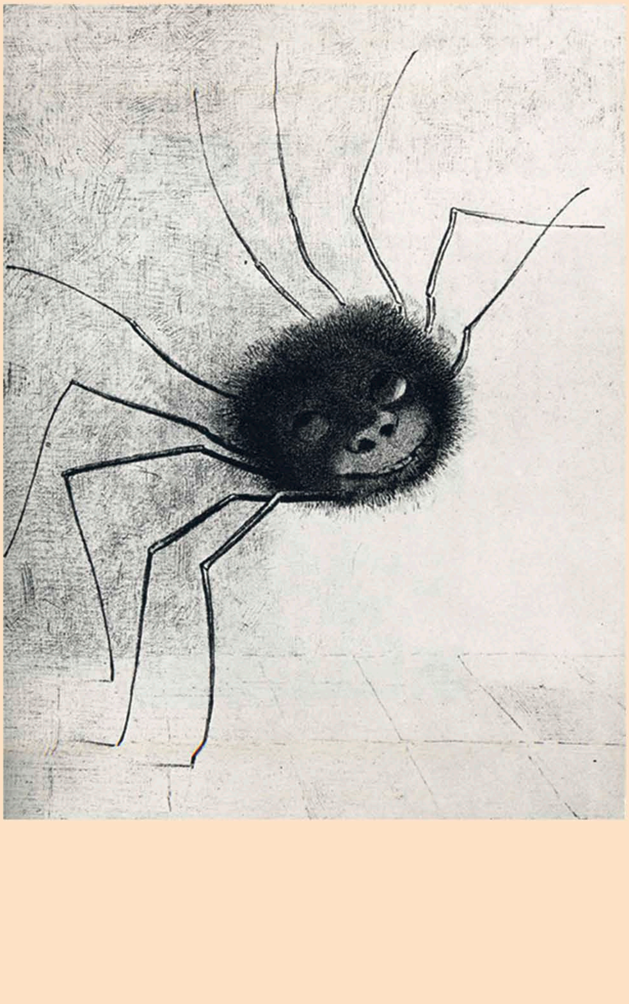
PERE ALBERCH

The Creative Trajectory of an Evo-Devo Biologist



Diego Rasskin-Gutman,
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The Creative Trajectory of an Evo-Devo Biologist

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Pere Alberch's research interest in possible versus impossible forms,
a question at the interface between science and the arts exquisitely
illustrated by Odilon Redon in this lithography, dated 1887, *Araignée souriante*.

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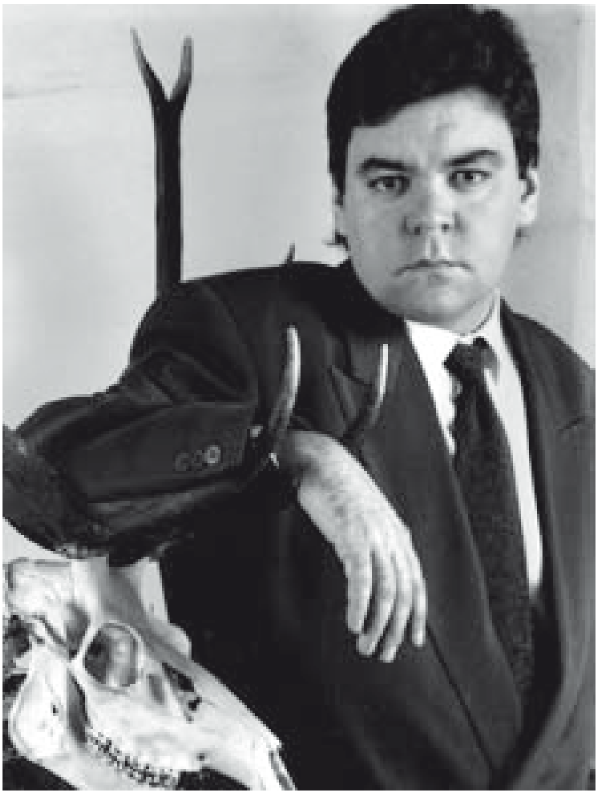

