It may further be mentioned here that all the chemicals used were examined separately in the same way to ascertain whether they contained radium; this appeared not to be the case. It is further noteworthy that every solution, both the acid and the alkaline ones, were boiled two or three times, and that the values inserted in the table are the average ones of the results obtained in the different experiments. By far the greater part of the radium is found in the acid solution; it was even often, — particularly for the rocks poor in radium — not to be demonstrated at all in the alkaline liquid.

It is seen that this investigation yields a similar result as the preceding ones: the rocks from Sumatra have a same relatively high content of radium of the order of magnitude 10^{-12} gr. per gramme.

We shall not yet draw any conclusions concerning the problems mentioned in the introduction, but postpone them till a number of rocks from *Borneo* have been discussed in a following communication.

Anorg. Chem. Laboratory University of Amsterdam.

Botany. — *"On the structure of the nucleus and karyokinesis in Closterium Ehrenbergii Men."* By Prof. C. VAN WISSELINGH. (Communicated by Prof. J. W. MOLL).

(Communicated in the meeting of June 25, 1910).

While the structure of the nucleus and the karyokinesis of *Spirogyra* have been repeatedly examined, the nuclei of the genus *Closterium* have rarely been the subject of investigation. This is the more remarkable, because the nuclei attain a considerable size. The few statements made in the literature about the structure of the resting nucleus of *Closterium* chiefly amount to this that the nucleus agrees with that of other algae, especially *Spirogyra*; thus for instance DE BARY [1]) states: Ein Zellkern von der für *Spirogyra, Zygnema* beschriebenen Structur nimmt die Mitte der Desmidieenzelle ein. DE WILDEMAN [2]) says: Le noyau des *Closterium* est du même type que celui des *Cosmarium* et des *Spirogyra.* The latter also gives some particulars of the nucleus. According to DE WILDEMAN the nucleus is formed by a rounded or rectangular mass, containing a large nucleolus at its centre. The nucleus contains hardly any

1) A. DE BARY, Untersuchungen über die Familie der Conjugaten, 1858, p. 40.

2) E. DE WILDEMAN, Recherches au sujet de l'influence de la température sur la marche, la durée et la fréquence de la caryokinèse dans le règne végétal, Extrait des Annales de la Société belge de microsc., t. XV, 1891, p. 47 and following.

chromatin, while the nucleolus stains very deeply, no matter what reagent is used. It is remarkable, that DE WILDEMAN in his investigations of living material, came to results somewhat different from those obtained with fixed material. In the living material he found considerable variation. In some cases the nucleolus was rounded, as in the fixed material, in other cases, however, the central mass was of a granular substance, and missed definite shape. DE WILDEMAN could often distinguish small globules, which were separate or united. The number of these small bodies decreased by fusion.

The accounts in the literature of the nuclear division of *Closterium* are as scarce as those of the structure of the nucleus. Some investigators, especially FISCHER [1]), HAUPTFLEISCH [2]) and LÜTKEMÜLLER [3]) have examined the division of *Closterium* in detail, but their investigations refer almost exclusively to the cell-wall. Several investigators, including FISCHER [4]) and DE WILDEMAN [5]) have directed attention to the division of the chromatophores, which begins before or during the nuclear- and cell-division, and to the movement of the daughter-nuclei along the cell-wall to the places where the chromatophores are divided into two. FISCHER further mentions that several nuclear bodies occur in the daughter-nuclei.

The most important data concerning the nuclear division have certainly been furnished by KLEBAHN [6]). His investigations refer to the germinating zygotes, in which he observed the union of both nuclei to one, the mitotic division of this nucleus into two equal daughter-nuclei and the subsequent mitosis of these daughter-nuclei, which by that process each produce two unequal nuclei. The figures of KLEBAHN clearly show that the nuclei divide by mitosis, and that in this division spindle formation takes place. The nuclear- and cell-division of the vegetative cells was not examined by KLEBAHN.

It is evident from the above summary of the results of various authors, that our knowledge about the nuclear structure of *Closterium* is still very incomplete. The accounts of some investigators of the resemblance of the nuclei of *Closterium* to those of

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[1]) A. FISCHER, Ueber die Zellteilung der Closterien, Bot. Zeitung, 1833, N⁰. 14, p. 225.

