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Consumer Life History and Demography in Dynamic Environments

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Consumer Life History and Demography in Dynamic Environments

by

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Abstract

Individual demographic performance is the outcome of a complex interaction between a set of physiological rules predetermined at a genetic level, the individual, and a particular context, the environment. Both are highly dynamical entities. The environment varies in response to many deterministic and stochastic forces, and individuals respond to their environment as a function of their own internal conditions. My thesis combines empirical and theoretical works on consumer-resource systems in order to study how genetic and environmental factors interact to generate patterns of phenotypic expression and assess their effects on resulting ecological and evolutionary dynamics.

I first present methods for carrying out sensitivity analysis of dynamic energy budget models. These techniques are then used to study how the feedback loop between consumer and resource dynamics affects individual life-history and emerging demographical patterns. Thirdly, I present the results of large manipulative experiments on *Daphnia*-algae systems aiming at characterizing how the combined effect of genetic variation and environmental fluctuations affect the population demographic patterns and their underlying energetic basis. The genomic response of individuals was further characterized through microarray experiments. Finally, I integrate some of these results within a simplified model framework to draw general implications for the dynamics of ecological systems.

Sensitivity analysis reveals that including the effect of the environmental feedback drastically alters the predictions that are made on the patterns of expression in individual life history, and tends to buffer genotypic variation. Experimentations decouple this environmental feedback to reveal that the demography of the different clones is impacted in a different way by environmental food fluctuations. This result arises because the relation between individual energetics and food density is qualitatively affected by dynamic variation in the food environment. The genomic analysis supports these results and also emphasizes the

importance of consumer-resource interactions for the functioning of biological organisms, as up to 84% of the *Daphnia* genome was found to respond to variation in food conditions.

All the results of this thesis converge to highlight the fact that dynamical aspects in the environment significantly affect patterns of genetic expression at both the individual and population level.

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Chapter 1

Introduction

Elucidating the factors that contribute to the life history of a species is a central tenet of ecology and evolution. Life history theory seeks to explain how natural selection and other evolutionary forces shape organisms to optimize their survival and reproduction in the face of ecological challenges posed by the environment (Stearns 1992, 2000, Roff 2002, Reznick 2010). Individual life history is however far from being a constant property of individuals but is a highly dynamical feature that varies under the influence of the environment and the evolutionary process (Stearns 1992, Roff 2002, Beckerman et al. 2002, De Roos et al. 2003). Due to their direct link to the processes of birth and death, understanding the mechanisms that produces particular life history is of paramount importance for both evolutionary biology and ecology (Saccheri and Hanski 2006). Individual life history is indeed both the product of, and has consequences, on ecological and evolutionary processes.

The expression of individual life history has both a genetic and an environmental basis which can be highly variable and show variation among individuals within the same population, among populations, and over time. Dynamic variation at the individual level is underlain by a map from genotype to phenotype which is highly complex, and environmental variation may still complicate this picture further (Sclichting and Smith 2002, Abouheif et al. 2013). By contrast, ecological and evolutionary theories have been built by making simplifications on the mechanisms underlying the interactions between the organisms and their environments. In its most classical vein, ecological theory assumes strongly simplifying assumptions on individual life history and studies their ecological implications. Conversely, classical life history evolution theory assumes strongly simplifying assumptions on the ecological context and studies how life history strategies evolve. Both theories are now striving

to include more realism and to move beyond these simplifying assumptions. Moving over these assumptions often leads to the emergence of qualitatively new phenomena that could otherwise not be explained, which I will discuss in this chapter. In this context, a major goal of this thesis is to clarify how these dynamic aspects of life-history processes affect and are affected by ecological and evolutionary processes.

My research uses one of the rare systems where we have sufficient knowledge about the biology of the consumer and their food supply, to study in details evolutionary processes and ecological dynamics - the *Daphnia*-algae system. *Daphnia* are common herbivores that feed on algae in lakes and ponds throughout the world, and are keystone species mediating the dynamics and stability of many freshwater systems (Lampert 2011). *Daphnia* populations are composed of clones that reproduce asexually with rare bouts of sexual recombination, making it ideal to control for genetic factors. Females usually grow by discrete increment by moulting every 2-3 and may release a brood of eggs at that time. These eggs can be easily counted while females carry them in their brood pouch as their carapaces are translucent. Development, fecundity and mortality are food-dependent, and their energetic basis has been thouroughly studied (McCauley et al. 1990, Nisbet et al. 2004). These results have been used to produce unique insights on the processes driving the dynamics of their populations (e.g. McCauley and Murdoch 1987, McCauley et al. 1999, 2008). The *Daphnia* genome as been sequenced and is being thoroughly studied as well (Colbourne et al. 2011, Miner et al. 2012). Combining this wealth of knowledge with the ease with which *Daphnia* populations can be cultured and manipulated in laboratory makes it one of the best models for integrating processes over biological scales and test the questions raised in this thesis.

In this introductory chapter, I start discussing the mechanisms at the interplay between individual life history and ecological processes and their implications. I then discuss the evolution of these life histories. Along this discussion, I highlight some of the current issues in our understanding of these processes that motivated my Ph.D. research.

1.1 State-dependent life histories and physiologically structured population dynamics

In purely age-dependent life-history theories, the life history expressed by individuals is fixed and does not depend on the conditions they experience (McNamara and Houston 1996). In most circumstances however, life history processes have a strong dependency on states, which may relate to both individual physiological states and environmental states. The primary consequence is that individuals experiencing different environmental histories may express very different life histories (De Roos et al. 2003). As individual demographic rates and interaction rates with other species are a direct product of the life history expressed by individuals, a number of consequences ensue for population and community processes that I will discuss here.

State-dependent life histories generate inter-individual heterogeneity that has consequences for population and community processes, as individuals in different states do not contribute to the same extent to the demographic process, and for interactions with other species. Structured population dynamics theory provides a quantitative link between the expression of individual life history and its consequences for the demographic process (Diekmann and Metz 2010). A key ingredient in this link is a model relating how the individual response to its environment generates a distribution of individuals at the population level, and how this distribution affects ecological interactions.

In this context, two key aspects of state-dependent life histories have been the main focus of ecological research: the dependence on individual size or stage; and the dependence on resource conditions. Ontogenetic development often leads organisms to show pronounced changes in size during their lifetime, which is associated with changes in ecological performances (De Roos and Persson 2013). This may include variation in the type and strength of interaction with other species, and life history processes, such as development, reproduction and mortality rates. Similarly, resource-dependence affects ecological performances. The

expression of life history traits is limited because organisms have a limited quantity of resources available (e.g. energy, time) to allocate among competing functions (e.g. growth, reproduction, survival) (Van Noordwik and De Jong 1986, Roff 2002). Thus, the phenotypic expression of a particular genotype associated with life history should ultimately depend on the environmental context experienced, and how these resources have been allocated among the competing functions.

In many, if not most circumstances, the energy acquired through feeding is a limiting resource, and also governs how an individual will grow during its lifetime. Energy budgets provide a mechanistic framework for relating individual feeding history with their life history (Kooijman 2010). They explicitly account for the mechanisms of energy acquisition, allocation and expenditure underlying individual trajectories of development, reproduction and mortality. The cornerstone of the approach is that these processes have a functional relationship on both the food environment (and possibly other environmental factors) and individual states. Given the variety of life histories found in natural systems, theory has strived to identify whether general features of population and community processes could be associated with general features of individual life history (Murdoch et al. 2003). Most dynamic novelties arising from structured population dynamics can be linked to the facts that state-dependent life histories may: (i) generate competitive asymmetry in individuals among the population: different individuals may respond very differently to common environmental conditions. (ii) Turn the expression of individual life history into a density-dependent process: population processes impact some aspects of the environment, which in turn affects the expression of individual life history (De Roos et al. 2003). This may be understood in the context of consumer-resource systems. Most aspects of consumer development, reproduction and mortality are food-dependent. Dynamics in the food environment are in turn largely mediated by consumer foraging activity. The effect of this ecological feedback on individual life history are however only assessable within a framework that accounts for the processes

underlying the expression of individual life history in a mechanistic way, in contrast to more classic studies that only incorporate life history effects in a phenomenological way (see e.g. Caswell 2001, Beckerman et al. 2002).

The effect of competitive asymmetry has been most studied in the context of consumer-resource interactions (De Roos and Persson 2013). In these systems, competitive asymmetry is related to the ability of individuals to produce biomass, which can be quantified as the mass-specific rates of biomass production as a function of individual states. For example, to illustrate the population implications of competitive asymmetry, we can consider its consequences under equilibrium conditions. At equilibrium, the net rate of biomass production in the population is, by definition, zero. Since individuals are not equivalent in the population, this implies that some individuals will be net biomass producers whereas others will be net biomass losers depending on their states. Certain classes of individuals may thus form a bottleneck for recruitment and population growth, whereas other stages may not be limited at all or to a limited extent (De Roos and Persson 2001). If we consider, for example, competitive asymmetry between juveniles and adults, population growth is limited by low adult fecundity when juveniles are the superior competitors, whereas in the opposite case, population growth is limited by the maturation from juvenile to adult stages. In term of individual life history, when juveniles are superior competitors, juvenile growth will be fast and survival high, whereas adult will have reduced lifespan and fecundity. The opposite is true when adults are superior competitors.

De Roos and Persson (2013) reviewed the novel phenomena associated with state-dependent life histories for the structure and dynamics of populations and communities. In particular, accounting for ontogenetic asymmetry and the ecological feedback on life histories provides explanations for the emergence of various kinds of population cycles (De Roos and Persson 2003, Murdoch et al. 2002, Claessen et al. 2000, 2002), coexisting dynamical attractors (McCauley et al. 1999, 2008), phenomena of biomass overcompensation, in which an increase

in mortality rates causes an unexpected increase in total population biomass (De Roos et al. 2007, Ohlberger et al. 2011, Schröder et al. 2009, Nilsonn et al. 2010), emergent Allee effects (Persson et al. 2007), emergent facilitation (Cameron and Benton 2004, De Roos et al. 2008), and other implications for community structure and dynamics, including coexistence mechanisms.

As my thesis focuses on the dynamical implications of state-dependent life histories, I will provide more details on the mechanisms underlying population dynamical processes by discussing the mechanisms involved in the emergence of population cycles. State-structured competitive interactions allow for the emergence of two major kinds of cycles depending on whether the effect of density dependence is direct or delayed: single-generation cycles and delayed-feedback cycles. Both these types of cycles are commonly observed in natural and laboratory systems (Murdoch et al. 2003).

Single-generation cycles are thus called because the period of the cycle is approximately equal to the generation time. They emerge as a consequence of a particular class of individuals being competitively dominant, thereby suppressing production in other cohorts. The mechanisms underlying these cycles are well exemplified by considering competitive asymmetry between juveniles and adults. When juveniles are competitively dominant, cyclic dynamics arise because a large dominant cohort of juvenile is produced and induces a strong level of density dependence. This results in fecundity suppression and/or strong mortality in adult stages. As this cohort progresses through life stages and matures into adults, density dependence is released and a new dominant cohort is produced. In contrast, when adults are superior competitors, the greater sensitivity of juveniles to density dependence induces variability in developmental delay and thus variation in recruitment to adult stages which ultimately lead to population cycles.

Delayed feedback cycles occur in a large range of scenario when density dependence operates on fecundity and/or survival with a delay. Such cycles are best illustrated with the

classical Nicholson-blowfly experiments (Nicholson 1957, Gurney and Nisbet 1985). In these experiments, blowfly larvae were fed ad libitum, while the food supply of adult population was limited. When adult density was high, density dependence caused total recruitment rate to larval stage to be strongly depressed. When this small group of newborns matured later into adults, they consequently experienced a high per-capita food availability which then caused a large production of newborns. Because larval food was not limited, the high number of descendants produced subsequently matured into a large number of adults causing fecundity to decrease again. Therefore, whenever a high number of newborns were produced, density dependence acted to decrease fecundity with a delay: once juveniles matured into adults. This caused successive generations of adults to alternate between high and low density with a period of approximately two developmental delays. Longer periods can occur when adults are relatively long lived. In these experiments, adults and juveniles differed in their sensitivity to density dependence because they experienced different environments. These mechanisms are however present in a larger class of systems, for example, when productivity depends on individual states.

All of these unique phenomena happen because of the combination of the functional dependence of ecological rates on individual states, and the population feedback on individual life history. Theory has been able to identify some of the major mechanisms associated with state-structured competitive interactions that explain major patterns at the population and community level (Murdoch et al. 2003, De Roos and Persson 2013). Despite this progress, our understanding of the interplay between individual life history and population and community processes remains limited. For example, *Daphnia* population dynamics show characteristics of both juvenile-driven and adult-driven single-generation cycles, which clearly suggests that the theory remains incomplete (De Roos and Persson 2013). Progress on these topics requires integrative approaches that simultaneously accounts for the mechanisms causing variation in an individual state along its life, the functional relationship between an individual state

and ecological performance, and the dynamical consequences of particular life histories.

1.2 Life-history evolution theory

In the previous section, I outlined some general principles underlying the expression of individual life history and their population and community consequences. I did not explicitly consider how these processes affect and are affected by the evolutionary process.

Classical life history evolution theory has simplifying assumptions on the mechanisms underlying the expression of life history. In this section, I will discuss the basic principles and assumptions underlying this theory and discuss the consequences of releasing some of these assumptions. In particular, I highlight three ideas linked to the fact that both the environmental dynamics and individual expressed life history are dynamic processes: (i) Dynamic variation in the environment affects qualitatively the selective process by affecting the determinants of an organisms fitness. (ii) Phenotypic expression responds dynamically to the environment, which has implications for the selective process. (iii) Evolution affects ecological processes, which feeds back to cause further change in the evolutionary processes

1.2.1 Natural selection and the notion of optimality in life-history

Life-history characteristics of living organisms are very diverse between species, but also within species. Life-history evolution theory tries to explain how such a large range of diversity emerges from the evolutionary processes, and most particularly from natural selection. Key issues traditionally studied by life-history theory are the determinants of age and size at maturity, of optimal body size, of reproductive effort, of life-span length, of whether it is optimal to produce few or many offspring (Stearns 1992, Roff 2002).

The process of natural selection affects the genetic composition of populations over time because different genotypes vary in their reproductive success, which leads to the central notion of fitness. Fitness, while usually hardly directly measurable, can be defined as the

capacity of a particular genotype to maximize its genetic contribution to future generations (Grafen 2007). It has been suggested that a limited number of key phenotypic traits of individual organisms may strongly determine their fitness. These traits have been collectively referred to as life-history traits, and usually describe the patterns of development, reproduction and survival. Thus, to determine the genetic dynamics generated by natural selection, one must first understand the selective mechanisms driving phenotypic evolution. If natural selection leads organisms to maximize their reproductive success, we can expect that the phenotypic response of an individual to its environment responds to a fitness maximization principle. The classical approach of optimization is to define a measure of fitness, define the relationship between traits and fitness, describe the trade-offs between traits, then find the combination of traits that maximizes fitness (Stearns 2000).

This approach has historically been central tenet around which life history theory has been developed, and most of current evolutionary thinking on the evolution of life histories stem from these assumptions. In its most classical vein, life-history theory assumes near-equilibrium dynamics, thus implying stationarity in the demographic process along with constant environmental conditions. This assumption is used to derive common fitness criteria used in life history analyses. In particular, this produces the result that the maximization of individual reproductive success coincides with the maximization of the population growth rate over the long term (Houston and McNamara 1999). It is also assumed that the environment has a single role: selecting among phenotypic variants. However, as discussed earlier, the environment may also affect the phenotypic expression of a particular genotype, i.e., life histories often include plastic responses to the environment. Finally, it is also assumed that evolutionary changes do not affect ecological processes. I now discuss the consequences of releasing these assumptions.

1.2.2 Environmental heterogeneity and variability

Evolution in constant environments is a particular case of the more general theory of evolution in fluctuating environments. Here, I present the consequences of relaxing the assumption of constancy in the environment.

Two observations justify the need to obtain a deeper understanding on the evolution of life-history in fluctuating environments. (i) Environments are strongly variable, which has important consequences on individual reproductive success. (ii) Evidence exists of adaptation to environmental stochasticity, particularly through the studies of phenotypic plasticity and genotype-environment interactions (Meyers and Bull 2002). In addition, the processes of selection in fluctuating environments have been largely invoked to understand the maintenance of genetic diversity in natural populations (Chesson 2000).

The environment is virtually never homogeneous nor static, but heterogeneous and dynamic. The environment displays spatial and temporal variability under the influence of biotic and abiotic factors, which may be caused by intrinsic (population dynamics) or extrinsic (environmental forcing) processes; these changes may be stochastic or deterministic, and act at different spatial and temporal scales. This poses a challenge for organisms, as they require relatively constant internal conditions for physiological processes to function correctly (Kooijman 2010). Moreover, virtually every aspect of an individual ecological performance (e.g. vital rates such as fecundity, survival, growth, feeding, etc.) may be conditioned by the environment, which will have direct consequences on the selective process. This has resulted in organisms evolving a large range of mechanisms to cope adaptively with such variability (reviewed in Meyers and Bull 2002).

The effect of variability on individual and evolutionary processes may largely differ as a function of the type, intensity, and predictability of the environmental component causing this variation (Roff 2002). A major distinction can be made between the different sources of variability based on the degree with which their effects are correlated across different

individuals (McNamara 2000). Variability acting independently on different individuals, e.g. demographic stochasticity, does not preclude populations from reaching stationarity and, as such, has been fully integrated within the framework of life-history theory. On the other hand, the environment may vary as a whole and cause a highly correlated response among individuals. The evolutionary processes involved in this last case add considerable complexity to the determinants of reproductive success, which are still not completely understood. Early works on the topic date from the 1960s with the pioneering works of Cohen (1966, 1967, 1968).

One of the main predictions from these lines of work is that individual optimization fails in fluctuating environments (Houston and McNamara 1999, Metz 2008, but see Grafen 2007 for an alternative interpretation). While the maximization of individual reproductive success in constant environment coincides with the maximization of the long-term growth rate of the genotypic population, this is usually not the case in fluctuating environments (Tuljapurkar 1989, Tuljapurkar et al. 2003). The fitness measure used in such environment is the geometric mean fitness of the population (Levins 1962, 1968). As environmental variance increases, the geometric mean fitness decreases. Therefore, early works on life-history evolution in fluctuating environments have emphasized the idea of a trade-off between the mean and variance in fitness, referred to as bet-hedging (Slatkin 1974, Philippi and Seger 1989). Some genotypes with a reduced mean reproductive success, but also a lower temporal variance, may therefore be at selective advantage under fluctuating environments. For example, this can be achieved by decreasing fecundity if juvenile are more sensitive to environmental stochasticity than adults, or if smaller clutches are buffered more against environmental conditions (Gillepsie 1977, Schaffer 1974). Moreover, under many circumstances, optimality is achieved through the use of randomized, instead of pure strategies (often referred to as adaptive coin flipping, Cohen 1966, Cooper and Kaplan 1982). That is, the rule prescribing the behavioral and physiological decisions to be taken by an individual are no longer described

by a deterministic solution, but is described by a probability distribution of behavioral decisions.

1.2.3 Phenotypic plasticity and state-dependent life histories

One of the consequences of individual adaptation to the different form of environmental variation is phenotypic plasticity (Via et al. 1995, de Witt and Scheiner 2004, Morris et al. 2014). The concept of phenotypic plasticity encompasses the early observation that a single genotype can produce a range of different phenotypes, and that conversely, a given phenotype can be produced by different genotypes. The range of phenotypes that a single genotype can produce is called the norm of reaction. The norm of reaction is a mechanistic response to the environment that mediates patterns of phenotypic expression. Since the fitness value of particular genotype depends on the phenotypic patterns expressed in response to the environmental conditions encountered, reaction norms can be considered as a trait to be optimized by selection processes (DeWitt et al. 1998, Morris and Rogers 2014). Organisms may thus be selected to cope with environmental variability. When it is adaptive, a great level of plasticity often comes with a cost (Callahan et al. 2008), leading to a trade-off between the degree of local adaptation (i.e. the degree of adaptation to a particular environmental state) and the degree of plasticity. Relative constancy in the environment might therefore be reflected by a low degree of plasticity, while more variable environments might be reflected by a high degree of plasticity, but not necessarily: because of the costs and limits of plasticity, no genotype has evolved plastic traits so flexible that it can dominate in all environments (Tollrian and Harvell 1999, Calahan et al. 2008, Svanback and Schluter 2012). This has direct implications in understanding the mechanisms maintaining genetic diversity in spatially or temporally variable environments: some genotypes are well adapted to a particular environmental state, while other are less well adapted, but best in another environmental state (Miner et al. 2005, Roff and Fairbairn 2007). Moreover the cost and benefits of the trade-off components are likely to change in a different fashion as a function

of the genetic identity of the individual, a process that gives rise to genotype-environments interactions (GxE). GxE can be interpreted as genetic variation in the reaction norm. GxE provide a source of relative nonlinearity in competitive interactions between genotypes, one of the fundamental mechanisms explaining the maintenance of genetic diversity in natural populations (Chesson 2000).

Further progress has been made by recognizing that phenotypic plasticity may also be interpreted as an organisms response to its state (McNamara and Houston 1996). I previously discussed that individual ecological performance may depend on the particular state of the individual. This has led to the idea that the behavioral and physiological decisions taken by an organism may themselves depend on an individual's state, and that natural selection may be acting on these state-dependent strategies to improve an organisms fitness (Houston and McNamara 1999). In other words, an organism's response to its state may be shaped by the selective process. The complexity of these analyses does not only lie in the question of determining what aspects of an individual's ecological performance is optimized by the selective process, but also on the fact that the actions taken by an organism cannot be considered in isolation. The *realized* fitness of a particular life history is the product of the particular sequence of actions made by an individual along its lifetime. However, if some form of variability is present in the environment, the *expected* fitness of a particular strategy is more complex to assess, as different environmental conditions may affect the state trajectories of individuals and may be further affected by dynamic individual responses to this variation. Thus, the action taken by an organism at a given time may influence its state in the future, which will in turn affect its residual reproductive value and hence the fitness value of this action.

In an evolutionary analysis, one would thus require an assessment of how the combination of particular strategies and particular environmental scenarios conditions an individual fitness, for a multitude of strategies and environmental scenarios. These results can only be

reached if we have a robust understanding of the physiological and demographic consequences of particular strategies, hence requiring a strong mechanistic linking as to how dynamic variation at the individual level and dynamic variation in the environment combine with each other to produce particular life histories with particular demographic consequences.

1.2.4 The ecological feedback

Since natural selection acts on state-dependent aspects of an individual demographic performance, and that these very processes condition population and community processes, evolutionary changes in the population entail ecological changes. These ecological changes in turn affect the selective process as the selective value of the genotypes is a function of their environment, thus creating a feedback between ecological and evolutionary processes (Dieckmann 1995, Vincent and Brown 2005). The complexity induced by this eco-evolutionary feedback is linked to the fact that in many circumstances, environmental changes may affect the fitness ranking of genotypes. This leads to the conclusion that, in general, no optimization principle applies (Metz et al. 2008).

The field of adaptive dynamics specifically focuses on the role of this eco-evolutionary feedback on the evolution of organisms' traits (Dieckmann 1995, Diekmann 2004). The main approach consists first in assuming that the environmental dynamics are set by a resident population. The fate of new genetic variants (often assumed to be produced through mutations, hence referred to as mutants) is then evaluated as their ability to grow under these conditions. Under the so-called infinitesimal model, mutations are assumed to results in infinitesimally small changes in an organisms strategy. By using local perturbation analysis, the linearization of the fitness function around the resident strategy makes it possible to assess whether a small genotypic change results in an increase or a decrease in fitness. The resulting metric is the selection gradient. As long as its value is different from zero, this indicates that the resident strategy is unstable in the sense that small mutations may increase an organism's fitness and thus replace the resident strategy. Under these assumptions, the

evolutionary process should, over the long term, result in a strategy where the selection gradient vanishes, i.e. no strategy close to the resident strategy may invade.

These types of analyses have brought new insights on the type of evolutionary phenomena produced by this interplay between ecological and evolutionary processes. First, the predicted outcome of the selective process may be very different from that predicted in the absence of the ecological feedback (Mylius and Diekmann 1995). Second, it can give raise to qualitatively new phenomena, such as evolutionary branching points, which may, for example, lead a monomorphic population toward a dimorphic state. The implications for the speciation process has been discussed elsewhere (Dieckmann and Doebeli 1999, Doebeli and Dieckmann 2000).

In most of these analyses, a time scale separation between ecological and evolutionary processes is assumed: the apparition of a new genetic variant results in a period of competitive interaction between the resident and mutant strategy that is short relatively to the time required for new genetic mutants to appear (Diekmann 2004). Thus, on the evolutionary time scale, if the mutant successfully invades and replaces the resident strategy, the environment sets more or less instantaneously to its long-term behavior. This coincides with the classic view that evolution is a slow process, acting on the scale of thousands if not millions of generations, whereas ecological processes acts on the scale of few generations (Slobodkin 1961). New evidences have however challenged this view and shown that evolution can be as fast as the ecological process (Thompson 1998, Carroll et al. 2007, Pelletier et al. 2009, Ellner et al. 2011, Schoener 2011). Once again, releasing simplifying assumptions comes with qualitatively new phenomena for both the ecological and evolutionary process, and we are only starting to understand the implications of this eco-evolutionary feedback on contemporary time scales (Ellner 2013).

All these observations highlight the fact that the dependency of ecological dynamics on the genetic composition of the population has influence on the evolutionary process and

vice-versa. The environment dynamically responds to genetic and phenotypic variation. Any analysis aiming at studying how these processes act in conjunction requires a mechanistic account of how ecological and evolutionary factors act to generate phenotypic variation at the individual level, and assess how this variation, in turn, conditions ecological and evolutionary processes.

1.3 Conclusions and structure of the thesis

I discussed how the combination of state-dependence in individual energetics and density-dependent processes could give rise to various population and community phenomena, and had implications for evolutionary processes. Linking this theory with empirical systems requires experimentations that assess and parameterize the functional dependency of individual performance on environmental state. Yet, in almost all the experiments that have been carried out, individuals were raised in isolation under various but constant environmental conditions. It is then assumed that the inferences drawn from these experiments remain valid under conditions of density dependence and when the environment varies in a dynamic fashion. This assumption has however never been tested, but as discussed in this introduction, there are logical reasons to expect that it may not always be valid.

As the *Daphnia* systems represents one of the systems in which individual energetics have been the most thoroughly studied, I conducted an exhaustive review of the literature to identify studies that looked specifically at how different clones respond to variation in their food environment (Deng 1996, Ebert 1991, 1993, Ebert et al. 1993, Epp 1996, Giebelhausen and Lampert 2001, Glazier 1992, Glazier and Callow 1992, Nelson 2004, Nelson and McCauley 2005, Olijnyk and Nelson 2013, Pietrzak et al. 2010, Weider 1985, Weider and Wolf 1991, Yampolsky and Kalabushkin 1991). Beside the studies of Nelson et al. (Nelson 2004, Nelson et al. 2005), these studies have invariably used individuals raised in isolation and under constant food conditions. It is thus hardly defendable that we can bridge indi-

vidual genetics with ecological and evolutionary patterns, when the presupposed effects of interclonal differences measured at the individual level have not even been tested in term of their effects at the population level.

In this introduction, I discussed how the individual is a highly dynamical unit, responding to physiological, ecological and evolutionary factors. Ecological and evolutionary theories have brought some insights on the novel phenomena emerging from this variability. Yet, they also give a hint of the complexity emerging when more and more realism on these processes is considered. There is still much theoretical and empirical knowledge that remain to be bridged to reach a better understanding of these processes. Reaching a comprehensive knowledge of how environmental variability and genetic variability combine with each other to affect the dynamics at the individual and population levels can only be achieved through a complementary effort of mathematical modeling, of empirical quantification of life-history, demography and environmental dynamics, and of mechanistic approaches aiming at characterizing the physiological responses of genetic variants to their environment and their impact on ecological and evolutionary dynamics. My Ph.D. research has been motivated by these observations and aims at bringing some elements that could help completing this knowledge. More specifically, I have been looking at the effects the ecological feedback entails on the expression of individual life history and how genetic and environmental variation combine each other to affect the demographic process. I used a mechanistic approach that link the genetic response of individuals to ecological and evolutionary factors to the population process through the consideration of individual energetic and life-history processes.

In the second chapter of this thesis, I present methods for carrying out sensitivity analysis of dynamic energy budget models. I make use of the 'direct method' from mathematical theory on a simple model for individual growth, fecundity and mortality. The method allows to quantify the effects and to decompose the chain of events that small genetic perturbation in energetics cause on individual realized patterns of life history. Sensitivity analyses are

of paramount importance in ecological and evolutionary theory, but their application to continuous time models has been virtually ignored from these fields until now. I discuss the benefits its application can bring for the study of many ecological issues.

In the third chapter, I make further use of these techniques to study how genetic variation affects individual life history under conditions of density dependence. In consumer-resource systems, regulation of the consumer population growth is achieved through resource adjustment, which is only possible if the resource conditions adjust dynamically to consumer biology. These mechanisms drastically alter the predictions made on realized patterns of individual life history and have important implications for life-history evolution theory, as these effects have been traditionally ignored from these lines of research.

In the fourth chapter, I present the results of large manipulative experiments on *Daphnia*-algae systems designed to study how the combined effect of genetic variation and environmental fluctuations affect the population demography and its underlying energetic basis. The previous chapter reports that the environmental feedback tends to buffer the effect of genetic variation on individual phenotype and demography. Thus, I used a unique experimental protocol that decoupled this environmental feedback and allowed for a better characterization of genetic differences among clones. It is shown that resulting population demographical patterns are affected by a genotype-environment interaction, and that environmental fluctuations affect qualitatively the dependence of individual energetics on food conditions. These results have important implications for the way we envision how genetic variation affects ecological processes at the population and higher levels.

In the fifth chapter, I investigate the molecular basis underlying observed demographical patterns observed in the previous chapter. Genome-wide gene expression patterns were quantified for two of the clones used in the previous experiment, using microarrays. This chapter represents only a first-step analysis of the wealth of data provided by this experiment and aims at providing a preliminary test of the patterns of gene expression under distinct

environmental dynamics and assess their coherence with observations at the population level. These results show that a large percentage of the *Daphnia* genome is sensitive to the cycle phase, cycle type, and clonal variation. Moreover, genotype-environment interactions could also be detected at the genomic level.

Finally, I explore further the consequences of one of the major findings drawn from these experiments, namely that environmental fluctuations may reverse the competitive ranking between differently sized individuals. I show that beyond potential genetic differences, common mechanisms drive the dynamics of these experimental systems. These results may help explaining seemingly contradictory evidences on the patterns of intraspecific competitions observed in *Daphnia* species, and they have major implications for understanding the mechanisms driving the dynamics of many consumer-resource systems.

Chapter 2

Sensitivity analysis of continuous-time models

2.1 Introduction

Sensitivity analysis is a powerful tool that allows assessment of the effect of parameter variation on model outputs. Its application to ecological and evolutionary theory involves a large range of issues and represents an important contribution to the fields of population dynamics, conservation biology, epidemiology, ecotoxicology, evolutionary biology (Benton and Grant 1999, Caswell 2000, Easterling et al. 2000, Metcalf and Pavard 2007). In these applications, the models being used are specified on a discrete-time basis, usually as a form of matrix population models. There is a well-developed framework for applying sensitivity analysis to these types of models, including models accounting for environmental variability, density-dependent growth and transient dynamics (Caswell 2000, 2007, 2008, 2009).

Whereas discrete-time models are adequate for modeling many systems, a large range of systems are best modeled within a continuous-time framework, with models specified as a set of differential equations (Gurney and Nisbet 1997). This includes a large class of population models, such as consumer-resources or epidemiologic models. Continuous-time models are also important for models of individual life-history, as they are often derived from the consideration of the energetics of growth, development and reproduction, processes that operate on a continuous-time basis (De Roos 2008).

Besides a handful of studies, sensitivity analysis has largely been omitted from the analysis of these ecological and life history models (Buzby et al. 2008, Tavener et al. 2008; as far as the authors are aware). The insights that sensitivity analysis can yield for understanding their behavior is potentially as large as their discrete-time cousins, and ecological and evolutionary theory would certainly benefit from a more spread application of sensitivity analyses.

De Roos (2008) presented numerical methods for deriving the sensitivity of the population growth rate based on finite differenciation methods, and this method may also be applicable to a larger range of issues. Mathematical theory possesses however an analytical framework with a comprehensive set of tools that makes this type of analysis applicable to almost any models commonly used by ecologists and evolutionary biologists.

Time-dependent models make predictions on the dynamics of the variables over time and these trajectories have often an intrinsic interest beyond their asymptotic behavior. This is particularly true when the time variable represents an individual age, such as in models for individual life-history, for example. The interest is then placed on the whole, age-dependent, realization of the schedule of growth, reproduction and mortality. In this case, parameter perturbation is likely to affect these trajectories all along an individual lifetime, and may manifest its effect in a contrasted way depending on the individual age. The sensitivities of these model outputs to parameter perturbation are thus themselves time-dependent trajectories, and have therefore been referred by some author as dynamic sensitivities (Shiraishi et al. 2009). The possibility of performing this kind analysis on systems of differential equations can thus prove particularly useful for the field of life history theory, as it enables dealing with issues that could previously be dealt only with models having closed-form solutions (e.g. Roff 2002). For example, it can be used to study how variation in the energy allocation function between growth and reproduction affects resulting patterns of individual development and fecundity. Doing so does not only yield insights on the mechanisms underlying patterns of phenotypic expression, but because of the direct link of these components with the demographical process, this allows to draw a number of implications for ecological and evolutionary processes. In particular, it makes it possible to calculate the selection gradient for physiologically structured populations, which is itself a sensitivity.

The two main methods allowing sensitivity analysis are the direct and the adjoint meth-

ods (Tortorelli and Michaleris 1994). The goal of this paper is to present and adapt the direct method to the analysis of ecological methods. We walk the reader through an application using a model that predicts individual lifetime patterns of growth, fecundity and survival based from the consideration of the mechanisms mediating individual energetics. Additionally, we also present method for analyzing the sensitivity of discrete events, such as the age at maturity, which are less easily accessible than the basic tools for sensitivity analysis, as this type of phenomena yields an hybrid system, which exhibit both continuous and discrete dynamic behavior.

2.2 Dynamic models and dynamic sensitivities

2.2.1 The direct method

Consider a general dynamic model given by the following system of ordinary differential equations:

$$\frac{d\mathbf{x}}{dt}(t,p) = \mathbf{f}(\mathbf{x}(t,p),p) \tag{2.1}$$

$$\mathbf{x}(t_0,p) = \mathbf{x}_0 \tag{2.2}$$

Where $\mathbf{x}(t,p)$ is a vector containing the n state variables, $x_i(t,p)$. Note that we made their values at time t to be a function of some parameter of interest p. $\mathbf{f}$ is a vector of n function f_i giving the rate of change in $\mathbf{x}(t,p)$ as a function of time.

Integrating such a system yields a solution $\mathbf{x}(t,p)$ that gives the trajectory of the states variables as a function of time, for a given value of the parameter p. The effects of small (finite) changes Δp in the parameter p on these trajectories can be assessed through linear extrapolation:

$$\mathbf{x}(t,p+\Delta p) \approx \mathbf{x}(t,p) + \frac{d\mathbf{x}(t,p)}{dp}\Delta p \tag{2.3}$$

The sensitivities are the terms $\mathbf{S}_p(t) \equiv \dfrac{d\mathbf{x}(t,p)}{dp}$, or expressed in scalar notation, $S_{ip}(t) \equiv$

$\frac{dx_i(t,p)}{dp}$. They can be interpreted as the expected displacement in $\mathbf{x}(t,p)$ relative to an arbitrarily small displacement in p.

Most systems of differential equations do not have any closed-form solution. Instead the dynamics are specified by a system of equations, such as (2.1, 2.2), and the solution is inferred by numerical integration. Doing so prevents any differentiation of the solution, and one has to resort on other methods in order to derive the sensitivities. One of these is the *direct method* (Tomovic and Vukobratovic 1972, Dickinson and Gelinas 1976). It simply requires the integration of an additional set of differential equations, together with the original system. Its derivation is straightforward and involves only basic calculus. The only potential difficulty one may encounter arises when discontinuities are present in the rate functions $\mathbf{f}$, but we give a method for dealing with these issues in part 5.

The derivation starts by noticing that since:

$$x_i(t,p) = \int_{t_0}^{t} f_i(\mathbf{x}(\tau,p),p)d\tau \tag{2.4}$$

then

$$S_{ip}(t) = \frac{d}{dp}\int_{t_0}^{t} f_i(\mathbf{x}(\tau,p),p)d\tau \tag{2.5}$$

Interchanging the order of differentiation and integration gives:

$$S_{ip}(t) = \int_{t_0}^{t} \frac{df_i(\mathbf{x}(\tau,p),p)}{dp}d\tau \tag{2.6}$$

This implies that the variable $S_{ip}(t)$ can be solved by integrating the differential equation

$$\frac{dS_{ip}(t)}{dt} = \frac{df_i(\mathbf{x}(t,p),p)}{dp} \tag{2.7}$$

along with the original system.

Further progress is made by applying the chain rule to (2.7):

$$\frac{dS_{ip}}{dt} = \frac{\partial f_i}{\partial p} + \sum_{j=1}^{n} \frac{\partial f_i}{\partial x_j} S_{jp} \tag{2.8}$$

Notice that a correct interpretation of the resulting sensitivities is only possible in the context of dynamical systems. Indeed, at a given time t, the perturbation in p affects only directly the rates at which the system changes. This eventually affects the state of the system at future time, but not at the focal time t.

2.3 Applying the sensitivity analysis

In what follows, we show an application of these methods on an example model. We first present the model, and then provide some basic, though relatively detailled, interpretation of the results of this analysis, with the hope of giving the reader a better sense as to what these dynamic sensitivities represent, how they arise, and to what purpose they can be used.

2.3.1 The model

The model we use is a basic model for the physiological ecology of individual consumers. It is derived from a simple account of basic rules of energy acquisition, allocation and expenditure; and predicts individual lifetime patterns of growth, reproduction and survival. Below, we give a brief description of the model and its derivation. The model is set using functions and parameter values that roughly corresponds to *Daphnia pulex* biology (Nisbet et al. 2004, 2010). All equations and default parameter values used are given in Tables 2.1 and 2.2.

Individual consumers feed on a single resource present at a constant concentration F. They grow all their life, as long as enough food is available. The relation between an individual weight and length is fixed: $W = \chi L^3$. We use length (L) as the state-variable describing the individual "size". Metabolic maintenance rate scales linearly with individual weight: $M = m\chi L^3$. Individuals have a type II functional response, with a half saturation constant F_h and a maximum assimilation rate that increases proportionally to the square of individual's length: $A_{max} = \nu L^2$. At any age, individual allocate energy toward growth. The rate of energy allocation is a fixed proportion θ of the net production rate (assimilation

minus maintenance rate). Taken together, these elements produce the following equation for an individual growth rate:

$$\frac{dL}{da} = \frac{\theta}{3\chi}\left(\frac{F}{F+F_h}\nu - m\chi L\right) \tag{2.9}$$

All remaining energy serves reproductive purposes: development of a reproductive structure in juveniles (maturation), and offspring production in adults. For simplicity, we assume no energy costs associated with the maintenance of the reproductive tissues. The dynamics in the cumulative amount of energy invested toward reproduction, $R(a)$, thus follow:

$$\frac{dR}{da} = (1-\theta)\left(\frac{F}{F+F_h}\nu L^2 - m\chi L^3\right) \tag{2.10}$$

In the juvenile stages, when this quantity hits some threshold R_m, the individual matures into an adult and starts reproducing.