Preface

Back in the mid-1990s, encouraged by a suggestion of the then vice-chancellor of research at the University of València, Juli Pereto, we started the process of creating a research institute on biodiversity and evolutionary biology. The first task was to look for a prominent evolutionary biologist; Pere Alberch was the immediate, most obvious choice. I was aware of his interest in resuming empirical scientific research, work that he had already done in the United States, as well as in giving up the managerial activities that he had been performing upon his return to Spain. The Institute Cavanilles, which at the time was just built, seemed for him an excellent chance for this dual objective, and his decision of joining us was for me the opportunity to start, led by him, a center that could attract researchers in the field of Evolutionary Biology from all over the world.

What I cherish most about Pere was the frank and passionate discussions we engaged in about the explanatory scope of the theory of evolution. We were in tune from the first moment we met, not only in our way of understanding science, but also in our way of understanding art and thought in general. As a result, we agreed that the Institute would be like a crucible in which scientific discussions will flourish, yes, but with the firm commitment to creating something new. I often wonder where we could have been today if Pere were still with us. Honestly, I have not recovered yet from his intellectual and human loss.

His family, aware of Pere's interest in coming to Valencia, deemed it appropriate to deposit with us his latest writings and courses in which he had been working. I promised them to pay him the proper homage and to claim on his behalf some of his ideas that have shaped the subsequent development of evo-devo. I thought that the tenth anniversary of his death was a good opportunity to do so. We organized a symposium as original and daring as his thinking, and we have compiled his fundamental work in the book that the reader has now in his hands. Nothing would be more satisfying than to find out that this work will serve to enhance the figure of a true innovator.

Valencia, December 2008

ANDRÉS MOYA
Head, Cavanilles Institute of Biodiversity
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PART II

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Foreword

Pere Alberch was an outstanding figure of developmental and evolutionary biology during the last quarter of the 20th century. He made important contributions to developmental biology and its relation to organic evolution in the framework of theoretical biology. However, his research activities extended beyond the summits of theory, with an important empirical work focused on amphibian metamorphosis and limb development in amphibians and reptiles. In addition, his awareness of the social function of science naturally led him to a genuine interest in the field of museology.

Art and artistic activities played an important role in his scientific thinking. Alberch used to make a distinction between a museum public façade and its true soul. The former would be centred on exhibitions, but the true soul of a museum consisted of its collections and its research activities, which together turned museums into living, dynamic institutions. Uniting a clear vision and his own affinity with the arts, Alberch became a true artist of museology, using the museum venues as a vast exhibition hall. It was a place where he was able to make scientific and artistic statements by proposing to the visitor installations where he would mix equal parts of science and art.

Born in Badalona, near the great city of Barcelona (Spain), on November 2, 1954, he soon showed interest in natural history, influenced by his mother and grandfather, writing his first two scientific papers about amphibians, his preferred group, when he was only 19 years old. He carried out studies of biology at the University of Kansas and worked on herpetology with professors Linda Trueb and William E Duellman. He pursued his career at the University of California at Berkeley, joining the group of David Wake, who became, along with mathematician George Oster, one of his thesis advisors.

Alberch was firmly committed to furnishing biology with more formal and theoretical frameworks; mathematics and philosophy were often used in the background of his contributions. When he met Stephen Jay Gould, his interest in the relationship between development and evolution was fostered to such an extent, that he soon developed a whole research program dealing with the role development needed to play in a comprehensive evolutionary theory. Later, he went on with his task at the University of Harvard, where he was appointed as Associate Professor, and performed curatorial functions at the Agassiz Museum of Comparative Zoology. He was a true Renaissance man, with eclectic, multi-faceted interests, in sharp contrast with the typical scientist of our days.

