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Mate-Guarding in the Australian Zebra Finch, Taeniopygia guttata: for Males, Protecting Paternity is More Important Than Extra-Pair Copulations

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Mate-guarding in the Australian zebra finch, *Taeniopygia guttata*: for males, protecting paternity is more important than extra-pair copulations

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Master of Science

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Birds are commonly sexually promiscuous, and conflict between the sexes has led to the evolution of paternity assurance strategies, including mate-guarding. Although adaptive explanations for mate-guarding have tended to focus on male fitness consequences, mate-guarding is likely adaptive for both males and females when there are costs to mate infidelity. Further, active female participation in male mate-guarding is also likely to be adaptive when there are female fitness costs to harassment by extra-pair males. While ecological variation may affect extra-pair copulation (EPC) behaviors, flexible mate-guarding may confound a simple relationship between a single ecological variable and patterns in extra-pair paternity among and within avian species. To better understand the adaptive explanations for mate-guarding as well as the observed variation in paternity patterns, it is necessary to explore how mate-guarding is generally structured by the costs and by the benefits of engaging in the behavior. To investigate (1) the costs and benefits of mate-guarding; and (2) the active role of the female, I conducted an experiment with the Australian zebra finch (Teniopygia guttata) in which I independently varied the opportunity for each member of a captive breeding pair to engage in EPC behavior. Within an experimental chamber, I exposed breeding pairs to extra-pair stimulus birds in five (male:female) sex ratios (1:0; 0:1; 1:1; 1:3; 3:1). Varying an individual’s EPC opportunity changed the cost of mate-guarding, and varying their mate’s EPC opportunity changed the benefit of mate-guarding. I predicted that both males and females would (1) mate-guard less intensely as their EPC opportunity increased; and (2) mate-guard more intensely as their mates’ EPC opportunity increased. My results indicate that female zebra finches are not nearly as active as males in mate-guarding. Further, it appears that for male zebra finches, protecting paternity was more important than engaging in EPC behavior; as the threat to paternity increased, males engaged in EPC behavior less. While there was not conclusive evidence of active female participation in male mate-guarding, there was a slight tendency for females to participate in being guarded as the threat to within-pair paternity increased. Investigations of mate-guarding, both in terms of its flexibility and efficacy, should focus separately and explicitly on male and female behavior and subsequent fitness returns. Only after individually evaluating each pair member’s inclination to pursue and prevent EPCs, can there be clear predictions about the relationship between the social and ecological context and (1) mate-guarding intensity; and (2) paternity patterns.
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CHAPTER 1. MATE-GUARDING: A COST-BENEFIT ANALYSIS

I. Extra-pair Paternity

A. Prevalence and variation

Most birds are socially monogamous, and in the vast majority of species (85%) there is some level of cooperative biparental care of the offspring (Bennett and Owens 2002). Until recently, it was assumed that easily observable social bonds were representative of underlying sexual fidelity. However, when the molecular tools to assign nestling paternity became readily available, sexual promiscuity was identified as a common feature of avian mating systems. Recent reviews indicate that in over 70% of avian species, some offspring are sired by a male other than the social father (Griffith et al. 2002). Extra-pair paternity (EPP) can affect the strength of sexual selection acting in populations (Webster et al. 1995), and in recent decades there has been much interest both in the adaptive function of EPP and in the sources of variation in extra-pair behaviors between species and populations (Petrie and Kempenaers 1998; Griffith et al. 2002; Westneat and Stewart 2003).

There is a great deal of variation in rates of EPP among avian taxa; among species, EPP can range from 0% (e.g. the Harlequin duck (*Histrionicus histrionicus*)) (Lazarus et al. 2004) to over 70% (e.g. the superb fairy-wren (*Malurus cyaneus*)) (Griffith et al. 2002). Phylogeny plays an important role in influencing interspecific variation in EPP (Bennett and Owens 2002). For example, passerines generally display greater EPP than non-passerines (15% ± 16% SD vs. 3% ± 5% SD (Westneat and Stewart 2003)). There are also marked differences in EPP among closely related species; within the
swallow subfamily (Hirundininae), EPP rates range from 5% to 50% (Westneat and Stewart 2003). Even within the same species there can be pronounced variation between populations; one population of willow warblers (Phylloscopus trochilus) was found to have no extra-pair offspring (EPO) while in another 50% of all broods had at least one EPO (Petrie and Kempenaers 1998).

There are several plausible and non-mutually-exclusive hypotheses to explain the current variation in EPP among and within avian species. The traditional ecological explanations for variation in EPP are breeding synchrony (Stutchbury and Morton 1995) and breeding density (Westneat and Sherman 1997; Stewart et al. 2010). Under higher levels of breeding synchrony there may be greater net benefits to seeking extra-pair fertilizations (EPFs) for both males and females. When females are synchronously fertile, males may experience less intrasexual competition for extra-pair partners (Stutchbury and Morton 1995). In addition, females may be better able to assess the quality of potential extra-pair partners when all males are simultaneously competing under similar energetic, environmental, and social conditions (Stutchbury 1998). Breeding density may increase EPP by increasing the encounter rate between extra-pair and pair individuals (Stewart et al. 2010).

There is conflicting evidence as to the strength and consistency of the impact of these ecological conditions on EPP (Bennett and Owens 2002; Griffith et al. 2002). There is more support for the hypothesis that breeding synchrony affects EPP from interspecific comparisons (Stutchbury and Morton 1995) than intraspecific manipulations (Arlt et al. 2004). However, there is more evidence that breeding density
influences variation in EPP rates between populations (e.g. eastern bluebird (*Sialia sialis*) Stewart et al. 2010) than between species (Westneat and Sherman 1997).

Interactions between breeding density and synchrony may confound a simple relationship between EPP and either ecological variable. Density may directly affect synchrony; for example, female lesser grey shrikes (*Lanius minor*) minimize synchrony under high densities (Kristin et al. 2008). Alternatively, density and synchrony could interact directly to influence paternity patterns; in the eastern bluebird, synchrony positively affects EPP at high densities (Stewart et al. 2010). In certain contexts, flexible paternity assurance strategies like mate-guarding may account for a discrepancy between a theoretical increase in the opportunity for extra-pair copulations (EPCs) and observed paternity patterns (Petrie & Kempenaers 1998).

Identifying universal patterns in EPP may only be possible if general predictions take into account how the relative costs and benefits of engaging in EPP are separately influenced by the specific social and ecological context. Westneat and Stewart (2003) point to the conceptual importance of identifying the parties involved in producing EPP and propose conflict theory as a means of understanding the interactions between those individuals. The parties that produce EPP are the pair female, pair male, and extra-pair individual(s), and these individuals may have conflicting reproductive interests (Lifjeld et al. 1994). If the pair female has been under selection to achieve EPFs, paternity patterns could be determined by the disparity between female choice and pair male paternity guards (e.g. Kempenaers et al. 1992). If the pair female has not been under selection to achieve EPFs, then paternity patterns could be determined by the
disparity between (1) extra-pair male pursuit and female resistance (e.g. Lifjeld and Robertson 1992); or (2) extra-pair male pursuit and pair male paternity guards (e.g. Komdeur et al. 2007). Each set of conflicts may be separately influenced by the social and ecological context.

There are situations in which the pair female, pair male, or extra-pair male is clearly seen to be in control of EPFs at the expense of at least one of the other parties. For example, female blue tits (*Cyanistes caeruleus*) evade pair male paternity guards to engage in EPCs (Lifjeld and Robertson 1992). However, it is not always apparent how the behavioral conflicts over paternity are resolved. Variation in the outcome of these conflicts could have profound effects on the relationship between a given ecological factor (e.g. resource availability) and EPP (Westneat and Stewart 2003). For example, when the female is constrained by the energetic demands of resisting her mate’s paternity guards, food supplementation will increase EPP (as in the serin (*Serinus serinus*)) (Hoi-Leitner et al. 1999). However, when the pair male is constrained by the energetic demands of paternity assurance (e.g. mate-guarding), food supplementation will decrease EPP (as in the red-winged blackbird (*Agelaius phoeniceus*)) (Westneat 1994).

Although similar mechanisms may not determine EPP rates in all avian systems, EPC behaviors should be universally structured by the relative costs and benefits of EPP for each individual. To identify how the behavioral conflicts over paternity may be resolved, it is necessary to clearly identify the cost and benefits of promiscuity for each player in the “game”.

4
B. Costs and benefits of promiscuity

Sexual promiscuity could be costly to individuals in three ways; there could be costs associated with (1) being promiscuous; (2) having a promiscuous mate; and (3) having promiscuous neighbors. Examples of each of these costs are summarized in Table 1. An individual may experience costs inflicted by any of the other parties involved in producing EPP (i.e. pair female, pair male, and extra-pair male). The relative importance of these costs likely varies among and within species as well as over time with changing ecological conditions.

Males benefit from sexual promiscuity to the degree that they are able to increase their total number of offspring through EPFs (Webster et al. 1995; Yezerinac et al. 1995). Even before there were the genetic tools to detect it, Trivers (1972) argued that where there is an initial imbalance in parental investment (i.e. a disparity in the energetic costs of sperm and egg production), selection will favor a mixed male reproductive strategy wherein males explore both pair and extra-pair matings. Indeed in socially monogamous species, variation in extra-pair mating success may be more important than variation in pair mating success in accounting for variation in total mating success (i.e. the strength of sexual selection) (Webster et al. 1995).

There are many ways by which a female might benefit from producing EPO. By engaging in EPCs, females may be guarding against mate infertility, maximizing the genetic diversity of their offspring, ensuring their genetic compatibility with the sire, seeking ‘good genes’ for their offspring, and obtaining direct, non-genetic resources
(reviewed in Griffith et al. 2002). While many of these reasons have been proposed independently, it is likely that different or multiple factors play a role in shaping the reproductive behavior of a particular species.

C. Avoiding the costs of promiscuity- mate-guarding

Of the costs outlined in Table 1, male loss of reproductive success has received the most attention for its role in shaping male behavior (Petrie & Kempenaers 1998). The two main paternity assurances strategies are frequent pair copulations and mate-guarding (Møller and Birkhead 1991). Mate-guarding may be the more effective strategy (Møller and Birkhead 1993). Mate-guarding behavior seems to be absent only in taxa where the specific ecological context renders effective mate-guarding impossible (e.g. when there is intense nest site competition in colonially breeding birds or when frequent male foraging trips are necessitated by courtship feeding in birds of prey, owls, and shrikes) (Møller and Birkhead 1991).