The fraction of individuals surviving up to age a is denoted as $s(a)$. Per-capita mortality rate is taken to depend on an individual length such that:

$$\frac{ds}{da} = -(\mu_0 + \mu_1 e^{\mu_2 L})s(a) \tag{2.11}$$

We chose a parameterization that makes it an increasing function of length.

For further purposes, we will also need to calculate the average number of descendants produced from birth to age a that we refer to as effective fecundity: $B(a) = \int_0^a \beta(\alpha)s(\alpha)d\alpha$, where $\beta(a)$ is the rate of offspring production of individuals of age a. Thus, the value $B(\infty)$ is the basic reproductive number R_0.

Our model therefore contains 4 state-variables: L, s, R, and B. We denote the corresponding rate functions as g_L, g_s, g_R, and g_B, respectively. Note that under these assumptions and under constant food concentration, the model results in von Bertalanffy growth with parameters: $L_\infty = \frac{\nu F}{m\chi(F+F_h)}$ and $r_B = \frac{\theta m}{3}$. We also note here that net production rate is a hump-shaped function of length, and so are individual growth rate and the fecundity rate since they are directly proportional to net production rate. It starts as an increasing

function of length, reaches a maximum at $L_p = \dfrac{2\nu F}{3m\chi(F+F_h)}$, and then decreases again with length, to become null at $L = L_\infty$. Under the model parameterization used in our example, $L_p = 4.72$ mm, a length that individuals do not reach within their lifetime. So, in the interpretation presented below, we simply consider net production to be an increasing function of length, and to remain always positive.

Examples of model runs for a constant food concentration of 100 μgC/mL and for different values of θ are shown in Figure 2.1, left pannel. Natural questions that can be asked in view of this figure are: what is the effects of a small change in the model parameters on these trajectories? And how do these effects come about? For example, does increasing energy investement toward reproduction necessarily increases lifetime offspring production? Figure 2.1e clearly shows that the response is no - most certainly because of the allocation trade-off with somatic growth. Can we then characterize the details about how this trade-off operates? All these questions can be enlightened through the help of a sensitivity analysis.

2.3.2 Model analysis

For this example, we choose θ - the fraction of net production allocated to growth - as the parameter to be perturbed. The system of differential equations used to solve the sensitivities is given in Table 2.3 in a generic form and as a function of model parameters and variables in Table 2.4. A diagramatic representation of the paths conveying the perturbation to the state variables is given in Figure 2.2. Model predictions for three values of θ, and the corresponding sensitivities are plotted in Figure 2.1.

Sensitivity of $L(a)$. Figure 2.1 shows that, not surprisingly, an increase in the fraction of net production allocated to growth increases an individual length at every age. It is also an increasing function of an individual age, reflecting the fact that the perturbation does not only affects an individual's growth rate at every age, but also accumulate over the growth trajectory. This intuitive result can also be explained in a more mechanistic fashion

by considering the differential equation for the sensitivity (Table 2.3 eq. 1). Variation in the function $S_{L\theta}$ is the result of two processes: the direct effect of parameter variation on the growth rate $\frac{\partial g_L}{\partial \theta}$; and variation resulting from the perturbed value of individual length consequent to the past effects of the perturbation $\frac{\partial g_L}{\partial L} S_{L\theta}$. Both $\frac{\partial g_L}{\partial \theta}$ and $\frac{\partial g_L}{\partial L}$ are positive (as long as $L < 4.72$ mm), leading $S_L(a)$ to be a monotonically increasing function of age.

Sensitivity of $s(a)$. Increasing θ decreases the fraction of individuals surviving at every age, and the survival curve is most strongly affected by the perturbation in θ when individuals are about 20 days old. In contrast to the previous case, the perturbation does not act directly upon the survival function (Figure 2.2 and Table 2.3). Mortality rates are impacted only because mortality rate depends on individual length, which is affected by the perturbation. Since the per-capita mortality rate is an increasing function of length, and the growth trajectory is affected positively by the perturbation, per-capita mortality increases at every age.

The other term mediating variation in the survival function is $\frac{\partial g_s}{\partial s} S_{s\theta}$ and results from the effect of the perturbation at previous age on $s(a)$. Since the focus is at an individual level, it is important to recognize that this is a mathematical effect resulting from the fact that we calculate an expectation which corresponds to the probability of survival from birth to age a, but individual survival remain unaffected *per se*. Because $\frac{\partial g_s}{\partial s}$ is negative, the resulting feedback is negative, and this eventually nullifies the effect of the perturbation.

Sensitivity of $R(a)$ Compared to the two previous cases, the pattern of sensitivity of the reproductive output appears more variable, and strongly depends on the value of θ. Increasing θ for small values of θ increases reproductive output at every age (green curve in Figure 2.1f), whereas the opposite is true for large values of θ (blue curve). In a crude way, this indicates that reproductive output is the greatest at some intermediate values of θ, as it indeed appears to be the case in Figure 2.1e. Intermediate values of θ result in $R(a)$ to be less sensitive to the perturbation. This produces more subtle patterns of variation, where

the perturbation affects the distribution of the reproductive output within the individual's lifetime, rather than simply causes an overall increase / decrease.

Perturbation in $R(a)$ arises from the direct negative effect of parameter perturbation on the rate of reproductive allocation, and from an indirect effect resulting from the dependency of net production on length (Table 2.3, Figure 2.2). Since net production increases with length, this last effect contributes positively to the $R(a)$. Again, these two processes oppose each other, much like it was the case for $S_{s\theta}(a)$. However, there is no negative feedback effect limiting the range of variation in $S_{R\theta}(a)$ and the effect of perturbation seems to grow unbounded with age.

Whereas the results from the two previous cases could have been roughly predicted on a purely logical ground, it is much harder to deduce the effects of the perturbation on R without the quantitative predictions provided by the sensitivity analysis.

In this section, we have shown how applying an analysis of sensitivity may reveal the effect of parameter perturbation on the different components of an individual's expressed life history. This effect is usually non-uniform over an individual's lifetime, reflecting the fact that the perturbation affects vital rates in a different fashion as a function of an individual age, but also that these effects accumulate at future ages. For example, the dynamics of all three curves ($S_{L\theta}$, $S_{s\theta}$, and $S_{R\theta}$) depend on a number of elements that are interdependent in time. Nonetheless, their rates of change at any given age depends on a sum of independent mechanisms as revealed by eq. (2.8) and this makes it possible to break apart the effect of the perturbation into the contribution of independent processes. The above discussion on the different curves exemplifies how the consideration of this property may be used to provide further insights on the mechanisms allowing the perturbation to propagate over the different compartments of an individual expressed phenotype.

The last aspect of the model analysis that has not been discussed so far concerns the sensitivities of $B(a)$. Its analysis presents an additional difficulty, which is exemplified the

jump in the function $S_{B\theta}(a)$ occuring at maturity that can be seen in Figure. 2.1h. It happens because the fecundity rate function is discontinuous at maturity. The way to deal with that peculiarity is a particular case of analyzing sensitivities associated with the timing of events, and we present the framework necessary to deal with this below.

2.4 Sensitivity analysis of events

In many circumstances, the timing of events matters, and perturbation in this timing may generate additional sources of perturbation. Formally, we define an event as a particular state of interest taken by the system. For example, an event may be associated with the moment at which an individual reaches maturation, or the moment at which the survival probability equates 0.5. Mathematically, this may be expressed as a condition on the state of the system, such as:

$$\int_{t_0}^{t_e} f(\tau, p)d\tau = h(x(t_e, p)) \tag{2.12}$$

Where t_e is the time at which the event happens, and h is some function defining the conditions on the state of the system.

If the function $f(\tau, p)$ is affected by parameter perturbation, the interval of time $(t_e - t_0)$ necessary for condition (2.12) to be met will most certainly be affected too, making it an implicit function of p. In order to assess the resulting displacement in t_0 and t_e, we resort to the following property of integrals with a variable domain of integration (Apostol 1991, Stewart 2007):

$$\frac{d}{dp}\int_{t_0(p)}^{t_e(p)} f(\tau, p)d\tau = \int_{t_0}^{t_e} \frac{\partial f(\tau, p)}{\partial p}d\tau - f(t_0, p)\frac{dt_0}{dp} + f(t_e, p)\frac{dt_e}{dp} \tag{2.13}$$

To see how this property can be used, we come back to our example. We saw that maturity was trigerred once the individual had gathered enough energy to this purpose. This condition is expressed as:

$$\int_0^{A_m} g_R(a, \theta)\, da = R_m \tag{2.14}$$

where A_m is the age at maturity.

Considering perturbation in θ and using (2.13), we deduce that:

$$\int_0^{A_m} \frac{\partial g_R(a,\theta)}{\partial \theta} da + g_R(A_m,\theta)\frac{dA_m}{d\theta} = 0 \tag{2.15}$$

And we conclude that:

$$\frac{dA_m}{d\theta} = -\frac{S_{R\theta}(A_m)}{g_R(A_m,\theta)} \tag{2.16}$$

Calculating such a measure may be useful because for its intrinsic interest, but it might also be a necessary step for further calculations. Indeed, in this particular example, the condition defining the event is posed on one variable of the system only (R). This implies that the perturbed timing of this event may be associated with further variation on the values of the other state-variables. For example, perturbation in the age at maturity entails additional perturbation in the length at maturity. Indeed, the length at maturity is defined by:

$$L_m(\theta) = \int_0^{A_m(\theta)} g_L(a,\theta) da \tag{2.17}$$

Using (2.13) once again, it follows that:

$$\frac{dL_m}{d\theta} = S_{L\theta}(A_m) + g_L(A_m,\theta)\frac{dA_m}{d\theta} \tag{2.18}$$

which is readily calculated once $S_{L\theta}$ (part 3.2) and $\dfrac{dA_m}{d\theta}$ (eq. 2.16) are known.

Hence, the perturbation affects the length at maturity in two different ways: because of its effect on the growth schedule - which is accounted for by the term $S_{L\theta}(A_m)$; and also because L_m depends on the age at which the individual matures, which may also be affected by the perturbation. This rises a call for warning: if no care is taken, one may conclude too quickly that the sensitivity simply equates $S_{L\theta}(A_m)$.

Discontinuities

A particular kind of events are those represented by discontinuities. Biologically, such discontinuities are often the result of abrupt changes in individual vital rates, as might happen when individuals start reproducing. In our example, the function $g_B(a)$ presents such a discontinuity and 'jumps' from 0 to $\beta(A_m)s(A_m)$ at maturity. Discontinuities may have important consequences in term of integration, and their effects must be carefully accounted for (Tolsma and Barton 2002).

Consider once again the following dynamical system:

$$\frac{dx(t,p)}{dt} = f(t,p) \tag{2.19}$$

and consider a discontinuity in the function $f(t,p)$ arising at some point in time t_d. Rather than considering the value of the function at this point, we look at the limits on the left (i.e. $t < t_d$) and on the right ($t > t_d$) of this point. As long as these limits are well defined and finite at t_d, this does not cause any major problem for integrating the function, as integrability only requires for the solution $x(t,p)$ to be continuous (Tolsma and Barton 2002). However, in the present context, a subtlety may arise because the location at which this discontinuity arises may be affected by parameter perturbation.

Indeed, when the sensitivity function is integrated over some interval $[0,t]$ containing the discontinuity ($0 < t_d(p) < t$), we get that:

$$\frac{dx(t,p)}{dp} = \frac{d}{dp}\int_0^{t_d(p)} f(\tau,p)d\tau + \frac{d}{dp}\int_{t_d(p)}^{t} f(\tau,p)d\tau \tag{2.20}$$

Since each term on the right hand side of (2.20) contains a variable domain of integration, we need to use (2.13) for their calculations, and acknowledge that the left and right limits of f at t_d may be different, to get the result that:

$$\frac{dx(t,p)}{dp} = \int_0^t \frac{\partial f}{\partial p}(\tau,p)d\tau + \left(f(t_d^-,p) - f(t_d^+,p)\right)\frac{dt_d}{dp} \tag{2.21}$$

where $f(t_d^-,p)$ is the limit on the left hand side, and $f(t_d^+,p)$, the limit on the right hand side. Notice that when these limits are equal, the last terms of eq. (2.21) cancel out, and

there is no effect of the discontinuity. This kind of discontinuities are accordingly classified as 'removable discontinuities'. Only when these two limits are unequal will there be an effect, and the discontinuity is called a 'jump discontinuity' (Stewart 2007).

In term of computation, the easiest way for accounting the effect of a jump discontinuity is to break the integration at t_d, update $S_{xp}(t)$ by adding up $\left(f(t_d^-, p) - f(t_d^+, p)\right) \frac{dt_d}{dp}$, and resume the integration.

In our example, $g_B(a)$ presents a jump discontinuity at maturity, with a limit on the left of $g_B(A_m^-) = 0$, and a limit on the right of $g_B(A_m^+) = \beta(A_m)s(A_m)$. Thus, for ages $a >= A_m$:

$$S_{B\theta}(a) = \int_{A_m}^{a} \frac{\partial g_B(\alpha, \theta)}{\partial \theta} d\alpha - g_B(A_m^+, \theta) \frac{dA_m}{d\theta} \tag{2.22}$$

The value $-g_B(A_m^+, \theta)\frac{dA_m}{d\theta}$ represents the extent by which the function S_B 'jumps' at maturity, as depicted in Figure 2.1h. Since $g_B(a)$ is always positive, it can be deduced that, with everything else fixed, a perturbation increasing the age at maturity has a negative effect on $B(a)$ around that age, whereas the opposite is true. This simply comes as a consequence from the fact that individuals maturating earlier also start producing offspring earlier, and they do so at a rate $g_B(A_m^+, \theta)$. More details on the sensitivity analysis of events and how to handle discontinuities can be found in Tolsma and Barton (2002) and ZivariPiran and Enright (2012). The curve depicted in Figure 2.1h is of particular interest for evolutionary analysis, as it predicts how reproductive success is affected over an individual lifetime, and its asymptotic value is an estimate of the selection gradient. For example, it shows here that the variation induced by the perturbed age at maturity may contribute to a substantial extent to the final value of the selection gradient if θ is small or large, but has barely any effect if θ takes intermediate values. Similarly, individuals of different ages will contribute to a different extent to variation in fitness, depending on the value of θ.

2.5 Discussion

Perturbation analysis is a major tool in the study of ecological and evolutionary processes (Benton and Grant 1999, Caswell 2000, Easterling et al. 2000, Metcalf and Pavard 2007). There is a well-established toolbox for matrix population models, but this type of approach is virtually absent from the analysis of continuous-time systems specified by a system of differential equations. In this paper we present a set of tools that makes this analysis possible based on the "direct method" from mathematical theory. It applies to any model specified by a set of ordinary differential equation, and is relatively simple to carry out. The mathematics are no more involved than those used for matrix population models and simply require to integrate an additional set of differential equations for the sensitivities along with the original system. The methodology is conceptually not much different from that presented by Hal Caswell (2007, 2008, 2009) for matrix population models, where the sensitivities are in essence evaluated by bookeeping the effect of the perturbation along a given trajectory of the system or along the stationary distribution of individual states. Both share the advantages that no assumption needs to be made on stationarity or linearity, making them applicable to both transient and non-linear dynamics.

All the framework presented in our study is based on the direct method, because we found it to be the most intuitive and the simplest to apply. There exist however variations around this methodology, which mostly aim at improving numerical accuracy and efficiency - as well as alternatives, such as the adjoint-method (Cacuci 1981, Tortorelli and Michaleris 1994, Chaniotis et al. 2001, Dunker et al. 2002, Wu et al. 2008, Perumal and Gunawan 2011, Zi 2011). We also note that, in contrast to discrete-time models, an additional kind of error arises when analysing periodic dynamics, because of the effect of parameter perturbation on the period of the cycles. There are however well established methods for dealing with this phenomenon (Larter 1983, Wilkins et al. 2009, Lu and Yue 2012), and we will present them in another paper.

Although we applied this methodology to a specific model about individual life history, its application may be valuable for studying a very large range of issues involving dynamic systems - as large as those of their discrete-time cousins. The sensitivity of the population growth rate, which has importance for many fields, can be readily derived. It may help in understanding the effects of natural or anthropological environmental variations on ecological systems, which includes applications in population, community, and ecosystem ecology, such as population management, conservation issues. It may be applied to issues in adaptive dynamics, epidemiology, parameter estimation, stability analyses, etc. The list could be quite long, see for example Caswell (2000, 2007) or Benton and Grant (1999), for more complete discussions on the topic. Note that the method is easily extendable for assessing elasticities ("relative sensitivities"), or higher order sensitivities, which are for example often used for assessing the dynamic stability of evolutionary singular points (Geritz et al. 1997, 1998).

The method also comes with qualitatively new phenomena associated with the sensitivity analysis of events. Events may be an important component of continuous time systems. In the simplest case, an event is simply a carrier of information and does not impact the dynamical behavior of the system. It essentially acts as a flag on the state of the system. For example, an event defined as the half-life of the variable modelled (e.g. individual) is often of interest, but does not impact in itself the dynamics of the system. At another extreme, an event may induce a jump in the state of the system and/or be the trigger for changes in the system dynamical behaviour. Both these kinds of events may be of interest in the biological world. Important issues involving events include studying the factor determining maturation - as exemplified in this study - and the dynamical consequences of this delay (see e.g. Day and Rowe 2002, De Roos and Persson 2003, Ernande et al. 2004, McCauley et al. 2009); threshold traits, where continuous variation in a trait called liability underlies discrete phenotypic variation (Hazel et al. 1990, 2004; Roff 1996); ecological threshold inducing major

changes in the dynamics of ecological systems, such as regime shifts (Groffman et al. 2006, Andersen et al. 2009); the period of cyclic dynamics (Kendall et al. 1999, Klausmeier 2008); and no doubt, one could think of more examples.

Ordinary differential equation models represent only a specific kind of continuous-time models, but other important one exists, such as delayed or partial differential equation models (DDE and PDE). For these models, direct methods also exists, which are essentially similar to those presented here (DDE: Rihan 2002, ZivariPiran and Enright 2012; PDE: Koda et al. 1979, Li et al. 2003, Petzold et al. 2006). The sensitivity analysis of DDE models involves only little additional complexity, but that of PDE is - not surprisingly - more tedious. PDE models are intrinsically more complex, but maybe more importantly, the discretization of the state space necessary for their computation induces an additional kind of error, which may be difficult to characterize (see e.g. Petzold et al. 2006 for a discussion on the topic). In any circumstance, the fact that a methodology already exists for these models is comforting and provides good support in the perspective of developping and applying the analysis for ecological models.

Table 2.1: Model definitions.

Balance equations

$\frac{dL}{da} = g_L(a) = \frac{\theta}{3\chi}\left(\frac{F}{F+F_h}\nu - m\chi L\right)$	Growth in length
$\frac{ds}{da} = g_s(a) = -(\mu_0 + \mu_1 e^{\mu_2 L})s(a)$	Survival function
$\frac{dR}{da} = g_R(a) = (1-\theta)(\frac{F}{F+F_h}\nu L^2 - m\chi L^3)$	Energy investement in reproduction
$\frac{dB}{da} = g_B(a) = \beta(a)s(a)$	Effective fecundity

Table 2.2: Parameter definitions and default values.

Parameter	**Value**	**Definition**
F_h	164	half saturation constant
ν	3.61	maximum assimilation rate per squared length unit
m	0.1	maintenance rate per weight unit
θ	varied	proportion of net production allocated toward growth
L_b	0.69	length at birth
W_e	1.6	egg mass
R_m	5.75	threshold for maturation
μ_0	0.07	mortality scalar
μ_1	0.005	mortality scalar
μ_2	1.5	mortality exponent
χ	1.93	parameter in the weight-for-length relationship

Table 2.3: Balance equations for the sensitivities in a generic form.

$$\frac{dS_{L\theta}}{da} = \frac{\partial g_L}{\partial \theta} + \frac{\partial g_L}{\partial L} S_{L\theta}$$
$$\frac{dS_{s\theta}}{da} = \frac{\partial g_s}{\partial L} S_{L\theta} + \frac{\partial g_s}{\partial s} S_{s\theta}$$
$$\frac{dS_{R\theta}}{da} = \frac{\partial g_R}{\partial \theta} + \frac{\partial g_R}{\partial L} S_{L\theta}$$
$$\frac{dS_{B\theta}}{da} = \frac{\partial g_B}{\partial \theta} + \frac{\partial g_B}{\partial L} S_{L\theta} + \frac{\partial g_B}{\partial s} S_{s\theta}$$

Table 2.4: Balance equations for the sensitivities as a function in an explicit form.

$$\frac{dS_{L\theta}}{da} = \frac{F\nu}{3(F+F_h)\chi} - mL + \frac{\theta m}{3} S_{L\theta}$$

$$\frac{dS_{s\theta}}{da} = -\mu_1\mu_2 e^{\mu_2 L} s S_{L\theta} - (\mu_0 + \mu_1 e^{\mu_2 L}) S_{s\theta}$$

$$\frac{dS_{R\theta}}{da} = -\frac{F\nu}{(F+F_h)} L^2 + m\chi L^3 + \left((1-\theta)(\frac{2\nu}{F+F_h} L - 3m\chi L^2) \right) S_{L\theta} +$$

$$\frac{dS_{B\theta}}{da} = -\frac{F\nu}{(F+F_h)W_e} s L^2 + \frac{m\chi s L^3}{W_e} + \left((1-\theta)(\frac{2\nu}{F+F_h} L - 3m\chi L^2) \right) \frac{s}{W_e} S_{L\theta} + \beta(a) S_{s\theta}$$

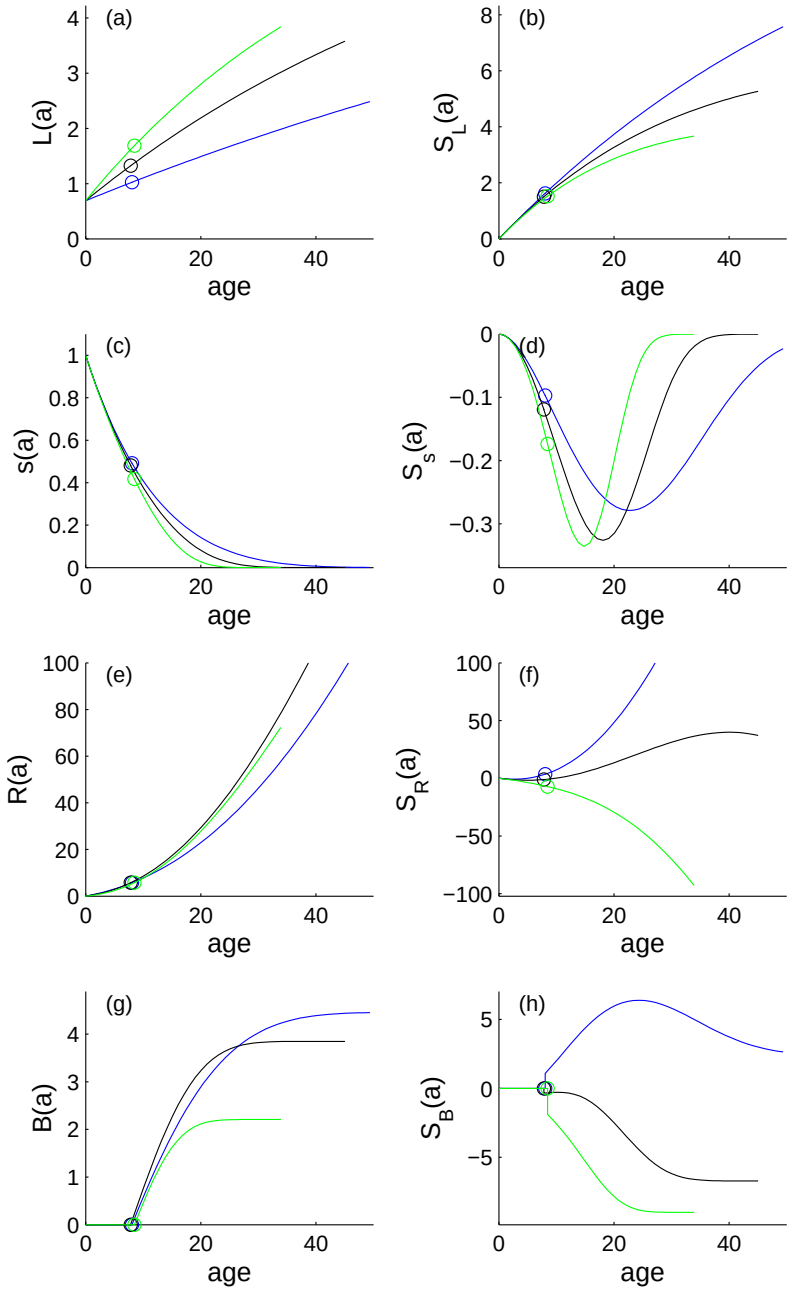

Figure 2.1: Trajectories of the state variables (left pannel), and sensitivities to θ (right pannel). Blue: $\theta = 0.2$; Black: $\theta = 0.4$; Green: $\theta = 0.6$. Circles indicates the age at which the individual matures. Food concentration is constant and equals 100 μgC/L.

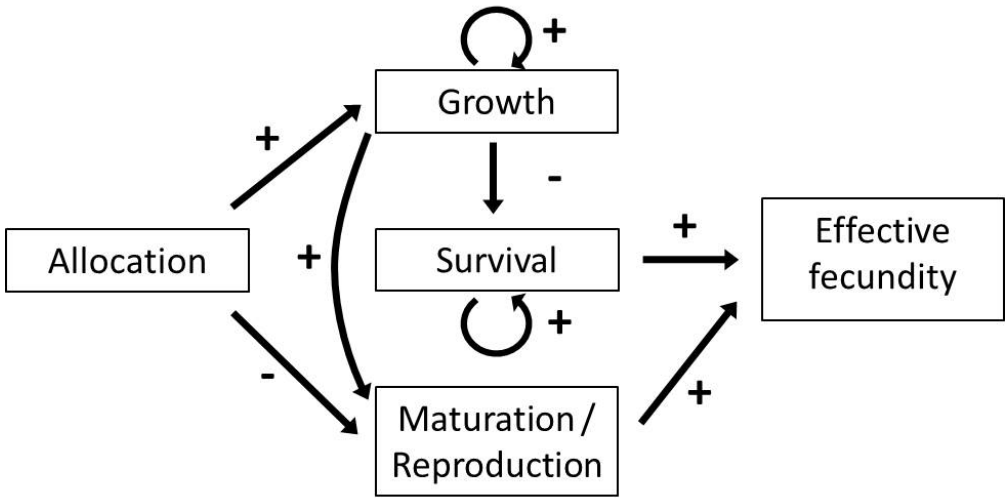

Figure 2.2: Diagramatic representation of the paths conveying the perturbation in the allocation function to model's state variables. The signs next to the arrows indicates whether increasing the value of a variable of the system (parameter or state-variable) increases or decreases the *rate* of change in another dependent variable.

Chapter 3

Unravelling the determinants of individual life history in the presence of ecological feedback: A sensitivity analysis for continuous-time models

3.1 Introduction

Individual life history relates how individual realized patterns of growth, survival and reproduction are expressed within an individual lifetime as a function of individual age or state (Roff 2002, Stearns 1999). Much interest has been placed in understanding the genetic and environmental determinants of these patterns (Noordwijk and de Jong 1986, Wade and Kalitz 1990, Falconer and Mackay 1996, Stearns 2000), as well as characterizing their ecological and evolutionary consequences (Metcalf and Pavard 2007, Coulson et al. 2010, De Roos and Persson 2013). Classical life history theory has been built from a static perspective, where the environment remains constant, or assume unchanging patterns of variability that do not depend on population genetic composition. Yet in many circumstances, changes in an organism's biology, such as those induced by the evolutionary process, entail changes in local ecological conditions.

Phenotypic evolution is primarily thought as the consequences of genetic changes that affect the internal functioning of organisms through a chain of physiological processes (Schlichting and Pigliucci 1998). Phenotypic changes also affect the functioning of ecological units, such as population and community processes, and these changes feed back on individual organisms to affect further patterns of phenotypic expression (Figure 3.1, Pelletier et al. 2009, Ellner et al. 2011). The ecological feedback loop hereby induced has consequences for predicting the effects of natural selection on patterns of phenotypic expression in natural setting

and the ecological effects of evolution, as well as for understanding how natural selection acts in these systems (Dieckmann 1997, Kisdi and Geritz 2010). These types of questions take particular importance at a time when ecologists and evolutionary biologist start realizing that evolution can act as fast a ecological processes (Pelletier et al. 2009, Ellner et al. 2011).

The ecological feedback loop is tightly linked to the concept of population regulation. Population do not grow without bounds, and this is achieved because density dependence acts on individual vital rates, such as fecundity and survival (Murdoch 1994, Turchin 1999, Meszéna and Metz 1999, Murdoch et al. 2003, Meszéna et al. 2006). A large body of ecological theory has focused on the mechanisms causing population growth to be regulated. In this context, understanding the mechanisms underlying patterns of phenotypic expression and vital rates is a fundamental question because it underlies virtually every aspect of ecological and evolutionary dynamics (De Roos et al. 2003).

In many circumstances, density dependence is not direct but mediated through relevant aspects of the environment, such as resource conditions. To understand the effects of variation in an organism's biology, one therefore needs to account for the modified patterns of interactions between individuals and their environments.

In the case of consumer-resource interactions, the feedback loop is made up by (i) the effect of consumer population on resource growth, which depends on the foraging pressure exerted by the consumers, and (ii) the effect of variation in resource density on consumer population growth, which occurs as a consequence of the resource dependence of consumer demographic rates. Understanding this interaction requires understanding the physiological intricacies mediating the individual energetics of acquisition, allocation and expenditure. The quantity of energy individual organisms can acquire through feeding is by nature limited. As a consequence, further partitioning of this energy among various physiological processes operates in a competitive fashion which limits the expression of individual phenotypic and life history traits (McNamara and Houston 1996, Zera and Harshman 2001, Roff 2002, Koiijman

2010). Physiological changes can affect all these processes simultaneously. It may therefore be difficult to apprehend how population regulation may be achieved and in what way perturbed patterns of individual growth, survival and reproduction contribute to that end.

Progress on these questions have been impeded because these processes operate dynamically, and most often, on a continuous time basis (Metz and Diekmann 1986, De Roos et al. 2008, Kooijman 2010) and their analysis requires the use of mathematical tools no readily available to the ecologist. For example, there is currently no tool to derive the selection gradient for models of individual energetics or to predict demographic and individual phenotypic changes under conditions of density dependence other than pure numerical simulations. Simulation studies are useful to characterize these effects, but their explanatory power on to what cause these changes to happen is limited because many of the intermediate processes underlying demography are affected simultaneously.

Analytical sensitivity analysis provides a way to characterize the effect of small variations in model parameters to the resulting outcome. Its explanatory power is greater than pure numerical simulations, because predictions are achieved by keeping track mechanistically of the effects of the perturbation along the different components of the model. This makes it possible to decompose easily the contribution of intermediate processes to an overall outcome. In an ecological context, it makes it possible to integrate information across biological scales. For example, it may be used to characterize how much phenotypic change arises from the direct effect of a perturbation in a genetic parameter and how much is induced by the environmental feedback loop. As another example, it may be used to decompose the contribution of perturbed schedules of fecundity, survival and development to the selection gradient, or to the reaching of population regulation.

Until now, the application of analytical sensitivity analysis in ecology and evolution has been almost entirely restricted to discrete-time models, and we presented a methodology for its application to ecological models specified in continuous time (Chapter 2). Our analysis

aims at applying this type of analysis to a model of physiological ecology and life history of a consumer species in order to draw implications for the ecological and evolutionary processes. We use the same model as in Chapter 2, which predicts individual lifetime patterns of growth, fecundity and survival based from the consideration of the mechanisms mediating individual energetics. The difference is that we now explicitly account for the mechanisms generating density dependence on individual vital rates, and explore how this incorporation modifies predictions.

In this chapter, we restrict our attention on equilibrium dynamics, because we are mostly interested on how the environmental variation induced by the feedback loop between consumer and their resources affects patterns of phenotypic expression. This issues has never been investigated before using sensitivity analysis, so we start with the simplest case, as an analysis under non-equilbrium conditions is likely to come with a burden of technical details that may obscur general conclusions from this analysis. The generality and limitations of these conclusions is considered in more details in the discussion.

3.2 Analysis of physiologically structured population at equilibrium

In chapter 2, our example involved the analysis of the consequences of a perturbation in a parameter on the physiological dynamics of an individual foraging under constant food conditions. In this context, parameter perturbation affects individual-level dynamics due to variation in the internal mechanisms underlying patterns of phenotypic expression. We will refer to these effects altogether as the *intrinsic* effects of the perturbation. In contrast, the effects arising from variation in resource density consequent to the ecological feedback will be referred to *feedback-mediated* effects.

In contrast to our previous analysis, the occurence of a feedback loop between consumer and resource dynamics requires the perturbation to be defined at a population level, not only at the individual level. Such kind of perturbation may, for example, result from an extrinsic

influence on the system such as an anthropogenic perturbation affecting individual survival (e.g. harvesting), or a change of mean temperature producing effects on physiological rates. It is also coherent with evolutionary interpretations, where the perturbation originates either from change in the mean genotypic value of a population, or from *both* the occurence of a mutation at an individual level, and the substitution of the genotype originally present in the population (resident) by the mutant one (invasion / replacement dynamics).

Because of this assumption, we need to raise our model for individual physiological dynamics (the i-state model) to the population level, and then assess the consequences of the perturbation on the interaction between the population and its environment. Raising the i-state model to a population level may come with its own difficulties (De Roos 1997, Diekmann and Metz 2010). In this paper, we restrict our attention to equilibrium demography, which makes things simpler, as the equilibrium distribution can be directly described from the i-state model (Gurney et al. 1996, Diekmann et al. 2003).

The model used in this example is thus the very same in structure as the one used in Chapter 2. It predicts both the physiological dynamics over an individual lifetime, as well as the equilibrium demography of the population. This close correspondence between individual dynamics and equilibrium demography is the source for many insights about the processes at play, but care must also be taken to keep in mind these two possible interpretations in order to keep a coherent reasoning.

To predict equilibrium demography, we need to add another ingredient to the model by imposing additional constraints on the dynamics in order to get an account of the ecological feedback and reach stationarity. At the level of the consumer population, the condition for equilibrium imposes that each individual produces on average one descendant during its lifetime. When there is no direct interaction between individual consumers, this can only happen if the resource density settle to the particular level $F = F^*$ verifying that condition (De Roos et al. 2010). Mathematically, F^* is therefore the particular food density verifying

that:

$$R_0(F^*) \equiv \int_0^\infty \beta(a, F^*) s(a, F^*) da = 1 \tag{3.1}$$

R_0 is the individual's lifetime offspring production, β is the fecundity rate of age a individuals, and s, their survival probability.

This relation implies that food density is not a parameter anymore, but instead, a variable which value is entirely determined by the consumer biology. Thus, we can write equilibrium food density as a function of the parameter(s) p underlying consumer biology: $F^* = F^*(p)$. Such a dependence highlights the coupling between consumer's individual-level dynamics and resource density, which is the source of the ecological feedback loop. To emphasis this state of affairs and the difference with the initial i-state model, we refer to the system now formed by the i-state model plus the condition for equilibrium, as the *coupled system*.

Since eq. (3.1) establishes the condition determining equilibrium food density, it also provides the ground for estimating the effect of parameter perturbation on food equilibrium density. Making R_0 a function of both the parameter p to be perturbed, and food equilibrium density, we get that:

$$\frac{dR_0(F^*(p), p)}{dp} = 0 \tag{3.2}$$

Applying the chain rule, we obtain:

$$\frac{\partial R_0}{\partial p} + \frac{\partial R_0}{\partial F}\frac{dF^*}{dp} = 0 \tag{3.3}$$

Thus,

$$\frac{dF^*}{dp} = -\frac{\partial R_0}{\partial p} \Big/ \frac{\partial R_0}{\partial F} \tag{3.4}$$

Without any loss of generality, these results could also be derived in term of the per-capita population growth rate, $r(F, p)$. One simply needs to replace R_0 by r in equations (3.2)-(3.4). The sensitivities of r can be obtained by differentiating the Lotka renewal equation

and evaluating the resulting expression for $r(F,p) = 0$ to get that $\frac{\partial r}{\partial p} = \frac{1}{T}\frac{\partial R_0}{\partial p}$, where $T = \int_{A_m}^{\infty} a\beta(a)s(a)da$ is the mean generation time.

Equation (3.3) is fundamental to establish the effect of the environmental feedback on demography and individual physiological dynamics. It states that, at equilibrium, any variation in R_0 induced by intrinsic changes in individual demography (first term on the LHS) is *exactly* countered by an effect associated with the ecological feedback loop (second term on the LHS). It is the dependence of individual-level processes on resource density that determines the appropriate adjustment in equilibrium resource density necessary to compensate for the intrinsic effect of the perturbation on R_0.

The effect of the perturbation on any variable X of the coupled system can be expressed as:

$$\frac{dX}{dp} = \frac{\partial X}{\partial p} + \frac{\partial X}{\partial F}\frac{dF^*}{dp} \tag{3.5}$$

This clearly distinguishes the effects of intrinsic physiological changes from those mediated by the environmental feedback. Note that the former kind of effects is the very same that those predicted from the perturbation analysis of the uncoupled i-state model.

We will examplify the consequences of these processes by applying sensitivity analysis of our example model.

3.3 Sensitivity analysis

The result of the sensitivity analysis of the coupled system are shown in Figure 3.2. The perturbed parameter is θ, the proportion of net production allocated to growth. The contributions of intrinsic changes, ecological-feedback mediated changes, and total changes are shown separately.

For the growth and survival curves, the environmental feedback either amplifies the effects of the perturbation, or has almost neglictible effects. On the other hand, its effect

on cumulative reproductive energy investment (R) is large, and may reverse that of intrinsic physiological changes depending on the value of θ. The last pannel of figures shows that the intrinsic and feedback-mediated effects on effective fecundity (B) indeed cancel each other out eventually. In all cases, the (positive) perturbation in θ always results in an earlier schedule of reproduction, which may seem counter-intuitive, and contrasts with the case with no environmental feedback.

A conclusion that can be drawn from these figures is that, even though the effect of the environmental feedback on R_0 is fully constrained by the requirement of matching the intrinsic effect of the perturbation (eq.(3.3)), its effect on the intermediate phenotypic and demographic rates leading to this state of affairs can be of any kind: negligible, amplifying, buffering, or reversing that associated with intrinsic organismal changes. Figure 3.3 illustrates this variety of effects still further, by showing the contributions of intrinsic versus feedback-mediated effects to the total variation of various demographic statistics and phenotypic traits in different regions of the parameter space. One can see that the contribution of the two kinds of effects are generally opposite, and the prevalence of one or the other is balanced, depending on the trait being looked at and the region of the parameter space.

