

REVIEW

Ecological Developmental Biology: Developmental Biology Meets the Real World

Scott F. Gilbert¹

Biology Department, Edward Martin Research Laboratories, Swarthmore College,
500 College Avenue, Swarthmore, Pennsylvania 19081

The production of phenotype is regulated by differential gene expression. However, the regulators of gene expression need not all reside within the embryo. Environmental factors, such as temperature, photoperiod, diet, population density, or the presence of predators, can produce specific phenotypes, presumably by altering gene-expression patterns. The field of ecological developmental biology seeks to look at development in the real world of predators, competitors, and changing seasons. Ecological concerns had played a major role in the formation of experimental embryology, and they are returning as the need for knowledge about the effects of environmental change on embryos and larvae becomes crucial. This essay reviews some of the areas of ecological developmental biology, concentrating on new studies of amphibia and *Homo*. © 2001 Academic Press

INTRODUCTION

As developmental biology matures, it finds itself increasingly interacting with other areas of biology, and helping each of these areas to solve some of its major questions. The integration of developmental biology with molecular genetics has enabled us to understand how gene regulation specifies tissues and regulates cell differentiation. The integration of developmental biology with human genetics has enabled us to understand the mechanisms by which mutant genes produce syndromes involving different organ systems and has explained why some genetic conditions are dominant while others are recessive. The new integration of developmental biology with evolutionary biology is allowing us to understand how changes in gene expression during development can alter the formation of body plans. There is now another field with which developmental biology is negotiating: ecology. Moreover, as in any good interaction (including the aforementioned ones), the effects are going to be reciprocal. In ecological developmental biology (“eco-devo”), we can expect that developmental biology will change as much as ecology does.

Ecological developmental biology is the meeting of developmental biology with the real world. It involves studying

development in its natural context rather than only in the laboratory. It means, as Mead and Epel (1995) state, that sea urchin development must be studied among breakers, as well as in beakers. The field of eco-devo investigates many problems that have traditionally been segregated into fields, such as immunology, larval ecology, pest management, life history strategies, neurobiology, and even dentistry. Table 1 lists some of the areas of eco-devo for animals.

Ecological developmental biology used to play a much larger role in our field. In the mid- and late 19th century, embryology had centered on ecological and evolutionary paradigms. Indeed, experimental embryology originated among those investigators who sought to understand how the environment determined phenotype (Nyhart, 1995). August Weismann (1875), for instance, noted that *Araschnia levana* butterflies eclosing from their pupae at different seasons had differently colored wings (Figs. 1A and 1B), and he could turn the summer morph into the spring form by cooling the pupae. Carl von Siebold (1854) showed that some parthenogenetic female aphids produced eggs that matured into both males and females late in the breeding season, to produce an overwintering egg (which would invariably hatch as a parthenogenetic female). Thomas Hunt Morgan's first chromosome studies (1909) demonstrated the cytological mechanism for this phenomenon. Several investigators studied environmental sex determina-

¹ Fax: 610-328-8663. E-mail: sgilber1@swarthmore.edu.

TABLE 1

Some of the Zoological Areas Covered by Ecological Developmental Biology

-
- I. Context-dependent normal development
- A. Morphological polyphenisms
1. Nutrition-dependent (*Nemoria*, hymenoptera castes, sea urchin larvae)
 2. Temperature-dependent (*Arachnia*, *Bicyclus*)
 3. Density-dependent (locusts, *Scaphiopus*)
- B. Sex determination polyphenisms
1. Location-dependent (*Bonellia*, *Crepidula*)
 2. Temperature-dependent (*Menidia*, turtles)
 3. Social-dependent (wrasses)
- C. Predator-induced polyphenisms
1. Adaptive predator-avoidance morphologies (*Daphnia*, *Hyla*)
 2. Adaptive immunological responses (*Gallus*, *Homo*)
 3. Adaptive reproductive allocations (ant colonies)
- D. Stress-induced bone formation
1. Prenatal (fibular crest in birds)
 2. Postnatal (patella in mammals; lower jaw in humans?)
- E. Environmentally responsive neural systems
1. Experience-mediated visual synapses (*Felix*, monkeys)
 2. Cortical remodeling (phantom limbs, learning)
- II. Context-dependent life-cycle progression
- A. Larval settlement
1. Substrate-induced metamorphosis (bivalves, gastropods)
 2. Prey-induced metamorphosis (gastropods, chitons)
 3. Temperature/photoperiod-dependent metamorphosis cues
- B. Diapause
1. Overwintering in insects
 2. Delayed implantation in mammals
- C. Sexual/asexual progression
1. Temperature/photoperiod-induced (aphids, *Megoura*)
 2. Temperature/colony-induced (*Volvox*)
- D. Symbioses/parasitism
1. Blood meals (*Rhodnius*, *Aedes*)
 2. Commensalism (Euprymna/Vibrio; eggs/algae; *Paleon/Alteromonas*)
 3. Parasites (*Wachtellia* in *Wollbachia*)
- E. Developmental plant-insect interactions
- III. Embryo and larval adaptations to environments
- A. Egg protection
1. Sunscreens against radiation (*Rana*, sea urchins)
 2. Plant-derived protection (*Utetheisa*)
- B. Larval protection
1. Plant-derived protection (*Danaus*, tortoise beetles)
- IV. Teratogenesis
- A. Chemical-induced teratogenesis
1. Natural compounds (retinoids, alcohol, lead)
 2. Synthetic compounds (thalidomine, warfarin)
 3. Hormone mimics (diethylstilbesterol, PCBs)
- B. Infectious agents
1. Viruses (*Coxsackie*, *Herpes*, *Rubella*)
 2. Bacteria (*Toxoplasma*, *Treponema*)
- C. Maternal conditions
1. Malnutrition
 2. Diabetes
 3. Autoimmunity
-

Note. This is one only person's view, and it is chiseled in 1% agarose. There is overlap in the categories, and this list is not to be thought of as inclusive. The entire areas of plant developmental plasticity and plant-animal interactions have not been included.

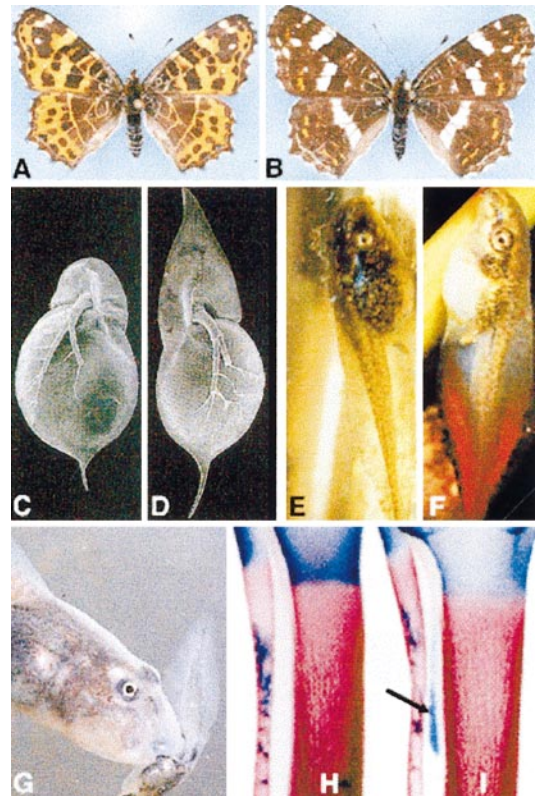


FIG. 1. Instructive induction of morphological phenotypes by the environment. (A, B) The spring (A) and summer heat-induced (B) morph of the European map butterfly, *Araschnia levana*. (C, D) The uninduced (C) and *Chaoborus* kairomone-induced (D) morphs of *Daphnia cucullata*. (E, F) The uninduced (E) and kairomone-induced (F) morphs of the tadpole of the grey treefrog *Hyla cryoscelis*. (G) *Scaphiopus* tadpoles, the uninduced morph in the jaws of the density-induced morph. (H, I) Uninduced (H) and movement-induced (I) tissue in the embryonic chick hindlimb. The arrow points to the movement-induced fibular crest, an important bone in bird evolution. (A and B, courtesy of H. F. Nijhout; C and D, courtesy of R. Tollrian; E and F, courtesy of J. Van Buskirk; G, courtesy of T. Wiewandt; H and I, courtesy of G. Müller.)

tion in *Bonellia* and in insect hives (see Hertwig, 1894). This first generation of experimental embryologists also investigated the effects of ion or nutrient deprivation on morphogenesis (Selenka, 1876; Born, 1881; Herbst, 1893). Thus, by the turn of the last century, embryologists already knew about temperature-dependent polyphenisms (and their adaptive significance), context-dependent sex determination, and environmental teratogenesis.