Although mate-guarding has traditionally been defined as the close following of a fertile female (Beecher and Beecher 1979; Birkhead and Møller 1992), experimental measurements of mate-guarding tend to encompass a wide range of pair activities (Table 2). These behaviors (following, interactions with intruding males, pair proximity in the territory or at the nest, and song) are similar in that they prevent or limit interaction between the female and extra-pair males within the particular study system.

If the presence of the pair male decreases EPCs (e.g. Alatalo et al. 1987), then male mate-guarding may not be adaptive solely in offsetting the costs of male
cuckoldry; female participation in male mate-guarding may also be adaptive in offsetting female costs of promiscuity such as harassment by intruding males (Kempenaers et al. 1995) and forced EPCs (Low 2005). Further, active female mate-guarding could be adaptive in offsetting the females costs of male infidelity in situations where (1) there are costs to male infidelity; and (2) female behavior inhibits male EPC behavior (Eens and Pinxten 1995).

Mate-guarding is particularly relevant to a discussion of the behavioral conflicts over paternity because mate-guarding intensity is related both to (1) the costs of mate infidelity; and (2) the benefits of achieving EPFs (time spent achieving EPFs is time not spent mate-guarding) (Figure 1). The role of the female in mate-guarding behavior should not be ignored. Female participation in male mate-guarding and active female mate-guarding can have profound effects on both male mate-guarding behavior (Lifjeld et al. 1994) and observed paternity patterns (Kempenaers et al. 1995). Mate-guarding behavior is shaped by the costs and benefits of engaging in the behavior. Further, the role of mate-guarding in the behavioral conflicts surrounding EPP will be influenced by the degree to which guarding intensity varies as those cost and benefits fluctuate.

II. Mate-Guarding

A. Costs

There are theoretical predictions (Dias et al. 2009) and empirical evidence (Westneat 1994; Komdeur 2001) that within a species, male mate-guarding is shaped by the costs of engaging in the behavior. As for any behavior, an obvious cost of guarding one’s mate
is the opportunity cost of not engaging in another mutually excluded activity (Dias et al. 2009). The two costs of male mate-guarding that have received the most attention are the fitness costs of not soliciting EPCs (e.g. Dickenson 1997) and the energetic costs of not foraging (e.g. Komdeur 2001).

Several researchers have noted a tradeoff between soliciting EPCs and mate-guarding (McKinney et al. 1983; Alatalo et al. 1987; Dickinson 1997). The implied cost of not soliciting EPCs generates a simple prediction: males should mate-guard more when there is little chance of achieving EPFs. Both this prediction and its opposite (that males should mate-guard less when there is a high chance achieving EPFs) can be confounded by individual male attractiveness. When female extra-pair behavior is driven by female choice of ‘good genes’, it may be difficult to distinguish between a male’s opportunity to achieve EPFs and his risk of cuckoldry. In these situations, attractive males may gain the most EPFs and loose the least paternity (Green et al. 2000). When possible, experiments should correct for the possible covariance of an individual male’s risk of being cuckolded and his attractiveness as a potential extra-pair partner.

The trade-off between foraging and mate-guarding is more straightforward. Males that mate-guard more suffer a decrease in weight and body condition (Komdeur 2001; Low 2006).

There has been little attention given to the costs of female mate-guarding, but it seems reasonable to suggest that female mate-guarding would be under similar selection pressures as male mate-guarding in situations (1) where females increase their total fitness through EPFs and mate fidelity; and (2) when female mate-guarding
conflicts with foraging demands. This latter trade-off may be more important given that females could be guarding their mates during potentially energetically demanding periods (i.e. before and during the egg laying period (Monaghan and Nager 1997)).

B. Benefits

Through active mate-guarding, males and females are able to affect their mates’ interactions with extra-pair individuals. Through participating in male mate-guarding, females may be able to affect the frequency and duration of their own interactions with extra-pair males.

a. Male mate-guarding

The chief fitness benefit of male mate-guarding is assumed to be the protection of within-pair paternity (Beecher and Beecher 1979; Birkhead and Möller 1992). The simplest indicator of cuckoldry risk may be the fertility status of the female, and there is substantial evidence that males follow their mates more closely during the female fertile period (e.g. Möller 1985; Birkhead et al. 1987). Male mate-guarding often stops abruptly at the onset of incubation (e.g. Komdeur 1999), and in situations where males guard non-fertile females (Marthinsen et al. 2005; Fedy and Martin 2009), the behavior is thought to serve purposes other than paternity protection (e.g. to aid in female foraging efficiency). While the guarding of fertile females is highly suggestive of an adaptive explanation of mate-guarding behavior (i.e. the protection of within pair-
paternity), there is conflicting data on the direction of the relationship between male mate-guarding intensity and within-pair paternity within a species (Table 3).

There is an inherent problem with evaluating the benefit of male mate-guarding through a correlation between mate-guarding intensity and within-pair paternity. If mate-guarding is flexible, then mate-guarding intensity could correlate with cuckoldry risk. If males mate-guard more intensely as cuckoldry risk increases, males that mate-guard more could still loose more within-pair paternity than their neighbors (e.g. male bluethroats (*Luscinia svecica*)) (Johnsen et al. 1998). Without knowing an individual's risk of being cuckoldry, it is impossible to say if an individual male would have lost even more paternity if he had guarded less (Westneat and Stewart 2003).

Assessing the efficacy of male mate-guarding among species is complicated by variation in the social and ecological context. There is a distinction between mate-guarding in order to prevent female pursuit of EPCs and mate-guarding in order to prevent extra-pair male intrusions (Westneat and Stewart 2003). Kokko and Morrell (2005) have suggested that since mate-guarding efficiency decreases as the female's tendency to seek EPCs increases, male mate-guarding intensity may be expected to be very low when females are very unfaithful. Therefore in highly promiscuous species, there may not be a strong relationship between measures of mate-guarding and within-pair paternity.

When EPFs are due to extra-pair male intrusions, experiments have clearly demonstrated a positive relationship between male presence and within-pair paternity within a species (e.g. Chaung-Dobbs et al. 2001a; Komdeur et al. 2007). It is worth
noting that in some of the cases outlined in Table 3, the suggested relationship between mate-guarding and EPP was inferred based on an assumed correlation between extra-pair male intrusions, EPCs, and EPFs. Although trends in EPC behavior do not always reflect trends in EPF (Westneat and Sherman 1997), mate-guarding may impact both. In the pied flycatcher (*Ficedula hypoleuca*), mate-guarding prevents extra-pair male intrusions (Björklund and Westman 1983), EPCs (Björklund and Westman 1983; Alatalo et al. 1987) and EPFs (Moreno et al. 2009).

While mate-guarding is not under selection in every ecological context, and there are potential problems with inferring the benefit of mate-guarding through a correlation of mate-guarding intensity and within-pair paternity, mate-guarding is beneficial to some males in some contexts.

**b. Female participation in male mate-guarding**

If EPC behavior is costly for the female (either in the form of harassment or forced EPFs) then females should behave in a way that aids her mate in following her (Lifjeld et al. 1994; Gowaty and Buschhaus 1998). However, female participation is rarely explicitly quantified. In the stitchbird (*Notiomystis cincta*), male mate-guarding prevents paternity loss due to forced EPCs (Low 2005). Although female participation was not measured, high levels of female resistance to forced EPCs suggest that females would have good reason to cooperate in male mate-guarding behavior.

In the blue tit, EPP was not correlated with male mate-guarding intensity but it was correlated with female following rates (Kempenaers et al. 1995). When pair males
were caged on the territory for short intervals (30 min), females were subjected to increased harassment by extra-pair males. In this species, it appears that females are in control of EPFs and may participate in mate-guarding behavior as a way to avoid the costs of extra-pair male intrusions (Kempenaers et al. 1995). Burley et al. (1994) suggested that female zebra finches (*Taeniopygia guttata*) might participate in male mate-guarding behavior more when they are mated to males experimentally manipulated to be more attractive.

Fedy and Martin (2009) investigated male mate-guarding behavior outside of the female’s fertile period. In both the red-faced warbler (*Cardellina rubrifrons*) and the grey-headed junco (*Junco hyemalis caniceps*), male mate-guarding allowed the female to forage more efficiently. Although this has little bearing on promiscuity, it does illustrate that being mate-guarded may have important energetic consequences for the female. It would be interesting to look for similar effects of male guarding on female foraging during the pre-laying and laying period.

c. Female mate-guarding

Although mate-guarding has primarily been explored as a male tactic, female mate-guarding behavior should be under complementary selection pressure when there are costs associated with male infidelity. When females incur costs from having promiscuous mates (Table 2), females should limit their mate’s promiscuity through active mate-guarding (i.e. through behavior that limits interaction between a mate and extra-pair individuals) (Petrie and Kempenaers, 1998).
In the facultatively polygynous European starling (*Sturnus vulgaris*), females that behaved more aggressively towards other females were more likely to remain monogamous and receive more paternal investment in their broods than less aggressive females (Sandell 1998). In addition to female-female conflict, there is also evidence that female starlings solicit pair copulations to keep their mates from singing to extra-pair females (Eens and Pinxten 1995).

In an experiment with red-winged blackbirds, females were more aggressive towards conspecific mounts in soliciting rather than perching positions (Yasukawa and Searcy 1992). Although red-winged blackbirds are polygynous (Yasukawa and Searcy 1995), females may be under selection to protect the nonsharable portion of their male’s parental investment by discouraging secondary females from nesting on their male’s territory (Yasukawa and Searcy 1992).

There is also evidence of female mate-guarding in socially monogamous species. In the harlequin duck, pair bonds are long-term and extra-pair copulations are very rare (Robertson and Goudie 1999). When presented with conspecific female mounts, harlequin duck females mate-guarded more as the risk of male infidelity (i.e. number of female mounts) increased (Lazarus et al. 2004). In this species, intense mutual mate-guarding may maintain sexual fidelity (Lazarus et al. 2004).

