Wir gehen jetzt zur ausführlicheren Beschreibung des vordersten Abschnittes des Mitteldarmes über. Ich bemerkte schon, dass hier die Darmwand zwei seitliche, geschwulstähnliche Ausstülpungen bildet; wir beobachteten sie von außen auf Fig. 1 bei *Md 1*, wo sie auf der abgebildeten Seite wieder in zwei sich trennende Massen zerfallen; jede Masse besteht ihrerseits aus kleineren gerundeten Ausstülpungen, die Zahl, Größe und Verteilung aller dieser Ausstülpungen ist unbeständig. Wenn wir die Wand dieses Mitteldarmabschnittes auf Schnitten untersuchen, so sehen wir, dass sie ebenfalls fast ausschließlich aus einer einfachen Epithelschicht besteht, welche noch dicker ist, als bei dem benachbarten Mitteldarmabschnitte. Hier (Fig. 4) unterscheiden wir auf den ersten Blick zwei Zellenarten des Epithels, welche sich sowohl durch ihre Form und Größe, wie auch durch ihren Inhalt schroff unterscheiden. Erstens sehen wir große annähernd isodiametrische Epithelzellen (Fig. 4, *f*), welche auf Flächenschnitten der Wand, wie auf Fig. 5, *f*, gerundete Umrisse zeigen und deren Inhalt grobkörnig erscheint, zweitens andere Zellen, welche in Bezug zu den ersteren als Stützzellen bezeichnet werden können (Fig. 4 u. 5, *st*), und welche typische Epithelzellen darstellen, ganz ähnlich den Epithelzellen des medianen Streifens und der übrigen Mitteldarmabschnitte. Auf Querschnitten der Darmwand (Fig. 4, *st*) erscheinen sie als hohe gebogene Zellen; wenn sie gruppenweise getroffen werden, wie auf der genannten Figur in der Mitte, so sieht die Gruppe auf dem Schnitte bikonkav aus, also die Gruppe erscheint in der Mitte gleichmäßig eingeschnürt. Bei Untersuchung von Flächenschnitten der Darmwand (Fig. 5) sehen wir, dass die „Stützzellen" ein Reticulum bilden, dessen Fenster einzelne körnertragende Zellen einschließen. Das Protoplasma der „Stützzellen" ist feinkörnig, es enthält auch Vakuolen, wie bei den Epithelzellen der übrigen Mitteldarmabschnitte, obschon nicht immer; die Zahl, sowohl wie die Größe der Vakuolen, ist auch hier eine inkonstante. Nicht alle „Stützzellen" sind so groß, wie die soeben beschriebenen und nicht alle reichen von der einen Oberfläche der Darmwand bis zur anderen; wie in den übrigen Mitteldarmabschnitten, giebt es auch hier kleine Kryptenzellen, welche die gleiche peripherische Lage und denselben Charakter haben; der abgebildete Schnitt (Fig. 4) hat sie nicht getroffen, dennoch sind sie leicht zu beobachten und haben ein eben solches Aussehen, wie auf Fig. 2 und 3.

(Schluss folgt.)

The Chromoplasts and the Chromioles.

By Prof. **Gustav Eisen**,
California Academy of Sciences, San Francisco.

Through the kindness of Dr. F. Doflein of the Zoological Museum of München, I have had the privilege of receiving a copy of Professor

Richard Hertwig's paper: „Ueber die Bedeutung der Nucleolen" (Separatabdr. aus den Sitzungsber. der Gesellschaft für Morphologie und Physiologie in München, 1898, Heft 1). The conclusions arrived at by the author interest me the more because they are in some points almost identical with those of my own, the latter being the results of a study of the spermatogenesis of *Batrachoseps attenuatus*, a batrachian common in California. A preliminary account of these investigations will appear this winter in the „Biological Bulletin", the forthcoming new organ of the Biological Station at Woods Holl.

