No fitness consequence of experimentally induced polyandry in a monandrous wasp

Fitness from polyandry in a monandrous wasp

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Summary

Tests of the effects of multiple mating by females on female fitness have been primarily with polyandrous species, where a benefit to multiple mating has usually been found. In contrast, no such benefit was found here for the parasitic wasp *Spalangia endius*, a highly monandrous species. Females that mated only once prior to oviposition exhibited a rapid decline in daughter production long before they died. The production of daughters, but not sons, is sexual in this species, i.e., requires sperm. Nevertheless, females with greatly decreased daughter production did not then remate. When such females were experimentally manipulated into copulating with a second male, additional sperm were stored in the females' sperm storage organs. However, this sperm increase had no significant effect on daughter production, total offspring production or longevity. There was no evidence that either immediate or delayed polyandry currently benefits females. The lack of benefit may be a general feature of highly monandrous species or a common feature of parasitic hymenopterans regardless of mating system.

Keywords: monandry; multiple mating; polyandry; receptivity; parasitoid wasp; sex ratio; sperm storage

Introduction

The number of times a female mates can affect the evolutionary fitness of at least three parties, the female, her first mate, and subsequent male suitors (Chapman et al., 2003; Wedell et al., 2006). However, the fitness effects of her mating multiply are unlikely to be equal among the parties, creating the potential for conflict (Trivers, 1974; Parker, 1979; Arnqvist & Rowe, 2005). A male benefits from mating with a female and by preventing subsequent male suitors from doing so. However, preventing her from remating will conflict with her interests if she benefits by mating multiply. Mating multiply (polyandry) may benefit her by ensuring more, better or more genetically diverse sperm or by ensuring more food for her or her offspring (e.g., references in Arnqvist & Nilsson, 2000). Alternatively, mating just once (monandry) may benefit her if she receives sufficient sperm with one mating and mating is costly, as when seminal fluids or male structures cause harm or when mating attempts interfere with a female's other activities, like feeding and oviposition (e.g., references in Arnqvist & Nilsson, 2000; Cordero & Eberhard, 2003). If a female benefits by mating just once, this may conflict with the interests of her subsequent suitors.

Here fitness effects of monandry versus polyandry are examined in a highly monandrous species, the nonsocial parasitic hymenopteran *Spalangia endius*. Fitness effects of multiple mating by females have been reviewed for insects in general (Arnqvist & Nilsson, 2000), as well as for social hymenopterans (Crozier & Fjerdingstad, 2001) and for lepidopterans (Torres-Vila et al., 2004). As Torres-Vila et al. (2004) note, most species in these reviews are polyandrous, as appears also to be the case for studies of fitness effects of monandry versus polyandry in other taxa (Simmons, 2005). These reviews show that a benefit to multiple mating in polyandrous species is common. However, whether this is equally true for monandrous species is less clear (Torres-Vila et al., 2004; Baer & Schmid-Hempel, 2001, 2005; Arnqvist & Andres, 2006). More studies of monandrous species are needed (Torres-Vila et al., 2004).

The dearth of studies of highly monandrous species has been at least in part for practical reasons: it is difficult to compare females that mate once to females that mate multiply in monandrous species (Torres-Vila et al., 2004). However, the biology of *S. endius* allowed the creation of females that had two fairly natural matings, even at two different times in their lives. Another reason for examining *S. endius* is that there have been relatively few evolutionary studies of mating in parasitic Hymenoptera, despite their being about 50,000 described species (Godfray, 1994).

S. endius is a small wasp that parasitizes the pupae of certain fly species (Rueda & Axtell, 1985). One wasp emerges per host. Male *S. endius* emerge before females and wait at the cluster of hosts for females to emerge (King et al., 2005). Both sexes are sexually mature at emergence, and neither sex is overtly aggressive. Males do not appear to provide females with any nutrition during mating (King, 2002a).

Upon emergence, less than 5% of females copulate with a second male, whereas males are polygynous (King et al., 2005). Males readily approach, and briefly fan their wings at, both virgin and mated females with no discrimination. However, males are then more likely to retreat without mounting and without mounted-courtship if the female is mated. Thus, mated females are described as unattractive. Unattractiveness here refers only

to the decrease in mounting, not to a decrease in initial attraction of males to females. Unattractiveness is relative to virgin females; mated females are still sometimes mounted. Besides being unattractive, mated females are also unreceptive, meaning that even if mounted, they rarely open their genital orifice (King et al., 2005). Opening is necessary for copulation. Copulation and the postcopulatory courtship that follow take about half a minute (King, unpublished work). Postcopulatory courtship is the period after copulation and prior to dismount, during which the male is vibrating; and it appears to be responsible for making females unattractive and unreceptive (King & Fischer, 2005).