However, in the early part of the 20th century, the physiological paradigm overtook the ecological one, and experimental embryology became *Entwicklungsmechanik*, the causal physiology of development. This approach directed the focus of attention on events occurring within the embryo rather than on how the environmental milieu

determined the embryo. The second upheaval came in the 1960s, when the paradigms of molecular genetics overtook those of physiology (Gilbert, 1996; Keller, 1995). One no longer studied the “whole organism.” Rather, differential gene expression became the dominant model. Neither of the earlier traditions entirely died. However, the practitioners of ecological developmental biology found themselves widely scattered into various disciplinary areas that did not communicate with one another. Meanwhile, developmental biology lost the concept of ecological regulation, and with the single exception of Waddington’s *Principles of Embryology* (1956a), the concept was absent from development textbooks² until the 1980s.

Developmental biology has matured enormously in the past decade, and it should now return to some of these older questions of environment, which have become increasingly important. Conservation biology needs to know about the survival and development of the embryonic and larval stages of development as much as it does about the adult stage (see, for example, Morreale *et al.*, 1982). Environmental chemicals that we had thought harmless (at least to adults) may be dangerous to developing organisms and may threaten the fertility of adults (Colburn *et al.*, 1996). Developmental biology can also aid ecology by examining the proximate causes for life history strategies. While ecologists have known for decades about developmental plasticity, developmental biologists have only recently begun to address the molecular mechanisms for these phenomena.

In addition to being critical for ecology and conservation biology, ecological developmental biology can enrich contemporary developmental biology by providing a wealth of new opportunities for research. Contemporary developmental biology has focused on six animal model species, all of which have converged on the same developmental phenotype. Each of our model systems for developmental biology—the frog *Xenopus laevis*, the nematode *Caenorhabditis elegans*, the fly *Drosophila melanogaster*, the chick *Gallus gallus*, the mouse *Mus musculus*, and the zebrafish *Danio rerio*—has been selected for small body size, large litter size, rapid embryonic development, early sexual maturation, the immediate separation of the germline from somatic lines, and the ability to develop within the laboratory (Buss, 1987; Bolker, 1995; Bolker and Raff, 1997). The last two criteria are very important because they eliminate the effects of the environment on development. While the model systems have two enormous advantages—

² The possible ecological aspect of developmental biology was also mentioned in the illustrations of some of Paul Weiss’ reviews (see, for example, Weiss, 1950). But for Weiss, ecology was more of a metaphor for the set of changing interactions needed to cause morphogenesis. While the concept of environmental regulation of gene expression declined in developmental biology, Waddington’s and Weiss’ use of this concept was (and is) continued by behavioral biologists concerned with environmental factors and learning (see Gottlieb, 1992) and by a school of philosophers of science called developmental systems theorists (see Oyama *et al.*, 2000).

they allow one to compare research from different areas of the world and they enable genetic regulation to be studied without any major variability coming from the environment—these animals have been selected for their suitability to the genetic paradigm of developmental biology. The shortcoming of our model systems is that they do not represent development in the real world. Most organisms probably will not develop well in the laboratory. There are environmental cues that regulate and permit development to occur.

This paper will explore some of the relationships between development and ecology. It will concentrate almost exclusively on animal development, for there is already an extensive and well-reviewed literature on environmental regulation of plant development (see, for example, West-Eberhard, 1989; Sultan, 1995; Dudley and Schmitt, 1996; Pigliucci *et al.*, 1996; Pigliucci *et al.*, 1998; Agrawal, 1998).

CONCEPTS OF ECOLOGICAL DEVELOPMENTAL BIOLOGY

Developmental plasticity (sometimes called phenotypic plasticity) is the notion that the genome enables the organism to produce a range of phenotypes. There is not a single phenotype produced by a particular genotype. The structural phenotype instructed by the environmental stimulation is referred to as a morph.³ When developmental plasticity manifests itself as a continuous spectrum of phenotypes expressed by a single genotype across a range of environmental conditions, this spectrum is called the norm of reaction (or reaction norm; Woltereck, 1909; Schmalhausen, 1949; Stearns *et al.*, 1991; Schlichling and Pigliucci, 1998). The reaction norm is thought to be a property of the genome and can also be selected. Different genotypes will be expected to differ in the direction and amount of plasticity that they are able to express (Gotthard and Nylin, 1995; Via *et al.*, 1995).

A related form of developmental plasticity, polyphenism,

³ Historically, geneticists have generally tried to avoid systems that have developmental plasticity and they tend to dismiss it as “developmental noise.” Phenotypic plasticity interferes with the ability to say that a particular allele is associated with a particular phenotype, and it introduces terms such as “incomplete penetrance” and “expressivity” into their analyses. “Penetrance” (the proportion of individuals carrying the mutant gene who express the mutant phenotype) and “expressivity” (the extent of a mutation’s phenotypic manifestation within an individual) were coined by Oskar Vogt (1926). Rather than accept that genotype did not deterministically produce phenotypes, he gave these new properties to the genes (Sarkar, 1999). The major proponent of norms of reaction among geneticists was Theodosius Dobzhansky. While a morph is the term usually given to alternate anatomical phenotypes instructed by the environment, there are often alternative behavioral phenotypes associated with these morphs (see Relyea, 2000c). These presumably have an anatomical basis, as well, but they are studied as alternative behavioral patterns.

refers to the occurrence in a single population of discontinuous (“either/or”) phenotypes elicited by the environment from a single genotype (Mayr, 1963). The example mentioned earlier is a temperature-dependent polyphenism in which the European map butterfly, *Araschnia levana*, has two seasonal phenotypes so different that Linnaeus classified them as two different species (Weele, 1999). The spring morph is bright orange with black spots, while the summer form is mostly black with a white band (Figs. 1A and 1B). The change from spring to summer morph is controlled by changes in both day length and temperature during the larval period. When researchers experimentally mimic these conditions, the summer caterpillars can give rise to “spring” butterflies (Weismann, 1875; Nijhout, 1991). In addition to temperature-dependent polyphenisms, there are polyphenisms based on nutrition, population density, or the presence of a predator.

One of the most interesting aspects of eco-devo concerns these predator-induced polyphenisms. To demonstrate predator-induced polyphenisms, one has to show that the phenotypic change is caused by the predator (usually from kairomones, soluble chemicals released by the predator). It has often been shown that the induced phenotypic modification increases the fitness of its bearers when the predator is present. For instance, juvenile *Daphnia* and other invertebrate species will alter their morphology when they develop in pond water in which their predators have been cultured. The water in which the predatory larvae of the dipteran *Chaoborus* have been cultured can induce a “neck spine” or a “helmet” during *Daphnia* development. These allow the *Daphnia* to escape from their predator more effectively (Figs. 1C and 1D). The induced *Daphnia* suffer lower mortality from these predators (Tollrian and Dodson, 1999; Agrawal *et al.*, 1999). This induction is even transferred to the parthenogenetic offspring of these *Daphnia*. Those *Daphnia* whose mothers had been exposed to predation cues were born with large helmets, even if the mothers had been transferred to water that lacked the caged predators. Thus, progeny born in a precarious environment (i.e., an environment where the kairomone concentration is high enough to induce helmet growth in their mothers) are thereby born with a defense against predation.

