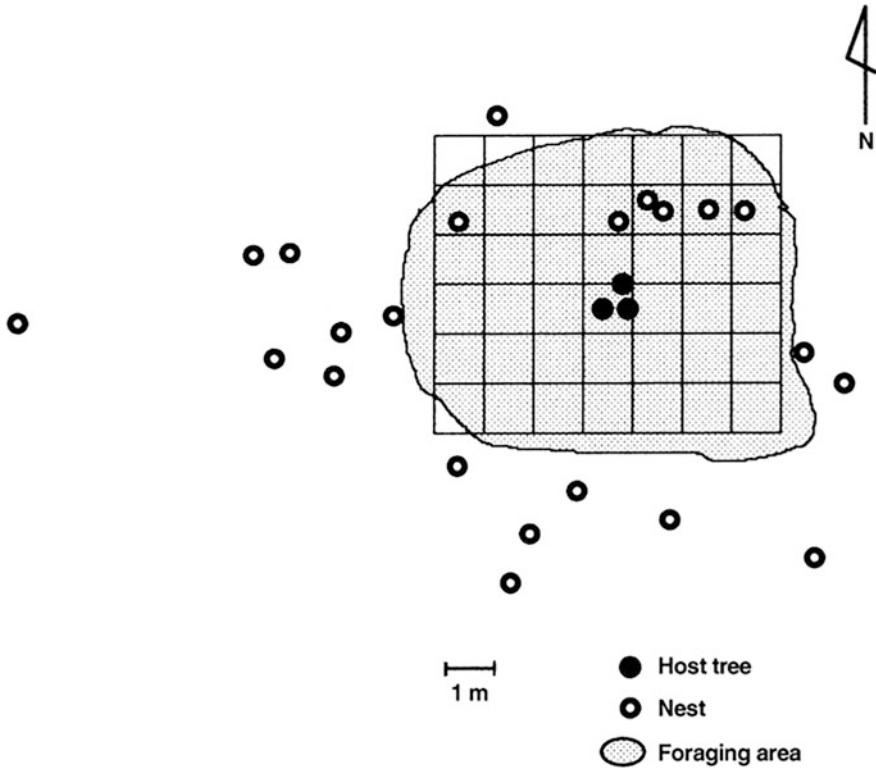
Fig. 4.26 Natural nesting and foraging areas at field site in 1995. Adapted from Filippi et al. (2002)

(Hironaka et al. 2005). The related Japanese cydnid provisioners do things quite differently. *Adomerus triguttulus* lays most trophic eggs in the original egg mass but continues to add trophic eggs during the egg guarding phase (Nakahira 1994). The timing and pattern of deposition of trophic eggs by *Adomerus variegatus* mothers are very similar to what is seen in *P. japonensis* (Mukai et al. 2010). *Canthophorus niveimarginatus* mothers, on the other hand, produce two types of trophic eggs based on timing of deposition. A small number of trophic eggs are deposited on the outside of the initial egg mass, and mothers also deliver trophic eggs to nymphs upon hatch. Thus, females produce both pre- and post-hatch trophic eggs (Filippi et al. 2009). As the mother is releasing the post-hatch trophic eggs, nymphs climb up to her abdomen and gather on the eggs to feed. Nymphs cannot survive the first stadium without access to trophic eggs because fragile first instars cannot penetrate the seed to access the endosperm (Baba et al. 2011). Mothers of *Adomerus rotundus* also produce these two types of trophic eggs, though the number of pre-hatch trophic eggs is apparently much smaller and not all females produce them (Inadomi et al. 2014). Sadly, mothers of *Sehirus cinctus*, the only provisioning cydnid in North America, do not produce trophic eggs (personal observation, L.F.).

One might speculate that the reason for pre- and post-hatch eggs allows for some flexibility in these iteroparous species. If the first egg batch should fail, then another one can quickly be produced. However, if the first batch is successful, the mother releases the less costly eggs upon hatch as food. The ecological significance of the different patterns of trophic egg production, and of no trophic egg production in *S. cinctus*, warrants further investigation.

4.19 Verifying Risk-Sensitive Decision-Making by Nesting Mothers

We discovered answers to some of the ultimate "whys" regarding the adaptiveness of maternal guarding of eggs and nymphs, progressive provisioning, and production of trophic eggs. We know that guarding protects against predation (Filippi-Tsukamoto et al. 1995, Filippi et al. 2000; Nomakuchi et al. 2001) and that in the absence of a provisioning female, even when predators are eliminated, the altricial offspring cannot gain enough access to good drupes to sustain growth and development, so careful choosing of good drupes by mothers is another adaptive function of progressive (Filippi et al. 2000; Nomakuchi et al. 2005). We found that survivorship of nymphs is positively correlated with duration of the nesting period, which is positively correlated with provisioning capacity of the female (Filippi et al. 2000; Nomakuchi et al. 2005). We discovered that trophic egg production by females is essential for success of offspring because often mothers cannot find good drupes for a few days after hatch (Hironaka et al. 2005). We also discovered that females manipulate the composition of their egg mass, producing more or fewer fertile and trophic eggs, based on resource availability, competition, and the mother's level of energetic stores (Filippi et al. 2012). However, a true understanding of the ecological constraints that lead to the evolution of behaviors, and the consequences of those behaviors on reproductive success, requires examination of the ecological parameters, such as resource availability, competition level, predation level, and weather effects, and their effect on patterns of behavior and reproductive success over several years. The natural behaviors observed in each of the previous field studies only provided a snapshot of the behaviors that occur in response to the specific conditions that prevailed during the year of the study. Particularly in environments where conditions are not stable and the resource is ephemeral, unpredictable, and scarce, evolution should favor parents that can use risk-sensitive decision-making to respond to the prevailing conditions in a way that would minimize costs and optimize their average fitness gains (Caraco 1980; Bulmer 1994). Because foraging and defense against predation are both essential but mutually exclusive behaviors, evolution should favor individuals that can assess and respond to the intensity of predation risk and balance their allocation of resources to these behaviors optimally (Lima and Dill 1990; Lima and Bednekoff 1999). In the case of nesting *P. japonensis*, the biggest defenses against predation are prolonging the duration

of the nesting period and guarding of eggs and nymphs by the mother, and duration of the nesting period is positively correlated with provisioning success of the female. Clearly, a careful balance between provisioning and guarding is required for the female to both be away from the nest often enough to provide sufficient drupes and be present in the nest often enough to ward off would-be predators. Provisioning capacity is determined by the abundance of drupes available to provisioning females, the intensity of competition, and the distance of the nest from the foraging site. Females invest considerable energy traveling over great distances to and from the foraging area, laden with heavy drupes. Moreover, as the provisioning season coincides with the rainy season, females regularly have the added constraint on provisioning capacity of navigating the rough terrain in a flooded environment. Are females adapting their behaviors to best mitigate all these constraints?