The very earliest large cells in the testes are the polymorphous spermatogonia. They are not only the largest cells in the testes but are also the most interesting. The nucleus of this cell is polymorphous as regards form; that is, the nuclear wall is deeply folded without any apparent irregularity. In this stage of the cell we can not speak of chromosomes as they do not exist in any form, shape, or manner. What we see is a large number of minute granules of even size and shape, and all capable of being stained intensely by the iron-haematoxylin method. By a proper afterstaining with congo-red, we find that there are other granules, small and large, and which are of a distinctly different nature from the dark-staining ones. By this method we also demonstrate without any difficulty the two kinds of nucleoli, as well as the various structures of the cytoplasm. The first-mentioned granules are the chromatin elements which in time will form the chromosomes. For these granules, which are of constant size, form and number, I have proposed the name chromioles. In the polymorphous spermatogonium these chromioles are scattered along the nuclear membrane and are connected with each other by threads of linin, like strings of pearls. The chromioles are perfectly round, and uniform in size. In the earliest stage of the spermatogonium they lie isolated from each other, later on, under certain conditions, they are grouped in threes. If we now turn to the other elements in the nucleus we find them to be as follows:

The linin granules already referred to, which vary in size only apparently, some being placed or distributed singly while others are bunched together. Their function is to connect the chromioles with each other, as well as to connect and support the chromosomes after the latter are formed.

With the method employed as stated above, the chromioles stain black or grey, while the linin granules stain brick red. The larger elements in the nucleus belong to two distinct structures known as nucleoli, true nucleoli and net-knots.

For reasons that will appear later on, and also for the sake of clearness, I have proposed to call the net-knot chromoplast, while for the true nucleolus I suggested the name of lininoplast.

The chromoplast stains intensely dark, just like the chromioles, while the lininoplast stains like the linin granules. There may be one or more chromoplasts, and there may be several lininoplasts. The lininoplasts are nearly always round and are never seen in division, while the chromoplasts are often seen to be in state of direct division. They may be either round or oblong, even or contracted in the centre. In the early stages the chromoplasts lie entirely free from the chromioles, surrounded only by a radiating halo of linin-threads. As far as mitosis is concerned the cell-stage just described is the one of absolute rest. The various constituents of the cell are at this stage simply in a state of metabolism, preparing themselves for the future work of mitosis. The first sign of an ushering in of the latter consists in a movement of the chromioles. These are apparently attracted by the chromoplast, and we soon see the chromoplast surrounded by a number of strings of chromioles, for which strings I have proposed the name of leaders. These leaders approach the chromoplasts from all sides and connect with it.

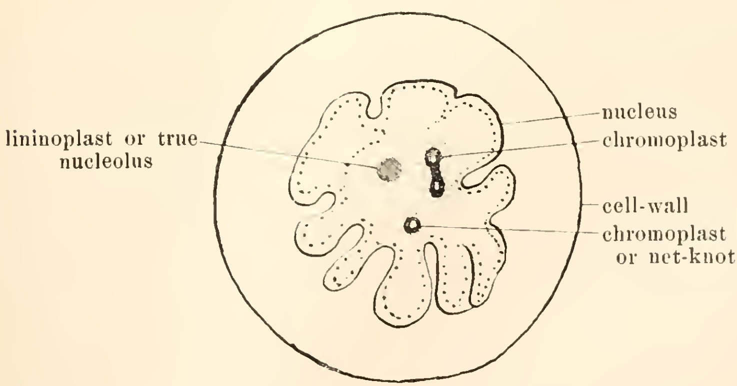
In the beginning, the number of leaders may be variable, but later on we find that there are as many leaders as there are to be chromosomes in the fully developed nucleus. At the end of this stage we find in *Batrachoseps* twelve leaders attached to one or two chromoplasts. All the chromioles are now connected to form leaders none being left free in the nucleus. In the early stage we find that each leader consists of a single row of chromioles, more or less surrounded by a thin film of a distinct and particular chromoplasma — probably derived from the chromoplast -- and strung together by a thread formed of linin granules.

As the leaders develop we find that the chromioles group themselves by threes, thus forming incipient chromomeres. The leaders now begin to contract more and more, and at the end of this stage we find in the nucleus, which by this time has become perfectly round, one or more chromoplasts connected with twelve leaders made up of a constant number of chromomers, each chromomere containing three chromioles. In the next stage of the nucleus the chromosomes become differentiated in the following manner:

The leaders split lengthwise, but the distal ends remain connected with, or rather are held together by the chromoplast. The splitting of the leader is caused primarily by the splitting into two of each chromiole and chromomere. Immediately preceeding the splitting of the leaders the chromomeres each contain six chromioles. This number is constant, and from now on in every fully developed chromomere we shwall always find six chromioles. An account of the process of the formation of the chromosomes is outside of the scope of this paper, but has been fully set forth in the preliminary report mentioned above.

