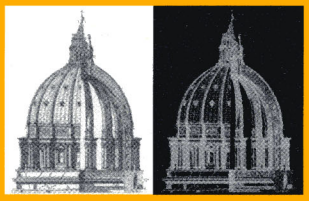
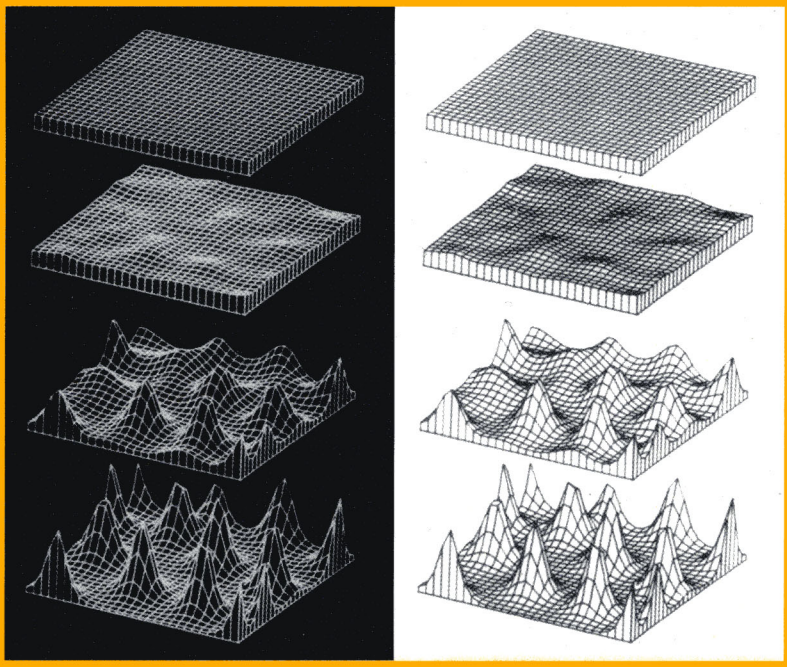
Alfred Gierer



HYDRA the model

- model for what ?



Alfred Gierer

Max-Planck-Institut für Entwicklungsbiologie

Tübingen/Germany

HYDRA the model - model for what?

Evening lecture

8th International Workshop on Hydroid Development

September 1999 - Tutzing/Germany

To begin with, I would like to thank the organizers of the meeting, Charles David and Thomas Bosch, for the kind invitation to give this evening lecture. It is intended as an essayistic talk, mingling personal, historical and - relatively few - systematic aspects. I apologize that it cannot, by any means, do justice to the scope of Hydra as a model system and to all the beautiful work that is being reported on here at our meeting.

How a physicist gets involved with Hydra - some personal reminescences

In the late sixties, two American postdocs, Hans Bode and Charles David (and, later, Harry McWilliams), as well as three German graduate students - Chica Schaller, Stefan Berking, and Ekkehard Trenkner - who were postdocs themselves not long thereafter, joined me in Tübingen in starting a project on the developmental biology of Hydra. Subsequently, other colleagues joined us to work on the project at various stages. We were intrigued by the problem of how spatial patterns are newly formed in tissues, for which Hydra regeneration is such a clear and spectacular model. We tried to push regeneration to the limit by studying the formation of animals from aggregates of previously isolated cells. We searched for activating and inhibiting factors affecting head formation and other patterning processes. Finally, we studied the regulation of stem cell renewal and cell

differentiation. There was an initial romantic phase of enthusiasm unchallenged by deeper knowledge, followed by a somewhat lengthy incubation phase before eventually results were achieved and published. Thereafter, the members of our original group were able to convert their achievements into job offers from various distinguished institutes and were very successful in developing their own projects, as seen at many Reisensburg/Tutzing meetings. For me, these meetings, with all the progress reported at them, have been some of the most delightful events in life ever since. In parallel with all this, Hans Meinhardt and I had started a program, which has continued up to the present, on theoretical biology, with particular emphasis on the formation of spatial patterns. I will say something about this a little later in this talk. Experimentally, my division in Tübingen has changed orientation since the seventies, tending more in the direction of neuroembryology.

After the emergence of our Tuebingen group in the sixties, developmental biology invaded more and more parts of our Max Planck Institute for Virus Research - to the extent that its name was changed to the Max Planck Institute for Developmental Biology in 1984. How does it come about that a virus lab mutates into a developmental biology lab? As far as I am concerned, some of the reasons for this are personal. However, the shift in emphasis, and the obstacles and controversies that accompanied it, reflect

some more general facets of biology as a science reaching far back into history. These include the balance between bottom-up and top-down approaches, holistic and reductionist concepts, the role of the parts in explaining the whole, and the relations between mechanisms and organisms.

Encouraged by Charles David, allow me to first make a few remarks of a personal nature. I studied physics from 1946 onwards, without the slightest trace of biology. There were two somewhat diffuse motivations when I was a teenager at High School that eventually lured me into physics: a fascination with physical chemistry: 'what is the physics of the periodic system and the chemical bond?' and a vague notion that Heisenberg was a genius. When rumours spread that he would settle in Göttingen in 1946, I did everything I could to enroll at Göttingen University and to find a way into his newly founded Max Planck Institute of Physics. Heisenberg was very stimulating for young scientists in several ways. He was willing to take young people seriously right at the start, though not always later. It was a very liberal style. He made it clear that the best phases in science are romantic phases such as he had experienced in the twenties, when quantum mechanics was invented and philosophical aspects of science were one of the main areas of discussion. Schrödinger's cats, half alive, half dead, roamed the institute.

