Int. J. Dev. Biol. 40: 89-92 (1996)

The origin of developmental physiology of plants in Germany

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Eine Frage, vor deren Angriff jeder zurückschreckt, wird nie zur Lösung gelangen. *Hofmeister 1867.*

In the middle of the nineteenth century the pioneering research of Wilhelm Hofmeister (1824-1877, Professor in Heidelberg and Tübingen) put the ontogenesis of plants in the centre of botany in Germany. The comparison of the alternation of generation of mosses, ferns, gymnosperms and angiosperms provided so much important, new information that no further ground was necessary to establish ontogenesis as a central field of plant research (Hofmeister, 1851). However, for the highly analytical intellect of Wilhelm Hofmeister, it was clear that the description of the formation of the adult plant could not satisfy the mind, if it were not possible also to describe the causal factors playing a role in the direction of development or morphogenesis (Bopp, 1994). In the second part of the first volume of his "Handbuch der Physiologischen Botanik", edited by himself (Hofmeister 1867), that deals with the general morphology of plants (Gewächse), he discussed the following example: On the vertical shoots of the chestnut the leaves are in five rows; however in the horizontal growing branches there are only two rows. In an experiment he could show that the position of the leaves and the number of rows is determined by gravity.

From this simple experiment Hofmeister raised the question of how far external forces have an influence on the final appearance of a plant. And he continued: "Selbstredend ist bei der Untersuchung der Beeinflussung der Gestaltung der von ihrer Umgebung in hohem Grade abhängigen, dazu auch dem Experiment leicht sich unterwerfenden Pflanzen eher ein Erfolg zu erhoffen, als bei der gleichen Untersuchung an Thieren." This sentence means: To study the effect of external factors on development it is easier and more successful to do the experiments with plants than with animals.

"The forces", said Hofmeister, "which determine the form of developing parts of plants are completely unknown, particularly those which are responsible for the specifically different, heritable constant formation processes." This means that Hofmeister clearly recognized that potentialities exist in the organism which combine with external influences to determine the direction in which it develops.

In a few very detailed, but almost descriptive chapters, Hofmeister explained the effect of gravity (the deviation of the plumb line), the effect of light or illumination and finally the consequences of penetration of parasites into host plants. The particular examples he used for gravity were the positions of leaves within a bud, and for light, the effects that we call today phototropism.

Repeatedly Hofmeister mentioned that experiments are necessary for final decisions, and only with experimental approaches could the effect of external factors be shown unequivocally. However he performed only few and very simple experiments, and accentuated at the end of the chapter: "At the present time we are not able to guess at the causes that determine the direction of branching, the position of leaves ... during the growth of plants."

The influence of Hofmeister on the following generation of scientists must be regarded as very important. They performed the

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0214-6282/96/$03.00

first steps towards establishing developmental physiology in Germany.

Also the influence of the two most eminent German plant physiologists in the nineteenth century should not be forgotten: Wilhelm Pfeffer (1845-1920, Professor in Bonn, Basel, Tübingen and Leipzig) and Julius Sachs (1832-1897, Professor in Bonn, Freiburg and Würzburg). However the details of their work are not discussed here because developmental physiology is only a quite peripheral aspect of the eminent work of both concerning general physiology.

Further advances in developmental physiology came from the physiological experiments on the formation of organs in plants, summarized in a booklet written by Hermann von Vöchting (1847-1917, Professor in Basel and Tübingen) and dedicated to Wilhelm Pfeffer (Vöchting, 1878). In its introduction Vöchting explained that the aim of the book is "to investigate by which forces, internal as well as external, the position of newly formed roots and shoots is determined during regeneration and how the same forces influence already existing parts of the plants." This sentence shows the direct relationship to Hofmeister's views mentioned above. Vöchting asked: "What are the real causes", and he argued that "this is not a question of the flow of saps within the plants" – because Julius Sachs had criticized the experiments of Vöchting, and suggested that all the results could be explained by two substances, both distributed in different ways within the plant. The direction of the flow should depend on the previous outer conditions. The consequence is that the shoot forming substance went upwards and the root forming substance downwards. These 'specific organ forming substances' were, however, hypothetical, and were never identified. Therefore they do not contribute to the understanding of further experiments.

In a short chapter of a second volume (Vöchting, 1884), Vöchting answered the objections of Sachs. This chapter is such a clear and comprehensive summary of his theoretical background that we can use it to understand his arguments.

