General discussion
Background of the project

The research on blue tit sex ratio adjustment and male plumage coloration described in this thesis was part of a larger research program on avian sex allocation, which included four subprojects in total. The research program, which was funded by the Netherlands Organisation for Scientific Research (NWO) and coordinated by Serge Daan (University of Groningen), aimed at elucidating functions and mechanisms of avian sex allocation. Each subproject focussed on different aspects of sex allocation in birds. One project (PhD research by Nikolaus von Engelhardt, supervision by Cor Dijkstra and Ton Groothuis, University of Groningen) concentrated on revealing the physiological and hormonal mechanisms generating shifts in the primary and secondary sex ratio (von Engelhardt 2004). This research was carried out on zebra finches (*Taeniopygia guttata*), which seem ideal for studying the proximate control of avian sex allocation. Zebra finches are easy to keep and breed in captivity and shifts of primary or secondary sex ratio in response to manipulated male attractiveness (Burley 1981, 1986) and feeding regime (Kilner 1998; Bradbury & Blakey 1998; Rutkowska & Cichoń 2002; Rutstein *et al.* 2004b) have previously been described, which makes sex ratio variation in zebra finches open to experimentation. A second project (by Tim Fawcett, Ido Pen and Franjo Weissing, University of Groningen) took a modelling approach to further develop the theory on adaptive sex allocation, with an emphasis on vertebrates with complex life-histories such as birds and mammals (*e.g.* Pen & Weissing, 2000; Fawcett *et al.* 2006). The two remaining projects studied the functional aspects of sex allocation in response to male attractiveness in wild bird populations. The blue tit (*Parus caeruleus*) was chosen as a model species because male sexual attractiveness in the form of UV coloration of the crown plumage is easy to measure and manipulate in the field and shifts in sex ratio in relation to male attractiveness (Sheldon *et al.* 1999) and quality (measured in terms of over-winter survival; Svensson & Nilsson 1996) had been previously shown. In addition, the blue tit is a very abundant bird species in western Europe including the Netherlands and readily accepts nestboxes for breeding (Perrins 1979), making it a very practical species for study.

One of the two blue tit projects (PhD research by Tobias Limbour, supervision by Kate Lessells, Netherlands Institute of Ecology, Heteren) investigated sex-biased parental care as a strategy to adjust parental investment in male versus female offspring (Limbour *et al.* 2004). The other project (supervision by Jan Komdeur, University of Groningen) investigated facultative adjustment of the sex ratio at laying (the primary sex ratio), which results are presented in this thesis.

In this last chapter, I first briefly review recent developments in avian sex allocation research and discuss our results in the light of these developments. Thereafter, I discuss the progress that has been made in understanding the role of the ornamental UV/blue crown plumage in sexual selection and sex allocation in the blue tit and compare our results to the findings in other blue tit populations. Finally, I suggest some directions for future research.
Avian sex allocation: function and mechanism

Mechanisms of primary sex ratio adjustment

It has long been thought that the chromosomal sex determination system of birds (and mammals) with random segregation of the sex chromosomes during meiosis poses a strong constraint that precludes facultative sex ratio adjustment at laying (Williams 1979; Clutton-Brock 1986). In the 1990s, however, a number of studies was published that showed significant, and probably also adaptive, shifts in the primary sex ratio (e.g. Dijkstra et al. 1990; Wiebe & Bortolotti 1992; Daan et al. 1996; Ellegren et al. 1996; Komdeur 1996; Svensson & Nilsson 1996; Appleby et al. 1997; Komdeur et al. 1997, Kilner 1998; Nager et al. 1999; Pen et al. 1999; Sheldon et al. 1999). These studies have stimulated a surge of interest in facultative primary sex ratio adjustment in birds and since then the number of published studies on the subject has been expanding continuously (see Figure 1.1, Chapter 1). Despite this large research effort, there is ongoing controversy about the generality of facultative primary sex ratio adjustment in birds (Koenig & Dickinson 1996; Radford & Blakey 2000; Krackow 2002) and recent meta-analyses have come to opposing conclusions on the general sex ratio patterns in the literature (West & Sheldon 2002; Ewen et al. 2004; Cassey et al. 2006). One important issue which is hindering further progress in this debate is the lack of a known genetic or physiological mechanism that birds use to influence the primary sex ratio (Krackow 1995; Pike & Petrie 2003; Krackow 2002).

