

**Box 3 Continued**

BMR as a yardstick to evaluate or interspecifically scale maximal metabolic performances, seems nevertheless quite valid as long as the appropriate, context-specific, values for BMR are used (McKechnie *et al.* 2006). However, even though BMR may (1) be repeatable within individuals (Hayes *et al.* 1998, Bech *et al.* 1999, Horak *et al.* 2002, Labocha *et al.* 2004, Rønning *et al.* 2005, Vézina and Williams 2005), (2) have a heritable component (Konarzewski and

Diamond 1995, Książek *et al.* 2004, 2009) and (3) be selected for in the wild (Broggi *et al.* 2005), BMR may be too much of an epiphenomenon to be a good trait for natural selection to work on (Fig. 45C). We suggest that natural selection first and foremost targets the size and capacity of different organs and organ groups, and that BMR will follow as a result (cf. Jackson and Diamond 1996, Williams and Tieleman 2000, Sadowska *et al.* 2008).

**CHAPTER 5**

## Phenotypic plasticity: matching phenotypes to environmental demands

### Adaptive arm-waving, and more...

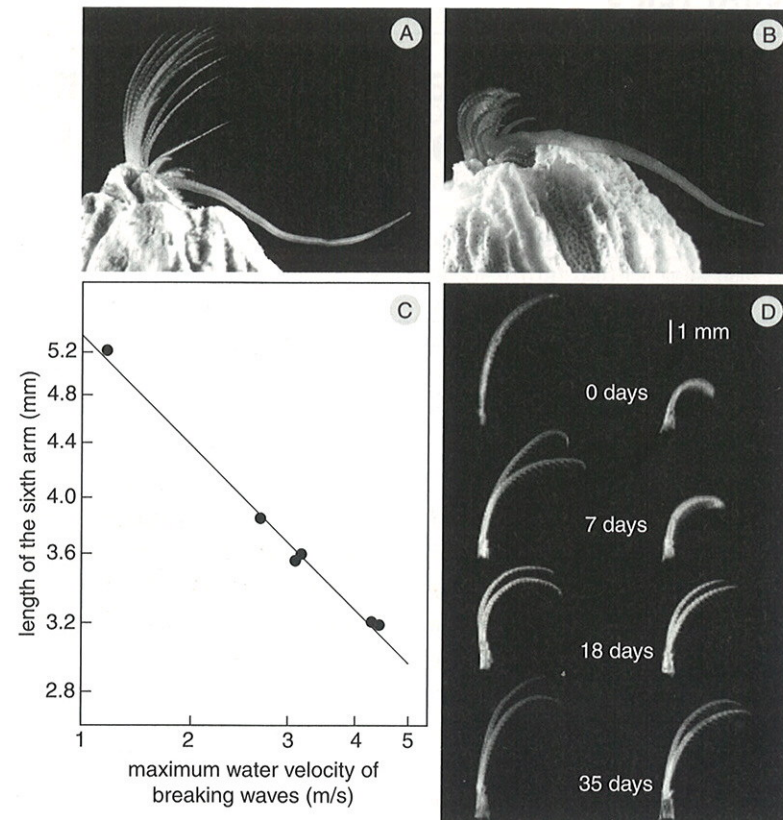
Barnacles are peculiar crustaceans. Protected in cone-shaped shells, firmly attached to rock and other hard substrates, they weather the pounding of waves, the attacks by predators, the occasional desiccation that comes with living on the high shore and the high temperatures on sunny days. They feed on small planktonic items, which they fish from the surrounding water by a 'net' made up of what their arthropod ancestry has provided them with: six pairs of strongly modified arms. These arms are called cirri, and the fishing net a cirral net (Fig. 46A, B).

It is easy to imagine that, when the water is calm, even wide and fragile nets will work, whereas under strong water flows such nets would be ripped to the side and become inefficient at best, and damaged at worst. Several species of intertidal barnacles indeed carry long arms on sheltered shores, and short arms on exposed shores (Fig. 46C; Arsenault *et al.* 2001, Marchinko and Palmer 2003). At similar densities of edible plankton, high water flows will compensate for the smallness of the cirral net. Under very exposed conditions, however, the adaptive miniaturization comes to a halt (Li and Denny 2004), as barnacles increasingly begin to limit cirral netting to times of relatively slow water flow (Marchinko 2007, Miller 2007). Barnacles are quite easily dislodged from where they live and can be artificially glued on to other pieces of rock or shell. This

enabled Kerry Marchinko (2003) to show that, when displaced to other flow regimes within Barkley Sound on the Pacific coast of Vancouver Island, they will quickly adjust the length and strength of their arms (Fig. 46D), even when mature. Thus, cirral nets appear fully reversibly plastic throughout life.

Their crustacean affiliation also shows when it comes to sex and reproduction. Unlike many marine invertebrates that engage in so-called 'free spawning'—jettisoning sperm and eggs in the water, where they find each other by chance (Luttikhuisen *et al.* 2004)—barnacles get intimate. Indeed, as documented by Charles Darwin (1854), at lengths up to eight times body length, barnacles possess the longest penises in the animal world (Neufeld and Palmer 2008). This goes together with the fact that most are hermaphrodites—combining maleness with femaleness—but apparently they do not do it on the cheap and self-fertilize. Glued to the substrate, barnacles cannot cuddle up; they have to use a long penis to find, reach, and then impregnate fastened but fertile neighbours (Klepal 1990, Murata *et al.* 2001). Now, what was true for the feeding arms forming the cirral net, would also be true for a penis extending out of the father's cone: at high water-flow rates, a long penis would be hard to manoeuvre properly. Building on the previously mentioned barnacle work at Barkley Sound, Chris Neufeld and Richard Palmer (2008) indeed found that penises of *Balanus*





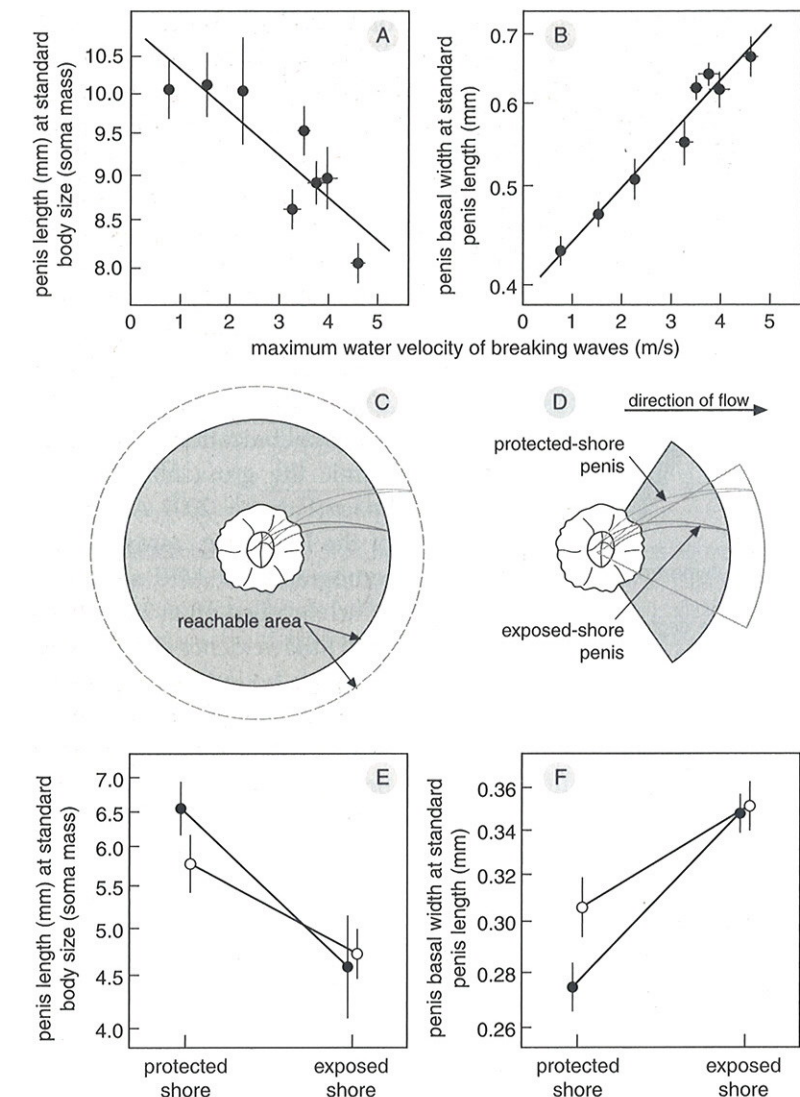
**Figure 46.** Barnacles *Balanus glandula* extend their arms to form cirral nets with which to withdraw small planktonic food items from the overlying water. Their cirral nets are cast wide in the calm waters of protected shores (A), but the arms are short in the waves of exposed shores (B), these examples representing opposing ends of a gradient (C). On the photos, in addition to folding out their cirral nets, the barnacles have also extended their considerable penises (see Fig. 47). Experimental displacement to protected shores from another protected (left) or a wave-beaten environment (right), shows the arms (in this case the sixth) to adjust adaptively within a couple of weeks.

Compiled from Neufeld and Palmer (2008) (A, B), Arsenault *et al.* (2001) (C), and Marchinko (2003) (D).

*glandula* became shorter and thicker at the base on the more exposed shores (Fig. 47A, B). For reasons just outlined, the variations in penis dimension made sense (Fig. 47C, D). Perhaps not surprisingly, penis length and thickness were also perfectly reversibly plastic (Fig. 47E, F). Not only do penis size and shape reflect the flow regime of the waters surrounding its owner, in the Eastern Atlantic, lonely barnacles have larger penises relative to crowded ones (Hoch 2008).

A big penis will come at a cost though, as it needs to be grown and maintained, and it may

interfere with foraging (Klepal *et al.* 1972, Hoch 2008). While barnacles do not shed sperm and eggs, rather they shed free-living larvae. At temperate latitudes, where larval food supply may vary during the year (Yoder *et al.* 1993, Dasgupta *et al.* 2009), the reproduction of barnacles, it turns out, is also strongly seasonal. Long before the reversibility of penis size was examined, Margaret Barnes (1992) had concluded that some barnacles get rid of their penises altogether outside the mating season. What is left during the 'off season' is a rather cost-free 'stump'.



**Figure 47.** With increasing wave action, the penis of the barnacle *Balanus glandula* becomes shorter (A) and thicker at the base (B). This makes sense because the area within reach of the penis, and therefore the number of potential mates, is a function of its length and strength; in calm water, more area would be reached by a long and slender penis (C) and in strongly flowing water, by a short and strong penis (D). Experimental displacements from either a protected shore (filled dots) or an exposed shore (open circles) shows penis length (E) and basal width (F) have mutually adjusted after 20 weeks. Means with SE.

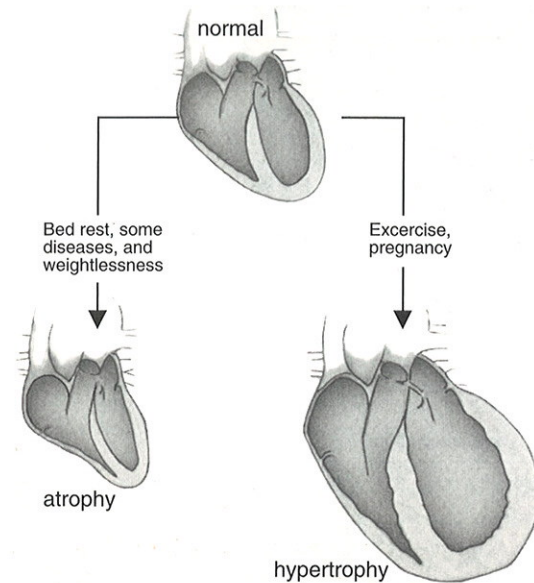
Compiled from Neufeld and Palmer (2008).

### Use it or lose it

Just as reproductively inactive barnacles do away with their penises, astronauts (who speak English) and cosmonauts (who speak Russian) quite unwillingly lose body parts

during their time in space (Lackner and DiZio 2000, Payne *et al.* 2007). Remember the sight of weakened astronauts returning from Apollo missions on the TV screen? Without gravity a human environment is less demanding in





**Figure 48.** Visualization of the degree of atrophy of human hearts after several weeks of space travel or bed rest, as well as the degree of hypertrophy that comes with exercise and pregnancy.

Modified after Hill and Olson (2008).

