Living on voles
– plastic life of the Ural owl

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Academic dissertation

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This thesis is based on the following articles, which are referred to in the text by their Roman numerals:


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# Contributions

The following table shows the major contributions of authors to the original articles or manuscripts:

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Summary

Abstract

Individuals face variable environmental conditions during their life. This may be due to migration, dispersion, environmental changes or, for example, annual variation in weather conditions. Genetic adaptation to a novel environment happens through natural selection. Phenotypic plasticity allows, however, a quick individual response to a new environment. Phenotypic plasticity may also be beneficial for individual if the environment is highly variable. For example, eggs are costly to produce. If the food conditions vary significantly between breeding seasons it is useful to be able to adjust the clutch and egg size according to the food abundance. In this thesis I use Ural owl–vole system to study phenotypic plasticity and natural selection using a number of reproduction related traits. The Ural owl (Strix uralensis) is a long-lived and sedentary species. The reproduction and survival of the Ural owl, in fact their whole life, is tied to the dramatically fluctuating vole densities. Ural owls do not cause vole cycles but they have to adjust their behaviour to the rather predictable population fluctuations of these small mammals. Earlier work with this system has shown that Ural owl laying date and clutch size are plastic in relation to vole abundance. Further, individual laying date–clutch size reaction norms have been shown to vary in the amount of plasticity. My work extends the knowledge of natural selection and phenotypic plasticity in traits related to reproduction. I show that egg size, timing of the onset of incubation and nest defense aggressiveness are plastic traits with fitness consequences for the Ural owl. Although egg size is in general thought to be a fixed characteristic of an individual, this highly heritable trait in the Ural owl is also remarkably plastic in relation to the changes in vole numbers, Ural owls are laying the largest eggs when their prey is most abundant. Timing of the onset of incubation is an individual-specific property and plastic in relation to clutch size. Timing of incubation is an important underlying cause for asynchronous hatching in birds. Asynchronous hatching is beneficial to offspring survival in Ural owl. Hence, timing of the onset of incubation may also be under natural selection. Ural owl females also adjust their nest defense aggressiveness according to the vole dynamics, being most aggressive in years when they produce the largest broods. Individual females show different levels of nest defense aggressiveness. Aggressiveness is positively correlated with the phenotypic plasticity of aggressiveness. As elevated nest defense aggressiveness is selected for, it may promote the plasticity of aggressive nest defense behaviour. All the studied traits are repeatable or heritable on individual level, and their expression is either directly or indirectly sensitive to changes in vole numbers. My work considers a number of important fitness-related traits showing phenotypic plasticity in all of them. Further, in two chapters I show that there is individual variation in the amount of plasticity exhibited. These findings on plasticity in reproduction related traits suggest that variable environments indeed promote plasticity.
1. Introduction

Natural selection is considered to act on phenotypic variation between individuals favoring those individuals who are best adapted to the local environment. Through the process we name natural selection the best fitting individuals will then produce most of the offspring surviving to reproduce. These individuals will contribute most copies of their genes to future generations. This chain of events potentially leads to evolution increasing the frequency of the selected genes in the population. In the process of natural selection the environment plays an important part as setting requirements and limits for the selected phenotypic expression (Southwood 1977). For evolution to occur through natural selection, phenotypic variation has to be heritable because, by definition, evolution is a change in population allele frequencies. From a quantitative genetics perspective only selection on heritable phenotypic variation is important for adaptive evolution (Fisher 1930, Schlichting & Pigliucci 1998). Thus, it can be, and often has been, viewed that genes are the driving force with phenotypes following their lead (Dawkins 1976). Consequently, selection on non-heritable phenotypic variation has long been considered unimportant for adaptive evolution (e.g. Williams 1966). Environmentally induced phenotypic variation has often been considered a nuisance rather than possibly important factor shaping evolution (e.g. Schlichting & Pigliucci 1998). This has led some to think that phenotypic plasticity would act to slow down the effects of natural selection on genes (Stearns 1982, Levin 1988). More recently, phenotypic plasticity has been considered to be important mechanism facilitating species adaptation to changing environments (Stearns 1989, Scheiner 1993, Schlichting & Pigliucci 1998, Räsänen & Kruuk 2007, Charmantier et al. 2008, Gienapp 2008).

1.1. Environment, selection and phenotypic expression

A phenotype is the product of a developmental program of an individual which is coded by the underlying genotype and affected by the environment (Scheiner 1993, Fig. 1). Environment is what an individual experiences during its development and lifetime (Southwood 1977). For example, in mammals the first environment experienced is the mother’s womb. Maternal effects (nutritional condition, health etc.) affect the development of the unborn. Later on the environment the individual experiences while growing to maturity (e.g. parental care, competition with siblings, conspecifics, and physical environment) affects directly the outcome of developmental process, the phenotype. Thus, the direction of phenotypic expression can be affected in multiple stages throughout the developmental processes and life history decisions of individuals (Scheiner 1993, Schlichting & Pigliucci 1998, Roff 2002). The change in the outcome (= phenotype) of the interaction of the environment and the developmental procedures is called phenotypic plasticity (Stearns 1989, Scheiner 1993, Schlichting & Pigliucci 1998, Pigliucci & Murren 2003).

On top of the environmental and genetic effects development (production of a phenotype) can be affected by random processes. In other words identical genotypes sharing the environment can produce different phenotypes...
Scheiner (1993). Environmental variation also affects evolution (and thus indirectly has an effect on the future phenotypes) by directing the process of natural selection (Fig. 1). A classic example of this comes from Galapagos Islands. In a widely cited paper Boag & Grant (1981) show how changing selection pressures acting on survival affect the morphological expression of bill size in a population of Darwin’s finches (*Geospiza fortis*) through changing environment, namely food quality and quantity.

1.2. The importance of phenotypic plasticity

The ability of a genotype to produce variable phenotypes in response to changing environment is called phenotypic plasticity (Stearns 1989, Schlichting & Pigliucci 1998). Phenotypic plasticity has received a lot of attention among biologists, and it has been widely studied from laboratory experiments to wild populations, and from plants to invertebrates to and vertebrates (e.g. Stearns 1989, Schlichting & Pigliucci 1998, Roff 2002, and references therein). As a potential example of interest for also non-scientific society serves the recent outburst of studies considering species adaptation to climate change (e.g. Brommer et al. 2005, Nussey et al. 2005a, Reed et al. 2006, Charmantier et al. 2008, Gienapp 2008). The change in spring temperature has been shown to be tracked by the plastic responses in the timing of breeding of wild birds (Charmantier et al. 2008). This is but one example of the importance of phenotypic plasticity for the species in responding to a changing environment. In a similar way phenotypic plasticity during the development or in the expression of behavioral and physiological characteristics can also be a means for a species to take a foothold in new environments (Schlichting & Pigliucci 1998).