Although trophic egg production is a major component of parental care that *P. japonensis* females use to enhance offspring success, and we have determined that females can adjust egg numbers and proportions using adaptive risk-sensitive decisions, we did not have long-term data on trophic egg production in the field. Therefore, this long-term study was to assess only the progressive provisioning component of parental care. To verify that females were using risk-sensitive decision-making to enhance provisioning success, we analyzed data on the prevailing ecological conditions (abundance of good drupes, intraspecific competition, and weather) and the behaviors and physiological responses that were under female control (timing of oviposition and distance of the nest from the foraging area, both of which affect access to the resource and intensity of competition, and the duration of provisioning activity during the provisioning seasons of 4 years <1995, 1997, 1998, 1999>). We correlated the variation in these parameters with provisioning success of females (number of drupes provisioned). We tested four hypotheses: (1) the availability of drupes would be the greatest determinant of female risk-sensitive behavior. We predicted that the intensity of both intraspecific competition and inclement weather would be negatively correlated with provisioning capacity. (2) The greater exposure to predation and disturbance by other foraging females of nests within the foraging area because of sparse leaf litter and vegetation (Tsukamoto and Tojo 1992) combined with the heavy rains that prevail during the provisioning season should make nesting in the foraging area a risky behavior. Females were predicted to opt for the risky nest site only under severe conditions (extreme scarcity of drupes, extreme weather that would make the repeated and lengthy trips of progressive provisioning particularly costly). (3) Successful provisioners would adapt physiologically by adjusting the timing of oviposition so that provisioning coincides with the period when good drupes are available in the foraging area. (4) Finally, we hypothesized that females nesting according to the parameters in the first three hypotheses would have the greatest provisioning success.

The site that was used for the 4 years surveyed contained three host trees in a triangular pattern about 30 cm away from each other with overlapping crowns at the center of the foraging area. The ground under the crowns was cleared somewhat to facilitate observations, and a 6 × 7 m quadrat was established. Each year we individually marked with liquid paint markers all females entering the foraging

area during the first 10 days or so of the provisioning season and labeled as many nests as we could find (Fig. 4.26). To assess the relative number of good drupes available to foraging females, we set two seed traps under the collective crowns of the host trees as described in Sect. 1.11. The traps were set on opposite sides of the trees, about 80 cm from the trunks. As an estimation of relative drupe number and quality, drupes that fell into the traps were collected weekly, counted, and bisected to determine the endosperm content. Drupes containing more than 40% endosperm were considered good (Nomakuchi et al. 1998).

4.19.1 Variation in Duration of Provisioning Season and Individual Provisioning Activity

The duration of the provisioning period over the 4 years ranged from 16 to 30 days and averaged 24.25 days. The start of the provisioning season varied over the 4 years by about 2 weeks, beginning on about June 18th in 1995, June 11th in 1997, June 4th in 1998, and June 19th in 1999 (Fig. 4.27; Filippi et al. 2002). The provisioning period lasted for 21, 27, 30, and 16 days in 1995, 1997, 1998, and 1999, respectively. To assess the intensity of intraspecific competition, hourly counts of all females, marked and unmarked, in the foraging area were made daily during the most active provisioning period of the day, from early afternoon until evening, and the peak value for each day was used as the relative number of females for the day. The numbers of foraging females rose sharply over several days, peaked for a few days, and then dropped sharply. There were dramatic fluctuations in the peak numbers of foraging females over the 4 years, ranging from 13 to 65. In 1997 we also observed and found that provisioning occurred throughout the night, but the numbers of females foraging at night were half those observed in the daytime.

Marked individuals were recorded during each count each day to determine how long individuals continued to provision. The number of days individual females provisioned ranged from 1 to 25 over the 4 years but varied widely from year to year (range; average $\pm$ SE, 1–10 days; 3.5 ± 0.21 days; $n = 114$ in 1995; 1–19 days; 6.72 ± 0.39; $n = 156$ in 1997; and 1–25 days; 6.05 ± 0.34; $n = 137$ in 1998). There were significant differences between individual provisioning durations of 1995 and 1997 and between 1995 and 1998 (Kruskal-Wallis, post hoc for multiple comparisons $p < 0.001$; Filippi et al. 2002). Because only one census per day was done in 1999, that year was not included in this calculation. However, among the 3 years, the shortest duration of the overall provisioning period was in 1995, and, perhaps not surprisingly, that year also had the shortest duration of individual provisioning. This suggests that individual provisioning of females in 1999, when the overall duration of the provisioning period was 5 days shorter than that of 1995, was probably even shorter than that of 1995.

Fig. 4.27 Variation in
seasonal changes in
numbers of female
P. japonensis and available
good drupes in the foraging
area during four different
years. Symbols above
arrows represent days of
torrential rain. Dotted line in
female count for 1995 is
estimation. Adapted from
Filippi et al. (2002)
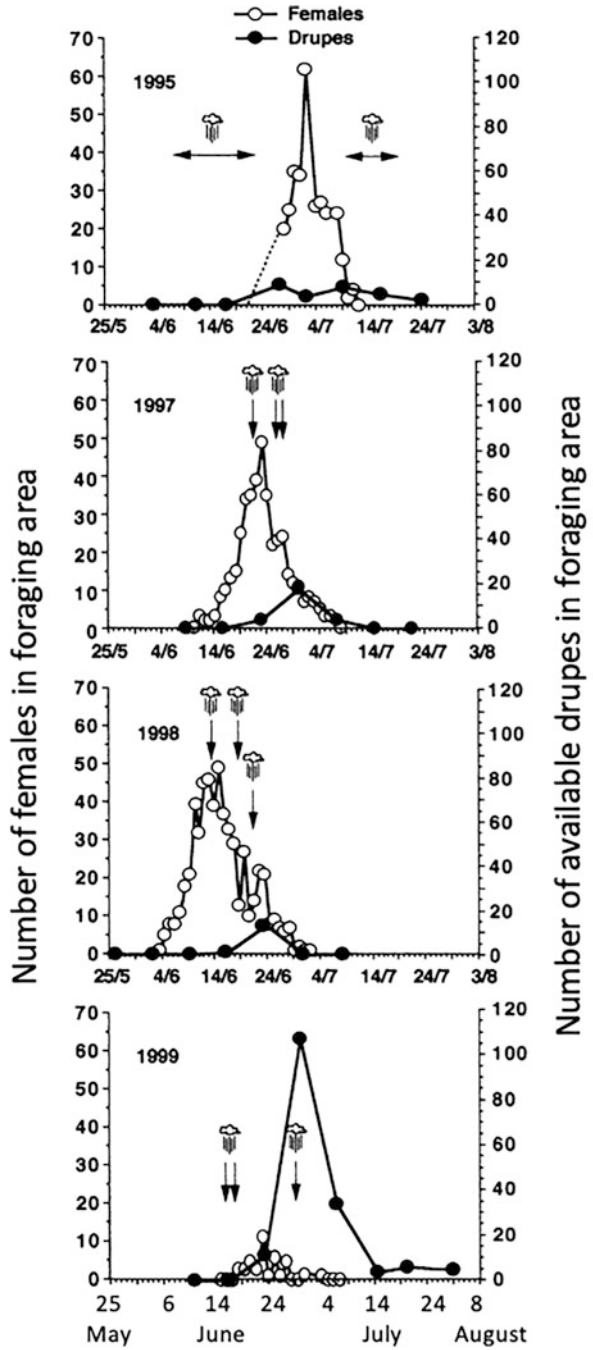
4.19.2   *Variation in* **S. jasminodora** *Drupe Availability and Weather During the Provisioning Season*

