The Meiotic Divisions in Pollen Mother Cells of the Sweet-pea (Lathyrus odoratus, L.) with special reference to the Cytological Basis of Crossing-over

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By

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With Plates V–XII and 7 Text-figures

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INTRODUCTION

While there are several interpretations of the mechanism of crossing-over, there is a certain weak point in these interpretations, namely that the cytological data employed for them are mostly those

obtained from plants or animals in which the phenomenon of crossing-over has not yet been adequately studied. To obtain a more correct idea of the mechanism, we should rather seek its cytological basis in those plants or animals with regard to which we have already a sufficient knowledge of the phenomenon.

The sweet-pea is a classical material for the study of linkage, and we have now the fact that there are in this plant most probably as many linkage groups as the chromosome number, exhibiting certain definite percentages of crossing-over between the groups (PUNNETT, 1923).

The present investigation had its start in the spring of 1925. It was suggested by Prof. Y. KUWADA, and was intended to deal specially with the mechanism of crossig-over. Meanwhile, in 1926, Miss LATTER's research on this material was published, but it seems not to be superfluous to publish here the new results we have obtained, which differ from her results in some important points.

I. MATERIAL AND METHODS

Material was taken from several garden varieties grown in the research field of the Botanical Institute, Department of Science, Kyoto Imperial University. No cytological differences could be found among these varieties.

Of the various fixatives employed, the following two were found to be the best for this material after trials in various ways :—

1. NAWASCHIN's fixative (1% chromic acid 10 c.c., glacial acetic acid 1 c.c., commercial formalin 4 c.c) was used in combination with CARNOY's mixture (absolute alcohol 6, chloroform 3, glacial acetic acid 1 in volume). Preparations made from material fixed in NAWASCHIN's fixative after having been treated with CARNOY's mixture for 1-2 minutes were found to be the best for observations of the earlier stages. For studies of more advanced stages a more prolonged previous treatment with CARNOY's mixture was required, for instance, 3 minutes for the heterotype metaphase and 5 minutes or more for

stages of the homotype division. The material, after being fixed for 3 hours in NAWASCHIN's fixative, was transferred to 30% alcohol directly without rinsing in water; dehydration and paraffin imbedding were done in the usual manner.

2. The Bonn modification of FLEMMING's solution was used with the previous treatment with CARNOY's mixture (the same as that mentioned above). This method was found to give good results too. A more prolonged previous treatment with CARNOY's mixture than the first method was, however, necessary in this second method; 5–10 minutes for stages of heterotype division, and 10–15 minutes for homotype division. After being fixed for 24 hours, the material was washed in running water for 24 hours, and then, having been dehydrated through graded alcohols beginning with 2.5%, was imbedded in paraffin in the usual manner.

In both these methods flower-buds in appropriate stages of development were cut at their apices by the aid of a sharp dissection needle to open out their calyxes so that the direct contact of the anthers with the fixatives to be applied might be secured, and then the buds were plunged into the fixative without any delay. An exhaust pump was used to drive away air bubbles and the surplus of chloroform from the material soon after it had been transferred from the first fixative to the second.

Sections were cut in various thicknesses varying from 7μ to 15μ to meet necessary requirements, and stained exclusively with HEIDENHAIN's iron alum haematoxylin.

II. MORPHOLOGICAL CHARACTERISTICS OF SOMATIC CHROMOSOMES

Root tips raised in wet saw dust and fixed in the Bonn modification of FLEMMING's solution were used as material.

14 chromosomes are found in the equatorial plate. They are rarely found situated in the equatorial plane throughout their whole length, stretching out straightly from one end to the other, but usu-

ally so oriented that the only part of the chromosomes that always lies in this plane is the region of the spindle fiber attachment and the free end parts are quite out of the plane, generally being directed more or less irregularly towards the poles (Fig. 1). This latter situation makes it very difficult to study their morphological characteristics exactly, but some closer observations in metaphase and anaphase made clear the following points :—

The 14 chromosomes can be classified into 7 pairs, according to their lengths and to the number and the position of the constrictions they carry, or the lengths of the segments between the constrictions. The paired arrangement of these morphological pairs of chromosomes in the equatorial plate could not be found in this material (Figs. 2 and 3).

Each chromosome has a constriction very near the proximal end (the end nearer to the point of spindle fiber attachment) which causes the formation of a very short end segment appearing like a spherical body attached to this end of the chromosome. This end segment very often splits longitudinally earlier than the other part of the chromosome and then becomes so much elongated that it loses the spherical appearance (comp. Fig. 2 with Figs. 3 and 4). Two pairs of chromosomes have 2 other constrictions and consequently they consist of 4 segments; all the other pairs of chromosomes have only one extra constriction, so that they consist of 3 segments. The length of the corresponding segments differs in different pairs, as the whole length of the chromosomes is different in different pairs. In Fig. 4 *a*—*c* all the chromosomes are arranged pairwise according to their lengths.

The spindle fiber is attached at the 2nd constriction in all the chromosomes (Fig. 5), so that the position of the spindle fiber attachment is in all of them submedian or subterminal according to the position of the constriction. In the anaphasic chromosomes the constrictions are not so clear as in those of the metaphase but the same relation as in the metaphase in length between the two parts on both

sides of the point of spindle fiber attachment is recognizable. The chromosomes are now bent over at the point of spindle fiber attachment into the form of a V with unequal arms, the apices of the V being directed towards the pole and the two unequal arms towards the equatorial region (Fig. 6 and Fig. 7).

When the root tips were treated before fixation with 0.75% solution of chlorallhydrate as directed by SAKAMURA (1920) in his paper, the constrictions in each chromosome became obscure except the first constriction, which is subterminal in its position (Fig. 8).

III. MEIOTIC PHASES IN POLLEN MOTHER CELLS

1) *Heterotype division*

Presynizetic stage and synizesis. Young pollen mother cells are rich in cytoplasm and of the polygonal shape. The nucleus at an early stage shows in it a faintly stained reticulum with more densely stained granules suspended in it (Fig. 9). More than one nucleolus may be found in a certain early stage, but when the reticulum becomes more conspicuous, there is always one only. The vacuoles and crystal-like bodies found by LATTER (1926) in this plant in the nucleolus in this stage could not be observed in the present material. Convoluted leptotene threads become then recognizable in the nucleus, some parts of the thread being thicker than the other part (Fig. 10). Soon the convolution of the leptotene threads begins to contract and become separated from the nuclear membrane to form the synizetic ball, enveloping the nucleolus in its center. After the synizetic contraction has reached its maximum, the nucleolus comes out of the convoluted threads (Fig. 11). In this stage the phenomenon of "nucleolar budding" is observed. One or more small nucleolar bodies are sometimes found on the mother nucleolus, sometimes free from it in the nuclear cavity (Fig. 11). From this time onward, the nucleolus undergoes changes in shape. Its clear spherical shape is now lost,

but an irregular amoeboid shape becomes conspicuous; in most cases the nucleolus is fragmented into more than two lumps of irregular shapes. These lumps take their positions tightly pressed agaist the nuclear wall so as to assume the shape of a concavo-convex lens or a crescent (Fig. 12 and Fig. 13). The vacuolated state is now very conspicuous in these nucleolar fragments.

A material connection between the nucleolus and the spireme thread has been described by some authors, and also in the sweet-pea by LATTER (1926). The result of the present observations, however, points to the conclusion that such can not be said to be of general occurrence, though an accidental contact by chance may be found.

As the stage proceeds further, the crescent nucleolus and its fragments gradually diminish in volume, contract into spherical bodies once again and become free from the nuclear wall, being suspended in the nuclear substance. In material fixed with NAWASCHIN's fixative the nucleolus is stained more densely than in material fixed with FLEMMING's solution. The vacuolisation in the nucleoli was observed in this advanced stage only in a few cases in both these different fixation methods (Figs. 15, 17 and 22).

Postsynizetic stage. From the stage of synizetic contraction onwards the nucleus shows a conspicuous growth in its volume as well as in the elements contained in it, as can be seen by comparison of Fig. 12 with Fig. 13. The synizetic knot now loosens by and by to have the spireme thread distributed throughout the nuclear cavity, and thus the nucleus becomes once more filled up with a loose convolution of pachytene threads (Fig. 13 and Fig. 15).

In the sweet-pea the leptotene stage is of very short duration and the nuclear threads are too thin to allow of a correct judgement about its behaviour. But, as the stage proceeds into the postsynizetic stages the present material appears to be appropriate for detailed observation. In observing, care was taken to have appropriate illuminating conditions. Characteristic features observed in this stage are: 1) the double nature of the spireme thread, and 2) the parallel

disposition of the threads two by two with crossing points at certain lengths.

The postsynizetic spireme thread consists of two strands twisted around each other which may be recognized as the longitudinally split halves. This double nature is observable even in so early a stage as that where the spireme is just beginning to spin out of the synizetic knot. Sometimes the twisting is very loose as is seen in a portion of the spireme shown in Fig. 13*b*, and sometimes it is so tight that not only is the double nature of the spireme completely concealded but also this tightly twisted spireme forms a spiral by further twisting, as is seen in the lower part of the spireme shown in Fig. 14*c*. The turns of the spiral often present a "pearl necklace" appearance which may be misunderstood as consisting of a row of chromomeres (Fig. 14*a*). The most prevalent appearance of the spireme is that of a rope or two spirals twisted around each other (Fig. 13 and Fig. 15). Very often the loose portion is found between the two portions where the twisting is tight, as shown in Fig. 13*a*. The direction of twisting is often found to be opposite in the portions on the two sides of the loosely twisted portion.

Parallel disposition of the spireme threads and their crossing at certain lengths may be recognized in the later stage of synizesis, but it can be clearly recognized only after the synizetic knot has become loose and filled up the whole nuclear cavity (Fig. 15). In this stage the spireme threads are found associated two by two running parallel to each other for some distance, at the close of which the two threads converge to form a cross and run again apart from each other. Some examples are illustrated in Figs. 16 *A*—*D*.

In Fig. 16 *A*, we see at *a* a crossing point of two spireme threads each of which consists of two strands. At the point of crossing, the four arms of the cross are found lying on one and the same optical plane, a situation which shows that the two threads are set in a more intimate relation to each other than mere contact or superposition. In Fig. 16 *B*, a similar feature of the cross to that observed at *a* in Fig. 16 *A* is found at *a*. At *b*, the cross feature itself is quite the same as at *a*, but here the portions of the two spiremes on both sides of the cross are found on different optical planes and the crossing point between

them. Figures b_1, b_2 and b_3 are drawings of what is seen in these three successive optical planes. They show that this cross is not a perspective, but a true cross. In Fig. 16 *C*, two crossing points are seen at *a* and *b*. At *c* the cross is only perspective; spireme *I* merely touches the upper side of spireme *II* with its lower side. In Fig. 16 *D*, two crossing points are seen at *a* and *d*. Between these two points there are two twistings (*b* and *c*). They look as if the two spiremes were superposed one on the other, but really they are some distance apart from each other.

These observations lead us to the conclusion that in these points of crossing the two spireme threads may be disposed in a more intimate relation to each other than mere superposition or contact.

The two characteristic features of the spireme threads described above, i. e. their double nature and their crossing at certain points, become more clear as the stage proceeds further.

Segmentation of spireme, diakinesis and metaphase. Segmentation of the spireme threads seems to occur step by step beginning from so early a stage as that when the spinning of the spireme out of the synizetic knot is not yet complete, for we can see already in this stage some free ends of the spireme threads lying in pairs (Fig. 16 *A*, upper end). Gradual contraction and thickening of the segmented spireme threads follow then, giving rise to the presentation of a different aspect of the nucleus from that of the immediately preceding stage (comp. Figs. 17 and 18 with Fig. 15). The spireme thread becomes untwisted by and by, probably as the result of the shortening and thickening of the thread itself. We can distinguish 7 bivalents, each consisting of the two parallel spireme segments with a cross-point or points between them. They are of different sizes and forms which will be described later in detail. In this stage most bivalents are found stretched across the nuclear cavity from one side to the other, forming a radiating figure. At both ends of these bivalents we can very often observe that the component spireme threads or the univalents, are double, a fact which shows the four-stranded nature of the bivalents. This stage seems to correspond to the 'second contraction' or 'brochonema' of LATTER (1926). The nucleolus is often found in the central region of the nuclear cavity, but no figure suggesting the

"central mass" or "knot" as described by LATTER (1926) in this material could be found. As the bivalents contract and thicken to assume compact forms characteristic in the diakinesis and metaphase, the parts or segments between the crossing points take up the form of rings which show a tendency to be disposed perpendicularly to each other successively (Figs. 18–21. Comp. also Figs. 52–54).

