ENDOCRINE CORRELATES OF SOCIAL AND REPRODUCTIVE BEHAVIOURS IN A GROUP-LIVING AUSTRALIAN PASSERINE, THE WHITE-BROWED BABBLER

A thesis submitted in partial fulfillment of the requirements

for the award of the degree

DOCTOR OF PHILOSOPHY

from

UNIVERSITY OF WOLLONGONG

by

Suzanne Oppenheimer
B.A., M.A.

Department of Biological Sciences
2005

ABSTRACT

To investigate the evolutionary rationale for the seemingly altruistic behaviours commonly seen in cooperatively breeding Australian passerines, I examined alloparental behaviour in the White-browed Babbler *Pomatostomus superciliosus* (WBBA). Toward this end, I analysed behavioural, hormonal, and genetic factors in both free-living and captive WBBAs. Studies of free-living birds examined social and reproductive behaviours and hormonal correlates to reproduction. With captive birds, I performed both observations and manipulative experiments focusing on intragroup social structure, social behaviours, and the endocrine correlates to such structure and behaviours.

The WBBA was selected as a study species as they live in sedentary, year-round social groups that engage in cooperative breeding. Field work was conducted in Back Yamma State Forest, in the central west region of New South Wales, Australia. In this population of WBBAs, groups included many close genetic relatives, and neighboring groups also shared several related individuals. There were multiple breeding pairs within most groups, and reproductive behaviours between breeding pairs were similar to those of many biparentally breeding songbirds. However, nest defense and post-fledgling care were undertaken by large cooperative groups.

In free-living WBBAs, plasma levels of testosterone (T), estradiol (E2), progesterone (P), prolactin (Prl), and corticosterone (B) were measured, and laparotomies were performed to ascertain gonadal condition. Endocrine profiles in WBBAs were similar to those reported for a number of passerines and likely reflected physiological changes necessary for breeding, such as spermatogenesis and ovulation. Males' T profiles resembled those of some polygynous passerines, in that plasma T

levels rose after the completion of the female partners' clutch. This may reflect the possibility for extra-pair copulations in WBBA groups with multiple breeding females. There was some indication that WBBAs' endocrine system may have been fine-tuned to support alloparental behaviour. In adult males that chaperoned fledglings and juveniles, elevated plasma Prl titres may have facilitated alloparental care. Furthermore, elevated plasma P levels in some adult females may have been related to non-breeding behaviour and perhaps also to care of post-fledging young. Unlike many temperate zone species, many WBBAs maintained recrudesced gonads for much of the year, reflecting their extended (if not perpetual) breeding season.

Studies on captive WBBAs were conducted in aviaries at the University of Wollongong. Extensive observations were undertaken to investigate intragroup social structure and associated allofeeding behaviour. Despite an absence of aggression, intragroup social structure appeared stratified and was maintained by ritualised behaviours and vocalisations. In particular, allofeeding behaviour appeared to act as an important social signal within groups, indicating high social standing of the feeder and low social status of the receiver. Plasma levels of B and Prl were measured and compared to social factors, but I found no hormonal correlates to WBBA social status or behaviour in groups with stable social structures.

To further examine the relationship between the endocrine system and social behaviours and structure, manipulative experiments were carried out on captive WBBAs. Removal of group members from socially stable groups elicited no overt aggression, and exchange of members between groups elicited little aggressive behaviour; however, both experiments resulted in significant social restructuring. Nevertheless, I found no significant hormonal correlates (T, E2, and B were measured) to social instability caused by these perturbations. Another social behaviour, roost nest

building, was correlated with elevated plasma T and E2 levels, in some months of the year but not others.

Field and captive studies of the WBBA supported hypotheses suggesting that (1) alloparental behaviours evolved via kin selection mechanisms and (2) alloparental behaviours are important signals of quality used to help select mates and/or attract collaborators. In WBBAs alloparental behaviours seem to be either directed toward kin or co-opted as a means of advertising social status.

TABLE OF CONTENTS

Page

Part 1: Literature Review

Part 2: Field Studies of Free-living White-browed Babblers

Chapter II. Natural History and Morphometrics of White-browed Babblers in Back Yamma State Forest

Chapter III. Gonadal and Hormonal Phenologies in Free-living White-browed Babblers

Part 3: Behavioural and Endocrine Studies of Captive White-browed Babblers

Chapter IV. White-browed Babbler Group Organisation and Social Behaviour

Chapter V. Physiological Correlates of Social Behaviour During Periods of Social Stability

Chapter VI. Physiological Correlates of Social Behaviour During Periods of Social Instability

Chapter VII. Physiological Correlates of Social Behaviour During Periods of Intense Social Interactions Associated with Roost Nest Building

List of Tables

List of Figures

List of Abbreviations

ABBBS	Australian Bird and Bat Banding Scheme
B	corticosterone
E2	estradiol
BYSF	Back Yamma State Forest
F	cortisol
P	progesterone
Prl	prolactin
T	testosterone
WBBA	White-browed Babbler

Acknowledgements

I would like to thank all those who made this study possible and enjoyable. Special thanks go to my advisers, Bill Buttemer and Lee Astheimer, who guided me through a jumble of questions into clear ideas.

I wish to thank my parents, Nancy and Paul Oppenheimer, who have always encouraged the pursuit of knowledge through schooling and travel. Without their support, none of this would have been possible.

By helping me in the field, as well as supporting me in life, Peter O'Brien provided limitless assistance. Thank you to my boys, Aden and Conor, who grudgingly accepted hours without mommy while I revised and revised.

To all those who helped with field and aviary work, Paul McDonald, Karen Fildes, Michael Jarmin, Geoff Graves, Ian, and probably a couple I'm forgetting, much thanks!! A special thank you to Karen Fildes, who not only helped in the field and aviary, but also provided much emotional support!

For providing not only a place to live while working in the field, but also a sense of family in the Australian bush, thank you Jo Hay, the Irwin family, Ann Walton, Bill Faulkner, and Andrew Dean.

Thank you to State Forests of NSW for allowing me to work in your forests, and especially to those in the Forbes office who took an interest in my work, Andrew Dean, Ann Walton, and "Speck."

To Andrew Cockburn and all those in his lab at Australia National University, thank you for the insightful discussions on cooperative breeding, as well as the much needed assistance with DNA fingerprinting.

Part 1: Literature Review

Chapter I. Aspects of Sociality and the Relationship Between Hormones and Behaviour

Sociality

1. Continuum of social organisation

Sociality, or social relationships among individuals in a group, exists in many forms throughout the animal kingdom. These diverse social relationships can be placed along a continuum, ranging from short-lived, loose associations between individuals to long-term, highly-organised societies. In an attempt to help explain this diversity, social systems have been classified into categories along this continuum. Among the most important aspects of social behaviour to consider are the degree of reproductive skew, the extent of helping behaviour, and the persistence of association. The degree of reproductive skew indicates the relative ratio of potential reproducers to non-reproducers in a population. For instance many species of social insects exhibit a high degree of reproductive skew, as many individuals in the population never breed (Sherman *et al.* 1995). Helping behaviour refers to the assistance an individual provides to offspring that are not their own. For instance, in some bird species, adults provision young that are not their own, while in other species, only the parents provision their young (Brown 1987). Persistence of association refers to the length of time a group of animals remain together. Some animals maintain a solitary existence, while others form stable, long-term associations (Lee 1994).

While any single one of these conditions may be used to categorise animals, Vehrencamp (1979) integrates reproductive skew, helping behaviour, and persistence of association into a useful model describing a continuous scale of social complexity. After solitary living, Vehrencamp's (1979) most basic category includes simple clusters of animals around a resource, such as food or nesting sites. For instance, vultures congregate around a carcass, and many sea birds nest in dense colonies. These are temporary associations, and few social behaviours are exhibited (beyond a reproductive pair). Next along Vehrencamp's (1979) continuum are groups that perform some type of communal activity, such as mobbing defense or communal roosting. This may entail some organisation and stability, but, importantly, all members benefit. Examples include crèches of young penguins huddling against the cold or squirrels screaming an alarm when a predator is detected. More complex still are societies that exhibit communal behaviours, but also unequally allocate reproductive opportunities or other tasks among members, either through a permanent or temporal division. For instance, cooperatively breeding birds cooperate in many aspects of daily life (e.g. feeding, roosting, nest building), but there are some members of these group that often do not breed. The most complex category along Vehrencamp's continuum includes animals living in complex, colonial societies with overlapping generations and a strict division of labour, including reproductive tasks. Many social insects fall into this category, in which there are distinct castes, only one of which ever breeds.

2. Benefits of sociality

i. Protection from predators and the environment

Simple proximity to conspecifics has an inherent anti-predation benefit: the chance of any one animal becoming prey is diluted by others in the group (Pulliam and Caraco 1984). As the number of potential prey increases, the likelihood that any one individual is preyed upon decreases. The probability that a bird is taken by a falcon from a group of five is one in five, but in a flock of 100, the probability is one in 100. This effect may be particularly useful to vulnerable members, which may escape notice by predators. All animals in a group are not equally likely to be recognised as prey items by predators; grouping may help potential victims, such as young or injured animals, escape detection (Bertram 1978, Hass and Valenzuela 2002).

There is likely to be improved detection of predators in group-living over independently-living animals. The “many-eyes” hypothesis explains this phenomenon: as group size increases, so do the number of eyes watching for predators (Pulliam 1973, Lima 1990). Studies have shown that some birds receive the benefit of improved predator detection by forming large groups. Kenward (1978) found that as the number of individuals increased in Wood Pigeon *Columba palumbus* flocks, attacks by a trained Goshawk *Accipiter gentilis* were increasingly unsuccessful. Also, detection of a stuffed predator took less time in large colonies of Bank Swallows *Riparia riparia* than in small colonies (Hoogland and Sherman 1976). However, Lima (1995) found little support for improved collective detection in his study on mixed flocks of Dark-eyed Juncos *Junco hyemalis* and American Tree Sparrows *Spizella arborea*. Unlike the aforementioned studies, Lima (1995) employed a ball rolling down a ramp as the “danger situation,” and this may have proved less threatening than a common predator.

Increased predator detection not only decreases the chance of becoming prey, but also allows for more time to devote to other activities. As flock size increases, individual birds spend proportionally less time scanning for predators and more time on other activities (Caraco *et al*. 1980). Heinsohn (1987) found that winter flocking in White-winged Choughs *Corcorax melanothamphos* allowed the birds to devote less time to predator vigilance and more to foraging; Powell (1974) found that group foraging facilitated predator detection and enabled Starlings *Sturnus vulgaris* to enhance their foraging capacity by increasing individual foraging time.

Coordinated group behaviours may give animals additional advantages. Mobbing and alarming are effective defenses against predators, especially if the predator is larger than the potential prey. Soldier castes in some social insects employ both of these defense mechanisms. Intruder detection results in the release of an alarm pheromone which stimulates soldiers to attack the intruder with a virtual armory, including shearing mandibles, piercing jaws, acidic sprays, and sticky glue (Alcock 2001). Whereas a single termite would pose little threat to a small mammal or large insect, an army of soldiers is sufficiently intimidating to force retreat.

In harsh conditions, such as extreme cold or aridity, communal roosting or huddling can protect animals from the environment by decreasing heat loss or desiccation. Woodlice avoid desiccation by huddling (Allee 1926), and Emperor Penguins *Aptenodytes forsteri* huddle in dense groups to help alleviate the effects of freezing temperatures (Bertram 1978). Kirkwood and Robertson (1999) found that thermoregulatory huddling in the Emperor Penguin can decrease total energetic costs by about 50% of the energy required to rest alone. Whereas a single animal may face death or extreme energy depletion in adverse conditions, a member of a group may be better able to deal with harsh conditions.

ii. Enhanced food acquisition

Animals living in groups may find food more efficiently and quickly and may capture prey more effectively than solitary animals. Groups may exchange information about food sources and can avoid feeding in areas which have been recently denuded. Experimental tests by Krebs *et al.* (1972) support this hypothesis by showing that groups of four Great Tits *Parus major* found more hidden food than individuals, and Pitcher *et al*. (1982) found that minnows and goldfish found food more quickly as group size increased. If resources are clumped rather than evenly dispersed, groups may more efficiently find and exploit resources (Beauchamp 2002). If one bird in a social group finds a good food source, others in the group are likely to benefit (Bugnyar and Kotrschal 2002). Also, it has been suggested that breeding colonies act as "information centers" where birds exchange information regarding food sources (Brown 1988, Barta and Szep 1995). However, this hypothesis has been criticized, and an alternate theory describes recruitment at the feeding site rather than at the colony (Richner and Heeb 1996). Both of these theories, however, support the idea that group interactions enhance communication regarding food sources.

Group foraging or hunting may help in capturing prey. For instance, Northern Rockhopper Penguins *Eudyptes chrysocomo museleyi* coordinate their underwater behaviour to catch prey, many large carnivores hunt cooperatively to kill larger prey than they could hunting individually, and Pelicans *Pelecanus erythrorhynchos* cooperate to drive fish toward shallow water where they are more easily captured (Elliot 1992, Krebs and Davies 1984, Tremblay and Chrel 1999, respectively). Groups can also more successfully defend carcasses from scavengers (Krebs and Davies 1984). Tight flocks of cooperatively breeding Hall's Babblers *Pomatostomus halli* engage in vigorous foraging bouts which may help flush prey items from cover (Balda and Brown 1977). Colonial Orb-web Spiders *Metabus gravidus* build webs spanning spaces which would be impossible for a single spider to spin, allowing them to utilise resource-rich areas that would otherwise be inaccessible (Vehrencamp 1979). Whereas an individual forager would have exclusive access to a prey item, it may not be able to kill or find as much food as a group. While group foraging requires that prey is shared by multiple mouths, it may also ensure that ample food is available.

Group members may benefit by learning where to find food and water, what items are suitable food, and how to acquire it by watching one another. Whether or not there is active teaching by the more experienced individuals, younger or less experienced group members may learn from the more experienced group members. For instance, juvenile Baboons *Papio ursinus* learn which foods are palatable by watching the reactions of others in the group (Cambefort 1981), and Red Squirrels *Tamiasciurus hudsonicus* decrease time and energy used to feed on a novel food (hickory nuts) if first allowed to watch experienced squirrels (Weigl and Hanson 1980). Also, African Elephants *Elephas maximus* learn from others where to find food and water sources during drought (Moss 1988), and Chimpanzees *Pan troglodytes* teach tool technologies to their young (Goodall 1970, Boesch 1991). Social transmissions such as these enable less experienced animals to utilise resources which would otherwise be much more difficult to obtain.

Larger groups can more successfully defend territories and food sources than can individuals or pairs. Large groups occupy large territories in a number of cooperatively breeding bird species (e.g. Arrowmarked babblers *Turdoides jardineii*: Monadjen *et al*. 1995, Florida Scrub-Jays *Aphelocoma coerulescens*: Woolfenden 1981, Galapagos Mockingbirds *Nesomimus parvulus*: Curry 1988). Except in homogeneous environments, large, exclusive territories will ensure a greater diversity of resources, which may be important in times of poor environmental conditions or in poor quality habitats (Gaston 1978).

iii. Facilitated reproductive success

Grouping increases the probability of intersexual contact (Lee 1994). This is beneficial for finding potential mates and may increase the opportunities for sexual selection. Rather than expending time and effort searching for a potential mate, group-living animals may mate with others in the group. Some ungulates and pinnipeds form harems, and males mate with all females in their group. In Elephant Seal *Mirounga angustirostris* colonies, a single harem master attempts to exclude all other males from mating with a number of females, as do Red Deer *Cervus elaphus* stags, which keep herds of females in compact groups during the breeding season (Halliday 1994).

In cooperatively breeding groups, members give and receive help rearing young. Group members may guard young and actively chase predators from the vicinity of nests or burrows. This protective behaviour has been reported in a number of cooperatively breeding birds and mammals; for instance, Acorn Woodpeckers *Melanerpes formicivorous* (Emlen and Vehrencamp 1985), Florida Scrub-Jays (Emlen and Vehrencamp 1985), White-throated Magpie-jays *Calocitta formosa* (Innes and Johnston 1996) colonial Mongooses *Helogale* spp. (Ewer 1973, Rasa 1977), and Silver-backed Jackals *Canis mesomelas* (Emlen 1984) are among those species that protect young within their groups. Also, multiple non-parental members may provision young with food, a situation which has been shown to increase nestling survival in the Alpine Accentor *Prunella collaris* (Nakamura 1998), Arabian Babblers *Turdoides squamiceps* (Wright 1998), and White-winged Choughs (Rowley 1978).

3. Evolution of Sociality and Alloparental Behaviour

i. Evolutionary mechanisms

Many avian species exhibit some form of social behaviour, ranging from temporary flocks to year-round, highly organised societies. Theories proposed to explain the evolution of sociality most often attribute the evolution of sociality to a variety of factors, including the benefits of group living, ecological constraints, individual selection, and/or kin selection (Hamilton 1964, West-Eberhard 1975, Vehrencamp 1979, Emlen 1984, Danchin and Wagner 1997, Dugatin 1997). Any discussion regarding the evolution of sociality relies on the assumption that behaviour has a genetic component that affects an animal's interactions with its environment and other animals (Alcock 2001). Not all behaviours are genetically "programmed," but the propensity to exhibit certain behaviours can be influenced by an animal's genotype. For example, many behavioural abnormalities have been linked to a single gene mutations in the fruit fly *Drosophila melanogaster* (Benzer 1973), male cricket *Teleogryllus gryllus* hybrids sing songs intermediate between the distinct songs of the parental species (Bentley and Hoy 1972), and some agonistic behaviour was found to be heritable in Silvereyes *Zosterops lateralis* (Kikkawa *et al*. 1986). While behaviours are rarely directed solely by genes, an animal's genotype predisposes it to respond to environmental cues in a certain manner. It follows that sociality has a genetic component that responds to evolutionary pressures.

Any heritable characteristic, including social behaviour, that increases an animal's inclusive fitness should result in the evolutionary selection of that characteristic. An animal's inclusive fitness takes both the individual's fitness and the fitness of its kin into account. As close relatives share many genes, selection should favour any behaviour that benefits kin. “Kin selection,” coined by Hamilton (1964), refers to this selection operating at the genetic level. Individual selection and kin selection are the most widely accepted mechanisms thought to guide the evolution of sociality (Hamilton 1964, West-Eberhard 1975, Vehrencamp 1979, Dugatin 1997).

Individual selection focuses on an individual’s fitness that can be maximised by increasing survival or reproductive success; therefore, social behaviours that increase an animal’s survival or fecundity will increase its individual fitness. As discussed earlier, group-living offers many advantages associated with enhanced survival and reproductive success. For instance, animals living in groups may increase their fitness by decreasing the risk of predation, increasing foraging efficiency, and improving protection for their young. Social behaviours associated with group living are perpetuated or become more common when they improve the fitness of those individuals exhibiting them (Vehrencamp 1979).

Environmental pressures, such as ecological constraints that restrict independent breeding, may also have an impact on individual selection and the evolution of sociality. The ecological constraint theory predicts that young will remain on their natal territories when there is a local shortage or absence of breeding openings in the population (Selander 1964). By remaining on their natal territory, individuals may enhance their survival and later reproductive success. Many studies have implicated shortages of suitable breeding openings as causal factors leading to the formation of social groups (e.g. Zahavi 1974, Craig 1979, Woolfenden and Fitzpatrick 1984, Koenig and Mumme 1987). Furthermore, manipulative studies of three species (Seychelles Warblers *acrocephalus sechellensis*: Komdeur 1992, Superb Fairy-wren *Malurus cyaneus*: Pruett-Jones and Lewis 1990, and Red-cockaded Woodpeckers *Picoides borealis*: Walters *et al.* 1992) have confirmed that when additional breeding opportunities are made available, dispersal and independent breeding are favoured over remaining with the natal group. Young members of cooperative groups, who forgo breeding while on their natal territories, may increase their chances of survival and may later inherit prime breeding habitat from their parents, thus increasing their fitness. This theory has received much support and has been used to help explain the formation of persistent family groups in many species (Brown 1974, Stacey 1979, Koenig and Pitelka 1981, Emlen 1982a, Woolfenden and Fitzpatrick 1984, Koford *et al.* 1986, Brown 1987, Koenig and Mumme 1987, Pruett-Jones and Lewis 1990, Komdeur 1992).

It is important to note that sociality may in fact be the ancestral condition in many species, and current ecological conditions may not help explain the incidence of social organisations and behaviours of extant species (Cockburn 1996, 2003, Heinsohn and Double 2004). Instead, historical conditions may better explain the evolution of sociality, while current conditions may help explain the secondary loss of sociality in some species (Nicholls *et al*. 2000). “Phylogenetic inertia” may play a role in maintaining social behaviours which evolved in the ancestors of members of present-day populations; in other words, in some taxa there may be a phylogenetic disposition to exhibit some form of sociality, despite the cessation of selective forces acting upon that behavioural trait (Edwards and Naeem 1993). Regardless, the aforementioned evolutionary mechanism is still valid to help explain the evolution of sociality, whether sociality evolved relatively recently or thousands of years ago.

A different hypothesis, the prolonged brood care hypothesis, stresses the benefit of offspring remaining on their natal territory. In this hypothesis suitable habitat may be available for dispersal, but those young that remain in close contact with their parents have higher survival than do young that disperse from their natal territory. Parental nepotism can enhance survival of offspring that remain on the natal territory beyond the typical fledgling stage (Ekman *et al*. 2000). As a result fitness of both the parents and the offspring improves. Parental nepotism has been documented in Mexican Jays *Amphelocoma ultramarina* and Siberian Jays *Perisoreus infaustus*, as parents allow offspring to gain access to food that was denied to non-kin. Similarly, in Bewick's Swans *Cygnus columbianus* and Belding's Ground Squirrels *Spermophilus beldingi*, parental nepotism has been noted, as parents protect their offspring from harm (Sherman 1977, Scott 1980, Barkan *et al*. 1986, Ekman *et al*. 2000). In such cases parents are likely to increase their fitness by improving survival rates of their offspring, while offspring improve their chances of survival by accepting aide from their parents.

ii. Alloparental behaviour

Regardless of whether ecological constraints or prolonged brood care is more influential in promoting natal philopatry, social groups often develop through the retention or inclusion of kin. Many social groups are composed of related individuals, and some of these groups exhibit seemingly altruistic behaviours (e.g. Florida Scrub-Jays: Emlen and Vehrencamp 1985, White-headed Vanga *Leptopterus viridus*: Nakamura *et al*. 2001, White-winged Choughs: Rowley 1978, Arabian Babblers *Turdoides squamiceps*: Wright 1998). In such groups, some individuals forgo breeding, while providing care for young that are not their own (alloparental behaviour).

The concept of kin selection is often evoked as a means of explaining behaviours such as alloparental feeding, grooming, and protection in cooperatively breeding animals (Emlen 1982, Brown and Pimm 1985, Russell and Rowley 1988, Mumme *et al*. 1989, Queller 1994). While non-breeding individuals within a cooperative group may experience lower individual fitness than their breeding cohorts, the net gain in inclusive fitness reaped from social interactions must be positive for the behaviours to persist. According to the kin selection model, helpers attempt to increase their inclusive fitness by providing alloparental assistance to close relatives, who share many of their own genes. Group-rearing of young has been shown to increase nestling survival in some species (Alpine Accentor: Nakamura 1998, Arabian Babblers: Wright 1998, Splendid Fairy-wrens *Malurus splendens*: Russell and Rowley 1988, White-fronted Bee-eaters *Merops bullockoides*: Emlen and Wrege 1989, and White-winged Chough: Rowley 1978). In such cases related helpers, whose breeding opportunities may be limited or absent, enhance their inclusive fitness by improving survival of young with whom they share many genes.

While kin selection provides a logical explanation for many instances of alloparental behaviour, this concept does not explain all cases. In a number of species, alloparental behaviours have also been observed in adults who are unrelated to the offspring they assist (reviewed in Stacey and Koenig 1990, Cockburn 1998). Furthermore, variations in helping behaviour cannot be explained by differences in relatedness in some species (Galapagos Hawks *Buteo galapagoensis*: Delay *et al.* 1996, Green Woodhoopoes *Phoeniculus purpureus*: Du Plessis 1993, Superb Fairy-wrens: Dunn *et al.* 1995, Venezuelan Stripe-back Wrens *Campylorhynchus nuchalis*: Piper 1994). In fact, in the White-browed Scrubwren *Sericornis frontalis*, help is preferentially directed toward unrelated offspring (Whittingham *et al.* 1997). Although animals do not improve their inclusive fitness in such instance, individuals may reap other benefits from alloparental behaviour directed toward unrelated kin. For instance, it has been suggested that helpers gain skills associated with parenting, which may be of use in future breeding attempts (Skutch 1961b, Lancaster 1971, Rowley 1977). Whereas naïve individuals may be relatively unsuccessful at rearing a brood of their own, experienced individuals (who have previously assisted with other broods) may be more successful (Brown 1987). Studies of Florida Scrub-Jays (Woolfenden and Fitzpatrick 1984) and Splendid Fairy Wrens (Rowley and Russell 1990) suggest that those breeders with past helping experience have higher breeding success than inexperienced individuals. Notably, however, several studies show no significant effect of prior helping experience (Acorn Woodpeckers: Koenig and Mumme 1987, White-fronted Bee-eaters: Emlen and Wrege 1989).

Reciprocal altruism has also been suggested as a mechanism promoting helping behaviour by unrelated individuals (Trivers 1971, Ligon and Ligon 1983, Wiley and Rabenold 1984). In the reciprocal altruism model, animals exchange services (Roberts 1998); in the case of cooperative breeding, helpers would provide assistance toward another's brood with the expectation that the parents or offspring would later assists with the helper's own brood. While this system has been demonstrated in Vampire Bats *Desmodus rotundus* (Wilkinson 1984), it also seems highly prone to cheating. Cheaters in a reciprocal altruism system would reap the greatest benefits; a recipient who fails to reciprocate would be better off than one who does reciprocate (Trivers 1971). As reciprocation may be difficult to enforce (Sigmund 1993), it seems unlikely that reciprocal altruism has contributed to the expression of alloparental behaviours. Competitive altruism (Roberts 1998) and the similar handicap principal (Zahavi 1975) offer another explanation for the seemingly altruistic behaviour exhibited by some unrelated animals. These theoretical explanations interpret altruistic behaviour as a signal of high quality (Zahavi 1995, Roberts 1998). It has often been suggested that helpers compete over allofeeding opportunities in an effort to attract mates or additional helpers (Brown 1978, Emlen 1978, Ligon and Ligon 1978, Brown and Brown 1980, Emlen 1984, Putland 2001). Individuals who exhibit a high degree of "altruistic" behaviour may be more successful at attaining mates or collaborators than are "more selfish" individuals (Boland *et al*. 1997a, Putland 2001, Hawkes and Bird 2002). Clarke's (1989) findings in the cooperatively breeding Bell Miner *Manorina melanophrys* support this theory: widowed females preferentially paired with the unmated male helpers who had assisted the most with her previous nesting attempt. Members of some species even seem to deter group members from demonstrating alloparental behaviours; for instance, Arabian Babblers (Carlisle and Zahavi 1986), Pied Kingfishers *Ceryle rudis* (Reyer 1990), and some mammals (MacDonald and Moehlman 1982) have been observed interfering with the helping behaviour of group members. Zahavi (1995) contends that interference can be explained by competitive altruism but not by any model of indirect selection. He argues that kin selection and reciprocal altruism would favour helping by collaborators, as individuals' gains would be greatest when others invest in helping behaviour. Rather than being altruistic, alloparental behaviour in non-kin seems to be a selfish act intended to increase an individual's fitness.

None of the preceding hypotheses (i.e. prolonged brood care, kin selection, reciprocal altruism, competitive altruism) are mutually exclusive. In fact, it is likely that all or a number of these evolutionary mechanisms have culminated in the observed incidences of alloparental behaviour. One theory, however, questions whether helping behaviour is even subject to evolutionary pressures. This alternate hypothesis suggests that helping may not be adaptive; instead, helping behaviour may be an unselected consequence of normal parental care (Jamieson 1989). According to this theory, selection for a high degree of parental behaviour insures that helping behaviour is perpetuated. Further, any discriminatory behaviour which could result in diminished parental care would be maladaptive; more specifically, the ability to discriminate between the stimuli of one's own begging young versus another's begging young might be unlikely to evolve (Rohwer 1986). The provisioning of nestlings of avian brood parasites, such as cuckoos, despite the detrimental fitness consequences to the parents, supports this assertion. Consequently, in cooperatively or communally breeding species, helping behaviour may simply be the result of strong selection for individuals that respond to begging calls and postures, regardless of the individual's relationship to the offspring (Jamieson 1989). Such "misplaced attention" would be most likely to occur in birds of close proximity to the breeding pair.

iii. Parental behaviour

Regardless of the mechanism, parental (or alloparental) behaviour seems to be important in the formation and/or maintenance of social groups, and parental care is thought to be an important pre-condition for eusociality (Queller 1994). The term "parental care" may be used to describe a wide range of parental-type behaviours; for instance, brooding, preening, feeding, protecting, leading, and sheltering are all parental behaviours when directed toward young individuals (Clutton-Brock 1991). Species employing some breeding systems exhibit a higher degree of parental care than those with other breeding systems. There is a continuum in avian parental care ranging from an absence of incubation and post-hatching parental care (e.g. Megapodes: Jones *et al*. 1995) to extensive care lasting through juvenile stages (e.g. White-winged Choughs: Heinsohn 1991).

Parental care may set the stage for helping behaviour (Queller 1994), and species with altricial young may be more predisposed towards a higher degree of sociality than are species with precocial young. As altricial young require more post-hatching care than do precocial young, multiple individuals may be better able to provide this care than can a solitary parent. Biparental care requires coordination of appropriate parental behaviours and may be considered a rudimentary form of sociality. As more individuals assist in rearing young, a greater degree of coordination is necessary, and a more complex social system may evolve. The evolution of cooperative breeding social structures, in which there is often high reproductive skew and a division of labour, may have been facilitated by helpers' potential to extend the period of offspring care (Langden 2000).

If parental behaviour has indeed played an important role in the evolution of sociality, it follows that some of the same physiological mechanisms affecting parental behaviour may also have influenced social behaviours. Many parental and other social behaviours are associated with or are influenced by the endocrine system. To understand the physiological bases for parental behaviour, the relationship between endocrine changes and reproductive phases should be examined.

Hormones and Reproductive Behaviour

During the breeding period, hormones promote physiological and behavioural changes necessary for successful breeding. Appropriate hormonal fluctuations are regulated via the hypothalamic-pituitary-gonadal (HPG) axis. External factors, such as photoperiodic cues (Wingfield and Farner 1980) and/or social interactions (Kroodsma 1976, Wingfield and Marler 1988) typically trigger the hypothalamus to secrete gonadotrophin releasing hormone (GnRH), which then stimulates the anterior pituitary to release the godadotropins, luteinizing hormone (LH) and follicle stimulating hormone (FSH). Together, LH and FSH orchestrate gonadal maturation or recrudescence and stimulate the gonads to release gonadal steroids: progesterone (P) in females, estradiol (E2) in females and in some males, testosterone (T) in males and females, and dihydrotestosterone (DHT) in males (Wingfield *et al.* 1987, Schlinger 1998). These gonadal steroids serve multiple physiological roles, including stimulating spermatogenesis and ovulation. In conjunction with the gonadal steroids, prolactin (Prl), a peptide hormone released from the anterior pituitary, also plays an important role in avian reproductive physiology (reviewed in de Vlaming 1979, Buntin 1996). Specifically, Prl is associated with the development of the brood patch in many species and the secretion of crop milk in Columbiformes (Eisner 1960, Silver 1984, Vleck *et al.* 1991). Concurrent with the physiological changes necessary for breeding, gonadal steroids and Prl also help direct appropriate behavioural changes.

In many passerines male-male aggression is common at the initiation of breeding attempts. Not only does T help promote spermatogenesis at this time, but it may also promote this aggressive behaviour (Harding 1981, Balthazart 1983). In support of this is the observation that T levels seem to be more closely related to some form of “challenge,” rather then to a specific nesting stage (Wingfield *et al.* 1990b). For instance, in males of many monogamous, multi-brooded species, agonistic interactions and elevations in T levels accompany only the initial courtship and nest building stages. After the initiation of the first brood in such species, territories are established and relationships between neighbors are relatively stable; as a result, second or later nesting attempts do not elicit aggressive behaviours, and typically, no concomitant increase in T is observed (e.g. Song Sparrow *Melospiza melodia*: Wingfield 1984). However, aggressive interactions may persist for long periods in polygamous species, where males defend receptive females and large territories for extended periods. In these males T levels remain elevated for extended periods (e.g. Pied Flycatcher *Ficedula hypoleuca*: Silverin and Wingfield 1982). These and other studies demonstrate that T helps direct agonism in birds (Ramenofsky 1984, Wingfield *et al.* 1990, Wikelski *et al.* 1999, Hau *et al.* 2004).

In many species a dramatic change in behaviour takes place after breeding pairs are established, and a suite of hormonal changes facilitates the induction and maintenance of these behaviours. Nest building is often a prerequisite of egg-laying and may also be used as a form of sexual or pair bonding display (Lehrman 1961, Ehrlich *et al.* 1988). There is a great variety of nest types, from simple scrapes to complex mud and/or stick constructs, and some birds invest much time and effort in nest building. The coincidence of nest-building behaviour with gonadal changes preceding egg laying suggests that nest building behaviour is induced by some of the same hormones as those released by or affecting the gonad (Lehrman 1961). Some studies have found a correlation between nest building and elevated plasma titres of P or Prl (Dawson and Goldsmith 1982, Vleck *et al.* 1991), and others have successfully induced nest building behaviour with the administration of E2 and P, E2 and Prl, or T alone (Crook and Butterfield 1958, Cheng and Silver 1975, Hutchinson 1975, Logan and Carlin 1991). While there does not seem to be an universal correlation between nest building and a single hormone (or specific combinations), nest-building behaviours are associated with the presence of reproductive hormones.

Following nest building and laying, most birds incubate their eggs. In many species only one parent incubates, while in others both parents or a number of members of the cooperative group incubate. Prolactin is the hormone most often associated with the onset and maintenance of incubation behaviour in birds (reviewed in Goldsmith 1983, Buntin 1996). In many species Prl levels rise at the onset of incubation and remain elevated throughout the incubation period. In species where the female provides the majority of parental care, her Prl levels typically exceed those of males, and the opposite is true when the male is the primary care provider (Goldsmith 1983, Oring *et al.* 1988, Gratto-Trevor *et al.* 1990). Furthermore, in the cooperatively breeding Red-cockaded Woodpecker, Prl levels are elevated in helpers who assist with incubation (Khan *et al*. 2001).

A decrease in T levels in males involved with incubation may also be important in promoting incubation behaviour. T levels are six times higher in non-incubating than incubating Wilson's phalaropes *Phalaropus tricolor* (Fivizzani *et al*. 1986), and plasma T levels decrease abruptly at the onset of incubation in the Spotted Sandpiper *Actitis macularia* and Rufous Whistler *Pachycephala rufiventris* (Fivizzani and Oring 1986, McDonald *et al*. 2001b, respectively). Furthermore, administration of exogenous T to male Rufous Whistlers greatly reduced their contribution to incubation (McDonald *et al*. 2001a). It seems that high T levels, which are typically associated with agonistic or mate acquisition behaviours, may be incompatible with incubation behaviour (Silverin 1980, Hegner and Wingfield 1987, Oring *et al*. 1988). Decreasing plasma T levels may be the result of increasing Prl levels at the onset of incubation. Many studies have indicated that Prl has an anti-gonadal action which inhibits the production and subsequent secretion of T (El Halawani *et al*. 1991, Seiler *et al*. 1992, Buntin 1996).

With the exception of brood parasites and megapodes, all birds exhibit some form of parental behaviour toward their newly-hatched young (Ehrlich *et al.* 1988). However, there is much variation in the pattern of parental care; for instance, species with precocial young simply shelter and lead their young, while those species with altricial young must feed and brood their nestlings. Regardless of the extent of parental care, there is a formidable change in parental behaviour upon hatching, and there appears to be a concomitant endocrine change.

While multiple hormones probably play a role in these changes, Prl and P are the hormones most often associated with parental behaviours in birds (Riddle 1963, Goldsmith 1983, Silver and Cooper 1983, Brown 1985, Buntin 1986 and reviewed in

Balthazart 1983 and Buntin 1996). The best evidence linking parental behaviour with specific hormones comes from changes in Prl and the secretion of crop milk. In columbiform birds, Prl stimulates growth of crop epithelial cells that produce crop milk, and engorgement of the crop sac stimulates feeding of the young (Riddle 1963, Goldsmith *et al.* 1981). Additional evidence also suggests a relationship between parental behaviour and elevated Prl in non-columbiform species. For example, in species with altricial young, Prl levels typically remain high until the young are able to thermoregulate (Goldsmith 1983, Rosenblatt 1992), and in galliforms Prl injection into either hens or roosters induces a full repertoire of parental behaviours, such as sheltering chicks, leading them to food and away from danger, and calling to the young (Nalbandov *et al.* 1945, Lehrman 1961, Buntin 1986). Furthermore, in some cooperatively breeding species, Prl has been associated with parental-type behaviours exhibited by non-breeding helpers (Florida Scrub-Jays: Schoech *et al.* 1996b, Harris' Hawks *Parabuteo unicinctus*: Vleck *et al.* 1991, Red-Cockaded Woodpeckers: Khan *et al.* 2001).

As with incubation behaviour, high T levels may be incompatible with care of young (Wingfield and Moore 1987, Ketterson *et al.* 1992, 1996, Van Duyse *et al.* 2002). In the males of a number of bird species, periods of elevated T and parental behaviour are temporally separated (Beletsky and Orians 1987, Wingfield and Moore 1987). Further, experimental studies have shown that T treatment acts to decrease parental provisioning rates (Silverin 1980, Hegner and Wingfield 1987a, De Ridder *et al.* 2000). Even in the cooperatively breeding Superb Fairy Wren, in which provisioning of nestlings is often concurrent with male courtship, T treatment was shown to depress this parental behaviour (Peters *et al.* 2002).

In many avian species behaviours such as “challenge-type” aggression, nest building, and parental care are confined to the breeding season. However, in some species these behaviours are exhibited throughout the year. For instance, in flocking or group-living species, challenge situations may arise at any time; some birds build roost nests year-round; prolonged parental or alloparental behaviour is common in some group-living or cooperatively-breeding species. When equivalent behaviours are exhibited in the context of breeding as well as during non-breeding periods, are the accompanying hormonal changes also comparable?

1. Hormones and Non-reproductive Social Behaviour

Non-reproductive social behaviours are thought to have evolved from reproductive and parental behaviours (Crews 1997). Thus, it might be expected that some of the same hormones associated with reproductive behaviours may also be involved with affiliative behaviours. By examining behavioural endocrinology outside the context of reproduction, specific hormone-behaviour relationships can be scrutinized without the possibly confounding factors associated with the physiological changes necessary for reproduction.

i. Testosterone and aggression

Outside the context of breeding, aggressive behaviours are common in animals establishing and maintaining dominance hierarchies. Some studies have demonstrated a relationship between plasma T levels and aggression associated with social dominance. For instance, a positive relationship between plasma T titers, aggression, and social status has been found in group-living male Rhesus Monkeys *Macaca mullata* and Olive Baboons *Papio anubus* (Rose *et al*. 1971, Sapolsky 1982, respectively). In the cooperatively breeding Florida Scrub-Jay *Aphelocoma coerulescens* and White-browed Sparrow Weaver *Plocepasser mahali*, dominant males have been found to have higher T levels than any other member of the group (Schoech *et al.* 1991, Wingfield *et al.* 1991, respectively). Further, exogenous T has been found to be successful at elevating the dominance status of male Gambel's White-crowned Sparrows *Zonotrichia leucophyrs gambelii* and Red-winged Blackbirds *Agelaius phoeniceus* (Baptista *et al*. 1987, Searcy and Wingfield 1980, respectively).

Nevertheless, the relationship between plasma T and social dominance may not be as clear-cut as it seems; an equivalent number of studies have found no relationship between plasma T and social status. In Dark-eyed Juncos *Junco hyemalis*, wintering Harris' Sparrows *Zonotrichia quereula*, and Rhesus Monkeys, there was no correlation between social rank and plasma T levels (Holberton *et al.* 1989, Monaghan and Glickman 1992, Rohwer and Wingfield 1981, respectively). Many factors may contribute to these inconsistencies. For instance, some studies have demonstrated that T is not a useful predictor of social status, except in newly formed groups or in situations where social position is challenged (Ramenofsky 1984, Schwabl *et al.* 1988, Wingfield and Lewis 1993). There may be species-specific differences or seasonal variability in the relationship between T and the aggressive assertion of social status. Perhaps the relevance of social dominance affects the reliance on T; for instance, T levels may correlate with aggression when social status reflects future breeding opportunities, but not when it only reflects access to resources, such as food or shelter.

Aggression during the non-breeding period has been observed in birds contesting or defending territories. While T seems to help regulate territorial aggression during the breeding season in many temperate zone passerines (see Balthazart 1983, Wingfield and Ramenofsky 1985, Wingfield 1994), elevated plasma levels of T are not typically found in species that exhibit year-round or winter territorial aggression (see Wingfield and Soma 2002, Hau *et al.* 2004, but see Wikelski *et al.* 1999). It seems that the same behaviour, territorial aggression, may be dependent on high plasma T titres in some birds when in breeding condition, but otherwise is independent of elevated plasma T levels (Wingfield *et al.* 2001, Wingfield and Soma 2002). For example, in male European Robins *Erithacus rubecula* and non-migratory song sparrows *Zonotrichia melodia morphna*, territory defense during the breeding season was associated with elevated T levels, while non-breeding territory defense was not (Schwabl and Kriner 1991, Schwabl 1992, Wingfield and Monk 1992). Furthermore, circulating T levels were low all year in the tropical Bay Wren *Thryothorus nigricapillus* and White-browed Sparrow Weaver, despite year-round territory defense (Levin and Wingfield 1992). As prolonged elevations of plasma T titres may incur physiological costs (Dufty 1989, Ketterson *et al.* 1991, Nelson and Demas 1996, Hillgarth and Wingfield 1997, Peters 2000, Wingfield *et al.* 2000), it may be maladaptive to maintain high plasma T levels for extended periods outside the breeding season.

Recent evidence, however, suggests that sex steroids may indeed support non-breeding aggressive behavior, but may not always be apparent in plasma measurements. Sex steroids may be produced *de novo* in the brain and have paracrine effects on CNS processing, or inactive hormone precursors (such as dehydroepiandrosterone (DHEA)) may be converted into active sex steroid hormones by target tissues (Soma *et al.* 2000, Hau *et al.* 2004). If this is the case, secretions of gonadal steroids may vary at exceptionally low levels, undetectable with most commonly used assays for plasma T. Alternately, other factors may adjust the sensitivity of target tissues to steroids, such as alterations in steroid receptor density (Soma and Wingfield 1999, Wingfield and Soma 2002).

ii. Prolactin and alloparental behaviour

Another well-founded hormone-behaviour relationship is that of Prl and parental behaviour (see above). Because the expression of alloparental behaviour is very similar (if not virtually identical) to parental behaviour, the relationship between Prl levels and alloparental behaviour has also been investigated. A positive relationship between alloparental behaviour and Prl titres has been found in both mammalian and avian species (Common Marmosets *Callithrix jacchus*: Mota and Sousa 2000; Florida Scrub-Jays: Schoech *et al.* 1996b; Harris' Hawk: Vleck *et al.* 1991; Mexican Jays: Brown and Vleck 1998; Red-cockaded Woodpeckers: Khan *et al.* 2001; Wolves *Canis lupus*: Asa 1997). While the mechanism of action remains unclear, it is thought that Prl facilitates alloparental behaviour in the same manner that it affects parental behaviour. Prolactin is thought to help mediate the expression of behaviours typically expressed by parents caring for young.

In some species, alloparental behaviour extends well into (and sometimes beyond) the juvenile life stage (Arabian Babblers: Zahavi 1974, Gray Jays *Perisoreus canadensis*: Waite and Strickland 1997, Siberian Jays: Ekman *et al.* 2000, White-winged Choughs: Heinsohn 1991). In such cases, alloparental behaviour may not solely reflect the need to provision young, but may also play an important role in affiliative behaviour (Mitani and Watts 2001, de Kort *et al.* 2003,). Several researchers have suggested that alloparental behaviour may reflect social position within groups (Craig 1980, Kemp and Kemp 1980, Verbeek and Butler 1981, Ligon and Ligon 1983, Craig 1988). Alloparental behaviour may have been co-opted as an expression of social position within a hierarchy; stereotypical "parental" behaviours may be used to advertise social dominance (Zahavi 1995). Also, alloparental behaviour may be used as a means of demonstrating parental ability and may be a factor in future mate selection (Boland *et al.* 1997a, Putland 2001). The role of Prl in such contexts has not been examined. Prolactin may mediate the expression of alloparental behaviour regardless of the social context; alternately, Prl may not play a role in alloparental behaviour when it is uncoupled from “caring” behaviour.

iii. Corticosteroids and social hierarchies

Affiliative behaviour carries with it certain social stresses. Even within the most egalitarian societies, competition for food, mates, breeding sites, and social position may arise. Social factors have been shown to affect animals’ stress response, specifically the hypothalamic-pituitary-adrenal (HPA) axis. Animals respond to stressors (both physical and psychological) through a series of reactions that involve the activation of the adrenal cortex through the HPA axis and results in the secretion of glucocorticoids (Harvey *et al.* 1984, Sapolsky 1993). Short-term elevations in glucocorticoid levels typically enable animals to survive challenges to homeostasis, but long-term elevations are often detrimental (Sapolsky 2002, Wingfield and Kitaysky 2002). In the short-term, elevated glucocorticoids can have behavioural and physiological effects that help animals respond to stressful situations (Wingfield *et al.* 1998, Sapolsky *et al.* 2000). Because of this causal relationship between stress and glucocorticoid release, glucocorticoid secretion has been widely used as a reliable measure of stress (Levine 1993).

Much attention has been paid to the relative levels of stress, as estimated by glucocorticoid levels, associated with social position within a hierarchy. However, studies investigating this relationship have not reached a consensus. Some studies have shown that dominant animals have lower glucocorticoid levels than their subordinates (e.g. fish: Ejike and Schreck 1980, Sloman *et al*. 2001; mice: Louch and Higgenbotham 1967; Olive Baboons: Sapolsky 1990; rats: Sakai *et al.* 1991; various bird species: Wingfield and Moore 1987; wolves: Fox and Andrews 1973). Greater psychosocial stress in subordinate animals than in dominants has been attributed to a number of factors; for instance, the effect of defeat (Louch and Higginbotham 1967), the risk of increased predation as subordinates are forced to forage over greater distances from the group than are dominants (Schwabl *et al*. 1988), decreased access to resources (Louch and Higginbotham 1967), and intimidation by dominants (Rohwer and Wingfield 1981) may all impose psychosocial stress and ultimately the elevation of subordinates' glucocorticoid levels. Nevertheless, others studies have found that dominant animals have higher glucocorticoid levels than subordinate animals (e.g. African Elephants: Foley *et al.* 2001; African Wild Dogs *Lycaon pictus*: Creel *et al.* 1996; Dwarf Mongooses *Helogale parvula*: Creel *et al.* 1992; female Common Marmosets: Saltzman *et al.* 1994; Meerkats *Suricata suricatta*: Carlson *et al.* 2004; Ring-tailed Lemurs *Lemur catta*: Cavigelli 1999; Squirrel Monkeys *Saimiri sciureus*: Coe *et al.* 1979). In this case, it has been suggested that social dominance is in fact more stressful than subordinance because dominants engage in more aggressive interactions than do subordinates (Creel *et al*. 1996).

These conflicting results may be due to a number of factors, including whether animals are in breeding or non-breeding condition (Wilson *et al.* 1978), variable criteria for assessing dominance (McGuire *et al.* 1986), whether animals are captive or free-living (Creel *et al.* 1996, Creel 2001), the stability of social relationships (Coe *et al.* 1983), and the organisation of the social structure (Creel 2001, Abbott *et al*. 2003). The presence of familiar social partners and stable social relationships may be one of the most important factors in determining the endocrine response to stress (Levine 1993, Sapolsky 2002). Recently formed groups or those undergoing reorganisation may not be comparable to long-standing social hierarchies. It has been suggested that the presence of stable social relationships, especially the presence of kinship bonds, may in fact ameliorate the stress response (Levine 1993, Abbott *et al*. 2003). High stability of social position is often correlated with low basal cortisol (F) levels, as it does in wild Baboons (Sapolsky 1992, 1993). Alternately, during unstable periods, psychological stress may be invoked through high rates of aggression, shifting alliances, and disruption of feeding and affiliative social behaviours (Levine *et al.* 1989). Instability associated with initial or disruptive grouping often stimulates glucocorticoid increase, as shown in Mice (Louch and Higginbotham 1967, Bronson 1973), Squirrel Monkeys (Mendoza *et al.* 1979), Rhesus Monkeys (Mendoza *et al.* 1979), and Chickens (Siegel and Siegel 1961).

Aims

In this thesis I examine several alternate, but not necessarily mutually exclusive, hypotheses regarding the evolution of alloparental behaviour in a bird species, the White-browed Babbler (WBBA) *Pomatostomus superciliosus*, using endocrine, genetic, and behavioural measures.

1. Study Species

White-browed Babblers are gregarious, sedentary passerines with a range covering much of Australia (Simpson and Day 1996) (Fig. 1-1). They are boisterous, bold, curious, and common, making them an ideal study species. They are often seen foraging on the ground in groups, preening en masse, following one another on flights, and roosting together at night, as well as during the heat of the day. Communal activities are performed year-round by all WBBA group members. Solitary birds are rarely observed; WBBAs are almost always seen in groups of more than three birds (pers. obs.).

Cooperative breeding is a phylogenetically conserved trait among members of the genus *Pomatostomus* (Edwards and Naeem 1993). Further, cooperative breeding has been documented in many members of two genetically unrelated (though behaviouraly similar) genera of babblers worldwide: *Turdoides* and *Pomatostomus* (Arabian Babbler: Zahavi 1990, Common Babbler *Turdoides caudatus*: Gaston 1978, Grey-crowned Babbler *Pomatostomus temporalis*: Brown *et al*. 1978, Jungle Babbler *Turdoides striatus*: Gaston 1977, Rufous Babbler *Pomatostomus isidori*: Bell 1982, White-browed Babbler: Cale 1999). In cooperative-breeding social systems, some individuals assist in rearing offspring that are not their own; this alloparental assistance may come at any time from the start of nest building through to the time when young reach independence (Rowley 1976). White-browed Babblers exhibit stereotypical alloparental behaviours throughout the year.

Figure 1-1. Four WBBAs photographed in the aviary at University of Wollongong.

2. Hypotheses explaining alloparental behaviour

Alloparental behaviour has intrigued behavioural ecologists because its seemingly selfless motivation is not consistent with evolutionary pressures. Attempts have been made to explain these seemingly altruistic behaviours, and a number of hypotheses have been proposed: the kin selection hypothesis, the unselected consequence of communal breeding hypothesis, and the competitive altruism/ handicap principle. All three endeavour to provide an evolutionarily sound rationale for these behaviours.

i. Kin selection hypothesis (Hamilton 1964):

The kin selection hypothesis suggests that alloparental behaviour is expressed toward relatives in an effort to maximise one's own inclusive fitness. "Altruistic" behaviour, such as alloparental behaviour, should be expressed preferentially toward close relatives, who share many of the same genes. As alloparental behaviour should improve the fitness of those who receive it, it should increase the proportion of the alloparent's genes in the population.

Is alloparental behaviour expressed preferentially toward kin in the WBBA? To answer this, I examined alloparental behaviour and relatedness in captive groups of WBBAs. Alloparental behaviours were identified, characterised, and quantified, and genetic analysis of relatedness was performed. If kin selection is a likely explanation for the evolution of cooperative breeding in WBBA, I would expect a high degree of relatedness between those individuals giving and receiving alloparental care.

ii. Unselected consequence of communal breeding hypothesis (Jamieson 1989, 1991):

Alloparental behaviour may not be adaptive, but instead may be an unselected consequence of communal breeding (Jamieson 1989). In communally breeding species, adults may come into contact with young that are not their offspring, and alloparental behaviour may be expressed in response to stimulation by these young. Evolutionary selection for provisioning begging young may be strong enough to induce “parental” behaviours by non-parental individuals (Jamieson 1989, 1991). In this case, the driving force behind alloparental behaviour is the stimuli of young birds.

In order to refute this hypothesis, it would be necessary to prove that alloparental behaviour has in some way been fine-tuned; this suggests that alloparental behaviour has indeed been selected for, rather than simply being a byproduct of selection for provisioning young (Jamieson and Marshall 1999). If the endocrine system is involved in the induction of alloparental behaviours, this would suggest that selection has favoured birds whose physiology promotes alloparental behaviour and, thus, alloparental behaviour is in fact adaptive (Vleck *et al.* 1991, Khan *et al.* 2001). As hormonal changes are known to facilitate the expression of parental behaviour, similar hormonal change might also be expected in alloparents. For example, depressed T levels and elevated Prl levels may facilitate the expression of alloparental behaviour.

Are “reproductive” hormones associated with the expression of alloparental behaviours in the WBBA? To examine this question, I first determined typical levels of reproductive hormones in breeding birds and identified hormones associated with certain parental behaviours. A field study provided this behavioural and endocrine data. Hormonal and behavioural studies of captive birds were then performed to examine whether hormones may facilitate the expression of alloparental behaviour.

iii. Competitive altruism/ Handicap principle (Zahavi 1975, Roberts 1998)

Altruistic behaviour may be a signal of an individual’s quality and may be used in competitive situations, such as mate and territory acquisition or the quest for improved social status. Although “altruistic” behaviour may seem distinct from agonistic signals, such as aggressive contests, both may be a form of competition and may be regulated by the same physiological mechanisms. Likewise, “altruistic” behaviour may impart psychosocial stress on the receiver if receipt implies subordinate status. Just as defeat can affect an animal’s stress response, so might receipt of “altruistic” actions in certain circumstances.

Behavioural measures were examined in captive groups to help discern whether “altruistic” behaviours could signal social standing within a hierarchy. As “altruistic” behaviours may be a subtle form of competition, hormones typically associated with aggressive contests were examined. Also, as psychosocial stress is known to affect the endocrine system, the stress responses of group members were evaluated. If imparting alloparental care is in effect an assertion of dominance, I would expect the receivers (subordinates) to be subject to psychosocial stress and exhibit higher basal glucocorticoid levels than the alloparents (dominants) and possibly show greater sensitivity to a stressor. Because a number of studies indicate that animals in stable associations differ hormonally from those in unstable groupings, I will also examine these hormonal relations during periods of experimentally induced social instability.

PART 2: Field Studies of Free Living White-Browed Babblers

Chapter II. Natural History and Morphometrics of White-browed Babblers in Back Yamma State Forest

INTRODUCTION

While avian cooperative breeding is rare worldwide, a relatively high proportion of Australian old endemic passerines are cooperative breeders (Russell 1989, Cockburn 2003, Heinsohn and Double 2004). In fact, cooperative breeding may be the ancestral state for many of Australia's avifauna (Cockburn 1996). In predominately cooperatively breeding taxa, group-living and extra-parental assistance with brood rearing are the norm. However, group-living does not necessitate helping, and there are variable patterns of helping across and within species (Gardner *et al.* 2004).

"Cooperative breeding" does not define a single set of behaviours, but instead a range of behaviours that involves care of young group members by non-parental individuals. A number of Australian passerines have been reported to exhibit singular cooperative breeding, wherein a single breeding pair and multiple non-breeding group members help rear young; for instance, in White-winged choughs (Rowley 1976) and Splendid Fairy-wrens (Rowley and Russell 1990), a single breeding pair receives help from non-breeding helpers during all nesting stages.

Other species are plural cooperative breeders, in which there are multiple breeding pairs within a social group, as well as multiple helpers (Brown 1978). While less common than singular cooperative breeding, plural breeding has been noted in a number of species; for instance, plural breeding has been observed in Acorn Woodpeckers (Koenig 1981), Bell Miners (Clarke 1984), Bushtits *Psaltriparus minimus* (Sloane 1996), Guira Cuckoo *Guira guira* (Macedo and Bianchi 1997), Mexican Jays (Brown and Brown 1990), and Pukekos *Porphyrio porphyrio* (Craig and Jamieson 1990).

Some species exhibit only a single type of cooperative mating system; for instance, White-winged Choughs are obligate cooperative breeders, in which helpers are necessary for breeding success (Boland *et al.* 1997a). However, other species may exhibit variable mating systems; for instance, White-browed Scrubwrens are facultative cooperative breeders, in which breeding success is possible with or without helpers (Magrath and Yezerinac 1997). Furthermore, some species, such as the Australian Magpie *Gymnorhina tibicen* and Bushtits employ different mating systems in different locales, suggesting that environmental or demographic factors may play an important role in determining the adoption of cooperative breeding strategies (Hughes *et al.* 1996, Sloane 1996).

Singular cooperative breeding has been recorded in each of the five babbler species within the genus *Pomatostomus* (Brown 1978, Bell 1982, Cale 1999). However, Chandler (1920) observed more than one simultaneously breeding pair per WBBA group, suggesting that this species might adopt plural cooperative breeding under some conditions. Since life history traits, such as breeding strategy, can have a profound interrelationship with endocrine factors (Jacobs and Wingfield 2000), it was important to verify life history traits of my study population before proceeding with my study of hormone-behaviour relations in the WBBA.

MATERIAL AND METHODS

Study Site

This study was conducted from June 1996 to July 1999 in Back Yamma State Forest (BYSF), 16 km ENE of Forbes, NSW, 148°E, 33°S. In addition, five WBBA groups caught in Warredary State Forest (NW of Grenfell, NSW, approximately 50 km from BYSF) were included in genetic analyses. Back Yamma State Forest is a 4330 ha forest that ranges in elevation from 260 m to 350 m, and it is surrounded by agricultural land. As there are few trees along bordering roads or in neighboring properties, there is little chance for WBBA dispersal out of BYSF. The forest has been heavily ring-barked and logged since 1880, creating an open woodland interspersed with cleared paddocks. The majority of my study was conducted in the western half of the forest, dominated by White Box *Eucalyptus albens*, Grey Box *E. microcarpa*, Yellow Box *E. melliodora*, and White Cypress Pine *Callitris glaucaphylla*. There was little under story in the forested areas, and open paddocks were covered with a mixture of native and exotic grasses.

Capture, Marking, and Measurements

Birds were caught using 8 X 12 m Japanese mist nets within the boundaries of their groups' territories. In order to catch nesting birds, nets were often placed close to a nest. Sometimes, I used playbacks of taped recordings of conspecific calls and a mounted decoy to entice birds into an area. Occasionally, I returned a bird to the net after banding it, in an effort to lure more group members. These live decoys were not left in the net for more than 10 minutes.

When captured, WBBAs were banded with an uniquely numbered, metal band, issued by the Australian Bird and Bat Banding Schemes (ABBBS). Also, each bird was banded with a distinct combination of three colour bands (permitted by ABBBS), allowing visual identification of individuals from a distance.

On capture, I measured tarsus, head, and culmen length to the nearest 0.01 mm with Vernier calipers, and noted fat and moult status of each bird. Flattened wing cord was measured to the nearest 0.5 mm from the bend of the wing to the tip of the longest primary using a wing rule. Body mass was measured to the nearest 0.5 g with a 50 g Pesola spring scale.

Moult was assessed on each bird by blowing on the body feathers and inspecting the remiges and rectrices. I recorded the location and number of feathers in pin and scored body molt as light, moderate, or heavy. I then compiled these data into presence or absence of moult (feathers in pin) in three body zones: wing, body, and rectrices. Wing moult concerned only primaries or secondaries, and body moult was restricted to head, neck, back, flank, and abdominal regions. Body moult was only recorded if more than five pin feathers were found on all regions examined .

Fat levels were scored by examining subcutaneous deposits in the furculum and on the abdomen. The following scale (based on Ralph *et al.* 1993) was employed:

0.0=no fat visible on furculum or abdomen

0.5=trace levels of fat on furculum and/or abdomen

1.0=thin layer of fat covering furculum and/or abdomen

1.5=thicker layer of fat covering furculum and/or abdomen

2.0=furculum half filled in some patches and/or patches of abdominal fat

2.5=furculum somewhat concave and/or large patches of abdominal fat

3.0=furculum filled to level of clavicles and fat covering abdominal region

White-browed Babblers were captured and banded with permission from State Forests of NSW (Special Purposes Permit No. 05341), National Parks and Wildlife Service (Scientific Investigation Licenses B1581and C415), and ABBBS (Authority No. 2186). All capture, handling, and sampling protocols were approved by the Animal Ethics Committee of the University of Wollongong (Ethics No. AE96/04).

Group Monitoring

Data were collected from one hundred, eighty-six free-living WBBAs in 41 groups over the course of my study. Colour bands enabled individual identification with the aid of 10X40 binoculars. However, WBBAs sometimes removed colour bands, making identification difficult. Repeated captures were used to replace colour bands, confirm identities (by the ABBBS numbered, metal bands), and determine gonadal condition. To examine group structure and breeding behaviour of WBBAs, I focused behavioural observations and repeated captures on seven neighboring groups. During the three years of my study, these seven groups were monitored to determine group composition and breeding status. These focal groups were closely followed for two to three months each breeding season and sporadically during the non-breeding season. When positive identification was possible (often difficult due to bands being obscured), I noted associations between individuals. I also recorded where individuals or groups were observed. I did not mark or measure territory boundaries, but I noted landmarks and mapped groups' locations.

Blood Sampling

Blood samples were collected for DNA fingerprinting (red blood cells) and hormone analyses (plasma). As soon as possible after capture (typically within 3-5 min), a small blood sample (approximately 400 μl) was taken by puncturing an alar vein with a 26 ga. needle and collecting the blood into microhaematocrit tubes. All blood samples were collected between 0700-1200h. Blood was taken no more frequently than once every ten days. Blood samples were kept on ice until centrifugation; plasma was withdrawn using a Hamilton syringe and transferred to an Eppendorf storage tube. Both plasma and blood cells were frozen for later laboratory analyses.

I collected blood from 100 WBBAs in 17 social groups for DNA fingerprinting, including most members of my seven focal groups (see below). Although group membership changed over time, individuals were considered part of a group if they were caught at the same time and location as a given group and/or were observed taking part in group activities.

Laparotomy

Gender and gonadal status were evaluated by unilateral laparotomy. Methoxyfluorane inhalation was used to lightly anaesthetise the birds. Birds' wings and legs were gently restrained with rubber bands and secured with push pins to a cardboard box. Alcohol was used to cleanse the left flank area and a few feathers were plucked. A small incision (5-7 mm) was made in the flank, the skin and intestines were gently reflected using forceps, allowing examination of the gonads. Left testis volume was calculated from length and width measurements, using the volume of an ellipsoid: $V=4/3\pi ab^2$, where a=1/2 longest diameter and b=1/2 diameter at the widest part of the testis (Blanchard 1941). Ovary condition was scored based on the following characteristics:

1.0=completely regressed, flat, grey ovary
1.5=ovary slightly bumpy, but no distinct ova
2.0=distinct ova, but no follicular hierarchy
2.5= some follicles differed in size, but only marginally
3.0= ova larger than 1 mm and follicular hierarchy evident
3.5= yolking of developing ova
4.0=egg in shell gland

Because of concerns that breeding females might abandon nests after being handled, I usually did not perform laparotomies on females when I could ascertain gender and reproductive status by observation (i.e. presence of a brood patch or incubation behaviour).

DNA Analysis

One hundred WBBAs in 17 social groups were genetically fingerprinted. Most members of each of my seven closely-monitored, wild groups were fingerprinted; the remaining fingerprints were those of wild-caught WBBAs that were transferred to aviaries for experimental purposes.

The multi-locus DNA fingerprinting protocol I used follows that described by S. Yezerinac, M. Double, and A. Higgins (1996) and later updated by G. Sargent, S. Legge, and M. Hall (1998). An overview of the method follows. First, DNA is extracted from a tissue sample and is then fragmented by enzymes that recognise specific sequences of base pairs. These fragments are then separated on an agarose gel, where they are segregated according to size and weight. The separated fragments are then transferred to a membrane, and radioactive probes are then used to label core sequences of 9-15 base pairs. Next, the location of these base pair sequences is revealed on x-ray film as a series of bands. The number of DNA fragments (i.e. bands) present in an individual's lane results in a unique DNA fingerprint, and the similarity between two such DNA fingerprints represents the genetic similarity between two individuals.

1. DNA Extraction

Frozen samples of packed red blood cells were thawed, and 30 µl of red blood cells were placed into test tubes. To each sample, I added 3 ml 1 X TNE buffer (0.1 M Tris-Cl, 0.1 M NaCl, 1 mM EDTA), 300 µl 1-M Tris-HCl (ph 8.0), 10 µl Proteinase K, and 80 µl 25% SDS (Sodium dodecyl sulfate); contents of test tubes were agitated overnight in a 37°C incubator. The following day, 1.3 ml of 6M NaCL were added, tubes were manually agitated for approximately 15 seconds, and centrifuged for 15 minutes at 3500 RPM. The supernatant was removed with a pipette, and 8 ml 100% EtOH were added to each of these samples. The tubes were gently inverted until the DNA precipitate appeared. The DNA was spooled onto glass rods, rinsed in 70% EtOH, allowed to dry, dissolved in 500 µl 1 X TE buffer (10 mM Tris, 0.1 mM EDTA), and agitated overnight at 37°C.

This extracted DNA was diluted 1:5 with TE buffer, and these diluted samples were used to determine the quality and concentration of the uncut DNA, as follows. To verify that each sample of DNA was not degraded, 1 µl of diluted, uncut DNA was run on a 0.8% agarose test gel, and the concentration of a 10 µl sample of the diluted, uncut

DNA was checked using a Genequant spectrophotometer. The DNA was then digested as described below.

2. DNA Digestion and Precipitation

Samples of DNA extract equivalent to 15 μg of DNA were aliquoted into test tubes, and the DNA was digested with *Hae* III enzyme in distilled H_2O and enzyme buffer at pH 7.5. Samples were then incubated overnight in a 37°C water bath. The following day, I added 30 μl sodium acetate and 600 μl 100% EtOH and chilled the tubes on ice for two hours. The samples were centrifuged for 20 minutes at 13,000 rpm and EtOH was poured off. The precipitate was rinsed again with 500 μl 70% EtOH, and tubes were centrifuged for 7 minutes at 13,000 rpm. After this final EtOH rinse was discarded, samples were allowed to dry at room temperature until all EtOH had evaporated. The DNA precipitates were then resuspended in 30 μl of 1 X TE buffer and incubated overnight at 37°C. To verify that the DNA was properly digested, another test gel was run, and the concentration of the DNA was then determined with a Genequant spectrophotometer.

3. Electrophoresis and Southern Blotting

Five μg samples of digested DNA were separated on a 35 cm, 0.8% agarose gel in 1 X TBE buffer (89 mM Tris, 2.5 mM Boric acid, 89 mM EDTA), with voltage set at 70 V. Gels were run for 72 h, and the buffer was replaced after 36 h. Dye was loaded into an outside well of each gel to measure its running rate and to maintain a standard run length across gels. Voltage was set at 70 V and was minimally adjusted to maintain standard run-length across gels.

To prepare the gel for Southern blotting, the top and bottom 5 cm of the gel were trimmed. Next, I immersed each gel in a series of solutions to depurinate, denature, and neutralise the DNA. Depurination was achieved by immersing the gels in 0.25 M HCl for 10 mins. To denature I soaked gels in a solution of 0.4 M NaOH and 1.5 M NaCl for 30 mins, and to neutralise I used 0.5 M Tris-Cl (0.5 M Tris, 1.5 M NaCl, adjusted to ph=7.5 with HCl) for 30 mins. The DNA from the gels was then transferred onto Millipore-NY$^+$ membranes via capillary action, using a Southern blot. After the transfer was complete, the DNA was fixed to the membrane by UV crosslinking with a Spectrolinker™.

