

## On the Configurations of Gemini in the Pollen Mother Cells of *Vicia faba*, L.

By

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*With 11 Text-figures*

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In the course of the cytological studies of the pollen mother cells in the sweet-pea attention was drawn to the fact that there is a wide range of variation in the configuration of gemini, which is due to the difference in the number of points of intersection or attachment between the component univalents. All the different configurations so far observed were given in the preceding paper with the explanation of their genesis (MAEDA, 1930). The pollen mother cells of *Vicia faba* were studied to supplement this result from the sweet-pea.

### MATERIAL AND METHODS

Material was taken from the variety *me garosperma*. Quite the same methods of fixation as those adopted in the case of the sweet-pea proved to be successful for this material too. Sections were cut 10-12 $\mu$  thick and stained exclusively with HEIDENHAIN'S iron alum haematoxylin.

### OBSERVATIONS

It has been made clear by SHARP (1914) and SAKAMURA (1915, 1920) that one pair of the 12 chromosomes in the cells of the root-tip of *Vicia faba* is about twice as long as the other 5 pairs, and that

the spindle fibers are attached to the longer one at the middle point and to the shorter ones at a point very near one of their ends. They have also found that in diakinesis and metaphase of the heterotype division in the pollen mother cells, there are 6 gemini which correspond in the difference in size and the position of the spindle fiber attachment with the pair of somatic chromosomes. The longer geminus is very conspicuous and is generally called the M-geminus, as it was first named by SAKAMURA. No conspicuous difference can be found among the shorter gemini, however, so that we may treat them merely in the morphological sense as belonging to one category and may call them all m-gemini. Thus we have in this plant gemini of two different types with regard to their morphological aspects, i. e. the M-geminus and the m-gemini.

The number of the points of intersection or attachment varies, so far as the present observation goes, from one to six in the case of m-gemini, and from two<sup>D</sup> to thirteen in the M-geminus. The segments between the intersection points open out into rings in a chain, each of which is disposed perpendicularly to the other (Fig. 1), and thus various configurations arise. In the case of the m-gemini

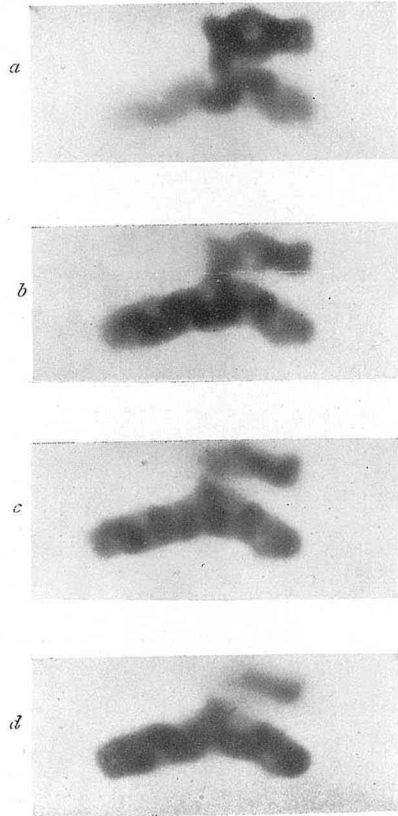


Fig. 1. Microphotographs of an M-geminus taken in four successive optical planes to show orientation of the rings which are disposed perpendicularly to the adjoining ones.

<sup>D</sup> See Fig. 8a.

all the configurations found can be classified into 7 different cases according to the number of the points of intersection or attachment. In Figs. 2-7, examples of these kinds of configurations are given. In the accompanying schema the one partner of a homologous pair is shown black and the other white so as to make clear the mode of intersection or attachment.