In preparations from material fixed by certain methods of fixation the spiral structure of chromosomes was observed in the stages of diakinesis and metaphase (MAEDA, 1928). The structure is to be considered as owing its origin to the contraction of the chromosome, or more properly speaking, to the contraction of its matrix accompanied by no or far slighter contraction of its chromatic part (cf. KUWADA, 1926, p. 5).

The fact that the spireme threads which are found in the early postsynizetic stage consisting of two twisted strands and running parallel two by two with a certain number of crosses between them develope into the four-stranded bivalents leads us to the conclusion that in this plant the mode of syndesis is parasyndesis.

Anaphase and telophase. There is some complication in the mode of disjunction of the bivalents, and details of the observations will be described in Section VI. But the general scheme of separation towards the poles may be mentioned here. The disjoined univalents are transported to both polar regions 7 by 7. They are pulled at their point of spindle fiber attachment, so that they are bent at this point into the shape of a V with unequal arms its apex directed towards the pole. Each arm is longitudinally split, and the chromosome assumes the form of an X or double V, the spilt halves remaining connected with each other only at the crossing point of the X or the apex of the V (Fig. 27, Fig. 28 and Fig. 29). These shapes of the anaphasic chromosomes are clearly observable in polar view (Fig. 30*a*) or in a somewhat oblique view (Fig. 30*b*). In these cases, we can

count $4 \times 7 = 28$ round bodies in an optical section (Fig. 30*a*) as described by WINGE (1919) in this material.

These chromosomes with 4 arms, are then tightly clumped together into a group at the pole, although individual arms are still clearly observable (Fig. 31). The formation of the nuclear membrane follows then soon and two daughter nuclei are organized without a cell plate being formed between them (Fig. 32).

Interkinesis. This stage is of very short duration in this material. The quadripartite figures of the chromosomes derived from their shape, the X or the double V, in the anaphase, is retained throughout this stage, and no anastomoses among the arms of the X's or V's are formed. One or more nucleoli are found in the nucleus in this stage.

As the nucleus increases in its volume, the chromosomes, which have been tightly clumped together, become loosened from one another and are distributed throughout the nuclear cavity. The 4 arms of each chromosome become elongated and show a clear spiral structure along their whole length. A slight change in the relative position of the 4 arms of each chromosome may occur, but such changes as to cause contact or twisting of the arms do not take place in any case (Fig. 33).

In a later stage the arms of the chromosomes become thinner but shorter, presenting the spiral structure very conspicuously (Fig. 34).

A sudden contraction in volume of the nucleus follows then, and around this contracted nucleus a clear zone becomes visible. The chromosomes contract again and assume once more clear X or double V shapes (Fig. 35 and Fig. 36).

2) *Homotype division*

Soon after the disappearance of the nuclear membrane, two spindles each containing 7 chromosomes in the nuclear plate are formed at right angles to each other in almost all cases. Every chromosome is now caught by the spindle fiber at the crossing point of its 4 arms, which is situated on the equatorial plate, each two corresponding arms being so disposed as to be symmetrical with reference

to the plane of the equator (Fig. 37). The sister longitudinal halves of the chromosomes then begin to separate from each other and are transported to different poles. These processes proceed very regularly and simultaneously in all the chromosomes in the two spindles (Fig. 38, Fig. 39 and Fig. 40).

The chromosomes in the homeotypic anaphase resemble in appearance very much those in the somatic anaphase (comp. Fig. 7 with Figs. 39 and 40). No longitudinal split is recognizable in these chromosomes. When all the chromosomes have reached the poles, the nuclear membrane is formed around each chromosome-group. In the tetrad nuclei individual chromosomes are observed showing the single spiral structure for a tolerably long period (Fig. 41 and Fig. 42).

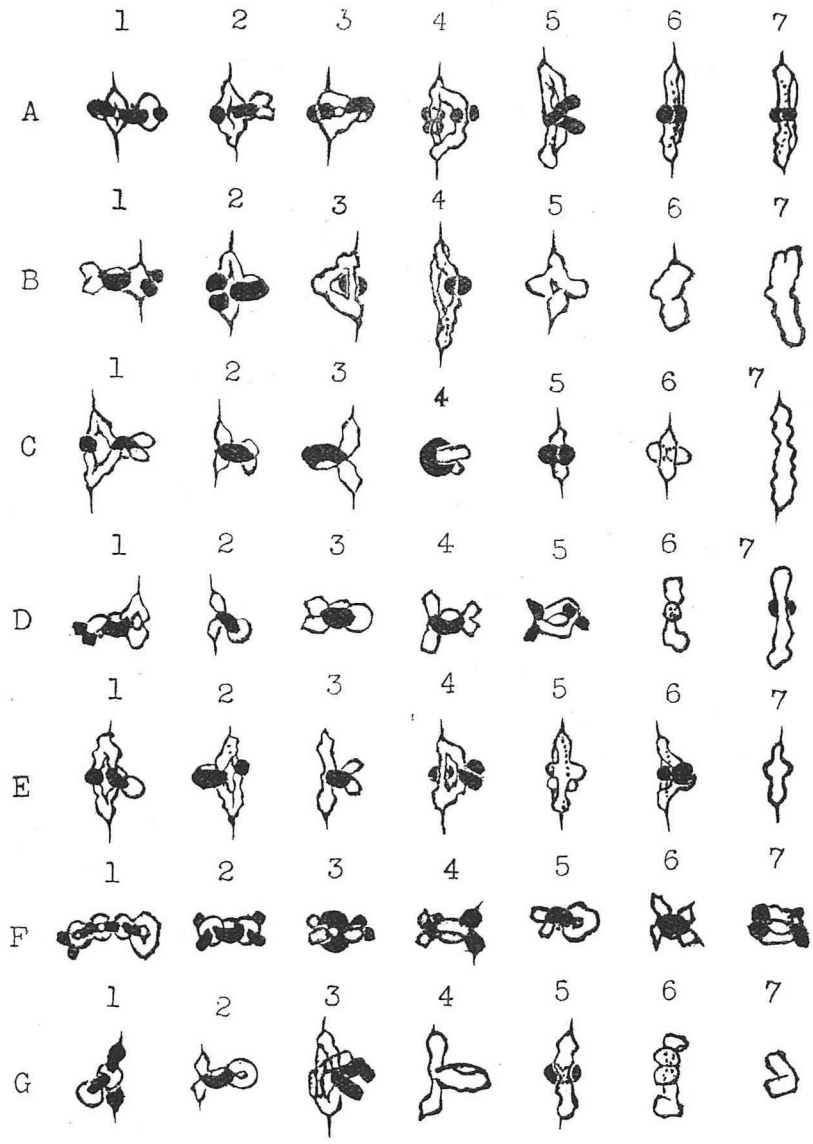
The cytokinesis is accomplished by the furrowing as described by LATTER (1926, p, 263).

IV. THE CONFIGURATION OF BIVALENTS IN THE HETEROTYPE DIVISION

There are many types of configuration in the bivalent chromosomes. Examples are given in Text-fig. 1 *A*—*G*, which were drawn from pollen mother cells that were found in one and the same pollen sac and escaped from being cut through by the microtome knife. Among these figures the groups *A*, *B* and *C* are those taken from one and the same section. The others are those from other sections. Each group consists of chromosomes found in one cell. In the figures, the bivalents are arranged in the order of complexity of their configurations to make it easy to compare those in the different groups with one another.

Group *A* (Text-fig. 1 *A*). 1, a quadruple ring[1] with terminal processes on one side (right in the figure); 2, a double ring with a vertical V on one side (right) and two short processes on the other (left); 3, a double ring with two processes on one side (left); 4, a

[1] By "quadruple ring" it is here meant that the bivalent assumes the shape of a chain consisting of four rings. We find in this plant simple, double, triple, quadruple and other more complicated rings in this sense. In the compound rings each component ring is disposed at right angles to the adjoining ones.



Text-fig. 1 *A—G*. Seven groups of bivalent chromosomes from one and the same pollen sac. Explanation in the text.

simple vertical ring or loop with two processes on each side; 5, a simple vertical ring or loop with horizontal V on one side (in the front in the figure); 6 and 7, a simple vertical ring or loop with two processes on one side (in the front in the figures).

Group *B* (Text-fig. 1 *B*). 1, a double ring with two terminal arms on each side (the ring on the right hand side in the figure to which the spindle fiber is attached appears like a spherical body); 2, a double ring with two processes on one side (left); 3 and 4, a simple vertical ring with two short processes on one end (in 3 on the right, in 4 in front); 5, a cross with an open space in the middle, suggesting a double cross; 6, a vertical rod slightly bent in the middle; 7, the same bent to a less extent in the middle.

Group *C* (Text-fig. 1 *C*). 1, a double ring with lateral processes on one side (left) and a vertical V on the other (right); 2, a horizontal simple ring with a vertical V on each side; 3, a horizontal simple ring with a vertical V on one side (right); 4, a vertical simple ring appearing like a compact mass, with a horizontal V on one side (right); 5 and 6, a vertical rod with two short processes which are attached to one side of the rod in its middle region; 7, a vertical rod showing constrictions.

Group *D* (Text-fig. 1 *D*). 1, a triple ring with a horizontal V with very short arms on one side (left); 2 and 3, a double ring, each part appearing like a compact mass, with a vertical V on one end of the chain (left); 4, a horizontal simple ring with a vertical V on each side; 5, a vertical simple ring with two processes on one side (right) and a V on the other (left); 6, a vertical rod, bent near both ends, with a chromatic lump in the middle; 7, a cross with a long vertical and a very short horizontal arm.

Group *E* (Text-fig. 1 *E*). 1, a double ring with a chromatic mass, perhaps a compact ring, on one side (right) and a small chromatic lump on the other (left); 2, a double ring with a small chromatic lump on one side (right); 3, a horizontal simple ring with a vertical rod on one side (left) and a vertical V on the other (right); 4, a vertical loop with two processes on one side (left) and a V on the other (right); 5, a vertical loop with two processes on each side; 6, a vertical loop with two processes on one side (right) and a chromatic lump on the other (left); 7, a cross.

Group *F* (Text-fig. 1 *F*). 1, a septuple ring with a small V on one side (left); 2, a triple ring with a V on each side; 3 and 4, a double ring with a V on each side; 5 a double ring with a vertical V on one side (left); 6, a horizontal simple ring with a vertical V on each side; 7, a vertical simple ring with two processes on each side.

Group *G* (Text-fig. 1 *G*). 1, a triple ring with a vertical V on one end of the chain (right); 2, a double ring with a vertical V on one side (left); 3, a vertical loop with a horizontal V on each side; 4, a horizontal simple ring with a vertical rod on one side (left); 5, a vertical rod with a lateral chromatic lump on each side (the rod is constricted in the middle); 6, a vertical rod, bent near both ends so as to assume the shape of a Z, with a constriction and two superposed chromatic lumps in the middle; 7, a rod bent into a V (oblique view).

This result of observations shows that, in the sweet-pea, the configuration may be different on different occasions even in one and

the same bivalent. A similar state of affairs has been reported by TISCHLER (1921–22) in *Tradescantia fluminensis*, by NEWTON (1926) in *Tulipa australis* and by BELLING (1926, 1927) in *Uvularia* and *Hyacinthus orientalis*.

To obtain some concrete knowledge about the origin of these configurations earlier stages in the development were followed back. The results obtained will be mentioned below beginning from the simplest configuration.

1. The case where the component univalents of a geminus are attached to each other only at one of the ends.

In the early diakinesis the component univalents which show something of a zigzag aspect in structure are conjoined end-to-end and run rather straight (Fig. 43 *a*, *b*, *c*), or sometimes more or less irregularly (Fig. 44 *a*, *b*). This relative position of the components is derived from the original side-by-side arrangement by their opening out. In the later diakinesis and metaphase they assume as the result of contraction the shape of a rod or a V (Fig. 43 *d*, *e*, *f*, *g*, *h*, *i*, *j*), or an S or a Z (Fig. 44 *c*, *d*, *e*, *f*, *g*), with or without a constriction in the middle. When they are pulled towards the poles they become elongated into such a form as C or ∫ (Fig. 43 *k*, *l*, *m*, Fig. 44 *h*, *i*).

2. The case where the component univalents cross each other at one point.

In early diakinesis the two component univalents intersect at various angles at some distance from the ends (Fig. 45 *a–d*). The arms of the cross may not run straight, but sometimes take irregular courses (Fig. 45 *d*, Fig. 46 *b*).