[2]) P. HAUPTFLEISCH, Zellmembran und Hüllgallerte der Desmidiaceen, Inaugural-Dissertation, 1888.

[3]) J. LÜTKEMÜLLER, Die Zellmembran der Desmidiaceen, Beiträge zur Biologie der Pflanzen (Cohn), VIII. Bd., 1902, p. 347.

[4]) l. c. p. 226, 232 and 233.

[5]) l. c. p. 50, 51 and 52.

[6]) H. KLEBAHN, Studien über Zygoten, I, Die Keimung von Closterium und Cosmarium, Pringsheim's Jahrb. für wiss. Botanik, XXII. Bd., p. 420 et seq.

Spirogyra and other *Conjugatae* have especially little value, because there is not even a definite consensus of opinion among botanists as to the structure of the *Spirogyra* nucleus, which has been so frequently examined. For instance, how different are the views about the nucleolus of *Spirogyra*. Some take it as identical with the nucleoli which occur in the vegetable kingdom generally, while others regard it as a small nucleus lying in a larger one. Hence the mere statement that the nucleus of *Closterium* agrees with that of *Spirogyra* means but little. Further investigations will have to show whether the nucleolus of *Closterium* indeed agrees with that of *Spirogyra*; i. e. whether it is an ordinary nucleolus, or something corresponding to a small nucleus, or something else. The variations in the nucleus of *Closterium*, mentioned by DE WILDEMAN certainly heighten the interest in this point of investigation.

The investigations of the vegetative nuclear division in *Closterium* have brought but little to light. Thus it is not even certain, whether the nuclear division is a mitosis. This may only be thought probable in connection with the results which KLEBAHN obtained with germinating zygotes and because the nuclei also divide by mitosis into other *Conjugatae*. Whether chromosomes arise, whether a spindle is formed what changes the nucleolus undergoes, of all these cardinal points in the investigation of the karyokinesis of *Closterium* nothing is known as yet.

Many years ago I intended to examine the nuclear and cell division of *Closterium*. More than once I had to give up my attempts for lack of sufficient material, until in March and April 1910 I was able to cultivate *Closterium Ehrenbergii Men.* for some time successfully so that at last I had at my disposal a very abundant and healthy material with numerous stages of division, which enabled me to examine repeatedly all occurring stages of division.

Fixed material had to be used for the investigation because not much is visible of the nuclear division in living material, even less than in *Spirogyra*; FLEMMING's mixture was used for fixation (1 g. chromic acid, 6 g. glacial acetic acid, 0.5 g. osmic acid, 120 c.c. distilled water). To bring the nuclear figures into prominence a solution of chromic acid was used. With the help of this the cytoplasm with the chromatophores and the starch was dissolved. When this has happened the flat nuclei turn over, which is of great advantage, because it enables one to examine microscopically the same nucleus in a horizontal and in a vertical position; this applies to the resting nucleus as well as to the various stages of division. After more prolonged action the chromic acid also dissolves the nuclei, but the different parts are dissolved to an unequal extent. This circumstance may therefore also contribute to a wider knowledge of the nuclear structure. Sometimes the material was examined after it had been acted on by chromic acid for some time, washed and stained with "Brillantblau extra grünlich". I will omit the details of this method because I have already described it in an earlier publication [1]). I only wish to point out that the material has to be very carefully treated with FLEMMING's solution. By this treatment the nuclei must obtain a great resistance to chromic acid; on the other hand the cytoplasm with the chromatophores and the starch must slowly dissolve in the chromic acid solution, without contracting or losing their definite outline. For this purpose the material was fixed with a small quantity of FLEMMING's mixture and was daily examined to see whether the action had been sufficient and if necessary more of the FLEMMING's mixture was added.

In this paper the results of my investigation will be mentioned, as far as they concern the structure and the division of the nucleus. Before doing so I must briefly indicate my stand-point with respect to the different views of the nuclear structure and the karyokinesis in *Spirogyra*, for otherwise it would not be clear what I mean by such expressions as corresponding to or different from *Spirogyra*. After my last publications on the karyokinesis of *Spirogyra* I have more than once returned to the subject, not only with species about which I had written before, but also with others. In no case did these investigations raise doubt as to the earlier results. On the whole the newly examined species differed very little from those that had been examined before. The investigation of a species received from England alone led to new results of which I hope to give an account later.