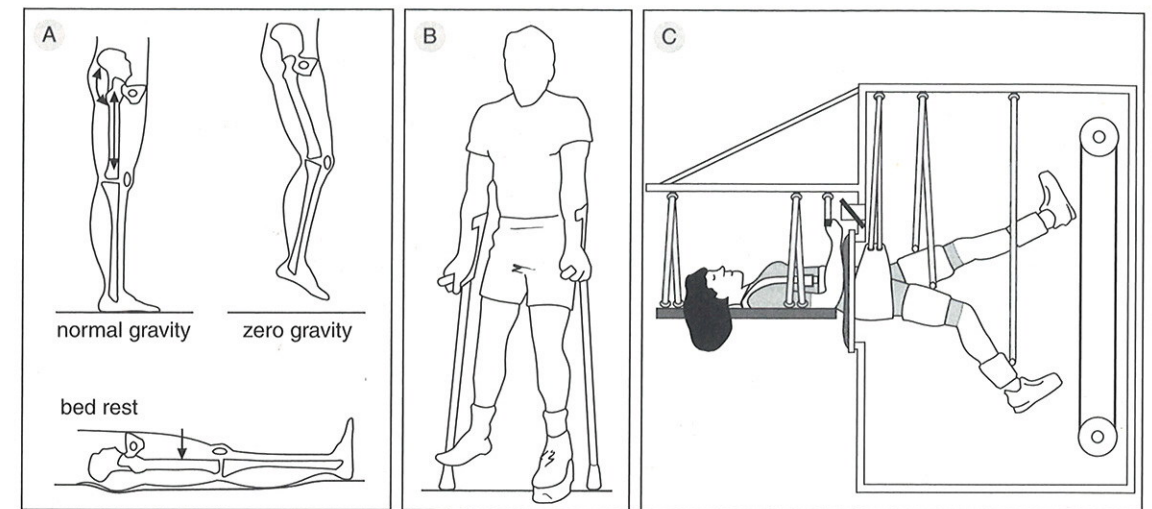
several different ways and this shows upon the return to Earth.

One of the first things to be affected is the heart (Fig. 48), which may shrink by as much as a quarter after one week in orbit (Hill and Olson 2008). Heart atrophy is correlated with decreases in blood and stroke volume, blood pressure, and reduced exercise capacity (Convertino 1997, Payne *et al.* 2007). Especially after several months in the International Space Station, and particularly in women, upon return to Earth, astro- and cosmonauts experience dizziness and blacking-out because blood does not reach the brain in sufficient quantities (Meck *et al.* 2001, Waters *et al.* 2002).

Six weeks in bed leads to similar atrophy of the heart as one week in space, suggesting that either weightlessness itself, or the concomitant reduction in exercise, causes heart atrophy (Perhonen *et al.* 2001). For the muscles and bones

of the limbs, the effects of weightlessness are not only easier to predict on the basis of first principles (Fig. 49A), but the predictions are also easier to verify experimentally (Fig. 49B, C). Importantly, some muscle groups are wasted more than others (Vico *et al.* 1998, Payne *et al.* 2007). Because they bear the body's weight, the 'anti-gravity' muscles of thigh and legs would be expected to show atrophy when they are pushed out of work during space flight or during experimental weightlessness on Earth (Fig. 49); and they do (Payne *et al.* 2007). Various types of immobilization experiments successfully mimic the gross effects of weightlessness in space (Fitts *et al.* 2001, Adams *et al.* 2003). Muscles of the lower leg, assayed either by computed tomography or MRI scanning, showed losses, which levelled off at 20% after 40 days (Fig. 50), with little evidence of differences between earth- or space-bound microgravity effects. Despite the best attempts at giving them replacement exercise, after six months aboard the International Space Station, crew members still had lost 13% of their calf muscle volume and 32% of the peak power of their legs (Trappe *et al.* 2009). Not only is muscle volume reduced during disuse, various metabolic changes occur that include a decreased capacity for fat oxidation. This can lead to the build-up of fat in atrophied muscle (Stein and Wade 2005).

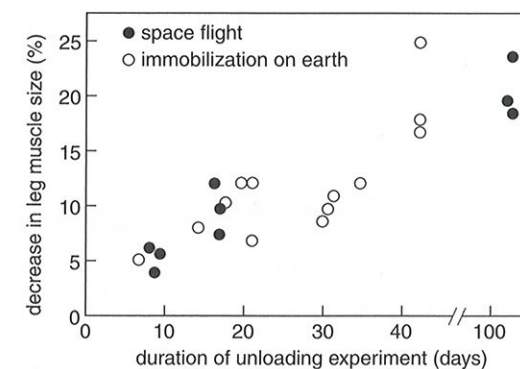
In addition to muscle loss, and apart from a deterioration of proper immune functioning during space travel (Sonnenfeld 2005, Crucian *et al.* 2008), arguably the most fearsome effect on bodies is bone loss. Although their hardness and strength, and the relative ease with which they fossilize, give bones a reputation of permanence, bone is actually a living and remarkably flexible tissue. In the late-nineteenth century, the German anatomist Julius Wolff (1892) formulated that bones of healthy persons will adjust to the loads that they are placed under (Fig. 51A). A decrease in load will lead to the loss of bone material at appropriate places, and an increase to thicker



**Figure 49.** How a change of gravitational forces (size of arrows indicates relative forces) affects the muscles and bones of the leg (A), and some of the contraptions used in human experiments on the effects of use and disuse on muscle and bone size, strength, and functioning (B, C). Diagram (B) shows how loads are taken off the lower limb without constraining the knee or ankle, whereas (C) shows how bed-rested women received compensatory treadmill running in a low-pressure chamber to mimic exercise for astronauts.

Compiled from Lackner and DiZio (2000), Tesch *et al.* (2004), and Dorfman *et al.* (2007).

bone (Fig. 51B). It is no surprise then that, in the gravity-free environments in space, bones will de-mineralize, especially the lower limbs that normally counteract the effects of gravity (Fig.

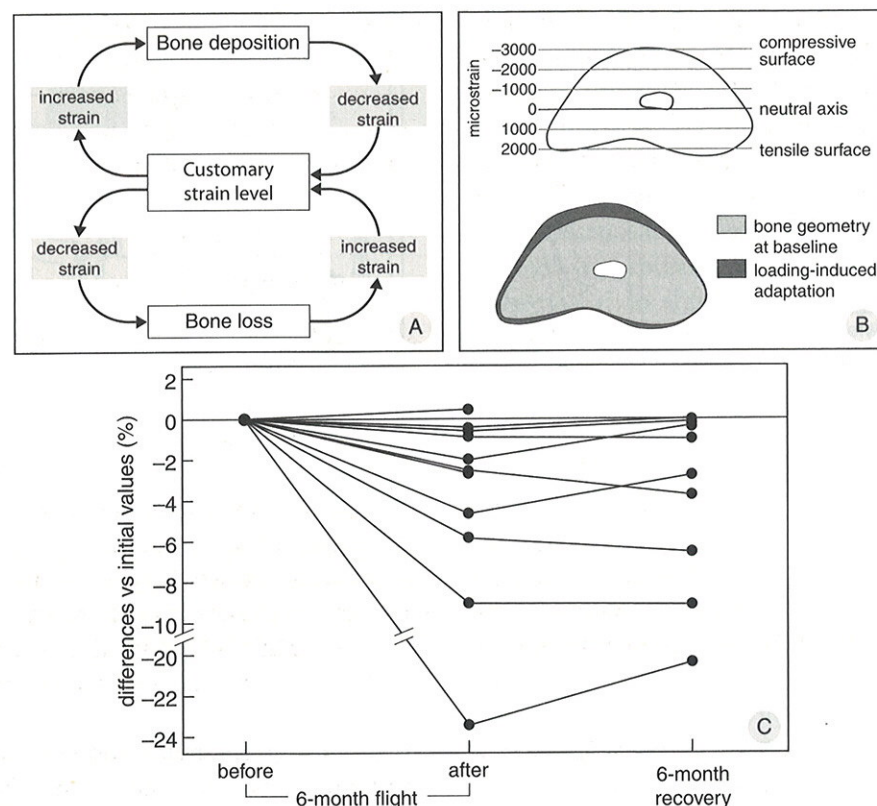


**Figure 50.** Relative loss of the lower leg muscles (triceps surae, gastrocnemius, soleus, or some combination thereof) as a function of the duration of unloading either during space flight or in immobilization experiments on Earth (this includes limb immobilization by casting, unilateral lower limb suspension as illustrated in Fig. 49 B, and bed rest).

Modified from Adams *et al.* (2003).

51C). With considerable individual variation, during half a year in space, cosmonauts lost up to a quarter of the tibial bone material (Vico *et al.* 2000). Although bone-formation continues under zero gravity conditions—this was established in chicken embryos sent to space (Holick 1998)—relative to bone-resorption, bone-formation rates decline (Holick 2000). What has been of greatest concern is that, unlike muscle loss that levels off with time (Fig. 50), bone loss seems to continue steadily with 1–2% a month of weightlessness. During a 2–3 year mission to Mars, space travellers could lose up to 50% of their bone material, which would make it impossible to return to Earth's gravitational forces. Thanks to Wolff's Law, space agencies will have to become very creative in addressing the issue of bone loss during flights to Mars (Lackner and DiZio 2000, Whedon and Rambaut 2006). So far, boneless creatures, such as jellyfish, are much more likely to be able to return safely to Earth after multi-year space trips than people. Gravity is a Mars bar.





**Figure 51.** The use-disuse principle formulated as a functional adaptation model for bones (A), an illustration of the principle for an ulna of a rat (B), and a test of disuse-use effects in human cosmonauts (C). In the latter case, the loss of bone mineral density during a 6-month space flight, and the degree of recovery during the subsequent half year on Earth, were established.

Compiled from Ruff *et al.* (2006) (A), Robling *et al.* (2002) (B), and Vico *et al.* (2000) (C).

Bone loss during space travel certainly brings home the point that 'if you don't use it, you lose it', but what about 'if you use it again, do you regain it'? Within six months of their return to Earth, cosmonauts did indeed show partial recovery of their tibial bone mass (Fig. 51C). Obviously most of them needed more time, but even after one year of recovery, bone losses in men who had been experimentally exposed to three months of permanent bed rest were not fully compensated, though their calf muscles had fully recovered much earlier (Rittweger and Felsenberg 2009).

### The dynamic gut

It is anybody's guess what happens to the innards of space travellers, but on the basis of what we see in other vertebrates (Starck 2003, Naya *et al.* 2007), in the course of journeys beyond the atmosphere, their digestive tracts may well show change. To get a feel for what is possible, let's look at snakes, animals that go through veritable 'gastrointestinal rebirths' (Pennisi 2005). Pythons, boas, rattlesnakes, and vipers are snakes that employ sit-and-wait foraging strategies (Greene 1997). This makes them go without meals for so long that their

stomachs, intestines, and accessory organs shrink and become 'dormant' (Secor *et al.* 1994, Secor and Diamond 1995, Starck and Beese 2001, Ott and Secor 2007). When these snakes do catch the occasional prey, it can be almost as big as themselves. Prey capture, killing by constriction, and swallowing through the extended gape (Fig. 52A), elicits a burst of physiological activity, with drastic upregulation of many metabolic processes (reviewed in Secor 2008). Immediately, the heart starts to grow (Fig. 52C). With a doubling of the rate of heartbeat, and as blood is shunted away from the muscles to the gut (Starck and Winner 2005, Starck 2009), blood flow to the gut increases by an order of magnitude. Within two days, the wet mass of the intestine more than doubles (Fig. 52D): this change is additional to growth, involves the rehydration of previously inactive cells, and the incorporation of lipid droplets (J.M. Starck pers. comm.). This is followed by 100% increases of liver and pancreas mass, and a 70% increase of kidney mass (Fig. 52C). At the same time, stomach acidity increases (a drop in pH from values of 7–8 to 1–2), ensuring that the skeleton of the ingested prey disappears within about six days (Fig. 52B). Many amazing changes, at several different levels of physiological action, take place in the intestine, including the instantaneous lengthening of microvilli (Fig. 52E).