In birds, in contrast to mammals, females are the heterogametic sex (females are ZW; males are ZZ). Consequently, female birds produce male (Z) and female (W) ova, which potentially gives them control over the sex of their offspring (Krackow 1995; Oddie 1998). Although it is far from clear how birds could bias the sex ratio at laying, several mechanisms are conceivable (Krackow 1995; Emlen 1997; Komdeur et al. 2002; Pike & Petrie 2003). These can broadly be separated into pre- and post-ovulation control mechanisms. Pre-ovulation, females could possibly influence the sex of the oocyte shed from the ovary either by non-random segregation of sex chromosomes during the first division of meiosis shortly before ovulation, by atresia of follicles of the ‘wrong’ sex, or by differential growth of follicles destined to become a specific sex (Krackow 1995; Komdeur et al. 2002; Pike & Petrie 2003). These can broadly be separated into pre- and post-ovulation control mechanisms. Pre-ovulation, females could possibly influence the sex of the oocyte shed from the ovary either by non-random segregation of sex chromosomes during the first division of meiosis shortly before ovulation, by atresia of follicles of the ‘wrong’ sex, or by differential growth of follicles destined to become a specific sex (Krackow 1995; Komdeur et al. 2002; Pike & Petrie 2003). Post-ovulation, oocytes of the ‘wrong’ sex could be re-absorbed at various stages before and after fertilisation. Such re-absorption of a ‘wrong’ sex oocyte after ovulation would probably lead to a gap in the laying sequence (Emlen 1997), given the strong hierarchy in the development of follicles in the ovary with a fixed interval between the maturation of the successive follicles (Sturkie 1986). Female manipulation of egg sex by post-ovulation absorption would probably be more costly than pre-ovulation control, because of the loss of time and invested resources (Pen & Weissing 2002). It is not unlikely that different sorts of mechanisms of sex ratio adjustment are at work in different avian taxa (Pike & Petrie 2003).
Recently, several findings have been published that may bring us a step closer to understanding the physiological basis of avian primary sex ratio adjustment. In several species, the sex and size of eggs have been found to be related (Mead et al. 1987; Anderson et al. 1997; Cordero et al. 2000, 2001; Magrath et al. 2003), which indicates that females may be able to discriminate between eggs of different sex in their reproductive tract and allocate resources accordingly. Moreover, studies have found egg sex-specific allocation of maternal hormones to the yolk (Petrie et al. 2001; Müller et al. 2002; but see Pilz et al. 2005), which must be the result of differential hormone allocation to the developing oocyte by the cells of the follicular wall. This suggests either that females are able to discriminate between follicles destined to become a specific sex, or that the maternal hormones allocated to the developing oocyte influence the segregation of the sex chromosomes during the first meiotic division shortly before ovulation. Furthermore, several studies have shown endocrine influences on primary sex ratio (Correa et al. 2005; Pike & Petrie 2006; but see von Engelhard et al. 2004). Particularly strong evidence for pre-ovulation control of egg sex was presented by Komdeur et al. (2002) who showed egg sex of Seychelles warblers (Acrocephalus sechellensis) to be significantly biased without a preceding laying gap, which implies that the bias already existed before ovulation. Very recently, the first evidence has been found for atresia of ‘wrong’ sex oocytes by pigeons (Pike 2005; pre-ovulatory follicle selection) as well as sex-specific growth patterns of oocytes of house finches that exhibit sex-biased laying orders (Young & Badyaev 2004). This long-needed progress in the elucidation of the physiological basis of primary sex ratio adjustment is both very exciting and promising, and will hopefully give the study of avian primary sex ratio adjustment a new impulse.