Species living in cyclic environments provide a different angle for the study of phenotypic plasticity. Cyclic changes in environment are often coupled with variable fitness expectations for a breeding animal. The reproductive value of an offspring may vary a lot from one season to another, as well as within reproductive season (e.g. Brommer et al. 2002b). Also the survival prospects of an individual are bound to vary according to the environment. Environmentally induced variation in the direction and strength of natural se-
lection favors different phenotypes in different seasons. Thus, variable environment may act to promote phenotypic plasticity in labile traits. Studies on species facing such environments in the wild are scarce (Brommer et al. 2003, Nussey et al. 2005a, b, Brommer et al. 2005), and quite little is currently known about the fitness consequences of phenotypic plasticity in the wild. Some studies provide population or species level evidence of the adaptive value of phenotypic plasticity in the face of environmental change (e.g. Charmantier et al. 2008) and rare studies have been able to show that phenotypic plasticity of a trait is in fact heritable and under selection (e.g. Nussey et al. 2005c) but such analyses require detailed long-term studies of individuals. Yet, such studies are fundamentally important for understanding phenotypic plasticity because they provide information on individual performance over a range of environments, and have the potential to explore the fitness consequences of phenotypic plasticity on individual level.

1.3. Phenotypic plasticity in maternal effects

The environment experienced during the development in early life may affect individual performance later (Sinervo & McEdward 1988, Scheiner 1993, Lindström 1999). Maternal effects are defined as the effects of mother’s phenotype (phenotypic and genetic) on offspring phenotype (Mousseau & Fox 1998). In animals these effects include such factors as timing of reproduction, number of offspring, nutritional and immunological resource allocation during gestation/egg development, and behavioral decisions during the dependency period of offspring, such as food provisioning and allocation or offspring defense. Maternal effects have been shown to have profound effects on offspring phenotype. Plasticity in maternal effects is likely to be adaptive, because by adjusting maternal effects the offspring phenotype can be primed to best meet the environmental requirements (Räsänen & Kruuk 2007).

The influence of environmental variance on maternal effects of timing of reproduction and offspring size are particularly well established in a wide variety of study systems (Lindström 1999, Charmantier et al.2008). However, our knowledge of the long-term consequences of such maternal effects as egg size, immunological transfer, incubation behaviour and maternal care are not ubiquitous. The plastic responses of maternal effects have potentially positive, negative or no effects on offspring phenotypes. Yet, maternal effects provide possibly one of the strongest cases of adaptive plasticity, as environmentally induced maternal effects often increase the offspring success in a variable environment (Mousseau & Fox 1998, Räsänen & Kruuk 2007).

1.4. Measuring phenotypic plasticity

"Reaction norm is a mirror that reflects environmental effects into phenotypes” (Stearns 1989), although it is not necessary to constrain the use of reaction norms to describe only phenotypic variation and plasticity as a function of environment (e.g. Schlichting & Pigliucci 1998, Roff 2002). Reaction norms can be used to describe individual (phenotype) – environment (I × E) or genotype – environment (G × E) interactions (Brommer et al. 2005, Nussey et
Individual reaction norms depicting for example phenotypic plasticity (I × E) can be viewed as independent traits. The shape of the reaction norm is not restricted to linear form, although most of the available studies on phenotypic plasticity use linear reaction norms to describe I × E relationships (Nussey et al. 2007).

Linear reaction norms can be modelled as random effects using a random regression linear mixed model (RR LMM, Nussey et al 2007). A linear reaction norm is defined by two terms: elevation and slope. The elevation term describes the individual phenotype in an average environment when the environmental variable is mean-centred. The slope term describes individual phenotypic response to the environmental gradient (Stearns 1989, Scheiner 1993, Schlichting & Pigliucci 1998, Roff 2002, Nussey et al. 2007 Fig. 2). In principal, a significant elevation term of a RR indicates the existence of consistent differences across individuals, and in the absence of individual variation in plasticity (i.e. a significant slope term of a RR), these estimates are consistent over the whole environmental gradient. The presence of individual variation in plasticity affects the generality of the elevation estimates of a trait and only the elevation in an average environment should then be considered (Dingemanse et al. 2009). Further, natural data often consists of individuals which have not been observed over the whole environmental gradient. This is likely to affect the elevation and slope estimates of a RR model (Dingemanse et al. 2009). The slope term estimates the amount of variance explained by differences among individual slopes of the trait–environment reaction norm after correcting for the mean slope of the population. A significant proportion of variance explained by the slope term indicates that individuals differ in the amount of plasticity they show in a response to changes in the environmental factor. A non-significant slope term means that all individuals have (statistically speaking) uniform responses (Nussey et al. 2007). It should be remembered that incomplete sampling of individuals over the environmental gradient may lead to this result as well (Dingemanse et al. 2009). The RR LMM method of measuring individual reaction norms is illustrated in Fig. 2 from chapter IV. Each line in Fig. 2 depicts an individual reaction norm of Ural owl female nest defense aggressiveness against the change in vole abundance. These reaction norms are linear approximations of I × E relationship.
1.5. Aim of the thesis

The Ural owl–voles study system is near to ideal to study plasticity of traits related to reproduction in a natural population. As a long-lived and site-tenacious species individual Ural owls face a wide range of environments they have to cope with. The work with this study population was started already in 1977 by Hannu Pietiäinen and Heikki Kolunen. Thanks to consistent work from the very beginning, the collected data make longitudinal analyses possible. The common factor in the chapters of my thesis, and also the connection to the previous work with Ural owls, is the phenotypic plasticity which is promoted by the variable environment generated by the generally predictably fluctuating vole dynamics. The essence of longitudinal studies lies in the individual level analyses. The chapters I, III and IV take the advantage of the long-term data sets in analyses of different aspects of reproduction on an individual level, while in chapter II I experimentally study how food-limitation affects maternal effects under variable natural food conditions. The aims of the first chapter were to study how fluctuating environment affects the phenotypic plasticity and quantitative genetics of egg size. The Ural owl–voles system allows new insights on this widely studied trait (Christians 2002) because of the fluctuating environment. Further, I wanted to link egg size to individual fitness estimates. The second chapter studies food-limitation of maternal effects (I concentrate mainly on egg size in this thesis) through experimental manipulation of food conditions prior to egg-laying. This 3-year study was designed to study food-limitation of maternal effects (i.e. egg size, immunity transfer) in all vole cycle phases. The careful monitoring of individual offspring from egg to fledging allowed evaluation of the possible effects of egg size to fledging condition. The aim of chapter III was twofold. Firstly, I studied the timing of the onset of incubation (a causal reason for asynchronous hatching) on individual level. Secondly, I studied the fitness consequences of hatching asynchrony to the survival of Ural owl nestling. In chapter IV I studied plasticity and fitness consequences of aggressive nest defense behaviour in Ural owl females on individual level. In this thesis I have studied phenotypic variation and plasticity of a number of traits related to reproduction in a naturally fluctuating environment. Further, I link my findings to natural selection in order to assess the relevance of the results in an evolutionary context.