4. DNA Probing

The membranes were first soaked in distilled H_2O and then in Westneat's pre-hybridisation solution at 65°C for 3 to 4 h. Sequentially, three DNA probes were radiolabeled by primer extension with ^{32}P dCTP and then hybridised to the membranes; Jefferys' DNA probes 33.15 and 33.6 (Jefferys *et al.* 1985a,b) were used, as well as Per DNA probe (Shinn *et al.* 1985). After hybridising overnight at 65°C, membranes were washed twice in a solution of 2 X SSC (0.3 M NaCl, 30 mM sodium citrate) and 0.1% SDS and four times in a solution of 1 X SSC (0.15 M NaCl, 15 mM sodium citrate) and 0.1% SDS. Membranes were then sealed in plastic and labeled. Before hybridizing with the next probe, membranes were stripped by soaking them in 0.5% SDS and rinsing them in 1 X SSC.

5. Autoradiography

Plastic-sealed membranes were taped into metal cassettes, an intensifier screen was inserted, a sheet of X-ray film was laid over top, and the cassette was closed. The cassette was placed in a –70°C freezer for 2-10 days and then removed and developed using standard film development.

6. Scoring of Autoradiographs of Gels

DNA samples from between 18 and 22 individuals were run on each gel. Fifteen unique autioradiographs were produced from probing each of 5 gels with 3 different probes. To facilitate intragroup bandsharing comparisons, members of the same WBBA group were run next to one another on a gel. Due to distortions inherent to most gels and slight variations in the running of gels, comparisons across multiple gels were problematic; therefore, I examined bandsharing only within a single gel, not across gels. The bands present in a given lane on a single gel were compared to those of the other lanes on the same gel, resulting in multiple pairwise comparisons. Either the Jefferys' 33.15 or Per probes produced a shared band in all or almost all individuals within each gel; this row of shared bands was used to judge distortion between lanes of a given gel (Fig. 2-1). No bands were scored from the top 4 cms or bottom 7 cm of the gels, where bands were either too faint or too clustered for accurate scoring.

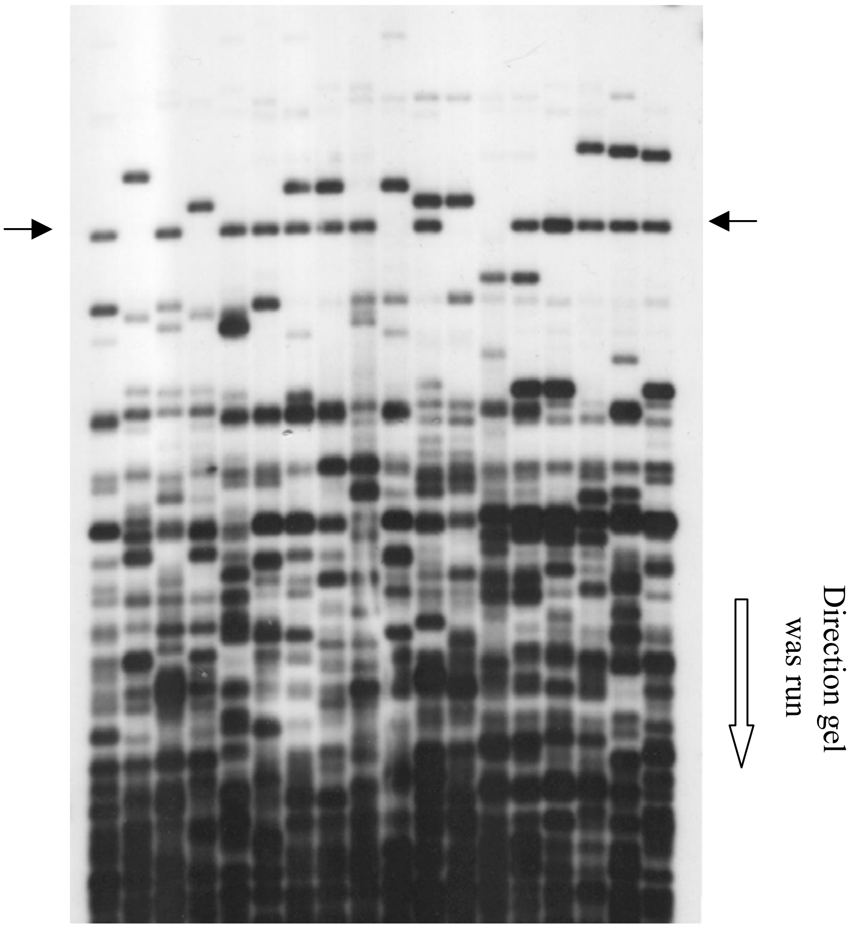


Figure 2-1. DNA fingerprints from 18 WBBAs probed with Per. Columns represent DNA bands from different individuals. A band common to 13 individuals is identified by the solid arrows and was used to ascertain that central lanes ran slightly slower than exterior lanes.

To determine the degree of bandsharing between individuals on a gel, I used the following equation:

$$D= 2N_{AB}/(N_A + N_B) \qquad \text{(Eqn. 1)}$$

Where D represents the degree of bandsharing, N_A is the number of bands scored for individual 'A', N_B is the number of bands scored for individual 'B', and N_{AB} is the number of bands shared by both individuals 'A' and 'B' (Wetton *et al.* 1987, Lynch 1990).

First, I calculated D for each probe separately. Then, assuming that bands exposed by each of the three probes were independent of one another, bandsharing coefficients were determined for all probes together (Burke and Bruford 1987). For this calculation I summed all bands produced by the 3 probes for the two birds being compared for the terms N_A and N_B in Eqn. 1 and used the sum of all bands shared by the two birds for the term $N_{AB.}$

Data Analysis

Although data collected from within a group of WBBAs are unlikely to be independent, they were treated as if they were for statistical analyses. This assumption follows Eberhart *et al.* (1983) who argued that such data better fit a model assuming independence than one acknowledging relatedness of observations.

Standard parametric statistics were applied to normally distributed data, and nonparametric statistics were used for data found not to be normally distributed. General statistics (e.g. mean and standard error) were used to describe many ecological and physical aspects of the WBBA population. Contingency tables were analysed with chi-squared tests and Fisher's exact test (when tables contained empty cells). Morphometric differences between males and females were tested with Student's t-tests. Kolmogorov-Smirnov two sample test was used to test for differences between the distribution of male and female intragroup relatedness. These statistical analyses were performed using SYSTAT™ 7.0 (1997).

Discriminate function analysis was used to develop a morphometric means of discriminating gender; this analysis derives a linear equation from a combination of independent variables which best discriminates between *a priori* groups (Dillon and Goldstein 1984). Morphometric measurements were acquired from birds whose gender had been determined by laparotomy.

Analyses conducted using bandsharing data involved multiple pairwise comparisons. The non-independence of these data invalidates the assumptions of conventional statistical methods. Therefore, I used Mantel tests (Mantel 1967) to determine the significance of differences among groups. The Mantel test compares two matrices of pairwise comparisons among individuals to test if they are significantly correlated. I compared two symmetric matrices; in one matrix, I included bandsharing coefficients, and in the other I coded each pairwise comparison as one of two categories. For these analyses, I used GENEPOP (1.2) software (Raymond and Rousset 1995), that calculated a rank correlation coefficient without approximation to test the significance of the Mantel test.

RESULTS

Description of Groups

1. Group Size

In BYSF, White-browed Babblers lived in groups of 3 to 12 adults. Group membership was easily discernable; group members followed one another on flights, preened and foraged en masse, and protected territory boundaries and breeding nests as a cohesive entity. In 41 groups mean size was 7.0±0.4 birds. Except for incubating females, solitary WBBAs were never observed. While large groups were often very cohesive, at times they split into smaller subgroups, and breeding pairs were frequently found foraging far from other members of their group. Also, I occasionally observed large flocks of between 20 and 30 WBBAs; these flocks were not included when calculating mean group size. Such flocks were observed in both the breeding and non-breeding seasons. I do not have sufficient sample sizes to statistically analyse inter-month or inter-year differences in group size, but there were no striking patterns in group size across seasons or years (Figs. 2-2 and 2-3).

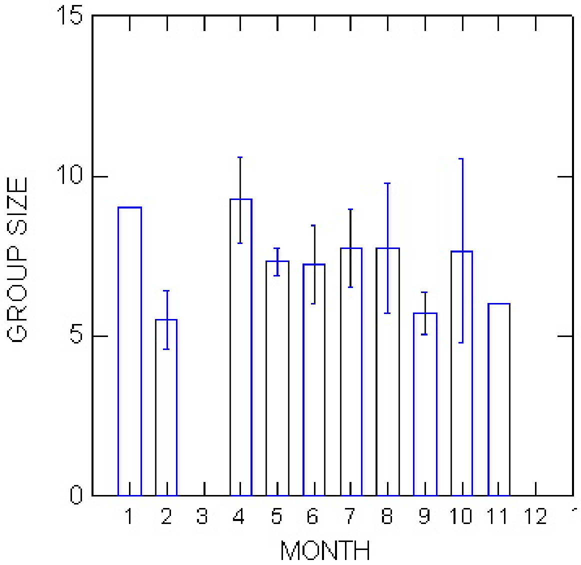


Figure 2-2. Group size across months in all years. Column height represents means, and error bars indicate standard error of the mean.

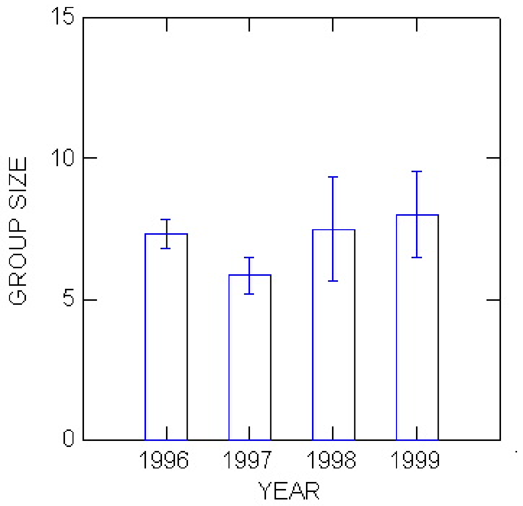


Figure 2-3. Group size across years. Column height represents means and bars indicate standard error.

2. Territories

Throughout the course of this study, groups could reliably be found in the same areas; most groups' territories remained fixed over at least three years. Of my seven closely-monitored groups, only one territory shifted markedly. Members of this group formed breeding pairs with two members from another group and founded a new territory along one edge of their original territory.

Groups' territories typically shared boundaries with those of other groups. Territory boundaries were loosely defined and often overlapped along edges with the territories of their neighbors'. However, confrontations rarely arose; I witnessed fewer than five territorial interactions in more than 1000 hrs of observation. During such interactions, WBBAs from both groups flew high into the tops of the trees and called noisily for up to five minutes. I never saw chases or other agonistic behaviour during these interactions, and after a few minutes, one group always moved away.

3. Gender Ratio

Group membership varied over the three years of my study, and one or more individuals invariably proved elusive when attempting to identify or capture all members of a given group. While these factors precluded accurate determination of gender ratios in groups, there tended to be more males than females per group in the majority of observations. This male-biased trend is substantiated by the overall gender ratio (in WBBAs banded in my study) of 1.6:1 males to females. Although this strongly suggests a male bias in most groups, there may be an inherent bias in sampling, as incubating females spend most of the day on the nest and thus may not have been captured or observed as frequently as males.

4. Breeding Structure

In each of the seven closely-monitored groups, I found more than one breeding female per group within each breeding season. Within groups' territories, active brood nests were built 30 to 100 m from each other. Two to three sequentially-breeding females were recorded in each of the seven groups. Multiple concurrent breeders were also confirmed: two females in each of six groups, and three in one group. From these and other behavioural observations, I suspect that it was common for most females within a group to breed concurrently, especially in the middle of the breeding season. Moreover, out of 47 females captured during peak breeding months, only five females had regressed ovaries and no brood patch (i.e. 11% of females in this group were non-reproductive). In each of two groups, there were two non-reproductive females, and a single non-reproductive female was found in another group.

5. Intergroup and Intragroup Relatedness

DNA fingerprinting was used to examine relatedness among WBBAs. All seven focal groups were fingerprinted, as well as seven other groups from BYSF and three from Warredary State Forest (approx. 50 km from BYSF). Hybridization with three DNA probes per gel resulted in an average of 90 scorable bands per individual (Table 2-1). The pattern of these bands made up an unique DNA fingerprint for each individual (Fig. 2-4). The similarities in banding patterns between individuals reflect similarities in their DNA base pair sequences. By comparing banding patterns between gel lanes, the genetic similarities between individuals could be evaluated by determining a bandsharing coefficient (D) using Eqn. 1.

Table 2-1. Mean number of bands scored per WBBA using three different DNA probes (Jeffreys' 33.15 and 33.6 and Per). "All" refers to a combination of all three probes.

Probe	Mean # bands	SE
33.15	27.4	1.4
33.6	32.2	2.9
Per	30.4	1.7
all	90.0	2.8

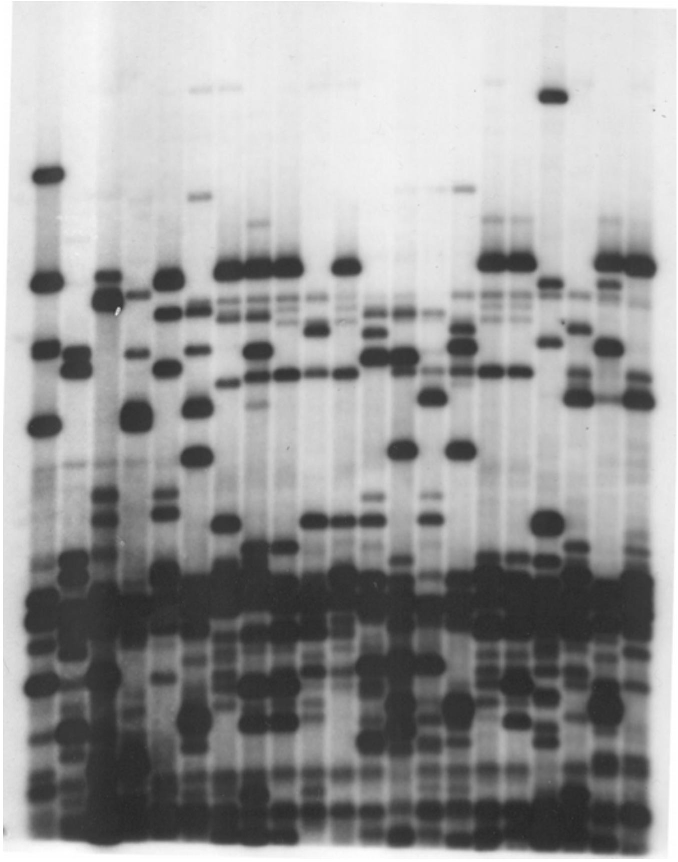


Figure 2-4. DNA fingerprints (Jeffreys' 33.15 probe) from 21 WBBAs in 3 neighboring WBBA groups. Each column depicts an individual's unique DNA fingerprint.

Each WBBA's fingerprint was compared to all others on the same gel; this resulted in approximately 190 pairwise bandsharing coefficients (D) per gel. White-browed Babblers' D ranged from 0 (very unique band patterns) to 0.947 (very similar), but the majority fell below 0.400 (Fig. 2-5). I was only able to analyse DNA bandsharing from one set of putative parents and their two chicks. Bandsharing coefficients for the mother and two offspring fell at 0.421 and 0.485, and D for the putative father and two offspring were 0.381 and 0.071 (Fig. 2-5).

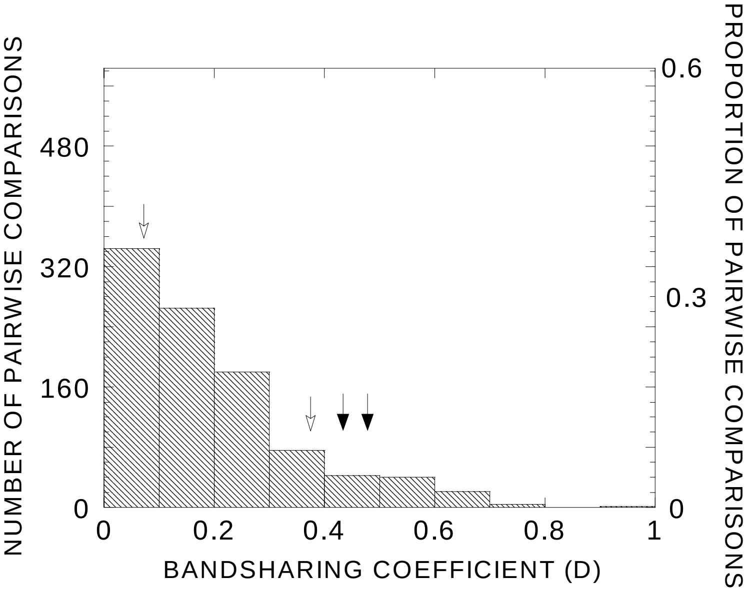
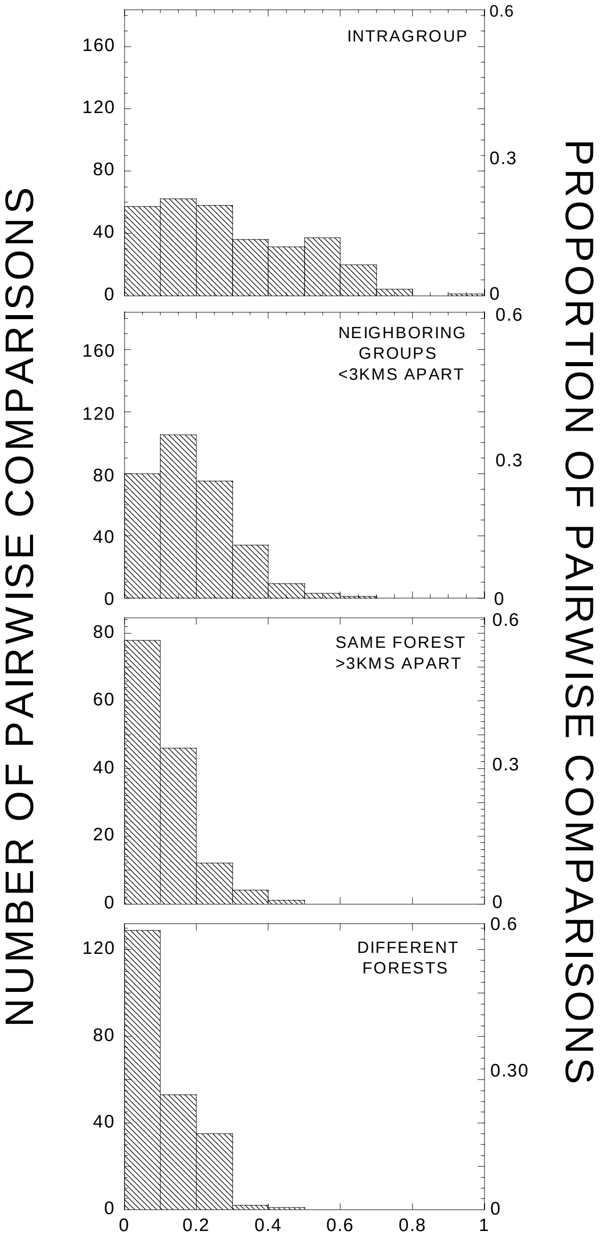


Figure 2-5. Distribution of DNA bandsharing coefficients for 100 WBBAs in 17 social groups. Open arrows indicate D values for putative father and 2 nestlings. Closed arrows indicate D values for mother and 2 nestlings.

There was a strong tendency for D values to be highest among birds in the same social group and next highest in birds from neighboring groups (Fig. 2-6). Bandsharing coefficients were much lower between birds captured more than 3 km apart in the same forest and between birds captured in different forests, with D distributions being highly skewed towards zero in both groups (Fig. 2-6).



BANDSHARING COEFFICIENT (D)

Figure 2-6. Comparison between the distribution of pairwise bandsharing coefficients and relative proximity of 100 WBBAs in 17 groups. Frequency and relative proportion of D between WBBAs from the same group, neighboring groups, distant groups from the same forest, and groups from different forests.

Bandsharing coefficients were markedly different from birds within a given social group compared to values for birds from different groups. I compared D from WBBAs in the same social group to D of birds in two different groups. In each of the five gels examined, D was significantly higher in the intragroup pairs than in the intergroup pairs (Table 2-2).

Table 2-2. Comparison of bandsharing coefficients between WBBAs in the same social group ("Intragroup") to those birds from different social groups ("Intergroup"). All P values derived from Mantel tests.

Gel	Intragroup mean ± SE	Intergroup mean ± SE	P
1	0.23±0.02	0.10±0.01	<0.001
2	0.29±0.02	0.18±0.01	0.008
3	0.32±0.03	0.16±0.01	<0.001
4	0.23±0.04	0.09±0.01	0.005
5	0.34±0.02	0.19±0.01	<0.001

6. Group Dynamics

Day-to-day structure of groups appeared somewhat fluid. Large groups often split into smaller groups for periods of the day, and breeding pairs frequently foraged separately from other group members. The membership of these smaller associations varied daily. However, overall group membership remained stable over many months.

Emigration/immigration between groups occurred occasionally. During the three years of my study, I confirmed 28 instances of intergroup movements. Of these, 13 intergroup movements occurred between the seven closely-monitored groups; the other 15 movements were of previously unbanded birds that immigrated into one of the seven focal groups. These figures underestimate group movements in that they do not include emigration of birds away from the focal groups. A number of birds that were originally part of a group later were not observed with that group, but it is unclear whether they were simply hidden from view, died, or moved to unmonitored groups.

Considering the 1.6:1 male bias in the population, intergroup movements were almost twice as common for females as for males: 15 females and 13 males changed groups. Furthermore, genetic evidence corroborates female-biased emigration from natal groups. Comparison of bandsharing values among females from the same group to bandsharing values among males from the same group shows that males have significantly higher D values than do females (Fig. 2-7; Kolmogorov-Smirnov test, P=0.007).

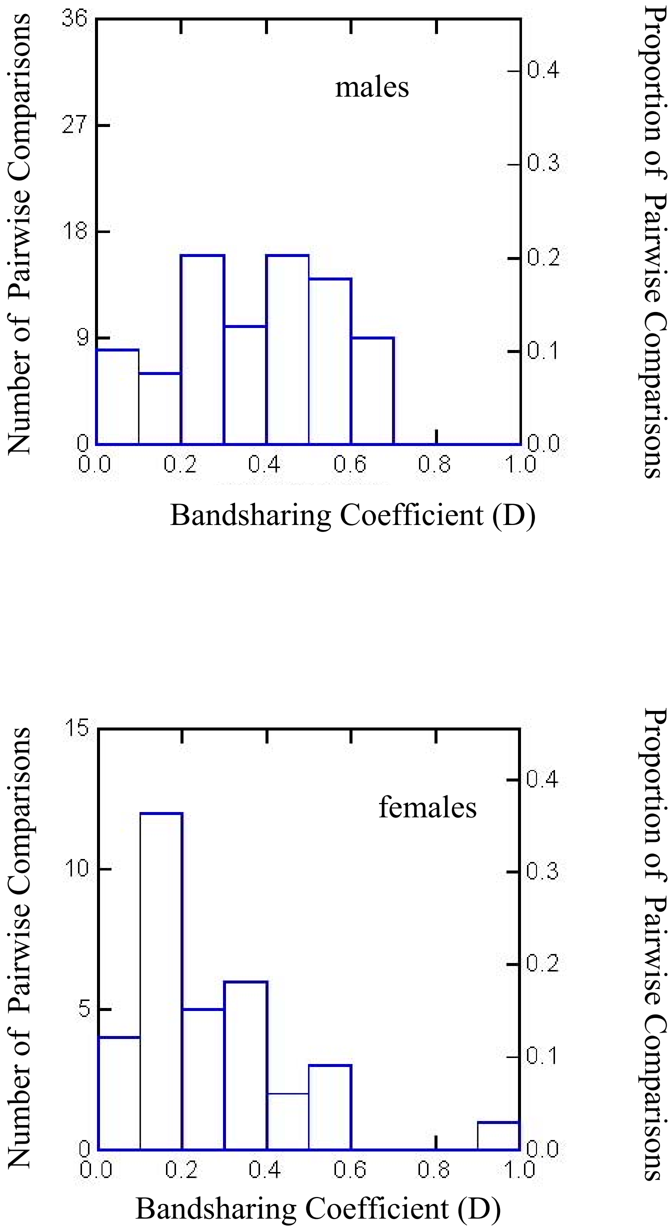


Figure 2-7. Comparison of intragroup relatedness of males versus females. Distribution of bandsharing values from pairwise comparisons of females from the same group and of males from the same group.

Life-History Stages

1. Non-breeding Season

Although I spent little time in the field outside WBBAs' typical breeding period, I gleaned pertinent information from the brief trips I made to my study site during non-breeding months (February-May). White-browed Babblers were found in groups year-round. During the non-breeding period, groups tended to range more widely than during breeding months; non-breeding groups often crossed into neighboring territories. Also, on two occasions, I observed winter flocks of 20-30 WBBAs; these flocks must have been composed of multiple groups. These large flocks were seen foraging and moving together as cohesive groups.

2. Courtship

Courtship behaviour began as early as May and was observed in all months up to February. From early courtship through incubation, a male constantly accompanied his mate. Often, courting pairs were observed separate from the rest of their group. The male frequently fed the female, and she often received the food and then performed a begging call and display in which she flattened her body and fluttered her wings. Pairs often sang duets, that were frequently accompanied by a wing flutter performed while birds of both gender stretched their bodies upwards. On a few occasions, I observed the courting pair repeatedly hopping over one another on a perch. Pairs were also frequently seen allopreening each other; however, this behaviour was not unique to breeding pairs.

3. Brood-nest Building

Brood-nest building activity began in June and extended through January, but only a few breeding attempts were made at the margins of this period. Almost all nesting attempts were initiated between the winter and summer solstices, when temperature and day length increased (Fig. 2-8). The actual number of nest attempts made per month could not be deduced, as the amount of time I spent in the field each month and the relative proportion of time I spent nest searching versus other field activities was too variable.

Both the male and female of a pair constructed brood nests without the aid of other group members. Brood nests were large, bulky, domed nests made of sticks and bark strips and lined with feathers and animal hair. The nests were ovoid, with the entrance always positioned at the narrow end of the nest. Entrances were approximately 8 cm in diameter. Nests were built at an average height of 4.7±0.4 m (n=29) almost exclusively in White Cypress Pines averaging 6.9±0.4 m (n=28) tall.

Please see print copy for Figure 2-8

Figure 2-8.

White-browed Babblers also construct nests for communal nighttime roosting. Each group of WBBAs had a number of roost nests (up to 20 per group in various states of repair) on their territory, and group members would congregate at a nest or cluster of nests toward dusk. Between two and seven (possibly more) birds would enter a nest and roost together at night. Roost nests were built year-round, and all group members actively took part in nest building. Groups performed nest maintenance and/or new nest construction daily. New material was sometimes added to old nests or sticks were rearranged. Roost nests were much bulkier than brood nests and could be simple platforms, partially domed, or completely domed. Roost nests were sometimes lined with bark strips, but never with feathers or hair. Sometimes, brood nests were converted into roost nests, and I know of one nest that was used as a brood nest, then a roost nest, and again as a brood nest.

4. Laying and Incubation

Clutch size was invariably two eggs (n=22 nests), and eggs were laid on consecutive days. Incubation began on the day the first egg was laid. Nest lining was added to the nest during laying and throughout incubation. A single female incubated each clutch, and the mated male (i.e. presumed mate) sometimes fed the female on the nest. The mated male often accompanied his mate to and from the nest; it was unusual for other group members to accompany the incubating female to or from the nest.

If the female ended her incubation bout before her mate arrived, she would fly to a high perch and give a repeated, high-pitched call ("pi-pi-pi-pi") that was returned by her group; she would then fly in the direction of the returned calls. When the male was in the vicinity of the brood nest, the incubating female would often give a two syllable call ("mee-neep") from inside the nest. For two incubating females observed over 272 mins between the hours of 0800-1100, incubation bouts lasted 44.5±9.2 mins, and females remained off the nest for 23.5± 5.3 mins. Because nests were discovered after the onset of incubation and eggs were lost before they hatched, I do not know whether observations took place during early or late stages of incubation.

5. Nestlings, Fledglings, and Juveniles

Of 30 active breeding nests found during the three years of my study, only four hatched young! As a result of such low breeding success, my observations of group behaviours relating to young are anecdotal, but relevant nonetheless. I never witnessed a nest depredation event, but eggs were removed from these failed nests, and no shell fragments were ever found. The failed nests were usually intact and the entrance holes were rarely enlarged, suggesting an avian predator of smaller or similar size to the WBBA or perhaps a snake. White-browed Babblers were seen to defend nests from Grey-crowned Babblers, and they were noticeably agitated by Pied Butcherbirds *Cracticus nigrogularis* in the area of their nests. I also suspected Grey Shrike-thrush *Colluricincla harmonica* of nest predation because of their size and ubiquitous presence in WBBAs' habitat at BYSF.

The hatching sequence was determined for only one nest: chicks hatched on consecutive days. While the exact duration of incubation was not determined for this nest, eggs were known to have hatched between 12-19 days after being laid. I observed activities at four nests with one to nine day-old chicks for a total of 28 h over 10 days. Nestlings were fed by both parents in three nests and by only the female in one. Only the suspected parents fed their nestlings; other WBBAs were noted near the nest, but they never carried food to the chicks. However, on a few occasions, five to ten WBBAs defended their cohort's nest from potential predators (me and Grey-crowned Babblers).

Of the four broods that hatched, two were depredated within five to ten days of hatching, and the outcome of the others was not determined. However, I did find one recently-fledged young from an undiscovered nest. This fledgling could not fly, but it could adeptly scramble into the undergrowth and hop into the lower branches of trees. This fledgling was in a large group of more than 11 adult WBBAs that were unbanded when first observed. I witnessed distraction displays where a WBBA approached me and then flew erratically around and up and down a nearby tree. All birds protected the fledgling by either performing distraction displays or scolding. There were also some older young-of-the-year WBBAs in the group; these birds could fly and were recognised by their yellow gape and relatively small body size. This group was captured and banded and observed to remain together for at least three months from my first observation of it. This group disbanded sometime within 12 months; group members joined nearby groups, dispersed outside my study area, or perhaps died.

Juvenile WBBAs were noted in two other large groups of more than 10 WBBAs in my central study area. On both occasions, there were a large number of unbanded WBBAs in the groups; as members of most groups in my central study area were banded, this suggested that the unbanded birds were not resident to the area. Groups with juveniles were very cohesive and defensive when approached. Some birds approached me with harsh scolding calls, and others performed distraction displays.

6. Renesting

Renesting after egg loss occurred throughout the breeding season. Some females began construction of new nests on the day after eggs were destroyed, others postponed renesting for a week or more, while others abandoned their nesting attempts for the season. Remarkably, one female laid at least five clutches; she initiated nesting in July and didn't hatch chicks until December.

Sexual Dimorphism

Although there are no sexually dimorphic plumage characteristics in WBBAs, there were morphometric differences between males and females. Adult males tended to be heavier and to have longer wings, culmens, and tarsi than adult females (Table 2-3).

Table 2-3. Morphometric measurements of male and female WBBAs, presented as mean and standard error for each measure. Statistical comparison of gender differences for each variable was tested using Student’s t-test.

	MALES			**FEMALES**				
	Mean	S.E.	N	Mean	S.E.	N	t	Prob.
Mass (g)	39.93	0.19	140	39.08	0.271	95	2.68	0.008
Wing (mm)	80.59	0.16	138	78.07	0.175	94	10.32	<0.001
Culmen (mm)	25.58	0.13	123	23.98	0.129	87	8.66	<0.001
Tarsus (mm)	26.67	0.10	123	25.91	0.095	87	5.52	<0.001

In an effort to develop a morphometric means of identifying gender in WBBAs, I performed a discriminant function analysis (DFA). I used the DFA to formulate a linear combination of wing, culmen, and tarsus, that could reliably predict gender; mass was not used as females’ mass varied with ovarian development (see Chapter III). After verifying the assumptions of the discriminant model (normality, equal variance and covariance within each group), I tested different combinations of variables to determine which most reliably classified gender, as determined by laparotomy. A combination of wing, culmen, and tarsus measurements correctly identified gender in the highest percentage of cases (Table 2-4).

Table 2-4. Morphological variables used in discriminant function analyses to identify gender of WBBAs, and the percent of birds correctly classified for gender using combinations of these variables.

Variables	% Correct
Wing	76
Wing + Tarsus	76
Wing + Culmen	81
Culmen	77
Culmen + Tarsus	77
Tarsus	66
Wing + Culmen + Tarsus	82

Using these three variables, the canonical discriminant score (DF1) was calculated with the following equation (Eqn. 2):

$$DF1= -46.416+ (0.367*\text{wing length})+ (0.261*\text{tarsus length})+(0.411*\text{culmen length}).$$

Pillai’s trace statistic verified that there was a significant difference between males and females when combining lengths of wing, culmen, and tarsus into a single DF1 (approx. $F_{3,245} = 58.75$, $P<0.001$). However, there is substantial overlap of male and female DF1s between –1 and 0 (Fig. 2-9). Reliability of correctly identifying 95% of cases can be achieved for males when DF1 exceeds 0.64 and for females when DF1 is below –1.34. Ninety-nine percent reliability can be achieved when DF1 exceeds 1.17 or is below –2.73 for males and females, respectively. The exact probability of correctly identifying gender from a combination of wing, tarsus, and culmen lengths measurements can be calculated using $1/(1+e^{-DF1})$ and $1-[1/(1+e^{-DF1})]$ for males and females, respectively (Fisher and van Belle 1993).

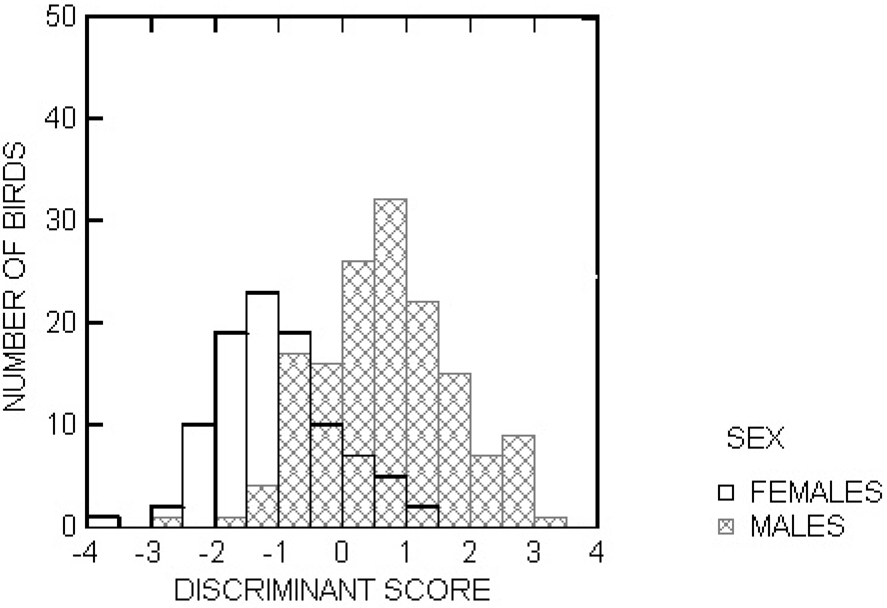


Figure 2-9. Distribution of discriminant scores among male and female WBBAs. Unfilled (females) and hatched (males) bars are overlaid.

Fat

White-browed Babblers of both genders were typically lean; no birds were found to have furcular fat scores above 2.5, and only three females had furcular fat scores greater then 2, one of which was a breeding female (unknown stage) and two were captured outside the breeding season and were not in breeding condition. There was a tendency for females to have more furcular fat than males, but this trend was not statically significant (χ^2=8.362, P=0.079). In wild-caught WBBAs, abdominal fat scores remained low year-round in all years in both genders; I never caught a bird with an abdominal fat level above one.

During the three years of my field study, there were significant year-to-year differences in furcular fat scores (χ^2=19.802, P=0.011). There were more birds with low fat scores (fat≤ 0.5) in 1996 and 1997 than in 1998 and more birds with high fat scores (fat≥ 1.5) in 1996 (Fig. 2-10).

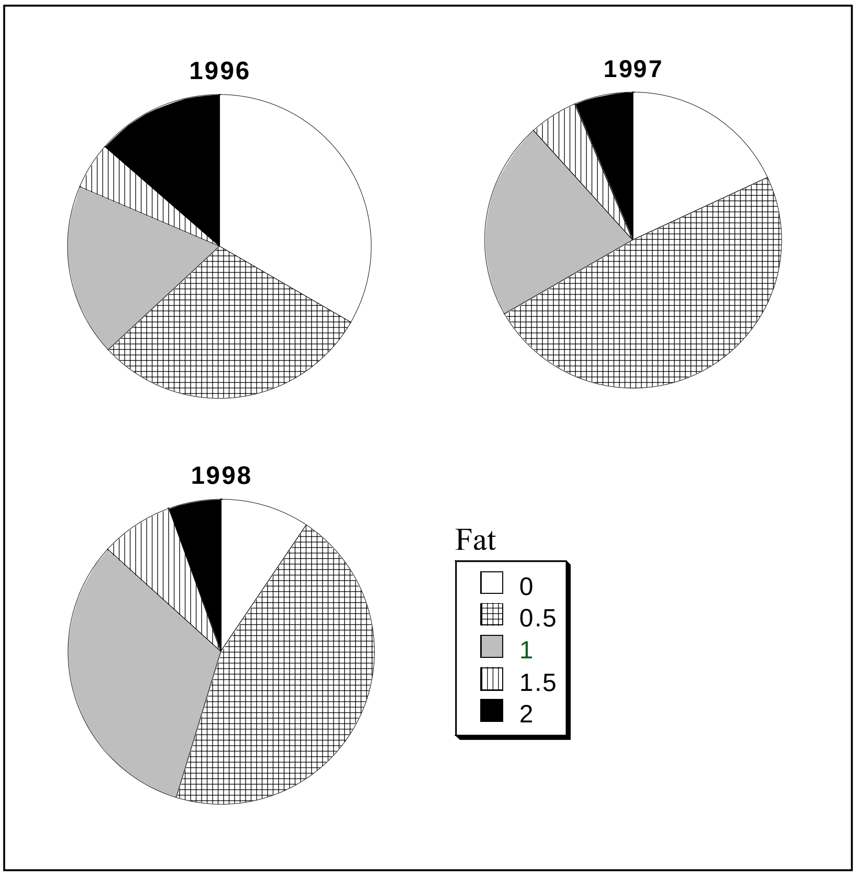


Figure 2-10. Comparison of WBBA fat levels among years. Pies represent years and slices represent proportion of birds having specific furcular fat scores.

Similarly, there were significant differences in month-to-month furcular fat levels (Fisher's exact test P=0.001), but scrutiny of the data suggested no discernable pattern. Furthermore, if the year was split into the non-breeding months and breeding months (Jan-Jun. and Jul.-Dec., respectively) there was no significant difference in furcular fat between seasons (χ^2=4.262, P=0.372).

Moult

There was a distinct annual cycle of moult in WBBAs (Fig. 2-11), and visual inspection of the data suggested no sexually dimorphic patterns. White-browed Babblers rarely moulted during the winter months; however, they did moult during the breeding season. In fact, body moult was found in the greatest proportion of birds during the height of the breeding season (Sept.- Nov.). Some birds were not moulting during this same period (Fig. 2-11), but I found no other differences between these birds and those undergoing moult. Of 109 WBBAs known to be in breeding condition (i.e. those whose breeding stages had been verified), 18% had no moulting feathers, 55% had light to moderate body moult, 12% had heavy body moult, and 15% had both wing and body moult. All birds caught in the post-breeding months (Jan.-Apr.) were undergoing some moult. Insufficient sample sizes in each month of every year prohibited analysis of inter-year differences in moulting patterns.

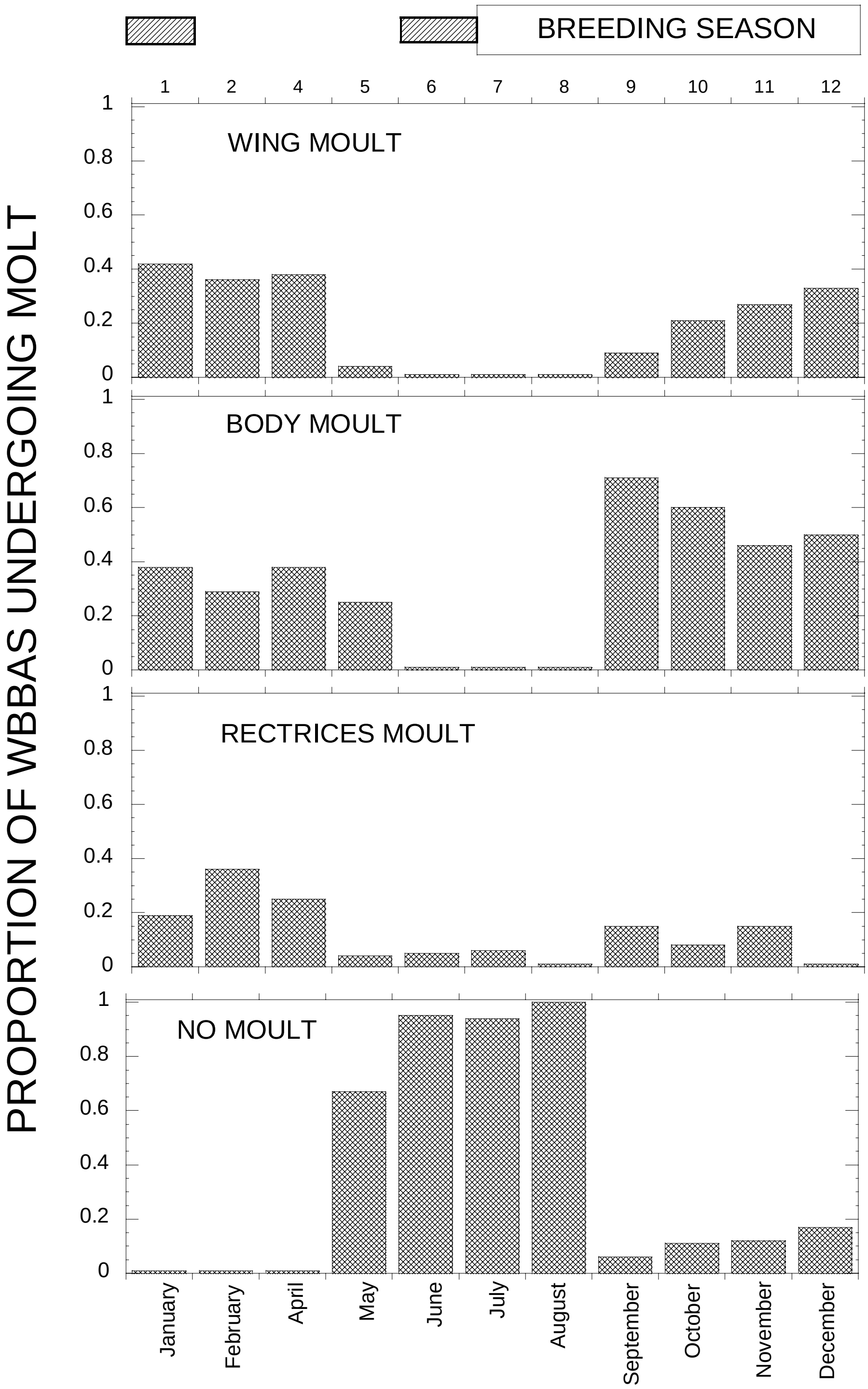


Figure 2-11. Seasonal moulting pattern in WBBAs. Proportion of birds moulting in each month. Smallest bars indicate 0. Timing of breeding season indicated by boxes above figure; smaller hatched rectangles indicate the margins of the breeding season, when only a few WBBAs bred (see Chapter III).

DISCUSSION

Cooperative Plural Breeding

Most reports identify WBBAs as singular-breeding cooperative breeders (Simpson and Day 1996, Cale 1999), but Chandler (1920) reported multiple breeding females per group. I also noted multiple breeding pairs in most BYSF groups. Geographic influences on social systems have been noted in other species: Acorn Woodpeckers (Stacey and Koenig 1990), Australian Magpies (Farabaugh *et al.* 1992), Bushtits (Sloane 1996), and Common Babblers (Gaston 1978). Regional variation in social behaviour may result from environmental differences; Wcislo and Danforth (1997) proposed that the presence or absence of various environmental cues may act to suppress or activate certain social traits. Moreover, Wcislo and Danforth (1997) suggested that natural selection may maintain variability in social organizations in response to temporally varying environmental condition. The social variability apparent in WBBAs may result from diverse ecological constraints imposed on populations in different locales or those subject to capricious environmental conditions.

White-browed Babbler groups that live in unfavourable habitats may be “forced” into a social system that allows only one breeding pair per group and requires the help of all group members to raise young. However, conspecific groups living in more favourable habitats may have sufficient resources to permit multiple breeding pairs per group. Cale (1999), who reported singular cooperative breeding in WBBAs, worked in fragments of natural vegetation amid farming land in the Kellerberrin area of the Western Australian wheat belt; there may have been greater ecological constraints imposed on these WBBAs than on those I studies within a 4000 ha State Forest. Ecological constraints and habitat saturation, such as scarce food resources and limited breeding vacancies, have often been cited as factors contributing to singular cooperative breeding (Emlen and Vehrencamp 1985, Ford *et al.* 1988, Sklepkovych 1997). More abundant resources and unsaturated habitat may have enabled multiple breeding pairs to attempt nesting in my study. Also, high rates of egg predation in the BYSF population may contribute to plural breeding; birds that may have otherwise helped with another's brood may be free to initiate their own nesting attempt due to the paucity of needy nestlings and fledglings.

Plural breeding is typified by multiple breeding pairs within cooperative groups, and has been well studied in only a few avian species: Acorn Woodpeckers (Koenig 1981), Bushtits (Slone 1996), Galapagos Mockingbirds (Curry 1988), Grove-billed Anis *Crotophaga sulcirostris* (Vehrencamp *et al.* 1986), Guira Cuckoos (Macedo and Bianchi 1997), Mexican Jays (Brown and Brown 1990), and White-fronted Bee-eaters (Emlen and Vehrencamp 1985). Emlen (1996) suggested that reproductive sharing may be expected at intermediate levels of ecological constraint, where breeding opportunities exist for subordinate individuals but when there are still benefits to group living. Despite the fact that WBBA helpers did not feed the incubating female or nestlings, helpers were present in this population and did assist with predator defense and rearing of fledglings and juveniles. In most cooperatively breeding species (e.g. Acorn Woodpeckers (Koenig 1981) and Splendid Fairy-wrens (Russell and Rowley 1988)), helpers assist with earlier reproductive stages, but others have found helping behaviour to be restricted to later stages; for instances, in the White-headed Vanga, helpers only assist with predator defense (Nakamura *et al.* 2001), and in Gray jays, helpers contribution only after chicks fledge (Waite and Strickland 1997).

As WBBAs were consistently observed in groups of more than three birds, it seems that there was an intrinsic value to group living. They may benefit from group foraging through greater feeding efficiency and flushing prey more effectively and from group predator detection and defense (Macedo and Bianchi 1997). Also, communal nighttime roosting may be important for WBBAs' survival. White-browed Babblers devoted a high proportion of their lives to nest construction and maintenance; presumably, WBBAs' roost nests offer thermal benefits and protection from predators, as they do in Green Woodhoopoes (Du Plessis and Williams 1994), Sociable Weavers *Philetairus socius* (White *et al.* 1975), and Verdin *Auriparus flaviceps* (Buttemer *et al.* 1987).

Although WBBAs cooperated in many aspects of their life, breeding opportunities existed for multiple pairs on many territories, and plural breeding resulted. Sloane (1996) found year-to-year and flock-to-flock differences in a long-term study of the plurally breeding Bushtit; she warned that long-term studies are necessary to ascertain the range of behaviours possible in species with complex mating systems. Birds must weigh the costs and benefits of independent versus cooperative breeding at each breeding opportunity. Independent breeding (i.e. plural cooperative breeding) may be the best bet in some years on some territories. Perhaps in other years or on other territories, WBBAs' breeding opportunities become more limited; in such cases, singular cooperative breeding, with multiple group members helping with a single nest, may be the norm.

Kinship

Because I don't have a large data set of individuals with known relatedness with which to calibrate bandsharing coefficients, I can not reliably assign relatedness based on my bandsharing data. However, assumptions regarding kinship of WBBAs in my study population can be made. There is a greater likelihood for close relatives to share a larger number of bands (from DNA fingerprints) than do unrelated individuals (Quinn *et al.* 1994). Among avian species, bandsharing coefficients from unrelated, conspecific typically fall below 0.25 and those for closely related individuals above 0.50, although exceptions are found (Table 2-5). It is valid for bandsharing coefficients among different species to be compared, as bandsharing coefficients represent the ratio of shared bands to total bands scored for both individuals, rather than an absolute number of scored bands. Three out of four putative parent-offspring bandsharing coefficients from my study fell close to those reported for closely related individuals in other studies (Table 2-5). As has been found in other species, bandsharing coefficients from unrelated WBBAs (those from distant groups) in my study were typically low and almost invariably below 0.30. Although more data is necessary to be certain, I expect that WBBA bandsharing coefficients that exceeded 0.40 indicate a high probability that the individuals were first order relatives.

Table 2-5. Mean bandsharing coefficients between unrelated and related individuals in six avian species.

Species	Unrelated (mean ± SE)	Related (mean ± SE)	Relationship
House Finch[1]	0.22 ± 0.03	0.57 ± 0.02	mother X nestling
Noisy Miner[2]	0.22 ± 0.01	0.56 ± 0.001	mother X nestling
Noisy Miner[2]	0.22 ± 0.01	0.57 ± 0.01	father X nestling
Long-eared Owls[3]	0.135 ± 0.014	0.502 ± 0.054	close relatives
Siberian Jays[4]	0.43 ± 0.08	all > 0.6	first order
House Sparrow[5]	0.151 ± 0.019	0.536 ± 0.012	mother X offspring
House Sparrow[5]	0.151 ± 0.019	0.598 ± 0.009	father X offspring
House Sparrow[5]	0.151 ± 0.019	0.580 ± 0.009	full siblings
White-browed Babbler[6]	0.19 ± 0.025	0.37 – 0.87[7]	mother X offspring
White-browed Babbler[6]	0.19 ± 0.025	0.41 – 0.82[7]	father X offspring
White-browed Babbler[8]	0.10 ± 0.01	0.421, 0.485[9]	mother X offspring

[1]*Carpodacus mexicanus*: Hill *et al.* 1994, [2]*Manorina melanocephala*: Poldman *et al.* 1995, [3]*Asio itus*: Galeotti *et al.* 1997, [4] Ekman and Tegelstrom 1994, [5]*Passer domesticus*: Wetton *et al.* 1992, [6] Dunlop 1999, [7] range of coefficients reported, [8]present study [9] coefficients from 2 offspring.

Of the two father-offspring bandsharing coefficients I calculated, one fell close to 0.40, but the other fell considerably lower (0.07). It appears that this male fathered only one of the chicks in this nest. Extra-pair fertilizations, once considered rare, are now known to be common among avian species (Avise 1996, Haydock *et al.* 1996). However, in a different population of WBBAs, in the Kellerberrin district of Western Australia, extra-pair fertilizations were rare, 1.6% (Dunlop 1999). More data are necessary to determine if the same holds true for WBBAs in BYSF.

White-browed Babblers in the same social group were more closely related to each other than to birds in different social groups. In many cooperatively breeding species, groups are composed of nuclear families (Stacey and Koenig 1990). White-browed Babbler groups in my population also seemed to be made up of family members; thirty percent of bandsharing coefficients between individuals in the same group were above 0.40. High intragroup bandsharing was most likely due to natal philopatry. In addition to the benefits of group living discussed earlier, natal philopatry may enable young WBBAs to learn from older birds, receive protection from predators and harsh environmental conditions, and more easily establish a breeding site (Strickland 1991, Sklepkovych 1997). Parents may also benefit from allowing their offspring to remain on their territories by increasing offspring survival and, thus, their own fitness (Zahavi 1990, Ekman and Tegelstrom 1994).

Individuals from neighboring groups were more closely related to each other (higher bandsharing coefficients) than distant groups, suggesting that dispersals regularly occurred between neighboring groups. Equally low relatedness (bandsharing coefficients) was found in individuals from distant groups (more than 3 kms from each other) within BYSF and in individuals from forests approximately 50 kms from each other. This also suggested that dispersal was largely limited to nearby groups. This observation is consistent with Cale's (2003a) description of WBBAs' population structure in Western Australia, in which groups were organized into "social neighborhoods." Interactions and dispersals were common between groups within these "social neighborhoods," and much less common beyond the "social neighborhoods" (Cale 2003a). A high degree of dispersal and perhaps a high incidence of extrapair copulations between neighboring groups could explain the high relatedness between WBBAs in neighboring groups within the BYSF population. Furthermore, my anecdotal behavioural observations also support the organisation of WBBAs into "social neighborhoods," as described by Cale (2003a).

The high degree of relatedness within the "social neighborhood" may help explain the low level of aggression and high degree of collaboration observed in WBBAs. Group activities, such as communal foraging and roosting, may benefit all group members. As group members tend to be genetically related, social behaviours that enhance fitness may be selected for, based on both individual and kin selection. Conversely, as both territorial and intragroup aggression could be detrimental, they may be selected against.

Group Stability

While WBBA groups often fractured into smaller associations for periods of the day, long-term group composition was largely stable. Groups maintained permanent territories, although territory boundaries were relaxed outside the breeding period. Groups retained ownership of a number of roost nests within their territories and spent a good deal of time maintaining these nests. As vigilant nest maintenance may be costly and the thermal benefits of communal roost nest use may be valuable, territory permanence may be important to protect groups' investment in building and maintaining nests and to insure nest availability. Year-round territories may also alleviate territorial confrontations and facilitate foraging efficiency.

Although group composition was largely stable, flocking and splitting did occur. Groups sometimes split into subunits for periods of the day; small subunits were observed preening, roosting, or foraging apart from their group. I also observed large flocks in the winter and during the breeding season. Winter flocking may enable WBBAs to exploit limited resources, as suggested for White-winged Choughs and Hoatzin *Opisthocomus hotazin* (Rowley 1978, Strahl and Schmitz 1990, respectively). During the breeding season, large flocks of WBBAs sometimes formed around fledgling or juvenile birds. These flocks probably included non-breeding birds and those whose nests had failed. Sloane (1996) noted flocks containing juvenile and fledgling Bushtits, and she hypothesized that these flocks enabled successful parents to attempt a second brood.

Dispersal was more common in female than male WBBAs, as has been noted in other cooperatively breeding species: Bushtits (Sloane 1996), Splendid Fairy Wrens (Rowley and Russell 1990), and Arabian Babblers (Zahavi 1974). Dispersing may be more costly and dangerous than staying at home, and the male bias in the BYSF WBBA population may be due to higher mortality in females than males. Higher rate of dispersal in one gender over the other is thought to be a means of avoiding inbreeding (Johnson and Brown 1980). As WBBA groups were made up of related individuals (see above), female dispersal may help prevent inbreeding by insuring that opposite gender siblings breed in different locales (Johnson and Brown 1980).

Sexual Dimorphism

Although WBBAs' gender can not be determined based on plumage characteristics, body size may be used to ascertain gender. Based on morphometric measurements and laparotomy data, I formulated an equation (Eqn. 2) that may be useful for those who wish to determine WBBA gender in a noninvasive manner. However, because there may be geographic variation in body size, this equation should be validated for use at other locales.

Patterns of Fat Storage and Moult

Unlike migratory bird species, that undergo seasonal patterns of fattening, WBBAs remain lean year-round. As there are numerous costs associated with excessive fattening, such as metabolic costs, transportation costs, reduced manoeuvrabilty, and increased exposure to predators, birds should only fatten to an extent that is necessary (Blem 1975, Jansson *et al.* 1981, Lima 1986, Witter and Cuthill 1993, Biebach 1996, Klaassen and Lindstrom 1996, Norberg 1996). When the risk of starvation is high, birds should carry larger fat reserves (Houston and McNamara 1993, Bednekoff and Houston 1994, McNamara *et al*. 1994); for instance, many birds, especially migratory birds, must build up fat reserves to protect against variable or low food supply (Biebach 1996, Gosler 1996, Pravosudov and Grubb 1997) or low temperatures that result in increased thermoregulatory costs (Biebach 1996, Nilsson and Svensson 1996). Alternately, when food is abundant or predictable, birds should minimize the costs of carrying fat loads (McNamara *et al*. 1994).

In the WBBA, the costs of carrying excess fat may exceed the benefits. White-browed Babblers are a largely sedentary species living in a mild climate with minimal seasonality. Under such conditions food is available year-round, and WBBAs may not need large fat reserves to hedge against low food supply or severe weather conditions. Furthermore, thermoregulatory costs during the cool nights are likely ameliorated by the use of communal roost nests by WBBAs. While I did not examine the energetic benefit of communal roosting, the use of communal roost nests by other avian species have been shown to confer considerable metabolic energy savings (Kendeigh 1961, Buttemer *et al.* 1987); I expect a similar energy savings in WBBAs utilising communal roost nests.

The variation in fat levels observed in WBBAs is probably related to insect availability and social factors, that may dictate priority of access to resources (Barkan *et al.* 1986). While there were no discernable patterns in fat levels associated with WBBAs' annual cycle, females' tendency to carry more fat than males may be due to females' storing fat for egg production. Also, courtship and incubation feeding by males may result in fatter females.

The seasonal pattern of moult parallels changes in ambient temperature. During the coldest months, WBBAs do not moult body or wing feathers. As WBBAs foraged on the ground, rectrices received much wear and were sometimes replaced in the winter. Many avian species refrain from breeding and moulting synchronously, as both are energetically costly (Wingfield and Farner 1978, Hemborg 1999). However, in some tropical, Australian temperate, and opportunistically breeding species, moulting and breeding occur simultaneously (Dittami and Gwinner 1990, Astheimer and Buttemer 1999, Jacobs and Wingfield 2000). Like such species, the majority of breeding WBBAs were found to be moulting. Some breeding WBBAs were moulting primaries, and body moult was recorded most often during the peak of the breeding season. Also, WBBAs appeared to have a protracted moult that lasted several months and progressed slowly. Whereas a short moulting period may be too energetically costly to undertake while breeding (Dittami 1986), the daily energetic costs associated with WBBAs' protracted moult may be sufficiently reduced to not have a deleterious effect on breeding. Furthermore, the small clutch size (2 eggs) of WBBAs may alleviate some of females' energy demands associated with breeding and may permit the overlap between moulting and breeding. Perhaps the energetic costs of moulting during the coldest months (e.g. loss of flight and thermoregulatory efficiencies), accompanied by decreased food availability, is more costly than moulting and breeding synchronously.

Low Nesting Success

Nest predation is common among Australian passerines (Poiani and Pagel 1997), and laying small clutches over an extended period may be an antipredator strategy (Poiani and Jermiin 1994). Nevertheless, the exceptionally high rate of egg loss in WBBAs in BYSF is intriguing. Although none of the nests I found succeeded in fledging young, WBBA numbers remained high within BYSF during the three years of my study. Nevertheless, WBBAs long life span (≥ 6 years, N. Schrader pers. comm.) may obscure a potentially declining population.

I suspect that low nesting success resulted from interspecific predation. Low nesting success due to predators was also noted in a concurrently breeding population of Rufous Whistlers in BYSF (McDonald pers. comm.). Perhaps in the years of my study, predator numbers were high, and WBBA nesting success suffered. Potential predators observed in the area of WBBA nests included: Grey Shrike-thrush, Pied Butcher Birds, Grey-crowned Babblers, and snakes.

Although I obtained no evidence to suggest that WBBAs destroyed each others eggs, intraspecific predation is another possible cause of the high mortality. Egg tossing or destruction of chicks is common in plurally breeding birds (Vehrencamp 1977, Trail *et al.* 1981, Mumme *et al.* 1983, Barkan *et al.* 1986, Curry 1988). For instance, dominant Galapagos Mockingbirds may interfere with the nesting attempts of subordinate birds in an effort to control subordinates' reproduction, decrease the costs of sharing territories, or perhaps to increase the number of potential helpers (Curry 1988). While interspecific predation seems more likely, some of the WBBA's observed nest failures may have resulted from intraspecific predation. It is also possible that my presence affected nest failure rates in WBBAs; predators may have learned to follow me to nests, or nests may have been abandoned after their discovery. However, if this were the case, I would have expected to have discovered some nests already at the nestling stage and to have encountered more groups with fledglings.

Life History Stages

In many areas of Australia, including BYSF, the mean annual variation in temperature is low (Fig. 2-8). In the absence of extreme fluctuations, conditions are potentially favourable for breeding over long periods of the year. Accordingly, many Australian bird species have long breeding seasons (Simpson and Day 1996), and I recorded WBBA breeding activity in eight months of the year. Nevertheless, I did find that the majority of WBBAs' breeding attempts (in BYSF) were made between the winter and summer solstices, while day length and ambient temperatures were increasing. This tendency may have been related to a presumed increase in food availability during warmer months; much of WBBAs diet likely consisted of ectothermic insects and small herptiles that would be more active as temperatures increased.

From courtship through the nestling stage, WBBA breeding pairs behaved similarly to many biparentally breeding songbirds; the breeding pair built the nest, the female incubated alone, the male sometimes fed the incubating female, and the pair fed the nestlings. Even though WBBA breeding pairs continued to associate with their group, breeding activities were confined to pairs. This is unlike a number of other cooperative species in which the group helps build the nest (e.g. Grey-crowned Babbler: King 1980), more than one bird incubates (e.g. Groove-billed Anis: Vehrencamp *et al.* 1986) and many birds feed the nestlings (e.g. White-fronted Bee-eaters: Emlen and Wredge 1989).

As there were multiple breeding pairs per WBBA group, the pool of potential helpers was smaller than in singular cooperatively breeding species. Even if all pairs were not nesting simultaneously, birds may have been busy courting or looking for mates or nest sites. However, the 1.6:1 male to female ratio indicates that some males must have been unmated and available to help with nest building or chick rearing. Perhaps these "free" males were actively seeking mates or extra-pair copulations instead of investing in helping behavior. Furthermore, non-breeding helpers may have been discouraged from assisting with nesting by the breeding pair. Because frequent renesting brings with it the possibility of mate-switching, WBBA non-breeding helpers may have posed a threat to either member of the breeding pair at any stage in the breeding cycle, and thus may have been discouraged from helping.

The high incidence of mate guarding suggested that the threat of extra-pair copulation or mate switching may have been common. Furthermore, for the one set of parents from which I have genetic data, the putative breeding male appears to only have fathered one of the two offspring. In many cooperatively breeding species, mate guarding was most common during the period of egg fertilization (Nakamura 1998b); however, WBBA males not only guarded their mates during nest building and laying, but also throughout incubation. Mate guarding during incubation may help strengthen pair bonds (in case of renesting) and decrease predation risk (of mates and eggs).

The high risk of nest predation in BYSF may also have influenced alloparental behaviour within groups of WBBAs. As nest predation may increase with parental activity (Martin *et al*. 2000), the absence of alloparental behaviour in WBBAs prior to the fledgling stage may be a predator avoidance strategy. As has been suggested by Strickland and Waite (2001), reducing (or eliminating) the frequency of nest visits by non-breeders may decrease the probability of nest detection by predators. Especially for WBBAs in BYSF, where nest predation rates were exceedingly high, the threat of predator detection may outweigh any benefits accrued from the feeding of incubating females and nestlings by non-breeders. Furthermore, intraspecific variation in allofeeding behaviour may be explained by differences in predator species or density among population (Strickland and Waite 2001). For example, low squirrel density in the habitat of the Florida Scrub-Jay may permit allofeeding of nestling, while high squirrel density in the habitat of the Western Scrub-Jay *Aphelocoma coerulescens* of Oaxaca may prohibit nestling allofeeding (Hall and Kelson 1959, Burt and Peterson 1993, Strickland and Waite 2001). Based solely on WBBA nest predation rates (100% in BYSF vs. 39% in the Kellerberrin region of W. Australia (P. Cale pers. comm.)), there may have been fewer predators in WBBAs' habitat in W. Australia than in BYSF; this may help explain the presence of nestling allofeeding in W. Australia and its absence in BYSF.

CONCLUSIONS

In BYSF, WBBSs are plural cooperative breeders, with few non-reproductive females in the population. Social groups are likely made up of extended families, and the population structure is consistent with the "social neighborhoods" described by Cale (2003a). In my study, individuals within groups are more closely related than between groups, and neighboring groups contain more close relatives than do distant groups. Short-distance dispersal is more common than long-distance dispersal, and there are frequent interactions (typically amicable) between neighboring groups. Day-to-day social associations are fluid (i.e. pairs or small cohorts form), but long-term group structure remains stable.

Pre-fledging reproductive stages in WBBAs resemble those of many biparentally-breeding passerines. There is a strong bond between the breeding pair, and only the pair takes part in nest building, laying, incubation, and feeding nestling. However, after the young fledge, multiple group members support the young. Sometimes, large groups of WBBAs, containing individuals from multiple groups, were found with young birds. These large groups protect (and I suspect provision) the young of the year, but these associations are short-lived and likely disband within a few months.

White-browed Babblers can moult and breed simultaneously. They both breed and moult over many months of the year (notably the warmer months). A protracted moult coupled with a small clutch size may ameliorate the energetic costs of moulting and breeding simultaneously.

Chapter III. Gonadal and Hormonal Phenologies in Free-living White-browed Babblers

INTRODUCTION

Gonadal state and hormone levels change over time in response to a variety of factors, including seasonal and behavioural cues. However, not all animals respond to the same cue in the same manner. Variations in temporal patterns of gonad and endocrine cycles can sometimes be explained by differences in a species' life history strategy (Dittami and Gwinner 1990, Logan and Wingfield 1995). For instance, breeding seasonality, mating system, and extent of parental investment may correspond to a species' distinct physiological cycles. In this chapter, I will explore temporal changes in reproductive hormones and gonad condition in the WBBA, a quasi-seasonal, plural cooperative breeder.

Temporal changes in reproductive hormones are thought to synchronise physiological changes with the time of year when conditions are most favourable for breeding (Balthazart 1983). Typically, environmental factors, such as photoperiod or food availability, are interpreted by the central nervous system, which can then trigger hormonal changes that help time reproductive events (Wingfield *et al.* 1992). Hormones probably play two roles: facilitation and coordination of males' and females' reproductive behaviours (e.g. singing, territoriality) and initiation of reproductive readiness (e.g. sperm production).

Many avian studies that have examined temporal changes in hormone levels have focused on species breeding in north temperate regions, where the breeding season is short, and breeding stages closely parallel the calendar date in highly synchronous populations (e.g. Morton and Allan 1990, Wingfield and Goldsmith 1990). In many such species, there is a short window of breeding opportunity, and it is advantageous for birds' endocrine systems to respond to "initial predictive information," such as photoperiod, in order to anticipate the arrival of the most suitable season for breeding (Wingfield *et al.* 1992). Recently, it has also been shown that species living in less seasonal environments, such as the tropics, also respond to "initial predictive information," but fine tune their breeding readiness to local environmental conditions, such as food availability (Hau *et al.* 2000b). However, in species living in minimally seasonal environments or those with protracted breeding seasons, hormones may play a less important role as signaling cues than for those living in highly seasonal environments with short breeding opportunities (Wingfield and Lewis 1993).