The data for the 4 years show interesting and varied patterns (Fig. 4.27; Filippi et al. 2002). The numbers of total and good drupes collected in the seed traps over the course of the provisioning season for each of the 4 years are presented in Fig. 1.10. The total numbers of drupes collected varied from 120 to 270 over the 4 years, but the percentage of good drupes varied from 5 to 95%, and the year that 270 drupes were collected (1998) had the lowest percentage of good drupes (5%). 1995 and 1999 had similar numbers of drupes (~180), but in 1995, only 19.4% were good, while in 1999, over 95% of the drupes were good! (Fig. 4.27; Filippi et al. 2002). Of course, females did not have provisioning access to drupes that were still on the host tree and could only provision drupes that had fallen to the ground; the torrential rains that preceded the peaks in the drupe numbers likely brought the drupes down and made them available for provisioning. An interesting pattern that would have been a serious constraint to provisioning females is that in most years the numbers of provisioning females were very high relative to the numbers of drupes and the peak number of females searching for drupes to provision occurred *before* the peak number of good drupe availability on the ground (Fig. 4.27; Filippi et al. 2002). Of course, we only collected drupes from a small portion of the entire crown, so there were surely more drupes for females to provision than the very small numbers presented in the graph; however, clearly the availability of good drupes is poor, at best, most years when females are provisioning. It was particularly poor in 1998 when females were provisioning for nearly 3 weeks before good drupes were collected in the seed traps. This would severely constrain provisioning capacity and offspring success. Good drupes were more abundant when nymphs were out foraging, but unless mothers can provision the nest to keep the young safe until the mid-third instar, offspring success will be severely limited. Notably, in 1999, the pattern was reversed. There were very few provisioning females, probably because of frequent nest failure the previous year, and a superabundance of good drupes. It is likely the abundance of drupes in 1999 while females were provisioning meant that females could find good drupes easily, had little competition for them, and were able to find and stockpile many drupes in a much shorter period of time. This would also explain the shorter duration of the provisioning period that year (Fig. 4.27). Interestingly, early females comprised between 60 and 66% of all provisioning females each year, and for each of the 3 years, the early provisioners always provisioned significantly longer than late females: (mean ± S.E.) 3.99 ± 0.27 days ($n = 73$) and 2.29 ± 0.25 days ($n = 41$) for early and late females, respectively, in 1995 (Mann-Whitney U test, $p < 0.001$); 8.5 ± 0.58 days ($n = 93$) and 4.0 ± 0.4 days ($n = 63$) for early and late females, respectively, in 1997 ($p < 0.001$); and 6.75 ± 0.48 days ($n = 91$) and 4.65 ± 0.37 days ($n = 46$) for early and late females, respectively ($p < 0.05$) (Filippi et al. 2002). It is likely that the paucity of good drupes earlier in the season meant that it took longer for females to find and provision their nests with enough drupes to sustain the young to a safer stage.

4.19.3   Intensity of Competition for the Resource, Weather, and Provisioning Capacity of Females

As a measure of the intensity of intraspecific resource competition for provisioning females in each of the 4 years, we divided the total number of good drupes collected over each provisioning season by the highest number of females counted among all the censuses in the foraging area for that season (female density index, FDI). A larger FDI indicates a lower intensity of intraspecific competition. FDI values ranged from 0.28 (1998) to 13.92 (1999) (Table 4.7). The findings indicate that there is enormous variation in intraspecific competition from year to year. Remarkably, intraspecific competition was 25 times greater in 1995 and 1997 and 50 times greater in 1998 than it was in 1999. These differences apparently drive population fluctuations from year to year. Interestingly, although the FDI values for 1995 and 1997 were very similar, the average provisioning capacity of females in 1995 was about 1/3 of that in 1997. In fact, the average provisioning capacity of females in 1995 was very similar to that of females in 1998, when the FDI was only 0.28, which was less than half that in 1995. We attribute the poor provisioning capacity in 1995 to the torrential rain that persisted for 2 weeks in early June, at least 1 week of which was during the time females would have been provisioning, and then again for about 5 days late in the provisioning season (Fig. 4.27). As mentioned, the entire nesting season from the egg stage through provisioning coincides with the rainy season. Daily rain is normal and expected, but persistent torrential rains are less common and certainly would disrupt provisioning behavior and even lead more directly to nest destruction because of flooding.

In fact, it is surprising that the population did not crash the following year because of the number of consecutive days with torrential rain in 1995. The first week of torrential rain occurred when females were sheltered in their nests under the leaf litter guarding their eggs. It seems not too many nests got washed away. Provisioning females during that second week in early June would not have fared as well. It is also of interest that despite the very short provisioning season in 1999 (16 days), females were able to provision nests with so many drupes. Undoubtedly, the very large FDI that year meant that females had a very easy time finding good drupes, making many

Table 4.7 Drupe availability, female density and provisioning capacity for four provisioning seasons

	Number of good drupes[a]	Number of females[b]	FDI	No. of drupes provided (mean $\pm$ SE) (n)
1995	36	62	0.58	10.9 $\pm$ 1.9 (11) a
1997	26	48	0.54	31.4 $\pm$ 4.6 (22) b
1998	14	50	0.28	11.2 $\pm$ 2.5 (34) a
1999	167	12	13.92	36.6 $\pm$ 9.2 (8) b

FDI female density index (no. good drupes/female). Numbers followed by different letters were significantly different (Kruskal-Wallis, $p < 0.001$). Adapted from Filippi et al. (2002)
[a] Total number of good drupes collected in seed traps (see text)
[b] Number of females at peak density (see text)

Fig. 4.28 *S. jasminodora* seedlings emerging in 2000 from drupes in a nest from the 1999 nesting season. Nest was 4.56 m outside the range of fallen drupes

round trips to the foraging area and bringing many to the nest in a very short period of time. At this field site, we have seen such a high proportion of good drupes available to provisioning females only twice, about once every 10 years. Anecdotally, and of considerable interest, is what we observed the following year (2000). We found several tiny *S. jasminodora* seedlings emerging from drupes in one of the 1999 nests that was 4.56 m away from the foraging area (Fig. 4.28)! Clearly, those drupes were not fed on and had been spared. Females keep provisioning until they literally drop dead, and with the great number of good drupes in 1999, they apparently provisioned more than was needed before dying of exhaustion and ending the season quite early. Now for the interesting question. Were there so many good drupes simply because there were so few females feeding on the host tree, thus leaving many more drupes undamaged and able to ripen fully before falling to the ground? Or, more intriguing, is the host tree deliberately manipulating the production of good drupes so that the *P. japonensis* population will crash every 10 years or so, allowing some of the over abundantly provisioned drupes that have been dragged far from the host tree to germinate? Coevolution? This is a tough question to answer as the life span of the tree is long and it is not amenable to cultivating outside its natural habitat (alas, we have tried!). It would be a brilliant strategy for the tree, because seedlings cannot emerge under the crown, likely because of allelochemicals produced by the mother tree, and we have never seen any other potential seed disperser, such as a bird or mammal, feed on the nasty tasting drupes.