**C. Evidence for flexible mate-guarding**

The benefits of engaging in mate-guarding behavior should trade-off with the costs. The degree to which individuals mate-guard flexibly will have a profound impact on the
intensity and resolution of the behavioral conflicts over extra-pair paternity; patterns in female EPC opportunity (i.e. cuckoldry risk) will not predict EPP patterns if there are flexible paternity assurance strategies. There is evidence that individuals modulate their mate-guarding behavior across several social and ecological contexts.

**Density**

Within a species, the rate of EPF tends to increase with breeding density (Westnest and Sherman 1997). This trend would suggest that if all else is equal, there is a greater risk of cuckoldry under higher densities, and therefore male mate-guarding intensity should be greatest under high density. Although breeding density may not have an impact on paternity patterns, western bluebird (*Sialia mexicana*) males follow their females more intensely when the nearest neighbor is closer (Dickinson and Leonard 1996). Similarly, when breeding density is experimentally reduced, male Seychelles warblers (*Acrocephalus sechellensis*) mate-guard less (Komdeur 2001).

**Synchrony**

The relationship between breeding synchrony and EPP is unclear (Bennett and Owens 2002; Griffith et al. 2002). Breeding synchrony could affect the parties involved in producing EPP (i.e. the pair female, pair male, extra-pair individuals) in ways that lead to conflicting predictions about the overall relationship between synchrony, risk of cuckoldry, and mate-guarding intensity (Hammers et al. 2009).
Breeding synchrony could affect the pair male’s opportunity to achieve EPFs. Under low synchrony, fewer fertile females could mean fewer potential EPC partners (Stutchbury 1998). Synchrony also could affect the behavior of the extra-pair male; asynchronously fertile females could attract proportionally more extra-pair male initiated EPCs since each extra-pair male will have fewer fertile potential extra-pair partners (Wagner et al. 1996). Synchrony could influence female behavior by making it easier for the female to assess extra-pair male quality (Stutchbury 1998). Under high synchrony, females may be more inclined to pursue EPFs.

While the suggested effects of breeding synchrony on the pair and extra-pair males would predict more intense male mate-guarding under lower synchrony, the suggested effect on female behavior would predict more intense male mate-guarding under higher synchrony. There is evidence of both trends. In the golden whistler (Pachycephala pectoralis) (van Dongen 2008) and the black-throated blue warbler (Dendroica caerulescens) (Chaung-Dobbs et al. 2001a), males mate-guard more when synchrony is lower. In the colonially breeding fairy martin (Petrochelidon ariel), males mate-guard more when synchrony is higher (Hammers et al. 2009).

To make sense of how breeding synchrony affects mate-guarding behavior (and consequently, EPP patterns), it is necessary to separately evaluate how synchrony affects the costs and benefits of mate-guarding for each of the involved parties.
Resource Availability

Mate-guarding is energetically demanding (Komdeur 2001; Low 2006); therefore males should mate-guard more when there are the resources to do so. In an experiment with red-winged blackbirds, Westneat (1994) found that males given supplemental food increased their mate-guarding intensity and consequently lost less paternity. Similar trends were observed in the great tit (Parus major) (Cuthill and Macdonald 1990) and Seychelles warbler (Komdeur 2001).

Relative attractiveness of the players

The costs and benefits of mate-guarding may depend on the individual characteristics of pair female, pair male, extra-pair individuals. The pair bond between the male and female likely influences EPC behavior and variation in its strength may affect mate-guarding behavior. Petrie and Hunter (1993) theorized that intraspecific variation in mating behaviors (specifically copulation frequency) is the result of a mismatch in partner quality; other models indicate that both a female’s inclination to pursue EPCs and male mate-guarding decisions are determined by male quality (Dias et al. 2009). Zebra finch females mated to males experimentally manipulated to be more attractive may participate more in male mate-guarding (Burley et al. 1994), and in an experiment where interspecific cross-fostering was thought to decrease pair affinity in great tits, males invested less time mate-guarding (Hansen et al. 2009).

The relative attractiveness of pair and extra-pair males may also affect male mate-guarding intensity. As previously noted, it is difficult to distinguish between an
increased cuckoldry risk and a decreased EPF opportunity when unattractive males experience both. For a relatively unattractive male, the benefit of mate-guarding may be great and the opportunity cost may be negligible. Males that are relatively less attractive than their neighbors have been found to mate-guard more intensely (Estep et al. 2005; Johnsen and Lifjeld 1995). This pattern could reflect an increased benefit of guarding (i.e. greater paternity threat), a decreased cost of guarding (fewer lost opportunities to achieve EPFs), or both.

III. Research Needs

Mate-guarding appears to affect mate infidelity and be limited by the costs of engaging in the behavior. Mate-guarding intensity is influenced by variation in the frequency and outcome of interactions between the pair female, pair male, and extra-pair individuals. Future research should account for this variation by explicitly viewing mate-guarding as the result of dynamic interactions between the parties involved in producing EPP. Discounting the role of the female and extra-pair individuals may limit the development of a complete theoretical model that relates mate-guarding behavior to paternity patterns.

A. Problems with removals

Removal experiments have offered crucial insight into the importance of male presence on female reproductive behavior; however, insofar as they offer insight into the importance of male mate-guarding, removal experiments can fail to adequately account for...
for the behavior of the female and extra-pair males. A removal, even a short one, is much longer than a natural break in male mate-guarding behavior. During a removal, female behavior may change not in response to a change in male mate-guarding behavior but in response to perceived mate death or desertion. For example, red-winged blackbird females behave differently during a natural length male extra-territorial foray (213 seconds) than during a much longer male absence (60 minutes) (Westneat 1994).

The behavior of extra-pair males in response to pair male removal may also be in danger of misinterpretation. If an extra-pair male's rate of intrusion trades-off with his own mate-guarding behavior (Alatalo et al. 1987; Dickinson 1997), then an extra-pair male's decision to solicit copulations from a neighboring female may be influenced by his perceived chance of success in achieving an EPF. In other words, if extra-pair males perceive a removal as a desertion rather than a decrease in mate-guarding, then the change in his intrusion/solicitation behavior may be greater than expected in response to a simple decrease in male mate-guarding intensity.

Some removal experiments have minimized the perception of mate desertion by caging the removed male on the territory (e.g. Chaung-Dobbs et al 2001a; Dickenson 1997). However, this approach does not control for how caged male behavior (i.e. song type or frequency) may change or how females or extra-pair males may perceive caged males.

Caging and removing the male certainly curtails male mate-guarding behavior, but it may have effects beyond the intended one. When the intent is to experimentally
modify just mate-guarding behavior, it may be better to indirectly target mate-guarding by manipulating the costs and/or benefits of engaging in the behavior (e.g. through food supplementation (Westneat 1994) or by changing male perception of his female’s fertility status (Komdeur et al. 1999). This type of approach would be possible only in situations where it was clear which cost and benefits structured mate-guarding behavior.

B. Problems with measurement

Experimental measures of male mate-guarding directly and indirectly disregard the importance of the female and extra-pair individuals in influencing mate-guarding. While common measure of mate-guarding (Table 2) encompass a wide range of interactions, they have mostly been interpreted as active male behaviors undertaken to prevent or limit female EPC behavior.

When male mate-guarding is quantified as a percentage of female movements from one location to another than the male follows, female behavior is indirectly ignored. The proportion of movements that a male follows might depend on the overall number of times that the female leaves if (1) female leaving behavior is indicative of female inclination to seek EPCs; and (2) males mate-guard according to the female’s inclination to seek EPCs (Dias et al. 2009). If a female is inclined to seek EPCs she might leave very frequently and her mate may follow her proportionally more than if she left him less frequently (i.e. there may not be a direct correlation between number of movements and proportion followed). Males that follow a higher percentage of female
movements may lose the same or more paternity than males that follow a lower percentage not because of differences in male behavior but because of differences in female behavior. When following behavior is reported solely as a proportion, there may be a misleading correlation between male mate-guarding intensity and paternity patterns.

Quantifying mate-guarding as a measure of pair proximity directly ignores female behavior. Unless proximity is measured at a location where one individual is more likely to be found (e.g. at the nest (Hammers et al 2009)), it is difficult to show intentionality on the part of either individual. Proximity quantifies male mate-guarding, female participation in male mate-guarding, and female mate-guarding but it is often interpreted solely as a measure of male mate-guarding. Unless there is a compelling reason to discount female participation (e.g. when the female initiates more moves away from the male then vice versa), proximity should be loosely interpreted as a measure of pair bonding or mutual mate-guarding.

IV. Conclusions

Extra-pair fertilization is an almost ubiquitous avian reproductive tactic. There are costs associated with sexual promiscuity and sexual conflict has led to the evolution of paternity assurance strategies. Adaptive explanations for EPP have tended to focus on female fitness (Hunter et al. 1993; Jennions and Petrie 2000; Griffith et al. 2002) and perhaps consequently, adaptive explanations for mate-guarding have tended to focus on male fitness (Birkhead and Møller 1992). However, mate-guarding is likely to be
under positive selection in both males and females, especially when there are fitness
costs to mate infidelity. Male and female mate-guarding limits mate promiscuity in
some contexts (e.g. Eens and Pinxten 1995; Komdeur et al. 2007), and females
participate in male mate-guarding when there are costs to interacting with extra-pair
males (e.g. Kempenaers et al. 1995).

Mate-guarding is structured by the dynamic interactions between all parties
involved in producing EPP, including the pair female, pair male, and extra-pair
individuals. Future discussion of the adaptive significance of mate-guarding may be
aided by reformulating the working definition of mate-guarding as a behavior that limits
interactions between a mate and extra-pair birds. This would generate clear testable
predictions about mate-guarding intensity from either the male or female perspective
as the costs of having an unfaithful mate varied. Where there are reasons to avoid
interactions with extra-pair individuals, participation in mate-guarding should not be
discounted. Mate-guarding should also be considered as having adaptive function even
in species with very low EPP rates (Lazarus et al. 2004).

While ecological variation may affect EPC behaviors, flexible mate-guarding may
confound a direct relationship between a single ecological variable and EPP patterns
between and among avian species. To better explore the adaptive explanations for
mate-guarding and the observed variation in paternity patterns, it is necessary to
understand how mate-guarding behaviors are generally structured by the costs and by
the benefits of engaging in the behavior.
Table 1. Suggested costs of promiscuity.