The proximate mechanisms regulating mating behaviour have not been well investigated for species with prolonged or continuous reproductive activity. Many of these species live in either unpredictable or highly constant environments and breed opportunistically in response to short-term environmental cues. While some species living in such environments show distinct seasonal cycles of gonadal maturation and regression (e.g. Wikelski *et al.* 2000), others maintain gonads in an advanced state of development for extended periods (Immelmann 1971, Serventy 1971, Immelmann 1973, Crews and Moore 1986). In central west NSW, I recorded breeding in WBBAs for eight months of the year, and breeding may even take place year-round. However, there did appear to be a seasonal bias, with most breeding attempts occurring between the winter and summer solstices (Chapter II).

Seasonality aside, there are many other aspects of a species' life history that may influence physiological cycles. For instance, differences in breeding strategies, such as pairing patterns (e.g. monogamy, polygamy, polygynandry) and style of offspring care (e.g. uniparental, biparental, cooperative), can be reflected in diverse levels and patterns of hormonal secretions (Gratto-Trevor *et al.* 1990, Vleck and Brown 1999). Some aspects of WBBAs' breeding are typical of monogamy: males and females form pair bonds for the duration of the breeding season, females incubate alone, and males feed their incubating mates. However, unlike most monogamous songbirds, WBBA pairs are also members of social groups containing multiple breeding pairs, as well as some non-breeding individuals. Females in the process of laying their clutch may be exposed to begging nestlings, males with incubating mates may come into contact with laying females, and non-breeding birds may forage along side fledglings. Stimuli from sexually receptive females, begging young, and male-male interactions can influence hormone secretions (Wingfield *et al.* 1987, Wingfield *et al.* 1989, Richard-Yris *et al.* 1998). In this chapter I will examine whether WBBAs' hormone and gonad cycles reflect their plural-breeding social structure. Are hormonal signals important in regulating reproductive activity in a species that shows low responsiveness to seasonal changes, little territorial behaviour, and a complex social system?

Some endocrine signals are probably universal across a wide range of animals, as they initiate or facilitate essential reproductive events, such as spermatogenesis and ovulation. However, hormonal fluctuations may also reflect distinctive behavioural patterns associated with a species' breeding strategy or social system. Many studies have shown that sexual and social behaviours in male birds are strongly influenced by T (Adkins-Regan 1981, Balthazart 1983, Wingfield and Ramenofsky 1985). Elevated T has often been associated with spring-time testicular recrudescence in seasonally-breeding species, territory establishment and defense, and mate-guarding behaviour (reviewed in Wingfield *et al.* 1994a). However, chronically elevated T can have detrimental effects (Silverin 1980, Dufty 1989, Zuk *et al.* 1995). Temporal variations in

T levels may reflect a trade-off between the costs and benefits of elevated plasma T (Vleck and Brown 1999). Also, mating strategies may influence cycles of T titres. For instance, in many monogamous species, in which male-female pair bonds endure for the entire breeding season (or longer) and aggressive male-male interactions are isolated to the beginning of the breeding season, males' T levels are high for only a short period at the onset of breeding (Wingfield and Farner 1993). However, in polygynous species, in which males interact with receptive females and other males for extended periods, T levels are often elevated for prolonged periods (Beletsky *et al.* 1990, Vleck and Brown 1999). This variation may reflect differences in the intensity of male-male interactions, males' exposure to receptive females, and the likelihood of copulations. In many avian species, males' T levels become elevated when females are receptive (Silverin and Wingfield 1982, Wingfield *et al.* 1989) and when males are challenged by another male (Wingfield *et al.* 1987). In the plurally breeding WBBA, where breeding males may be exposed to multiple breeding females and other potentially breeding males within the group throughout the breeding season, do males' T levels reflect his mates' breeding stage?

While T has received a great deal of attention in male birds, other hormones are also involved in the coordination of the reproductive effort. Estradiol and P are involved in reproductive physiology and behaviour in female birds. Estradiol regulates follicular maturation and vitellogenesis (Balthazart 1983) and stimulates courtship behaviour in some passerines (Moore 1982, Searcy and Capp 1997). Progesterone stimulates oviduct development, ovulation, and nest building behaviour in some avian species (Johnson and Tienhoven 1980, Silver 1990, Logan and Wingfield 1995). Given the effects of E2 and P on breeding physiology, endocrine cycles in breeding female WBBAs may be expected to appear similar to those of other breeding passerines.

However, some female WBBAs do not breed. These non-breeding females are exposed to equivalent environmental stimuli as are the breeding females, but they do not pair with males or proceed with typical reproductive behaviours, such as brood nest-building, laying, or incubation. Do environmental changes affect the physiology of breeding and non-breeding females equally, or do hormonal patterns more closely correlate with behaviour? How do patterns of secretion and levels of E2 and P compare between breeding and non-breeding WBBA females?

The endocrine system's influence extends beyond sexual physiology and mating behaviour. Hormones are also associated with parental behaviours, and there is an extensive literature that implicates Prl as the "parental" hormone (reviewed in Buntin 1996). Perhaps the most well established relationship is Prl's stimulus of crop milk production in columbiform birds (Silver 1984). Prolactin secretion has also been linked to the induction and maintenance of incubation behaviour and the feeding and brooding of newly hatched young (Silverin and Goldsmith 1984, Vleck *et al.* 1991, 2000). Alloparental behaviour, where an individual provides care (e.g. allofeeding, protection, or brooding) to young that are not their own, is identical in appearance to stereotypical parental behaviours. If parental behaviours are induced by Prl secretion, alloparental behaviours also may be affected by Prl levels. Recent studies examining the relationship between alloparental behaviour and Prl titres in cooperatively breeding birds suggest that Prl may play a role in the expression of alloparental behaviour. In Florida Scrub-Jays, alloparental feeding of nestlings was correlated with elevated Prl titres (Schoech *et al.* 1996b), and in Harris' Hawk, the member of the group that provided the greatest proportion of provisions (adult-plumaged male helper) had elevated Prl levels (Vleck *et al.* 1991). Do WBBAs' cycles of Prl secretion resemble those found in other bird species? Is there any evidence to suggest that Prl mediates alloparental behaviour?

MATERIAL AND METHODS

Data Collection

Data described in this chapter were collected from a population of free-living WBBAs resident in BYSF from June 1996 to July 1999. One hundred and eighty-six WBBAs were individually identified with numbered ABBBS metal bands and coloured leg bands. Breeding status of individuals was determined by observations of behaviour. When appropriate, males and females were assigned a breeding stage: non-breeding, nest building, laying, incubating, with nestling, with fledgling, or with juvenile. I labeled males as "laying" or "incubating" when their mates were in these stages. Males were considered paired with a female if they helped build a brood nest, mate guarded, or fed the incubating female; other group members were never observed partaking in these activities. I assigned breeding stages conservatively; if I was unsure of an individual's status, I recorded its stage as "unknown." In this chapter "nest building" refers to the building of brood nests only, not the building of roost nests (examined in Chapter VII).

Description of the study site and monitoring and capturing techniques are detailed in Chapter II.

Hormonal and Gonadal Analyses

To examine hormone and gonad profiles in relation to season and breeding stage, I collected and analysed blood samples and performed unilateral laparotomies. Blood collection and laparotomy procedures are described in Chapter II. Radioimmunoassays (described below) were used to measure levels of T, E2, Prl, and P in blood samples. Sample sizes for hormone and gonad analyses are reported in Tables 3-1 and 3-2. Capture effort was not evenly distributed across the year.

Table 3-1. Number of WBBAs assessed for hormone levels and gonad condition in each month.

MEASURE

	Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.	Sep.	Oct.	Nov.	Dec.
Testes	8	2	0	9	7	9	7	10	19	21	10	2
Ovary	3	2	0	4	10	4	4	3	12	14	2	0
T ♂	8	2	0	6	9	7	7	10	25	31	12	2
E2 ♀	0	0	0	0	0	0	1	0	7	10	1	1
Prl ♂	2	0	0	2	3	6	3	10	12	27	4	2
Prl ♀	1	0	0	1	1	0	0	2	6	13	1	0
P ♀	5	3	0	3	4	4	6	3	17	23	13	2

Table 3-2 Number of WBBAs assessed for hormone levels and gonad condition in each breeding stage.

BREEDING STAGE

MEASURE

	Non-breeding	Nest Building	Laying	Incubating	With Nestling	With Fledgling	With Juv
Testes	26	4	2	8	4	8	1
Ovary	17	3	2	3	0	6	0
T ♂	25	4	2	8	4	8	2
E2 ♀	0	1	2	4	0	5	0
Prl ♂	8	1	1	8	4	7	1
Prl ♀	2	3	1	6	2	2	0
P ♀	15	7	3	11	3	11	0

Hormone concentrations in plasma samples were measured using radioimmunoassay (RIA) techniques, reviewed below (Chard 1987): First, specific antibodies are raised against the hormone of interest in animals unrelated to the species of interest. An unknown plasma sample is mixed with fixed amounts of radiolabelled hormone and antisera containing the hormone-specific antibodies and allowed to equilibrate. Both the radiolabelled and the unlabelled hormone present in the plasma sample compete for binding sites on the antibodies. All unbound hormone (both radiolabelled and unlabelled) is removed, and the level of radiolabelled hormone bound to the antisera in the plasma sample is determined on a gamma or beta counter, and this is compared to a standard curve generated with standard concentrations of unlabelled hormones in the same manner. The concentration of hormone in the plasma samples is then determined by comparing the level of bound label in plasma samples to the level of bound label in the standard concentrations.

1. Corticosterone

Extraction and assay methods follow procedures described in Ball and Wingfield (1987) and represent methods used in Professor Wingfield's lab.

i. Steroid extraction

Distilled water (dH_2O) was added to 5-20 µl of plasma samples to make a total volume of 20 µl in glass extraction tubes. Recoveries were determined by spiking all samples before extraction with 20 µl of tritiated B (approx. 2000 cpm), obtained from Amersham TRK 406 [1,2,6,7-^{3}H]. Tritiated B (20 µl) was added to three, 7 ml scintillation vials to later verify total recovery counts per minute. Tubes were mixed and allowed to equilibrate overnight at 4°C. Next, I extracted the steroid from each plasma sample in 4 mls redistilled dichloromethane (Sigma-Aldrich, reagent grade) for 2 hrs at room temperature. The dichloromethane fraction was then removed into clean glass tubes using glass disposable pipets. These fractions (containing B) were placed in a water bath at 37°C and dried under nitrogen gas using an Evaporack. PBSG assay buffer (550 µl) was added to each tube, and tubes were vortexed. Duplicate aliquots of this extracted sample were used in the RIA (200 µl). A single aliquot (100 µl) was used for recovery counts; these were aliquoted directly into 7 ml scintillation vials; 4.5 ml scintilation fluid was added and vials counted for 5 min each the following day on a beta counter (LKB Wallace Scintillation Counter).

ii. Radioimmunoassay

In each RIA, in addition to samples, two blank tubes (20 ml distilled water) and two tubes with standard amounts of B (1000 pg) were included. Standard B (Sigma C-2505) was made up in PBSG. The standard curve was set up with serial dilutions ranging from 7.8 to 2000 ng/ml. Antisera was obtained from Endocrine Sciences, Tarzana, CA, Code B21-42. Tritiated B (100 μl) and B antisera (100 μl) were then added to tubes, the tubes were mixed and refrigerated overnight. The following day distilled water was added to total counts tubes and dextran-coated charcoal in PBSG (500 μl) to other tubes. These tubes were allowed to equilibrate for 12 min and then centrufuged for 10 min (2000 rpm) at 4°C to separate bound from free B. The supernatant was decanted into 7 ml scintillation vials, and 4.5 mls of scintillation fluid (Omnifluor made up in toluene) were added. The vials were held in the dark overnight, and then the radioactive counts per minute (cpm) were counted on a beta counter (LKB Wallace Scintillation Counter) for 3 min each.

iii. Calculations of B concentration

By determining the percent of radiolabelled B bound to the antisera in the standard curve, the amount of B in the unknown plasma samples could be calculated. With the aide of a computer program, a cubic spline smoothed curve was fitted to the standards for each assay. This standard curve plotted the log of the standard concentrations versus the percent of bound tritiated B. Corticosterone concentrations in the unknown samples were then estimated from the curve.

The following equation was used to determine the final concentration of B in each sample: [(B value from standard curve X 2.75)/ (% recovery)] X (1/ µl plasma). Recovery counts indicated the proportion of B recovered by the extraction process and were used to adjust the final concentration. Percent recovery was calculated as recovery cpm/(total cpm/5.5). Because original volumes of plasma samples varied (dependent upon available sample volumes), final B concentrations were also adjusted for plasma volume.

iv. Sensitivity and variation

The minimum concentration of B that could be detected in the plasma samples was 7.8 ng/ml. By including two samples with standard amounts of B in each assay, the intra and inter-assay variation could be determined. The inter-assay variation in my assays was 18.8%, and the intra-assay variation was 11.8% for a total of eight assays.

2. Testosterone, Progesterone, and Estradiol

Assay methods for RIA for Double Antibody Assays follow those described by the kit manufacture,r Pantex (Santa Monica, CA, USA).

i. Assay

For T, P, and E2 I used Pantex Direct ^{125}I kits (catalog no. 135, 137, 174M, respectively). These kits contained all the reagents in liquid form needed for each assay, including standards for a standard curve, ^{125}I -labeled steroid, antiseras, and buffer. The range of standards included with the kits were as follows for T, P, and E2, respectively: 0.1-25.6 ng/ml, 0.2-80 ng/ml, and 0.01-3 ng/ml. First, standards and plasma samples (20 µl) were aliquoted into tubes, then radiolabelled steroid was added.

Primary antisera was pipetted into each tube, and tubes were incubated in a water bath at 37°C. A second antisera was then added to precipitate steroid bound to the first antisera, and, after centrifugation, the supernatant was decanted and discarded. Because hormone extraction was not required, estimates of recovery were not necessary. The precipitate, which contained the total hormone bound to antisera, was counted on a gamma counter (LKB Wallace).

Because I could only obtain between 100-150 µl of plasma from individual WBBAs (from approximately 300 µl whole blood) and because reproductive steroid levels were generally rather low (< 5 ng/ml), I modified the kit assay protocol. I made the following modifications to the recommended assay procedure (Pantex, CA.): only one half the recommended dose of all reagents (standards, primary and secondary antisera, and label) was used for each full volume plasma sample. This not only doubled the capacity of the kit, but also amplified the sensitivity of the assay, by effectively doubling my sample volume. This was necessary as it allowed me to use small sample volumes and thus use each blood sample for a number of hormone assays. Prior to attempting half dose assays, I verified the accuracy of the standard curves using half volumes. The curves for half-dose and full-dose assays were similar in shape and level of accuracy. Also, for the T assay I diluted the lowest concentrations standard (0.1 ng/ml) with distilled water in order to include an additional standard (0.05 ng/ml) in the curve.

ii. Calculation of steroid concentrations

Percent binding relative to the total potential binding was determined for each standard. Using a computer program, standard concentrations and percent binding were plotted on a logit-log graph, and a cubic spline smoothed curve was fitted to the points.

Next, percent binding was determined for each plasma sample, and steroid concentrations in the plasma samples were determined from the curve. Because initial plasma volumes were double that of the standards, plasma steroid concentrations read from the fitted lines were halved to calculate final steroid levels in each plasma sample.

iii. Sensitivity and variation

The minimum levels of T, P, and E2 that could be determined (after the modifications to the recommended assay procedure) were 0.025 ng/ml, 0.1 ng/ml, and 0.005 ng/ml, respectively. In my assays intra-assay variation was 10.5%,10.9%, and 4.2% for the same three steroids. Finally, inter-assay variation for T (n=4 assays), P (n=3 assays), and E2 (n=3 assays) was 12.0%, 7.9%, and 7.1%, respectively.

3. Prolactin

i. Assay

This assay was performed by Dr. Jeff Downing at the University of Sydney, Camden NSW and was based on an assay developed for sheep Prl, described by J. Downing (1995). Plasma samples were aliquoted into tubes, 50 µl of antisera containing normal rabbit serum (NRS) was added, tubes were vortexed, and incubated overnight at 4°C. Next, 50 µl of ^{125}I-chicken Prl label (iodinated by Dr. Downing) was added, and the assay was incubated for two days at 4°C. Donkey anti-rabbit serum (DARS) was added, tubes were vortexed and allowed to equilibrate overnight at 4°C. Next, tubes were centrifuged, the supernatant aspirated and the radioactivity in the precipitate determined using a gamma counter (Ortec 770).

ii. Calculation of Prl concentrations

Prolactin concentrations in the unknown samples were determined by comparing the amount of bound label in the unknown samples to the level of bound label in the standards (AFP-10328B obtained from the National Institute of Diabetes and Digestive and Kidney Diseases, Torrance, CA U.S.A.). A software package, built into the gamma counter, performed these calculations by plotting the log of the standard concentrations versus the percent of bound label, fitting a cubic spline smoothed line, and estimating the Prl concentration in each of the unknown samples. Final Prl concentrations were adjusted according to the volume of the initial plasma samples.

iii. Sensitivity and variation

The minimum amount of Prl that could be detected by this assay was 0.07 ng/ml. Prolactin samples were analysed in two assays, and inter-assay variation was 11.8%, and intra-assay variation was 7.7% (Downing pers. comm.).

Data Analysis

Analysis of Variance and Student's t-tests were applied to normally distributed data. Normality was examined visually using normal probability plots. General statistics (e.g. mean and standard error) were used to describe various hormone, gonad, and morphometric measures. Correlations between continuous variables were examined, and Pearson product-moment correlation coefficients are reported. Fisher's exact test was used to examine contingency tables with small sample sizes or missing cells. These statistical analyses were performed using SYSTAT™ 7.0 (1997).

As the field study was only a portion of my entire study, limited time was allocated to field data collection. As a result sample sizes for some variables are smaller than desirable for some statistical analyses. Because of such limitations in my data set (e.g. I did not have large sample sizes for each hormone examined in multiple social groups in all months of all years), I was forced to restrict data sets or pool data. As I was most interested in seasonal changes in gonads and hormone levels, I typically pooled data from multiple social groups and multiple years. Before pooling this data I first examined the validity of doing so. Because there is a great deal of evidence that gonad condition and hormone levels fluctuate seasonally (e.g. Dittami 1986, Dufty and Wingfield 1986a,b, Chandola-Salklani *et al.* 1990), I restricted these analyses to the smallest number of concurrent months in which I had sufficient samples; as I spent the most time in the field during peak breeding months, these data were typically drawn from birds in breeding condition. Once satisfied that data could be pooled across years and/or social groups, I proceeded with analyses examining temporal changes in gonad condition and reproductive hormones. While these attempts were made to reduce potential biases, this methodology may have introduced some bias.

RESULTS

Testes

Testis condition varied markedly among males, from fully regressed to fully enlarged, with calculated volumes ranging from 1.9 to 35.5 mm^3. I investigated patterns of variation in testes volume across years, months, breeding stages, and WBBA social groups. To establish the validity of pooling data across three years (June 1996 to July 1999), I first tested for an effect of year on testes volume. To minimise the effect of season, I restricted my data set to two consecutive months in which I had multiple samples in each of three years, September and October 1996 through 1998 (see methods). There was no significant difference in testes size among years ($F_{2,37}$=2.601, P=0.088), but this relatively low P value warranted further investigation. Testes volume tended to increase from 1996 to 1997 to 1998. However, I suspect this trend may have been due to more males being caught while tending young in 1996 and 1997 than in 1998. When I removed those males tending young from the analysis, an ANOVA indicated no significant differences among years ($F_{2,28}$=1.321, P=0.283). With this higher P value, I was comfortable pooling data from all years for subsequent analyses.

Next, I examined intergroup differences and the validity of pooling data across groups. To minimise the possible influence of season and to maximise the amount of data available, I restricted my analyses to four consecutive months (August-November), and I chose five groups in which I had data from more than five individuals per group. There was no significant difference in testes volume among groups ($F_{4,33}$ =0.574, P=0.683).

When I combined data from all groups in all years, a seasonal pattern in testes volume emerged; testes were largest July through February and smallest April through

June (Fig. 3-1). The period of enlarged testes coincided with the breeding season and partially regressed testes with the non-breeding season. An ANOVA verified significant differences among months ($F_{10,93}$=5.910, P<0.001), and Tukey's pairwise comparisons indicated the following differences (P<0.05): testes measured in May were smaller than in August through October and testes measured in April and June were smaller than in August through November.

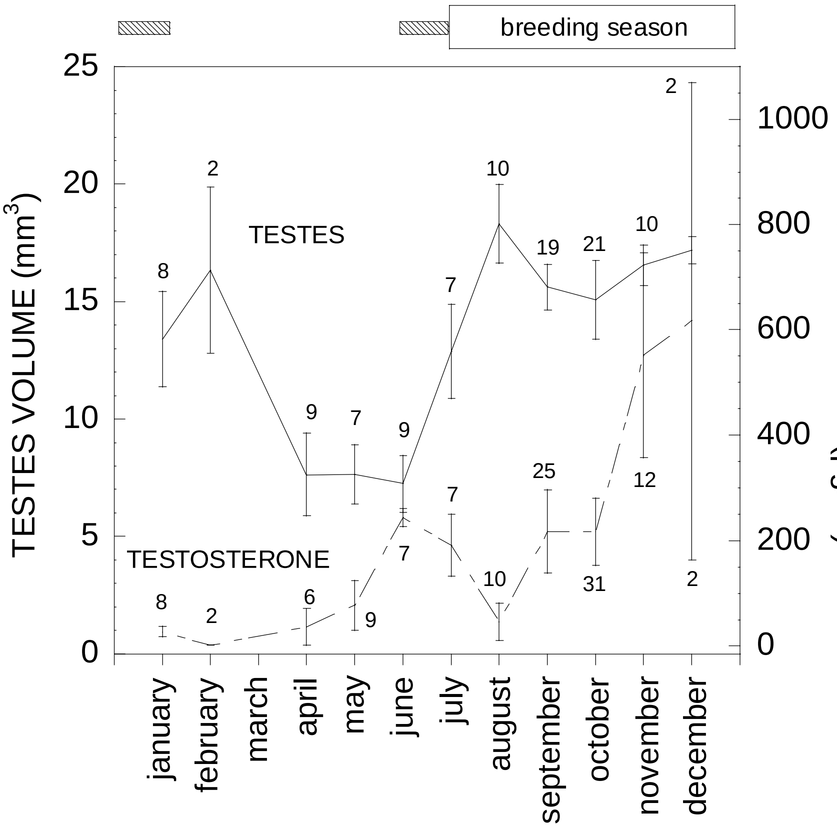


Figure 3-1. Annual variation in testes volume and plasma T in male WBBAs. Mean testes volume (solid line) and plasma testosterone (dotted line) in each month, except March. Points indicate means, and bars represent one standard error. Hatched rectangles indicate the margins of the breeding season, when only a few WBBAs bred. Numbers inside graph indicate sample sizes.

Although testes were typically small in non-breeding months, they were only partially regressed in many cases. Based on an observation of a juvenile male (recognized by his yellow gape) with a testis volume of 3.70 mm^3 and on a thorough examination of my data, I consider testes volumes less then 5 mm^3 to be "fully regressed." Of 25 males whose testes were measured in April, May, or June, only nine had fully regressed testes (Fig. 3-2). As mean volume of fully recrudesced testes during the height of the breeding season (September, October, and November) was 16.65 ± 0.63 mm^3 , fully regressed testes were approximately 30% of their fully recrudesced size, and average testes size during the non-breeding months of April, May, and June (7.49±0.81) was approximately 45% of their fully recrudesced size.

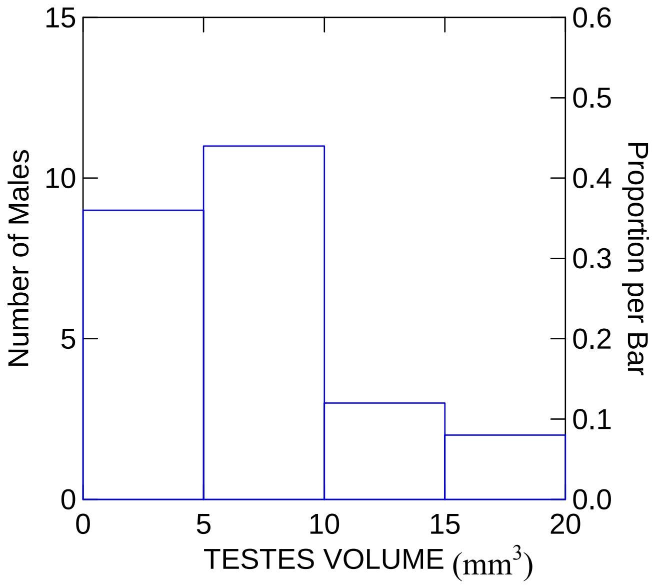


Figure 3-2. Distribution of testes volumes in male WBBAs in non-breeding months (April, May, and June).

Testes volume also varied in relation to breeding stage; testes were largest while the breeding pair was building a nest and the female was laying her eggs, then testes declined in size with each sequential breeding stage (Fig. 3-3). A striking reversal to this pattern was the large testes of a single male accompanying a juvenile (Fig. 3-3). When comparing breeding stages between nest building and fledgling care, testes volume varied significantly with breeding stage ($F_{4,21}$=7.765, P<0.001). Tukey's pairwise comparisons indicate a significant difference (P<0.05) between birds caught with fledgling young and those males involved in nest building, with laying or incubating females. Some adults caught with fledglings or juveniles were not the young birds' parents, but were included in the breeding stage categories "with fledgling" and "with juvenile."

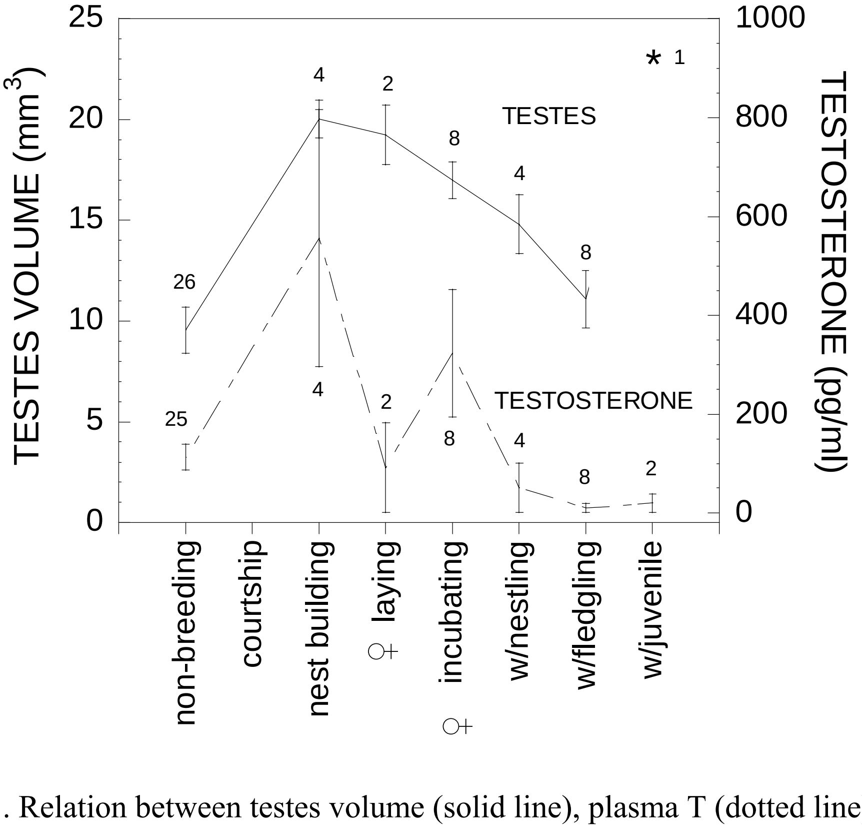


Figure 3-3. Relation between testes volume (solid line), plasma T (dotted line), and breeding stage in male WBBAs. Points indicate means, and bars represent one standard

error. Asterisk (*) indicates testes volume of a single male caught accompanying a juvenile. Numbers inside graph indicate sample sizes.

I also tested whether overall size of the male was associated with testis size. To investigate this I tested for correlations between testes volume and morphometric measurements during the height of the breeding season (September, October, and November) and during the non-breeding season (April, May, and June). At the height of the breeding season, testes volume was not significantly correlated with tarsus length (ρ=-0.089, P=0.543) or bill length (ρ=0.256, P=0.080), but it was significantly correlated with wing length (ρ=0.431, P=0.002) and body mass (ρ=0.338, P=0.016). During the non-breeding season, testes volume was not significantly correlated with any morphometric measure (tarsus length: ρ=0.087, P=0.679, wing length: ρ=0.334, P=0.103, body mass: ρ=0.324, P=0.115, bill length: ρ=0.387, P=0.062.

Testosterone

Variation in male WBBAs' T levels was examined in relation to a number of factors. First, I examined whether it was valid to pool data across years. As I am missing data from some months of each year and because of suspected seasonal variation, this presents a potential bias. To compensate for this, I chose to restrict my analysis of inter-year variation to only September and October; during each of these months, I have at least four T samples from males in each of the three years of my study. An ANOVA indicated no significant difference in T levels in September and October among years ($F_{,50}$=1.282, P=0.286). From this finding, I inferred that differences among years were negligible, and subsequent analyses pooled data across years.

Next, I examined the variation in T levels in relation to social groupings. For this analysis I chose two breeding months (September and October) and five groups with sufficiently large sample sizes (n>5 birds). I found that T did not vary among groups ($F_{4,35}$=0.526, P=0.717), thus subsequent analyses pooled data from all social groups.

Examination of seasonal variation in T uncovered a pattern (Fig. 3-1). There were two annual T peaks, a small peak in June and a much higher one in December. Notably, the first rise in T occurred prior to full recrudescence of the testes and prior to the winter solstice; all June T samples were taken at least 5 days before the winter solstice. Testosterone levels seemed to decrease in August when testes volume reached a plateau. Month-to-month variability in T was statistically significant ($F_{10,104}$=2.312, P=0.017); Tukey's posthoc comparisons indicated differences (P<0.05) between November and January and between November and February. The large variation in T levels in December was likely a factor of a small sample size; only two males were caught in December. Both males were moulting, and both were members of groups including an incubating female. Though their testes volumes differed by only 1 mm^3, their T levels were remarkably different: a particularly small male, possibly a young bird, had a T titre of 165 pg/ml, while an average sized male, likely an adult, had a T titre of 1070 pg/ml.

Plasma T levels also varied with breeding stage, but differences were only marginally significant ($F_{5,22}$=2.673, P=0.049). Tukey's pairwise comparisons indicated a significant difference in T only between males in nest building and fledgling stages (P=0.049). Testosterone levels were highest during nest building, declined abruptly in two males with laying females, and then rose while the female incubated; testosterone levels remained low while males cared for young (Fig. 3-3).

Ovaries

As ovary condition did not differ significantly among years (Fisher's exact test, P=0.244), data were pooled across years. Data were also pooled across social groups, as there were no significant differences among groups (Fisher's exact test, P=0.461).

Although some females showed potential for ovarian development (scores of >1.5) year-round, active ovaries with yolk deposition and follicular hierarchies (scores of >2.5) were only observed between August and November (Fig. 3-4). Furthermore, across all months the level of ovarian development ranged widely (Fig. 3-4). Nevertheless, ovary condition varied significantly across months (Fisher's exact test, P=0.001) and between the breeding and non-breeding season, demarcated as July-December and January-June, respectively (Fisher's exact test, P<0.001). Keep in mind that ovarian condition of some breeding females was not examined surgically when behaviours (e.g. nest building or incubation) could accurately be used to ascertain breeding stage.

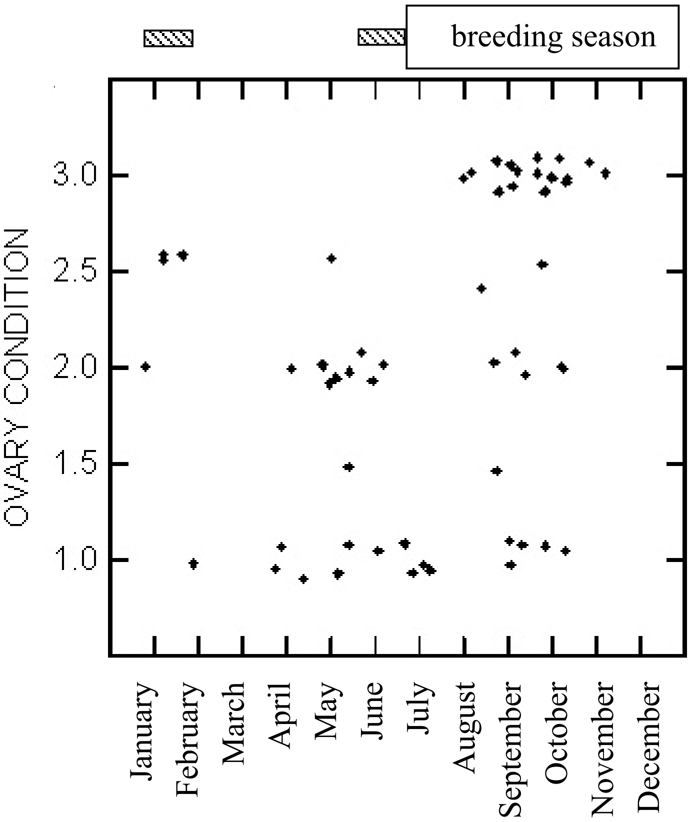


Figure 3-4. Annual variation in ovary condition. 1=completely regressed, flat, gray ovary, 1.5=ovary bumpy, but no distinct ova, 2=distinct ova, but no follicular hierarchy, 2.5= some follicles differed in size, but only marginally, 3= ova larger than 1 mm and follicular hierarchy evident. Overlapping points are randomly offset to indicate number of individuals. Hatched rectangle indicate the margins of the breeding season, when only a few WBBAs bred.

As expected, there was a significant association between ovary condition and breeding stage (Fisher's exact test, P=0.001). Ovaries were consistently well developed (score of 3) in all stages between and including nest building and incubation. There was considerable variation in the condition of ovaries in non-breeding females and in females with fledglings (Fig. 3-5). When I caught adults females with fledglings, it was unclear which female was the mother of the young, and thus I could not always determine which female had bred.

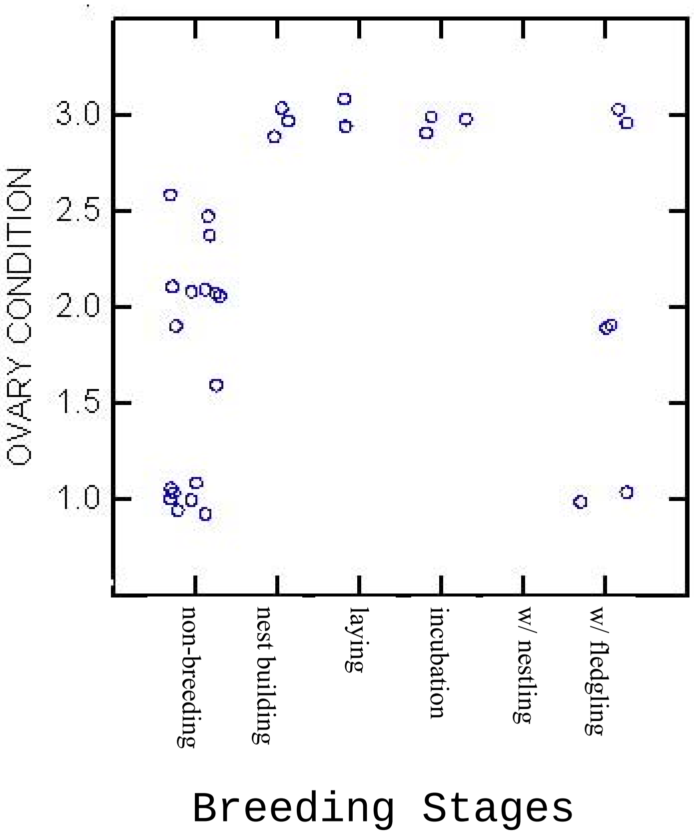


Figure 3-5. Relation between ovary condition and breeding stage. Ovary condition labeled as above (Fig. 3-4). Overlapping points were randomly offset to indicate number of individuals.

Most females with well-developed ovaries (those with follicular hierarchy, scores of 2.5 and 3) had brood patches, while those with regressed ovaries (those with no distinct ova, scores of 1 and 1.5) typically had no brood patch (Fig. 3-6). Roughly half of females with ovaries in an intermediate state of development had brood patches, while the other half did not (Fig. 3-6). In almost all months that WBBAs were sampled, some females had fully feathered breasts, while others had brood patches (Table 3-3).

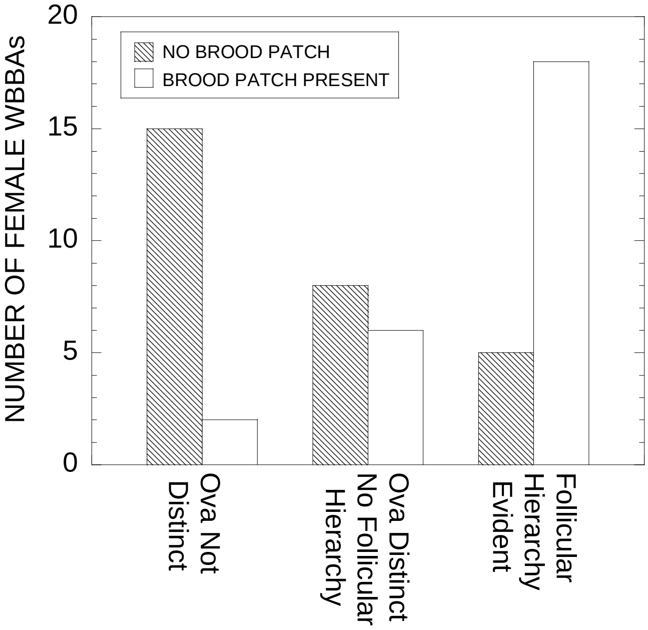


Figure 3-6. Comparison between presence or absence of brood patches in female WBBAs and the condition of the ovary.

Table 3-3. Number of females caught each month with a brood patch and without a brood patch.

	Jan.	Feb.	Mar.	Apr.	May	Jun.	Jul.	Aug.	Sept.	Oct.	Nov.	Dec.
No Brood Patch	3	1	0	3	5	3	4	1	6	2	0	0
Brood Patch Present	1	2	0	1	2	0	2	2	10	14	7	2

I also examined ovary condition in relation to body size, mass, and fat level by comparing morphometric parameters between females with developed (ovary condition ≥ 2) versus undeveloped (ovary condition=1) ovaries. Mean body mass of females with completely regressed ovaries was significantly less than that of females with more developed ovaries ($t_{71}=-3.689$, $P<0.001$); body mass of females with regressed ovaries was 37.2 ± 2.0 g (mean ± S.D.) and that of females with more developed ovaries was 39.8 ± 2.6 g (mean ± S.D.).

There were no significant differences in wing ($t_{71}=-0.528$, $P=0.599$), tarsus ($t_{71}=-0.054$, $P=0.957$), or bill length ($t_{71}=0.079$, $P=0.938$) between females with regressed and developed ovaries. Furcular fat levels did not differ between females with regressed and active ovaries (Fisher exact test, $P=0.085$). Abdominal fat levels were not compared; accurate assignment of abdominal fat classes was often impossible in breeding females, as edematous brood patches obscured abdominal features.

Estradiol

Of 20 females sampled, only three had detectable (>5 pg/ml) E2 levels. The highest levels were found in a laying female (67.6 pg/ml), and an incubating female measured 12.0 pg/ml. However, another laying and three other incubating birds had undetectable E2 levels.

Progesterone

Plasma levels of P were examined in relation to a number of factors. First, I considered annual variation in P. Because I did not collect samples from every month in each year, I restricted my analysis to September and October 1996 to 1998, as I have at least four samples for each of these month during this three year period. There were no significant differences in P levels among years ($F_{2,34}$=0.452, P=0.640). Subsequent analyses pool P data across years. Again, data were restricted to September and October and to three social groups in order to examine variation among social groups. No significant differences were found among social groups ($F_{2,16}$=3.428, P=0.058), and data were pooled for further analyses.

Progesterone levels remained low from December through April, but the pattern was irregular in other months (Fig. 3-7). There were no significant differences in P levels among months ($F_{10,57}$=0.623, P=0.788), nor was there a significant difference in P between non-breeding months (January-June) and breeding months (July-December) (t=-1.2184, P=0.227). From July through September, when most females were breeding, there seemed to be an increased variability in P levels over other months. The high mean in September is partially due to an unusually high P titre (6398 pg/ml) recorded in a single female; this female will be discussed in relation to breeding stage below.

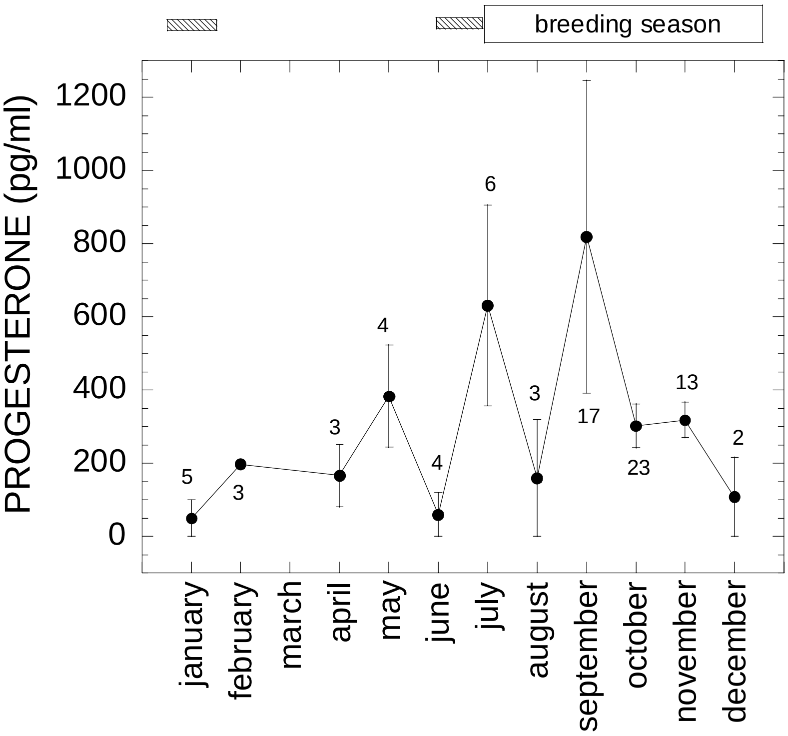


Figure 3-7. Patterns of plasma P in female WBBAs. Bars represent one standard error. Hatched rectangles indicate the margins of the breeding season, when only a few WBBAs bred. Numbers inside graph indicate sample sizes.

Progesterone levels varied significantly among breeding stages ($F_{5,37}$=3.505, P=0.011) and were highest (2308 ± 2044 pg/ml (mean ± S.E.)) in laying females (Tukey's pairwise comparison, P<0.031). However, the high mean P in laying females was heavily weighted by the high value from a single female with P of 6398 pg/ml; this female had one egg in the nest and was caught and bled when flushed from her nest in the early morning, perhaps just prior to laying her second egg. While there was no doubt that this female was in the laying stage, perhaps she was a special case, as she may have been caught immediately prior to laying. When this unusually high P value was removed from the analysis, mean P in laying females dropped from 2308 ± 2044 pg/ml (mean ± S.E.) to 264 ± 16 pg/ml (mean ± S.E.), and an ANOVA indicated no significant variation among stages ($F_{5,36}$=1.014, P=0.424). Nevertheless, P levels tended to be highest in females building nests, laying, and those caught with fledglings (Fig. 3-8).

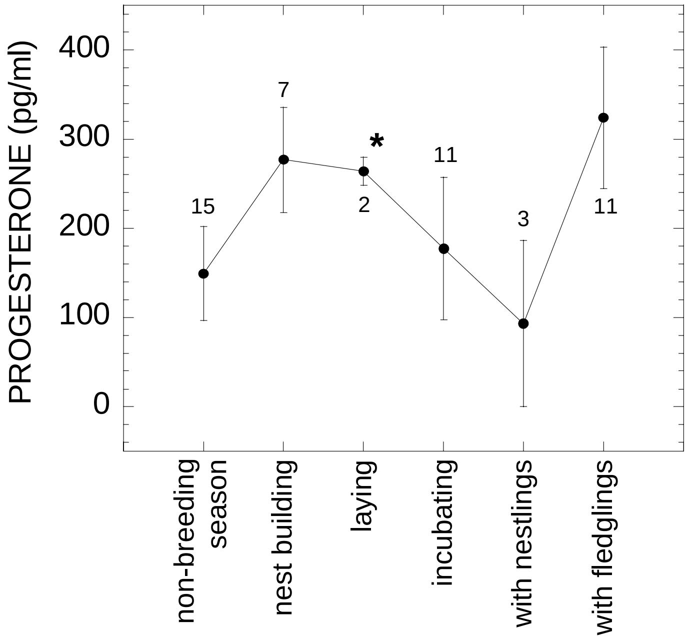


Figure 3-8. Relation between female WBBAs' P levels and breeding stage. Asterisk (*) indicates adjusted mean; a laying female with P measuring 6398 pg/ml was removed from this analysis (see text). Points indicate means, and bars represent one standard error. Numbers inside graph indicate sample sizes.

Not all females bred during the breeding season (Chapter II); these females showed no behavioural signs of pairing to males, had no brood patch (breast completely feathered), and had completely regressed ovaries, suggesting that they were not (and had not been) breeding. Nonetheless, some of these non-breeding females had surprisingly high P levels. To more closely explore the association between breeding condition and P levels, I examined the relationship between ovarian condition and P titres during breeding months from July through December. Progesterone levels were highest in females with regressed ovaries and remained relatively constant in females with ovaries in other conditions (Fig. 3-9). This trend was marginally significant ($F_{4,39}$=2.608, P=0.050).

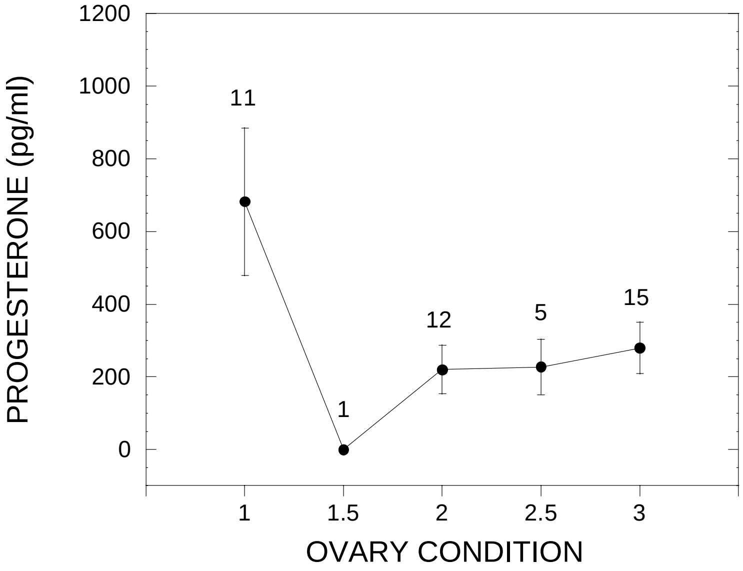


Figure 3-9. Mean P levels relative to ovary condition. Ovary condition labeled as in Fig. 3-4. Points indicate means, and bars represent one standard error. Numbers inside graph indicate sample sizes.

Prolactin

First, I explored the validity of pooling Prl data across years, and I restricted my analyses to September and October for males and to October for females. In these months, I have multiple samples in each of three years from each gender. Plasma Prl levels did not vary significantly with year for either males ($F_{2,36}$=1.883, P=0.167) or females ($F_{2,10}$=1.641, P=0.242). Subsequent analyses pool Prl data across years. Next, I explored variation in Prl titres that might be attributed to social group. Small sample sizes prohibited analysis in females, but I examined male Prl levels in four social groups in September and October. There was no significant difference among social groups ($F_{3,23}$=0.551, P=0.653), and data were pooled for further analyses.

Seasonal variation in plasma Prl levels were examined in WBBAs (Fig. 3-10). In both males and females, Prl tended to be higher in breeding months than non-breeding months; this difference reached statistical significance in males (t_{69}=-2.958, P=0.004), but too few females' Prl levels were measured in winter months for a meaningful test. In males Prl levels tended to be highest in two birds caught in December and lowest in three caught in May. Both males caught in December had incubating mates, and males caught in May were not breeding. Statistically significant differences in males' Prl levels were found among months ($F_{9,61}$=2.691, P=0.011), and Tukey's pairwise comparisons indicated a significant difference between May and December (P=0.010) and marginally non-significant differences between May and September (P=0.077), January and December (P=0.080), and June and December (P=0.069). In females sample sizes in the first half of the year were small, but seasonal changes in Prl levels were evident (Fig. 3-10). Prolactin levels were low in females caught in April and May, moderately high in August, September, and October, and tended to be highest in females caught in November and January. Perhaps due to small sample sizes in some months, there were no statistically significant differences in females' Prl levels among months ($F_{6,18}$=1.815, P=0.153).

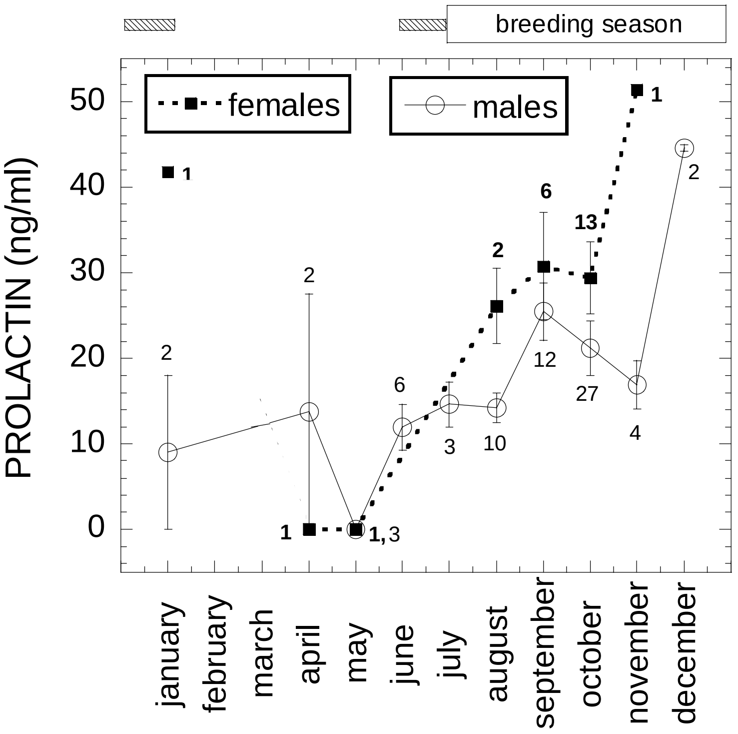


Figure 3-10. Annual patterns of Prl titres in male and female WBBAs. Points represent means, and bars indicate standard errors. Hatched rectangles indicate the margins of the breeding season, when only a few WBBAs bred. Numbers inside graph indicate sample sizes; darker numbers refer to females and lighter to males.

Prolactin levels were also examined in relation to breeding stage (Fig. 3-11). In males Prl levels did not vary significantly among stages ($F_{6,23}$=1.223, P=0.331). However, Prl titres seemed to be higher in males paired to laying and incubating mates and when accompanying fledglings and juveniles than in males building nests and attending nestlings. In females Prl levels varied significantly with stage ($F_{5,10}$=3.932, P=0.031), and Tukey's pairwise comparisons identified two marginally non-significant differences between non-breeding and nest building females (P=0.059) and between non-breeding and incubating females (P=0.052). Prolactin levels were lowest in females during the non-breeding period and tended to remain high from nest building through nestling care, though a single laying female had low levels. Two females caught with fledglings tended to have lower Prl levels than during other breeding stages.

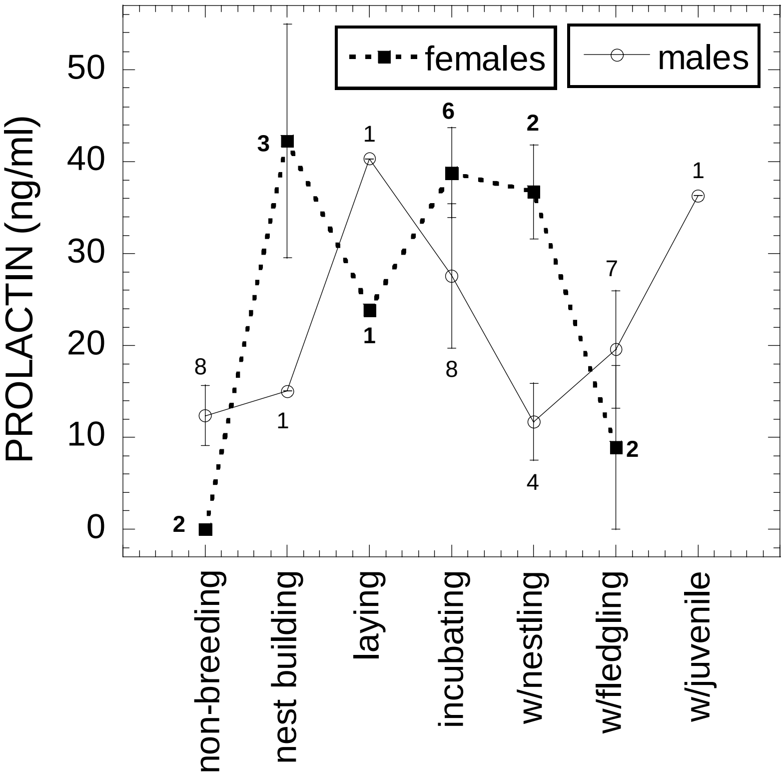


Figure 3-11. Relation between plasma Prl and breeding stage in male and female WBBAs. Points indicate means, and bars represent standard error. Numbers inside graph indicate sample sizes; darker numbers refer to females and lighter to males.

DISCUSSION

Gonadal Cycles

In WBBAs, fully recrudesced testes and well-developed ovaries were observed in most months of the year, unlike in many species in which gonads are enlarged for only a few months of the year (e.g. Song Sparrows: Wingfield 1984, Starlings: Dawson and Goldsmith 1982, White-crowned Sparrows *Zonotrichia leucophrys*: Wingfield and Farner 1978a). These findings are consistent with my observation that WBBAs can breed in at least eight months of the year. In species that can breed for much of the year, gonadal recrudescence may be initiated in the absence of conditions favourable for breeding, and mature gonads may be maintained for long periods, allowing mating to take place as soon as conditions become appropriate (Crews and Moore 1986). For instance, the Zebra Finch *Taeniopygia guttata* is an opportunistic breeder, that initiates breeding in response to conditions favourable for grass germination (Davies 1977, Zann 1996); Zebra Finch males can maintain spermatogenically active testes year-round, and females can perpetually maintain ovarian follicles in an advanced resting state (Crews and Moore 1986). A comparison of the approximate percent decrease in testes size reveals a striking similarity between the Zebra Finch and the WBBA. In the WBBA, testes receded to approximately 45% of their fully recrudesced size, and Zebra Finch testes receded to approximately 50% of their fully recrudesced size (Davies 1977). In contrast, in more seasonally breeding species, including the Rufous Whistler (studied concurrently in the same forest as the WBBA) and the tropical Baya Weaver *Ploceus philippinus*, regressed testes measured approximately 2% and 8% of their fully recrudesced size (McDonald *et al.* 2001b, Chandola-Saklani *et al.* 1990, respectively). Presumably, testes maintained in a partially regressed state would be able to initiate spermatogenesis more rapidly than fully regressed testes. This may allow opportunistically breeding species to maintain a moderate to high level of reproductive readiness year-round, whereas seasonally breeding species do not.

Male WBBAs seemed to initiate gonadal recrudescence in the absence of widespread breeding opportunities. Even though only a few WBBAs initiated nesting as early as July, most males' testes began to enlarge in this month and had reached maximum size by August. Most males had fully recrudesced gonads well before the peak of the breeding season, and they maintained mature gonads beyond the end of the breeding season. The only months WBBAs' testes were partially regressed coincided with the months approaching the winter solstice, when temperature declined and day length shortened. In many avian species living at mid to high latitudes, and even some tropical species, annual cycles of day length affect gonadal maturation (Chandola-Saklani 1990, Wingfield 1990, Hau *et al.* 1998, Gwinner and Scheuerlein 1999). While I speculated that breeding could take place year-round in the WBBA (Chapter IIa), April, May, and June may be the months least suitable for breeding. While I have no data on the energetic costs of maintaining large gonads, there may be costs associated with constantly maintaining fully recrudesced testes. In the winter months, when environmental conditions are typically unfavourable for breeding, energy may be better appropriated elsewhere than for maintaining fully recrudesced testes.

Testes volume varied among males, and in breeding months there was a positive relationship between wing length and testis size and between body mass and testis size. As there were more males than females in the BYSF WBBA population, some males could not breed. Perhaps large males obtained the greatest proportion of matings, initiated gonadal recrudescence first, and maintained large gonads for longer than small males. Some studies have found that testes size is correlated with social status in cooperatively breeding birds (Ambrose 1985, Wingfield *et al.* 1991), and King (1980) found that testes size was positively correlated with age in Grey-crowned Babblers. Perhaps larger WBBAs (with large testes) held a higher social status within the group and/or were older than smaller males. During non-breeding months, when testes were partially regressed, large males did not maintain larger testes than small males. Testes size in non-breeding months may be factor of when a given male last bred.

Among females, ovarian development was highly variable. In all months of the year, ovarian development ranged from completely regressed to highly developed. In every month sampled, some females had well-developed ovaries while others had completely regressed ovaries. Furthermore, in most months of the year, some females had a brood patch (suggesting they were presently or had been recently incubating), while others had fully feathered breasts. The cooperatively breeding White-browed Sparrow Weaver showed a similar pattern, with some fraction of females in the population having ovarian follicles at an advanced level of development at all times of the year (Wingfield *et al.* 1991). In the WBBA and White-browed Sparrow Weaver, some individuals may be able to initiate final follicular maturation and egg-laying at any time of the year. The observation of WBBA females with brood patches in all months except March (when no WBBAs were captured) and June supports this assertions. At the same time, some females were not ready to breed, even at the peak of the breeding season. In many cooperatively breeding species, it is common for some group members to act as non-breeding helpers (e.g. Acorn Woodpecker: Koenig 1981, Florida Scrub-Jay: Schoech *et al.* 1996a, Splendid Fairy-wren: Russell and Rowley 1988). Furthermore, non-breeding helpers have been noted in WBBA populations in other locations (Chandler 1920, Cale 2003b). In the BYSF WBBA population, several females were able to breed concurrently in each group, and I did not observe helpers at the nest (Chpt II). However, the observation that some females had regressed ovaries and fully feathered breasts during the breeding season, suggests that there was a non-breeding component of some groups. These non-breeding group members may have been utilised as helpers under some conditions (although not observed during my study) or as chaperones for fledglings (see Chpt II).

Early and late in the breeding season, many females were not breeding. Although age could not be determined morphologically, as it can by eye colour in Grey-crowned Babblers (Counsilmann and King 1977), and social rank was not examined in wild WBBAs, both these factors may influence a females' propensity to breed. Older WBBA females or those of high social rank may maintain breeding readiness year-round, while young birds or those of moderate or low social position either may not breed or only attempt broods in some months of the year. This hypothesis is supported by my finding that females with regressed ovaries weighed less (but were similar in other morphological measures) than those with more developed ovaries. Younger birds or those with low priority of access to resources (low social rank) may have weighed less than older birds or those with high priority to food resources (high social rank). However, this difference in body mass between these two groups of females may also have been amplified by the greater mass of the developed than the regressed ovary and/or an increase in body mass prior to breeding. Such a scenario was recorded in female White-crowned Sparrows *Z. l. gambelii*, in which body mass increased during the final maturation phase of ovarian follicles and was maximal at ovulation and oviposition (Wingfield and Farner 1978a).

As expected, gonadal cycles were correlated with breeding stage. Testes volume peaked when males were nest building and tended to decrease with each subsequent breeding stage until chicks fledged; however, differences were only significant between males in pre-hatching breeding stages and males with fledglings. For the most part, testes tended to be largest at the time just before and during the period of female fertility. Once the female laid her clutch of eggs, mean testes volume tended to be lower. Others have found that testes weight decreased throughout breeding cycles (Wingfield and Farner 1978a), and it has been suggested that Prl, which was high during incubation in the WBBA, as in many other species, has an antigonadal effect (Dawson and Goldsmith 1982). A single WBBA male caught with a juvenile was the exception to the common pattern; this male had exceptionally large testes and high Prl. While this male seemed to be part of a large group protecting the young bird, he may also have been pursuing another breeding opportunity; if this were the case, his testes were within the normal range of males caught at the beginning of their breeding cycle.

All females caught between nest building and nestling stages had ovaries in an advanced state of development. During the nest building and laying stages, ovarian follicles were large as females prepared to lay eggs. Size of ovarian follicles declined after laying, but only slightly. This pattern was unlike that found in White-crowned Sparrow females, where ovarian follicles decrease in size after incubation begins (Wingfield and Farner 1978a). Because of frequent renesting during their protracted breeding season, it may be efficient for WBBAs to maintain an advanced state of ovarian readiness. Females caught with fledglings were found with ovaries in various stages of development; however, it was unclear which of these females mothered the young. Presumably, females with completely regressed ovaries were non-breeding females, those with gonads in an intermediate stage of development may not have had an immediate breeding opportunity, while females with fully developed ovaries may have been the mother of the young and/or about to initiate a nesting attempt.

Testosterone

In birds T is necessary for spermatogenesis and the expression of some breeding behaviours, such as territory establishment and mate acquisition (Vleck and Brown 1999). However, a number of deleterious effects have been associated with chronically elevated T levels: decreased survival, decreased reproductive success, decreased tendency to provide parental care, immunosuppression, elevated energy turnover, disruption of social relationships, and increased risk of injury (Wingfield *et al.* 1997, 1999, Vleck and Brown 1999). Testosterone levels may vary temporally as a result of these costs and benefits (Vleck and Brown 1999, Jacobs and Wingfield 2000).

In the WBBA, T levels were basal from January through April, when birds did not initiate breeding attempts. As in other species (e.g. Song Sparrow: Wingfield 1984, Wingfield and Goldsmith 1990), WBBAs' T levels increased prior to testicular recrudescence. Testosterone levels tended to rise in May and June, when courtship and the first nesting attempts were initiated. The elevated T titres in June were not a post-solstice effect, as all June T samples were collected before the winter solstice.

Pair formation has been correlated with increased T levels in a number of species (Pied Flycatcher: Silverin and Wingfield 1982, Pigeons *Columba livia*: Haase *et al.* 1976, White-crowned Sparrow: Moore and Wingfield 1980). Even though few WBBA pairs began nesting as early as June, all males in the groups may have responded with increased T secretion in response to the first receptive females. There is also ample evidence that social challenge may elicit increases in T secretion (see Wingfield *et al.* 1990). As pairs began forming at the beginning of WBBAs' breeding season, intragroup social relationships may have been challenged. The elevated T levels in May and June may reflect these social challenges associated with pair formation within the group.

After an initial increase in T levels in May and June, T levels tended to decrease in July and August, followed by an increase in subsequent months (Fig. 3-1). A similar pattern was seen in Brown-headed Cowbirds *Molothrus ater* and Song Sparrows: T increased during the period when males competed for access to females or while territories were being established, then decreased, and later increased when females began to lay (Dufty and Wingfield 1986, Wingfield 1984, respectively). Unfortunately, I collected little behavioural data on the males I caught in July and August, and I can not be certain of their breeding status. However, a similar scenario to that reported in the Brown-headed Cowbird and Song Sparrow may help explain the small T peak in June followed by a decline. The initial May and June increase in T levels may have corresponded to the period of pairing, but then pairs may have postponed breeding for a couple months, resulting in declining T levels in July and August. Testosterone levels may have begun to rise again in the population when pairs began to build nests, which was accompanied by a surge in T titres. As a high proportion of the WBBA population bred between September and November, T levels were high in these months. Fewer WBBAs bred in December, but the mean T level remained high; however, this mean was biased as a result of the small sample (2 birds) and the fact that one of these was unusually high (1070 pg/ml).

While there does appear to be a general seasonal pattern in T secretion, WBBAs' T levels correspond more closely to breeding stage than to calendar date, as has been shown for a number of other species (Wingfield 1983, Wingfield and Moore 1987, Wingfield *et al.* 1990). Levels tended to be highest when birds were building brood nests. This coincided with a period of both mate guarding and the period of female fertility. Others studies have also found that T levels were high during nest building, and some have suggested that T helps regulate nest building behaviour, mate guarding behaviour, and spermatogenesis (Wingfield *et al.* 1989, Logan and Carlin 1991, Seiler *et al.* 1992, Saino and Moller 1995).

When their mates were laying eggs, T levels were low in 2 WBBA males. Testosterone levels were also shown to decrease from peak levels in male White-crowned Sparrows, Florida Scrub Jays, Bengalese Finch *Lonchura striata*, and Pied Flycatchers when their mates were laying eggs (Silverin and Wingfield 1982, Morton and Allan 1990, Schoech *et al.* 1991, Seiler *et al.* 1992). Morton and Allan (1990) suggested that T levels in male White-crowned Sparrows mated to laying females decreased from peak levels because the females solicited increasingly fewer copulations. As both WBBA males were caught after the first egg had been laid, they may have already fertilised their mates second (and last) egg, and she presumably solicited few, if any, copulations. While I suspect that many males sought extra-pair copulations, there may have been a brief lull between the laying of his mate's eggs and his search for another receptive female.

While female WBBAs incubated their clutches, their mate's T levels tended to rise again. In monogamous species with short breeding seasons, a male's breeding opportunities end when most females in the population, including his mate, have laid their clutch of eggs. In such species, males' T levels are typically low while females are incubating (e.g. Barn Swallows *Hirundo rustica*: Saino and Moller 1995, Song Sparrows: Wingfield 1984, White-crowned Sparrows: Wingfield and Farner 1978). However, in plurally breeding species with long breeding seasons, breeding opportunities may exist for males after their mates begin incubating (Vleck and Brown 1999). Even though WBBAs formed apparently monogamous pairs, all males in a group were exposed to several breeding females throughout the protracted breeding season. Elevated T levels in WBBA males during the period of their mates incubation may reflect these circumstances.

Testosterone levels were low when WBBA males were caring for young. There is considerable evidence that elevated T is incompatible with parental behaviour in birds. For instance, nestling care was correlated with low T levels in male Yellow-headed Blackbirds *Xanthocephalus xanthocephalus*, Harris' Hawks, and Mexican Jays (Beletsky *et al.* 1990, Mays *et al.* 1991, Vleck and Brown 1999, respectively); treatment with T resulted in significantly lower rates of incubation in male Rufous Whistlers (McDonald *et al*. 2001a); nestling feeding rates were drastically reduced by T-treatment in male House Sparrows, Dark-eyed Juncos, and Pied Flycatchers (Hegner and Wingfield 1987a, Ketterson *et al.* 1992, Silverin 1980, respectively); nestling feeding rates rose in male House Sparrows following treatment with flutamide, an antiandrogen (Hegner and Wingfield 1987a). The low T levels in WBBAs caring for young are consistent with these findings. Of the four males caught attending nests with nestlings (at two different nests), only the single reputed father had measurable T levels (200 pg/ml); the other 3 males were not paired to the breeding females. Perhaps this difference was more related to paternity and/or social status within the group than to breeding stage.