When the crossing occurs at a point very near one end, the shorter arms are often found in the metaphase fused into a lump which is situated in the region of the equator, and the two longer arms very much shortened and thickened, so that the whole bivalent presents a tripartite appearance (Fig. 45 *h–k*). When the longer arms are pulled towards the poles, a vertical rod, with the lump or two unfused processes of the shorter arms superposed on or juxtaposed to

each other in the middle, is produced (Fig. 46 *h–k*). When the crossing occurs near the middle of the univalents (Fig. 47 *a–c*) they condense gradually to take in the metaphase the shape of an X which is vertically disposed with arms situated symmetrically on both sides of the equatorial plane (Fig. 47 *d*, *e*, *g*, *j*). When the pulling forces operate from both poles, a vertical rod with a horizontal V or two fairly long processes (Fig. 47 *f*, *h*, *i*, *p*, *q*, *r*), or a vertical cross with or without an open space in the region where the crossing takes place (Fig. 47 *k–o*) is produced.

The disjunction is attained by the horizontal division plane as shown in Figs. 47 *s*, 57, 64. The result of this mode of disjunction will be discussed later.

3. The case where the component univalents are attached to each other at their both ends to form a ring.

In diakinesis, the univalents form loops of various shapes (Fig. 48 *a–h*). In the metaphase they are contracted into rings with or without an appreciable clear space in the center (Fig. 48 *i–k*). In the early anaphase, while in some cases the attachment is undone at one end, in others it remains as it is at both ends for a fairly long period. In this latter case the ring, being elongated by being pulled towards the poles, gives an appearance of two V's attached to each other at the free ends of their arms which present a coil appearance (Fig. 48 *l–o*).

4. The case where the two component univalents are attached to each other at one of their ends and cross each other at some distance from the other.

In the early stage of diakinesis the univalents form a loop with two free portions of various lengths (Fig. 49 *a–d*). In a later diakinesis the loop contracts into a ring and the two free portions make a lump or two short processes when they are very short (Fig. 49 *e*, *f*, *g*, *i*, *j*), and when comparatively long, they form arms diverging into a V which may be transformed into a rod by the opening out of the angle of the V (Fig. 49 *h*, *k*, *l*, *m*).

In the metaphase is produced a bivalent of the shape of a vertical ring with one or two processes (Fig. 49 *n*, *o*, *p*) or a horizontal V on one side of the ring (Fig. 49 *q*), or a horizontal ring with a vertical rod attached to one side of the ring (Fig. 49 *r*, *s*). Whether the ring is vertical or horizontal is determined by the position of the point of spindle fiber attachment. When the spindle fiber is attached to the portion forming the ring, or the ring is vertical, a figure of two V's inversely conjoined with each other at both free ends of the arms is produced in anaphase (Fig. 49 *t*, *u*).

5. The case where the component univalents cross each other at two points some distance from the ends.

In diakinesis the univalents form a loop with two free arms at each crossing point (Fig. 50 *a–c*). In the metaphase, when the spindle fiber is attached at a point on the ring, a vertical ring with two processes or free arms attached on the equatorial level to each side of the ring is produced (Fig. 50 *f–h*), and when it is attached to the free arms, a horizontal ring with a vertical rod on one side and a horizontal V on the other results (Text-fig. 1, *C* 2, *E* 3).

6. The case where the two component univalents are attached to each other at their two ends and cross at one point between the ends.

In the earlier diakinesis the univalents form a bivalent of the shape of two loops connected in the form of a figure 8 (Fig. 51 *a–c*). In a later diakinesis the loops contract and thicken to compose a chain of two rings disposed perpendicularly to each other,—the "double ring" (Fig. 51 *f–k*). In the metaphase, the formation of the double rings is completed (Fig. 51 *l–o*).

7. The case when the two component univalents are attached to each other at one end and cross each other at two points.

In diakinesis the univalents form a chain of two loops with two free arms at one end of the chain (Fig. 52 *a*, *b*). In the later diakinesis and metaphase these three segments contract into more compact forms, the loops now being better called rings. Each segment is disposed

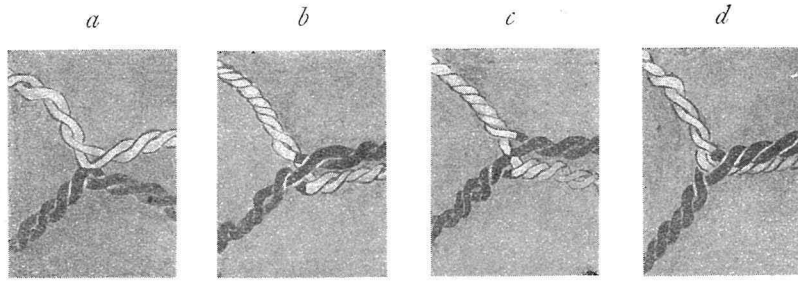
perpendicularly to the other, the segment to which the spindle fiber is attached taking the vertical position (Fig. 52 *f–n*).

From what has been described above, we can see that there is a common method of procedure in the formation of the various configurations of the bivalents, and we are able to infer how more complicated configurations can arise. The method of procedure we can postulate is: 1) in earlier stages the component univalents may cross each other at some points. When the crossing points are more than two, a loop or a chain of loops is formed, the ends of the univalents remaining free from each other as "arms": 2) as the stage proceeds, each loop or segment begins to be disposed in right angles to the others successively: 3) the loops contract into rings and the free segments or the arms into V's: 4) in the metaphase a chain of as many rings as the number of loops, with or without the arms attached to the ends of it is produced, each segment being disposed perpendicularly to the other. As the crossing may occur at any corresponding level of the univalents, the length of the segments cut off by the crossing points varies in every case. When the end segment forming the arms is very short, it is often contracted into a lump (Text-fig. 1, *D* 6) or two processes which are attached to one side of the adjoining segment. These two processes are found arranged side by side, sometimes in the same plane as that of the next segment (Text-fig. 1, *B* 2, *F* 7, and *G* 6), and sometimes in the plane perpendicular to it.

In Figs. 53 and 54 more complicated bivalents in successive stages than those described above are shown. Such cases are of relatively rare occurrence, and their configurations are often rendered indiscernible by the contraction of the segments into more or less compact textures as shown in Fig. 55. When however, the bivalents become looser in their texture in late metaphase and meta-anaphase, it is possible to discriminate details in behaviour of their disjoining components. The largest number of segments counted in this stage was eight.

V. THE CROSSING OF THE SPIREME THREADS

The following different types were observed in the mode of crossing of the spireme threads.



Text-fig. 2. Schemas showing four different types in the mode of crossing of the spireme threads. *a*, Type I; *b*, Type II a; *c*, Type II b; *d*, Type III.

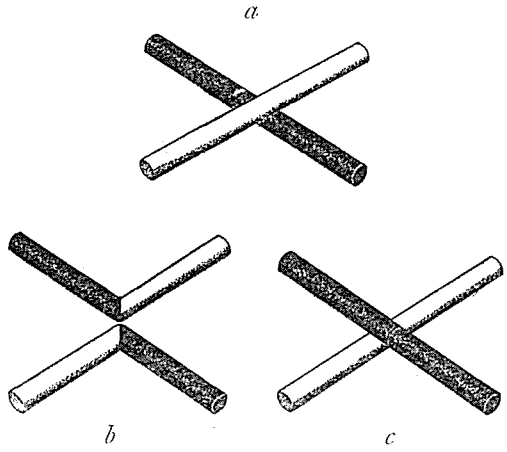
Type I. The two stranded spireme threads are attached to or fuse with each other with their inner strands at a point, the outer strands being quite free from each other (Fig. 51 *a*, Text-fig. 2 *a*, Text-fig. 4 *a*, *a'*). In this case the cross appearance is only apparent. It does not represent a real cross but merely a "node". When it is due to a simple attachment of the threads it will become lost as the threads contract into the definitive double chromosome or geminus.

Type II. The two inner strands cross each other (a) merely perspectively (Text-fig. 2 *b*, Text-fig. 5 *a*, *a'*) or (b) actually forming a fusion, the two outer strands being disposed in the shape of an x and not forming a cross between each other (Text-fig. 2 *c*, Text-fig. 6 a_1, *a'*). Types I and II b are not distinguishable from each other in the spireme stage but the results of observations of bivalents in the metaphase or in early anaphase, which will be described later, show both to have a real existence.

Type III. Each strand of a spireme thread crosses each of the other, one strand of the former being interlaced with that of the latter successively. In this case the cross is only perspective and not an actual cross or fusion (Fig. 47 *a*, Text-fig. 2 *d*, Text-fig. 7 *a*, *a'*).

Types I and II are veay often met with, but type III is of very rare occurrence. Type I corresponds to that observed by NEWTON in *Tulipa* where "the chromatids touch without interlacing" (NEWTON, 1926. p. 348, Fig. 31 a). Tyye II corresponds to the "chiasma" figures often found in animal spermatocytes. In plants such a case has been reported only in a few cases, for example, by BELLING in *Uvularia* (1926), *Hyacinth* (1927) and *Lilium* (1928), by NEWTON in *Tulipa* (1926) and by BABCOCK and CLAUSEN in *Crepis* (1929). There are two opposing opinions about the origin of the figure. GRANATA (1910), MCCLUNG (1914), ROBERTSON (1919) WENRICH (1919), WILSON (WILSON and MORGAN, 1920) and others maintain the view that this "chiasma" owes its origin to the segmental opening out of the 4 chromatids or strands which have been tightly associated side-by-side into two rings disposed perpendicularly to each other, that is to say, through the reductional plane in one segment and through the equational plane in the next. JANSSENS (1909, 1924), on the other hand, explains the origin as follows: In some early stage of meiosis, at the point where the two two-stranded component threads of bivalents cross each other, fracture occurs and then recombination which results in the segmental interchange or crossing-over takes place in the two strands which are actually in contact. Thus they no longer form a cross, but are disposed in the shape of an x. As a result of torsion, which subsequently takes place at this point of contact or "node," the two segments of the bivalent on both sides of the "node" become so disposed as to lie in different planes perpendicular to each other. By this torsion, the two intact or non-cross-over strands are made to form either a cross which is merely perspective or an actual cross or fusion according to the direction of the torsion, but the cross-over strands run in any case along both sides of this "node" without crossing or fusing with each other (JANSSENS, 1909, Schéma XIII-XV. See also Text-fig. 5 a, a' of this paper). In the sweet-pea, as mentioned in Sect. III, each component spireme thread of the bivalent is composed of two strands twisted about each other. Thus, the former

opinion that the "chiasma" figure is due to segmental opening out of the 4 strands through the two different planes can not be applied in the present case. The origin of the figure is only apprehensible by accepting JANSSENS' explanation that it is due to fracture and recombination occurring between the two of the 4 strands in some earlier stage of meiosis, and to the subsequent torsion that takes place on the level of the point of fracture and recombination (Text-fig. 2 *b*, Text-fig. 3 *b* and Text-fig. 5 *a*, *a'*). Type III is only explicable when we assume that fracture and refusion have occurred in the two inner strands in some earlier stage, not causing a segmental interchange, but only resulting in inversion of the relative position of the two strands, the outer strands being kept in their original disposition (Text-fig. 3 *c*).



Text-fig. 3. Schemas explaining the process of fracture and recombination or refusion of the two strands; *a*, original disposition, *b*, the recombination for Type II, *c*, the refusion for Type III.

VI. THE DISJUNCTION OF THE COMPONENT CHROMOSOMES OF BIVALENTS AND THEIR LONGITUDINAL SPLITTING IN ANAPHASE

Now we are faced by the question of how the disjunction of the component univalents can be attained in such a complicated state of pairing as described in Sect. V. In the following paragraphs, only those cases where accurate observations were possible, and only those facts which may have an important bearing upon the question will be mentioned.

In cases where the component univalents are attached to each

other only at one of their ends the matter of disjunction is very simple, it being attained at the very commencement of anaphase by the horizontal separation of the components.

In the case where the bivalents are composed by the more complicated methods of pairing, matters are somewhat different from this.

One of the conspicuous features in these cases with which we often meet is a clear space which appears in the center of the crossing point of the component univalents. This is sometimes clearly shown even by the bivalents in an earlier stage (Fig. 47 *o*). The end-parts of the univalents which are free from the spindle fiber, split longitudinally earlier than the other parts when they are very short, and present an appearance of globules surrounding this clear space (Fig. 46 *l–o*). When these end-parts are comparatively long they are disposed horizontally and when the chromosomes are pulled towards the poles, become widened out so as to take a rhombic shape as shown in Figs. 47 *s*, 58 and 64. Very soon the longitudinal splitting becomes thoroughly completed, and such figures as those shown in Fig. 27 (the chromosome in the middle) and Figs. 59–63 are produced. In these figures the parallel disposition of the two homologous horizontal parts is clearly seen. There is no doubt that figures such as those represented in Figs. 59, 61–63 are derived from those of a double X or a vertical ring or a rod each with a horizontal V on one side (Fig. 47 *p*, *q* and *s*, Fig. 49 *q* and Fig. 57), and figures such as those represented in Fig. 60 are derived from those of a vertical ring with a horizontal V on each side (Fig. 50 *h*). The latter is the case that has been emphasized by JANSSESN (1909, 1924) and by CHODAT (1925) to be one where the bivalent is divided reductionally in its vertical parts and equationally in its horizontal parts. It is clear that in the present material these figures can only arise from the relative position of the 4 chromatid strands shown as Type II a in Sect. V.