**Primary sex ratio adjustment to male attractiveness**

A wide array of factors have been found to influence primary sex ratio in birds, including for example position in the laying order (Badyaev et al. 2002), season (Dijkstra et al. 1990), prey abundance (Appleby et al. 1997), habitat quality (Komdeur et al. 1997), feeding regime (Kilner 1998), maternal condition (Nager et al. 1999), male quality (Svensson & Nilsson 1996) and male sexual attractiveness (Sheldon et al. 1999). Although these studies show that birds modify the primary sex ratio in response to a wide variety of factors, it is often difficult to make a clear a priori prediction on the relationship between primary sex ratio and a specific factor. The sex ratio shift to be expected often depends on the exact details of an animal’s life-history, which are often not precisely known in birds (Pen & Weissing 2002; West & Sheldon 2002). One factor for which it is possible to give a clear a priori prediction of the expected effect on sex ratio is male sexual attractiveness (West & Sheldon 2002). Females are expected to produce more sons when mated to an attractive male (Burley 1981, 1986; Pen & Weissing 2000; Fawcett et al. 2006). This is predicted because in birds males show generally greater variation in reproductive success than females (Møller & Ninni 1998). Therefore, sons will benefit more
from having an attractive father, given that attractiveness is heritable. Burley (1981, 1986) originally developed this verbal argument, and she showed that offspring sex ratio at fledging was related to manipulated male attractiveness in zebra finches. The validity of the verbal argument has recently been confirmed by analytical (Pen & Weissing 2000) and simulation models (Fawcett et al. 2006).

Several correlational studies have attempted to find a relationship between primary sex ratio and male attractiveness or quality, yielding mixed evidence for such a link, with results varying between different years (e.g. Radford & Blakey 2000; Griffith et al. 2003) and populations (e.g. Svensson & Nilsson 1996; Leech et al. 2001; Rosivall et al. 2004; Griffith et al. 2003; Dreis et al. 2006). Relatively few experimental studies in which some aspect of male attractiveness was manipulated have found an effect on primary sex ratio (collared flycatcher [Ficedula albicollis], Ellegren et al. 1996; blue tit, Sheldon et al. 1999; spotless starling [Sturnus unicolor], Polo et al. 2004; peafowl [Pavo cristatus], Pike & Petrie 2005). The results on blue tit primary sex ratio variation in relation to manipulated male UV coloration presented in this thesis add another experimental example of primary sex ratio adjustment to male attractiveness. Several other experimental studies in different species, have failed to find an effect of male attractiveness on primary sex ratio (barn swallow [Hirunda rustica], Saino et al. 1999; mallard [Anas platyrhynchos], Cunningham & Russell 2000; dark-eyed junco [Junco hyemalis], Grindstaff et al. 2001; zebra finch [Taeniopygia guttata], Zann & Runciman 2003; Rutstein et al. 2004a). Note that Burley (1981, 1986) in her seminal study on offspring sex ratio and male attractiveness in zebra finches investigated the sex ratio of surviving offspring after fledging (i.e. the secondary sex ratio).

Given the clear a priori prediction that females paired to attractive males should produce more sons, the inconsistency between studies is difficult to explain. It is interesting in this respect that the modelling study of Fawcett et al. (2006) showed that, although females should indeed produce more sons when mated with an attractive male, the selective pressure for such sex ratio adjustment to evolve is very weak. This may explain why studies find such different patterns in different years, populations and species. Given the generally weak selection pressure for sex ratio adjustment to evolve, the occurrence of sex ratio adjustment to male attractiveness may be very sensitive to the specific circumstances of each particular population and study species. Their specific ecological and social environment may easily shift the balance of the costs and benefits of sex ratio control in relation to male attractiveness. Further study of the fitness consequences of sex ratio adjustment would be needed to better predict the patterns of sex ratio variation to be expected (Komdeur & Pen 2002).