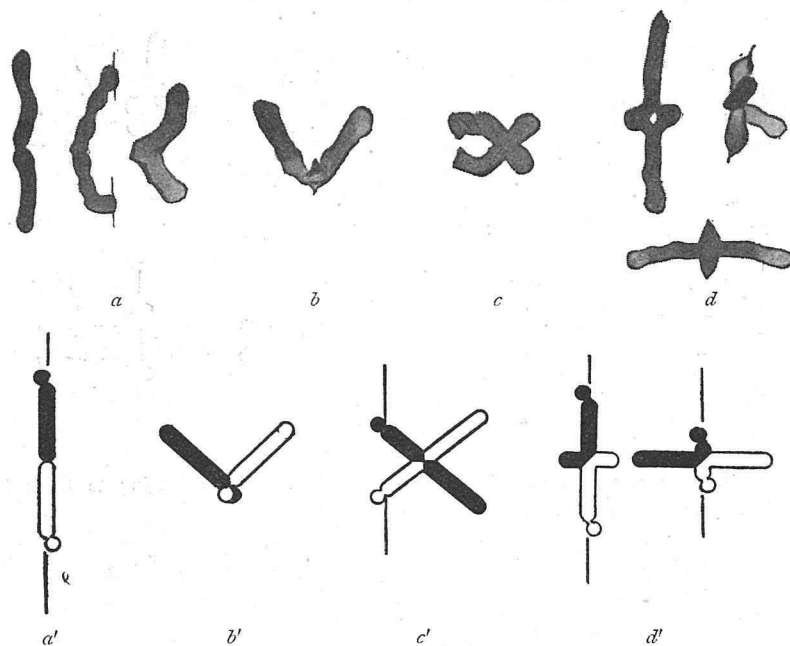


Fig. 2. Examples and schemas of configurations of m-gemini belonging to Classes I and II. Explanation in the text.

### Class I

There is no point of intersection between the two component univalents of a geminus (Fig. 2 a, a').

The components are attached to each other only at their distal ends or the ends opposite to the one near which the spindle fibers are attached to the chromosomes, and form a vertical V or rod more or less markedly bent into the shape of a longitudinally drawn out S or C at points near the free ends (Fig. 2 a, a').

### Class II

There is one point of intersection between the two component univalents.

1) The components intersect each other at their proximal ends near which the spindle fibers are inserted in the chromosome, and form a horizontal V with the spindle fibers attached at the apex of the V (Fig. 2 *b*, *b'*).

2) When the components intersect at an intermediate point between the two ends, we have an X (Fig. 2 *c*, *c'*) or a cross, or a vertical rod with a horizontal V in its middle, the length of the rod or the arms of the V varying according to the position of the intersection point on the chromosomes (Fig. 2 *d*, *d'*).

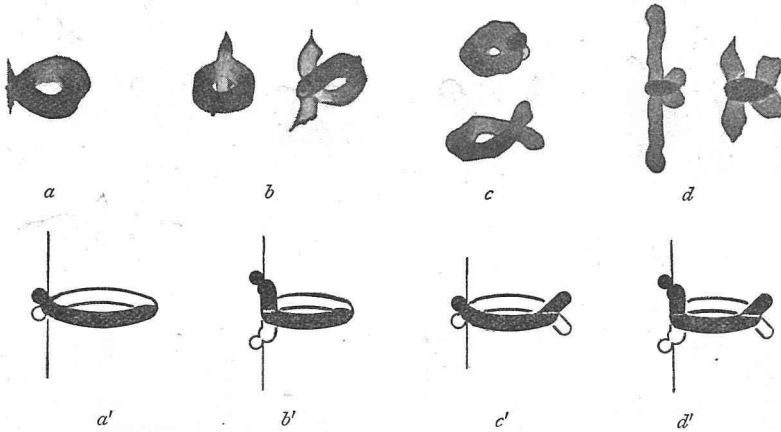


Fig. 3. Examples and schemas of configurations of m-gemini belonging to Class III. Explanation in the text.

### Class III

There are two points of intersection or attachment (Fig. 3).

1) When the two components are attached to each other at their distal ends and intersect at a point very near their proximal ends, we have a horizontal ring to the intersection side of which the spindle fibers are attached (Fig. 3 *a*, *a'*).

2) When the two components are attached to each other at their distal ends and intersect at a recognizable distance from the proximal ends, we have a horizontal ring with a vertical rod, the spindle fibers being found attached to the ends of this rod (Fig. 3 *b*, *b'*).