My thesis advisor, Karl Wirtz, although a nuclear physicist, was also fascinated by the physical foundations of biology as a result of his time in Berlin; Timofejef was there. He confessed to me that, if he were young, he would not work on nuclear reactions but would study proteins. I worked on protons jumping across hydrogen bonds, vaguely motivated by the importance of such bonds in proteins, the substance of life as most of us still thought at that time. I got hold of Linus Pauling's book "*The Nature of the Chemical Bond*," and there I found my new hero, especially after I heard him lecture on the newly discovered alpha helix in 1952.

After getting my PhD in Göttingen, I was lucky enough to be awarded a Fulbright fellowship, though not lucky enough to be assigned to Caltech where Pauling worked. Instead, I got assigned to MIT because I was a physicist and not a chemist. Altogether, it was a rewarding year. After returning to Germany, I worked at the newly founded Max Planck Institute for Virus Research in Tuebingen, which turned out to be an ideal place for a young physicist interested in biology. At the time Tobacco Mosaic Virus became a model system for studying basic biological processes, including the role of nucleic acids as genetic material and the mutations induced in them by nitrous acid. In fact, the fifties and the early sixties were the romantic decade of molecular biology (often compared, for good reasons, with the golden twenties when quantum physics was developed), when so many

exciting facts emerged from laboratories in distant places, often within months rather than years. The TMV work got me an early appointment as head of a division of molecular biology in our Institute; I continued to work in this field for a few years, the next topic being polyribosomes. By 1963, the central dogma of molecular biology was established: DNA makes RNA and RNA makes proteins. Fine, but what next? Perhaps even: now what?

Such questions encouraged a radical change in my research interests and lead to my taking up work on the developmental biology of multicellular organisms. This field was not yet fashionable in the sixties. It had an old-fashioned flavour, said to be frustrating by lack of specificity of effects, such as those of Spemann's organizer. To be sure, we had at first the attitude that it might now be up to us molecular biologists to tell those embryologists what their field was all about, with all their fuzzy notions of morphogenetic fields, polarity, competence, gradients and whatnot. But very soon our wonder at the marvellous, if holistic and phenomenological, world of embryology reached the highest levels, and I became particularly intrigued with what is perhaps the most holistic of the problems, the generation of spatial patterns. Anyhow, it became more and more obvious that it was up to us molecular biologists to become a little more modest and, first of all, to learn from them, the development people.

Experts representing various biological systems were invited to the Institute and the advocate putting the case for Hydra was Werner Müller, who worked in the Zoology Department at Tuebingen University. We decided on Hydra and, soon after that, Richard Campbell, visiting us from Irvine, was wading into muddy ponds all over the Schoenbuch forest to search for good specimens. Howard Lenhoff sent us Kanaev's book on the history and biology of Hydra that he had edited, with the most warm-hearted wishes for our new group, and Pierre Tardent provided us with *H. attenuata* and also with generous advice - altogether most gracious support for our start.

What followed I have already told you, with one exception: the theoretical biology of pattern formation. The central question is, how can physical laws and processes account for the *de novo* production of spatial patterns in cells and tissues starting from near-uniform conditions? Obviously, a full explanation requires knowledge of the molecules involved, but this would not be sufficient. Even a complete list of all these molecules would not in itself explain the resulting spatial structure, say, of a mouse. In general, pattern formation is a systems' feature. A cloud is condensed water, a snowflake is frozen water, H_2O; there are no mysteries about the molecules involved and yet this fact is not enough to make any of us understand the form of clouds or the beauty of snowflakes. Ultimately, it is a combination of material knowledge and systems theory that is required. Likewise,

investigating biological pattern formation is a two-way process - bottom up starting from interactions of molecules and cells, and top down starting from phenomena, such as patterns and proportions. And since the two approaches are often correlated with different outlooks of the scientists involved, not excluding their philosophical and metaphysical ideas, it is not surprising that their coming together may be retarded by psychological obstacles.

Mechanisms, organisms and the origins of developmental biology in the 18th century

Since different mental attitudes of this type can be traced far back into the history of science, let me spend a few minutes following such historical traces at this point. In a sense, Aristotle may be considered the founder of biology as a science, because it was he who first postulated that reproduction and metabolism - and not features such as breathing - define life. And he took life processes, with their holistic and goal-directed features, as a model for physics as a whole; here was harmony, not conflict between physics and biology. Only in modern times, when Galileo, Kepler and Newton laid the foundations of modern physics with mathematical laws governing the movements of bodies, did the relation between mechanisms and organisms, between the living and the non-living world, become such very

challenging and puzzling problems. Only then did the question arise: how can a physics developed exclusively from studies in the inorganic world claim validity for all events in space and time, which do, after all, also contain living organisms? Is the living body just a machine, as Descartes postulated, with perhaps some vague allowance for effects of the soul mediated in man by a small part of the brain, the pituary gland? Is the seemingly new formation of the organism in each generation just an illusion, whereas actually it preexisted even in the egg? Does this mean that all future generations of an organism are contained in the body, like the ever-smaller Russian dolls within dolls? Is there nothing but unfolding of preexisting, invisibly small structures? If so, this would imply that there is no real development at all and then, of course, no developmental biology and no developmental biologists either.