The central part of Vöchtings research work included quite simple but very instructive experiments on isolated willow branches, which regenerate in a strongly polar manner according to the polar structure of the branch. Figures based on the results of these experiments have been reproduced in all German textbooks since then. Vöchting said: "As soon as the branch is separated from the tree, 'special forces' arise. The effect of these forces is different in the apical and basal part of the plant." Therefore the experiments showed that the formation of roots and shoots on isolated branches depends firstly on internal causes and only secondly on external ones, such as gravity, light, etc.

The internal causes are expressed as "polarity of the isolated shoots". It is important to note, that the "special forces" have nothing to do with vitalism; Vöchting emphasised that physical laws are valid for all the processes in question. One of the objections of Sachs was that long lasting external conditions could be responsible for the state of the internal factors – taking for example his "specific substances", gravity determines the direction of the flow of these substances.

This argument was settled by another simple experiment of Vöchting . The branches of the weeping willow behave exactly like those of the normal willow although their tips have been growing downwards for long enough to change the flow of "specific substance" by gravity.

Vöchting refused in all his papers to define the character of the 'internal forces', independent of the fact that it was impossible at this time to characterise substances chemically or physically. He said: "Whether it is possible to characterise the unknown structure of the plant organism or not is less important, if we hold strongly in mind that the law of conservation (Erhaltung der Kraft) must be valid!"

Vöchting included also the "heritable structures" amongst the internal forces. According to his arguments all internal factors are based on the physico-chemical structures of the cell. Therefore it is impossible to distinguish between the two groups, the variable and the invariable structures. "Both groups of forces are separated only by the fact that the first group is known (polarity) and the second one is unknown."

Georg Klebs (1857-1918, Professor in Basel, Halle and Heidelberg) was the first to recognize the necessity of separating both components, to understand really the interaction between outer and inner factors, and he also substantiated this theoretically in a long and painstaking work (Bopp, 1985). With this work (Klebs, 1903) he became the real "father of developmental physiology" in Germany. He started his experiments with algae (Klebs, 1881), which appear as polymorphic forms of one species. The question was whether the forms became separate species by modification dependent on external factors, or whether the modifications disappeared as soon as the external factors were changed.

This discrimination is only possible if the original material is uniform and cultivated in controlled, pure conditions. Obviously to solve this problem pure cultures had to be grown under defined external factors and then the development had to be studied in detail and over time. This component, time dependence, was first studied by Klebs. His experimental work on the extremely variable mosses, different algae and some fungi provided the base for his book "Willkürliche Entwicklungsänderung bei Pflanzen" (arbitrary change of development in plants) (Klebs, 1903). After this he concentrated most of his experimental work on the succulent and very variable species *Sempervivum funkii* (Klebs, 1913, 1918).

For this species the arbitrary changes of development concern the formation of a rosette, the elongated shoot axis, the shape of the leaves, the formation of flowers etc. With many different variations of the culture conditions Klebs was able to produce nearly every form of development he wanted.

From these experiments he concluded, that the development of plants is not fixed by a rigid and invariable process, but that heredity determines "only" the form of reaction to external conditions. However, to be able to reach a definite state, particular essentials are needed.

With this concept, Klebs discovered several, for the future of developmental physiology important things: the dependence of particular developmental events on lightning conditions. His examples included the transition from the filamentous moss protonema to the leafy moss shoot, and, more important, the different patterns of growth adopted by fern prothallia in blue and red light (Klebs, 1916). These experiments signal the origin of the important field of photomorphogenesis in the twentieth century. Another essential step was the discovery of "Blühreife" (ripeness to flower). Klebs had the opinion that a definite relation between carbon and nitrogen is the prerequisite for this ripeness, and without the ripeness plants are not able to flower (Klebs, 1918).

The most important consequence of the concept of 'ripeness to flower' was the discovery of the dependence of flower formation on day length. In a recent book about photoperiodism, Vince-Prue (1975) has mentioned this important result: "Georg Klebs was carrying out carefully controlled experiments on flowering in *Sempervivum funkii.* He succeeded in inducing the rosettes to flower in the middle of winter by giving a few days of continuous illumination from incandescent lamps; non-illuminated rosettes always remained vegetative. He concluded that *'in nature flowering is probably determined by the fact that from the equinox (21st March) the length of day increases ... and ... when it reaches a certain length flowering is initiated. Light probably acts as a catalytic rather than a nutritive factor.'* Klebs thus recognized that flowering could be accelerated by long days."