All in all, the stomach operates for less than a week, but the intestines carry on for another week to finish the job. Interestingly, part of the winding down of the gut includes the generation and banking of new cells lining the intestine, thus preparing the pythons for the next meal (Starck and Beese 2001). And what digestively goes for pythons also goes for another reptile, the caiman *Caiman latirostris* from South America (Starck *et al.* 2007). The precise mechanism of the restructuring of the gut and associated organs may differ between mammals, birds, and the sauropsid reptiles to which

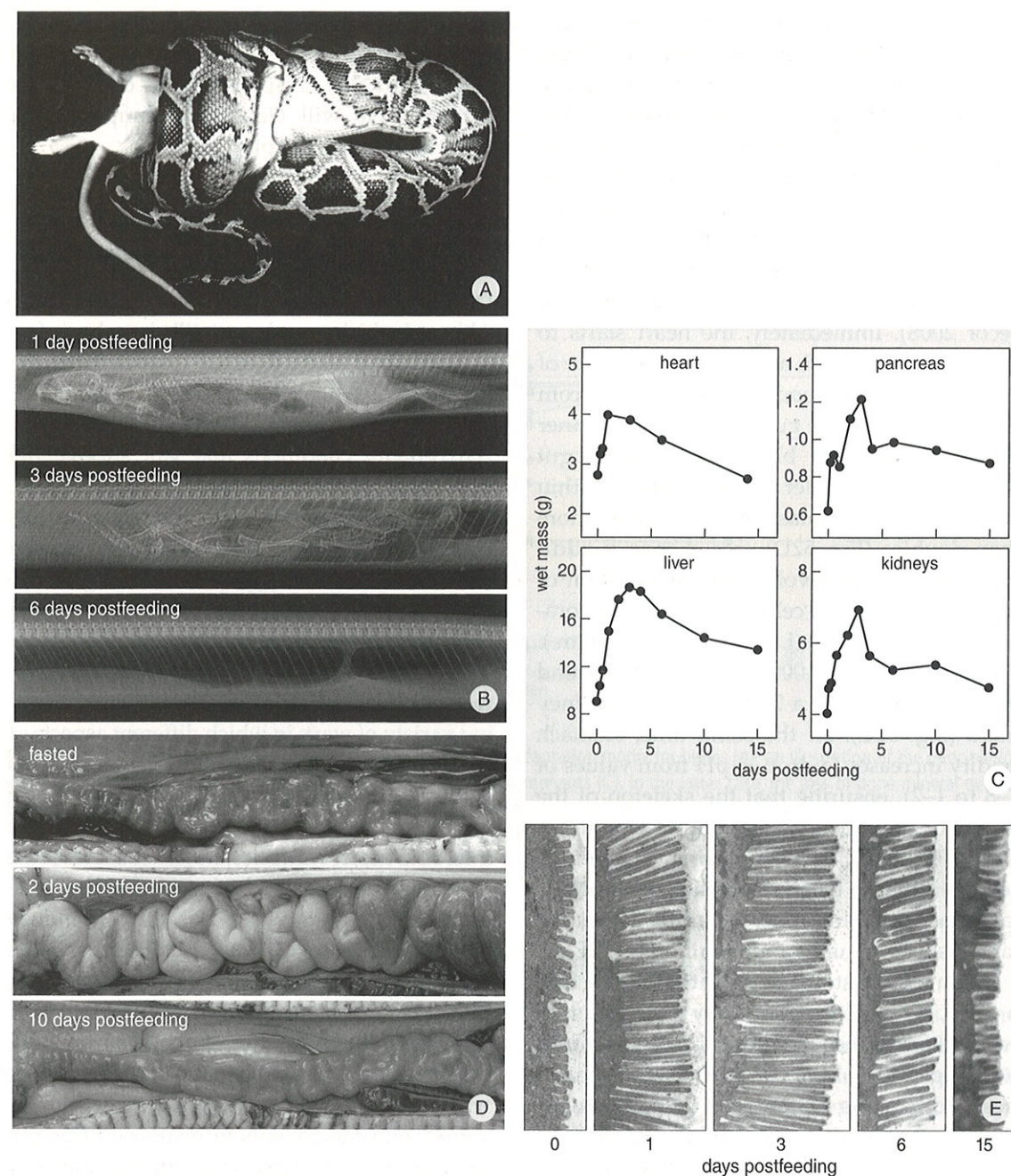
snakes and crocodiles belong (Starck 2003), but in all these groups, organ size and capacity are remarkably responsive to changes in demand. We will elaborate examples from birds later in this chapter.

### 'Classical' phenotypic plasticity: developmental reaction norms

Let's just briefly think about space travel again. Although chicken embryos still show bone formation when taken to space (Holick 1998), on the basis of what we now know it would be impossible for a human child to grow up under zero gravity conditions and still survive the functional demands of a return to Earth. 'Developmental plasticity' is the name for this category of phenotypic plasticity, the one in which environmental conditions during ontogeny determine the size, shape, construction, and behaviour of the eventual mature phenotype. There are innumerable examples of developmental phenotypic plasticity, and this exhilarating variety of ways in which different aspects of environments shape different organismal traits is illustrated in Fig. 53. Picking up the barnacle theme from earlier in this chapter, some of the intertidal acorn barnacles *Chthamalus anisopoma* develop a bent shell when grown in the presence of predatory snails; this form is indeed more resistant to predation than the normal morph (Lively 1986, 1999, Lively *et al.* 2000, Jarrett 2008).

A classic case of developmental phenotypic plasticity is the variation in the degree to which water fleas *Daphnia* show hoods, helmets, spines, and longer tails in response to predators, or to the chemical identifiers (the 'kairomones') that they leave in the water (e.g. Tollrian and Dodson 1999, Caramujo and Boavida 2000, Petrušek *et al.* 2009, Riessen and Trevett-Smith 2009). This variation was first described by the enlightened German zoologist Richard Woltereck (1909) in clones of water





**Figure 52.** After a Burmese python *Python molurus* has captured a prey item, killed it by asphyxiation, and started swallowing it (A), much of the snake's internal physiology gears into action. (B) Shows how the bones of a rat are digested in the acidic stomach within 6 days post-feeding. In the first few days after ingestion, the internal organs show growth spurts and then a slow return to the old levels. Within 2–3 days, the intestine has grown to full capacity (D), including a drastic lengthening of the microvilli (E, the bars in this image represent 1  $\mu$ m).

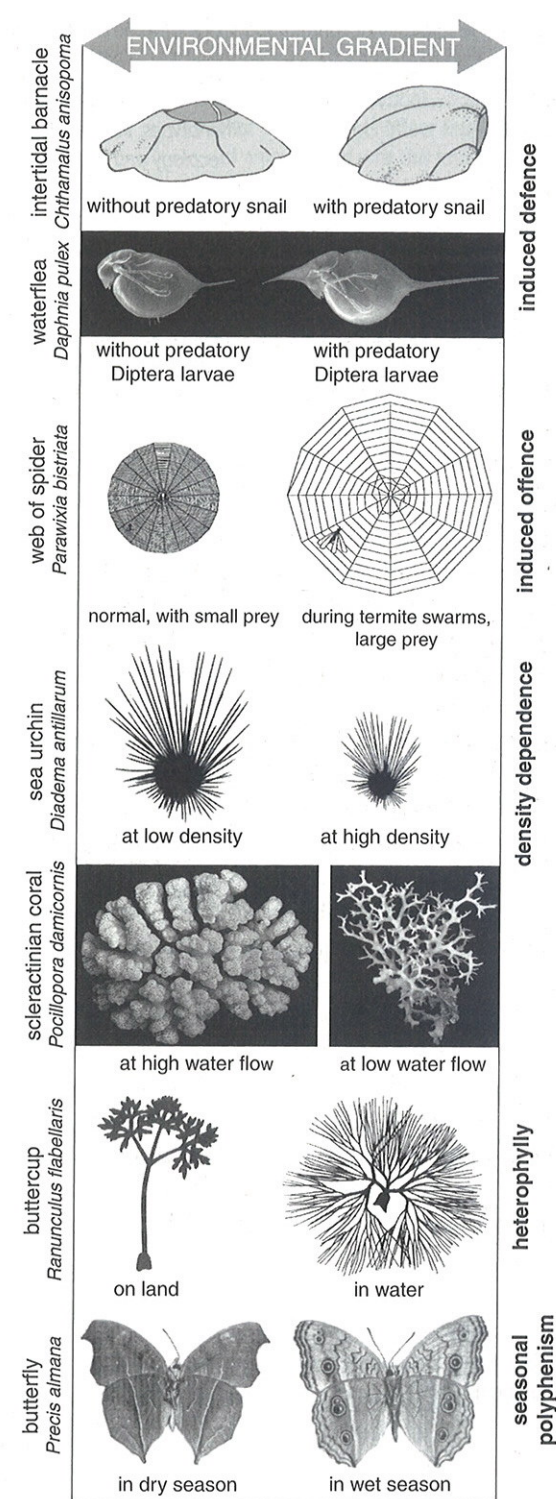
Compiled from Lignot *et al.* (2005) and Secor (2008).

fleas (see Harwood 1996, Sarkar 2004). Finding that different pure strains of *Daphnia* show similar variation in hood size, variation that converged under particular environmental conditions, Woltereck began to doubt the distinction between genotype and phenotype, a contrast that was just becoming part of the mental armoury of biologists at the time. Woltereck was straightened out by the Danish plant physiologist and budding geneticist Wilhelm Johannsen (1911). Johannsen suggested, first in two books in Danish and then in an address to the American Society of Naturalists in 1910 (with which he reached the world), that what genotypes do is not to instruct for a single phenotype, but to instruct for the ways that phenotypes respond to environmental variation (Roll-Hansen 1979). This relationship between trait values and specific environmental conditions has since been called what Woltereck named it: 'Reaktionsnorm' or 'reaction norm' or 'norm of reaction' (Pigliucci 2001a, 2001b). The norm of reaction of barnacles to predatory snails, and of water fleas to predatory insects or fish, is now also known as an example of the category of 'induced defences' (Tollrian and Harvell 1999).

Figure 53 shows five more reaction norms reflecting several other types of developmental plasticity. Spiders weaving a coarser grained and bigger web when they can catch large prey (in this case flighted termites), could be said to show 'induced offence' (cf. Padilla 2001, Kopp and Tollrian 2003, Miner *et al.* 2005, Kishida *et al.* 2009). Caribbean sea urchins *Diadema antillarum* have a hard calcite

**Figure 53.** Examples of phenotypic plasticity in invertebrate animals and in plants, along with some of the common denominators of the particular class of plasticity.

Compiled from (from top to bottom): Lively (1999), Lampert *et al.* (1994), Sandoval (1994), Levitan (1989), Kaandorp (1999), Schlichting and Pigliucci (1998, p. 56), and Nijhout (1999).





**Table 6.** A summary of content and scope of some recent books dedicated to the phenomena of phenotypic plasticity.

Reference	Book title	No. of pages	Content and scope
Rollo (1995)	<i>Phenotypes, their epigenetics, ecology and evolution</i>	463	An early, but very wide-ranging, synthesis by a molecular biologist of the genetic background and possible hierarchical organization of phenotypic construction, or organismal design, with due recognition of the role of ecological factors.
Schlichting and Pigliucci (1998)	<i>Phenotypic variation: a reaction norm perspective</i>	387	The textbook on phenotypic evolution from combined genetic and developmental perspectives. Well-illustrated. Norms of reaction are the core idea to explore the basics and evolutionary consequences of phenotypic responses to environmental variation.
Tollrian and Harvell (eds) (1999)	<i>The ecology and evolution of inducible defenses</i>	383	Explores and reviews a wide range of cases of developmental plasticity that fall under the banner of 'induced defence mechanisms': from the use of armour, via unpalatability, to immunity and behaviour as environmentally induced mechanisms for bodily defence.
Pigliucci (2001a)	<i>Phenotypic plasticity: beyond nature and nurture</i>	328	A solid attempt to develop mainly genetic interpretations of phenotypic plasticity as the core concepts in evolutionary biology.
West-Eberhard (2003)	<i>Developmental plasticity and evolution</i>	794	Almost encyclopaedic review of the importance of developmental processes for evolutionary innovation. Develops the notion that in evolution phenotypes change first, and that genes follow.
DeWitt and Scheiner (eds) (2004)	<i>Phenotypic plasticity: functional and conceptual approaches.</i>	247	With a loose focus on developmental processes and their genetic interpretation (reaction norms), the chapters explore a wide range of issues to do with phenotypic plasticity.
Turner (2007)	<i>The tinkerer's accomplice: how design emerges from life itself</i>	282	Formulates the role of self-organization, in interaction with replicators on one side and the environment on the other, in phenotypic expressions. It develops the idea that organisms show design, not because their genes instruct them to, but because agents of homeostasis build them that way.
Blumberg (2009)	<i>Freaks of nature: what anomalies tell us about development and evolution</i>	326	This is mostly a book about development, but one in which the importance of interactions between developing bodies and their environments is strongly emphasized.