Hymenopterans such as *S. endius* exhibit haplodiploidy, whereby females develop from fertilized eggs, and males from unfertilized eggs. Thus, even completely sperm-depleted females can produce sons. Nevertheless, being able to also produce daughters should increase a female's fitness by allowing her to adjust the sex of her offspring in response to environmental conditions (Charnov, 1982; Napoleon & King, 1999; King, 2002b).

The present study examines: 1) whether monandrous females exhibit a decline in daughter production as they oviposit, as might be expected from their using up their sperm supplies; 2) whether as their daughter production declines, they regain attractiveness and receptivity and thus remate, allowing them to resume earlier levels of daughter production; 3) whether copulating twice, either early in life or after daughter production has dropped, would increase the fitness of females; 4) whether remating after daughter production has dropped would result in additional sperm storage. This study showed that daughter production declines as mothers oviposit, but they do not remate and would not benefit from doing so.

Materials and methods

The *S. endius* were from a colony established from wasps collected in 1996 from Florida, U.S.A. and maintained using a natural host, *Musca domestica* pupae (King, 1988). Parasitized host pupae were individually isolated in glass test tubes prior to the wasps' emergence in order to obtain virgin wasps. To minimize variation in wasp body size, only "large" hosts as defined in King (1988) were used; and wasps were randomly assigned to treatments. Mated wasps were generated by placing a 0 d old virgin male and a 0 d old virgin female into a test tube and watching for copulation to occur. The hosts had pupated within the last 2 d when they were presented to females. Two-tailed P values are presented.

Decreased daughter production and remating

These two experiments were to determine if and when females begin to decrease their production of daughters and whether doing so causes females to remate. Mated males were used in one experiment and virgin males in the other because they respond differently (King et al., 2005); virgin males seem to be less sensitive than mated males to female unattractiveness and so are more likely to attempt to mate.

The first experiment included both once-mated females ($N = 19$) and virgin females ($N = 12$) and had two phases. In the first phase females were allowed to

oviposit for about a week in order to prepare them for remating tests in the second phase of this experiment. In addition, the once-mated females from the first phase were used to determine whether the daughter production of mated females declines over time. Each female was given hosts for life: 20 hosts for each of the first 3 d, 15 for each of the next 4 d, and then 10 hosts daily. These numbers of hosts were chosen because they were just more than the maximum number of offspring produced by females in a previous experiment (Napoleon & King, 1999). The adult offspring of the mated females were counted. There is no differential mortality of the sexes in this species (Napoleon & King, 1999). Thus, observed sex ratio patterns, as determined by counting emerged adult offspring, reflect the sex ratio at oviposition. Offspring sex ratio was regressed against maternal age. The offspring of the virgin females were not sexed and counted because virgin females can produce only sons, and virgin females do not differ from mated females in number of offspring or longevity (King, 2002a). However, these virgin females were used in the second phase of this experiment.

The second phase was to determine whether once-mated females and virgin females would mate after a period of ovipositing when tested with mated males. For testing, a 0 d old mated male was placed with the female in a test tube and his sexual response was examined, i.e., whether he fanned, mounted, courted while mounted, and copulated. Ten minutes was chosen because in a previous study 96% of virgins were mounted within less than 3 min, with no further increase in mountings by 6 min (King & Fischer, 2005). After 10 min, if the male had neither mounted nor fanned and retreated from the female, an interaction was forced by tapping the test tube until the male landed near the female. Male responses both without and with the forced interaction are reported. Of the 19 mated females, 15 were still alive to be tested with a male after about a week of oviposition. The exact day of testing turned out not to matter (see Results). Ten were tested with a male on their seventh day, right before providing the female with that day's hosts. The other females were tested on their ninth or tenth day if they were still alive ($N = 5$ females tested). Of the twelve virgin females, four were tested when they were 7 d old and eight of them on their ninth or tenth day.

The second experiment was to determine whether once-mated females would mate after a period of ovipositing when tested with virgin males. The experimental setup was otherwise the same as in the first experiment except that offspring were not counted since the relationship between age and daughter production had already been established in the first phase of the first experiment. Females were tested on their seventh day ($N = 23$) or on their tenth day ($N = 22$).

Monandry versus immediate polyandry

This experiment tested the effects of single versus double copulation on female fitness, specifically on longevity and offspring production. There were three treatments ($N = 17$ per treatment) and females were randomly assigned to treatment: monandrous females, polyandrous females and interrupted monandrous females. The monandrous females were each presented with a single virgin male and allowed to mate. In the polyandrous treatment, the female was allowed a first mating that went through and included copulation, with the male removed just before postcopulatory courtship. Such females are

still attractive and receptive to subsequent males (King & Fischer, 2005). The female was then immediately presented with a second male and the second mating was allowed to go to completion. There was no obvious difference in male or female behavior during second matings versus during normal matings.