It appears rather strange to us that this doll-within-doll concept was a dominant theory in the 18th century, propagated especially by Bonnet and Haller. The alternative is that there is real epigenesis, as Aristotle had postulated 2000 years earlier; but if this is so, does it require new physical laws or extraphysical concepts? Yes, was the assumption of one of the early pioneers of modern developmental biology in the 18th century, Caspar Friedrich Wolff. His PhD thesis of 1759, entitled "Theoria generationis", strongly supported *de novo* generation. There is only one extant picture of

Caspar Friedrich Wolff (Fig. 1). Characteristic of his thorough experimental research is a beautiful drawing of the 36 h chick embryo he observed with a microscope and reported in his dissertation in 1759 (Fig. 2). Wolff insisted that the visible features and sequences of pattern formation during chick embryogenesis were inconsistent with the unfolding of preexisting structures. Unfortunately, it was the dogmatic preformationist Haller who dominated the scientific establishment at the time, making life difficult for Wolff. Eventually he accepted an invitation from Catherine the Great to become a member of the newly founded Russian Academy in St. Petersburg. Only after the turn of the 18th century was Wolff rediscovered, especially by Goethe, who wrote about "our outstanding compatriot, whom a dominant school with which he could not agree had driven out of his native country."

Figure 1

Caspar Friedrich Wolff (1734-1794), pioneer of developmental biology

Fifteen years before Wolff presented his "Theoria generationis", in 1744, Trembley had published his discoveries on the regeneration of Hydra: an animal dissected into two pieces will develop into two complete animals. These findings were refined by Reaumur, who showed that virtually any small seemingly uniform section of body tissue developed into a complete polyp, and by Roesel von Rosenhoff, the editor of a Journal called *'Insekten-belustigungen'*, *'Amusement about Insects'*, an 18th century version of popular journals of the type of "Scientific American". In this journal, in 1755 Roesel himself described his discovery that random aggregates of the tiniest pieces of Hydra tissue he could obtain by cutting polyps would eventually regenerate into multi-headed Hydra monsters.

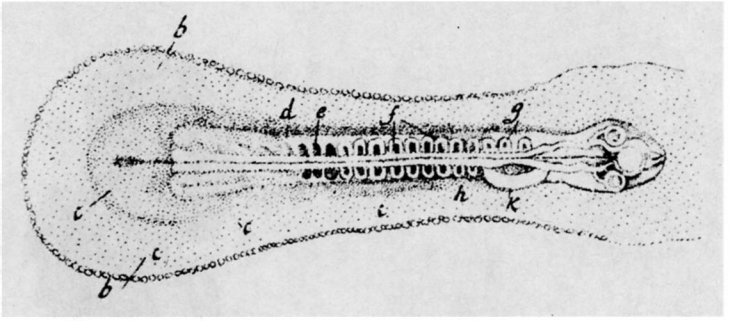


Figure 2

Early stage of chick development, as reproduced from the dissertation by Caspar Friedrich Wolff "Theoria Generationis", Halle 1759.

The impact of Trembley's discovery sounds almost unbelievable in our times. It is described in Kanajew's book on Hydra, in the edition prepared by Howard Lenhoff that I have already mentioned. In the 18th century, regenerating polyps were talked about in literary, philosophical and theological circles. Embassies considered it their duty to keep their governments informed about progress in Hydra research, and an observer declared that the discovery of polyp regeneration and that of electricity were the two outstanding achievements of the 18th century. But even with less enthusiasm, giving modern scientific scepticism its due, it is hard to overlook the importance of Trembley's work. He can be considered the first experimental developmental biologist in the history of science. His marvellous book makes good reading even today, which can hardly be said of most other works of his time. And his attitude towards science was also most remarkable. In the last pages of his book he writes that

"what we actually know is still very little in relation to the innumerable wonders of nature. The best method to understand known facts is to discover new facts. Nature is to be understood with the help of nature, not by our preconceptions which are too limited for grasping such great objects of research as a whole."

MÉMOIRES
POUR L'HISTOIRE
DES POLYPES.

QUATRIÈME MÉMOIRE.

Opérations faites ſur les Polypes, & les ſuccès qu'elles ont eu.

LA première opération, que j'ai faite ſur les Polypes, a été de les couper *transverſalement*. On a vu en general au commencement du premier Mémoire *, quel en a été le ſuccès. J'ai renvoié à celui-ci le détail de cette Expérience. * Pag. 13. &c.

POUR

Figure 3

Abraham Trembley (1710-1784), with two pupils, doing experiments on Hydra. The vignette is on the first page of the fourth memoir in Trembley's centennial book on polyps *"Mémoires pour l'histoire des polypes d'eau douce"* of 1744.

Fig. 3, taken from Trembley's book of 1744, shows him as a private tutor with two pupils doing experiments on the polyps. By the way, Trembley did not go on working on Hydra for long; he changed fields and became a prominent author on child development and education.

On the physical basis of biological pattern formation

Thus, Caspar Friedrich Wolff and, even more, Abraham Trembley left us with the question of how the generation of patterns in biological systems, i.e. the development of spatial order by internal processes within cells and tissues, can be explained on a physical basis. Many scientists, up to the time of Spemann in the thirties, had thought or guessed that this might not be possible at all. A new physics, or some extraphysical principles might be required. However, we now know that this is not the case. Combinations of rather conventional molecular interactions and movements, even passive movements by diffusion, are good at pattern generation. This was discovered by Turing in 1952. He designed and discovered equations for reaction-diffusion systems that generate spatial concentration patterns starting from near-uniform initial distributions. His deduction was based on Fourier methods, that is on the analysis of destabilization of uniform distributions by concentration waves of certain wave lengths. Thus, normal chemical reactions in liquid media are able to generate concentration

patterns. Does this have biological significance for morphogenesis? To answer this question it is necessary to explore conditions for pattern formation in molecular terms and, most important, to explain the impressive self-regulatory features of developing biological systems, such as proportion regulation - the adaptation of the size of a part to the size of the whole. With these aims in mind, Hans Meinhardt and I proposed in the seventies a theory of pattern formation based on two concepts: autocatalytic activation and lateral inhibition. Our starting point was a line of thought originally introduced into the field of pattern formation by Kuffler and by Kirschfeld and Reichardt in our neighbouring Institute for Biological Cybernetics.

