ANALYSIS OF DEVELOPMENT

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ILLUSTRATED

W. B. SAUNDERS COMPANY

Philadelphia & London · 1955

Made in United States of America

Press of W. B. Saunders Company, Philadelphia

Library of Congress Catalog Card Number: 55-5565

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PREFACE

THIS BOOK owes its inception to informal gatherings, seminar fashion, of a small group of embryologists who for several summers (1933–1940) periodically retired from the busy scene of the Marine Biological Laboratory at Woods Hole to the peace and quiet of the sand dunes along the northern coast of Cape Cod near Barnstable. With the sea as background and the sand for a blackboard the "Sandpipers" (a name derived from our alert and ever-searching avian companions on the beach) discussed at length the problems of development and groped for a better understanding of the mechanisms of embryogenesis.

To those who took part in them, these group discussions were a valuable experience. The satisfaction that came from the exchange and conciliation of conflicting views aroused our urgent desire to broaden the experience and share it with a far wider circle of biologists. Jointly the hope was engendered that future accounts of embryological knowledge would emphasize the dynamic and causal aspects of embryogenesis rather than mere description and seriation of developmental stages, a practice still too common in the lecture room and textbook. To transcend descriptive embryology and blend experimental data with "Beobachtung und Reflexion" was clearly set as our goal. Only by such an account could younger students be challenged and influenced in their future research and teaching in this important field. Above all, the need was felt for helping to overcome the trends of overspecialization by encouraging a wider, interdisciplinary perspective and by integrating the ever-growing volume of accumulated information into a broad conceptual framework. The need for a well-balanced account of the developmental process was apparent. But how was such a plan to be translated into action?

It was evident from the start that the subject matter had grown in volume and intricacy to the point where it seemed futile for any one individual to attempt to cope with such a task. The alternative was to call on many specialists for authoritative presentations of their respective subjects. We realized that by this procedure much of the desired unity and integration would be sacrificed, and the present volume bears plainly the stigmata of these imperfections. Yet, despite our hesitations on this score, the three of us, encouraged by the urging of many colleagues, outlined in 1947 a plan for a collaborative work on the analysis of the developmental process.

The original blueprint contained an outline and table of contents of the subject matter to be covered in hierarchical divisions, as well as specifications for their serial order and relative proportions. For this basic pattern the three Editors take full responsibility. Yet, within that general frame, the individual contributors were given no more than a general topical guide that left full scope to their personal preferences in the choice of samples, style, and manner of presentation, the only provision being that they conform to the general spirit and objectives of the undertaking. The guiding aims were expressed to them in the following commentary.

The purpose of this book is to present a modern *synthesis* of our knowledge of the principles and mechanisms of development. In these days of rapidly expanding information, it becomes increasingly difficult to keep perspective. It is urgent, therefore, that this book provide not just another source of information, but that it view the phenomena of development from a common perspective so that the reader may recognize the great main lines and inner coherence of the field above the multiplicity of often unrelated details of which the field seems composed when viewed too closely. There is perhaps need for a comprehensive compilation of all the experimental data that have been amassed in the field of Experimental Embryology in the past. However, this book is *not* intended to fill that need. It is not to be a handbook. It does not aim at a complete and exhaustive review of the field. Each contributor is asked to make a critical and, in a way, subjective selection of the special field to be covered in his article. He should give a clear outline of the general problems, concepts, and lines of investigation of his topic and illustrate them with selected examples from experimental data. Only those experiments should be presented that are crucial and analytically strong and convincing. Repetitiveness should be avoided. Use should be made of tabulations and graphs wherever possible. Since the book addresses itself mainly to active or potential investigators (particularly in the experimental branches of embryology, pathology, histology, endocrinology, and developmental genetics), it would be of value to point out gaps in our knowledge, the lack of critical experimental data in unexplored or controversial fields, and lines of research which would deserve being followed up. In summary, the book has as its major objective the synthesis and evaluation of pertinent material selected from the whole field of animal growth and development, with emphasis upon recognized principles and mechanisms as well as on unsolved and new problems.