3) When the two components intersect at a point very near their proximal ends and also at a certain distance from the distal ends we have a horizontal ring with a vertical V on its distal side (Fig. 3 *c*, *c'*). In most cases this ring is disposed not precisely horizontally, but only nearly so. Very often chromatic lumps or processes are found on the side of the ring to which the spindle fibers are attached.

4) When the two components intersect at two points apparently away from both ends, we have a horizontal ring with a vertical V on one side of it and a vertical rod on the opposite to the ends of which the spindle fibers are attached (Fig. 3 *d*, *d'*).

Each two chromosomes reproduced in Fig. 3 *b-d* show that the point of intersection may occur at any corresponding levels of the component univalents.

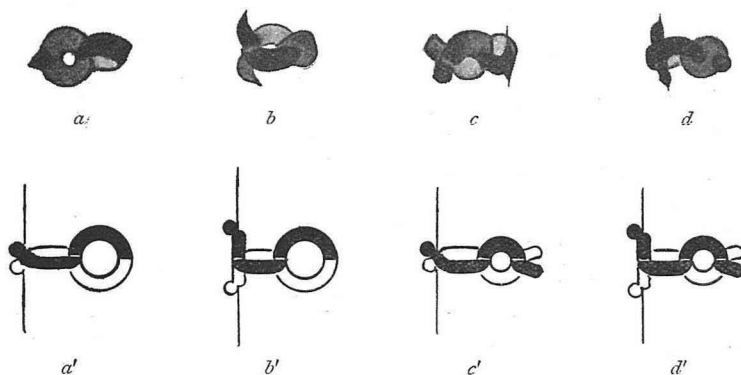


Fig. 4. Examples and schemas of configurations of m-gemini belonging to Class IV. Explanation in the text.

#### Class IV

There are 3 points of intersection or attachment (Fig. 4).

1) When the two components are attached to each other at their distal ends and intersect at a point very near the points of spindle fiber attachment, and also at a third point between these attachment and intersection points, we have a chain of two rings disposed perpendicularly to each other, the proximal ring being horizontal (Fig. 4 *a, a'*).

2) When the two components are attached to each other at their distal ends and intersect at two points recognizably away from both ends, we have a chain of two rings disposed perpendicularly to each other with a vertical rod on the free side of the horizontal ring, the spindle fibers being found attached to the ends of this rod (Fig. 4 *b, b'*).

3) When the two components intersect at a point very near the proximal end and also at two other points, we have a chain of two rings disposed perpendicularly to each other with a horizontal V on the free side of the vertical ring, the spindle fibers being found attached to that of the other ring (Fig. 4 *c, c'*). In most cases the latter ring is disposed not precisely but only nearly horizontally and we find very often two chromatic lumps or processes at the point of spindle fiber attachment.

4) When the two components intersect at 3 points quite away from both ends, we have a chain of two rings disposed perpendicularly to each other with a horizontal V on the free side of the vertical ring and a vertical rod on that of the horizontal ring, the spindle fibers being found at the ends of the rod (Fig. 4 *d, d'*).

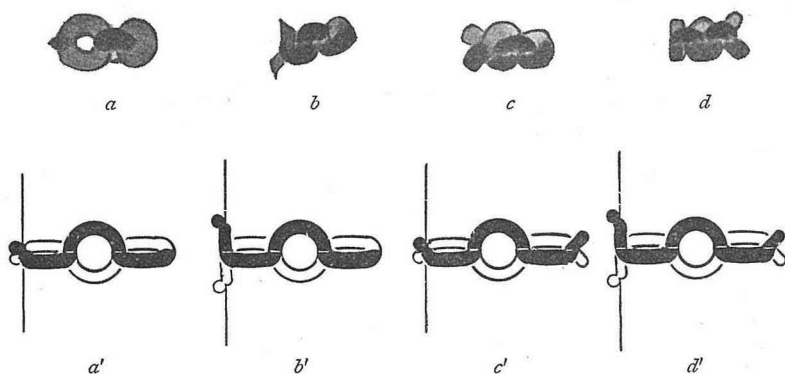


Fig. 5. Examples and schemas of configurations of *m*-gemini belonging to Class V. Explanation in the text.