3.4 Discussion

3.4.1 Individual dynamics and environmental coupling

Our study aims at providing further clarifications about the contribution of the ecological feedback to the expression of phenotypes and its consequences for the demographic process by looking in depth at the individual-level processes mediating the interaction between organisms and their environment. In this prospect, equations (3.3) and (3.5) are very useful because they explicit the nature of the elements involved in this interaction. Their derivations result from a simple recognition of the dependence of R_0 on consumer biology and environmental conditions (Mylius and Diekmann 1995), and an application of the chain

rule. It is essentially similar to those carried out by De Roos (2008), which applied it to the Lotka renewal equation for analyzing density-independent models and derived the sensitivity of the intrinsic growth rate. Here, we show that the application of these principles to density-dependent models yields powerful insights on the mechanisms mediating phenotypic expression and population regulation, which we will discuss below.

These equations show that phenotypic and demographic changes can be compartementalized into the contribution of three major components: the "intrinsic" changes in individual life history, variation in ecological conditions, and a plastic response to these changes ($\frac{\partial X}{\partial p}$, $\frac{dF^*}{dp}$, and $\frac{\partial X}{\partial F}$, respectively). Though these equations are derived under a restricted set of assumptions and formulated in term of perturbation analysis, they carry more generality and may be interpreted phenomenologically as a decomposition of phenotypic changes into intrinsic and plastic components, with concomitant changes in ecological conditions.

The effects of the ecological feedback come as a consequence of imposing constraints on population growth, such that long-term population dynamics are regulated. The primary implication is that the effect of the ecological feedback to the overall demographic performance of individuals (i.e. R_0) buffers that of the original perturbation: it has the exact same magnitude but an opposite direction. This also defines the effect of the perturbation on the environmental component: both the intrinsic effect of the perturbation and the plastic response to the environmental conditions are already fully constrained by intrinsic properties of the individual (i.e. the structure of the i-state model), the condition for long-term stationary dynamics is thus reached through an adjustement of the ecological variable.

This fundamental constraint does however not necessarily hold for the other phenotypic and demographic traits involved in the process. Nonetheless, because of this final result, we may still expect the ecological feedback to be of the same importance than that of the intrinsic effect of the perturbation in determining total phenotypic changes in the population, and most often, to act in an opposite direction. In part 3.3, we confirmed this inference by

studying in details the relative contribution of both these effects on the traits involved (Figures 3.2 and 3.3). This analysis also showed that, even though these effects often act in an opposite direction, all possible scenarios are possible, and the ecological feedback can either buffer, reverse, or amplify the intrinsic effect of the perturbation.

3.4.2 Beyond the stationarity assumption

Because we focused on the role of the ecological feedback, we considered exclusively variation in the ecological component underlying this feedback, and assumed stationarity of the process with associated steady-state dynamics. The prevalence of equilibrium demography in natural systems may certainly be deemed circumscribed (Hasting 2004, 2010), so we discuss this assumption first, argue that it does not impede much on the generality of the results discussed below, and propose solutions to move beyond that assumption.

First, population regulation does not necessarily implies equilibrium dynamics, it only needs population abundance to remain bounded over time. In any circumstance, if population growth is effectively regulated, an ecological feedback loop must still be at play for population abundance to remain bounded over the long term (Murdoch 1994, Turchin 1995, Murdoch et al. 2003). In particular, the general result that any variation in demographic performance induced by a perturbation must be eventually compensated for through the ecological feedback still holds. In other words, an equation similar to (3.3), integrated over an appropriate period of time, still holds under non-equilibrium situation (e.g. one period for cyclic dynamics, its limit toward infinity for chaotic dynamics). Admittedly, the details as to how and when regulation kicks in may vary and the mathematical analysis more complex.

Secondly, these results are about the *outcome* of the process. There is no explicit account for the dynamics happening between the time the perturbation is applied, and the time at which the adjustement of ecological conditions is reached. These details are certainly of importance for understanding the contemporary dynamics of ecological systems (Hasting 2010). Though we only looked at what happened once stationarity was reached, it is only

an assumption specific to the focus of our study and does not constitute a limitation of the general methodology presented. These transient dynamics can be fully accounted for by including a model for the dynamics of the ecological variable. For example, these dynamics could be modelled by assuming some function for resource production (e.g. logistic) and accounting for the effect of consumption on resource density.

3.4.3 Implications for life history theory

Life histories evolve in the presence of ecological feedback (Bassar et al. 2010, 2012, 2013, De Roos and Persson 2012, Travis et al. 2013). Arguably, the main result of this study in this respect, is that ecological feedback-mediated effects bring about as much changes on patterns of phenotypic expression than genetic changes do. Predictions on patterns of phenotypic evolution that omit this inclusion may therefore be misleading and be the source for mismatches between the phenotypic and demographic characteristics expected to emerge from the evolutionary process, and observations of organisms' biology in the wild. This mismatch may be thought as the difference between two qualitatively different measures: what natural selection *favors* versus what natural selection *results in*. In an evolutionary context, the term $\frac{\partial R_0}{\partial p}$ can be interpreted as the selection gradient, as it reflects the variation in population growth consequent to a small genetic change in individual physiological parameters (Lande and Arnold 1983). This measure relates to the life history that is selected for or against, and is only expressed during the early stages of the selective processes. However, in the case these characteristics are actually selected (i.e. become the resident strategy or the mean genotypic value in the population), it becomes clear from equation (3.5) and the examples shown above that, in general, these are not the phenotypic and demographic characteristics that will eventually be expressed by the individuals. Clearly, *when the focus is at a phenotypic level, what natural selection favours, and what natural selection results in are two very different things*. Both measures may lead to contrasting and sometimes opposite predictions, because in numerous instances, the effect of the environmental feedback may be

stronger. As the effect of the environmental feedback is defined in a way that oppose the effect of natural selection, it has often a buffering effect on the variation induced by natural selection on phenotypic and demographic components, particularly those closely related to fitness. This may be a reason for instances of selection that remain undetected in the wild, and provides further grounds for speculating that rapid evolution may be more prevalent than expected from the observation of phenotypic changes alone (Ellner et al. 2011, Palkovacs et al. 2012).

Previous analyses have been developed to partition the phenotypic variation following environmental changes into the contribution of physiological, ecological and evolutionary components (Collins and Gardner 2008, Ellner et al. 2011). Our approach differs from these previous works in that environmental change is not considered as given here, but is instead a consequence of combining genetic changes in the focal population with a feedback loop between individuals and their environment. Additionally, our framework of analysis investigates the effect of genetic variation on the whole age-dependent trajectories of individuals' life history, rather than single valued traits. Interestingly, both these analyses also found that ecological and evolutionary opposed each other and were often similar in magnitude. The context of their studies is slightly different, because these authors mostly considered case scenarios where the environmental perturbation originated first, and evolution followed, whereas we considered the opposite. Our analysis is nonetheless supportive of these results, and our analysis strongly suggests that these findings may be the consequences of the ecological feedback loop that cause population to be regulated, because, irrespectively of the nature of the perturbation affecting population demography, other mechanisms need to act in an opposite way to limit population growth and maintain their persistence.

3.4.4 Implications for population persistence and regulation

Our results are not only relevant in an evolutionary context. The perturbation may originate from an extrinsic influence on the system. This could, for example, reflect an increased mortality due to harvesting, or a contrario, an effect following the application of conservation measures. In many circumstances, human-induced perturbations act negatively on population growth, and thus, the ecological feedback, which acts in an opposite direction, may prevent population from extinction by improving (some aspects of) the environmental conditions. Basically, similar predictions can be made from even the simplest consumer-resource models (e.g. Lotka-Volterra like models), though they lack the necessary realism to make predictions on individual life histories. Compensatory response of life histories to perturbations have been documented empirically (Metcalfe and Monoghan 2002, Moe et al. 2002, Schröder et al. 2009, Nilsson et al. 2010, Ohlberger et al. 2011).

The mechanisms underlying the population feedback on individual realized life histories may therefore have an important role for population persistence. At an individual level, population persistence relies essentially on the plastic phenotypic response following the adjustement in the ecological variable. Nonetheless, in contrast to the commonly held view, the plastic response may not be qualified as adaptive per-se, because it is dictated by the physiological mechanisms driving the response of individuals to their environments. None of these two mechanisms involved bear any relation to the adaptive process, and they may be more adequately interpreted as components mediating the resilience of ecological systems.

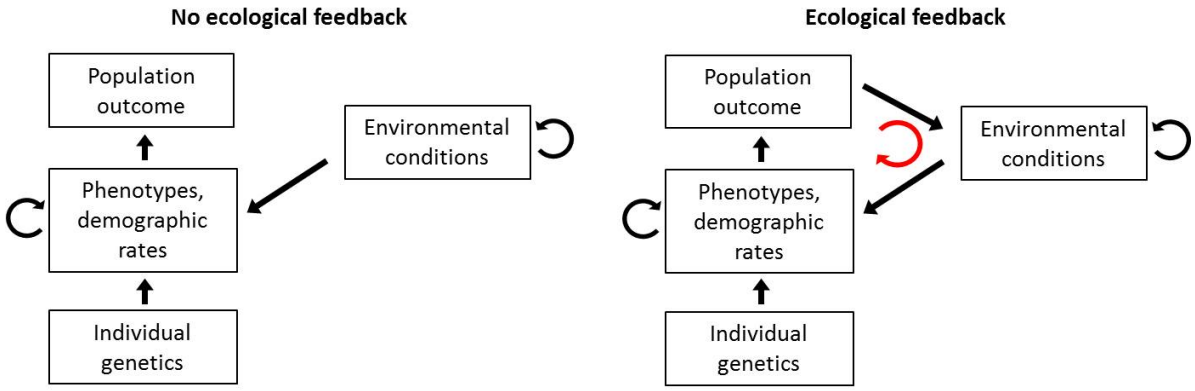

Figure 3.1: Diagrammatic representation of the interdependency between individual genetics, phenotypes, environment and population processes. Individual expressed phenotype is the product between the genetically based components mediating the individual physiological response to their environment, and the state of their environment. The left pannel shows what happens in the abscence of ecological feedback and the right pannel what happens in its presence. The ecological feedback loop is represented by the red arrow: individual expressed phenotypes condition the population process which contributes to environmental dynamics to eventually feed back on patterns of phenotypic expression.

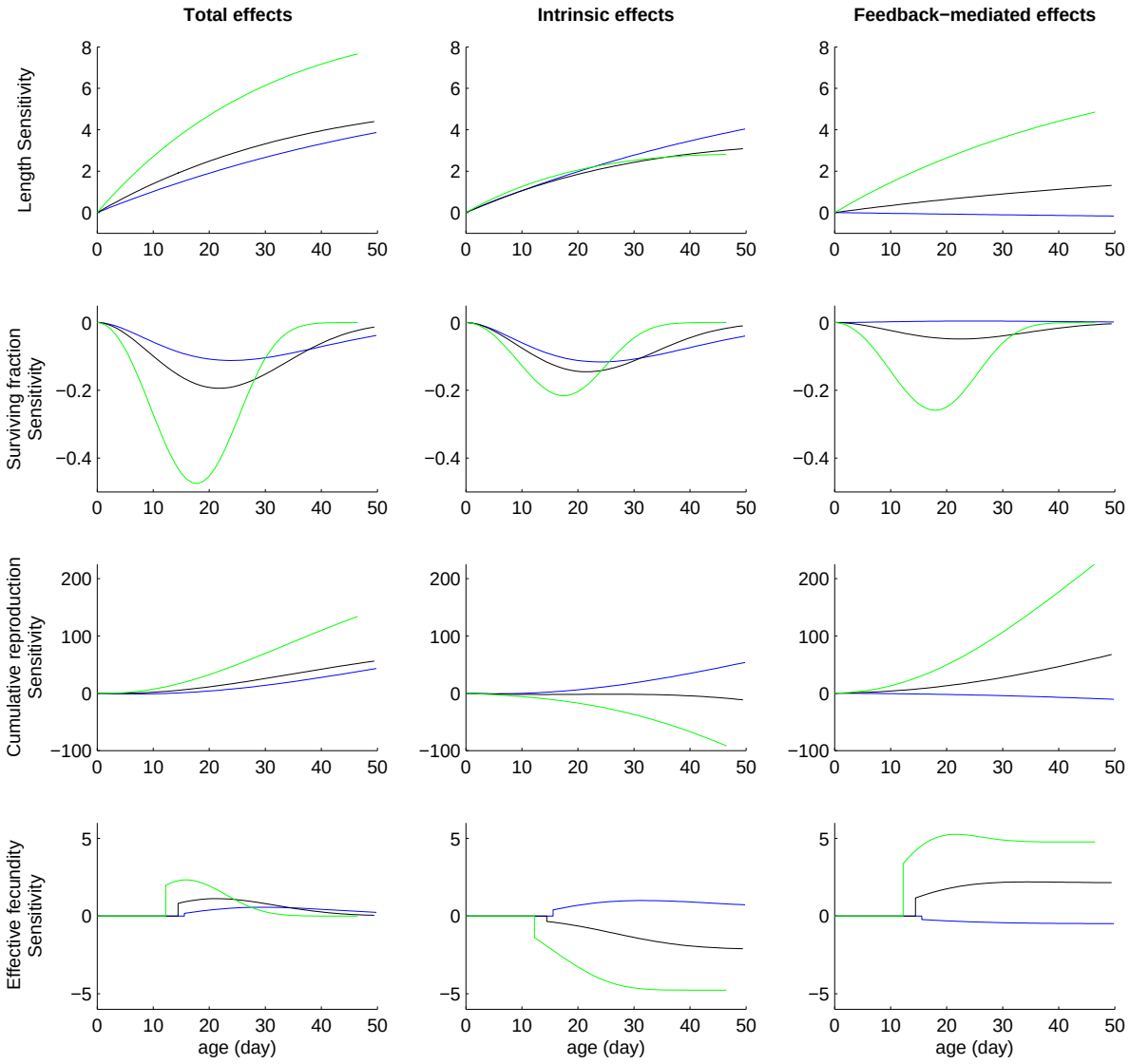

Figure 3.2: Sensitivities of the model state-variables to perturbation in θ, the fraction of the net production allocated to growth. The different columns show the total, intrinsic effect and feedback-mediated effects. Blue: $\theta = 0.2$; Black: $\theta = 0.4$; Green: $\theta = 0.6$.

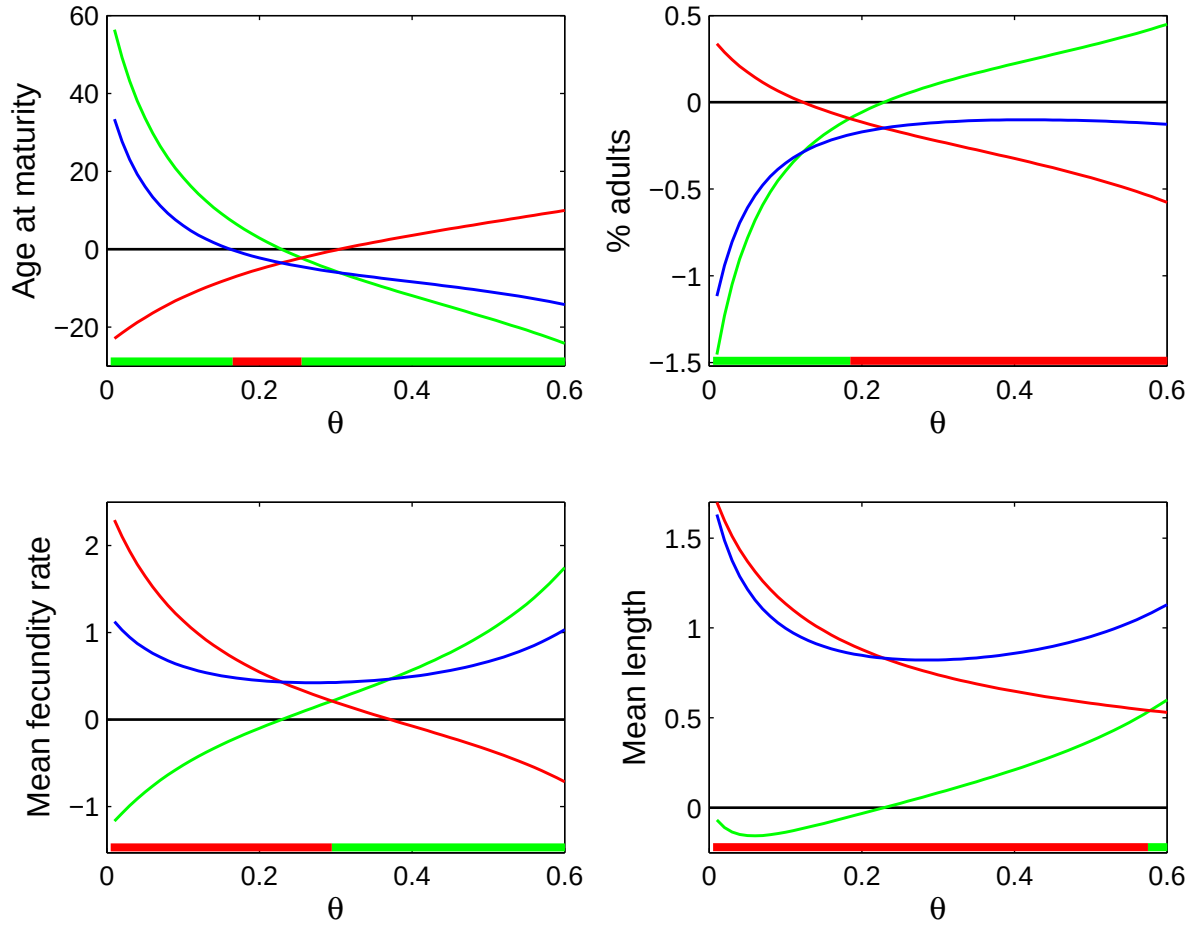

Figure 3.3: Contributions of intrinsic physiological changes (Red) and environmental feedback (Green) to variation in some demographic statistics and phenotypic traits as a function of θ, the fraction of the net production allocated to growth. The blue line represents the total effect of the perturbation. The bars at the bottom of each graph indicates which of the two effects contributes the most to total change. Note that the sign of $\frac{dF^*}{dp}$ changes from negative to positive around $\theta = 0.23$, which is always associated with a change in the sign of the contribution of feedback-mediated effects.

Chapter 4

Genotype-environment interactions at a population level: The expression of genetic variation depends on the patterns of environmental fluctuations

4.1 Introduction

Understanding how organisms and populations respond to environmental variability is a fundamental question in ecology. Ecological interactions, such as consumer-resource or intraspecific competitive interactions, can generate various kinds of cycles, and therefore have implications for understanding the dynamics of natural systems (Murdoch et al. 2003, De Roos and Persson 2013). The variation generated by these cycles may affect ecological rates at the individual level to generate qualitatively new phenomena compared to constant food conditions. For example, decreasing food levels may induce major change in reproductive allocation and trigger starvation (Kirk 1997, Peeters et al. 2010). The energetics of individual feeding, growth, fecundity and mortality is one of the cornerstones that makes it possible to establish a connection between the organisms' food environment and their demography (Gurney et al. 1996, Nisbet et al. 2010, Kooijman 2010). Yet research that focused on linking individual energetics with demographic patterns has been dominated by experiments on individuals subjected to constant food conditions (Chapter 1). Inferences to the population level are then drawn using a modeling approach under the assumption that energy allocation rules and functional dependency of vital rates on resource conditions and individual state remain constant while the conditions otherwise vary (Nisbet et al. 2000). This assumption has rarely been tested and may explain some of the limitations of this approach to predict and understand non-equilibrium dynamics.

Natural systems also contain large amounts of genetic variability which may affect population and higher-level ecological processes (Mousseau et al. 2000, Nevo 2001, Petchey and Gaston 2006, Hughes et al. 2008). There has been a recent surge of interest for characterizing the effects of intraspecific genetic variability on ecological functioning, and studies have looked at its effects on processes such as biomass productivity, response to disturbances, or fluxes of energy and nutrients, see for example reviews (Petchey and Gaston 2006, Hughes et al. 2008). For example, it has been demonstrated in a poplar system that the genotype of individuals impacts the strength and nature of trophic interactions with birds, insects and beavers; the composition of the arthropod and microbial community hosted; and organic decomposition and nitrogen mineralization (Whitham et al. 2003, 2006, 2008). Despite some differences, many of the effects of genetic diversity parallel those of species diversity, and studies have shown that intraspecific genetic variability can have as much effects on ecological functioning as interspecific variability does (Hooper et al. 2005, Petchey and Gaston 2006, Hughes et al. 2008). This interest for the effect of intraspecific genetic diversity has been further enforced with the recognition that ecological and evolutionary processes can act on the same time scale and generate eco-evolutionary dynamics (Pelletier et al. 2009, Ellner et al. 2011, Schoener 2011).

The effects of genetic variability are likely to depend on the environmental context, i.e. genetic and environmental factors can interact to generate genotype $\times$ environment interactions (Roff 1997). Genotype $\times$ environment interactions have been the topic of many studies which focused on their effects on individual phenotypic expression and their implications for the evolutionary process (Gillespie and Turelli 1989, Via and Lande 1985). Nonetheless, individual phenotypic response is also a major feature conditioning ecological processes, and this type of interaction has consequences for the functioning of ecological units (Miner et al. 2005). These consequences of genotype-environment interactions remain poorly characterized. This is even truer when dynamical variation in the environment is considered (Miner

and Vonesh 2004, Miner et al. 2005). Only a handful of studies have looked specifically at how dynamical variation in the environment (in contrast to variation in mean environmental conditions) affects individual-level phenotypic response, and still fewer have looked at genetic variation in this response (Siems et al. 1998, Miner and Vonesh 2004, Engelmann and Schlichting 2005, Schoeppner and Relyea 2008, Rodriguez 2012). Studies using natural setting include dynamical variation in the environment, but do rarely make a specific or mechanistic account of the key environmental features affecting individual phenotypic expression, because the number of factors potentially involved is usually far too great. By contrast, laboratory experimental works allow for the control of both genetic and environmental factors. These studies used almost invariably constant environmental conditions (Chapter 1). Thus we still lack as of today comprehensive studies.

Our study aims at characterizing how genetic variability interacts with dynamical features of the environment to affect the demography of a consumer species using experimentations and modeling on a number of monoclonal *Daphnia*-algae systems. Population response to its environment is largely conditioned by genetically-based mechanisms of energy acquisition and expenditure, which govern how an individual grows, reproduces and dies (Nisbet et al. 2000, Kooijman 2010). In order to clarify how genetic variation in these mechanisms affects the response of populations to their food environments, we used a series of experiments involving different clonal populations of the Cladoceran *Daphnia pulicaria*. The dynamics of *Daphnia*-algae systems are well known for being driven by consumer-resource interaction. In particular, this interaction generates various kinds of population cycles (McCauley and Murdoch 1987, McCauley et al. 1999, 2008). A fundamental property of these systems is that the dynamics of both population are interlocked through a feedback loop: consumer foraging activity affects the dynamics of the resources, and resource dynamics affect the dynamics of the consumer population. While primordial for understanding the dynamics of these systems, the feedback loop induced by the interaction represents an impediment for characterizing

the response of the consumer population to their food environment, because this latter is not under full experimental control. Allowing for the consumer-resource interaction to take place would imply that different consumer populations are not subjected to the same environmental conditions. Therefore, we subjected the populations to carefully controlled variations in food supply in order to decouple the dynamics in food production from the consumer population demography and control the level of variation in the food environment. This allowed us to make direct comparisons as to how different clonal populations respond to the same patterns of fluctuations in their food environment. Indeed populations are monoclonal, so that the genetic basis conditioning individual and population response to their environment is genetically-based and remains the same across different environmental conditions. In these experiments, food was supplied either in a constant fashion, or in fluctuating fashions, in a way that mimics both large and small amplitude cycles that can be observed in *Daphnia* population dynamics (McCauley et al. 1999). Moreover, these cycles are produced by different mechanisms: small-amplitude cycles result from competitive interactions between differently sized individuals, whereas large-amplitude cycles result from the instabilities inherent to consumer-resource interactions (McCauley et al. 1999, 2008).

Individual energetics are likely to depend on individual physiological states and on the feeding conditions they experience, resulting in a substantially complex interplay between individual and population processes. In particular, the functional dependency of individual performances on these states has been shown to be critical in mediating patterns of population variation (De Roos et al. 2003, De Roos and Persson 2013). To bring further clarifications on how these mechanisms interact with genetic variability, we carefully parameterized models for each clone directly from population data. This type of approach, known as inverse analysis, present a number of advantages compared to a full model parameterization from individual data only. The measurement of key processes rates from individual experiments is indeed a tedious task, stretching over extended periods, which makes it a

possibility for only a few model organisms. In addition, getting a sufficiently comprehensive and accurate account on these processes is a challenge in itself. By contrast, inverse approaches allow the data to 'speak for themselves'. Despite this advantage, non-trivial issues are associated with the approach. Most notoriously, these problems are often of an underdetermined nature: many competing hypotheses on underlying individual processes can fit a particular set of data equally well (see e.g. Wood 2001, Nelson et al. 2004). Fortunately, statistical solutions have been proposed to overcome this problem (Wood 2001, Nelson et al. 2004). In this study, we make use of an experimental design that enables the implementation of successive steps of model selection and independent validations. Predictive ability of the models is thus assessed with respect to conditions that are not only independent, but also qualitatively different from those that have been used for parameterizing them.

4.2 Methods

4.2.1 Experimental methods

We used a series of experiments involving four clones of *Daphnia pulicaria* sampled from shallow lakes in Alberta, Canada. We confirmed these clones to be genetically different by sequencing 8 microsatellite markers. Details on the microsatellite analysis are given in chapter 5. We then quantified the demography of clonal populations separately grown in a set of controlled food environments. Populations were fed specified quantities of the green alga *Chlamydomonas reinhardtii*. These experiments involved constant food supply (CF), followed by starvation, called later starvation data; and small and large amplitude (SA and LA) fluctuations in food supply, detailed below.

Algae was grown separately in axenic cultures using COMBO medium (Kilham et al. 1998). Algal concentration was estimated by diluting two replicate samples in lugol's solution and counting 16 squares in a Sedgewick-Rafter counting chamber. Cell concentration was converted to carbon concentration assuming a carbon content of 2.0 μgC.cell^{-1} (Porter et

al. 1982).

In all experiments, *Daphnia* populations were reared at 22 °C in the dark, in order to inhibit algal growth. Populations were censused by transferring individuals on a 35 μm mesh, counting and sizing them under a dissecting microscope. Seven size classes were used (I: <0.8mm; II: 0.8 - 1mm; III: 1 - 1.2mm; IV: 1.2 - 1.4mm; V: 1.4 - 1.6mm; VI: 1.6 - 2mm; VII: >2mm). Individuals in class VII rarely exceeded 2.5 mm. Individuals greater than 1.4 mm were considered as adults. The number of eggs in their brood pouch were counted as well. After the census, populations were placed in new medium and fed algae, as specified below.

In CF experiment, populations were reared in a 275mL volume, censused and fed 3 times a week, on Monday, Wednesday and Friday, for a quantity equalling 1.5 mgC/L/day. Two to four replicates per clones were used. Total population biomass was considered stable when the biomass growth rate remained $\leq 1\%$ per day for at least 7 days. Feeding was then stopped in order to induce starvation. Populations were then censused every day, and replaced in fresh medium, until the last individual died.

In the SA and LA experiments, populations were reared in 800mL volume. They were transferred in new medium and fed every day. Food amounts ranged from 0.3 to 1.5 mgC/L/day in SA experiments, and from 0.3 to 5 mgC/L/day in LA experiments. The paterns of variation chosen roughly reflect those seen in large and small amplitude cycles observed in coupled algae-*Daphnia* systems (McCauley et al. 1999). More details on these specific feeding patterns are given in the appendix (Figure A.1). Populations were censused twice a week, and 3 replicates were used for each clone.

4.2.2 Modeling and parameter estimation

One way to circumvent the problem of underdetermination is to use independent validations, which tests how well a model can predict experimental data that are independent from the ones used for its parameterization. Having in our hands experiments in CF, SA and LA

environments, and data on starvation, we used the following strategy to parameterize and validate the models:

First, we fitted models for each clone to the data from the CF experiments, assuming that no starvation mortality occurred. For each clone, we obtained a number of different models that led to equally good fits of the data. These models were obtained using either (i) different model structures: we used a range of models that varied in their complexity and level of details on *Daphnia* biology. We report here only the final form selected for these models, as other, simpler, models were enable to predict well the dynamics under fluctuating food treatments. (ii) The likelihood function usually presented a number of local minima that led to equally good fits (ΔAIC$<$2), which we found by using different initial conditions. (iii) Given the uncertainty on the expected distribution of errors associated with this type of data, we also fitted these models using various criteria for optimizing model fits. The technical aspects on the fitting procedure are described in details in the section *Model fitting and objective function*.

Second, we parameterized a starvation mortality function from the starvation data.

Third, we compared the ability of the models previously obtained to predict the dynamics in the LA experiments, and selected the best model for each clone. We chose to do so rather than fitting all the data at once, as this allowed for an independent *test* of the robustness of the model inferences obtained. The set of data used for this test is thus not only independent from the one used for their parameterization, but it is also the demographic product of populations subjected to qualitatively different resource dynamics. This left us with four models, one for each clone.

Finally, we used the data provided by the SA experiments as a means to validate the estimates selected for each clone. For the purpose of this study, we needed models that not only capture well the dynamics, but also reflect clonal differences. However, these differences being relatively small, we found that the dynamics of a given clone were sometimes predicted

better by the model parameterized for another clone. When that happened, we rejected the parameterization, and came back to step one in order to refine those estimates, and proceeded with subsequent steps again. We considered our estimates as final only once the parameterization for each clone provided the best match with its own dynamics in SA experiments.

Models derivations

To capture subtle differences in clonal biology, we wanted to use a model that adheres as much as possible to what is known about *Daphnia* biology, and that included mechanistic details on *Daphnia* physiological ecology. Some functions of the model were also represented non-parametrically, using cubic spline functions, in order to release the structure of the model from too restrictive assumptions on *Daphnia* biology (see Wood 2001 for a discussion on semi-mechanistic models in ecology). We first sketch out some basic elements of the models. Details requiring specific attention are dealt with individualized headings. The resulting model and parameter values and range are given in Tables 4.1 and 4.2.

Three state variables are used to represent an individual : L, its length (mm); W, its weight (μgC); and W_r, the cumulative mass of carbon invested to reproduction (μgC). In addition, other variables of the model include cohort abundance n and food density $F(\mu\text{gC/L})$. Since the experiments involved transfer cultures, F is reset at a frequency given by the experimental design, and only declines in between two transfers, due to *Daphnia* feeding.

Daphnia individuals have a type II functional response, with a constant half saturation constant (F_h) and a maximum ingestion rate that increases with individual length $I_m(L)$. We used the relation reported in McCauley et al. (1990). We assumed the assimilation efficiency ϵ_a to be constant. Metabolic maintenance rate is given by a function $M = m(L)W$. We used a 3-knots spline function to parameterize $m(L)$. We assumed that individuals were subjected to a level of mortality independent of food, but dependent on individual length

($\delta_i(L)$). That function was again parameterized with a 3-knots spline function.

Growth, reproduction and allocation

Though models on *Daphnia* usually assume simpler forms for the growth and fecundity function, we found that the inclusion of the details regarding molting described below was critical to obtain predictions that compare well to the experimental data of the fluctuating food treatments, particularly with respect to the timing of reproduction.

Growth Individuals grow in length by discrete increment every T_m days, typically 1.5-3 days (McCauley et al. 1990). Under non-starving conditions, the length reached at the time of the molt is typically food-independent. The relation between individual weight and length at that time is well described by an allometric function: $W_l = \xi L^q$ (Lynch 1989). We call this particular weight the nominal weight-for-length of an individual, and its reciprocal $L_w(W)$, the nominal length-for-weight.

There are several scenario to consider for calculating the growth increment between two molts $\Delta L = L(t) - L(t - T_m)$. Under good feeding conditions, $L(t - T_m)$ and $L(t)$ are given by the nominal length-for-weight relationship. In contrast, under poorer food conditions, an individual weight may be less than its nominal weight-for-length at any of these times. An individual cannot shrink however, so its length remains unchanged. This results in four combinations for the growth increment that can be accounted for using the following relation:

$$\Delta L = \max(L_w(W(t)), L(t)) - \max(L_w(W(t - T_m)), L(t - T_m)) \tag{4.1}$$

To make this results usable in the continuous time framework in which our model is set, we use an approximation that makes individual length continuously updated according to the mean growth increment per unit time:

$$\frac{dL}{dt} = \frac{\Delta L}{T_m} \tag{4.2}$$

Egg production During the intermolt period, adult females also allocate some of the energy acquired toward egg production. New eggs are formed in female's ovaries at the begining of a molt, and packed with nutrients some time before the following molt (McCauley et al. 1990). At molting, these eggs are transferred into the brood pouch, where they incubate and develop until the mother molts again. This implies that the energy allocated to reproduction during an instar is not released directly at the following molt, but only after a subsequent second molt.

Denoting W_r the cumulative mass of carbon invested toward reproduction, and applying again a continuous-time approximation, the mean rate at which a female releases eggs is:

$$\beta(t) = \frac{W_r(t - T_m) - W_r(t - 2T_m)}{(1 + \gamma)W_b T_m} \tag{4.3}$$

where W_b is the mass of a juvenile at birth, and γ is the overhead cost of egg production.

Allocation function to reproduction There has been a multiplicity of model for representing the allocation function. They either assume a direct dependency on energy fluxes, in the form of net production or net assimilation models, a dependency on individual states only, or both (see Nisbet et al. 2004 for a discussion).

Under sufficiently good and constant food conditions, individual weight varies smoothly and remains over the nominal weight for length between two molts. In contrast, if food conditions vary, the relation between an individual weight and its length may be much more variable. Since a weight below W_l can only happen if net energy balance remains negative for some time, we can use this fact to define a measure of individual condition. We define the relative weight-for-length as the ratio between an individual weight and its nominal weight-for-length: $\omega = W/W_l(L)$ so that a value smaller than 1 indicates nutritive stress, whereas good feeding conditions result in values that are greater or equal to one. Taking these elements together, we modelled the rate of energy allocation to reproduction assuming dependence on individual size, condition and food conditions, resulting in a function of the form: $\Theta(t) = \alpha_0 f_W(W, \alpha_1) f_\omega(\omega, \alpha_2) X(t)$.

$X(t)$ reflects the influence of current food conditions on allocation. We tried three possibilities, where $X(t)$ was either the net acquisition rate, the net production rate, or was simply set to 1. This last possibility, which makes the allocation process to depend on individual states only, did not prove fruitful with this framework. The other terms reflect the dependency of allocation on individual states. f_W reflect the dependency of allocation on individual weight, and the f_ω, on its relative weight-for-length. We scaled these functions between 0 and 1 over the intervals $[W_m\ W_{sup}]$ and $[\omega_0\ \omega_{sup}]$, respectively. The lower bounds are biologically meaningful: individuals start to allocate toward reproduction only once they reach a specific weight $W_m = W_l(1.4)$, which reflects the onset of maturity, and they stop allocating whenever ω is smaller than $\omega_0 = 1$. The upper bounds are used to reduce multicolinearity in parameter estimation, but the specific values chosen are of no consequence. For the functions f_W and f_ω, we used either power functions or exponential functions:

$$f(x) = a(w - b)^{\alpha_i} \tag{4.4}$$

$$\text{or} \quad f(x) = ae^{\alpha_i x} + b \tag{4.5}$$

where x is either W or ω. The parameters estimated are $\alpha_i = \alpha_1$ or α_2, whereas a and b are chosen appropriately to produce the scaling discussed above.

The starvation function

We assume that individuals are subjected to a mortality rate due to starvation that decreases with decreasing ω, following:

$$\delta_s(L) = \max(0, \alpha(L)e^{-\beta(L)\omega(t)} - \delta_0(L)) \tag{4.6}$$

$\alpha(L)$ and $\beta(L)$ are splines functions with 3 knots and $\delta_0(L)$ is an offset in starvation mortality rate. The offset takes either the value of a constant δ_c to be estimated, or $\delta_0(L) = \alpha(L)e^{-\beta(L)}$ if $\delta_c < \alpha(L)e^{-\beta(L)}$. This condition insures that individual do not starve as long as $\omega > 1$. The resulting function is given by $\delta_0(L) = \max(\delta_c, \alpha(L)e^{-\beta(L)})$.

We also assumed a food-independent component of mortality, and the dynamics in $\omega(t)$ depend on the basal metabolic rate. Both are estimated from the CF experiments. The parameterization of the starvation function is therefore dependent on the estimates from CF experiments.

Numerical simulations

We implemented the model using the escalator boxcar train (EBT) algorithm (De Roos 1988). An advantage of this method is that does not make any assumption about the nature of the model for the dynamics in individual state other than requiring that all individuals born at the same time behave in exactly the same way. As far as we know, the combination between the EBT algorithm and delay partial differential equations as never been used before. We therefore carefully checked for any sign of inadequacy from the model outputs, but none was apparent.

Model fitting and objective function

The demographic models generate predictions on the abundance of individuals in each of the size-classes used in the experiements, and on the number of eggs they carry. We derived a criterion for optimizing model using nonlinear regression, based on the likelihood of the multivariate normal distribution (Bates and Watts 2007) in combination with data transformation in order to improve normality and stabilize variance. Our strategy consisted in trying a set of transformations, involving power transforms ($g(x) = x^{\lambda}$), as well as the inverse hyperbolic sine function ($g(x) = \sinh^{-1}(\lambda x)/\lambda = \log(\lambda x + \sqrt{\lambda^2 x^2 + 1})/\lambda$). This latter transformation function has been showed to be more powerful than the former when assessed (Burbidge et al. 1988). It tends toward the identity function as λ tends toward 0, and to a log-transformation for large values of λx. An important strength of this transformation lies in the way it handles zero values, which allows for the likelihood function to be defined *on the original scale of measurement*, and hence directly select for the optimal value of λ in the optimization process.

We used this property to produce an objective criterion for assessing the match between model predictions and data of fluctuating food treatments, i.e. for the steps of independent validations. More specifically, we applied an inverse hyperbolic sine transformation on each category of observation and selected the λ values that maximized the likelihood. This produced likelihood values for each model that can be directly compared to each other.

All the estimates were obtained using global optimization algorithms (the direct search algorithms from matlab (Mathworks) global optimization toolbox), and were further refined using standard gradient search method (the fmincon function from matlab optimization toolbox)

4.3 Results

Our experiments produced a large volume of data, involving the response of different demographic variables to different food regimes for different clones. We organize these results by presenting how the different clones respond to the different food regimes. Each part contains a discussion of the data first, and then a discussion on the agreement between models and data.

4.3.1 Constant food supply

Figure 4.1 shows the trajectory of juvenile, adult and egg biomasses over the course of the experiment. Juvenile biomass first showed a very strong increase, generally followed by a more or less accentuated decline. Adult biomass followed a more steady increase until it stabilized. Most populations exhibit an early peak in fecundity followed by a period of lower fecundity. There was some variation in the timing and intensity of this peak among clones, and some populations also showed a second fecundity peak. By regressing biomass growth rate on total population biomass, we estimated that population reached steady values at 3.9 mgC/L for clone A; 3.2 mgC/L for clone B; 2.8 mgC/L for clone C; 3.2 mgC/L for

clone D. Moreover, over the course of the experiment, the biomass of clone A was almost always greater than that of the other clones, whereas the opposite was true for clone C. This variation in total population biomass seems to be mostly linked to variation in juvenile biomass which exhibit strong differences among clones (Figure 4.1). The differences in adult biomass trajectories among clones are much less pronounced. Regarding population structure (Figure 4.2), it remained relatively stable after the growth phase, and adults made up most of the total population biomass, typically around 60-80%.