Gilbert and Epel (2009)	<i>Ecological developmental biology: integrating epigenetics, medicine, and evolution</i>	480	Fantastically comprehensive and well-illustrated textbook on the ways that environments affect development of animals, and why this knowledge is important in medicine and evolutionary biology.
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skeleton but are able to shift body size in response to variations in local food conditions. Levitan (1989) experimentally altered competitor densities, and showed that body size increased and decreased to levels predicted from field-based food abundance–body size relationships. By adjusting structural body size, the urchins reduced maintenance costs and thereby optimized reproduction and survival according to local food availability (Levitan 1989). Although body size is altered, the mouth structures (Aristotle’s lantern) of the urchins, and thus their capacity to eat, remained unchanged (Levitan 1991).

Depending on the type of water flow, scleractinian corals build either denser or more spaced out structures (Kaandorp 1999, Todd 2008, who still owe us a term for this category of phenotypic plasticity). Buttercups *Ranunculus* have ‘proper’ wide leaves when growing on land and almost filamentous leaves when growing in water, representing a category of plant plasticity called heterophylly (Cook and Johnson 1968). The last example in Fig. 53 is quite typical of the insect world. Depending on season, a tropical butterfly *Precis almana* may either show angular wing shapes and a dull brown colour, so that it resembles a dead leaf (in the dry season), or show rounded and colourful wings with eye-spots (in the wet season) (Nijhout 1999). This category of phenotypic plasticity is known as (seasonal) polyphenism. A final category of

phenotypic plasticity is the size-, age-, condition-, and/or context-dependent sex change that occurs in some plants, and in animals as diverse as annelids, echinoderms, crustaceans, molluscs, and, most famously, in fish (Polikansky 1982, Munday *et al.* 2005, 2006).

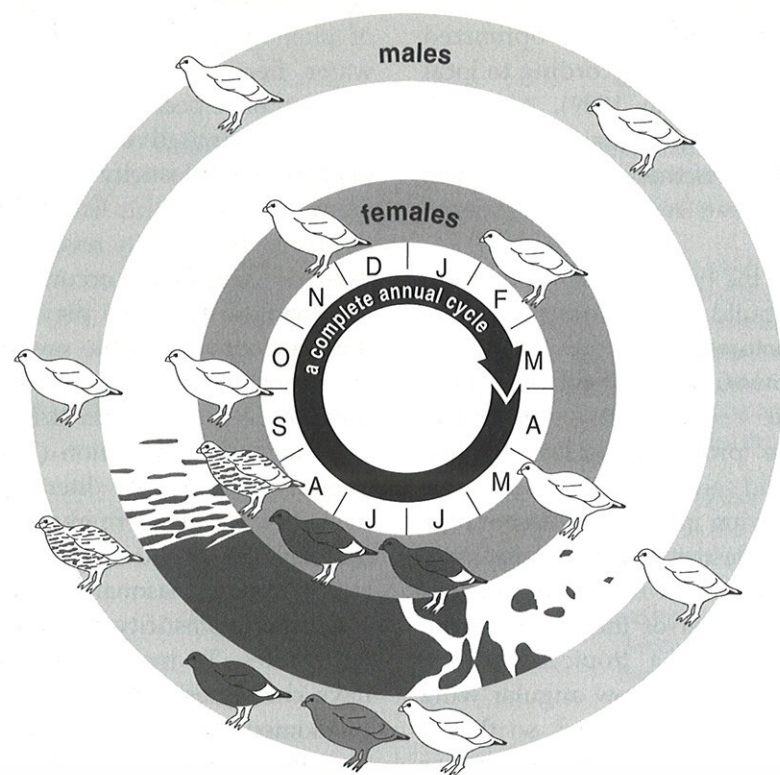
All the ‘textbook’ examples amalgamated in Fig. 53 represent relatively well-studied cases of phenotypic plasticity. Some, such as the water fleas of Woltereck and subsequent authors, have been important in the development of quantitative genetic-related theories of phenotypic plasticity (Pigliucci 2001a). What are missing from this list of examples are the extensive, but usually reversible, internal and external changes that occur in vertebrate bodies, changes that have played major roles in the previous and in the present chapter, but are almost absent from the burgeoning literature on phenotypic plasticity (see Table 6). With a notable exception (Turner 2007), the phenotypic plasticity literature has mostly passed by the staggeringly large body of phenomena related to internal (physiological), external, and seasonally cyclic (moult) and behavioural plasticity, especially in the higher vertebrates. To make sure that readers will have encountered the full range of plasticity phenomena by the end of this chapter, let’s now look at the reaction norms of male and female rock ptarmigan *Lagopus mutus*, showing phenotypes that vary with time of year, snow cover, and, in the case of males, the availability of mates.



### Seasonal phenotype changes in ptarmigan, deer, and butterflies

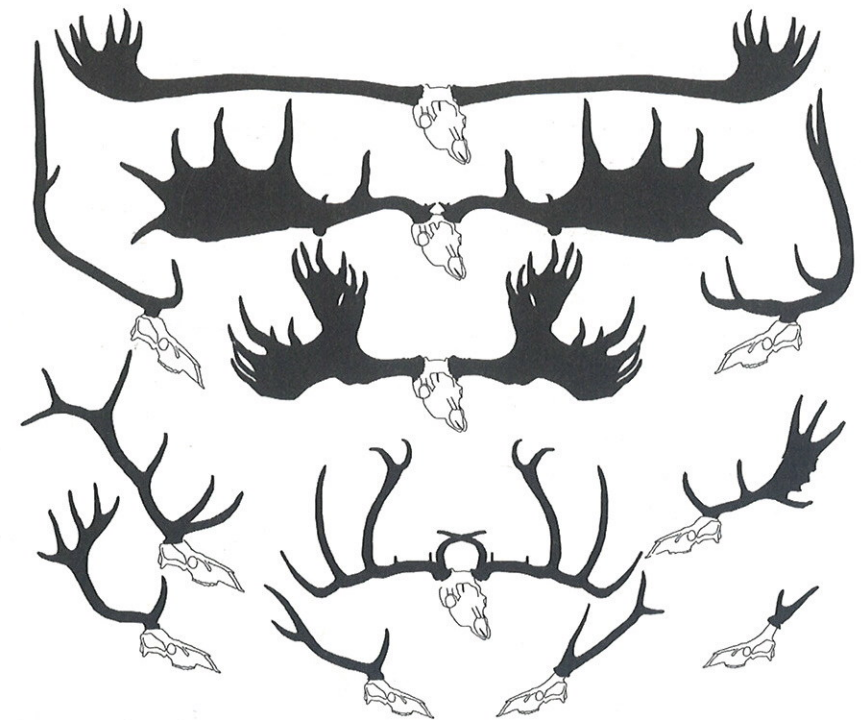
Rock ptarmigan are a species of grouse that lives on the arctic tundra. For most of the year the tundra is covered by snow, and both sexes are camouflaged by white plumage (Montgomerie *et al.* 2001). In the brief arctic spring, when the snow melts and is replaced by summer tundra, the females moult their white plumage in favour of green and brown feathers (Fig. 54). However, the males do not immediately moult their white feathers and are exceptionally conspicuous for some time, being attractive not only to prospecting females, but also easily discovered targets for

hunting gyrfalcons *Falco rusticolus*. When opportunities for mating disappear, the male ptarmigan camouflage themselves by soiling their white plumage and so are dirtiest at the start of incubation (Montgomerie *et al.* 2001). Only when a clutch is lost to predators and their mates become fertile again, do the males clean up. They eventually moult into a cryptic summer plumage once the season with mating opportunities really has ended. Thus, the visual aspect of the plumage in females is determined by its role for camouflage only. In males, plumage not only serves the dual role of camouflage in winter (when white) and in late summer (when comprising a cryptic patterning of browns), but also



**Figure 54.** Annual cycle of the external appearance of male and female rock ptarmigan in the Canadian Arctic in relation to changes in the camouflage afforded by the tundra habitat (indicated by the texture of the middle circle). This illustrates the power with which such intra-individual changes can be interpreted in a functional context. The changes in plumage represent a change in feathers, except for the change from a pure white to a dirty white colour of males in late June–July, which is the result of soiling.

From Piersma and Drent (2003) based partly on data from Montgomerie *et al.* (2001).



**Figure 55.** Variation in antler size and shape of extant and extinct 'true' deer Cervidae.

Compiled from Emlen (2008).

the sexually selected, ornamental role (white) in the mating season. The soiling of the white plumage when mating opportunities are no longer present (but when the moult into a cryptic plumage has not yet begun) provides a behavioural 'quick fix' to a critical phenotypic allocation problem.

Showing off is especially important in males that are in a position to monopolize more than one female during their fertile periods, and this is what deer do, at least as long as they live in semi-open habitats. The true deer (Cervidae) are unique among animals in having males that show temporary bony outgrowths on their heads, the antlers (Fig. 55).<sup>1</sup> Antlers are ornamental weapons that should impress both sexual competitors and prospective mates (Geist 1998,

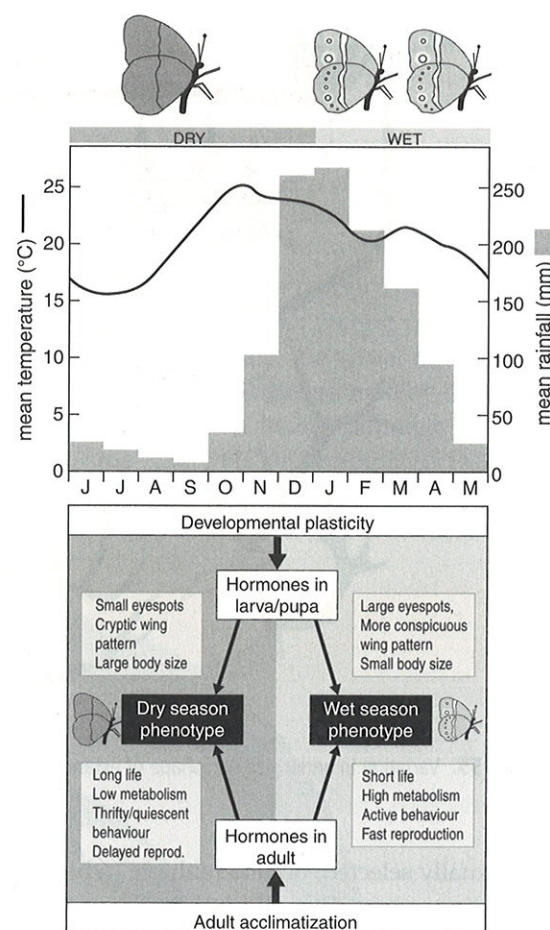
Emlen 2008). What makes deer antlers so different from functionally similar horns and tusks in other ungulates is that they are as seasonal as the plumages of ptarmigan. Antlers first grow at puberty, and successive sets of antlers developed by individuals tend to increase in size and complexity (Goss 1983, Bubenik and Bubenik 1990, Lincoln 1992). Whereas the small forest deer of the tropics may regenerate antlers at any time of the year, at temperate latitudes deer show synchronized seasonal cycles of growth, calcification, cleaning, casting, and regeneration (Lincoln 1992, Goss 1995). Regeneration of antlers occurs outside the reproductive season, in a period of low testosterone, through a process that is different from wound healing in that it is a stem cell based process (Kierdorf *et al.* 2007). Growing

<sup>1</sup> In reindeer and caribou *Rangifer tarandus*, females also have antlers, as they have to compete with the males and amongst each other for scarce fodder under arctic winter snow (Lincoln 1992).



antlers are covered by a 'velvet skin', which is shed when the underlying bone calcifies and dies, yielding the insensitive antlers that can be used as weapons during the rut. Hard antlers remain attached to the skull as long as the stags have raised testosterone levels (3–9 months, depending on species), after which the connection is severed and the antlers are cast (Lincoln 1992). Overall, the phenotype of individual male deer is as seasonally variable as it is in male ptarmigan.