2. Species and Methodology

2.1. Ural owl – a top predator tied to voles

The Ural owl is a monogamous and site-tenacious bird of prey (4% move > 5 km; Saurola 1987). The Ural owl is long-lived, with an average age at first breeding of 2.9 years and an average breeding lifespan of 3.3 years (Brommer et al. 1998). The oldest females may reach an age of > 20 years. The main prey of Ural owls consist of field voles (Microtus agrestis) and bank voles (Myodes glareolus; Lundberg 1981, own obs.). During the nestling and fledgling periods also water voles (Arvicola terrestris) and migratory birds, especially thrushes have an important role as prey items (Karell et al. 2009, own obs).
Fennoscandian Ural owls live and breed in a variable environment (Lundberg 1981, Pietiäinen 1989, Brommer et al. 2002a). Field and bank vole dynamics undergo a rather regular three-year cycle with distinct phases: low, increase and decrease (Sundell et al. 2004, Brommer et al. 2009, Fig. 3). These vole population dynamics determine to a large extent Ural owl reproductive success and survival (Lundberg 1981, Pietiäinen 1989, Brommer et al. 2002a). Vole abundance can vary up to 50-fold between the lowest and the highest numbers. Many individuals therefore experience very different environmental conditions during their breeding career.

A low phase is characterized by low vole abundance in the preceding autumn with a continuous depression in vole numbers over the winter and breeding season. Very few Ural owls (10–20%) decide to breed in low phases, and those which breed are usually more experienced females (Brommer et al. 2002a). After a low phase vole numbers start to increase during summer and autumn, and stay relatively stable over the winter (Fig. 3). Increase phase is then followed by decrease phase characterized by peaking autumn vole numbers which crash practically to zero during the nestling or fledgling period. Up to 85% of active pairs breed during the increase and decrease phases (Brommer et al. 2002a).

Ural owl laying date is tied to the vole dynamics (Pietiäinen 1989, Brommer 2002a). The earliest breeders generally lay the largest clutches (Pietiäinen 1989), although the individual clutch size–laying date trends vary (Brommer et al. 2003). Ural owls usually start laying eggs in March–April, all time median laying date being 31 March (Brommer et al. 2002a). Median laying date varies from 9 March (decrease phase) to 19 April (low phase, Brommer et al. 2002a and later observations). Ural owls may lay between one to eight eggs, smallest clutches being laid in the low phase and largest clutches in the decrease phase. Eggs are laid on average in two day intervals and they start to hatch after ca. 32 days of incubation (Pietiäinen 1989). The recruitment probability for Ural owl young is highest in the increase phase when offspring have three times higher chances to be recruited than in a decrease phase (Brommer et al. 1998). Ural owl reproductive life span is affected by the age at first breeding, which in turn is affected by the phase of the vole cycle an individual was born in (Brommer et al. 1998).
Ural owl sex roles are very distinct, and typical for birds of prey (Newton 1979, Cramp 1985). Ural owl males feed females prior to breeding (Lundberg 1980). During the courtship feeding female Ural owls gather energy resources for laying and incubating the clutch, and they may gain several hundred grams of weight (Pietiäinen & Kolunen 1993, own obs.). After the onset of laying, the female stays in the nest while the male provides her with food. Later on males also provide food for the nestlings and fledglings. Male Ural owls rarely take part in active nest defence but they are often observed hooting when approaching the nest box (own observations). Females incubate the eggs and feed the young after hatching. Females will stay brooding in the nest for at least two weeks after the hatching of the first chick. After the nestlings grow older females move outside to guard the nest and later on the fledglings. Ural owl young are flightless when they fledge approximately at the age of four weeks. At that stage they are still flightless and totally dependent on their parents for care and food. They become independent after about three months after fledging.

2.2. General methodology of data collection

Study population
Ural owls have been studied in our study population since 1977. The study area is situated in southern Finland (Päijät-Häme) in an area of about 1500 km$^2$. Ural owls breed in nest boxes, which are 3–4 km apart from each other. There are about 180 nest boxes in the study area (the accurate number slightly varies across years). Study area consists of a mosaic of managed forests, clear cuts, agricultural landscape, lakes and human settlement. For more details of the study area see Pietiäinen (1989).

Basic field protocol
Ural owl females were caught during the laying/incubation/nestling period from the nest box using a hand-net that was placed in front of the nest box opening. Handling has not caused any obvious stress to the females and nest abandonments due to handling have been extremely rare in this species, although some protocols have required intense handling of the birds. All the caught females have been ringed as nestlings or at the first capture as adults. This enables lifelong individual recognition.

When caught, females were weighed with a Pesola spring balance (accuracy 5 g) and their arm (radius-ulna) length was measured with a ruler to the nearest 1 mm. All eggs have been consistently individually numbered with a pencil to establish laying and hatching order during the visits to clutches. The laying interval of Ural owls is approximately 2 days which enables accurate determination of the laying sequence of most eggs. Ural owl eggs have white egg shell. In cases when more than one egg has been laid between consecutive visits to a clutch the laying order can be deduced on the basis of darkening of the egg shell caused by fine particles in the sawdust getting attached to it. The presumed laying order has been later checked and confirmed at hatching. Egg length and breadth have been measured with a calliper with the accuracy of 0.05 mm. Egg volume has been obtained from a species-specific formula (Pietiäinen et al. 1986). Unhatched eggs remain in the nest, and could thus be
identified easily. In 1983, 1986 and 2000–2006 some nests were visited regularly around hatching with two to three day intervals in order to assess the hatching order and hatching spans, and match the eggs with the chicks (I, II, III).

**Vole census data**
Vole abundance in the study area was estimated by snap-trapping voles biannually in late September/early October and in early June since autumn 1986. The small-square method (Myllymäki et al. 1971) where traps (n = 300) baited with rye bread were set out for two consecutive nights in 25 quadrates (15 m × 15 m) at three localities (8, 8, and 9 quadrates per locality) which represented the main components in the landscape (variously aged re-plantations and mature forest) was used to estimate the vole abundance. Three traps were set in every corner of these quadrates approximately 1 m apart from each other. Traps were checked and re-set after the first night.