With these suggestions we approached twenty-five biologists prominent in the subject areas to be covered in the volume. They readily accepted the invitation to collaborate despite the tribulations and obligations inherent in such undertakings. The Editors are very grateful to all of them not only for their contributions to this book but also for the spirit of cooperation and patience which they exhibited during the years of arduous labor that went into its preparation. As in all concerted creative efforts of this kind, progress in realization was slow and at times faltering. Contrary to the development of an organism, no forces were at work to coordinate the separate creative efforts, and the Editors did not see fit to weld the different contributions into a uniform mold. Each contributor is finally responsible for the organization, scope, and content of his text. The Editors, on the other hand, must bear the responsibility for the plan and the scope of the book, and assume the blame for any defects in its structure.

Whatever its imperfections and limitations, the book represents a first-hand portrayal of present-day views of animal development. As such, we hope it may provide a basis of departure for future endeavors of this kind. The science of embryology, like the embryo, is governed by the principles of progressive differentiation, its present status only a transitory moment between past and future—its full potentialities yet to be realized. It is to the pioneering spirit of those students who hereafter will enter the field of development and growth that this volume is primarily dedicated. In no lesser degree we inscribe these pages to students and investigators in other fields of the biological sciences, including medicine and agriculture, who are constantly confronted with problems of a developmental nature and must deal with them.

The Editors have been fortunate indeed in the cordial relationship which has existed between them and the publishers from the beginning of this undertaking. We are most grateful to them for their unlimited patience, resourcefulness, and splendid cooperation in making a book such as this all that it should be in style and typography.

B. H. Willier
Paul Weiss
Viktor Hamburger

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Section I

PROBLEMS, CONCEPTS AND THEIR HISTORY

JANE M. OPPENHEIMER*

"Is cell-differentiation inherent or induced?

"A thoughtful and distinguished naturalist tells us that while the differentiation of the cells which arise from the egg is sometimes inherent in the egg, and sometimes induced by the conditions of development, it is more commonly mixed; but may it not be the mind of the embryologist, and not the natural world, that is mixed? Science does not deal in compromises, but in discoveries. When we say the development of the egg is inherent, must we not also say what are the relations with reference to which it is inherent? When we say it is induced, must we not also say what are the relations with reference to which it is induced? Is there any way to find this out except scientific discovery?"

W. K. Brooks ('02, pp. 490–491)

It is the self-imposed task of the present compendium to review and evaluate the past and present accomplishments of the science of embryology in order more intelligently to facilitate progress into its future. The separate contributions which make up the main body of the volume must necessarily concentrate on particular fields of investigation. It is the purpose, therefore, of the first two chapters to provide a general background against which these more special subjects may be considered. Out of convenience, rather than from logical necessity, these two chapters will concern themselves first with concepts, and secondly with techniques, though the nature of the scientific method is such that these two aspects of the problem are inextricably interrelated. Arbitrarily, too, the topics chosen for discussion will be selective rather than exhaustive; since it is not possible in a few pages to do justice to even a few of the great contributors of the past, only those have been chosen whose writings are most relevant to the sequel, and even of these, many can enjoy only the barest mention.

* The writing of Sections I and II was carried out both at the Osborn Zoological Laboratory, Yale University, and at Bryn Mawr College. I owe especial gratitude to the Library of the College of Physicians of Philadelphia for the use of their collections and for generous assistance.

THE EARLY EMBRYOLOGY OF THE GREEKS: ARISTOTLE

Since it was the Greeks who performed the great *tour de force* of freeing science from magic and elevating it into the realms of pure reason, it is sensible to begin by examining a few of their contributions to embryology. They were early to develop an interest in beginnings; their very word for nature (*φύσις*, physis) according to some, including Aristotle (*Parts of Animals,* 1945 edition, pp. 74–75), implies growth, genesis or origin (*φύεσθαι*), and Anaximander, who flourished in the sixth century B.C., spoke of the *γόνιμον*, the germ or fetus of the world. They recognized early that change was an essence of existence, as we know from Herakleitos' emphasis on flux, and as is evident from their mythological conception of cosmos evolving from chaos. And from the beginning they compared cosmos to the organism, witness Plato (*Timaeus,* [1944] edition, p. 117):

> Its composing artificer constituted it from *all* fire, water, air, and earth; leaving no part of any one of these, nor any power external to the world. For by a reasoning process he concluded that it would thus be a whole animal, in the highest degree perfect from perfect parts.