Another concept associated with ecological developmental biology is context dependency. Since phenotype is not predictable from the genotype, it depends upon the context in which the organism is developing (Gilbert and Sarkar, 2000). At one temperature, the snapping turtle embryo becomes male; at another temperature it becomes female. Fed one diet, a female ant larva becomes a sterile worker; fed another diet, the same larva becomes an enormous fertile queen. This introduces the concept of the reactive genome, an idea of C. H. Waddington (1956a). The genome not only acts, but it reacts. It responds to the environment by changing its expression patterns. This, of course, has been known since the discovery of the lactose-inducible operon, but the *lac* operon has rarely been associated with the environmental regulation of gene expression. The abil-

ity of the genome to respond to environmental inducers has been termed tertiary induction (Gilbert, 2000) to emphasize that while secondary induction refers to the interactions of competent cells to inducers within the embryo, the same heuristic can be applied to inducers from outside the embryo.

The relationship of eco-devo to evo-devo is something that needs to be studied intensively. Two principles, though, have already been seen as being very important. One concerns trade-offs. In a predator-induced polyphenism, the induced phenotype can better survive the predator, but the phenotype may be less adaptive in other ways. For instance, the carp *Carassius carassius* is able to respond to the presence of a predatory pike if the pike has already eaten a carp. The induced carp grows into a pot-bellied, hunched-back morph that will not fit into the pike’s jaws. However, the induced morphology results in increased drag when swimming and consequently the fatter fish cannot swim as efficiently (Brönmark and Petterson, 1994). In *Daphnia*, the production of helmets appears to lessen the amount of resources that can provision eggs (Riessen *et al.*, 1984; 1992). If the induced phenotype not only were more successful in avoiding predators but also had no significant trade-offs, one might expect that it would become the dominant morph of the population. For this to happen, the more fit phenotype would have to be formed even in the absence of the environmental inducer. In other words, the same phenotype would be induced by internal rather than external factors. This replacement of external inducers by internal inducers has been called genetic assimilation (Waddington, 1942; 1953; 1956b; see Schmalhausen, 1949). Recent molecular evidence (Gibson and Hogness, 1996; Rutherford and Lindquist, 1998) has shown that given selection for the trait, such genetic assimilation of environmentally induced phenotypes occurs readily in the laboratory. As Waddington emphasized, this is in no way Lamarckian. Rather, by orthodox Darwinism and orthodox embryology, one could explain evolutionary phenomena (such as the appearance of calluses on ostrich’s skin before they are born) that *appear* Lamarckian.

HOW ECO-DEVO CAN ENRICH DEVELOPMENTAL BIOLOGY: THE AMPHIBIAN EXAMPLE

As an example of how ecological developmental biology can enrich our discipline, I will look at an area that developmental biologists know well: amphibian development. [Another reason to look specifically at amphibian ecological developmental biology is that an excellent account of environmental regulation during insect development has recently been published by Nijhout (1999).] The standard account of amphibian embryology is a detailed and exciting scientific story starting with Spemann and his laboratory. It shows how the major axes of the frog can be traced from fertilization through gastrulation, how the

germ layers become specified, how organs form, and how metamorphosis is achieved. It is a standard paradigm for introducing experimental techniques, conceptual frameworks, and how our science has progressed from tissue transplantation to gene manipulation, from “organizer molecules” to specific paracrine factors and their regulators. It’s a story internal to the embryo. The frog egg can develop perfectly well in a jar of pond water. All that’s needed for *Xenopus* to develop appears to reside inside the embryo.

But *Xenopus* is hardly one’s representative amphibian. Indeed, *Xenopus* became the frog king precisely because it was so well buffered from environmental cues. The fact that *Xenopus* gamete production and mating are not confined to a particular breeding season made it possible for investigators to have amphibian embryos at any time of year (Gurdon and Hopwood, 2000). This is a rare and useful situation and allowed *Xenopus* to displace *Rana*, *Triturus*, and even *Ambystoma* as the model amphibian for embryological studies. If we go into the field and look at development of several amphibian species in the wild, we get a rich picture of environmental and genomic interactions. Moreover, these interactions may be critical in the scientific evaluation of agricultural and industrial policies.

Context-Dependent Development: Predator-Induced Polyphenisms

Predator-induced polyphenism is abundant among amphibia, and tadpoles found in ponds or in the presence of other species may differ significantly from those tadpoles reared by themselves in aquaria. For instance, when newly hatched wood frog (*Rana sylvetica*) tadpoles are reared in tanks containing the predatory larval dragonfly, *Anax* (confined in mesh cages so that they cannot kill the tadpoles), the tadpoles in the predator-filled tanks grow smaller than those in similar tanks without the caged predators. Moreover, their tail musculature deepens, allowing faster turning and swimming speeds to escape predator strikes (McCollum and Leimberger, 1997; Van Buskirk and Relyea, 1998). In fact, what initially appeared to be a polyphenism may be a reaction norm that can assess the number (and type) of predators. The addition of more predators to the tanks causes a continuously deeper tail fin and tail musculature.

The tadpoles of related species produce different phenotypic changes, depending on the predator. The tadpole of the gray treefrog (*Hyla cryoscelis*) responds to predator kairomones both by size change and by developing a bright red tail coloration that deflects predators (Figs. 1E and 1F; Relyea and Werner, 2000a; McCollum and Van Buskirk, 1996). The trade-off is that the noninduced tadpoles grow more slowly and survive better in predator-free environments (Van Buskirk and Relyea, 1998; Relyea, 2000b).

But the story doesn’t end here. In addition to responding to cues from predators, *Rana* tadpoles also respond to cues from competitors. Wood frog and leopard frog tadpoles compete for the same food. The presence of the leopard frog

tadpoles changes the responses of the wood frog tadpoles to predator-derived cues (Relyea, 2000c). In some instances, the competitor-induced phenotypes go in opposite directions than the predator-induced phenotypes (making shallower tails, for instance). In these cases, the competitor-induced phenotypes are more competitive (against other organisms competing for the same food source), but they suffer a higher predation.

While these predator-induced changes appear ubiquitous, they develop slowly and are quantitative traits. However, immediate and qualitative predator-induced developmental changes can also be found in amphibians. The developing stages of the red-eyed tree frog *Agalychnis callidryas* are exposed to two types of predators. The eggs are attached to vegetation overhanging ponds. The embryos of these arboreal eggs are prone to predation by wasps and snakes. The tadpoles hatch from these eggs and fall into the water. There, they can be eaten by predatory fish and shrimp. When the eggs are attacked by snakes, the embryos sense the presence of the snake and vigorously shake in their egg cases. Within seconds, the embryos (having achieved gill circulation) hatch prematurely into the water. Embryos that do not hatch quickly are eaten. While they have escaped the terrestrial predator, the trade-off is that, once in the water, these earlier stages are at higher risk for being eaten by the aquatic predators (Warkentin, 1995, 2000).

Context-Dependent Development: Abiotic Conditions

The spadefoot toad *Scaphiopus* has a remarkable strategy for coping with a particularly harsh environment. The toads are called out from hibernation by the thunder that accompanies the first spring storm in the Sonoran desert. (Unfortunately, motorcycles produce the same sounds, causing these toads to come out from hibernation and die in the scorching Arizona sunlight.) The toads breed in the temporary ponds caused by the rain, and the embryos develop quickly into larvae. After the larvae metamorphose, the young toads return to the desert, burrowing into the sand until the next year’s storms bring them out.

The desert ponds are ephemeral pools that either dry up quickly or persist, depending on the initial depth and the frequency of the rainfall. One might envision only two alternative scenarios confronting a tadpole in such a pond: either (1) the pond persists until it has time to metamorphose and it lives or (2) the pond dries up before metamorphosis, and it dies. These toads (and several other amphibians), however, have evolved a third alternative. The time of metamorphosis is controlled by the pond. If the pond does not dry out, development continues at its normal rate, and the alga-eating tadpoles eventually develop into juvenile spadefoot toads. However, if the pond is drying, overcrowding occurs, and some of the tadpoles embark on an alternative developmental pathway. They develop a wider mouth and more powerful jaw muscles which enable them to eat,

among other things, other *Scaphiopus* tadpoles (Fig. 1G). These carnivorous tadpoles metamorphose quickly, albeit into a smaller version of the juvenile spadefoot toad.