Another conspicuous feature often found in the process of disjunction of bivalents is shown in Fig. 27 (the chromosome on the left), Fig. 28 (the chromosome on the right) and Figs. 74–81. In all these

figures, while three arms of the four of each disjoining double V-shaped univalent have already clearly separated from the corresponding arms of the other, the fourth arms of both univalents still remain attached to each other at a point at some distance from their free ends, or they are connected at this point of each by a short filament which is pulled out very long towards both poles across the equatorial region. This figure may be maintained for so long a period that it can be regarded as affording evidence of the fact that in the prophase the two arms or strands can be actually fused with each other at the point of crossing and hence, it leads us to the conclusion that this relation between the two strands in the earlier stages is maintained throughout the succeeding stages just before the completion of the process of disjunction. As pointed out by JANSSENS in 1909 (p. 393), such figures as those just mentioned are of frequent occurrence in the animal spermatocyte division. In the plant kingdom we find a fine example in *Vicia faba* studied by SAKAMURA (1920, p. 8). He states: "In der Metakinese der hetrotypischen Kernteilung verhalten sich einige gewöhnliche Chromosomen so, wie es in den Fig. 17–23 dargestellt ist. Unterwegs hört das Auseinanderziehen der homologen Chromosomen plötzlich auf, indem sie noch mit je einen gewissen Teil aneinander haften, ... Meistens aber trennen sich die beiden Schenkel eines V-förmigen Chromosoms nicht zu gleicher Zeit von denen des homologen Schwesterchromosoms, da ein Schenkel sich früher ablöst als der anderer; aus diesem Grunde zieht sich der eine sogleich zusammen, während der andere auffallend weiter in die Länge gezogen wird. Die M-Tochterchromosomen verhalten sich in der Metakinese ähnlich wie oben erwähnt. ....."

When the latter feature is the case, we have two different cases. The first case is shown in Figs. 66 and 67. The longitudinal halves of each component univalent of a bivalent are attached to each other at the free ends of their longer arms to form a loop. The loop of each univalent is attached to that of the other at a point on the equator. Figs. 68 and 69 show a slightly advanced stage where the

loop of the lower univalent is clearly opened out by the disattachment of the ends, and that of the upper is just beginning. Figs. 74–76 show these in still further advanced stages. If we consider these figures together with that reproduced in Fig. 51 *a* and the schema Text-fig. 2 *a* we can percieve that all these figures represent successive stages in the process of disjunction of the bivalents of the same origin in their configuration (cf. Text-fig. 4). The origin of such figures has been interpreted by JANSSENS in another way (JANSSENS, 1924, p. 275, Schéma XXXV).

In the second case one of the longitudinal halves of a component univalent of a geminus crosses and fuses with that of the other at a point at some distance from the end, while the other half of the univalent is attached to that of the other simply at the end (Figs. 70–73). Figs. 77–79 show a slightly later stage. It will be clear from the examination of Text-fig. 6 that such figures are derived from those earlier figures which belong to the type described as Type II b in Sect. V. It was mentioned in the foregoing that the origin of the figure of Type II b is only explicable by the assumption that in a certain stage of prophase the segmental interchange must have occurred between two of the four chromatid strands, while the other two remain fused with each other on this level of crossing or interchange (cf. Text-fig. 2 *c*, Text-fig. 6). Type I and Type II b are quite similar in appearance in the prophase, but their real existence can be proved by comparing those figures in the stage of disjunction shown in Figs. 66–69 with those in Figs. 70–73, or Text-fig. 4 *b*, *c*, *b'*, *c'* with Text-fig. 6 a_2, *b*, *a'* *b'*.

Finally there is a case such as that shown in Figs. 84–87. In these figures the 4 chromatids are interlaced so as to have each chromatid on one side of the longer arms of the two sister chromatids tightly attached to each other, and remain in this state up to a later stage, giving an appearance like a chain of chromatid loops as is seen in Figs. 84–86. In Fig. 87 a slightly later stage is represented. Comparing these figures with those shown in Fig. 47 *a* (cf. Text-fig.

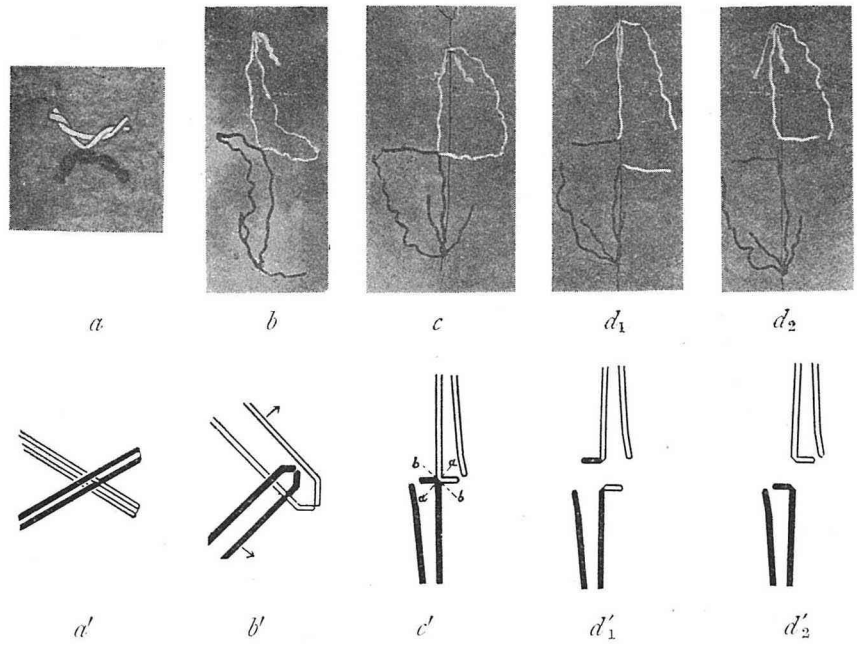
2 *d*), we are led to the conclusion that this disposition of chromatids must have been derived from that shown in Fig. 47 *a*, or conversely it may be taken as proving the actual existence of such a disposition of the 4 chromatids in the prophase, i. e. the case shown as Type III in Sect. V. (cf. Text-fig. 7). The origin of these figures has been interpreted by JANSSENS in another way (JANSSENS, 1924, p. 276, Schéma XXXVI).

VII. CYTOLOGICAL BASIS OF CROSSING-OVER

As mentioned above, the twisted state of the two component double strands of spireme threads which is observed in a relatively early prophase becomes lost gradually as the condensation of the strands proceeds further, and thus the strands may become free from each other. While the twisted strands are being untwisted by their continual condensation, they themselves are compelled at the same time to take the spiral aspect by the condensation. If the spiral begins to be formed during the state when the untwisted strands are still tightly associated side-by-side, one double spiral will be formed, and where they have been clearly separated from each other, two free single spirals will be produced. In the first case, which is of rather less frequent occurrence than the second case, there may be a certain difficulty in the separation of the coiled strands (cf. KUWADA, 1926, 1927. ALEXANDER and BRIDGES 1928), but this has nothing to do with the disjunction in the bivalent itself, or the phenomenon of crossing-over.

Besides the twisting mentioned above there is found another complexity between the two component double strands. This is the occurrence of the " nodes " or the crosses of the component univalents, the details of which were described in Sect. V. The rings formed with segments of the double strands between the nodes or crossing points become less obscure in the late metaphase, and the bivalent sometimes appears like a rod as shown in Fig. 55 *f* and *g*. This obscurity may involve in it the disappearance of the " nodes " (Type

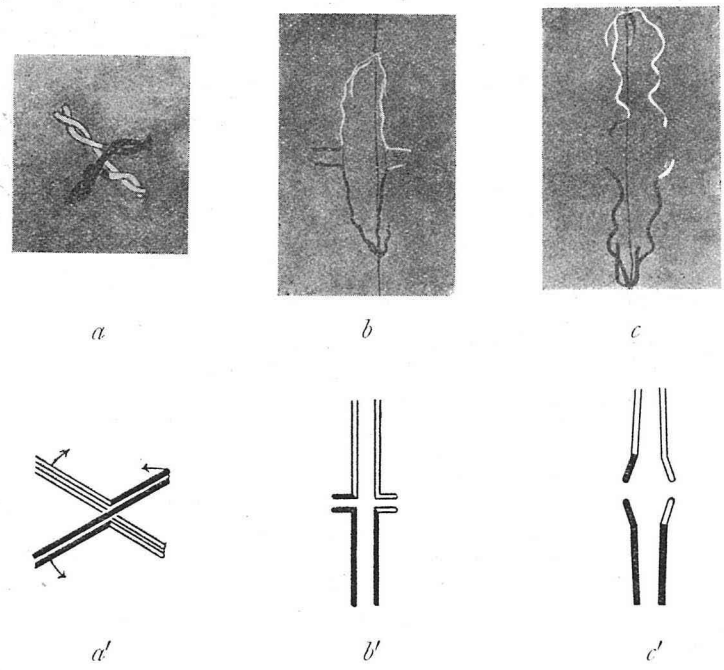
I in Sect. V) which is due to contraction of the strands in length. If all the nodes of a bivalent are constructed by mere attachment of four strands (Type I in Sect. V), the strands may now separate from one another without much difficulty. If, on the other hand, some of the nodes are so constructed as shown in Sect. V as Type I (in the case of true fusion) and Types II a, b, the segmental interchange between the strands will result. The possibilities of crossing-over which we can infer from the results of our observation are as follows :



Text-fig. 4. Models and schemas to show the disjunction process of a bivalent having a crossing point belonging to Type I in Sect. V. When the attached two strands separate in *a*–*a* as is mentioned in *c'* we shall have d_1 and d_1' in which a segmental interchange between two strands is expected : when it is separated as in *b*–*b*, we shall have d_2 and d_2' in which no segmental interchange is expected.

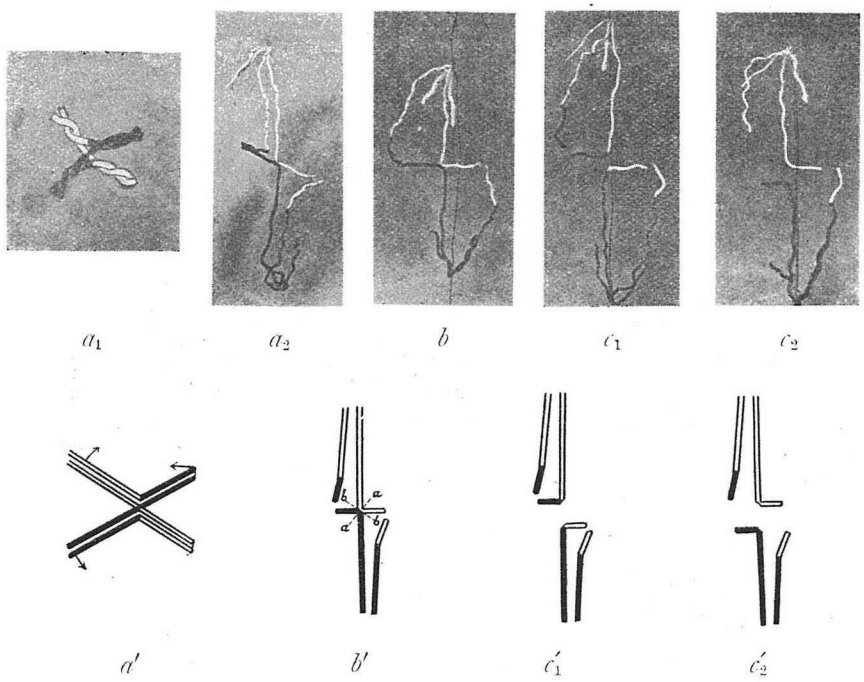
1) The case when the disposition of the four strands at the crossing point is of Type I. In this case the possibitity of the segmental interchange is to be found only when the strands in contact with each other actually fuse at the point of contact. When these

strands disjoin through the plane *a–a* the interchange will occur, but when they disjoin through the plane *b–b*, no such interchange will result (Text-fig. 4. cf. Figs. 80–83). In this case the interchange may be expected to take place between only a pair of the strands.