Although WBBAs' patterns of T secretions appeared similar to those found in many other species, there was a striking difference. In WBBAs that can breed for at least eight months of the year, breeding T levels were substantially lower than those found in many other species. In fact, peak T levels in male WBBAs were almost an order of magnitude lower than those typically found in many north temperate species (e.g. Red-winged Blackbird: Johnson 1998, Song Sparrow: Wingfield 1984, White-crowned Sparrow: Wingfield and Farner 1978).

Similarly low peak T levels are typical in male Bell Miners (approx. 550 ± 70 pg/ml) and White-browed Sparrow Weaver (approx. 330 ± 90 pg/ml), both cooperative breeders with a protracted breeding season, similar in length to the WBBAs' (Poiani and Fletcher 1994, Wingfield *et al.* 1991, respectively). One possible reason for maintaining low background or permissive levels of T in these species may be related to the duration of the breeding season and the potential negative effects of having chronically high plasma T. If T levels were maintained at high levels for extended periods, deleterious effects, such as immunosuppression or increased risk of injury, could eventuate (reviewed in Vleck and Brown 1999).

Furthermore, low plasma levels of sex steroids have been correlated with minimal territorial behaviour, while high T levels have been associated with aggression and intense territoriality (Rohwer and Wingfield 1981, Schwabl *et al.* 1985, Mays *et al.* 1991).). Territorial aggression and the associated elevated T levels are often observed during periods of territory establishment (Wingfield *et al.* 1990). As WBBAs maintain year-round territories, there may be little need for aggressive behaviour to establish territories, and thus no elevation in T levels. Low T levels in WBBAs are consistent with the minimal aggressive behaviour (territorial or otherwise) observed in this species. In highly social species with low peak T levels, such as the WBBA, White-browed Sparrow Weaver, Superb Fairy-wren, Harris' Hawk, and Pied Kingfisher, high T levels may be incompatible with their gregarious societies (Mays *et al.* 1991, Peters *et al.* 2001, Reyer *et al.* 1986, Wingfield *et al.* 1991, respectively). In highly social species, minor adjustments in behaviour may be better fine-tuned by modest fluctuations in T levels, than by large changes that could result in drastic behavioural modifications (Peter *et al.* 2001).

Finally, there is some evidence that in Australian passerine species, in which opportunistic breeding and extended breeding seasons are common, peak plasma T levels are considerable lower than in their Northern Hemisphere counterparts (Schmidt *et al.* 1991, Poiani and Fletcher 1994, Astheimer and Buttemer 1999, McDonald *et al.* 2001b, Peters *et al.* 2001). While this may indicate a phylogenetic constraint, there also appears to be a correlation between life history characteristics and the maximum breeding T levels observed in Australian species (Astheimer and Buttemer 1999). Social species with opportunistic and/or protracted breeding seasons may not require abrupt hormonal signals to change their behavioural and physiological state. Highly social species may not need to radically change their behaviour to encourage pair bonding, and some may maintain a moderate to high level of gonadal readiness year-round. These species may be able to achieve the appropriate behavioural and physiological modifications through minimal hormonal changes in response to environmental cues. Among species that maintain affiliative relationships year-round, there may be a relatively higher importance of cognitive cues and long-term memory than hormonal cues.

Estradiol

In birds E2 is required for follicular maturation and vitellogenesis (Balthazart 1983), and elevated E2 is thought to be necessary for the expression of courtship, nest building, and copulatory behaviours (Moore 1982, Moore and Kranz 1983, Balthazart 1983, Wingfield and Moore 1987). In many species E2 in measurable throughout the breeding cycle (e.g. Brown-headed cowbirds: Dufty and Wingfield 1986b, Florida Scrub-Jays: Schoech *et al.* 1991, White-crowned Sparrows: Wingfield and Farner

1978a), but in others E2 falls below assay delectability, except at select stages of the breeding cycle. For instance, in the Pied Flycatcher, E2 was only detected in females sampled during the first part of egg laying and the latter part of incubation (Silverin and Wingfield 1982), and in the White-browed Sparrow Weaver, E2 was only detected in three samples from egg-laying females (Wingfield *et al.* 1991).

In the WBBA, E2 reached detectable levels in only a few breeding females; of those whose breeding stage was known, one was laying and the other was incubating. A surge in E2 at egg laying is consistent with a role in yolk formation and in sexual behaviour (Wingfield *et al.* 1991). In the WBBA, other typically estrogen-dependent behaviours may be controlled by hormones other than E2, or E2 may exert its influence at very low levels.

Progesterone

In the months following the peak breeding season, female WBBAs' P levels tended to be low, but there was no other discernable seasonal pattern. As P plays a role in avian reproductive physiology and behaviour (Silver 1990), low levels may be expected when birds are in post-breeding condition. Although there was minimal seasonal predictability in P secretions, there was a pattern associated with reproductive stage.

During nest building and egg laying periods, P tended to be elevated. This is consistent with findings in Northern Mockingbird *Mimus polyglottus*, Bengalese Finch, and Ring Doves *Streptopelia risoria*, where P levels were elevated in nest building and/or laying females (Logan and Wingfield 1995, Seiler *et al.* 1992, Silver 1990, respectively). These temporal changes reflect the role of P in oviduct development, ovulation, and the induction of nest building (Silver 1990). Based on the very elevated measurements in one female WBBA just prior to egg laying, a transitory surge in P may be associated with laying. As this datum was collected immediately prior to a female laying the final egg of her two egg clutch, the P surge could not be attributed to the pre-ovulatory surge found in some species (Tanaka and Inoue 1990). The high P observed in the egg laying female is consistent with findings in domestic Canaries *Serinus canaria* (Sockman and Schwabl 1999) and may induce the transition from active courtship to sexual refractoriness and incubatory behaviour (Leboucher *et al.* 2000).

Surprisingly, female WBBAs caught with fledglings also tended to have high P levels. While some have speculated that P may be associated with alloparental behaviour, detailed studies have found no evidence to support this (Schoech *et al.* 1991, Vleck *et al.* 1991). However, it may be possible that P facilitates alloparental care in female WBBAs with fledglings, either directly or indirectly through an inhibitory effect on breeding behaviour. Non-breeding females (those with completely regressed ovaries) had higher P titres than females in breeding condition, suggesting that there may be a relationship between non-reproductive behaviour and P. Progesterone has been found to suppress female sexual behaviour in birds, mammals, and reptiles (El Halawani *et al.* 1986, Gonzalez-Mariscal *et al.* 1993, Goodwin *et al.* 1996). Prolonged elevated levels of P may inhibit the release of gonadotrophic hormones, leading to decreased follicular development, a depression of E2 secretion, and, consequently, an inhibition of courtship behaviour (Harvey *et al.* 1986, Sharp 1996). Also, Leboucher *et al.* (2000) demonstrated that P treatment in domestic Canaries led to a direct inhibition of the copulation solicitation displays by females. High P may act to suppress follicular development and breeding behaviours in female WBBAs with fledglings and in other non-breeding individuals.

Prolactin

Although seasonal differences in Prl levels only reached statistical significance in males, a seasonal pattern was also apparent in females. As has been found in other avian species, Prl levels tended to be higher during the breeding months than the non-breeding months (Dawson and Goldsmith 1982, Hector and Goldsmith 1985, Silverin 1991). This pattern may be attributed to Prl's relationship to avian parental physiology and behaviour (Buntin 1996) as well as its secretion in response to photoperiodic cues (Hiatt *et al.* 1987, Chakraborty 1995).

Elevated levels of Prl are typically associated with some type of parental behaviour in birds, and most commonly with incubation and nestling feeding. In species with altricial young, Prl levels typically remain high through incubation and at least the early stages of chick rearing (Dawson and Goldsmith 1982, Goldsmith 1982, Myers *et al.* 1989). In contrast, in species with precocial young, Prl usually decreases by the time of young hatching (Goldsmith and Williams 1980, El Halawani *et al.* 1990, Richard-Yris *et al.* 1998). In columbiform species, that feed their young crop milk, development of the crop gland is controlled by Prl, and Prl levels do not surge until just before hatching (Goldsmith *et al.* 1981, Silver 1984). In species where the female is the sole incubator, Prl is typically higher in females than in males (Goldsmith and Williams 1980, Goldsmith 1982). In sex-role reversed species or those with biparental care, where males perform equivalent or more parental duties than females, males have Prl levels equal to or higher than females (Oring *et al.* 1989, Seiler *et al.* 1992).

Some aspects of female WBBAs' pattern of Prl secretion were typical of altricial species in which the female is the sole incubator. As in the Florida Scrub-Jay, Great Tit, and Pied Flycatcher, Prl levels were high while female WBBAs incubated and cared for nestlings (Schoech *et al.* 1996b, Silverin 1991, Silverin and Goldsmith 1984, respectively). Tactile stimulus from the eggs may stimulate Prl secretion in many species (reviewed in Silverin 1991), and Prl is thought to control development of the brood patch and to maintain incubation and brooding behaviour (Eisner 1960, El Halawani *et al.* 1986). Furthermore, Prl stimulates pre-migratory hyperphagia and fattening in some species (reviewed in Wingfield *et al.* 1990). Schoech *et al.* (1996b) suggested that the relationship between Prl and increased feeding behaviour may help explain high Prl levels in birds feeding nestlings.

Unlike the pattern observed in many other passerines (Goldsmith 1982, Silverin and Goldsmith 1984, Seiler *et al.* 1992), female WBBAs tended to have high Prl titres during nest building. These high Prl levels may be related to brood patch development or repeated nesting attempts (El Halawani *et al.* 1986, Hiatt *et al.* 1987). As WBBAs' clutch consisted of only two eggs laid on consecutive days, the transition period between nest building and incubation is short, and elevated Prl levels in the nest building stage may facilitate brood patch development prior to incubation. In some multi-brooding species, Prl levels remain high between broods or between nesting attempts (Song Sparrow: Wingfield and Goldsmith 1990, White-crowned Sparrow: Hiatt *et al.* 1987). Similarly, elevated Prl levels may have persisted between repeated breeding attempts in female WBBAs. Due to the high incidence of nest failure and renesting in the WBBA (Chapter II), nest building often occurred immediately after incubation; the high Prl levels recorded in nest building females may have been maintained from high levels in incubating birds.

Only one Prl sample was obtained from a laying WBBA female, and the level of Prl was intermediate between non-breeding and peak levels but lower than in nest building females. In other species Prl titres typically rose steadily from non-breeding until incubation (Oring *et al.* 1986, Schoech *et al.* 1996b). More data would be necessary to determine if this female's Prl titre was anomalous or indicative of a common pattern.

In all pre-fledging nesting stages except laying, WBBA Prl levels tended to be lower in males than in females. In species where the female provides a greater proportion of parental care than the male, Prl levels are typically higher in females than males (Goldsmith and Williams 1980, Dittami 1981, Goldsmith 1982). The same seems to hold true for the WBBA, as the female is the sole incubator.

The single male sampled that had a laying mate had a high level of Prl. However, the highest three Prl titres were recorded in males with incubating mates. In other species elevated Prl levels have been found in males while their mates were incubating, and it has been suggested that Prl may facilitate provisioning behaviour (Dawson and Goldsmith 1982, Schoech *et al.* 1996b). Male WBBAs were observed provisioning their mates from courtship through incubation, and elevated Prl levels may have promoted this behaviour in males with laying and incubating mates.

In four male WBBAs attending nestlings, Prl titres were close to non-breeding levels; one of these males was paired to the breeding female, while the other three were observed accompanying the breeding pair to and from the nest and protecting the nest from a potential predator (me!). While only the male paired to the breeding female was observed feeding nestlings, the "non-breeding" males may also have provisioned nestlings when I was not present. In some species with altricial young, Prl levels in the attending parents remained elevated for the first few days after hatching and then decreased (Goldsmith 1982, Silverin 1991, Seiler *et al.* 1992). Male WBBAs sampled for Prl in this period were caught at two nests, one with a nestling more than six days old, and one with nestlings of unknown age. Perhaps male WBBAs' Prl levels were high for the first few days post-hatching and then decreased to the titres recorded. As “non-breeding” males attending nestlings may have played more of a protecting role than a provisioning role, low Prl titres might be expected; however, Prl titres in the male feeding nestlings was similar to those of the “non-breeding” males. At the same time, female WBBAs’ Prl was high, but Prl levels in females may be more closely associated with brooding of nestlings than feeding behaviour.

In many species with altricial young, Prl levels were low by the time young fledged (Silverin and Goldsmith 1984, Myers *et al.* 1989, Seiler *et al.* 1992), but male WBBAs captured with post-fledging young (that may not have been their own) tended to have elevated Prl levels. In other cooperatively breeding species, high Prl levels have also been found in group members caring for young. For instance, in the Harris’ Hawk, Prl surged in adult-plumaged male helpers when provisioning nestlings and fledglings (Vleck *et al.* 1991), and some Mexican Jay helpers and breeders retained elevated Prl levels well past the date of fledging (Brown and Vleck 1998). It has been suggested that helping behaviour has a physiological basis (Brown and Vleck 1998), an hypothesis supported by the aforementioned studies, as well as findings of elevated Prl levels in Florida Scrub-Jay helpers feeding nestlings (Schoech *et al.* 1996b). Elevated Prl levels in WBBA males caught with fledglings and juveniles may facilitate behaviours associated with the care of young.

While the feeding of nestlings by their fathers seems to be independent of high Prl, the feeding of fledglings by multiple group members may be facilitated by elevated Prl. After a successful breeding attempt, mated males may attempt another brood. As described previously, low Prl and high T levels are thought to help coordinate behavioural and physiological modifications associated with the initiation of breeding attempts. On the other hand, unmated males or those that had abandoned nesting attempts for the season may have helped rear group members' offspring. In these non-breeding birds, low T and high Prl may facilitate the expression of alloparental care.

CONCLUSIONS

As hormones help bring about many of the physiological changes necessary for breeding, such as spermatogenesis and ovulation, many, if not all, species share some aspects of their endocrine cycles. White-browed Babblers are no exception to this rule. However, patterns of breeding activity vary among species, and these patterns are reflected in specific endocrine profiles. For the most part, hormonal changes associated with different reproductive stages in breeding male WBBAs appear most similar to those of polygynous passerines. While WBBAs' social system is best described as plural cooperative breeding, some aspects of their breeding behaviour resembles polygyny. Breeding males are paired to a single female, but these males are also exposed to multiple breeding females, and they may seek extra-pair copulations. The possibility of additional breeding opportunities, after completion of a male's social partner's clutch, might provide the stimulus necessary for the protracted elevation of T levels in male WBBAs. Stimuli from receptive extra-pair females may prolong T elevations, promote spermatogenic activity, and encourage extra-pair copulations during the period when a male's mate is incubating.

Patterns of hormone secretions associated with the reproductive stages of breeding female WBBAs were similar to those recorded in other monogamous or polygynous passerines. Except for the possibility of renesting, WBBA females' breeding opportunities ended with the completion of her clutch. It appears that in many passerines, including the WBBA, endocrine and behavioural changes corresponded to behavioural and physiological changes necessary for fertilisation, egg laying, incubation, brooding and feeding of young.

While breeding birds are constrained by behavioural and physiological requirements associated with reproduction, non-breeding birds may be afforded more flexibility. As I could not be certain that I discovered all breeding attempts, I could not confirm non-breeding status for many birds. However, I was confident in assigning non-breeding status to adult-sized females with regressed ovaries during the height of the breeding season. These females had higher P levels than their breeding cohorts. It seems that there was a relationship between P and non-breeding behaviour in female WBBAs; however, it was unclear whether behavioural suppression of breeding behaviour induced elevations in P or if high P suppressed breeding behaviour and predisposed birds to adopting a non-breeding or helping role within a group, Also, male WBBAs caught with fledglings and juveniles tended to have elevated Prl and depressed T levels. While some males caught with young may have been their fathers, other undoubtedly were not. As elevated Prl levels are thought to promote parental-type behaviour (Schoech 1998) and low T is thought to play a permissive role in the expression of parental behaviour (Beletsky *et al.* 1990, Vleck and Brown 1999), hormone titres found in these male WBBAs may have predisposed them to alloparental (or parental) care.

This endocrine fine-tuning of social behaviour in WBBAs suggests that non-breeding and/or helping behaviour may in fact be adaptive, at least in this species. As suggested by Vleck *et al.* (1991), if alloparental behaviour is an adaptive trait, then the physiological basis of the behaviour should be modified to promote alloparental behaviour. Elevated P levels in some WBBA female group members may provide a physiological means of promoting the availability of non-breeding group members to assist in the rearing of young, and depressed T and elevated Prl in some WBBA males may promote alloparental behaviours. These findings in the WBBA add to a small body of evidence (Vleck *et al.* 1991, Schoech *et al.* 1996b, Vleck and Brown 1999) suggesting that alloparental behaviour is adaptive and is not an unselected consequence of group living, as suggested by Jamieson (1989, 1991). These studies suggest that selection has favoured birds whose endocrine systems promote alloparental behaviour.

While WBBAs' endocrine and gonadal profiles seemed to closely match reproductive physiology and behaviour, there was also a seasonal pattern, which did not exactly correlate with photoperiod or solstices. In the coldest months of the year, when few WBBAs bred, reproductive hormone levels were depressed; conversely, in warmer months, when much of the population was in breeding condition, reproductive hormone levels were elevated. For WBBAs, the energetic expense of thermoregulation in cold months coupled with presumed lowered food availability may have been incompatible with breeding, and these constraints may have resulted in the observed seasonal fluctuation. I expect that in some years or in other environments (with a more reliable food supply) breeding in WBBAs can take place year round.

In highly seasonal environments in which resources are unevenly and predictably distributed across the year, cues, such as day length, are necessary to coordinate different life stages, such as breeding or migration, with the appropriate time of year. In such cases, physiological systems have adapted to take advantage of these predictive cues. Compared to many species living in highly seasonal environments, WBBAs exhibit only a minimal seasonal pattern in hormonal fluctuation and modifications to gonadal condition. Distinct seasonal endocrine patterns may enable some species living in highly seasonal environments to anticipate breeding opportunities and take advantage of narrow windows of time affording prime breeding conditions. For WBBAs there is a large window of breeding opportunity, during which birds may base their "decision" to breed on social factors or small scale environmental factors (such as recent rains or local food availability). In such cases birds should be physiologically prepared to breed for extended periods, but should not base their physiological readiness solely on seasonal cues, such as day length.

Part 3: Behavioural and Endocrine Studies of Captive White-browed Babblers

Chapter IV. Babbler Group Organisation and Social Behaviour

INTRODUCTION

Social structure is important among group-living animals, particularly as it influences priority of access to resources (Craig and Douglas 1986, Schwabl *et al.* 1988). A stable social organisation helps ensure that dominant individuals control limited resources and may enhance their survival and fitness, as has been found in Willow Tits *Parus montanus* (Ekman 1990). Subordinates benefit by being tolerated by dominant individuals within a stable social group, while all members of the group reap the benefits of group-living, such as improved food finding and predator defense (Bertram 1978). In a stable social structure, all group members benefit from decreased aggression and can direct their attention away from potentially injurious fighting and toward foraging and watching for predators (Holberton *et al.* 1989). The maintenance of long-term hierarchies within social species allows animals to allocate more time to endeavours that may enhance survival and/or fitness rather than on aggressive contests over each and every resource.

In groups exhibiting a social framework, aggressive and non-aggressive behaviours may be associated with the attainment and maintenance of social position. In many species aggressive contests are relied upon to establish and defend a social order, but ultimately social status dictates priority of access to resources. In other species aggression may play little or no role in attaining social position, and contests are replaced by more ritualistic displays of dominance and submission, such as vocalisations and postures. In order to examine the establishment and maintenance of social hierarchies, some investigators have focused on aggressive contests. For instance, social dominance in birds has often been examined in relation to aggressive contests over food (Carlisle and Zahavi 1986, Craig and Jamieson 1990, Ekman 1990, Ekman and Sklepkovych 1994). However, in group-living species that exhibit minimal aggression, other non-aggressive signals of social status have been identified and examined. For instance, Jungle Babblers advertise high social status by acting as sentinels (Gaston 1977); allopreening may replace aggression as a means of establishing and reinforcing social rank in a variety of avian species (Harrison 1965); Arabian Babblers display social dominance by allofeeding subordinate individuals (Carlisle and Zahavi 1986); subordinate Galapagos Mockingbirds crouch and call in a ritualised display similar to that of begging juveniles (Curry 1988).

Even in non-aggressive, stable groups, social dominance often reflects a competitive advantage and may be influenced by a number of factors (Sklepkovych 1997). Gender, body size, age, and kinship are known to affect social position within groups of birds. Typically, males dominate females (Craig and Douglas 1986), larger birds dominate smaller (Nakamura 1998b), and older individuals dominate younger (King 1980). In some species, however, kin-directed tolerance may effectively elevate the social rank of young group members above that of older individuals. For instance, in the Siberian Jay (Ekman and Tegelstrom 1994, Sklepkovych 1997) and Mexican Jay (Barkan *et al.* 1986), retained offspring are subject to relaxed competition and are afforded preferential access to resources.

In this chapter, I examine social dynamics in captive groups of WBBAs. Free-living WBBAs live in long-term social groups, usually composed of four or more individuals. Groups remain stable for extended periods, and members infrequently display intragroup aggression (Chapter II). Thus, this species affords an opportunity to examine the types of behaviour associated with hierarchical maintenance in a highly social avian species. Furthermore, multiple variables, such as extent of relatedness, intragroup gender ratios, and morphometric traits, permits examination of how such variables affect social interactions among group members.

MATERIAL AND METHODS

Capture and Living Conditions

Groups of WBBAs were caught in BYSF and Warredary State Forest and transported to Wollongong NSW soon after capture (described in Chapter II). Most groups were caught in the evening, driven to Wollongong overnight, and released into the aviary in the morning. Prior to being released into the aviary, birds were marked with coloured and numbered bands, measured, and weighed as described in Chapter II. Laparotomies, also detailed in Chapter II, were performed on capture when time constraints and field conditions permitted or soon after removal to the aviaries in Wollongong.

Each group was housed separately and consisted of three to seven birds (mean of 5.2 ± 0.4). Aviary group composition was the same as that when caught in the wild, unless specified. No breeding activity took place in captivity. Initially, two separate groups were maintained concurrently, and subsequently, four separate groups were maintained concurrently. Concurrent, captive groups are referred to as “Assemblages.” Between February 1997 and August 1998, four assemblages of WBBAs (14 groups in total) were housed in the aviary (Table 4-1). After observations and experiments with each group, birds were released at the point of capture. No groups were help captive for more than 4.5 months.

Table 4-1. Dates of captivity and gender composition of groups of WBBAs held in the aviary at Wollongong NSW. Groups held concurrently are identified by the same "Assemblage" number, and "Group" number identifies each group.

Assemblage	Group	Dates of Captivity	Males	Females
1	13	26 Feb. 1997- 11 May 1997	2	5
1	14	3 Apr. 1997- 11 May 1997	3	1
2	15	14 May 1997- 10 Aug. 1997	3	4
2	16	16 May 1997- 10 Aug. 1997	2	2
2	17	18 May 1997- 10 Aug. 1997	5	2
2	18	20 May 1997- 10 Aug. 1997	1	3
3	20	1 Oct. 1997- 10 Feb. 1998	2	2
3	21	4 Oct. 1997- 10 Feb. 1998	1	2
3	22	16 Oct. 1997- 10 Feb. 1998	4	3
3	23	18 Oct. 1997- 10 Feb. 1998	2	3
4	24	10 Apr. 1998- 9 Aug. 1998	2	2
4	25	9 Apr. 1998- 9 Aug. 1998	3	1
4	26[1]	16 Apr. 1998- 9 Aug. 1998	3	1
4	27	16 Apr. 1998- 9 Aug. 1998	6	1

[1] Birds in group 26 fell ill, and data from this group was not used in the analyses described in this chapter.

note: There was no captive "group 19." I used this designation (group 19) for a free-living group.

The aviary complex consisted of four large aviaries: two cells faced north and two south, separated by a central corridor. Each group of birds was housed in a single aviary; groups were visually, but not aurally separated. Aviaries measured 4.5 m wide, 2.3 m high, and 3.7 m deep. The outward face was wire mesh, while the sides and back were solid aluminium. Half of the roof was covered with corrugated roofing, while the other half had wire mesh. Birds could seek shelter from inclement weather, but were exposed to ambient temperature.

The floor was concrete and was covered by wood chips. Tree cuttings were supplied for cover, and a variety of branches were hung for perches. Old nests were brought from the forest, and small sticks were provided for additional nesting material.

I supplied the birds with fresh food every morning, and in hot weather food was refreshed in the afternoon to prevent spoilage. During cooler months food was supplied only once a day. Birds were provided a mixture of mashed, hard-boiled eggs blended with “Insectivore Rearing Mix” *ad libitum* (Wombaroo Food for Wildlife, Glen Osmond, South Australia). As an occasional supplement (approx. once a week), I added raw mince meat, grated cheese, and meal worms (*Tenebrio* larvae) to the egg mixture. I supplied fresh water daily in bowls large enough for bathing. All food and water dishes were cleaned daily.

Initial Observations

The first group of birds brought into the aviary (Group 13, Table 4-1) was observed for more than 15 hours over three weeks. I observed the birds at various times of the day and sometimes provided them with supplemental food, such as meal worms. All behaviours were noted, and observation sessions were reviewed to establish quantifiable behaviours.

A high frequency of allofeeding behaviour was noted during these initial observations. Allofeeding was especially common when meal worms were fed; one bird would often feed another, but it seemed that some individuals fed or received more than others. In an effort to further investigate allofeeding behaviour and to study the implications of preferential access to a preferred food (meal worms), a series of observational and manipulative experiments was undertaken (see below).

I also noted a high frequency of allopreening; all members of the group participated by grooming others, being groomed, or both. However, quantification proved difficult as it was an extremely common yet variable interaction; often, multiple pairs would allopreen at once, sometimes two birds would preen another bird simultaneously, and the interactions were variable in duration ranging from seconds to over five minutes. I recorded allopreening behaviours, but did not detail direction or duration.

Adjustment and Training Periods

Based on the above observations, I developed experimental protocols for observational studies on captive WBBAs. After their arrival at Wollongong, birds were allowed to adjust to the aviary environment for 10 days, during which time I visited them at least once a day to provide food and water and to monitor their health. After this adjustment period, they were trained to find meal worms in a small dish. This dish was attached to the wire mesh wall of the aviary, allowing provision of meal worms without entering the aviary. At first I put many meal worms in the dish and walked out of view; as birds learned to associate my actions with food, I remained within their view after replenishing the dish with worms. The training period lasted for 5-10 days and was considered complete when birds came to the dish without hesitation, while being observed from a 5-10 m distance.

Feeding Competition Observations

After the birds were trained, I performed series of observations on each group. Observations were undertaken in the morning after the birds were fed and watered. Each group was observed for a single, 20-30 minute period per day, and a voice recorder was used to note behaviours.

Two meal worms at a time were placed in the dish. After the meal worms were consumed, two more were added to the dish. Meal worms were usually taken in rapid succession, and the dish was repeatedly replenished for the period of observation. Between 15 and 50 meal worms were eaten by each group during daily observation sessions.

During these sessions, I noted which birds took meal worms from the tray and which ones got the first 10 worms supplied on that day. I recorded whether birds ate the worms themselves or fed them to another bird and any instances of a WBBA stealing a meal worm from another WBBA. Calls and conspicuous displays, such as wing flutters, begging, and dueting were recorded, as were the few instances of chases, pecks, and supplants (one bird displacing another from a perch or the feeding dish). Tape recorded observation sessions were transcribed, and occurrences of particular behaviours were tallied for each session (Table 4-2).

Table 4-2. Definitions of behavioural idioms used to identify common behaviours exhibited by captive WBBAs during feeding competition observations.

Behavioural Idiom	Definition of Behaviour
got 1	took a meal worm from the dish
first 10	took one of the first 10 meal worms offered that observation session
fed	actively fed a meal worm to another bird that proceeded to eat it
got fed	accepted and ate a meal worm offered by another bird
stole	stole a meal worm from another bird
stolen from	had a meal worm in its possession stolen by another bird
beg	gave a begging call and/or performed a fledgling-like wing flutter
raspy call	harsh-sounding call often given when feeding another bird
duet	series of short calls given alternately by two birds with or without an erect wing flutter

Quantification of Behaviours

There was substantial variation in frequency of particular behaviours among groups over the course of this study. Some of this variation may have resulted from temporal and environmental differences among these observation sessions. Accordingly, I decided to standardise behavioural observations across sessions by evaluating frequencies of individuals' behaviours in relation to the occurrence of such behaviours among members of the same group. This was achieved by calculating a "Behavioural Quotient" for members of each group using the following relations: Behavioural Quotient= number of times an individual performed a given behaviour (as listed above) ÷ number of times all members of the individual's group performed that behaviour.

These Behavioural Quotients were used in further analyses as a measure of an individual's behaviour relative to that of other group members.

Classification of Behaviours

I used Behavioural Quotients to separate birds into distinct categories. Two different classifications were created: Access Class and Allofeeding Class. These Behavioural Classes were used to describe intragroup behaviours. More specifically, Access Class was a measure of a bird's access to a preferred resource (meal worms), and Allofeeding Class indicated whether individuals typically fed others, were fed by others, performed both, or abstained from both. Three Access Classes were designated according to the proportion of an individual's intragroup "got 1" and "first 10" behaviours, and four Allofeeding Classes were distinguished by the proportion of "fed" and "got fed" behaviours (see Table 4-2 for definitions of idioms). Access Classes ranged from 1=greatest access to meal worms to 3=least. Allofeeding classes were designated as follows: F=feeder, R=receiver, N=non-allos (neither fed nor received meal worms), and B=both (fed and received meal worms).

To formulate a standard set of criteria to assign Access and Allofeeding Classes, I used cluster analyses as a guide to differentiate groups. Using individuals' Behavioural Quotients, I separated birds into groups using an agglomerative cluster analysis (K-means clustering, SYSTAT 7.0, 1997) based on Euclidian distances. Cluster analyses identify groups that have characteristics that are more similar to each other than to those in another group; clusters have smaller within cluster variation than between cluster variation (Dillon and Goldstein 1984, Anderson 1999).

In an effort to minimise possible temporal biases, only WBBAs held in the aviary concurrently (“Assemblages,” Table 4-1) were analysed together in each cluster analysis. Two cluster analyses were performed on each Assemblage; one grouped birds based on their access to meal worms, and the other clustered birds based on their allofeeding behaviour. However, 3 out of 13 groups exhibited very low or no allofeeding, and, as a result, these groups could not be included in the allofeeding cluster analysis.

For cluster analyses and subsequent analyses on stable group behaviour, I used only observational data that were collected from birds before any manipulative experiments were undertaken or blood samples were collected, as described in subsequent chapters.

Statistical Analyses

Standard descriptive statistics were used to summarize data. Unless stated otherwise, means ± SE are reported. Contingency tables were analysed with chi-squared tests or Fisher’s exact test (when tables contained empty cells or small sample sizes). ANOVA was used to examine the relationship between kinship and preferential access to resources. Variations in body size and mass among different Behavioural Classes were examined using ANCOVA, with gender as the covariate. Tukey-Kramer posthoc analyses were used following both ANOVAs and ANCOVAs. To examine differences in the distribution of DNA bandsharing values (i.e. kinship) among birds exhibiting differences in allofeeding behaviour, Kolmogorov-Smirnov two sample test was used. These statistical analyses were performed using SYSTAT™ 7.0 (1997).

RESULTS

Description of Behaviours

Based on initial observations when meal worms were placed in large feeding dishes accessible to multiple birds concurrently, all birds eagerly sought meal worms. However, during feeding trials, when access to the meal worms was limited by the size of the dish to one or two birds, there seemed to be a strict order as to which birds were given priority of access to the meal worms. This order seemed to have been well-defined prior to the feeding trials, and birds did not seem to physically prevent one another from approaching the dish.

After meal worms were placed in the small feeding dish, birds quickly approached, sometimes before I backed away. Often, multiple birds approached the feeding dish at once, but seldom did the entire group. Even when multiple birds approached the dish, squabbles rarely erupted, and usually a single bird was allowed to take the meal worms without contest.

Allofeeding occurred frequently during almost all of the observations sessions. One bird would take a worm and feed it to another bird. Occasionally, the receiving bird would solicit the food by begging and following the feeder from perch to perch, but more often allofeeding was initiated by the feeder. The feeder would approach another bird (the recipient) and lean towards it with the worm, and the receiver would usually open its mouth and accept and eat the worm. This interaction was often followed by vocalisations: a harsh, raspy, repeated, two-syllable call from the feeder and a begging call from the receiver. This begging call was reminiscent of a begging fledgling; the bird gave a whiney chatter that was accompanied by a flattening of the body and a fluttering of the wings. Both of these calls were also occasionally given in the absence of food.

Sometimes, a meal worm would be refused by the receiver, accepted but deliberately dropped, or accepted but passed on to another bird. On occasion, pairs of WBBAs passed a meal worm back and forth more than five times, and I've witnessed a single worm passed down a line of five birds.

During observation sessions aggressive behaviour was very rare. In fact, over the course of my study (118 hours of observations between March 1997 and July 1998), I witnessed fewer than 15 instances of aggressive behaviour; these took the form of supplants, pecks, and chases. These behaviours occurred too infrequently to analyse statistically

Behavioural Classifications

In 13 groups the duration of observation sessions per group totaled 294.2 ± 18.0 mins over 9.8 ± 0.7 sessions. Behaviours recorded during these observation sessions were examined using cluster analyses, which helped establish behavioural criteria to assign birds to behavioural classes. Table 4-3 lists the results from eight separate cluster analyses performed on four Assemblages.

Results from the cluster analyses indicated some inconsistencies among Assemblages (Table 4-3); for instance, in Access Class 1 the mean "Got 1" was 32.5% for Assemblage 2 but was 48.3% for Assemblage 3, and in Access Class 2 the mean "Got 1" for Assemblage 1 was 34.0% but was 19.7% for Assemblage 2. Despite these and other inconsistencies, a general pattern emerged: birds with the highest priority of access to the first 10 meal worms provided in any observation session (First 10) also got the most meal worms overall (Got 1), and birds that allofed others rarely were fed by others and vice versa.

Table 4-3. Mean and Standard Errors of Behaviour Quotients for Behavioural Classes generated by cluster analyses.

	Access Cluster Class	Got 1 (%) ± SE	First 10 (%) ± SE		Allofeeding Cluster Class	Got Fed (%) ± SE	Fed (%) ± SE
Assemblage 1	1	37.9 ± 0.6	73.3 ± 3.3		N/A[1]	N/A[1]	N/A[1]
	2	34.0 ± 4.5	16.5 ± 7.0		N/A[1]	N/A[1]	N/A[1]
	3	3.73 ± 2.5	0.67 ± 0.4		N/A[1]	N/A[1]	N/A[1]
Assemblage 2	1	32.5 ± 5.9	46.4 ± 9.8		Receivers	49.7 ± 7.0	11.3 ± 4.6
	2	19.7 ± 1.4	15.4 ± 2.3		Feeders	9.6 ± 4.3	49.8 ± 7.7
	3	7.4 ± 2.4	1.6 ± 0.9		Non-allos	8.6 ± 2.2	4.0 ± 2.1
Assemblage 3	1	48.3 ± 6.3	69.5 ± 3.6		Receivers	42.8 ± 2.8	5.8 ± 3.2
	2	28.5 ± 3.0	17.5 ± 4.5		Feeders	5.8 ± 4.9	60.3 ± 10.3
	3	4.0 ± 1.9	1.9 ± 0.8		Non-allos	4.8 ± 2.4	8.7 ± 3.9
Assemblage 4	1	45.2 ± 16.2	55.4 ± 6.9		Receivers	41.1 ± 7.2	1.3 ± 1.3
	2	23.1 ± 0.7	25.2 ± 2.6		Feeders	3.2 ± 3.2	78.4 ± 15.2
	3	6.8 ± 3.0	1.9 ± 0.7		Non-allos	5.9 ± 2.6	7.6 ± 2.6

[1] None of the birds in Assemblage 1 exhibited substantial allofeeding behaviour

Only in Assemblage 1, which consisted of only two WBBA groups, was there a somewhat equitable division of meal worms among birds in two of the cluster classes ("Got 1," Table 4-3). In all other divisions there was a clear distinction between classes and a consistent pattern of relative proportions of behaviours in each class, as designated by the cluster analyses. Because my intention was to examine actual intragroup behaviours and not the statistical groupings as described by the cluster analyses, the data presented in Table 4-3 were used as a guide only for categorising birds from their behaviours. Based on the class separations indicated by the cluster analyses, I established a standardised set of criteria for differentiating Standard Behavioural Classes.

Table 4-4 outlines the criteria I used to designate Standard Behavioural Classes guided by the cluster analyses. I set cutoff points along the range of “Got 1” and “First 10” values to assign Access Classes. Birds that captured a high percentage of all meal worms offered (“Got 1”≥ 30%) as well as a high proportion of the first meal worms offered (“First 10”≥ 30%) were assigned Access Class 1. Those birds that had moderate access to all meal worms offered (20-30% “Got 1”), as well as to the first worms offered that day (10-30% “First 10”) were placed in Access Class 2. Finally, those birds that obtained few meal worms (≤ 20% “Got 1” and ≤ 10% First 10”) were assigned Access Class 3.

The assignment of Allofeeding Classes was also guided by the cluster analysis. Because I wanted to distinguish between birds that acted as both allofeeders AND receivers and those that acted as only one or the other (allofeeders OR receivers), Allofeeding Classes were based on both the absolute frequency of behaviours and the differences between relative frequencies of behaviours. Group members that frequently received allofeeds, but rarely allofed (“Got Fed” at least 20% more than “Fed”) were designated “Receivers.” Those birds that often allofed, but rarely accepted meal worms from others (“Fed” at least 20% more than “Got Fed”) were deemed “Feeders.” Those that rarely offered or accepted allofeeds (“Fed” and “Got Fed” ≤ 20%) were assigned “Non-allos” status, while those that both offered and accepted allofeeds (“Fed” and “Got Fed” ≥ 10%, but less than 20% difference between “Fed” and “Got Fed”) were assigned “Both” status.

Table 4-4. Criteria for assigning Standard Behavioural Classes.

Access Class	Criteria		Allofeeding Class	Criteria
1	Got1≥30% First10≥30%		Receiver (R)	Got Fed≥20% more than Fed
2	Got1= 20-30% First10=10-30%		Feeder (F)	Fed≥20% more than Got Fed
3	Got 1≤20% First10≤10%		Non-allos (N)	Fed and Got Fed<20%
			Both (B)	Fed and Got Fed > 10%, but less than 20% difference

Of 68 birds, 14 could not be classified into Access Classes based on the above criteria, and of 54 that exhibited allofeeding behaviour, three could not be assigned Allofeeding Classes based on these criteria. Based strictly on the above criteria, behavioural data of some birds placed them in between classes. As the Behavioural Classes were used as a means of summarising intragroup behaviours, and because criteria for assigning standardised Behavioural Classes were subjective (albeit guided by the cluster analyses), I permitted leniency in the classification of birds whose behaviours did not strictly adhere to set criteria. I closely examined behaviours of each of the outliers in relation to behaviours of its group members, and if the bird seemed to belonged in a specific class, I assigned it to that class. More specifically, if one of the two measures used to classify a bird (e.g. "Got One" or "First 10") was at least 5% inside the set cut off point for placement in a specific Behavioural Class and the other measure was less than 5% outside the set cut off, I allowed classification of that bird as within former class. For example, bird "89506" obtained 28.7% of all meal worms fed ("Got One") and 37.2% of the first 10 meal worms fed ("First 10"); because "Got One" was less than 5% below the cut off for Access Class 1 (≥30%) and "First 10" was more than 5% above the cut off for Access Class 1 (≥30%), I assigned bird “89506” to Access Class 1. Using these criteria, only three birds remained unassignable for Access Class and one for Allofeeding Class, and their data were removed from further behavioural analyses.

Intragroup Structure

Groups often differed in both number of individuals per group and in the proportion of group members falling into each Access and Feeding Class. However, most groups had at least one bird in each Access Class, and in groups exhibiting allofeeding, most had at least one bird in all but the “Both” Allofeeding Class (Table 4-5).

Table 4-5. Numbers of WBBAs per group that were categorised into each Access and Allofeeding Class.

	Access Class			Allofeeding Class			
Group	1	2	3	R	F	N	B
13	1	2	4	-	-	-	-
14	1	1	2	-	-	-	-
15	2	1	4	1	1	4	1
16	1	-	3	2	1	1	-
17	-	4	3	1	2	4	-
18	2	1	1	1	1	1	1
20	2	-	2	2	1	-	1
21	1	1	1	2	1	-	-
22	1	2	4	-	1	5	1
23	1	2	2	2	2	1	-
24	1	1	2	2	1	1	-
25	-	4	1	-	-	-	-
27	1	2	4	2	1	4	-

Access and Allofeeding Classes were significantly associated with one another (Fisher’s exact test, $P<0.001$). White-browed Babblers were more likely to fall into particular combinations of Access and Allofeeding Classes than they were into others

(Table 4-6). For instance, Feeders were equally likely to be in Access Class 1 or 2, but never fell into Access Class 3, while Receivers were usually in Access Class 3 and never in 1. Although sample sizes were small, there appeared to be a trend for males to be Feeders in Access Class 1 and 2 more often than females, and females tended to be Receivers in Access Class 2 and 3 more often than males (Table 4-6).

Table 4-6. Interrelationship between Access and Allofeeding Classes in groups of WBBAs. Contingency table indicates numbers of WBBAs, numbers of males, and numbers of females in each Behavioural Class. As not all birds could be categorized into Allofeeding Classes, sample sizes are smaller than in other tables.

Access Class

Allofeeding Class

both	1	2	3		males	1	2	3		females	1	2	3
F	6	6	0		***F***	5	4	0		***F***	1	2	0
R	0	2	13		***R***	0	0	4		***R***	0	2	9
B	2	0	2		***B***	1	0	1		***B***	1	0	1
N	3	5	11		***N***	2	3	6		***N***	1	2	5

When independently examining Access and Allofeeding Classes, gender did not influence an individual's Behavioural Class (Table 4-7). There was not a significant association between gender and Behavioural Class, for either Access Class (χ^2=1.627, P=0.443) or Allofeeding Class (χ^2=5.567, P=0.135); however, upon closer examination, males allofed a greater number of meal worms than females, and the most common interaction was males feeding females (Fig. 4-1). By contrast, females seemed equally likely to feed males or other females (Fig. 4-1).

Table 4-7. Number of male and female WBBAs in each Access and Allofeeding Class.

Class: Access Allofeeding

	1	2	3		F	R	B	N
Males	9	10	16		9	5	2	11
Females	4	8	17		3	11	2	8

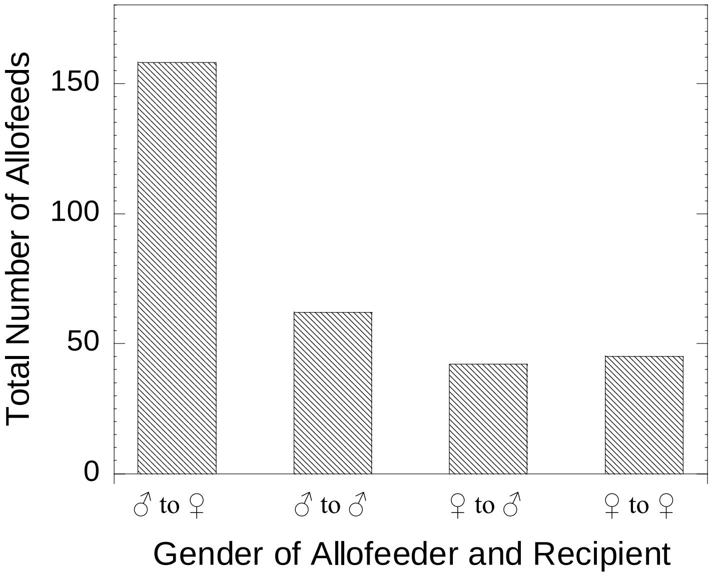


Figure 4-1. Number of meal worms fed by males to females, males to males, females to males, and females to females. Data collected from 26 male and 23 female WBBAs during observation sessions.

I also considered whether body size and/or mass might affect a bird's access to meal worms and/or propensity to allofeed or receive. Because groups had different proportions of males and females (Table 4-1) and males were generally larger and heavier than females (Chapter II), I compensated for this potential bias by designating gender as a covariate when statistically analysing the relationship between size or weight and Behavioural Class. In effect, this removed variability in size or weight that could be attributed to gender. There was not a significant relationship between wing, culmen, or tarsus lengths and Access or Allofeeding Class (Table 4-8, Figs. 4-2 and 4-3); however, body mass differed significantly among Access Classes, but not among Allofeeding Classes (Table 4-8). Birds in Access Class 2 tended to be lightest (Fig. 4-2). Tukey pairwise comparison indicated a significant difference between Access Class 2 and 3 (P=0.016) and a non-significant difference between Access Class 1 and 2 (P=0.077); mean weights between both Access Class 1 and 2 and between 2 and 3 differed by almost 2 g. Differences in wing length among Access Classes were only marginally non-significant (Table 4-8). Wing length tended to be shortest in WBBAs in Access Class 2 (Fig. 4-2).

Table 4-8. Variation in body size and mass among Access and Allofeeding Classes. Results of 8 ANCOVAs are presented. In each case gender was used as the covariate and varied significantly with Behavioural Class (P≥ 0.04). Superscripted asterisk **(*)** indicates significant difference.

	Access Class				Allofeeding Class		
	df	F-ratio	Prob		df	F-ratio	Prob
Wing	2,60	3.006	0.057		3,46	0.125	0.945
Mass	2,60	4.154	**0.020***		3,46	0.022	0.996
Tarsus	2,60	0.066	0.936		3,46	1.087	0.364
Bill	2,60	0.100	0.905		3,46	1.240	0.306

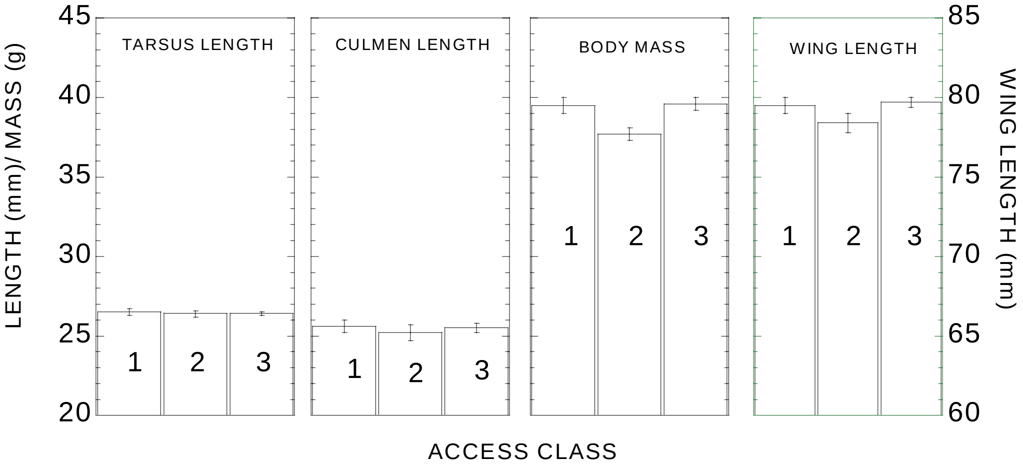


Figure 4-2. Morphological traits among WBBAs of different Access Classes. Left axis pertains to all but wing length.

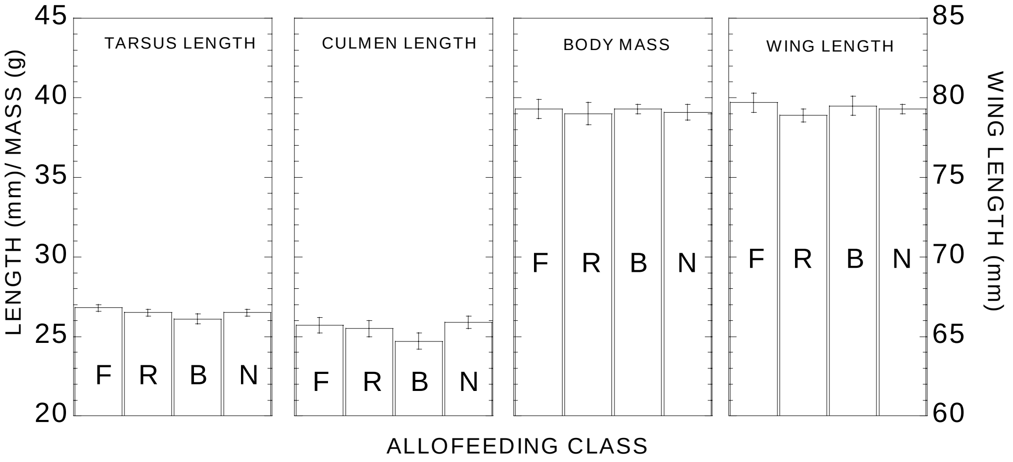


Figure 4-3. Morphological traits among WBBAs of different Allofeeding Classes. Left axis pertains to all but wing length.

Vocalisations

White-browed Babblers frequently gave calls that accompanied particular behaviours. Begging calls and a fledgling-like wing flutter were often given by a bird being fed by another. Raspy calls were frequently given by a bird feeding another, usually immediately after or sometimes just before the allofeeding events. Both begging and raspy calls were sometimes given in the absence of food. Duets were performed by a male and female perched side-by-side and were sometimes accompanied by a flutter of wings, given while the birds stretched their heads and necks upwards.

Begging calls and gestures, duets, and raspy calls were observed with varying frequency among WBBAs, and birds were categorised based on these behaviours. In order to differentiate between birds that exhibited a given behaviour once or very occasionally and birds that frequently exhibited a given behaviour, I deemed birds beggars, raspy callers, or dueters only if they gave the associated call five or more times during the series of observations. No bird fell into more than one of these behavioural categories. Five groups contained a single dueting pair. Six groups contained a single beggar, while one group had two beggars. In five groups there was a single raspy caller, and in another group there were two.

Neither males nor females were more or less likely to be beggars or raspy callers (Table 4-9, Fisher's exact test: P=1.000). However, beggars, raspy callers, and dueters were more likely to fall into some Behavioural Classes than in others (Table 4-9). Fisher's exact test indicated a significant relationship between Access Class and vocalisations (P=0.006) and between Allofeeding Class and vocalisations (P<0.001). Beggars were most often Receivers in Access Class 3, and raspy callers were most often Feeders in Access Class 1. Dueters fell into each Access Class, and in four groups that exhibited allofeeding behaviour, the male (of the dueting pair) acted as the Feeder and the female the Receiver (Table 4-10).

Table 4-9. Number of males and females classified as raspy callers or beggars.

	Males	Females
Raspy Callers	4	3
Beggars	5	3

Table 4-10. Number of WBBAs that gave begging calls, raspy calls, and duets in each Access and Allofeeding Class.

Class:	**Access**				**Allofeeding**			
	1	2	3		F	R	B	N
Beggar	0	1	7		0	6	0	0
Raspy caller	5	2	0		6	0	1	0
Dueter	3	2	5		4	4	0	0
None of the above	6	9	22		4	8	3	14

n.b. Not all birds were assigned vocalisation classifications, and not all groups were partitioned into Allofeeding Classes, but could still contain beggars, raspy callers, or dueters

Allofeeding Interactions

To determine if there was an allofeeding "hierarchy" or if there was a high degree of allofeeding reciprocity, I examined intragroup meal worm allofeeding patterns for each group. Group members were arranged in matrices in order to analyse the linearity of interactions (Table 4-11). These matrices were organised with allofeeders arranged in descending order of suspected allofeeding rank in the vertical columns and all potential recipients arranged in the same order along each row. If hierarchies were strictly linear, no interactions would be recorded below the diagonal, nor could any two birds be transposed in rank order with the same result. Interactions recorded below the diagonal represented reversals to the common pattern of behaviour and indicated a violation of the linear trend of the hierarchy.

In most groups there was a high degree of linearity and few reversals; of five groups with reversals, three had only one instance of a reversal (Table 4-11). Males ranked above females in the hierarchy in five groups, females above males in three groups, and in two groups males and females could be transposed without increasing the number of reversals.

Despite the apparent linearity of these hierarchies, each had more than 5% probability of occurring by chance. In fact, in any group of less than six individuals, the probability that a hierarchy appears linear by chance is high, and this probability always exceeds accepted levels of statistical significance (P>0.05) (Appleby 1983). Even in the three WBBA groups with six or more individuals, the absence of allofeeding relationships between some combinations of individuals (0's in the analysis, Table 4-11) rendered the linear trends non-significant (Appleby 1983).

Table 4-11. Number of allofeeding events in 10 groups of WBBAs. Numbers following the letter "G" indicate group identities. Otherwise, letters represent colour bands of individuals, and numbers indicate the number of allofeeds. Allofeeders are read vertically, and Receivers are read horizontally. Bold, italicised numbers indicate reversals (interactions violating the linear trend in the allofeeding "hierarchy").

G-20	BO	RW	MB	YG
BO ♂	--	12	15	0
RW ♂	***1***	--	1	15
MB ♀			--	1
YG ♀				--

G-23	R	Y	DB	M	LG
R ♀	--	0	0	9	5
Y ♂		--	0	2	5
DB ♂			--	0	4
M ♂				--	0
LG ♀			***1***		--

G-24	Y	W	R	DB
Y ♂	--	38	9	22
W ♀		--	1	0
R ♂			--	1
DB ♀				--

G-18	OW	GO	MY	YG
OW ♂	--	11	2	2
GO ♀	***1***	--	2	4
MY ♀			--	4
YG ♀		***9***		--

G-15	OB	BM	BO	WY	MB	YR
OB ♀	--	3	0	1	0	0
BM♀		--	2	0	4	0
BO ♂		***1***	--	1	1	3
WY♀	***1***			--	0	0
MB ♂					--	0
YR ♀						--

G-16	RW	YG	MY	GB
RW ♂	--	24	16	0
YG ♀		--	1	0
MY ♂			--	1
GB ♀				--

G-17	OB	BM	YW	RB	BG	GY
OB ♂	--	0	7	0	1	0
BM ♀		--	6	2	1	2
YW ♂			--	0	0	0
RB ♀	***1***			--	0	0
BG ♀					--	0
GY ♂						--

G-22	MO	RB	WM	BO	GR	YM
MO ♀	--	4	0	0	0	0
RB ♂		--	0	0	3	0
WM ♂			--	0	1	0
BO ♂				--	1	0
GR ♂					--	1
YM ♀						--

G-21	RW	GO	LG
RW ♀	--	12	10
GO ♀		--	0
LG ♂			--

G-27	Y	RW	DB	MY	M	R	LG
Y ♂	--	1	1	0	0	0	1
RW ♂		--	0	1	10	0	0
DB ♂			--	3	0	2	0
MY ♂				--	0	0	0
M ♀					--	0	0
R ♂						--	0
LG ♂							--

Kinship and Behaviour

To test hypotheses pertaining to the influence of relatedness on behaviour, I investigated the degree of DNA bandsharing (which implies relatedness) in relation to some of the behaviours quantified above. Both bandsharing and behavioural data were collected from nine captive groups (see Chapter II for DNA fingerprinting methods). First, I examined the relationship between priority of access to resources (Access Class) and relatedness (bandsharing). Were some birds given high priority of access to meal worms because of their kinship to particular group members? Bandsharing coefficients were calculated for all combinations of two birds in each of nine groups, and each pair of these birds was placed into one of six categories based on the birds' combination of Access Classes (Fig. 4-4). Bandsharing values varied significantly among the six categories ($F_{5,56}$=2.755, P=0.027). Post-hoc analysis (Tukey's pairwise comparison) did not identify any significant differences between categories. However, two marginal, but non-significant, differences were revealed: when both members of a pair were in Access Class 3, pairwise bandsharing tended to be lower than when one member was in Access Class 1 and the other in Access Class 2 (P=0.082) or when one member was in Access Class 2 and the other in Access Class 3 (P=0.084). Most WBBAs that were closely related to a group member in Access Class 1 (D close to 0.4) were themselves in Access Class 2 or 1. In other words, WBBAs in Access Class 1 shared more bands (i.e. had higher bandsharing coefficients) with other group members in Access Class 1 and others in Access Class 2 than they did with group members in Access Class 3 (t_{23}=2.112, P=0.046).

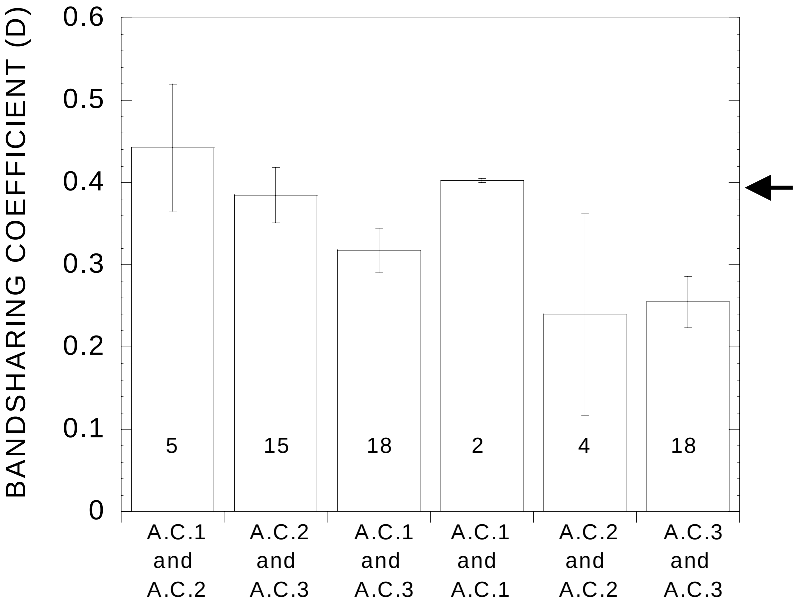


Figure 4-4. Pairwise bandsharing coefficients (an index of relatedness) in WBBAs in different combinations of Access Classes. Height of columns indicates means, and bars represent standard errors. Numbers inside columns indicate number of WBBA pairs, and the arrow indicates estimated mean bandsharing coefficient for first order relatives (see Chapter II).

I also examined bandsharing between dueting pairs. Bandsharing information was available from only four groups with dueting pairs, prohibiting statistical analysis. However, all dueting pairs had low pairwise bandsharing (D between 0.200-0.260), but their bandsharing values were not always the lowest among male-female pairs in their group (Fig. 4-5).

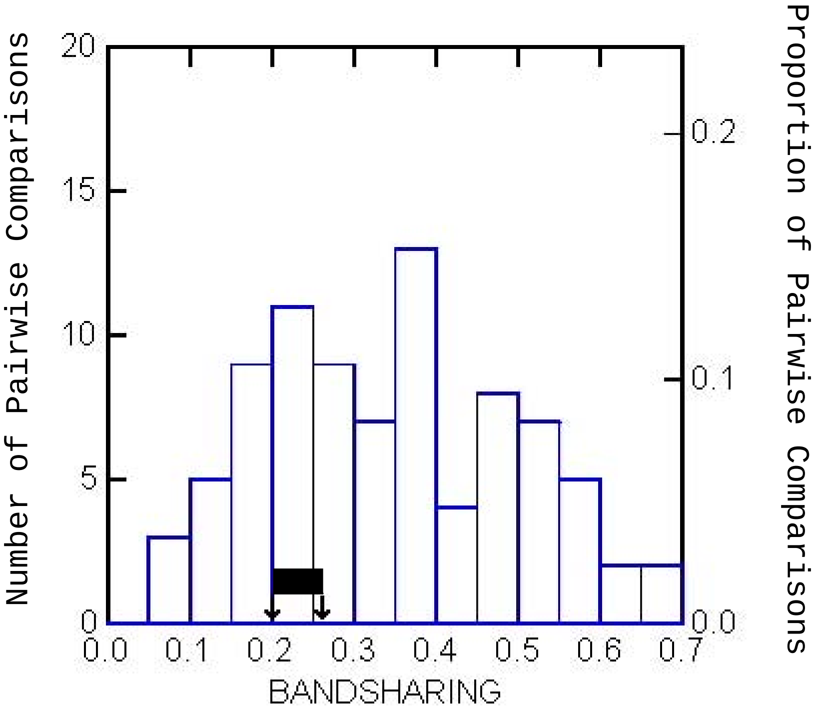
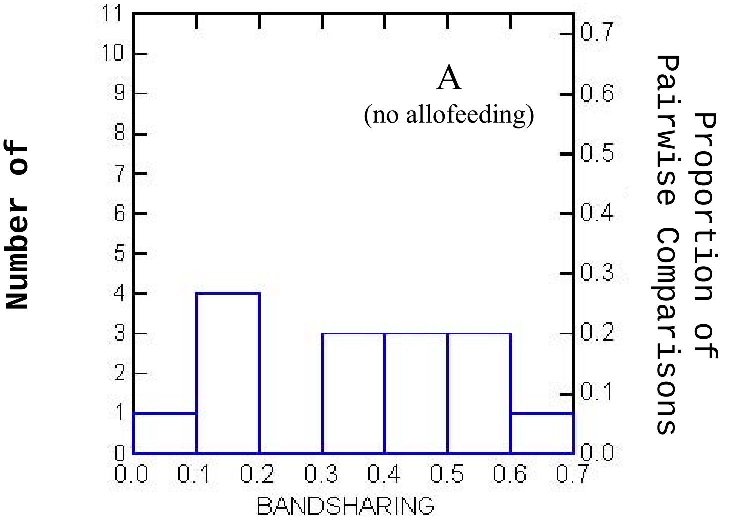


Figure 4-5. Distribution of bandsharing values from 83 intragroup pairwise comparisons in 9 groups of WBBAs. Bandsharing values of dueting pairs fell between 0.200 and 0.260 (filled rectangle and arrows on X axis delineate this range).

I examined the relationship between allofeeding behaviour and genetic relatedness. Analysis was confined to intragroup pairwise comparisons within five groups exhibiting the highest frequencies of allofeeding behaviour. Two categories were considered: A (n=15 pairs) comprised pairs of birds that didn't allofeed one another, and B (n=11 pairs) consisted of pairs in which frequent allofeeding was recorded (more than 5 unidirectional allofeeds during observation sessions). Figure 4-6 illustrates the distribution of bandsharing values in each of these groups. There was no significant difference between these two distributions (Kolmogorov-Smirnov test, P=0.509). However, it is striking that six of the eleven allofeeding pairs had bandsharing values between 0.2 and 0.3, all of which were heterosexual pairs. Of allofeeding pairs in the 0.4 to 0.6 bandsharing range, three were same-gender and two were heterosexual pairs.



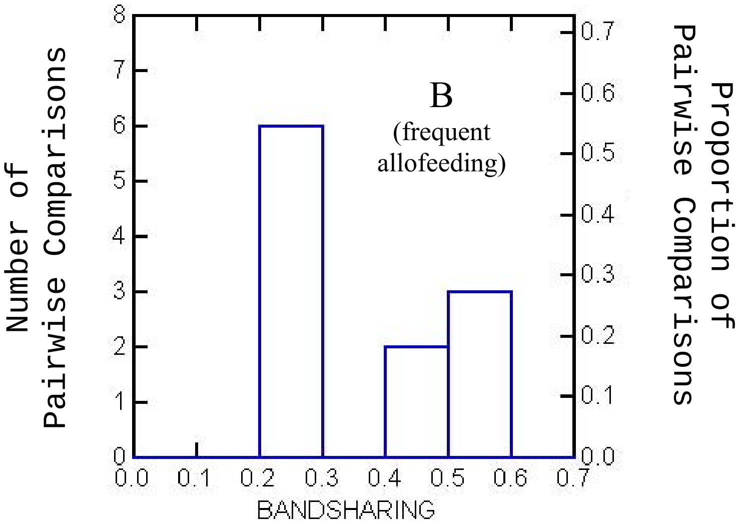


Figure 4-6. Distribution of pairwise bandsharing values in two categories: pairs of WBBA that do not exhibit allofeeding (A) and those that do allofeed (B).

DISCUSSION

Dominance

Animals living in social groups often organise themselves in ways that effectively partition resources while minimising aggression. These organisations can take many forms, from loose aggregates, such as wintering bird flocks, to assemblages with highly specialised divisions of labour, such as many Halictinae bees (Crespi and Yanega 1995). In many social groups that maintain more than a transitory association, some group members dominate over others in gaining access to resources. A dominance hierarchy, often established and/or maintained by aggressive contests, is frequently used to describe patterns of intragroup relationships (e.g. Rohwer and Rohwer 1978, Chase 1982, Craig and Douglas 1986, Ekman 1990).

To investigate the social dynamics within groups of WBBAs, I performed behavioural studies in a controlled aviary setting. In an attempt to identify a dominance hierarchy, I tried to provoke aggressive intragroup interactions by providing a preferred food (meal worms) in a restricted manner; this technique had been used by other investigators to determine dominance hierarchies in a number of social bird species (Arabian Babblers: Carlisle and Zahavi 1986, Pukeko: Craig and Jamieson 1990, Siberian Jays: Ekman and Sklepkovych 1994). However, I witnessed virtually no aggressive interactions in the WBBA.

The absence of such intragroup aggression is not uncommon among social bird species and has also been noted within long-standing groups of White-fronted Bee-eaters, Jungle Babblers, adult Bell Miners, and Groove-billed Anis (Emlen and Wredge 1989, Gaston 1977, Poiani 1993, Vehrencamp *et al.* 1986, respectively). In the WBBA, as well as in other social species, the use of aggression to assert dominance may be counterproductive because it could dissuade potential collaborators from joining (or remaining with) the group (Carlisle and Zahavi 1986). Researchers examining social interactions in non-agonistic species have used a variety of behavioural characteristics to evaluate social status within groups. Poiani (1993) assigned dominance status to Bell Miners based on how long they queued before feeding chicks. Nakamura (1998a) described status that was based on priority of access to resources in Alpine Accentors, while Curry (1988) used calls and postures to determine dominance in Galapagos Mockingbirds.

Although it has been persuasively argued by Drews (1993) that "dominance" should refer only to agonistic behaviour, "dominant" and "subordinate" are convenient terms to describe relative intragroup behaviours. There are no other terms that convey the same concept but omit aggression from their meanings. Furthermore, in comparing my findings with other studies it becomes necessary to rely on a definition of "dominance" that includes non-aggressive interactions. Thus, I will relate dominance in WBBAs to studies that assess dominance in terms of aggression, but I will also describe non-aggressive behaviours similar to those observed in the WBBA.

Behavioural Classes

White-browed Babbler group members behaved differently from one another. My evaluation of these differences focused on two behavioural classifications: Access Class and Allofeeding Class. Access Class reflected priority of access to a preferential food item (meal worms), and Allofeeding Classes described patterns of accepting and offering meal worms. Although these two Classes described different behaviours, they revealed a consistent interaction between rankings of birds within their classifications.

In the following paragraphs, I argue that relative rank within both Access and Allofeeding Classes reflect dominance status within groups of WBBAs.

1. Access Class

Priority of access to resources has often been used to identify dominance of members within a group (reviewed in Drews 1993). In the group-living Mexican Jay, Siberian Jay, and Alpine Accentor (Barkan *et al.* 1986, Ekman and Sklepkovych 1994, Nakamura 1998a, respectively), birds were trained to find an artificial food source, and agonistic interactions over access to food were used to construct dominance classifications. Aggression was used by these birds to insure priority of access to the food source.

Although WBBAs did not act aggressively toward one another when obtaining meal worms, one or two birds in each group routinely took most of the first meal worms offered each day, as well as a high proportion of the total. Live food was highly sought after by all WBBAs, yet some birds never even approached the feeding tray. It seemed that there was an accepted dominance order within the group that conferred preferential feeding access to some birds without the need for aggressive interactions. Birds in Access Class 1 could be viewed as winners of contests over resources and birds in Access Class 3 the losers; in other words, Access Class 1 described the most dominant position and Access Class 3 the least.