Nevertheless all the species examined agree in this, that the nucleolus or the nucleoli must be regarded as small nuclei inside a

[1]) Ueber den Nucleolus von Spirogyra, Ein Beitrag zur Kenntnis der Karyokinese, Bot. Zeitung, 56. Jahrg. 1898, 1. Abt. p. 199.

Ueber das Kerngerüst, Zweiter Beitrag zur Kenntnis der Karyokinese, Bot. Zeitung, 57. Jahrg., 1899, 1. Abt., p. 155.

Ueber Kernteilung bei Spirogyra, Dritter Beitrag zur Kenntnis der Karyokinese, Flora oder Allgem. Bot. Zeitung, 1900, 87. Bd. 4. Heft, p. 356.

Ueber abnormale Kernteilung, Fünfter Beitrag zur Kenntnis der Karyokinese, Bot. Zeitung, 61. Jahrg., 1903, 1. Abt. p. 210.

Ueber die Karyokinese bei Oedogonium, Sechster Beitrag zur Kenntnis der Karyokinese, Beihefte zum Botan. Centralblatt, Bd. XXIII, 1908, Abt. 1, p. 138, 139, 148 et seq.

large one, a view which agrees with the opinion of CARNOY [1]), who first drew attention to the interesting structure of the nucleolus of *Spirogyra*. As I have found, [2]) all the details which can be distinguished in a nucleus can also, by suitable means, be demonstrated in the nucleolus of *Spirogyra*, namely: a wall and contents, containing one or two threads, or a network, such as nuclei usually have, in addition to a substance which may be compared to that of ordinary nucleoli. As a rule these elements of the contents do not entirely fill up the space inside the nucleolus and cavities containing fluid may further be distinguished inside it. Also in their division the nucleoli of *Spirogyra* show very important points of agreement with nuclei, for instance the dissolving of the wall and of the substance agreeing with that of ordinary nucleoli and the longitudinal splitting of bodies which are comparable to chromosomes.

The nuclei of *Spirogyra* are, as far as the research extends, distinguished from all vegetable nuclei by their remarkable nucleoli. It is self-evident therefore that in examining the nuclei of *Closterium*, which are still so little known, I paid special attention to the nucleoli, the peculiar appearance of which had already attracted the attention of investigators. The answer to the question, whether *Closterium* possesses as remarkable nucleoli as those of *Spirogyra* and whether therefore both these Conjugatae agree in this respect, was thus an important point of investigation for me. In other respects also I have, however, endeavoured to bring to light as much as possible concerning the nuclear structure and karyokinesis.

Resting nucleus. The unicellular plants possess a single nucleus. As a rule it is found near the centre of the cell, i.e. it is about equidistant from both apices of the cell and everywhere about equidistant from the cell-wall, which with regard to the nucleus is concave on one side, convex on the opposite side. Often, however, the nucleus is somewhat nearer to one end than to the other and it occasionally lies considerably nearer to that part of the wall, which turns its concave side towards the nucleus.

As far as its shape is concerned the nucleus of *Closterium* agrees with that of *Spirogyra*, for it is flattened, appearing oval when viewed from above and circular when viewed sideways. The position of the nucleus in the cell also agrees with that seen in *Spirogyra*. The flattened poles are turned towards the apices of the

[1]) J. B. CARNOY, Biologie cellulaire, fasc. 1, p. 236.

[2]) Ueber den Nucleolus von Spirogyra, l. c. p. 220 et seq. Ueber Kernteilung bei Spirogyra l. c. p. 374 et seq. p. 359 and 360. Ueber abnormale Kernteilung, l. c. p. 215 et seq. and 241.

cell. The size of the nuclei surpasses that of the nuclei of *Spirogyra.* Observations on the diameter of nuclei of *Closterium Ehrenbergii* and of some thick species of the genus *Spirogyra* are given below.

Closterium Ehrenbergii Men. from 37 to 66 μ, average 53 μ, found near Groningen.

Spirogyra crassa Ktz. from 40 to 44 μ, average 42 μ, found near Utrecht and determined by MOLL [1]).

Spirogyra maxima (Hass.) Wittz. from 31 to 40 μ, average 36 μ, found near Groningen.

Spirogyra triformis n. sp. (with 6 chromosomes in the equatorial plate [2]) from 27 to 31 μ, average 28,5 μ, found near Steenwijk.