Specialization was not part of his vocabulary; this was probably the main reason why he failed to obtain tenure at Harvard. As noted by David Wake, the staff at Harvard did not know where to place him: Herpetologist? Developmental biologist? Evolutionary biologist? Harvard failed to fully appreciate the multiple dimensions of Pere Alberch, and thus he returned to Spain.

Upon his arrival to Spain in 1989, he obtained right away the position of Research Professor at the Consejo Superior de Investigaciones Científicas (CSIC) and he was appointed immediately as Director of the Museo Nacional de Ciencias Naturales (MNCN) in Madrid. His curatorial experience at the Agassiz Museum led him to restructure the exhibition philosophy of this institution, which was absolutely outdated. Some years after, he was offered the opportunity to lead the recently created Institute Cavanilles for Biodiversity and Evolutionary Biology (ICBiBE) at the University of Valencia. Alberch was already in the process of moving to his new position in Valencia, when he died in his sleep on 13 March 1998 from heart failure.

Ten years after his premature death, the ICBiBE organized an international meeting that was held in Valencia from the 21st to the 24th of May, 2008. More than thirty speakers debated for three days about his legacy in biology and museology. In September 2008, the Konrad Lorenz Institute for Evolution and Cognition hosted yet another meeting to honour Alberch's legacy. Now, we present this book, to further his memory by re-editing Alberch's most important contributions as well as three original studies illustrating the many ways he was a decisive actor in the origination of evo-devo research.

The volume is divided into two parts. The first one consists of introductory papers by Diego Rasskin-Gutman, Miquel De Renzi, Arantza Etxeberria y Laura Nuño de la Rosa. These works serve as introductions by placing Alberch in a historical-philosophic framework and by analyzing how he influenced –and was influenced by– several fields, such as evolutionary palaeobiology or developmental evolutionary biology.

The second part of the book is a compilation of facsimile reproduction of 20 selected papers of Pere Alberch. This part is subdivided into three sections. The first section contains only one paper, which we consider to be crucial, because it exposes the main lines of his future research program. The next section, the largest of this selection, groups together seventeen papers dealing with the common subject of evolution and development. Here we find the main topics on which Alberch focused his research: developmental constraints, heterochrony and modelling. The third section is devoted to museology.

We are at exciting times in biological research. Several disciplines are starting to coalesce and the full integration between development and evolution—both at molecular and morphological levels—is at reach with the aid of new concepts, formalisms, as well as mathematical and computational techniques. With the publication of this book, we pay a well deserved homage to the many ideas of Pere's great mind, which we are sure will shine new light on the future integration of the biological sciences.

Diego Rasskin-Gutman
Miquel De Renzi

PART I

The international meeting in Valencia, Spain (May 2008) featured 34 lecturers. In this picture, taken at the meeting venue, the Colegio Mayor Rector Peset of the University of Valencia, we see some of the lecturers as well as some participants. The editors of this book appear in the upper row: Miquel De Renzi (far left side) and Diego Rasskin-Gutman (far right side).

A world of opportunity within constraint: Pere Alberch's early evo-devo

Arantza Etxeberria
Laura Nuño de la Rosa

Introduction

The work of Pere Alberch is crucial for studying the early stages of evo-devo. In particular, it illustrates very persuasively why developmental systems have so much to say about the course of evolutionary change. In addition to an important empirical work, he elaborated a stimulating framework of theoretical ideas on biological form, morphological variation, and how developmental processes establish possible evolutionary paths previous to the action of natural selection. In this framework, the study of development and evolution are related through the notion of possible morphologies. In his view, the morphology of organisms shows internal coherence and structure, emergent from complex non linear interactions among parts and with the environment. This constitutes a source of determinism absent from other accounts in which novelty is considered to appear only from random mutations. In his words: "In evolution selection may decide the winner of a given game but development non-randomly selects the players" (Alberch, 1980, p. 665).