We counted the number of drupes in marked nests at the end of the nesting season. The range and degree of variation among females in provisioning capacity varied considerably within a year and over the 4 years (Fig. 4.29; adapted from
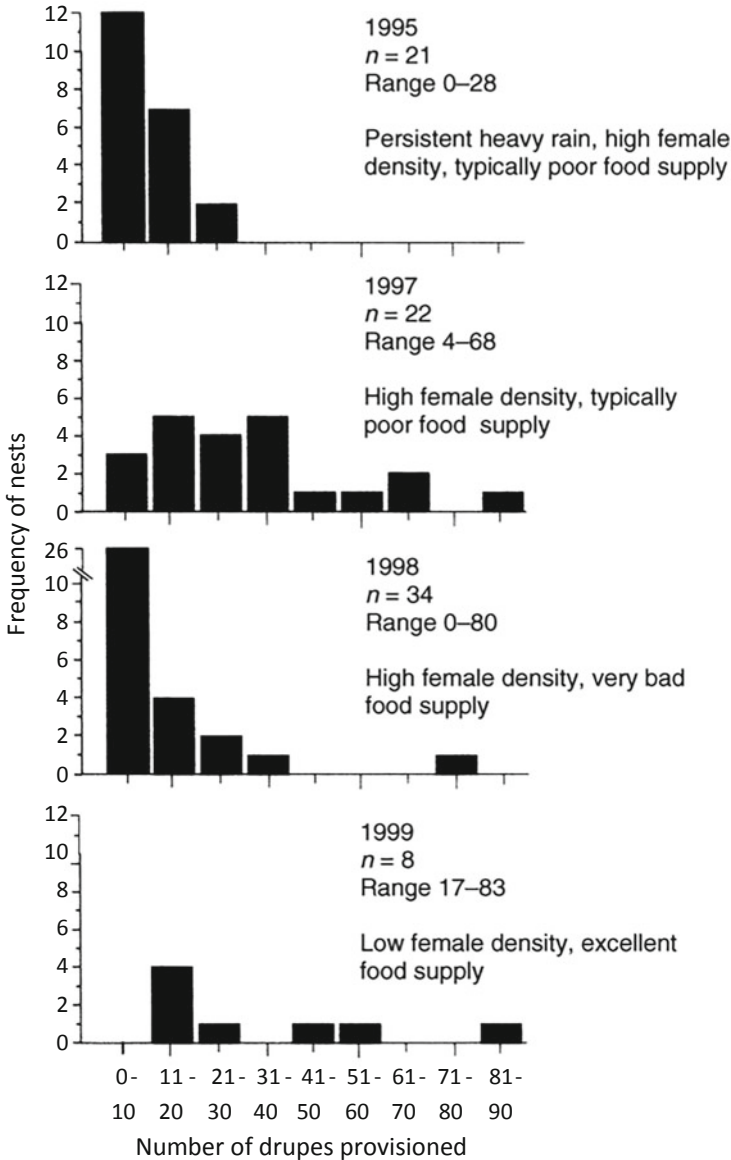
Fig. 4.29 Frequency distribution of drupe provisioning success of females at the end of four provisioning seasons. Adapted from Filippi et al. (2002)

Filippi et al. 2002). In 1995 and 1998, 57% and 76%, respectively, of females provisioned less than or equal to ten drupes. Moreover, three females in 1995 and two females in 1998 were unable to provision any drupes at all. Except for one super successful female in 1998, the range and pattern of variation in provisioning capacity

were similar in both years. No females in 1995 and only two females in 1998 managed to provision more than 30 drupes. These 2 years had severe, identifiable constraints (weather in 1995 and very intense competition because of very poor resource level in 1998) that were likely responsible for the poor provisioning.

The range of provisioning capacity in both 1997 and 1999 was much broader and more evenly distributed (Fig. 4.29). In 1997, a typically poor year with no extreme weather issues, 86% of the females provisioned greater than or equal to 11 drupes, and nearly half of the females provisioned more than 30 drupes. Remarkably, in 1999, 100% of the females provisioned more than 17 drupes, and nearly half provisioned more than 40 drupes.

4.19.4   Variation of Nest Distribution over the Four Nesting Seasons

As predicted, females had an overwhelming preference for nesting outside the foraging area (Fig. 4.26; Table 4.8), particularly when there were no severe stressors. Preference in some years was also apparently dependent on the stage in the provisioning season (early or late) that the female was active. A similar variation in nest site preference was seen in the trophic egg study (Filippi et al. 2012). As predicted, most females in all years nested outside the foraging area, and females only nested inside the foraging area in the two particularly stressful years (1995 and 1998). There were significantly different nesting patterns (inside/outside for early and late nesters) for each of those 2 years (Fisher's exact test, $p < 0.01$ for both years).

Table 4.8 Frequency of early and late nests inside and outside the foraging area

	Stage in provisioning season		
	Nest site	Early	Late
1995[a]	Inside	6	0
	Outside	5	10
1997	Inside	0	0
	Outside	14	8
1998[a]	Inside	0	6
	Outside	20	8
1999	Inside	0	0
	Outside	6	2

[a] Fisher's Exact test, $p < 0.01$. Adapted from Filippi et al. (2002)