**Primary sex ratio adjustment in the blue tit**

Several correlational and experimental studies have now investigated the relationship between the primary sex ratio and male attractiveness or quality in blue tits (Table 8.1). The overall picture that emerges from these studies is rather mixed, with some evidence for primary sex ratio adjustment, but patterns vary among
study years and populations. Svensson and Nilsson (1996) provided the first evidence for primary sex ratio adjustment in the blue tit. In a Swedish population, they found females to bias the sex ratio of their broods towards sons if paired to a supposedly high-quality male that survived to the following breeding season. As a follow up on this finding, Sheldon et al. (1999) carried out their influential study (>150 citations) in a Gotland population of blue tits that provided both correlational and experimental evidence for primary sex ratio adjustment in relation to male crown UV reflectance – which is probably an important determinant of male attractiveness (Andersson et al. 1998; Hunt et al. 1998; Delhey et al. 2003). Sheldon et al.’s (1999) correlational data showed that females produced a greater proportion of male eggs if paired with a male with higher natural UV reflectance, which is consistent with the straightforward prediction by sex allocation theory (Trivers & Willard 1973; Burley 1981, 1986; Fawcett et al. 2006).

The experimental data of Sheldon et al. (1999) were more difficult to interpret. Experimental reduction of male UV reflectance – making males less attractive – did not result in a lower proportion of male offspring, as predicted by sex allocation theory (e.g. Fawcett et al. 2006). Instead, the positive correlation between the sex ratio and natural male UV reflectance was reversed so that the proportion of sons decreased with increasing pre-treatment UV reflectance, which was also indicated by a significant ‘UV treatment x pre-treatment UV reflectance’ interaction effect on sex ratio (Sheldon et al. 1999). To test the generality of this unexpected pattern we replicated the Sheldon et al. (1999) study in two years in our Vosbergen blue tit population, while closely following Sheldon et al.’s (1999) experimental procedures (Korsten et al. 2006; Chapter 4). Like Sheldon et al. (1999), we found no difference in the overall sex ratio between the UV-reduced and control group, whereas in one of two years, the ‘UV treatment x pre-treatment UV reflectance’ interaction was significant (Korsten et al. 2006; Chapter 4). This result is important, because it shows the unexpected sex ratio pattern found by Sheldon et al. (1999) to be repeatable among populations, which provides the basis for moving on to the next step of elucidating the adaptive value of these complex patterns of blue tit sex ratio variation.

After the benchmark study of Sheldon et al. (1999), several correlational studies on blue tit primary sex ratio, male attractiveness and survival have been carried out in different populations (Table 8.1). The positive correlation between the proportion of sons and male UV reflectance reported by Sheldon et al. (1999), which was based on data of a single breeding season, was confirmed by additional data of the same population collected in a second year, but not by the data of a third year (Griffith et al. 2003). Furthermore, these data showed that males surviving to the following breeding season tended to have a higher proportion of sons in their broods than males that did not survive (Griffith et al. 2003), which confirms the study by Svensson and Nilsson (1996). In contrast, in a large dataset (comprising three study years) on a blue tit population in the UK no correlations among primary sex ratio, (extra-pair) paternity and male survival were found (Leech et al. 2001). Unfortunately, no data on
male crown coloration are available for this population. Finally, a recent study on a French blue tit population found no significant correlation between offspring sex ratio and male crown coloration. Instead, the proportion of male offspring was positively correlated with a quality aspect of male song (Dreiss et al. 2006). It is unclear why correlational patterns of blue tit sex ratio differ between populations and more work is needed to understand the proximate and ultimate causes for this variability. A first step would be to compare the role of crown UV coloration in mate choice and competition, as well as its heritability, between the different populations.