But more than this, perhaps even because of it, they were able even as early as the time of Anaximander to conceive of the organism as emergent, and indeed of animals as related to man: a fragment concerning the teachings of Anaximander reads that "living creatures arose from the moist element, as it was evaporated by the sun. Man was like another animal, namely, a fish, in the beginning" (Burnet, '30, p. 70).

No attempt can be made here to enumerate the many Greek philosophers to build upon these beginnings, or to evaluate the contributions of those who did. It will have to be sufficient here to name a few, and the interested reader is referred to Balss ('36) for additional details. Suffice it here to comment that theirs was the task of the first early and perhaps random collection of data, which must precede even the primitive classification which many consider to represent the first stage of scientific inquiry.

Of some, we know only from the meager extant fragments, that they recorded what they thought to be observed fact; for instance, from Parmenides a fragment remains implying that males are generated on the right and females on the left. In the case of others, even before Aristotle, it is clear that they believed that around the observed facts they could elaborate theory. Empedokles, for example, believed the fetus to arise partly from male and partly from female semen, the children resembling most the parent who contributed most to the offspring; he spoke of the influence of pictures, statues and so forth in modifying the appearance of the offspring, of twins and triplets as due to "superabundance and division of the semen" (Burnet, '30, p. 244); he knew there was a regular sequence of events in development and spoke of the heart as formed first in development, the nails last, sowing seeds of concepts, which, right or wrong, were destined often to recrudesce in subsequent ages.

A Hippocratic treatise on generation went further in developing theories, formulating an early expression of the doctrine of pangenesis, and, relating to it, what seems to be on *post hoc* reasoning a doctrine of the inheritance of acquired characters. This treatise, before Aristotle, recognized the importance of methodology, and advocated systematic daily observation of chicken eggs: "Take twenty or more eggs and let them be incubated by two or more hens. Then each day from the second to that of hatching remove an egg, break it, and examine it. You will find," continues the writer, illustrating an apparent dependence of concept on method and inferring the great generalization, "exactly as I say, for the nature of the bird can be likened to that of man" (Singer, '22, p. 15).

Aristotle's own accomplishment was none the less impressive, for all he drew on his predecessors and contemporaries. "There was a wealth of natural history before his time; but it belonged to the farmer, the huntsman, and the fisherman—with something over (doubtless) for the schoolboy, the idler and the poet. But Aristotle made it a science, and won a place for it in Philosophy" (Thompson, '40, p. 47). And in establishing it as scientific, he set its standards higher than hitherto by far.

He followed, in embryology, the method of the Hippocratic writer *On Generation,* to perform and record most of the available observations, many in error but also many correct, thus to constitute a collection of knowledge on the development of the chick which became the foundation on which all embryology was to build; and it has been said, with much justice, of his account that "almost two thousand years were to roll by before it was to be equaled or surpassed" (Adelmann, *ed.,* in Fabricius, 1942 edition, p. 38). He concerned himself not only with the development of the chick but also with the generation of many other forms, and elaborated a kind of classification (though not in the modern sense; cf. Thompson, '40) of animal forms according to their mode of reproduction. By so doing, he both established embryology as an independent science, and he fitted embryological knowledge into a pattern larger than its own, with great clarity of vision and imagination.

On the theoretical side, he followed his predecessors by adopting a modified view of pangenesis, and concurred with them in supporting the doctrine of the inheritance of acquired characters. He broke away from his predecessors, however, in developing a new and erroneous yet highly influential concept of the relative roles of male and female in development, postulating the former as providing the form, at once formal, efficient and final cause, and the latter the substance, the material cause, for the new organism.

By thus undervaluing the egg, he paid embryology the obvious immediate disservice; but in formulating his conception of biological form as inseparable from matter he laid the way open for ultimate progress in biological science. The argument is metaphysical to the taste of the modern scientist; but Aristotle will be found not to be the last embryologist to be so tainted. We concur with his intent, after all, every time we speak of "animal forms" as a euphemism for "animal species." And Aristotle, with the natural historian's innate feeling for natural form, by envisioning form as a part of actuality rather than something above it, brought biological material to be directly investigable by the sense organs.