4.19.5 Variation in Provisioning Capacity Based on Nest Location and Active Timing

As predicted, opting for the riskier behavior led to greater provisioning capacity under stressful conditions (Fig. 4.30; Filippi et al. 2002). In terms of timing of provisioning activity during the provisioning season, later should be riskier because the provisioning female would have more intraspecific competition and her young would be competing with older nymphs when they got to the foraging area, so foraging later in the season should be the preferred option under severely constrained conditions. The rain in 1995 was an unpredictable event that a female would not have had information about before she laid her eggs. However, in 1998, females would have been able to assess food availability and intraspecific competition while feeding on the host tree to make their eggs. In 1998, good drupes were not available until late in the season, so females nesting later had an advantage while provisioning, and they had a strong tendency to provision more drupes; however, the difference was not significant (Fig. 4.30; range 0–22 and 1–80 for early and late females, respectively). Interestingly, in 1999 all females nested just outside the foraging area. They apparently did not perceive a problematic degree of intraspecific competition and disturbance to their nests because of the low density of females, and there was a
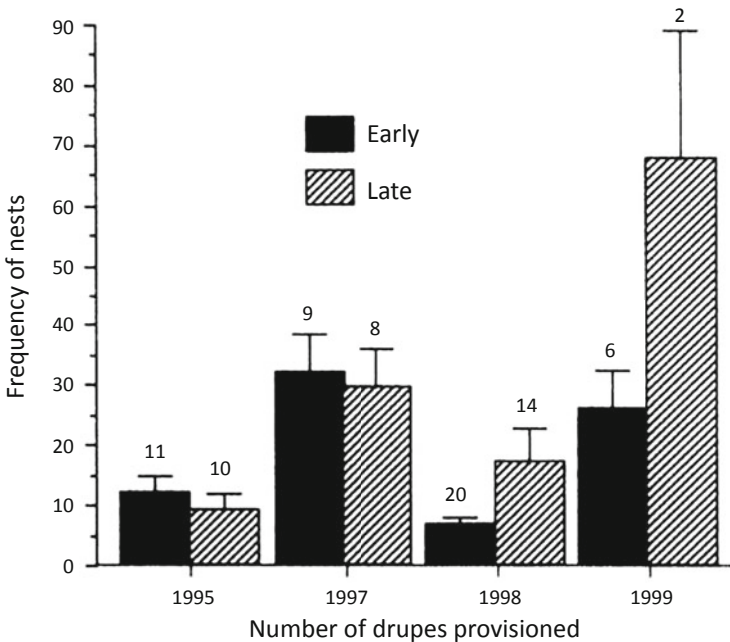

Fig. 4.30 Impact of active stage in provisioning season (early or late) and provisioning success of females in four provisioning seasons. Numbers above bars indicate sample size. Error bars represent S.E. Adapted from Filippi et al. (2002)

Table 4.9 Mean no. of drupes provisioned per early and late nest inside and outside the foraging area

		Average number of drupes provisioned per nest			
		Early	n	Late	n
1995	Inside	14.9 ± 4.0	6	–	–
	Outside	9.0 ± 3.2	5	9.4 ± 2.7	10
1998	Inside	–	–	6.8 ± 1.9 a	6
	Outside	6.8 ± 1.2 a	20	25.4 ± 8.9 b	8

Means ± SE. Numbers within group followed by different letters are significantly different (Kruskal-Wallis with *post hoc* Bonferroni adjustment $p < 0.01$. Adapted from Filippi et al. (2002)

huge surge in drupes later in the season (Fig. 4.27), so provisioning later was the best strategy that year. Late females provisioned more than twice as many drupes on average than early nesting females, but because of the small sample size, statistical analysis was not possible (Fig. 4.27). There was no difference in the number of drupes provisioned by early and late females when there was no severe resource constraint (1995 and 1997) (range, 0–27 and 0–28 for 1995 early and late females, respectively; 2–68 and 4–57 for 1997 early and late females, respectively). Apparently, risk-sensitive nesting in terms of timing in the provisioning season is only realized under extreme resource conditions.

There were only 2 years to compare where nests had been found both inside and outside the foraging area (1995 and 1998). We compared the provisioning success of early and late females both inside and outside the foraging area for those years (Table 4.9). We predicted that females nesting and provisioning according to the risk-sensitive constraints proposed would provision more drupes. While there was no significant difference in 1995 between early and late females (Fig. 4.30; Table 4.9), physiological constraints that we did not assess (fat body content, etc.) could have made the strategy used optimal. In 1998, there was no difference in provisioning capacity between early/outside and late/inside nests. However, there was a significant difference between late inside and outside nests. In 1998, there were very few good drupes available early in the season. It appears that provisioning late was optimal that year and females nesting inside the foraging area might have suffered from thieving females. We will cover this in detail in Chap. 6; however, it bears mentioning here that females were observed entering other females' nests and removing drupes that the resident female had provisioned. We expect that females nesting within the foraging area should experience thievery more often because of the high density of females foraging there and the lesser amount of leaf litter and ground cover to conceal the nests.

4.19.6 Conclusions Regarding Risk-Sensitive Decision-Making

We conclude that *P. japonensis* females show flexibility in their behavioral and physiological patterns and apparently use risk-prone behavior under *extreme* ecological conditions. In other words, hypothesis 1 was supported under the constraints of severe ecological conditions of severe weather (persistent torrential rain) and extreme resource constraint, assessed by both absolute abundance and intraspecific competition (Figs. 4.28 and 4.29; Tables 4.7, 4.8, and 4.9), but also under extreme resource abundance. Females opted for risk-prone responses and provisioned more when exposed to these conditions. The flexibility was still present under moderate conditions; however, it resulted in no difference in provisioning capacity. Adaptive variation among individuals can be maintained in a population, particularly in species with as little dispersal as *P. japonensis*. Risk-sensitive behavior could become prevalent over evolutionary time and be maintained when the benefits allow individuals to respond favorably to unpredictable and stressful conditions (Clutton-Brock 1991; Budaev 1999).

Hypothesis 2 was also supported: the riskier nest site (inside the foraging area) was only chosen when conditions were constrained and led to good provisioning success (Tables 4.8 and 4.9). In support of the third hypothesis that females would make physiological adjustments such that they were provisioning when drupes were most abundant, females that nested later when the resource was not available until late in the provisioning season provisioned more drupes than early females (Fig. 4.30 and Table 4.9). Support for the first three hypotheses results in support for the fourth hypothesis that females adjusting the timing and location of their provisioning activities according to the constraints outlined provision the most drupes (Figs. 4.27, 4.28, and 4.29 and Tables 4.7, 4.8, and 4.9).

The extreme behavior of progressive provisioning is essential to *P. japonensis* offspring success (Filippi et al. 2000), and females seem to have an "on" switch to begin provisioning triggered by the hatching of their eggs but no "off" switch. They provision until they die of exhaustion, even when their offspring have already left the nest. In this semelparous species, which literally puts all its eggs in one basket, evolution should favor any physiologically feasible adaptation that would enhance the provisioning capacity of females because of the overwhelming impact it has on offspring success. A major evolutionary innovation would be the ability to respond to the unpredictable and highly variable conditions that provisioning females encounter, in other words, risk-sensitive decision-making. We have presented drupe data for only 4 years here (Figs. 1.10 and 4.27) but collected drupe data for 8 years. The resource is not only ephemeral, but good drupe availability fluctuates from very poor to abysmal and is only occasionally (every 10 years) very good. It is under the backdrop of these poor resource and weather conditions that the physiology and behaviors of *P. japonensis* have evolved. We have described in Chap. 2 other ways that *P. japonensis* optimizes use of the resource, which is both rich in content when it is "good" and poor in terms of abundance. Only inseminated females

feed, the metabolic rate is dramatically suppressed, in females, especially, and males don't ever feed as adults but use their remaining reserves to mate with numerous females before they perish when those reserves are depleted. In the face of these extreme traits, it seems a matter of course that risk-sensitive decision-making, the ability to respond adaptively to the conditions one is faced with, should also evolve in this species.

In response to the great variation and unpredictability of the ecological conditions *P. japonensis* females have faced during the nesting season over their evolutionary history, it appears that natural selection has favored risk-sensitive decision-making in nesting behaviors as one additional mechanism to maximize fitness (Luttbeg and Warner 1999).