**Pre-laying feeding experiment (II)**
A pre-laying feeding experiment was carried out during 2004–2006 (II). The aim of this experiment was to study the role of food-limitation on maternal effects on egg size and immunity. We delivered extra food straight to nest boxes prior to feeding (fed group), while other nest boxes were only visited (control group). This protocol imitates the natural behaviour of Ural owls, as in birds of prey males courtship feed females prior to laying (Newton 1979, Meijer et al. 1990). Beginning in mid-February each year, Ural owl territories that had been active in past years were included in the protocol. The protocol consisted of delivering dead rooster chicks every fifth day to nest boxes of the fed group. Approximately 500g of rooster chicks were delivered on each visit. During the visits all uneaten chicken from the previous visit were removed and replaced with fresh chicken. Feeding was kept going until the female started laying or the territory was determined unoccupied.

**Hatching asynchrony and timing of the onset of incubation (III)**
In 1983, 1986 and 2000–2006, the Ural owl nests were visited regularly around the time of laying and the expected time of hatching in order to establish from which eggs individual hatchlings came. Hatching takes about 2 days from the appearance of the first cracks on the egg shell. This allows estimating the hatching time at least with the accuracy of one day. This accurate data on hatching was used to estimate the timing of the onset of incubation and realized hatching asynchrony. Hatchlings were individually marked by dyeing their head with a felt-tip marker pen. This made individual recognising possible also later and allowed me to study the effects of asynchronous hatching on nestling survival on individual level.

**Nest defense aggressiveness study (IV)**
Nest defense aggressiveness of Ural owl females was studied during visits to nest boxes to ring the chicks from 1983 to 2006 (excluding 1984 and 1985). Ural owls are medium-sized birds of prey which are known to be very aggressive nest defenders (Cramp 1985). Ural owl females are larger (c. 1,000g) than males (c. 700g), and the sexes have distinctly different roles in reproduction. Males do most of the hunting to provide food for the female and the off-
spring. Females incubate, feed, brood, and guard the offspring (Cramp 1985). A Ural owl female may take considerable risks while defending her brood. Aggressive females will hit a human intruder with force, and a female may occasionally die because of the impact of the blow (Saurola 1987, personal observation).

Statistics

This thesis is based on one experimental study (II) and analyses of long-term data (chapters I, III, IV). Analyzing longitudinal data with repeated measures combined with variables with sometimes non-normal distributions involves rather complicated modeling. My approaches have been linear mixed models (LMM) and generalized linear mixed models (GLMM) which have been used according to the requirements of data in all the chapters (Pinheiro & Bates 2000, Crawley 2002, Bolker et al. 2009). Details of the statistical methodology can be found in the referred chapters. I have conducted my analyses using R statistical software (R Core Team 2007). In chapter I the heritability analysis was performed using ASReml software (VSN international).

The phase of the vole cycle vs. vole abundance – reasons for methodological choices

An important characteristic of this system is that Ural owls are dependent on the voles but the cycles are driven, not by Ural owls, but by the predator community (e.g. Hanski et al. 1991, Korpimäki et al. 2004). Many individual Ural owls skip breeding every now and then. This skipping coincides most of the time with the low vole phases after decrease phases (Pietiäinen 1989, Brommer et al. 2002a). The survival of adults and young after decrease phases is lowered significantly (Brommer et al. 2002a) and the surviving individuals may be too weak to even attempt breeding. However, to skip breeding every now and then may also serve as an important ‘self-maintenance break’ for Ural owls (Pietiäinen et al. 1984) and the cyclic vole dynamics with distinct phases provide reliable cues for the timing of this break. The most striking of these cues is the near disappearance of the voles. A natural ‘break-year’ is followed by the most profitable (in terms of recruitment probability) breeding year in the increase phase (Brommer et al. 1998, 2002a). Thus, in increase phases Ural owls are ‘all charged up’ for breeding. Another important characteristic of increase phases is the presumably high vole abundance throughout the following winter. This is especially important for overwinter survival and recruitment of the young (Brommer et al. 2002a).

In the introduction and in chapter IV I advocate the reaction norm approach to study I×E interactions (phenotypic plasticity) according to Nussey et al. (2007) by using a continuous environmental variable (changes in vole abundance in chapter IV). In chapters I and II, on the other hand, I study plasticity in egg size using a rough generalization of vole dynamics, the vole phases. It is clear from Fig. 3 that cycle phases are not alike. Thus, some residual noise will be present by the use of this classification. However, as explained above, the phase-definition contains some important biology. Hence, the choice of method is balancing between the biology, the tools and the data available. There are reasons for why I have made the choices of using different analytical methods in different chapters. First, the vole trappings started only in the autumn 1986 whereas I had in my use owl breeding data (laying
date, clutch size, egg size etc.) from 1977 onwards. Second, cyclic vole dynamics are an inherent and important part of the study system. Thus, it was intuitive to me to use the vole cycle phases in the analyses of the first chapter. In chapter IV, on the other hand, I used a change in vole abundance from autumn to spring as a rough generalization of the dynamics. This method allowed me to study plasticity using the reaction norm approach (Nussey et al. 2007), and did not force me to exclude data points from the years of irregular vole dynamics (1999–2003) which were excluded from chapter I. The measure of environment in chapter IV was then a compromise between the data available and the methodology.

3. Results & Discussion

3.1. Ural owl egg size – plasticity, food limitation and selection

Egg size is thought to be an evolutionarily important trait because the early environment (quality and quantity of resources) may have profound consequences on offspring development (Sinervo & McEdward 1988, Mousseau & Fox 1998). Egg size is an offspring trait as it is bound to affect the growth and development (Williams 1994, Mousseau & Fox 1998, Christians 2002), but it is also simultaneously a maternal trait as it both reflects the female’s genotype and maternal allocation decision (Williams 1994, Christians 2002, I, II). In birds, egg size has been found to be highly repeatable and generally a fixed character (Christians 2002). The studies on the importance of egg size to offspring quality have produced controversial results, some presenting positive effects on fledging condition of maternal allocation of nutrients in eggs and others finding no effects (Sinervo & McEdward 1988, for birds see the reviews of Williams 1994 and Christians 2002, and references therein). Perhaps it is because of the above mentioned reasons that plasticity of egg size has received little attention in birds (although see Pietiäinen et al. 1986, Hakkarainen & Korpimäki 1994a, Valkama et al. 2002). Maternal adjustment of egg size has been shown to have profound consequences on offspring phenotype and subsequently to offspring fitness in other taxa (Sinervo & McEdward 1988, Sinervo 1990). Mothers have been suggested to be able to adjust the egg size in relation to the environment for maximizing their fitness over variable environments (Sinervo 1990). In chapters I and II I studied the plasticity, food limitation and fitness consequences of egg size from both an offspring perspective and from a maternal perspective. In chapter II I also study maternal transfer of immunity to offspring via eggs.