The polyandrous treatment was not entirely equivalent to two entire matings because the first male was not allowed to perform postcopulatory courtship. If the monandrous and polyandrous treatments had simply been compared, any difference could have been due to the absence of the first postcopulatory courtship in the polyandrous treatment rather than due to the second mating. Therefore, whether there was an effect of the first male's postcopulatory courtship on fitness was tested by including an interrupted monandrous treatment. The interrupted monandrous females did not receive postcopulatory courtship because the male was removed immediately after copulation was completed. If the fitness of monandrous females did not differ from the fitness of interrupted monandrous females, then this would show that postcopulatory courtship has no effect on a female's fitness, and so any difference between the monandrous treatment and the polyandrous treatment could be attributed to the second mating in the latter.

After the mating(s), each female was presented with hosts daily for life: 20 hosts for each of her first 4 d, 15 for each of the next 4 d, and then 10 hosts daily. Adult offspring were counted from these hosts. We were interested in comparing the monandrous females to the polyandrous females. However, in all analyses, monandrous females were first compared to interrupted monandrous females to test for effects of postcopulatory courtship, as explained above. When there were no differences, the two were combined to increase statistical power, prior to comparison to the polyandrous females.

In this experiment and in the following experiment, each replicate consisted of one of each treatment performed nearly simultaneously. However, to increase power, the replicate effect was excluded from the analyses because it was not statistically significant (Zar, 1999). Thus, data on longevity, daughter production and total offspring production were analyzed by independent t-tests rather than by paired tests. Longevity was log-transformed to achieve normality prior to analyses, but transformation was unnecessary for the other variables. In addition to comparing fitness means, variances were also compared because polyandry could be selected for as a means of reducing fitness variance (reviewed in Jennions & Petrie, 2000). Variances were compared using Levene's test for equality of variances.

Monandry versus delayed polyandry

This experiment examined the effects on a female of copulating a second time after 4 d of oviposition. There were two treatments ($N = 30$ per treatment): polyandrous females and monandrous females. In the polyandrous treatment, each female mated once prior to receiving any hosts, but the mating was interrupted after copulation, prior to postcopulatory courtship, so that she would remain receptive. After the first mating, she was given 4 d of 20 hosts daily. Then she was exposed to an additional male, i.e., on her fifth day. If the female did not remate within 10 min, the male was tapped next to the female to encourage mating. For a subset of the females that still did not remate, a second male was introduced. If a female did not remate, this was noted. However, regardless of

whether she remated, she was subsequently given another 4 d of 20 hosts daily. The effect of remating was tested after 4 d because by then a female's daughter production had dropped (see Results). Each monandrous female mated normally with a single virgin male prior to receiving 20 hosts daily for a total of 8 d.

For all females in both treatments, adult offspring were counted from hosts from the last 4 d, i.e., after most females in the polyandrous treatment had mated a second time. First, daughter production and offspring production were compared between females in the monandrous treatment ($N = 30$) and only those females in the polyandrous treatment that failed to remate ($N = 8$). Because the two types of singly mated females did not differ in either daughter production ($t_{36} = 0.66$, $P = 0.52$) or total offspring production ($t_{36} = 0.67$, $P = 0.51$), they were combined prior to comparison to females that mated twice. Offspring from the first 4 d of hosts were counted from a subset of females ($N = 28$) in order to test whether the daughter production of singly mated females dropped from the first 4 d to the last 4 d under conditions of this experiment. It did (see Results).

Sperm counts with delayed polyandry

To test whether females increase the number of sperm in their spermathecae by remating, sperm counts were compared between females that remated after 4 d of oviposition ($N = 16$) and females that did not ($N = 19$). The experimental set up was as above except that after 4 d of oviposition, about half of the females were given the opportunity to remate. All females that had the opportunity to remate did so, although two females did so only after being presented with a third or a fourth male. All females were dissected. The sperm in the spermathecae were dispersed in a drop of saline solution and counted under fluorescence microscope after ethanol fixation and DAPI staining (Bressac & Chevrier, 1998).

Results

Decreased daughter production and remating

Mated females lived up to 21 d when given hosts. Mothers were still producing female-biased sex ratios on their seventh day (Figure 1), but the proportion of daughters was significantly less than on the first day (mean ± SE: 0.63 ± 0.07 versus 0.81 ± 0.03; paired t-test: $t_{16} = 3.15$, $P = 0.006$). This was because the number of daughters decreased by three-fourths, whereas the number of sons decreased by less than half. Across all mothers, there was a statistically significant negative relationship between proportion of daughters and age (regression: $F_{1, 214} = 60.0$, $P < 0.001$; $R^2 = 0.22$, $y = -0.036x + 0.86$). Looking at the relationship within individual mothers, the direction of the relationship was negative for 18 of 19 mothers. The 4 of 19 females that were still alive by day 17 had begun producing only sons.