In many studies investigating social dominance, investigators have found that males had the highest dominance rank (Craig and Douglas 1986, Barkan *et al.* 1986, Zahavi 1990, Poiani 1993). Surprisingly, there was no significant gender bias in any of the WBBA Access Classes. Some studies have shown that the largest birds command the most resources; for instance, larger, heavier dominant male Alpine Accentors had more mating opportunities than did smaller, subordinate males (Nakamura 1998b), and larger adult male Mexican Jays competed more successfully at feeding stations than smaller females (Barkan *et al.* 1986). In the WBBA, there appeared to be a trend toward a relationship between some measures of body size and Access Class. White-browed Babblers in Access Class 1 and 3 tended to have longer wings and greater body mass than those in Access Class 2. Although indicative of body condition, mass also reflects body size (Freeman and Jackson 1990). Comparatively larger WBBAs in Access Class 1 did indeed command more resources than smaller birds in Access Class 2; however, smaller WBBAs in Access Class 2 were permitted greater access to meal worms than were larger birds in Access Class 3. Perhaps young birds in Access Class 2, that may have had shorter wings and lower body mass, were tolerated at the feeding dish by dominants more so than were older, larger group members in Access Class 3. This scenario has been found in Piñon Jays *Gymnorhinus cyanocephalus*, Mexican Jays, and Siberian Jays, where young were given priority of access to food over adults (Balda and Balda 1978, Barkan *et al.* 1986, Ekman and Tegelstrom 1994, respectively).

A number of other studies have noted that age can influence dominance interactions in group-living birds (Woolfenden and Fitzpatrick 1977, Craig 1979, King 1980, Ekman 1990, Emlen 1996, Magrath and Yezerinac 1997, Wright 1997, Nakamura 1998a,b). Since I had no information on age of the WBBAs in my study, it was possible that both gender and size affected behaviour, but that age was the overriding factor influencing priority of access to resources. Perhaps larger male WBBAs did in fact dominate same-aged females, but some younger females dominated older males. This scenario could result in the observed equivalent proportions of males and females in each Access Class in WBBA and also may have confounded an examination of size effects.

Kinship might also have influenced priority of access to resources. Parental tolerance of their offspring's aggressive behaviour over food has been implicated in the high dominance rank of young birds in some species (Mexican Jays: Brown and Brown 1984, Siberian Jays: Ekman and Tegelstrom 1994). However, the relationship between foraging competition and kinship has only been directly tested in one species, the Siberian Jay. Adult Siberian Jays tolerated kin at foraging sites much more readily than non-kin (Ekman and Tegelstrom 1994, Sklepkovych 1997). The same kin bias may have occurred in WBBAs. Genetic evidence suggested that close kinship ties to a group member in Access Class 1 may have conferred greater access to meal worms than did distant or unrelated ties. In other words, if a bird's parent or sibling was in Access Class 1, it was more likely to be in Access Class 2 or 1 than in Access Class 3. The reverse did not hold true: a high degree of relatedness (bandsharing) to a bird in Access Class 2 did not seem to confer special meal worm privileges. Although more specific experiments would be required to verify this hypothesis, it appears that kinship does influence priority of access to resources (and thus dominance rank) in WBBAs.

2. Allofeeding Class

Observations on allofeeding birds have led to the same conclusion in a few different species: dominant birds allofeed subordinates more often than the reverse (Arabian Babbler: Carlisle and Zahavi 1986; Green Woodhoopoes: Ligon and Ligon 1983; Northwestern Crow *Corvus caurinus*: Verbeek and Butler 1981). Furthermore, allofeeding is thought play a role in the establishment and/or reinforcement of social bonds in a number of other species (Ground Hornbills *Bucorvus leadbeateri*: Kemp and

Kemp 1980, Jackdaws *Corvus monedula*: de Kort *et al.* 2003, Pied Starlings *Spreo bicolor*: Craig 1988, Pukekos: Craig 1980).

As hypothesised for the above species, WBBAs also seem to use allofeeding as a means of establishing and/or maintaining social position within their groups. White-browed Babblers in Access Class 1 and 2 most often fed birds in Access Class 3, and birds in Access Class 3 never fed birds in Access Class 1 or 2. In other words, the more dominant WBBAs fed birds subordinate to themselves. The apparent linearity of the allofeeding hierarchies also suggested that allofeeding was a structured interaction and was not performed randomly or reciprocally by all members of the group. White-browed Babblers that received meal worms rarely returned the favour; subordinate birds seldom fed their superiors. Furthermore, WBBAs that received meal worms often gave a submissive display and call, much like a begging fledgling. Though difficult to test conclusively, acceptance of meal worms by WBBAs appears to be an indication of their subordinance.

Similar fledgling-like behaviours exhibited by adults were also used by Curry (1988) to assign social status in the plurally breeding Galapagos Mockingbird. When a bird crouched and called in a manner similar to a begging juvenile upon being approached or threatened by another group member, he assigned a subordinate position to the former and a dominant position to the latter. Also, Gaston (1977) used a similar line of reasoning when he suggested that allopreening interactions reflected dominance relationships within groups of Jungle Babblers. Gaston (1977) judged dominance in terms of reproductive opportunity, and subordinates with no opportunity to reproduce (young birds) rarely allopreened, as opposed to dominant, reproductively active birds that frequently allopreened.

Allofeeding Class in WBBAs was not influenced by either gender or size. Also, when examining the allofeeding hierarchy, there was no clear trend of male or female domination in allofeeding rank. However, when I examined absolute number of allofeeding events, I found that males fed more than twice as many meal worms as females and that the most common interaction was for males to feed females. Also, among dueting pairs, it was always the male that fed the female. Dueting accompanies courtship and pair bonding in a number of species (Morse 1970, Wickler 1980, Arrowood 1988, Levin 1996), and the low bandsharing coefficient between WBBA dueting pairs suggested that they were unrelated and perhaps predisposed for courtship and breeding. In these dueting WBBAs, allofeeding may have served as an indicator of social status as well as a pair bonding ritual. In free-living WBBAs, males often brought food to females during courtship (Chapter II). The more frequent occurrence of male versus female allofeeding events during observation sessions of captive WBBAs might have reflected this dual purpose. In many instances allofeeding, a typical breeding behaviour in WBBAs, seems to have been co-opted into an expression of social status within the year-round groups.

Although sample sizes were small and patterns between relatedness and allofeeding were statistically non-significant, the distribution of bandsharing values among allofeeding pairs is intriguing. Values clustered within two separate bandsharing intervals: between 0.2 and 0.3 and between 0.4 and 0.6. As mentioned previously, dueting pairs were unrelated birds that exhibit a high level of allofeeding. All allofeeding pairs with bandsharing between 0.2 and 0.3 were dueting pairs, and allofeeding was probably related to pair bonding behaviour. The other pairs of frequently allofeeding birds were closely related (bandsharing between 0.4 and 0.6), probably parents and offspring or siblings (Chapter II).

Even when allofeeding took place between parent and offspring or between sibling, it may still have reflected dominance relationships. By acting like young birds, adult offspring emphasised their submission, and allofeeding parents asserted their dominance by maintaining their role as provider of resources. Parents may have provisioned their young well past fledging in an effort to encourage their young to stay with the group as subordinate members and share such costs as territory defense, vigilance, and possibly alloparental care. Meanwhile, offspring seem to accept the role of the submissive juvenile by begging for and accepting food. Not only do such young-behaving birds reap nutritional benefits, but they may also profit from prolonged parental care and observational learning (Lawton and Lawton 1986, Heinsohn 1991).

Sibling allofeeding can be described as a means of redirecting aggression while asserting social dominance. Allofeeding a sibling may help establish or maintain social dominance without the potential costs associated with agonistic actions. Carlisle and Zahavi (1986) reported that same-aged sibling Arabian Babblers fought to establish dominance soon after fledging, but that aggression became increasingly subtle and ritualised with time. Further investigation is necessary to determine whether a similar sequence of events may take place among sibling WBBAs.

In two other *Pomatostomus* babbler species (Grey-crowned and Hall’s), investigators speculated that peer allofeeding was simply a means of maintaining group cohesion (King 1980, Balda and Brown 1977, respectively). These studies reported that food sharing occurred between all members of the group. However, King (1980) also described a begging display that was performed during allofeeding interactions and was used as a submissive display. While I do not dispute the suggestion that allofeeding helps maintain group cohesion, I expect that the birds receiving the proffered food were also signaling their subordinate role to the feeder. Although impossible to glean from the reported data, I expect that further investigation would show that dominant Grey-crowned and Hall's Babblers feed birds that are subordinate to them. Furthermore, through my cursory examination of allo*preening* behaviour, I speculate that a closer evaluation of its social implications may reveal that dominant WBBAs allopreen subordinate birds more often than the reverse, as has been shown in Jungle Babblers (Gaston 1977).

Behaviours as Signals

While the mechanisms of hierarchy formation in the WBBA remain unidentified, uncontested access to meal worms and allofeeding behaviour serve to reinforce the established social structure. Similarly, allopreening and various vocalisations may indicate social position. Such behaviours act as signals that help maintain a group's social structure. Using signals rather than aggressive contests to indicate social status may be advantageous to both dominants and subordinates as it avoids costly fighting (Rohwer and Rohwer 1978). This may be especially true for group-living animals, that interact frequently. Furthermore, in cooperative societies, threats and fights are not appropriate strategies for attracting collaborators (Zahavi 1990).

White-browed Babblers in Access Class 1 asserted their social position by monopolising access to meal worms, but they also advertised their dominance and foraging ability when they retreated to a perch or flew about the aviary with the worm. In such a manner they displayed their prowess to group members. Lotem *et al.* (1999) suggested that when individuals altered their behaviour when watched by others, they were using that behaviour as a signal. By showing their takings to others, WBBAs advertised their access to a preferred resource and, thus, their high social status.

Allofeeding can also be described as a means of advertising social status. The handicap principle (Zahavi 1975, Carlisle and Zahavi 1986) suggests that altruism serves as a signal of quality used to gain social prestige. For instance, if a bird is able to find enough food to feed both itself and another, it must be of high quality. By demonstrating the ability to bear a cost, the demonstrator reveals its hidden quality (Hawkes and Bird 2002). Under natural conditions, where food sources are not as abundant as in the aviary, allofeeding may be a much more costly action and thus have a more meaningful social impact than in captivity. In fact, White-winged Choughs sometimes pretend to allofeed nestlings, but consume the food themselves; in this way, they advertise their "quality" but avoid the cost (Boland *et al.* 1997a). In the Arabian Babbler, Zahavi (1990) interpreted all seemingly altruistic behaviours, including allofeeding, as selfish means of displaying social status. Furthermore, Carlisle and Zahavi (1986) suggested that the most effective means to demonstrate quality was to perform behaviours that would be valuable to potential collaborators. Putland (2001) further suggests that alloparental behaviour may be a sexually selected display influencing mate choice. Allofeeding by WBBAs explicitly demonstrated foraging ability and willingness to provision others, both of potential benefit to mates or other members in cooperatively breeding groups.

Furthermore, the raspy and begging calls, that often followed an allofeed, drew attention to the interaction; this suggested that the allofeeding interaction had social implications, not only for the participating pair, but also for all group members. These vocalisations may not only have acted to attract attention to allofeeding interactions, but may also have been direct advertisements of social status, especially for the raspy callers. The begging call was almost exclusively given by birds of low social position (Access Class 3, Receivers) and the raspy call was predominately given by those of high social status (Access Class 1, Feeders). These calls may have acted as inexpensive means of asserting social standing within groups.

CONCLUSIONS

Captive groups of WBBAs lived within a stable, stratified social structure. Agonism did not play an important role in the maintenance of their social structure. Instead, this structure was maintained, at least in part, by ritualized behaviours, such as allofeeding and certain vocalisations. Some of these behaviours (such as allofeeding and begging) may have been co-opted from breeding behaviours. In addition, body size and kinship may influence social standing within groups of WBBAs.

The functional significance of the social structure may be more apparent when extrapolated to conditions faced by wild WBBAs. Free-living WBBAs seem to rely on group-living for survival (Chapter II). Therefore, it should be of utmost importance to amicably partition resources within the group, so as to facilitate maintenance of group cohesion. At times, free-living WBBAs may be faced with scarce food resources and/or limited breeding opportunities. Under such conditions a stable social structure may help coordinate group members' priority of access to resources. A stable social structure may help ensure priority of access to resources for dominants, while subordinates can direct their efforts away from competition and toward finding alternate resources.

Chapter V. Physiological Correlates of Social Behaviour During Periods of Social Stability

INTRODUCTION

In group-living animals social factors can both influence and be influenced by hormone levels. In some species social dominance, social change, and specific behaviours are correlated with high or low levels of certain hormones. For example, dominant male Harris' Sparrows were found to have higher T levels than subordinates (Rohwer and Wingfield 1981), plasma F levels increase in Squirrel Monkeys during group formation (Mendoza *et al.* 1979), and allofeeding is associated with elevated Prl levels in Harris' Hawks (Vleck *et al.* 1991). However, specific hormone-behaviour relationships are inconsistent among species. For instance, an inverse relationship between social status and glucocorticoid levels was found in mice and Olive Baboons (Louch and Higginbotham 1967, Sapolsky 1990), a positive relationship was found in African Wild Dogs and Dwarf Mongooses (Creel *et al.* 1996), while no relationship was found in Florida Scrub-Jays and White-throated Sparrows *Zonotrichia albicollis* (Schwabl *et al.* 1988, Schoech *et al.* 1997). Such discrepancies have sometimes been attributed to differences in the stability of the social structure being examined. It has been suggested that the relationship between hormone levels and social status are only apparent during the establishment of structured social relationships (Ramenofsky 1984, McGuire *et al.* 1986, Hegner and Wingfield 1987b), while others have suggested that there are endocrine correlates to social status only in established social groups (Ely and Henry 1978, Sapolsky 1990).

Most studies examining hormonal correlates of social position in birds have focused on agonistic-based dominance relationships in flocks of wintering birds (e.g.

Hegner and Wingfield 1987b, Schwabl *et al.* 1988) or breeding status in cooperatively breeding birds with a single breeding pair per group (e.g. Wingfield *et al.* 1991, Vleck *et al.* 1991, de la Cruz *et al.* 2003). The WBBAs I studied do not fall into either of these categories. They live in year-round groups and are plural cooperative breeders with multiple pairs per group (Chapter II). Because there were multiple breeding pairs in most social groups, I could not differentiate individuals based on breeding role. Further, there was minimal aggressive behaviour, so dominance could not be assigned based on agonistic interactions. Instead, I focused my studies on social position based on resource competition and allofeeding behaviour (Chapter IV).

Aspects of establishing, maintaining, and occupying different social positions can result in variability in individuals' stress levels. While the definition of "stress" is subject to some debate (James *et al.* 1989), herein it will refer to conditions that give rise to a departure from homeostasis, including both physical and psychological factors. Animals respond to stressors through a series of reactions, including those involving the hypothalamo-pituitary-adrenal (HPA) axis that result in the secretion of glucocorticoids (Harvey *et al.* 1984, Sapolsky 1993, 2002). As a result glucocorticoids are often regarded as "stress hormones" and their levels are frequently used to evaluate the degree of stress experienced by animals (Baum *et al.* 1982, Harvey *et al.* 1984, Pollard 1995). Baseline glucocorticoid concentrations are an indicator of an animal's unstressed (or chronically stressed) levels, and serially sampled glucocorticoid levels taken during the course of a stressful episode (e.g. capture and handling protocol) can reflect the sensitivity of the HPA axis to stress (Wingfield *et al.* 1998). Both measures are useful in quantifying the degree of stress perceived by animals.

Previous studies have shown that the degree of stress, measured by increases in glucocorticoid levels, often varies in relation to social position. In some species subordinate individuals exhibit higher baseline plasma glucocorticoid levels and/or more rapid rises in plasma levels when exposed to stress than more dominant members (e.g. fish: Ejike and Schreck 1980, Sloman *et al*. 2001; mice: Louch and Higgenbotham 1967; Olive Baboons: Sapolsky 1990; rats: Sakai *et al.* 1991; wolves: Fox and Andrews 1973; various bird species: Wingfield and Moore 1987). Elevated glucocorticoid levels in subordinates are thought to result from reduced access to resources and intimidation or harassment from dominants (Bronson 1973, Eberhart *et al.* 1983, Schwabl *et al.* 1988). Nevertheless, other studies have suggested the opposite, that dominant individuals are more stressed (i.e. have higher glucocorticoid levels) than subordinate group members (e.g. African Elephants: Foley *et al.* 2001; African Wild Dogs: Creel *et al.* 1996; Dwarf Mongooses: Creel *et al.* 1992; female Common Marmosets: Saltzman *et al.* 1994; Ring-tailed Lemurs: Cavigelli 1999; Squirrel Monkeys: Coe *et al.* 1979). In some cases social dominance may be more stressful than subordinance because dominants engage in more aggressive interactions than do subordinates (Creel *et al.* 1996).

As aggression plays little or no role in maintaining social structure in stable groups of WBBAs and as all birds in captive groups have ample access to necessary resources, neither subordinate nor dominant individuals may experience chronically high stress levels; therefore, I expected no correlation between social status and baseline B levels in stable, captive groups of WBBAs. However, there may be a more pronounced stress response, measured by serial samples taken during a capture stress series, in subordinate individuals than dominant birds. As dominant individuals have the capacity to command more resources than subordinates, subordinate individuals may perceive a higher level of stress under adverse conditions than do dominants. Subordinates may sense less predictability and control than dominants, and thus their HPA axis may be "primed" to respond to emergency situations more so than dominants.

The HPA axis is not likely the only endocrine system associated with social position and social behaviour. While allofeeding behaviour was used as an indicator of social position (Chapter IV), and, as just discussed, social position may correlate with aspects of B secretion, allofeeding behaviour may also be related to Prl secretion. As allofeeding behaviour is identical in appearance to parental feeding of offspring, allofeeding behaviour may be facilitated by the same hormones as those associated with nurturing young. Prolactin is associated with parental behaviour in a wide range of species (e.g. Bengalese Finch: Seiler *et al.* 1992; Common Marmoset: Mota and Sousa 2000; Cotton-top Tamarin *Saguinus oedipus*: Ziegler *et al.* 2000; Florida Scrub-Jay: Schoech *et al.* 1996b; Golden Hamster *Mesocricetus auratus*: McCarthy *et al.* 1994; Macaroni *Eudyptes chrysolophus* and Gentoo *Pygoscelis papua* Penguins: Williams and Sharp 1993; mice: Voci and Carlson 1973; rabbit: Gonzalez- Mariscal *et al.* 2000; Spotted Sandpiper: Oring *et al.* 1986). As long as parents are caring for young, Prl levels often remain higher than basal, non-breeding levels (Dawson and Goldsmith 1982, Vleck *et al.* 1991). My measurements of high Prl titres in a some free-living WBBAs caught with a post-fledging young (Chapter III) is consistent with this pattern. Persistence of high Prl titres may facilitate prolonged feeding of young birds, and this may influence the retention of young within groups. Also, allofeeding behaviour seems to have been co-opted into an advertisement of social status (Chapter IV), and Prl potentially plays a role in facilitating allofeeding in this context as well.

It has also been suggested that alloparental behaviour is simply an unselected consequence of group-living (Jamieson 1989, 1991). If this is the case, there may be no relationship between plasma Prl titres and allofeeding behaviour, as allofeeding would be a direct response to stimuli from young birds (Jamieson 1989). However, if the endocrine system is involved in the facilitation of alloparental behaviour, it suggests that alloparental behaviour is in fact adaptive (Vleck *et al.* 1991). High Prl levels in WBBAs that exhibit a high frequency of allofeeding behaviour would support the hypothesis that allofeeding is adaptive. In this chapter I explored this possibility by examining Prl levels of captive birds in relation to their allofeeding class.

MATERIAL AND METHODS

Housing, Capture, and Blood Sampling

In order to examine the relationship between hormone levels and behavioural classes, I captured free-living WBBAs, held them in aviaries, performed behavioural observations, and collected blood samples. White-browed Babblers were housed and captured as described in Chapter IV. Blood samples for hormone analyses were taken more than 25 days after WBBAs were first released into the aviary and only if there was no change to group membership during this period. When blood was sampled, all birds in aviary groups were caught at the same time. Because all birds in a given group were disturbed upon entering the cage, both time of entry and time of blood collection were recorded. Because alarm calls from one group tended to agitate other groups, we either caught multiple groups concurrently with the aide of several people or single groups on different days. Blood samples were collected from Assemblage 1 in March, from Assemblage 2 in June (prior to the winter solstice), from Assemblage 3 in November, and from Assemblage 4 in May.

For comparative purposes, B levels of wild-caught WBBAs were also assessed. These samples were collected from breeding birds in September and October 1997 and non-breeding birds in April 1997. As collecting timed serial samples from multiple birds concurrently proved difficult for me when I worked alone, these samples were collected when only one or two birds were caught at a time (instead of large groups). Capture methods are outlined in Chapter II, and the blood collection protocol is described below.

Birds were bled as described in Chapter II. In the field blood samples for B measurements were taken between 0800 and 1100. In the aviary blood was sampled between 0900 and 1100. To assess B secretory responses to capture and handling, birds had blood sampled three times over an hour. The first B sample was taken as soon as possible after capture, and these bleeding times ranged from 3 –15 min post-disturbance. The second B sample was taken 30 ± 5 min post-disturbance, and the third 60 ± 5 min post-disturbance. Between sampling events, the birds were held in cloth bags. Plasma used for measuring Prl was taken from the initial 3-15 min post-disturbance blood sample.

Behavioural Analyses

To quantify social position, Behavioural Classes were designated as described in the previous chapter (IV). Behavioural observations were performed in the weeks immediately prior to blood sampling events. Each bird was classified into an Access Class and Allofeeding Class based on almost five hours of observation per group (20-30 min of observation per day). Behavioural observations also verified that there was little intragroup conflict and that relationships appeared stable. This study used groups from all four Assemblages of concurrently held groups.

Statistical Analysis

As only four WBBA groups could be housed concurrently, Assemblages (of groups) were held at different times over two years. Attempts were made to maintain a constant aviary environment, but ambient conditions varied among Assemblages. Therefore, when examining hormone levels in relation to Behavioural Classes, I first tested for an effect of Assemblage and, when significant, examined factors within Assemblages only (i.e. not across all four Assemblages). Access Class and Allofeeding

Class were analysed separately from one another. Because so few birds fell into the Allofeeding Class "Both" category, data from these birds were omitted from analysis.

When examining the relationship between Access or Allofeeding Classes and hormone levels, gender was taken into account, since gender is known to influence circulating hormone levels in birds (Adkins-Regan *et al.* 1990, Vleck *et al.* 1991, Wingfield and Lewis 1993). With some data sets I had sufficient sample sizes to use two-way designs with gender and Behavioural Class (Access or Allofeeding Class) as main effects. In other analyses certain combinations of Behavioural Class and gender were missing, and one-way designs were used to examine gender and Behavioural Class separately.

Analyses of B secretory responses to the capture and handling protocol were based on the three serial blood samples. These included (Fig. 5-1): initial, 30 and 60 min B levels, peak B value (the highest plasma B level of the three samples collected from each bird), rate of change in plasma B from initial to 30 min samples, 30 to 60 min samples, and initial to peak level, and total B response. Total B response was calculated as the integrated area under the curve. I approximated the area using Kaleidagraph™ graphical software, which calculated the cumulative area of the trapezoids formed by the data points. All samples could not be collected at precisely the same time; as a result, initial samples ranged from 3 to 15 min after entry into the cage, 30 min samples ranged from 29 to 34 min after entry into the cage, and 60 min samples ranged from 57 to 63 min after entry into the cage. To standardise the time intervals for the total B response analysis, initial and "60 min" values were interpolated to 15 and 57 minutes. I chose these time points because they allowed me to avoid extrapolating points beyond the endpoints of some bird's capture stress profiles.

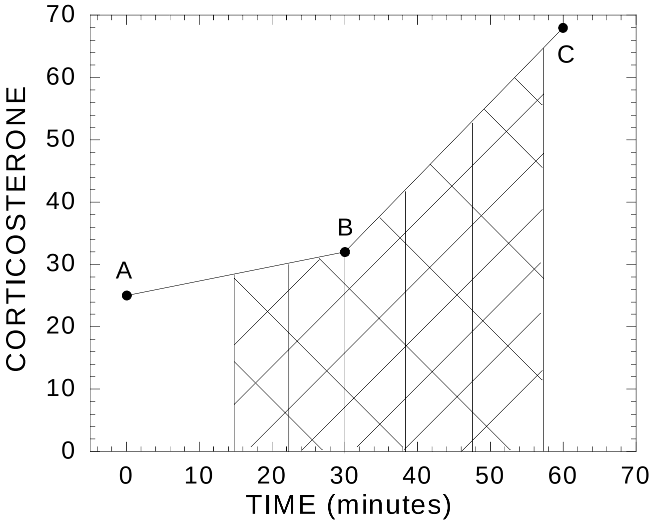


Figure 5-1. Depiction of factors used to describe the B response. Initial sample=A, 30 min sample=B, 60 min sample=C, peak=C, rate initial-30 min=B-A/change in time, rate 30-60 min=C-B/change in time, rate to peak=C-A/change in time, Area under the curve= regions bounded by cross-hatched lines. Note: Area under the curve was calculated from 15-57 mins for all birds (see text for details).

Student's t-tests were used to examine the hormonal differences between free-living and captive WBBAs. For most other hormonal analyses, ANOVA was used to examine the relationships between hormone levels and gender and Behavioural Class. One-way designs were used, except when there were sufficient data to include gender and another main effect in a two-way design (see Results). Because initial B samples were collected over a 12-min time range and because vertebrate B levels typically increase with duration of exposure to a given stressor (see Schoech *et al.* 1991), I compensated for the large variation in initial sample times in my analyses of initial B. To examine relationships between initial B levels and gender and Behavioural Class, initial B samples were analysed using ANCOVA, with time since capture as a covariate. ANCOVA is similar to ANOVA but corrects for variance attributable to a covariate. Prior to performing ANCOVAs, I verified the assumption that the slopes of the regression lines were equivalent across main effects (methods described in Zar 1999) using SYSTAT 7.0 for WINDOWS.

To illustrate initial B levels in graphical analyses, I plotted the individual residual values from a regression of initial B on time since capture for all birds sampled (following Schoech *et al.* 1991). For this graphical analysis, I first determined the common regression line and then determined the residual value associated with each individual's initial B values in relation to this common regression line. Residuals that were greater than zero indicated that the individual was "over-responsive" to capture stress, and residuals that were less than zero suggested the opposite; relative levels of the residuals reflected the degree to which individuals were "over or under-responsive." Residuals were used for graphical representation only; ANCOVAs were used to statistically analyse initial B levels (see above).

RESULTS

Corticosterone

1. Individual Variation

The B responses to capture and handling were surprisingly variable among captive WBBAs. The shapes of B capture-stress profiles included nearly all possible three point curves (Fig. 5-2). Nonetheless, in 79% of 62 WBBAs sampled, there was a positive slope between the initial and 30 min sample (Fig. 5-2 A, B, C, D); there was a negative slope in 16% (Fig. 5-2 F, G, H) and approximately a zero slope in 5% of cases (Fig. 5-2 E). Because capture-stress profiles were so variable, I used the five B measures described in Figure 5-1, in addition to initial, 30 and 60 min B levels, to describe individuals' responses to capture and handling.

2. Comparison of Aviary vs. Field Capture Stress Response

None of eight measures used to quantify the capture stress response differed significantly between captive and free-living WBBAs (Table 5-1).

Table 5-1. Comparison of eight measures of the capture stress response between free-living and captive WBBAs. [1]indicates that an ANCOVA was performed with time since disturbance as a covariate. All other analyses were performed with Student's t-tests. [2] indicates that scale is in ng/ml/min.

Capture Stress Measure	Free-living (Mean ± SE (ng/ml)	n	Captive (Mean ± SE (ng/ml)	n		
Initial B[1]	-1.233±1.360	14	0.254±0.702	68	$F_{1,81}= 2.405$	P=0.125
30 min B	24.612±1.903	17	23.841±1.293	64	$t_{79}= 0.286$	P=0.776
60 min B	24.571±5.637	19	28.021±2.013	57	$t_{74}= -0.932$	P=0.354
Rate initial-30min[2]	0.693±0.103	14	0.471±0.061	64	$t_{76}= 1.587$	P=0.114
Rate 30-60 min[2]	-0.023±0.068	17	0.039±0.050	53	$t_{68}= -0.631$	P=0.530
Peak B	29.456±2.063	14	31.388±1.527	53	$t_{65}= -0.611$	P=0.544
Rate to Peak B[2]	0.594±0.102	14	0.860±0.070	53	$t_{65}= -1.810$	P=0.075
Total B response	693.334±48.300	14	697.624±38.054	53	$t_{65}= -0.055$	P=0.957

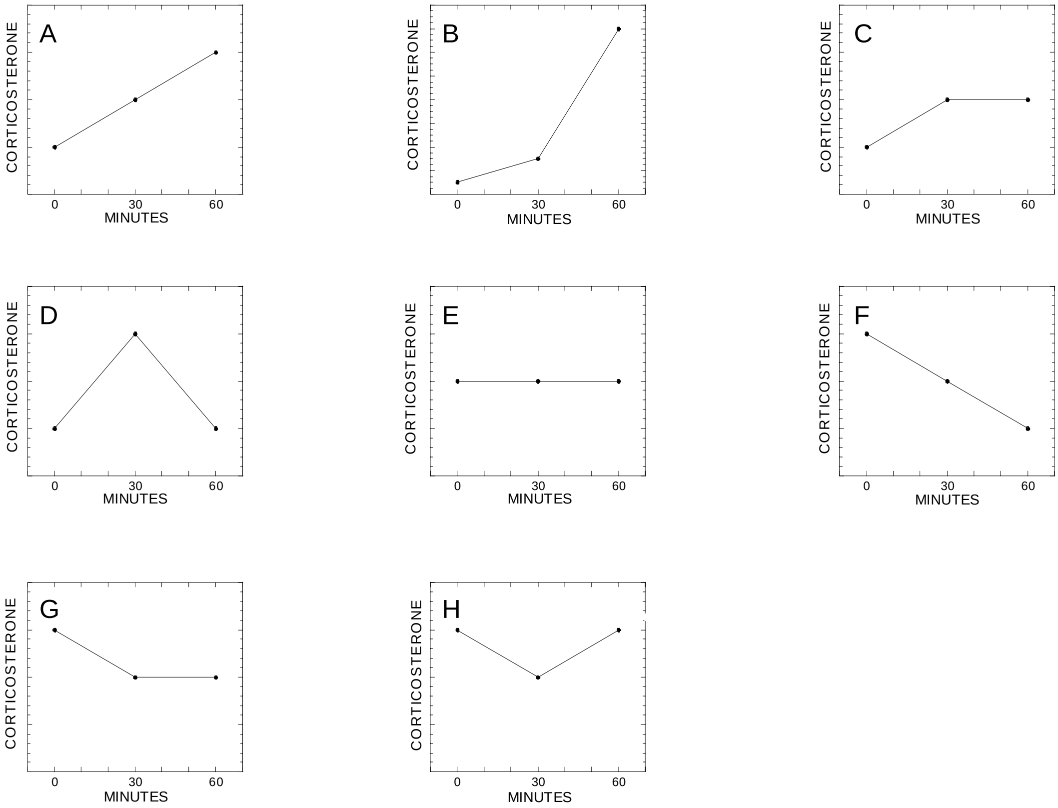


Figure 5-2. Range of individual capture-stress response patterns measured for members of stable WBBA groups. No scale is shown on the Y-axis because absolute levels of B differed among individuals exhibiting similarly shaped responses. Individuals' actual sample times varied, but 0, 30, and 60 min are used as stylistic approximations.

3. Body Condition

Neither body mass ($F_{3,64}$=1.279, P=0.289), furcular fat (Fisher's exact test, P=0.229), nor abdominal fat (Fisher's exact test, P=0.306) varied significantly with Assemblage; therefore, these data were combined across all Assemblages. None of the eight measures of the capture stress response were significantly associated with body mass, furcular fat, or abdominal fat (Table 5-2), nor were any trends apparent.

Table 5-2. Comparisons between 3 measures of body condition and 8 measures of the capture stress response in WBBAs. [1]indicates that corrections were made to account for an effect of time since capture on initial B levels.

Body Mass **Furcular Fat** **Abdominal Fat**

Corticosterone Measure	Pearson Correlation Coefficient	Bonferroni Probability	F Value	Prob-ability	F Value	Prob-ability
Initial B[1]	-0.238	0.080	$F_{6,45}=1.407$	0.233	$F_{3,60}=0.435$	0.729
30 min B	-0.041	0.747	$F_{6,56}=1.508$	0.192	$F_{3,59}=2.279$	0.089
60 min B	-0.047	0.728	$F_{6,50}=1.764$	0.126	$F_{2,54}=1.280$	0.286
Rate initial to 30 min	0.067	0.607	$F_{6,54}=0.344$	0.910	$F_{3,57}=1.906$	0.139
Rate 30 to 60 min	-0.053	0.705	$F_{6,46}=1.878$	0.105	$F_{2,50}=0.900$	0.413
Peak B	-0.122	0.393	$F_{6,46}=0.739$	0.621	$F_{2,48}=2.192$	0.122
Rate to Peak	-0.012	0.935	$F_{6,46}=0.765$	0.601	$F_{2,48}=0.219$	0.804
Area Under Curve	0.037	0.797	$F_{6,46}=1.065$	0.397	$F_{2,48}=1.618$	0.209

4. Inter-Assemblage Variation in Plasma B Levels

Corticosterone response to capture stress varied markedly among Assemblages. There were significant differences among Assemblages in four of eight measures of the capture-stress response and marginally non-significant differences in three more (Table 5-3). Due to these differences among Assemblages, further statistical analyses compare B levels within Assemblages only.

Table 5-3. Inter-Assemblage variation in B secretion during the serial capture stress protocol. Six ANOVAs and one ANCOVA[1] examining the effect of Assemblage on B measures. Asterisk (*) indicates significant differences (P<0.05).

	Corticosterone Measure	
Initial B[1]	$F_{3,63}=10.677$	$P<0.001$*
30-minute B	$F_{3,62}=5.195$	$P=0.003$*
60-minute B	$F_{3,55}=4.685$	$P=0.006$*
Rate initial-30 minutes	$F_{3,60}=2.046$	$P=0.117$
Rate 30-60 minutes	$F_{3,51}=2.071$	$P=0.116$
Peak B	$F_{3,49}=2.099$	$P=0.112$
Rate to Peak B	$F_{3,49}=3.107$	$P=0.035$*
Total B Response	$F_{3,42}=0.636$	$P=0.596$

5. Plasma B levels in relation to Access Class

I examined capture-stress responses in relation to Access Class within each Assemblage. Figures 5-3, 5-4, and 5-5 depict intra-Assemblage patterns of the eight measures of B-secretory patterns during the capture-stress response in each Access Class. Only one B measure exhibited a consistent pattern across all Assemblages: the rate of change from “0-PEAK” was consistently lowest in Access Class 1 (Fig. 5-5). This trend was not significant in any of the four Assemblages (see below).

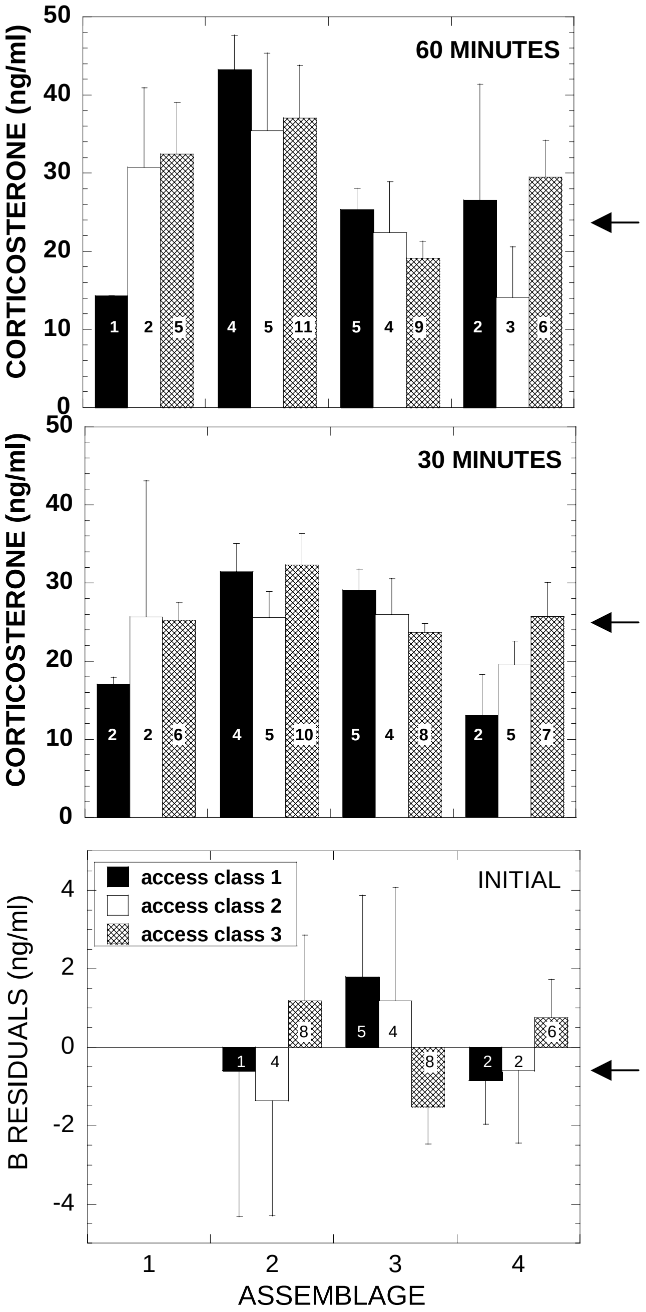


Figure 5-3. Comparisons of 3 B measures among 3 Access Classes in 4 Assemblages. Corticosterone measures include: B levels at 30 and 60 min and the residuals from a regression of initial B level on time since disturbance. In Assemblage 1 this regression was not significant, and no data are shown. Height of columns indicates means, and bars represent one standard error. Numbers inside columns indicate sample sizes. Arrows indicate mean values from free-living WBBAs.

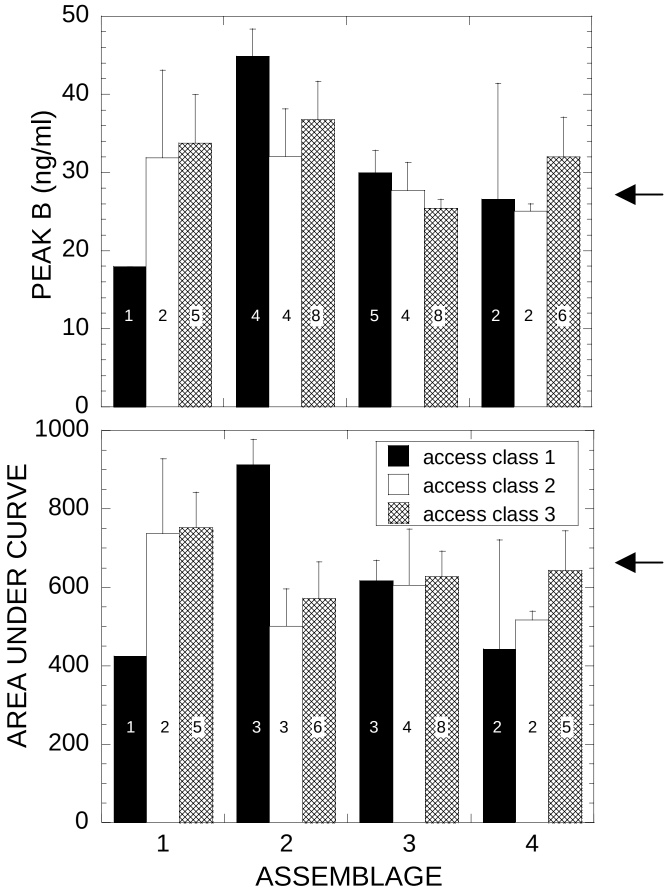


Figure 5-4. Mean peak B and mean total B response (measured by area under the curve formed by 3 serial B samples) in each Access Class in each Assemblage of WBBAs. Height of columns indicates means, and bars represent one standard error. Numbers inside columns indicate sample sizes. Arrows indicate mean values from free-living WBBAs.

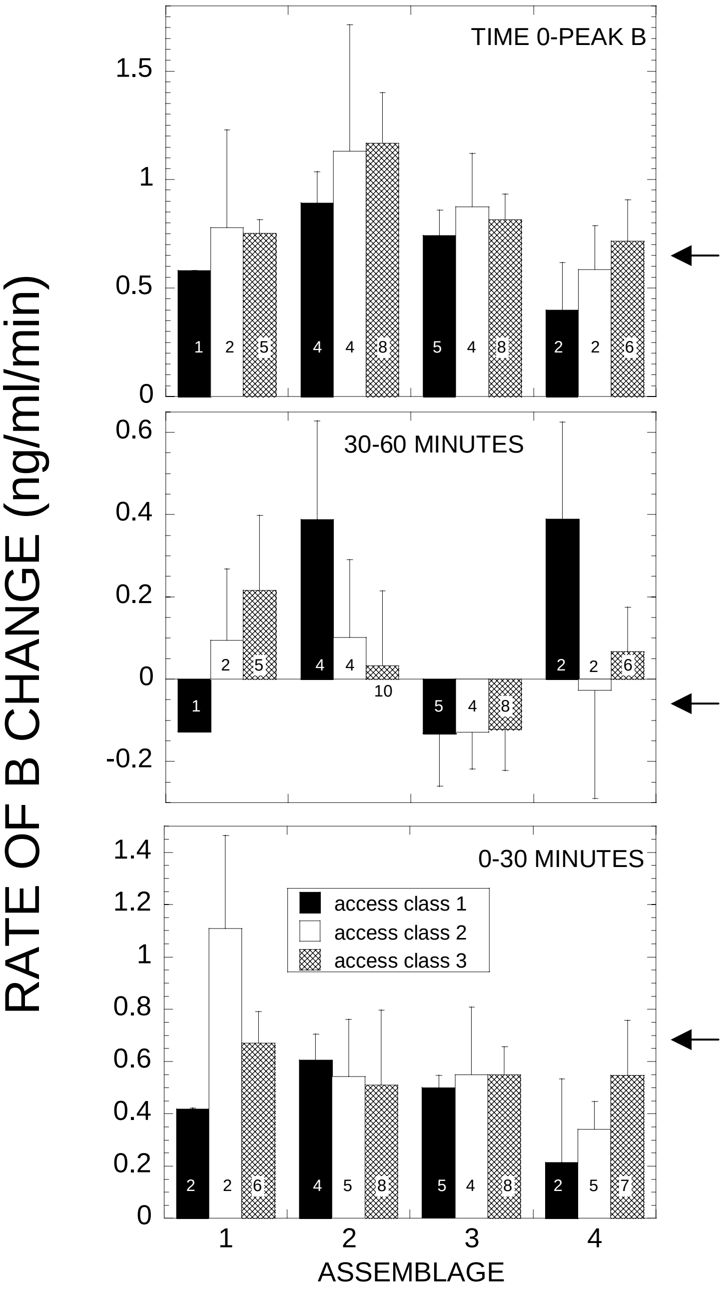


Figure 5-5. Mean rate of change of B levels at 3 intervals along the capture-stress profile in each Access Class in each Assemblage of WBBAs. Height of columns indicates means, and bars represent one standard error. Numbers inside columns indicate sample sizes. Arrows indicate mean values from free-living WBBAs.

Although I found only one consistent pattern across all Assemblages, I examined the intra-Assemblage statistical variation of each B measure associated with Access Class and gender. Two-way designs with gender and Access Class were possible only in Assemblages 2 and 3. In Assemblage 1 and 4, in which there was insufficient data for a two-way design, each main effect was examined independently.

To examine the effect of gender or Access Class on the eight measures of the capture-stress response, 36 one-way ANOVAs and 4 one-way ANCOVAs were performed on Assemblages 1 and 4. All yielded non-significant P values (all $P \geq 0.120$). To examine the combined effects of gender and Access Class on the eight measures of the capture-stress response, 14 two-way ANOVAs and 2 two-way ANCOVAs were performed on Assemblages 2 and 3. Only one significant effect ($P<0.05$) was identified: in Assemblage 2 initial B (with time since capture as a covariate) was significantly higher in males than females ($F_{1,12}=8.060$, $P=0.015$).

6. Plasma B levels in relation to Allofeeding Class

I examined the capture-stress response in relation to Allofeeding Class within each of three Assemblages (WBBAs in Assemblage 1 did not exhibit sufficient allofeeding behaviour to allow analysis). Comparisons of Intra-Assemblage patterns of the eight measures of the capture-stress response among three Allofeeding Class revealed some consistent trends (Figs. 5-6, 5-7, and 5-8).

In all Assemblages mean initial B levels (represented by the residuals from a regression of B with time since capture) tended to be higher in Feeders than in birds that neither fed nor received (Nonallos) (Fig. 5-6). Mean B levels measured at 60 min post-capture tended to be lowest in Nonallos (Fig. 5-6). Feeders tended to have higher total B response than Nonallos (Fig. 5-7). Nonallos tended to have higher mean rates of change from basal levels to peak B levels than Feeders (Fig. 5-8), and Nonallos also tended to have the lowest mean rate of B change from 30 to 60 min (Fig. 5-8).

To examine the statistical significance of these relationships, one-way and two-way ANOVAs and ANCOVAs were used. Of the trends described above, only two approached statistical significance: differences in initial B levels among Allofeeding Classes in Assemblage 3 ($F_{2,10}$=3.635, P=0.065) and differences in 60 min post-capture B in Assemblage 2 ($F_{2,12}$=3.0217, P=0.087). All other relationships were non-significant ($P \geq 0.169$). However, in Assemblage 2 there was a significant interaction in initial B levels between gender and Allofeeding Class ($F_{2,11}$=4.913, P=0.030). Among males Nonallos had the highest initial B levels, but among females Feeders had the highest B levels.

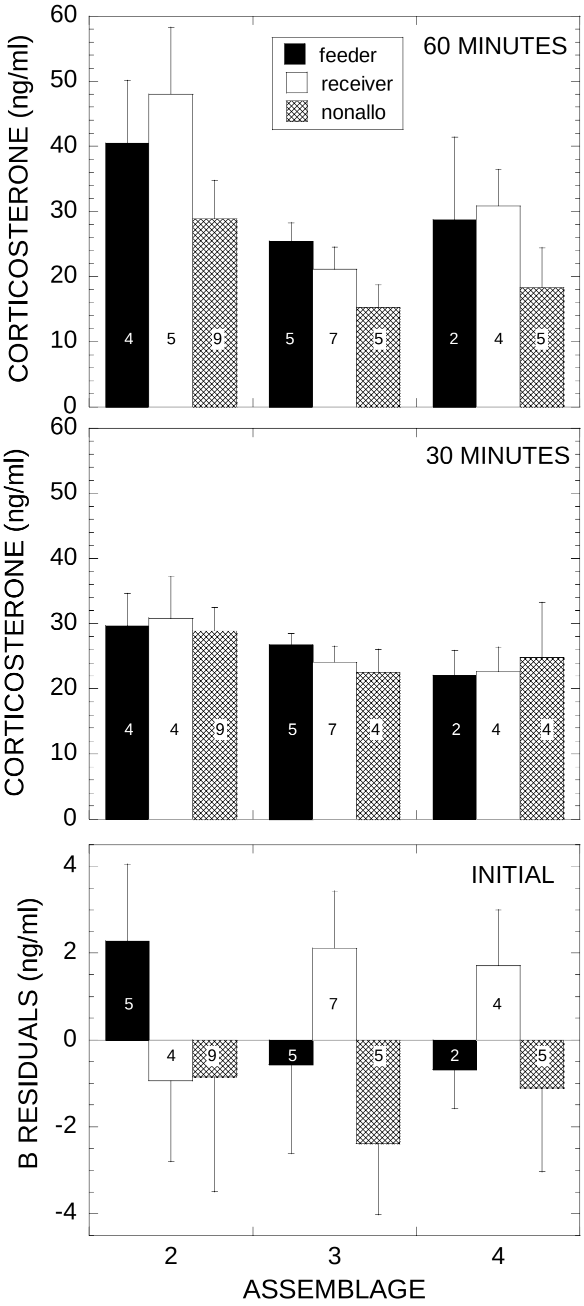


Figure 5-6. Comparison of initial, 30 min, and 60 min B levels among Allofeeding Classes in 3 Assemblages. Initial B levels were regressed on time since capture, and residuals are shown. Numbers inside columns indicate sample sizes. Height of columns indicates means, and bars represent one standard error.

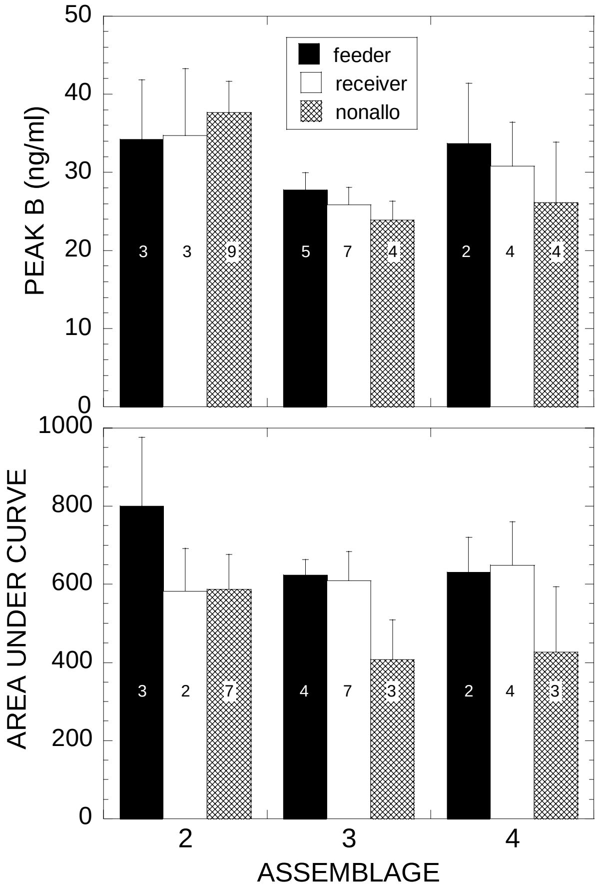


Figure 5-7. Comparison among Allofeeding Classes of peak B and total B response (measured as area under the curve formed by 3 serial B samples) in 3 Assemblages. Numbers inside columns indicate sample sizes. Height of columns indicates means, and bars represent one standard error.

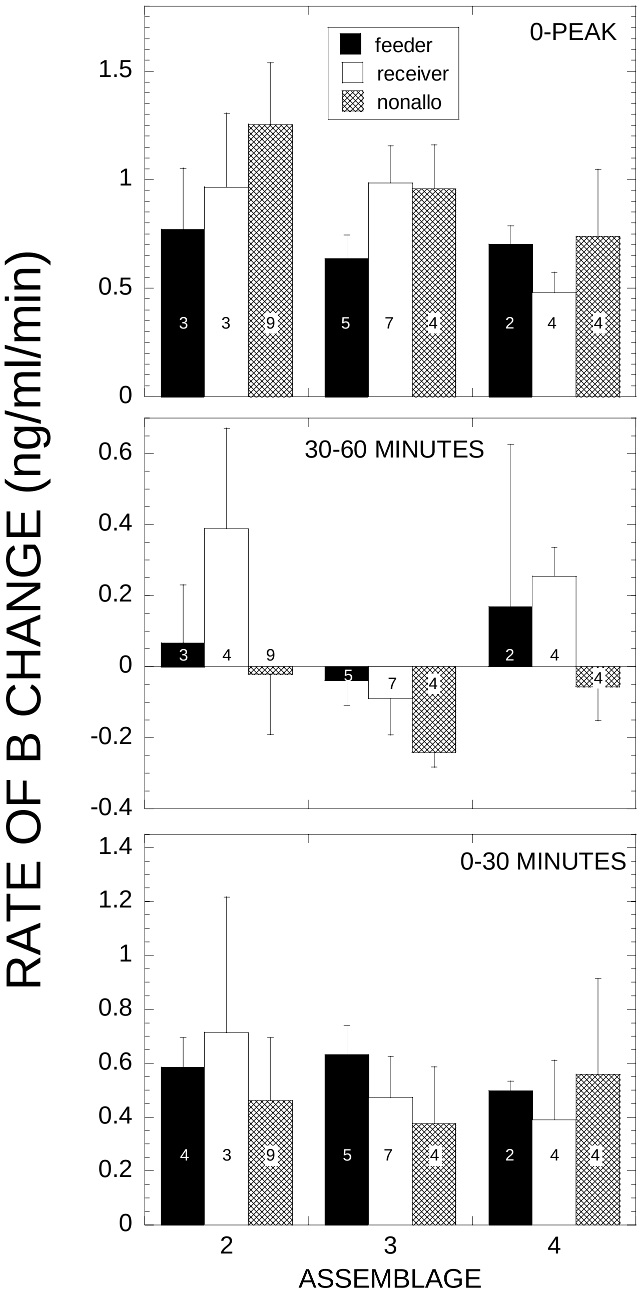


Figure 5-8. Comparisons among Allofeeding Classes of mean rate of change of B levels at 3 intervals during the one-hour capture-stress protocol in 3 Assemblages. Numbers inside columns indicate sample sizes. Height of columns indicates means, and bars represent one standard error.

Prolactin

There were significant differences in Prl levels among Assemblages held at different times of the year ($F_{3,41}$=4.794, P=0.006). In male WBBAs, differences in Prl levels among months (or Assemblages) were marginally non-significant ($F_{3,17}$=2.975, P=0.061), but in females Prl levels were significantly variable among months ($F_{3,20}$=6.954, P=0.002). In captive females Prl titres were higher in November than in March and May (Tukey's pairwise comparison P=0.005, P=0.008, respectively). In captive males Prl levels tended to be higher in May and November than March and June (Fig. 5-9).

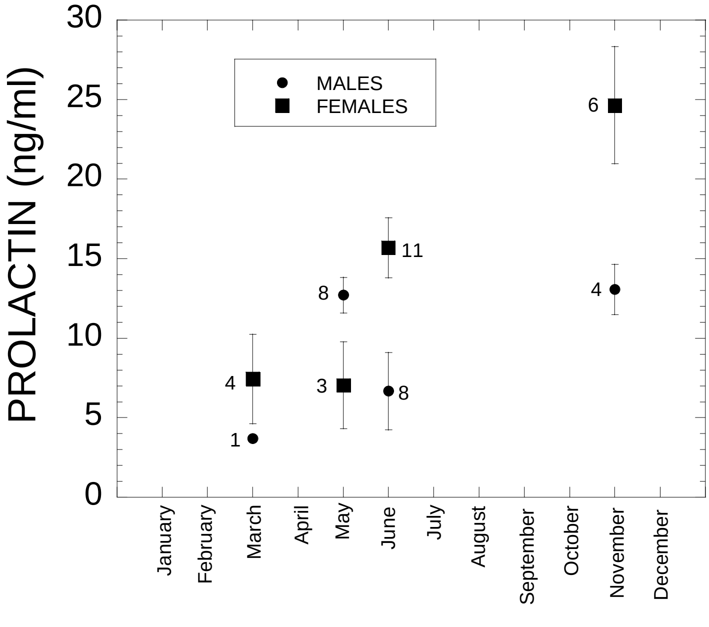


Figure 5-9. Plasma Prl levels in captive male and female WBBAs in relation to time of year. Points represent means, and bars indicate one standard error. Assemblage 1 was sampled in March, Assemblage 2 in June, Assemblage 3 in November, and Assemblage 4 in May. Numbers inside graph indicate sample sizes.

Because of inter-assemblage variability (see above), Prl levels were examined separately within each Assemblage. Data limitations prevented application of two-way models analysing the combined effect of gender and Access or Allofeeding Class on Prl levels. However, one-way designs indicated significant differences between males and females in each of three Assemblages, but none among Access or Allofeeding Classes (Table 5-4). Because of small sample sizes for Prl in Assemblage 1 (only five samples), I omitted data from Assemblage 1 from these and further analyses.

Table 5-4. Comparisons of Prl titres between genders, Access Classes, and Allofeeding Classes. In each Assemblage one-way ANOVAs were used. Superscripted asterisk (*) indicates significant difference ($P<0.05$).

Assemblage	Mean ± SE (ng/ml)		Gender	Access Class	Allofeeding Class
	♂	♀			
2 (June)	6.675 ± 2.418	15.682 ± 1.881	$F_{1,17}=8.914$ P=0.008*	$F_{2,16}=0.681$ P=0.520	$F_{2,14}=0.194$ P=0.826
3 (Nov.)	13.068 ± 1.584	24.647 ± 3.687	$F_{1,8}=5.879$ P=0.042*	$F_{2,7}=1.012$ P=0.411	$F_{2,5}=0.715$ P=0.533
4 (May)	12.704 ± 1.113	7.037 ± 2.715	$F_{1,9}=5.552$ P=0.043*	$F_{2,8}=1.502$ P=0.279	$F_{2,8}=0.380$ P=0.696

Because interactions between gender and Behavioural Classes could not be examined in a 2-way design, I graphed data to investigate whether gender differences may have obscured patterns among Behavioural Classes (Figs. 5-10 and 5-11). There were no consistent patterns within either the male or female subset of data. Furthermore, these data revealed inconsistent patterns within Behavioural Classes; for instance, in Assemblage 2 females in Access Class 3 had higher Prl than males in the same Access Class, but in Assemblage 4 this pattern was reversed (Fig. 5-10).

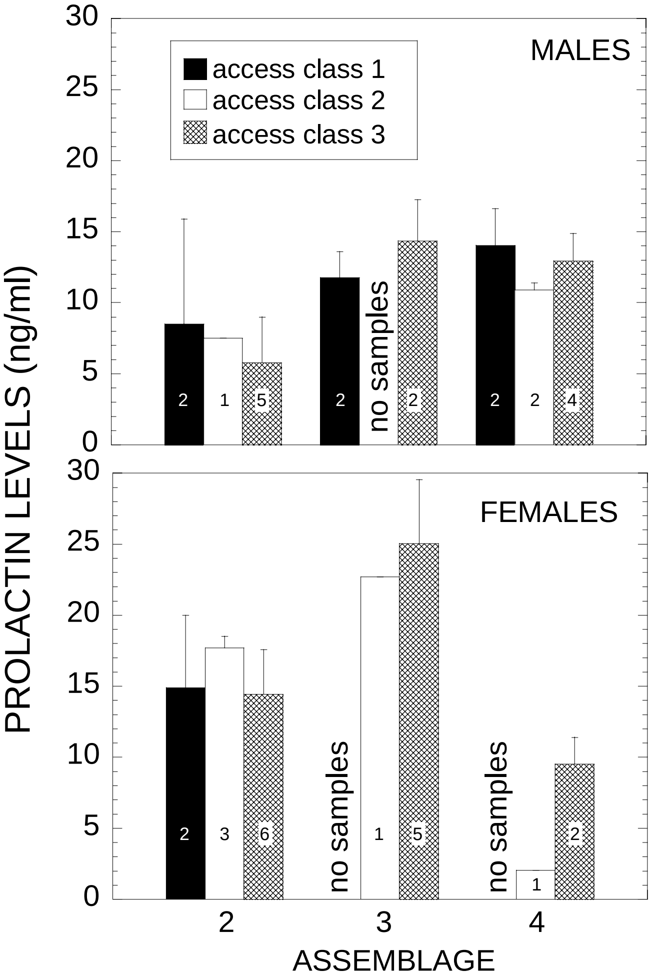


Figure 5-10. Comparison of Prl levels among Access Classes. Males and females examined separately within each Assemblage. Numbers inside columns indicate sample sizes. Height of columns indicates means, and bars

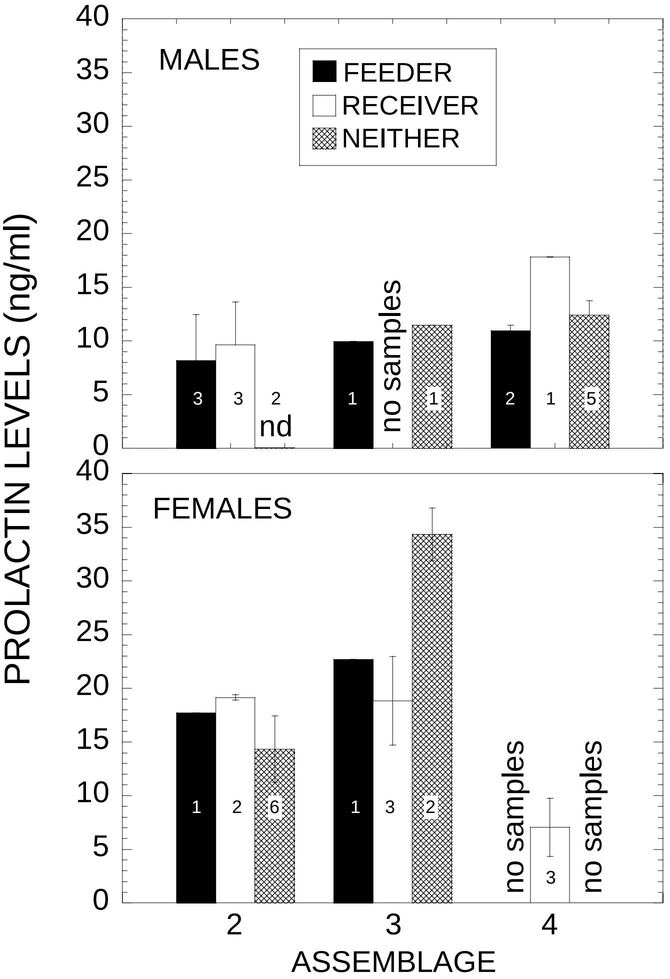


Figure 5-11. Comparison of Prl levels among Allofeeding Classes. Males and females examined separately within each Assemblage. Height of columns indicate means, and bars represent one standard error. “nd” indicates that Prl was not detected in the samples that were analysed. Numbers inside columns indicate sample sizes.

DISCUSSION

Corticosterone Levels and Capture Stress

Among WBBAs sampled, there was remarkable variability in capture stress profiles. This variability is not likely an artifact of captivity, as multiple measures of the B response to capture and handling were not significantly different between captive and free-living WBBAs. Many passerines exhibit a stereotypical pattern of B secretion in response to capture and handling stress: baseline B levels are low, followed by a rapid increase within the first 10 minutes of capture, and B levels plateau within 30 to 60 minutes of capture (e.g. Florida Scrub-Jays: Schoech *et al.* 1997; Gambel's White-crowned Sparrow: Astheimer *et al.* 1994; Lapland Longspurs *Calcarius lapponica*: Astheimer *et al.* 1995). In captive WBBAs, almost every imaginable pattern was exhibited over the 60-minute sampling period, although the majority of birds exhibited an increase in B from the initial to the 30-minute sample.

Detailed examination of these disparate patterns revealed variation that could be attributed to Assemblage. Each Assemblage of WBBA groups was held in the aviary at a different times during the three years of study. Accordingly, each Assemblage experienced differences in temperature, day length, and rainfall. Environmental variables, such as temperature, atmospheric pressure, and inclement weather are know to affect the avian stress response (Siegel 1980, Wingfield 1985). White-browed Babblers held during the autumn and winter months (those in Assemblages 2 and 4) seemed to exhibit a greater stress response than those held during the spring and summer. Perhaps low temperature and/or short days heightened the stress response. It is unlikely that these differences were influenced by breeding readiness, as the breeding season began in the winter months and extended through the beginning of summer (Chapter II), although captive birds did not breed.

Even within Assemblages WBBAs' capture-stress responses were highly variable. Body condition has been shown to affect the adrenal's response to stress in some species; heavier birds often have lower adrenocortical secretion when compared to lighter birds (Wingfield 1994b, Wingfield *et al.* 1995, Schoech *et al.* 1997). In the WBBA, there was a suggestion of a trend (Table 5-2, P=0.080) for heavier birds to have lower initial B measurements (adjusted to account for the effect of time since capture). However, there were no significant differences in the capture stress responses that could be attributed to body mass or fat levels. Body condition did not help to explain the variable stress response in WBBAs.

Social factors may have influenced WBBAs stress response. While the exact nature of the relationship between social status and glucocorticoid levels remains equivocal, there is considerable evidence that adrenocortical activity is influenced by psychosocial stimulation. Some studies have suggested that reduced access to resources and/or intimidation or harassment by dominants acts to heighten the HPA stress response in subordinate individuals (e.g. mice: Bronson 1973; Talapoin Monkeys: Eberhart *et al.* 1983; White-throated Sparrows: Schwabl *et al.* 1988). While other studies have found that dominants exhibit a greater HPA axis response to stress than subordinates because dominants engage in more aggressive contests than subordinates (e.g. African Elephants: Foley *et al.* 2001; African Wild Dogs and Dwarf Mongooses: Creel *et al.* 1996; female Common Marmosets: Saltzman *et al.* 1994). In an attempt to address such confounding findings, Abbott *et al*. (2003) examined multiple social and kinship factors in 10 primate species and found that rank related differences in F levels relied on two important factors. Subordinates had higher F levels than dominants (1) when they experienced higher rates of stressors and (2) when they had lower levels of social support (Abbott *et al*. 2003).

In captive WBBAs that were provided with abundant food resources, exhibited minimal aggressive behaviour, and seemed to have strong, stable social bonds, individuals' social status did not seem to be related to their adrenocortical response to capture and handling stress. While some trends were uncovered, the ambiguity of my findings yielded no conclusive evidence suggesting a relationship between social status and adrenocortical responsiveness. My findings did concur with some studies that suggested that there was little association between adrenocortical response and social rank in stable groups of animals (e.g. Florida Scrub-Jays: Schoech *et al.* 1997; House Sparrows: Hegner and Wingfield 1987b; Talapoin monkeys: Yodyinyuad *et al.* 1982; Vervet monkeys *Cercoppithecus aethiops*: McGuire *et al.* 1986; White-browed Sparrow Weavers: Wingfield *et al.* 1991). Hegner and Wingfield (1987a) suggested that stable social relationships may be maintained by non-hormonal mechanisms, such as social inertia and/or social recognition. As captive groups of WBBAs were initially caught as cohesive, free-living groups and membership remained stable in captivity, social inertia and/or social recognition may well have contributed to the maintenance of social relationships.

Furthermore, the extent to which glucocorticoid levels reflect social rank may be related to how adverse subordinance is (Sapolsky 1993, Abbott *et al*. 2003). Depending on rates of aggression, being subordinate can be worse in some social groups than in others. Captive WBBAs in stable groups did not engage in potentially costly and stressful exhibitions of social rank, such as fighting, but rather seemed to reinforce their social position with calls and ritualized behaviours, such as allofeeding. Subordinate birds did not seem to suffer as a result of their social position. Perhaps rank-related differences in WBBAs' adrenocortical responsiveness would only become apparent if dominance were asserted aggressively or if subordinate birds were excluded from necessary resources, such as food or shelter.

It has also been suggested that in complex social groups and/or in animals in stable groups, dominance may be less intense and the hormone-behaviour relationships may be less apparent than in animals in unstable, dyadic relationships (Coe *et al.* 1979). Whereas the relative dominance rank may be obvious in winners and losers of paired dyadic encounters, such relationships may be far more ambiguous in larger groups of animals. In groups of WBBAs, there is a high degree of social complexity. For instance, there may be alliances between courting or parent-offspring pairs, increased competition between same sex, same age siblings, and/or large birds may hold high social positions (Chapter IV). Whereas fighting prowess may be the primary contributor to establishing rank in animals in unstable, dyadic relationships, multiple factors likely contribute to WBBAs' social relationships. Further, Carlson *et al.* (2004) points out that multiple variables, such as access to unrelated breeding partners, weight, and age may all factor into an animal's endocrine response and must be considered when examining differences among group members. The relationship between hormones and behaviour may be less pronounced or more difficult to discern when multiple factors contribute to social relationships, as they do in WBBAs.