Ural owl egg size and the vole dynamics

Due to the cyclic vole dynamics the resources available to reproduction vary from year to year. This variation in food abundance is reflected, besides clutch size and laying date (Pietiäinen 1989, Brommer et al. 2002a), also in the egg size (I, II). Approximately 60% of the variation in Ural owl egg size is heritable (I). This is similar to heritability estimates of egg size in birds in general (Christians 2002). I show with the Ural owl that, despite the high heritability, a significant proportion of variation in egg size (> 22%) is due to intra-indi-
individual adjustments (i.e. plasticity) of egg size to environmental conditions, namely vole abundance (I, Fig. 4). Ural owl females lay the largest eggs in the decrease phase (I, Fig. 4), when the voles are most abundant both during the winter preceding reproduction, and presumably during the laying period. In general, there is plenty of food during decrease phases which enables laying of large clutches and eggs (Pietiäinen et al. 1986, Pietiäinen 1989, Brommer et al. 2002a, I). During increase phases the food conditions before reproduction are good but not nearly as good as during decrease phases (see Fig. 3 for illustration of vole dynamics). Egg size is a costly trait (Perrins 1996), as is also shown in chapter II. Costs of laying large eggs may be illustrated in the breeding attempts of increase phases (when the smallest eggs are laid, Fig. 4). It may be that Ural owl females are constrained from laying both large clutches and eggs in other but decrease phases. In low phases voles are scarce, during and before the laying, and only experienced individuals with, presumably, good mates and territories attempt breeding. Therefore clutch sizes are markedly smaller in low phases than in the two other phases (Pietiäinen 1989, Brommer et al. 2002a) and females lay small eggs (I).

**Egg size and contents: plastic and food-limited traits which potentially affect offspring quality**

In the pre-laying feeding experiment (II), we show that egg size and quality (concentration of maternal antibodies) is food-limited in all phases of a vole cycle. Females which were fed prior to laying were in better somatic condition and laid larger eggs with higher immunoglobulin concentration than females in the control group. Large eggs hatch with a higher probability than small ones (I). Increased egg size also has positive effects on fledging weight (3.8 grams/egg cm³, II) of the offspring. This may be important for future survival. In a closely related species, the Tawny owl (Strix aluco), offspring mortality is higher in lighter fledglings (Overskaug et al. 1999). Maternal priming of offspring immune system may be beneficial for further survival because during the first days after hatching nestlings cannot produce own immunoglobulins and are totally dependent on maternally derived immunity (e.g. Grindstaff et al. 2003). Thus, egg size and quality are costly (food-limited) characters potentially affecting offspring quality.
The hatchability of Ural owl eggs improves with increasing egg size (I), although the viability of eggs is relatively stable over a range of sizes corresponding for more than 95% of the laid eggs (Fig. 5A, B, I). Thus, laying smaller than maximum-sized eggs is probably not highly detrimental for the viability of the egg. The decision of laying larger eggs than necessary may be an illustration of food abundance. In chapter II egg size had profound positive consequences on offspring size at fledging. This may be important for performance later in life (Lindström 1999, Overskaug 1999). The position in a brood has a strong effect on the fledging weight of an individual nestling (Karell et al. 2009). However, in chapter III, I show that the fledging position in the brood does not affect the ultimate measure of success, the recruitment probability, of a young. Hence, potential long-lasting fitness benefits from fledging in prime condition remain so far unexplored and need to be addressed in future studies.

Other potential, but speculative benefits of hatching from a large egg may relate to the early survival in a brood but we find no evidence for that. Ural owl chicks hatching from large eggs may have energy reserves that last longer if not fed properly. Food-shortage during the early development may have irreversible consequences for offspring development (Arnold et al. 2007). Appleby et al. (1999) have shown that nutritional shortage during early development may affect parasite resistance in adulthood in Tawny owl. Ural owl young are most likely to experience food-shortage in the decrease phases when the largest clutches are laid if the voles crash early (Pietiäinen 1989, Brommer et al. 2002a). It may also be that in the increase phase, when the smallest eggs are laid, it pays off to invest resources to other reproductive decisions (clutch size) or somatic maintenance, as the food conditions remain stable over the breeding season and also after offspring fledge.
Canalizing selection on Ural owl egg size

Ural owl egg size is under canalizing selection both on the level of individual eggs and on the level of females (I). Extremely large and small eggs do not hatch as well as intermediate-sized ones (Fig. 5A, I). Whereas the negative fitness consequences of small eggs have been described previously (e.g. Perrins 1996), this is the first evidence that extremely large eggs have reduced hatchability (Fig. 5A, I). It is unknown why very large Ural owl eggs have a reduced hatchability, but our finding does suggest that there is a limit to the benefit of producing larger eggs on the level of the individual egg. Furthermore, females that lay the smallest and the largest eggs also show a reduced lifetime production of fledglings. The reduction in lifetime fledgling production is due to a reduced lifespan of these females, rather than reduced fecundity.

One striking aspect of the canalizing selection on Ural owl egg size is that only relatively few egg sizes are actually selected against (Fig. 5A) and that majority of the eggs laid are on a selectively neutral size range (Fig. 5B). Similar pattern is found in the selection on lifetime fledgling production on individual-level (I). Only a minority of females (laying extreme-sized eggs) is selected against while most of the females have more or less equal fitness. The females that are selected against have shorter life-spans, resulting to lower lifetime fledgling production (I). A fitness plateau that I have described on egg and individual levels probably facilitates the evolution of plasticity in egg size. When a clear single fitness optimum in egg size is lacking, individuals have many possibilities for making individual adjustments to their current environmental conditions without this leading to a reduction in fitness. The patterns of selection on Ural owl egg size that we have here described help to explain how a high heritability can be maintained without an expectation of evolution leading to an increasing egg size or a single optimal egg size. We show that variation in egg size can both be constrained by selection against extremely-sized eggs at both ends of the continuum, and be maintained by a broad plateau in fitness across a wide range of egg sizes.