In the 1970s and 1980s many biologists argued against the prevalent gene-centred view of biology and demanded to study how the organization of multicellular organisms, their *Bauplan*, influences morphological variation and, consequently, evolutionary change. In Alberch's work the link between development and evolution is regained in several ways: (1) by challenging the static hierarchical theories of development in favour of a dynamic, cyclical, and interactive conception that he pursued through experiments and dynamical models; (2) by stressing the relevance of studies of ontogenetic phenomena at the appropriate morphogenetic level, so that morphologies might be regarded as products of complex genetic and epigenetic interactions; and (3) by suggesting that some developmental properties constrain possible paths of evolution in specific directions, because they define the set of variations associated with certain forms (e.g., the tetrapod limb, the pattern of digits, or the form of teeth). Thus, Alberch questioned the theoretical framework that assumes a direct correspondence between genetic structure and morphology with natural selection acting on the effects of random changes.

Although Alberch's work flourished precisely before and during the expansion of developmental genetics, he was more interested in the mathematical techniques of dynamical systems than in the new molecular techniques appearing at the time. His approach was that of a theoretical biologist who considered that experimental and theoretical work together could allow for an a priori and predictive knowledge of living form and its evolution. Thus, Alberch's peculiarity with respect to others investigating development from a dynamical systems approach, like Goodwin (1994) or Kauffman (1993), is the use of experiments in addition to simulations.

In his relatively short life, Alberch wrote many articles on many subjects (he published his first paper when he was only 19 years old), but probably his most important, and most cited, work was done during the decade of the 1980s. Many of the advancements in evo-devo these days are a consequence of ideas and projects in which he, along with many others, was involved at that time.

In this paper we focus on what we consider to be the main early ideas of evo-devo. First we see that Alberch defends the importance of doing research at the morphological level, which he considers as non reducible to lower level molecular factors and that appears as a realm of discrete themes. Then, we focus on the relevance of developmental mechanisms, the role they acquire for studying evolution and how they are conceptualized as developmental constraints to reflect the intrinsic abilities of developmental systems to produce some forms and not others, with consequences for evolutionary opportunities. Finally, in the last section we summarize some conceptual elements of Alberch's views on how to integrate different approaches to explain the morphological organization of living beings.

The themes of the discrete morphospace

The phenomenological departure point of Alberch's research is the recognition of the ordered character of morphospace, i.e. the space of biological forms: "Nature is not chaos, neither is it a boundless continuum of forms." (1982a, p. 315). Morphological variation is not random and continuous, as the standard view in evolutionary biology maintains, but shows a discrete organization. This "morphological order" has the following properties:

> " (1) Phenotypes are discrete. That is, points are not distributed uniformly over phenotype space, but tend to cluster around major "themes", corresponding to taxa or classes of teratologies. (2) While there may be considerable dispersion around a morphological theme, the variability in any trait is definitely limited. (3) When new morphological themes arise, either in ontogeny or phylogeny, the transitions between themes are not random. (4) These properties are largely the result of epigenetic interactions during development."(Oster and Alberch 1982, p. 444).

The neo-Darwinian approach cannot fully account for the discrete character of morphospace: if evolution depends strongly on the action of natural selection on gradual variations, any living form must be theoretically possible, and the space of biological morphologies, continuous. From this perspective, discrete characters must be the result of functional convergence: unity of type in non-related taxa is seen as the outcome of similar selective pressures.

In Alberch's view, internal (or developmental) and external (or selective) explanations of evolutionary change are not contradictory, but they deal with different problems: the origin and the fixation of evolutionary change. Standard evolutionary biology has been traditionally concerned with the fate, not with the origin, of variation. The neglect of the origin of variation is linked to the beliefs of the isotropy of molecular variation and the direct correspondence of genotype and phenotype. Alberch challenged both assumptions and vindicated the study of the patterns of variation (the existent and possible order) precisely to understand the mechanisms responsible for their generation, i.e., the variability of developmental systems, as morphological parallelisms suggest that the same mechanisms are in action (Alberch, 1983). Thus, in his view, evolutionary research needs to proceed in two steps: the first is to elucidate an ordered pattern of variation, i.e., to characterize invariance, and the second, to look for the mechanisms behind those patterns (Alberch, 1985b). Without characterizing morphological invariance -he argued- it would be impossible to find the mechanisms responsible for its generation. That is why in Alberch's work morphology recovers the significance as a biological discipline that it lost in the Modern Synthesis: comparative anatomy has a fundamental role in evolutionary theory, since its job is "to determine regularities of structural organization that enable a classification and understanding of the ordered diversity of form" (Shubin and Alberch 1986, p. 377).