References

Agrawal, A.F., E.D. Brodie III, and J. Brown. 2001. Parent-offspring coadaptation and the dual genetic control of maternal care. *Science* 292: 1710–1712.

Alexander, R.D. 1974. The evolution of social behavior. *Annual Review of Ecology and Systematics* 5: 179–183.

Baba, N., M. Hironaka, T. Hosokawa, H. Mukai, S. Nomakuchi, and T. Ueno. 2011. Trophic eggs compensate for poor offspring feeding capacity in a subsocial burrower bug. *Biology Letters* 7: 194–196.

Baur, B. 1992. Cannibalism in gastropods. In *Cannibalism: Ecology and evolution among Diverse Taxa*, ed. M.A. Elgar and B.J. Crespi, 102–126. Oxford: Oxford Univ. Press.

Becqueart, J. 1935. Presocial behavior among the Hemiptera. *Bulletin of the Brooklyn Entomological Society* 30: 177–191.

Boos, S., J. Meunier, S. Pichon, and M. Kölliker. 2014. Maternal care provides antifungal protection to eggs in the European earwig. *Behavioral Ecology* 25: 754–761. https://doi.org/10.1093/beheco/aru046.

Bradshaw, A.D. 1965. Evolutionary significance of phenotypic plasticity in plants. *Advances in Genetics* 13: 115–155.

Brussard, L., and W.J.F. Visser. 1987. Dung exploitation by the dung beetle *Typhaeus typhoeus* (Col., Geotrupidae). *Oecologia (Berlin)* 72: 21–27.

Budaev, S.V. 1999. Individual differences in parental care and behaviour profile in the convict cichlid: A correlation study. *Animal Behaviour* 58: 195–202.

Bulmer, M. 1994. *Theoretical evolutionary ecology*. Sunderland, MA: Sinauer Associates.

Caraco, T. 1980. On foraging time allocation in a stochastic environment. *Ecology* 61: 119–128.

Choe, J.C. 1989. Maternal care in *Lahidomera suturella* Chevrolat (Coleoptera: Chrysomelidae: Chrysomelinae) from Costa Rica. *Psyche* 96: 63–67.

Choe, J.C., and B.J. Crespi. 1997. *The evolution of social behaviour in insects and arachnids*. Cambridge, UK: Cambridge University Press.

Clutton-Brock, T.H. 1991. *The evolution of parental care*. Princeton, NJ: Princeton University Press.

Costa, J.T. 2006. *The other insect societies*. Cambridge, MA and London, England: The Belknap Press of Harvard University Press.

Creighton, J. 2005. Population density, body size, and phenotypic plasticity of brood size in a burying beetle. *Behavioral Ecology* 16: 1031–1036.

Crespi, B.J. 1992. Cannibalism and trophic eggs in subsocial and eusocial insects. In *Cannibalism: Ecology and evolution among Diverse Taxa*, ed. M.A. Elgar and B.J. Crespi, 176–213. Oxford: Oxford University Press.

Davies, N.B., J.R. Krebs, and S.A. West. 2012. *An introduction to behavioral ecology*. 4th ed. Chichester, West Sussex, UK: Wiley-Blackwell, Blackwell Publishing.

Diesel, R. 1992. Maternal care in the bromeliad crab, *Metopaulias depressus*: Protection of larvae from predation by damselfly nymphs. *Animal Behaviour* 43: 803–812.

Eggert, A.-K., and S.K. Sakaluk. 1995. Female-coerced monogamy in burying beetles. *Behavioral Ecology and Sociobiology* 37: 147–153.

Eggert, A.K., and J.K. Müller. 1997. Biparental care and social evolution in burying beetles: Lessons from the larder. In *Social behavior in insects and arachnids*, ed. J.C. Choe and B.J. Crespi, 216–236. Cambridge: Cambridge University Press.

Eggert, A.K., M. Reinking, and J.K. Müller. 1998. Parental care improves offspring survival and growth in burying beetles. *Animal Behaviour* 55: 97–107.

Eickwort, G.C. 1981a. Presocial Insects. In *Social Insects*, ed. H.R. Hermann, vol. 11, 199–280. New York: Academic Press.

Eickwort, G.C. 1981b. Presocial insects. In *Social insects*, ed. H.R. Hermann, vol. II, 199–280. New York: Academic Press.

Filippi-Tsukamoto, L., S. Nomakuchi, K. Kuki, and S. Tojo. 1995. Adaptiveness of parental care in *Parastrachia japonensis* (Hemiptera: Cydnidae). *Annals of the Entomological Society of America.* 88: 374–383.

Filippi, L., M. Hironaka, S. Nomakuchi, and S. Tojo. 2000. Provisioned *Parastrachia japonensis* (Hemiptera: Cydnidae) nymphs gain access to food and protection from predators. *Animal Behaviour* 60: 757–763.

Filippi, L., M. Hironaka, and S. Nomakuchi. 2002. Risk-sensitive decisions during nesting may increase maternal provisioning capacity in the subsocial shield bug, *Parastrachia japonensi*s. *Ecological Entomology* 27: 152–162.

Filippi, L., M. Hironaka, and S. Nomakuchi. 2005. Kleptoparasitism and the effect of nest location in a subsocial shield bug Parastrachia japonensis (Hemiptera: Parastrachiidae). *Annals of the Entomological Society of America* 98: 134–142.

Filippi, L., N. Baba, I. Kyoichi, M. Hironaka, and S. Nomakuchi. 2009. Pre- and post-hatch trophic egg production in the subsocial burrower bug, *Canthophorus niveimarginatus* (Heteroptera: Cydnidae). *Naturwissenschaften* 96: 201–211.

Filippi, L., M. Hironaka, and S. Nomakuchi. 2012. Individual variation in trophic egg production: Evidence for maternal manipulation in response to resource and competition levels. *Ethology* 118: 503–510.

Fordyce, J.A. 2006. The evolutionary consequences of ecological interactions mediated through phenotypic plasticity. *The Journal of Experimental Biology* 209: 2377–2383.

Gilbert, J.D.J., and A. Manica. 2015. The evolution of parental care in insects: A test of current hypotheses. *Evolution* 69: 1255–1270.

Godfray, H.C.J. 1995. Evolutionary theory of parent-off-spring conflict. *Nature (London)* 376: 133–138.

Grimaldi, D., and M.S. Engel. 2005. *Evolution of the insects*. New York, NY: Cambridge University Press.

Gyôtoku, N., and S. Tachikawa. 1980. Life history of *Parastrachia japonensis* (Scott) (Cydnidae: Sehirinae). *Rostria* 33: 359–368. (in Japanese).

Halffter, G. 1997. Subsocial behavior in Scarabaeinae beetles. In *Social behavior in insects and arachnids*, ed. J.C. Choe and B.J. Crespi, 237–259. Cambridge, UK: Cambridge University Press.