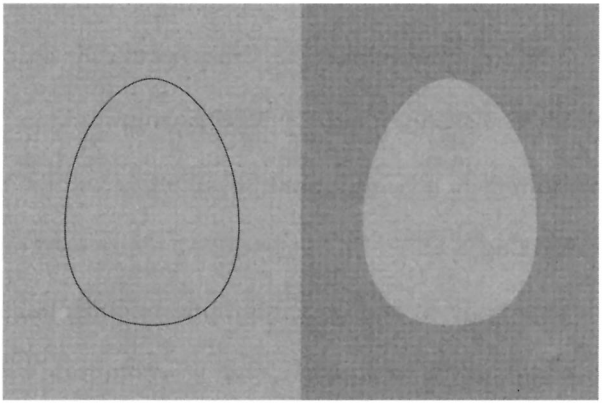


Figure 4

Lateral inhibition in pattern recognition. Drawing an egg means drawing the contour of an egg (left) which is abstracted from the primary image of the egg on the retina (right) by mechanisms involving lateral inhibition.

The key concept is lateral inhibition. Let us draw an egg (Fig. 4 left). Does our drawing really represent an egg? No, it is the contour of an egg. The image of a real egg on the retina looks different (Fig. 4 right). To obtain the contour, the local intensities projected onto the retina are processed there by local activation in conjunction with an inhibitory effect extending into the environment of activation. Inside the bright area, high inhibition cancels high activation, while inside the dark area, low inhibition cancels low activation. But at the edge, high activation is not cancelled because the inhibition extending from the neighbouring dark area is low. Therefore the edge is enhanced and the contour of the egg is outlined. This mechanism is well established in both psychophysical and neurobiological terms, and it shows an intriguing general feature: it allows the generation of striking patterns, starting with very shallow ones. For instance, our eye can recognize the edges of areas that are very slightly less grey than the areas surrounding them.

We applied the principle of lateral inhibition in modified form to pattern formation in the course of development, now of course using molecules interacting and moving instead of neurons firing. Our theory shows that patterns are formed by local self-enhancing reactions which are controlled, spatially limited, and disciplined by a wider ranging inhibitory effect: The

ranges of activation and inhibition are defined by the mean distance between production and decay or removal of a molecule. Starting from an initially near-uniform distribution, local activation is self-enhancing, but activation at one location can proceed only at the expense of de-activation elsewhere. Non-linear interactions are required for the generation of reproducible, stable patterns. Power laws for the order of reactions can be introduced to analyse general conditions for which type of systems would generate patterns and which would not; one of the simplest ones has been used by us as a model, but the general conclusions would apply to other versions as well.

Hydra regeneration is a particularly clear example of *de novo* pattern formation and helps to get the logic straight. What do we want to explain? Any isolated section of the body column regenerates an animal with head and foot (Fig. 5). The *orientation* of the pattern is determined by the polarity of the regenerating tissue. We attribute this to the slope of a source gradient extending across the tissue from head to foot. However, the absolute value of this graded cue cannot determine whether and where a head is formed because the same part of the body column with the same local level of the polarity-defining graded source can lead to formation of a head, or a foot, or nothing, depending on how the section is cut. In other words, the pattern formed is oriented by previous polar cues. But, aside from that one single bit of information - deciding on orientation to the left or to the right - the

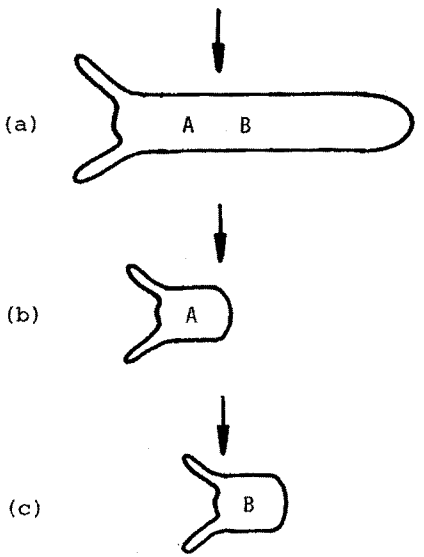


Figure 5

***De novo* pattern formation in Hydra regeneration**. Any section cut from the gastric column of Hydra regenerates an animal with head and foot. Thus, the same part of the body column (arrow) may produce nothing (a), a foot (b), or a head (c), depending on whether and where the section is cut. It follows that no pre-existing local property of the tissue (such as a polarity-defining gradient determining the orientation of regenerates) can *per se* decide whether and where a head is formed; this can be decided only by the formation of a new morphogenetic gradient after the onset of regeneration.

pattern itself, namely a head-activating morphogenetic gradient, is newly formed. It is such internal *de-novo*-pattern generation that the interaction of activation and inhibition is capable of explaining, as shown by computer simulations (Fig. 6). The resulting pattern is self-regulating and is a product of molecular interactions and movements within the initially near-uniform tissue; it requires no dolls-within-dolls, however hidden. And this type of mechanism gives rise to the striking regulatory features that are so characteristic of biological development, ensuring reliability despite complexity. In particular, details of initial conditions don't matter. Regeneration is possible, as is induction, inhibition and, under certain conditions,