Text-fig. 5. The same for a bivalent having a crossing point belonging to Type II a in Sect. V. Segmental interchange is expected to occur as mentioned in *b*, *b′*, *c* and *c′*.

2) The case when the disposition of the four strands is of Type II a. In this case there will take place no interchange on disjoining, because there are no strands which fuse with each other. But the disposition of this type is comprehensible, as discussed above, only by assuming that a segmental interchange has occurred between two strands in some early stage of prophase. Thus we should expect as the final result an interchange between two of the strands, and not between the other two (Text-fig. 5). This type is the one with which we meet most frequently in the anaphase.

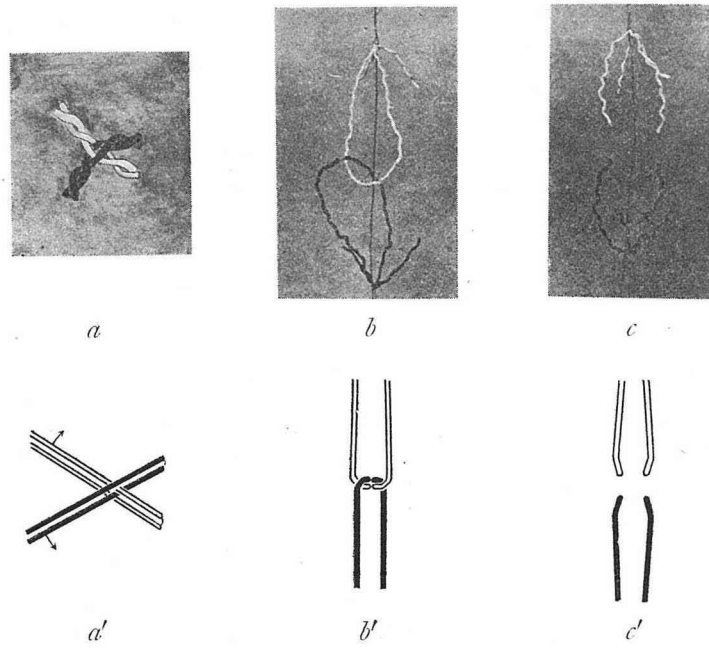


Text-fig. 6. The same for a bivalent having a crossing point belonging to Type II b in Sect. V. When the attached two strands separate in a–a we shall have c_1, c_1' in which segmental interchange between 4 strands is expected: when it is separated in b–b we shall have c_2 and c_2' in which segmental interchange between two strands is expected.

3) The case when the disposition is of Type II b. In this case too, as in Case 2) (Type II a), it is necessary to assume that an interchange in the early prophase has taken place between two strands, so that we must expect an interchange between these two strands. In this type, moreover, the other two strands actually fuse with each other, and therefore there is a possibility of interchange between these two strands, too, at the time of disjunction (Text-fig. 6 cf. Figs. 80–83). This case is in its result a combination of Cases 1) and 2). All the four strands may have been interchanged—the four strand crossing-over.

In the case when the disposition of the four strands is of Type III, no interchange can be expected to take place in the prophase as

discussed above, and there is no actual fusion between the strands, and consequently no interchange is to be expected at all (Text-fig. 7).



Text-fig. 7. The same for a bivalent having a crossing point belonging to Type III in Sect. V. In this case no segmental interchange is expected as the final result.

On this cytological basis, we may conclude that in the sweet-pea the crossing-over may occur in an early stage of prophase as considered by WILSON, MORGAN, JANSSENS and others, as well as at the time of disjunction, as was concluded to be the case by CHODAT (1925) from his cytological studies of *Allium*.

In the present investigation no stage was found which corresponds to the "brochonema" of LATTER (1926), a stage which she has found in this plant and assumes to form the cytological basis of crossing-over.

VIII. SUMMARY

In the somatic mitosis in root tips of the sweet-pea, there are 7 pairs of chromosomes of different lengths. Two pairs are the chromosomes with three constrictions, and the others are those with two

constrictions. The spindle fiber is attached at the second constriction in all the chromosomes.

In the nucleus of the young pollen mother cell a faintly stained reticulum is found with densely stained granules imbedded in it. Leptotene threads become then recognizable.

The synizetic contraction takes place. A nucleolus is found lapped by the convolution of the threads. It sends out buddings in this stage. When the synizetic knot begins to be spun out, the nucleolus goes to more than two pieces which are found pressed against the nuclear wall into crescent forms.

A material connection between the nucleolus and the spireme threads is not found to be of general occurrence.

The syndesis takes place after the mode of parasyndesis.

The double nature of the spireme threads and the crossing between two of these threads which run approximately parallel are found from the later synizetic stage up to shortly before the metaphase.

The spireme threads undergo torsion, and according to the degree of it the different portions of them present different appearances.

There are different types in the mode of crossing between the spireme threads or the component univalents of gemini. In these modes of crossing two features are important from the point of view of the bearing upon the cytological basis of crossing-over. In the one, an assumption is necessary to explain its origin, that a segmental interchange must have occurred in an early prophase, and the other indicates that the interchange is to be expected to take place on disjunction. This latter expectation is supported by the observation of the chromosome behaviour on disjunction.

In the later anaphase the univalent chromosomes assume the shape either of double V's or X's.

In the interkinesis no anastomoses are formed between the chromosomes, but each chromosome is clearly distinguishable from the others throughout this stage. The nucleus contains a nucleolus or nucleoli in it.

The homeotypic division is carried on in the regular manner.

In the tetrad formation the cytokinesis is accomplished by furrowing.

In conclusion the author wishes to express his cordial thanks to Prof. Y. KUWADA for his guidance and encouragement throughout the course of this investigation.

IX. LITERATURE CITED

ALEXANDER, J. and BRIDGES, C. B. (1928). Some Physico-Chemical Aspects of Life, Mutation and Evolution. (in ALEXANDER, J: Colloid Chemistry, Theoretical and Applied. Vol. 2. New York.)

BABCOCK, E. B. and CLAUSEN, J. (1929). Meiosis in Two Species and Three Hybrids of Crepis and its Bearing on Taxonomic Relationship. Univ. California Publ. in Agric. Sci. Vol. 2, No. 15.

BELLING, J. (1926). Single and Double Rings at the Reduction Division in *Uvularia*. Biol. Bull., Vol. 50.

—— (1927). Configuration of Bivalent of *Hyacinthus* with Regard to Segmental Interchange. Biol. Bull., Vol. 52.

—— (1928). Nodes and Chiasmas in the Bivalents of *Lilium* with Regard to Segmental Interchange. Biol. Bull., Vol. 54, No. 6.

CHODAT, R. (1925). Le Chiasmatypie et la Cinèse de Maturation dans *l'Allium ursinum*. Bull. Soc. Bot. Genève 1925.

GRANATA, L. (1910). Le cinesi spermatogenitiche di *Phamphagus marmaroatus* (*Burm*). Arch. f. Zellforsch., Vol. 5.

JANSSENS, F. A. (1909). La Theorie de la Chiasmatypie. La Cellule, t. 25.

—— (1924). Le Chiasmatypie dans les Insects. La Cellule, t. 34.

KUWADA, Y. (1926). On the Structure of the Anaphasic Chromosomes in the Somatic Mitosis in *Vicia Faba*, with special Reference to the so-called Logitudinal Split of Chromosome in the Telophase. Mem. Coll. Sci. Kyoto Imp. Univ. Ser. B. Vol. 11, No. 1.

—— (1927). On the Spiral Structure of Chromosomes. Bot Mag., Tokyo, Vol. 41, No. 483.

LATTER, J. (1926). The Pollen Development of *Lathyrus odoratus*. Ann. Bot., Vol. 40.

MAEDA, T. (1928). The Spiral Structure of Chromosomes in the Sweet-pea (*Lathyrus odoratus, L.*). Bot. Mag., Tokyo, Vol. 42, No. 496.

McCLUNG, C. (1914). A comparative study of chromosomes in Orthopteran spermatogenesis. Journ. Morph., Vol. 47.

NEWTON, W. C. F. (1926). Chromosome Studies in *Tulipa* and related Genera. Journ. of Linnean Soc., Vol. 47.

PUNNETT, R. C. (1923). Linkage in the Sweet pea (*Lathyrus odoratus*). Journ. Gen., Vol. 13.

ROBERTSON, W. R. B. (1916). Chromosome Studies. I. Journ. Morph., Vol. 27.

SAKAMURA, T. (1920). Experimentelle Studien über die Zelle- und Kernteilung mit besondere Rücksicht auf Form, Grösse und Zahl der Chromosomen. Journ. Coll. of Sci. Imp. Univers. Tokyo, Vol. 39, Art. 11.

TISCHLER, G. (1921–22). Allgemeine Pflanzenkaryologie. Berlin.

WENRICH, D. H. (1916). The Spermatogenesis of *Phrynotettix magnus*, etc. Bull. Mus. Comp. Zool. Harvard Coll., Vol. 60.

WILSON, E. B. and MORGAN, T. H. (1920). Chiasmatypie and crossing-over. Amer. Natur., Vol. 54.

WINGE, O. (1916). On the relation between number of chromosomes and number of types, in *Lathyrus* especially. Journ. Gen., Vol. 8.

X. EXPLANATION OF PLATES

All figures have been drawn by the aid of an ABBE's camera lucida using ZEISS' apochr. imm. 2 mm., and comp. oc. K 18 for outlines and comp. oc. K 12 for the studies in details, except where otherwise indicated.

Fixation: Figs. 1–8, the Bonn modification of FLEMMING's solution; Figs. 9, 18–22, 24, 35, 36, NAWASCHIN's fixative; Figs. 10–13, 14 *a*, *b*, *c*, 15, 16 *B*, 17, 23, NAWASCHIN's fixative in combination with previous treatment with CARNOY's mixture for 1 minute; Figs. 25, 26, 28, 29, 37–42, NAWASCHIN's fixative with the previous treatment with CARNOY's mixture for 3 minutes; Figs. 14 *d*, *e*, 16 *A*, *C*, *D*, 32–34, the same with the previous treatment for 5 minutes; Fig. 31, the Bonn modification of FLEMMING's solution with the same previous treatment as NAWASCHIN's fixation for 5 minutes; Figs. 27, 30 *A*, *B*, the same with the previous treatment for 15 minutes; Figs. 43–55, NAWASCHIN's fixative with the previous treatment for several different numbers of minutes; Figs. 56–90, the Bonn modification of FLEMMING's solution with the previous treatment for 10 or 15 minutes.

Plate V

From cells in root tips

Fig. 1. Side view of metaphase.

Figs. 2 and 3. The same in polar view.

Fig. 4. 14 metaphasic companion chromosomes arranged in the order of their length; *a*, from an early, *b* and *c*, from a later stage.

Fig. 5. Individual chromosomes in their process of separation, each showing the point of spindle fiber attachment at the 2nd constriction.

Fig. 6. Side view of anaphase.

Fig. 7. 14 pairs of anaphasic chromosomes individually drawn from the same group as that reproduced in Fig. 6. They are arranged in the order of their length.

Fig. 8. A cell from a chloralized root tip with 14 contracted chromosomes; in each of them only one of the constrictions is clearly manifested.

Plates VI—XII

From pollen mother cells

Plate VI

Fig. 9. Two young pollen mother cells, one with three nucleoli and the other with one nucleolus.

Fig. 10. Leptotene stage.

Fig. 11. Synizesis in its height of contraction; nucleolus has a nucleolar budding on it.

Fig. 12. Later synizesis. Double nature of spireme threads and their crossing are recognizable; nucleolus is lugged into two crescent-shaped pieces which are pressed against the nuclear membrane.

Fig. 13. Late synizesis. Double nature of spireme threads is very conspicuous. *A* and *B* are portions of the threads drawn with 1.5mm.×K18.

Fig. 14. Portions of the double spireme threads in the late synizesis showing some different aspects of doubleness. Detailed explanation in the text. Magnification: 1.5 mm. ×K18.

Fig. 15. Hollow spireme stage. The double nature of the spireme threads and their parallel disposition and crossing at certain points are conspicuous throughout the whole length of the threads.

Fig. 16. Portions of the spireme threads in the hollow spireme stage more highly magnified (1.5 mm.×K18); detailed explanation in the text.

Fig. 17. A later stage; 7 bivalent chromosomes can be distinguished from one another.

Fig. 18. Still later stage.

Fig. 19 and 20. An early diakinesis. Some different configurations of the bivalent chromosomes are seen.

Plate VII

Figs. 21 and 22. Diakinesis. In Fig. 22 the multipolar spindle is visible outside the nuclear membrane.

Fig. 23. Early metaphase with multipolar spindle.

Fig. 24. Metaphase with bipolar spindle.