Butterflies only live a couple of months. However, just like ptarmigan and most other organisms, butterflies experience seasonal environments and, in response, many butterfly species show seasonal polyphenism: within single genotypes, but in successive generations and depending on the time of year, they may show strikingly different phenotypes (Kingsolver 1995a, 1995b, Nijhout 1999). Well-studied examples are the *Bicyclus* butterflies from East Africa. Like other tropical butterflies, they are exposed to an alternation of dry and wet seasons. From June to October it is dry and mostly relatively cold, but from November through April it is wet and initially quite hot (Fig. 56). In the dry season, the butterflies are large and cryptically coloured. During the wet season, they show abundant eye-spots and a somewhat smaller body size (Windig *et al.* 1994, Brakefield *et al.* 2007). The alternative morphologies can be generated from split broods reared at either high or low temperatures (Kooi and Brakefield 1999). The butterflies of the dry season emerge around the end of the rains and have to survive almost half a year before they can lay eggs on fresh green grass at the start of the next wet season. To survive that long, they rely on their cryptic colouration and a quiescent lifestyle, including low metabolic rates (and a long life span under laboratory conditions). The ornamented butterflies of the wet season come in two generations. The first, which hatch from eggs laid by their cryptic parents, immediately mate and lay again, thus



**Figure 56.** Dry and wet seasonality in Malawi, East Africa, and the phenotypic responses by *Bicyclus anynana* butterflies that have one generation of inconspicuous large morphs in the dry season, and two generations of more conspicuous and smaller morphs in the wet season. Larvae of the last wet-season cohort develop with declining temperatures and produce the cohort of dry-season morphs without eye-spots. The diagram illustrates that this seasonal polyphenism is steered by both developmental plasticity (leading to the conditional metamorphosis induced in the larvae and expressed at pupation) and adult acclimatization (maternal, e.g. hormonal, contributions to the eggs laid in whatever season).

Compiled from Brakefield and Reitsma (1991), and Brakefield *et al.* (2007).

producing a slightly longer lived cohort, that then produces the cryptic cohort that endures the dry conditions, and so on. The wet season

butterflies show the distinct eye-spots; these may thwart attacking birds (Kodandaramaiah *et al.* 2009) and are important during mate choice (Constanzo and Monteiro 2007, Oliver *et al.* 2009). The colourful wet season butterflies are also much more active, have a higher metabolism and a shorter life-span (even when not exposed to predation in the laboratory). A wonderful combination of field and laboratory research enabled Paul Brakefield and co-workers (2007) to establish that the seasonal plasticity shown by *Bicyclus* butterflies represents aspects that are typical of both 'classical' developmental plasticity (hormonal effects in larva and pupa) and physiological acclimatization (induced in the reproductive adult stage and maternally contributed to the eggs) (Fig. 56).

The extents of phenotypic change shown by ptarmigan and butterflies are rather similar, but their life spans (and life-cycle characteristics) are not. Individual ptarmigan may experience many seasons, individual butterflies a single season at best. What is called seasonal polyphenism in insects is called life-cycle staging in birds and mammals. And whereas the genetic-developmental architectures behind the phenotypic expressions are likely to be very different in birds and butterflies, in both groups seasonally varying ecological shaping factors to do with feeding, the avoidance of predation, and mating, can functionally explain their strikingly variable phenotypes.

### Environmental variability and predictability, and the kinds of phenotypic adjustments that make sense

Different activities related to reproduction and survival (breeding, moult, migration, hibernation, etc.) are usually separated in time within individuals, tend to occur at predictable times of the year in seasonal environments, and are accompanied by changes in the reproductively

active phenotype (Murton and Westwood 1977). To perform optimally under a wide range of environmental conditions (variations that are often cyclic), a long-lived individual must track or anticipate the external changes by regulating gene expression to adjust its morphology, physiology, and behaviour (Willmer *et al.* 2000). The cyclically varying phenotypic expressions of these adjustments within an individual rock ptarmigan, for example, are thus called 'life-cycle stages' (Jacobs and Wingfield 2000, Piersma 2002, Ricklefs and Wikelski 2002). The seasonal template for such sequences might be provided by the natural photoperiodic rhythm and/or by an endogenous circannual pacemaker (Gwinner 1986). In addition, temperature, rainfall, food, or densities of conspecifics might give supplementary information, which individuals could use to 'fine-tune' the timing of their phenotypic transformations (Wingfield and Kenagy 1991). The polyphenisms of short-lived organisms, as in many insects, reflect a similar strategy (Shapiro 1976, Danks 1999).

The accuracy with which future environmental conditions can be predicted would determine the kind of phenotypic plasticity that one might expect to evolve (Levins 1968, Moran 1992, Padilla and Adolph 1996). In unpredictable environments, where there is insufficient environmental information, or if the wrong environmental information is used, an organism might end up with a phenotype that does not quite match its current environment (Hoffman 1978). Indeed, such mismatches are considered to be an important cost of developmental plasticity (see Table 7). However, this potential cost would disappear for organisms that are capable of fast and reversible phenotypic change. We will get back to this issue after we have addressed 'trade-offs'.

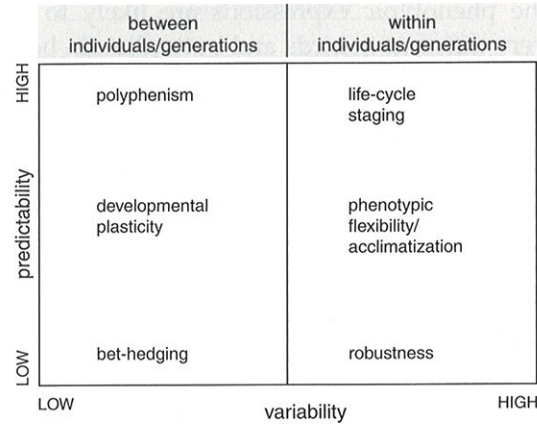
Carrying on from our introductory classification in Chapter 1 (Table 1), Fig. 57 represents the best attempt at present to place the different



**Table 7.** Inventory of costs of, and limits to, phenotypic plasticity. Developed from DeWitt *et al.* (1998), Pigliucci (2001a), Lessells (2007), Valladares *et al.* (2007), and Vézina *et al.* (2010).

Costs of plasticity	Limits to plasticity
<i>Maintenance:</i> energetic costs of sensory and regulatory mechanisms, and of any additional structures.	<i>Phylogenetic 'constraint':</i> however adaptive a kind of plasticity would be, in any organism some forms of plasticity are impossible because of historic genetic, developmental or sensory constraints.
<i>Production:</i> excess costs of producing structures plastically rather than as fixed genetic responses.	<i>Damage-related constraints:</i> in plants, phenotypic plasticity may be limited by herbivory because grazing damage prevents achievement of the optimal phenotype.
<i>Information acquisition:</i> investments (risk, time, energy) for sampling the environment, including lost opportunities (e.g. mating, foraging).	<i>Phenotypic compromise:</i> resource constraints may bias functional adjustment to one trait at the cost of another.
<i>Developmental instability:</i> phenotypic imprecision may be inherent for environmentally contingent development, which can result in reduced fitness under stabilizing selection.	<i>Information reliability:</i> the environmental cues may be unreliable or changing too rapidly.
<i>Genetic:</i> due to deleterious effects of plasticity genes through linkage, pleiotropy, or epistasis with other genes.	<i>Lag time:</i> the response may start too late compared with the time schedule of the environmental change, leading to maladaptive change.
	<i>Developmental range:</i> plastic genotypes may not be able to express as wide a range of adaptive phenotypes as a polytypic population of specialists would, e.g. because of limitations in the workings of neuroendocrine control system.
	<i>Epiphenotype problem:</i> the plastic responses could have evolved recently and still function like an 'add-on' to the basic developmental machinery rather than as an integrated unit; this may compromise their performance.

classes of phenotypic plasticity within a two-dimensional space, bounded by axes that represent (1) the degree of environmental variability (relative to the organism concerned) and (2) the degree to which this variability is predictable (Drent 2004). Highly predictable changing environments would select for *polyphenism* in short-lived organisms and *life-cycle staging* in long-lived organisms. The lower the predictability of environmental variation, the better it is for organisms to respond opportunistically, rather than seasonally scheduled. *Developmental plasticity* would then describe the kind of variable responses: organisms encountering unpredictably variable environments in the course of their life would benefit from plasticity being reversible, i.e. showing *phenotypic flexibility*. If environmental variation cannot be predicted, organisms might go into *bet hedging* (generating differently adaptive phenotypes at random) in shorter-lived organisms. In theory at least, especially longer-lived animals could also cope with extravagant changeability of the environment by not adjusting the phenotype at all, i.e. show *robustness*.



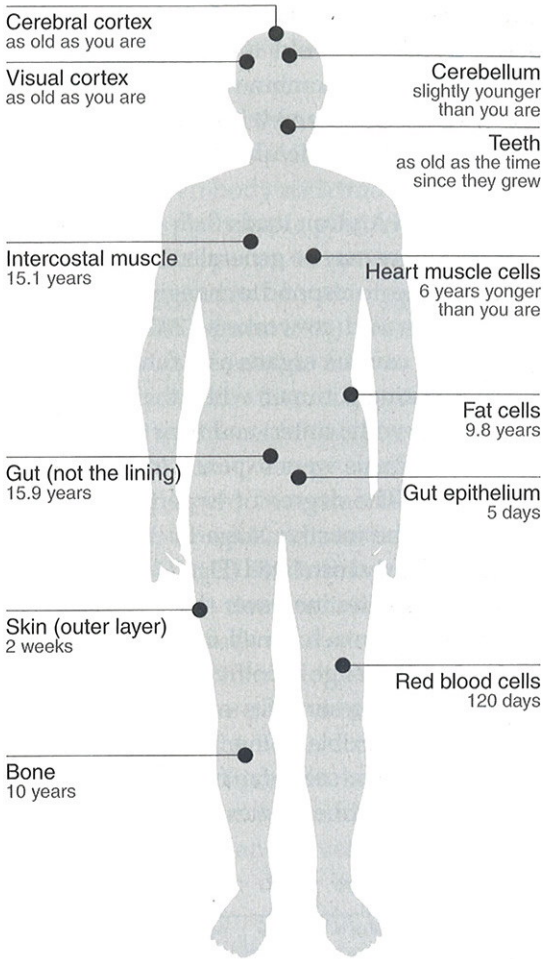
**Figure 57.** Different classes of phenotypic plasticity (or the lack of it in the case of robustness) arranged according to an axis of environmental variability over time and an axis of environmental predictability over time. The categories on the left show plasticity between individuals (and thus, between generations), and on the right within individuals. Inspired by Meyers and Bull (2002) and adapted from Drent (2004).

Degrees of flexibility

In the previous chapter we encountered sled dogs that, along with human polar explorers, were the true champions of endurance exercise. Arctic sled dogs hard at work would be expected to show many physiological and internal morphological changes compared to sled dogs on holiday. In summer, the absence of snow and ice puts sled dogs out of work. Nadine Gerth and co-workers (2009) used the contrast between summer and winter for a comparison of muscle size and ultrastructure in Greenlandic sled dogs. During the summer holiday, dogs had thinner muscle, the slimming representing changes in mitochondrial numbers, lipid droplet sizes, and numbers of contractile myofibrillar elements. Gerth *et al.* (2009) were surprised that capillary networks in the muscles did not change with season, and did not explain it. Could there be inherent differences between tissue types in the degree to which they can adjust to changing functional demands (Lindstedt and Jones 1987)? And what measure would best reflect a capacity for change? Cell turnover perhaps?

A fruitful approach to look at cell turnover in different tissues, especially the longer-lived ones, was the serendipitous use of peaks in <sup>14</sup>C in the atmosphere generated by nuclear bomb testing in the 1960s. This <sup>14</sup>C is incorporated into body tissues. Spalding *et al.* (2005a) used DNA from stored tissues to estimate their age at the moment of sampling (often after death). They verified the method with the enamel of teeth that, once formed, are not rebuilt (Spalding *et al.* 2005b). Whereas the lining of the gut and the outer skin show fast DNA turnover, indicating fast cell-replacement (Fig. 58), the DNA in fat cells, muscle cells, and bone cells is not replaced in many years. The oldest tissues in any person are the heart (cardiac muscle cells) and various components of the brain.

The high turnover of gut epithelium tallies well with the high flexibility of the intestine



**Figure 58.** Most of your cells are younger than you are (at least the DNA in the cells is), but there is a lot of variation between cell types.