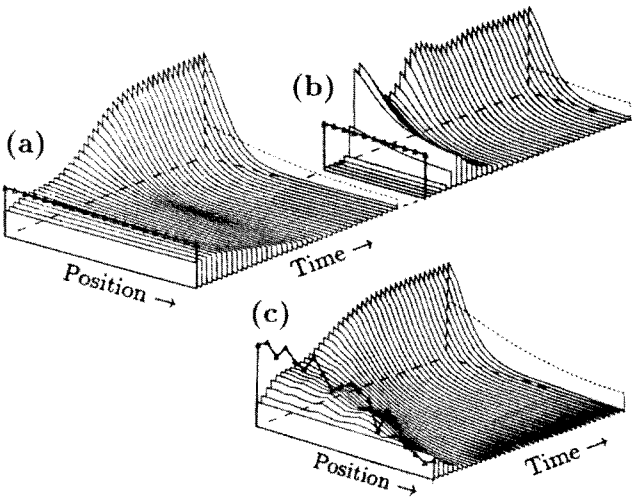


Figure 6

Pattern formation by autocatalysis and lateral inhibition explains characteristics of developing biological systems, in particular self-generation of patterns and their robustness against distortions, as shown by these computer simulations: (a) A striking pattern (that is a morphogenetic gradient specifying positional information, for instance for head formation in Hydra) is formed starting from near-uniform initial distributions. (b) A section cut from (a) regenerates a new pattern. (c) Different, rather bizarre initial conditions lead to essentially the same graded distribution as in (a). Thus, only the orientation of the pattern is determined by initial conditions, whereas its form is self-regulating, and the pattern-forming mechanism is capable of correcting distortions to a considerable extent.

proportion regulation. Not only gradients but also symmetrical and periodic distributions, stable or pulsing in time can be generated in this manner. It is, of course, not the purpose of this lecture to summarize the theory that was often a topic, especially in Hans Meinhardt's talks, in the Reisensburg/Tutzing meetings.

I would like to mention only a few aspects here. One is the relation of our activator-inhibitor approach, which is directly linked to biological developmental regulation, to Turing's Fourier-type stability analysis, i.e. to the detection of spatial wave lengths of distributions towards which the uniform distribution is unstable. It can be shown that the mathematical content of the lateral inhibition approach and Turing's criterion are very similar; but the demonstration is by no means straight forward. It takes time and patience to prove. Indeed, the lateral inhibition rules can be shown mathematically to be the *only* mode of pattern generation for the simplest two factor case.

But what about pattern formation in systems with more of two variables? This, after all, is the biologically most likely case: feedback loops, for instance, consisting of a chain of reactions. Consider schemes of, say, seven or ten reactions. What is now our criterion for pattern formation? We might think of collecting those compounds that have autocatalytic effects and analysing them for themselves, but this leads nowhere. For instance, activation can result just from inhibition of inhibition, allowing for pattern formation even if there is not a single directly activating reaction. The more useful approach is a different one: apply the lateral inhibition concept from the outset. Begin the analysis by sorting the molecules involved into short-range ones on the one hand, and those subject to wider distribution in the

tissue on the other. Then, check whether the short-range subset, taken together, is in itself autocatalytic as a system, and whether the long-range molecules prevent an overall autocatalytic explosion. In this way, the concept "pattern formation by the interaction of activation and inhibition" can be generalized into multi-component systems with activation and inhibition as features of subsystems rather than of individual substances. And yet, the basic regulatory capabilities that characterize biological development are maintained. Applied to Hydra it means that a satisfactory physical explanation of *de novo* pattern formation would require detection of those molecules that operate more locally, in order to demonstrate the autocatalytic features of this subset, as well as the detection of inhibiting effects of the long range components. In view of exciting new results, such as those on *brachyury* and *wnt* genes, it appears that such a consistent physical account of *de novo* pattern formation may not be out of reach for research on Hydra.

Now, after perhaps a bit too much mathematics and systems theory for an evening talk, let us relax and turn to more general issues. What is the relation between the activation-inhibition type of biological pattern formation and other processes of generating structures? Self-enhancement is involved in many systems, in the formation of crystals and sand dunes, for instance, and of galaxies and stars. It is involved in socioeconomic processes, such

as the formation of towns, and in psychological processes - success generates success, frustration leads to even more frustration. It might be involved in the generation of socioeconomic inequalities. And, there may be unexpected relations between pattern formation and pattern recognition, beyond edge enhancement.

Fig. 7 shows a multiple peak pattern formed following initiation by random fluctuations. Though the resulting pattern is irregular, it is not truly random; it shows a "granularity" by avoiding small distances due to the effects of

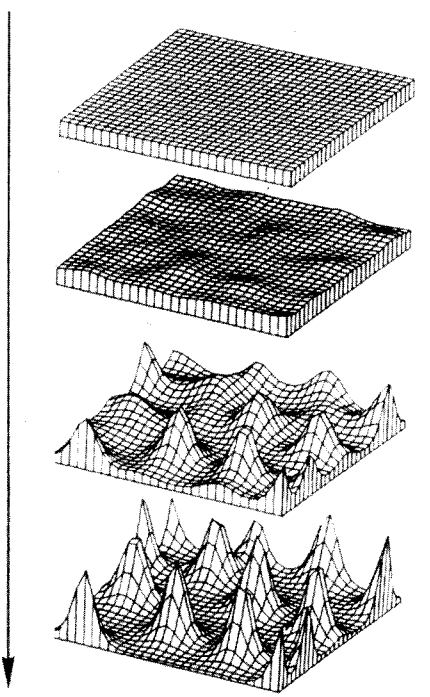


Figure 7

Formation of multiple peak patterns. Even if initiation of pattern formation occurs by random fluctuations, the pattern of peaks formed, though irregular, is not completely random. Due to lateral inhibition, small distances between peaks are avoided, giving rise to a 'granular' texture.