3.2. Timing of the onset of incubation and hatching asynchrony in the Ural owl

In bird species which have a variable clutch size, the beginning of full incubation is thought to be linked to the determination the final clutch size (Meijer et al. 1990). However, the mechanisms of clutch size determination vary across different bird taxa (Haywood 1993). The larger the clutch laid, the longer the females usually delay the onset of incubation (Meijer et al. 1990, Valkama et al. 2002b, Fig. 6, I). The timing of the onset of incubation is also a highly relevant female decision in determining emerging hatching patterns – or hatching asynchrony (HA). Brood size/age hierarchy in asynchronously hatching species is to a large extent established through hatching order (e.g. Hahn 1981, II). The brood a chick hatches in is thus a part of the environment a chick experiences during its early development. Interactions in the brood potentially affect the nestling’s development and survival in future. For example, an intense food competition can lead to periods of malnutrition which can seriously affect nestling’s further development (Wiebe & Bortolotti
1994) and performance in later life (e.g. Arnold et al. 2007). A lot of hypotheses on the adaptive value of HA have been proposed (see e.g. Slagsvold 1986, Nilsson 1993). However, it has rarely been studied, how the timing of the onset of incubation varies across individuals (but see Wiebe et al. 1998a, Wang & Beissinger 2009).

Given that there is variation in nest attendance before the onset of full incubation (Wiebe et al. 1998a, Wang & Beissinger 2009), there are probably differences in the timing of the onset of incubation as well. If there is variation in the timing of the onset of incubation independent of the final clutch size, contrary to what was suggested by Meijer et al. (1990), it means that females can affect the realized hatching patterns, and thus the environment the offspring are experiencing. The aim of chapter III was first to study, whether such differences in the incubation behaviour exist in the Ural owl, and second, to explore the consequences of hatching asynchrony for the Ural owl nestlings.

**Ural owl females differ in the timing of the onset of incubation**

The hatching span is correlated with brood size. I found that, on average, the onset of incubation was delayed for 0.29 (± 0.07 SE) days per egg laid (Fig. 6, III). The female component explained 25% of the variation in the timing of the onset of incubation after the effect of clutch size was removed (III). Thus, the timing of the onset of incubation is an individual–specific property in the Ural owl. Timing of the onset of incubation is plastic in relation to clutch size. Ural owls begin incubating later when they produce larger clutches (Fig. 6, III). Unfortunately, the available data did not allow me to study whether the timing of the onset of incubation in relation to variable clutch sizes (i.e. incubation schedule–clutch size reaction norm) differs across individuals.

The pre-laying feeding experiment (II) revealed that in the Ural owl the timing of the onset of incubation is not affected by the extensive surplus feeding effort (III). I did not find any effects of vole phase, laying date or female age on the timing of the onset of incubation. Temperature during the laying sequence did not appear significant in the analyses. This may be due to the link
between the timing of the onset of incubation and clutch size, the latter being strongly affected by the vole dynamics (Pietiäinen 1989, Brommer et al. 2002a) and temperature (Lehikoinen et al. unpublished manuscript). Further, Meijer et al. (1990) suggest that timing of the onset of incubation is linked to the cessation of the ovarian development. Thus, clutch size, presumably a primary decision, affects the realized timing of the onset of incubation (Meijer et al. 1990, Valkama et al. 2002b, Fig. 6). There was annual variation in the timing of the onset of incubation because presumably years differ in quality in other terms than vole dynamics as well. The data of individual observations is also spread out over many years; naturally, in such a way that different cohorts have bred in different years. There may thus be individual–environment interactions which remain unnoticed due to the fact that not all the environments are shared by all individuals.

Hatching asynchrony – a pattern emerging from female decision and egg hatchability

The timing of the onset of incubation is in my mind fundamental for understanding the causes of hatching asynchrony (HA). Many theories of the adaptive nature of HA have been put forward (Slagsvoldt 1986, Nilsson 1993) but underlying mechanisms have rarely been studied even in a descriptive way (although see Wiebe et al. 1998a, Wang et Beissinger 2009). As illustrated in Fig. 7 the HA in Ural owl is primarily a result of the timing of the onset of incubation. However, the relatively large proportion of eggs that do not hatch (13%, I) affect the final HA significantly when compared to the fully hatched broods (Fig. 7, III). This phenomenon is not uncommon in birds. Spottiswoode & Möller (2004) compared 99 species of birds and found that across species on average 10% of the eggs laid do not hatch. Thus, it is likely that this is not just a species specific phenomenon but that it is actually a more general pattern across bird species.
Asynchronous hatching is favored in the Ural owl

I analyzed the effect of HA on the brood level mortality and the effects of HA on survival of individual offspring (III). In the brood level analysis I took the female perspective to offspring mortality (number of young dying) whereas the individual level approach looks at survival from an individual offspring perspective. The brood level analysis shows that increasing synchrony leads to more chicks dying, irrespective of brood size or year. This is in accordance with the results of Wiebe & Bortolotti 1994 (but see Wiehn et al. 2000 for Eurasian kestrel). Asynchronous hatching establishes a quite clear size hierarchy to a Ural owl brood. It has been suggested that asynchronous hatching would be preferable due to this fact (Hahn 1981). By creating clear hierarchy to a brood the female may cut down the need for siblings to further negotiate their rankings, thus lowering the energy expenditure and improving the chances in the brood. Asynchronous hatching is also spreading out the peak energy requirement periods of the nestlings (Hussel 1972), and in this way reducing the risk of starvation. The main reason for deaths during nestling period in Ural owl broods is starvation. I further found that laying date interacts with HA in such a way that late broods suffer from synchronous hatching more than early broods. Late breeders are in poorer condition (Pietiäinen & Kolunen 1993) and may occupy poorer territories. Synchronous hatching is detrimental to Ural owl nestlings, presumably, through energetic costs. It is intuitive that late broods end up paying greater costs from synchronous hatching than early broods.

The individual level analysis of offspring survival until fledging underlines the importance of the position in the brood hierarchy. HA did not explain the survival of individual offspring (marginally non-significant). In contrast, hatching position had a large effect on survival until fledging. Last hatched chicks in larger broods make it rarely out alive and may even be left to starve in the nest box if the older siblings fledge earlier. An interesting observation on offspring fitness is that recruitment probability of female offspring is not affected by the position in a brood as long as the young manage to fledge (III). Thus, laying large broods is not an entirely exaggerated outcome of good environmental conditions but may act against the high probability of eggs not hatching in the Ural owl clutches (I).