**Ornamental plumage coloration in the blue tit**

The popularity of the blue tit and its structurally based UV/blue ornamental crown plumage as a model system in sexual selection and sex allocation research leads to a very favourable situation for behavioural ecologists. In this situation there is a large,
and still growing, amount of data on a single model system, while a number of investigations and experiments have also been replicated in different populations (e.g. Andersson et al. 1998; Hunt et al. 1998; Sheldon et al. 1999; Örnborg et al. 2002; Delhey et al. 2003; Foerster et al. 2003; Griffith et al. 2003; Alonso-Alvarez et al. 2004; Limbourg et al. 2004, Johnsen et al. 2005; Dreiss et al. 2006; Delhey et al. 2006; Delhey & Kempenaers 2006; Korsten et al. 2006). Here, I will summarize the present knowledge of this well-studied plumage ornament and compare the results presented in this thesis with findings by other studies.

**Sexual dichromatism of UV coloration**

Blue tits have long been regarded as largely monochromatic (Cramp & Perrins 1993), with males having perhaps only slightly brighter blue crown plumage than females (Perrins 1979). In 1998, however, two studies using spectrophotometry showed independently that the reflectance of the blue crown feathers was markedly different between males and females in the UV part of the spectrum (Andersson et al. 1998; Hunt et al. 1998; see also Figure 1.2, Chapter 1). It was shown that males have on average more UV-shifted and UV-chromatic crown plumage. This finding has been confirmed in a number of other populations (Delhey et al. 2006; Hadfield et al. 2006; Chapters 1 and 7). Moreover, it has now become clear that UV reflecting plumage as well as sexual dichromatism in the UV is in fact present in numerous bird species (Eaton & Lanyon 2003; Eaton 2005).

Blue tits have UV vision (Hart et al. 2000), like most passerine birds (Cuthill et al. 2000), and are thus able to perceive variation in UV reflectance of the plumage of conspecifics. The clear sexual dichromatism in blue tits must be the result of sex-specific selection on crown UV coloration, which strongly suggests that the blue tit crown plumage is a sexually selected ornament that functions as a signal in female mate choice or male-male competition. Interestingly, the sexual dichromatism is far from complete, and there is large individual variation in crown UV coloration (Chapter 7), with substantial overlap of the distributions of male and female crown colour (Chapters 1 and 7). This suggests that the UV crown plumage may have a signalling function in females as well, and play a role in male mate choice or inter-individual competition. The sexual dichromatism could then be viewed as the result of a different balance of the costs (e.g. increased predation risk) and benefits (e.g. more successful mate attraction) of crown UV coloration in males and females.

Alternatively, female UV coloration may be the result of a non-adaptive genetic correlation with male ornamentation, while sex-limitation of ornament expression is incomplete (Kraaijeveld et al. submitted). According to this scenario selective pressures on male and female crown colour expression may be counteracting each other to some extent. Such a situation has been described for the ornamental red bill coloration of zebra finches (Price & Burley 1993, 1994; Price 1996). In zebra finches, males with redder bills are more successful in mate attraction and have higher fitness, while in females individuals with more orange (less red) bills have
higher fitness (Price & Burley 1994). This leads to a situation in which there is sexually antagonistic selection (Rice 1992) on bill coloration, with males selected to become more red and females to become more orange (Price & Burley 1994). A genetic correlation between bill coloration of males and females subsequently prevents the evolution of more pronounced sexual dichromatism (Price & Burley 1993, 1994; Price 1996). It is not unlikely that a similar situation exists for the blue tit’s crown coloration. It is interesting in this respect that we found a preliminary hint of sex-linked inheritance (or a maternal effect) affecting crown colour expression (Chapter 2), which might be expected in case of such sexually antagonistic selective pressures (Merilä & Sheldon 2001). The possibility that there is sexually antagonistic selection on blue tit crown coloration certainly deserves further attention, and this idea could be tested by a quantification of the fitness consequences of variation in crown coloration in both males and females.