lateral inhibition. A biological example of such textures is the surface of the Fugu-fish (Fig. 8a), a potentially poisonous Japanese delicacy for gourmets who live by the rule "no risk, no fun". Bela Julesz has found that this texture or 'granularity' is subject to immediate perception, i.e. it is recognized directly by the pattern recognition system of our brain. This can be seen by looking at Fig. 8b: the granularity in the center excludes small distances while in the outer areas the average density of spots is the same but with a truly random distribution. You recognize immediately, without conscious thought, that the texture in the center is distinct. In much more general terms, the

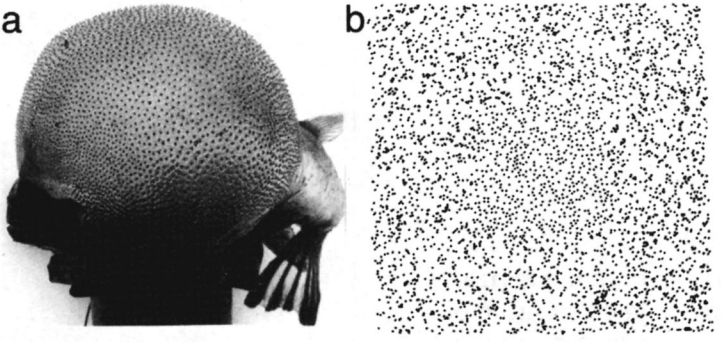


Figure 8

The surface of Fugu-fish (a) shows the 'granular' texture corresponding to the model calculation of Figure 7 for pattern formation by activation and lateral inhibition. This type of texture (center of Figure b) is recognized immediately by the pattern recognition system of our brain and distinguished from truly random distributions with the same average density (outer parts of Figure b).

relationship between mechanisms of pattern formation and the aesthetics of patterns is illustrated in Hans Meinhardt's book *'The algorithmic beauty of sea shells'*, explaining the development of beautiful visual patterns by formally beautiful developmental mechanisms.

On morphogenetic fields, real form and the role of mathematics

Pattern formation, as I have discussed it up to now, concerns invisible concentration patterns of morphogenetic substances. How do they elicit morphogenesis proper, that is, the generation of real form? Real form means curvature, for instance, that resulting from evagination of initially flat cell sheets as in the case of budding Hydra. Cytoskeleton, intercellular junctions, cell surface molecules, and intercellular matrix, all have their roles. Again, some general systems' aspects may be helpful in studying and understanding the processes involved.

To begin with, in terms of physical cell interaction, it is not that difficult to envisage a cell sheet evaginating upon local activation; the more difficult issue to grasp is why there is a stable cell sheet in the first place (Fig. 9). If cells like contact with other cells, they clump. If not, they segregate. A cell sheet, however, is a compromise, and this requires non-linear interactions beyond Steinberg's schemes. Cells that form sheets do like contact between cells, but not too much of it; they want to see the waterfront as well. And

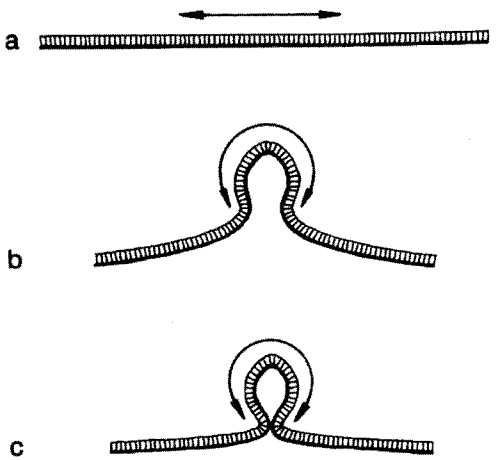


Figure 9

Stability and evagination of cell sheets. Stability requires non-linear cellular interactions which prevent clumping as well as dissociation. Activation of part of the sheet by a morphogenetic field (double arrow) may cause bending moments, curvature and thus evagination of the sheet.

cells are polar, giving rise to cell sheets differing in and near their two opposing surface areas. Once the conditions of cell sheet stability and polarity are met, however, almost *any* local morphogenetic signal would change bending moments, thus giving rise to local tissue evagination. And this applies also for multiple sheets made up of ectoderm, mesoglea, endoderm. Thus, understanding the stability of this arrangement in the first place goes a long way towards understanding morphogenesis.

There is a non-molecular aspect of evagination that I would like to mention briefly - the intricate interaction of the curvatures of the two surface dimensions upon evagination. We have adapted and applied shell theory for computer models of such processes. In Fig. 10 computer simulations are shown of rotationally symmetric evagination or invagination of cell sheets, induced by a spot of activation causing a local bending moment to arise there. Shell theory is the theory used by architects to construct thin curved concrete roofs covering wide areas; the difference is that in our biological applications the emphasis is not on tangential forces but on bending moments, which architects avoid like hell because they would cause their roofs to collapse.

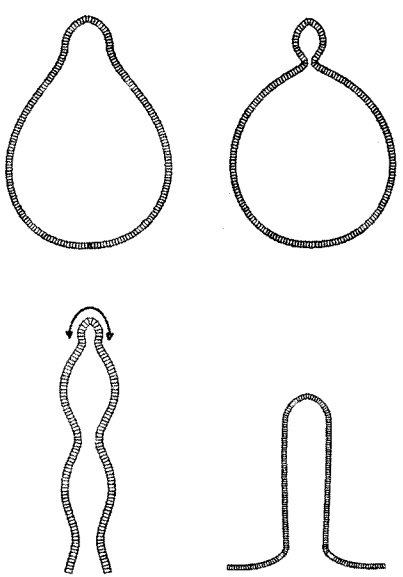


Figure 10

Generation of form, modeled on the basis of shell theory. Activation of a central area of the sheet is assumed to induce bending moments and evagination. The figure shows computer simulations of sections cut along the axis of rotationally symmetric structures.