Spirogyra setiformis (*Roth.*) *Kg.* from 27 to 31 μ, average 27 μ, found near Steenwijk.

In the nucleus of *Closterium Ehrenbergii* there may be distinguished the same component parts, as generally occur in nuclei namely the nuclear wall, the network, the nucleolus or the nucleoli and the nuclear fluid. The wall of the nucleus is thin; it seems to be thinner than that of *Spirogyra.* It cannot long resist the influence of chromic acid. The network has a delicate, regular, reticulate structure. The nucleolus has a peculiar appearance. It consists of a collection of more or less rounded polyhedral bodies, which are mostly attached to one another, but still may quite well be distinguished separately. When the network has dissolved in chromic acid, it may easily be observed that many of the small bodies are joined. Each body may be regarded as a separate small nucleolus and the whole as a collection of small nucleoli. Amongst these occasionally one or a few occur which are considerably larger, and also more or less spherical. It seems to me that the small nucleoli lie in the meshes of the network which probably prevents their fusion to one great nucleolus. In dealing with the karyokinesis we will show that there are good reasons for this view, as well as for the conclusion that they consist of a fluid substance. The small nucleoli agree with those which are generally found in the vegetable kingdom and not with those normally occurring in *Spirogyra.* They are not at all to be compared with small nuclei. They have no wall, neither is the collection of nucleoli surrounded by a wall, nor can threads be distinguished in it as integral elements or be liberated from it by means of chromic acid, as is the case with the nucleoli in the nuclei of *Spirogyra.*

1) J. W. MOLL, Observations on Karyokinesis in Spirogyra, Verhandelingen der Koninkl. Akad. van Wetensch. te Amsterdam, 2e sect. D. 1. N⁰. 9, p. 16.

2) C. VAN WISSELINGH, Ueber Kernteilung bei Spirogyra, l. c. p. 356 and 362.

In *Closterium Ehrenbergii* I have not found important variations of the nucleoli in different nuclei, such as should occur according to DE WILDEMAN. It is true that the small nucleoli in the nucleus seem to form a more compact mass in the one than in the other which probably has led DE WILDEMAN to distinguish two different types. I have not however found essential points of difference.

Karyokinesis. When in *Closterium Ehrenbergii* division is about to take place, modifications occur in the cytoplasm as well as in the nucleus. At some distance from the nucleus both chromatophores show a constriction as the beginning of a division into two. Cytoplasm collects near the nucleus and the latter also shows considerable modifications. The nucleoli become distributed in the nucleus. The nuclear wall is dissolved and the network forms visible threads. The most striking of these three processes is the distribution of the numerous nucleoli in the nucleus. The threads arising from the network are at first rosary-like. They slowly contract to form a great number of short thick threads or chromosomes. Meanwhile many nucleoli unite so that often large globules arise. The modifications which the network undergoes, seem to go hand in hand with the union of the nucleoli. By the contraction of the network to broad threads the nucleoli seem to have more opportunity for fusion. The numerous nucleoli sometimes prevent the distinct observation of the chromosomes. When the action of the chromic acid somewhat disintegrates the nuclear figure the chromosomes become distinctly visible. The fusion of the nucleoli to globules shows that they consist of a fluid substance. A large proportion of the nucleoli get outside the nucleus; in consequence of this a great number of globules of different sizes are seen on either side of the nucleus. Gradually these dissolve in the cytoplasm.

When the nucleus has undergone the above mentioned modifications, the formation of the equatorial plate begins. The chromosomes move to the plane passing through the equator of the nucleus and finally they all lie in that plane. Thus the equatorial plate has been formed. The latter has the following peculiarities. It is flat, seen sideways it is almost round. It is of a considerable size. The diameter is from 26 to 40 μ on an average 35 μ. Although the structure of the equatorial plate is rather favourable for the determination of the number of chromosomes, the latter are so numerous that I have not succeeded in counting them exactly. There are more than sixty. Just as in other cases (*Spirogyra* [1]), embryosac [2]) of

1) Ueber den Nucleolus von Spirogyra, l.c. p. 209.
2) Ueber das Kerngerüst, l.c. p. 168.