The signal for this accelerated metamorphosis appears to be the change in water volume. *Scaphiopus* tadpoles are able to sense the removal of water from aquaria, and their acceleration of metamorphosis depends upon the rate at which the water was removed. The stress-induced corticotropin-releasing hormone signaling system appears to modulate this effect (Denver *et al.*, 1998; Denver, 1999). The two morphs can be obtained by feeding tadpoles the appropriate diets, and the rapid development of the cannibalistic tadpoles may be being driven by the thyroxine they acquire from their prey. While the large cannibalistic morphs survive under these conditions, and the other *Scaphiopus* tadpoles perish from desiccation or ingestion by their pond-mates, there is a trade-off. The trade-off is that the toads generated by fast-metamorphosing tadpoles lack the fat reserves of those toads produced from the more slowly growing tadpoles, and their survival rate after metamorphosis is not as high as that of those toads developing from slower growing larvae (Newman, 1989, 1992; Pfennig, 1992a,b).

Cannibalistic larvae are also seen among Ambystomid salamanders (see Collins and Pfennig, 1993). These salamanders also have larval morphs that are controlled by the environment, especially the temperature of the water in which they swim (Wakahara, 1996; Safi *et al.*, 1997). Environmental context plays a very significant role in producing the phenotype of many amphibians.

Teratogenesis

Teratogenesis forms an important area for the interaction of environment and development (Gilbert, 2000; Stocum, 2000). For amphibian embryos, three important teratogens appear to be ultraviolet radiation, pesticides, and trematodes. Many eggs and early embryos lie in direct sunlight. This radiation is harmful to DNA, and eggs of several species survive the ultraviolet radiation by having evolved natural sunscreens. The eggs of many marine organisms possess high concentrations of mycosporine amino acid pigments that absorb UV-B. Moreover, just like our melanin pigment, these pigments can be induced by exposure to ultraviolet (UV-B) irradiation (Jokiel and York, 1982; Siebeck, 1988; Adams and Shick, 1996). Many frog eggs have evolved high levels of DNA repair enzymes. Blaustein and colleagues (1994) found that levels of the UV-damage-specific repair enzyme photolyase (which excises and replaces the UV-damaged thymidine residues) vary 80-fold between the eggs of the tested amphibian species and are correlated with the site of egg laying. Those eggs more exposed to the sun had higher levels of photolyase. Interestingly, the highest photolyase levels were in species such as the Pacific treefrog (*Hyla regilla*) whose populations were not seen to be in decline. The lowest levels were seen in those species (such as the Western toad, *Bufo boreas*, and

the Cascades frog, *Rana cascadae*) whose populations had dramatically declined. Filtering the UV light raises the percentage of hatched tadpoles from these eggs.

Pesticides may also present developmental problems that are exacerbated by environmental factors. Throughout the United States and southern Canada, there has been a dramatic increase in the number of deformed frogs and salamanders in what seem to be pristine woodland ponds (Oulette *et al.*, 1997; Burkhart *et al.*, 2000; Meteyer *et al.*, 2000). These deformities include extra or missing limbs, missing or misplaced eyes, deformed jaws, and malformed hearts and guts. Water from these lakes can cause malformations in laboratory-reared *Xenopus* (Burkhart *et al.*, 1998). It is not known what is causing these disruptions, but there is speculation (see Hilleman, 1996; Oulette *et al.*, 1997; Gilbert, 2000) that pesticides (sprayed for mosquito and tick control) might be interfering with normal amphibian development. The abnormalities seen in these frogs resemble those malformations caused by exposing tadpoles to known teratogens such as retinoic acid (Crawford and Vincenti, 1998; Gardiner and Hoppe, 1999).

New research has shown that some pesticides may be relatively harmless to amphibians developing in the laboratory but potentially devastating to the same developing amphibians in the wild. One such compound may be methoprene, a juvenile hormone mimic that inhibits mosquito pupae from metamorphosing into adults. Since vertebrates do not have juvenile hormone, it was assumed that this pesticide would not harm fish, amphibians, or humans. This has been found to be the case: methoprene, itself, does not have teratogenic properties. However, upon exposure to sunlight, methoprene breaks down into two products that have significant teratogenic activity in frogs. These compounds have a structure similar to that of retinoic acid and will bind to the retinoid receptor (Harmon *et al.*, 1995; La Claire *et al.*, 1998). When *Xenopus* eggs are incubated in water containing these compounds, the tadpoles are often malformed, and show a spectrum of deformities similar to those seen in the wild (La Claire *et al.*, 1998). Another pesticide, carbaryl, may also be dangerous to amphibian populations in the wild but not in laboratory tests. It appears that very low concentrations of this compound usually do not affect tadpoles under the 4-day exposure conditions used in the laboratory. However, in ponds, this compound may become concentrated and the exposure time longer. Gray treefrog (*Hyla versicolor*) tadpoles exposed 4 days to a concentration of carbaryl that was only 4% the LD₅₀ almost always survived. However, if exposure was for 10–16 days, 10–60% of the tadpoles died (Relyea and Mills, 2001).

Moreover, if the tadpoles are exposed to these low doses in the presence of predatory kairomones, the “low” concentration of carbaryl actually kills 60–98% of tadpoles. Thus, Relyea and Mills (2001) conclude that, under the more realistic conditions of increased exposure times and predatory stress, the current application protocols for carbaryl use could devastate gray treefrog populations. Moreover,

since predator-induced stress appears to be very widespread among amphibians, and carbaryl's mode of action is common to many pesticides, the use of pesticides once thought relatively harmless might be causing widespread devastation of amphibian populations.

Trematode parasites are also a source of developmental anomalies in amphibians. Trematode eggs can divide tadpole limb fields just like a foil barrier and thereby create frogs with supernumerary limbs or autopods (Sessions and Ruth, 1990; Johnson *et al.*, 1999). Interestingly, several investigators (Sato, 1961; Eguchi, 1980; Okada, 2000) have speculated that Wolffian regeneration (of lens from the dorsal iris) evolved as an adaptation to trematode parasitism of the lens cortex. Catching tadpoles in the wild, these investigators documented both lens parasitism and the formation of new lenses in those amphibians and fish that have evolved Wolffian regeneration. T. S. Okada (2000) explicitly states that: "These extraordinary facts are the first suggestion of lens regeneration as a kind of adaptive phenomenon. Taking a broader view, this is one of first examples indicating the interaction of two different species in controlling the development and differentiation of a particular organ system, and thus implies a link between the disciplines of developmental biology (embryology) and ecology."

HUMAN NORMS OF REACTION

Nutrition and Exercise

Of all the animals, humans may have the largest norms of reaction. Our examples of developmental plasticity are so prevalent that we have given their study different names: nutrition, immunology, neurobiology, and perhaps even orthodontic dentistry. Were I to reside long in the Andes or Alps, I would develop red blood cells more efficiently, due to a developmental system that produces more erythropoietin in response to oxygen deprivation (Brunn *et al.*, 1998; Wenger *et al.*, 1998). Our bones and our muscles are remodeled through exercise and diet throughout development. If we were prevented from receiving sunlight as children, our etiolated bodies would succumb to rickets, the deficiency of activated vitamin D. Vitamin D is activated when the skin is exposed to sunlight, and its active form is needed to initiate the transcription of those genes whose protein products regulate intestinal calcium absorption and the levels of calcium and phosphate in our bones. As adults, a lack of calcium or vitamin D in our diet can cause osteoporosis, a disease that is determined both by genetic parameters and by "lifestyle" (see Jones *et al.*, 1998; Cymet *et al.*, 2000). Bone density is also regulated by mechanical stress, and several genes for osteoblast and osteocyte functions are known to be regulated through physical load (Nomura and Takano-Yamamoto, 2000; Ogasawara *et al.*, 2000; Zaman *et al.*, 2000). Astronauts experiencing weightlessness are at risk for such negative bone remodeling (losing about 1% of heel bone mineral density

per month in space), and studies on the space shuttles have shown that several genes, including the gene for the vitamin D receptor, are dramatically downregulated in microgravity (Hammond *et al.*, 2000; Wassersug, 2000). Muscle development is another obvious place where environment plays a role in human phenotype production, and were I to exercise daily, I could no doubt hypertrophy my pectoral, abdominal, and biceps muscles into strong and sculptured wonders.