Most males fanned in response to females that had been ovipositing for roughly a week, regardless of both the female's and the male's mating status (Tables 1, 2).

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However, mounting, mounted courtship, and copulation were almost completely absent.

Monandry versus immediate polyandry

Comparison of monandrous females versus interrupted monandrous females revealed that postcopulatory courtship did not significantly affect longevity (independent t-test: t_{32} = 0.00, P = 1.00), daughter production (independent t-test: t_{32} = 0.68, P = 0.50) or total offspring production (independent t-test: t_{32} = 0.34, P = 0.74). Comparison of females that copulated twice to females that copulated once showed that copulating a second time did not significantly affect longevity (independent t-test: t_{49} = 0.63, P = 0.53), daughter production (independent t-test: t_{49} = 1.09, P = 0.28) or total offspring production (independent t-test: t_{49} = 1.26, P = 0.22).

Variance did not differ significantly for any of the comparisons. Variances did not differ significantly between monandrous and interrupted monandrous females for longevity, daughter production or total offspring production (Levene's tests: $F_{1,32}$ = 0.05, P = 0.83; $F_{1,32}$ = 0.014, P = 0.91; $F_{1,32}$ = 0.36, P = 0.55). Variances also did not differ significantly between females that copulated once versus twice for longevity, daughter production or total offspring production (Levene's tests: $F_{1,49}$ = 0.24, P = 0.63; $F_{1,49}$ = 1.77, P = 0.19; $F_{1,49}$ = 0.21, P = 0.65).

Monandry versus delayed polyandry

Among females that mated only once, daughter production dropped from the first 4 d of oviposition to the last 4 d (Figure 2; paired t-test: t_{27} = 7.14, P < 0.001) and corresponded to a drop in the proportion of daughters from 0.81 ± 0.01 to 0.72 ± 0.02 (paired t-test: t_{26} = 4.65, P < 0.001). However, over their last 4 d, females that copulated once did not differ significantly from females that copulated twice in terms of daughter production (Figure 2; t_{58} = 1.01, P = 0.32) or total offspring production (t_{58} = 0.25, P = 0.80). Variance was not significantly less, or even in that direction, for females that copulated twice versus once in terms of daughter production or total offspring production over their last 4 d (Levene's tests: $F_{1,58}$ = 3.02, P = 0.088; $F_{1,58}$ = 2.25, P = 0.14).

Sperm counts with delayed polyandry

Females that had been ovipositing for 4 d increased the number of sperm in their spermathecae when allowed to remate (Figure 3; mean ± SE, range: 121.42 ± 14.11, 14 - 214 to 277.81 ± 27.04, 81 - 485; t_{33} = 5.36, P < 0.001).

Discussion

Decreased daughter production and remating

S. endius females that mated only once started to decrease their daughter production within

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a week of beginning to oviposit. However, they did not remate at that point, primarily because males did not mount them. In other words, mated females did not regain their attractiveness as they oviposited and stopped producing daughters. In fact, their attractiveness further decreased: 0% of mated males mounted mated females after roughly a week of ovipositing, whereas more than 30% mount newly emerged mated females (King et al., 2005; King & Fischer, 2005). The loss of attractiveness over time was not unique to mated females. A virgin female's attractiveness to mated males also dropped to 0% after roughly a week of ovipositing. In contrast more than 85% of mated males will mount a newly emerged virgin female (King et al., 2005; King & Fischer, 2005). One explanation for this loss of attractiveness is that almost all females may mate within a week under natural conditions, thereby removing selection to remain attractive.

The possibility that females remate later than they were tested cannot be completely ruled out, but it seems unlikely given that virgin females also had become almost completely unattractive by testing time. Furthermore, fitness returns from remating late in life will be low because very few offspring are produced then (Figure 1). These results suggest that females exhibit lifetime monandry.

In contrast, in most animal species, including most insects, females are polyandrous (Arnqvist and Nilsson, 2000). Solitary species of parasitic hymenopterans have been reported to be an exception, but based on extremely limited data (Ridley, 1993). Solitary species are, by definition, those in which only one offspring develops per host, as in *S. endius*. Willingness to remate was usually only examined fairly soon after emergence (Ridley, 1993), whereas remating later in life could be common because low levels of sperm then is common (Henter, 2004). When females of apparently-monandrous solitary species have been tested when they exhibit a drop in daughter production or sperm stores, results have been mixed (studies with n > 5). *Trichogramma minutum* females remate and resume some daughter production, although much less than early in life (Leatemia et al., 1995); *Lariophagus distinguendus* and *S. endius* females do not remate (Steiner et al., 2008; present study). Thus, some solitary species exhibit lifetime monandry despite substantial drops in daughter production and/or sperm stores.