Models did a fairly good job at capturing patterns of variation in juvenile and adult biomass, and fecundity (Figure 4.1). Biomass levels tend however to be slightly underestimated. Recall that the data presented are made up from a number of sub-categories, 7 size classes and 3 fecundity classes. When there is a consistent bias in the estimation of a particular category (e.g. juvenile biomass is consistently underestimated in clone A past day 15), the fit of these subcategories is more balanced, where a mix of over- and under-estimation is present (Figures A.2-A.5). Nonetheless, the models correctly predict the relative compartmentalization of biomass between juvenile and adult stages in the population (Figure 4.2). Models also clearly capture the fact that clone A maintains higher biomass levels and a higher proportion of juvenile biomass in the population.

4.3.2 Starvation

The patterns of starvation survival among stage classes exhibit some variation among clones (Figure 4.3, Figures A.6-A.9). Adults tended to withstand starvation better than juvenile, but there was some variation among clones as to how much difference there was between juvenile and adult survival. In all cases, it is noteworthy that medium-sized adults (1.6 - 2 mm) always survive better than larger adults, and except for clone D, small sized-adults (1.4 - 1.6 mm) do too. The model for starvation did a good job overall at capturing mortality patterns among size classes, particularly in clone A and D (Figure 4.3). In clone B, it tended to predict higher mortality levels than observed in the juvenile stages. In clone C, the sharp

change in survival between large-juvenile and small-adult survival is not well accounted for, resulting in an underestimate of small-adult survival.

4.3.3 Fluctuating food supply

In the LA experiments, the dynamics of every clone followed very regular patterns of oscillation (Figure 4.4). In all clones, there was almost no juvenile present during the nadir of population abundance, and individuals survived the period as adults. A closer look at adult classes (Figures A.10-A.13) shows that these adults were almost exclusively medium sized (1.6-2mm). Models predictions matched the data very well. The phase of the fluctuations (timing growth-peak-decline-nadir) is well captured. The amplitude of the fluctuations is relatively well captured too, though it tends to be underestimated for juvenile biomass. Models were however unable to capture effectively the transient dynamics that the data display during the first cycle.

In comparisons to large-amplitude fluctuations, the dynamics under small-amplitude fluctuations (SA) were less regular, and exhibited more interclonal variation (Figure 4.5). The dynamics had more variation between different periods of the cycle, and clone D also exhibited a high level of variation among replicates. The regularity of the oscillations greatly varied among clones. Clone A displayed regular oscillations with well-marked peak and troughs, whereas the dynamics in clone C and D were less regular. In particular the dynamics of the adult stages in these clones lacked any clear cyclical patterns. Similarly to large amplitude fluctuations, individuals tended to get through the nadir mostly as medium-sized adults, but this pattern was less pronounced as a fraction of individuals also survive in other size classes. Models did a poorer job at capturing the dynamics in small amplitude treatments. Patterns in juvenile biomass and fecundity are relatively well captured, but it seems that there are always some imperfections for predicting adult biomass. A closer look into the details (Figures A.14-A.17) reveals that the problem almost always lies in overestimating the abundance of large adults (>2mm). In clone A, the levels of adult biomass are overestimated during the

nadir. In clone D, beside the strong overestimation of large adults, the dynamics in other classes are actually well captured (Figure A.17). The predictions associated with clone C are more problematic, as the model predicts regular oscillations in adult stages when the actual data show much more irregular patterns and smaller biomass levels. This irregularity suggests a stronger involvement of stochastic processes than in the other clones, and this cannot be captured by the deterministic model.

4.3.4 Model analysis

Next we analyze whether the patterns of food-dependence in individual energetics are affected by the type of cycle, and whether this type of relation may affect the expression of genetic variation as a function of the type of the feeding regime. The rationale here is that individual feeding history - which for example depends on the type of cycle and the location within the cycle, e.g. growing versus declining phase - may affect the relation between individual length and individual weight, with subsequent implications for the ecological rates that depends on them. The variable we examine is individual energetic efficiency, which we measure as the quantity of biomass produced or lost for each unit of individual biomass per unit time. New biomass is produced through reproduction, lost through mortality, and either gained or lost through variation in body weight, which depends on the balance between energy acquisition and expenditure. Since these relations also contains dependence on individual states, and in particular on their developmental stage, we show these results for both juveniles and adults using midpoint length of $L = 1.05$ mm and $L = 1.8$ mm, respectively. This representation also provides a synthetic way for summarizing parameter estimates, for which a complete list is given in the appendix (Figures A.18-A.21).

Note that the feeding protocole consisting in food transfers induces food concentration to vary strongly in between two feeding interval. We do not focus on these short-term variations in the food environment, but rather focus on integrated effect by averaging individual energetic efficiency over a feeding interval (i.e. 2 days in the CF, and 1 day in SA and LA).

When plotted against food abundance, individual energetic efficiency forms a single line in the CF treatments, whereas it forms a closed loop in SA and LA treatments (Figure 4.6 and 4.7). This indicates that individual energetics are asymetric with respect to the directions of change in food in the fluctuating treatments. In other words, individual energetics depend on previous feeding history, in technical term, individual energetics present a hysteretic behavior. This behavior is much more pronounced for LA cycles than it is for SA cycles, as shown by the larger range of values spanned by the function and the increased difference in individual energetics as a function of whether food concentration increases or decreases. Starvation mechanisms have much influence on these trajectories, as shown by the strong asymmetry at low food levels in the LA treatments. Changes in the allocation function contribute to hysteresis too, as shown by the greater level of asymmetry in adults compared to juveniles under medium to high food conditions. The remaining asymmetry may be attributed to direct variation in the association between individual length, on which depends the assimilation process, and individual weight, on which depends metabolic maintenance rate. Note that all asymmetry present is ultimately a consequence of variation in the relation between individual length and weight, as further energetic processes depend on these variables.

4.4 Discussion

4.4.1 Modeling insights

Daphnia-algae dynamics are largely driven by consumer-resource type interactions and their underlying energetic basis (Nisbet et al. 2010). This is epitomized in an experimental system such as ours, where all other factors are under control. In order to make inferences on the energetics and characterize the variation existing between clones, we used a modeling approach. The availability of demographic data in qualitatively different food environments made it possible to use a series of steps of model selection and independent validations. This

provided a powerful way to overcome the problem of underdetermination intrinsic to these types of issues.

Because populations are monoclonal, the genetic basis conditioning individual and population response to their environment is genetically-based and remains the same across different environmental conditions. The differential expression of genetic variation between the LA and SA treatments therefore implies that the different environmental regimes affect the mapping between individual and population response to their environment. Model analysis provides two lines of evidence supporting the fact that the dynamics are more sensitive to clonal variation in energetics in SA than they are in LA.

First, there was a good agreement between model and data in the large amplitude experiment, in contrast to the small amplitude experiments, where the lack of fit is stronger. This implies that some degree of error in individual energetics does not matter as much under large amplitude treatments than it does under small amplitude treatments.

Secondly, differential expression of genetic variability between SA and LA may be caused by any one of two (non-exclusive) possibilities. First, the different kinds of cycles may affect the association between individual performances and resource conditions, and do it in a different fashion for the different clones. Alternatively, these relations may or may not remain the same, but population dynamics may not be equally sensitive to interclonal variation in different regions of these functions. For example, the dynamics may be very sensitive in low food regions, where small interclonal differences in energetics may make the differences as to whether an individual starve or not, whereas it may be much less sensitive to variation happening under high food conditions. Different environmental regimes would then cause these different regions to be visited at different frequencies.

Model analysis supports both hypotheses. Cycle type induces much change in the relation between individual energetic efficiency and food conditions (Figures 4.6 and 4.7). In addition, individual energetics have a hysteretic behaviour: the relation between individual

energetics and food concentration is asymmetric with respect to the direction of change in food concentration, and it also depends on the type of cycle considered. Individual behaviour is thus conditioned by the rate at which food concentration changes. The hysteretic behavior is more pronounced in LA cycles than it is in SA cycle because food concentration changes at a faster rate. This implies that large amplitude fluctuations impose stronger directionality in energetics than small amplitude fluctuations do. Therefore, the dynamics under large amplitude fluctuations are more subjected to the dynamics of the food environment. Taken together with the fact that the dynamics of the food environment is under experimental control and common to all clones, this explains why genetic variation affect less the dynamics in LA than in SA. Basically, large amplitude fluctuations causes much more variation in individual vital rates over a cycle than genetic differences do, effectively washing out the effect any clonal differences may have on the resulting population dynamics.

The hysteretic behavior is tied to the fact that short-term variation in food concentration affects the relation between individual length and weight, which conditions the processes of individual growth, reproduction and survival. This suggests that models aiming at studying the dynamics of structured populations under non-equilibrium conditions should include variables that reflect the influence of an individual's physiological condition on energetics, and not just the size- and resource- dependence of these processes. This dependency of individual energetics is not unique to our model and has also been considered in other frameworks, most notably the DEB theory from Kooijman (2010). Indeed, various lines of evidence have suggested that such considerations are important to maintain consistency at the individual level between energetics and empirical patterns of physiological behaviour (Kooijman 2010).

4.4.2 Implications

First, these results have implications for the detection of genetic variability effects in natural systems and for inferences that can be drawn from laboratory experiment. In particular, no

perceivable effect of genetic variability does not necessarily implies its absence, i.e. environmental fluctuations may elicit cryptic genetic variation. Conversely, if no significant effect of genetic variation is detected in an experiment, this does not necessarily imply that this variation will not be expressed in natural settings. This is particularly relevant to the level of inference that can be drawn from experiments that use constant environmental settings.

Secondly, genetic variability and genotype $\times$ environment interactions have role for the functioning of ecological units, the maintenance of genetic diversity and the selective process (Gillespie and Turelli 1989, Via and Lande 1985, Miner et al. 2004). Studying these issues involves the consideration of two different mappings: that from genotype to phenotype, and that from phenotype to population. The mapping between genotypes and phenotypes is a notoriously complex one. Not only do environmental conditions matter, but the patterns of variability in the environment may also affect that mapping. Works at the individual level have reported mixed evidence as to whether environmental variability *per se* affects patterns of phenotypic expression at the individual level (Miner and Vonesh 2004, Schoeppner and Relyea 2008, Siems et al. 1998, Engelmann and Schlichting 2005, Rodriguez 2012). Here we show that this type of variability may not only matter for phenotypic expression, but also for the way genetic variability affects population response to their environment. Most interestingly, different patterns of variation may either inhibit or exarcerbate these differences at the population level. Thus a specific account on the type of variation, and on how it interacts with individual and demographic processes, is required. Note that in natural systems, algae dynamics are interlocked with that of *Daphnia* populations due to the consumer-resource feedback loop, which we decoupled in our experiments. Our results thus apply for the way population *respond* to particular patterns of variations in their environment, not necessarily to the total effect of genetic variation on those dynamics. In any circumstance, tackling these issues will require joint appreciation of individual and population which may benefit further from both theoretical and empirical studies. For example, McCauley et al. (2008)

used bioassay experiments that made it possible to record individual performance, together with an assessment of population dynamics in a *Daphnia*-algal system to show that different types of population cycles affected key aspects of individual realized life history, and was a primary determinant of population dynamical behavior.

Our results also indicate that if environmental fluctuations act as a strong driver in natural systems, standing genetic variation may simply not matter in term of their effects on ecological processes. Fluctuation based coexistence theory requires both environmental variation, and a differential response of populations to this variation (Chesson and Huntly 1997, Chesson 2000), so that these types of systems may be less likely to host large amount of genetic diversity if other mechanisms are not present. This echoes findings from Nelson et al. (2005, 2007) who found that small amplitude fluctuations provided equalizing mechanisms, hence promoting the maintenance of genetic diversity, in comparison to large amplitude fluctuations, where competitive exclusion was much faster. More generally, we may speculate that the genetic composition of populations may have more importance for understanding the dynamics of systems subjected to moderate amount of environmental variability, whereas it may have a lesser importance for widely fluctuating systems. In our experiments, we used non-random patterns of fluctuations that are representative to the type of food fluctuations *Daphnia* populations may be subjected to. The dynamics of *Daphnia*-algae systems have parallels with many other systems, because they are driven by broadly similar mechanisms associated with consumer-resource interactions, and structured competitive interactions (McCauley and Murdoch 1987, Murdoch et al. 2004, De Roos and Persson 2013). Thus, the results presented in this study not only demonstrate the potential importance of environmental variability in mediating the interplay between individual and population processes, but they are also likely to reflect mechanisms that are common to many natural systems.

Based on our results, we may speculate on the relative importance of genetic variation in affecting ecological dynamics. For example, we found that large amplitude environmen-

tal fluctuations increased the contribution of environmental dynamics in driving population demography relatively to genetic factors. When fluctuations are sufficiently large, standing genetic variation may therefore not matter in term of their effects on ecological processes. This may also have implications for the mechanisms that promote the maintenance of genetic diversity in natural systems: fluctuation based coexistence theory requires both environmental variation, and a differential response of populations to this variation (Chesson and Huntly 1997, Chesson 2000), so that systems presenting large amplitude fluctuations may be less likely to host large amount of genetic diversity if other mechanisms are not present. This echoes findings from Nelson et al. (2005, 2007) who found that small amplitude fluctuations provided equalizing mechanisms, hence promoting the maintenance of genetic diversity, in comparison to large amplitude fluctuations, where competitive exclusion was much faster. In our experiments, we used non-random patterns of fluctuations that are representative to the type of food fluctuations *Daphnia* populations may be subjected to. The dynamics of *Daphnia*-algae systems have parallels with many other systems, because they are driven by broadly similar mechanisms associated with consumer-resource interactions, and structured competitive interactions (McCauley and Murdoch 1987, Murdoch et al. 2004, De Roos and Persson 2013). Thus, the results presented in this study not only demonstrate the potential importance of environmental variability in mediating the interplay between individual and population processes, but they are also likely to reflect mechanisms that are common to many natural systems.

Table 4.1: Model definitions.

Individual level dynamics	
$\frac{dn}{dt} = -(\delta_s(t) + \delta_i(t))n(t)$	Cohort number dynamics
$\frac{dL}{dt} = \frac{\max(L_w(t), L(t)) - \max(L_w(t - T_m), L(t - T_m))}{T_m}$	Growth in length
$\frac{dW}{dt} = \epsilon I(t) - m(L)W(t) - \Theta(t)$	Individual weight dynamics
$\frac{dWr}{dt} = \Theta(t)$	Cumulative energy invested in reproduction
Population level dynamics	
$B(t) = \sum \beta(t)n(t)$	Birth rate
$\frac{dF}{dt} = -\sum I(t)n(t))$	Algae dynamics between transfer
Model functions	
$\beta(t) = \frac{W_r(t - T_m) - W_r(t - 2T_m)}{(1 + \gamma)W_b T_m}$	Egg production rate
$W_l(t) = \xi L^q$	Nominal weight for length
$L_w(t) = W_l^{-1}(t)$	Nominal length for weight
$\omega = W/W_l(L)$	Ratio between an individual weight and its nominal weight for length
$I(t) = \frac{F(t)}{F(t) + F_h} I_{max}(L)$	Ingestion rate
$I_{max}(L) = \iota_0 L^{\iota_1}(\, 1 - \exp(-(L/\iota_2)^{\iota_3})\,)$	Maximum ingestion rate
$\delta_s(t) = \max(0, \alpha(L)e^{-\beta(L)\omega(t)} - \delta_0(L))$	Starvation mortality rate
$\delta_0(L) = \max(\alpha(L)e^{-\beta(L)}, \delta_c)$	Offset in the starvation mortality rate
$\Theta(t) = \alpha_0 f_W(W) f_\omega(\omega) X(t)$	Energy allocated to reproduction (see text for details)
Spline functions	
$m(L)$	Weight specific maintenance rate
$\delta_i(L)$	Intrinsic mortality rate
$\alpha(L)$	Component of the starvation mortality function
$\beta(L)$	Component of the starvation mortality function

Table 4.2: Parameter values and definitions.

Parameter	Value or range	Description	Source
F_h	164 μgC/L	Half saturation constant	McCauley et al. 1990
ι_0	9.64 μgC/d/mm$^{-\iota_1}$	Parameter in the maximum ingestion rate function	McCauley et al. 1990
ι_1	1.76	Parameter in the maximum ingestion rate function	McCauley et al. 1990
ι_2	0.95 mm	Parameter in the maximum ingestion rate function	McCauley et al. 1990
ι_3	2.14	Parameter in the maximum ingestion rate function	McCauley et al. 1990
ξ	2.62 mm	Parameter in the weight for length relationship	McCauley et al. 1990
q	2.4	Parameter in the weight for length relationship	McCauley et al. 1990
T_m	1.5-3 d	Intermolt duration	This study
ϵ_a	0.4 -0.9	Assimilation efficiency	This study
γ	0.4 -2	overhead cost of egg production	This study
α_0	> 0	parameter in the allocation function	This study
α_1	unconstrained	parameter in the allocation function	This study
α_2	unconstrained	parameter in the allocation function	This study

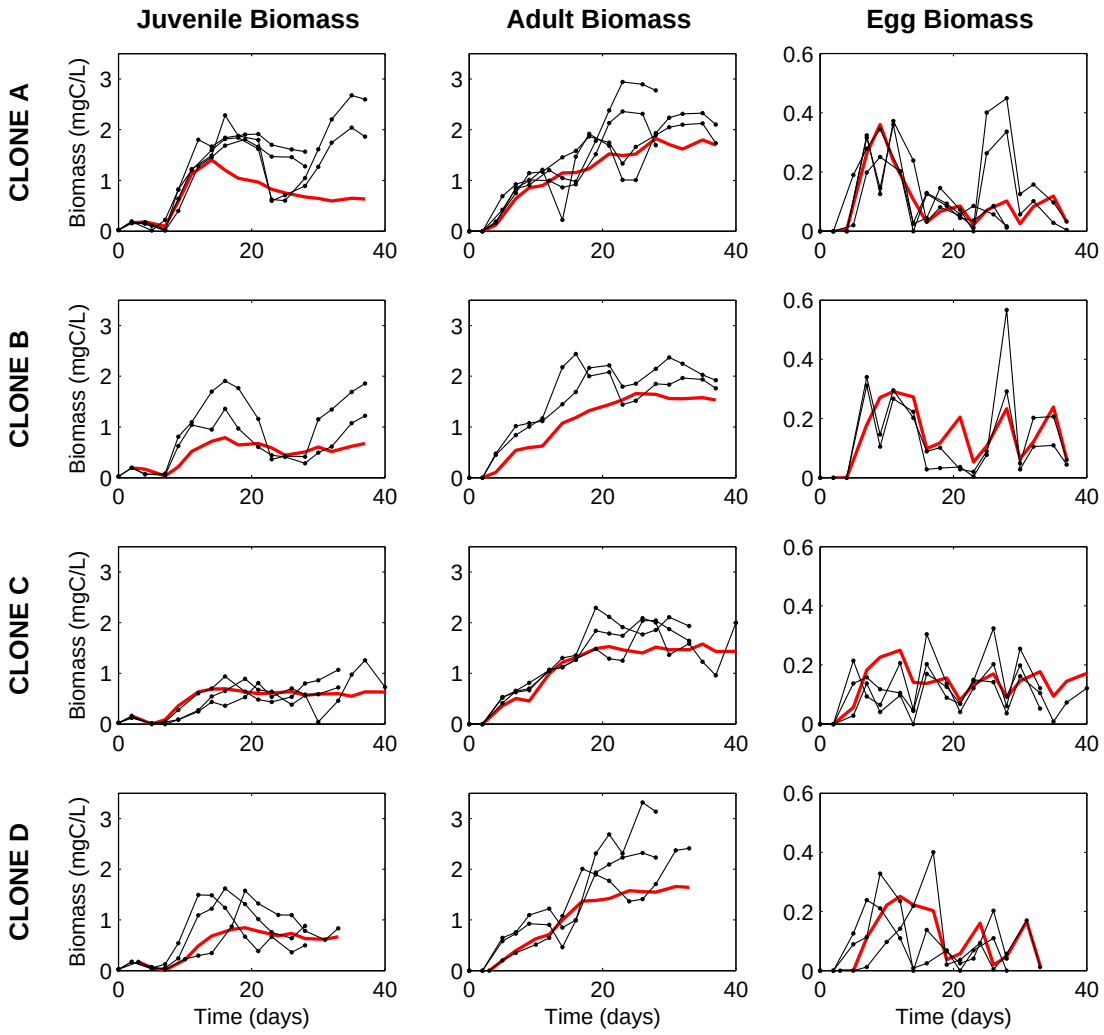

Figure 4.1: Biomass of the different stages in the constant food supply experiments. Black: experimental data; Red: model fits to the data.

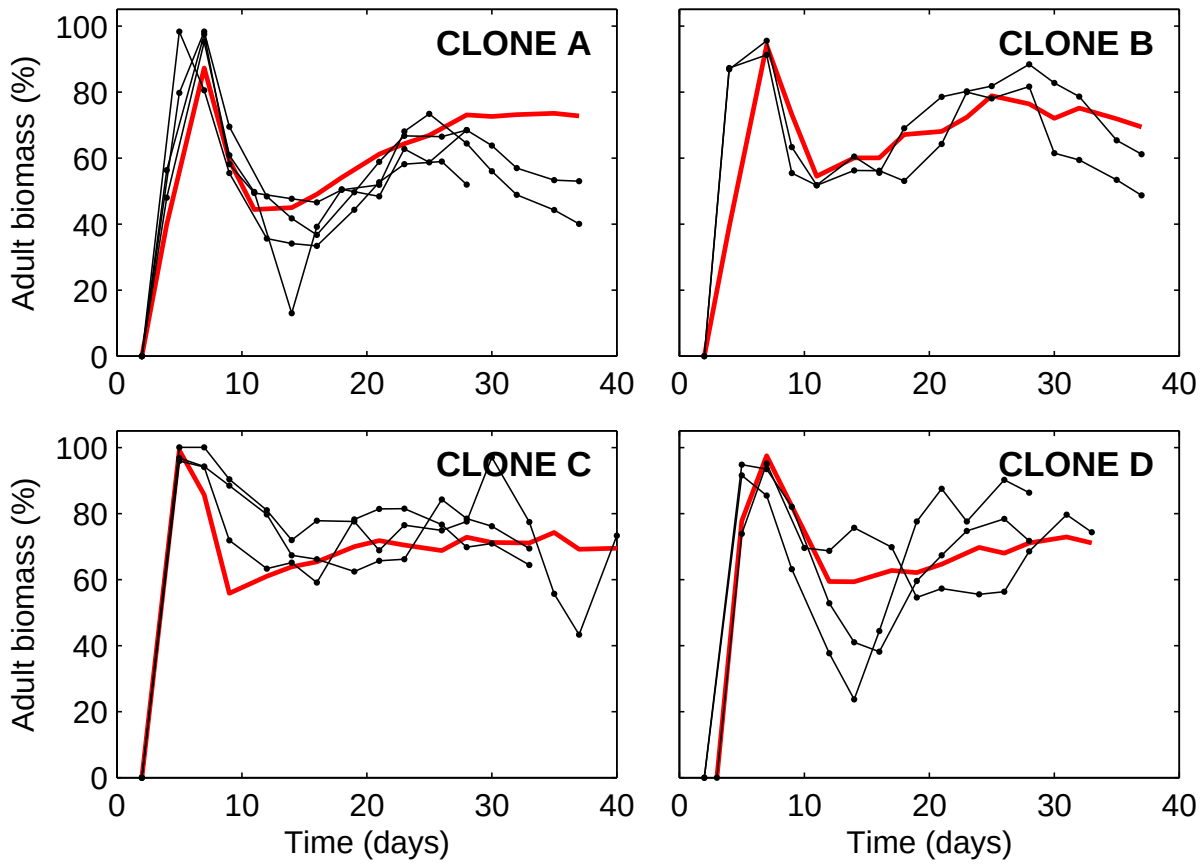

Figure 4.2: Proportion of adult biomass in the population in the constant food supply experiments. Black: experimental data; Red: model fits to the data.

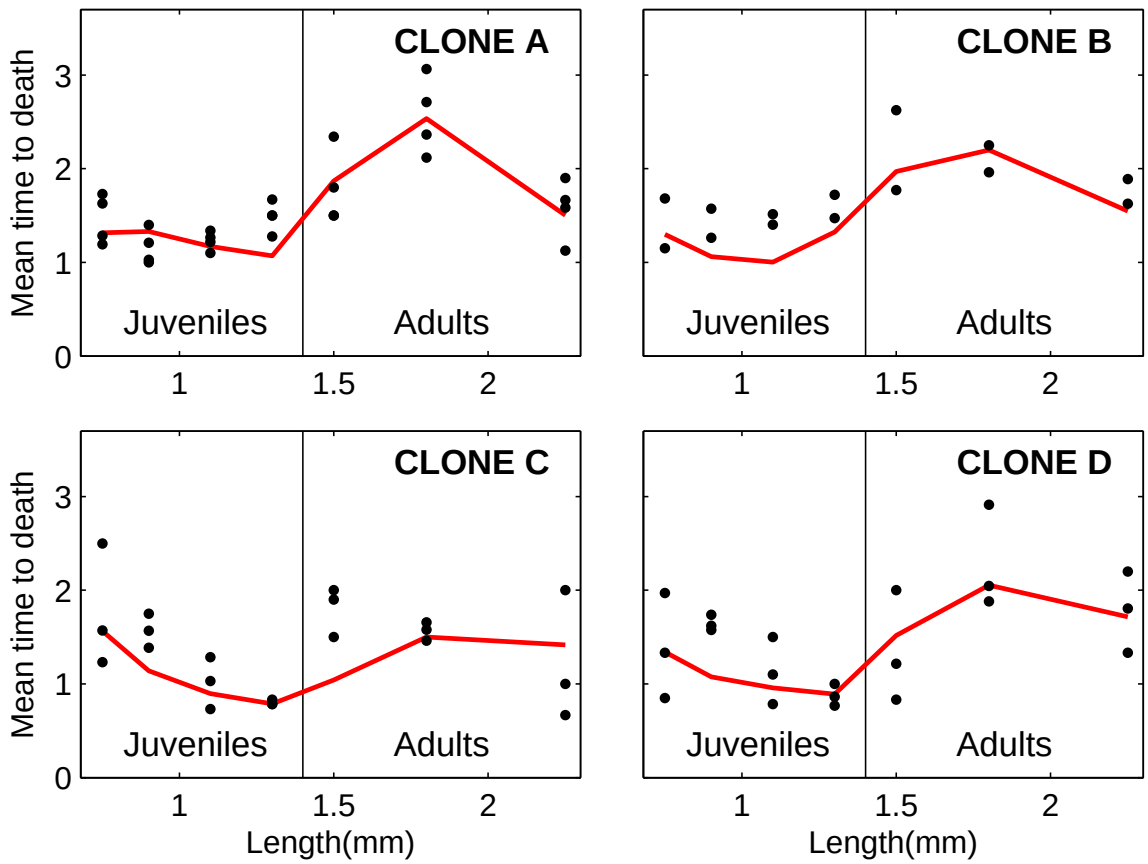

Figure 4.3: Mean time to death until starvation as a function of individual length. Each black dot represents the value measured in the different replicates. Red lines are model fits to the data.

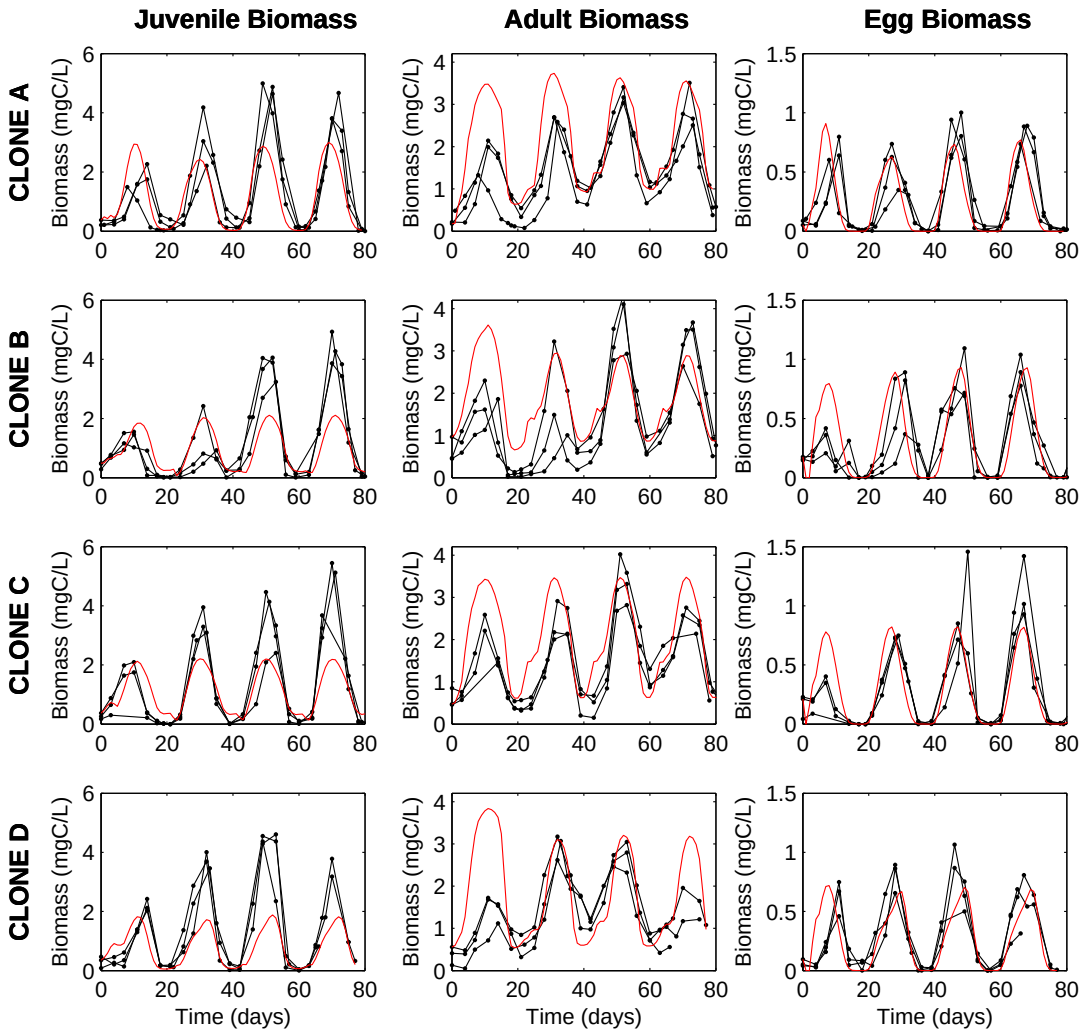

Figure 4.4: Biomass of the different stages in the large-amplitude food supply experiments. Black: experimental data; Red: model predictions.

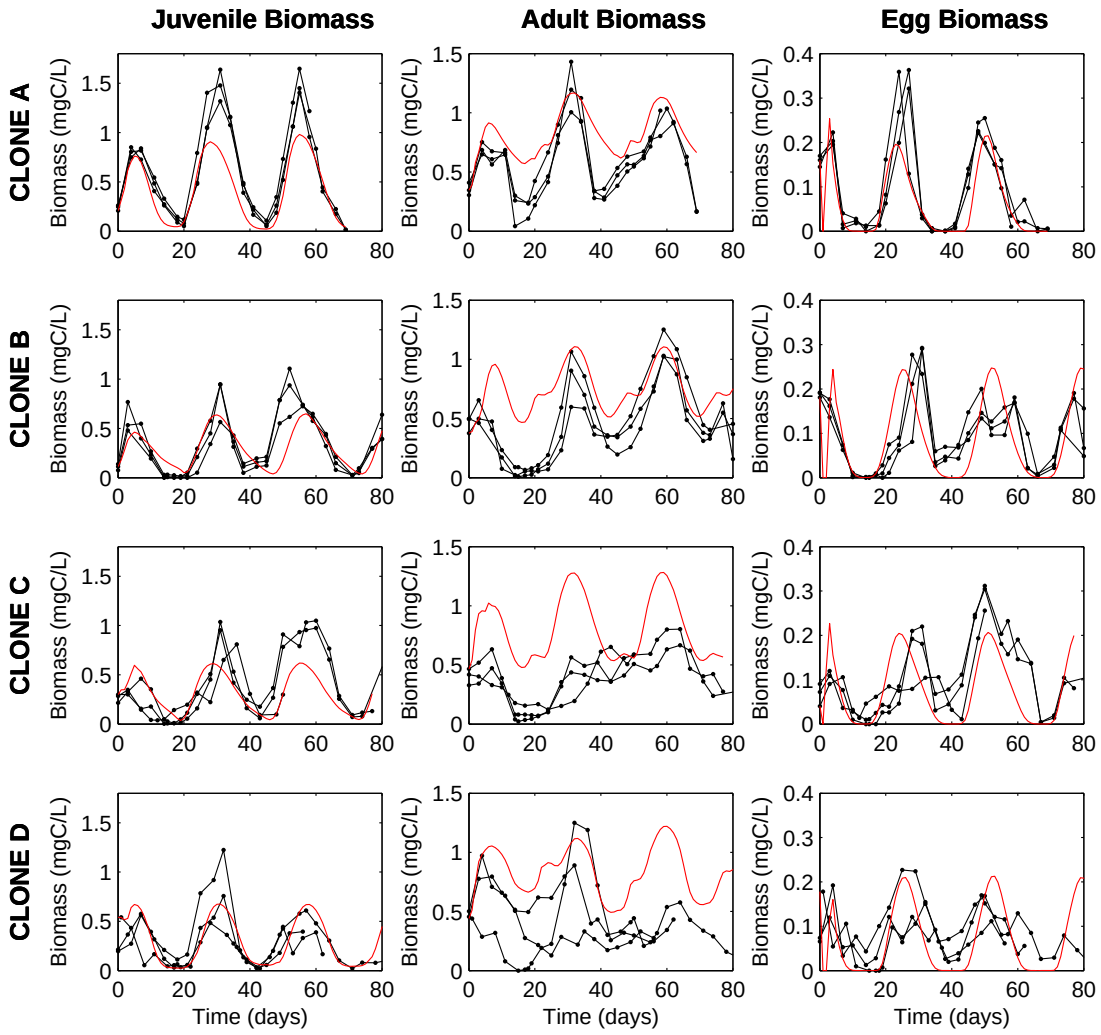

Figure 4.5: Biomass of the different stages in the small-amplitude food supply experiments. Black: experimental data; Red: model predictions.

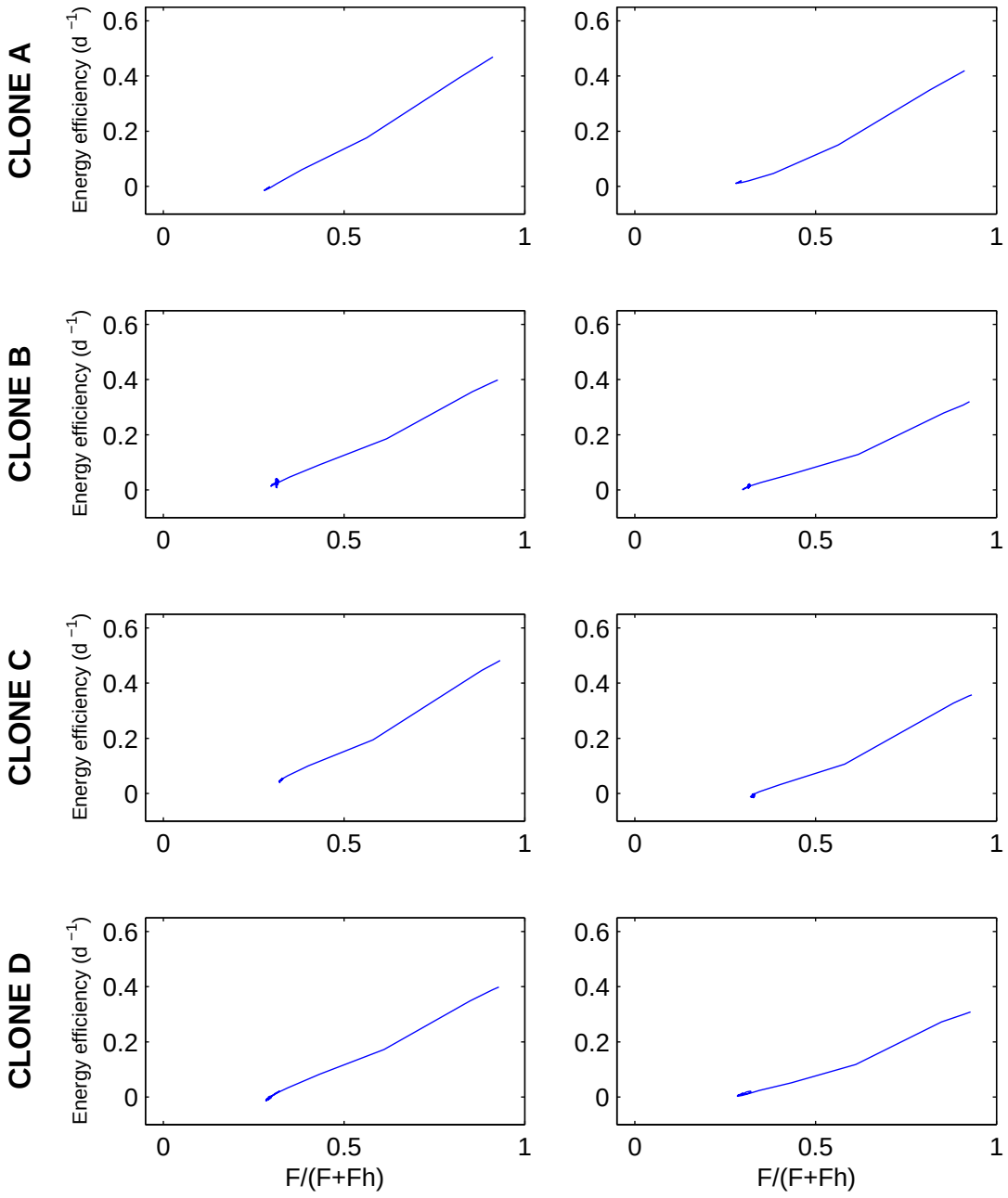

Figure 4.6: Juvenile and adult energy efficiency as a function of scaled food density in the constant food supply experiments, i.e. the saturating term of the functional response, F/(F+Fh).