**Seasonal variation in UV coloration**

An extremely important source of variation in blue tit crown coloration is the time of year (Örnborg et al. 2002; Delhey et al. 2006; Chapters 2 & 7). The reflectance of the crown plumage is maximally UV-shifted just after the yearly moult in autumn (Örnborg et al. 2002). Thereafter, over the year, crown coloration becomes gradually less UV-shifted, and towards the next breeding season also UV chroma decreases, while overall brightness increases (Örnborg et al. 2002). Changes in crown coloration are especially rapid during the breeding season between nest building and chick feeding (Örnborg et al. 2002). The decline in UV reflectance is probably due to feather wear as well as accumulation of fat and dirt (Örnborg et al. 2002). In particular the abrasion of the feather barbs and barbules containing the microscopic light-scattering structures could lead to a change in feather colour (Örnborg 2002). Also the action of feather-degrading bacteria and other ecto-parasites may play a role in the deterioration of the feathers (Kose & Møller 1999; Shawkey & Hill 2004).

The magnitude of the seasonal colour changes is surprisingly large (Örnborg et al. 2002; Delhey et al. 2006; Chapter 2). For example, Delhey et al. (2006) showed that changes in UV chroma and hue between early winter and late spring were similar to the differences in UV coloration due to sex and age respectively. It is unknown what the functional consequences of these striking colour changes are. The decline in UV coloration may be adaptive, and related to a shifting balance between benefits of a strong UV signal used in mate attraction during winter and early spring and costs such as the risk of detection by predators. For example, male rock ptarmigans (*Lagopus mutus*) soil their conspicuous white breeding plumage with dirt as soon as mating has taken place to increase crypsis (Montgomerie et al. 2001). On the other hand it has been hypothesised that the magnitude of the decrease in UV coloration may be related to individual quality (Delhey et al. 2006). According to this idea high quality individuals are better able to prevent their plumage from degradation, because they can invest more time and energy in main-
tenance of their feathers (e.g. in the form preening behaviour). Indeed, a measure of body size (tarsus length) was negatively related to the seasonal change in blue tit hue, whereas the magnitude of the decline in UV chroma was related to the loss of body mass between winter and spring, supposedly indicating a cost of feather maintenance (Delhey et al. 2006). However, female extra-pair mate choice was not related to variation in the magnitude of the seasonal declines (Delhey et al. 2006). Clearly, further research is needed to better understand the causes and consequences of the sharp seasonal decline in blue tit UV coloration.

Whether or not there is an adaptive explanation for the seasonal changes in crown coloration, it is obviously extremely important to control for this variation in analyses of crown colour variation. Especially, when estimates of crown coloration collected over long time frames are included or when comparisons of crown coloration are made between individuals measured at different periods (Chapter 2). If not adequately controlled, the large seasonal variation could easily obscure interesting patterns or lead to spurious relationships.

**Age-related variation in UV coloration**

A small part of the individual variation in blue tit UV coloration is related to age, both in males and females (Chapters 2 and 7). Older birds have significantly more UV-shifted and UV-chromatic crown plumage (Chapter 2). This supports the idea that the UV coloration is an indicator of individual quality, as condition dependent ornaments are typically more developed in older individuals (Andersson 1994; Siefferman et al. 2005). However, the age differences in UV coloration appear to be very subtle when compared to the variation due to sex differences as well as the seasonal changes (Delhey et al. 2006; Chapters 2 and 7). Indeed, we could assign the correct age based on crown UV coloration of only 56.0% of males and 72.0% females measured during winter (Chapter 7), which makes it a rather poor indicator of age.

The increase in UV coloration with age could have two different causes: 1) individuals may increase in UV coloration during their life; 2) individuals with less intense UV coloration may have a lower survival probability, leading to a positive correlation between age and UV colour due to selective disappearance of low-UV individuals at older ages (Siefferman et al. 2005). So far, there is evidence that both processes could be responsible for the positive correlation between UV coloration and age in blue tits. In the Gotland population, males, but not females, with more UV-chromatic crown plumage had a higher chance of surviving to the following breeding season (Sheldon et at. 1999; Griffith et al. 2003). In a blue tit population in Austria, however, over-winter survival was not dependent on crown coloration. Instead, in this population UV coloration was shown to increase with age within individuals (Delhey & Kempenaers 2006). In conclusion, evidence suggests that the sexually dichromatic UV coloration can function as a signal of individual quality or age, but it remains to be explained why the observed age effects are very subtle in comparison to overall individual variation.
Consequences of UV colour variation for mate choice and sexual selection