Daughter production decreases quickly with maternal age in *S. endius*. Traditionally in parasitic hymenopterans, any decrease in daughter production as the mother ages has been assumed to result from decreasing sperm stores (e.g., Henter, 2004 and references therein). This assumption has been based on both theoretical and empirical grounds. Sperm are necessary for daughter production; the number of sperm in the female's spermathecae decreases as she oviposits in correspondence with a drop in daughter production (Chevrier & Bressac, 2002); and experimental studies have demonstrated an association between females containing few sperm and the production of few daughters (Gordh, 1976; Nadel & Luck, 1985; Leatemia et al., 1995).

The drop in daughter production by *S. endius* starts very early in life. Low sperm stores could potentially explain the drop if sperm usage is inefficient. Sperm usage is especially inefficient in vertebrates (Birkhead & Møller, 1993), but even in insects it usually takes more than one stored sperm to achieve a single fertilization (e.g., for other parasitoid wasps: 4.08 calculated from Bressac & Chevrier, 1998, though only 1.33 for another species (Bressac et al., 2009)). *S. endius* females contained about 121 sperm at day 5; and from day 5 on, females produced 20.68 ± 3.10, n = 19, more daughters. This

translates to 5.84 sperm per daughter. From day 1 to 4 an *S. endius* female produced about 37 daughters (Figures 1, 2). Thus, at this rate, a female would have needed 37 * 5.84 = 216 sperm on day 5 just to stay at the same high number of daughters over the next 4 d. Remating at day 5 would supply more than that (discussed below). Females were ovipositing alone in these experiments, which is expected to maximize daughter production (Hamilton 1967; King 2002b). Thus, 5.84 sperm per daughter may be the highest sperm efficiency that females ever achieve.

If the offspring sex ratio in *S. endius* is constrained by sperm numbers, why has selection not resulted in males transferring more sperm or in polyandry? After monandry evolved, there would be no sperm competition. In this situation, selection may favor males that inseminate each female only partially so that they can inseminate more females. Partial insemination may be particularly advantageous to males if female density is high or female mortality rates are high. Such partial insemination has also been reported for the parasitoid wasp *Dinarmus basalis* (Chevrier and Bressac, 2002). However, *D. basalis* is polyandrous, which should reduce sperm limitation. Why has polyandry not evolved in *S. endius*? Perhaps receptive males are of limited availability or perhaps females lack the physiology to make use of the larger numbers of sperm that multiple males would provide (discussed below). Either of these two factors could reduce or eliminate the current benefit of polyandry.

Although sperm limitation has been the traditional explanation for decreases in daughter production with maternal age in parasitic hymenopterans, that the pattern is adaptive also remains a possibility. However, why the optimal sex ratio would change with maternal age in a parasitoid wasp such as *S. endius* remains to be determined. One possibility is that older females produce smaller eggs, as in another parasitic hymenopteran (Giron and Casas 2003), and large egg size is more important to the fitness of daughters than of sons. Egg size has been shown to affect fitness in an ectoparasitic hymenopteran (Mayhew and Heitmans, 2000), but whether it does so differentially for females versus males and in endoparasitoids remains to be tested.

Effect of remating on fitness

Despite a rapid drop in daughter production among *S. endius* females, copulating twice did not significantly increase a female's fitness in the present study. This was true regardless of whether the second copulation was immediately after the first or after daughter production had begun to drop and regardless of whether fitness was estimated by longevity, offspring production, daughter production, or the amount of variance in these estimates. Although whether a female was polyandrous or monandrous appeared to have no effect on her own fitness, her monandry may benefit her first mate through decreased sperm competition for him.

The results with *S. endius* are consistent with findings for the two other species that have been examined that are as highly monandrous, the bumble bee *Bombus terrestris* and house flies. When manipulated into a second mating, females of neither had greater fitness (Baer & Schmid-Hempel, 2001, 2005; Arnqvist & Andres, 2006). This similarity of results suggests that the differences among the studies in methodology may be unimportant. The bumble bee study relied on artificial insemination; the house fly study and the present *S.*

endius study did not. The technique used to achieve multiple mating prevented females from receiving a double dose of accessory fluids along with the double dose of sperm in both the bumble bee study and the house fly study, but not in the present study.

Obviously, not every cost and benefit of monandry versus polyandry can be examined in a single study, and a benefit to polyandry may yet be found in those species where it appears lacking. However, the lack of an increase in fecundity with multiple mating in the highly monandrous species that have been tested (Baer & Schmid-Hempel, 2001, 2005; Arnqvist & Andres, 2006; present study) contrasts with the usual increase in polyandrous species and in less highly monandrous species (Arnqvist & Nilsson, 2000; Torres et al., 2004).