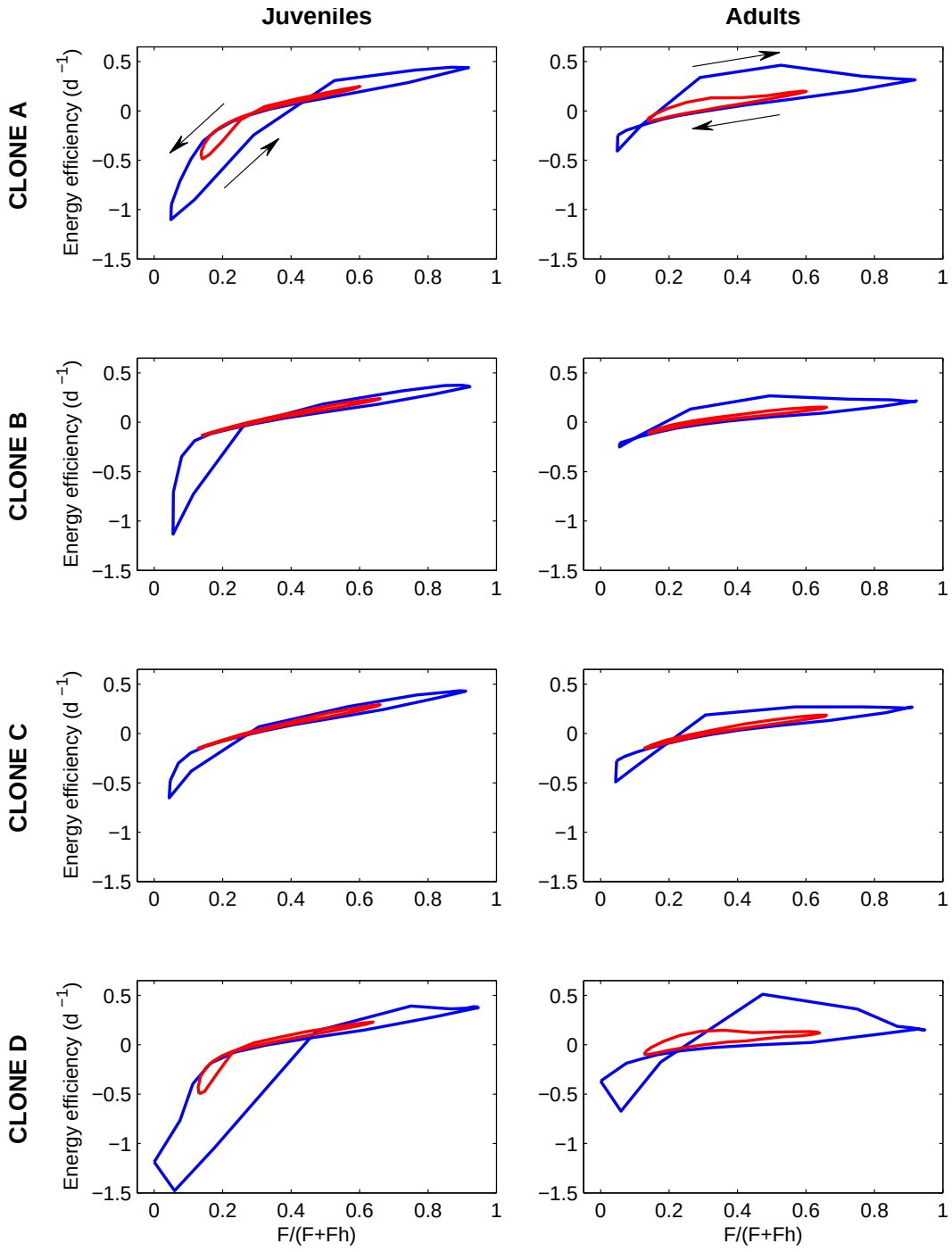

Figure 4.7: Juvenile and adult energy efficiency as a function of scaled food density. The blue line corresponds to large amplitude cycles, whereas the red lines correspond to small amplitude cycles. The arrows indicate the direction of time.

Chapter 5

The functional genomics of consumer-resource cycles: Linking demographic patterns with patterns of gene expression

5.1 Introduction

Considering the role of genes with reference to the ecological context in which populations evolve provides an integrative framework that can shed light into genetic, ecological and evolutionary issues, not reachable otherwise (Ungerer et al. 2008). This increased explanatory power is enabled by the detection of genes of functional significance for ecological processes, facilitating the study of the full chain of events linking the individual response to their environment to ecological patterns over the different biological scales involved (genes, physiology, life history, population dynamics) (Dalziel et al. 2009). It is indeed a longstanding issues of ecological and evolutionary sciences to understand the origin of phenotypic variation. For example, population dynamics are dictated by the way fecundity and mortality rates are affected by environmental conditions, and similarly, it is the variation in phenotypic patterns of expression among genetic variants that allows natural selection to operate. The first feature of an individual response affected by environmental variation are the quantity of RNA transcripted, which, through a chain of physiological events, translate to higher components of the individual phenotype. Yet, there is no organism in which we fully understand the mechanistic linking between environment and phenotypic expression. Identifying those genes that systematically respond to environmental conditions experienced by individuals may help adressing some of these caveats by identifying physiological pathways of importance and studying them in more depth. Integrating genomic and ecological sciences has,

for example, made it possible to make progress in elucidating gene functions (e.g. Mihola et al. 2009), on the origin of genomic architecture (e.g. Rogers et al. 2013), on which part of the genome is under selective pressure (e.g. Rogers and Bernatchez 2007, Linnen et al. 2013), and on the aspects of genetic variation that matter for ecological processes (e.g. Sorria-Corrasco et al. 2014). Yet, in all of these investigations, elucidating the links between the genomic basis of phenotypic variation and a functional mechanistic explanation of how this variation interacts with the environment remains challenging (Colbourne et al. 2011, Latta et al. 2012).

Understanding the mechanisms underlying organisms' response to their food environments is a fundamental question in ecology (Murdoch et al. 2003, Olff et al. 2009, Kooijman 2010, Jeyasingh et al. 2011). Feeding provides the nutrients and energy necessary to achieve growth, reproduction and survival, with direct implications for the demographic process and ensuing consequences on higher levels of biological organization. Natural communities present large levels of genetic variation which may affect how different individuals and populations respond to similar food conditions (Vellend and Geber 2005, Hughes et al. 2008). The food environment is also usually highly variable over time and space, once again, with consequences on these processes. Given the prevalence of consumer-resource interactions in nature and their importance for many physiological processes (Murdoch et al. 2003, Kooijman 2010), we may expect the genome to be strongly responsive to variation in the food environment over an ecological time scale. Similarly, patterns of genomic expression are of importance for evolutionary analyses that study the process of selection under the influence of resource dynamics. Natural selection results from differential patterns of phenotypic expression among genetic variants, particularly those traits closely related to fecundity and mortality. Since patterns of resource availability experienced by individuals are one of the major determinants of patterns of phenotypic expression, we may expect a fine tuning of patterns of phenotypic expression in response to resource conditions. In order to identify

and study genes subjected to selection, it is may be be helpful to identify those genes that show diverging patterns of expression among genotypic variants.

In our previous works, we investigated the role of environmental and genetic variation in driving population demographic patterns, using populations of the cladoceran *Daphnia pulicaria*. In these experiments, populations involving different clones were raised in isolation, and environmental variation was induced by providing the population with a controlled amount of food every day (Chapter 4). The patterns of food supply mimicked two different types of cycles observed in natural and laboratory conditions, and resulted in either small- or large-amplitude fluctuations in the food environment (McCauley et al. 1999, Figure 5.1). We found that whereas small-amplitude (SA) fluctuations induced strong differences in the population dynamics of the different clones, the dynamics under large-amplitude (LA) fluctuations were very similar among clones. We concluded that emerging demographical patterns were likely the result of genotype x environment interactions. Using a modeling approach, we hypothesized that these patterns were underlain by a hysteretic behavior in individual energetics (i.e. dependence on past conditions) that was related to the intensity of environmental fluctuations. Therefore, the influence of environmental variation was greater in LA treatments in driving the dynamics relatively to the effect of genetic variation. Yet, the functional genomic response to these cycles remains unknown.

In this chapter, we analyze how patterns of gene expression of two of the clones used in the experiment varied over a population cycle, in both SA and LA cycles. Given these noticeable effects of clonal- and cycle type at the population level, we are testing in this chapter the general question as to whether observed variations in demographic patterns resulting from differences in clone and cycle type can be associated with characteristic patterns of expression at the genomic level. The results of this analysis are for now purely descriptive, and linking patterns of gene expression with phenotypic and environmental components will be provided in future research.

5.2 Methods

5.2.1 Experimental design

The individuals used for genomic analysis were sampled in the context of the demographic experiment presented in Chapter 4. We used two clones of *Daphnia pulicaria* (clone A and B), which were confirmed to represent distinct genotypes with 8 microsatellite markers (Table B1, Colbourne et al. 2004). In these experiments, different clonal populations were fed a daily ration of the green alga *Chlamydomonas reinhardtii* in a way that produced either small- or large-amplitude cycles in their food environment (respectively abbreviated as SA and LA). More details on the rearing protocol and the experimental methods used can be found in Chapter 4.

For each type of cycle and each clone, we sampled individuals at four points within the cycle, corresponding to different events in *Daphnia* population biomasses: the period of maximal growth, the peak, the period of maximum decline, and the nadir, respectively abbreviated as G, P, D, N (Figure 5.1). The individuals were sampled at the end of the demographic experiments, and the timing of these events was calculated from data on previous cycles (Chapter 4). These expectations matched well the actual timing of the events in large amplitude cycles, but there were some mismatches in the small-amplitude treatments (Figure 5.1). This mismatch was somewhat expected, as small-amplitude cycles displayed irregular patterns of variation (Chapter 4), which caused some differences in the phase of these events among the different cycles of the time series.

For each point of the cycle, a group of usually twenty adults were sampled from a donor population. These individuals were then replaced with identically sized individuals from another replicate population in order to insure that the sampling of these individuals did not significantly affect the demographic process. Sampled individuals were preserved in RNA later (Qiagen) and then kept at -80°C until extraction.

5.2.2 RNA extraction, amplification and array hybridization

Five individual were pooled per biological sample in order to ensure sufficient RNA yield. All samples were shipped on dry ice to Funomics (Funomics Global, Inc., Saskatoon, Saskatchewan, Canada) who performed the RNA isolation. *Daphnia* were homogenized and total RNA was isolated using the Trizol (Invitrogen) -chloroform method (Chomczynski and Sacchi 1987). Total RNA was suspended in RNA Secure (Ambion, Inc). The quality of total RNA preparations was assayed by electrophoresis and measured on a Thermo Scientific NanoDrop 2000c UV Spectrophotometer (for RNA abundance and contamination) and the Bio-Rad Experion (for RNA degradation). Only samples with a A260/A280 ratio close to 2 and A260/A230 ratio greater than 1.8 were retained for analysis, and every individual yielded high quality RNA (Table B2).

RNA amplification, microarray processing, and normalization of signal intensity were performed at Notre-Dame university (Indiana, USA) by Dr. Jacqueline Lopez and Dr. Erliang Zeng. The NimbleGen (Roche-NimbleGen, Inc., Madison, WI, USA) *Daphnia pulex* expression array 12x135k platform (GEO Accession GPL11278; Colbourne et al. 2011) is a high-density NimbleGen gene expression microarray of 12 identical arrays prepared by Maskless Array Synthesizer. Each array contained 137,000 isothermal probes representing 35,665 genes, with each gene represented by as many as three unique probes, while the remaining probes were designed from transcriptionally-active regions (TARs) whose gene models are not yet known. RNA amplification, hybridization and microarray analysis followed Colbourne et al. (2011). Beginning with at least 0.2μg of total RNA, a single round of amplification using MessageAmpTM II aRNA kit (Ambion, Austin, TX, USA) was performed to generate cRNA (10 μg) that was converted into double strand cDNA with random primers using the SuperScript Double-Stranded cDNA Synthesis kit (Invitrogen, Carlsbad, CA, USA). Double-stranded cDNA (1 μg) was labeled with NimbleGen's Dual-color Labeling Kit (Roche NimbleGen, Inc., Madison, WI). Replicates of each treatment for each genotype

were alternatively labeled and a dye swap was included among the replicate experiments. Dual-color hybridization, post-hybridization washing and scanning were done according to the Roche NimbleGen's instructions. Images were acquired using a Roche Nimblegen MS200 scanner (Roche Nimblegen, Inc., Madison, WI).

5.2.3 Statistical analysis

The NimbleGen array image data were processed using NimbleScan version 2.5 (Roche Nimblegen, Inc., Madison, WI) to extract probe intensity values. Gene expression values (i.e. gene intensity value) were obtained from a summarization of intensity values of all corresponding probes using the RMA (Robust Multi-array Average) method. The pre-processed microarray data were imported into an in-house analysis pipeline using Bioconductor for normalization and analysis (Gentleman et al. 2004). All genes were quantile-normalized across arrays, samples, and replicates (Bolstad et al. 2003).

Differential expression was assessed by contrast analysis in order to characterize the effect of cycle type, clonal type, and within-cycle variation on gene expression. Mean of gene expression measurements was compared between two conditions using an F-test, whereby p-values were adjusted for multiple comparisons using a false discovery rate procedure (Storey and Tibshirani 2003). This analysis was carried out on the 29,212 transcriptional units with confirmed gene models (Colbourne et al. 2012 ; J. Lopez, personal communication).

Gene ontology analysis was performed using the genome annotation reported in Colbourne et al. (2011), and available at the JGI portal. Among the 6,643 genes annotated for biological processes, we evaluated whether certain functional categories were over-represented by comparing the observed frequency of differentially expressed genes in each category with their expected frequency, based on the reference microarray gene set. In addition, this analysis would highlight potentital genes exhibiting parallel responses among treatments and cycles. Statistical significance was assessed using Fisher exact tests with a false discovery rate correction (Bluthgen et al. 2005). The significance level was set at the recommended

0.01, with a minimum of five associated genes required in each gene set.

5.3 Results

5.3.1 Global patterns of gene expression

The proportion of the genes responding to within cycle variation varied from 58.2% in clone A / LA cycle to 68.2% in clone B / SA cycle (Table 5.1). More details on the proportion of genes differentially expressed by groups of treatments is provided by a Venn diagram in Figure 5.2. Considering both kinds of cycles, 80.7% of the clone A genome was differentially expressed in at least one within-cycle comparison. This proportion was 84.2% in clone B. Considering both kinds of clones, the LA cycles induced gene expression variation for 78.7% of the genes, and the SA cycles, 85.6%. We examined the effect of clone and cycle type on transcript abundance. There are two ways to examine the variation induced by cycle or clonal type: (i) examining how much transcript abundance is affected by these factors at each point in the cycle, (ii) examining how much these factors affect transcript abundance at one point of the cycle relatively to another (e.g. by examining whether the difference in transcript abundance between the growth and the decline phase is affected by cycle type). We refer to these two case scenarios as the effects of the factors on transcript abundance, and their effects on within-cycle variation in transcript abundance, respectively. Table 5.1 reports both the proportion of comparisons resulting in a significant effect of the factor being tested, and the proportion of genes that showed a significant effect in at least one of the comparisons being tested. The first type of measurements aims at reflecting how prevalent these effects are for the dynamics, whereas the second type of measurements provides information on the proportion of the genome that has the ability to respond to these factors.

Both types of assessment show that cycle type has a greater impact on the expression of clone B genome than in clone A, both in term of transcript abundance and in term of within-cycle variation in transcript abundance: 16.4% of the comparisons showed a significant effect

of cycle type on transcript abundance in clone A, whereas this proportion was 27.7% in clone B. In regard to within-cycle variation in transcript abundance, these values are 18.6% and 24.1%, respectively.

The effect of clonal type on transcript abundance was more pronounced in LA cycles than in SA cycles (31.8% vs 26.6% respectively). On the other hand, both type of cycles induced a comparable effect of clone on the within-cycle variation in transcript abundance (14.2% vs 14.1%).

Finally, 21.1% of the comparisons tested showed a significant interaction between the effects of clonal type and the effects of cycle type on transcript abundance, and 15.8% of the comparisons regarding within-cycle variation in transcript abundance. All in all, this resulted in 44.2% and 46.2% of the genes showing a significant effect of the interaction in at least one comparison.

5.3.2 Candidate genes

As the previous results indicate, large percentages of the genome were associated with clonal and cycle type, we examined the gene ontology categories among these gene sets involved in the clone x cycle interaction, both in term of the effect on transcript abundance and on within-cycle variation in transcript abundance (Table 5.2, the other comparisons are given in Table B3). Almost all categories involved in the clone x cycle interaction and over-represented in term of their effect on transcript abundance are also over-represented in term of their effect on within-cycle variation in transcript abundance. These categories were mostly related to protein processing and metabolism. These same categories were also frequently involved in causing clonal differences in both types of cycles (Table B3). Concerning gene categories underlying the differences caused by cycle type, 4 categories were common to both clones in term of their effect on transcript abundance, and 10 of those in term of cycle effect on within-cycle variation in transcript abundance.

5.4 Discussion

The food dependence of individual physiological and life-history processes and its importance for the population process and in shaping the evolution of Cladoceran species has long been recognized and studied (Lynch 1984, Nisbet et al. 2004). Our results suggest that the variation in these phenotypic processes is underlain by the variation of a very large proportion of the genome: up to 84.2% of all identified protein-encoding genes responded to the variation induced by the food environment. This echoes findings of Colbourne et al. (2011), which found that a large proportion of the genome responded to variation in ecological factors such as the presence of kairomones (79% of the genome), or cadmium concentration (72% of the genome). Moreover, patterns of genomic expression strongly varied among clones. Given the importance of phenotypic variation, and more particularly individual energetics, in determining the ecological performance of individuals, this suggests that a large portion of the genome may be under selection as a result of variation in the food environment, and that the consideration of the mechanisms involved is of paramount importance to understand the evolution of the genomic architecture of these species.

Considering the effect of clonal variation and cycle type, some aspects of the genomic response are well in line with the conclusions drawn from the analysis of the demographical consequences, whereas others are more surprising.

We found that cycle type had a notable effect on patterns of gene expression. The mechanisms driving the dynamics of the populations in these two kinds of cycles are of a qualitatively different nature (McCauley et al. 1999, 2008), and it is interesting to see that they leave a specific signature at the genomic level. An application of these results my involve using patterns of gene expressions as an ecological marker. For example, it has been found in *Arabidopsis* that wounding and insect feeding could be distinguished by the transcript profile induced by each treatment (Reymond et al. 2000). In our present study, this signature of population dynamics at a genomic level suggests that genomic tools may

be similarly used as a substitute from longitudinal survey data, for detecting the dynamical state and regime populations are subjected to in the wild. Gene ontology analysis suggests that a number of gene categories are over-represented in response to variation in cycle type, and we will investigate these issues more deeply in order to identify specific genes that can be used as robust ecological markers for identifying cycle type directly from gene expression data.

Variation induced by the type of cycle is also important in an evolutionary context, as qualitatively different selective regime operate in both kinds of cycles (Nelson et al. 2007). Cycle type has been shown to affect the selective regime in term of their integrated effect on fitness. In particular, small amplitude cycles provide equalizing mechanisms that promote the maintenance of genetic diversity. Concurrent differences in patterns of gene expression may be suggestive that these different types of cycles affect not only the selective regime, but that they may also have an effect on which genes, and to which extent these genes, are subjected to selection. This is further supported by the fact that many genes were significantly affected by clone x cycle interaction, which suggests that cycle type does not induce mere quantitative differences in gene expression, but qualitative changes as a function of clones.

A large number of genes were significantly affected by clonal type. Overall, clonal type induced more differences in the transcript abundance in LA cycles than SA cycles. This may come as a surprise, as we found the exact opposite in term of the effect of clonal type in demographical patterns. Several hypotheses may be involved in explaining this divergence. (i) Variation in gene expression does not necessarily translate into phenotypic variation of ecologically important traits. (ii) The timing at which genes are expressed and the identity of these genes may be more important in explaining emerging interclonal at the phenotypic and population level. A few genes may, for example, have a disproportionate effect on the resulting outcome. (iii) In the present context, it may be more important to consider

the relative change in transcript abundance over the cycle (i.e. the effect of clonal type on within-cycle variation in transcript abundance), rather than the absolute variation in transcript abundance caused by clonal type. (iv) Finally, variation in gene expression may still be expressed at a phenotypic level, but food fluctuations may canalize this variation to a greater extent in large-amplitude treatments than small amplitude treatments. This is the hypothesis we drew in our previous study to explain the emergence of genotype x environment interaction at the demographic level (Chapter 4).

Contrast analysis suggests a number of candidate genes involved in these differences, and gene ontology analysis suggests further that these genes are mostly related to protein processing and metabolism. Nonetheless, GO categories encompass processes that can hardly be related to life history processes. Further analysis will be required to assess whether the patterns of expression of these genes align to the expectation of being linked with resource use and the energetics of growth, reproduction and mortality. In particular, the recent analysis of *Daphnia* genome not only showed that there is about a third of the genome that does not present homology with any other known genome (Colbourne et al. 2011). Moreover, it is these *Daphnia*-lineage specific genes are the most differentially expressed in response to ecological variation (Colbourne et al. 2011). The analysis provided in this paper is only a first step aiming at describing general patterns of the functional genomic response of *Daphnia* in response to the variation induced by dynamic food variation, by different types of environmental regimes, and by genetic variation. Our analysis of chapter 4 generated large amounts of data on relevant phenotypic, demographic and environmental variables, and we also drew many inferences on individual energetics through modeling. A next step in the analysis of these genomic data will consist in studying how variation in gene patterns affect the expression of individual life history traits and respond to ecological variables, through correlative analysis. This type of analysis is however non-trivial due to possible non-linear relationship between patterns of gene expression and phenotypic expression (Shannon et al.

2002, Daub et al. 2004, Chen et al. 2010). Nonetheless, it will make it possible to identify candidate gene underlying phenotypic variation in ecologically important traits.

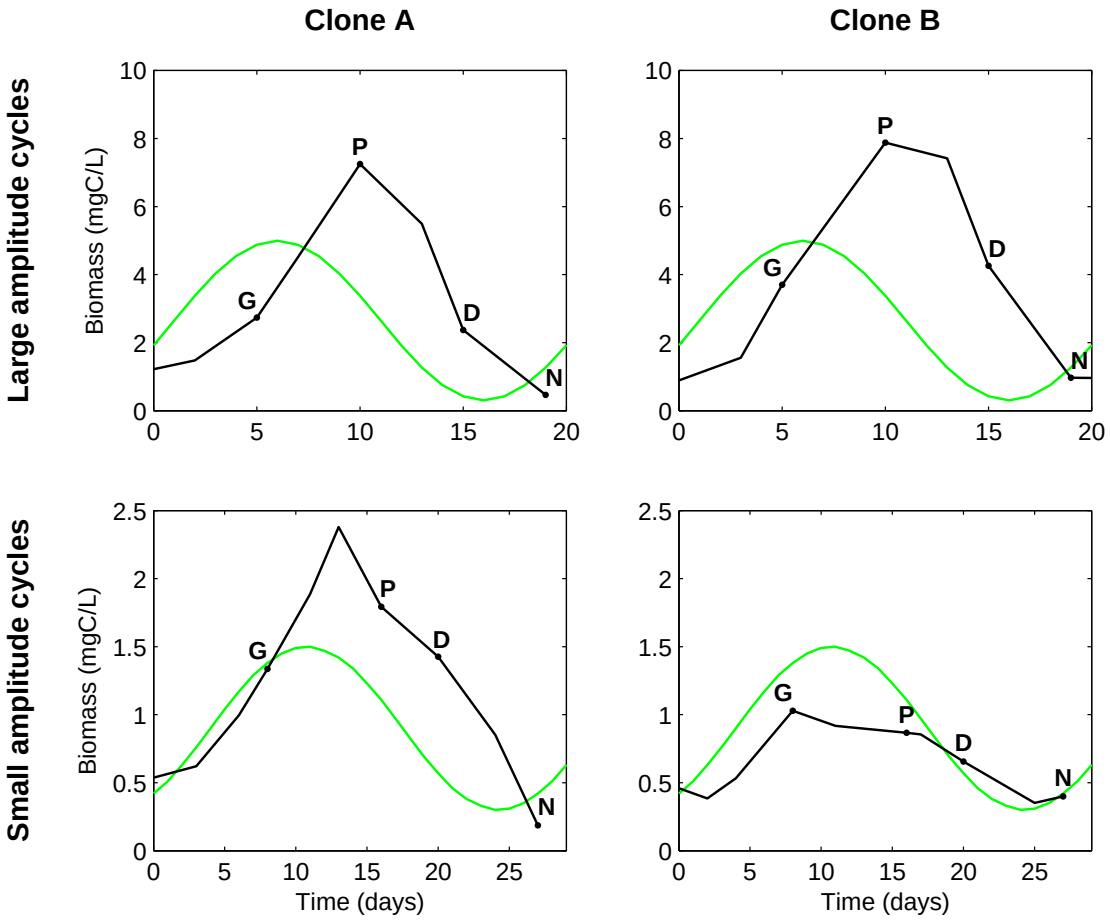

Figure 5.1: Variation in total population biomass (black lines), and daily quantity of algae fed to the populations (green lines), for both clones and both type of cycles. G: growth phase, P: peak; D: decline phase; N: Nadir.

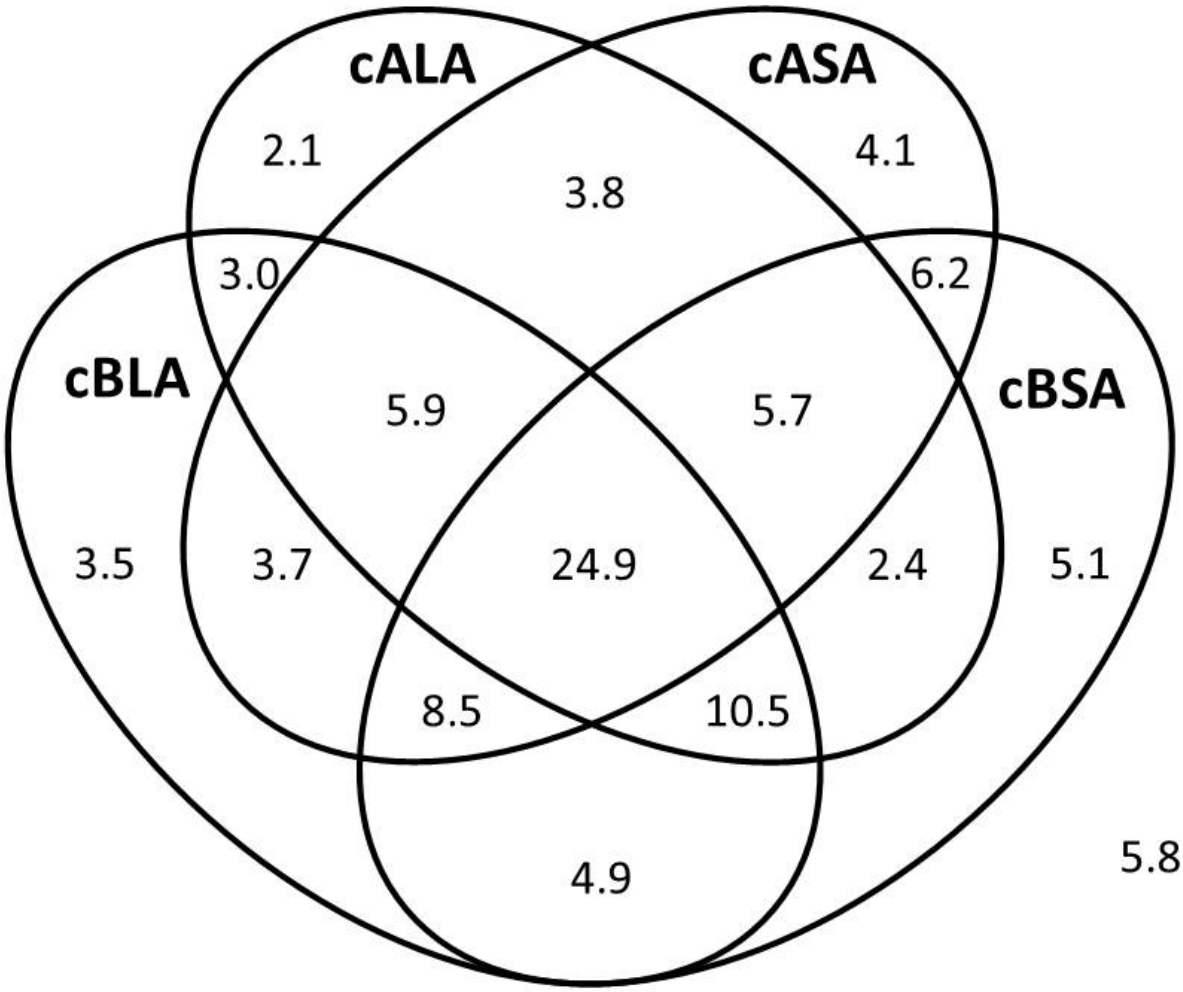

Figure 5.2: Venn diagram showing the percentage of genes being differentially expressed in at least one within cycle comparison. The numbers at the intersection between two or more area show the percentage of genes that are differentially expressed in the different treatments.

Table 5.1: Results of the contrast analysis.

			% of significant comparisons	% of genes with at least one significant comparison
Variation in transcript abundance	Effect of clone type	LA	31.8	67.9
		SA	26.6	63.4
	Effect of cycle type	cA	16.4	49.7
		cB	27.7	69.9
	Cycle x clone interaction	-	21.1	44.2
Within-cycle variation in transcript abundance	direct comparisons	cALA	22.6	58.2
		cASA	24.8	62.7
		cBLA	26	64.8
		cBSA	27.7	68.2
	Effect of clone type	LA	14.2	45.4
		SA	14.1	44
	Effect of cycle type	cA	18.6	54.8
		cB	24.1	61.4
	Cycle x clone interaction	-	15.8	46.2

Table 5.2: GO categories over-represented among the gene sets involved in the clone x cycle interaction.

	GO Accession number	Term	p-value
Variation in transcript abundance	6457	protein folding	2.22E-07
	6072	glycerol-3-phosphate metabolism	0.00010
	6470	protein amino acid dephosphorylation	0.00013
	59	protein-nucleus import. docking	0.00037
	44267	cellular protein metabolism	0.00126
	6468	protein amino acid phosphorylation	0.00131
	5975	carbohydrate metabolism	0.00149
	8152	metabolism	0.00361
	6885	regulation of pH	0.00509
	6511	ubiquitin-dependent protein catabolism	0.00552
	8299	isoprenoid biosynthesis	0.00725
Within-cycle variation in transcript abundance	59	protein-nucleus import. docking	9.67E-10
	44267	cellular protein metabolism	9.21E-09
	6468	protein amino acid phosphorylation	9.55E-08
	8152	metabolism	1.12E-07
	6457	protein folding	2.23E-07
	6470	protein amino acid dephosphorylation	1.95E-06
	9117	nucleotide metabolism	9.24E-05
	6511	ubiquitin-dependent protein catabolism	0.00021
	6207	de novo' pyrimidine base biosynthesis	0.00035
	6885	regulation of pH	0.00109
	17000	antibiotic biosynthesis	0.00143
	7018	microtubule-based movement	0.00192
	6072	glycerol-3-phosphate metabolism	0.00468
	6510	ATP-dependent proteolysis	0.00468
	6783	heme biosynthesis	0.00468
	6177	GMP biosynthesis	0.00510
	16568	chromatin modification	0.00544
	18149	peptide cross-linking	0.00624
	7600	sensory perception	0.00813
	51258	protein polymerization	0.00969

Chapter 6

Environmental fluctuations can reverse size-dependent competitive ability

6.1 Introduction

A major challenge in ecology is to explain the rarity of large amplitude population cycles in nature when theory on consumer-resource interactions predicts their emergence (Murdoch 1994). Theory has mostly focused on the mechanisms allowing model's equilibrium to remain stable in spite of the destabilizing effect of enrichment. Another possible type of explanation is that competitive interactions among individuals dominate over consumer-resource interactions to generate alternative kinds of cycles with smaller amplitude (Murdoch et al. 2003, McCauley et al. 2008, De Roos and Persson 2013). These 'stage-structured' cycles have been characterized in many natural and experimental systems (Murdoch et al. 2002, 2003). They arise as a result of density dependence acting differentially on individual vital rates as a function of their state, and have been best described in the context of stage- or size-structured interactions (De Roos et al. 2003). The mechanisms involved in these cycles place this question at the interplay between individual and population processes. A particular aspect of this interplay that remains poorly characterized is how individual processes interact with dynamically varying environments to affect the dynamics of populations. The purpose of this chapter is to demonstrate that an explicit consideration of these processes may help explaining the relative stability of populations in nature.

The hypothesis that stage-structured dynamics dominate over consumer-resource dynamics has been supported by research on *Daphnia*-algae systems. Experiment and theory have confirmed that both large- and small-amplitude cycles can coexist under the very same

conditions (McCauley et al. 1999, 2008). In these experiments, patterns of population dynamics were recorded for many populations subjected to the same conditions. The vast majority of populations displayed stage-structured small-amplitude cycles, whereas only a small portion of them displayed large-amplitude consumer-resource cycles (McCauley et al. 2008). Whereas, these authors have been able to show that the various sources of instabilities provided by both consumer-resource and intraspecific competitive interactions were fundamental to the presence of these coexisting attractors, much remains to be clarified on the mechanisms underlying these dynamics.

Understanding the mechanisms generating these dynamics, and in particular why stage-structured cycles prevail over consumer-resource cycles, requires a thorough investigation of the intricacies associated with both size-dependent competitive interactions and consumer-resource interactions. In consumer-resource systems, most aspects of individual demographic performance are food-dependent (Gurney et al. 1996). In addition, individual state, such as developmental stage or size, may affect how much individuals are sensitive to variation in the food environment, and how they contribute to the demographic process (Meszéna et al. 2006; De Roos et al. 2003). In this context, the dependency is best understood by considering the mechanisms of energy acquisition and allocation underlying patterns of individual realized patterns of growth, reproduction and mortality. One of the greatest difficulties associated with these kinds of questions is however to reach an understanding on how individual level processes translate to the population level.

These state-dependent aspects of an individual life history bear direct relationships with their competitive abilities. Individuals compete for a common pool of resources that limit their demographic rates. Different individuals are however not limited to the same extent which may be assessed by their ability at producing or loosing biomass (Persson et al. 1998). De Roos and coworkers (2007) have characterized some of the major mechanisms involved in these types of interactions by considering the dynamical consequences of asymmetry in

competitive ability between juveniles and adults. In the models used by these authors, it is assumed that either juveniles or adults are superior competitors, and that this ranking is fixed and maintained over varying food conditions. They have been able to show that this ontogenetic asymmetry has important consequences for the structure and dynamics of natural communities. In particular, they found that various kinds of cycles with different properties emerge depending on the extent by which adult and juvenile competitive ability differs. These theoretical predictions account for broad patterns of population variation in a number of empirical systems including fish populations, indian meal moths, soil mites, and unicellular organisms (Townsend et al. 1990, Sanderson et al. 1999, Briggs et al. 2000, Massie et al. 2010, Persson and De Roos 2013).

This theory comes nonetheless with its limitations, and our understanding of the mechanisms causing these cycles may be still sharpened. For example, *Daphnia* population dynamics display many features characteristic of stage-structured dynamics, but they also cannot fully be explained within this framework because different lines of evidences suggest either juveniles or adults to be the superior competitors (reviewed in De Roos and Persson 2013). Works on individual energetics suggest that additional mechanisms may be at play. Indeed, individual contribution to the dynamics of the system is directly related to the (state-dependent) feeding, development, reproduction, and mortality rates. These processes may be affected very differently by variation in the food environment, and cause the relative competitive ability of different stages to be affected non-uniformly. For example, there is no particular reason to expect that the individuals surviving best under poor food conditions will also be the most productive ones under high food conditions. In this chapter, we build upon results drawn from our previous works on *Daphnia*-algae systems to show that a possible consequence of dynamical food regimes is to reverse the competitive ranking between juveniles and adults, and more generally cause food-dependent competitive asymmetry between individuals in different states. We then discuss the implications of these results for

the population dynamical process.

6.2 Evidences for alternating competitive rankings

In Chapter 4, we implemented population models for four different *Daphnia* clones based on individual energetics and parameterized them from data on their population dynamics under different regimes of food supply. We previously discussed the implications of the differences found between clones for ecological and evolutionary processes. Here, we reverse the question and aim at distinguishing the major mechanisms conditioning the demography of *Daphnia*-algae beyond any clonal difference.

Competitive ability of individuals can be related to their efficiency at producing biomass. We measure individual energy efficiency as the quantity of biomass produced or lost for each unit of individual biomass per unit time (De Roos and Persson 2013). Figure 6.1 shows the efficiency of juveniles and adults at producing biomass over a food cycle. This figure was obtained from one of the models we parameterized in chapter 4. It is shown for a given clone, but it is representative of the general patterns we found for other clones. This graph clearly shows that under low food availability, adults are superior competitors, whereas juveniles take over during periods of high food availability. The reason for that is two-fold. First, adults have a greater resistance to starvation during periods of food shortage (Figure 6.1). Secondly, under higher food conditions, adults allocate an increased proportion of the assimilated food to reproduction (Figure 6.2). Reproduction is however a costly process, we estimated that only 33% to 50% of the energy invested in reproduction actually results in the production of newborn biomass, and literature reports values ranging from 33% to 71% (Nisbet et al. 2004). Therefore, for a given rate of assimilation, an increased fraction of energy allocated to reproduction implies a decrease in total biomass production.

The general mechanism mediating alternative competitive ranking is linked to the fact that, at an individual level, biomass production rate is a sum of different processes: new

biomass is gained through reproduction, lost through mortality, and either gained or lost through variation in body weight, which depends on the balance between energy acquisition and expenditure. These processes scale differently with individual state, and dynamic variation in food concentration causes the relative contribution of these processes to vary over time.

We found no other study explicitly reporting alternating competitive rankings in the literature. Nonetheless, we explored the model in McCauley et al. (2008) in depth to find that this mechanism was actually included in it. It is an important finding, as this model has been parameterized from robust empirical data and encountered major success in explaining *Daphnia* population dynamical behavior, notably the presence of coexisting cyclic attractors. In figure 6.3, we plot the energy efficiency of juveniles and adults assumed by that model as a function of food conditions. Adults are more efficient at very low food levels, whereas juveniles are better under higher food concentrations. This result comes from the fact that juveniles are estimated to be intrinsically more efficient in their energy use, but adults are less sensitive to starvation mortality. There are some differences with our own findings as the superior competitiveness of juveniles under high food conditions was due to an increased reproductive effort in adults, which comes with a cost. In contrast, McCauley et al. (2008) assume that a constant fraction of assimilated energy is invested to reproduction. Beyond these differences, both parameterizations result in the qualitatively similar result that food variation may reverse competitive ranking between adults and juveniles.

6.3 Dynamical effects of variable competitive asymmetry

In the analysis that follows, our goal is to characterize the dynamical consequences of variable degree of competitive asymmetry between juveniles and adults as a function of food density, including the possibility of reversing competitive rankings. We use the model of McCauley et al. (2008) as a baseline, as the energetics of individuals are modeled in a simple and easily

apprehensible way. Model equations are summarized in Table 6.1, and parameter definition in Table 6.2. Briefly, in this model the consumer population is divided into two stages, juveniles and adults, which feed on the same resource environment. Juvenile developmental delay, reproduction rate, and mortality rates are all food-dependent. More details on its derivation can be found in McCauley et al. (2008) and Nelson et al. (2007). Basically, in this model, biomass loss and production result from the balance between two processes: energy assimilation, which enables the production of new biomass through growth and reproduction, and mortality. Essentially, the assimilation process largely prevails under moderate to high food conditions, whereas starvation mortality prevails under low food conditions. Only within a narrow range of food densities do these two processes have a similar magnitude effect on biomass production.

In order to assess the consequences of varying the relative competitive ability of juveniles and adults at low versus high food conditions, we need to slightly reformulate the model of McCauley et al. (2008). Indeed, in the original formulation, starvation mortality rate is considered to be inversely proportional to the assimilation rate. Therefore varying any assimilation-related parameter affects the response at both high and low food level. We re-express the starvation mortality functions using two new compounds parameters, δ_J and δ_A, that set juvenile and adult mortality rates independently from assimilation rates. Since we do not aim at characterizing the dependency of model behaviour on parameter values *per se*, but rather characterize the dynamical consequences of variable degree of competitive asymmetry between juveniles and adults, we can safely ignore the dependency of δ_J and δ_A on assimilation-related parameters in the subsequent steps. By doing so, we can now independently vary the degree of competitive asymmetry at low food density by varying δ_J and δ_A, - the juvenile and adult mortality scalars - and vary it at high food concentrations by varying I_J and I_A - the maximum ingestion rates of juvenile and adult.