It must suffice to state here that in time the leaders separate from each other, but to each leader, which now may be termed a chromosome, remains attached a part of the original chromoplast. It may naturally be asked how do we know that such is the case. The answer is this: The chromoplast can nearly always be recognized by its structure. It always contains certain highly refractive granules, which are seen in the chromoplasts no matter in what stage of development the nucleus may be. These granules are the landmarks by which the chromoplasts may always be recognized. Thus, when the chromosomes are in the equator of the cell, the chromoplast is seen in one side of the centre in the ring-like chromosome. The subsequent division of the chromosome is through a simple equation, and passing through the chromoplast, gives each new or daughter-chromosome an equal share of the chromoplast. The chromoplast is now situated at the end of one of the limbs of the horse-shoe-shaped chromosome. The next stage is an umbrella or ring-like nucleus. This stage, far from being caused by imperfect fixing methods, is an absolute necessity, as being the only means by which the chromoplasts can change place. Let us understand this clearly. Before the chromosomes enter the ring-like nucleus the chromoplast is found at the very point of one of the limbs. When in the next cell — the spermatocyte — the chromosomes reappear, we find that the chromoplast has changed its position. It is no longer found at the apex of one of the limbs, but is seen to be located at the very angle where the two limbs meet. This change of locality takes place in the ring-shaped nucleus, in which the chromomeres and chromosomes loose their individuality, the chromioles and chromoplasts remaining.

1.

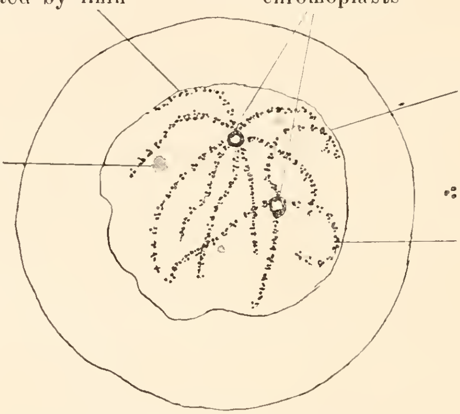


1. Polymorphous spermatogonium. The black, dark granules are the chromioles. The light colored granules are the linin granules, here represented as a thread only. The cell is in perfect rest. Only a few chromioles are indicated.

I have found the chromioles and the chromoplasts in all cells capable of mitosis which I have investigated, and I have come to the conclusion that they are constant elements in all cells. The leucocyte is of almost identical structure with the polymorphous spermatogonium. We find in it the scattered chromioles, and among them a varying number of chromoplasts, the latter of larger size and more intense staining quality.

The lininoplast, or true nucleolus, is, according to my conclusions, principally a storage reservoir of linin granules, and has somewhat the same relative influence on the linin granules as the chromoplasts have on the chromioles.

2.



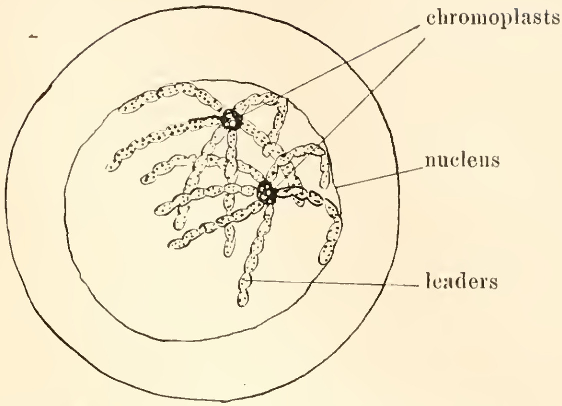
2. Polymorphous spermatogonium. Two chromoplasts connected with 12 leaders or incipient chromosomes. Each leader with chromomeres containing 3 chromioles each. Three lininoplasts.

Again, regarding the chromioles:

These minute bodies are the only constant parts of the chromosomes. The formation of the chromosomes is due to an independent process, so to say, presided over by the chromoplast, and in every respect independent of the centrosomes. The mitosis as a whole is the result of two parallel but independent processes, which run parallel for a time, but touch at certain points and aid each other in accomplishing their mutual ultimate purpose. In other words, the chromoplasts prepare and form the chromosomes, the centrosomes accomplish their proper mitosis.