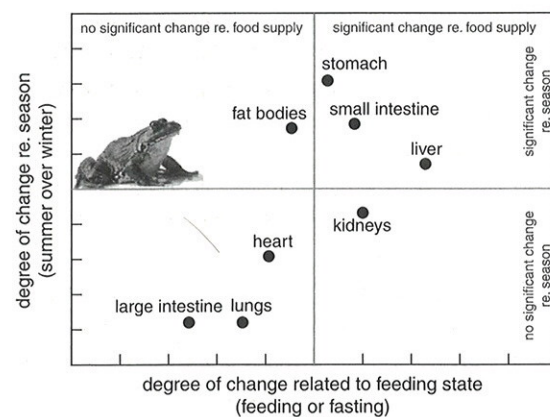
Modified from Vince (2006), partly based on Spalding *et al.* (2005a, 2005b 2008) and Bergmann *et al.* (2009).

that we discussed earlier. However, we have also shown that hearts show fast changes in size in response to changes in workload, so how does that tally with the cardiac muscle cells living so long (Bergmann *et al.* 2009)? The discrepancy is due to cell components other than the nucleus being built and broken-down, rather than whole cells being renewed and removed. Myofibrillar volume may change, and the densities and total volume of mitochondria may quickly change too (Hoppeler



and Flück 2002). Note that the brain is mainly composed of very old cells (Fig. 58). Brains thus beautifully combine permanence (constancy of cell numbers) with change (new neural connections made all the time) (Edelman 2004).

A study on Andean toads *Bufo spinulosus* suggests that there may be generalizable, tissue-specific, capacities to respond to changing conditions. Daniel Naya and co-workers (2009) compared the sizes of various organs as a function of season (comparing summer, when the animals are most active, with winter) and their feeding state (summer animals were exposed to a two-week fast or fed). The degree of hypertrophy of the active over the inactive stage in the two situations was clearly correlated (Fig. 59): heart, lungs, and the large intestine never showed significant change, but stomach, small intestine, and liver showed great change in both contexts. The highly flexible small intestine fits our expectations, as does the inflexible lung (see Chapter 3). Nevertheless, we seem far from clearly understanding why some tissues, or components of



**Figure 59.** Degree of change in the dry mass of various organs of mature Andean toads *Bufo spinulosus* as a function of either feeding and fasting (X-axis) or season (summer compared with winter, Y-axis). The degrees of change reflect the first two principal components in a factor analysis.

Based on Naya et al. (2009).

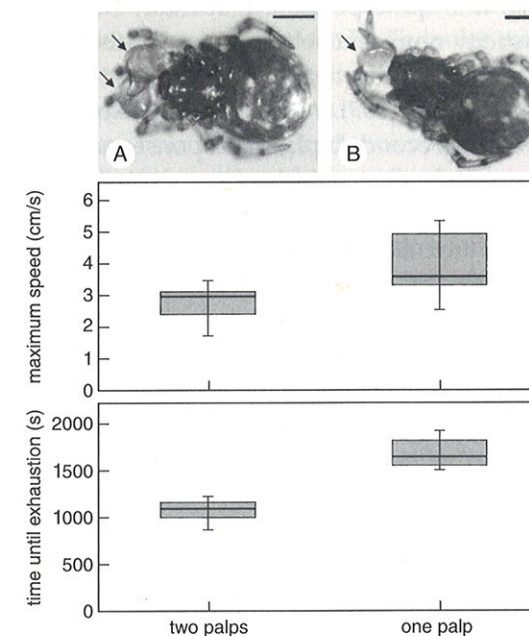
these tissues, show greater and faster responses to changes in demand than others.

### Direct costs and benefits, their trade-offs, and other layers of constraint

Do you still remember that a big penis might hinder the waving of cirral nets to catch planktonic food items by barnacles (Hoch 2008)? Then here is a somewhat similar example, where the trade-off between benefits and costs of a sizeable reproductive organ has been mapped in much more detail. Enter arachnids, or spiders. In most species, males are smaller than females, but in some species, males are dwarfs (Vollrath 1998). This might be because such small males can use ballooning on silky threads to find distant females, or because they would be no threat or competitor for the females, which would ease the access to what, in spiders, is the discerning and ruling sex. In cobweb spiders of the genus *Tidarren*, the males are so tiny, about 1% of the female's mass, that it raises 'a delicate problem' (Vollrath 1998): if the mating organs, a paired structure with the name pedipalps (which they fill with the sperm to be pumped into the female), have shrunk in proportion to the body, they would no longer fit snugly—key to lock—into the female's receptive organ, the epigyne. *Tidarren* spiders have solved this problem by the tiny males carrying relatively massive copulatory organs, to the extent that this makes the spiders pretty clumsy and reduces their chances in the scramble to get to a fertile female before others (Ramos et al. 2004). The tiny *Tidarren* males have a radical solution, as they actively break off the carrying arm of one pedipalp before filling it with sperm and before they approach a female. The loss of one pedipalp speeds them up considerably, and also makes them more enduring athletes (Fig. 60). This partial emasculation has clear dividends and may not even be as costly as it would seem at first sight.

Female *Tidarren* tend to consume males they have mated with and mostly give them the chance to insert only a single pedipalp. Still, these spiders may trade insemination capacity for speed and endurance, providing an example of a trade-off that leads to the severing of a body part that gets in the way at some stage.

Trade-offs are about alternative investments, the allocation problem of where, and in which parts of the phenotype, to invest scarce resources (e.g. Stearns 1989, 1992, Gervasi and Foulfopoulos 2008). Many dung beetle males develop large horns on the head or on the thorax, with which they compete for females. In a manipulative study where larvae of *Onthophagus nigriventris* were prevented from



**Figure 60.** Minute males of the cobweb spider *Tidarren sisypoides* before (A) and after (B) removal of one half of the paired ejaculatory organs, the pedipalps. The pedipalps are indicated by the two (A) or single (B) arrows; the scale bar represents 1 mm. Box plots of maximum speed before and after pedipalp removal ( $n = 16$ ) and time till exhaustion in two groups of spiders still having two pedipalps ( $n = 15$ ), or a single ( $n = 17$ ) one, are shown below the photos.

Compiled from Ramos et al. (2004).

developing such horns, Leigh Simmons and Douglas Emlen (2006) could show that the growth of these horns actually occurred at the expense of an organ that also helps to secure progeny—large testes. All the forms of phenotypic plasticity embody such trade-offs. Theory has it that for phenotypic plasticity to evolve, no single phenotype can be optimal in all environments experienced by the organism (Via and Lande 1985, Moran 1992). Thus, there *must* be trade-offs (and reliable cues to inform the organism about the state of its variable environment). Investments in one direction necessarily, due to time, energy, and nutritional limitations, prevent investments in another direction (for a model and a test with *Daphnia*, see Hammill et al. 2008). Only fantasy creatures called 'Darwinian demons' would be able to respond continuously and at many levels to changes in trade-offs by adjusting phenotype. Dung beetles that are able to predict that they can monopolize the female once they have secured it, would invest in horns; those that predict they would not, would play the sperm-competition game and invest in testes. The Darwinian demonic dung beetles would be able continuously to adjust the relative sizes of their horns and testes in response to fluctuating ecological and social environments. Although dung beetles show considerable developmental versatility (Emlen et al. 2007, Rowland and Emlen 2009), Darwinian demonic dung beetles do not exist. There are always limits to plasticity.

This is all we might have said on trade-offs and phenotypic plasticity, except that in the pertinent literature there exists another layer of concern: the trade-off between being phenotypically plastic, or not being plastic at all (Newman 1992, Via et al. 1995, DeWitt et al. 1998, Pigliucci 2001a); this is related to the idea that plasticity itself is a trait with a genetic basis (Relyea 2005). Perhaps there are true costs to plasticity itself (Table 7). Such costs



could include the energetic costs of the maintenance of sensory and regulatory structures enabling plasticity, or the excess costs of producing a plastic rather than a fixed (robust) structure. Adaptive plasticity makes it necessary to acquire information about the environment, and such acquisition will cost time. Plastic structures may (or may not) be more unstable than developmentally fixed structures, and if there were to be such things as plasticity genes, in some combinations with other genes they *could* be deleterious. In a review, Pigliucci (2001a) could not come up with much evidence in favour of any such costs. But perhaps all these theoretically possible costs of plasticity are not as far-fetched as they might at first sight appear. In the first specific study on the costs of plasticity (Relyea 2002a), evidence for such costs appears 'pervasive'. Comparing sibships of larval frogs that possessed different degrees of phenotypic plasticity, depending on the trait examined and the environment experienced, increased plasticity could either have positive, negative, or no effects on fitness components, such as degrees of trait development, mass gains, and survivorship. Thus, now that the appropriate studies are being done (Lind and Johansson 2009), there is increasing evidence that plasticity per se may come at a cost.

The mere fact that Darwinian demons do not exist, actually demonstrates that there *are* limits to plasticity, and we provide a list of possibilities in Table 7. Let us quickly run through them. Phylogenetic constraints are obvious, damage through herbivory may prevent plants from achieving locally optimum phenotypes (Valladares *et al.* 2007), organisms may have to compromise between different adjustments in the light of resource limitations (Vézina *et al.* 2010), environmental information is bound to be unreliable at times, and lag-times between environmental changes and phenotypic responses must be an issue.

However, the last two limits-categories are somewhat controversial. As Massimo Pigliucci (2001a) points out, for an intra-individual phenomenon it is odd to juxtapose individual constraint with population variation, as DeWitt *et al.* (1998) did in their formulation of a 'developmental-range' limit. We left it in Table 7 because it is quite possible that control mechanisms (e.g. of a neuroendocrine nature) limit the way in which environmental information is fed into the functionally adjusting phenotype (Lessells 2007).

The 'epiphenotype problem', the issue that plastic responses may be recent add-ons that are not yet genetically and developmentally hard-wired and may therefore compromise performance, is interesting on two accounts. First, it is quite possible that many forms of plasticity represent self-organized rather than developmentally tightly orchestrated structures (Turner 2007). We shall get back to that in a minute. Second, 'a plastic response may start as an epiphenotype and subsequently be integrated by natural selection attempting to reduce the costs and limits of the new response' (Pigliucci 2001a, p. 176). In our final chapter we will return to plasticity as a creative force in evolution (West-Eberhard 2003). In a celebrated class of developmental plasticity that so far has escaped notice, all these elements of costs, benefits, and limits nicely combine. Let's look now at frogs and newts, and especially their tadpoles!

### Phenotypes of fear

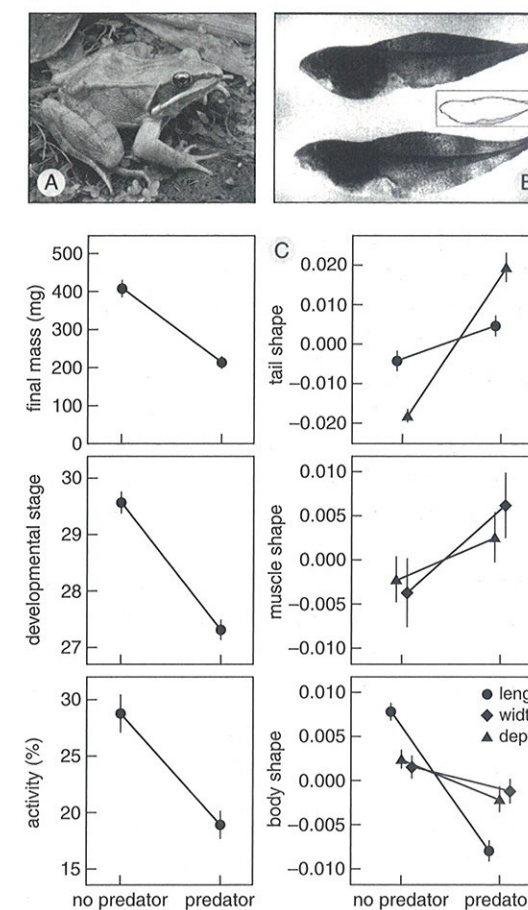
That amphibians live up to their name, that they never fully made the transition from living in water to living on land, actually gives them a lot of ecological opportunity. By their enormous life-history versatility, they can capitalize on the resources offered by ponds, pools, and puddles, and even the underlying soil (Newman 1992). Toads can survive years in dry desert soil, frogs

can survive deep-frozen ground, and when the rains come, or the spring arrives, the sleeping beauties wake up, display, and quickly mate and lay eggs, so that the larvae can use the temporary, ephemeral, pool- and puddle-habitats to mature and metamorphose in a life-history stage that can endure the subsequent lean times (Wilbur and Collins 1973, Wilbur 1990, Relyea 2007). Amphibians in Paradise, salamanders that have found permanent and predator-poor, deep-water bodies, forego that robust land-living stage altogether. Most famous among these reproductively active larvae is the Mexican axolotl *Ambystoma mexicana* (Gould 1977, Ryan and Semlitsch 1998).