Prolactin

Variation in Prl levels among the WBBA Assemblages may have been due to seasonal influences. Photoperiod affects Prl levels in many birds, and vernal increases in Prl secretion are common (Ebling *et al.* 1982, Hiatt *et al.* 1987, Silverin and Goldsmith 1997). Prolactin levels in free-living WBBAs' showed seasonal variation; Prolactin levels began to increase above basal levels in June and increased throughout the summer (Fig. 3-20). It follows that WBBAs held in the aviary at different times of the year would also show seasonal variation in Prl titres.

Gender differences in Prl secretion found in captive WBBAs have been noted in many other birds. In species in which females provide more parental care than males, Prl levels are typically higher in females (Dawson and Goldsmith 1982, Hiatt *et al.* 1987). In free-living WBBAs, females are the sole incubator, and their mean Prl levels are higher than those of males in every breeding month when I had samples from both genders (Fig. 3-10). This pattern held true in all but one of the three Assemblages of captive WBBAs; females had significantly higher Prl levels than males. Although WBBAs did not breed in captivity, photoperiodic cues coincident with the breeding season may still have affected Prl levels, as has been shown in Starlings (Chakraborty 1995). In Assemblage 4 males had significantly higher Prl levels than females, but only three females were sampled in this month and all were Receivers in Access Class 2 or 3. This subset of data may not have provided an accurate representation of the population.

Access Class did not correlate with Prl levels in WBBAs. While there is little evidence of a relationship in birds, in some mammals Prl secretion is correlated with elevated stress levels that often accompany low social position (Meyerhoff *et al.* 1988, Gala 1990, Dijkstra *et al.* 1992). It is thought that elevations in Prl levels following stress may stimulate the immune system and help counter the immunosuppressive effects of adrenocortical hormones released in response to stress (Hirschhorn *et al.* 1963, Spangelo *et al.* 1985). As social position does not seem to be correlated with stress levels (as measured by B secretion) in WBBAs, social position may not induce stress-related variation in Prl titres.

Numerous studies have demonstrated a relationship between parental behaviour and Prl levels (Lea *et al.* 1981, Gratto-Trevor *et al.* 1990, Richard-Yris *et al.* 1998). Behaviours associated with feeding brooding mates or young may be maintained or stimulated by Prl (Lehrman 1961, Silverin and Goldsmith 1984, Vleck *et al.* 1991, Schoech *et al.* 1996b), and alloparental behaviour, including feeding of young, has been associated with elevated Prl levels in the Harris' Hawk and Florida Scrub-Jay (Vleck *et al.* 1991, Schoech *et al.* 1996b). However, Allofeeding Class in captive WBBAs did not seem to be related to Prl levels. Prolactin levels did not vary significantly among Allofeeding Classes; in fact, mean Prl levels in Feeders, tended to lower than one or both of the other Allofeeding Classes in all Assemblages (see Fig. 5-11).

As discussed in Chapter IV, some allofeeding may have taken place between parents and offspring. Even though Prl probably facilitated parental feeding when their offspring were in the nest or perhaps when recently fledged (Chapter III), this hormone-behaviour relationship did not seem to persist into adulthood. Although an allofeeding event appeared identical to a parent provisioning its offspring, this behaviour may have held distinct purposes depending on the context in which it was exhibited. Allofeeding may not have been related to the nutritional needs of the recipient, but rather to social factors. As discussed in Chapter IV, allofeeding may have acted to help establish and reinforce social position. High Prl levels may only be associated with feeding behaviour when coupled with parental care and not when dissociated from breeding condition. Although some behaviours appear similar, it is likely that different mechanisms help regulate these behaviours at different stages in an animal's life cycle (Wingfield *et al.* 1997).

Furthermore, in stable groups of animals, non-hormonal mechanisms, such as social inertia and social recognition, may be more important in maintaining group structure and cohesion than hormonal cues (Hegner and Wingfield 1987). While allofeeding behaviour likely acts as a signal to reinforce the established social structure (Chapter IV), social inertia and social recognition may also play a large role in maintaining group structure. As a result there may be little need for hormonal signals to help maintain stable social structures in persistent groups of WBBAs.

CONCLUSIONS

In stable groups of WBBAs, there seems to be no relationship between stress and social position. Neither dominance nor subordinance were associated with significant or consistent variation in the B secretory response to capture and handling stress. As social position does not carry with it an inequitable division of necessary resources (such as food or shelter) or a significant amount of aggressive behaviour (either expressed or received), it may not be stressful to be dominant or subordinate in stable groups of WBBAs. Furthermore, among stable groups of WBBAs, group stability is likely maintained by non-hormonal factors such as social recognition and social inertia and reinforced by behavioural (rather than hormonal) signals. The high degree of inter-individual variation in B response to capture and handling stress in WBBAs may be related to a myriad of factors, including age, body size, length of time with the group, kinship to others in the group, or presence or absence of a stable pair bond within the group. The complexity of WBBAs' social system may preclude an accurate examination of the factors contributing to the variation in WBBA's B stress response.

I found no evidence to suggest that allofeeding behaviour in captive, stable groups of WBBAs is associated with Prl titres. This finding neither refutes the unselected consequence theory, proposed by Jamieson (1989), nor provides support for an adaptive advantage to allofeeding behaviour among adult WBBAs. Allofeeding behaviour in WBBAs could simply be an unselected consequence of group living (Jamieson 1989); however, my behavioural observations suggest that it acts as an important social signal. Vleck *et al.* (1991) suggested that a behavioural trait, such as allofeeding, may be considered adaptive if the endocrine system has been modified via evolutionary processes to promote the given behaviour. As Prl secretion does not appear to facilitate allofeeding behaviour in captive WBBAs, this lends no evidence to support the hypothesis that allofeeding behaviour is an adaptive trait in WBBAs.

However, as I’ve hypothesized that allofeeding behaviour is a signal of social status co-opted from parental behaviour, rather than an extension of parental behaviour, there may be no basis for expecting a relationship between Prl levels and allofeeding behaviour when it is practiced between adults WBBAs. As suggested by Wingfield *et al.* (1997), different mechanisms are probably responsible for regulating the same behaviour when performed at different life cycle stages.

Chapter VI. Physiological Correlates of Social Behaviour During Periods of Social Instability

INTRODUCTION

During long-term stable conditions, hormone-behaviour relationships may be less apparent than during the establishment of social relationships (Coe *et al.* 1979). Some studies have shown that when social relationships are forming, hormone levels correlate with emerging social positions, but that once relationships are established, hormone levels are independent of these stable social relationships (Ramenofsky 1984, Schwabl *et al.* 1988). It has been suggested that stable social relationships are maintained by learned response biases, social inertia, and social recognition, instead of by hormonal mechanisms (Ramenofsky 1984, Hegner and Wingfield 1987b). However, when social relationships are unstable or social positions are contested, hormonal mechanisms may play a role in establishing social roles (Wingfield 1984b).

In WBBA groups with well-established social organisations, I found no relationship between hormone levels and social position (Chapter V), as has been found in a number of species in stable social groups (Mcguire *et al.* 1986, Hegner and Wingfield 1987b, Schoech *et al.* 1997). In the current chapter, I examine the relationship between hormones and behaviours during times of experimentally-induced social instability. Such experimentally-induced social changes mimic those that may be experienced by free-living WBBAs. While group membership was largely stable, members did die or emigrate and new members were integrated into the group (Chapter II). Presumably, such occasions instigated some social restructuring within the group.

Hormone levels were measured under two different conditions: (1) in groups where dominant members had been removed and (2) in groups where some members were removed and replaced with unfamiliar individuals. These conditions were chosen because they mimic events that occur in the wild and because there is evidence from studies on other species that one or both of these conditions might reveal a hormonal association to social reestablishment (Hegner and Wingfield 1987b, Gust *et al.* 1993, Saltzman *et al.* 1994).

To gauge hormonal responses to social instability, I measured changes in steroid hormones: B, T, and E2. Corticosterone, the major glucocorticoid in birds, was measured to gauge WBBAs' physiological response to social stress that was provoked by social perturbations. Testosterone and E2 were measured to investigate the interaction between reproductive steroids, social position, and behaviour in an unstable social environment.

Glucocorticoids are known to rise in response to both physical and psychological stress (Harvey *et al.* 1984, Myerhoff *et al.* 1988). In particular, psychological stress during hierarchy formation or social instability elicits an increase in glucocorticoid levels in a number of animals (e.g. mice: Ely and Henry 1978; Rhesus Monkeys: Gust *et al.* 1993; Squirrel Monkeys: Levine 1993; Vervet Monkeys: McGuire *et al.* 1986). Furthermore, social relationships seem to affect an individual's ability to cope with stress (Levine 1993). Typically, the presence of familiar social partners and stable social relationships ameliorate an individual's HPA response to a stressor (Levine 1993, Abbott *et al.* 2003). Alternately, separation from familiar social partners or the formation of new social relationships activate the HPA axis and result in increased glucocorticoid levels (Levine 1993). To examine how social instability affects the stress response in WBBAs, I compared the B-response to capture and handling stress in stable groups to groups in which I had altered membership.

Many studies of male birds have found that social behaviours, as well as sexual behaviours, are under strong control by T (e.g. Saino and Moller 1995). However, results of studies examining the relationship between T and social status have proven inconsistent. Some studies have found a positive correlation between T and dominance (e.g. Rose *et al.* 1971, Yodyingyuad *et al.* 1982, Ramenofsky 1984), while others have not (e.g. Rohwer and Wingfield 1981, Creel *et al.* 1992). A closer examination of these studies suggests that T levels are elevated and correlate with dominance while relationships are being established, but that T levels decrease and do not correlate with social position after relationships have stabilised (Ramenofsky 1984, Wingfield 1984, Hegner and Wingfield 1987b). In social groups of WBBAs', do high T levels correlate with social dominance during times of social restructuring?

Comparatively little attention has been paid to the function of E2 in influencing social status. However, there is evidence that females' plasma levels of E2 may also be correlated with social factors. For example, Dwarf Mongoose alpha females typically had higher E2 than subordinates, and this difference was magnified during estrus (Creel *et al.* 1992). In female Florida Scrub-Jays, E2 levels varied according to breeding status (i.e. helper vs. breeder) and stage in the breeding cycle. Furthermore, Florida Scrub-Jay female helpers caught away from their home territories had E2 levels that were almost 10 times higher than those caught on their home territories (Schoech 1998). In social groups of WBBAs', do E2 levels vary in relation to social position and behaviour?

MATERIAL AND METHODS

Study Design

To examine the effect of social perturbations on WBBAs' hormone levels, I manipulated group composition. Because I was interested in situations that could occur in the wild, I chose to look at both removal of members from a group (mimicking death or emigration in the wild) and introduction of new group members (mimicking immigration). I designed two different protocols to examine these situations. In the Removal Experiment, I examined the effect of removal of group members on behaviours and hormone levels of the remaining members. In the Exchange Experiment, I explored the behavioural and hormonal response to the exchange of birds between groups. . Because both males and females emigrated in the wild (Chapter II), I did not discriminate between genders when deciding which birds to remove or exchange.

The protocol for capturing birds and sampling blood was identical to that described in Chapters II and V, except when otherwise noted. Plasma used for measuring T and E2 was taken from the first blood sample of the 3-sample capture stress series.

1. Removal Experiment

In this experiment I removed high ranking members (based on my assessment of Behavioural Classes) from groups in an effort to elicit social instability and subsequent restructuring. The Removal Experiment was performed on four concurrently held groups of WBBAs (Assemblage 3), and blood samples were collected from each group on three occasions (Table 6-1). In an attempt to avoid ambiguity and stem confusion, I will use the term "bleed date" to refer to all blood samples taken at the same point in the experiment (e.g. 1 day post-removal). The protocol for capturing birds and sampling blood was identical to that described in Chapters II and V. Two of the four concurrently housed groups in Assemblage 3 served as controls, and their membership remained stable throughout the course of the experiment.

Table 6-1. Schedule of blood sampling, treatment, and hormones evaluated in the Removal Experiment. Each group was bled only once per bleed date, but samples were collected from only a single group on each day, thus a range of bleed dates is reported.

	Bleed Date1	**Bleed Date 2**	**Bleed Date 3**
WBBA Group	13-14 Nov.	26-29 Nov.	15-16 Dec.
20	Control B	Control B,T	Control B,T
21	Control B	Control B,T	Control B,T
22	Pre-Removal B	1 Day Post-Removal B,T	18 Days Post-Removal B,T
23	Pre-Removal B	1 Day Post-Removal B,T	19 Days Post-Removal B,T

In the two experimental groups, I removed the two to three “highest ranking” members in each group, thus reducing group membership to sizes equivalent to control groups (3 and 4 members per group). I removed one male and one female, both in Access Class 1, from group 23 on 26 November 1997, and I removed two males in Access Class 1 and one female in Access Class 2 from group 22 on 28 November. Members removed from their groups were housed in a separate aviary approximately 100 m from the main aviary to prevent auditory and visual contact. Blood samples were collected from the remaining group members (i.e. those not removed from their groups) 24 hours after removal and again 17 or 18 days later (Table 6-1).

In order to quantify the behavioural effects of group manipulations, I observed behaviours for 20-30 min per day (as described in Chapter IV). In the days preceding group manipulations, each WBBA group was observed for 120-150 min. After members were removed from two groups, each group was again observed for 180-300 min. Access and Allofeeding Classes were assigned to each WBBA in the period before Bleed Date 1 and also for the period between Bleed Dates 2 and 3, following the criteria outlined in Chapter IV (Table 4-4). This allowed me to compare the incidence of behavioural changes in control groups to that in manipulated groups over the course of the experiment.

2. Exchange Experiment

In this experiment, I evaluated the hormonal response to social instability by exchanging members between groups. Two individuals were removed from each of the four groups in Assemblage 4 and replaced with two from a different group. On 8 July, 1998 two birds were exchanged between groups 24 and 27, and on 27 July, 1998 two birds were exchanged between groups 25 and 26 (Table 6-2). To provide an incentive for sexual competition, I insured that at least one female remained in each group and that established dueting pairs were separated. When possible, I exchanged individuals in Access Class 2, whose social status could either improve or diminish with the exchange. Between groups 24 and 27, I exchanged one female in Access Class 2 and one male in Access Class 3 for two males in Access Class 2 and 3 (Table 6-2). In groups 25 and 26, I exchanged two males in Access Class 2 for one male in Access Class 1 and one male in Access Class 2 (Table 6-2). White-browed babblers introduced into an unfamiliar group will be referred to as “intruders,” and those that remained with their original group will be referred to as “residents.”

Table 6-2. Identity, gender, and Access Class of WBBAs used in the Exchange Experiment. Double arrow (↔) indicates that birds were exchanged from one group to another.

WBBA Group	Band	Gender	Access Class	Ex-changed	WBBA Group	Gender	Band	Access Class
24	05291	♀	2	↔	27	♂	05276	2
24	05292	♂	1		27	♂	05272	3
24	05293	♂	3	↔	27	♂	05274	3
24	05294	♀	3		27	♀	05271	3
					27	♂	05273	1
25	89594	♂	2	↔	26	♂	05263	1
25	89595	♂	2	↔	26	♂	05267	2
25	89597	♂	3		26	♂	05264	1
25	89598	♀	1		26	♀	05268	3

Although I was interested in the endocrine and behavioural changes occurring after disturbance, the time courses required for social or endocrine reestablishment were unknown. Behavioural observations were undertaken throughout the course of the experiment. However, because frequent blood collection could be detrimental to the health of the birds, I restricted blood sampling events (Table 6-3). A pre-exchange blood sample was necessary for intra-individual comparisons for all WBBAs. To examine the acute endocrine response to social perturbation, blood samples were taken six hours after member exchange in two groups; birds were bled at 1400h. To examine the short-term response, blood was sampled three days post-exchange in all groups. Because I expected groups to have re-stabilised by 3 weeks, I sampled 2 groups at 23-day post-exchange. The exact schedule of blood sampling events for each group is outlined in Table 6-3. To firther protect the health of birds that were bled repeatedly over a short time period, I restricted the number of serial samples taken for B analysis at each sampling event. For groups 24 and 27, two serial samples were taken for B analysis at each sampling event: initial and 30 min. For groups 25 and 26, that were bled more frequently, only a 30 min B sample was taken. Except for the aforementioned sampling times and number of serial samples, the protocol for capturing birds and sampling blood was identical to that described in Chapters II and V.

Table 6-3. Schedule of blood taking and hormones evaluated in the Exchange Experiment.

WBBAGroup	13 May	11-13 July	27 July	30-31 July
24	Pre-Exchange B	3 Days Post-Exchange T,B,E2	-	23 Days Post-Exchange T,B,E2
25	-	Pre-Exchange T,B,E2	6 Hrs Post-Exchange T,E2	3 Days Post-Exchange T,B
26	-	Pre-Exchange T,B,E2	6 Hrs Post-Exchange T,E2	3 Days Post-Exchange T,B
27	Pre-Exchange B	3 Days Post-Exchange T,B,E2	-	23 Days Post-Exchange T,B,E2

Behavioural observation sessions (described in Chapter IV) were undertaken on each group. Prior to the exchange, groups were observed for 70-80 min. Immediately after the exchange, groups were observed continuously for 60 min. In the two days following the exchange, I watched each group for 45-55 min. In the two groups allowed to re-stabilise for 23 days (Groups 24 and 27), I observed behaviours for 130-150 min in the 6-21 days after the exchange. Because of an unrelated scheduling conflict, members from Groups 25 and 26 were returned to their initial groups following the 3-day post-exchange bleed; there was no extended re-stabilisation period for these groups.

Multiple constraints contributed to the limited design of this experiment. The capacity of the aviary (only four groups could be held concurrently), limited time availability, and difficulties associated with maintaining healthy WBBAs in captivity prohibited a more robust experimental design. In an endeavour to maximize the number of experimental groups, appropriate controls were eliminated. While this resulted in a complicated analysis, it also maximised the number of perturbed groups in which to examine the effects of social manipulations.

Statistical Analysis

For the Removal Experiment I used repeated measures ANOVAs and ANCOVAs (for initial B levels) to examine the effects of experimental manipulations on hormone levels and body mass. Because the repeated measures design compares changes occurring for each individual, I did not include gender differences in the model. Since experiments were not designed across multiple Assemblages, all analyses examined only pre and post-manipulation for a given Assemblage. For the Removal Experiment, eight B measures were analysed (as described in Chapter V): initial B, 30 and 60 min B levels, peak B value (the highest plasma B level of the three samples collected from each bird), total B response, rate of change in plasma B from initial to 30 min samples, 30 to 60 min samples, and initial to peak level. To illustrate initial B levels in graphical analyses, I plotted the residuals from a regression of initial B on time since capture, as described in Chapter V. To examine variation in furcular and abdominal fat levels, I used Fisher's exact test because it allowed for small samples sizes and/or missing cells.

For the Exchange experiment, the B response to social manipulations was examined using repeated measures ANOVAs and ANCOVAs (for initial B). In two groups (25 and 26), only a single B sample was taken at each "bleed date," thus only the 30 min B levels were reported. In the other two groups (24 and 27), the first two serial samples were taken at each bleeding event during the Exchange Experiment, and three B measures were examined: initial B levels, 30 min B levels, and rate of change from initial to 30 min samples. To illustrate initial B levels in graphical analyses, I plotted the residuals from a regression of initial B on time since capture, as described in Chapter V. Differences among behavioural classes in B levels at the 30 min sample, 3 days post-exchange were analysed using an ANOVA. Variation in T levels were examined using a repeated measures ANOVA when there were multiple comparisons over time, a t-test when comparing two groups at a single sampling event, and an ANOVA to examine differences among multiple groups at a single sampling event. Changes in body mass in response to experimental manipulations were analysed with paired t-tests in the Exchange experiment.

RESULTS

Removal Experiment

1. Behavioural Response

Throughout the course of the experiment, I observed no agonistic or agitated behaviour in any of the WBBA groups. Removal of high ranking group members seemed to elevate the social status of the remaining birds, especially those in Access Class 3. White-browed Babblers in experimental groups (those in which group members were removed) tended to change Access and or Allofeeding Classes with higher frequency than those in control groups (Table 6-4). Perhaps due to small sample sizes, this trend was not siginificant for either Access Class (Fisher's exact test P=0.070) or Allofeeding Class (Fisher's exact test P=0.103). When comparing behavioural observations before and after removals, only one WBBA in a control group changed Behavioural Class; the bird that neither fed nor received meal worms, "Nonallo," became a "Receiver." In the experimental groups, five WBBAs changed Allofeeding Classes and four changed Access Classes (Table 6-4). In one group a WBBA classified in Access Class 3, Allofeeding Class Receiver changed to Access Class 1, Feeder and another changed from Access Class 2 to 1. In the other experimental group, two birds changed from Nonallo to Receiver, one from Access Class 3, Nonallo to 1, Feeder, and one from Access Class 3, Nonallo to 1, Both.

Table 6-4. Comparison of Behavioural Class instability in control and experimental (removal) groups of WBBAs. Access and Allofeeding Classes were compared before and after member removal.

WBBA Group	Treatment	Number of WBBAs Post-Removal	Number of WBBAs that Changed:	
			Access Class	Allofeeding Class
20	control	4	0	1
21	control	3	0	0
22	removal	4	2	4
23	removal	3	2	1

2. Body Condition

During the Removal Experiment, body mass of WBBAs was measured at each of three bleed dates. There were no significant differences in body mass between control and experimental birds over the experimental period ($F_{1,12}$=0.210, P=0.655) nor did body mass vary among the three bleed dates ($F_{2,24}$=0.546, P=0.587). However, there was a significant interaction with body mass between bleed date and treatment ($F_{2,24}$=6.546, P=0.005). In the control groups, body mass seemed to remain stable from the first blood sample to the second (bleed date 1 to 2), and then tended to increase by the third sample (bleed date 3). In the experimental groups, body mass tended to progressively decrease slightly from bleed date 1 to 2 to 3 (Fig. 6-1).

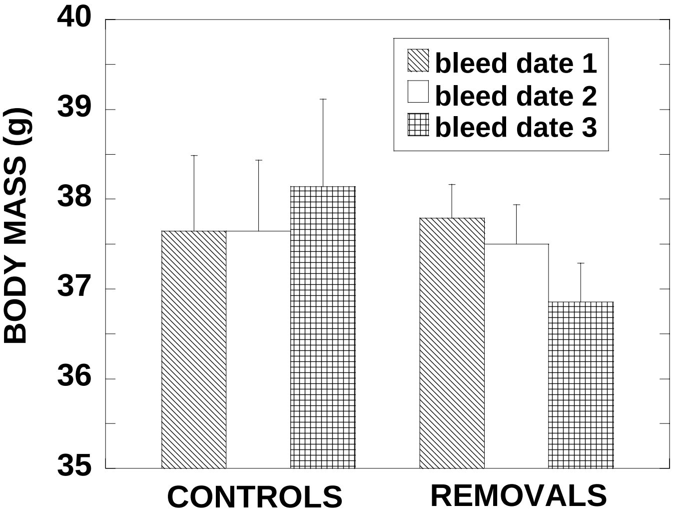


Figure 6-1. Body mass in control and experimental WBBAs during the Removal Experiment, during which WBBAs were weighed at each “bleed date.” Height of columns indicates means, and bars represent one standard error. “Removals” refers to individuals belonging to groups in which members had been removed. Seven birds in each of the control and removal groups were weighed at each bleed date.

Body fat scores remained low in WBBAs throughout the Removal Experiment; furcular fat reached a level of 3 in only two birds out of the 14 studied, and abdominal fat never exceeded a level of 1. Fat levels tended to decrease more often in WBBAs in experimental groups than in control groups (Table 6-5). However, these relationships were not significant for furcular fat levels between bleed date 1 and 2 (Fisher’s exact test P=0.069) or bleed dates 2 and 3 (Fisher’s exact test P=0.153), nor for abdominal fat levels between bleed dates 1 and 2 (Fisher’s exact test P=0.466) or bleed dates 2 and 3 (Fisher’s exact test P=0.496).

Table 6-5. Changes in furcular and abdominal fat levels in control and experimental WBBAs during the Removal Experiment. Number of WBBAs whose fat levels increased, decreased, or remained unchanged between blood sampling periods.

	Bleed date 1 to 2			**Bleed date 2 to 3**		
Furcular fat	in-crease	de-crease	no change	in-crease	de-crease	no change
control	2	3	2	2	0	5
removals	0	7	0	2	3	2
Abdominal fat						
control	1	0	6	2	2	3
removals	0	2	5	2	0	5

3. Corticosterone Response

Throughout the Removal Experiment, there were no significant differences between control groups and groups with removed members in any of the eight B measures that described the capture stress response (Table 6-6). However, there was a significant effect over time for five of the eight B measures (Table 6-6); in each of these five B measures (B titres at 30 and 60 min, peak B, area under curve, rate of change initial-30 min), levels decreased with each subsequent bleed date (Figs. 6-2, 6-3, and 6-4). Of the remaining three B measures, two (rate of change to peak and initial B) tended to decrease in control groups with each bleed date (Figs. 6-2 and 6-4).

Table 6-6. Effect of treatment (control vs. removal) and repeated measures (bleed date) on WBBA capture-stress response. To examine all B measures, seven repeated measures ANOVAs and one repeated measures ANCOVA[1] were used. Asterisks (*) indicate significant difference ($P<0.05$). All interaction terms were non-significant ($P\geq0.072$). [2] degrees of freedom are 2 and 21.

	Between Treatments			**Across** Repeated Measures	
	$F_{1,11}$	Prob.		$F_{2,22}$	Prob.
Initial B[1]	1.689	0.220		3.085[2]	0.067
30 min B	0.530	0.482		26.123	**<0.001***
60 min B	0.420	0.530		10.639	**0.001***
Rate initial-30 min	1.741	0.214		11.356	**0.007***
Rate 30-60 min	0.103	0.754		0.623	0.546
Rate to Peak B	0.869	0.371		1.336	0.284
Peak B	0.481	0.503		17.914	**0.001***
Area Under Curve	1.380	0.265		26.335	**<0.001***

Although not significantly different, the reduction in B levels between bleed dates appeared greater in control group than in manipulated groups in B levels at 30 and 60 mins (Fig. 6-2) and in peak B and area under the curve measures (Fig. 6-3). This trend is most evident when comparing bleed dates 1 and 3.

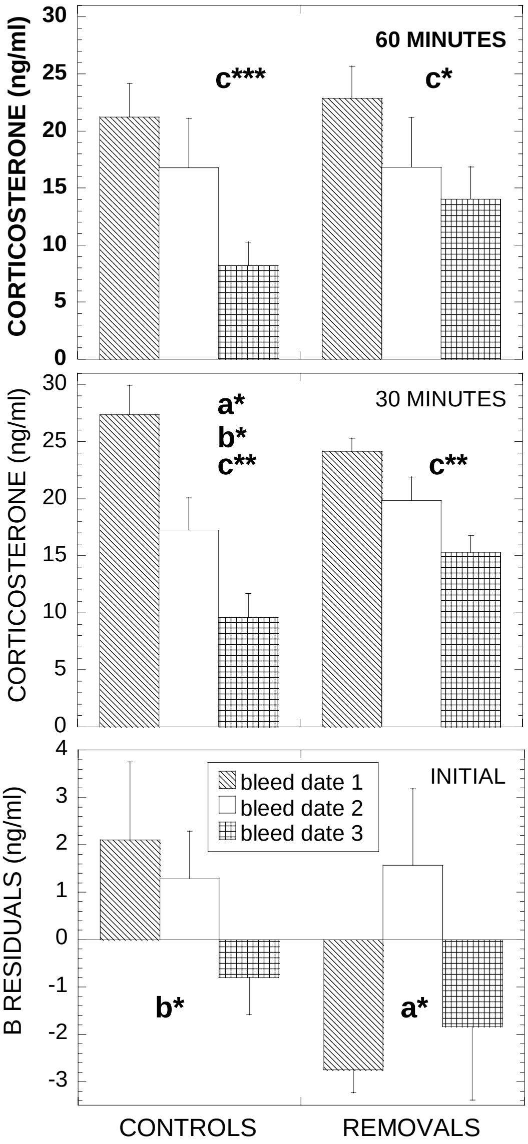


Figure 6-2. Corticosterone levels at initial, 30 min and 60 min serial samples in control groups and experimentally manipulated groups (removals) across three bleed dates. Initial B levels are expressed as the residuals from a regression of time on initial B to account for variable timing of the initial sample. Height of columns indicates means, and bars represent one standard error. “a,b,c” indicate significant differences between bleed date 1 and 2, bleed date 2 and 3 , and bleed date 1 and 3, respectively. Number of asterisks (*) indicates significance levels: *P<0.05, **P<0.01, ***P<0.001. Seven control birds and seven removals were sampled at each bleed date.

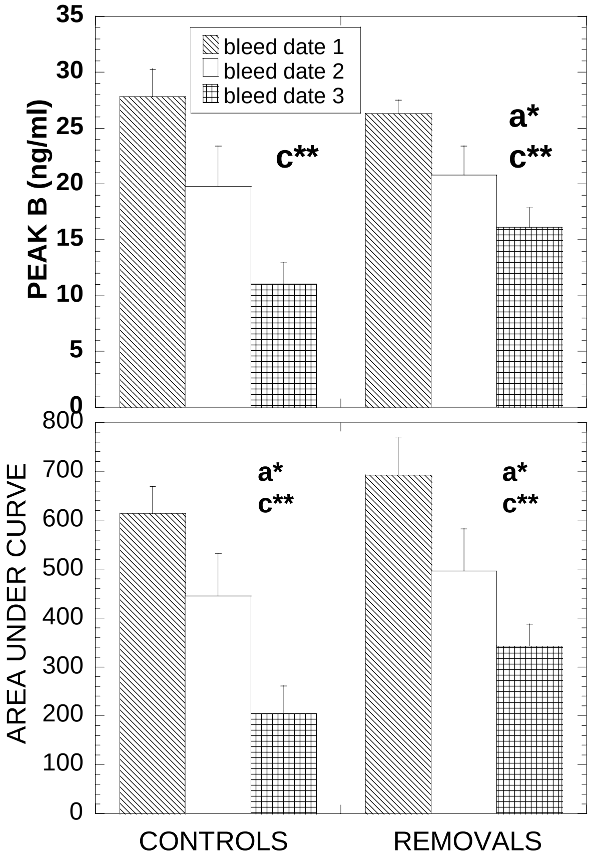


Figure 6-3. Peak B levels and total B response (area under curve) in control groups and experimentally manipulated groups (removals) across three bleed dates. Height of columns indicates means, and bars represent one standard error. “a and c” indicate significant differences between bleed date 1 and 2 and bleed date 1 and 3, respectively. Number of asterisks (*) indicates significance levels: *P<0.05 and **P<0.01. Seven control birds and seven removals were sampled at each bleed date.

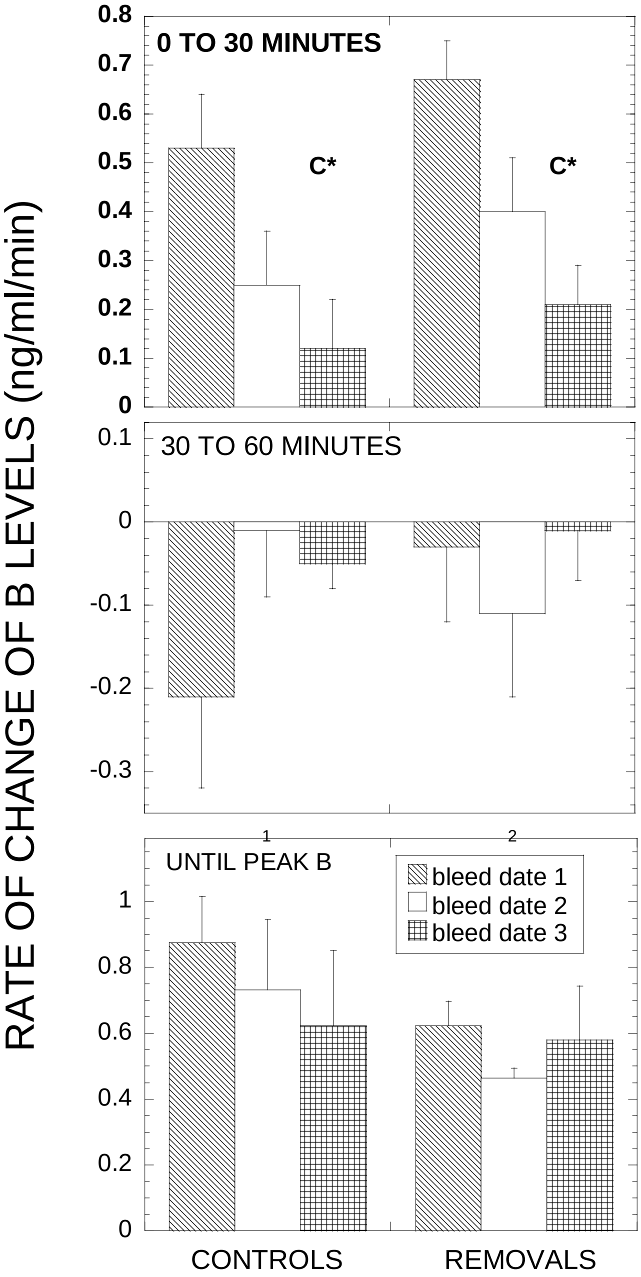


Figure 6-4. Rate of change of B levels in control groups and experimentally manipulated groups (removals) at three time intervals: 0 to 30 min, 30 to 60 min, and initial sample (“0”) to time at peak B. Height of columns indicates means, and bars represent one standard error. “c*” indicates significant differences (P<0.05) between bleed date 1 and 3. Seven control birds and seven removals were sampled at each bleed date.

4. Testosterone

During the first sampling period of the Removal Experiment (bleed date 1), samples were not collected for testosterone analysis. During the subsequent two bleed dates, T did not differ between treatments ($F_{1,10}$=0.222, P=0.648) or between bleed dates ($F_{1,10}$=0.374,P=0.555), nor was there an interaction between these two factors ($F_{1,10}$=1.005, P=0.340). Furthermore, out of 24 samples, only two had T levels measuring more than 50 pg/ml, and 15 WBBAs had T titres below detectable limits of the assay (T<25pg/ml).

Exchange Experiment

1. Behavioural Response

During the first 60 minutes after introduction of novel group members, birds exhibited the full repertoire of previously noted behaviours. In addition, both residents and intruders pecked at perches. Also during this 60 minute period, up to six displacements occurred, in which one WBBA would take another's place on a perch. In one group an intruder flew erratically around the cage, while in another an intruder was chased by a resident for almost five minutes. Birds familiar with one another (those from the same group before the manipulation) seemed to spend more time in close proximity to one another than did those from different home groups. Allo*preening* occurred most frequently when residents allopreened each other or when intruders allopreened residents; intruders were rarely allopreened by either intruders or residents (Table 6-7). Allo*feeding* also occurred with varying frequency among residents and intruders (Table 6-7); all but one of the observed allofeeds were offered by residents, and the most common interaction was for residents to allofeed intruders. A begging call was usually given by intruders in response to being fed, and intruders also begged when approached by a residents. Within a week of the member exchange, residents and intruders seemed to mix freely, and there were increasingly fewer displacements, chases, and perch pecks, while incidences of allofeeding and allopreening occurred within the ranges noted in persistently stable groups.

Table 6-7. Number of allopreening and allofeeding events that took place between residents and intruders during the first 60 min after introduction of novel group members during the Exchange Experiment.

Direction	# of Allopreens	# of Allofeeds
Resident → Resident	24	4
Intruder → Intruder	2	0
Resident → Intruder	5	7
Intruder → Resident	14	1

Behavioural Classes were assigned after each of three observation periods: "Pre-Exchange" refers to the Class assignments based on observations made before the exchange, "Exchange" to those Class assignments based on observations made in the two days following the exchange, and "Post-exchange" to Class assignments based on observations conducted from days six through 23 after exchange. Many WBBAs changed Access and Allofeeding Classes from one observation period to the next (Table 6-8).

Table 6-8. Number of WBBAs that changed Access and/or Allofeeding Class during the Exchange Experiment. Asterisks (*) indicate that data is not available, as Post-exchange observations were not undertaken on groups 25 and 26. Numbers below “Pre-Exchange to Exchange” and “Exchange to Post-Exchange” indicate the number of WBBAs that changed Behavioural Class from one observation period to the next.

		Changes in Access Class		**Changes in Allofeeding Class**	
WBBA Group	Number in Group	Pre-Exchange to Exchange	Exchange to Post-Exchange	Pre-Exchange to Exchange	Exchange to Post-Exchange
24	4	2	2	2	1
25	4	1	*	2	*
26	4	1	*	2	*
27	5	2	0	1	3

Intruders were more likely to change Behavioural Classes than were residents (Table 6-9). When comparing Access Classes before (pre-exchange) and immediately after member exchange (exchange), only one resident changed Access Class, while all but one intruder changed Access Classes. Of these birds, three intruders attained a higher Access Class, two assumed lower Access Class status, and the single resident fell from Access Class 2 to 3.

Table 6-9. Number of residents and intruders that maintained or changed Behavioural Classes in the pre-exchange to exchange period of the Exchange Experiment.

	Maintained Access Class	**Changed** Access Class	Maintained Allofeeding **Class**	Changed Allofeeding **Class**
Residents	8	1	6	2
Intruders	1	5	1	7

Changes in Allofeeding Class mimicked those in Access Class; more intruders' Allofeeding Class changed between pre-exchange and exchange periods than residents' (Table 6-9). Three intruders changed from Nonallo (neither receiver nor feeder) to Receiver, three from Receiver to Nonallo and one from Both (feeder and receiver) to Nonallo, while one resident changed from Nonallo to Receiver and the other from Nonallo to Feeder.

In the two groups with both exchange and post-exchange observations, intruders were the only ones to change Access Class; one fell from Access Class one to two, and the other rose from three to two. Changes in Allofeeding Class were also noted. An intruder was classified as Nonallo during the exchange observations, but changed to Feeder. Of the three residents that changed Allofeeding Classes, two changed from Nonallo to Receiver and one from Nonallo to Feeder.

2. Body Condition

Due to small sample sizes and an inconsistent protocol, I was unable to analyse changes in body condition of all birds in all sampling periods or analyse differences between residents and intruders. However, in Groups 24 and 27, comparisons could be made between pre-exchange and 23 days post-exchange periods, and in Groups 25 and 26, comparisons could be made between 6 hrs post-exchange and 3 days post-exchange periods (Table 6-10). Body mass was significantly lower when measured 3 days post-exchange than 6 hrs post-exchange (paired t=4.324, P=0.023), but there was no significant difference between pre-exchange and 23 days post-exchange measurements (paired t=-1.472, P=0.191). In groups 25 and 26, mean body mass decreased 2.25 g between 6 hrs post-exchange and 3 days post-exchange. Both furcular and abdominal fat also decreased in all but one bird from the 6 hrs post-exchange to 3 days post-exchange periods, but there appeared to be no consistent pattern from pre-exchange to 23 days post-exchange measurements (Table 6-10).

Table 6-10. Changes in body mass, furcular fat, and abdominal fat in WBBAs during the Exchange Experiment. Numbers indicate the number of birds whose body mass or fat increased, decreased, or remained unchanged between the specified periods.

	6 hours post-exchange to 3 days post-exchange			**Pre-exchange to** 23 days post-exchange		
	increased	decreased	no change	increased	decreased	no change
Body Mass	0	4	0	4	1	2
Furcular Fat	0	3	1	3	2	2
Abdominal Fat	0	3	1	1	4	2

3. Corticosterone Response

In Groups 24 and 27, B levels were measured before the exchange, 3 days after, and 23 days after the exchange. In Groups 25 and 26, B levels were measured only twice: before the exchange and 3 days after (Table 6-3). Exchanging WBBAs between groups did not have a significant effect on individuals’ capture-stress response (Table 6-11). However, all three measures of the capture stress response analysed for this experiment (initial B, 30 min B, and rate of change) tended to increase from pre-exchange to 3 day post-exchange levels (Figs. 6-5 and 6-6). After a 23 day period of stabilisation, capture stress measures tended to remain stable or to decrease (Figs. 6-5 and 6-6).

Table 6-11. Repeated measures ANOVAs and an ANCOVA (for initial B) examining the effect of treatment period (pre-exchange, 3 days post-exchange, and 23 days post-exchange) on WBBAs' capture stress response. [1] pre-exchange and 3 days post-exchange treatment periods only.

Initial B	$F_{2,20}=0.673$	P=0.521	2 groups
30 min B[1]	$F_{1,12}=2.396$	P=0.148	4 groups
30 min B	$F_{2,10}=2.214$	P=0.160	2 groups
Rate initial-30 min	$F_{2,6}=0.327$	P=0.733	2 groups

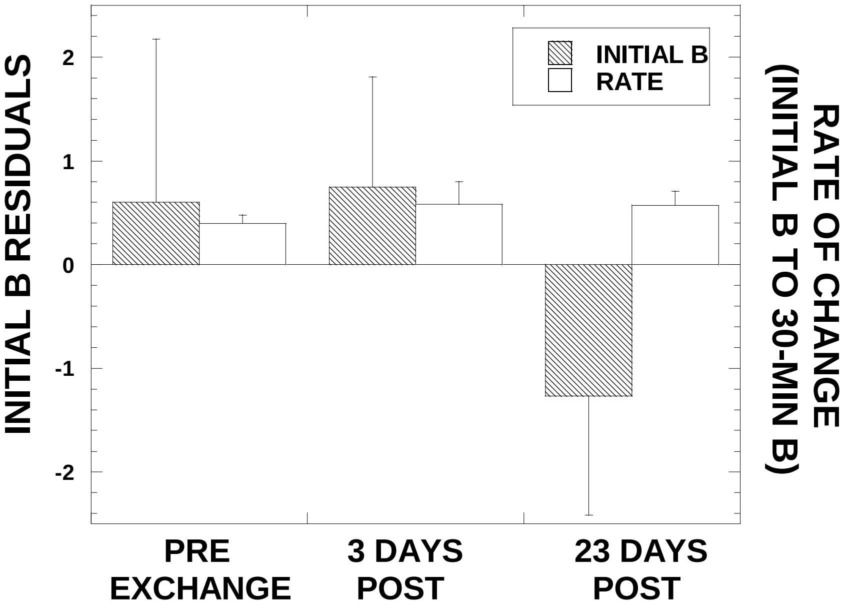


Figure 6-5. Comparison of WBBAs' capture stress response before member exchange (pre-exchange), soon after exchange (3 days post), and after a period of stabilisation (23 days post). Residuals from a regression of initial B levels on time since capture were plotted to correct for variation in the time course of the first samples. Data are from Groups 24 and 27 only, and n=9 for each column. Height of columns indicates means, and bars represent one standard error.

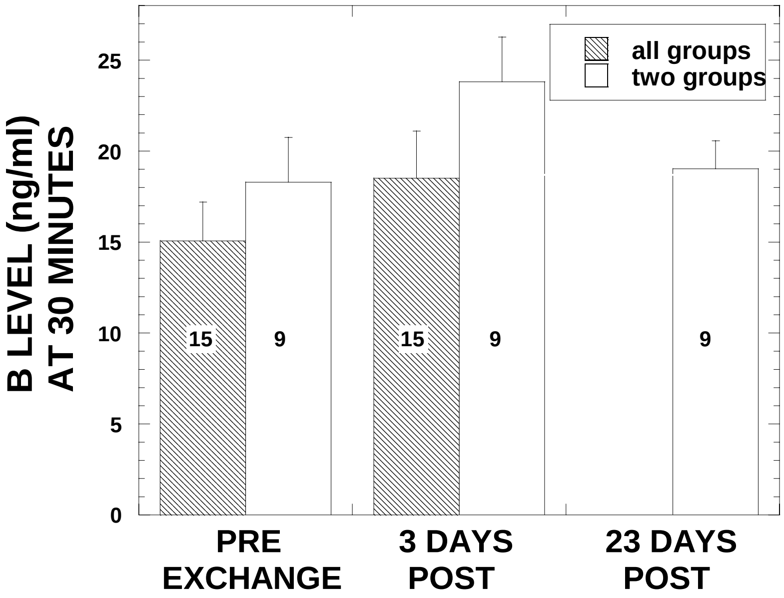


Figure 6-6. Comparison of B levels at 30 min post capture throughout the Exchange Experiment. Samples were taken from all groups at the pre-exchange and 3 days post- exchange periods, but from only Groups 24 and 27 at the 23 days post-exchange period. Height of columns indicates means, and bars represent one standard error. Numbers inside columns indicate sample sizes.

To examine whether residents and intruders responded differently to the Exchange Experiment, I compared their capture stress profiles three days after exchange of group members. As sample sizes were small and there were no significant differences in any of the capture stress measures between males and females (P>0.05), genders were analysed together. In residents initial B tended to be lower, B at 30 min tended to be higher, and rate of change from initial to 30 min B tended to be marginally higher than in intruders (Fig. 6-4). However, none of these differences were significant (Table 6-12).

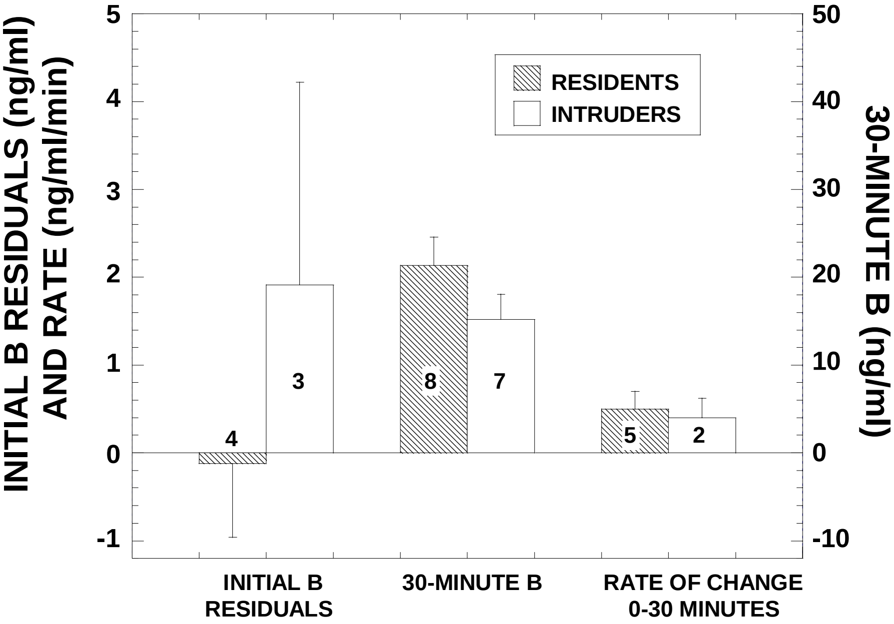


Figure 6-7. Comparison of residents' and intruders' capture stress response 3 days after member exchange. Residuals from a regression of initial B levels on time post-capture correct for variable timing of the initial samples. Height of columns indicates means, and bars represent one standard error. Numbers inside columns indicate sample sizes.

Table 6-12. Analyses of differences between residents' and intruders' capture stress responses at 3 days post-exchange. ANCOVA was used for initial B levels to account for variability in time since capture. ANOVAs were used to examine B levels at 30 min and rate of change of B from initial to 30 min levels.

Initial B	$F_{1,4}$=0.879	P=0.402
30 min B	$F_{1,13}$=1.966	P=0.184
Rate of B Change	$F_{1,5}$=0.091	P=0.776

Finally, I examined the relationship between Behavioural Classes and B levels in groups of WBBAs with manipulated group membership. Due to small samples sizes at other sampling periods during the Exchange Experiment, I only present data from the 30 min sample at three days post-exchange. Mean B levels tended to be lower in WBBAs in Access Class 2 and Allofeeding Class Feeder than in other Behavioural Classes (Figure 6-8); however, this relationship was not significant for either Access Class ($F_{2,12}$=1.065, P=0.375) or Allofeeding Class ($F_{2,12}$=0.868, P=0.445).

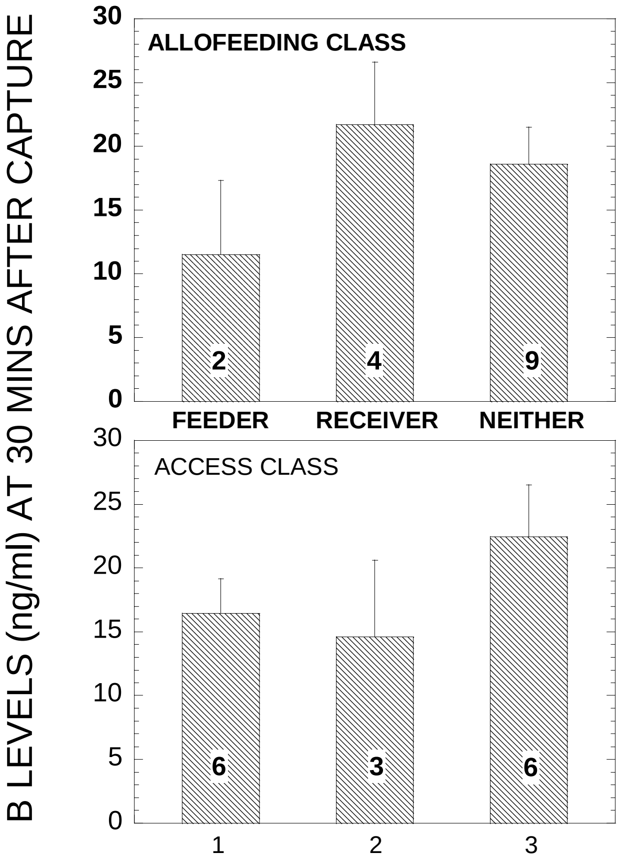


Figure 6-8. Comparison of WBBAs' B levels among Behavioural Classes during the Exchange Experiment. Mean B levels at 30 min post-capture, 3 days post-exchange are presented. Sample sizes are indicated by numbers inside columns. Height of columns indicates means, and bars represent one standard error.

4. Testosterone

During the Exchange Experiment, T levels were only measured in male WBBAs. In Groups 24 and 27, T levels were measured 3 days after the exchange and again 23 days after. In Groups 25 and 26, T levels were measured before the exchange, 6 hrs after, and three days after (Table 6-3). Three days after the exchange, T levels in male WBBAs tended to be higher and more variable than during other sampling periods (Figure 6-9). The elevated T levels and large standard error 3 days post-exchange was largely due to two males with unusually high T levels (see below). While inconsistent sampling frequency among groups prohibited analysis of all sampling periods in each WBBA group, some statistical comparisons were possible. In Groups 25 and 26, there were no significant differences among individuals in the following treatment periods: 14-15 days pre-exchange, 6 hrs post-exchange, and 3 days post-exchange (repeated measures ANOVA $F_{2,10}$=0.166, P=0.849). In the Groups 24 and 27, there was no significant difference in males' T levels between 3 days post-exchange and 23 days post-exchange (paired t_5=1.670, P=0.156).

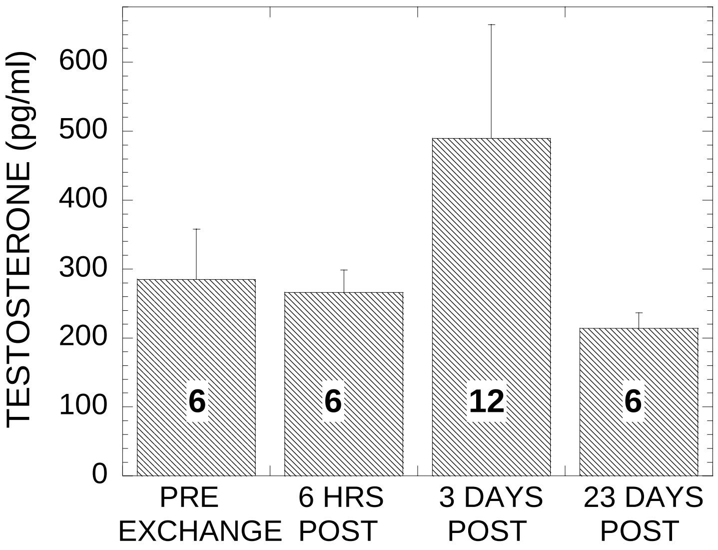


Figure 6-9. Comparison of mean male T levels across 4 sampling periods during the Exchange Experiment. Sample sizes are indicated by numbers inside columns. Height of columns indicates means, and bars represent one standard error. Testosterone levels were measured in two groups at each sampling period, except at 3 days post-exchange, when T levels were measured in 4 groups (see text).

In post-exchange sampling periods, I also examined differences in T levels between residents and intruders. Male residents tended to have higher mean T levels 6 hrs post-exchange and 23 days post-exchange than intruders, but this pattern was reversed 3 days post-exchange (Figure 6-10). None of these relationships were significant (Table 6-13). The large standard error and high mean T level 3 days post-exchange in intruders was largely due to two males with T titres of 1150 pg/ml and 2100 pg/ml. One of these intruding males avoided all other group members as much as possible in the days following the exchange and maintained his Behavioural Class (Access Class 3, Allofeeding Class Nonallo) for the duration of the experiment. The other intruding male WBBA with high T levels exhibited no obviously distinct behaviours following member exchange, and his Behavioural Class changed from pre-exchange (Access Class 3, Receiver) to 3 days post-exchange (Access Class 2, Nonallo). Corticosterone levels in these two birds were not remarkably different from those of other WBBAs. Even when these outliers were removed, mean T level of intruder still tended to exceed that of residents (284.00 ± 47.71 pg/ml vs. 240.24 ± 24.85 pg/ml).

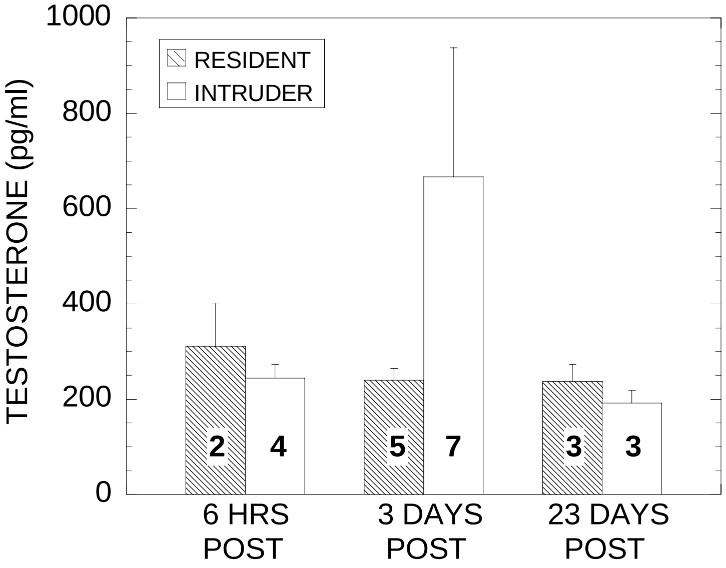


Figure 6-10. Mean T levels in male residents and intruders at three sampling periods during the Exchange Experiment. Numbers inside bars indicate sample sizes. Height of columns indicates means, and bars represent one standard error.

Table 6-13. Analyses of the differences in T levels between residents and intruders at three sampling periods during the Exchange Experiment. [1] A separate variance t-test was used because variances in the two groups were unequal.

Time Post-Exchange	df	t value	Prob.
6 Hours	4	0.950	0.396
3 Days [1]	6.1	-1.315	0.218
23 Days	4	1.022	0.365

Finally, I examined the relationship between Behavioural Classes and T levels of male WBBAs in groups during the Exchange Experiment. Due to small samples sizes during other sampling periods, I only examined T levels at three days post-exchange; testosterone titres were measured in four groups at three days post-exchange compared to two groups at other sampling times (Table 6-2). Mean T levels tended to be lowest in males in Access Class 1, Allofeeding Class Feeder and highest in Access Class 3, Allofeeding Class Nonallo (Fig. 6-11). However, these trends were not significant for either Access Class ($F_{2,9}$=1.290, P=0.322) or Allofeeding Class ($F_{2,9}$=0.530, P=0.606). Furthermore, the high T values in Access Class 3 and Allofeeding Class Nonallo were largely due to two males (mentioned previously) with T levels greater than 1000 pg/ml. When these outliers were removed, mean T levels more closely resembled those in other Behavioural Classes: 238.33 ± 39.83 pg/ml for Access Class 3 and 245.00 ± 23.08 pg/ml for Allofeeding Class Feeder.

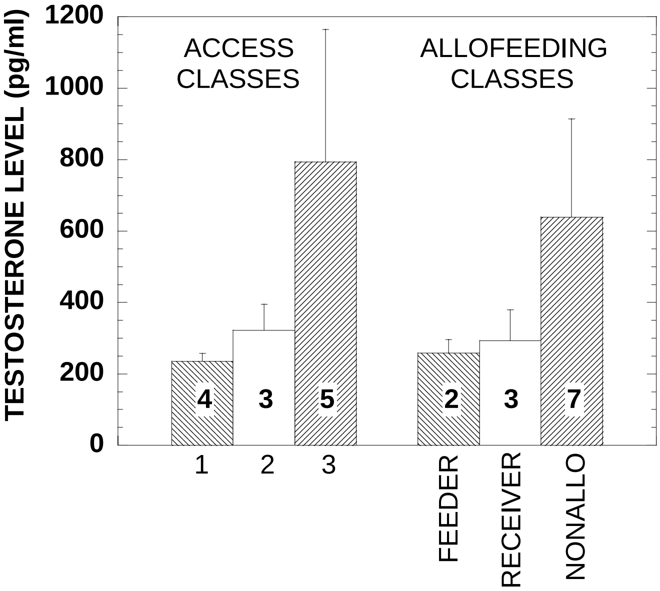


Figure 6-11. Comparison of male T levels among Behavioural Classes at 3 days post-exchange. Numbers inside columns indicate sample sizes. Height of columns indicates means, and bars represent one standard error.

5. Estradiol

Female WBBAs' E2 levels were measured at pre-exchange, 6 hrs post-exchange, 3 days post-exchange, and 23 days post-exchange (Table 6-3). In all cases E2 levels were below the level of detection of the radioimmunoassay (5 pg/ml). Estradiol was not measured in male WBBA.

DISCUSSION

Behavioural Response

Experimental manipulations resulted in changes in WBBA social structure. In the Removal Experiment, more than 70% of birds from experimental groups changed Behavioural Class, while only a single bird from a control group exhibited a change. In the Removal Experiment, WBBAs in Access Class 3 tended to increase in rank to Access Class 1 most often. Relatedness within the group may have influenced this change. In stable groups WBBAs in Access Class 1 seemed to confer high priority of meal worm access to their close relatives, and these relatives typically occupied Access Class 2 (Chapter IV). In some cases WBBA parents may have tolerated their offspring at the feeding dish more so than they did non-relatives, as has been shown in the Siberian Jay (Ekman and Tegelstrom 1994, Sklepkovych 1997). It is possible that WBBAs in Access Class 1 awarded preferential treatment to their offspring, that resulted in their offspring's attainment of Access Class 2; when the "parents" in Access Class 1 were removed from their groups during the experiment, preferential treatment was no longer extended to these "offspring," and they were surpassed in rank by older (or larger or more assertive) members that had previously occupied Access Class 3. There is considerable evidence that age, size, and personality affect dominance interactions in many group-living species (Craig 1979, Johnson 1988, Ekman 1990, Sapolsky 1990, Emlen 1996), and it is likely that these factors influence social status in WBBAs as well.

The Exchange Experiment appeared to elicit a more intense behavioural response than the Removal Experiment; for instance, aggressive behaviours were apparent during the Exchange Experiment, while none were observed in the Removal Experiment. Further, birds appeared agitated and exhibited displacement behaviours upon introduction of novel group members, behaviours that were not observed during the Removal Experiment. In the Exchange Experiment, birds not only faced social restructuring due to removal of members, but they also were forced to integrate new members into the group. Introduction of unfamiliar group members may also have initially elicited conflicts over territories (i.e. aviaries).

The lack of aggression I observed in WBBAs during the Removal Experiment appears to be unusual among social birds described to date. In many free-living social species, intragroup aggression following social disruption is common. For instance, Wingfield *et al.* (1991) reported increased aggression in White-browed Sparrow Weavers after removal of group members. Hannon *et al.* (1991) observed conspicuous contests following the death of breeders in groups of Acorn Woodpeckers, and Curry (1988) reported aggressive chases between males following the death of the alpha males' mates in Galapagos Mockingbirds. In captive groups of WBBAs, previously established social roles, age, or kinship may have influenced the emergence of new social positions when members were removed; aggressive contests may not have been needed to establish a new social structure among WBBAs familiar with one another. However, in the Exchange Experiment, the introduction of unfamiliar, unrelated individuals precludes the formation of a social structure based on previously established roles or kinship. In this case, aggressive behaviour (such as chases and supplants) may have helped establish social positions. Furthermore, non-aggressive behaviours such as allofeeding and allopreening may have contributed to the establishment of social position. Immediately following introduction of new group members, residents were more likely to offer allofeeds than intruders, and residents may have used allofeeding to assert their dominance (see Chapter IV). Residents were also most likely to receive allopreens from other residents and intruders, and this behaviour may have acted to help strengthen existing or establish new social bonds.

In the Exchange Experiment, residents maintained their Behavioural Class with greater regularity than did intruders. However, intruders were equally likely to attain a higher or lower social position than they had held in their resident groups. It appears that immigration to a new group acts as an impetus for change in social position, but more so for the immigrant than for the long-term residents. Of course, in the wild the impetus to emigrate may be experienced by certain individuals more so than others, and their previous rank may affect how they are treated in a new group.

Body Condition

Social manipulations tended to have some effect on both body mass and fat levels in WBBAs. In the Removal Experiment, there was no significant effect of either treatment or sampling period on body mass, but there was a significant interaction term between these two factors. Body mass remained stable or increased with each subsequent sampling period in control birds, while it decreased slightly with each period in experimental birds. Also, in the two weeks between bleed dates 1 and 2 of the Removal Experiment, fat levels tended to decrease more often in groups in which members had been removed than in control groups. Exchange of group members also seemed to result in a decline of body condition. While the experimental design prevented statistical analysis, a precipitous drop in body mass (more than 2 g or approximately 5% of body mass) and an uniform decrease in fat levels in the three days after member exchange suggested an influence of social change on body condition. A decrease in feeding in response to social instability may have contributed to the observed effect on body condition. White-browed Babblers in socially unstable groups may spend proportionally less time feeding than those in stable groups. The reliability and abundance of food in the aviary setting may allow WBBAs to devote more time to behaviours relevant to restructuring the social system than to feeding. A similar reaction may be predicted for wild birds; however, because food is more limited and/or requires more time to obtain in the wild than in captivity, the adjustment in body condition may be accentuated but take place over a longer time course.

Furthermore, the observed trends may reflect a physiological response to social dissonance resulting from the social manipulations. Increased stress levels during social instability may have contributed in the observed decline in body condition. For example, stress-induced changes in metabolic rate (Senar *et al.* 2000) or stress-related hormonal changes resulting in protein catabolism (Siegel 1980) may influence body condition. While a number of studies suggest the opposite (a lipogenic effect of stress), the subjects of such studies typically exhibited a substantial elevation in glucocorticoid levels (Baum and Meyer 1960, Wingfield and Silverin 1986, Gray *et al.* 1990). Although there was some indication that WBBAs' B levels increase slightly in response to social stress (see below), such minimal changes in WBBAs' glucocorticoid levels did not seem to have the same catabolic effect (protein breakdown and lipogenesis) as do more pronounced glucocorticoid elevations.

Corticosterone

Corticosterone, the major glucocorticoid in birds, is a sensitive measure of an individual's state of psychological arousal (Mendoza *et al.* 1979). Heightened psychosocial stress, resulting from social instability or hierarchy formation, has been shown to stimulate the HPA axis and result in increased secretion of glucocorticoids (Bronson 1973, McGuire *et al.* 1986, Levine 1993). Furthermore, a number studies have demonstrated that social rank is related to the plasma level of glucocorticoid secretions during times of instability but not in stable hierarchies (Coe *et al.* 1979, McGuire *et al.* 1986, Levine 1993). In order to investigate the effects of social instability and newly acquired social position on the HPA axis, I examined B levels in WBBAs with manipulated group membership.

When examining the influence of social manipulation on the capture-stress response, an unexpected effect of repeated sampling of captive individuals became apparent. In the Removal Experiment, there was a significant influence of bleed date on many measures of the capture-stress response. With each successive sampling event, there was a diminished adrenal response to the stressor (capture and handling). Other studies have shown that the avian stress response can become habituated to various stressors, including heat, cold, underfeeding, and treadmill exercise (see Harvey *et al.* 1984). Furthermore, Harvey *et al.* (1984) suggested that when animals were repeatedly subjected to aversive stimuli that did not lead to physiological insult, the animals developed expectancies that modified the hypothalamic signal and the adrenal responsiveness to the stimulus. While capture and handling stress of Pied Flycatchers has been shown to increase B levels more so than does exposure to various predators and conspecific challengers (Silverin 1998), WBBAs seem to habituate to this potent stressor. With each successive sampling event, WBBAs may have become accustomed to capture and handling, and they may have learned to expect that no real harm would come to them. This cognitive appraisal may have resulted in the diminished B release observed with successive sampling throughout the Removal Experiment.

Neither group member removal nor member exchange had a statistically significant effect on WBBAs' adrenocortical response to capture and handling. However, there were some trends suggesting that social manipulations may have influenced the adrenocortical response in WBBAs. For instance, in the Removal

Experiment, basal B in experimental groups tended to increase from levels before removal to 1 day after removal; meanwhile, basal B fell with each subsequent sample in control groups (Fig. 6-2). Also, when comparing B measures before removal to those 18 and 19 days post-removal, those of control groups tended to decrease more than those of experimental groups (Figs. 6-2 and 6-3).

In the Exchange Experiment, basal and 30 min B levels tended to rise from pre-exchange to 3 day post-exchange levels. Because this experiment did not include control groups, I do not know if there was a similar habituation to the capture and handling protocol as that seen in the Removal Experiment. However, if it were valid to assume that all WBBAs became accustomed to capture and handling, then the slight increase in B levels after the exchange of group members might have biological significance.

These trends suggest an effect of psychosocial stimulation on the HPA axis in WBBAs, in accordance with a vast literature suggesting that social factors can modify adrenocortical activity (Harvey *et al.* 1984, Sapolsky 1992, Levine 1993). In many species aggression plays a role in the establishment of social relationships (Bronson 1973, Ramenofsky 1984, Schwabl *et al.* 1988); however, in the WBBA little agonistic behaviour was observed. Perhaps the HPA axis is more responsive (or influential) in animals with social systems that rely more heavily on aggression to determine social position than in the largely non-aggressive WBBA.

Some studies have suggested that it is the relative social significance of instability that affects HPA activity, as opposed to instability itself. For example, Sapolsky (1992) found that the hormonal response to social instability in Olive Baboons differed depending on whether an individual's rank increased or decreased during social restructuring. Saltzman *et al.* (1994) determined that F levels in female Common

Marmoset corresponded more closely to changes in reproductive function than to shifts in rank without accompanying changes in ovarian function. Although I can only hypothesize about the functional significance of social position in the WBBA (see Chapter IV), I would guess that the observed social instability resulting from experimental manipulations may not have caused a uniform response among WBBAs. The resulting variability may have led to the observed statistically indistinguishable adrenocortical responses before and after the social manipulations.