<table>
<thead>
<tr>
<th></th>
<th>Being promiscuous</th>
<th>Having a promiscuous mate</th>
<th>Having promiscuous neighbors</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Male Costs</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Disease¹</td>
<td></td>
<td>Loss of reproductive success⁴</td>
<td>Loss of reproductive success⁶</td>
</tr>
<tr>
<td>Possible increased risk of cuckoldry²</td>
<td></td>
<td>Energetic demands of mate-guarding⁵</td>
<td>Energetic demands of territory defense⁷</td>
</tr>
<tr>
<td>Sperm depletion³</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Female Costs</strong></td>
<td></td>
<td>Disease¹</td>
<td>Forced EPC¹²</td>
</tr>
<tr>
<td>Disease¹</td>
<td></td>
<td>Loss of paternal investment⁸,⁹</td>
<td>Harassment¹³</td>
</tr>
<tr>
<td>Loss of paternal investment⁸,⁹</td>
<td></td>
<td>Possible loss of paternal investment¹¹</td>
<td></td>
</tr>
<tr>
<td>Retaliation from mate⁹</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Searching for extra-pair mates¹⁰</td>
<td></td>
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</tr>
</tbody>
</table>


*there is conflicting evidence of this cost (Sheldon 2002; Yezerinac et al. 1996; Dixon et al. 1994).
Table 2. Experimental measures of mate-guarding behavior.

<table>
<thead>
<tr>
<th>Species</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Male following behavior</strong></td>
<td></td>
</tr>
<tr>
<td>Black-throated blue warbler</td>
<td>Chuang-Dobbs et al. 2001a</td>
</tr>
<tr>
<td>Blue tit</td>
<td>Kempenaers et al. 1995</td>
</tr>
<tr>
<td>Great tit</td>
<td>Hansen et al. 2009</td>
</tr>
<tr>
<td>Purple martin</td>
<td>Wagner et al. 1996</td>
</tr>
<tr>
<td>Western bluebird</td>
<td>Dickinson and Leonard 1996</td>
</tr>
<tr>
<td>Zebra finch</td>
<td>Birkhead et al. 1989</td>
</tr>
<tr>
<td><strong>Male interaction with intruding males</strong></td>
<td></td>
</tr>
<tr>
<td>Black-throated blue warbler</td>
<td>Chuang-Dobbs et al. 2001a</td>
</tr>
<tr>
<td>Harlequin duck</td>
<td>Lazarus et al. 2004</td>
</tr>
<tr>
<td>Pied flycatcher</td>
<td>Moreno et al. 2009</td>
</tr>
<tr>
<td>Zebra finch</td>
<td>Birkhead et al. 1989</td>
</tr>
<tr>
<td><strong>Proximity</strong></td>
<td></td>
</tr>
<tr>
<td>Black-throated blue warbler</td>
<td>Chuang-Dobbs et al. 2001a</td>
</tr>
<tr>
<td>Blue grosbeak</td>
<td>Estep et al. 2005</td>
</tr>
<tr>
<td>Bluethroat</td>
<td>Johnson et al. 2003</td>
</tr>
<tr>
<td>Eastern Bluebird</td>
<td>Gowaty and Bridges 1991</td>
</tr>
<tr>
<td>Golden whistler</td>
<td>van Dongen 2008</td>
</tr>
<tr>
<td>Hooded warbler</td>
<td>Fedy et al. 2002</td>
</tr>
<tr>
<td>Purple-crowned fairy-wren</td>
<td>Hall and Peters 2009</td>
</tr>
<tr>
<td>Reed Bunting</td>
<td>Marthinsen et al. 2005</td>
</tr>
<tr>
<td>Stitchbird</td>
<td>Low 2005</td>
</tr>
<tr>
<td>Western bluebird</td>
<td>Dickinson and Leonard 1996</td>
</tr>
<tr>
<td><strong>Together at nest</strong></td>
<td>Fairy martin</td>
</tr>
<tr>
<td><strong>Male song</strong></td>
<td></td>
</tr>
<tr>
<td>Great reed warbler</td>
<td>Hasselquist and Bensch 1991</td>
</tr>
<tr>
<td>Great tit</td>
<td>Cuthill and Macdonald 1990</td>
</tr>
</tbody>
</table>
Table 3. Suggested relationships between mate-guarding intensity and within-pair paternity.

<table>
<thead>
<tr>
<th>Relationship</th>
<th>Species</th>
<th>Test</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>Barn swallow</td>
<td>EPC</td>
<td>Møller 1987</td>
</tr>
<tr>
<td></td>
<td>Black-throated blue warbler</td>
<td>EPP</td>
<td>Chaung-Dobbs et al. 2001a</td>
</tr>
<tr>
<td></td>
<td>Bluethroat</td>
<td>EPP</td>
<td>Johnson et al. 2008</td>
</tr>
<tr>
<td></td>
<td>Eastern bluebird</td>
<td>EPP</td>
<td>Macdougall-Shackleton et al. 1996</td>
</tr>
<tr>
<td></td>
<td>House martin</td>
<td>EPP</td>
<td>Riley et al. 1995</td>
</tr>
<tr>
<td></td>
<td>House wren</td>
<td>EPP</td>
<td>Brylawski and Whittingham 2004</td>
</tr>
<tr>
<td></td>
<td>Pied flycatcher</td>
<td>EPP</td>
<td>Alatalo et al. 1987</td>
</tr>
<tr>
<td></td>
<td>Pied flycatcher</td>
<td>EPC, I</td>
<td>Björklund and Westman 1983</td>
</tr>
<tr>
<td></td>
<td>Red-winged blackbird</td>
<td>EPP</td>
<td>Moreno et al. 2009</td>
</tr>
<tr>
<td></td>
<td>Reed bunting</td>
<td>EPP</td>
<td>Marthinsen et al. 2005</td>
</tr>
<tr>
<td></td>
<td>Seychelles warbler</td>
<td>EPP</td>
<td>Komdeur et al. 2007</td>
</tr>
<tr>
<td></td>
<td>Stitchbird</td>
<td>FEPC</td>
<td>Low 2005</td>
</tr>
<tr>
<td></td>
<td>Western bluebird</td>
<td>EPC, I</td>
<td>Dickinson 1997</td>
</tr>
<tr>
<td></td>
<td>Wheatear</td>
<td>EPC, I</td>
<td>Currie et al. 1999</td>
</tr>
<tr>
<td></td>
<td>Yellowhammer</td>
<td>I</td>
<td>Sundburg 1994</td>
</tr>
<tr>
<td></td>
<td>Zebra finch</td>
<td>EPC</td>
<td>Birkhead et al. 1989</td>
</tr>
<tr>
<td>Neutral</td>
<td>Blue tits</td>
<td>EPP</td>
<td>Kempenaers et al. 1995</td>
</tr>
<tr>
<td></td>
<td>House Martin</td>
<td>EPP</td>
<td>Riley et al. 1995</td>
</tr>
<tr>
<td></td>
<td>Tree Swallow</td>
<td>EPP</td>
<td>Lifjeld and Robertson 1992</td>
</tr>
<tr>
<td>Negative</td>
<td>Bluethroat</td>
<td>EPP</td>
<td>Johnsen et al. 1998</td>
</tr>
<tr>
<td></td>
<td>Eastern bluebird</td>
<td>EPP</td>
<td>Gowaty and Bridges 1991</td>
</tr>
<tr>
<td></td>
<td>Purple Martin</td>
<td>EPP</td>
<td>Wagner et al. 1996</td>
</tr>
</tbody>
</table>

*Test is **bolded** if data came from a removal experiment; EPP= extra-pair paternity; EPC= extra-pair copulation; FEPC= forced extra-pair copulation; I= extra-pair male intrusion
Figure 1. Individual mate-guarding behavior trades off with EPC behavior. For both the male and the female, the intensity of mate-guarding (and participation in mate-guarding) is shaped by (1) the cost and benefits of achieving EPFs; (2) the costs of mate infidelity; and (3) a tradeoff between the engaging in EPC behavior (i.e. benefit of EPFs) and mate-guarding (i.e. cost of mate infidelity).
CHAPTER 2. MATE-GUARDING PLASTICITY IN THE AUSTRALIAN ZEBRA FINCH

I. Introduction

A. Mate-guarding

Extra-pair fertilization (EPF) is an almost ubiquitous avian reproductive tactic (Griffith et al. 2002). Sexual conflict over EPF has led to the evolution of paternity assurance strategies, including frequent copulation and mate-guarding (Birkhead and Møller 1992). When defined as the close following of a fertile female, male mate-guarding is absent only in those ecological contexts where effective guarding would be impossible (e.g. when there is intense nest site competition in colonially breeding species or when frequent male foraging trips are necessitated by courtship feeding in birds of prey, owls, and shrikes) (Møller and Birkhead 1991). Mate-guarding behavior may decrease mate infidelity (e.g. Møller 1987), but engaging in mate-guarding will trade-off with the fitness benefits of achieving EPFs as time spent mate-guarding is time not spent soliciting extra-pair copulations (EPCs) (Chuang-Dobbs et al. 2001a).

Although adaptive explanations for mate-guarding have tended to focus on male fitness consequences (Birkhead and Møller 1992), mate-guarding is likely adaptive for both males and females when there are costs to mate infidelity (Petrie and Kempenaers, 1998). Mate infidelity is costly for males in that it decreases total fitness (Webster et al. 1995) and for females in that it might decrease paternal investment in offspring (Yasukawa and Searcy 1992). In at least some contexts, both males and females are able to affect the promiscuity of their mates through a change in their mate-guarding behavior (e.g. Sandell 1998; Komdeur et al. 2007). Active female participation in male
mate-guarding has been largely disregarded, but it may be adaptive when there are fitness costs to interacting with extra-pair males (Lifjeld et al. 1994). By aiding her male in his mate-guarding efforts, females may avoid both forced EPFs (Low 2005) and harassment by extra-pair males (Kempenaers et al. 1995).