Klebs can be regarded as the discoverer of photoperiodism, although he never expressed this in a general way, demonstrated by Garner and Allard (1920) in USA after his death. The same is true for the dependence of flower-formation on cold-treatment in perennial plants. Also in this case the general rule was described by other scientists. The conclusion that external factors have a catalytic character - nowadays we would say the function of a "signal" – and not a nutritional function was first expressed by him and marks a very big step in the field of plant physiology.

All these details are important enough to preserve the name of Klebs as one of the pioneers in developmental physiology, but more important are his efforts to find clear definitions to understand experimental results (Klebs, 1913): How do external factors interact with the heritable substance that he called "specific structure" ("spezifische Struktur"). For this purpose "mediators" are necessary, and he recognized that these mediators are, or can be, the "internal factors". They react directly with the external factors; however, they also can be changed or modified by them. The group of inner factors activate or inactivate the "specific structure".

This knowledge appears repeatedly in the research work of Klebs in many different variations, and it is expressed very clearly in the following sentence: "The real fundamental fact is the dependence of the inner factors on the outside world: through this alone can the outside world influence specific structures, and thus determine which potentialities are brought to realization, and which remain hidden." ("Die fundamentale Tatsache ist die Abhängigkeit der inneren Faktoren von der Außenwelt, dadurch allein wird ermöglicht, daß die Außenwelt auf die spezifischen Strukturen einwirken kann, daß sie bestimmt, welche Potenzen zur Verwirklichung kommen, welche verdeckt bleiben"). This means, the inner factors are on one side the points of impact of the external factors, and at the same time the transmitters of the changes to the "specific structures", which themselves are constant. Therefore one has to assume that these structures are something definite, though Klebs knew that with time also the specific structures have to be changed by mutation to allow the evolution of species.

It seems clear that this concept of Klebs, which can also explain the controversial discussion between Sachs and Vöchting (Vöchting, 1884), about the effect of outer conditions on the growing plant, is the base for all recent ideas of the regulation of developmental processes by external factors like light, temperature, gravity, water, but also hormones etc.

At this time Georg Klebs was the only botanist engaged with these fundamental concepts. One of Klebs' contemporaries was Karl von Goebel (1855 - 1932, Professor in Rostock, Marburg and München). First he was interested in the more descriptive aspects of plant development, extensively demonstrated in his "Organographie der Pflanzen". However, his book "Einleitung in die experimentelle Morphologie der Pflanzen" (Goebel, 1908) placed him among the fathers of developmental physiology of plants. He wanted to comprehend and describe the facts that are important for the *living* plant and in the first chapter of his book he notes that the morphological description refers preferentially to 'dead bodies' (Leichen) of the plant, as preserved in the herbaria! "Mere everyday observation shows us however, that this can in no way provide a full understanding of the relationships of structures within any particular plant." ("Schon die alltägliche Beobachtung zeigt uns aber, daß man damit keineswegs eine vollständige Kenntnis der Gestaltungsverhältnissse einer bestimmten Pflanzenart erhalten kann.").

At first he characterized the function of experimental morphology and demonstrated this with many examples, mostly quite simple to perform, which explain the basic laws of developmental processes of higher plants. He examined the influence of external factors on leaf-shape, the branching of main and lateral axes, and the processes of regeneration. And he examined the consequences of polarity, the possibility of passing over particular phases, and finally the reversibility of developmental steps. All his results are summarized in the following sentence: "The particular states of development are dependent on different internal conditions, which are under the influence of the surrounding world." This sentence is however, in comparison to the definitions of Klebs, a simplification. It is not much more than a description of the facts, while Klebs' definition can be regarded as trend setting and stimulating for further research work.

Georg Klebs was not the founder of a school of developmental physiology. However, his influence on the further progress of the field is obvious. Botanical developmental physiology after Klebs did not, maybe, reach the same high standard as in zoology, where H. Spemann (1869 - 1941, Professor in Rostock, Berlin and Freiburg), received the Nobel prize for his research work in 1935. Nevertheless the general suggestions of Klebs on botanical developmental physiology remain without doubt. Only few cases should be mentioned, where the interaction between nucleus and plasma is demonstrated, something that can be regarded as the continuation of the question about the relation between the specific structure and the internal factors. In this context we have to remember the important research work of Fritz von Wettstein (1924) on the moss *Funaria hygrometrica*, the papers of Richard Harder (1927) about the function of nuclei in hybrid mycelia of higher fungi, the very fascinating and still stimulating experiments of J. Hämmerling (1934) with the unicellular giant alga *Acetabularia*, and finally the reciprocal breeding experiments of F. Oehlkers (1938) with unifoliate species of the genus *Streptocarpus*.