The greater expression of the UV coloration in males than in females (Andersson et al. 1998; Hunt et al. 1998) is a strong indication that the UV/blue crown plumage is a sexually-selected ornament in blue tits (Andersson 1994). However, the direct evidence for a role of UV coloration in mate choice is not as strong and straightforward as is sometimes assumed (Sheldon et al. 1999; Korsten et al. 2006).

A mate choice experiment with a relatively small number of captive females (n = 7) has shown that females preferred males with greater brighter (achromatic brightness), but not more UV-chromatic, crown feathers (Hunt et al. 1998). In another mate choice experiment both males and females were allowed to choose between two potential mates of the other sex that were each placed behind a transparent screen; one screen was transparent to UV light (UV+), whereas the other screen was opaque to UV (UV–) (Hunt et al. 1999). In this experimental set-up males and females had a preference for the UV+ individual. However, this result does not necessarily demonstrate that blue tits specifically prefer potential mates that have more UV-reflecting crown feathers, because the entire appearance and background are different between UV+ and UV– birds (cf the effect of looking through blue sunglasses which filter out red light; see also Hunt et al. 2001 for further discussion). In fact, this experiment only demonstrates that blue tits are capable of visual discrimination in the UV part of the spectrum, and that they select against odd looking (UV–) mates.

In a wild blue tit population in Sweden assortative mating for crown UV chroma was found (Andersson et al. 1998), which implicates mutual mate choice with both males and females preferring more UV-chromatic mates. However, in some other populations including ours (Box B) no such assortative mating was found (T. Limbourg, personal communication; but see Alonso-Alvarez et al. 2004). It is unclear what causes this difference between populations. A possible explanation for the difference may be the fact that northerly populations of blue tits in Sweden are less sedentary than temperate populations and often leave their breeding areas during harsh conditions in winter. This may lead to increased ‘freedom’ of mate choice during pair-formation in early spring when the birds return to the breeding area in northern populations, whereas in more temperate populations pairs tend to remain in the breeding area all year round, thereby restricting the choice options for unpaired individuals.

Analyses of genetic parentage in several blue tit populations have revealed that ca. 10–15% of all offspring are sired by a male outside the social pair bond, which is normally formed by a single male and female (Kempenaers et al. 1997; Krokene et al. 1998; Leech et al. 2001; Delhey et al. 2003; Box D). The occurrence of extra-pair paternity increases the variance in male reproductive success in socially monogamous bird species, such as the blue tit, and thereby the potential for sexual selection on male ornaments. Comparative analyses have suggested that the evolution of structurally based plumage coloration, such as the blue tit’s crown coloration, is associated with high extra-pair paternity rates on the species level (Owens &
At present only one published study has investigated the relationship between extra pair paternity and male crown plumage in blue tits in more detail (Delhey et al. 2003; see also Delhey et al. 2006). This study showed that males with a more UV-shifted crown hue are less cuckolded (Delhey et al. 2003), which is consistent with a female preference for more ornamented males. Remarkably, however, males with less UV-shifted hue sired more extra-pair young (Delhey et al. 2003). This may indicate a potential trade-off between guarding the paternity of the own brood and the pursuit of extra-pair copulations (Kokko & Morrell 2005). In that case, the opposite correlations between UV coloration and within- and extra-pair paternity may reflect alternative male mating strategies, where more UV-ornamented males maximize within-pair success and less UV-ornamented males maximize extra-pair success (Delhey et al. 2003). However, an experimental UV reduction of males did not result in the predicted lower within-pair paternity in our study population, which refutes such a causal relationship between within-pair paternity and UV coloration (Box D). Assignment of extra-pair males would be an adequate next step to further investigate the causal links between UV coloration and male extra-pair success in our blue tit population.