### Class V

There are 4 points of intersection or attachment (Fig. 5).

We have in this class also 4 different cases which are quite similar to those found in Class IV, with the exception that in this case the number of rings of the chain is 3 and consequently the terminal V found at the distal end of the chain takes the vertical position.

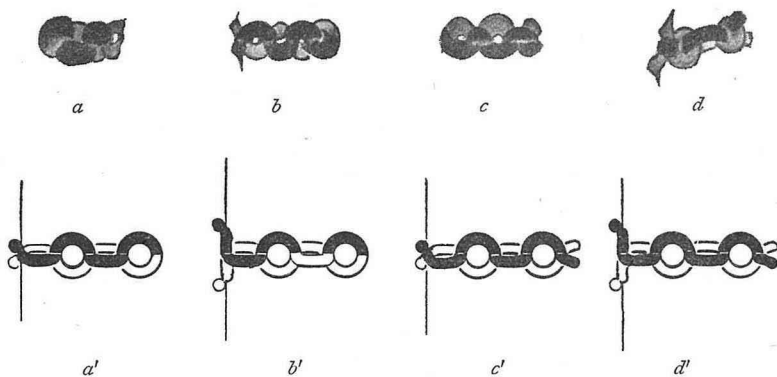


Fig. 6. Examples and schemas of configurations presented by the *m*-gemini belonging to Class VI. Explanation in the text.

### Class VI

There are 5 points of intersection or attachment (Fig. 6).

We have here also quite similar configurations to those described in Class IV, except that in this case the number of rings composing the chain is 4.

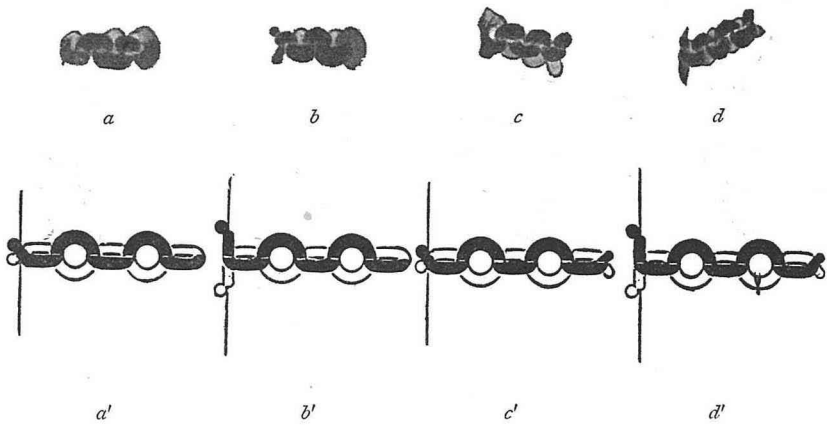


Fig. 7. Examples and schemas of configurations of m-gemini belonging to Class VII. Explanation in the text.

### Class VII

There are 6 points of intersection or attachment (Fig. 7).

Except that the number of rings of the chain is 5 all the configurations are quite similar to those in Class IV.

The M-geminus takes up also various configurations fundamentally in the same way as in the case of the m-gemini, but we have two points which must be mentioned here. 1. In most cases the number of points of intersection or attachment is considerably larger in this case than in the case of the m-gemini. No case has been met with where the number of points of intersection or attachment is less than 2. The configuration of a vertical loop with a horizontal V on one side was the simplest of all that were observed (Fig. 8 *a*). 2. The vertical ring to which the spindle fibers are attached is often drawn out into a rhomboid shape, and in cases where there are only a few points of intersection or attachment this ring is usually larger than the others. In Fig. 8 examples of the various configurations are given.