Figs. 25 and 26. Early anaphase just at the commencement of disjunction.

Fig. 27. Later anaphase; separation of the longitudinal halves is clear in each disjoining chromosome. Some bivalents delay in disjoining, the corresponding arms of disjoining homologous univalents still remaining in some way or other attached to each other in the equatorial region.

Fig. 28. A slightly later stage than that reproduced in Fig. 27. All bivalent chromosomes except one have disjoined and the disjoined univalent chromosomes with their separated longitudinal halves have reached their respective pole, each assuming the shape of a double V or an X.

Plate VIII

Fig. 29. Late anaphase. Each four arms of disjoined univalent chromosomes have further contracted.

Fig. 30 *a* and *b*. Chromosome groups in the polar region in the same stage as that reproduced in Fig. 29; *a*, polar view, *b*, the same view somewhat oblique.

Fig. 31. Later stage. Though the 7 univalent chromosomes are tightly clumped together with each other in the polar region, each individual chromosome can be clearly recognized.

Fig. 32. Early interkinesis nuclei with clearly separated 7 chromosomes of the shape of X or double V. Nucleoli are found in each nucleus.

Fig. 33. Advanced stage of interkinesis nuclei. The chromosomes are in the most elongated state.

Fig. 34. A still further advanced interkinesis; only one of the nuclei in polar view is shown. The 4 arms of each chromosome have become slender and some show a clear spiral structure.

Fig. 35. Late stage of interkinesis, just before the dissolution of the nuclear membrane. Each nucleus is diminished conspicuously in its volume, and pressed into the shape of an ellipsoid, the longitudinal axis of which is at right angles to the axis of the cell from pole to pole; a clear zone is visible surrounding each nucleus.

Fig. 36. The same in polar view.

Fig. 37. Homotype metaphase; the two spindles are at right angles to each other, each with 7 chromosomes of the shape of a double V or an X.

Plate IX

Fig. 38. Early homotype anaphase; all chromosomes are separated from their sister halves and assume the shape of a single V.

Figs. 39 and 40. Later homotype anaphase.

Fig. 41. Last stage of homotype anaphase.

Fig. 42. Tetrad: each nucleus with 7 chromosomes of a clear spiral structure.

Fig. 43. Bivalent chromosomes in successive stages of development into the shape of a rod or V at heterotype metaphase and those in early anaphase; *a–c*, early diakinesis, *d*, *e*,

late diakinesis, *f–j*, metaphase and *k–m*, early anaphase.

Fig. 44. The same developed into the shape of S or Z; *a*, *b*, very early diakinesis, *c–e*, late diakinesis, *f*, *g*, metaphase and *h*, *i*, early anaphase.

Plate X

Fig. 45. Bivalent chromosomes in successive stages of development which assume a tripartite appearance at heterotype metaphase; *a–d*, early diakinesis, *e–g*, late diakinesis and *h–k*, metaphase.

Fig. 46. The same, but here the bivalents assume at metaphase the shape of a rod with two short transverse arms or processes; *a–d*, early diakinesis, *e–g*, late diakinesis, *h–k*, metaphase and *l–o*, early anaphase.

Fig. 47. The same for those bivalents which assume the shape of a cross or X at heterotype metaphase; *a–c*, early diakinesis, *d–i*, late diakinesis, *j–q*, metaphase and *r*, *s*, early anaphase.

Fig. 48. The same for those bivalents which assume the shape of a ring; *a*, *b*, early diakinesis, *c–f*, middle diakinesis, *g*, *h*, late diakinesis, *i–k*, metaphase and *l–o*, early anaphase.

Fig. 49. The same for those bivalents which assume the shape of a ring with a V or a vertical rod at heterotype metaphase; *a–d*, early *e–m*, late diakinesis, *n–s*, metaphase and *t*, *u*, early anaphase.

Plate XI

Fig. 50. Bivalent chromosomes in diakinesis and metaphase of the shape of a ring with two V's or processes on both sides of the ring; *a*, *b*, early diakinesis, *c–e*, late diakinesis and *f–h*, metaphase.

Fig. 51. The same of the shape of a chain of two rings: *a–e*, early diakinesis, *f–k*, late diakinesis, and *l–o*, metaphase.

Fig. 52. The same of the shape of the double ring chain with a V on one side; *a–e*, early diakinesis, *f–j*, late diakinesis and *k–n*, metaphase.

Fig. 53. The same of the shape of a triple ring chain; *a–d*, early diakinesis, *e*, *f*, late diakinesis and *g–j*, metaphase.

Fig. 54 *A*. Bivalent chromosomes in early diakinesis which assume in heterotype metaphase more complicated configurations than those reproduced in Figs. 43–53.

Fig. 54 *B*. The same in late diakinesis.

Plate XII

Fig. 54 *C*. Bivalent chromosomes at heterotype mteaphase of more complicated configurations than the triple ring chain.

Fig. 55. The same; the details in the configurations are obscure on account of contraction of chromosomes.

Figs. 56 and 57. Bivalent chromosomes of the shape of a rod with two processes in the middle, just at beginning of the disjunction (only their equatorial parts are drawn); the clear space seen in the center of the figures is very conspicuous.

Fig. 58. A bivalent chromosome in the same stage of the shape of a vertical ring with two processes on one side. On this side the ring shows the same appearance as those shown in Figs. 56 and 57, while on the opposite side it is opened out, the disjunction having taken place earlier than on the other side.

Fig. 59. A similar chromosome to that reproduced in Fig. 58 in a slightly later stage. Note the parallel disposition of the equatorial parts of the chromatid strands.

Fig. 60. A figure in the same stage of a bivalent chromosome which is in metaphase of the shape of a ring with two arms on each side.

Figs. 61–63. The bivalent chromosomes reproduced in Figs. 59, 60 in a still further advanced stage; in these figures one or two arms of the disjoinning V's still remain undisjoined from the corresponding arms of the other.

Fig. 64. A bivalent chromosome just at the commencement of separation of the longitudinal halves (1.5 mm.×K18).

Fig. 65. The same in an advanced stage of separation of the longitudinal halves (1.5 mm.×K 18).

Figs. 66–69. Figures which are to be judged as having been derived from those bivalent chromosomes in which the mode of crossing between the components belongs to Type I mentioned in the text. Compare these figures with Text-fig. 4 b, c, b′, c′.

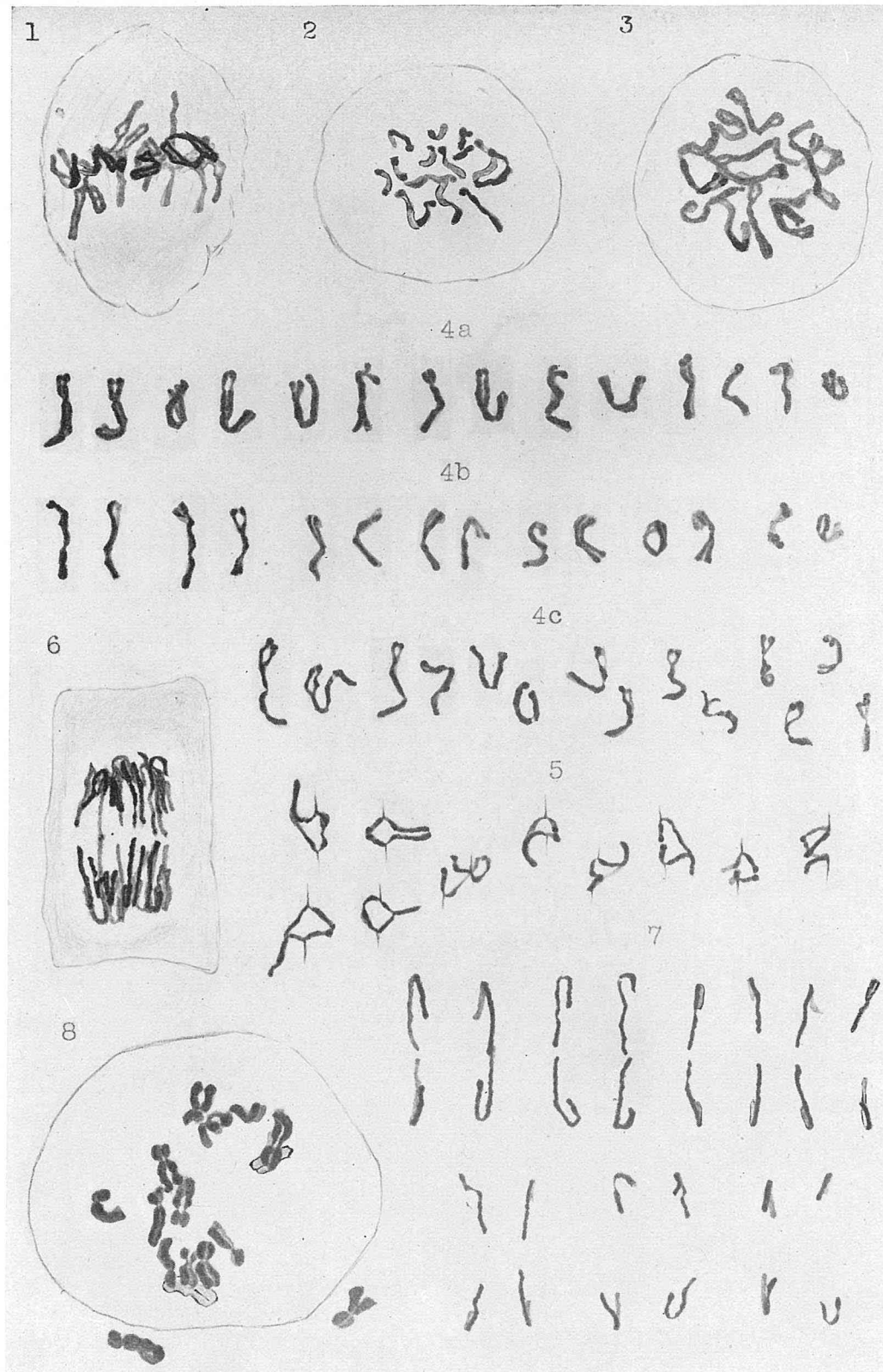
Figs. 70–73. The same in which the bivalent chromosomes are regarded as having been derived from those whose mode of crossing belongs to Type II b in the text. Compare these figures with Text-fig. 6 a_2, b, b'.

Figs. 74–79. Figures in later stage of the bivalent chromosomes reproduced in Figs. 66–73.

Figs. 80–83. Figures in later stage than Figs. 74–79, in which the disjunction is complete or is almost so. Compare these figures with Text-fig. 4, d_1, d_2, d_1', d_2' and Text-fig. 6 c_1, c_2, c_1', c_2'.

Figs. 84–87. Figures which are to be judged as having been derived from those bivalent chromosomes in which the mode of crossing is of Type III mentioned in the text.

Figs. 88–90. Figures in later stage of those reproduced in Figs. 84–87. Compare these figures with Text-fig. 7 c, c'.