One of the most critical cases of the dietary regulating of our phenotype involves the disease gulonolactone oxidase deficiency (hypoascorbemia; OMIM 240400). Homozygosity for a mutation in the gulonolactone oxidase gene on the short arm of chromosome 8 produces a syndrome that produces death in childhood due to poor collagen crosslinking. Interestingly, this lethal syndrome affects 100% of the human population. Gulonolactone oxidase is the final enzyme in the pathway leading to ascorbic acid, and we are all homozygous at this mutant locus (Nishikimi *et al.*, 1994). While most mammals have this enzyme and can synthesize vitamin C, our genes for this enzyme are mutated, and we cannot make this necessary compound. This is why we need ascorbic acid (vitamin C) in our diet. Another name for this disease is scurvy. Without this replacement therapy from the environment, we are all dead.

Immunology

Humans have specific predator-induced developmental plasticity on a scale unimaginable in invertebrates. Our major predators, of course, are microbes. We respond to them through an immune system based on the clonal selection of lymphocytes that recognize specific predators and their products. Our immune system recognizes a particular microbe such as a cholera bacterium or a poliovirus by expanding precisely those lymphocytes that can defend the body against them. When a B-cell binds its foreign substance (the antigen), it enters a pathway that causes that B-cell to divide repeatedly and to differentiate into an antibody-secreting cell that secretes the same antibody that originally bound the antigen. Moreover, some of the descendants of that stimulated B-cell remain in the body as sentinels against further infection by the same microorganism. Thus, identical twins are not identical with respect to their immune systems. Their phenotypes (in this case, the lymphocytes in their lymph nodes and their ability to respond against an infectious microorganism) have been altered by the environment. Moreover, our immune system also provides transgenerational immunity against common predators. The IgG antibodies produced by our mothers during pregnancy can cross the placenta and give us passive immunity when we are born. In birds, a similar antibody is placed into the eggs. The cells of our respective immune systems are not specified solely by our genetic endowment. (Even the genes for the antibodies and T-cell receptors aren't present in the zygote.) Rather, experience is added to

endowment. The environment, in this case, antigens, directs the development of our lymphocytes.

Neurobiology

Our immune system responds by monitoring our environment and changing with it. Similarly, our other sensory network, the nervous system, also changes with development. Competitive synapse formation based on experience (in both neuromuscular junctions and the optic nerve pathways) is crucial to the formation and maintenance of our nervous system (Hubel, 1967; Kennedy *et al.*, 1981). Neuronal plasticity is seen in many animals, and it is often associated with learning and/or memory (for instance, Rasika *et al.*, 1999; Tramontin *et al.*, 2000). Although difficult to study in humans, neuronal plasticity becomes manifest in some clinical situations. One such example is the “phantom limb phenomenon” (Flor *et al.*, 1995; Davis *et al.*, 1998; Montoya *et al.*, 1998), wherein the cortex becomes reorganized in a manner that sensory inputs from the stump of the amputated arm triggers neurological activity in the thalamic region that had formerly been innervated by neurons from the missing arm. Another case may be epilepsy, where reduced synaptic plasticity has been correlated with declarative verbal memory dysfunction (Beck *et al.*, 2000). Neural plasticity allows our muscles to perform properly when they receive sensory signals to contract, permits us to see in three dimensions, and enables us to learn and to recall experiences. These are critical portions of the human phenotype. As Purves and Lichtman (1985) concluded: “The interaction of individual animals and their world continues to shape the nervous system throughout life in ways that could never have been programmed. Modification of the nervous system by experience is thus the last and most subtle developmental strategy.”

Orthodontics

Physical stress is needed to produce bones such as the mammalian patella and the bird fibular crest (Figs. 1H and 1I; Müller and Streicher, 1986; Wu, 1996). Corruccini (1984) and Varrela (1992) have speculated that the reason nearly a quarter of our population needs orthodontic appliances is that our lower jaw has failed to develop normally. Such jaw anomalies (malocclusions wherein the teeth cannot fit properly in the jaw) are relatively new to European populations. Well-preserved skeletons from the 15th and 16th centuries show almost no malocclusion in the population. Instead, severe grinding is seen (Mohlin *et al.*, 1978; Helm and Prysödö, 1979; Corruccini, 1984; Varrela, 1990). The most popular explanation (see Proffit and Fields, 1993) for the increased rate of malocclusion in modern populations is that the current high incidence of malocclusion is due to increased migration. Genetically specified differences in jaw size do not match with genetically defined differences in tooth size. However, this purely genetic model does not

explain the increase in malocclusions seen in stable populations such as those in central Finland or southern Sweden. Corruccini and Varrela have hypothesized that the change in children’s meals from a coarse diet to a mild textured diet has resulted in decreased mastication and a decrease in jaw skeleton and muscle development. Increased chewing causes tension that stimulates mandible bone and muscle growth (Kiliardis, 1986; Weijs and Hillen, 1986), and placing young primates on a soft diet will cause malocclusions in their jaws, similar to those in humans (Corruccini and Beecher, 1982, 1984).

CONCLUSION

The environment is not merely a permissive factor in development. It can also be instructive. A particular environment can elicit different phenotypes from the same genotype. Development usually occurs in a rich environmental milieu, and most animals are sensitive to environmental cues. The environment may determine sexual phenotype, induce remarkable structural and chemical adaptations according to the season, induce specific morphological changes that allow an individual to escape predation, and induce caste determination in insects. The environment can also alter the structure of our neurons and the specificity of our immunocompetent cells. We can give a definite answer to the question posed by Wolpert in 1994:

Will the egg be computable? That is, given a total description of the fertilized egg—the total DNA sequence and the location of all proteins and RNA—could one predict how the embryo will develop?

The answer has to be “No. And thank goodness.” The phenotype depends to a significant degree on the environment, and this is a necessary condition for integrating the developing organism into its particular habitat.

Unfortunately, the environment can also be the source of chemicals that disrupt normal developmental processes. If only a fraction of what books such as *Our Stolen Future* (Colburn *et al.*, 1996) are saying is true, then developmental biologists are going to have to go to the forefront of conservation science. Ecological developmental biology must become a critical part of normative developmental biology if we are to base agricultural and industrial policies on scientifically accurate data.

As we become aware of the complexity of development, we are realizing that development is critically keyed to the environment. Ecologists have known about “life history strategies” of organisms for over a century. However, the proximate causes of these histories (such as how a fish becomes male in one environment and female in another) are just beginning to be understood. Progress in kairomone isolation is just starting, and the genes responsive to kairomones and larval settlement cues remain to be isolated (see Okazaki and Shizuri, 2000). There is important research to be done, and whole new worlds for developmental biology

to explore. "Eco-devo" can also play a balancing role in "evo-devo." Most discussions of evolutionary developmental biology have focused on the phylogenetic, nonadaptive, and macroevolutionary parts of evo-devo (e.g., the *Hox*, *Pax*, and *Distal-less* gene families and the origin of phyla, classes, and orders). Eco-devo would complement this, focusing upon the ecological, adaptive, and microevolutionary aspects of evolutionary developmental biology. Van Valen (1973) claimed that evolution can be defined as "the control of development by ecology." We are at the point where we can give some specific instances and mechanisms of where and how this happens.