Why was daughter production not increased by mating twice given that sperm numbers increased? In species in which mated females rarely remate, such as *S. endius,* selection to be able to benefit from additional sperm is not expected. Physiological explanations for why more sperm do not translate into a resurgence in daughter production merit further study. Females may produce compounds that are needed to make use of sperm, e.g., to release sperm from the spermatheca (Iida & Cavener, 2004), but may produce these compounds only in quantities sufficient for the number of sperm that they normally receive.

Jones (2001) and Montrose et al. (2004) propose that the cost of monandry occurs when females mate with already mated males. However, at least in the parasitic wasp, *A. calandrae*, females contain the same amount of sperm regardless of whether they mate with a virgin male or a male that already mated five times (Bressac et al. 2009).

Comparisons with insect species in general suggest that the lack of benefit to polyandry in female *S. endius* is related to its being highly monandrous. However, the lack of benefit could also be related to being a parasitic hymenopteran. Mating multiply instead of singly has no significant effect on daughter production in most parasitic hymenopterans that have been studied, even most polyandrous ones, although clearly more data are needed. Among polyandrous parasitic hymenopteran, mating multiply significantly increases daughter production in one (Chevrier & Bressac, 2002) but not in three others (Wilkes, 1966; Allen et al., 1994; Cheng et al., 2004; Jacob & Boivin, 2005). In the other "monandrous" parasitic hymenopteran besides *S. endius* that has been examined, 79% of females mate twice when exposed to two males simultaneously but not otherwise; and daughter production is not greater for these doubly mated females (Do Thi Khanh et al., 2005). The lack of an effect of multiple mating on daughter production occurs despite sperm from the additional males being transferred and used in daughter production along with the sperm of the first male (Wilkes, 1966; Allen et al., 1994; Do Thi Khanh et al., 2005; Bressac et al. 2009).

In conclusion, *S. endius* females appear to exhibit lifetime monandry. Monandry appears not to benefit a female, but may instead have evolved through its benefit to her first mate. What data are available suggest that a current lack of benefit to polyandry could be a common feature of highly monandrous species generally or a feature of parasitic hymenopteran regardless of mating system.

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Table 1. Percent of mated males that fanned, mounted, courted and copulated when presented with 7 – 10 d old females.

	Fanned	Mounted	Courted	Copulated
Mated females (*N* = 15)				
Within 10 min	80%	0%	0%	0%
Within 10 min and forced interaction	100%	7%	0%	0%
Virgin females (*N* = 12)				
Within 10 min	92%	0%	0%	0%
Within 10 min and forced interaction	92%	0%	0%	0%

Table 2. Percent of virgin males that fanned, mounted, courted and copulated with mated females.