Varying a parameter one at a time generally affects equilibrium food density, which

has direct consequences on system stability (Murdoch et al. 2003). Indeed, changes in resource equilibrium density affect the strength of density-dependence in resource growth rate. This induces concomitant changes in system stability that are associated with changes in resource dynamics rather than changes in consumer biology *per se*. In order to get a better characterization of the effects of competitive asymmetry on system stability, it is therefore preferable to maintain food equilibrium density to the same level, irrespective of the changes imposed on consumer biology. To do so, we varied juvenile parameters (I_J or δ_J) independently from one another, but the corresponding adult parameters (I_A and δ_A, respectively) were, in contrast, adjusted such that food equilibrium density remains the same under any of the parameter combination tested.

To assess the effects of parameter change on the prevalence of the different kind of dynamics, we simulated the dynamics of the system using a set of 1000 initial conditions and recorded the number of these simulations resulting in either equilibrium, small- or large-amplitude cycles. Note that the resulting proportion estimated should not be interpreted as a measure of the absolute tendency of the system to converge toward the different possible kinds of dynamics. Rather they indicate how parameter variation affects the relative prevalence of these different dynamics.

Figure 6.4 shows these results for three different levels of enrichment in resource carrying capacity (K = 0.6 mgC/L; 1.1 mgC/L; 1.6mgC/L) and Figure 6.5 shows the shape of the energy efficiency functions for some of the parameter combinations tested. The lower-left part of Figure 6.5, and of each panel of Figure 6.4, corresponds to situations with minimal asymmetry; going up along the I_J-axis increases asymmetry at higher food levels; going right along the δ_J-axis increases asymmetry at low food levels.

Richer environments increase the tendency of the system to display large amplitude cycles. However, enrichment does not qualitatively affect the effects parameter variation has on the prevalence of the different kinds of dynamics.

When there is almost no asymmetry a low food density (Figure 6.5, First column), the system always display large amplitude consumer-resource cycles, hence causing the disappearance of coexisting attractors (Figure 6.4). Increasing juvenile starvation mortality rate relatively to the adult one always tends to stabilize the system (Figure 6.4). This favors the emergence of small-amplitude stage-structured cycles at first, and further increase causes the system to stabilize to an equilibrium. Asymmetry in the response of adults and juveniles to starvation is thus an important feature for stabilizing consumer-resource cycles and for maintaining the coexistence of cyclic attractors. Similarly, increasing juvenile ingestion rate tends to favor small-amplitude cycles over large-amplitude cycles and further increase causes the system to stabilize to an equilibrium. Thus, increasing juvenile performance has quite opposite effects on system stability depending on whether it is increased under low food conditions or high food conditions.

This can be explained as follows. First, the asynchronous entry in starvation of the different stages in the population causes juveniles to start dying sooner. This releases the foraging pressure on resource and ultimately prevents massive mortality in adults which would otherwise enforce the consumer-resource mediated instabilities. This mechanism is further enforced when adult competitiveness is increased relatively to juvenile competitiveness under low food conditions, i.e. when δ_J is increased.

Secondly, increasing juvenile competitiveness under high food conditions causes the per-capita transfer rate of biomass to become greater from juvenile to adult stages than from adult to juvenile stages. In contrast to the case where the relative efficiency of adults and juvenile would remain fixed, this favors an increased proportion of adults in the population until the food starts declining again.

Overall, a counter-intuitive consequence of the alternation of competitive rankings between low and high food conditions is that it promotes more constancy in the adult stages. At low food density, there is almost no transfer of biomass between stages. The superior

competitive ability of adults is due to their better survival. This favors the maintenance of adult biomass. In contrast, under high food conditions, variation in individual performance results from variation in foraging performance (assimilation rate), and this causes differential transition rates of biomass between stages. The per-capita transfer rate of biomass through development (from juvenile to adults) is thus greater than through reproduction (from adult to juvenile). All these processes favor relative stability in adult biomass levels, and partially decouple the dynamics of adult stages from resource dynamics. This may contribute to attenuate the destabilizing effect of juvenile developmental delay on the dynamics of the population.

It has been previously suggested that the continuous presence of adults and their rather constant density over the cycle seem to be indeed an essential feature for stabilizing the interaction between *Daphnia* and algae (McCauley and Murdoch 1987, Ananthasubramaniam et al. 2013). In particular, the survival of a substantial number of adults during the nadir of population density allows for the population to resume reproduction shortly after algae population begins to recover (Ananthasubramaniam et al. 2013). The rapidly increasing foraging pressure induced by newborn production then allows to limit algae population growth rate and thus stabilizes the interaction. Here, we have shown that both increasing adult performance under low food conditions, or increasing juvenile performance under high food conditions can favor this mechanism.

6.4 Conclusions and implications

In view of these results, a natural question to be asked is why the fact that food conditions can reverse competitive rankings has never been previously characterized in the literature despite the tremendous number of studies on individual energetics?

First, this question is relevant to issues at the interplay between the individual and population processes, which is nowadays more rarely dealt with in the literature. Individual

studies have been primarily used to predict how individual energetics generates patterns of individual life history. But how these processes affect the overall efficiency of individuals at producing or losing biomass mostly takes importance within a population context, so that this result may have been overlooked.

Secondly, many of these aspects may have been missed because works on individual energetics are almost always based from experiments in constant environments. This study has been motivated by the model inferences we drew in chapter 4. One important mechanism contributing to food-dependent competitive asymmetry is starvation mortality, which is a specific response to variable environments. Starvation mortality is however rarely included within population models, and when it is, this is usually done in an overly simplistic way. Starvation mortality has been the focus of many studies on individuals (Tessier et al. 1983, Bradley et al. 1991, Gliwicz and Guisande 1993), but the connections with individual energetics, let alone the population process, are usually weaker. The other major feature mediating the reversing of competitive ranking was the positive association between reproductive effort and food density. Empirical works on *Daphnia* have however unequivocally reported that energy allocation rules depend on size but not on food density (Nisbet et al. 2004). A fundamental limitation of these previous works is that indeed their findings come from experiments on individuals raised under various, but constant, food levels. First, doing so does not guarantee consistency with population level behavior. Secondly and much more importantly, dynamically varying food conditions are likely to affect qualitatively the energetics of individuals (Chapter 4). In particular, constant food conditions result in weak homeostasis (Kooijman 2010). According to dynamic energy budget theory, the ratio between reserve mass and structural mass and hence the quantity of energy allocated to reproduction - then becomes directly proportional to energy assimilation rate. Under dynamically varying food conditions this ratio becomes much more variable and has strong consequences on reproductive allocation patterns.

In consumer-resource systems, size-specific competitiveness is often measured as the food density at which an individual with a specific size can just meet its maintenance needs (Persson et al. 1998). The focus remains however on moderately low food conditions, close to equilibrium conditions. A description of near equilibrium energetics is useful to understand what causes an equilibrium to be stable or unstable, but carry only limited insights to understand what causes the system to exhibit small or large amplitude fluctuations. By definition, the processes mediating these kinds of behaviors happen away from equilibrium conditions, and more focus on the energetic response over the whole range of conditions spanned over a cycle is required. Measuring size-dependent competitive ability as the ability of differently sized individuals at converting food to consumer biomass and evaluating it over the range of possible food conditions is, in this context, more insightful. We have shown that doing so may reveal hitherto unsuspected relations in the energetic mechanisms underlying intraspecific competitive interactions. Accounting for the specific mechanisms underlying individual energetic response to variable environments opens the way for a whole range of dynamical processes for structured consumer-resource systems that is not accounted for by current theory. As shown in this study, they have general consequences on the demographic process, including the stabilization of consumer-resource interactions, the emergence of stage-structured dynamics, and the coexistence of dynamical attractors and their prevalence. Moreover, dynamic variation in the environment can affect qualitatively individual energetics, such that size- of stage- specific efficiency may not only depend on current food conditions, but also on their feeding history (Chapter 4), and this is likely to complexify this picture furthermore. We conjecture that such mechanisms may be able to generate population cycles with new properties from those reported in the literature (i.e. consumer-resource, delayed-feedback, and the different kinds of generation cycles), and this may help explaining population patterns that cannot be fully accounted for by current theory, such as *Daphnia* population dynamics.

Table 6.1: Model equations.

$\frac{dF}{dt} = qF(t)\left(1 - \frac{F(t)}{K}\right) - \frac{F(t)}{F(t)+F_h}(I_J J(t) + I_A A(t))$	Resource dynamics
$\frac{dJ}{dt} = \frac{\chi I_A \sigma_A}{\gamma}\frac{F(t)}{F(t)+F_h}(A(t) - A(t-\tau(t))S(t)) - \delta_J \frac{F(t)+F_h}{F(t)}J(t)$	Juvenile dynamics
$\frac{dA}{dt} = \frac{\chi I_A \sigma_A}{\gamma}\frac{F(t)}{F(t)+F_h}A(t-\tau(t))S(t) - \delta_A \frac{F(t)+F_h}{F(t)}A(t)$	Adult dynamics
$w = \sigma_J I_J \int_{t-\tau(t)}^{t} \frac{F(\xi)}{F(\xi)+F_h} d\xi$	Condition for juvenile development
$S(t) = \exp\left(-\delta_J \int_{t-\tau(t)}^{t} \frac{F(\xi)}{F(\xi)+F_h} d\xi\right)$	Juvenile survival

Table 6.2: Variable and parameter definitions.

Variables	**Description**
F(t)	Resource density (mgC/L)
J(t)	Juvenile density (#/L)
A(t)	Adult density (#/L)
$\tau(t)$	Developmental delay (days)
S(t)	Juvenile through-stage survival

Parameter	**Description**	**Default Value**
q	Maximum per-capita resource growth rate	1 (d^{-1})
K	Resource carrying capacity	Varied (mgC/L)
I_J	Maximum juvenile ingestion rate	5.23×10^{-3} (mgC/L/d)
I_A	Maximum adult ingestion rate	1.91×10^{-2} (mgC/L/d)
F_h	Half saturation constant	0.164 (mgC/L)
χ	Proportion of utilized carbon allocated to reproduction	0.77 (-)
γ	Carbon required to produce one new offspring	1.51×10^{-3} (mgC/L)
σ_J	Proportion of ingested carbon utilized by juveniles	0.49 (-)
σ_A	Proportion of carbon utilized by adults	0.43 (-)
δ_J	Juvenile mortality scalar	4.25×10^{-3} (mgC/L/d)
δ_A	Adult mortality scalar	7.21×10^{-4} (mgC/L/d)
w	Mass gain required to complete juvenile development	4.8×10^{-3} (mgC/L)

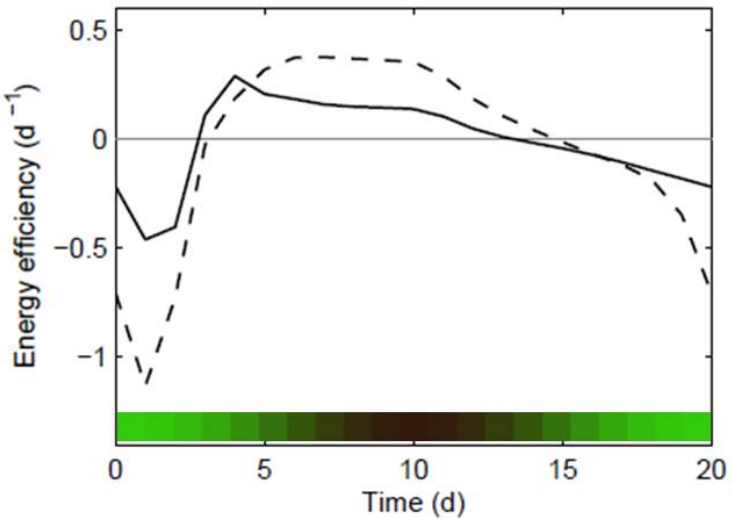

Figure 6.1: Energy efficiency of juveniles (L = 1.05mm; dashed line) and adults (L = 2mm; solid line) over a large amplitude cycle, as described in chapter 4. Time within the cycle is scaled such that t=0 corresponds to the nadir in food density, and T=10 corresponds to the peak. The green bar is an indicator of the food supplied, with darker color indicating greater amount of food supply.

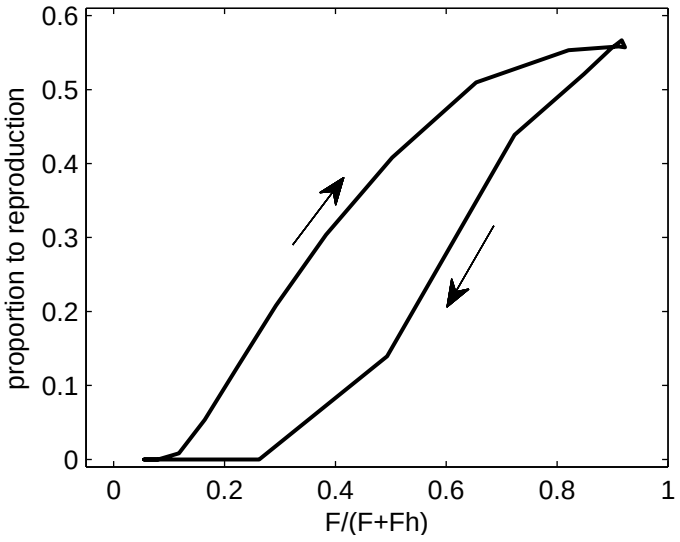

Figure 6.2: Proportion of net assimilation allocated to reproduction as a function of scaled food density (the saturating term of the functional response). Data is shown for 1.8mm individuals over a large amplitude cycle as described in Chapter 4. Arrows indicate the direction of time. Note that within the modeling framework used, the allocation fraction is not a direct function of food conditions but depends only on individual states. The significant association between allocation and food density results from non-random patterns of association between food density and individual states.

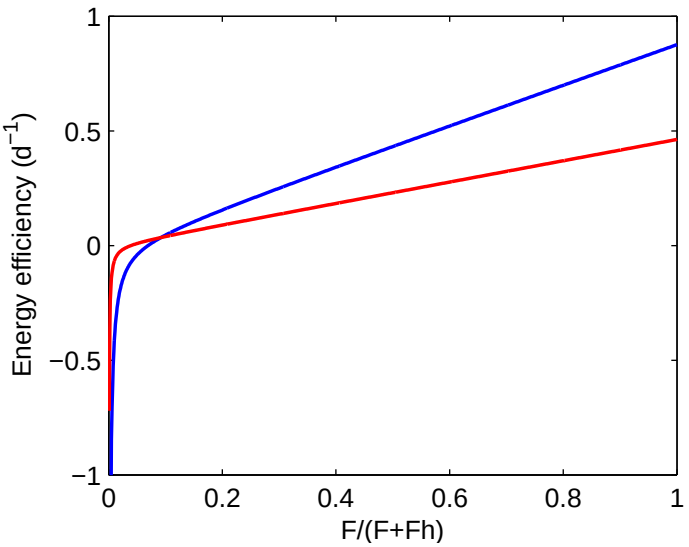

Figure 6.3: Energy efficiency of juveniles (blue) and adults (red) as a function of scaled food density, as predicted from McCauley et al (2008)'s model.

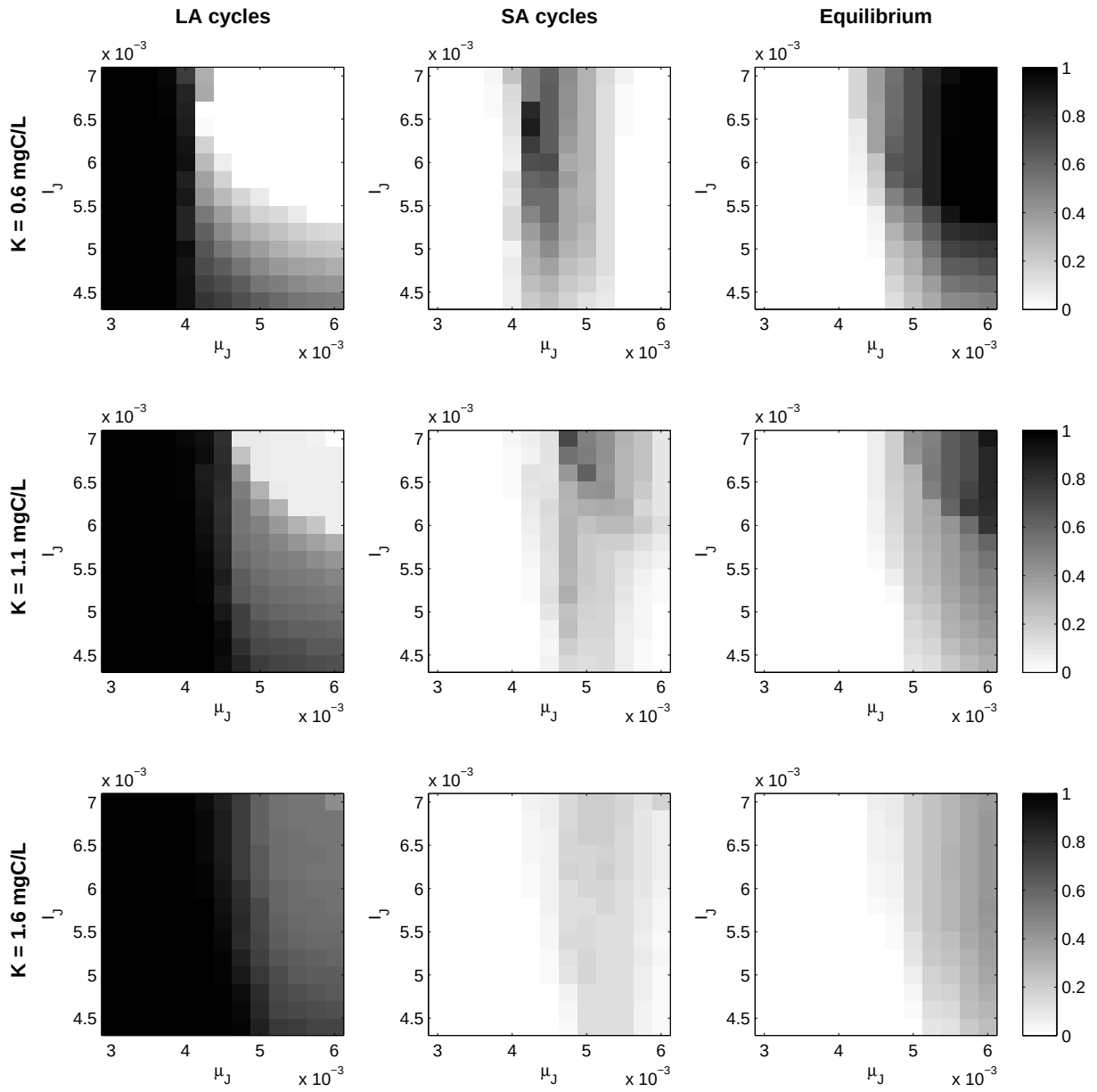

Figure 6.4: Proportion of simulations resulting in large-amplitude consumer-resource cycles, small-amplitude stage-structured cycles, or equilibrium. Data are shown for 3 different levels of enrichments.

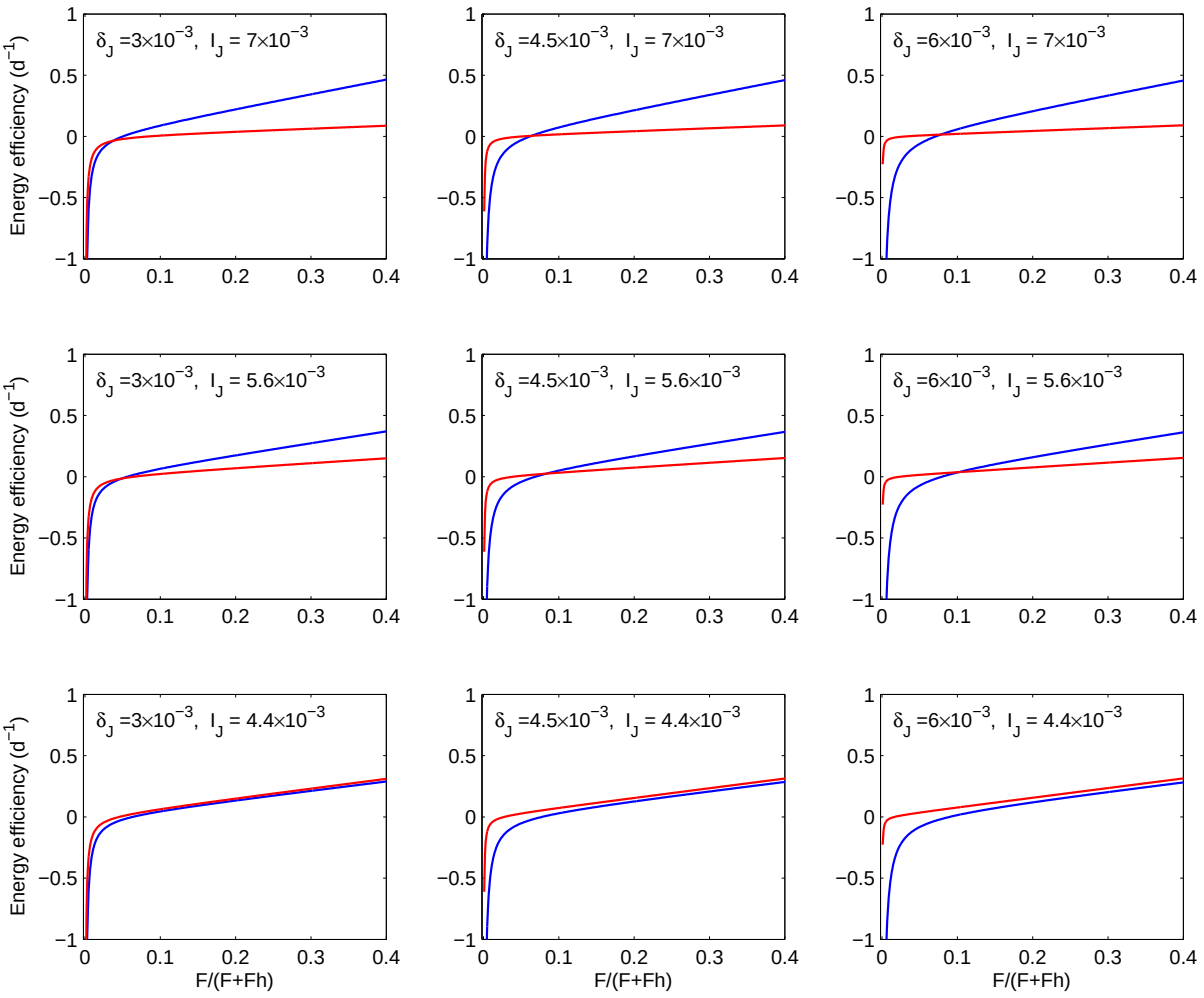

Figure 6.5: Energy efficiency of juveniles (blue) and adults (red) as a function of scaled food density for different combinations of I_J and δ_J.

Chapter 7

Concluding discussion

The work carried out in this thesis tackles issues related to the fact that the factors determining the expression of individual life history, and their consequences at higher levels of biological organization, emerge from a highly dynamical interplay between individuals and their environments. The mechanisms involved in this interplay span multiple levels of biological organizations. Genetic factors determine the energetics and the life history of individuals in response to their environments, which cascade further to populations, communities and ecosystems, to finally feed back to the individual level. Understanding the resulting implications for ecological and evolutionary processes thus requires the integration of mechanisms and feedback loops across biological scales. In this context, I focused more specifically on the role genetic and environmental variation take in this interplay.

The goal of theory is to propose mechanistic explanations for particular phenomena by evaluating the consequences of particular assumptions through modeling and assessing their coherence with observed patterns of variation (Levin 1992). In the context of ecological and evolutionary sciences, dynamic energy budgets (DEBs) provide a way of relating processes occurring over a range of biological scales (Nisbet et al. 2000). They make predictions on the life history expressed by individuals in response to specific environmental conditions, which can be used further to make predictions at the population level. Understanding the mechanism underlying model behavior implies being able to predict what happens to model output when something changes as well as to providing the reasons why.

Variation is of importance not only because of its omnipresence in the biological world, but also because it provides a fundamental avenue of testing the robustness of putatively explanatory mechanisms. Variation is often non-random: the evolutionary process may

cause directionality in genetics, ecological interactions causes specific patterns of variation in ecological components... Non-random variation may cause a linking between both the origin and the consequences of this variation, ultimately causing feedbacks in the dynamics of the various components involved. These reciprocal interactions can blur our ability to distinguish causal links between the processes involved in this interplay, and this is epitomized in consumer-resource systems. Our ability to understand the processes underlying the dynamics of these systems and predicting them thus depends on our ability to disentangle the contribution of the various components involved to the final outcome. This poses both empirical and theoretical challenges. In chapters 2 and 3, genetic variation is induced theoretically, and its effects studied through analytical sensitivity analysis. In chapters 4 and 5, this variation is induced empirically, both at the genetic level and at the environmental level.

The processes in DEB models operate on a continuous time basis, which, to some extent, has caused an impediment in the analysis of model behavior. Indeed, no method was readily available to the ecologist until now to carry out the otherwise widely used analytical sensitivity analysis (Caswell 2001). In chapter 2, I present a set of tools that makes this analysis possible for continuous-time models using the direct method from mathematical theory. This makes it possible to relate how small, genetically-based, variation in energetics affects individual phenotypic and life-history outputs. Beyond the predictive ability provided by this type of analysis, an important asset of this approach is that it makes it possible to reach some clarification on the contributions of the various processes linking the dynamics from one level of organization to another. Introducing this analysis to ecological and evolutionary sciences opens new exciting prospects for future research. In chapter 2, I have mentionned a number of fields that could benefit from this type of analysis. As I worked on this analysis and other parts of my research, I have often thought of applying this analysis on many, more specific, issues associated with state-dependent life histories and physiologically structured

population dynamics, and I hope to be able to tackle these issues in my future research. Many of these are related to the ability of this analytical framework to decompose biological variation into the contribution of their underlying components. For example, this may include decomposing the selection gradient into the contribution of fitness components, such as survival, reproduction, and age at maturity. Or again, this analysis may be very useful for studying further the effect of competitive asymmetry on consumer-resource dynamics following Chapter 6. Competitive asymmetry indeed affects the dynamics of the system because different individuals are not equally sensitive to food variation, but also because they do not impact their resource environment to the same extent due to their different foraging rates. It could be interesting to study how the contributions in these two components varies among different systems, and study whether systematic variation in these contribution may be associated with characteristic patterns in the emerging dynamics of these systems.

Among the potential issues that can be tackled with this type of analysis, I have studied how the inclusion of a feedback loop between consumer and resource dynamics modifies patterns of phenotypic expression. This ecological feedback has indeed a fundamental contribution in driving the dynamics of ecological systems, as it establishes, in the long-run, fundamental constraints that allows for population regulation (Metz et al. 1992, Murdoch 1994). It operates by altering ecological conditions and individual vital rates in the population. Although its implications for the population process have been frequently considered, its effects on the expression of individual life history have been much less so (Bassar et al. 2010). My analysis suggests logical reasons to expect a strong implication of the ecological feedback in driving the expression of individual life history. A next step in this investigation would be to test these inferences experimentally using controlled laboratory experiments under conditions of density dependence. From a more theoretical perspective, it would be interesting to study as well these issues under non-equilbrium conditions. I would then expect that similar conclusions on the symmetry between direct effects of the perturbations and

feedback-mediated effects hold in the long run. Nonetheless, this result would not necessarily apply directly to cohorts of individuals born at different time, as they may be subjected to these effects to a different extent. Understanding the resulting implications for the dynamics of the systems, the mechanisms of expression of individual life history and the selective process would benefit from this analysis.

The ecological feedback induces qualitative changes in the expression of individual life history because it causes environmental variation associated with changes in consumer biology. Another type of environmental variation inducing qualitative changes is when the environment varies in a dynamic fashion on a time-scale comparable to the individual lifetime. Chapter 4 shows that environmental fluctuations induce hysteresis in individual energetics. This conclusion was only possible because I used manipulative experiments that decoupled the dynamics of resource production from the consumer dynamics, effectively disentangling the effect of the energetic response of the population to environmental fluctuations from the feedback loop induced by the consumer-resource interaction. This hysteretic effect was dependent on the patterns of environmental fluctuations. The relative contribution of the environmental driver versus consumer biology to emerging demographic patterns was found to vary as a function of the intensity of the environmental fluctuations. This caused an emerging genotype-environment interaction on population demographical patterns, which has implications for both the selective regime and ecological processes, such as the mechanisms maintaining genetic diversity in the wild. This works focused on how different genetic variants respond to similar patterns of food variation, but does not allow predictions on the total effect because in natural systems, resource dynamics are linked to those of their consumers through a feedback loop. I do not consider the limitations as a drawback of my experimental set-up, as they makes it possible to reach conclusions on individual processes that would have been hardly reachable otherwise. Nonetheless, it would be exciting to follow these works with similar type of experiments when the feedback loop is operational. This

work also showed that much can be learnt on individual functioning from population data. The classical approach to linking individual processes with population patterns of variation is to carry out physiological experiments on individuals and make predictions on populations patterns. Yet, since populations patterns are the product of individual processes, it is possible to do the opposite, an approach known as 'inverse analysis'. This approach remains seldom used in ecology, despite the fact that population data are much more easily available than data on individuals, particularly under natural conditions. My study provides further support for the usefulness of this approach. The main problem with this approach, that I have been confronted to all along this analysis, is non-uniqueness: there are a number of possible inferences on individual processes that may predict the data equally well. Statistical solutions to this problem have been proposed, and I showed that carefully designed experimental set-up may also provide a powerful way to validate those estimates. Another possible alternative would have been to record the demography of the populations and also follow concurently the fate of some individuals over their lifetime, which would largely constrain the range of possible solutions.

The genomic analysis made it possible to refine these results further by showing that the mapping between genotype and phenotype is not the only component involved in explaining the genotype-environment interaction found in demographic pattern, but the map linking individual to population behavior is also a likely contributor. Indeed, genetic variation induced more differential gene expression in large-amplitude cycles than in small-amplitude cycles, which may seem otherwise paradoxical. These results are however only suggestive without further analysis and additional experiments, as several hypotheses could explain these patterns. In any circumstance, a large number of genes were found to be subjected to a genotype-environment interaction in their patterns of expression. This analysis also produced strong support for the importance of consumer-resource interactions for the functioning of biological organisms: up to 84% of all the protein-encoding genes were differentially

expressed as a result of food variation. Much analysis remains to be done on these data, and in particular, correlating patterns of gene expression with phenotypic and environmental components is likely to bring further insights on these issues. It will help identifying the role of genes in term of their ecological effects, and discriminate those of greater importance. This may then open the way for further investigations on the interplay between genetic and ecological processes, and may also open the way for a number of pratical applications, by for example, using gene expression measurements as markers of the ecological history of individuals.

Competitive asymmetry between individuals in different states has been shown to be able to explain major patterns in population and community dynamics and structure. Nonetheless, the dynamics the dynamics of *Daphnia*-algae systems could not be fully explained by this theory as yet because there were seemingly contradictory evidences as to whether juveniles or adults are competitively dominant (De Roos and Persson 2013). One aspect of ontogenetic asymmetry suggested by the previous analysis, and previously overlooked from these lines of research, is that environmental variation may induce alternative competitive rankings among individuals in the population, and more generally induce a variable degree of competitive asymmetry as a function of the conditions experienced. Through the analysis of a simplified model, I showed that such consideration may solve for these contradictory evidences on the dynamics of *Daphnia*-algae systems and bring some insights on the mechanisms involved in the stabilization of consumer-resource dynamics and the prevalence of various kinds of dynamics. I have showed that such consideration is likely to bring insights on the prevalence of small-amplitude stage-structured cycles over large-amplitude consumer-resource cycles. Another remaining open question is how frequent alternative competitive rankings happens in other systems. Studying these issues may require an extensive analysis of previously published data on individual energetics, but the specificity of the data required for this analysis makes it more likely for experiments that address directly these issues to be

more productive.

All these lines of work give further support for the importance of environmental and genetic variation on the ecological and evolutionary processes. A comprehensive ecological and evolutionary theory needs to account for the different ways this variation is expressed. Given the numerous ways variation can be expressed in the wild, we certainly cannot hope to make a systematic account of all these patterns, and abstraction need to be done at some level in order to reach some form of generality. The issues investigated in this thesis show that these effects span different levels of generality, depending on which aspect of the dynamics is being looked at. Rather than asking whether genetic and environmental variability matter for the dynamics of ecological systems, this suggests that progressing on these issues will rather depend upon our ability to disentangle and compartmentalize which aspects of the dynamics require an account of variability to be understood, and which aspects can be understood despite this variability; and for those responding to variability, whether predictions require specific accounts of the patterns and nature of the variability or whether generic conclusions can be drawn.

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Appendix A

Chapter 4 Appendices

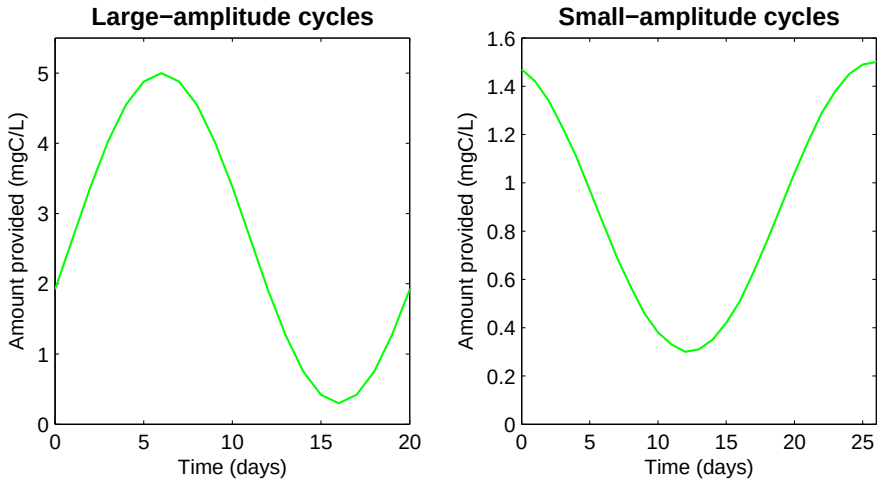

Figure A.1: Daily amount of algae provided in large- and small-amplitude treatments.

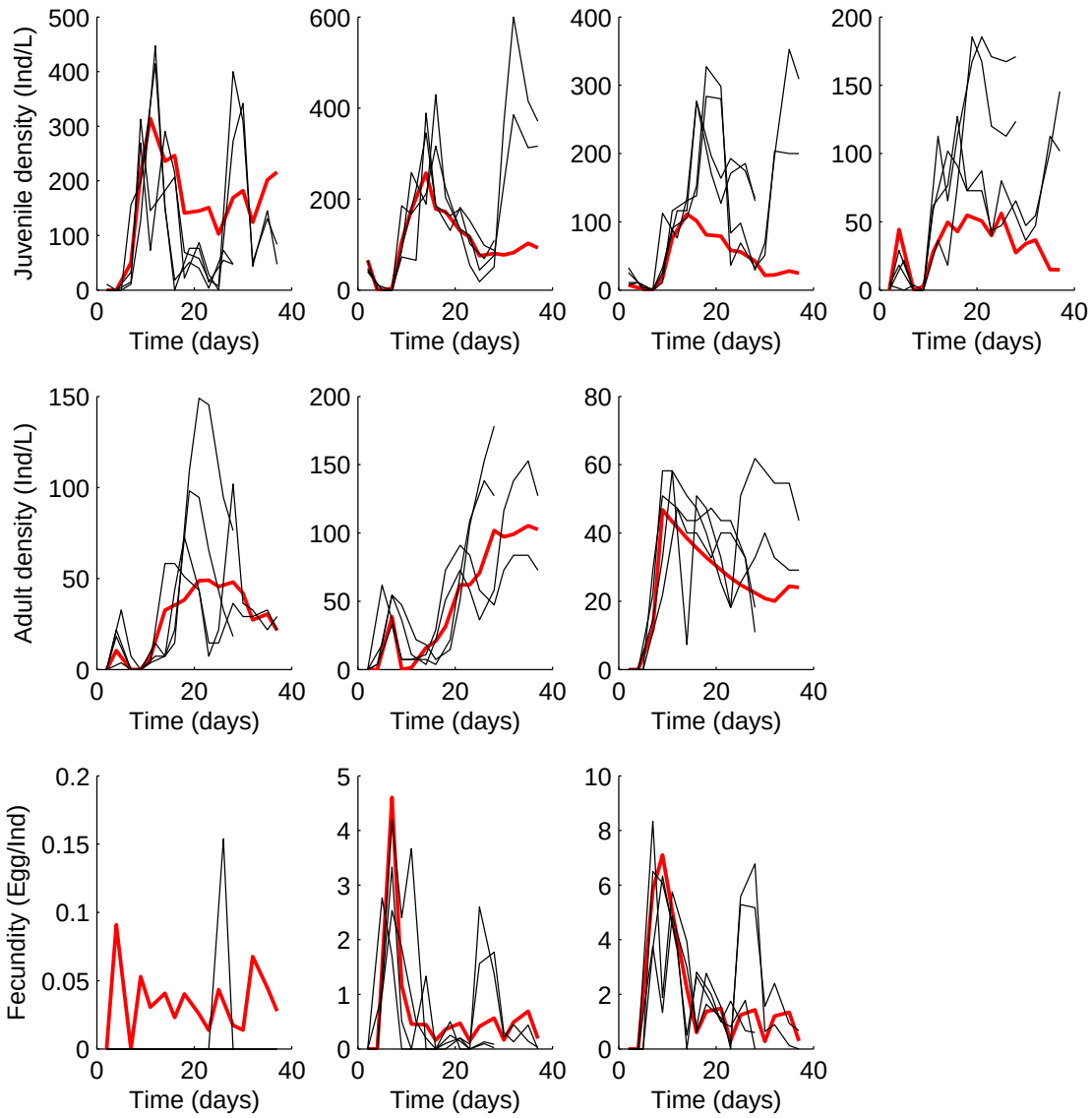

Figure A.2: Dynamics of the different classes in the constant food supply experiment for clone A. Starting from the upper-left graph and going to the bottom-right, size classes are < 0.8mm, 0.8-1mm, 1-1.2mm, 1.2-1.4mm, 1.4-1.6mm, 1.6-2mm, > 2mm. The last 3 graphs are the fecundities of the corresponding adult classes. Black lines are experimental data, red lines are model fits to the data.