ACKNOWLEDGMENTS

I thank the scientists and photographers who let me use their photographs in the figure. I also thank J. Bolker, A. Agrawal, and the anonymous reviewers for their suggestions. Support came from a faculty research grant from Swarthmore College and from National Science Foundation Grant IBN-0079341.

REFERENCES

- Adams, N. L., and Shick, J. M. (1996). Mycosporine-like amino acids provide protection against ultraviolet radiation in eggs of the green sea urchin *Strongylocentrotus doebachiensis*. *Photobiochem. Photobiol.* **64**, 149–158.
- Agrawal, A. A. (1998). Induced responses to herbivory and increased plant performance. *Science* **279**, 1201–1202.
- Agrawal A.A., Laforsch, C., and Tollrian, R. (1999). Transgenerational induction of defenses in animals and plants. *Nature* **401**, 60–63.
- Beck, H., Goussakov, I. V., Lie, A., Helmstaedter, C., and Elger, C. E. (2000). Synaptic plasticity in the human dentate gyrus. *J. Neurosci.* **20**, 7080–7086.
- Blaustein, A. R., Hoffman, P. D., Hokit, D. G., Kiesecker, J. M., Walls, S. C., and Hays, J. B. (1994). UV repair and resistance to solar UV-B in amphibian eggs: A link to population declines? *Proc. Natl. Acad. Sci. USA* **91**, 1791–1795.
- Bolker, J. A. (1995). Model systems in developmental biology. *BioEssays* **17**, 451–455.
- Bolker, J. A., and R. A. Raff. (1997). Beyond worms, flies and mice: It's time to widen the scope of developmental biology. *J. NIH Res.* **9**, 35–39.
- Born, G. (1881). Experimentelle untersuchungen über die entstehung der geschlechtsunterschiede. *Jahres-bericht d. Schlesischen gesell. f. väterländ. Culture* **21**, 2–23.
- Bridges, C. M. (2000). Long-term effects of pesticide exposure at various life stages of the southern leopard frog (*Rana sphenoccephala*). *Arch. Environ. Contam. Toxicol.* **39**, 91–96.
- Brönmark, C., and Pettersson, L. (1994). Chemical cues from piscivores induce a change in morphology in crucian carp. *Oikos* **70**, 396–402.
- Brunn, H. F., Gu, J., Huang, L. E., Park, J. W., and Zhu, H. (1998). Erythropoietin: A model system for studying oxygen-dependent gene regulation. *J. Exp. Biol.* **201**, 1197–1201.
- Burkhart, J. G., Helgen, J. C., Fort, D. J., Gallagher, K., Bowers, D., Propst, T. L., Gernes, M., Magner, J., Shelby, M. D., and Lucier, G. (1998). Induction of mortality and malformation in *Xenopus laevis* embryos by water sources associated with field frog deformities. *Environ. Health Perspect.* **106**, 841–848.
- Burkhart, J. G., Ankley, G., Bell, H., Carpenter, H., Fort, D., Gardiner, D., Gardner, H., Hale, R., Helgen, J. C., Jepson, P., et al. (2000). Strategies for assessing the implications of malformed frogs for environmental health. *Environ. Health Perspect.* **108**, 83–90.
- Buss, L. (1987). "The Evolution of Individuality." Princeton Univ. Press, Princeton, NJ.
- Colburn, T., Dumanoski, D., and Myers, J. P. (1996). "Our Stolen Future." Dutton, New York.
- Corruccini, R. S. (1984). An epidemiologic transition in dental occlusion in world populations. *Am. J. Orthod.* **86**, 419–426.
- Corruccini, R. S., and Beecher, C. L. (1982). Occlusal variation related to soft diet in a nonhuman primate. *Science* **218**, 74–76.
- Corruccini, R. S., and Beecher, C. L. (1984). Occlusofacial morphological integration lowered in baboons raised on soft diet. *J. Craniofac. Genet. Dev. Biol.* **4**, 135–142.
- Crawford, K., and Vincenti, D. M. (1998). Retinoic acid and thyroid hormone may function through similar and competitive pathways in regenerating axolotls. *J. Exp. Zool.* **282**, 724–738.
- Cymet, T. C., Wood, B., and Orbach, N. (2000). Osteoporosis. *J. Am. Osteopath. Assoc.* **100** (Suppl.), S9–S15.
- Davis, K. D., Kiss, Z. H., Luo, L., Tasker, R. R., Lozano, A. M., and Dostrovsky, J. O. (1998). Phantom sensations generated by thalamic microstimulation. *Nature* **391**, 385–387.
- Denver, R. J. (1999). Evolution of the corticotropin-releasing hormone signaling system and its role in stress-induced phenotypic plasticity. *Neuropeptides: Structure and function in biology and behavior. Ann. N. Y. Acad. Sci.* **897**, 46–53.
- Denver, R. J., Mirhadi, N., and Phillips, M. (1998). Adaptive plasticity in amphibian metamorphosis: Response of *Scaphiopus hammondi* tadpoles to habitat desiccation. *Ecology* **79**, 1859–1872.
- Dudley, S. A., and Schmitt, J. (1996). Testing the adaptive plasticity hypothesis: Density-dependent selection on manipulated stem length in *Impatiens capensis*. *Am. Nat.* **147**, 445–465.
- Eguchi, G. (1980). "Regeneration of Lens: A Switch in Differentiation of Tissue Cells." Iwanami Shoten, Tokyo. (In Japanese).
- Flor, H., Elbert, T., Knecht, S., Wienbruch, C., Pantev, C., Birbaumer, N., Larbig, W., and Taub E. (1995). Phantom-limb pain as a perceptual correlate of cortical reorganization following arm amputation. *Nature* **375**, 482–484.
- Gardiner, D. M., and Hoppe, D. M. (1999). Environmentally induced limb malformations in mink frogs (*Rana septentrionalis*). *J. Exp. Zool.* **284**, 207–216.
- Gibson, G., and Hogness, D. S. (1996). Effect of polymorphism in the *Drosophila* regulatory gene *Ultrabithorax* on homeotic stability. *Science* **271**, 200–203.
- Gilbert, S. F. (1996). Enzyme adaptation and the entrance of molecular biology into embryology. In "The Philosophy and History of Molecular Biology: New Perspectives" (S. Sarkar, Ed.), pp. 101–123. Kluwer Academic, Dordrecht.
- Gilbert, S. F. (2000). "Developmental Biology," 6th ed. Sinauer, Sunderland, MA.
- Gilbert, S. F., and Sarkar, S. (2000). Embracing complexity: Organicism for the 21st century. *Dev. Dyn.* **219**, 1–9.
- Gotthard, K., and Nylin, S. (1995). Adaptive plasticity and plasticity as an adaptation: A selective review of plasticity in animal morphology and life history. *Oikos* **74**, 3–17.