Mate-guarding behavior is also shaped by the costs of engaging in the behavior. The two most frequently discussed costs of male mate-guarding are (1) the energetic costs (Komdeur 2001); and (2) the opportunity costs of not achieving EPFs (Dickinson et al. 1997). Males suffer a decrease in weight and body condition during periods of intensive mate-guarding (Komdeur 2001), suggesting an energetic cost. Males with many opportunities to achieve EPFs may mate-guard less than those who do not (Chaung-Dobbs et al. 2001a), suggesting a cost-benefit trade-off between mate-guarding and pursuing EPFs. The costs of female mate-guarding have received little attention, but it seems reasonable to suggest that female mate-guarding would be under similar selection in situations where (1) females increase their total fitness through EPFs and mate fidelity; and (2) female mate-guarding conflicts with foraging demands. As other activities such as EPC behavior and foraging compete for an individual’s time, mate-guarding intensity should reflect (1) an individual’s risk of being cuckolded and (2) the cost of being cuckolded.

Separating the costs and benefits of mate-guarding is difficult, especially in situations where an individual’s EPC opportunity and their mate’s inclination to engage in EPC behavior are related. When female EPC behavior is driven by female choice, the same individual males should experience a small cuckoldry risk and a large opportunity
for EPFs (e.g. Green et al. 2000). The relative importance of each cost and benefit in shaping the selection pressures on mate-guarding behavior is likely affected by phylogeny and ecological context. A first step in untangling the costs and benefits would be to separately quantify the importance of each in a single social and ecological context.

The costs and benefits of mate-guarding are influenced by the interactions between the pair female, pair male, and extra-pair individuals, and they likely vary among and within species as well as over time with changing ecological conditions. There is observational and experimental evidence of substantial plasticity in the expression of mate-guarding behavior, and this plasticity is presumably indicative of the genetic variance of the trait. Within a species, males mate-guard more when their mates’ EPC opportunity increases (e.g. Dickinson and Leonard 1996); less when their mates’ EPC opportunity decreases (e.g. Komdeur 2001); more when their own EPC opportunity decreases (e.g. Johnsen and Lifjeld 1995); and less when their own EPC opportunity increases (Chuang-Dobbs et al. 2001a). There is also variation in mate-guarding intensity across changing ecological conditions (e.g. density (Komdeur 2001); synchrony (Hammers et al. 2009); and resource availability (Cuthill and Macdonald 1990)).

There is not a viable theoretical framework for modeling the evolution of mate-guarding that takes into account the costs and benefits for both males and females. Although mate-guarding behavior is structured by the dynamic interactions between multiple individuals, it is often only discussed from the perspective of the pair male.
Further, common field measures of male mate-guarding tend to discount any active role of the female. Measures of pair proximity (e.g. Chuang-Dobbs et al. 2001a; van Dongen 2008) do not account for female involvement, and some measures of male following behavior (e.g. Birkhead et al. 1989) do not account for female leaving behavior.

To investigate (1) the relative importance of the costs and benefits in structuring mate-guarding behavior and (2) the active role of the female, I conducted an experiment with the Australian zebra finch (*Teniopygia guttata*) in which I varied independently the opportunity for each member of the pair to engage in an EPC. Varying an individual’s EPC opportunity changed the cost of mate-guarding, and varying an individual’s mate’s EPC opportunity changed the benefit of mate-guarding.

**B. Study system- Australian zebra finch**

The zebra finch is a small (~12g) estrildid finch endemic to Australia (Zann 1996). Breeding easily in captivity, zebra finches have become a model system in several biological fields, including behavioral and evolutionary ecology. Renowned for their strong pair bonds (Zann 1996), rates of EPP are higher in captivity (28% of offspring (Burley et al. 1996)) than in the wild (1.7% of offspring (Griffith et al. 2010)). As a colonial and opportunistic breeder, zebra finch populations experience varying degrees of breeding density and synchrony. As both breeding density and synchrony may influence the opportunity for either member of the pair to engage in EPC behavior, birds may regularly experience variation in the both the costs and the benefits of engaging (and participating) in mate-guarding behavior.
As in other species, EPP is produced and prevented though the behavioral conflicts between (1) pair members; and (2) pair and extra-pair individuals. Frequent pair copulations and mate-guarding are both important paternity assurances in the zebra finch (Birkheak et al. 1989). In the wild and in captivity, males guard their females most intensely during the female’s fertile period (Birkhead et al. 1989). Males mate-guard by attacking intruding extra-pair males, alarm calling when the female is out of sight, and following the female’s movements (Birkhead et al. 1988; Birkhead et al. 1989). There is evidence that females may participate in being mate-guarded by delaying departure from the nest when the male is absent (Birkhead et al. 1988). Females may participate even more intensely in male mate-guarding when mated to males experimentally manipulated to be more attractive (Burley et al. 1994).

Despite male paternity guards and female participation in being guarded, EPFs do occur. Both males and females commonly solicit EPCs both in captivity and in the wild (Birkhead et al. 1988; Birkhead et al. 1989; Burley et al. 1994). There is evidence that females seek EPCs with males that are more attractive than their mate (Houtman 1992; Burley et al. 1996), and that females are largely in control of EPC through female choice (Birkhead et al. 1989; Forstmeier 2004). Forced EPCs are common (Birkhead et al. 1989; Burley et al. 1994), but as they are unlikely to result in successful EPFs, they may represent a form of harassment rather than a paternity threat (Birkhead et al. 1989). About 40% of all EPCs fail because the pair male attacks the extra-pair male (Birkhead et al. 1989).
These data indicate several important ideas: in the zebra finch, (1) low levels of
EPP are maintained though male (possibly mutual) mate-guarding; (2) forced EPCs may
constitute a form of harassment to the female; (3) the pair male can prevent EPCs and
EPFs if he is physically present; (4) females possibly select extra-pair partners based on
their relative attractiveness.

To explore the (1) relative importance of costs and benefits in structuring mate-
guarding behavior; and (2) role of the female, I exposed breeding pairs of zebra finches
to varying numbers of extra-pair stimulus birds in varying sex ratios. This allowed me to
manipulate separately the opportunity for each member of the pair to engage in an EPC;
as an individual’s EPC opportunity increased, their mate’s EPC opportunity remained
constant and vice versa. Varying an individual’s EPC opportunity changed the cost of
mate-guarding and varying an individual’s mate’s EPC opportunity changed the benefit
of mate-guarding.

After assessing the pair bond between the experimental pairs, I investigated
male mate-guarding, female mate-guarding, and female participation in male mate-
guarding. I divided mate-guarding behaviors into four main categories: (1) “pair
behavior” where intentionality could not be attributed either individual; (2) “individual
behavior” in which I separately evaluated an individual’s inclination to evade and follow
their mate; (3) “interactions with opposite sex stimulus birds” in which I evaluated each
individual’s inclination to engage in EPC behavior and their opportunity to engage in
EPCs (i.e. the efficacy of their mate’s mate-guarding efforts); and (4) “male vs. female
behavior” in which I compared the pair members relative inclinations to evade and follow their mates.

C. Predictions

Pair Bond - Are pair bonds affected by the experiment?

If pair bonds were unaffected by the experimental manipulations, I predicted that pairs would display stereotypic zebra finch pair bonding behaviors such as allopreening. As I expected stimulus birds to represent a potential source of harassment, I predicted that if pair copulations occurred, they would occur away from the stimulus birds.

Pair Behavior - Do pairs spend more or less time together when there is a greater EPC opportunity for one member of the pair? (Table 4)

Given the high cost of female infidelity, I predicted the behavior of the pair male would drive the general patterns of pair “togetherness”. As female EPC opportunity increased (i.e. the threat to within-pair paternity increased), I expected the pair to spend more time together and shorter periods of time apart. As male EPC opportunity increased, I expected opposite trends.

Individual Behavior - How is individual leaving and following behavior affected by (1) EPC opportunity; and (2) mate’s EPC opportunity? (Table 4)

Female leaving and active male mate-guarding
As female EPC opportunity increased, I predicted that females would participate in being mate-guarded (i.e. leave their mates less). In response to the greater paternity threat, I expected males to follow their mates more.

As male EPC opportunity increased, I predicted that females would engage less in other activities (like EPCs) and would consequently leave their males less. Due to the increasing opportunity cost of forgoing EPC behavior, I predicted that males would actively mate-guard (i.e. follow their mates) less.

Male leaving and active female mate-guarding
As female EPC opportunity increased, I predicted that males would engage less in other activities (like EPCs) and that they would leave their mates less frequently; in response to being left less, I expected that females would actively mate-guard (i.e. follow their mates) less.

As male EPC opportunity increased, I predicted that males would leave their females more frequently in order to actively solicit EPCs from a stimulus female. In response to being left more frequently, I expected females to follow their mates more.

**Interactions with Opposite Sex Stimulus Birds** - Are individuals less inclined to engage in EPC behavior as their risk of being cuckolded increases? When an individual’s EPC opportunity increases, do individuals mate-guard less effectively (i.e. allow their mate to be alone with potential EPC partner for longer periods)? (Table 4)
As female EPC opportunity increased, I expected males to engage less in their own EPC behavior and consequently to visit a stimulus female less. Similarly, as male EPC opportunity increased, I also expected females to engage less in their own EPC behavior and visit a stimulus male less.

Despite a predicted decrease in active female mate-guarding as female EPC opportunity increased, I predicted that due to the high costs of female infidelity, focal males would spend less time alone with the stimulus female. In contrast, as male EPC opportunity increased, I predicted that females would spend more time alone with the stimulus male due to the decrease in male mate-guarding intensity.

**Male vs. female behavior** - *Is one member of the pair more inclined to follow or evade their mate than the other?*

Given the higher costs of female infidelity, I predicted that males would follow females mates more frequently than females would follow males.
Table 4. Predictions for each measured behavior.