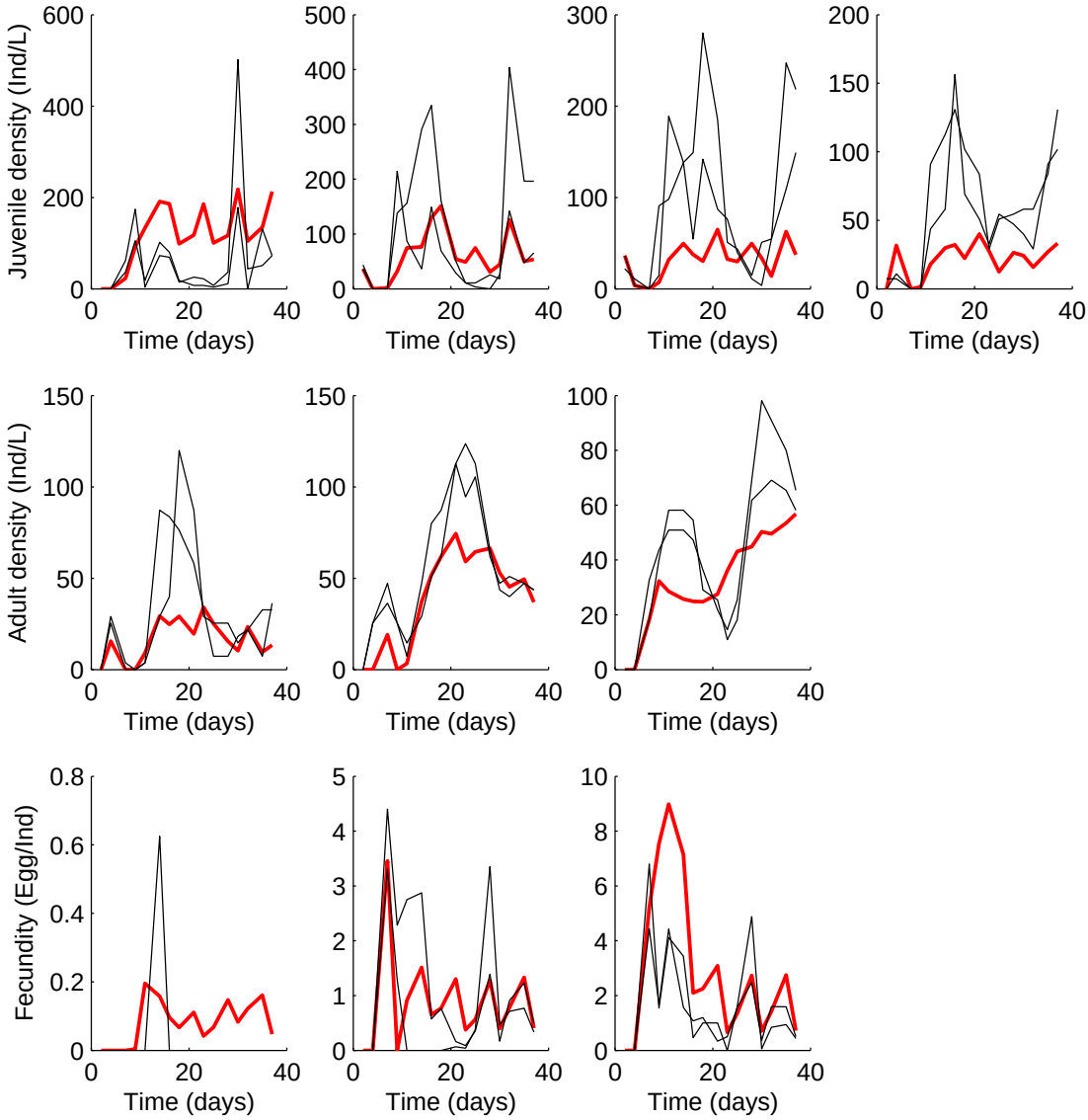

Figure A.3: Dynamics of the different classes in the constant food supply experiment for clone B. The graphs are ordered as in Figure A.2.

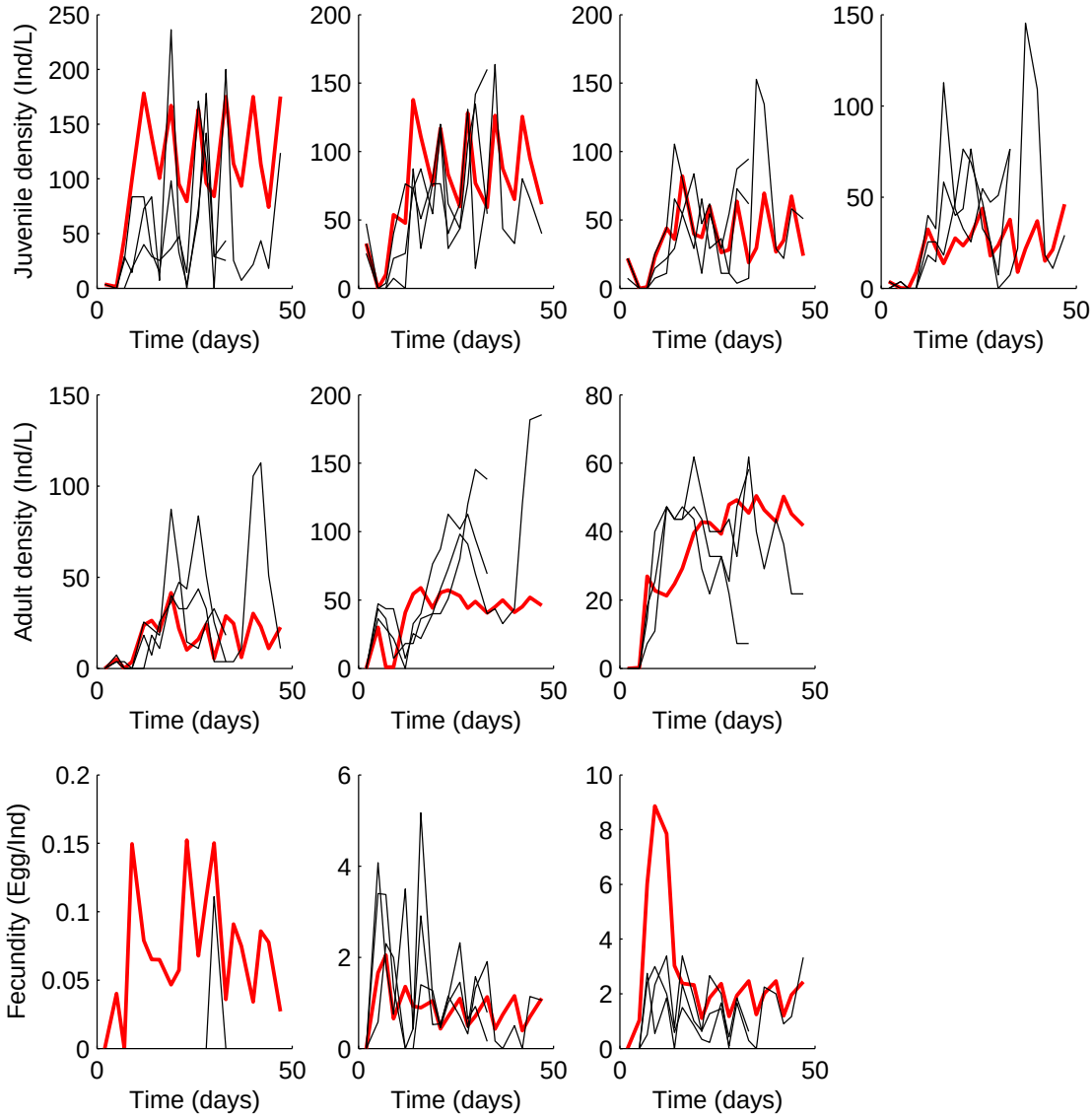

Figure A.4: Dynamics of the different classes in the constant food supply experiment for clone C. The graphs are ordered as in Figure A.2.

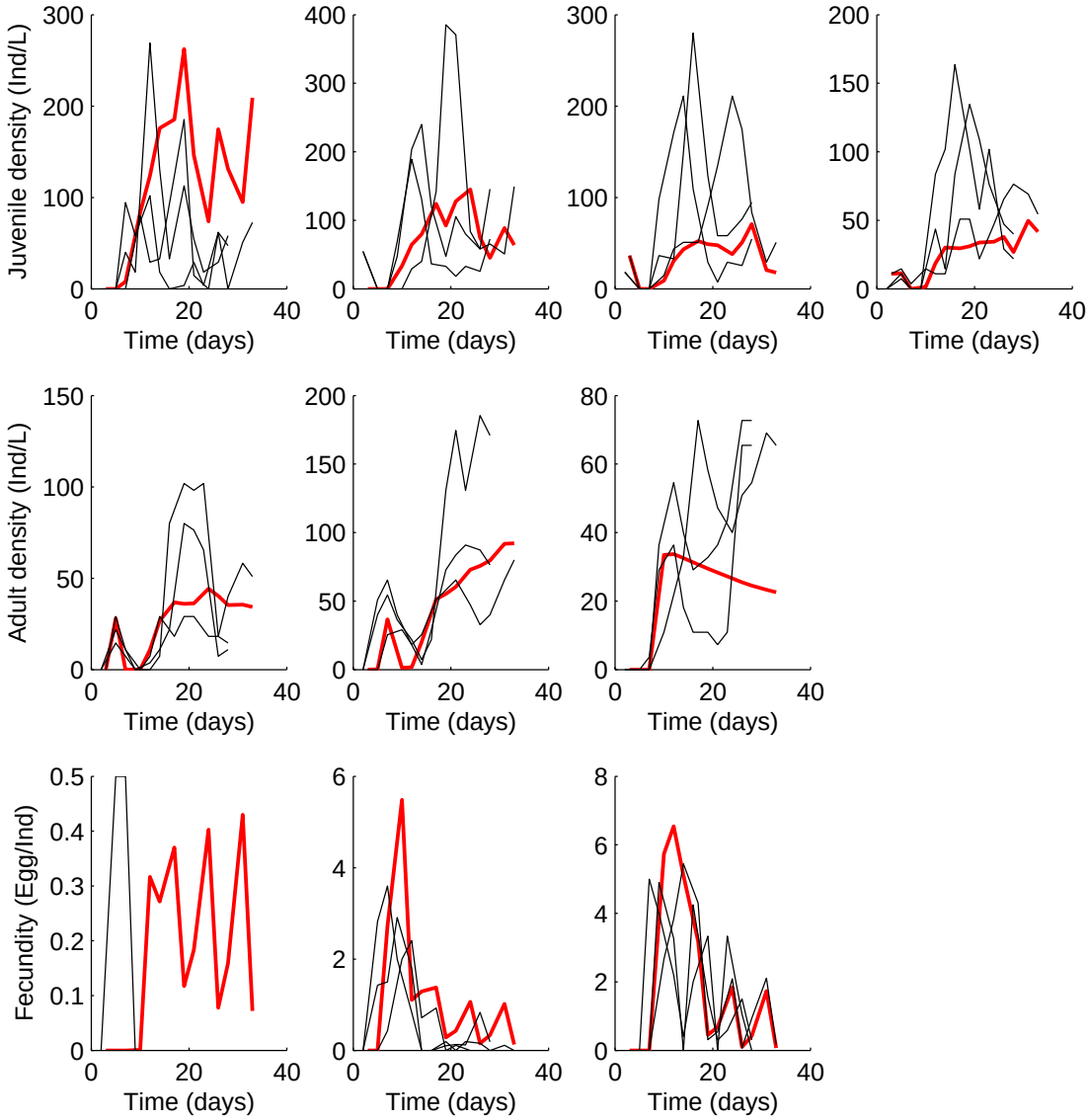

Figure A.5: Dynamics of the different classes in the constant food supply experiment for clone D. The graphs are ordered as in Figure A.2.

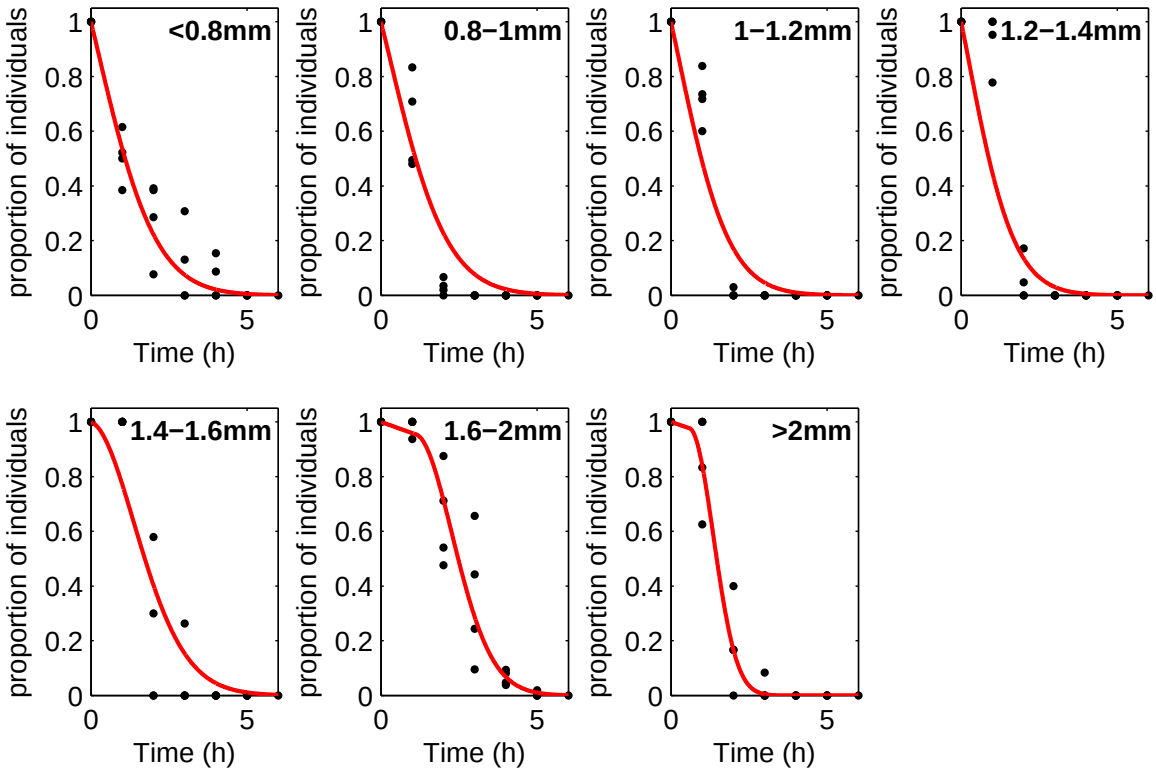

Figure A.6: Proportion of surviving individuals as a function of time under starving conditions for clone A. Black lines are experimental data, red lines are model fits to the data.

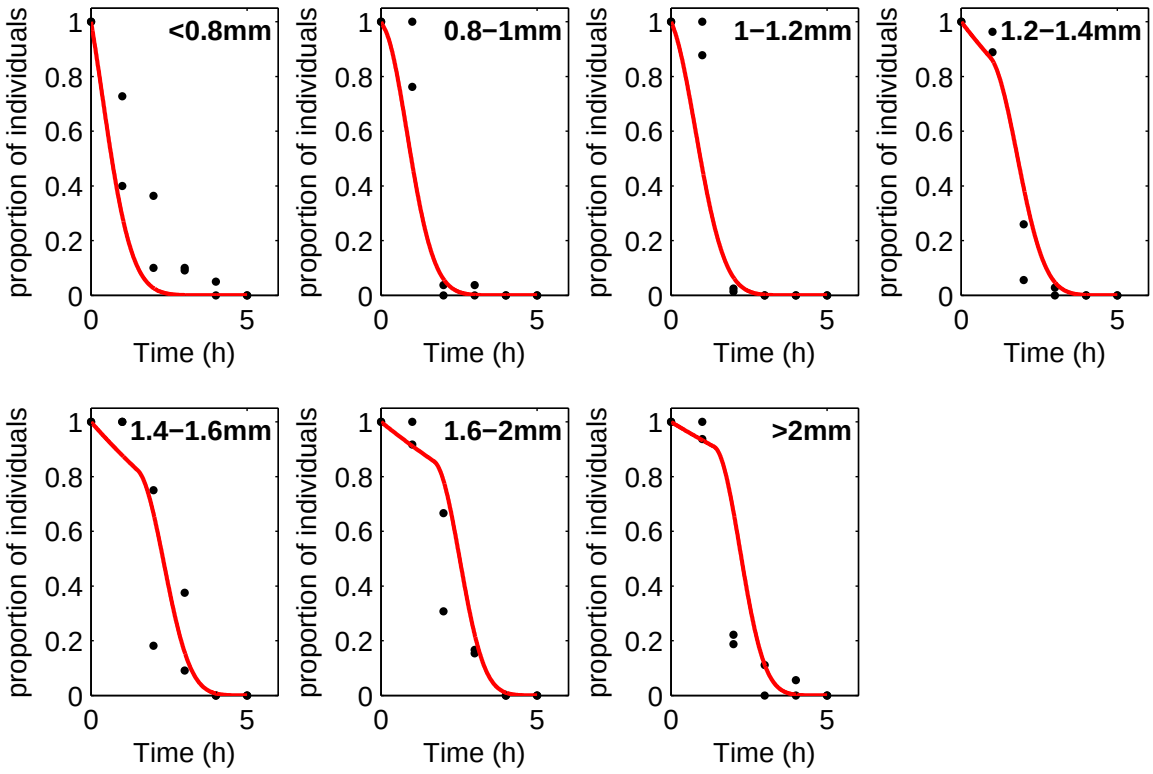

Figure A.7: Proportion of surviving individuals as a function of time under starving conditions for clone B. Black lines are experimental data, red lines are model fits to the data.

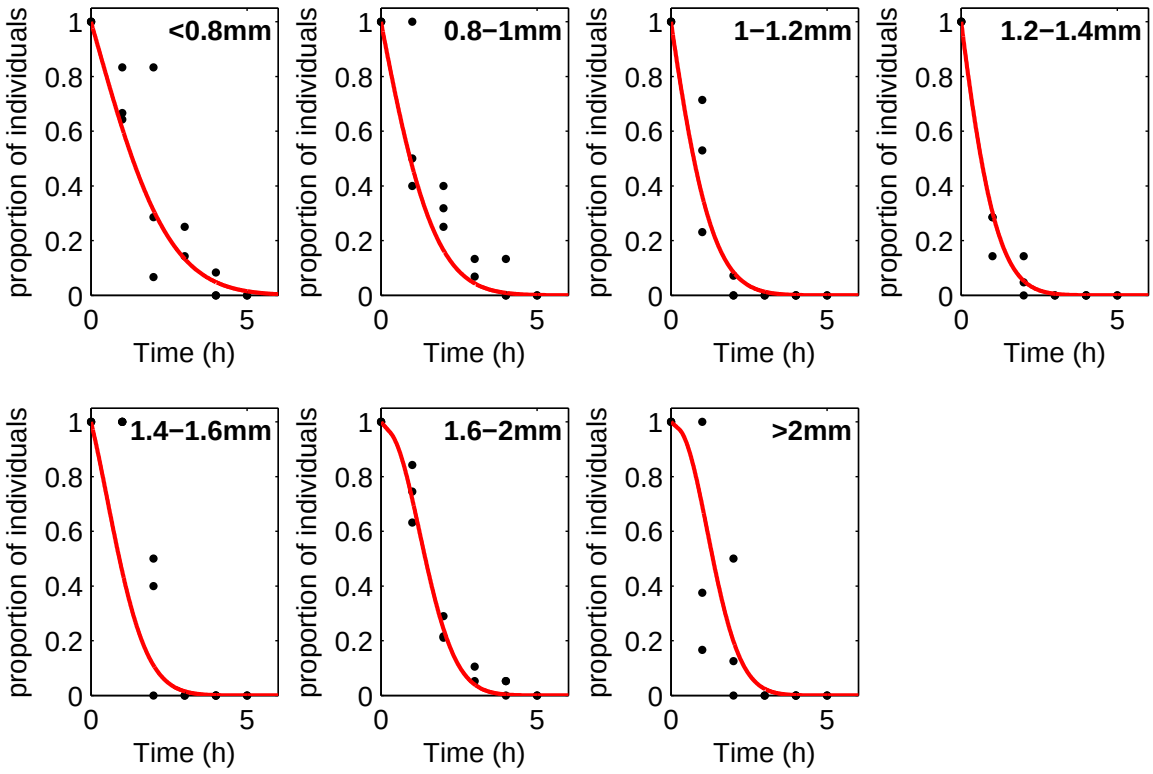

Figure A.8: Proportion of surviving individuals as a function of time under starving conditions for clone C. Black lines are experimental data, red lines are model fits to the data.

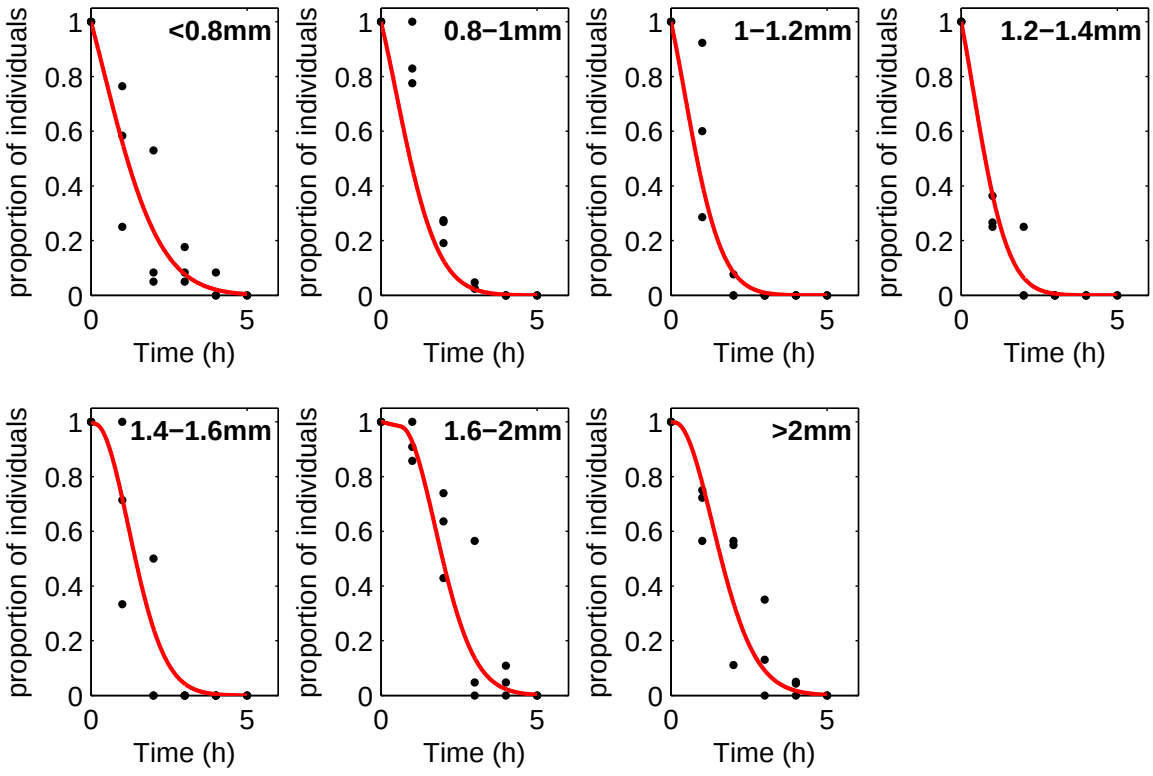

Figure A.9: Proportion of surviving individuals as a function of time under starving conditions for clone D. Black lines are experimental data, red lines are model fits to the data.

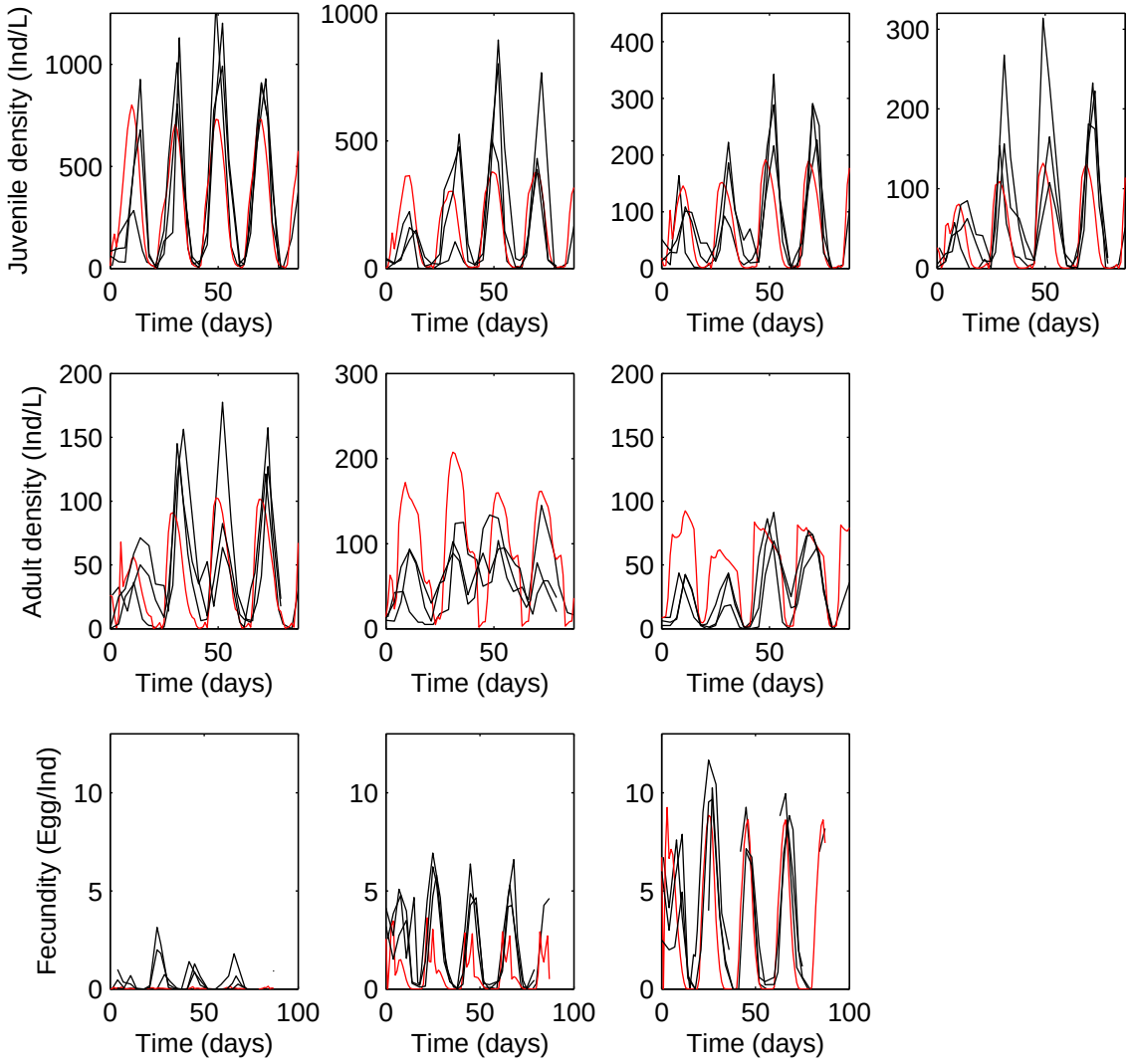

Figure A.10: Dynamics of the different classes in the large-amplitude food supply experiment for clone A. The graphs are ordered as in Figure A.2. Black lines are experimental data, red lines are model predictions.

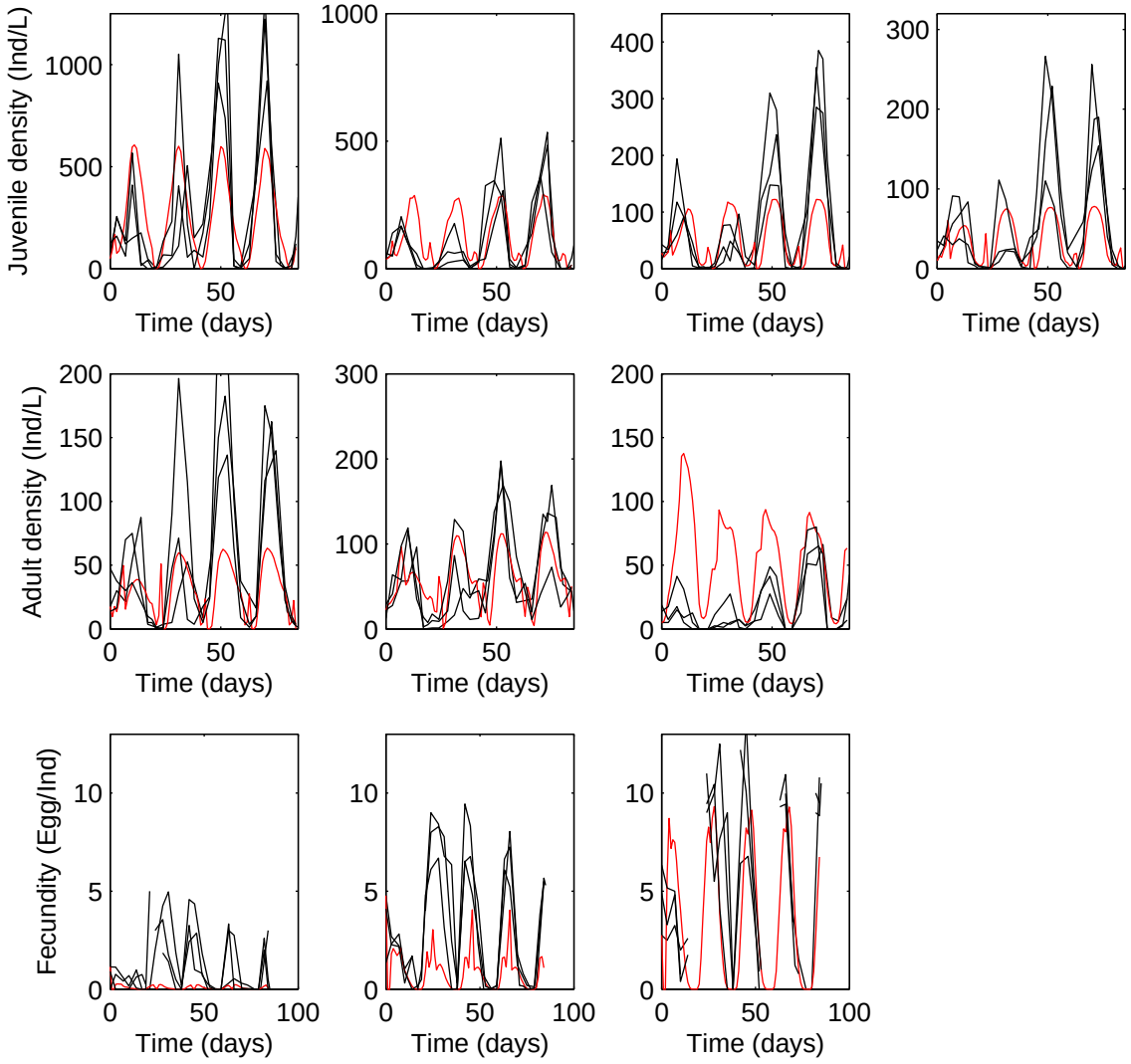

Figure A.11: Dynamics of the different classes in the large-amplitude food supply experiment for clone B. The graphs are ordered as in Figure A.2.

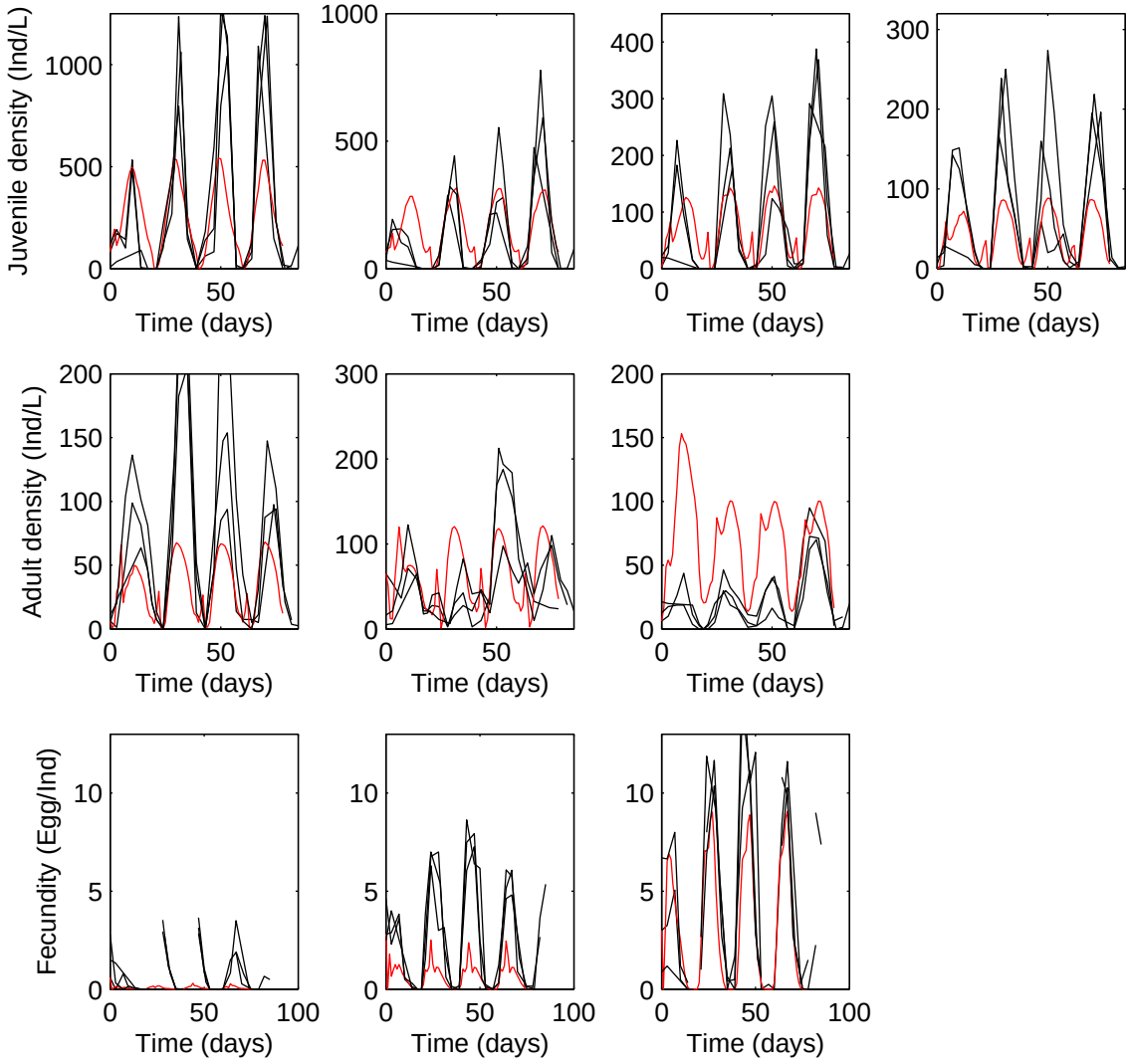

Figure A.12: Dynamics of the different classes in the large-amplitude food supply experiment for clone C. The graphs are ordered as in Figure A.2.

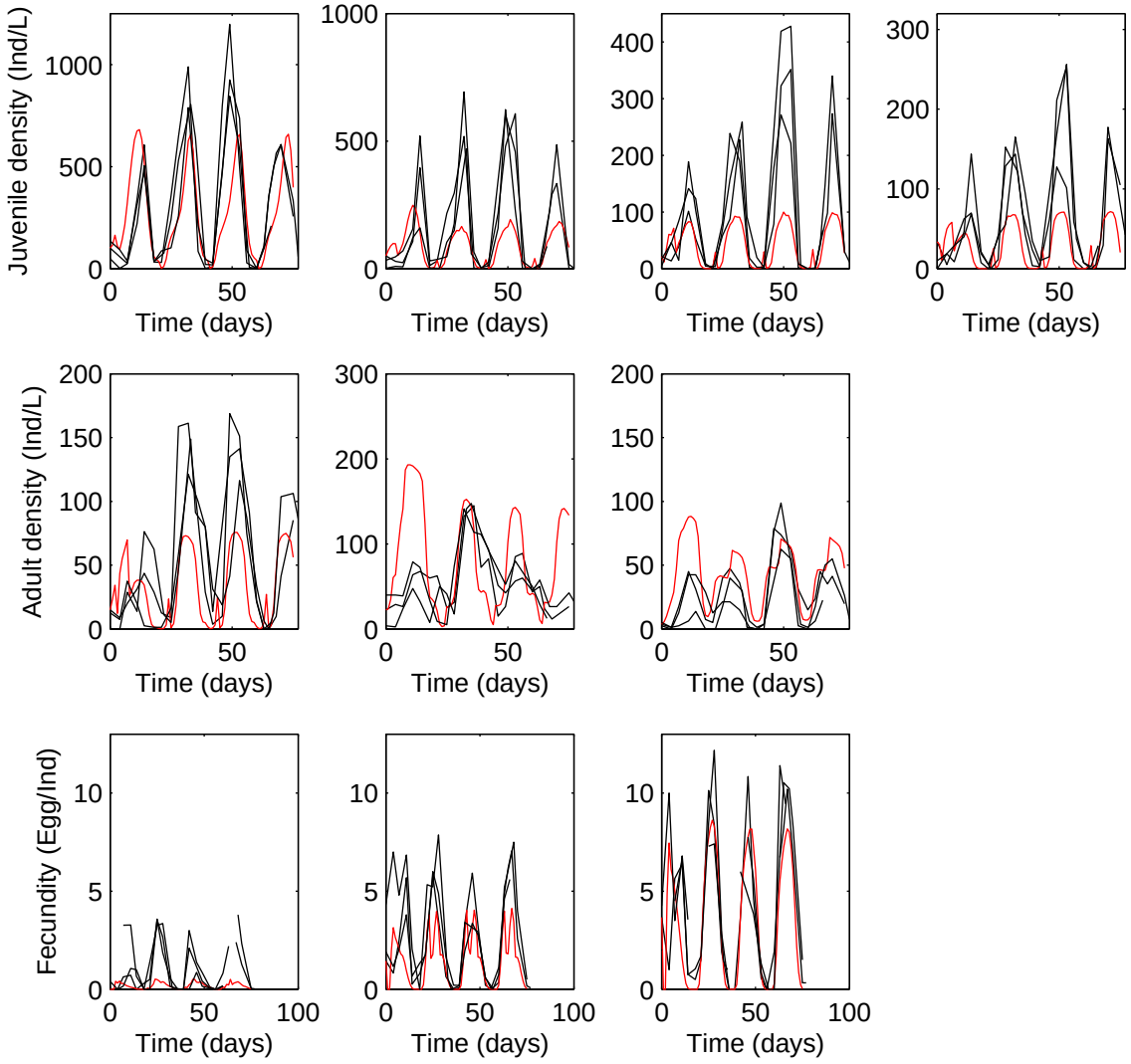

Figure A.13: Dynamics of the different classes in the large-amplitude food supply experiment for clone D. The graphs are ordered as in Figure A.2.