Small sample sizes prohibited analyses of many of the factors that may have influenced the social significance of group instability and ultimately affected B levels. However, I was able to examine whether HPA activity was greater in birds that remained in their resident cages or in those that were moved into another group. In the Exchange Experiment, there was no significant difference in the capture stress response, but intruders tended to show higher and more variable initial B levels than did residents. At least for some intruders, chronic stress levels (indicated by elevated initial B titres) seemed to be higher than in residents. Social support is thought to decrease the glucocorticoid response to some stressors (Abbott *et al.* 2003). Familiar surroundings with familiar social partners may have ameliorated the stress levels of WBBA residents, while unfamiliar surroundings and a dearth of well-known social partners may have imposed chronic psychological stressors on some intruders. Furthermore, before being held in captivity, most WBBAs probably had experienced immigration of new group members into their resident groups, but not all birds had emigrated from their natal groups. Past experience with social changes may have influenced an individual’s response to the Exchange Experiment, as was suggested by Gust *et al.* (1993) concerning social manipulations in groups of Rhesus monkeys. Resident WBBAs that had experience with novel members joining their groups may be subject to less stress than intruders that had no prior experience with emigration. It is likely that in the wild many WBBAs had experienced immigration of new members into their groups, but that many had remained on their natal territories and had no prior experience emigrating to a new group.

A number of studies have suggested that subordinate animals are subjected to pronounced psychosocial stress, due to decreased access to resources and intimidation by dominants (Louch and Higgenbotham 1967, Rohwer and Wingfield 1981, Silverin *et al.* 1984, Schwabl *et al.* 1988). I found some evidence suggesting a relationship between WBBAs' social position and psychosocial stress during the Exchange Experiment. Three days after the exchange of group members, subordinate WBBAs, those in Access Class 3 and Allofeeding Class "Receivers," tended to have the highest B levels at 30 min post-capture. It seemed that WBBAs in Access Class 3, those that had the lowest priority of access to preferred resources, and those in Allofeeding Class "Receiver, " that accepted food as a submissive ritual, responded to social instability with higher adrenocortical activity than did birds of higher social rank. In effect, subordinate WBBAs seemed more prone to psychosocial stress than did dominant birds.

Testosterone

During the Removal Experiment, T levels were low in all WBBAs. This was in contrast to elevated T titres in wild-caught birds at the same time of year (Chapter III). On the other hand, T levels in WBBAs during the Exchange Experiment were similar to those found in wild birds at the same time of year. The Removal Experiment was undertaken during November and December, toward the end of the natural breeding season. The Exchange Experiment was performed in July, at the beginning of the breeding season. Perhaps seasonal changes elicited a rise in captive WBBAs' T titres in July; conversely, the low levels measured in November and December may have been due to the lack of coincident breeding activity in conjunction with seasonal changes associated with the cessation of the breeding season.

Nevertheless, removal of WBBAs from stable groups did not stimulate a rise in T levels. Likewise, exchange of group members tended to have little effect on T levels. The only striking changes were in two males that exhibited pronounced increases in T levels three days after their introduction into new groups. Unlike many studies that demonstrated a relationship between dominance and/or aggression and high T levels (Rohwer and Wingfield 1981, Hegner and Wingfield 1987b, Schwabl 1992), these two males occupied subordinate positions and exhibited no obvious aggressive behaviour. Perhaps other factors, such as presence of potential mates, stimulated increased T levels in these two males.

A number of studies have demonstrated that T levels were influential during initial encounters with unfamiliar individuals, hierarchy formation, or territory establishment or defense (Ramenofsky 1984, Wingfield 1984b, Hegner and Wingfield 1987b, Wikelski *et al.* 1999). However, another cooperative breeder with year-round territories also seemed to exhibit no relationship between territorial aggression or social change and T titres. In the cooperatively breeding White-browed Sparrow Weaver, removal of breeding males from groups resulted in increased aggression but failed to elicit a concomitant increase in T levels (Wingfield *et al.* 1992). Also, simulated territory intrusions were ineffective in eliciting a rise in T levels in the cooperatively breeding White-browed Sparrow Weaver, despite a marked increase in aggressive behaviour (Wingfield and Lewis 1993). This difference in the reliance on T for social behaviours may be related to the degree of seasonality in the environment and in the behaviours exhibited throughout the year. If social instability or territorial disputes are seasonally predictable, as they are for many north temperate species, it may be beneficial to utilise hormonal cues to enhance readiness for social challenges. However, as chronically high T levels are potentially harmful (Dufty 1989, Moss *et al.* 1994, Beletsky *et al.* 1995, Zuk *et al.* 1995), it may be advantageous for the WBBA and White-browed Sparrow Weaver, that both have long breeding seasons and maintain year-round group territories, to disassociate territorial behaviour and social changes from T. Moreover, in species that maintain social groups year-round, it may be advantageous to disassociate T from social challenges, as social contests (however minor or ritualised) may be almost constant among group members.

Estradiol

Studies of some social species have shown that E2 levels are higher in dominant females than in subordinate females (Mays *et al.* 1991, Schoech *et al.* 1991, Creel *et al.* 1992, but see Schoech *et al.* 1996a). No such pattern was apparent in female WBBAs at any stage of the Exchange Experiment; in fact, all E2 levels were below the sensitivity of the RIA. Neither social instability nor the maintenance or establishment of social position seems to be related to substantial elevations of E2 levels in WBBAs. These findings were consistent with the low levels found in free-living WBBA females; detectable E2 levels were only recorded in a 15% of free-living females (Chapter III). As hypothesized in Chapter III, E2 may have an effect at very low levels (below the detectable limits of the assay I used) or E2 surges may be transient and not easily measured (but see Chapter 7). Perhaps female WBBAs' social behaviour was more closely associated with a different hormone (possibly another estrogenic compound) than it was to E2, and thus no relationship with E2 was observed.

CONCLUSIONS

Changes to WBBA group membership resulted in social restructuring. In groups with familiar members (Removal Experiment), overt, aggressive behaviour did not seem to be necessary for the re-establishment of a social structure following perturbations. However, when novel members were introduced into groups (Exchange Experiment), aggression did seem to play a role. Non-aggressive behaviours, such as allopreening and allofeeding also seemed to be important to the re-establishment of social order in groups of WBBAs in both experiments.

While there were no statistically significant findings indicating a physiological effect of social manipulations, there were a few suggestions in support of such an effect. Body condition tended to decline in response to social manipulations, and some B measures seemed to increase after the perturbations. Social instability is likely psychologically stressful to WBBAs, and a more robust experimental design may have indicated that this psychosocial stress did indeed have a physiological effect. There was little evidence suggesting that social status was related to hormone levels in WBBA groups with stable or unstable social structure. As hypothesized in Chapter V, the complexity of WBBAs social structure may interfere with an accurate analysis of the relationship between social position and hormone levels and/or non-hormonal mechanisms may be more important in the establishment and maintenance of WBBAs' social structure.

Chapter VII. Physiological Correlates of Social Behaviour During Periods of Intense Social Interactions Associated with Roost Nest Building

INTRODUCTION

My investigations of the endocrine relations to WBBAs' social structure have demonstrated few hormonal correlates. The large array of variables associated with WBBAs' complex social structure may have masked differences among group members. In this chapter, instead of focusing on the hormonal and behavioural differences among group members, I chose to examine whether a specific cooperative behaviour, roost nest building, has endocrine correlates. Because WBBAs' roost nest building behaviour is very similar to brood nest building behaviour (pers. obs.), I chose to examine whether the same hormones known to be associated with nest building in breeding birds might be evident in birds stimulated to build roost nests.

It has been shown that reproductive hormones are involved with brood nest building behavior (Lehrman 1961, Silverin 1991). Social and/or environmental stimuli likely result in endocrine changes which then facilitate brood nest building behavior. Although outside the context of reproduction, a different set of social and/or environmental stimuli probably also prompt roost nest building. The functions of brood and roost nests are distinct; brood nests typically protect eggs and provide a site for incubation, and roost nests act as a site for communal nighttime roosting. Nevertheless, nest building behavior is, in essence, the same regardless of nest function; birds bring nesting material to a specific site and construct the nest. If the endocrine system is involved in facilitating the construction of brood nests, might the same mechanisms also play a role in the cooperative building of roost nests?

During the course of my field and aviary observations of WBBAs, I observed entire groups taking part in building new roost nests, and all group members used these nests as communal nighttime roosts. Presumably, WBBAs' roost nests offer thermal benefits and protection from predators, as they do in Sociable Weavers (White *et al.* 1975), Verdin (Buttemer *et al.* 1987), and Green Woodhoopoes (Du Plessis and Williams 1994). White-browed Babblers' roost nests I examined were bulky, stick nests which were lined with bark strips. Nest designs varied, and active roost nests could be partially or completely domed or simple platforms. Some nests were large and could accommodate many birds; up to seven birds used a single roost nest in the aviary. In the closely related Chestnut-crowned Babblers *Pomatostomus ruficeps*, up to nine free-living birds were captured roosting in a single nest (Astheimer pers. comm.). Within some free-living WBBA groups' territories, there were up to three clusters of roost nests, with more than five nests per cluster; however, nests were in various states of repair. Groups were frequently seen cooperatively repairing old roost nests or building new ones. Although the active construction of brood and roost nests was similar, all group members built roost nests, but only breeding pairs built brood nests. The cooperative construction of nests, as well as the use of communal roosts, appears to be an important social activity which may strengthen social bonds.

The endocrine system may facilitate coordination of nest building behaviour. Numerous correlative and experimental studies have found that nest building is associated with changes in reproductive hormones in breeding birds. In various avian species, P, Prl, E2, T, and/or LH have been shown to either correlate with the nest building stage or stimulate nest building when injected or implanted (Lehrman 1961, Silverin 1991). For example, elevated Prl levels were recorded in breeding female

Harris' Hawks and Starlings during brood nest construction (Vleck *et al.* 1991, Dawson and Goldsmith 1982, respectively). Intensive brood nest building was induced in ovariectomised Ring Doves by E2 and P treatment (Cheng and Silver 1975) and in ovariectomised Budgerigars *Melopsittacus undulatus* by injection of E2 and Prl (Hutchinson 1975). Brood nest building was also induced by injection of T in Weaver Birds *Quela quela* and Black-crowned Night Herons *Nycticorax nycticorax* (Crook and Butterfield 1968, Noble and Wurm 1940, respectively). Furthermore, when building brood nests, T levels were high in breeding male WBBAs, and P titres were elevated in breeding females (Chapter III).

While these correlations suggest a relationship between nest building behaviour and certain hormones, a number of other physiological and behavioural changes take place concurrently with brood nest building. This makes the relationship between nest building and hormone levels difficult to tease apart from the behavioural and hormonal changes associated with breeding events, such as territory establishment and defense, spermatogenesis, and ovarian development. By examining WBBAs' hormone levels in relation to roost nest building, I sought to reveal whether sex steroids serve a coordinating role in nest building behaviour without the confounding influence of reproductive behaviour.

To investigate whether sex steroids were associated with nest building behaviour when dissociated from breeding, I examined levels of hormones in captive groups of WBBAs building roost nests. I chose to examine some of the hormones which are thought to be associated with nest building in breeding birds: T, E2, and P. Because of limited plasma volumes, other hormones (such as Prl) were not be evaluated. In this chapter, I report on experiments in which I induced nest building behaviour by destroying existing nests. Roost nest building behaviour is described, and hormone levels are compared between building and non-building WBBA groups.

MATERIAL AND METHODS

Housing and Capture

Aviary conditions and capturing techniques were identical to those described in Chapter IV.

Experimental Protocol

Nest building experiments were performed on six captive groups of WBBAs. Groups were made up of varying numbers of adult males and females (Table 7-1). Experiments were undertaken on three groups in January and February (Trial 1) and on three groups in May and June (Trial 2). All June experiments were conducted before the winter solstice.

Table 7-1. Number of male and female WBBAs in each group in each trial of the Nest Building Experiment.

Group	Number of Males	Number of Females	Trial	Months
1	1	2	1	Jan/Feb
2	4	3	1	Jan/Feb
3	2	3	1	Jan/Feb
4	2	2	2	May/June
5	3	1	2	May/June
6	3	1	2	May/June
Total	15	12	---	---

Upon introduction to the aviary, each WBBA group was provided with an intact nest taken from the field, as well as loose nesting material consisting of branches and twigs. Of their own volition, all groups disassembled the supplied nest and reconstructed a nest of their own within a few weeks of captivity. This nest was used as a communal nighttime roost by all group members.

All groups were videotaped twice to compare nest building activity before nest removal (non-building) and after nest removal (building). First, a complete nest was used as the focal point and was videotaped for roughly 75% of daylight hours. Two weeks after the initial taping, I destroyed the nest and scattered the sticks and nesting material throughout the aviary. This stimulated nest building activity, and as soon as the birds had constructed a small nest cup, a video camera was trained on the nest. Again, I videotaped activity at the nest for 75% of daylight hours. From the video footage, I quantified the amount of time the birds undertook nest building activities.

Because colours were "washed out" in the video footage (due to poor videotaping skills and low quality tapes), it was difficult to discern colours. As a result individual birds could not reliably be recognised based on their colour bands or any other distinctive features. Thus, although I intended on correlating hormone levels with nest building intensity of individuals, this was not possible.

Blood Sampling and Hormone Measurements

Before and after nest destruction, blood (400 μl) was collected from each bird the morning following videotaping between the hours of 0800 and 1100. Procedures for collection and storage of blood for measurement of T, E2, and P are as detailed in Chapters II and III.

Statistical Analysis

Standard parametric statistics were employed. "Nest building intensity" was determined from the videotape and defined as the total number of minutes the group actively engaged in nest building activities divided by the total number of minutes the group was videotaped. To analyse seasonal variation (Jan./Feb. vs. May/June) in nest building intensity, I used a Student’s t-test. A two-way ANOVA was used to examine the effects of trial and gender on T levels in building and non-building WBBAs. Paired Student’s t-tests were used to analyse differences in T or E2 titres between building and non-building WBBAs. Correlation analysis was used to explore the relationship between nest building intensity and T or E2 levels, and Pearson’s product-moment correlation coefficients are reported.

RESULTS

Roost Nest Building Behaviour

Roost nest building was stimulated by nest destruction, and construction of all new nests was begun within eight days of nest demolition. All groups whose nests were destroyed in Trial 2 (May/June) initiated building within a day of nest destruction; those whose nests were destroyed in Trial 1 (January/February) postponed building for three to eight days after demolition. In all groups a substantial nest was present within a day of the start of building, birds continued to add sticks and lining for many days, and nest maintenance was performed indefinitely.

Once initiated, nest building was undertaken vigorously by all group members. Because I was unable to differentiate colour bands, I could not assess the relative nest building contribution of individuals. However, in each group I frequently observed all group members building simultaneously during the period of videotaping (approximately 8.5 hrs in May/June and 11.5 hrs in January/February). Although fighting never erupted among group members during nest construction, there was a good deal of squabbling over access to nesting material or position on or around the nest. When multiple birds were collecting nesting material or building simultaneously, scolding-type vocalisations were often used, and sometimes a birds would attempt to steal nesting material from another.

Although all groups built in an almost frenzied manner, relative nest building intensity varied among groups (Fig. 7-1). Most groups built for roughly 40% of videotaped hours, while one group built for more than 80% and one for only 25% of videotaped hours (Fig. 7-1). Nest building intensity tended to be higher in Trial 2 (May/June) than in Trial 1 (January/February) (Fig. 7-1), but this trend did not reach statistical significance. White-browed Babblers in Group 5 worked on their old nest (before destruction) for approximately the same amount of time as they constructed their new nest (after destruction) (approx. 37% intensity: Fig. 7-1). Because of this, I have no "non-building" data for Group 5 (see analyses below).

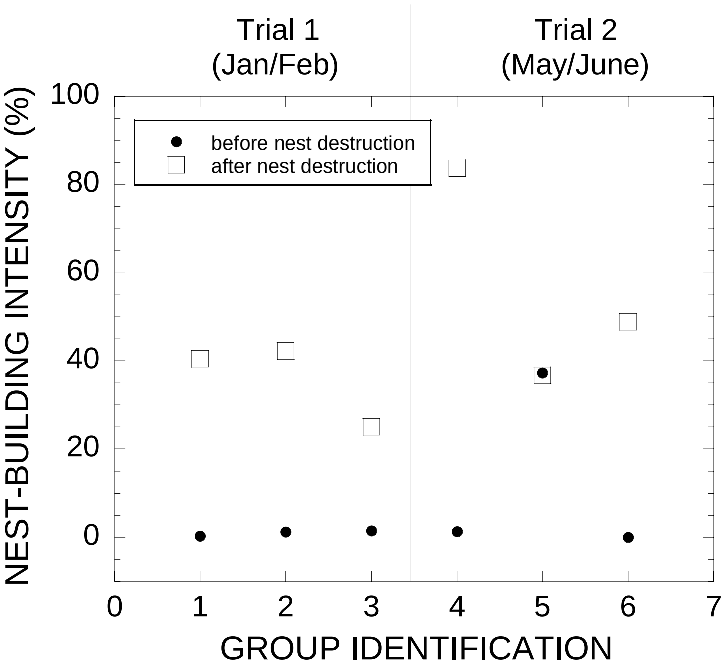


Figure 7-1. Relative nest building intensity before and after nest destruction in six WBBA groups.

Testosterone

Testosterone levels were higher in both building and non-building WBBAs in Trial 2 than in Trial 1, but there were no significant difference between genders (Fig. 7-2, Table 7-2). Because of these findings, further analyses used combined data from both genders, but separate data between Trials.

Table 7-2. Factors potentially associated with T levels during the Nest Building Experiment. Two, two-way ANOVA examining the effects of trial and gender on plasma T levels in building and non-building WBBAs.

Factors	Building		Non-building	
Trial	F-$_{1,22}$=54.780	P<0.001	$F_{1,18}$=42.335	P<0.001
Gender	F-$_{1,22}$=0.656	P=0.427	$F_{1,18}$=0.543	P=0.471
Interaction	F-$_{1,22}$=0.130	P=0.722	$F_{1,18}$=0.469	P=0.502

During Trial 1 T levels were higher in building than non-building WBBAs (paired t_{12}=4.381, P=0.001), but during Trial 2 T levels were similar in building and non-building WBBAs (paired t_7=-0.348, P=0.738) (Fig. 7-2). In Trial 2, plasma T levels increased from non-building to building samples in 50% the birds, while the opposite was true for the other 50%.

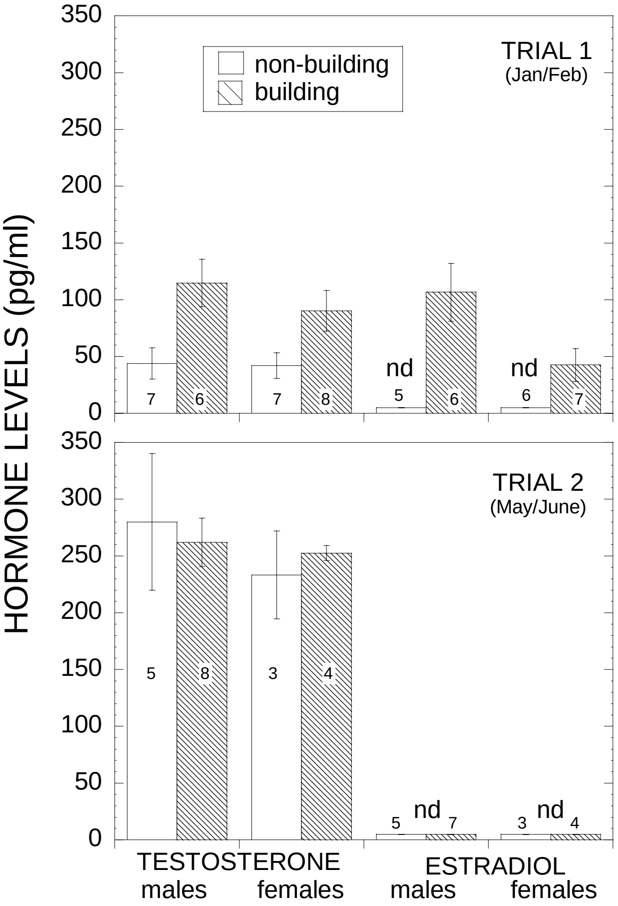


Figure 7-2. Comparisons of plasma titres of T and E2 in experimentally induced roost-nest building and non-building WBBAs in Trial 1 and 2. “nd” indicates that the hormone was not detected in the plasma samples. Height of columns indicates means, and bars represent one standard error. Numbers inside columns indicate sample sizes.

Next, I examined whether the intensity of nest building could be correlated with T levels. Because I could not determine individuals’ nest building intensity, I compared the groups’ nest building intensity to the groups’ mean T titre. Intensity of nest building seemed to be positively correlated with T levels in Trial 1 but not in Trial 2 (Fig. 7-3). However, this trend in Trial 1 was not significant (r=0.041, P=0.938).

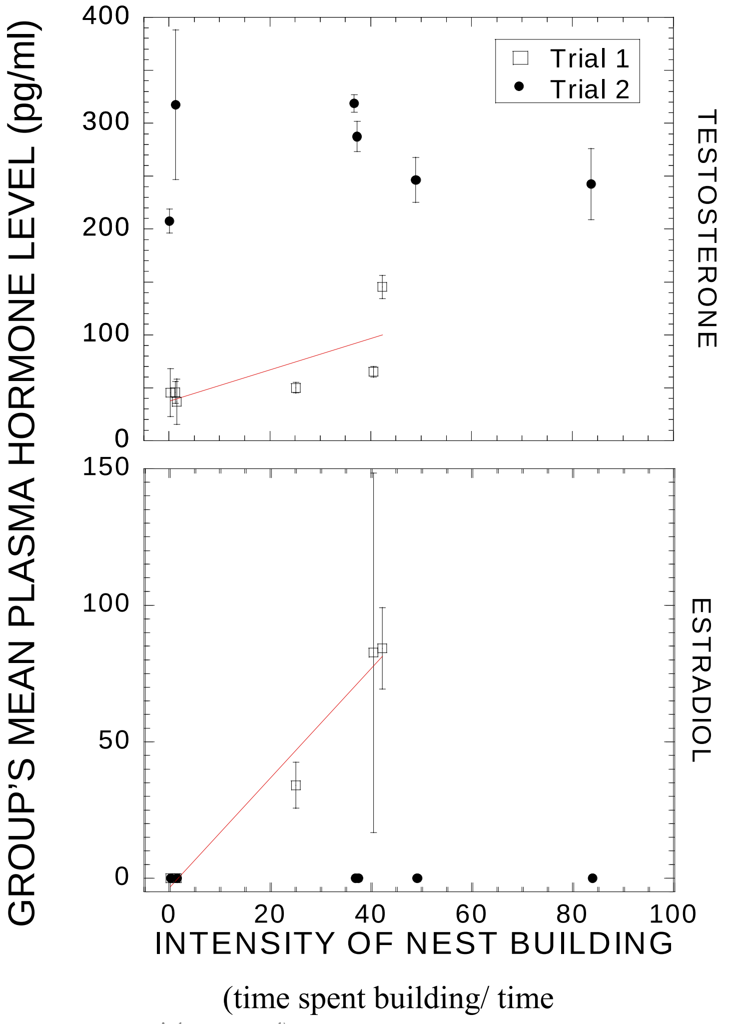


Figure 7-3. Intensity of nest building in relation to WBBA groups' mean plasma T and E2 levels in Trial 1 and Trial 2. Points indicate means, and bars represent one standard error of the mean.

Estradiol

Estradiol only reached measurable levels when WBBAs were building roost nests in Trial 1 (Fig. 7-2); at all other times during this experiment, E2 levels were below detectability of the assay. During Trial 1 building males had significantly higher E2 than both building females (t_{11}=2.272, P=0.044) and non-building males (paired t_3=3.289, P=0.046). Building females also tended to have higher E2 than non-building females in Trial 1, but this trend did not reach significance (paired t_4=2.302, P=0.083).

When males and females were grouped in Trial 1, building WBBAs had significantly higher E2 than non-building WBBAs (paired t_8=3.086, P=0.015). Although sample sizes were small, group’s nest building intensity was positively correlated with group’s mean E2 level in Trial 1 (Fig. 7-3, r=0.988, P<0.001).

Progesterone

Because of limited plasma levels in Trial 2, P levels were measured only in Trial 1. Of 15 WBBAs in Trial 1, only three females and one male had measurable P levels. Of these, the male and two females had higher P when they were not building than when they were building nests. A single female had higher P levels when she was building than when she was not building nests. All measurable P titres were close to non-breeding levels recorded in free-living females (Fig. 3-7).

DISCUSSION

Nest Building

In captive WBBAs, there was a positive relationship between roost nest building behaviour and plasma levels of T and E2, but not P in some months of the year. Similar correlations have been noted in other species building brood nests. For example, during the brood nest building period, T levels were high in male Northern Mockingbirds, Florida Scrub-Jays, and White-crowned Sparrows (Logan and Wingfield 1995, Schoech *et al.* 1991, Wingfield and Farner 1978a, respectively), and E2 levels were high in brood nest building female Starlings, Florida Scrub-Jays, and White-crowned Sparrows (Dawson 1983, Schoech *et al.* 1991, Wingfield and Farner 1978a,b, respectively). Furthermore, T treatment led to an increase in brood nest building activity in male Weaver Birds and Northern Mockingbirds (Crook and Butterfield 1968, Logan and Carlin 1991, respectively), and exogenous E2 stimulated brood nest building behaviour in Peach-faced Lovebirds *Agapornis roseicollis* (Orcutt 1967). Just as T and E2 are involved with nest building in breeding birds, they may also serve roles in nest building behaviour in non-breeding WBBAs (at least in some months of the year).

When this experiment was first conducted (Trial 1), both T and E2 seemed to be significantly associated with roost nest building. However, when the experiment was repeated (Trial 2), no hormonal correlates were observed. This seasonal difference in hormonal changes associated with nest building suggests several hypotheses. During Trial 2 (late Austral autumn), days are shorter and nights cooler than during Trial 1 (mid-summer). Thus, destruction of the nest may have substantially increased discomfort and night-time energy expenditure required to maintain body temperature. These conditions may have directly affected the central nervous system, which then may have stimulated nest building activity via cognitive pathways. The very rapid nest building response in Trial 2 (building was initiated within 24 hrs) provides suggestive evidence for this. In the warmer months (Trial 1), the stimulus to build may not have been as intense as in the colder months (Trial 2). The inability to roost together in a nest may have stimulated nest building via cognitive and hormonal pathways, resulting in a delayed nest building response (building was initiated 3-8 days post-demolition) and the observed hormonal changes.

In another possible scenario, elevated T levels in Trial 2 may have masked subtle changes associated with nest building. Small increases in plasma T may have occurred in response to nest destruction or nest building activity; there was even a trend to suggest this in female WBBAs in Trial 2. Perhaps nest destruction, the inability to roost in a nest at night, or nest building behaviour may have stimulated a slight increase in T levels in both trials, but this change was only measurable when basal T levels were low in Trial 1. However, this does not explain the uniformly low E2 levels in Trial 2. Because WBBAs in Trial 2 began building new nests soon after nest destruction and blood was sampled approximately 24 hrs after nest initiation, the time course between nest destruction and blood sampling was less than 48 hrs. It is possible that there was a surge in E2 levels in Trial 2, but that it did not occur until many days after nest destruction and after the blood sampling event.

Finally, it is possible that the HPG axis was more active or sensitive to social factors in January and February than in May and June. In accordance with this hypothesis, the social stimuli associated with nest building may have affected the HPG axis to a greater extent in Trial 1 than in Trial 2. The associated hormonal fluctuations observed in WBBAs may reflect the variable response of the HPG axis to social perturbations and may hold no functional significance to nest building behaviour per se.

Comparison of Plasma Hormone Levels in Wild and Free-living WBBAs

Seasonal differences in T levels in captive WBBAs paralleled those found in free-living male WBBAs. Testosterone levels measured in non-building, captive WBBAs in January and February (Trial 1) were close to the low levels recorded in wild-caught WBBAs at the same time of year (Fig. 3-1). In the wild most pairs had ceased breeding attempts by late December, and sex steroids were low during this post-breeding period. Some free-living WBBAs began pairing as early as May, and nesting activity typically began in June, often prior to the solstice (Chapter II). Concomitantly, T levels rose in free-living male WBBAs at the beginning of the breeding season (Fig. 3-1). In May and June (Trial 2), T levels in both building and non-building, captive WBBAs were elevated to similar levels as those found in wild males caught during June. Perhaps WBBAs' HPG axis responds to the same internal and environmental cues in both wild and captive WBBAs.

In captive WBBAs, E2 increased to detectable levels only in roost nest building birds during Trial 1 (January and February). In free-living WBBAs, only females were sampled for E2 and only from July through December; most had levels too low to detect in our assay (Chapter III). More data would be necessary to determine if the plasma E2 levels recorded in captive WBBAs are typical of free-living WBBAs at the same time of year.

Gender Comparison

Elevated levels of T are traditionally associated with male behaviour and reproductive function and high E2 and P levels with female reproductive behaviour and physiology. However, T is also secreted by females and E2 and P by males. Relatively few studies have examined the levels of T in females and E2 and P in males, and these have revealed large variations in relative levels. For example, in breeding Harris' Hawks and breeding Northern Mockingbirds, T levels are higher in males than females and E2 levels are higher in females than males (Mays *et al.* 1991, Logan and Wingfield 1995). However, E2 and T levels are similar in breeding male and female Western Gulls *Larus occidentalis* (Wingfield *et al.* 1980, Wingfield *et al.* 1982), and E2 levels are similar in male and female Zebra Finches (Adkins-Regan *et al.* 1990).

In non-breeding, nest building WBBAs, I found equivalent levels of T in male and female WBBAs, higher E2 titres in males than females, and consistently low P levels in both genders. In males elevated T is often associated with spermatogenesis and secondary sexual characteristics (Vleck and Brown 1999). In female vertebrates androgens have been shown to be involved with a variety of roles, including neuronal growth, immune functions, communication, and aggressive and sexual behaviour (Staub and De Beer 1997). Testosterone may function in some or all of these roles in WBBAs as well. Not only was T measured in female WBBAs, but E2 was also found in male WBBAs. While E2 serves a role in follicular maturation and vitellogenesis in females (Balthazart 1983), in males estrogens may regulate seasonal cycles of T (Mak *et al.* 1983) and/or influence behaviour (Balthazart 1990, Schlinger and Callard 1990). Unlike both T and E2, P was not notably elevated in either male or female WBBAs in Trial 1 (January and February). As P is typically associated with oviduct development and ovulation (Silver 1990, Tanaka and Inoue 1998), it was not surprising that P levels were uniformly low at the time of year when free-living WBBAs were concluding breeding behaviour. Progesterone may not play a large role in non-breeding physiology or behaviour in WBBAs.

CONCLUSIONS

Just as in many north temperate birds living in highly seasonal environments, WBBAs seemed to respond to seasonal changes with dependable fluctuations in plasma T levels. Even when held in captivity and not subject to the full brunt of environmental vagaries (i.e. captive birds were provided with dependable food resources and ample shelter), WBBAs' endocrine system still responded to the relatively minimal seasonality of the Australian environment. While interesting, this seasonal effect also added unwanted variability to my study of the hormonal correlates of nest building behaviour.

In some months of the year, there did seem to be a measurable hormonal response associated with roost nest building behaviour or to the social stimuli associated with building a group nest. At least in January and February, T and E2 may help direct nest-building behaviour in non-breeding, roost nest building WBBAs. Further study would be needed to determine whether the endocrine system responds to or is stimulated by roost nest building behaviour in WBBAs and why this association is not apparent in some months.

While T is typically considered a "male" hormone and E2 a "female hormone," comparable levels of both hormones were recorded in male and female WBBAs. Especially in non-breeding birds, T and E2 may serve some of the same functions in both males and females. Relatively few avian studies have examined T levels in females and E2 levels in males, and this study suggests a need to further investigate these "cross-gender" endocrine relations.

Chapter VIII. General Conclusions

Evolution of Alloparental Behaviour

Because “altruistic” behaviour, such as alloparental behaviour, seems inherently maladaptive, it is particularly intriguing. I set out to test some of the principal hypotheses that endeavour to explain the evolution of alloparental behaviour by using a multi-faceted approach that employed behavioural, genetic, and endocrinological measures. In Chapter I, I advanced three hypotheses that attempt to explain the motivation behind the expression of alloparental behaviour. While not necessarily mutually exclusive, these hypotheses propose different mechanisms to help rationalise this seemingly altruistic behaviour. The *kin selection hypothesis* (Hamilton 1964) stresses the inclusive fitness benefits of “altruistic behaviours,” such as alloparental behaviour. It posits that behaviours which benefit kin will also enhance the inclusive fitness of the individual demonstrating the behaviour. The *competitive altruism/ handicap principle* (Zahavi 1975, Roberts 1998) suggests that seemingly altruistic behaviours are in fact advertisements of an individual’s quality and may be used by group members to judge and choose potential collaborators or mates. Finally, the *unselected consequence of communal breeding hypothesis* (Jamieson 1989, 1991) doubts that “altruistic behaviour,” such as alloparental behaviour, is adaptive. Instead, it suggests that when non-parental individuals are in close contact with young, innate pressures prompt alloparental care; these innate pressures result from strong evolutionary selection for parental care, and these innate pressure stimulate provisioning behaviour regardless of kinship relations.

My data best support the kin selection and competitive altruism hypotheses explaining alloparental behaviours. In WBBAs alloparental behaviours seem to be either directed toward kin or co-opted as a means of advertising social status. Further, I put forth some evidence which challenges Jamieson's (1989) unselected consequence hypothesis. Some of my hormonal data suggests that the endocrine system has been fine-tuned to promote the expression of alloparental behaviour, which suggests that such behaviour is, in fact, an evolutionarily derived trait.

Kinship appears to play a large role in WBBA social behaviour, and WBBAs seem to behave nepotistically toward their kin. White-browed Babbler social groups include many genetically related individuals; members of the same social group are more closely related to each other than are members of different social groups. Cooperative behaviours, such as communal foraging and roosting, likely benefit all group members, resulting in enhanced individual and kin fitness. Further, parents (or other close relatives) frequently allofeed adult kin and allow kin to have preferred access to limited resources. (i.e. meal worms in the aviary). Some alloparental behaviours are clearly directed toward kin and, under field conditions, would likely result in enhanced inclusive fitness.

In many instances, alloparental behaviour in WBBAs acts as a social signal. Extensive observations and manipulative experiments on captive birds leads me to believe that allofeeding is an assertion of social status and can also function as a sexual display. In WBBAs allofeeding seems to take the place of aggression (seen in many other species) and is used as a means of establishing and maintaining social status among group members. The exchange experiment (Chapter VI) highlights this, as aggression was rarely observed during the integration of new group members, but there was a high frequency of dominant residents that allofed intruders. The resident birds seemed to establish their social dominance by allofeeding intruding birds. In stable groups the most frequent allofeeding interaction was males feeding unrelated females.

In such cases, allofeeding may be a form of sexual advertisement or a pair-bonding ritual. These finding support Zahavi's (1975) "handicap principle" which interprets seemingly altruistic behaviours as a means of displaying quality. In the same manner as a Peacock's tail, altruistic behaviour is a costly display (or handicap) that advertises an animal's ability to overcome this handicap and thus demonstrates its high quality. By feeding another adult bird, the allofeeder signals its quality by demonstrating its ability to nourish itself, despite giving up some food to others. Furthermore, allofeeding also acts as a direct signal of provisioning ability directed at attracting potential mates; the allofeeder displays its ability and willingness to provision others, and thus its potential to provision offspring.

Jamieson's (1989) "unselected consequence hypothesis" suggests that alloparental behaviour is not adaptive, but instead is an unselected consequence of communal breeding. However, there is accumulating evidence (e.g. Vleck *et al.* 1991, Schoech *et al.* 1996b, Brown and Vleck 1998) that alloparental behaviours have endocrine-based antecedents, therefore appear to be reinforced hormonally. This suggests that these behaviours provide fitness benefits for the species displaying them and are therefore selected traits. Some of my findings add to this body of evidence. Elevated Prl levels and depressed T levels may promote alloparental behaviour in free-living adult male WBBAs that chaperone fledglings and juveniles. Just as low T and high Prl titres help promote parental behaviour, they may also play a role in promoting alloparental behaviour. Furthermore, there seems to be a correlation between elevated P and non-reproductive behaviour in female WBBAs. In the WBBA, it was not determined whether high P titres inhibited reproductive behaviour or a lack of reproductive behaviour resulted in elevated P levels. However, it is possible that P plays a role in suppressing reproduction, as it does in a number of animals (see Chapter 3) and may even support alloparental behaviour in some female WBBAs. As it seems that the endocrine system has been modified to support alloparental behaviour, at least in some species, alloparental behaviour probably is adaptive, rather than simply being an unselected consequence of group living. Both the kin selection hypothesis and competitive altruism/ handicap principle are compatible with the assertion that alloparental behaviour is adaptive. In the kin selection model, alloparental care enhances inclusive fitness, and according to the handicap principle, alloparental care serves as an honest signal of quality which can be used to select mates or collaborators.

Phylogenetic Inertia and Social Plasticity

As suggested by Edwards and Naeem (1993) and Cockburn (1996, 2003), there seems to be a strong phylogenetic disposition toward cooperative breeding. Furthermore, the selective pressures which have impacted upon the evolution of cooperative breeding have likely changed over time, but cooperative breeding has endured in many taxa, suggesting that cooperative breeding persists due to "phylogenetic inertia" (Edwards and Naeem 1993). The WBBA seems to be an example of such a species, and, in fact, Edwards and Naeem (1993) found that cooperative breeding has persisted in members of the genus *Pomatostomus*, despite their invasion of a wide range of habitats. Cooperative breeding likely evolved under different ecological conditions than are present today, and thus current conditions may not help explain the distribution of cooperative breeders or the evolution of cooperative behaviours. However, current ecological conditions may play a role in maintaining and modifying cooperative behaviours.

Because WBBAs have a large range that encompasses many different types of habitats, different populations are subject to varying degrees and types of ecological constraints. These variable conditions can influence the social behaviours exhibited by different populations. While all WBBAs probably tend toward cooperative breeders, there does seem to be some plasticity to their social structure. Some conditions promote singular cooperative breeding (e.g. Kellerberin, Western Australian: Cale 2003a), while others appear to support plural cooperative breeding (e.g. Central West, NSW: present study). It remains unclear what drives this variation in social structure, but predation pressures and variable degrees of habitat fragmentation may be factors. In my study a high degree of predation and presumed good-quality habitat in a continuous tract of vegetation may encourage multiple pairs within social groups to make separate nesting attempts. Furthermore, increased activity around the nest (e.g. allofeeding by multiple group members) may be discouraged due to high predation pressures. In the highly fragmented landscape of the Kellerberin region of Western Australia, the habitat may only support a single breeding attempt per social group, predation pressures may be lower, and help by all group members may be crucial for successful rearing of young.

While pair breeding is considered the norm among many northern hemisphere passerines and singular cooperative breeding is recognized as a typical breeding system among many Australian species, there is a continuum of breeding strategies between these two extremes. Multiple factors help position birds along this continuum (Fig. 8-1). The benefits of group living and a phylogenetic disposition toward cooperative behaviours encourage cooperative breeding, while high habitat predictability and richness, a high degree of seasonality, high predation pressures, and a phylogenetic disposition toward pair breeding promote pair breeding (Fig. 8-1). In species that breed across a large geographical range (such as the WBBA), some of these factors vary across breeding sites. In such cases, members of the same species may exhibit alternate breeding strategies depending on the factors associated with the breeding locale.

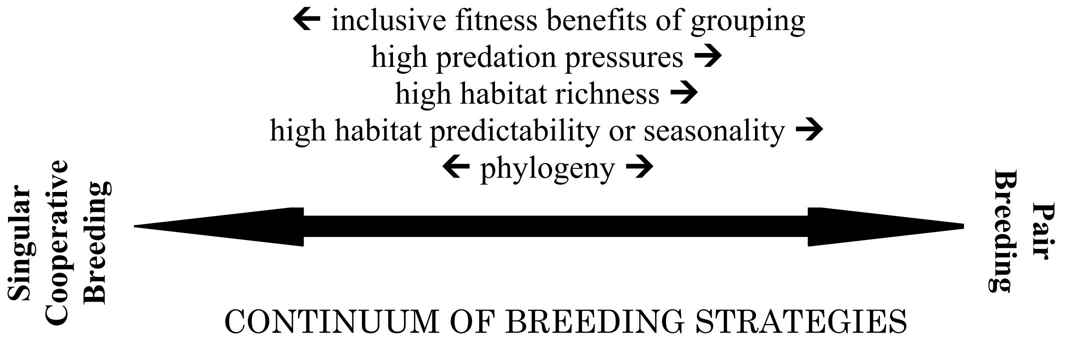


Figure 8-1. Pressures affecting the placement of species along a theoretical continuum of breeding strategies.

Endocrine Correlates to Reproduction

There is a wealth of information on the relationship between the endocrine system and reproduction in northern temperate regions, where there is high seasonality and predictability of resources. In such situations, photoperiodic cues stimulate the endocrine system to initiate breeding readiness in many species. Much less in know about species that breed aseasonally or for extended periods. It is likely that WBBAs are capable of breeding year-round and that local conditions are more important in promoting breeding readiness than is “initial predictive information,” such as photoperiod (Wingfield *et al*. 1992). In fact, male WBBAs’ T levels increase prior to the winter solstice, when day length is shortening. During the non-breeding period, testes only partially regress, and many females can have well-developed ovaries year-round. Maintaining gonads in an advanced state of reproductive readiness may enable WBBAs to initiate breeding whenever local conditions permit.

While WBBAs’ extended (or continuous) period of breeding readiness appears distinct from the highly seasonal breeding season of many northern temperate species, the physiological and behavioural changes associated with reproduction seem to be influenced by some of the same hormone secretions. Male WBBAs' T profiles correspond to breeding stages and resemble those of many northern hemisphere polygynous passerines. Mated male's T levels are elevated during the period of their mate's fertility, but also tend to rise after the completion of her clutch. This second peak in T levels suggests that male WBBAs seek extra-pair matings. Male WBBAs are exposed to multiple breeding females within their social groups, as well as to females in neighboring groups; there are probably many opportunities for extra-pair forays.

While the overall patterns of T secretions in male WBBAs are not remarkably different from those of many northern temperate species, the absolute levels of T are considerably lower. As various detrimental effects, including injury resulting from aggressive behaviour, are attributed to prolonged elevations of T, WBBAs' extended breeding period and their gregarious society may be incompatible with high levels of T. Alternately, their long term relationships within a sedentary group may rely more on cognitive choices than endocrine signals.

Endocrine Correlates to Social Behaviours

Many researchers have examined the relationships between hormone levels and non-reproductive social behaviours, but results have been ambiguous. Some studies have uncovered relationships between corticosteroid secretion and social position or Prl and nurturing behaviour, while others have found an absence of such relationships. In the WBBA, I found few hormonal correlates to non-breeding social behaviours. In stable groups, there is no association between behavioural measures and Prl titres or stress responsivity. Although allofeeding in a nurturing context may be facilitated by elevated Prl titres, allofeeding in a purely social context does not seem to be related to Prl levels. Further, I found no evidence that holding either dominant or subordinate positions within the group was more or less stressful (measured by the B capture stress response) than the other. In the captive setting, where there are no breeding opportunities and abundant food resources are available, there may be little social significance to dominance or subordinance; as a result, social status may not impact on the endocrine system. However, there was considerable inter-individual variation in the capture stress response. Multiple factors, such as age, intragroup social and kinship bonds, body mass, and body condition, likely contribute to this variation. Furthermore, the effects of captivity and repeated handling probably acted as confounding variables, as demonstrated by the habituation of the stress response in the “Removal Experiment” (Chapter 6).

While there were obvious behavioural responses to social manipulations and some indication that social manipulations effected a decline in body condition, there were no significant effects on the levels of hormones that I measured. Even in socially unstable groups, neither dominant nor subordinate social positions shared a pattern in B elevations. Likewise, the reproductive steroids, T and E2, did not play a clear role in social restructuring following social manipulations.

There was some evidence that elevated T and E2 contributed to the cooperative social activity of roost nest building. However, there was a distinct seasonal variation in this relationship. Testosterone and E2 levels were associated with nest building activity during late Austral autumn but not mid-summer. This relationship begs further investigation. Further study would examine whether the endocrine system responds to or is stimulated by roost nest building behaviour in WBBAs and could explore the seasonal variation in the relationship between roost nest building and the endocrine system.

My research on WBBAs suggests a number of broad research questions for further study:

1. While there is a great deal of information on social and agonistic behaviours among reproducing animals, there is relatively little on such behaviours outside the context of breeding. While reproductive behaviours are undoubtedly important, behavioural studies performed outside the breeding season should also provide interesting and valuable information, particularly on animals that are sedentary residents of their habitat.
2. What degree of influence do various factors have on the placement of species along the continuum of sociality? Recent studies highlight the importance of a phylogenetic disposition toward a particular social structure. Under what situations do environmental factors override phylogeny? What makes some species, such as the WBBA, plastic in their social organisation for reproduction?
3. For various animal behaviours, at what point does the influence of the endocrine system end and cognitive appraisal begin? While endocrine cues may often help direct behaviour, cognitive interpretation, often based on long-term associations and memory, should not be underestimated in the study of behaviour.

To pursue these issues, an integrative approach is needed. A wealth of molecular and biochemical techniques are now available to behavioural biologist, and such techniques should be utilized for a truly integrative approach when examining animal behaviour.

LITERATURE CITED

Abbott, D.H., E.B. Keverne, F.B. Bercovitch, C.A. Shively, S.P. Mendoza, W. Saltzman, C.T. Snowdon, T.E. Ziegler, M. Banjevic, T. Garland, Jr., and R.M. Sapolsky. 2003. Are subordinates always stressed? A comparative analysis of rank differences in cortisol levels among primates. Horm. Behav. 43: 67-82.

Adkins-Regan, E. 1981. Hormone specificity, androgen metabolism, and social behavior. Am. Zool. 21: 257-271.

Adkins-Regan, E., M. Abdelnabi, M. Mobarak, and M.A. Ottinger. 1990. Sex steroid levels in developing and adult male and female Zebra Finches (*Poephila guttata*). Gen. Comp. Endo. 78: 93-109.

Alcock, J. 1975. Animal Behavior: An Evolutionary Approach. Sinauer Associates, Inc. Sunderland, MA.

Alcock, J. 2001. Animal Behavior: An Evolutionary Approach. Sinauer Associates, Inc. Sunderland, MA.

Allee, W.C. 1926. Studies in animal aggregation: causes and effects of bunching in land isopods. J. Exp. Zool. 45: 255-277.

Ambrose, S.J. 1985. Aspects of the physiological and behavioural ecology of the White-browed Scrubwren, *Sericornis frontalis* (Acanthizidae), in Western Australia. Ph.D. thesis, University of Western Australia.

Appleby, M.C. 1983. The probability of linearity in hierarchies. Anim. Behav. 31: 600-608.

Arrowood, P.C. 1988. Duetting, pair bonding and agonistic displays in parakeet pairs. Behav. 106: 129-156.

Asa, C.S. 1997. Hormonal and experiential factors in the expression of social and parental behavior in canids. In: Cooperative Breeding in Mammals. N.G. Solomon and J.A. French (eds.). Cambridge University Press, Cambridge. pp. 129-149.

Astheimer, L.B., W.A. Buttemer, and J.C. Wingfield. 1994. Gender and seasonal differences in the adrenocortical response to ACTH challenge in an arctic passerine *Zonotrichia leucophrys gambelii*. Gen. Comp. Endo. 94: 33-43.

Astheimer, L.B., W.B. Buttemer, and J.C. Wingfield.1995. Seasonal and acute changes in adrenocortical responsiveness in an arctic-breeding bird. Horm. Behav. 29: 442-457.

Astheimer, L.B. and W.A. Buttemer. 1999. Gonadal and hormonal patterns in the annual cycle of an Australian honeyeater. In: Proc. 22 Intl. Ornith. Congr., Durban. N.J. Adams and R.H. Slotow (eds.) BirdLife South Africa, Johannesburg. pp. 1768-1783.

Avise, J.C. 1996. Three fundamental contributions of molecular genetics to avian ecology and evolution. Ibis 138: 16-25.

Balda, R.P. and J.L. Brown. 1977. Observation on the behaviour of Hall's Babbler. Emu 77: 111-114.

Balda, R.P. and J.H. Balda. 1978. The care of young Piñon Jays (*Gymnorhinus cyanocephalus*) and their integration into the flock. J. Ornith. 119: 146-171.

Ball, G.F. and J.C. Wingfield. 1987. Changes in plasma levels of LH and sex steroid hormones in relation to multiple-broodedness and nest site density in male Starlings. Physio. Zool. 60: 191-199.

Balthazart, J. 1983. Hormonal correlates of behavior. In: Avian Biology, Vol. 7. D.S. Farner, J.R. King, and K.C. Parkes (eds.). Academic Press, New York. pp. 221-365..

Balthazart, J. 1990. Aromatization of testosterone in the preoptic area and contol of male sexual behavior in the Japanese Quail. In: Endocrinology of Birds: Molecular to Behavioral. M. Wada, S. Ishii, and C.G. Scanes (eds.) Japan Sci. Soc. Press, Tokyo. pp. 297-311.

Baptista, L. F., B.B. DeWolfe, L. Avery-Beausoleil. 1987. Testosterone, aggression, and dominance in Gambel's White-crowed Sparrows. Wilson Bull. 99: 86-91.

Barkan, C.P.L., J.L. Craig, S.D. Strahl, A.M. Stewart, and J.L. Brown. 1986. Social dominance in communal Mexican Jays *Amphelocoma ultramarina*. Anim. Behav. 34: 175-187.

Barta, Z. and T. Szep. 1995. Frequency-dependent selection on information-transfer strategies at breeding colonies: a simulation study. Behav. Ecol. 6: 308-310.

Baum, A., N.E. Grunberg, and J.E. Singer. 1982. The use of psychological and neuroendocrinological measurements in the study of stress. Health Psychol. 1: 217-236.

Baum, G.J. and R.K. Meyer. 1960. Effect of adrenal steroids and diethystilbesterol on growth and fat content of Cockerels. Am. J. Physio. 19: 1263-1266.

Beauchamp, G. 2002. Higher-level evolution of intraspecific flock-feeding in birds. Behav. Ecol. Sociobio. 51: 480-487.

Bednarz, J.C. and J.D. Ligon. 1988. A study of the ecological bases of cooperative breeding in the Harris' Hawk. Ecology 69: 1176-1187.

Bednekoff, P.A. and A.I. Houston. 1994. Dynamic models of mass-dependent predation, risk-sensitive foraging, and premigratory fattening in birds. Ecology 75: 1131-1140.

Beletsky, L.D. and G.H. Orians. 1987. Territoriality among male Red-winged Blackbirds. II. Removal experiments and site dominance. Behav. Ecol. Sociobiol. 20: 339-49.

Beletsky, L.D., G.H. Orians, and J.C. Wingfield. 1990. Steroid hormones in relation to teritoriality, breeding density, and parental behavior in male Yellow-headed Blackbirds. Auk 107: 60-68.

Beletsky, L.D., D.F. Gori, S. Freeman, and J.C. Wingfield. 1995. Testosterone and polygyny in birds. In: Current Ornithology, Vol. 12. D.M. Power (ed.). Plenum, New York. pp.1-41.

Bell, H.L. 1982. Social organization and feeding of the Rufous Babbler *Pomatostomus isidori*. Emu 82: 7-11.

Bentley, D.R. and R.R. Hoy. 1972. Genetic control of the neuronal network generating Cricket (*Teleogryllus gryllus*) song patterns. Anim. Behav. 20: 478-492.

Benzer, S. 1973. Genetic dissection of behavior. Sci. Am. 229: 24-37.

Bertram, B.C.R. 1978. Living in groups: predators and prey. In: Behavioural Ecology: an Evolutionary Approach. Krebs, J.R. and N.B. Davies (eds.) Blackwell Scientific Publications, Oxford. pp 64-96.

Biebach, H. 1996. Energetics of winter and migratory fattening. In: Avian Ecology and Nutritional Ecology. C. Carey (ed.). Chapman and Hall, Inc. New York. pp. 280-323.

Blanchard, B. D. 1941. The White-crowned Sparrows, *Zonotrichia leucophrys*, of the Pacific Seaboard. Environment and annual cycle. University of California Publications in Zoology 46:1-178.

Blem, C.R. 1975. Geographic variation in wing-loading of the House Sparrow. Wilson Bull. 87: 543-549.

Boland, C.R.J., R. Heinsohn, and A. Cockburn. 1997a. Deception by helpers in cooperatively breeding White-winged Choughs and its experimental manipulation. Behav. Ecol. Sociobiol. 41: 251-256.

Boland, C.R.J., R. Heinsohn, and A. Cockburn. 1997b. Experimental manipulation of brood reduction and parental care in cooperatively breeding White-winged Choughs. J. Anim. Ecol. 66: 683-691.

Bronson, F.H. 1973. Establishment of social rank among grouped male mice: relative effects on circulating FSH, LH, and corticosterone. Physio. Behav. 10: 947-951.

Brown, C. R. 1988. Enhanced foraging efficiency through information centers: a benefit of coloniality in Cliff Swallows. Ecology 69: 602-613.

Brown, J.L. 1974. Alternate routes to sociality in Jays- with a theory for the evolution of altruism and communal breeding. Am. Zool. 14: 63-80.

Brown, J.L. 1978. Avian communal breeding systems. Ann. Rev. Ecol. Syst. 9:123-155.

Brown, J.L. 1987. Helping and Communal Breeding in Birds. Princeton University Press, Princeton, NJ.

Brown, J.L., D.D. Dow, E.R. Brown, and S.D. Brown. 1978. Effects of helpers on feeding of nestlings in the Grey-crowned Babbler (*Pomatostomus temporalis*). Behav. Ecol. Sociobiol. 4: 43-59.

Brown, J.L. and E.R. Brown. 1980. Reciprocal aid-giving in a communal bird. Z. Tierpsychol. 53: 313-324.

Brown, J.L. and E.R. Brown. 1984. Parental facilitation: parent-offspring relations in communally breeding birds. Behav. Ecol. Sociobiol. 14: 203-209.

Brown, J.L. and S.L. Pimm. 1985. The origin of helping. The role of variability in reproductive potential. J. Theor. Biol. 112: 465-477.

Brown, J.L. and E.L. Brown. 1990. Mexican Jays: Uncooperative breeding. In: Cooperative Breeding in Birds: Long-Term Studies of Ecology and Behavior. Stacey, P.B. and W.D. Koenig (eds.). Cambridge University Press, Cambridge. pp. 269-288.

Brown, J.L. and C. M. Vleck. 1998. Prolactin and helping in birds: has natural selection strengthened helping behavior? Behav. Ecol. 9: 541-545.

Brown, R.E. 1985. Hormones and parental behavior in vertebrates. Am. Zool. 25: 895-910.

Bugnyar, T. and K. Kotrschal. 2002. Scrounging tactics in free-ranging Ravens, *Corvus corax*. Ethology 108: 993-1009.

Buntin, J.D. 1986. Role of prolactin in avian incubation behavior and care of young: is there a causal relationship? In: Reproduction: a Behavioral and Neuroendocrine Perspective. B.R. Komisaruk, H.I. Siegel, M-F Cheng, and H.H. Feder (eds.). Annals of The NY Academy of Science, Vol. 474, New York. pp. 252-267.

Buntin, J.D. 1996. Neural and hormonal control of parental behavior in birds. In: Advances in the Study of Behavior, Vol. 25. Rosenblatt and Snowdon (eds.). Academic Press, San Diego, CA. pp. 161-213.

Burke, T. and M. Bruford. 1987. DNA fingerprinting in birds. Nature 327: 149-152.

Burt, D.B. and A.T. Peterson. 1993. Biology of cooperative-breeding Scrub-jays (*Amphelocoma coerulescens*) of Oaxaca, Mexico. Auk 110: 207-214.

Buttemer, W.A., L.B. Astheimer, W.W. Weathers, and A.M. Hayworth. 1987. Energy savings attending winter-nest use by Verdins (*Auriparus flaviceps*). Auk 104: 531-535.

Cale, P. 1999. The spatial dynamics of the White-browed Babbler in a fragmented agricultural landscape. Ph.D. thesis, Univ. of New England, Armidale.

Cale, P.G. 2003a. The influence of social behaviour, dispersal and landscape fragmentation on population structure in a sedentary bird. Biol. Conserv. 109: 237-248.

Cale, P.G. 2003b. The spatial dynamics of White-browed Babbler groups in a fragmented agricultural landscape. Pacific Conserv. Biol. 8: 271-280.

Camberfort, J.P. 1981. A comparative study of culturally transmitted patterns of feeding habitats in the Chacma Baboon *Papio ursinus* and the Vervet Monkey *Cercoppithecus aethiops*. Folia Primat. 36: 243-263.

Caraco, T., S. Martindale, and H.R. Pulliam.1980. Avian flocking in the presence of a predator. Nature 285: 400-401

Carlisle, T.R. and A. Zahavi. 1986. Helping at the nest, allofeeding and social status in immature Arabian Babblers. Behav. Ecol. Sociobiol. 18: 339-351.

Carlson, A.A., A.J. Young, A.F. Russell, N.C. Bennett, A.S. McNeilly, and T. Clutton-Brock. 2004. Hormonal correlates of dominance in Meerkats (*Suricata suricatta*) 2004. Horm. Behav. 46: 141-150.

Cavigelli, S.A. 1999. Behavioural patterns associated with faecal cortisol levels in free-ranging Ring-tailed Lemurs, *Lemur catta*. Anim. Behav. 57: 935-944.

Chakraborty, S. 1995. Plasma prolactin and luteinizing hormone during termination and onset of photorefractoriness in intact and pinealectomized European Starlings *(Sturnus vulgaris*). Gen. Comp. Endo. 99: 185-191.

Chandler, L.G. 1920. With camera and field glasses in north-west Victoria. Emu 20: 4-13.

Chandola-Saklani, A., K. K. Sharma, M.S. Bisht, and P. Lakhera. 1990. Ecophysiology of seasonal reproduction in the tropics: the Baya Weaver. In: Endocrinology of Birds: Molecular to Behavioral. M. Wada, S. Ishii, and C.G. Scanes (eds.). Japan Sci. Soc. Press, Tokyo. pp. 207-224.

Chard, T. 1987. An introduction to radioimmunoassay and related techniques. 3rd edition. Elsevier, Oxford.

Chase, I. D. 1982. Dynamics of hierarchy formation: the sequential development of dominance relationships. Behav. 80: 218-240.

Cheng, M-G and R. Silver. 1975. Estrogen-progesterone regulation of nest-building and incubation behavior in ovariectomized Ring Doves (*Streptopelia risoria*). J. Comp. Physio. Psych. 88: 256-263.

Clarke, M.F. 1984. Co-operative breeding by the Australian Bell Miner *Manorina melanophrys* Latham: a test of kin selection theory. Behav. Ecol. Sociobiol. 14: 137-146.

Clarke, M.F. 1989. The pattern of helping in the Bell Miner (*Manorina melanophrys*). Ethology 80: 292-306.

Clutton-Brock, T.H. 1991. The evolution of Parental Care. Princeton Univ. Press. Princeton, N.J.

Cockburn, A. 1996. Why do so many Australian birds cooperate: social ecolution in the Corvida?. In: Frontiers of Population Ecology. R.B. Floyd, A.W. Sheppard, and P.J. Debarro (eds.). CSIRO Pub., Melbourne. pp.451-472.

Cockburn, A. 1998. Evolution of helping behavior in cooperatively breeding birds. Ann. Rev. Ecol. Syst. 29: 141-177.

Cockburn. A. 2003. Cooperative breeding in oscine passerines: does sociality inhibit speciation? Proc. R. Soc. Lond. B 270: 2207-2214.

Coe, C.L., S.L. Mendoza, and S. Levine. 1979. Social status constrains the stress response in the Squirrel Monkey. Physio. Behav. 23: 633-638.

Coe, C.L., E.R. Smith, S.P. Mendoza, S. Levine. 1983. Varying influence of social status on hormone levels in male Squirrel Monkeys. In: Hormones, Drugs, and Social Behavior in Primates. H.D. Steklis and A.S. Kling (eds.). S.P. Medical, New York. pp. 7-32.

Counsilmann, J.J. and B. King. 1977. Ageing and sexing the Grey-crowned Babbler (*Pomatostomus temporalis*). Babbler 1: 23-41.

Craig, A.J.F.K. 1988. Allofeeding and dominance interactions in the cooperatively breeding Pied Starling. Anim. Behav. 36: 1251-1253.

Craig, J.L. 1979. Habitat variation in the social organization of a communal gallinule, the Pukeko, *Porphyrio porphrio melanotus*. Behav. Ecol. Sociobiol. 5: 331-358.

Craig, J.L. 1980. Pair and group breeding behaviour of a communal gallinule, the Pukeko, *Porphyrio porphrio melanotus*. Anim. Behav. 28: 593-603.

Craig, J.L. and M.E. Douglas. 1986. Resource distribution, aggressive asymmetries and variable access to resources in the nectar feeding Bellbird. Behav. Ecol. Sociobiol. 18: 231-240.

Craig, J.L. and I.G. Jamieson. 1990. Pukeko: different approaches and some different answers. In: Cooperative breeding in birds. Stacy, P.B. and W.D. Koenig (eds.). Cambridge University Press, Cambridge. pp. 387-412.

Creel, S. 2001. Social dominance and stress hormones. TREE 16: 491-497.

Creel, S., N. Creel, D.E. Wildt, S.L. Monfort. 1992. Behavioural and endocrine mechanisms of reproductive suppression in Serengeti Dwarf Mongooses. Anim. Behav. 43:231-245.

Creel, S. N. Creel, and S.L. Monfort. 1996. Social stress and dominance. Nature 379: 212

Crespi, B.J. and D. Yanega. 1995. The definition of eusociality. Behav. Ecol. 6: 109-115.

Crews, D. 1997. Species diversity and the evolution of behavioral controlling mechanisms. Ann. N.Y. Acad. Sci. 807: 1-21.

Crews, D. and M.C. Moore. 1986. Evolution of mechanisms controlling mating behavior. Science 231: 121-125.

Crook, J.H. and P.A. Butterfield. 1968. Effects of testosterone propionate and luteinizing hormone on agonistic and nest building behaviour of *Quelea quelea*. Anim. Behav. 16: 370-384.

Curry, R.L. 1988. Group structure, within-group conflict and reproductive tactics in cooperatively breeding Galapagos Mockingbirds, *Nesomimus parvulus*. Anim. Behav. 36: 1708-1728.

Danchin, E. and R. H. Wagner. 1997. The evolution of coloniality: the emergence of new perspectives. TREE 12: 342-347.

Davies, S.J.J.F. 1977. The timing of breeding by the Zebra Finch *Taeniopygia castanotis* at Mileura, Western Australia. Ibis 119: 369-372.

Dawson, A. 1983. Plasma gonadal steroid levels in wild Starlings (*Sturnus vulgaris*) during the annual cycle and in relation to the stages of breeding. Gen. Comp. Endo. 49: 286-294.

Dawson, A. and A.R. Goldsmith. 1982. Prolactin and gonadotropin secretion in wild Starlings (*Sturnus vulgaris*) during the annual cycle and in relation to nesting, incubation, and rearing young. Gen. Comp. Endo. 48: 213-221.

de Kort, S.R., N.J. Emery, and N. S. Clayton. 2003. Food offering in Jackdaws (*Corvus monedula*). Naturwissenschaften 90: 238-240.

de la Cruz, E. Solis, J. Balencia, O. Chastel, and G. Sorci. 2003. Testosterone and helping behavior in the Azure-winged Magpie (*Cyanopica cyanus*): natural covariation and an experimental test. Behav. Ecol. Sociobiol. 55: 103-111.

de Ridder, E., R. Pinxten, and M. Eens. 2000. Experimental evidence of a testosterone-induced shift from paternal to mating behaviour in a facultatively polygynous songbird. Behav. Ecol. Sociobiol. 40: 32-42.

de Vlaming, V.L. 1979. Actions of prolactin among the vertebrates. In: Hormones and Evolution, Vol. 2. E.J.W. Barrington (ed.). Academic Press, New York. pp. 561-642.

Delay, L.S., J. Faaborg, J. Naranjo, S.M. Paz, T. DeVries, and P.G. Parker. 1996. Paternal care in the cooperatively polyandrous Galapagos Hawk. Condor 98: 300-311.

Dijkstra, H., F.J.H. Tilders, M.A. Hiele, and P.G. Smelik. 1992. Hormonal reactions to fighting in rat colonies: Prolactin rises during defence, not during offense. Physio. Behav. 51: 961-968.

Dillon, W.R. and M. Goldstein. 1984. Multivariate Analysis. John Wiley & Sons, Inc., New York.

Dittami, J.P. 1981. Seasonal changes in the behaviour and plasma titers of various hormones in Bar-headed Geese. Z. Tierpsychol. 55: 289-324.

Dittami, J.P. 1986. Seasonal reproduction, moult and their endocrine correlates in two tropical Ploceidae species. J. Comp. Physio. B 156: 641-647.

Dittami, J.P. and E. Gwinner.1990. Endocrine correlates of seasonal reproduction and territorial behavior in some tropical passerines. In: Endocrinology of Birds: Molecular to Behavioral. M. Wada, S. Ishii, and C.G. Scanes (eds.). Japan Sci. Soc. Press, Tokyo. pp. 225-233.

Downing, J. 1995. Ovulation rate and the concentration of gonadotrophic and metabolic hormones in ewes fed lupin grain. J. Reprod. Fert. 103: 137-145.

Drews, C. 1993. The concept and definition of dominance in animal behaviour. Behav. 125: 283-313.

Du Plessis, M.A. 1993. Helping behaviour in cooperatively-breeding Green Woodhoopoes: selected or unselected trait? Behav. 127: 49-95.

Du Plessis, M.A., W.W. Weathers, W.D. Koenig. 1994. Energetic benefits of communal roosting by Acorn Woodpeckers during the nonbreeding season. Condor 96: 631-637.

Dufty, A.M. 1989. Testosterone and survival: a cost of aggressiveness? Horm. Behav. 23: 185-193.

Dufty, A.M. and J.C. Wingfield. 1986a. Temporal patterns of circulating LH and steroid hormones in a brood parasite, the Brown-headed Cowbird, *Molothrus ater* I. Males. J. Zool., Lond. A 208: 191-203.

Dufty. A.M. and J.C. Wingfield. 1986b. Temporal patterns of circulating LH and steroid hormones in a brood parasite, the Brown-headed cowbird, *Molothrus ater*. II. Females. J. Zool., Lond. A 208: 205-214.

Dugatkin, L.A. 1997a. The evolution of cooperation. Biosci. 47: 355-362.

Dugatkin, L.A. 1997b. Cooperation Among Animals: an evolutionary perspective. Oxford University Press. Oxford.

Dunlop, D.J.E. 1999. Co-operative breeding in White-browed Babblers (*Pomatostomus superciliosus).* Thesis submitted for the degree of B.Sc. (Hons.). University of Stirling.

Dunn, P.O., A. Cockburn, and R.A. Mulder. 1995. Fairy-wren helpers often care for young to which they are unrelated. Proc. R. Soc. Lond., Ser. B 259: 339-343.

Eberhart, J.A., E.B. Keverne, and R.E. Meller. 1983. Social influences on circulating levels of cortisol and prolactin in male Talapoin Monkeys. Physio. Behav. 30: 361-369.

Ebling, J.P., A.R. Goldsmith, and B.K. Follett. 1982. Plasma prolactin and luteinizing hormone during photoperiodically induced testicular growth and regression in Starlings (*Sturnus vulgaris*). Gen. Comp. Endo. 48: 485-490.

Edwards, S.V. and S. Naeem. 1993. The phylogenetic components of cooperative breeding in perching birds. Am. Nat. 141: 754-789.

Ehrlich, P.R., D.S. Dobkin, and D. Wheye. 1988. The Birder's Handbook, A Field Guide to the Natural History of North American Birds. Simon and Schuster Inc., New York.