Fritillaria and *Leucojum*, *Oedogonium*[1]) the chromosomes are not quite free in the cytoplasm but form a connected whole by means of delicate fibres. The cytoplasm may be dissolved and the entire equatorial plate isolated by treating material, fixed with FLEMMING's mixture, with chromic acid solution. The overturned equatorial plate is found floating and at first the chromosomes keep their original position with respect to each other. Only after prolonged action they become loose and separate. The chromosomes differ in length. In general they are short, most of them very short; the longer ones protrude from one of the sides of the equatorial plate. Their shape varies; some, especially the smaller ones are straight or slightly bent. Others are bent in different ways and form J-, S-, L-, U-, V-shaped and other figures. Longitudinally they show a line which indicates the place where they will split into two.

As follows from what has been said above, *Closterium* again supplies an example of a nucleus with chromosomes of varying length. Formerly this phenomenon attracted little attention in the vegetable kingdom. In 1898 I[2]) pointed out that in *Spirogyra* two of the twelve chromosomes differ from the others in having a thinner end from which a small thread-like body could be isolated which was rather resistant to the action of chromic acid, when the rest of the chromosomes had already been dissolved. The two aberrant chromosomes were often a little longer than the others. Later on I noticed two corresponding chromosomes in a *Spirogyra*-species in which the whole number of chromosomes was six[3]). In *Closterium* the chromosomes only differ in length. In 1905 this phenomenon was noticed in the vegetable kingdom by ROSENBERG[4]) namely in *Listera* and in 1908 by myself[5]) in *Oedogonium*; later it has also been noticed in other plants.

The division of the equatorial plate into two halves and the separation of these halves takes place in the ordinary way. The chromosomes split longitudinally. When the halves of the plate separate the ends of the chromosomes remain together longest. Consequently both halves often form rhomboidal figures and later on when separated V-formed ones with the arms pointing to each other. This especially occurs with the smaller chromosomes. With the longer chromosomes first the parts that are

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1) Ueber die Karyokinese bei Oedogonium, l.c. p 140.
2) Ueber den Nucleolus von Spirogyra, l.c. p. 205 et seq.
3) Ueber Kernteilung bei Spirogyra, l.c. p. 147.
4) Zur Kenntnis der Reduktionsteilung in Pflanzen, Botan. Notiser, 1905, Separatabdr. p. 9.
5) Ueber die Karyokinese bei Oedogonium, l.c. p. 141.

united with other chromosomes split and afterwards the free protruding portions. When the halves of the equatorial plate separate the halves of these chromosomes remain connected for a longer time at their free ends than at the ends which are not free. Finally these halves of the chromosomes also separate completely. During the action of the chromic acid the above mentioned particulars are readily observable. The rhomboidal, V-shaped and other figures which arise from the splitting chromosomes become entirely free.

The halves of the equatorial plate become smaller while they separate. At the same time the free parts of the longer chromosomes turn more or less outwards; thus there is also developed in *Closterium* the typical aspect peculiar to the phase known as diaster.

As I have remarked above, the nucleoli get into the cytoplasm, where they form on both sides of the nuclear-figure a number of greater and smaller globules. It not infrequently happens that at first a portion of the nucleoli remains behind attached to the equatorial plate between the chromosomes and sometimes considerable globules and masses are found between and on the separating halves of the equatorial plate.

When the equatorial plate has been formed the formation of the transverse wall also begins. The transverse wall develops in a way corresponding to that of *Spirogyra*. The process begins at the cell-wall and continues inwards until the cell is divided into two daughter-cells by a flat diaphragm. In *Closterium* this process is followed by another one, namely by the splitting of the cell-wall. There where the transverse wall arises, the cell acquires a constriction, which becomes deeper and deeper and is accompanied by a fission of the wall of the mother cell and with a splitting of the transverse wall, the halves of which develop strongly after splitting.

The equatorial plate is surrounded by the spindle. This arises from the cytoplasm surrounding the nucleus. In accordance with the size of the nucleus and of the equatorial plate the spindle is wide but it is not strongly developed, far less than in *Spirogyra*. The spindle fibres are delicate and thin. I have pointed out before [1]) that the spindle of the nucleus in *Spirogyra* most probably contributes to the regulation and acceleration of the separation of the daughter-nuclei; that when no spindle or an imperfect spindle is formed, this separation goes much more slowly and that through the development of the spindle the daughter nuclei are also driven apart, when the nucleus has been forced from its place by centrifuging, and with

[1]) Zur Physiologie der Spirogyrazelle, Beihefte zum Botan. Centralblatt, Bd. XXIV (1908), Abt. 1, p. 147.

chromatophores and protoplasm has been pressed against the wall. On account of what has been said above I am inclined to connect the inferior development of the spindle in *Closterium*, with the way in which the daughternuclei go to their appointed places in the daughtercells.