His theories concerning special developmental phenomena, related to his primary philosophy as they were, are deep in much of the embryological and indeed the wider biological thinking both of the past and the present. His description of the heart as the first organ of the embryo to be formed, both in time and in primacy, tied as it was to the conception of the soul as formal and final cause and of vital heat in the blood as the agent of the soul, dominated the notions not only of the developing but also of the adult circulation, and hence all physiology, through to the nineteenth century and the downfall of the phlogiston theory. His concept of organ as related to final cause epitomizes teleology, and with all the weight of Galen's authority in support still permeates much of the thought of modern biology. Matter with form inseparable from it as opposed to the more material matter postulated by Leucippus' and Democritus' atomic theory, which implied preformation, in a way made possible the whole doctrine of epigenesis, first clearly formulated by Aristotle and still central in all embryological thinking today. Form as inseparable from matter makes possible a conception of pattern emergent, an analogy of development and the *process* of plaiting a net or the *process* of painting a picture; for Plato, the Ideal mesh would have been already woven, the Ideal portrait previously complete. Aristotle (*Generation of Animals,* 1943 edition, pp. 147, 149, 225) could frame the modern question:

> How, then, are the other parts formed? Either they are all formed simultaneously—heart, lung, liver, eye, and the rest of them—or successively, as we read in the poems ascribed to Orpheus, where he says that the process by which an animal is formed resembles the plaiting of a net. As for simultaneous formation of the parts, our senses tell us plainly that this does not happen: some of the parts are clearly to be seen present in the embryo while others are not. . . . Since one part, then, comes earlier and another later, is it the case that A fashions B and that it is there on account of B which is next to it, or is it rather the case that B is formed after A? . . .
>
> In the early stages the parts are all traced out in outline; later on they get their various colours and softnesses and hardnesses, for all the world as if a painter were at work on them, the painter being Nature. Painters, as we know, first of all sketch in the figure of the animal in outline, and after that go on to apply the colours.

The metaphor will speak for itself to modern experimental embryologists. Aristotle, however, for all his natural acuity, was strangely double-minded. In his dynamic feeling for form, derived from direct study of living biological material, he was modern, and was to lead eventually straight to the inductive biology of modern times. But his conceptions of the wider Universe, based on pure reason, because statically and structurally interpreted and thus transmitted by medieval commentators, deluded posterity, and it was unfortunately the static Aristotle, the Aristotle of a sterile cosmogony, crystal clear but crystal rigid, who dominated the thought of the Middle Ages. So far as even the embryology was concerned, the Middle Ages transmitted his concepts, and occasionally amplified them, as in the case of Albertus Magnus, but devitalized them and thereby hardly improved them. Appreciation of their dynamic qualities awaited the Renaissance and later ages.

EMBRYOLOGY AND THE RENAISSANCE: FABRICIUS, HARVEY

When the Renaissance came under way it accelerated its course into the new thought by taking strength from the Greek past through all the resources of Humanism; and a "reconstruction of the Greek spirit" (cf. Singer, ['41], p. 166) was an essential part of the rebirth. Even Galileo has been called a "typical Paduan Aristotelian" in method and philosophy at least, if not in physics (Randall, cited by Adelmann, *ed.*, in Fabricius, 1942 edition, p. 55), and Whitehead ('25, p. 17) reminds us that Galileo "owes more to Aristotle than appears on the surface of his *Dialogues:* he owes to him his clear head and his analytic mind." Vesalius' interpretations of his observations were as teleological as those of Galen after which they were modelled (cf. Singer, '44, p. 81, who called him "a disciple of Galen by training, by inclination, and by his whole cast of thought"); his method, however, was also in part that of Aristotle. Copernicus, who was accused by Kepler of interpreting Ptolemy, not nature, at least challenged the Aristotelian cosmogony; Vesalius imitated the method of the Aristotle who is so rarely remembered as having written about an embryological problem (*Generation of Animals,* 1943 edition, pp. 345, 347):

> This, then, appears to be the state of affairs . . . so far as theory can take us, supplemented by what are thought to be the facts about their behaviour. But the facts have not been sufficiently ascertained; and if at any future time they are ascertained, then credence must be given to the direct evidence of the senses more than to theories.