Although the direct evidence for a role of blue tit crown UV coloration in (extra-pair) mate-choice is still not particularly strong, two recent studies on differential allocation of parental investment in blue tits provide additional evidence for female-driven sexual selection of male crown UV coloration (Limbourg et al. 2004; Johnsen et al. 2005). These studies show that females decrease their reproductive investment in terms of nestling feeding or nest defence against predators (Johnsen et al. 2005) if the UV reflectance of their male is experimentally reduced. One of these studies shows that the decrease in female parental investment in response to male UV-reduction had a negative effect on the growth of the chicks (Limbourg et al. 2004) and may thus reduce male fitness. These results are predicted by the ‘differential allocation hypothesis’ (Burley 1988; for a review see Sheldon et al. 2000). According to this hypothesis a decrease in maternal investment in response to decreased male UV reflectance would be expected if broods of low UV males are of lower reproductive value to the female, for example because the young inherit the male’s low UV reflectance, making them less successful in (extra-pair) mate attraction when adults.

While there is growing evidence for a role of blue tit crown UV coloration in mate-choice, the results presented in this thesis clearly suggest that UV coloration is not important in male-male competition during the breeding season (Chapter 6; but see Alonso-Alvarez et al. 2004) or competition for food during winter (Chapter 7).

**Future perspectives**

*Adaptive sex ratio variation in birds*

The very recent small steps towards a revelation of the physiological mechanism
underlying primary sex ratio adjustment in birds are promising. First evidence sug-
gests that female birds are able to bias egg sex ratio before ovulation (Komdeur et al. 2002), either through biased segregation of sex-chromosomes (Correa et al. 2005), differential growth patterns of oocytes destined to become either male or female (Young & Badyaev 2006), or through atresia of ‘wrong’ sex oocytes (Pike 2005). The female’s endocrine system may play an important role in the regulation of these processes and it is even possible that steroid hormones deposited by the follicular wall cells into the developing oocyte influence the segregation of sex chromosomes during the first meiotic division, shortly before ovulation (Petrie et al. 2001; Correa et al. 2005; but see Pilz et al. 2005). Elucidation of the physiological basis for primary sex ratio adjustment would greatly facilitate new progress in the study of adaptive primary sex ratio variation in birds. Knowledge about the underly-
ing mechanism will help to define the costs and constraints that are involved in fac-
cultative primary sex ratio modification in birds, which would make it possible to develop more realistic models explaining and predicting avian sex ratio variation (Pen & Weissing 2002).

Although the number of case studies on avian sex ratio variation is quickly expanding, there remains an urgent need for experimental studies testing a priori predictions. Despite several calls for such experimental work (e.g. Sheldon 1998; Komdeur & Pen 2002), the majority of published sex ratio studies is still correla-
tional. In addition, it is extremely important that well-documented and influential case studies (e.g. Sheldon et al. 1999), are replicated to assess the generality of the patterns found. Another approach that will help to get a grip on the generality of adaptive sex ratio variation is the use of meta-analyses, especially since a large enough number of case studies has now been accumulated. The chromosomal sex determining system may not pose such a strong constraint on the evolution of adaptive primary sex ratio adjustment in birds as was previously thought (West & Sheldon 2002; Griffin et al. 2005). It would be a fruitful future approach to try to understand patterns of avian sex ratio adjustment based on the relative costs and benefits of sex ratio manipulation, together with the constraints.

**Blue tit crown coloration as a model in sexual selection research**

We are just starting to get a grip on the significance of the blue tit’s UV crown plumage as a sexually selected ornament. The basic patterns of sex, age, and season-
related variation in UV coloration are now well-described. Their significance in mate-choice and competition, as well as the inheritance of crown colour, need now to be further investigated in the several blue tit populations studied. There is no doubt that the blue tit will continue to serve as an excellent model species for many studies of sexual selection and sex allocation still to come.