- Gottlieb, G. (1992). "Individual Development and Evolution: The Genesis of Novel Behavior." Oxford Univ. Press, New York.
- Gurdon, J. B., and Hopwood, N. (2000). The introduction of *Xenopus laevis* into developmental biology, of empire, pregnancy testing, and ribosomal genes. *Int. J. Dev. Biol.* **44**, 43–50.
- Hammond, T. G., Benes, E., O'Reilly, K. C., Wolf, D. A., Linnehan, R. M., Taher, A., Kaysen, J. H., Allen, P. L., and Goodwin, T. J. (2000). Mechanical culture conditions effect gene expression: Gravity-induced changes on the space shuttle. *Physiol. Genomics* **3**, 163–173.
- Harmon, M. A., Boehm, M. F., Heyman, R. A., and Mangelsdorf, D. J. (1995). Activation of mammalian retinoid-X receptors by the insect growth regulator methoprene. *Proc. Natl. Acad. Sci. USA* **92**, 6157–6160.
- Helm, S., and Prysödö, U. (1979). Prevalence of malocclusion in medieval and modern Danes contrasted. *Scand. J. Dental Res.* **87**, 91–97.
- Herbst, C. (1893). Experimentelle Untersuchungen über den Einfluss der veränderten chemischen Zusammensetzung des umgebenden Mediums auf die Entwicklung der Thiere. II. Wierteres über die morphologische Wirkung der Lithiumsälze und ihre theoretische Bedeutung. *Mitt. Zool. Station. Neapel.* **11**, 136–220.
- Hertwig, O. (1894). "The Biological Problem of To-Day: Preformation or Epigenesis?" (P. C. Mitchell, transl.) Macmillan, New York.
- Hilleman, B. (1996). Frog deformities pose a mystery. *Chem. Eng. News* **74**, 24.
- Johnson, T. J., Lunde, K. B., Ritchie, E. G., and Launer, A. E. (1999). The effect of trematode infection on amphibian limb development and survivorship. *Science* **284**, 802–804.
- Jokiel, P. L., and York, R. H., Jr. (1982). Solar ultraviolet photobiology of the reef coral *Pocillopora damicornis* and symbiotic zooxanthellae. *Bull. Mar. Sci.* **32**, 301–315.
- Jones, G., Strugnall, S. A., and DeLuca, H. F. (1998). Current understanding of the molecular actions of vitamin D. *Physiol. Rev.* **78**, 1193–1231.
- Keller, E. F. (1995). "Refiguring Life: Metaphors of Twentieth Century Biology." Columbia Univ. Press, New York.
- Kiliardis, S., Engström, C., and Thilander, B. (1985). The relationship between masticatory function and craniofacial morphology. *Eur. J. Orthod.* **7**, 273–283.
- La Claire, J. J., Bantle, J. A., and Dumont, J. (1998). Photoproducts and metabolites of a common insect growth regulator produce developmental deformities in *Xenopus*. *Environ. Sci. Technol.* **32**, 1453–1461.
- Mayr, E. (1963). "Animal Species and Evolution." Harvard Univ. Press, Cambridge, MA.
- McCollum, S. A., and Leimberger, J. D. (1997). Predator-induced morphological changes in an amphibian: Predation by dragonflies affects tadpole color, shape, and growth rate. *Oecologia* **109**, 615–621.
- McCollum, S. A., and Van Buskirk, J. (1996). Costs and benefits of a predator induced polyphenism on the gray treefrog *Hyla chrysoscelis*. *Evolution* **50**, 583–593.
- Mead, K. S., and Epel, D. (1995). Beakers versus breakers: How fertilisation in the laboratory differs from fertilisation in nature. *Zygote* **3**, 95–99.
- Meteyer, C. U., Loeffler, I. K., Fallon, J. F., Converse, K. A., Green, E., Helgen, J. C., Kersten, S., Levey, R., Eaton-Poole, L., and Burkhart, J. G. (2000). Hind limb malformations in free-living northern leopard frogs (*Rana pipiens*) from Maine, Minnesota, and Vermont suggest multiple etiologies. *Teratology* **62**, 151–171.
- Mohlin, B., Sagna, S., and Thilander, B. (1978). The frequency of malocclusion and the craniofacial morphology in a medieval population in Southern Sweden. *OSSA* **5**, 57–84.
- Montoya, P., Ritter, K., Huse, E., Larbig, W., Braun, C., Topfner, S., Lutzenberger, W., Grodd, W., Flor, H., and Birbaumer, N. (1998). The cortical somatotopic map and phantom phenomena in subjects with congenital limb atrophy and traumatic amputees with phantom limb pain. *Eur. J. Neurosci.* **10**, 1095–1102.
- Morgan, T. H. (1909). Sex determination and parthenogenesis in phylloxerans and aphids. *Science* **29**, 234–237.
- Morreale, S. J., Ruiz, G. J., Spotila, J. R., and Standora, E. A. (1982). Temperature-dependent sex determination: Current practices threaten conservation of sea turtles. *Science* **216**, 1245–1247.
- Müller, G. B., and Steicher, J. (1989). Ontogeny of the syndesmosis tibiofibularis and the evolution of the bird hindlimb: A caenogenetic feature triggers phenotypic novelty. *Anat. Embryol.* **179**, 327–339.
- Newman, R. A. (1989). Developmental plasticity of *Scaphiopus couchii* tadpoles in an unpredictable environment. *Ecology* **70**, 1775–1787.
- Newman, R. A. (1992). Adaptive plasticity in amphibian metamorphosis. *BioScience* **42**, 671–678.
- Newman, S. A., and Müller, G. B. (2001). Epigenetic mechanisms of character origination. In "The Concept Character in Evolutionary Biology" (G. Wagner, Ed.), pp. 559–579. Academic Press, San Diego.
- Nijhout, H. F. (1991). "The Development and Evolution of Butterfly Wing Patterns." Smithsonian Institution Press, Washington, DC.
- Nijhout, H. F. (1999). Control mechanisms of polyphenic development in insects. *BioScience* **49**, 181–192.
- Nishikimi, M., Fukuyama, R., Minoshima, S., Shimizu, N., and Yagi, K. (1994). Cloning and chromosomal mapping of the human nonfunctional gene for L-gulonolactone oxidase, the enzyme for L-ascorbic acid biosynthesis missing in man. *J. Biol. Chem.* **269**, 13685–13688.
- Nomura, S., and Takano-Yamamoto, T. (2000). Molecular events caused by mechanical stress in bone. *Matrix Biol.* **19**, 91–96.
- Nyhart, L. K. (1995). "Biology Takes Form: Animal Morphology and the German Universities, 1800–1900." Univ. of Chicago Press, Chicago.
- Ogasawara, A., Arakawa, T., Kaneda, T., Takuma, T., Sato, T., Kaneko, H., Kumegawa, M., and Hakeda, Y. (2000). Fluid shear stress-induced cyclooxygenase-2 expression is mediated by C/EBP β , CREB, and AP-1 in osteoblastic MC3T3-E1 Cells. *J. Biol. Chem.* **275**, www.jbc.org.
- Okada, T. S. (2000). Lens studies continue to provide landmarks of embryology (developmental biology). *J. Biosci.* **25**, 133–141.
- Okazaki, Y., and Shizuri, Y. (2000). Effects of inducers and inhibitors on the expression of bcs genes involved in cypris larval attachment and metamorphosis of the barnacles *Balanus amphotrite*. *Int. J. Dev. Biol.* **44**, 451–456.
- Ouellet, M., Bonin, J., Rodrigue, J., DesGanges, J. L., and Lair, S. (1997). Hindlimb deformities (ectromelia, ectrodactyly) in free-living anurans from agricultural habitats. *J. Wildlife Dis.* **33**, 95–104.