Hironaka, M., S. Nomakuchi, S. Iwakuma, and L. Filippi. 2005. Trophic egg production in a subsocial shield bug, and its functional value. *Ethology* 111: 1089–1102.

Hunt, J., and L.W. Simmons. 1998. Patterns of parental provisioning covary with male morphology in a horned beetle (*Onthophagus taurus*) (Coleoptera: Scarabaeidae). *Behavioral Ecology and Sociobiology* 42: 447–451.

Inadomi, K., M. Wakiyama, M. Hironaka, H. Mukai, L. Filippi, and S. Nomakuchi. 2014. Post-ovipositional maternal care in the burrower bug, *Adomerus rotundus* (Heteroptera: Cydnidae). *Canadian Entomologist* 146: 211–218.

Iwanishi, S., E. Hasegawa, and K. Ohkawara. 2003. Worker oviposition and policing behaviour in the myrmicine and *Aphaenogaster smithiesi* japonica Forel. *Behaviour* 66: 513–519.

Kam, Y.C., Y.H. Chen, T.C. Chen, and I.R. Tsai. 2000. Maternal care of an arboreal breeder, *Chirixalus eiffingeri* (Anura: Rhacophoridae) from Taiwan. *Behaviour* 137: 137–151.

Kight, S. 1997. Factors influencing maternal behaviour in a burrower bug, *Sehirus cinctus* (Heteroptera: Cydnidae). *Animal Behaviour* 53: 105–115.

Kim, K.W., and C. Roland. 2000. Trophic egg laying in the spider, *Amaurobius ferox*: Mother–offspring interactions and functional value. *Behavioural Processes* 50: 31–42.

Koedam, D., M.H.W. Velthuis, M.R. Dohmen, and V.L. Imperatriz-Fonesca. 2001. The behaviour of laying workers and the morphology and viability of their eggs in *Melipona bicolor bicolor*. *Physiological Entomology* 26: 254–259.

Kudo, S. 1990. Brooding behavior in *Elasmucha putoni* (Heteroptera: Acanthosomatidae), and a possible nymphal alarm substance triggering guarding responses. *Applied Entomology and Zoology* 25 (4): 431–437.

Kudo, S.-I., M. Satô, and M. Ôhara. 1989. Prolonged maternal care in *Elasmucha dorsalis* (Heteroptera: Acanthosomatidae). *Journal of Ethology* 7: 75–81.

Kudo, S., and T. Nakahira. 2004. Effects of trophic-eggs on offspring performance and rivalry in a sub-social bug. *Oikos* 107: 28–35.

Kukuk, P.F. 1992. Cannibalism in social bees. In *Cannibalism: Ecology and evolution among Diverse Taxa*, ed. M.A. Elgar and B. Crespi, 214–237. Oxford: Oxford Univ. Press.

Lack, D. 1947. The significance of clutch size. *Ibis* 89: 302–352.

Leigh, D., and P.T. Smiseth. 2012. Parent-offspring conflict over the transition to independence in *Nicrophorus vespilloides*: Parental chemical cues and offspring begging. *Ethology* 118: 460–465.

Lima, S.L., and P.A. Bednekoff. 1999. Temporal variation in danger drives antipredator behavior: The predation risk allocation hypothesis. *American Naturalist* 153: 649–659.

Lima, S.L., and L.M. Dill. 1990. Behavioral decisions made under the risk of predation: A review and prospectus. *Canadian Journal of Zoology* 68: 619–640.

Luttbeg, B., and R.R. Warner. 1999. Reproductive decision-making by female peacock wrasses: Flexible versus fixed behavioral rules in variable environments. *Behavioral Ecology* 10: 666–674.

Machado, G., and S. Trumbo. 2018. Parental care. In *Insect behavior, from mechanisms to ecological and evolutionary consequences*, ed. A. Cordoba-Aguilar, D. Gonzalez-Tokman, and I. Gonzalez-Santoyo. Oxford, UK: Oxford University Press.

Matsuura, K., T. Tamura, N. Kobayashi, T. Yashiro, and S. Tatsumi. 2007. The antibacterial protein lysozyme identified as the termite egg recognition pheromone. *PLoS One*. https://doi.org/10.1371/journal.pone.0000813.

Miyamoto, S. 1965. *The red stinkbug. Primary Color Insect Great Encyclopedia, III*. Hokuryukan, Tokyo. (in Japanese).

Meunier, J., and M. Kölliker. 2012. Parental antagonism and parent-offspring co-adaptation interact to shape family life. *Proceedings of the Royal Society B: Biological Sciences* 279: 3981–3988.

Michener, C.D. 1969. Comparative social behavior of bees. *Annual Review of Entomology* 14: 299–342.

Mock, D.W., and G.A. Parker. 1997. *The evolution of sibling rivalry*. Oxford, UK: Oxford University Press.

Monaghan, P., and R.G. Nager. 1997. The significance of clutch size. *Trends in Ecology & Evolution* 12: 270–274.

Mukai, H., M. Hironaka, N. Baba, T. Yanaki, K. Inadomi, L. Filippi, and S. Nomakuchi. 2010. A report of maternal care behavior in *Adomerus variegates* (Heteroptera: Cydnidae). *Canadian Entomologist* 142: 52–56.

Nafus, D.M., and I. Schreiner. 1988. Parental care in a tropical nymphalid butterfly Hypolimnas anomala. *Animal Behaviour* 36: 1425–1431.

Nakahira, T. 1992. Reproductive history and parental behaviour in the cydnid bug *Adomerus triguttulus*. M.Sc. thesis, Department of Agriculture, Hokkaido University, Sapporo, Japan.

———. 1994. Trophic egg production in the subsocial burrower bug *Admerus* (sic) *triguttulus*. *Naturwissenschaften* 81: 413–414.

Nakahira, T., and Si Kudo. 2008. Maternal care in the burrower bug *Adomerus triguttulus*: Defensive behavior. *Journal of Insect Behavior* 21: 306–316. https://doi.org/10.1007/s10905-008-9129-0.

Nalepa, C.A., and W.J. Bell. 1997. Postovulation parental investment and parental care in cockroaches. In *The evolution of social behavior in insects and arachnids*, ed. J.C. Choe and B.J. Crespi, 26–51. Cambridge, U.K.: Cambridge University Press.

Nalepa, C.A., K. Maekawa, K. Shimada, Y. Saito, C. Arellano, and T. Matsumoto. 2008. Altricial development in subsocial wood-feeding cockroaches. *Zoological Science* 25: 1190–1198.

Nomakuchi, S., L. Filippi, and S. Tojo. 1998. Selective foraging behavior in nest-provisioning females of *Parastrachia japonensis* (Hemiptera: Cydnidae): Cues for preferred food. *Journal of Insect Behavior* 11: 605–619.

Nomakuchi, S., L. Filippi, and M. Hironaka. 2001. Nymphal occurrence pattern and predation risk in the subsocial shield bug, *Parastrachia japonensis* (Heteroptera: Cydnidae). *Applied Entomology and Zoology* 36: 209–212.