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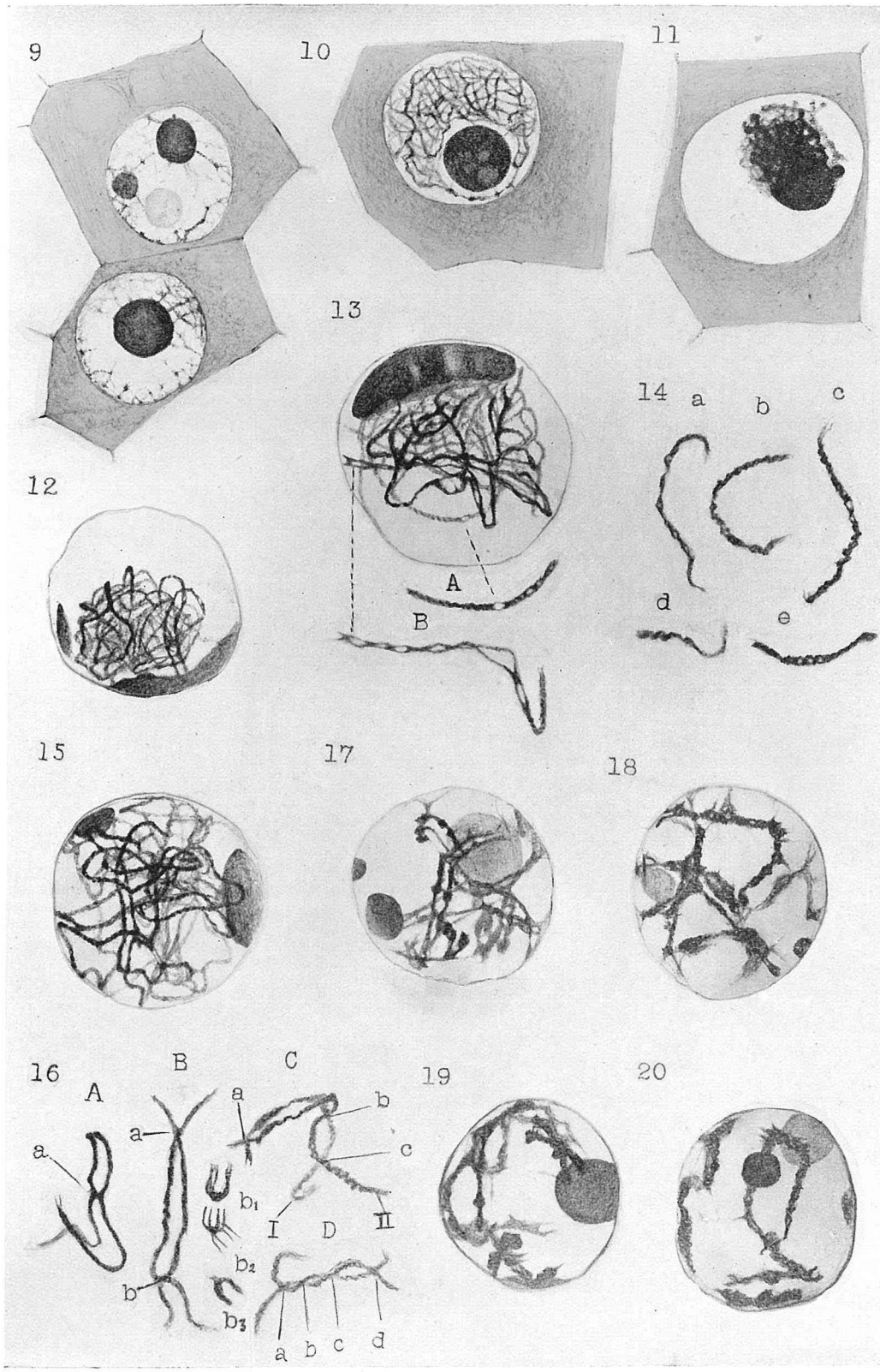
MAEDA: The Meiotic Divisions in the Sweet-pea



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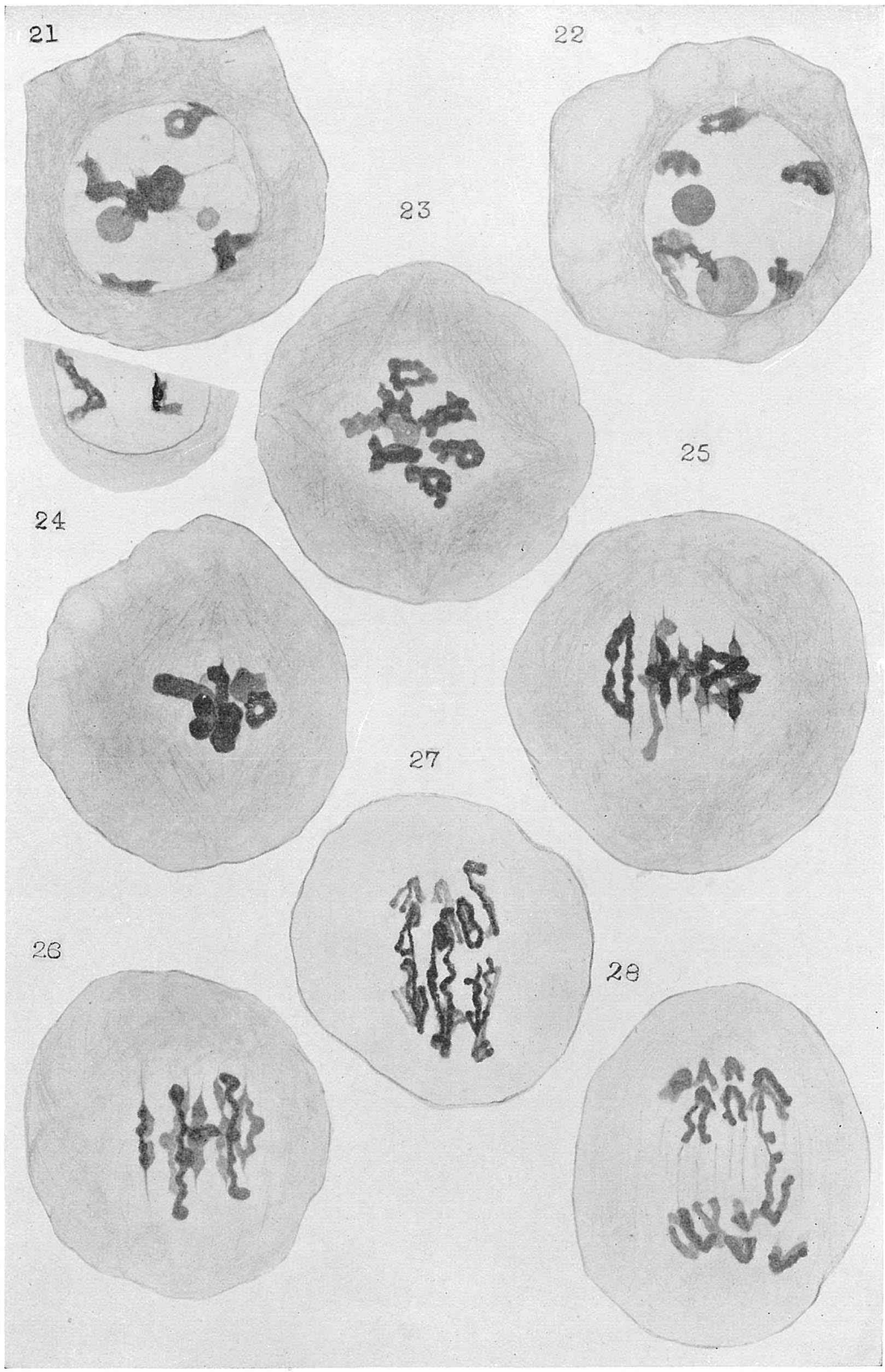

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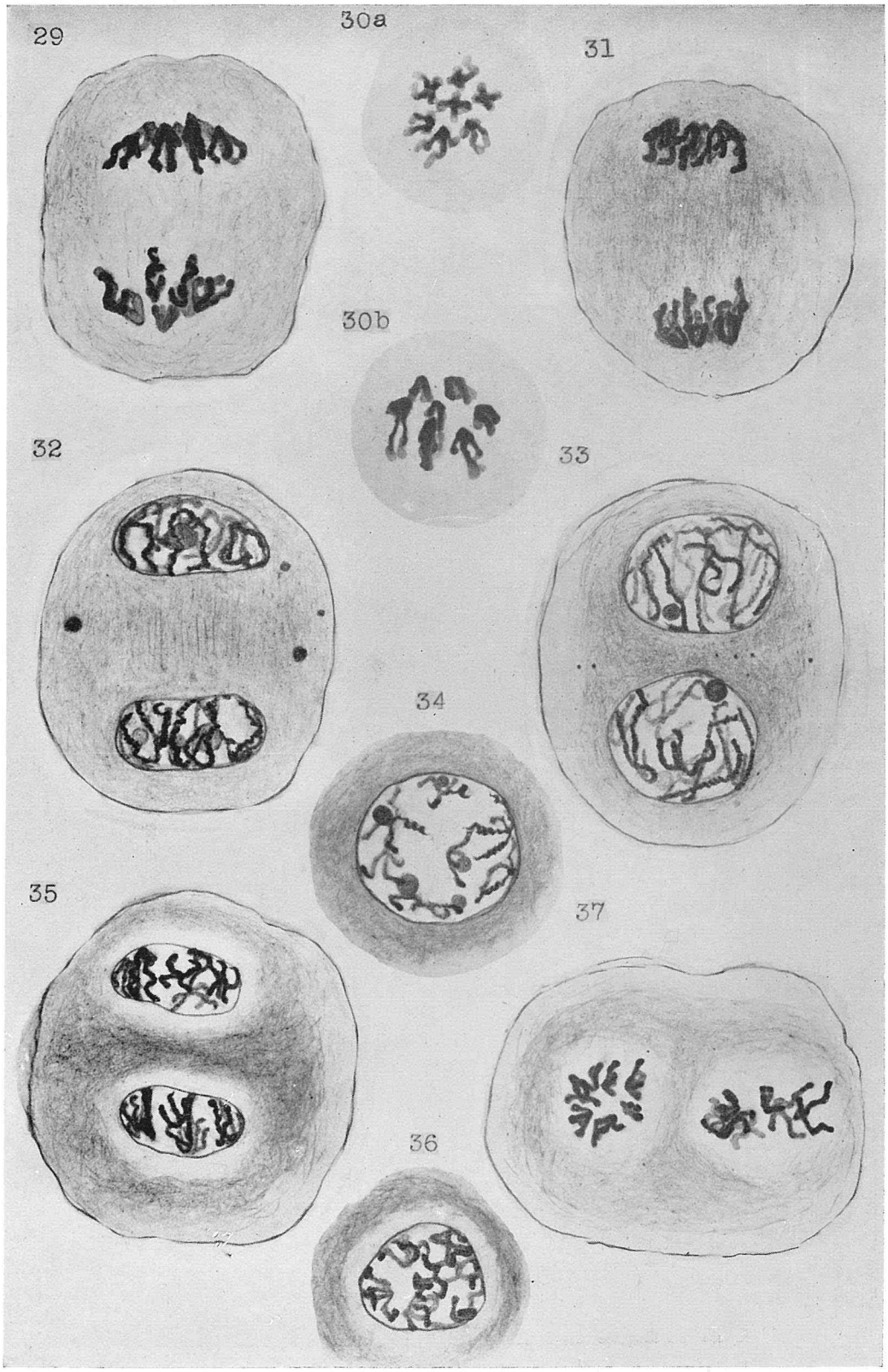

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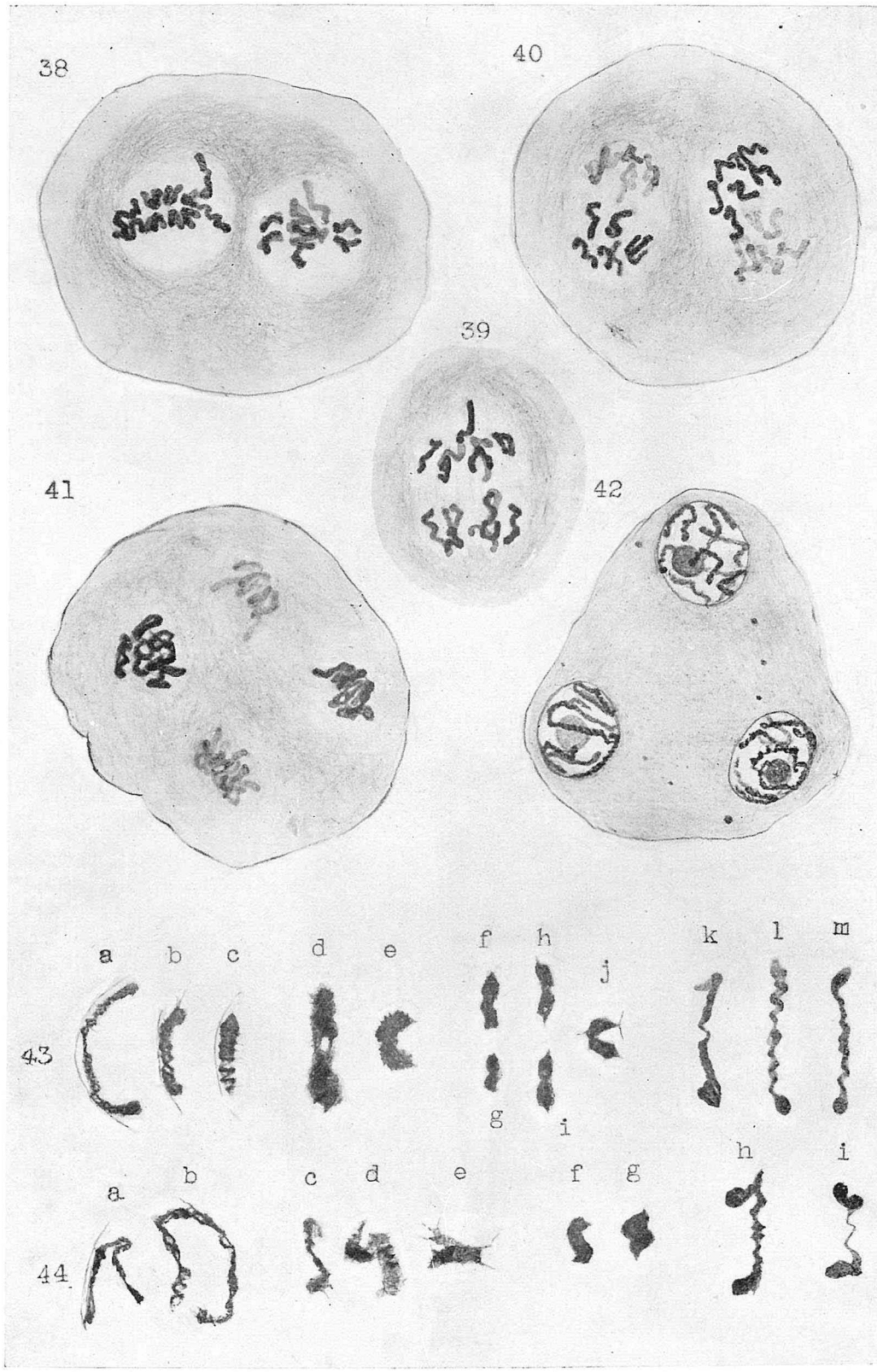