What is of most concern here though, is not the metamorphosis into the adult stage, but life as a larva. Look at a tadpole: it is a cute little package of developing protein. Tadpoles make high-quality, easily digested food for anybody that is interested. Apart from hiding (and thereby refraining from foraging and thus not growing), their only defence is an ability to flee fast, to confuse, or, in the case of many toads, possess a distasteful skin. Under threat it helps to have a relatively big and muscular tail (Fig. 61), as it may enable the tadpole to swim fast (Dayton *et al.* 2005). A large tail fin could also act as a lure to attract the attention of predators away from vulnerable parts of the body (Doherty *et al.* 1998, Johnson *et al.* 2008). In many species of frog, tadpoles from ponds with predators are smaller, with relatively shorter bodies and deeper tail fins than tadpoles from predator-free environments (Relyea 2001a). Indeed, one of the most common tadpole predators, dragonfly larvae, preferentially kill tadpoles with relatively shallow and short tail fins, and narrow tail muscles (Van Buskirk and Relyea 1998). Tadpoles lend themselves well to experimentation, as artificial ponds, either stocked or not stocked with enclosed or free-swimming tadpole-predators of various kinds, and with different levels of food or food-competitors, are easily set up. The

work by Rick Relyea and his team has turned the American wood frog *Rana sylvatica* (Fig. 61) into the best-understood example of what it means to have a very plastic larval stage.

Relyea (2003a) showed that when tadpoles were exposed to the smells and actions of dif-



**Figure 61.** An adult wood frog *Rana sylvatica* (A), typical responses of wood frog tadpoles to either the presence (B, bottom) or the absence (B, top) of predatory dragonfly larvae, with a drawing by Rick Relyea, where the difference in tail depth between the two morphs is shaded (B, inset). The compilation in (C) shows the multiple phenotypic effects of caged dragonfly predators on growth, development, activity, and relative morphology of wood frog tadpoles. The morphological traits were size-adjusted by taking mean residual values ( $\pm$  SE) from regressions on mass.

Compiled from Relyea (2001b) (B) and Relyea (2002a) (C).



ferent predators (there was a choice of four) or combinations thereof, they discriminated and produced predator-specific larval phenotypes. When predators were combined, tadpoles generally developed phenotypes that were typical of the more risky type, suggesting that they perceived the risk of combined predators as the risk of the most dangerous one. Tadpoles not only have to survive, they also have to grow. Wood frogs that face high levels of competition from other tadpoles increase their foraging activity and develop larger bodies. These larger bodies contain relatively larger scraping mouthparts and longer guts, which improve the animal's ability to find, consume, and digest food, but this ability comes at the cost of a large tail, which makes tadpoles more susceptible to predators (Relyea and Auld 2004, 2005).

The extraordinary ability of tadpoles to sense changes in their environment and respond in precise and often appropriate ways, is illustrated, for example, by wood frog tadpoles in experimental ponds that appeared to respond differentially to variations in the *per capita* food levels and to changes in conspecific density alone (Relyea 2002b). When European common frog tadpoles *Rana temporaria* were exposed to non-lethal caged predators (dragonfly larvae) at low tadpole densities they showed morphological as well as behavioural defences. At high densities and with severe competition for food they remained active and only adjusted their morphology (Teplitsky and Laurila 2007). Similarly, when tadpoles were exposed to fish (that pursue their prey), they built deeper and longer tails and bigger tail muscles. In the presence of dragonfly larvae (sit-and-wait predators), they built only deeper tails (Teplitsky *et al.* 2005a, Wilson *et al.* 2005), suggesting that tails as lures were the selective force behind oversized tail fins. Indeed, in a Neotropical tree frog *Dendrosophus ebraccatus*, the presence of dragonfly larvae induced

highly visible, large, and colourful tail fins, whereas the presence of fish induced achromatic tails (Touchon and Warkentin 2008). Tadpoles of the tree frog *Hyla versicolor* altered their defences when 10 different prey (themselves being one of the types) were either crushed by hand or consumed by dragonfly larvae. Across all prey types, crushing induced only a subset of the defences that were induced by consumption, suggesting once more that tadpoles can assess quite precisely the degree and type of threat and respond accordingly (Schoeppner and Relyea 2005).

Nevertheless, even the plasticity of tadpoles finds a limit. When Schoeppner and Relyea (2008) exposed wood frog tadpoles, and when Teplitsky *et al.* (2005b) exposed tadpoles of a European frog *Rana dalmatina*, to a gradient of densities of a particular predator, all measured traits exhibited graded responses that levelled off with increased predation risk. This suggests that there is either an organizational limit to plasticity or a 'functional limit' that reflects the high costs of the defensive phenotype. Part of these costs might be incurred later in life. Wood frog tadpoles reared with caged dragonfly larvae, rather than without, showed relatively large limbs and narrower bodies as fully grown frogs (Relyea 2001b); the survival value of these later adjustments was not established. However, four months after metamorphosis, common frogs that developed from predator-exposed, rather than predator-free, tadpoles swam more slowly with less endurance: the opposite of what happened in the larval stage (Stamper *et al.* 2009). This suggests that the larval adjustments may come with a long-term fitness cost.

We round up with the amazing story of two competing amphibian larvae studied in Japan—a frog *Rana pirica* and a salamander, or newt, called *Hynobius retardatus*. Tadpoles of many a salamander or toad have tendencies to become cannibalistic, but they only do so at high con-

specific densities (Pfennig 1992, Michimae 2006), and, preferably, if these conspecifics are not closely related (Pfennig and Frankino 1997). They then develop a broad head and gape large enough to swallow tasty nieces and nephews. They also turn predatory when there are many frog tadpoles around, as the mechanical vibrations of flapping tadpole-tails are sufficient for this transformation to take place (Michimae *et al.* 2005). In a wonderful example of a phenotypic arms race, in response to salamanders becoming broad-headed and dangerous, frog tadpoles developed thicker bodies and a comb on their backs to prevent ingestion by the salamander larvae; to which salamanders responded by further enlarging gape size (Kishida *et al.* 2009)! Interestingly, frog tadpoles do not become bulky and high-bodied solely in the presence of salamander tadpoles, they only adjust when the salamander tadpoles become dangerous, i.e. express the predacious phenotype (Kishida *et al.* 2006). A final subtlety is that when the threat of either predatory larval salamanders or dragonflies is taken away, the specific morphological defences are lost (Kishida and Nishimura 2006). This reversal indicates once more that induced defences can come with serious costs (Hoverman and Relyea 2007), and thus that flexibility, or reversibility, pays if it is still an option developmentally (Relyea 2003b).

### Plasticity: the tinkerer's accomplishment?

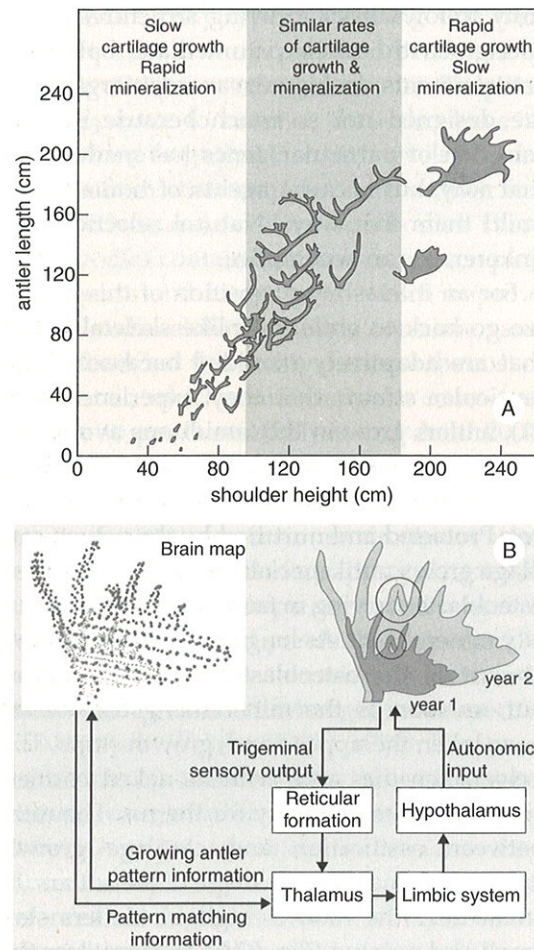
With a few exceptions (e.g. Newman and Müller 2000, Blumberg 2009), the literature on phenotypic plasticity is permeated with the idea that adaptive phenotypic responses, be they reversible or irreversible, cyclic or not, are instructed from the genome. Of course, they will be, at some level at least. In his eye-opening book *The tinkerer's accomplice*, J. Scott Turner (2007) builds on early lines of reason-

ing, including the famous 'spandrels'-paper by Stephen Jay Gould and Richard Lewontin (1979), and assembles many convincing arguments that good adaptive design is usually the result of the self-organization that logically follows from growing structures interacting with their immediate physical environments. In his own words: 'organisms are designed not so much because natural selection of particular genes has made them that way, but because agents of homeostasis build them that way'. Natural selection, the tinkerer, has an accomplice.

For an interesting exposition of this notion we go back to antlers. Unlike skeletal bones that are adaptively moulded because of the particular strains that they experience (Fig. 51), antlers grow in the 'assiduous avoidance of strain' by virtue of a specialized, blood-rich, and highly innervated type of skin called velvet. Protected and nurtured by the velvet, cartilage grows until specialized bone cells called osteoblasts, moving in from the blood, eventually mineralize it. As long as the cartilage stays abreast of the osteoblasts, the antlers grow, but, as soon as the mineralizing osteoblasts have taken the upper hand, growth stops. The velvet then dies and, with its naked ossified antlers, the stag is ready for the rut. The ratio between ossification and cartilage growth determines the size and shape of the antlers. In small deer the ratio is high, so antlers stay small and pointed (Fig. 62A). In large deer the ratio between ossification and cartilage growth is low, so antlers can grow as 'expansive broad plates' (Turner 2007). After the rut, the antlers are shed. The new set usually resembles the old set except that, if a deer is well fed, it is a bit larger. That antlers grow larger with age is probably self-organized. Reproductive and stress hormones combine with direct diet cues to affect the relative rates of ossification and cartilage growth, and thereby antler size and shape.



Now comes a surprise. When, during some stage in a deer's life, antlers incur damage at one side because a bit of velvet got damaged during antler growth, this damage will be vis-



**Figure 62.** (A) Antler length is a function of the shoulder height of the deer stag that carries it. The outcome is determined by a 'race' between how rapidly cartilage grows and how rapidly it mineralizes, a ratio that is body-size dependent. In small deer, mineralization relative to cartilage growth rates is high, which leads to small, single-point antlers. In the largest deer, the ratio is small and this produces the broad plate-like antlers carried by moose *Alces alces* and the extinct Irish elk *Megaloceros giganteus*. (B) A diagram outlining the ways in which antler shape may be memorized in the brain and how this memory feeds back into the shape of the antler during the subsequent growth cycle.

Put together from figures in Turner (2007).

ible again the next year, even if the velvet is fine then! The injured antlers are cast, but the memory of their shape apparently is not. Turner (2007) argues that the rich infiltration of the velvet with both sensory fibres (routed through the trigeminal nerve and the thalamus) and so-called autonomic fibres, which control a whole range of physiological functions, suggests that 'antlers are in the middle of a feedback loop in which the trigeminal sensory nerves convey information to the brain about the growing antler's shape, while pattern of growth is mediated by information streaming out on autonomic nerve fibres, perhaps through controlling patterns and rates of blood supply to the various parts of the growing antler' (Fig. 62B). This interaction would not only explain why growth deformities are copied in successive years, but also why antlers show such exquisite bilateral asymmetry.

There are no genes that directly instruct for antler size and shape. Antler form naturally results from variations in the relative rates of cartilage growth and ossification (which may partly be under genetic instruction), and the build-up of shape memories in the brain. Now think of the amphibian phenotypes of fear. Could the tail shape of tadpoles 'simply' result from differences in the kinds of exercise induced by the presence of particular tadpole predators?