The study of patterns of variation is thus the first step of Alberch's research and it is also the sole protagonist of many of his papers, where he tries to distinguish between the order of forms internally generated by development and the role of natural selection to favour subsets of them. With this goal, Alberch studied three main morphological phenomena where the patterns of variation cannot be explained by Natural Selection but by the structure of developmental processes.

One case is that of *morphological convergence* where a similar morphology appears linked to different adaptive requirements. In a series of experiments, the functional significance of structural modifications in hand and foot morphology (such as reduction and loss of phalangeal elements, development of interdigital webbing and fusion of tarsal and carpal elements) is explored. The goal is to analyse how to integrate adaptation and ontogeny in a case where a morphological convergence does not imply functional convergence (Alberch, 1981).

A second phenomenon of morphological order is that revealed by patterns of *intraspecific variation*. This is the subject of Alberch's study of phenotypic variation in osteological characters of populations of species of the neotropical salamander genus *Bolitoglossa* (Alberch, 1983). The main finding is a recurrence of the same variants in widely unrelated species, a fact that again, cannot be explained from an externalist approach.

Finally, a constant source of evidence for discrete variation comes from his studies of *teratologies*. Throughout his work, Alberch (like transcendental morphologists Etienne and Isidore Geoffroy Saint Hilaire) demonstrated a special fascination with the development of malformations (Alberch, 1989). Non-functional teratologies were especially appealing for the study of the generative properties of developmental systems. These deviations from "normal" development result in forms which are often lethal or less well adapted than their predecessors. Therefore, following the adaptationist logic, natural selection should prevent their appearance or at least contribute to their disappearance. But in spite of being strongly selected against, teratologies are still being generated in a recurrent way.

All these evidences of the discrete character of morphological order, the regularities of these patterns of variation, indicate the fundamental role of development in specifying the possible forms available for selection: morphological convergence, intraspecific morphological diversification and teratologies show a logic emerging from internal developmental systems.

Discovering developmental mechanisms

We have seen that the properties of morphospace demand that morphological evolution be studied from an ontogenetic perspective (Alberch, 1980). This requires defining developmental phenomena at the morphogenetic level: despite the standard position of population genetics according to which all evolutionary novelty comes from random mutations, in a direct one-to-one genotype-to-phenotype relation, the study of ontogeny requires special attention to "epigenetic regulation" and, thus, to a level higher than that of genetics to explain development.

The morphogenetic level is characterized in two ways. On the one hand, developmental form is not a static entity defined by the spatial position of its component parts, but the result of a dynamical process involving interactions, integration, and regulation among them. On the other hand, the morphogenetic level may be studied in physico-chemical terms. These two points converge in a conceptualization of developmental dynamics as susceptible to mathematical formalization and experimental treatment.

Here we describe Alberch's conceptualization of the morphogenetic level and the global properties of developmental processes. Then, both work on the mathematical formalization of the dynamics of developmental systems (and the role of mathematical biology) and of their biophysical properties (and the role of experimental embryology) are considered. Both approaches define a mechanistic viewpoint on development and evolution that may provide some degrees of determinism and predictability.