And this reference to shell theory brings me back to an interesting historical episode I would like to recount. The first application of mathematical mechanics to curved roofs was in the middle of the 18th century, about the time Hydra regeneration was discovered by Trembley. At that time, dangerous cracks appeared in the cupola of St. Peter´s Cathedral in Rome. Methods of saving the building were proposed, involving clumsy reinforcements of the walls or removal of the beautiful lantern that Michelangelo had placed on top of the cupola. At this stage, Pope Benedict XIV asked three mathematicians in one of his monasteries to look at the problem from a theoretical mathematical point of view. They proposed the addition of an iron chain around the base of the cupola, calculated to bear a load of at least 110 tons. This was done and it saved the building. The nice picture (Fig. 11) is taken from the original article "Parere di tre mattematici ..." of 1742. Historically, this was the beginning of constructional engineering based on modern physics. However, of even greater interest in the context of my talk is the psychology accompanying the discussion of which proposal to impliment. The "tre mattematici" were defensive. The three monks wrote: "We express our apologies to those who do not only prefer practice to theory but consider practice as exclusively adequate, theory perhaps as detrimental" - but then they ask for forgiveness in the case of St. Peter because of its huge and unique dimension. Their opponents, however,

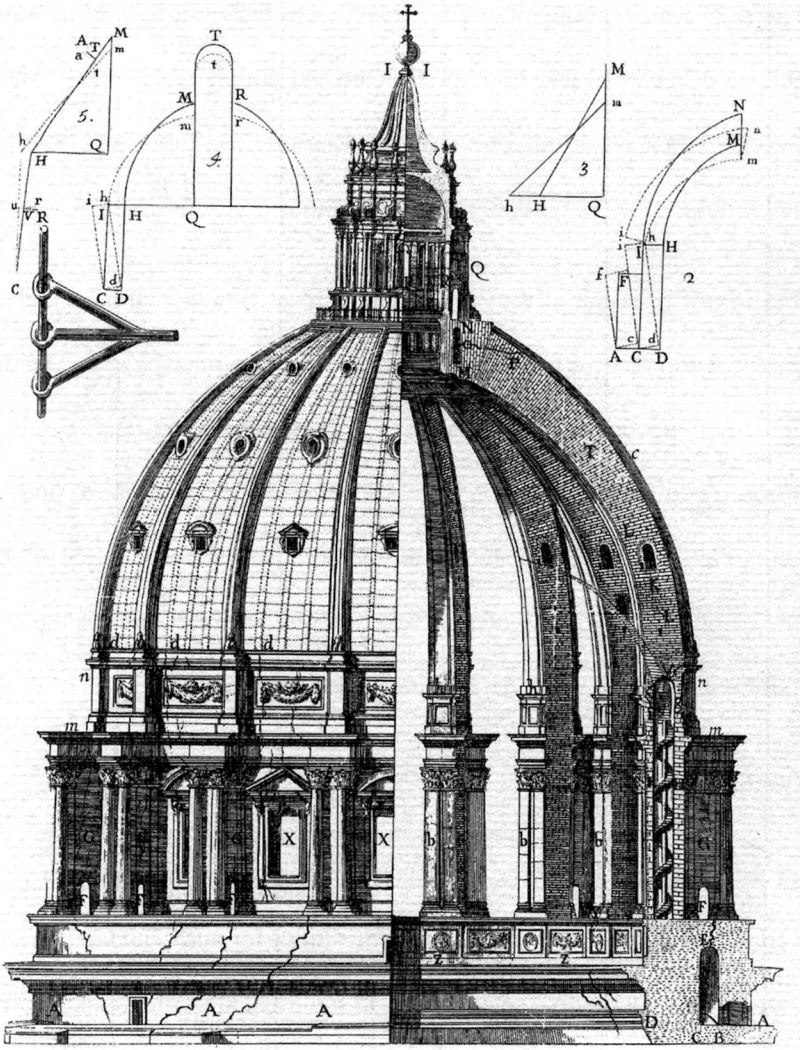


Figure 11

St. Peter´s Cathedral 1742: Damage in the base of the cupola, as shown in a reproduction of the original drawing in 'Parere di tre mattematici...'. The three monks were pioneers of constructional engineering and forerunners of shell theory (see Figure 10). Their mathematical analysis lead to successful measures for saving the cupola.

were aggressive: anonymous pamphlets were distributed in Rome saying that since Michelangelo had constructed the cupola of St. Peter without mathematics it would also be possible to repair it without any help from mathematics and from those mathematicians.

This is not to suggest that a serious conflict between theoretical and empirical approaches exists nowadays in developmental biology. We don't hear anything about anti-mathematics pamphleteering. But, there are rumours of a milder form of reluctance: stop reading a paper at once if a mathematical equation is encountered. What the story about St. Peter's tells us, in any case, is that scepticism about reasonable applications of mathematics can be a transient phenomenon.

Hydra - model for what?