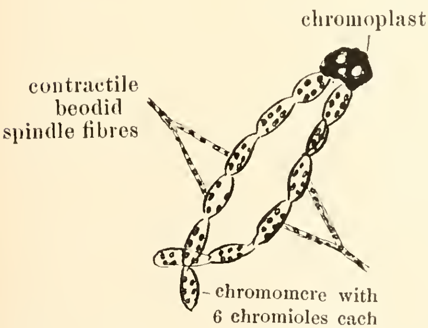
The number of the chromioles is constant in every species. In *Batrachoseps* the perfect chromosome contains six chromomeres, three in each limb of the chromosome. Each chromomere contains six chromioles, which makes for each chromosome 36 chromioles, and for the whole nucleus 432 chromioles.

3.

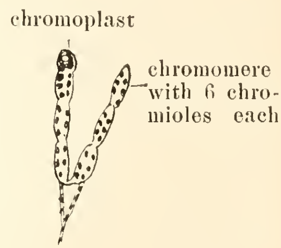


3. The two chromoplasts are connected with 6 leaders each. Each leader is made up of 6 chromomeres with 6 chromioles each. Linin and lininoplasts not figured.

4



5



4. Split chromosome just before being seperated into two doughter chromosomes.

5. A chromosome just after separation from its sister chromosome. Chromoplast attatched to the limb. Beaded contractile fibre at the angle of the two arms.

Finally, a word regarding the technique. It is useless to endeavor to study these minute bodies without the aid of the very best optical appliances possible, and with the most careful treatment of the material. A Zeiss — or equally good — Apochromate of 2 mm, with an Aperture of 1.40, is an absolute necessity. A lesser apperture will not suffice. Moreover an oil immersion condenser in the substage must be used, the common Abbe condensers do not give a sufficiently perfect light to enable one to see the necessary details. And lastly, the ordinary daylight is not suitable, being neither sufficiently pure, nor steady enough to always give the same results. In these investigations I have used exclusively the achromatic light-filter described in the „Zeitschrift f. wiss. Mikroskopie", Bd. XIV, p. 444. The fixative used was the iridium-chloride-acetic, described in the same volume, pages 195—196.

Diagrams.

1. Polymorphous spermatogonium. The black granules are the chromioles. The lighter granules are the linin granules, in the diagram represented as a thread. This cell is in a state of perfect rest, only a few of the chromioles are figured. Chromoplasts, or net-knot. Linino-plast or true nucleolus.

2. First maturation cell. Two chromoplasts connected with 12 leaders or incipient chromosomes. Each leader is made up of incipient chromomeres containing three chromioles each.

The Chromioles are supported by linin. Lininoplasts. Chromoplasts. Nucleus. Leaders.

3. The same cell as in diagram 2 in a more advanced stage. The two chromoplasts are connected with 6 leaders each. Each leader is made up of 6 chromomeres containing 6 chromioles each. Linin and lininoplasts are not figured. Chromoplasts. Nucleus. Leaders.

4. A chromoplast to which is attached a split chromosome, just before separation of the two daughter chromosomes in the equatoreal stage. The chromosome which originally consisted of a leader with 6 chromomeres has split lengthwise, the chromioles have doubled and the two halves are now being pulled appart by the beaded contractile fibres of the spindle. Contractile beaded, spindle fibres. Chromoplast with refractive granules. Chromomere with 6 chromioles.

5. A daughter chromosome immediately after separation. The chromoplast is attached to the apex of one of the limbs. The chromosome is V-shaped and consists of 6 chromomeres each one with 6 chromioles. At the junction of the two limbs is seen attached the beaded contractile fibre. Chromoplast. Chromomeres with 6 chromioles each.

28. Oct. 1898. [14]

Prof. Gilson's „Cellules musculo-glandulaires".

Von Dr. **J. Ogneff** in Moskau.

In einem der letzten Hefte der bekannten Zeitschrift „la Cellule" (Bd. XI VI. fasc.) giebt Prof. Gilson eine sehr interessante Beschreibung des Baues der von ihm gesehenen Zellen, welche die Leibeshöhle des Wurmes *Owenia fusiformis* bedecken. Im Gegensatz zu den gewöhnlichen Befunden bei den Anneliden trifft Gilson in der Haut von *Owenia* nach innen von dem äußeren Epithel nicht zwei — die muskulöse und peritonealen Schichten an, sondern nur eine, welche aus be-