The morphogenetic level.- One of the main difficulties for Alberch's theoretical stance was to establish the reality of the morphogenetic level as well as the necessity of working at it despite the success of the genome-centred approach in evolution and development. In his opinion, the latter favours a "hierarchical scheme" of development, which portrays an extreme version of the neo-Darwinist view of genes as directly prescribing developmental processes that, in turn, specify morphology in a straightforward way. This view reduces both morphological evolution and development to purely genetic problems: development to a sequence of gene expression and evolution to a change in gene frequencies.

However, this scheme presents several problems (Alberch, 1991). First, such an "open loop" system would be extremely unstable against the random disturbances that accompany normal development. Second, we do not find a one-to-one correspondence between certain regions of DNA and a given morphological trait (Alberch, 1983). On the contrary, the effect of genes on morphology is mostly indirect: they code for molecules which either regulate the expression of other genes or confer properties on cells (e.g., cell division rates, apoptosis, differentiation timing or cytoskeletal properties), which then construct organs and structures largely in accordance with physico-chemical laws. Thus, the complexity and nonlinearity of the genotype-phenotype mapping increases at higher levels of interaction. Due to the highly context-sensitive character of gene expression (dependent on embryonic stage and local environment), similar genetic changes may yield different morphological effects, and the other way around. In fact, at the morphogenetic level, genetic mutations may not even be expressed, since developmental interactions have properties that emerge from the dynamics of the system and are not encoded in the genome (Oster and Alberch, 1982; Oster et al., 1988; Alberch, 1991). Therefore, phenotypic diversity is not so much the product of new genes as of permutations in context (i.e., the timing and location of expression) of existing genes. The evolutionary consequences of this asymmetry are obvious: there are qualitative differences between modes of evolution at the genetic and epigenetic levels, and therefore there is often no direct correspondence between genetic and morphological divergence (Alberch, 1983).

In order to capture the properties of developmental systems and their evolutionary consequences, Alberch proposes an alternative view of development which he calls the "*cyclical/feedback scheme*". According to this scheme, developmental processes are divided in three interacting levels (1982a, p. 320): interactions among genes within a highly structured genome, proteins and enzymes generating cell properties involved in morphogenesis, and tissue interactions. Following Waddington, Alberch considered that these regulatory interactions specify the epigenetics according to which phenotypes are well buffered systems with respect to both genetic and environmental perturbations during ontogeny (Alberch, 1980). Therefore, in the cyclical view of development "genes are just one step in the chain of interactions, gene expression is both the cause and the effect of a morphogenetic process" (Alberch, 1991, p. 6).

Thus, Alberch's message is that the morphogenetic level of description deserves special attention in evo-devo because it vastly governs both the dynamics of morphogenesis and the appearance of novelty. For Alberch it is at this level where the range of possible variation for evolution is established, introducing a deterministic factor in the production of variation (which was largely left to random mutations in the Modern Synthesis) that allows for some form of predictability, in so far as the variability of a given trait (its potential for producing variations) can be determined.

Developmental dynamics: from heterochrony to construction rules.- Despite the efforts of authors like de Beer, Waddington or Schmalhausen, the role of development was absent from standard evolutionary theory since the decline of the recapitulation theory. However, it received a renewed interest by the scientific community after the publication of Gould's Ontogeny and Phylogeny (1977). Two years later, Alberch and co-workers published together with Gould a celebrated paper where they presented a quantitative method for describing the relationship between heterochronic changes and phyletic trends (Alberch et al., 1979). The aim was to improve Gould's "clock model," which was essentially qualitative and "static." Thus, Alberch and co-workers quantified Gould's model, by characterizing those modifications in the developmental processes that produce relative changes in size and shape. Furthermore, they provided a dynamic expression of heterochrony, defined in terms of shifts in specific processes such as onset, cessation, or rate of growth, rather than of end results (see De Renzi, this book). This approach is followed in later publications, where we find empirical work on heterochrony in *Bolitoglossa occidentalis* (Alberch and Alberch, 1980)