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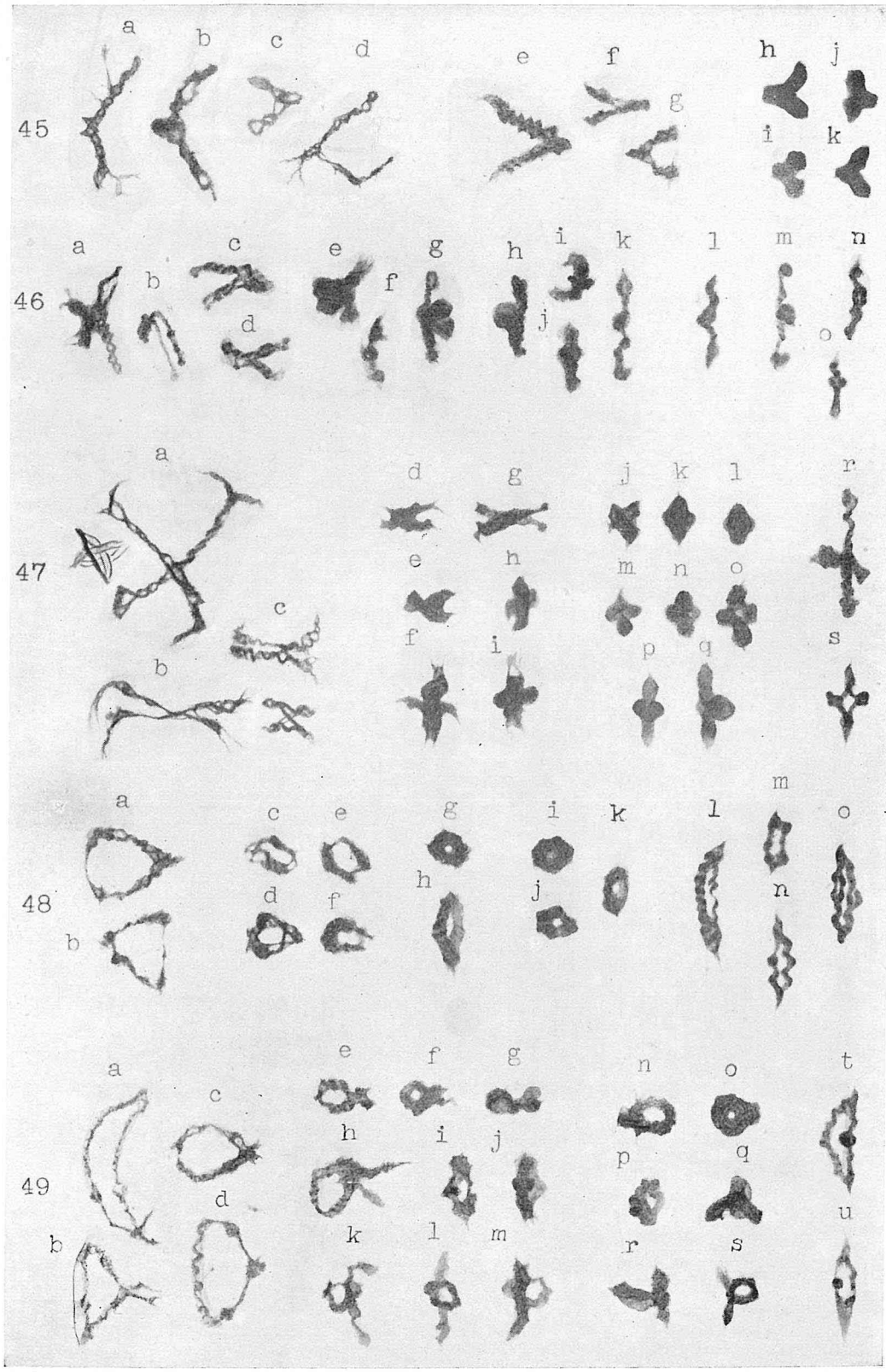

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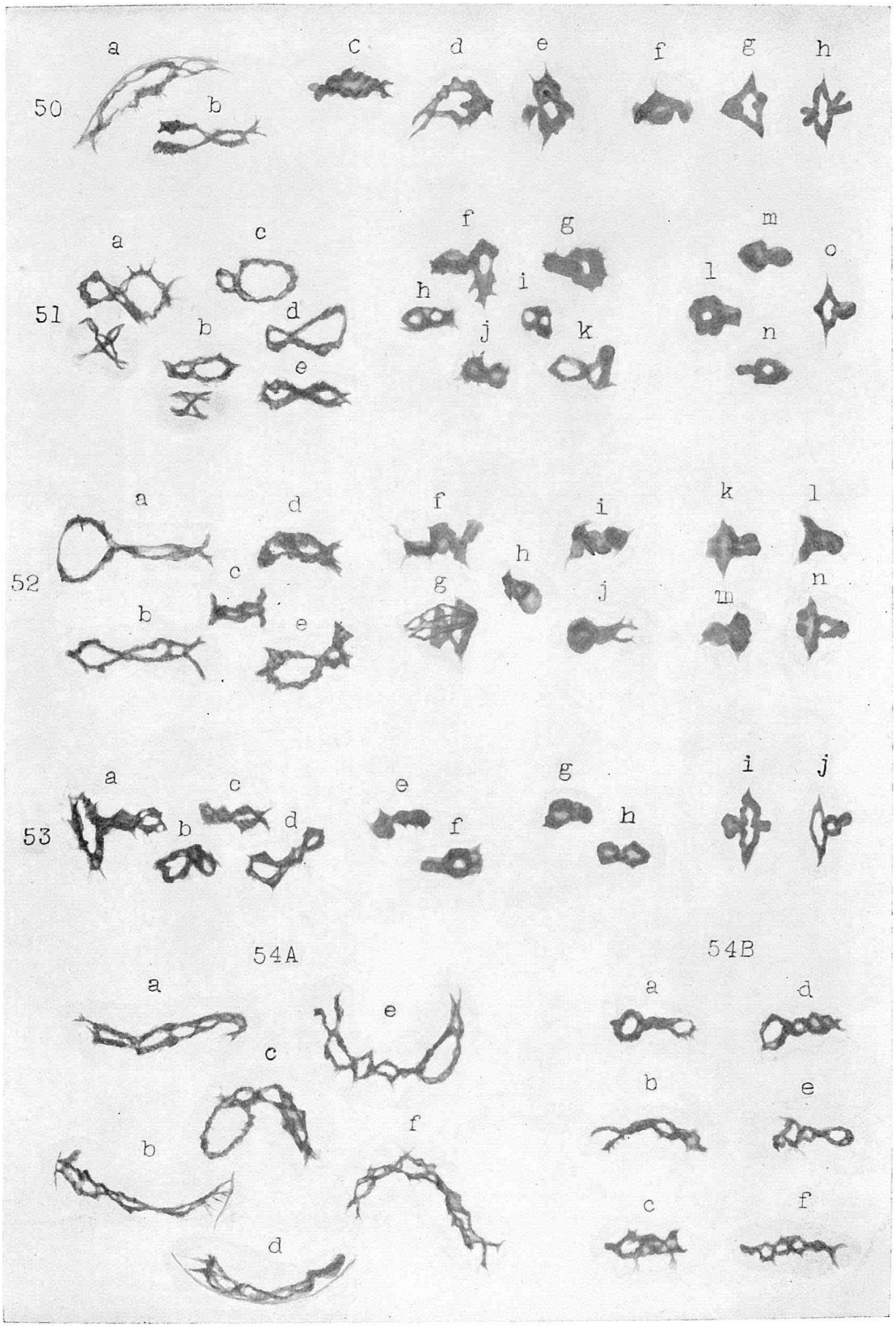
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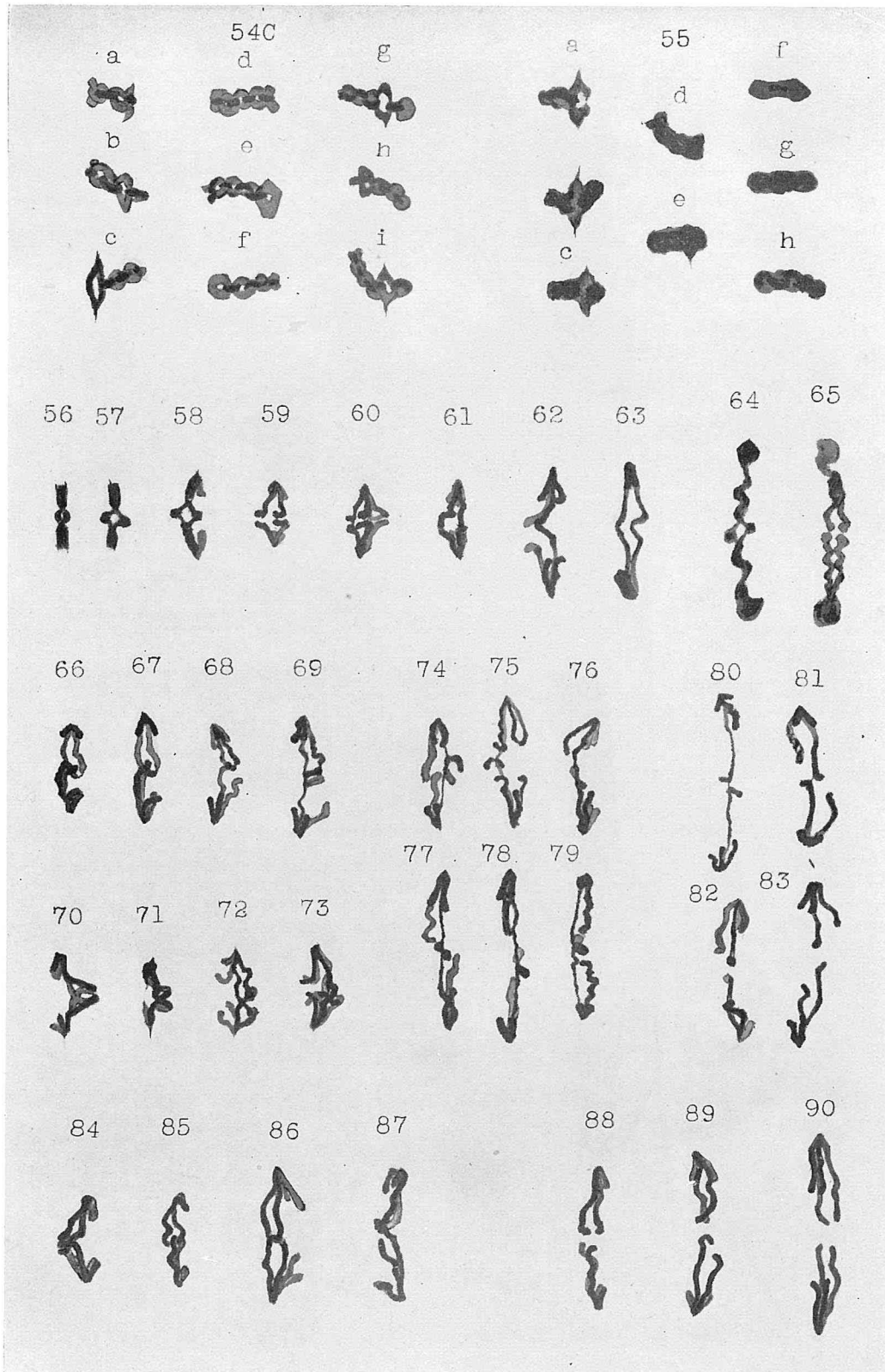

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MAEDA: The Meiotic Divisions in the Sweet-pea