<table>
<thead>
<tr>
<th>Behavior</th>
<th>Increasing stimulus males (1 stimulus female)</th>
<th>Increasing stimulus females (1 stimulus male)</th>
</tr>
</thead>
<tbody>
<tr>
<td>PAIR BEHAVIOR</td>
<td></td>
<td></td>
</tr>
<tr>
<td>% time in close proximity</td>
<td>↑</td>
<td>↓</td>
</tr>
<tr>
<td>% time in eyesight</td>
<td>↑</td>
<td>↓</td>
</tr>
<tr>
<td>Average time spent out of eyesight</td>
<td>↓</td>
<td>↑</td>
</tr>
<tr>
<td>INDIVIDUAL BEHAVIOR</td>
<td></td>
<td></td>
</tr>
<tr>
<td>FEMALE leaves mate (interest in EPC)</td>
<td>↓</td>
<td>↓</td>
</tr>
<tr>
<td>MALE follows mate (active MG response to being left)</td>
<td>↑</td>
<td>↓</td>
</tr>
<tr>
<td>MALE leaves mate (interest in EPC)</td>
<td>↓</td>
<td>↑</td>
</tr>
<tr>
<td>FEMALE follows mate (active MG response to being left)</td>
<td>↓</td>
<td>↑</td>
</tr>
<tr>
<td>INTERACTIONS WITH OPPOSITE SEX STIMULUS BIRDS</td>
<td></td>
<td></td>
</tr>
<tr>
<td>MALE visits to stimulus female (interest in EPC)</td>
<td>↓</td>
<td></td>
</tr>
<tr>
<td>FEMALE visits to stimulus male (interest in EPC)</td>
<td></td>
<td>↓</td>
</tr>
<tr>
<td>FEMALE time alone with stimulus male (efficacy of male MG / interest in EPC)</td>
<td></td>
<td>↑</td>
</tr>
<tr>
<td>MALE time alone with stimulus female (efficacy of female MG / interest in EPC)</td>
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</table>
II. Methods

A. Breeding colony and pair formation

Our captive outbred breeding colony consisted of 200-300 wild type zebra finches. All birds were maintained on a constant 14:10 light:dark photoperiod at approximately 19°C with a nutritionally complete seed mix, water, cuttlebone, and grit available ad libitum. Excluding siblings and half-siblings, I randomly paired 18 adult males and females. Each pair was housed in free flight cage measuring approximately 50cm x 30cm x 40cm and was provided with a plastic hooded nest box and nesting material.

If the randomly assigned pair did not begin nesting behavior within 3 weeks, I repaired the female a new male. I considered the pair bond to be established when the pair built a nest and began laying eggs. Female zebra finches are fertile between day -11 and day +3 (where day 0 is the day the first egg is laid) (Birkhead et al 1989). To ensure the fertility of the experimental female, I timed the use of the pair so that the female was laying immediately prior to when I began the experiment.

B. Experimental chamber

In order to investigate how mate-guarding behavior varied in response to changes in the opportunity for each member of the pair to engage in an EPC, it was necessary to design an experimental chamber where this variable could be manipulated. Within the cross-shaped experimental chamber (Figure 2), a pair could be exposed to up to four extra-pair stimulus birds at a time. There were five main areas within the chamber: one central area with no line of sight to the extra-pair cages, and four arms (each designated...
When present, the extra-pair stimulus birds were confined to small individual cages located at the ends of each arm of the chamber. While the stimulus birds were physically separated from the focal pair by thin metal bars, all birds were able to interact visually and acoustically. The chamber was 1.54m across, had 0.61m² of floor space and was 0.42m tall (total volume of 0.27 m³). The outer walls of the chamber were constructed of opaque plastic and the ceiling was Plexiglas in order to allow birds in the chamber to be video recorded from above. To facilitate identification, the focal male in each trial was painted with a small (under 3mm²) drop of white correction fluid on his cap.

When an experimental pair had constructed a nest and begun to lay eggs, I moved them to the experimental chamber. The chamber was in a separate room from the rest of the colony. I gave the pair seven days to acclimate to the space before commencing the experiment. Based on a preliminary study using fecal droppings as evidence of visitation, seven days was enough time for most pairs to explore all arms of the chamber.

C. Experimental trials

After the acclimatization period, I conducted six trials, where the number and sex ratio of stimulus birds defined a trial (Table 5). The first trial was always the control trial (“trial 0”) in which no stimulus birds were presented. The following five trials were conducted in random order as determined by a random numbers table. Each trial lasted 3 hours and was video recorded from above with a Sony HDR-SR1 digital video camera affixed...
with a wide-angle lens attachment. One to two trials were conducted each day. The first trial of the day was conducted 1 - 3 hours after the lights came on. The second trial of the day was conducted 1 hour after the first trial ended. The pair was in the chamber for a total of 11 - 13 days (7 acclimation days and 4 - 6 experimental days).

The stimulus birds presented in each trial were selected from a group of 28 adult females and 35 adult males set aside from the breeding colony. Over the course of the 5 trials, each focal pair was exposed to 6 stimulus males and 6 stimulus females. The same stimulus bird was never presented twice to the same focal pair, and the same 12 stimulus birds were not presented to every pair. I used a random numbers table to select stimulus birds and to determine in which arm of the chamber to place each stimulus bird in a particular trial.

To score the video recording of each trial, I divided the experimental chamber into 13 regions (Figure 3) and created a transcript of each focal bird’s movements. Each time a bird moved from one region to another I recorded the movement along with the time stamp.

D. Pair bond in the experimental chamber

Are pair bonds affected by the experiment?

To evaluate if the pairs were behaving appropriately (i.e. not overtly stressed and consequently disinterested in mating or courtship activities), I documented the occurrence of pair bonding activities. In each trial I recorded each copulation, allopreening event, and bout of nest building behavior - although pairs did not have a
nest box, males would rip up the newspaper lining the floor of the chamber in an attempt to nest build.

To test for an overall effect of the stimulus birds on pair copulation behavior, I recorded the location of each copulation as (1) within eyesight of a stimulus male; (2) within eyesight of a stimulus female; or (3) out of eyesight of a stimulus bird.

E. Measurements of mate-guarding behavior

a. Pair behavior

Do pairs spend more or less time together when there is a greater EPC opportunity for one member of the pair?

I used the relative positioning of the focal male and female in the 13 regions of the test chamber (Figure 3) to calculate pair proximity. A pair was considered to be in close proximity if individuals were in the same region or in adjacent regions. Using this method of calculation a pair couldn’t be considered “close” if the birds were more than 0.63m apart.

To calculate the percentage of each trial that the pair spent within eyesight of each other, I divided the experimental chamber into 5 areas based on line-of-sight demarcations (Figure 4). Birds were considered out of eyesight when they were in different areas. Using these 5 areas I also calculated the average period of time the birds spent out of one another’s eyesight.
b. Individual behavior

How is individual leaving and following behavior affected by (1) EPC opportunity; and (2) mate’s EPC opportunity?

I counted the total number of times that each individual left and followed their mate in each trial. I understood “leaving” to represent an individual’s lack of participation in mate-guarding (or their level of interest in another activity) and “following” to represent active mate-guarding.

Leaving and following behavior was scored based on line-of-sight demarcations (Figure 4); an individual was scored as leaving when they flew out of their mate’s line-of-sight and as following when they restored line-of-sight within 27 seconds of their mate leaving. I felt that it was important to assign a time cut-off for following behavior. Without assigning a cutoff, the maximum latency to follow was 1 hour and 7 seconds; I didn’t feel that a move after such a long period of time was truly a “following” response to being left. I chose 27 seconds as a cut-off because 90% of all “following” moves occurred within this timeframe.

While an individual’s “leaving” movement was independent of their mate’s behavior, a “following” movement depended, by definition, on their mate’s leaving behavior. I evaluated leaving and following behavior separately (instead of evaluating the percentage of moves followed) in order to preserve the independence of the leaving score. When there were significant differences in the focal bird’s leaving score, I compared the overall proportion of moves that were followed.
c. Interactions with opposite sex stimulus birds

*Are individuals less inclined to engage in EPC behavior as their risk of being cuckolded increases? When an individual’s EPC opportunity increases, do individuals mate-guard less effectively (i.e. allow their mate to be alone with potential EPC partner for longer periods)?*

For each trial in which there were stimulus birds of the opposite sex to the focal bird present I calculated (1) the number of times the focal bird visited the opposite sex stimulus bird; and (2) the total amount of time the focal bird spent alone with that stimulus bird. Visits to the stimulus bird were calculated by counting the times the focal bird ventured to either the near or far region of the stimulus arm (when the focal bird was within 0.42m of the stimulus cage). Time alone with the stimulus birds was calculated by summing the time the focal bird spent in the far region of an arm when the bird’s mate was neither in that region nor in the adjacent region. For example, a focal bird would be counted as visiting the north arm if he ventured to ‘north_{far}’ or ‘north_{near}’, and a focal male would be considered to be alone in the north arm if he was in ‘north_{far}’ and his mate was not in ‘north_{far}’ or in ‘north_{near}’ (Figure 3).

For this set of measurements, scores for each trial were corrected based on the focal bird’s behavior in that arm in the control trial. For example, if a focal female visited the north arm 3 times in the control trial and 10 times in a trial where there was a male stimulus bird in the north arm, the female would be reported as visiting male stimulus bird +7 times. If she had spent 20 minutes alone in the north arm during the control trial and 8 minutes alone in that arm when a male stimulus was present, she would be
reported as spending -12 minutes alone with the male stimulus bird. Calculating these metrics relative to the control trial (where there were no stimulus birds present) helped to adjust my measurements for any inherent bias in movement or arm preference by birds in the chamber.

d. Male vs. female behavior

*Is one member of the pair more inclined to follow or evade their mate than the other?*

To contrast male and female behavior I compared male and female leaving and following behavior in trial 3 where I presented the pair with 1 stimulus male and 1 stimulus female. I chose to compare pair member behavior in this trial because it was the closest to natural conditions; extra-pair individuals of each sex were present but not in a skewed sex ratio.

F. Statistical analyses

*Pair bond in the experimental chamber*

I tested for an affect of stimulus bird presence on pair copulation behavior with a chi-square goodness of fit test comparing the observed locations of copulation events against the expected locations based on a random distribution.

*Measurements of mate-guarding behavior*

To examine the measures of mate-guarding behavior, I separated the 5 experimental trials into 2 separate groups for analysis. In the first group (trials 2,3,5), only the number
of stimulus males increased; in the second group (trials 1, 3, 4), only the number of stimulus females increased (Figure 5).

Pair behavior
Using a repeated-measures ANOVA where pair identity was the repeated measure, I compared the proportion of time in close proximity, proportion of time in eyesight, and average period of time out of eyesight (1) as the number of stimulus males increased; and (2) as the number of stimulus females increased.