Eisner, E. 1960. The relationship of hormones to the reproductive behaviour of birds, referring especially to parental behaviour: a review. Anim. Behav. 8: 155-179.

Ejike, C. and C. Schreck. 1980. Stress and social hierarchy rank in Coho Salmon. Trans. Am. Fisheries Soc. 109: 423-429.

Ekman, J. 1990. Alliances in winter flocks of Willowtits; effects of rank on survival and reproductive success in male-female association. Behav. Ecol. Sociobiol. 26: 239-245.

Ekman, J. and B. Sklepkovych. 1994. Conflict of interest between the sexes in Siberian Jay winter flocks. Anim. Behav. 48: 485-487.

Ekman, J. and H. Tegelstrom. 1994. Offspring retention in the Siberian Jay (*Perisoreus infaustus*): the prolonged brood care hypothesis. Behav. Ecol. 5: 245-253.
Ekman, J., A. Bylin, and H. Tegelstrom. 2000. Parental nepotism enhances survival of retained offspring in the Siberian Jay. Behav. Ecol. 11: 416-420.

El Halawani, M.E., J.L. Silsby, E.J. Behneke, and S.C. Fehrer. 1986. Hormonal induction of incubation behavior in ovariectomized female Turkeys (*Meleagris gallopavo*). Biol. Reprod. 35: 59-67.

El Halawani, M.E., O.M. Youngren, L.J. Mauro, and R.E. Phillips.1990. Incubation and the control of prolactin secretion in the Turkey. 1990. In: Endocrinology of Birds: Molecular to Behavioral. M. Wada, S. Ishii, and C.G. Scanes (eds.). Japan Sci. Soc. Press, Tokyo. pp. 287-295.

El Halawani, M.E., J.L. Sibley, O.M. Youngren, and R.E. Phillips. 1991. Exogenous prolactin delays photo-induced sexual maturity and suppresses ovariectomy-induced luteinizing hormone secretion in the Turkey (*Meleagris gallopavo*). Biol. Reprod. 44:420-424.

Elliot, A. 1992. Family Pelecanidae (Pelicans). In: Handbook of the Birds of the World. Vol. 1. J. del Hoya, A. Elliot, and J. Saargatal (eds.). Lynx Edicions, Barcelona. pp. 290-311.

Ely, D.L. and J.P. Henry. 1978. Neuroendocrine response patterns in dominant and subordinant mice. Horm. Behav. 10: 156-169.

Emlen, S.T. 1978. The evolution of cooperative breeding in birds. In: Behavioural Ecology: An Evolutionary Approach. J.R. Krebs and N.B. Davies (eds.). Blackwell Scientific Publications, Oxford. pp.245-281.

Emlen, S.T. 1982. The evolution of helping. I. An ecological constraints model. Am. Nat. 119: 29-39.

Emlen, S.T. 1984. Cooperative breeding in birds and mammals. In: Behavioural Ecology: An Evolutionary Approach, 2nd edition. Krebs, J.R. and N.B. Davies (eds). Blackwell Scientific Publications, Oxford. 305-335.

Emlen, S.T. 1996. Living with relatives: Lessons from avian family systems. Ibis 138: 87-100.

Emlen, S.T. and S.L. Vehrencamp. 1985. Cooperative breeding strategies among birds. In: Perspectives in Ornithology. A.H. Brush and J.G.A. Clark (eds.). Cambridge University Press, Cambridge. pp. 93-120.

Emlen, S.T. and P.H. Wrege. 1989. A test of alternate hypotheses for helping behavior in White-fronted Bee-eaters of Kenya. Behav. Ecol. Sociobiol. 25: 303-319.

Ewer, R.F. 1973. The Carnivores. Cornell University Press, Ithaca, NY.

Fivizzani, A.J., M.A. Colwell, and L.W. Oring. 1986. Plasma steroid hormone levels in free-living Wilson's Phalaropes, *Phalaropus tricolor*. Gen. Comp. Endo. 62: 137-144.

Fivizzani, A.J. and L.W. Oring. 1986. Plasma steroid hormones in the polyandrous Spotted Sandpiper, *Actitis macularia*. Biol. Reprod. 35:1195-1201.

Foley, C.A.H., S. Papageorge, S.K. Wasser. 2001. Noninvasive stress and reproductive measures of social and ecological pressures in free-ranging African Elephants. Conserv. Biol. 15: 1134-1142.

Ford, H.A., H. Bell, R. Nias, and R. Noske. 1988. The relationship between ecology and the incidence of cooperative breeding in Australian birds. Behav. Ecol. Sociobiol. 22: 239-249.

Fox, M. and R. Andrews. 1973. Physiologic and biochemical correlates of individual differences in behavior of Wolf cubs. Behav. 46: 129-136.

Freeman, S. and W.M. Jackson. 1990. Univariate metrics are not adequate to measure avian body size. Auk 107: 69-74.

Gala, R.R. 1990. The physiology and mechanisms of the stress-induced changes in prolactin secretion in the rat. Life Science 46: 1407-1420.

Galeotti, P., A. Pilastro, G. Tavecchia, A. Bonetti, and L. Congru. 1997. Genetic similarities in Long-eared Owl communal winter roosts: a DNA fingerprinting study. Mol. Ecol. 6: 429-435.

Gardner, J.L., R.D. Magrath, and P. Olsen. 2004. Speckled Warblers break cooperative rules: absence of helping in a group-living member of the Pardalotidae. Anim. Behav. 67: 719-728.

Gaston, A.J. 1977. Social behaviour within groups of Jungle Babblers (*Turdoides striatus)*. Anim. Behav. 25: 828-848.

Gaston, A.J. 1978. The evolution of group territorial behavior and cooperative breeding. Am. Nat. 112: 1091-1100.

Goldsmith, A.R. 1982. Plasma concentrations of prolactin during incubation and parental feeding throughout repeated breeding cycles in Canaries (*Serinus canarius*). J. Endo. 94: 51-59.

Goldsmith, A.R. 1983. Prolactin in avian reproductive cycles. In: Hormones and Behaviour in Higher Vertebrates. J. Balthazart, E. Prove, and R. Gilles (eds.). Springer-Verlag, Berlin. pp. 375-387.

Goldsmith, A.R. and D.M. Williams. 1980. Incubation in Mallards (*Anas platyrhynchos*): Changes in plasma levels of prolactin and luteinizing hormone. J. Endo. 86: 371-379.
Goldsmith, A.R., C. Edwards, M. Koprucu, and R. Silver. 1981. Concentrations of prolactin and luteinizing hormone in plasma of Doves in relation to incubation and development of the crop gland. J. Endo. 90: 437-443.

Gonzalez-Mariscal, G. A.I. Melo, and C. Beyer. 1993. Progesterone, but not LHRH or prostaglandin E2, induces sequential inhibition of lordosis agents. Neuroendocrinology 57: 940-945.

Goodall, J. 1970. Tool using in primates and other vertebrates. Adv. Study Behav. 3: 195-249.

Goodwin, J., V. Hartman, M. Grammer, and D. Crews. 1996. Progesterone inhibits female-typical receptive behavior and decreases hypothalamic estrogen and progesterone receptor messenger ribonucleic acid levels in Whiptail Lizards (Genus *Cnemidophorus*). Horm. Behav. 30: 138-144.

Gosler, A.G. 1996. Environmental and social determinants of winter fat storage in the Great Tit *Parus major*. J. Anim. Ecol. 65: 1-17.

Gratto-Trevor, C. L., L. W. Oring, A.J. Fivizzani, M.E. El Halawani, and F. Cooke. 1990. The role of prolactin in parental care in a monogamous and a polyandrous shorebird. Auk 107: 718-729.

Gray, J. M., D. Yarian, and M. Ramenofsky. 1990. Corticosterone, foraging behavior, and metabolism in Dark-eyed Juncos, *Junco hyemalis*. Gen. Comp. Endo. 79: 375-384.

Gust, D.A., T.P. Gordon, and M.K. Hambright. 1993. Response to removal from and return to a social group in adult male Rhesus Monkeys. Physio. Behav. 53: 599-602.

Gwinner, E. and A. Scheuerlein. 1999. Photoperiodic responsiveness of equtorial and temperate-zone Stonechats. Condor 101: 347-359.

Haase, E., E. Paulke, and P.J. Sharp. 1976. Effects of seasonal and social factors on testicular activity and hormone levels in domestic Pigeons. J. Exp. Zool. 197: 81-88.

Hall, E.R. and K.R. Kelson. 1959. The mammals of North America, Vol. 1. Ronad Press, New York.

Halliday, T. 1994. Animal Behavior. University of Oklahoma Press. Tulsa, OK.

Hamilton, W.D. 1964. The genetical evolution of social behaviour, I, II. J. Theor. Biol. 7:1-52.

Harding, C.F. 1981. Social modulation of circulating hormone levels in the male. Amer. Zool. 21: 223-232.

Harrison, C.J.O. 1965. Allopreening as agonistic behaviour. Behav. 24:161-209.

Harrison, C.J.O. 1969. Helpers at the nest in Australian passerine birds. Emu 69: 30-40.

Harvey, S., J.G. Phillips, A. Rees, and T.R. Hall. 1984. Stress and adrenal function. J. Exp. Zool. 232: 633-645.

Harvey, S. C.G. Scanes, and J.G. Phillips. 1986. Avian reproduction. In: Fundamentals of Comparative Vertebrate Endocrinology. I. Chester-Jones, P.M. Ingleton, and J.G. Phillips (eds.). Plenum Press, New York. pp. 125-185.

Hass, C.C. and D. Valenzuela. 2002. Anti-predator benefits of group living in White-nosed Coatis (*Nasua narica*). Behav. Ecol. Sociobiol. 51: 570-578.

Hau, M., M. Wikelski, J.C. Wingfield. 1998. A Neotropical forest bird can measure the slight changes in tropical photoperiod. Proc. R. Soc. Lond. B. 265: 89-95.

Hau, M., M. Wikelski, and J.C. Wingfield. 2000b. Visual and nutritional food cues fine-tune timing of reproduction in a neotropical rainforest bird. J. Exp. Zool. 286: 494-504.

Hau, M., S.T. Stoddard, and K.K. Soma. 2004. Territorial aggression and hormones during the non-breeding season in a tropical bird. Horm. Behav. 45: 40-49.

Hawkes, K. and R.B. Bird. 2002. Showing off, handicap signaling, and the evolution of men’s work. Evo. Anthro. 11: 58-67.

Haydock, J., P.G. Parker, K.N. Rabenold. 1996. Extra-pair paternity uncommon in the cooperatively breeding Bicolored Wren. Behav. Ecol. Sociobiol. 38: 1-16.

Hector, J.A.L. and A.R. Goldsmith. 1985. The role of prolactin during incubation: comparative studies of three *Diomedea* Albatrosses. Gen. Comp. Endo. 60: 236-243.

Hegner, R.E. and J.C. Wingfield. 1987a. Effects of experimental manipulation of testosterone levels on parental investment and breeding success in male House Sparrows. Auk 104: 462-469.

Hegner, R.E. and J.C. Wingfield. 1987b. Social status and circulating levels of hormones in flocks of House Sparrows, *Passer domesticus*. Ethology 76: 1-14.

Heinsohn, R. and M.C. Double. 2004. Cooperate or speciate: new theory for the distribution of passerine birds. TREE 19: 55-57.

Heinsohn, R.G. 1987. Age-dependent vigilance in winter aggregations of cooperatively breeding White-winged Choughs (*Corcorax melanorhamphos*). Behav. Ecol. Sociobiol. 20: 303-306.

Heinsohn, R.G. 1991. Slow learning of foraging skills and extended parental care in cooperatively breeding White-winged Choughs. Am. Nat. 137: 864-881.

Hemborg, C. 1999. Sexual differences in moult-breeding overlap and female reproductive costs in Pied Flycatchers, *Ficedula hypoleuca*. J. Anim. Ecol. 68: 429-436.

Hiatt, E.S., A.R. Goldsmith, and D.S. Farner. 1987. Plasma levels of prolactin and gonadotropins during the reproductive cycle of White-crowned Sparrows (*Zonotrichia leucophrys*) Auk 104: 208-217.

Hill, G.E., R. Montgomerie, C. Roeder, and P. Boag. 1994. Sexual selection and cuckoldry in a monogamous songbird: implications for sexual selection theory. Behav. Ecol. Sociobiol. 35: 193-199.

Hillgarth, N. and J.C. Wingfield. 1997. Parasite-mediated sexual selection: endocrine aspects. In: Host-Parasite Evolution: General Principles and Avian Models. D.H. Clayton and J. Moore (eds.). Oxford Univ. Press, New York. pp. 78-104.

Hirschhorn, K., F. Bach, R. Kolodny, I. Firschein, and N. Gashen. 1963. Immune response and mitosis of human peripheral blood lymphocytes in vitro. Science 142: 1185-1187.

Holberton, R.L., K.P. Able, and J.C. Wingfield. 1989. Status signalling in Dark-eyed Juncos, *Junco hyemalis*: plumage manipulations and hormonal correlates of dominance. Anim. Behav. 37: 681-689.

Hoogland, J.L. and P.W. Sherman. 1976. Advantages and disadvantages of Bank Swallow *Riparia riparia* coloniality. Eco. Mono. 46: 33-58.

Houston, A.I. and J.M. McNamara. 1993. A theoretical investigation of the fat reserves and mortality levels of small birds in winter. Ornis Scand.24: 205-219.

Hughes, J.M., J.D.E. Hesp, R. Kallioinen, M. Kempster, C.L. Lange, K.E. Hedstrom, P.B. Mather, A. Robinson, and M.J. Wellbourn. 1996. Differences in social behaviour between populations of the Australian Magpie *Gymnorhina tibicen*. Emu 96: 65-70.

Hutchinson, R.E. 1975. Effects of ovarian steroids and prolactin on the sequential development of nesting behaviour in female Budgerigars. J. Endo. 67: 29-39.

Immelmann, K. 1971. Ecological aspects of periodic reproduction. In: Avian Biology, Vol. 1. D.S. Farner and J.R. King (eds.). Academic Press, New York. pp. 341-389.

Immelmann, K. 1973. Role of the environment in reproduction as source of "predictive" information. In: Breeding Biology of Birds. D.S. Farner (ed.). National Academy of Sciences, Washington, DC. pp. 121-157.

Innes, K.E. and R.E. Johnston. 1996. Cooperative breeding in the White-throated Magpie-jay. How do auxilliaries influence nesting success? Anim. Behav. 51: 519-533.

Jacobs, J.D. and J.C. Wingfield. 2000. Endocrine control of life-cycle stages: a constraint or response to the environment? Condor 102: 35-51.
James, G.D., D.E. Crews, and J. Pearson. 1989. Catecholamines and stress. In: Human Population Biology. M.A. Little and J.D. Haas (eds.). Oxford University Press, New York. pp. 280-295.

Jamieson, I.G. 1989. Behavioral heterochrony and the evolution of birds' helping at the nest: an unselected consequence of communal breeding? Am. Nat. 133: 394-406.

Jamieson, I.G. 1991. The unselected hypothesis for the evolution of helping behavior: too much or too little emphasis on natural selection? Am. Nat. 138: 271-282.

Jamieson, I.G. and L.J. Marshall. 1999. Helping behaviour in the Arabian Babbler: was Wright's dismissal of the unselected hypothesis warrented? Behav. Ecol. Sociobiol. 46: 435-436.

Jansson, C., J. Eckman, and A. von Bromssen. 1981. Winter mortality and food supply in tits *Parus* spp. Oikos 37: 313-322.

Jefferys, A.J., V. Wilson, and S.L. Thein. 1985a. Hypervariable 'minisatellite' regions in human DNA. Nature 314: 67-73.

Jefferys, A.J., V. Wilson, and S.L. Thein. 1985b. Individual-specific 'fingerprints' of human DNA. Nature 316: 76-79.

Johnson, A.L. and A. van Tienhoven. 1980. Plasma concentrations of sex steroids and LH during the ovulatory cycle of the hen, *Gallus domesticus*. Biol. Reprod. 23: 386-393.

Johnson, K. 1988. Sexual selection in Pinyon Jays I: female choice and male-male competition. Anim. Behav. 36: 1038-1047.

Johnson, K. and N.T. Burley. 1998. Mating tactics and mating systems of birds. Ornith. Mono. 49: 21-60.

Johnson, M.S. and J.L. Brown. 1980. Genetic variation among trait groups and apparent absence of close inbreeding in Grey-crowned Babblers. Behav. Ecol. Sociobiol. 7: 93-98.

Johnson, T.S. 1998. Behavioural correlates of testosterone and seasonal changes of steroids in Red-winged Blackbirds. Anim. Behav. 55:957-965.

Jones, D.N., R.W.R.J. Dekker, C.S. Roselaar. 1995. The Megapodes: Megapodiidae. Oxford University Press, Oxford, UK. 262pp.

Kemp, A.C. and M.I. Kemp. 1980. The biology of the southern Ground Hornbill *Bucorvus leadbeateri* (Vigors) (Aves: Bucerotidai). Ann. Transc. Mus. 32: 65-100.

Kendeigh, S.C. 1961. Energy of birds conserved by roosting in cavities. Wilson Bull. 73: 140-147.
Kenward, R.E. 1978. Hawks and doves: attack success and selection in Goshawk flights at Wood-pigeons. J. Anim. Ecol. 47: 449-460.

Ketterson, E., V. Nolan, L.Wolf, C. Ziegenfus, A. Dufty, and T. Johnsen. 1991. Testosterone and avian life histories: the effects of experimentally elevated testosterone on corticosterone and body mass in Dark-eyed Juncos. Horm. Behav. 25: 489-503.

Ketterson, E.D., V. Nolan, Jr., L.Wolf, and C. Ziegenfus. 1992. Testosterone and avian life histories: Effects of experimentally elevated testosterone on behavior and correlates of fitness in the dark-eyed junco (*Junco hyemalis*). Am. Nat. 140: 980-999.

Ketterson. E.D., V. Nolan Jr., M.J. Cawthorn, P.G. Parker, and C. Ziegenfus. 1996. Phenotypic engineering: using hormones to explore the mechanistic and functional bases of phenotypic variation in nature. Ibis 138: 70-86.

Khan, M.Z., F.M.A. McNabb, and J.R. Walters. 2001. Patterns of testosterone and prolactin concentrations and reproductive behavior of helpers and breeders in the cooperatively breeding Red-cockaded Woodpecker. Horm. Behav. 40: 1-13.

Kikkawa, J., J.N.M. Smith, R. Prys-Jones, and C. Catterall. 1986. Determinants of social dominance and inheritance of agonistic behavior in an island population of Silvereyes, *Zosterops lateralis*. Behav. Ecol. Sociobiol. 19: 165-169.

King, B.R. 1980. Social organization and behaviour of the Grey-crowned Babbler *Pomatostomus temporalis*. Emu 80: 59-76.

Kirkwood and Robertson. 1999. The occurrence and purpose of huddling by Emperor Penguins during foraging trips. Emu 99: 40-45.

Klaassen, M. and A. Lindstrom. 1996. Departure fuel loads in time-minimizing migrating birds can be explained by the energy costs of being heavy. J. Theor. Biol. 183: 29-34.

Koenig, W.D. 1981. Reproductive success, group size, and the evolution of cooperative breeding in the Acorn Woodpecker. Am. Nat. 117: 421-443.

Koenig, W.D. and F.A. Pitelka. 1981. Ecological factors and kin selection in the evolution of cooperative breeding in birds. In: Natural selection and social behavior- recent research and new theory. R.D. Alexander and D.W. Tinkle (eds.). Chiron Press, New York. pp. 261-280.

Koenig, W.D. and R.L. Mumme. 1987. Population ecology of the cooperatively breeding Acorn Woodpecker. Princeton University Press. Princeton, N.J.

Koford, R.R., B.S. Brown, and S.L. Vehrencamp. 1986. Habitat stauration in Groove-billed Anis (*Crotophaga sulcirostris*). Am. Nat. 127: 317-337.

Komdeur, J. 1992. Importance of habitat saturation and territory quality for evolution of cooperative breeding in the Seychelles Warbler. Nature 358: 493-495.

Krebs, J.R., M.H. MacRoberts, and J.M. Cullen. 1972. Flocking and feeding in the Great Tit *Parus major*- an experimental study. Ibis 114: 507-530.

Krebs, J.R. and N.B. Davies. 1984. Behavioural Ecology: an evolutionary approach. Sinauer Associates Inc., Sunderland, MA.

Kroodsma, D.E. 1976. Reproductive development in a female songbird: differential stimulation by quality of male song. Science 192: 574-75.

Lancaster, J. 1971. Play-mothering: the relations between juvenile females and young infants among free-ranging Vervet Monkeys (*Cercopithecus adthiops*). Folia Primat.15: 161-182.

Langden, T.A. 2000. Prolonged offspring dependence and cooperative breeding. Behav. Ecol. 11: 367-377.

Lawton, M.F. and R.O. Lawton. 1986. Heterochrony, deferred breeding, and avian sociality. In: Current Ornithology, Vol. 3. R.F. Johnston (ed.). Plenum Press, New York. pp. 187-222.

Lea, R.W., A.S.M. Dods, P.J. Sharp, and A. Chadwick. 1981. The possible role of prolactin in the regulation of nesting behaviour and the secretion of luteinizing hormone in broody Bantams. J. Endo. 91: 89-97.

Leboucher, G., N. Beguin, A. Lacroix, M. Kreutzer. 2000. Progesterone inhibits female courtship behavior in domestic Canaries (*Serinus canaria*). Horm. Behav. 38: 123-129.

Lehrman, D.S. 1961. Hormonal regulation of parental behavior in birds and infrahuman mammals. In: Sex and Internal Secretions, Vol II. W.C. Young (ed.). The Williams and Wilkins Co., Baltimore, MD. pp. 1268-1382.

Levin, R. 1996. Song behaviour and reproductive strategies in a duetting wren, *Thryothorus nigricapillus*: I. Removal experiments. Anim. Behav. 52: 1093-1106.

Levin, R.N. and J.C. Wingfield. 1992. The hormonal control of territorial aggression in tropical birds. Ornis Scand. 23: 284-291.

Levine, S. 1993. The influence of social factors on the response to stress. Psychother. Psychosom. 60: 33-38.

Ligon, J.L. and S.H. Ligon. 1978. Communal breeding in the Green Woodhoopoe as a case for reciprocity. Nature 176: 496-498.

Ligon, J.D. and S.D. Ligon. 1983. Reciprocity in the Green Woodhoopoe *Phoeniculus purpureus*. Anim. Behav. 31: 480-489.
Ligon, J.D. and S.H. Ligon. 1990. Green Woodhoopoes: Life history traits and sociality. In: Cooperative Breeding in Birds. P.B. Stacey and W.D. Koenig (eds.). Cambridge Univ. Press, Cambridge. pp. 33-65.

Lima, S.L. 1986. Predation risk and unpredictable feeding condition: determinants of body mass in birds. Ecology 67: 366-376.

Lima, S.L. 1990. The influence of models on the interpretation of vigilance. In: Interpretation and Explanation in the Study of Animal Behaviour: Vol. 2. Explanation, Evolution and Adaptation. M. Bekoff and D. Jamieson (eds.). Westview Press, Boulder, Colorado. pp. 246-267.

Lima, S.L. 1995. Back to the basics of anti-predatory vigilance: the group-size effect. Anim. Behav. 49: 11-20.

Logan, C.A. and C.A. Carlin. 1991. Testosterone stimulates reproductive behavior during autumn in Mockingbirds (*Mimus polyglottos*). Horm. Behav. 25:229-241.

Logan, C. A. and J.C. Wingfield. 1995. Hormonal correlates of breeding status, nest construction, and parental care in multiple-brooded Northern Mockingbirds, *Mimus polyglottos*. Horm. Behav. 29: 12-30.

Lotem, A., R.H. Wagner, and S. Balshine-Earn. 1999. The overlooked signaling component of nonsignaling behavior. Behav. Ecol. 10: 209-212.

Louch, C.D. and M. Higginbotham. 1967. The relation between social rank and plasma corticosterone levels in mice. Gen. Comp. Endo. 8: 441-444.

Lynch, M. 1990. The similarity index and DNA fingerprinting. Mol. Biol. Evol. 7: 478-484.

MacDonald, D.W. and P.D. Moehlman. 1982. Cooperation, altruism, and restraint in the reproduction of carnivores. In: Perspectives in Ethology, Vol. 5. P. Klopfer and P. Bateson (eds.). Plenum Press, New York. pp. 433-466.

Macedo, R.H. and C.A. Bianchi. 1997. Communal breeding in tropical Guira Cuckoos *Guira guira*: sociality in the absence of saturated habitat. J. Avian Bio. 28: 207-215.

Magrath, R.D. and S.M. Yezerinac. 1997. Facultative helping does not influence reproductive success or survival in cooperatively breeding White-browed Scrubwrens. J. Anim. Ecol. 66: 658-670.

Mantel, N. 1967. The detection of disease clustering and a generalized regression approach. Cancer Res. 27: 209-220.

Martin, T.E., J. Scott, and C. Menge. 2000. Nest predation increases with parental activity: separating nest site and parental activity effects. Proc. R. Soc. Lond. B: 267:2287-2293.

Mays, N.A., C.M. Vleck, and J. Dawson. 1991. Plasma leuteinizing hormone, steroid hormones, behavioral role, and nest stage in cooperatively breeding Harris' Hawks (*Parabuteo unicinctus*). Auk 108: 619-637.

McCarthy, M.M., G.H. Curran, and H.I. Siegel. 1994. Evidence for the involvement of prolactin in the maternal behavior of the hamster. Physio. Behav. 55: 181-184.

McDonald, P.G., W.A. Buttemer, and L.B. Astheimer. 2001a. The influence of testosterone on territorial defence and parental behavior in male free-living Rufous Whistlers, *Pachycephala rufiventris*. Horm. Behav. 39: 185-194.

McDonald, P.G., W.A. Buttemer, and L.B. Astheimer. 2001b. Gonadal and hormonal phenology in free-living male Rufous Whistlers, *Pachycephala rufiventris* (Passeriformes: Pachycephalidae). Australian J. Zool. 49: 651-662.

McGuire, M.T., G.L. Brammer, M.J. Raleigh. 1986. Resting cortisol levels and the emergence of dominant status among Vervet Monkeys. Horm. Behav. 20: 106-117.

McNamara, J.M., A.I. Houston, and S.L. Lima. 1994. Foraging routines of small birds in winter: a theoretical investigation. J. Avian Bio. 25: 287-302.

Mendoza, S.P., C.L. Coe, E.L. Lowe, and S. Levine. 1979. The physiological response to group formation in adult male Squirrel Monkeys. Psychoneuroendo. 3: 221-229.

Meyerhoff, J.L., M.A. Oleshansky, E.H. Mougey. 1988. Psychologic stress increases plasma levels of prolactin, cortisol, and POMC-derived peptides in man. Psychosomatic Medicine 50:295-303.

Mitani, J.C. and D. P. Watts. 2001. Why do chimpanzees hunt and share meat? Anim. Behav. 61: 915-924.

Monadjem, A., N. Owen-Smith, and A.C. Kemp. 1995. Aspects of breeding biology of the Arrowmarked Babbler *Turdoides jardineii* in South Africa. Ibis 137: 515-518.

Monaghan, E.P. and S.E. Glickman. 1992. Hormones and aggressive behavior. In: Behavioral Endocrinology. J.B. Becker, S.M. Breedlove, and D. Crews (eds.). MIT Press, Cambridge, MA. pp.261-285.

Moore, M.C. 1982. Hormonal response of free-living male White-crowned Sparrows to experimental manipulation of female sexual behavior. Horm. Behav. 16: 323-329.

Moore, M.C. and J.C. Wingfield. 1980. Hormone-behavior interactions during reproduction in White-crowned Sparrows (*Zonotrichia leucophrys*). Am. Zool. 20: 788.

Moore, M.C. and R. Kranz. 1983. Evidence for androgen independence of male mounting behavior in White-crowned Sparrows (*Zonotrichia leucophrys gambelii*). Horm. Behav. 17: 414-423.

Morse, D.H. 1970. Territorial and courtship songs of birds. Nature, London 226: 659-661.

Morton, M.L. and N. Allan. 1990. Effects of snowpack and age on reproductive schedules and testosterone levels in the male White-crowened Sparrow in a montane environment. In: Endocrinology of Birds: Molecular to Behavioral. M. Wada, S. Ishii, and C.G. Scanes (eds.). Japan Sci. Soc. Press, Tokyo. pp. 235-249.

Moss, C. 1988. Elephant memories. William Morrow, New York.

Mota, M.T. and M.B.C. Sousa. 2000. Prolactin levels of fathers and helpers related to alloparental care in Common Marmosets, *Callithrix jacchus*. Folia Primatol. 71:22-26.

Mumme, R.L., W.D. Koenig, and F.A. Pitelka. 1983. Mate guarding in the Acorn Woodpecker: within-group reproductive competition in a cooperative breeder. Anim. Behav. 31: 1094-1106.

Mumme, R.L., W.D. Koenig, and F.L.W. Ratnieks. 1989. Helping behaviour, reproductive value, and the future component of direct fitness. Anim. Behav. 38: 331-343.

Myers, S.A., Millam, J.R., and M.E. El Halawani. 1989. Plasma LH and prolactin levels during the reproductive cycles of the Cockatiel (*Nymphicus hollandicus*). Gen. Comp. Endo. 73: 85-91.

Nakamura, M. 1998a. Multiple mating and cooperative breeding in polygynandrous Alpine Accentors. I. Competition among females. Anim. Behav. 55:259-275.

Nakamura, M. 1998b. Multiple mating and cooperative breeding in polygynandrous Alpine Accentors. II. Male mating tactics. Anim. Behav. 55:259-275.

Nakamura, M. S. Yamagishi, and I. Nishiumi. 2001. Cooperative breeding of the White-headed Vanga *Leptopterus viridis*, an endemic species in Madagascar. J. Yamashina Inst. Ornithol. 33: 1-14.

Nalbandov, A.V., M. Hochhauser, and M. Dugas. 1945. A study of the effect of prolactin on broodiness and on cock testes. Endo. 36: 251-258.

Nelson, R. and G. Demas. 1996. Seasonal changes in immune function. Q. Rev. Biol. 71: 511-548.

Nelson, R.J. 2000. An Introduction to Behavioural Endocrinology. Sinauer Associates, Inc., Sunderland, MA.

Nicholls, J.A., M.C. Double, D.M. Rowell, and R.D. Magrath. 2000. The evolution of cooperative and pair breeding in thornbills *Acanthiza* (Pardalotidae). J. Avian Bio. 31: 165-176.

Nilsson, J.-A. and E. Svensson. 1996. The cost of reproduction: a new link between current reproductive effort and furture reproductive success. Proc. R.. Soc. Lond. B 263: 711-714.

Noble, G.K. and M. Wurm. 1940. The effect of testosterone propionate on the Black-crowned Night Heron. Endo. 26: 837-850.

Norberg, U.M. 1996. Energetics of flight. In: Avian Energetics and Nutritional Ecology. C. Carey (ed.). Chapman and Hall, New York. pp. 199-249.

Orcutt, F.S. 1967. Oestrogen stimulation of nest material preparation in the Peach-faced Lovebird (*Agapornis roseicollis*). Anim. Behav. 15: 471-478.

Oring, L.W., A.J. Fivizzani, and M.E. El Halawani. 1986. Changes in plasma prolactin associated with laying and hatch in the Spotted Sandpiper. Auk 103: 820-822.

Oring, L.W., A.J. Fivizzani, M.A. Colwekk, and M.E. El Halawani. 1988. Hormonal changes associated with natural and manipulated incubation in the sex-role reversed Wilson’s Phalarope. Gen. Comp. Endo. 72: 247-256.

Oring, L.W., A.J. Fivizzani, and M.E. El Halawani. 1989. Testosterone-induced inhibition of incubation in the Spotted Sandpiper (*Actitis mecularia*). Horm. Behav. 23:412-423.

Peters, A. 2000. Testosterone treatment is immunosuppressive in Superb Fairy-wrens, yet free-living males with high testosterone are more immunocompetent. Proc. R. Soc. Lond. Ser. B 267:883-889.

Peters, A., L.B.Astheimer, and A. Cockburn. 2001. The annual testosterone profile in cooperatively breeding Superb Fairy-wrens, *Malurus cyaneus*, reflects their extreme infidelity. Behav. Ecol. Sociobiol. 50: 519-527.

Peters, A., A. Cockburn, and R. Cunningham. 2002. Testosterone treatment suppresses parental care in Superb Fairy-wrens, *Malurus cyaneus*, despite their concurrent investment in courtship. Behav. Ecol. Sociobiol. 51: 538-547.

Piper, W.H. 1994. Courtship, copulation, nesting behaviour and brood parasitism in the Venezuelan Stripe-backed Wren. Condor 96: 654-671.

Pitcher, T.J., A.E. Magurran, and I.L. Winfield. 1982. Fish in larger shoals find food faster. Behav. Ecol. Sociobiol. 10: 149-151.

Poiani, A. 1993. Social structure and the development of helping behaviour in the Bell Miner (*Manorina melanophrys*, Meliphagidae). Ethology 93: 62-80.

Poiani, A. and T. Fletcher. 1994. Plasma levels of androgens and gonadal development of breeders and helpers in the Bell Miner (*Manorina melanophrys*). Behav. Ecol. Sociobiol. 34: 31-41.

Poiani, A. and L.S. Jermiin. 1994. A comparative analysis of some life-history traits between cooperatively and non-cooperatively breeding Australian passerines. Evo. Ecol. 8: 471-488.

Poiani, A. and M. Pagel. 1997. Evolution of avian cooperative breeding : comparative tests of the nest predation hypothesis. Evo. 51: 226-240.

Poldman, T., R. Montgomerie, and P. Boag. 1995. Mating systems of the cooperatively breeding Noisy Miner *Manorina melanocephala*, as revealed by DNA profiling. Behav. Ecol. Sociobiol. 37: 137-143.

Pollard, T.M. 1995. Use of cortisol as a stress marker: Practical and theoretical problems. Am. J. Human Bio. 7: 265-274.

Powell, G.V.N. 1974. Experimental analysis of the social value of flocking by Starlings (*Sturnus vulgaris*) in relation to predation and foraging. Anim. Behav. 22: 501-505.

Pravosudov, V.V. and T.C. Grubb, Jr. 1999. Effects of dominance and vigilance in avian social groups. Auk 116: 241-246.

Pruett-Jones, S.G. and M.J. Lewis. 1990. Sex ratio and habitat limitation promote delayed dispersal in Superb Fairy-wrens. Nature 348: 541-542.

Pulliam, H.R. 1973. On the advantages of flocking. J. Theor. Biol. 38: 419-422.

Pulliam, H.R. and T. Caraco. 1984. Living in groups: is there an optimal group size? In: Behavioural Ecology: An Evolutionary Approach, 2nd ed. J.R. Krebs and N.B. Davies (eds.). Sinauer, Sunderland, MA. pp. 122-147.

Putland, D. 2001. Has sexual selection been overlooked in the study of avian helping behaviour? Anim. Behav. 62: 811-814.

Queller, D.C. 1994. Extended parental care and the origin of eusociality. Proc. R. Soc. Lond. B 256: 105-111.

Queller, D.C. 2001. W.D. Hamilton and the evolution of sociality. Behav. Ecol. 12: 261-268.

Quinn, J.S., R.Macedo, and B.N. White. 1994. Genetic relatedness of communally breeding Guira Cuckoos. Anim. Behav. 47: 515-529.

Ralph, C.J., G.R. Geupel, P. Pyle, T.E. Martin, D.F. DeSante. 1993. Handbook of field methods for monitoring landbirds. Gen. Tech. Rep. PSW-GTR-144. Albany, CA. Pacific Southwest Research Station, Forest Service, U.S. Department of Agriculture. 41pp.

Ramenofsky, M. 1984. Agonistic behaviour and endogenous plasma hormones in male Japanese Quail. Anim. Behav. 32: 698-708.
Rasa, O.A.E. 1977. The ethology and sociology of the Dwarf Mongoose (*Helogale undulata rufula).* Z. Tierpsychol 43: 337-406.

Raymond, M. and F. Rousset. 1995. GENEPOP (Version 1.2): Population genetics software for exact tests and ecumenicism. J. Heridity 86: 248-249.

Reyer, H.-U. 1990. Pied Kingfishers: ecological causes and reproductive consequences of cooperative breeding. In: Cooperative Breeding in Birds. P.B. Stacey and W.D. Koenig (eds.). Cambridge Univ. Press, Cambridge. pp. 527-557.

Richard-Yris, M-A, P.J. Sharp, A-M. Wauters, D. Guemene, J-P. Richard, and M. Foraste. 1998. Influence of stimuli from chicks on behavior and concentrations of plasma prolactin and lueteinizing hormone in incubating hens. Horm. Behav. 33: 139-148.

Richner, H. and P. Heeb. 1996. Communal life: honest signaling and the recruitment center hypothesis. Behav. Ecol. 7: 115-119.

Riddle, O. 1963. Prolactin or progesterone as key to parental behaviour: a review. Anim. Behav. 11: 419-432.

Roberts, G. 1998. Competitive altruism: from reciprocity to the handicap principle. Proc. R. Soc. Lond. B. 265: 427-431.

Rohwer, S. 1986. Selection for adoption versus infanticide by replacement “mates” in birds. In: Current Ornithology, Vol 3. R.F. Jonhston (ed.). Plenum, New York. pp. 353-395.

Rohwer, S. And F.C. Rohwer. 1978. Status signalling in Harris’ Sparrows: experimental deceptions achieved. Anim. Behav. 26: 1012- 1022.

Rohwer, S. and J.C. Wingfield. 1981. A field study of social dominance, plasma levels of luteinizing hormone and steroid hormones in wintering Harris’ Sparrows. Z. Tierpsychol. 57: 173-183.

Rose, R.M., J.W. Holaday, and I.S. Bernstein. 1971. Plasma testosterone, dominance rank and aggressive behaviour in male Rhesus Monkeys. Nature 231: 366-368.

Rosenblatt, J.S. 1992. Hormone-behavior relations in the regulation of parental behavior. In: Behavioural Endocrinology. J.B. Becker, S.M. Breedlove, and D. Crews (eds.). MIT Press, Cambridge, MA. pp. 219-259.

Rowley, I.A. 1976. Co-operative breeding in Australian birds. In: Proc. 16th Intl. Ornith. Cong. H.J Frith and J.H. Calaby (eds.). Griffin Press, Ltd., S. Australia. pp. 657-666.

Rowley, I. 1977. Communal activities among White-winged Choughs *Corcorax melanorhamphus*. Ibis 120: 1-20.

Rowley, I. 1978. Communal activities among White-winged Choughs, *Corcorx melanorhamphus*. Ibis 120: 178-197.

Rowley, I. and E.M. Russell. 1990. The Splendid Fairy-wren *Malurus splendens*. In: Cooperative Breeding in Birds: Long-term Studies of Ecology and Behaviour. P.B. Stacey and W.D. Koenig (eds.). Cambridge University Press, Cambridge. pp. 1-30

Russell, E.M. 1989. Co-operative breeding- a Gondwanan perspective. Emu 89: 61-62.

Russell, E. and I. Rowley. 1988. Helper contribution to reproductive success in the Splendid Fariy-wren (*Malurus splendens*). Behav. Ecol. Sociobiol. 22: 131-140.

Saino, N. and A.P. Moller. 1995. Testosterone correlates of mate guarding, singing and aggressive behaviour in male Barn Swallows, *Hirundo rustica*. Anim. Behav. 49: 465-472.

Sakai, R., S. Weiss, C. Blanchard, R. Blanchard., R. Spencer, B. McEwen. 1991. Effects of social stress and housing conditions on neuroendocrine measures. Soc. Neurosci. Abst.17: 621.1.

Saltzman, W., N.J. Schultz-Darken, G. Scheffler, F.H. Wegner, and D.H. Abbott. 1994. Social and reproductive influences on plasma cortisol in female Marmoset Monkeys. Physio. Behav. 56: 801-810.

Sapolsky, R.M. 1982. The endocrine stress-response and social status in the wild Baboon. Horm. Behav. 16: 279-292.

Sapolsky, R.M. 1989. Hypercortisolism among socially subordinate wild Baboons originates at the CNS level. Arch. Gen. Psychiatry 46: 1047-1051.

Sapolsky, R.M. 1990. Adrenocortical function, social rank, and personality among wild Baboons. Biol. Psych. 28: 862-878.

Sapolsky, R.M. 1991. Testicular function, social rank and personality among wild Baboons. Psychoneuroendo. 16: 281-293.

Sapolsky, R.M. 1992. Cortisol concentration and the social significance of rank instability among wild Baboons. Psychoneuroendo. 17: 701-709.

Sapolsky, R.M. 1993. Endocrinology alfresco: psychoendocrine studies of wild Babbons. Recent Progr. Horm. Res. 48: 437-468.

Sapolsky, R.M. 2002. Endocrinology of the stress-response. In: Behavioural Endocrinology, 2nd edition. J.B. Becker, S.M. Breedlove, D. Crews, and M. McCarthy (eds.). MIT Press, Cambridge, MA. pp. 409-450.

Sapolsky, R.M., L.M. Romero, A.U. Munck. 2000. How do glucocorticoids influence stress responses? Integrating permissive, suppressive, stimulatory, and preparative actions. Endocr. Rev. 21: 55-89.

Sargent, G., S. Legge, and M. Hall. 1998. Multi-locus DNA fingerprinting protocol, update. Botany and Zoology Department, Australian National University, Canberra.

Schlinger, B. and G. Callard. 1990. Aromatization mediates aggressive behavior in quail. Gen. Comp. Endo. 79: 39-53.

Schlinger, B.A. 1998. Sexual differentiation of avian brain and behavior: current views on gonadal hormone-dependent and independent mechanisms. Annual Rev. Physio. 60: 407-429.

Schmidt, L. G., S.D. Bradshaw, and B.K. Follett. 1991. Plasma levels of luteinizing hormone and androgens in relation to age and breeding status among cooperatively breedng Australian Magpies (*Gymnorhina tibicen* Latham). Gen. Comp. Endo. 83: 48-55.

Schoech, S.J. 1998. Physiology of helping in Florida Scrub-jays. Am. Sci. 86: 70-77.

Schoech, S.J., Mumme, R.L., and M.C. Moore. 1991. Reproductive endocrinology and mechanisms of breeding inhibition in cooperatively breeding Florida Scrub-jays (*Aphelocoma c. coerulescens*). Condor 93: 354-364.

Schoech, S.J., R.L. Mumme, and J.C. Wingfield. 1996a. Delayed breeding in the cooperatively breeding Florida Scrub-jay (*Aphelocoma coerulescens*): inhibition or the absence of stimulation. Behav. Ecol. Sociobiol. 39: 77-90.

Schoech, S.J., R.L. Mumme, and J.C. Wingfield. 1996b. Prolactin and helping behaviour in the cooperatively breeding Florida Scrub-jay, *Apehlocoma c. coerulescens*. Anim. Behav. 52: 445-456.

Schoech, S.J., R.L. Mumme, and J.C. Wingfield. 1997. Corticosterone, reproductive status, and body mass in a cooperative breeder, the Florida Scrub-jay (*Apehlocoma c. coerulescens*). Physio. Zool. 70: 68-73.

Schwabl, H. 1992. Winter and breeding territorial behaviour and levels of reproductive hormones of migratory European Robins. Ornis Scand. 23: 271-276.

Schwabl, H. J.C. Wingfield, D.S. Farner. 1985. Influence of winter on endocrine state and behavior in European Blackbirds (*Turdus merula*). Z. Tierpsychol. 68: 244-252.

Schwabl, H., M. Ramenofsky, I. Schwabl-Benzinger, D.S. Farner, and J.C. Wingfiled. 1988. Social status, circulating levels of hormones, and competition for food in winter flocks of the White-throated Sparrow. Behav. 107: 107-121.

Schwabl, H. and E. Kriner. 1991. Territorial aggression and song of male European Robins (*Erithacus rubecula*) in autumn and spring: effects of antiandrogen tratment. Horm. Behav. 25: 180-194.

Scott, D.K. 1980. Functional aspects of prolonged parental care in Bewick's Swans. Anim. Behav. 28: 938-952.

Searcy, W.A. and J.C. Wingfield. 1980. The effects of androgen and antiandrogen on dominance and aggressiveness in male Red-winged Blackbirds. Horm. Behav. 14: 126-135.

Searcy, W.A. and M.S. Capp. 1997. Estradiol dosage and the solicitation display assay in Red-winged Blackbirds. Condor 99: 826-828.

Seiler, H.W., M.Gahr, A.R. Goldsmith, and H-R. Guttinger. 1992. Prolactin and gonadal steroids during the reproductive cycle of the Bengalese Finch (*Lonchira striata* var. *domestica*, Estrilididae), a nonseasonal breeder with biparental care. Gen. Comp. Endo. 88: 83-90.

Senar, J.C., V. Polo, F. Uribe, M. Camerino. 2000. Status signalling, metabolic rate and body mass in the Siskin: the cost of being subordinate. Anim. Behav. 59: 103-110.

Serventy, D.L. 1971. Biology of desert birds. In: Avian Biology. D.S. Farner, J.R. King, and K.C. Parkes (eds.). Academic Press, New York. pp. 287-339.

Sharp, P.J. 1996. Strategies in avian breeding cycles. Anim. Reprod. Sci. 42: 505-513.

Sherman, P.W. 1977. Nepotism and the evolution of alarm calling. Science 197: 1246-1253.

Sherman, P.W., E.A. Lacey, H.K. Reeve, L. Keller. 1995. The eusociality continuum. Behav. Ecol. 6: 102-108.

Siegel, H.S. 1980. Physiological stress in birds. Biosci. 8: 529-534.

Siegel, H.S. and P.B. Siegel. 1961. The relationship of social competition with endocrine weights and activity in male chickens. Anim. Behav. 9: 151-158.

Sigmund, K. 1993. Games of Life. Explorations in Ecology, Evolution, and Behaviour. Oxford University Press, Oxford. 256 pp.

Silver, R. 1984. Prolactin and parenting in the pigeon family. J. Exp. Zool. 232: 627-625.

Silver, R. 1990. Avian behavioral endocrinology: status and prospects. In: Endocrinology of Birds: Molecular to Behavioral. M. Wada, S. Ishii, and C.G. Scanes (eds.). Japan Sci. Soc. Press, Tokyo. pp. 261-272.

Silver, R. and M. Cooper. 1983. Avian behavioral endocrinology. Biosci. 33: 567-572.

Silverin, B. 1980. Effects of long-acting testosterone treatment on free-living Pied Flycatchers, *Ficedula hypoleuca*, during the breeding period. Anim. Behav. 28: 906-912.

Silverin, B. 1991. Annual changes in plasma levels of LH, and prolactin in free-living female Great Tits (*Parus major*). Gen. Comp. Endo. 83: 425-431.

Silverin, B. 1998. Behavioural and hormonal responses of the Pied Flycatcher to environmental stressors. Anim. Behav. 55: 1411-1420.

Silverin, B. and J.C. Wingfield. 1982. Patterns of breeding behaviour and plasma levels of hormones in a free-living population of Pied Flycatchers, *Ficedula hypoleuca*. J. Zool. London 198: 117-129.

Silverin, B. and A. Goldsmith. 1984. The effects of modifying incubation on prolactin secretion in free-living Pied Flycatchers. Gen. Comp. Endo. 55: 239-244.

Silverin, B., P.A.Viebke, and J. Westin. 1984. Plasma levels of luteinizing hormone and steroid hormones in free-living winter groups of Willow Tits (*Parus montanus*). Horm. Behav. 18: 367-379.

Silverin, B. and A. Goldsmith. 1997. Natural and photoperiodically induced changes in plasma prolactin levels in male Great Tits. Gen. Comp. Endo. 105: 145-154.

Simpson, K. and N. Day. 1996. Field Guide to the Birds of Australia. Penguin Books Australia Ltd. Ringwood, Victoria, Australia. pp 400.

Sklepkovych, B. 1997. The influence of kinship on foraging competition in Siberian Jays. Behav. Ecol. Sociobiol. 40: 287-296.

Skutch, A.F. 1961a. The nest as dormitory. Ibis 103: 50-70.

Skutch, A.F. 1961b. Helpers among birds. Condor 63: 198-226.

Sloane, S.A. 1996. Incidence and origins of supernumeraties at Bushtit (*Psaltriparus minimus*) nests. Auk 113: 757-770.

Sloman, K.A., N.B. Metcalfe, A.C. Taylor, K.M. Gilmour. 2001. Plasma cortisol concentrations before and after social stress in Rainbow Trout and Brown Trout. Physio. Biochem. Zool. 74: 383-389.

Smith, R.J.F. and W.S. Hoar. 1967. The effects of prolactin and testosterone on the parental behavior of the male Stickleback *Gasterosteus aculeatus*. Anim. Behav. 15: 342-352.

Sockman, K.W. and H. Schwabl. 1999. Daily estradiol and progesterone levels relative to laying and the onset of incubation in Canaries. Gen. Comp. Endo. 114:257-268.

Soma, K.K. and J.C. Wingfield. 1999. Endocrinology of aggression in the non-breeding season. In: Proc. Intl. Ornith. Cong. 22. N.J. Adams and R.H. Slotow (eds.). Birdlife, Durban, South Africa. pp. 1606-1620.

Soma, K.K., K.A. Sullivan, A.D. Tramontin, C.J. Saldanha, B.A. Schlinger, and J.C. Wingfield. 2000. Acute and chronic effects of an aromatase inhibitor on territorial aggression in breeding and nonbreeding male Song Sparrows. J. Comp. Physio. 186: 759-769.

Spangelo, B.L. N.R. Hall, A.L. Goldstein. 1985. Evidence that prolactin is an immunomodulatory hormone. In: Prolactin: Basic and Clinical Correlates. R.M. MacLeod, M.O. Thorner, U. Scapagnini (eds.) Liviana Press, Padova, Italy. pp. 343-349.

Stacey, P.C. 1979. Habitat saturation and communal breeding in the Acorn Woodpecker. Anim. Behav. 27: 1153-1166.

Stacey, P.C. and W.D. Koenig. 1990. Cooperative Breeding in Birds: Long Term Studies of Ecology and Behaviour. Cambridge University Press, Cambridge. 615 pp.

Staub, N.L. and M. De Beer. 1997. The role of androgens in female vertebrates. Gen. Comp. Endo. 108: 1-24.

Strahl, S.D. and A.Schmitz. 1990. Hoatzins: cooperative breeding in a folicorous neotropical bird. In P.B. Stacey and W.D. Koenig (eds.) Cooperative Breeding in Birds: Long-term Studies of Ecology and Behaviour. Cambridge Univ. Press, Cambridge. pp. 131-156.

Strickland, D. 1991. Juvenile dispersal in Gray Jays: dominant brood member expels siblings from natal territory. Canadian J. Zool. 69: 2935-2945.

Strickland, D. and T.A. Waite. 2001. Does initial suppression of allofeeding in small jays help to conceal their nests? Canadian J. Zool. 79: 2128-2146.

SYSTAT 7.0 for Windows. 1997. SPSS, Inc.

Tanaka, K. and T. Inoue. 1990. Role of steroid hormones and catecholamines in the process of ovulation in the domestic fowl. In: Endocrinology of Birds: Molecular to Behavioral. M. Wada, S. Ishii, and C.G. Scanes (eds.) Japan Sci. Soc. Press, Tokyo. pp. 59-68.

Trail, P.W., S.D. Strahl, J.L. Brown. 1981. Infanticide in relation to individual and flock histories in a communally breeding bird, the Mexican Jay (*Amphelocoma ultramarina*). Am. Nat. 118: 72-82.

Tremblay, Y. and Y. Chrel. 1999. Synchronous underwater foraging behavior in Penguins. Condor 101: 179-185.

Trivers, R.L. 1971. The evolution of reciprocal altruism. Q. Rev. Biol. 46: 35-57.

Van Duyse, E., R. Pinxten, M. Eens. 2002. Effects of testosterone on song, aggression and nestling feeding behavior in male Great Tits *Parus major*. Horm. Behav. 41: 178-186.

Vehrencamp, S.L. 1977. Relative fecundity and parental effort in communally nesting Anis, *Crptophaga sulcirostris*. Science 197: 403-405.

Vehrencamp, S.L. 1979. The roles of individual, kin, and group selection in the evolution of sociality. In: Handbook of Behavioral Neurobiology, Social Behavior and Communication, Vol. 3. P. Maler and J.G. Vandenbergh (eds.). Plenum Press, New York. pp. 351-394

Vehrencamp, S.L., B.S. Bowen, and R.R. Koford. 1986. Breeding roles and pairing patterns within communal groups of Groove-billed Anis. Anim. Behav. 34: 347-366.

Verbeek, N.A.M. and R.W. Butler. 1981. Cooperative breeding of the Northwestern Crow *Corvus caurinus* in British Columbia. Ibis 123: 183-189.

Vleck, C.M., N.A. Mays, J.W. Dawson, and A.R. Goldsmith. 1991. Hormonal correlates of parental and helping behavior in cooperatively breeding Harris' Hawks (*Parabuteo unicinctus*). Auk 108: 638-648.

Vleck, C.M. and J.L. Brown. 1999. Testosterone and social and reproductive behaviour in *Aphelocoma* jays. Anim. Behav. 58: 943-951.

Vleck, C.M., L.L. Ross, D. Vleck, T.L. Bucher. 2000. Prolactin and parental behavior in Adelie Penguins: effects of absence from nest, incubation length, and nest failure. Horm. Behav. 38: 149-158.

Voci, V.E. and N.R. Carlson. 1973. Enhancement of maternal behavior and nest building following systemic and diencephalic administration of prolactin and progesterone in the mouse. J. Comp. Physio. Psych. 83: 388-393.

Wagner, R.H., E. Danchin, T. Boulinier, and F. Helfenstein. 2000. Colonies as byproducts of commodity selection. Behav. Ecol. 11: 572-537.

Waite, T.A. and D. Strickland. 1997. Cooperative breeding in Gray Jays: philopatric offspring provision juvenile siblings. Condor 99: 523-525.

Walters, J.R., P.D. Doerr, and J.H, Carter III. 1992. Delayed dispersal and reproduction as a life-history tactic in cooperative breeders: fitness calculations from Red-cockaded Woodpeckers. Am. Nat. 139: 623-643.

Wcislo, W.T. and B.N. Danforth. 1997. Secondarily solitary: the evolutionary loss of social behavior. TREE 12: 468-474.

Weigl, P.D. and E.V. Hanson. 1980. Observational learning and the feeding behavior of the Red Squirrel, *Tamiasciurus hudsonicus*: the ontogeny of optimization. Ecology 61: 213-218.

West-Eberhard, M.J. 1975. The evolution of social behavior by kin selection. Q. Rev. Bio. 50: 1-33.

Wetton, J.H., R.E. Carter, D.T. Parkin, and D. Walters. 1987. Demographic study of a wild House Sparrow population by DNA fingerprinting. Nature 327: 147-149.

Wetton, J.H., D.T. Parkin, and R.E. Carter. 1992. The use of genetic markers for parentage analysis in *Passer domesticus* (House Sparrows). Heredity 69: 243-254.

White, F.N., G.A. Bartholomew, and T.R. Howell. 1975. The thermal significance of the nest of the Weaver *Philetairus socius*: winter observations. Ibis 117: 171-179.

Whittingham, L.A., P.O. Dunn, and R.D. Magrath. 1997. Relatedness, polyandry and extra-group paternity in the cooperatively-breeding White-browed Scrubwren (*Sericornis frontalis*). Behav. Ecol. Sociobiol. 40: 261-270.

Wickler, W. 1980. Vocal duetting and the pair bond. I. Coyness and partner commitment. A hypothesis. Z. Tierpsychol. 52: 201-209.

Wikelski, M., M.A. Hau, and J.C. Wingfield. 1999. Social instability increases plasma testosterone in a year-round territorial neotropical bird. Proc. R. Soc. Lond. B 266: 551-556.

Wikelski, M., M.A. Hau, and J.C. Wingfield. 2000. Seasonality of reproduction in a neotropical rain forest bird. Ecology 81: 2458-2472.

Wiley, R.H. and K.N. Rabenold. 1984. The evolution of cooperative breeding by delayed reciprocity and queuing for favorable social positions. Evo. 38: 609-621.

Wilkinson, G.S. 1984. Reciprocal food sharing in the Vampire Bat. Nature 308: 181-184.

Williams, T.D. and P.J. Sharp. 1993. Plasma prolactin during the breeding season in adult and immature Macaroni (*Eudyptes chrysolophus*) and Gentoo (*Pygoscelis papua*) Penguins. Gen. Comp. Endo. 92: 339-346.

Wilson, M.I., G.M. Brown, and D. Wilson. 1978. Annual and diurnal changes in plasma androgen and cortisol in adult male Squirrel Monkeys (*Saimiri sciureus*) studied longitudinally. Acta Endo. (Copenhagen) 87: 424-433.

Wingfield, J.C. 1983. Environmental and endocrine control of avian reproduction: An ecological approach. In: Avian Endocrinology- Environmental and Ecological Perspectives. S. Mikami, K. Homma, and M. Wada (eds.). pp. 265-288.

Wingfield, J.C. 1984. Environmental and endocrine control of reproduction in the Song Sparrow, *Melospiza melodia*. I. Temporal organization of the breeding cycle. Gen. Comp. Endo. 56: 406-416.

Wingfield, J.C. 1990. Interrelationships of androgens, aggression, and mating systems. In: Endocrinology of Birds: Molecular to Behavioral. M. Wada, S. Ishii, and C.G. Scanes (eds.). Japan Sci. Soc. Press, Tokyo. pp. 187-205.

Wingfield, J.C. 1994a. Control of territorial aggression in a changing environment. Psychoneuroendo. 19: 709-721.

Wingfield, J.C. 1994b. Modulation of the adrenocortical response to stress in birds. In: Perspectives in Comparative Endocrinology. K.G. Davies, R.E. Peter, and S.S. Tobe (eds.). National research Council Canada, Ottawa. pp. 520-528.

Wingfield, J.C. and D.S. Farner. 1978a. The annual cycle of plasma irLH and steroid hormones in feral populations of the White-crowned Sparrow, *Zonotrichia leucophrys gambelii*. Biol. Reprod. 19: 1046-1056.

Wingfield, J.C. and D.S. Farner. 1978b. The endocrinology of a natural breeding population of the White-crowned Sparrow (*Zonotrichia leucophrys pugetensis*). Physio. Zool. 51: 188-205.

Wingfield, J.C. and D.S. Farner. 1980. Control of seasonal reproduction in temperate-zone birds. Prog. Reprod. Biol. 5: 62-101.

Wingfield, J.C., A. Newman, G. L. J. Hunt, and D.S. Farner. 1980. Androgen in high concentrations in the blood of female Western gulls, *Larus occidentalis*. Naturwissenschaften 67: 514.

Wingfield, J.C., A.L. Newman, G.L. Hunt, and D.S. Farner. 1982. Endocrine aspects of female-female pairing in the Western gull (*Larus occidentalis wymani*). Anim. Behav. 30: 9-22.

Wingfield, J.C. and M. Ramenofsky. 1985. Testosterone and aggressive behaviour during the reproductive cycle of male birds. In: Neurobiology. R.Gilles and J. Balthazart (eds). Springer-Verlag, Berlin. pp. 92-104.

Wingfield, J.C. and B. Silverin. 1986. Effects of corticosterone on territorial behaviour of free-living male song sparrows *Melospiza melodia*. Horm. Behav. 20: 405-417.

Wingfield, J.C., G.F. Ball, A.M. Dufty, Jr., R.E. Hegner, and M. Ramenofsky. 1987. Testosterone and aggression in birds. Am. Sci. 75: 602-608.

Wingfield, J.C. and M.C. Moore. 1987. Hormonal, social, and environmental factors in the reproductive biology of free-living male birds. In: Psychobiology of Reproductive Behavior: An Evolutionary Perspective. D. Crews (ed.). Prentice-Hall, Englewood Cliffs, NJ. pp. 149-175.

Wingfield, J.C. and P. Marler. 1988. Endocrine basis of communication in reproduction and aggression. In: The Physiology of Reproduction. E. Knobel and J. Neill (eds.). Raven, New York. pp. 1647-77.

Wingfield, J.C., E. Ronchi, A.R. Goldsmith, and C. Marler. 1989. Interactions of sex steroid hormones and prolactin in male and female Song Sparrows, *Melospiza melodia*. Physio. Zool. 62: 11-24.

Wingfield, J.C. and A.R. Goldsmith. 1990. Plasma levels of prolactin and gonadal steroids in relation to multiple-brooding and renesting in free-living populations of the song sparrow, *Melospiza melodia*. Horm. Behav. 24: 89-103.

Wingfield, J.C., T.E. Hegner, A.M. Dufty, Jr., and G.F. Ball. 1990. The "challenge hypothesis": theoretical implications for patterns of testosterone secretion, mating systems, and breeding strategies. Am. Nat. 136: 829-846.

Wingfield, J.C., Hegner, R.E., and Lewis, D.M. 1991. Circulating levels of luteinizing hormone and steroid hormones in relation to social status in the cooperatively breeding White-browed Sparrow Weaver, *Plocepasser mahali*. J. Zool., London 225: 43-58.

Wingfield, J. C., T. P. Hahn, R. N. Levin, and P. Honey. 1992. Environmental predictability and control of gonadal cycles in birds. J. Exp. Zool. 261: 214-231.

Wingfield, J.C. and D. Monk. 1992. Control and context of year-round territorial aggression in the non-migratory Song Sparrow *Zonotrichia melodia morphna*. Ornis Scand. 23: 298-303.

Wingfield, J.C. and D.S. Farner. 1993. Endocrinology of reproduction in wild species. *In* D.S. Farner, J.R. King, and K.C. Parkes (eds.) Avian Biology. Vol IX. pp. 163-327. Academic Press. New York.

Wingfield, J.C. and D.M. Lewis. 1993. Hormonal and behavioural responses to simulated territorial intrusion in the cooperatively breeding white-browed sparrow weaver, *Plocepasser mahali*. Anim. Behav. 45: 1-11.

Wingfield, J.C., C.S. Whaling, and P. Marler. 1994. Communication in vertebrate aggression and reproduction: the role of hormones. *In* E. Knobil and J.D. Neill (eds.) *Physiology of Reproduction*. pp. 303-342. Raven Press, New York.

Wingfield, J.C., K.M. O'Reilly, and L.B. Astheimer. 1995. Ecological bases of the modulation of adrenocortical responses to stress in Arctic birds. Am. Zool. 35: 285-294.

Wingfield, J.C., J. Jacobs, and N. Hillgarth. 1997. Ecological constraints and the evolution of hormone-behavior interrelationships. In: The Integrative Neurobiology of Affiliation. S. Carter, I.I. Lederhendler, and B. Kirkpatrick (eds.). NY Academy of Sciences, New York. pp. 22-41.

Wingfield, J.C., D.L. Maney, C.W. Breuner, J.D. Jacobs, S. Lynn, M. Ramenofsky, and R.D. Richardson. 1998. Ecological bases of hormone-behavior interactions: the "emergency life history stage." Am. Zool. 38: 191-206.

Wingfield, J.C., J. Jacobs, K.K. Soma, D. Maney, K. Hunt, D. Wisti-Peterson, S. Meddle, M. Ramenofsky, K. Sullivan. 1999. Testosterone, aggression, and communication: ecological bases of endocrine phenomena. In: The Design of Animal Communication. M. Hauser and M. Konishi (eds.). MIT Press, Cambridge, MA. pp. 255-284.

Wingfield, J.C., J.D. Jacobs, A.D. Tramontin, N. Perfito, S. Meddle, D.L. Maney, and K. Soma. 2000. Toward an ecological basis of hormone-behavior interactions in reproduction of birds. In: Reproduction in Context. K. Wallen and J. Schneider (eds.). MIT Press, Cambridge, MA. pp. 85-128.

Wingfield, J.C., K.K. Soma, M. Wikelski, S.L. Meddle, and M. Hau. 2001. Life cycles, behavioural traits and endocrine mechanisms. In: Avian Endocrinology. A. Dawson and C.M. Chaturvedi (eds.). Narosa Publishing House, New Delhi. pp. 3-17.

Wingfield, J.C. and A.S. Kitaysky. 2002. Endocrine responses to unpredictable environmental events: stress or anti-stress? Integ. Comp. Biol. 42: 600-609.

Wingfield, J.C. and K.K. Soma. 2002. Spring and autumn territoriality in Song Sparrows: same behavior, different mechanisms? Integr. Comp. Biol. 42: 11-20.

Witter, M.S. and I.C. Cuthill. 1993. The ecological costs of avian fat storage. Philosophical Transactions of the R. Soc. Lond. B 340: 73-92.

Woolfenden, G.E. 1981. Selfish behavior by Florida Scrub-Jay helpers. In: Natural Selection and Social Behavior- Recent Research and New Theory. R.D. Alexander and D.W. Tinkle (eds.). Chiron Press, New York. pp. 257- 260.

Woolfenden, G.E. and J.W. Fitzpatrick. 1977. Dominance in the Florida Scrub-Jay. Condor 79: 1-12.

Woolfenden, G.E. and Fitzpatrick, J.W. 1984. The Florida Scrub-Jay. Demography of a Cooperative-Breeding Bird. Princeton University Press. Princeton, NJ. 426 pp.

Wright, J. 1997. Helping-at-the-nest in Arabian babblers: signalling social status or sensible investment in chicks? Anim. Behav. 54: 1439-1448.

Wright, J. 1998. Helping-at-the-nest and group size in the Arabian Babbler *Turdoides squamiceps*. J. Avian Biol. 29: 105-112.
Yezerinac, S., M. Double, A. Higgins. 1996. Multi-locus DNA fingerprinting protocol. Botany and Zoology Department, Australian National University, Canberra. 25 pp.

Yezerinac, S., M. Double, A. Higgins, G. Sargent, S. Legge, and M. Hall. 1996. Multi-locus DNA fingerprinting protocol. Botany and Zoology Department, Australian National University, Canberra. 30 pp.

Yodyingyuad, U., J.A. Eberhart, and E.B. Keverne. 1982. Effects of rank and novel females on behaviour and hormones in male Talapoin Monkeys. Physio. Behav. 28: 995-1005.

Zahavi, A. 1974. Communal nesting by the Arabian Babbler- a case of individual selection. Ibis 116: 84-87.

Zahavi, A. 1975. Mate selection- a selection for a handicap. J. Theor. Biol. 53: 205-214.

Zahavi, A. 1990. Arabian Babblers: the quest for social status in a cooperative breeder. In: Cooperative Breeding in Birds- Long Term Studies of Ecology and Behavior. P.B. Stacey and W.D. Koenig (eds.). Cambridge University Press, Cambridge. pps. 103-130.

Zahavi, A. 1995. Altruism as a handicap- the limitations of kin selection and reciprocity. J. Avian Bio. 26: 1-3.

Zann, R.F. 1996. The Zebra Finch: a synthesis of field and laboratory studies. Oxford University Press. Oxford.

Zar, J.H. 1999. Biostatistical Analysis. Prentice-Hall, Inc. Upper Saddle River, NJ. 663 pp.

Zarrow, M.X., R. Gandelman, and V.H. Denenberg. 1971. Prolactin: Is it an essential hormone for maternal behavior in the mammal? Horm. Behav. 2: 343-354.

Ziegler, T.E., F.H. Wegner, A.A. Carlson, C. Lazaro-Perea, and C.T. Snowdon. 2000. Prolactin levels during the periparturitional period in the biparental Cotton-top Tamarin (*Saguinus oedipus*): Interactions with gender, androgen levels, and parenting. Horm. Behav. 38: 111-122.

Zuk, M., T.S. Johnsen, and T. Maclarty. 1995. Endocrine-immune interactions, ornaments and mate choice in Red Jungle Fowl. Proc. R. Soc. Lond. B. 260: 205-210.