The scientist, who customarily characterizes the Renaissance as a movement for freedom with respect to authority, often neglects to remember that it was in part from "authority" that the inspiration to achieve freedom derived.

It was Fabricius, student of Fallopius, himself a student of Vesalius, who first exhaustively applied the rigorous "new" Vesalian method of direct observation to the study of embryos, though he had many predecessors who had made isolated observations on embryonic material (among them Columbus, Fallopius, Eustachius, Arantius, Aldrovandus, Coiter et al. Cf. Needham, '34, and Adelmann, *ed.*, in Fabricius, 1942 edition, for full discussion; see also Adelmann for full critical treatment of Fabricius himself).

On the observational side, he was the first to publish illustrations based on systematic study of the development of the chick, and this, though he neglected to describe them in detail, was probably his most significant contribution. He made the way easier for the later preformationists by drawing the supposed three and four day chicks much too advanced for their normal chronological age; among his other fallacies, the most notable was his ascription to the chalazae of the role of forming the embryo. Among his improvements to the existing embryological knowledge was his emphasis that the *carina* (whose metaphysics he discussed more completely than its embryological fate) is formed before the heart, controverting Aristotle, and before the liver, taking issue with Galen in both fact and philosophy. He studied the fetal anatomy of various vertebrates, that of many mammals, including man, and presented illustrations of the comparative anatomy of the placenta, showing his special interest in the umbilical and the fetal circulation, though he devoted himself to Galenic principles in his interpretations of these. Even Fabricius, then, as late as the sixteenth century was exemplifying the conflict of the Renaissance between allegiance to authority and confidence in direct personal observations. But though in one sense his position represents an inevitable retreat, even behind the position of Aristotle, in that he emphasized the anatomy of embryos rather than the process of development, yet his work looked forward to the new embryology in the influence it exerted on William Harvey.

Fabricius' name, as Adelmann points out (*op. cit.*, p. 115) begins the first sentence of Harvey's text on generation; and Harvey, too, like his preceptor, looked back to Aristotle in his interpretations, for all that his demonstration of the circulation in method, fact, and conception, was to lead to the whole experimental and analytical biology of the future. Harvey followed Bacon's principle of explaining nature by observation and experiment, and Galileo's of measuring what is measurable and making measurable what is not. Harvey's contemporaries believed, with Fracastorius, that "the motion of the heart was to be understood by God alone" (Harvey, *De motu*, 1931 edition, p. 25). Harvey proved it to be a mechanical function. Yet he could speak of the motion of the blood, after Copernicus, Kepler and Galileo, as "circular in the way that Aristotle says air and rain follow the circular motion of the stars" (*ibid.*, p. 70) and, like a good Aristotelian, he left the vital spirits remaining in the blood. "Whether or not the heart," he wrote, "besides transferring, distributing and giving motion to the blood, adds anything else to it, as heat, spirits, or perfection, may be discussed later and determined on other grounds" (*ibid.*, p. 49). Harvey may have surmised how to treat the organ as a machine, but he was in some ways too Aristotelian to appreciate the implications of his own advanced experiment.

He was not so bound by authority, however, as to be unable to free himself from some of the old embryological errors. He refuted on an observational basis, for instance, the notion that right and left represent maleness and femaleness, and he corrected the idea of Fabricius concerning the role of the chalazae by demonstrating the *cicatricula* (our blastoderm) as the source of the embryo; he corrected, too, various specific observational errors of Aristotle. Most important, he abolished for all time the Aristotelian conception of female as substance and male as form. Galen to be sure had seemed to localize both material and efficient causes in both male and female semen, as had Fabricius after him in a confused sort of way; but it was Harvey, for all his fanciful speculation concerning the significance of fertilization, who finally elevated the egg to its full and ultimate dignity. The processes of development can obviously hardly be investigated before the object that is developing is at least defined as their residence, and Harvey's contribution here was therefore a significant one.

It is abundantly clear, however, that by egg Harvey meant something different than we do. He knew there was necessary for de-