Individual behavior
Using a repeated-measures ANOVA where pair identity was the repeated measure, I compared female leaving, male following, male leaving, and female following (1) as the number of stimulus males increased; and (2) as the number of stimulus females increased. When there were significant differences in the number of times one pair member left, I compared the overall percentage of moves that their mate followed using a repeated-measures ANOVA where pair identity was the repeated measure. For this analysis, I excluded pairs for which there were no leaving movements in one or more trials.

Interactions with opposite sex stimulus birds
Using a repeated-measures ANOVA where pair identity was the repeated measure, I compared (1) individual visits to the opposite sex stimulus bird as the mate’s EPC
opportunity increased and (2) individual’s total time alone with the opposite sex stimulus bird as the mate’s EPC opportunity increased.

**Male vs. female behavior**

I compared leaving and following behavior between the sexes with a paired samples Student’s t-test.

All analyses were conducted with PASW Statistics v17 (Chicago, IL, USA) employing two-tailed tests of probability and an alpha level of 0.05. Statistical tests in which there was no significant difference detected but the $P$-value was 0.05-0.01 will be referred to as “trends” or “tendencies.”
Table 5. Number of stimulus birds presented in each experimental trial.

<table>
<thead>
<tr>
<th>Trial</th>
<th>Number of Stimulus Females</th>
<th>Number of Stimulus Males</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>0</td>
<td>1</td>
</tr>
<tr>
<td>2</td>
<td>1</td>
<td>0</td>
</tr>
<tr>
<td>3</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>4</td>
<td>3</td>
<td>1</td>
</tr>
<tr>
<td>5</td>
<td>1</td>
<td>3</td>
</tr>
</tbody>
</table>
**Figure 2.** Experimental chamber from above. Dark lines represent perches. Dashed lines represent wire barrier. Extra-pair stimulus birds are confined to cages represented by shaded areas. Food and water were provided in the center of the chamber.

**Figure 3.** Chamber regions used to generate movement transcripts. Chamber divided into 13 regions based on line of sight and proximity to extra-pair stimulus cages. Each arm is sub-divided into 3 regions. (Perches omitted.)
**Figure 4.** Chamber divided into 5 areas based on line-of-sight demarcations. (Perches omitted).

**Figure 5.** Independently manipulating male and female EPC opportunity. Moving from bottom left to top right, the number of stimulus males increases, but the number of stimulus females remains the same. Moving from top left to bottom right, the number of stimulus females increases, but the number of stimulus males remains the same.
III. Results

A. Pair bond in the chamber

Most pairs showed clear evidence of maintaining their pair bond while in the experimental chamber. Of the 18 experimental pairs, 15 were observed allopreening at some point during the experiment (Table 6). Over two-thirds of the pairs that allopreened also copulated or attempted to nest build. As allopreening is considered a stereotypic sign of pairing in zebra finches (Zann 1996), I excluded the 3 pairs that did not allopreen from subsequent analysis, reducing my sample size to 15 experimental pairs. Although the excluded pairs had been nesting prior to the experiment, their introduction into the experimental space may have disrupted either their pair bond or the female’s fertility status.

In the 324 hours of recorded observation, 9 of the 15 pairs copulated a total of 19 times. Two of these copulations (10.5%) occurred within eyesight of a stimulus bird; one occurred within eyesight of a male stimulus bird and the other within eyesight of a female stimulus bird. Copulations were more likely to occur out of eyesight of a stimulus bird than would be expected from a distribution based solely on the proportion of the experimental chamber within eyesight of stimulus birds ($\chi^2 = 6.88, N = 19, P = 0.032$).

B. Mate-guarding behavior

a. Pair behavior

Pairs stayed close together in all trials; they spent 72-76% of time in close proximity and 79-85% of the time within eyesight of each other.
The number of stimulus males had little influence on the percent of time a pair spent in close proximity ($F_{2,28} = 0.03, P = 0.968$), the percent of time a pair spent within eyesight ($F_{2,28} = 0.15, P = 0.864$), or the average period of time a pair spent out of eyesight ($F_{2,28} = 0.76, P = 0.480$). Similarly, the number of stimulus females had no detectable effect on proximity ($F_{2,28} = 0.26, P = 0.772$), eyesight ($F_{2,28} = 0.91, P = 0.415$), or the average time out of eyesight ($F_{2,28} = 0.28, P = 0.757$).

b. Individual behavior

Female leaving and active male mate-guarding

The number of stimulus males did not greatly influence the number of times the female left the male ($F_{2,28} = 2.42, P = 0.107$), although there was a tendency for the female to leave less as the number of stimulus males increased from 1 to 3 (a priori contrasts $F_{1,14} = 3.75, P = 0.073$, partial $\eta^2$ (effect size) = 0.202) (Figure 6). However, the number of stimulus males did appear to influence the number of times that the male followed the female ($F_{2,28} = 4.20, P = 0.025$) (Figure 6). As the number of stimulus males increased from 1 to 3, males followed their mates significantly less (a priori contrasts: $F_{1,14} = 8.16, P = 0.013$, partial $\eta^2 = 0.368$).

The number of stimulus females had no noticeable effect on female leaving ($F_{2,28} = 0.15, P = 0.857$) or male following ($F_{2,28} = 0.33, P = 0.722$) behavior.
Male leaving and active female mate-guarding

The number of stimulus males influenced both the number of times the male left the female ($F_{2,28} = 3.99$, $P = 0.030$) and the number of times that the female followed the male ($F_{2,28} = 6.99$, $P = 0.003$) (Figure 7). As the number of stimulus males increased from 0 to 3, males left their mate’s eyesight less (a priori contrasts: $F_{1,14} = 7.45$, $P = 0.016$, partial $\eta^2 = 0.347$) and females followed their mates less (a priori contrasts: $F_{1,14} = 12.30$, $P = 0.003$, partial $\eta^2 = 0.467$). However, the overall percentage of moves that the female followed was not influenced by the number of stimulus males ($F_{2,24} = 0.07$, $P = 0.932$).

The number of stimulus females had no effect on male leaving ($F_{2,28} = 0.98$, $P = 0.386$) or female following ($F_{2,28} = 2.22$, $P = 0.127$) behavior.

c. Interactions with opposite sex stimulus birds

Visits to potential EPC partners

The number of stimulus males had an effect on the focal male’s tendency to visit a stimulus female ($F_{1,35, 19.0} = 5.03$, $P = 0.028$) (Figure 8). Mauchley’s test indicated that these data had violated the assumption of sphericity ($W = 0.524$, $P = 0.015$) so degrees of freedom were corrected using Greenhouse-Geisser estimates of sphericity ($\varepsilon = 0.678$). When the number of stimulus males increased from 0 to 3, males visited the female stimulus birds less (a priori contrasts: $F_{1,14} = 7.47$, $P = 0.016$, partial $\eta^2 = 0.347$).

The number of stimulus females did not have a detectable effect on the number of times the female visited the stimulus male ($F_{2,28} = 0.20$, $P = 0.822$).
*Time alone with potential EPC partners*

The number of stimulus females did not greatly influence the female’s total time alone with the stimulus male ($F_{2,28} = 0.68, P = 0.513$), and the number of stimulus males did not greatly influence the male’s total time alone with the stimulus female ($F_{2,28} = 0.39, P = 0.679$).

**d. Male vs. Female Behavior**

When exposed to stimulus birds in an even sex ratio, the focal male and female were equally likely to leave one another ($t_{14} = 0.33, P = 0.743$), although there was a slight tendency for the male to follow the female more than the female followed the male ($t_{14} = 1.94, P = 0.073$) (Table 7).
Table 6. Pair bond behavior in the experimental chamber. Due to a lack of stereotypic pair bonding behaviors, pairs 1, 4, and 11 were excluded from subsequent analysis.

<table>
<thead>
<tr>
<th>Pair</th>
<th>Nest build</th>
<th>Allopreen</th>
<th>Copulate (at least once)</th>
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<tbody>
<tr>
<td>1</td>
<td>X</td>
<td>X</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>X</td>
<td>X</td>
<td>X</td>
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<td>X</td>
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<td>6</td>
<td>X</td>
<td>X</td>
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<td>7</td>
<td>X</td>
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<td>9</td>
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<td>10</td>
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<tr>
<td>18</td>
<td>X</td>
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Table 7. t-test results: male vs. female behavior in trial 3. There were no significant differences in male and female behavior when there the pair was exposed to 1 stimulus male and 1 stimulus female.

<table>
<thead>
<tr>
<th></th>
<th>Male</th>
<th>Female</th>
<th>df</th>
<th>P-value</th>
</tr>
</thead>
<tbody>
<tr>
<td># of leaves (mean ±SE)</td>
<td>38.07 ± 9.44</td>
<td>34.67 ± 7.55</td>
<td>14</td>
<td>0.743</td>
</tr>
<tr>
<td># of follows (mean ±SE)</td>
<td>22.47 ± 5.91</td>
<td>12.85 ± 3.32</td>
<td>14</td>
<td>0.105</td>
</tr>
<tr>
<td>% of mate’s moves followed (mean ±SE)</td>
<td>62.57 ± 6.69</td>
<td>43.81 ± 7.40</td>
<td>14</td>
<td>0.073</td>
</tr>
</tbody>
</table>
Figure 6. Female leaving and male mate-guarding as number of stimulus males increased. Mean ± SE number of times that the female left the male (□) and the male followed the female (■). As the number of stimulus males increased, females did not significantly change the rate at which they left their mates line-of-sight, but males followed their mates significantly fewer times per trial.
Figure 7. Male leaving and female mate-guarding as number of stimulus males increased.
Mean ± SE number of times that the male left the female (■) and the female followed the male (□). As the number of stimulus males increased, males left females significantly fewer times per trial, and females followed males significantly fewer times per trial.
Figure 8. Focal male EPC behavior as number of stimulus males increased. Mean ± SE number of times that the male visited the stimulus female arm compared to the number of times he visited that arm when it was empty in the control trial. Males visited the stimulus female less as the number of stimulus males increased.
IV. Discussion

My results indicate that the female zebra finches in my experimental chamber did not actively mate-guard, but that males did. Further, male mate-guarding behavior was affected more by the relative threat to within-pair paternity than by the relative opportunity to achieve an EPF. While there was not conclusive evidence of female participation in male mate-guarding, there was a tendency for females to leave males less as female EPC opportunity increased.