Nomakuchi, S., L. Filippi, S. Iwakuma, and M. Hironaka. 2005. Variation in the start of nest abandonment in the subsocial shield bug *Parastrachia japonensis* (Hemiptera: Parastrachiidae). *Annals of the Entomological Society of America* 98: 134–142.

Odhiambo, T.R. 1959. An account of parental care in *Rhinocoris albopilosus* Signoret (Hemiptera-Heteroptera: Reduviidae) with notes on its life history. *Proceedings of the Royal Entomological Society of London A* 34: 175–182.

Parker, A.H. 1965. The maternal behavior of *Pisilus tipuliformis* Fabricus (Hemiptera: Reduviidae). *Entomologia Experimentalis et Applicata* 8: 13–19.

Perry, J.C., and B.D. Roitberg. 2006. Trophic egg laying; hypotheses and tests. *Oikos* 112: 706–714.

Polis, G.A. 1981. The evolution and dynamics of intraspecific predation. *Annual Review of Ecology and Systematics* 12: 225–251.

———. 1984. Intraspecific predation and infant killing among invertebrates. In *Infanticide. Comparative and evolutionary perspectives*, ed. G. Hausfater and S. Blaffer Hrdy, 87–104. New York: Aldine.

Ralston, J.S. 1977. Egg guarding by male assassin bugs of the genus *Zelus* (Hemiptera: Reduviidae). *Psyche* 84: 103–107.

Royle, N.J., P.T. Smiseth, and M. Kölliker. 2012. *The evolution of parental care*. Oxford, UK: Oxford University Press.

Royle, N.J., A.F. Russell, and A.J. Wilson. 2014. The evolution of flexible parenting. *Science* 345: 776–781.

Sato, H. 1997. Two nesting behaviours and life history of a subsocial African dung-beetle, *Scarabaeus catenatus* (Coleoptera: Scarabaeidae). *Journal of Natural History* 31: 457–469.

Sites, R.W., and J.E. McPherson. 1982. Life history and laboratory rearing of *Sehirus cinctus* (Hemiptera: Cydnidae) with descriptions of immature stages. *Annals of the Entomological Society of America* 75: 210–215.

Smiseth, P.T., and A.J. Moore. 2002. Does resource availability affect offspring begging and parental provisioning in a partially begging species? *Animal Behaviour* 63: 577–585.

Stork, N.E. 2018. How many species of insects and other terrestrial arthropods are there on the Earth? *Annual Review of Entomology* 63: 31–45.

Tachikawa, S., and C.W. Schaefer. 1985. The biology of *Parastrachia japonensis* (Hemiptera: Pentatomoidea: ?-idae). *Annals of the Entomological Society of America*. 78: 387–397. https://doi.org/10.1093/aesa/78.3.387.

Tachikawa, S. 1991. *Subsocial terrestrial heteropterans of Japan: Parent-offspring relationships in stink bugs*. Tokyo, Japan: Tokyo Agricultural University Press.

Tallamy, D.W., and R.F. Denno. 1981. Maternal care in *Gargaphia solani* (Hemiptera: Tingidae). *Animal Behaviour* 29: 771–778.

———. 1982. Life history trade-offs in *Gargaphia solani* (Hemiptera: Tingidae): The cost of reproduction. *Ecology* 63: 616–620.

Tallamy, D.W., and T.K. Wood. 1986. Convergence patterns in subsocial insects. *Annual Review of Entomology* 31: 369–390.

Tinbergen, N. 1963. On aims and methods of ethology. *Zeitschrift für Tierpsychologie* 20: 410–433.

Trivers, R.L. 1974. Parent-offspring conflict. *American Zoologist* 14: 249–263.

Trivers, R. 1985. *Social evolution*. Menlo Park, CA: Benjamin/Cummings.

Trumbo, S.T. 1996. Parental care in invertebrates. *Advances in the Study of Behaviour* 25: 3–51.

———. 2012. Patterns of parental care in invertebrates. In *The evolution of parental care*, ed. N.J. Royle, P.T. Smiseth, and M. Kölliker. Oxford, UK: Oxford University Press.

Tsukamoto, L., and S. Tojo. 1992. A report of progressive provisioning in a stink bug, *Parastrachia japonensis* (Hemiptera: Cydnidae). *Journal of Ethology* 10: 21–29.

West, M.J., and R.D. Alexander. 1963. Subsocial behaviour in a burrowing cricket *Anurogryllus muticus* (De Geer). Orthoptera: Gryllidae. *The Ohio Journal of Science* 63: 19–24.

Whitman, D., and A. Agrawal. 2009. What is phenotypic plasticity, and why is it important? In *Phenotypic plasticity of insects: Mechanisms and consequences*, ed. D. Whitman and T.N. Ananthakrishnan, 1–63. Enfield, NH: Science Publishers.

Wilson, E.O. 1971. *The insect societies*. Cambridge, Mass: Harvard University Press.

———. 1975. *Sociobiology: The new synthesis*. Cambridge, MA: Harvard University Press.

Wong, J.W.Y., J. Meunier, and M. Kölliker. 2013. The evolution of parental care in insects: The roles of ecology, life history and the social environment. *Ecological Entomology* 38: 123–137.

Wood, T.K. 1976. Biology and presocial behavior of *Platycotis vitritta* (Homoptera: Membracidae). *Annals of the Entomological Society of America* 69: 807–811.

Wyatt, T.D., and W.A. Foster. 1989. Leaving home: Predation and the dispersal from the maternal burrow of *Bledius spectabilis*, a subsocial intertidal beetle. *Animal Behaviour* 38: 778–785.

Lisa Filippi initiated the work on *Parastrachia japonensis* at Mt. Hinokuma Park in Kanzaki Town, Japan, where she earned her M.S. and doctoral degrees elucidating many of the amazing physiological and behavioral traits of this fascinating insect through field and laboratory studies. She is currently a Professor of Biology at Hofstra University in New York.

Mantaro Hironaka in fact applied to Saga University in order to eventually join the *P. japonensis* research team. He proved to be a brilliant, enthusiastic, energetic, and creative member of the team who contributed significantly to a variety of behavioral studies. Mantaro went on to get his doctoral degree in Dr. Tojo's laboratory from the United Graduate School of Agriculture, Kagoshima University, focusing on the neurological processes involved in the complex navigation that *P. japonensis* uses when provisioning. He is currently an Associate Professor at the Department of Applied Entomology, Faculty of Bioproduction Science, Ishikawa Prefectural University.

Shintaro Nomakuchi an ecologist focusing on insect behavior, came to Saga University in 1993 as an Associate Professor, and was keen to share his very valuable expertise and join the *P. japonensis* research team, which he eventually headed. The work on clarifying the quality of the habitat, the techniques for monitoring the subpopulations and for carrying out a variety of ecological and behavioral studies, as well as the methods for performing robust data analyses, all got a hefty boost, and we were off and running. He is a Professor Emeritus of the Faculty of Agriculture at Saga University.