Let me now come back to our main topic: what is the future of Hydra research in general? Of course, nobody knows for sure, but we can make more or less educated guesses. One of the most important insights in the last twenty years of developmental biology has been the realization that the basic molecular mechanisms underlying the development of multicellular organisms must have been 'invented' very early in evolution and can be found throughout vast domains of the animal kingdom, though differing in how highly modified and elaborated they are. Therefore, research on Hy-

dra, which is itself such an unwilling organism for genetics, can draw on the molecular genetics of other organisms, e.g. *Drosophila,* while still taking advantage of the fact that Hydra is one of the most puristic models that exists for certain basic processes of development such as of *de novo* pattern formation and tissue evagination. This includes the two approaches I have discussed: one aims at understanding the mechanisms underlying head determination and regeneration in Hydra, looking for autocatalytic subsystems and loops, while the other relates tissue evagination to physical parameters of cell interactions and movements. There are further basic features and mechanisms presented at our meetings, for which Hydra is a good model; forgive me for not discussing them all. In particular, as shown in the first days of our present meeting, Hydra is an interesting and revealing model for early evolutionary events.

Let me recall in this context, a talk by Andrew Spencer in our Tutzing meeting four years ago, on features of the neural network of Cnidaria as an indication for early stages of the evolution of nervous systems in the animal kingdom. Neural networks process information mainly by electric signals transferred across and conducted along membranes. Basic inventions of such mechanisms were made by single cells. Calcium channels allowed for the rapid transmission of electric stimuli, to be converted rapidly into biochemical and behavioural responses; calcium, after all, is capable of throwing

metabolic switches in the cell. But when it comes to neural networks, requirements for optimizing information processing change: signals now have to be conducted across many cells and they are not supposed to throw major switches in the cells on their way. They do so, if at all, at the end of processing. Ca^{2+}, however, is good neither for rapid signal conduction nor for safeguarding metabolic stability, whereas Na^{+} is suitable for both. It seems that in this context evolution has introduced (or at least elaborated) Na^{+} channels. Cnidarians seem to represent - this is the line of thought of Spencer and Hille - interesting models for evolutionary intermediates between single cells and higher brains.

Now, since my talk here is vaguely reminiscent of an elder statesman's speech, I think it should end with some remarks on foreign policy, more specifically the relationship of the Hydra community to outside groups working in biology. Hydra research is a small field. If you search in the internet according to Current Contents for articles in the last five years with "hydra" in the title or keywords, you get 319 responses. About seventy of them refer to some huge astrophysical object in outer space and a few other items, and some 250 to our cute little animal. If you enter "mouse" instead of "hydra", you get 66 526 papers! Per capita, I think the scientific results of the small Hydra community may very well compete with those in other fields. What is important, however, is that the results are noticed by

the developmental and evolutionary biologists' groups at large and not just by those working on Hydra. Therefore, above average efforts are worthwhile to make our work known to other subcultures of biology as well as to take notice of theirs, not only by representations at general meetings but also on the Internet. It helps biology, helps others and helps those in our own field when applying for grants and positions. So let me conclude here, with a fervent plea for *extrovert behaviour* by Hydra researchers.

Notes: My talk is reproduced here virtually unchanged and therefore contains no references. Our early work on Hydra is summarized in my articles *'Hydra as a model for the development of biological form'* in Scientific American 231 (1974), pp. 44-54, and in *'Biological features and physical concepts of pattern formation exemplified by Hydra'*, Curr. Top. Dev. Biol. 11 (1977), pp. 17-59. The second article contains many references. Our first publications on the theory of pattern formation by local activation in conjunction with long-range (lateral) inhibition were A. Gierer und H. Meinhardt, *'A theory of biological pattern formation'*, Kybernetik (continued as Biological Cybernetics) 12 (1972), pp. 30-39, and H. Meinhardt and A. Gierer, *'Applications of a theory of biological pattern formation based on lateral inhibition'*, J. Cell Sci. 15 (1974), pp. 321-346. It is elaborated and reviewed in H. Meinhardt's book *'Models of biological pattern formation'*, Academic Press, London /New York (1982), and in my article *'Generation of biological patterns and form: Some physical, mathematical, and logical aspects'*, Progr. Biophys. Molec. Biol. 37 (1981) pp. 1-47. This article also contains on pp. 21-24 a demonstration that the set of general rules underlying the lateral inhibition approach is mathematically nearly equivalent to Turing's criteria of destabilization of uniform reaction-diffusion-systems by Fourier waves. The extension of the lateral inhibition theory to more than two components with activation and inhibition representing systems'

features rather than properties of individual molecules is described in the quoted article on pp. 26-33.

As for the historical episodes the story of the origin of developmental biology in the 18th century and the pioneering role of Caspar Friedrich Wolff, author of the dissertation *'Theoria Generationis'* (1759) in Halle, a center of enlightenment in Germany, is treated in Shirley A. Roe's excellent book *'Matter, Life and Generation. 18th century embryology and the Haller-Wolff debate'*, (1982), Cambridge University Press. With respect to the history of research on Hydra I would like to refer to E.E. Kanaev's (1952) book on *'Hydra'*, translated into English and edited by H.M. Lenhoff (1969). For those working on Hydra, it is most rewarding to look into A. Trembley's beautiful original book *'Mémoires pour servir à l'histoire d'un genre de polypes d'eau douce à bras en forme de cornes'*, Jean & Herman Verbeek, Leide (1744). As for the story of the origins of modern constructional engineering, including shell theory in connection with the repair of St. Peter in the middle of the 18th century, I would like to refer to the book by István Szabó *'Geschichte der mechanischen Prinzipien'* (1976), Birkhäuser, Basel/Stuttgart. There, the story of the discussions about damage and repair of St. Peter including the work of the *'Tre mattematici'* is told and nicely illustrated. In particular, the book contains a reproduction of the drawing of the cupola of St. Peter included in this paper.