A. Pair behavior

When exposed to stimulus birds in an even sex ratio, pairs in the experimental chamber behaved similarly to pairs breeding in an open aviary (Birkhead et al. 1989). In both situations, males followed a higher proportion of their mate’s moves than females did, although the trend was not statistically significant in the chamber ($P = 0.073$).

This trend in following behavior and the frequency of pair bonding activities such as copulation, allopreening, and nest building indicated that pairs were behaving normally in the experimental chamber. As pair behaviors are similar for pairs caged alone, pairs breeding in an open aviary, and pairs in the wild (Birkhead et al. 1988; Birkhead et al. 1989), I have reason to believe that patterns in mate-guarding within the experimental chamber should reflect patterns in mate-guarding in a more natural setting.
B. Flexible male behavior

As the threat to within-pair paternity increased (i.e. the number of stimulus males increased), males left their mates less and visited an extra-pair stimulus female fewer times. Although the change in male behavior did not affect the total amount of time that the pair spent in close proximity or within eyesight, these results suggest that males are less inclined to pursue EPCs when there is a high risk of being cuckolded. To maintain a constant (and possibly optimal) level of mate-guarding behavior, other activates (e.g. solicitation of extra-pair females) may decrease as the cuckoldry risk increases.

Contrary to predictions, males followed their mates less as the paternity threat increased; however, as males followed the same overall proportion of female moves, this pattern in male behavior was likely driven by the non-significant tendency ($P = 0.073$) for females to leave their mates less as the number of stimulus males increased. While female behavior could explain the downward trend in male following behavior, it does not explain why males did not follow their mates more closely as the paternity threat increased.

Male following behavior may not be sensitive to an increase in the paternity threat if males consistently mate-guard at a maximum intensity. Regardless of the relative risk of cuckoldry, a fertile female may always warrant close guarding; in other words, the even intensity of active male following may reflect the high cost of not mate-guarding under all treatment conditions. When breeding densities are at their highest, zebra finch nests may rest side-by-side (Zann 1996), and there may be high selection
pressure for males to mate-guard intensely. Although maintaining high intensity mate-guarding may be costly when cuckoldry risk is low, the costs of under-guarding a female may far outstrip the costs of over-guarding a female. In the zebra finch, consistently high intensity male mate-guarding may be more adaptive than plastic mate-guarding according to cuckoldry risk.

An experimental increase in the perceived EPC opportunity for males did not affect male behavior in the chamber. Although males seemed to be less inclined to pursue EPCs when the paternity threat was greatest (i.e. when there were more stimulus males), males were not more inclined to pursue EPCs when there was presumably a greater opportunity to achieve EPFs (i.e. when there were more stimulus females). As the number of stimulus females increased, males left and followed their mates with similar frequencies. Across these trials, mate-guarding intensity was likely influenced by the one stimulus male that was present in every trial. If EPFs are under female control (Forstmeier 2004), then it may not make adaptive sense to actively pursue EPCs when there is any chance at all of being cuckolded (i.e. when the female is fertile.)

These results support the idea of a trade-off between mate-guarding and pursuing EPFs (e.g. Alatalo et al. 1987); as the paternity threat increased, males were less inclined to pursue EPCs. However, these results do not wholly corroborate findings from other species that the intensity of male mate-guarding fluctuates according to the paternity threat (e.g. Komdeur 2001) and the opportunity to solicit EPCs (e.g. Chaung-Dobbs et al. 2001a). Contrary to previous findings, zebra finch males did not mate-guard
more intensely as the threat to paternity increased, but rather maintained relatively 
constant mate-guarding behavior. They also maintained similar mate-guarding behavior 
as their EPC opportunity increased.

In the zebra finch, male mate-guarding seems to be structured by the benefits of 
mate-guarding more than by the costs; in other words, for male zebra finches, the 
fitness costs of losing paternity are relatively more important than the fitness benefits 
of pursuing EPFs. These results could imply that for minimally promiscuous species, 
paternity assurance is more important than EPCs. In these species, EPFs may be the 
result of EPCs occurring when the extra-pair male’s mate is not fertile.

C. Flexible female behavior

There was little evidence of female participation in male mate-guarding and no evidence 
of active female mate-guarding. As the perceived opportunity for the female to achieve 
EPCs increased (i.e. when there were more stimulus males), there was a non-significant 
tendency for females to leave their mates less (P = 0.073), suggesting that females assist 
male mate-guarding. This tendency could reflect the high frequency of extra-pair male 
intrusions in open aviaries and the wild. Although forced EPCs may not represent a 
viable paternity threat (Birkhead et al. 1989), they could constitute a form of female 
harassment. Although the data do not fully support a claim of female participation, they 
do show that females do not attempt to elude their mates more as their opportunity to 
achieve EPFs increases. In the zebra finch, a greater opportunity to achieve EPFs does 
not seem to influence female inclination to engage in EPC behavior.
My results indicate that female zebra finches did not actively mate-guard in the experimental chamber. As perceived risk of male infidelity increased, there was no change in female leaving or following behavior. This could reflect a low cost of male infidelity for females.

D. Alternative explanations

My experimental design necessitated preventing physical interaction between pair and stimulus birds; this physical separation may have prevented the stimulus individuals from being perceived as either a threat to within-pair paternity or as a potential EPC partner. The best evidence that the stimulus birds were perceived as a threat to paternity is that pair copulations were more likely to take place out of eyesight of stimulus individuals. In open aviaries, pair copulations are frequently unsuccessful due to interruptions by extra-pair birds (Burley et al. 1994; Birkhead et al. 1988); therefore there is reason to expect that, when possible, pair copulations should occur away from potential disruptions. In this experiment, the observed locations of pair copulations suggest that pairs did perceive stimulus birds as potentially disruptive. However, physical separation may still have decreased the effect of extra-pair male harassment on female behavior. Females may have participated minimally in male mate-guarding simply because the wire barrier prevented physical harassment by the extra-pair males. Given the limitations of this experimental design and the modest sample size, female participation in male mate-guarding may warrant further examination; I am not proposing that we rule out an active role for females in mate-guarding.
Another alternative explanation for the patterns I found could be the individual variation in the stimulus birds’ attractiveness as extra-pair partners, specifically the fertility status of stimulus female and the relative attractiveness of stimulus male. The stimulus females were selected from a larger pool of unpaired females housed in single sex cages. Although these females often laid eggs, I could not be sure of an individual female’s fertility status when she was used as a stimulus bird. A non-fertile stimulus female may not have represented a sufficiently enticing potential EPC partner for the pair male. This could have led to an underestimate of the effect of male EPC opportunity on male and female mate-guarding behavior.

I did not attempt to quantify stimulus male attractiveness, but there is evidence that variation in extra-pair male attractiveness is important in affecting the outcome of extra-pair encounters (Forstmeier 2007). In this experiment, I manipulated female EPC opportunity by varying the number of potential EPC partners, but females may perceive EPC opportunity as a function of extra-pair male quality; in other words, female behavior in the chamber may have been influenced more by the quality of the stimulus males that were present and less by the overall number of stimulus males. If my experimental manipulations did not have the intended effect, I may have misinterpreted the effect of female EPC opportunity on male and female mate-guarding behavior. Future studies should consider the effect of stimulus bird quality on both male and female mate-guarding decisions.
D. Conclusions

It appears that for zebra finch males, protecting paternity is more important than pursuing EPFs. As the threat to paternity increases, there is a trade-off between mate-guarding and soliciting EPCs. Even though female behavior did not change as female EPC opportunity increased, aspects of male behavior did change in response to the threat that this female EPC opportunity posed. If mate-guarding is effective in limiting female interaction with extra-pair males (Birkhead et al. 1989), then an increase in male mate-guarding may serve more to prevent extra-pair male intrusions than to prevent female forays.

If zebra finch behavior in the chamber is similar to behavior in the wild, then it is possible that low levels of EPP are maintained through high levels of male mate-guarding, possible female participation in male mate-guarding, and low levels of female pursuit of EPFs. If male pursuit of EPFs is highest where there is little risk of cuckoldry, the paternity threat a male faces may be greatest under conditions of lower breeding synchrony (i.e. when extra-pair males are not guarding their own mates). EPFs may occur either when the female evades or extra-pair male overwhelms the pair male’s paternity guards.

Patterns in female EPC opportunity (i.e. cuckoldry risk) will not always predict EPP patterns if there are flexible paternity assurance strategies. Further, patterns in mate-guarding intensity will not always predict patterns in EPP if a limited measure of mate-guarding is employed. Although general measures of pair proximity did not vary in this study, changes in the perceived risk of cuckoldry did alter male leaving behavior.
The theoretical framework underlying the evolution of mate-guarding behavior should be expanded to include explicit reference to the active role of the female. Just as the adaptive function of EPP is explored from the male and female perspective, the adaptive function of mate-guarding behavior should also be explored from both perspectives. A more inclusive theoretical framework would help to generate clear testable predictions about mate-guarding intensity as the costs of having an unfaithful mate varied. In order to test these predictions, future investigations of mate-guarding, both in terms of flexibility and efficacy, should focus separately on males and females. When there are reasons to avoid interactions with extra-pair individuals, active participation in mate-guarding should not be discounted. Only after individually evaluating male and female pursuit and prevention of EPCs can there be clear predictions about the relationships between (1) ecological variables and mate-guarding; and (2) ecological variables and EPP patterns.
REFERENCES


VITA

I was born in Lansing, MI in 1984. Growing up, I spent time in Michigan, New Zealand, Illinois, Maryland, and Spain. I first became interested in birds and their many behaviors while studying biology at Oberlin College in Oberlin, OH. After graduating with a Bachelor of Arts in 2006, I worked as a field assistant on several projects that focused on the demography and social behavior of different species, including the black-throated blue warbler and the red-backed fairy-wren. I entered the master's program at the College of William & Mary in 2008, and defended my thesis in July 2010. In Fall 2010, I will enter the doctoral program in Evolution, Ecology, and Behavior at Indiana University-Bloomington.