All these papers, published in the twenties and thirties of this century, are steps along the parts to our recent concept of developmental physiology, which allows the study of the switching on and off of genetic information by different types of signals (internal as well as external), responsible for externally regulated morphogenesis.

A summary of our knowledge of developmental physiology and the main German contributions to this knowledge up to 1950 is given by Erwin Bünning (1906-1990, Professor in Königsberg, Köln and Tübingen) who has contributed by his own research work many important details in his fundamental textbook "Entwicklungsphysiologie der Pflanzen" (Bünning, 1948). This book (appeared in three editions) can be regarded as a certain conclusion of the "classical developmental physiology". Therefore we will finish this short chapter with a definition from Erwin Bünning: "Developmental physiology itself regards the genetical constitution as a given fact, and it is asked how the real course of processes within plants can be explained from the interaction of the genetical constitution with their surroundings. We can see that a clear border between inner and outer factors cannot be drawn. For example, if we analyze the process of restitution, one starts with inner conditions, which are produced by the interaction of inheritance and environment."

References

BOPP, M. (1985). Georg Klebs. In *Semper Apertus-Sechshundert Jahre Universität Heidelberg. 1386-1986.* Vol. III. Springer Verlag, Berlin, pp 73-96.

BOPP, M.(1994). Beiträge Heidelberger Botaniker zum Fortschritt ihrer Wissenschaft. 1833-1933. *Heidelberger Jahrbücher Bd. 38.* Springerverlag, Heidelberg, pp. 77-98.

BÜNNING, E. (1948). *Entwicklungs- und Bewegungsphysiologie der Pflanzen.* Springer Verlag, Berlin.

GARNER W.W. and ALLARD, H.A. (1920). Effect of the relative length of day and night and other factors of the environment on growth and reproduction in plants. *J. Agric. Res. 18:* 553-606.

GOEBEL, K. (1908). *Einleitung in die experimentelle Morphologie der Pflanzen.* B. G. Teubner, Leipzig.

HÄMMERLING, J. (1934). Über formbildende Substanzen bei *Acetabularia mediterranea*, ihre räumliche und zeitliche Verteilung und ihre Herkunft. *W. Roux'Arch. Entw.Mech. Org. 131:* 1-81.

HARDER, R. (1927). Zur Frage nach der Rolle von Kern und Protoplasma im Zellgeschehen und bei der Übertragung von Eigenschaften (nach mikrochirurgischen Untersuchungen an Hymenomyceten). *Z. Botanik 19:* 337-407.

HOFMEISTER, W. (1851). *Vergleichende Untersuchungen der Keimung, Entfaltung und Fruchtbildung höherer Kryptogamen.* F. Hofmeister Verlag, Leipzig.

HOFMEISTER, W. (1867). Die Lehre von der Pflanzenzelle. In *Handbuch der Physiologischen Botanik*, Vol. 1 (Ed. W. Hofmeister). Wilhelm Engelmann Verlag, Leipzig.

KLEBS, G. (1881). Beiträge zur Kenntnis niederer Algenformen. *Bot. Zeit. 39*: 249-257.

KLEBS, G. (1903). *Willkürliche Entwicklungsänderung bei Pflanzen. Ein Beitrag zur Physiologie der Entwicklung.* Gustav Fischer Verlag, Jena.

KLEBS, G. (1913). Über das Verhältnis der Außenwelt zur Entwicklung der Pflanzen. Sitzungsberichte Heidelb. Akad. Wissensch., Jahrgang 1913.

KLEBS, G. (1916). Zur Entwicklungsphysiologie der Farnprothallien. Sitzungsberichte Heidelb. Akad. Wissenschaft Jahrgang 1916, 4. Abhandlung.

KLEBS, G. (1918). Über die Blütenbildung in *Sempervivum. Flora 11*: 128-151.

OEHLKERS, F. (1938). Bastardisierungsversuche in der Gattung *Streptocarpus Lindl. Z. Botanik 32:* 305-393.

VINCE-PRUE, D. (1975). *Photoperiodism in Plants.* McGraw Hill, London.

VÖCHTING, H. (1878). *Über Organbildung im Pflanzenreich.* Erster Teil. Verlag Max Cohen und Sohn, Bonn.

VÖCHTING, H. (1884). *Über Organbildung im Pflanzenreich.* Zweiter Teil. Verlag von Emil Strauss, Bonn.

WETTSTEIN, F. von (1924). Morphologie und Physiologie des Formenwechsels der Moose auf genetischer Grundlage. *Z. Ind. Abst. und Vererbungslehre 33:* 1-236.