	Fanned	Mounted	Courted	Copulated
7 d old females (N = 23)				
Within 10 min	65%	4%	4%	0%
Within 10 min and forced interaction	96%	17%	17%	4%
10 d old females (N = 24)				
Within 10 min	83%	8%	8%	4%
Within 10 min and forced interaction	92%	17%	17%	4%
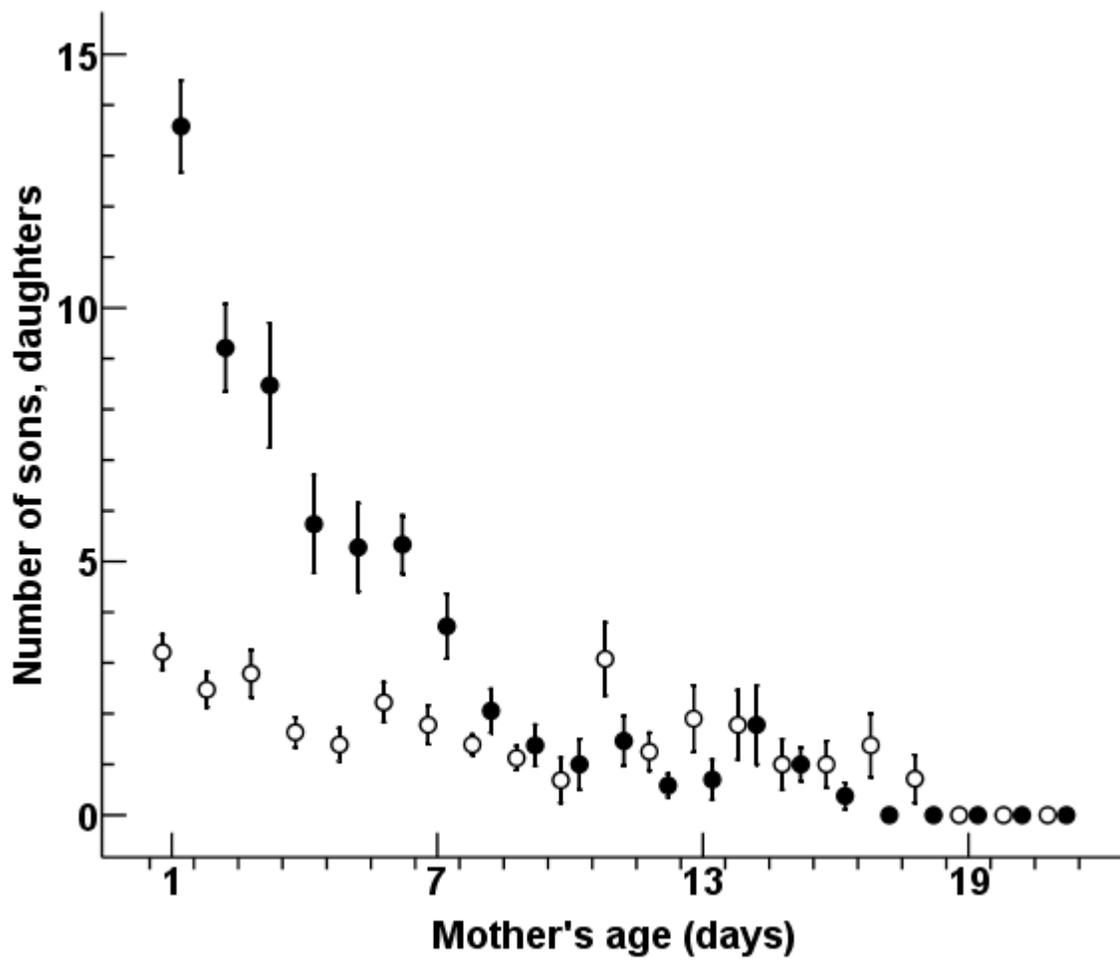
**Figure 1** Mean ± SE number of sons (○) and of daughters (●) versus maternal age.
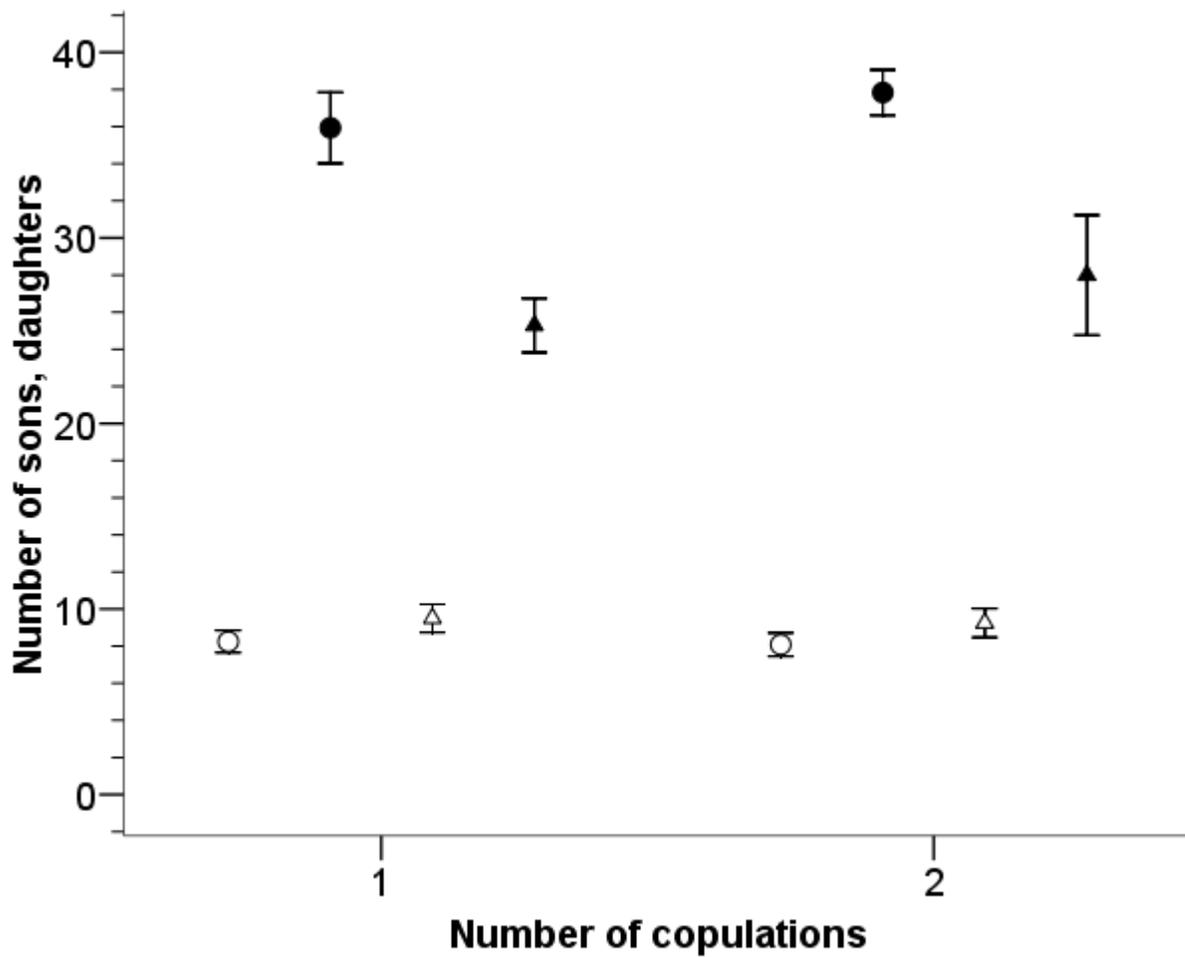