This treatment of heterochrony (and its focus on patterns of relative growth) was very influential, defining the way the concept is used in current evo-devo (Smith, 2001; 2002). However, after 1985 Alberch found that this approach had serious problems, and revised his views in accordance with his more dynamical and mechanistic conception of development (Alberch, 1985a; Alberch and Blanco 1996). Alberch opposes two conceptions of ontogeny and phylogeny: a kinetic one, which only classifies and compares developmental stages, and a dynamic one based on an understanding of the underlying mechanisms of development and the comparison of developmental events (Alberch, 1985a; Oster et al., 1988). The conception of development as a sequence of discontinuous morphological stages conserved throughout evolution and the associated mechanism of heterochrony is rooted in the Haeckelian view. But, taking into account the critique made by von Baer to the law of parallelism of Meckel-Serres, Alberch insists that comparative embryology does not show any linear recapitulation, concluding that all the intermediate stages postulated by heterochronic models are meaningless (Alberch, 1985a, p. 51). Instead, changes between two related morphologies "must be searched for in terms of changes in the developmental rules of interaction or initial conditions, rather than in intermediate ontogenetic stages" (p. 51). He uses several examples, including experiments of previous papers (Alberch and Gale, 1983; 1985) to "illustrate how evolutionary models based on heterochrony can be embedded within a more mechanistic and dynamic framework" (Alberch, 1985a, p. 55). He had to admit then that the work done in 1979 was "no longer valid" (p. 55), because this perspective did not fit well with his later views of development as a dynamical system governed by a set of "construction rules" that are able to generate a global pattern.

This new conceptualization of development has important consequences for the definition of homology, a hot question in current evolutionary biology. The distinction between developmental stages and developmental events leads Alberch to "champion the view that many of the difficulties in establishing homologies could be avoided or resolved by basing comparisons between elements on the developmental processes which created them, rather than on their final geometric form." (Oster et al., 1988, p. 877). The best empirical example of this approach is the study of the morphogenesis of the vertebrate limb. The early stages of the skeletal patterning were explained by a mechanistic model of embryonic branching and segmentation in initial chondrogenesis. In this model, the loss of a digit may result from the failure of a branching bifurcation, and then, it is not sensible to ask "which" digit was lost, since it is the basic sequence that has been altered. Thus, not the morphological elements but the morphogenetic processes are the units of comparison (Oster et al., 1988).

In order to capture this new view on developmental evolution, Alberch develops the notion of *construction rules,* which crystallized in publications on the morphogenesis of teratologies and tetrapod limbs. The comparative analysis and the experimental manipulation show how these rules determine how a limb can be modified by evolution:

> "An understanding of these, very often simple, rules can provide valuable insights into the apparent complexity of many developmental processes and, furthermore, allow us to determine the relationships among different phenotypes, since the set of possible phenotypic transformations will be constrained by the generative potentialities of the morphogenetic rules involved in the process." (Alberch, 1982, p. 321)

Development is viewed by Alberch as a dynamical system governed by certain rules or constraints which remain stable during long periods of time, but permit a certain range of variation through the alteration of certain (genetic and non-genetic) parameters. These rules arise from the interaction of different "resources" at different levels and direct or channel the possible evolution of the system.

The formal properties of developmental systems and the role of mathematical biology.- In order to explain the mechanics of developmental programs, Alberch used the conceptual and mathematical tools developed in the framework of dynamical system theory.

Developmental processes are viewed as "complex dynamical systems, where a small set of simple rules of cellular and physico-chemical interaction can interact to generate a complex morphology" (Oster and Alberch, 1982, p. 455).