Evolutionary importance of the timing of the onset of incubation and hatching asynchrony

From maternal perspective it may be advantageous to commence incubation as early as possible. There are however a few possible constraints in this. The primary reproductive decision about clutch size may set limits to the timing of the onset of incubation through the physiological processes linking it to cessation of egg production (Meijer et al. 1990). Therefore, females may not be able to begin incubation too early in the egg-laying period because it would decrease their final clutch size. Female Ural owls may also consistently differ in the egg-laying interval. This is a dimension my current study could not explore. It has also been documented that last-laid eggs usually develop faster (e.g. Viñuela 1997) which has not been taken into account in my study. However, I show in Fig. 7 (and in III) that estimated timing of the onset of incubation correlates rather well with the exact observations of hatching asynchrony in clutches where all the eggs hatched. In the light of my findings I can con-
clude, that timing of the onset of incubation is a trait which may well be evolvable because there are consistent phenotypic differences across individuals. The evidence showing the benefits of asynchronous hatching to offspring indicate that there may be selection acting on this trait. In a more general perspective the findings of this study underline the importance of future exploration of hatching asynchrony from two distinct perspectives: the female perspective and the offspring perspective since these two are different phenomena.

3.3. Plasticity and selection on Ural owl nest defense aggressiveness

Animal behaviour was long considered ultimately plastic in such a way that individuals behave optimally in each situation (Sih et al. 2004). Later on increasing attention has been drawn to the study of individual animal behaviour. It has been shown that many behavioral traits are, in fact, relatively fixed within individuals (Sih et al. 2004, Réale et al. 2007), and that they have a genetic basis (Réale et al. 2000, van Oers et al. 2003, Sih et al. 2004, Dingemanse & Réale 2005, Bell 2005). Behavioral traits have also been shown to affect selection on morphological traits (Duckworth 2006a) and dispersal patterns (Duckworth & Badyaev 2007) as well as reproductive success (Dingemanse et al. 2004). Thus, behavioral traits may have multiple implications on micro-evolution. Behavioral or personality traits can be roughly categorized into a few main axes. Main categories where most of the observed behaviors can be explored are aggressive–non-aggressive, proactive–reactive, bold–shy (e.g. Sih et al. 2004). Apart from the genetical basis of behavioral traits (Réale et al. 2000, van Oers et al. 2003, Sih et al. 2004, Dingemanse & Réale 2005, Bell 2005), it has also been shown that environmental factors often have a profound effect on their expression (Bell 2005).

Many aspects of maternal behaviour prior to the independence of the offspring represent important maternal effects. Chapter III discusses the importance of the timing of the onset of incubation, an example of a behavioral maternal effect, in relation to hatching asynchrony and offspring fitness. Parental food allocation decisions during the nestling period are important, even vital, from the offspring perspective (e.g. Humphries et al. 2006). Similarly, potentially costly nest and offspring defense behaviors (Ricklefs 1969) are likely to be important (Wiklund 1990, IV). In chapter IV I studied the nest defense aggressiveness of Ural owls. Nest defense aggressiveness was quantified with a scale from 1 to 6, following the guidelines of Hakkarainen & Korpimäki (1994b). This behavioral trait links strongly to animal personality studies. The Ural owl is a notoriously aggressive nest defender (Cramp 1985, IV). Ural owl females sometimes literally place their lives and health at risk for protecting their offspring (own observations). Chapter IV is based on data on the nest defense aggressiveness of Ural owl females which has been gathered on a long and sometimes bloody path from the late seventies until recent days. In this chapter I investigate the nest defense behaviour on individual and population levels. My aim was to explore the causes for aggressive nest defense behaviour and further pin-point the consequences in respect to natural selection.
Nest defense aggressiveness affects recruitment success in the Ural owl

Ural owl females show pronounced nest defense aggressiveness when the recruitment probability of the young is at its highest (Fig 8, IV). Previous work with Ural owl has shown that recruitment of the young is tightly linked to the vole dynamics, increase phases being the most productive years (Brommer et al. 1998).

Similar observations have been made in another boreal forest owl species, Tengmalm’s owl (*Aegolius funereus*). Hakkarainen & Korpimäki (1994b) showed that Tengmalm’s owls defend their offspring most vigorously in years when offspring had the highest recruitment probability. Tengmalm’s owl young have the highest chances of recruiting in the increase phase of a vole cycle (Korpimäki & Lagerström 1988). However, a causal relationship between nest defense behaviour and recruitment has not been shown before. Analyzing causality with descriptive natural data contains some problems because often important reproductive traits, such as laying date and clutch size, correlate strongly with each other. In the Ural owl laying date and clutch size are also highly correlated, and to a large extent determined by vole dynamics (Pietiäinen 1989, Brommer et al. 2002a, b). I found that both of these traits, as well as the changing vole dynamics, also explain the observed nest defense aggressiveness (IV). I used a path model to tease apart the correlations and causalities of these traits and offspring recruitment (Fig. 9), as suggested by Mitchell (1992). In the model all the pre-hypothesized causal pathways from vole dynamics to recruitment are depicted by arrows. From this set of options our model suggests a causal path laying date–brood size–nest defense aggressiveness–recruitment (thick arrows). Hence, there is evidence that nest defense aggressiveness plays an important role in offspring recruitment, independently from the effects of its correlates, reproductive timing (laying date) and reproductive output (brood size).

Inter-individual variation in the elevation and plasticity of nest defense aggressiveness

Vole dynamics, laying date and brood size affect the way Ural owls defend their offspring. Intuitively, nest defense aggressiveness increases with in-
creasing brood size and decreases with advancing laying date. The same trend
with advancing laying date exists for the reproductive value of Ural owl off-
spring (Brommer et al. 2002b). Further, aggressiveness is plastic in relation to
the fluctuating vole population size. I estimated the change in vole population
size as a difference between previous autumn vole trappings, which are reli-
able predictors for the future clutch sizes and laying dates and spring vole
trappings (at the time when offspring have already fledged (Pietiäinen 1989,
Brommer et al. 2002a).

I found that 52% of the observed variation in nest defense aggressiveness
was related to the variation across individual Ural owl females. Hence, there
were consistent differences across individuals in their nest defense aggres-
siveness. Individual Ural owl females also differentially adjusted their nest
defense aggressiveness in response to over-winter changes in vole abundance.
Individual-specific adjustment of nest defense aggressiveness in re-
sponse to the over winter change in vole numbers explained 6.8% of the vari-
ance in aggressiveness (IV). Thus, there are differences in the plasticity of ag-
gressive nest defense behaviour. Moreover, aggressiveness and plasticity of
Ural owl females are positively correlated ($\tau = 0.45$, IV). Thus, individuals
which are most aggressive in the mean environment tend to be most plastic in
their nest defense behaviour as well.