Figure 2 Mean ± SE number of sons and daughters produced over days 1-4 (○ sons, ● daughters) and days 5-8 (Δ sons, ▲ daughters) by females that copulated once or twice on day 1 prior to oviposition.
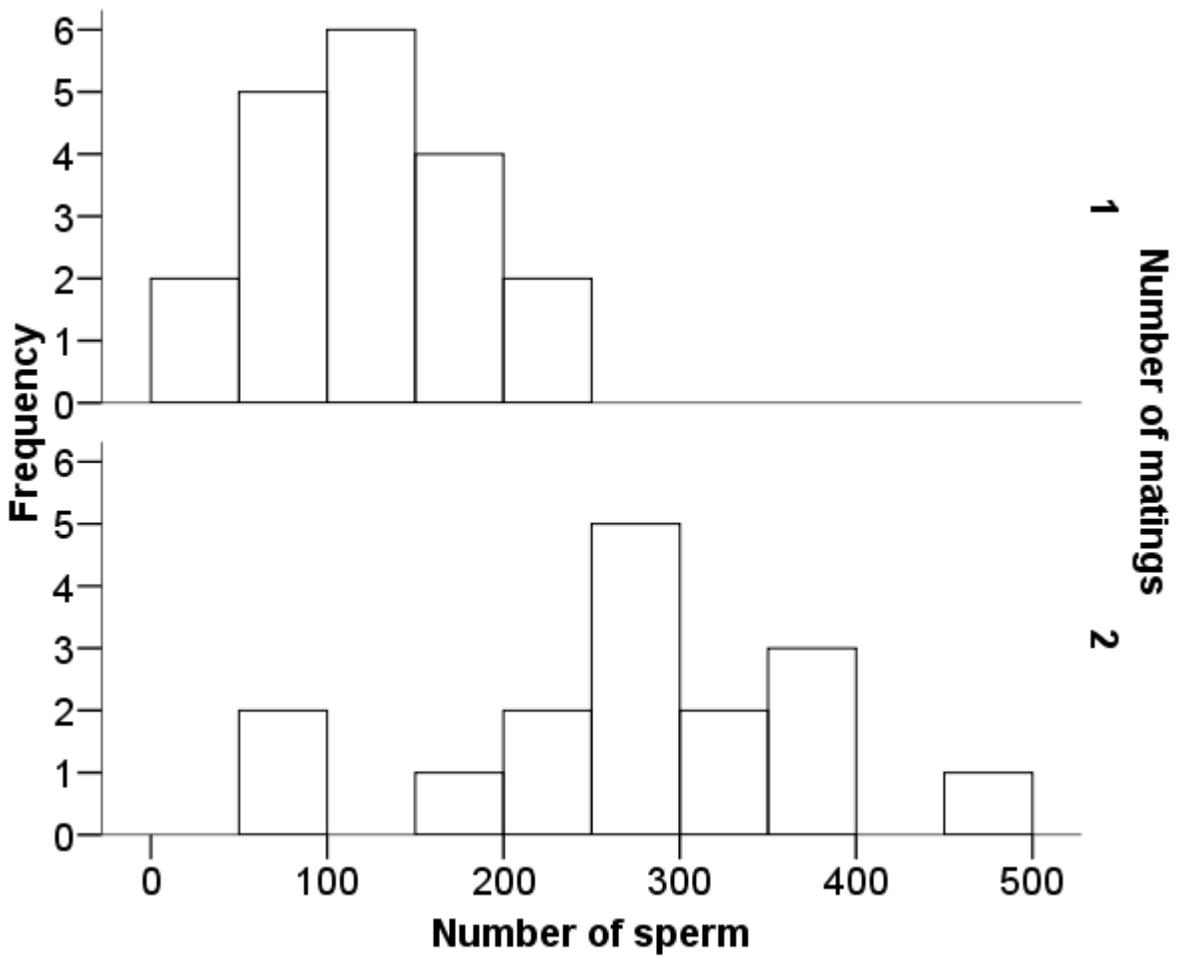
Figure 3 Number of sperm in mated females that had been ovipositing 4 d and had mated once (on day 1 prior to oviposition) or twice (on day 1 prior to oviposition and again with a new male on day 5)