These rules of construction, probably inspired in the conceptual framework of cellular automata, are formally captured as developmental parameters. Alberch postulates that developmental processes, as any pattern-generating system, have an associated parameter space that captures the global properties of the system, defining its "evolutionary potential" or evolvability:

> "This approach views form and pattern emerging as the combination of a set of specific pattern-generating algorithms with a set of initial boundary conditions. Diversification occurs as the result of changes in the developmental parameters of the algorithm (a "program" composed of a set of local rules of biochemical, cell-cell, or tissue interactions). The basic structure of a pattern formation model is to quantitatively state some local rules of interaction that are able to generate a global pattern." (Alberch, 1985a, p. 50)

The effects of genetic or environmental alterations in the basic developmental properties are mathematically abstracted as parameter perturbations in a dynamical system (Alberch, 1982, p. 323). Thus, "morphological diversity is generated by perturbations in parameter values (such as rates of diffusion, mitotic rate, cell adhesion, etc.) while the structure of the interactions among the components remains constant" (Alberch, 1989, p. 27). In order to visualize all possible pathways of transformation among phenotypes that some specific rules of construction generate, Alberch imported a tool from dynamical systems theory: *transformational diagrams* (Alberch, 1991). Each species or trait will have a unique transformation diagram dependent on its position in the parameter space, and smooth perturbations of the parameters can result in only certain given phenotypes. These properties of developmental systems are illustrated by teratologies, which are not only generated in an organized and discrete way, but also exhibit general transformational rules. In the case of the salamander limbs, the perturbation of developmental paths through a genetic mutation or experimental manipulation can only produce a limited set of transformations.

Alberch considers that the stabilities and bifurcations of Waddington's epigenetic landscape can be now formalized with the language and mathematical tools developed in dynamical system theory: phenotypic stabilities are treated as *dynamical attractors*, regions in parameter space where small perturbations do not disrupt the basic organization. In the same way, Müller and Wagner state that "ontogenies can be understood as systems of temporary equilibria or steady states between developmental entities" (Müller and Wagner, 1991, p. 249). *Bifurcations* are developmental thresholds (such as critical cell number or inductive or spatial relationships) with the opposite result. The way to explain how mechanistic rules can constrain processes of morphological evolution has to do with the presence of mathematical bifurcations and how it "implies discontinuity and directionality in phyletic transformations" (Oster and Alberch, 1982, p. 455). Thus, developmental thresholds explain the nonlinear character of development and the discontinuity of evolution outlined by the theory of punctuated equilibrium (Eldredge and Gould, 1972): modifications that go beyond such thresholds can cause nonlinear effects.

The physical properties of developmental systems and the role of experimental embryology.- Although modelling captures the structural properties of developmental systems, the study of the mechanical and chemical laws to which the morphogenetic processes are subjected is necessary (Oster et al., 1988). In addition to his conceptual work as a theoretical biologist, Alberch proved to be a brilliant experimental embryologist. The papers published in collaboration with Emily Gale since 1983 illustrate this work (Alberch and Gale 1985, 1986; Alberch, 1986; Alberch et al., 1986). Here the influence of perturbations of some developmental parameters in the generation of new forms is studied experimentally. The authors compared the results of treating the limb buds of a frog and a salamander with colchicine, a mitotic inhibitor. This treatment results in various abnormal morphologies such as limbs of smaller size that have lost some skeletal elements. Their results were used to contrast the hypothesis that the digital pattern is affected by reduction in the number of mesenchyme cells. Changes in pattern formation took place when the size and the number of cells of the limb bud were reduced under a critical value, an experimental fact consistent with the mathematical models studied with Oster (Oster et al., 1988). However, the main result of these experiments was that the malformations produced by colchicine were not chaotic, but they exhibited a high degree of order. Most of the patterns of diversity of digital morphology in amphibians can thus be explained as a reflection of developmental properties (Alberch and Gale, 1985).

The consequences of the experimental manipulation of developmental systems are twofold. (1) The experimentally generated pattern of variation can be compared with the patterns of natural variation, allowing phylogenetic inferences and to trace possible evolutionary pathways. For example, salamanders seem to develop their limbs in a way very different from the rest of tetrapods because the sequence of digit formation seems to be inverted (Alberch and Gale, 1983). Digit reduction is a phenomenon that has taken place several times independently in amphibian evolution. Frogs usually lose their most