**Natural selection and phenotypic plasticity on nest defense aggressiveness**

I show in chapter IV that, from year to year, more aggressive individuals are
favored by natural selection in terms of local recruit production. It may be that
the less aggressive females could produce offspring that disperse longer and
would not be recorded in our study population. However, running the same
analyses with data consisting of all the recruitment records from our study po-
population (c.a. twice the sample size) gives qualitatively similar results. On the
ultimate level, i.e. survival, there are no consistent costs from aggressive nest
defense behaviour. Thus, there is directional phenotypic selection for more

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**Fig. 9. Hypothe-
sized (arrows)
and estimated re-
relationships be-
tween overwinter
change in vole
density ($\Delta$-voles),
laying date,
brood size, ag-
gressiveness and
recruitment. Par-
tial regression co-
efficients from the
path model are
given with their
significance, * =
P<0.05, ** =
P<0.01, *** =
P<0.001.
aggressive individuals with no apparent opposing selective forces. Interestingly, more aggressive individuals tend also to be more plastic in their nest defense behaviour. Thus, phenotypic selection on the elevated nest defense aggressiveness may also promote the plasticity of nest defense aggressiveness among Ural owls. This would be intuitive, when considering the environment Ural owls are living in. Vole abundance and reproductive value of the offspring vary drastically across and within season (Brommer et al. 2002b). Although I did not find any cost of aggressiveness on the level of survival, maintaining elevated levels of aggressiveness may be costly through the hormonal control of this behavior (Silverin 1998). Aggressive individuals are also more prone to injuries when facing human intruders (own observation) or natural enemies.

Surprisingly many Ural owl females, however, showed a low amount of aggressiveness (IV). This provokes a relevant question about the evolution of nest defense aggressiveness in the Ural owl. Why are a large part of the females still non-aggressive if there is selection for aggressiveness, and there are no apparent costs involved in aggressiveness? There are a few open questions limiting our understanding of this process. Although the repeatability of nest defense aggressiveness is substantial (52%), confirming that there are real differences between individuals, I could not analyze the heritability of this trait. Heritability analysis requires, besides repeated observations of nest defense aggressiveness on individual level, also sufficient knowledge of the relatedness of the individual Ural owls (Kruuk 2004). Unfortunately, our data does not meet the last requirements. It thus remains unclear whether aggressive mothers really produce aggressive offspring. Another issue is the role of the male. Ural owls have distinct sex-roles (Cramp 1985). In the Ural owl males provide food for the young (Cramp 1985). Although male Ural owls in most of the cases do not participate in active nest defense there are some individuals exhibiting extreme aggressiveness against intruders. If there is a trade-off between nest defense and food-provisioning in male Ural owls, this would act as an opposing selective force for nest defense aggressiveness. There is evidence from other bird species that aggressive nest defense correlates negatively with brood provisioning in males (Duckworth 2006b).

Nest defense aggressiveness has likely been evolved against ground predators (silent dive and hit tactic). Ural owl nestlings fledge before they are fully developed. After fledging the offspring move around from tree to tree. However, at fledging they are unable to fly and cover the distances on ground. Further, they are dependent on their parents for another three months. The aggressiveness in Ural owl females is promoted during the fledgling period (Cramp 1985, own observation), and female Ural owls seem to discriminate quite strongly between the fledged young and nestlings (own observation). According to Sunde (2005) over 50% of the tawny owl (Strix aluco, a closely related species with similar ecology) fledgling mortality was caused during the first week after fledging. Thus, the post-fledging time is the most hazardous period for the offspring, and it may well be that the aggressiveness levels are in general lower during the nestling periods when our data has been gathered.
4. Conclusions

A variable environment is thought to promote phenotypic plasticity (Schlichting & Pigliucci 1998, Roff 2002). In the Ural owl–vole system, fluctuating vole dynamics are the source of drastic environmental variation causing offspring reproductive value and individual survival to vary across breeding seasons (Brommer 2002b). We have similar findings from other boreal bird species breeding in cyclic environments (Korpimäki & Lagerström 1988, Hakkarainen & Korpimäki 1994b) Further, Ural owls are long-lived and sedentary birds (Cramp 1985). Hence Ural owls have to adjust to the environmental variability they face during their life, and therefore one would expect that these birds show plasticity in their reproductive decisions.

Chapters I and II show how egg size, a trait generally thought to be fixed characteristic of an individual (Christians 2002), is in fact plastic in respect to vole dynamics. This is in accordance with earlier findings on avian egg size in cyclic food conditions (Hakkarainen & Korpimäki 1994a) Further, egg size is canalized and under stabilizing selection with positive effects on hatchability and fledging condition of the offspring. These results back up the earlier claims of egg size being important reproductive trait (e.g. Valkama et al. 2002a). In chapter III I study hatching asynchrony in Ural owls. I show that the underlying, and possibly evolvable, cause for asynchronous hatching in Ural owl is the timing of the onset of incubation. The novelty of my results compared to earlier work (e.g. Wiebe et al. 1998a, Valkama et al. 2002b) is that this is the first time to assign repeatability for timing of the onset of incubation. This trait is plastic in relation to clutch size, but not sensitive to environmental variables such as vole dynamics or temperature which is contrary to earlier findings on the subject (Wiebe & Bortolotti 1994, Wiebe et al. 1998b). The timing of the onset of incubation may be under selection because asynchronous hatching is beneficial for the survival of the Ural owl nestlings. Chapter IV explores the nature, causes and consequences of nest defense aggressiveness in the Ural owl. I show that individuals do not only differ in the level of nest defense aggressiveness but also in the level of plasticity shown. Aggressive behaviour is selected for because it best explains the recruitment of the young. Most aggressive and consequently most plastic individuals are selected for on the phenotypic level.

Throughout the chapters I have explored phenotypic variation and plasticity in relation to vole abundance fluctuations, and the evolutionary potential of the findings. Traits such as egg size or aggressiveness are costly to produce and annually varying food conditions require energetic adaptation. Further, clutch size is plastic in the Ural owl (Pietiäinen 1989, Brommer et al. 2002a) and it is likely to affect the realized incubation behaviour (Meijer et al. 1990), thus filling the gap between chapter III and vole dynamics. Vole population changes or vole cycles are the driving force of Ural owl life. The response, or adaptation, to this variable environmental quality is the phenotypic plasticity of practically any important reproductive trait, as predicted by textbook examples (Schlichting & Pigliucci 1998, Roff 2002).
Acknowledgements

I had a thought of just writing down the word ‘Kiitos’ in here. I have, however, read comments like ‘this needs to be explained in a more general way…’ just so many times in my manuscripts that I now show appreciation to this lesson from my supervisors. Thus, I will open up s bit the word ‘Kiitos’.

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