### Phenotypic flexibility in birds

In the most celebrated case of evolution in the world of birds, the radiation of Darwin's finches on the Galápagos Islands (Lack 1947, Grant 1986, Weiner 1994, Grant and Grant 2008), the trait of most interest is beak size. Depending on rainfall, plant growth, and the production of seeds of different sizes, in different series of years either big-beaked or small-beaked finches have survival advantages. As beak size has a genetic underpinning, selection events show up in the beak-size trajectories of the relatively

isolated island populations. Immersed, as we are here, in examples of phenotypic plasticity, one cannot help but wonder why small-beaked Darwin's finches, living during times when bigger beaks would be advantageous, do not themselves build bigger bills?

Although oystercatchers *Haematopus ostralegus* can and do modify bill size and shape (Hulscher 1985, van de Pol *et al.* 2009), Darwin's finches apparently cannot. In their very advanced synthesis of 34 years of work on natural selection in action in Darwin's finches, Peter and Rosemary Grant (2008) do not even use the word plasticity! Perhaps the material with which, and the way that, bills are built only allows for limited plasticity (Gosler 1987). This contrasts with most other parts of bird bodies that do show plasticity, mostly in the categories of life-cycle staging (Murton and Westwood 1977, Wingfield 2008) and phenotypic flexibility (this book). In fact, one of the few examples of developmental plasticity in birds is the one for oystercatcher bills where, to an extent at least, youngsters learn from their parents to feed on a particular diet (Sutherland 1987, Sutherland *et al.* 1996). Depending on the prey types that they select, their bill is worn and strengthened in particular ways (yes, self-organization!), even though there remains flexibility later in life.

Rather than structural body size, what predominantly varies in birds are body mass (an indicator of nutrient stores) and the size of most of the organs (Table 8), with the exception of the lungs (Piersma *et al.* 1999b). This is especially the case in those birds exposed to seasonally widely different conditions, either because they migrate from arctic or temperate to tropical climates, or precisely because they stay put at highly seasonal northern or southern latitudes. There is also considerable intra-individual variation in correlated metabolic attributes (Vézina *et al.* 2006, 2007) and aspects of the immune system (Buehler *et al.* 2008a).

Female red knots even show seasonal variation in the mass of their skeleton: they store calcium before going to the trouble of laying a clutch of four eggs (Piersma *et al.* 1996b). A phenotypic aspect not studied in shorebirds, but carefully documented in songbirds, is the seasonal waxing and waning of song nuclei in the brain (Nottebohm 1981, Smith *et al.* 1997, Tramontin *et al.* 2000; note that even modern humans leading domesticated lives show seasonal change in parts of the brain, Hofman and Swaab 1992). Within a week after shifting Gambel's white-crowned sparrows *Zonotrichia leucophrys gambelii* from short to long days, the number of neurons in a brain area called the High Vocal Centre increased by 50,000, or 70%, as did the volume of this nucleus (Tramontin *et al.* 2000). Perhaps surprisingly, the morphological changes were faster than the development of the song control circuitry and the degree of stereotypy of the songs themselves. These took some extra weeks to mature fully.

In view of the focus of much of the rest of this book on foraging, distributional decisions, and time-allocation problems, we round up by examining in greater detail the machinery for digestion and assimilation in birds, and their plasticity. The digestive system of birds can be divided into several main components (Table 9). In any one taxon, each of these digestive organs will have morphological characteristics that functionally reflect the evolutionary history and contemporary ecology of the taxon (Stevens and Hume 1995).

A large gut may provide the ability to use foods of low quality that are difficult to handle mechanically or chemically. The size of the digestive tract should, therefore, reflect the benefits of having organs of that size, in balance with the costs of maintaining that system. Before assessing degrees of variation in organ size, it is worth considering some general factors that may set upper bounds to the size of digestive systems. For example, it is obvious



**Table 8.** Many phenotypic traits of shorebirds (red knot, great knot, bar-tailed godwit, and others) show considerable intra-individual phenotypic change (C; relative to minimum) in the course of the seasons (S) and/or in relation to ambient environmental conditions (E).

Trait	C	S/E	References
Body mass	2.5 ×	S, E	Piersma <i>et al.</i> (2005)
Skeleton	1.4 ×	S	Piersma <i>et al.</i> (1996b)
Fat stores	150 ×	S, E	Piersma <i>et al.</i> (1999b)
Flight muscles	3 ×	S, E	Dietz <i>et al.</i> (2007)
Blood haematocrit	1.5 ×	S, E	Landys-Ciannelli <i>et al.</i> (2002)
Heart	1.4 ×	S, E	Piersma <i>et al.</i> (1999b)
Liver	2.5 ×	S?, E	Piersma <i>et al.</i> (1999b)
Gizzard	7 ×	E	Piersma <i>et al.</i> (1993b)
Intestine	4 ×	E	Battley <i>et al.</i> (2005)
Skin	1.7 ×	S	Battley <i>et al.</i> (2000)
Testicle	30 ×	S	T. Piersma unpubl. data
Feather mass	1.3 ×	S	Piersma <i>et al.</i> (1995a)
Basal metabolic rate	2.5 ×	S, E	Piersma <i>et al.</i> (1995a)
Peak metabolic rate	1.6 ×	S?, E	Vézina <i>et al.</i> (2006)
Basal corticosterone level	20 ×	S	Piersma <i>et al.</i> (2000)
Daily peak melatonin titer	10 ×	S	Buehler <i>et al.</i> (2009c)
Total leukocyte count	4 ×	S, E	Buehler <i>et al.</i> (2008a)
Microbial killing efficiency	10 ×	S, E	Buehler <i>et al.</i> (2008a)
Preen wax composition	Complete qualitative shift	S	Reneerkens <i>et al.</i> (2002, 2007)

**Table 9.** Subdivision of the major components of the digestive system in birds. Based on Klasing (1998), Battley and Piersma (2005), and J.M. Starck (pers. comm.).

Organ	Alternative name/components	Primary functions
Oesophagus + crop		Food storage, movement of food toward proventriculus
Proventriculus	Glandular stomach	Gastric secretion
Gizzard	Muscular stomach	Crushing or grinding food, mixing food with gastric secretions
Small intestine	<i>Intestinum tenue</i> , composed of <i>duodenum</i> and the combination of <i>jejunum</i> and <i>ileum</i> (the 'jejunoileum')	Enzymatic digestion, absorption of digestive end products
Ceca		Microbial fermentation, water and nitrogen absorption, immunosurveillance
Rectum	Colon, large intestine	Electrolyte, water and nutrient absorption
Cloaca		Storage and excretion of urine and faeces
Liver		Metabolism of absorbed nutrients, production of bile acids and bile salts
Pancreas		Secretion of digestive enzymes

that there is a physical limit to the volume available for organs within the abdominal cavity. Fat deposits will further reduce the abdominal space for the digestive tract. For example, migratory shorebirds deposit a thick layer of fat around the gizzard, covering the intestines and rectum, and pressing up against the tail end of the skeleton. In great knots *Calidris tenuirostris* about to depart on northward migration from Australia, fat in the abdominal cavity made up 38% of the total abdominal tissue mass (Battley and Piersma 2005). At such times the profile of the abdomen changes, with fat birds showing a 'bulge' behind the legs (Owen 1981, Wiersma and Piersma 1995). Conflicts for space between fuel stores and organs may limit the maximum size of the digestive organs.

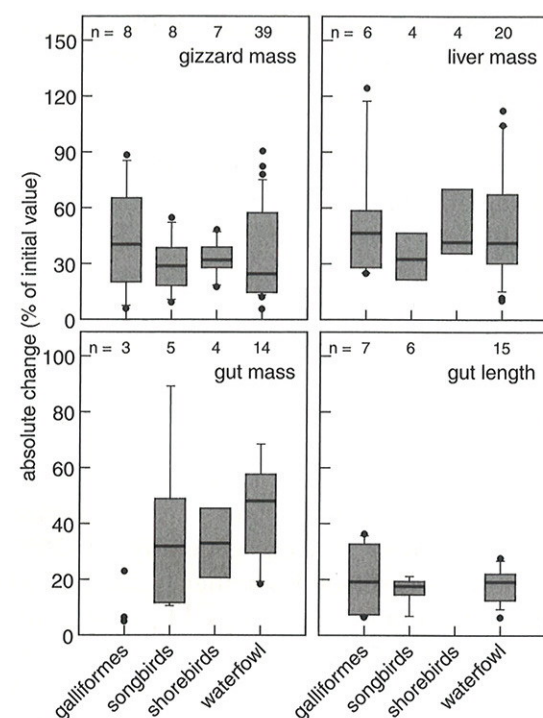
Internal organ mass could directly affect flight costs and performance of birds. Because the costs of flight increase with body mass (Kvist *et al.* 2001), and as manoeuvrability is impaired at higher masses (Dietz *et al.* 2007), minimization of digestive organ mass will be an important consideration. In a comparative study of raptors, Hilton *et al.* (1999) showed that 'pursuing' species (that require fast and manoeuvrable flight) had shorter guts than 'searching' species, presumably reflecting the pursuers' need to minimize mass. The shorter retention time of these species would reduce the period of time they had food in their guts, also keeping body mass low, though at a cost of lowered digestive efficiency. Sedinger (1997) showed that grouse have caeca 4–5 times longer than waterfowl feeding on the same diet, and suggested that the small caeca in waterfowl reflect a balance between the costs of flight for waterfowl and the benefits of the caeca for nutrient balance. And even in animals at rest, mass-specific maintenance costs of the intestine and liver are greater than those of muscle and adipose tissue (Krebs 1950, Blaxter 1989, Scott and Evans 1992). Reductions in metabolically active tissues, therefore, dramatically

reduce basal energy requirements (Battley *et al.* 2001, Konarzewski and Diamond 1995).

Under the simplifying assumption that size (or mass) indicates functional capacity, Fig. 63 summarizes the magnitude of changes in three major gastrointestinal components, the gizzard, gut or small intestine, and liver. Data are separated into four taxonomic groups: galliformes (primarily grouse and quail), songbirds, shorebirds, and waterfowl. Even though only few of the studies in the review explicitly focused on documenting such changes, organ mass variations of 20–80% were common across a wide range of species; length changes of the intestine typically were lower, generally 10–20%. Earlier we discussed whether tissue-related differences in flexibility could be due to differences in cell turnover, based on carbon-isotope signatures of historically spiked DNA (Spalding *et al.* 2005a). A method that informs about the whole cell content, rather than DNA only, may be more appropriate, and measurements of retention times of carbon-isotopes supplied in food fulfil this condition. In a study on zebra finches *Taeniopygia guttata*, Bauchinger and McWilliams (2009) found retention times varied between 8 days for small intestine, 10–13 days for stomach, kidney, liver, and pancreas, 17–21 days for heart, brain, blood, and flight muscle, and 26–28 days for leg muscle and skin. Thus, they found that tissues with a tendency to show large changes (notably the small intestine and liver) had fast turnover rates, whereas less dynamic tissues, such as the flight and leg muscles, had slower turnover rates. Tissue-specific turnover rate may thus partially determine the magnitude of organ flexibility.

Clearly, if we keep an open mind and our eyes open, pretty much all parts of bodies appear to vary with time, age, demand, and environment. To ascertain the functionality of these changes





**Figure 63.** Absolute change (either increase or decrease) in digestive organs of birds. Data represent maximum percentage changes recorded in studies for age- or sex-classes of birds, due to factors including diet type or quality, migration, breeding, and food intake. Measurements from wild and captive birds are included. The gut category represents data for both the entire gut (including caeca and rectum) and the small intestine only. Sample sizes for each taxonomic group are given above the box or points. Boxes represent the 25–75 percentiles (divided by the median), whiskers represent the 10 and 90 percentiles, and outliers are shown as dots. For groups with only a few values, individual data points are plotted.

From Battley and Piersma (2005).

we need to do much more than 'just' record the morphological changes. In the next few chapters we considerably expand our view beyond bodily structures to examine active free-living organisms, especially birds, as an integrated whole.

### Synopsis

'A single genotype can produce many different phenotypes, depending on the contingencies encountered during development. That is, the phenotype is the outcome of a complex series of developmental processes that are influenced by environmental factors as well as by genes. Different environments can have an effect on the outcome of development that is as profound as that produced by different genes'. To this succinct description of phenotypic plasticity by Nijhout (1999), we would like to add the possibility that much of the adaptedness shown by phenotypes is actually driven by self-organizational processes (Turner 2007). Reproductively successful phenotypes will satisfy ecological demands by optimizing the balance between various cost and benefit functions of specific kinds of phenotypic (trait) variation. In brief, most traits of all organisms are responsive to particular environment features over shorter or longer time scales. Consequently taking ecological context into account will enrich biology on all fronts.

## Part III

### Adding behaviour