In *Spirogyra* the daughter nuclei are widely separated by the development of the spindle, so that each almost immediately takes up its appointed place, while in *Closterium*, in which the spindle does not develop as strongly as in *Spirogyra* and dissolves more quickly in the cytoplasm, the daughter nuclei at first do not separate so far. After the transverse wall has been formed, the daughter-nuclei in *Closterium* are at a small distance from this on either side. Next they move along the cell wall to the places, where the chromatophores divide into two. First they move along the transverse wall and next along the wall of the mother-cell, on that side which is bent most. In the daughter-nuclei they take a place between the two chromatophores, which have arisen by division from a single chromatophore of the mother-cell.

As a rule the nuclear spindle in *Closterium* is developed regularly. If, however, the nucleus is not situated in the middle of the transverse plane of the cell, but more on that side which is most strongly curved, the spindle-fibres extend more on the opposite side.

The halves of the equatorialplate develop to daughter-nuclei in the following way. They become surrounded by a wall and the chromosomes gradually develop to a fine network. At first these get a looser structure and more and more resemble threads of beads; soon they can no longer be distinguished from one another; they seem to form a tissue of fine threads of beads and in the end they constitute a delicate network. It is difficult to indicate the exact moment at which the young daughter-nuclei become surrounded by a wall. Very soon numerous little nucleoli appear in the daughter-nuclei between the threadwork. They come nearer to each other, form some small masses and finally one great central whole or a collection of nucleoli. When the young daughter-nuclei are still found near the transverse wall, the nucleoli are still spread in great quantities all over the nucleus. When the nucleus arrives between the two chromatophores of the daughter cell, they form a few masses. This has led to the conclusion that at first the daughter-nuclei have not one, but several nucleoli. The daughter-nuclei are flat, like the resting nucleus. On their way along the cellwall their shape more or less adapts itself to the circumstances; in making curves the nuclei are bent.

Above I have briefly described the structure of the nucleus and the details of the karyokinesis in *Closterium Ehrenbergii.* As appears from what has been said the nucleus, as far as the nucleoli are concerned, does not agree with the nuclei of *Spirogyra,* as earlier investigators have supposed. In this respect the nucleus of *Closterium* differs on an important point from those of *Spirogyra,* namely, it does not possess a nucleolus which may be identified as or compared with a nucleus. The nuclei agree with nuclei, which are generally found among plants, especially the higher plants. Nevertheless they show one peculiarity: the nucleoli which are indeed present in great numbers, form in the middle of the nucleus a conglomeration.

The nucleus divides by karyokinesis or mitosis. All phenomena which generally occur, also take place in *Closterium.* In *Closterium* the nuclear division presents the following particulars: the distribution of the nucleoli in the nucleus and their extrusion into the cytoplasm, the great flat equatorial plate, the great number of chromosomes which is more than 60, the different length of the chromosomes, which in general are short and of which the longer ones only have free ends, protruding sidewards, the wide, feebly developed spindle and the translocation of the daughter-nuclei along the cellwall.

Later I hope to give a more detailed account of the karyokinesis in *Closterium* and to illustrate with figures the above mentioned results. In this paper hardly anything has been said about the cell division and the growth of the cellwall. To this I also hope to refer later.

Mathematics. — *"On the relation between the vertices of a definite sixdimensional polytope and the lines of a cubic surface".* By Prof. P. H. Schoute.

1. In his investigation about semiregular polytopes, and polytopes possessing a higher degree of regularity Mr. E. L. Elte, whose dissertation is to appear shortly has met with a sixdimensional polytope of degree of regularity $\frac{3}{4}$ with 27 vertices. Our aim here is to point out the complete correspondence in relations of position between the 27 vertices of this polytope and the 27 lines of a cubic surface.

The symbol of the characteristic numbers of this polytope is

$$(27,\ 216,\ 720,\ 1080,\ 432 + 216,\ 72 + 27),$$