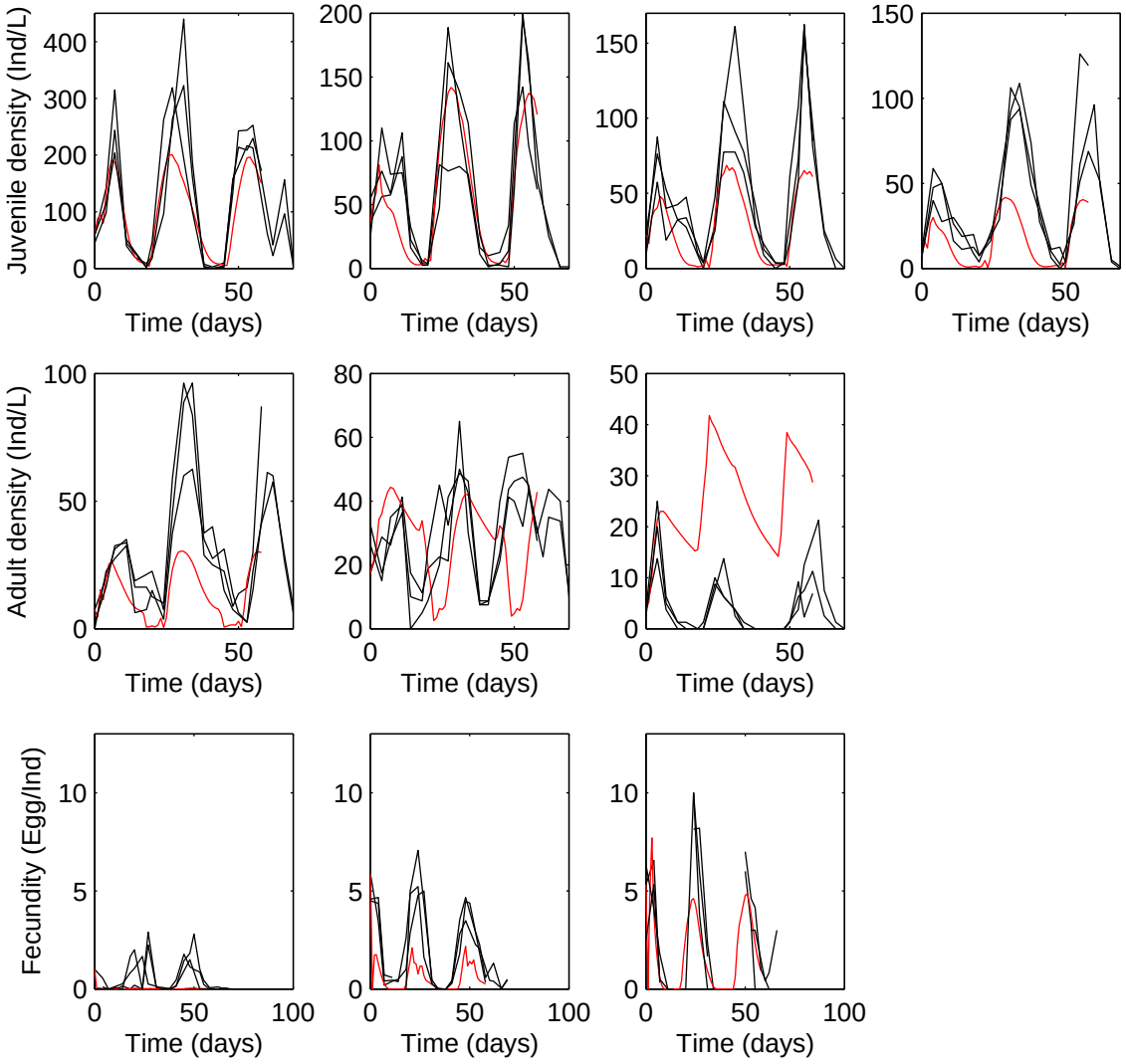

Figure A.14: Dynamics of the different classes in the small-amplitude food supply experiment for clone A. The graphs are ordered as in Figure A.2.Black lines are experimental data, red lines are model predictions.

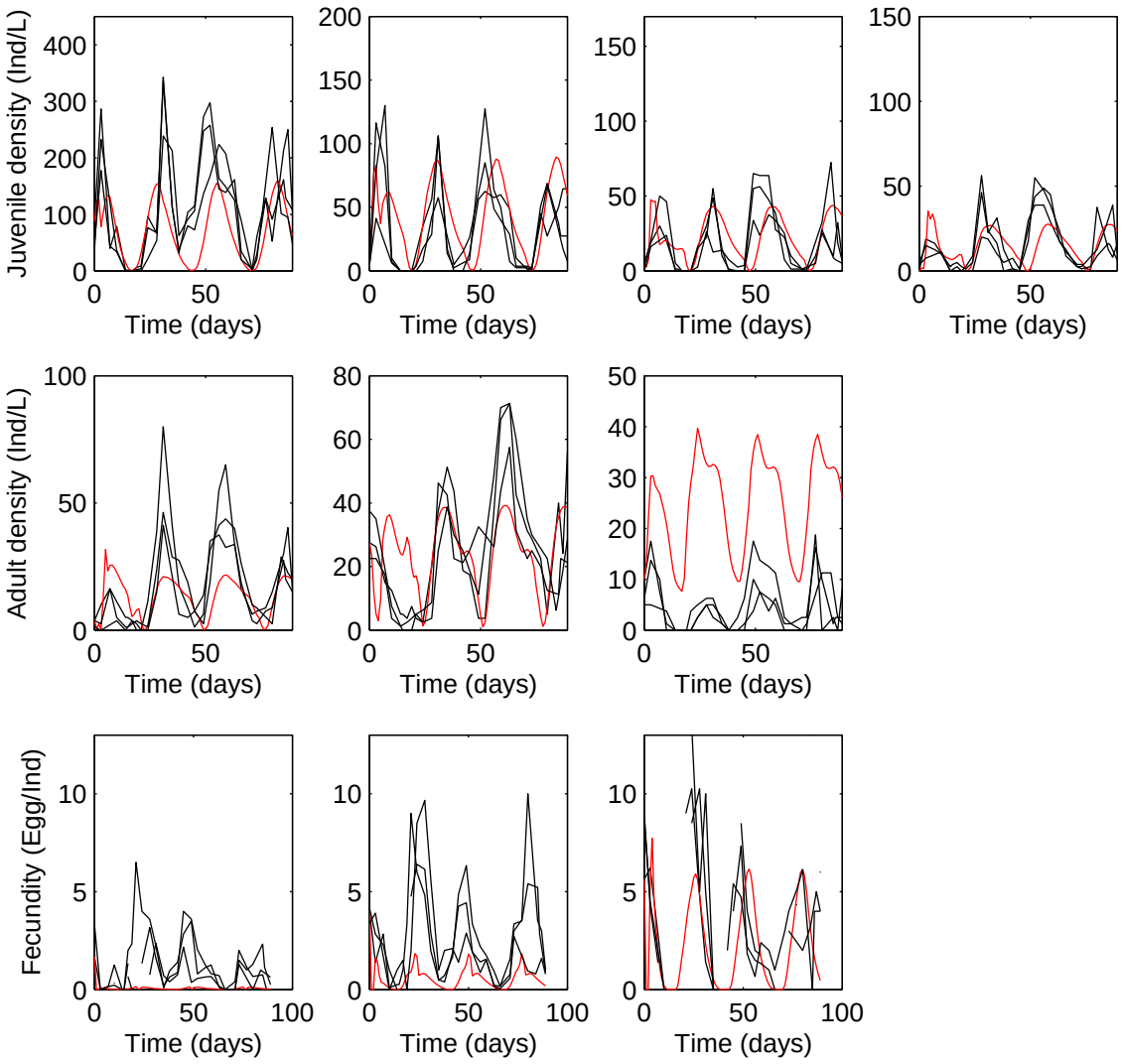

Figure A.15: Dynamics of the different classes in the small-amplitude food supply experiment for clone B. The graphs are ordered as in Figure A.2.

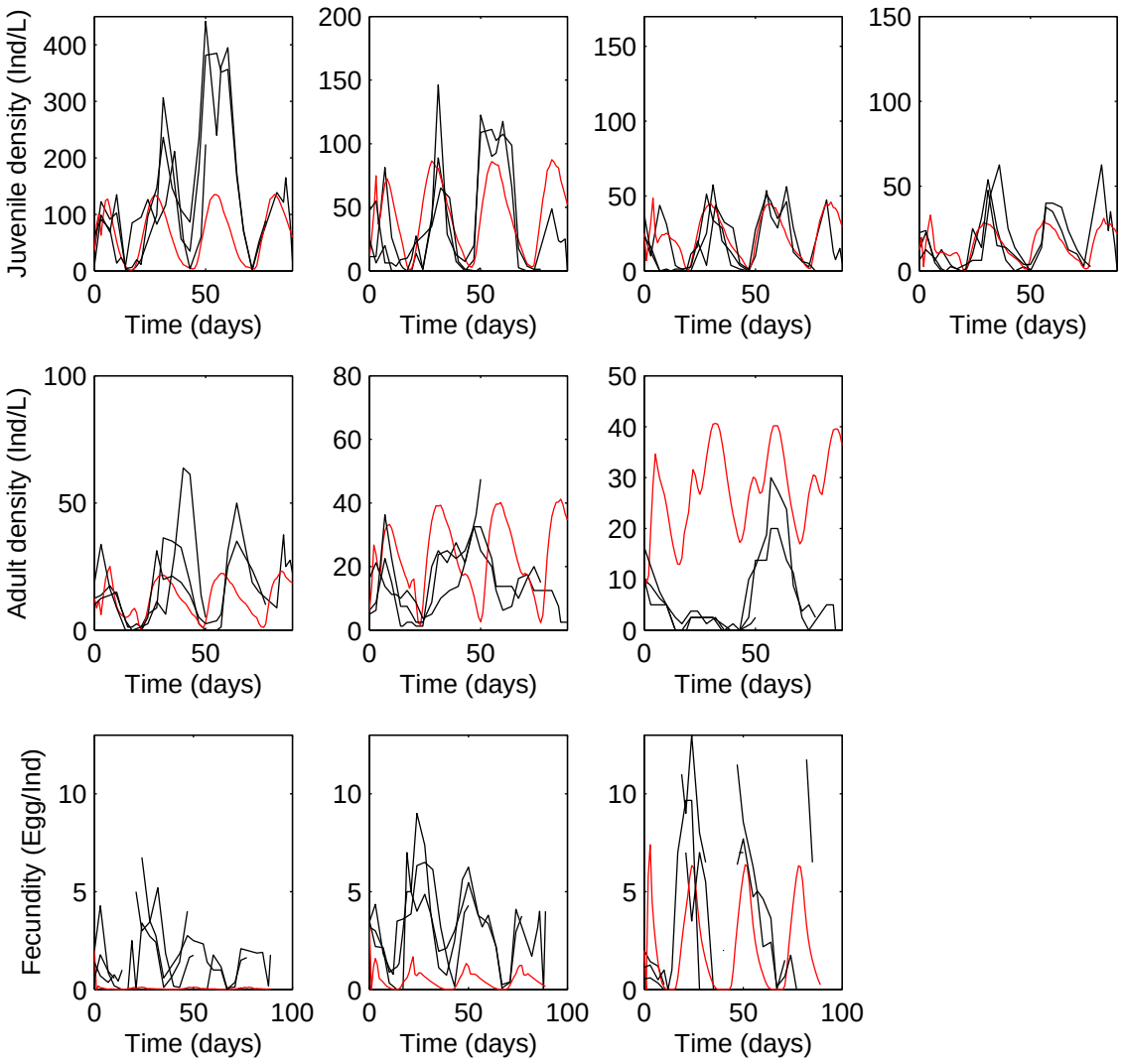

Figure A.16: Dynamics of the different classes in the small-amplitude food supply experiment for clone C. The graphs are ordered as in Figure A.2.

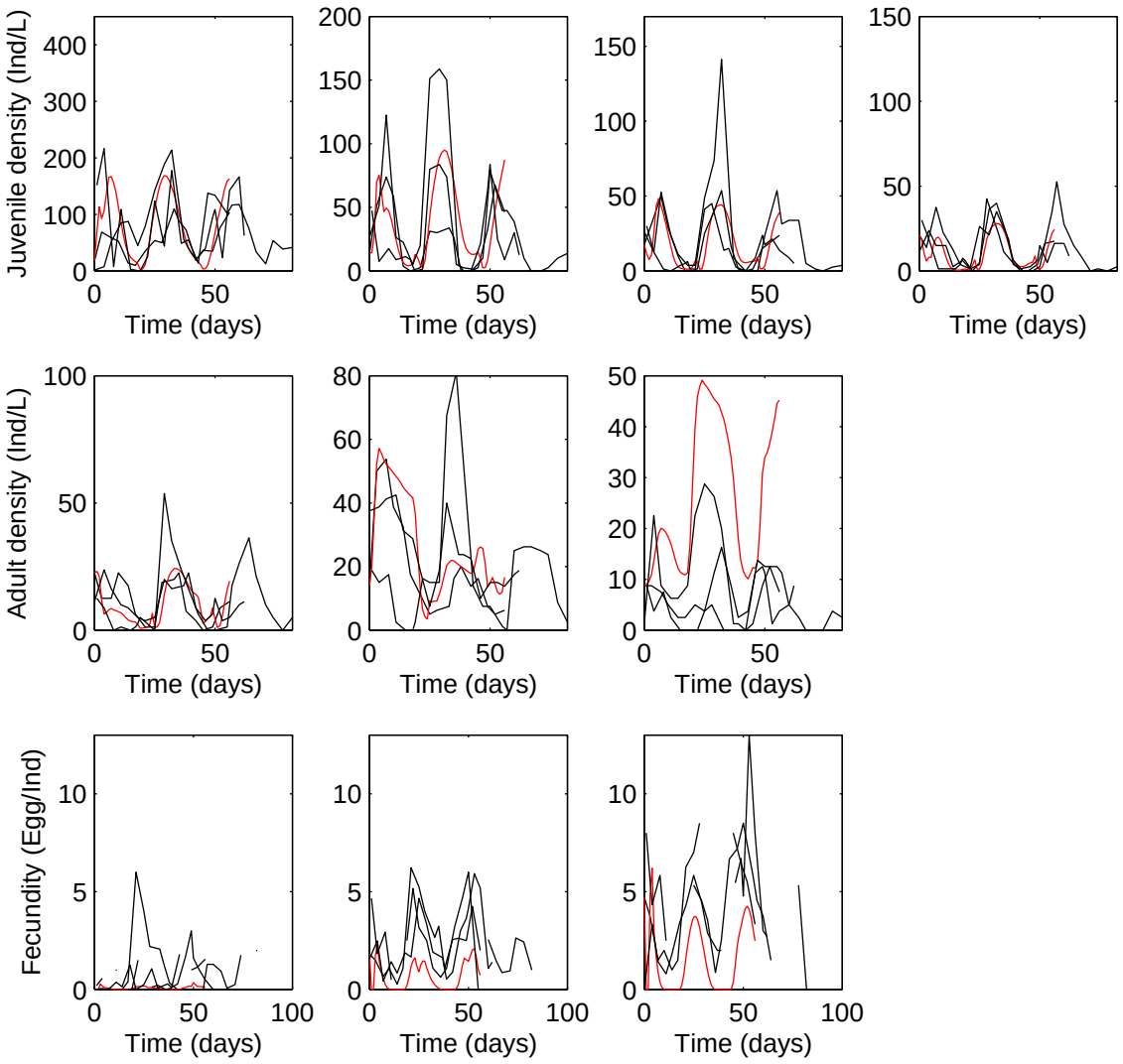

Figure A.17: Dynamics of the different classes in the small-amplitude food supply experiment for clone D. The graphs are ordered as in Figure A.2.

Parameter Values

ϵ_A	0.55
γ	2
T_m	1.5 d

Model Functions

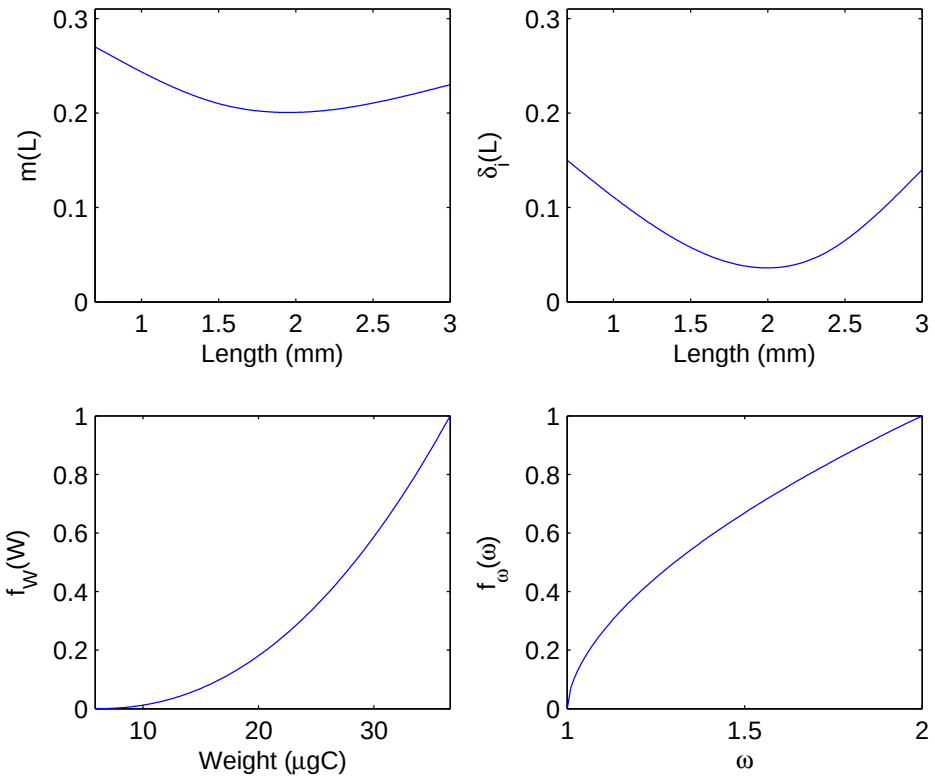

Figure A.18: Parameter values and estimated model functions for clone A (see main text for details).

Parameter Values

ϵ_A	0.53
γ	1.48
T_m	2.05 d

Model Functions

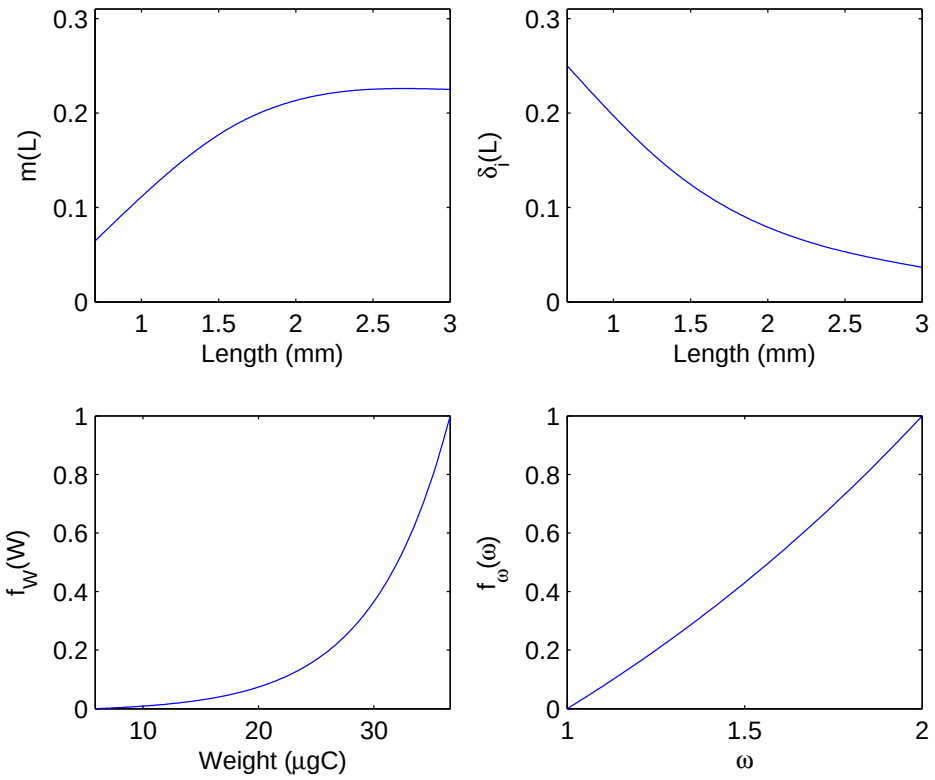

Figure A.19: Parameter values and estimated model functions for clone B (see main text for details).

Parameter Values

ϵ_A	0.58
γ	1.29
T_m	1.64 d

Model Functions

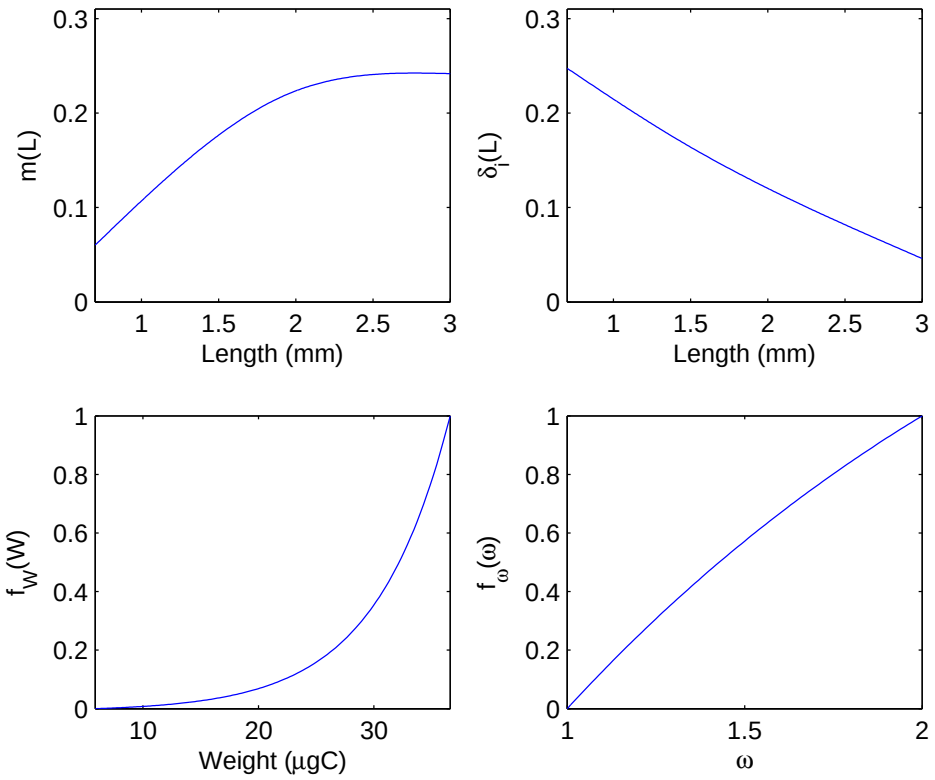

Figure A.20: Parameter values and estimated model functions for clone C (see main text for details).

Parameter Values

ϵ_A	0.53
γ	1.98
T_m	2.15 d

Model Functions

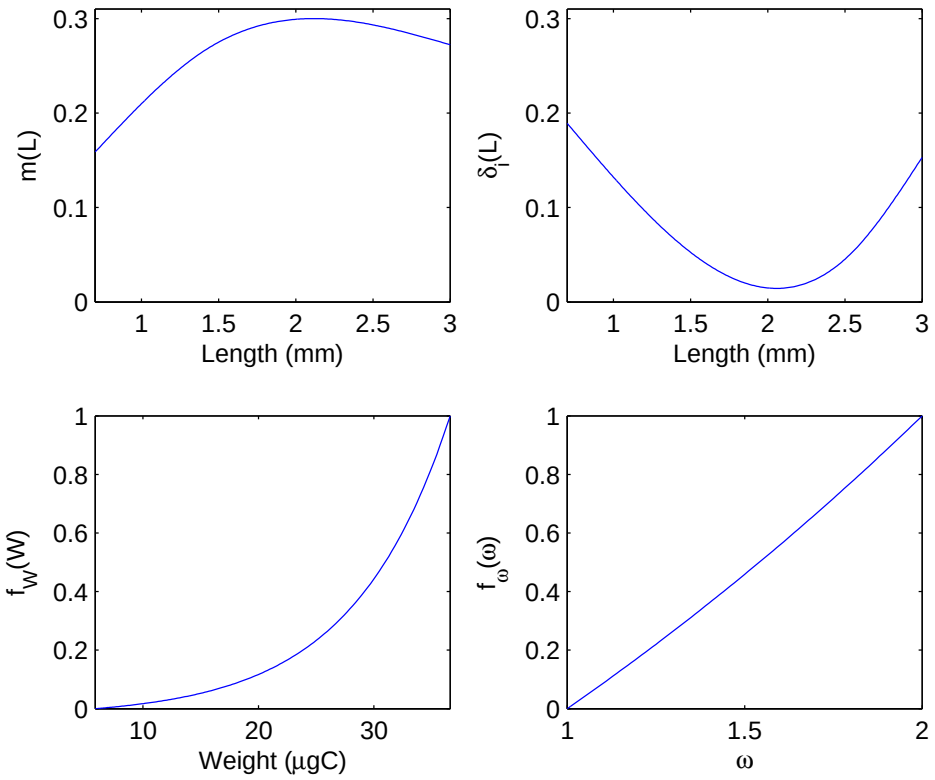

Figure A.21: Parameter values and estimated model functions for clone D (see main text for details).

Appendix B

Chapter 5 Appendices

Table B.1: Microsatellite loci used to characterize clones. Names correspond to those reported in Colbourne et al. (2004). Accession numbers are for the *Daphnia* Genomics Consortium website (wFleaBase). More details on the methods for characterizing these loci can be found in the original publication (Colbourne et al. 2004).

Name	Locus	Accession #	Size (base pairs)
Dp70	P1-M12	WFms0000072	239
Dp126	P2-B21	WFms0000129	163
Dp142	P2-H17	WFms0000146	319
Dp143	P2-H21	WFms0000147	364
Dp156	P2-N17	WFms0000160	136
Dp300	P4-K10	WFms0000309	329
Dp308	P4-M19;P4-L19	WFms0000318	223
Dp395	P5-M14	WFms0000409	207

Table B.2: Quantity and quality of total RNA extracted.

See text for abbreviations.

Clone	Cycle	Phase	OD260/280	OD260/230	Concentration (µg/µl)	Volume (µl)
A	LA	P	2.06	2.21	329.2	14
		P	2.06	2.19	408.2	14
		P	2.03	2.28	351	14
		D	2.03	2.24	172.5	14
		D	2.02	2	196.5	14
		D	2.04	2.23	64.6	40
		N	1.97	1.98	97.8	14
		N	1.92	1.82	142.1	14
		N	1.98	2.08	103.3	14
		G	2.03	2.18	695.1	14
		G	2.03	2.05	651.9	14
		G	2.07	2.3	564.1	14
	SA	P	1.96	1.85	49.6	40
		P	1.8	1.38	17.2	40
		D	2	1.89	249.5	14
		D	2.04	1.96	71.7	40
		D	2.01	2.05	198.9	14
		N	1.95	1.85	66.8	14
		N	1.99	2	123.7	14
		N	1.95	1.22	25.7	40
		G	2.05	2.28	420	14
		G	2.06	2.25	386.1	14
		G	2.04	2.09	281.6	14

Table B.2: Continued.

Clone	Cycle	Phase	OD260/280	OD260/230	Concentration (µg/µl)	Volume (µl)
B	LA	P	2.06	2.31	104.4	40
		P	2.02	2.11	290	14
		P	2.08	2.32	128.3	40
		D	1.99	2.23	56.2	40
		D	1.97	2.28	45.5	40
		D	2.03	2.18	43.1	40
		N	2.02	2.16	178.3	14
		N	2.02	2.04	170.7	14
		N	1.96	2.23	33	40
		G	2.09	2.15	75.9	40
		G	2.1	2.32	308.4	40
		G	2.09	2.32	756.6	14
	SA	P	1.84	1.28	23.6	40
		P	1.95	1.95	22	40
		D	1.91	1.85	110.3	14
		D	2.08	2.36	614.6	14
		D	1.94	1.84	29.2	40
		N	2.02	2.34	41.6	40
		N	1.9	2.42	20.7	40
		N	2.07	2.5	42.1	40
		G	2.07	2.27	183.4	40
		G	2.05	2.01	437	14
		G	2.06	2.38	368.7	14

Table B.3: GO categories over-represented among the gene differentially expressed in the different type of comparisons.

			Term	GO Accession number	p-value
Variation in transcript abundance	Effect of clone type	LA	protein amino acid phosphorylation	6468	7.39E-07
			protein-nucleus import. docking	59	2.06E-05
			protein folding	6457	0.00014
			cysteinyl-tRNA aminoacylation	6423	0.00039
			lipoprotein metabolism	42157	0.00205
			protein ubiquitination	16567	0.00443
			protein modification	6464	0.00467
			sensory perception	7600	0.00553
			steroid metabolism	8202	0.00553
		SA	protein amino acid phosphorylation	6468	4.68E-06
			transport	6810	0.00161
			proteolysis and peptidolysis	6508	0.00193
			sensory perception	7600	0.00432
			phototransduction	7602	0.00462
			protein complex assembly	6461	0.00687
			oxygen transport	15671	0.00801
			lipoprotein metabolism	42157	0.00834
	Effect of cycle type	cA	phototransduction	7602	2.53E-05
			sensory perception	7600	0.00033
			oxygen transport	15671	0.00033
			metabolism	8152	0.00073
			cellular protein metabolism	44267	0.00106
			cation transport	6812	0.00171
			protein folding	6457	0.00413
			peptide cross-linking	18149	0.00548
			de novo' pyrimidine base biosynthesis	6207	0.00653
			antibiotic biosynthesis	17000	0.00927
			protein-nucleus import. docking	59	0.00963

Table B.3: Continued.

			Term	GO Accession number	p-value
Variation in transcript abundance	Effect of cycle type	cB	protein amino acid dephosphorylation	6470	4.52E-06
			protein amino acid phosphorylation	6468	4.30E-05
			carbohydrate metabolism	5975	0.00038
			DNA metabolism	6259	0.00045
			mismatch repair	6298	0.00074
			protein-nucleus import. docking	59	0.00082
			protein folding	6457	0.00165
			regulation of pH	6885	0.00299
			regulation of translational initiation	6446	0.00424
			cellular protein metabolism	44267	0.00511
			signal transduction	7165	0.00574
			ubiquitin-dependent protein catabolism	6511	0.00590
			cation transport	6812	0.00779
			regulation of GTPase activity	43087	0.00835
			steroid metabolism	8202	0.00894
			microtubule-based movement	7018	0.00938
Within-cycle variation in transcript abundance	direct comparisons	cALA	ubiquitin-dependent protein catabolism	6511	1.49E-07
			DNA replication initiation	6270	3.44E-07
			cellular protein metabolism	44267	7.17E-07
			protein amino acid phosphorylation	6468	7.36E-06
			nucleobase. nucleoside. nucleotide and nucleic acid metabolism	6139	1.09E-05
			protein-nucleus import. docking	59	1.70E-05
			protein folding	6457	3.51E-05
			DNA replication	6260	0.00022
			chromatin modification	16568	0.00053
			D-ribose metabolism	6014	0.00085
			cell-matrix adhesion	7160	0.00180
			translational termination	6415	0.00388
			mitosis	7067	0.00388
			frizzled-2 signaling pathway	7223	0.00518
			protein polymerization	51258	0.00566
			nucleoside metabolism	9116	0.00650
			transmembrane receptor protein tyrosine kinase signaling pathway	7169	0.00668
			glycine biosynthesis	6545	0.00669
			tetrahydrobiopterin biosynthesis	6729	0.00669
			trehalose biosynthesis	5992	0.00714
			de novo' pyrimidine base biosynthesis	6207	0.00714
			nucleotide biosynthesis	9165	0.00714

Table B.3: Continued.

			Term	GO Accession number	p-value
Within-cycle variation in transcript abundance	direct comparisons	cASA	protein biosynthesis	6412	0
			regulation of transcription. DNA-dependent	6355	3.55E-10
			development	7275	4.34E-08
			protein modification	6464	4.44E-06
			glycolysis	6096	5.31E-06
			signal transduction	7165	3.11E-05
			phototransduction	7602	3.94E-05
			protein-mitochondrial targeting	6626	4.90E-05
			ribosome biogenesis and assembly	42254	4.90E-05
			mitochondrial inner membrane protein import	45039	4.90E-05
			GTP biosynthesis	6183	5.53E-05
			UTP biosynthesis	6228	5.53E-05
			CTP biosynthesis	6241	5.53E-05
			translational initiation	6413	0.00017
			ubiquitin cycle	6512	0.00029
			copper ion transport	6825	0.00039
			mRNA processing	6397	0.00046
			monovalent inorganic cation transport	15672	0.00064
			cell organization and biogenesis	16043	0.00064
			ATP biosynthesis	6754	0.00091
			glycerol-3-phosphate metabolism	6072	0.00147
			ATP synthesis coupled proton transport	15986	0.00149
			mitochondrial electron transport. ubiquinol to cytochrome c	6122	0.00195
			cell surface receptor linked signal transduction	7166	0.00195
			protein targeting	6605	0.00282
			frizzled-2 signaling pathway	7223	0.00324
			small GTPase mediated signal transduction	7264	0.00355
			nuclear mRNA splicing. via spliceosome	398	0.00393
			lipid transport	6869	0.00546
			cell adhesion	7155	0.00618
			cell cycle arrest	7050	0.00622
			homophilic cell adhesion	7156	0.00901
		cBLA	protein amino acid phosphorylation	6468	0
			phototransduction	7602	5.80E-07
			protein-nucleus import. docking	59	1.59E-06
			DNA replication initiation	6270	2.70E-06
			ubiquitin-dependent protein catabolism	6511	6.19E-06
			protein amino acid dephosphorylation	6470	2.81E-05
			mismatch repair	6298	0.00014
			cellular protein metabolism	44267	0.00014
			protein folding	6457	0.00019

Table B.3: Continued.

			Term	GO Accession number	p-value
Within-cycle variation in transcript abundance	direct comparisons	cBLA	regulation of pH	6885	0.00035
			oxygen transport	15671	0.00082
			steroid metabolism	8202	0.00104
			visual perception	7601	0.00108
			chromatin modification	16568	0.00131
			DNA metabolism	6259	0.00223
			glycerol-3-phosphate metabolism	6072	0.00286
			transport	6810	0.00382
			metabolism	8152	0.00482
			signal transduction	7165	0.00690
			fatty acid beta-oxidation	6635	0.00749
			ATP-dependent proteolysis	6510	0.00830
			microtubule-based movement	7018	0.00963
		cBSA	proteolysis and peptidolysis	6508	1.33E-10
			carbohydrate metabolism	5975	1.22E-08
			protein amino acid phosphorylation	6468	1.46E-07
			phototransduction	7602	2.92E-06
			lipid transport	6869	2.32E-05
			peptide cross-linking	18149	2.93E-05
			microtubule-based movement	7018	0.00012
			protein amino acid dephosphorylation	6470	0.00022
			cellular protein metabolism	44267	0.00064
			cell-matrix adhesion	7160	0.00207
			regulation of translational initiation	6446	0.00264
			nucleotide-sugar metabolism	9225	0.00304
			regulation of pH	6885	0.00460
			nucleocytoplasmic transport	6913	0.00554
			N-linked glycosylation via asparagine	18279	0.00554
			visual perception	7601	0.00658
			nucleotide metabolism	9117	0.00775
			nucleotide-sugar transport	15780	0.00775
	Effect of clone type	LA	protein amino acid phosphorylation	6468	3.47E-11
			protein amino acid dephosphorylation	6470	2.51E-08
			protein folding	6457	1.17E-07
			regulation of GTPase activity	43087	0.00030
			protein-nucleus import. docking	59	0.00034
			regulation of pH	6885	0.00041
			glycerol-3-phosphate metabolism	6072	0.00090
			regulation of translational initiation	6446	0.00146
			metabolism	8152	0.00183
			protein complex assembly	6461	0.00205

Table B.3: Continued.

			Term	GO Accession number	p-value
Within-cycle variation in transcript abundance	Effect of clone type	LA	cation transport	6812	0.00281
			intracellular protein transport	6886	0.00338
			neuropeptide signaling pathway	7218	0.00492
			protein modification	6464	0.00544
			small GTPase mediated signal transduction	7264	0.00795
			development	7275	0.00817
			ubiquitin-dependent protein catabolism	6511	0.00838
			protein-nucleus import	6606	0.00913
			isoprenoid biosynthesis	8299	0.00913
		SA	heme biosynthesis	6783	1.52E-05
			cellular protein metabolism	44267	4.44E-05
			de novo' pyrimidine base biosynthesis	6207	7.83E-05
			oxygen transport	15671	0.00018
			protein folding	6457	0.00024
			sensory perception	7600	0.00062
			protein complex assembly	6461	0.00090
			glycolysis	6096	0.00090
			glycerol-3-phosphate metabolism	6072	0.00127
			nucleotide-sugar transport	15780	0.00127
			carbohydrate metabolism	5975	0.00131
			protein polymerization	51258	0.00150
			GMP biosynthesis	6177	0.00220
			pyrimidine nucleotide biosynthesis	6221	0.00220
			phototransduction	7602	0.00221
			cell wall catabolism	16998	0.00288
			nucleotide metabolism	9117	0.00431
			metabolism	8152	0.00467
			rRNA processing	6364	0.00827
			purine ribonucleoside salvage	6166	0.00918
			tRNA aminoacylation for protein translation	6418	0.00994
	Effect of cycle type	cA	protein-nucleus import. docking	59	1.89E-07
			cellular protein metabolism	44267	4.35E-06
			phototransduction	7602	8.05E-06
			peptide cross-linking	18149	9.75E-06
			sensory perception	7600	2.95E-05
			protein amino acid phosphorylation	6468	7.01E-05
			carbohydrate metabolism	5975	0.00014
			de novo' pyrimidine base biosynthesis	6207	0.00085
			DNA replication initiation	6270	0.00088
			protein-nucleus import	6606	0.00098
			metabolism	8152	0.00124
			phosphatidylserine biosynthesis	6659	0.00148

Table B.3: Continued.

			Term	GO Accession number	p-value
Within-cycle variation in transcript abundance	Effect of cycle type	cA	protein polymerization	51258	0.00161
			translational initiation	6413	0.00212
			D-ribose metabolism	6014	0.00266
			fatty acid metabolism	6631	0.00266
			peptidoglycan catabolism	9253	0.00266
			glycolysis	6096	0.00288
			oxygen transport	15671	0.00288
			protein folding	6457	0.00312
			chromatin modification	16568	0.00327
			antibiotic biosynthesis	17000	0.00343
			visual perception	7601	0.00540
			regulation of translational initiation	6446	0.00587
			cell-matrix adhesion	7160	0.00806
			L-phenylalanine catabolism	6559	0.00849
		cB	protein amino acid dephosphorylation	6470	6.91E-10
			oxygen transport	15671	6.09E-09
			protein amino acid phosphorylation	6468	1.07E-08
			carbohydrate metabolism	5975	6.20E-08
			protein-nucleus import. docking	59	1.27E-07
			cellular protein metabolism	44267	1.71E-05
			protein folding	6457	1.89E-05
			microtubule-based movement	7018	2.23E-05
			glycerol-3-phosphate metabolism	6072	2.98E-05
			mismatch repair	6298	4.96E-05
			signal transduction	7165	5.01E-05
			DNA metabolism	6259	0.00018
			regulation of pH	6885	0.00038
			threonyl-tRNA aminoacylation	6435	0.00069
			regulation of translational initiation	6446	0.00164
			DNA replication initiation	6270	0.00183
			ubiquitin-dependent protein catabolism	6511	0.00197
			regulation of GTPase activity	43087	0.00221
			alanyl-tRNA aminoacylation	6419	0.00408
			chromatin modification	16568	0.00408
			phosphatidylserine biosynthesis	6659	0.00447
			glycine catabolism	6546	0.00685
			N-linked glycosylation	6487	0.00895
			trehalose metabolism	5991	0.00920
			cell surface receptor linked signal transduction	7166	0.00920