- Oyama, S., Griffiths, P. E., and Gray, R. D. (Eds). (2000). "Cycles of Contingency: Developmental Systems and Evolution." MIT Press, Cambridge, MA.
- Pfennig, D. W. (1992). Polyphenism in spadefoot toad tadpoles as a locally adjusted evolutionarily stable strategy. *Evolution* **46**, 1408–1420.
- Pfennig, D. W. (1992b). Proximate and functional causes of polyphenism in an anuran tadpole. *Funct. Ecol.* **6**, 167–174.
- Pfennig, D. W., and Collins, J. P. (1993). Kinship affects morphogenesis in cannibalistic salamanders. *Nature* **362**, 836–838.
- Pigliucci, M. (1998). Developmental phenotypic plasticity: Where internal programming meets the external environment. *Curr. Opin. Plant Biol.* **1**, 87–91.
- Pigliucci, M., Schlichting, C. D., Jones, C., and Schwenk, K. (1996). Developmental reaction norms: The interactions among allometry, ontogeny and plasticity. *Plant Species Biol.* **11**, 69–85.
- Proffit, W. R., and Fields, H. W., Jr. (1993). "Contemporary Orthodontics," 2nd ed. Mosby, St. Louis.
- Purves, D., and Lichtman, J. W. (1985). "Principles of Neural Development." Sinauer, Sunderland, MA.
- Rasika, S., Alvarez-Buylla, A., and Nottebohrm, F. (1999). BDNF mediates the effects of testosterone on the survival of new neurons in the adult brain. *Neuron* **22**, 53–62.
- Relyea, R. A., and Werner, E. E. (2000a). Morphological plasticity of four larval anurans distributed along an environmental gradient. *Copeia* **2000**, 178–190.
- Relyea, R. A. (2000b). The lasting effects of adaptive plasticity: Predator-induced tadpoles become long-legged frogs. *Ecology*, in press.
- Relyea, R. A. (2000c). Trait-mediated indirect effects in larval anurans: Reversing competitive outcomes with the threat of predation. *Ecology* **81**, 2278–2289.
- Relyea, R. A., and Mills, N. (2001). Predator-induced stress makes the pesticide carbaryl more deadly to grey treefrog tadpoles (*Hyla versicolor*). *Proc. Natl. Acad. Sci. USA* **98**, 2491–2496.
- Riessen, H. P. (1984). The other side of cyclomorphosis: Why *Daphnia* lose their helmets. *Limnol. Oceanogr.* **29**, 1123–1127.
- Riessen, H. P. (1992). Cost-benefit model for the induction of an antipredator defense. *Am. Nat.* **140**, 349–362.
- Rutherford, S. L., and Lindquist, S. (1998). Hsp90 as a capacitor for morphological evolution. *Nature* **396**, 336–342.
- Safi, R., Begue, A., Hanni, C., Stehelin, D., Tata, J. R., and Laudet, V. (1997). Thyroid hormone receptor genes of neotenic amphibians. *J. Mol. Evol.* **44**, 595–604.
- Sarkar, S. (1999). From the *Reaktionsnorm* to the adaptive norm: The norm of reaction, 1909–1960. *Biol. Philos.* **14**, 235–252.
- Sato, T. (1961). Über die Linsregeneration bei den Cobitiden Fischen I. *Misgurnus anguillicaudatus* (Cantor). *Embryologia* **6**, 251–290.
- Schlichting, C. D., and Pigliucci, M. (1998). "Phenotypic Evolution: A Reaction Norm Perspective." Sinauer, Sunderland, MA.
- Schmalhausen, I. I. (1949). "Factors of Evolution: The Theory of Stabilizing Selection." Univ. of Chicago Press, Chicago.
- Selenka, E. (1876). Zur entwicklung der holothurien, ein beitrage zur keimblättertheorie. *Zeitschr. wissensch. Zool.* **27**, 155–178.
- Sessions, S. K., and Ruth, S. B. (1990). Explanation of naturally occurring supernumerary limbs in amphibians. *J. Exp. Zool.* **254**, 38–47.
- Siebeck, O. (1988). Experimental investigation of UV tolerance in hermatypic corals. *Mar. Ecol. Prog. Ser.* **43**, 95–103.
- Stearns, S. C., de Jong, G., and Newman, R. A. (1991). The effects of phenotypic plasticity on genetic correlations. *Trends Ecol. Evol.* **6**, 122–126.
- Stocum, D. L. (2000). Frog limb deformities: An "eco-devo" riddle wrapped in multiple hypotheses surrounded by insufficient data. *Teratology* **62**, 147–150.
- Sultan, S. E. (1995). Phenotypic plasticity and plant adaptation. *Acta Bot. Neerl.* **44**, 363–383 (Special Jubilee Symposium Volume).
- Tollrian, R., and Dodson, S. I. (1999). Inducible defenses in cladocera: Constraints, costs, and multipredator environments. In "The Ecology and Evolution of Inducible Defenses" (Tollrian, R., and Harvell, C. D., Eds.), pp. 177–202. Princeton Univ. Press, Princeton, NJ.
- Tramontin, A. D., and Brenowitz, E. A. (2000). Breeding conditions induce rapid and sequential growth in adult avian song control circuits: A model for seasonal plasticity in the brain. *J. Neurosci.* **20**, 854–861.
- Van Buskirk, J., and Relyea, R. A. (1998). Natural selection for phenotypic plasticity: Predator-induced morphological responses in tadpoles. *Biol. J. Linn. Soc.* **65**, 301–328.
- van der Weele, C. (1999). "Images of Development: Environmental Causes in Ontogeny." State Univ. of New York Press, Albany.
- Van Valen, L. (1973). Festschrift. *Science* **180**, 488.
- Varrel, J. (1992). Dimensional variation of craniofacial structures in relation to changing masticatory-functional demands. *Eur. J. Orthod.* **14**, 31–36.
- Via, S., Gomulkiewicz, R., De Jong, G., Scheiner, S. M., Schlichting, C. D., and Van Tienderen, P. H. (1995). Adaptive phenotypic plasticity: Consensus and controversy. *Trends Ecol. Evol.* **10**, 212–217.
- Vogt, O. (1926). Psychiatrisch wichtige tatsachen der zoologisch-botanischen systematik. *J. Psychol. Neurobiol.* **101**, 805–832.
- von Siebold, C. T. (1854). "On a True Parthenogenesis in Moths and Bees: A Contribution to the History of Reproduction in Animals." W. S. Dallas, London.
- Waddington, C. H. (1942). Canalization of development and the inheritance of acquired characters. *Nature* **150**, 563–565.
- Waddington, C. H. (1953). Genetic assimilation of an acquired character. *Evolution* **7**, 118–126.
- Waddington, C. H. (1956a). "Principles of Embryology." Macmillan, New York.
- Waddington, C. H. (1956b). Genetic assimilation of the bithorax phenotype. *Evolution* **10**, 1–13.
- Wakahara, M. (1996). Heterochrony and neotenic salamanders: Possible clues for understanding the animal development and evolution. *Zool. Sci.* **13**, 765–776.
- Warkentin, K. M. (1995). Adaptive plasticity in hatching age: A response to predation risk trade-offs. *Proc. Natl. Acad. Sci. USA* **92**, 3507–3510.
- Warkentin, K. M. (2000). Wasp predation and wasp-induced hatching of red-eyed treefrog eggs. *Anim. Behav.* **60**, 503–510.
- Wassersug, R. J. (2000). Vertebrate biology in microgravity. *Am. Sci.* **89**, 46–53.
- Weijts, W. A., and Hillen, B. (1986). Correlations between the cross-sectional area of the jaw muscles and craniofacial size and shape. *Am. J. Physiol. Anthropol.* **70**, 23–431.
- Weismann, A. (1875). Über den saison-dimorphismus der schmetterlinge. In "Studien zur Descendenz-Theorie." Engelmann, Leipzig.
- Weiss, P. (1950). The outlook in morphogenesis. *Ann. Biol.* **26**, 563–582.

- Wenger, R. H., Kvietikova, I., Rolfs, A., Camrenisch, G., and Gassmann, M. (1998). Oxygen-inducible erythropoietin gene expression is dependent on a CpG hypoxia-inducible factor-1 DNA-binding site. *Eur. J. Biochem.* **253**, 771–777.
- West-Eberhard, M. J. (1989). Phenotypic plasticity and the origins of diversity. *Annu. Rev. Ecol. Syst.* **20**, 249–278.
- Wolpert, L. (1994). Do we understand development? *Science* **266**, 571–572.
- Woltereck, R. (1909). Weitere experimentelle untersuchungen über artveränderung, speziell über das wesen quantitativer artunterscheide bei daphniden. *Versuch. Deutsch. Zool. Ges.* 110–172.
- Wu, K. C. (1996). Entwicklung, stimulation, und paralyse der embryonalen motorick. *Wien. Klin. Wochenschr.* **108**, 303–305.
- Zaman, G., Cheng, M. Z., Jessop, H. L., White, R., and Lanyon, L. E. (2000). Mechanical strain activates estrogen response elements in bone cells. *Bone* **27**, 233–239.

Received for publication November 2, 2000

Revised December 23, 2000

Accepted December 23, 2000

Published online March 27, 2001