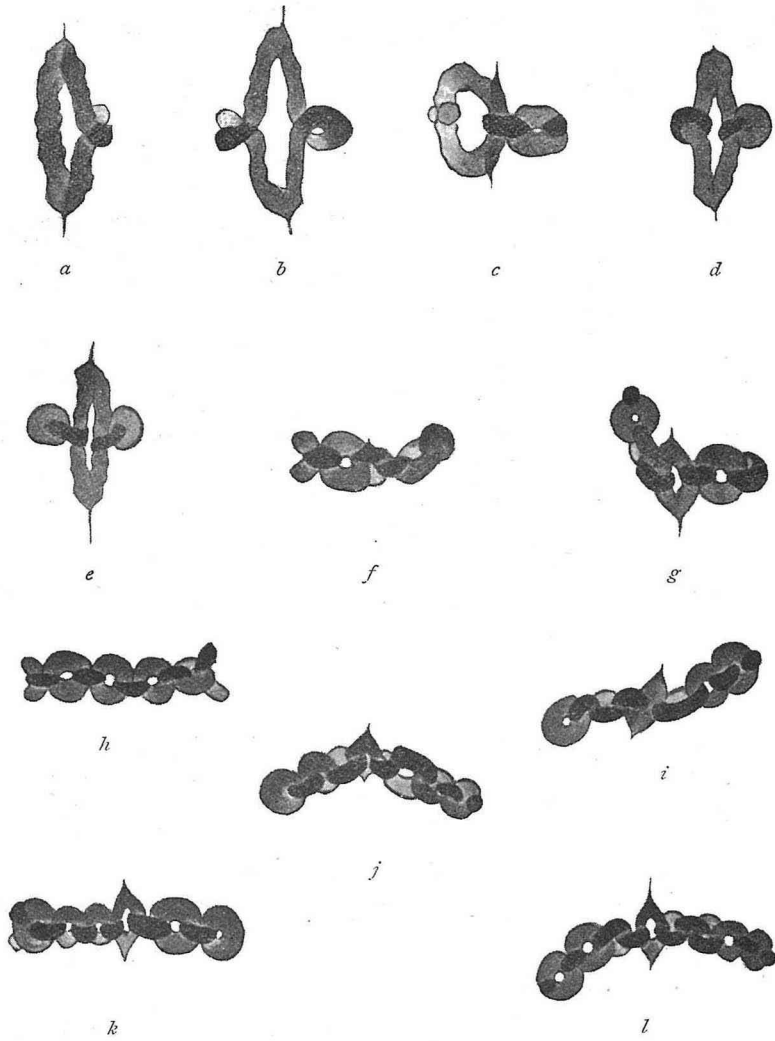


Fig. 8. Examples of configurations taken up by the M-gemini. The number of points of intersection or attachment is 2 in *a*, 3 in *b*, 4 in *c*, 5 in *d*, 6 in *e*, 7 in *f*, 8 in *g*, 9 in *h*, 10 in *i*, 11 in *j*, 12 in *k* and 13 in *l*.

In Fig. 9 some examples of earlier configurations of the m-gemini with different numbers of points of intersection or attachment are shown, and in Fig. 10 those of the M-geminus are given.



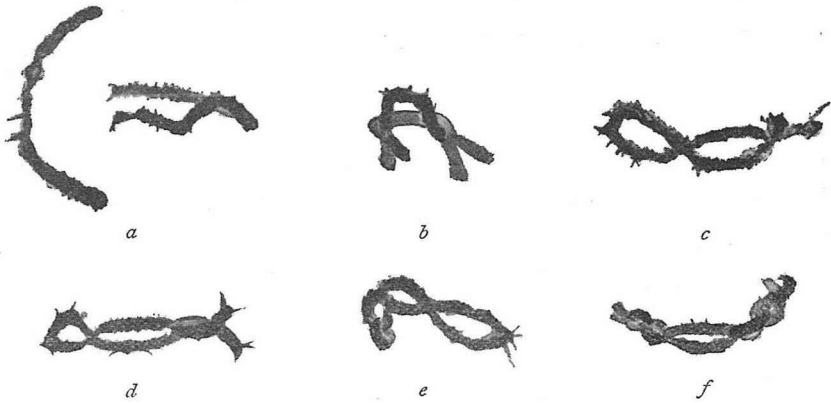


Fig. 9. Examples of configurations taken up by the m-gemini in the diakinesis. The number of points of intersection or attachment is 1 in *a*, 2 in *b*, 3 in *c*, 4 in *d*, 5 in *e* and 6 in *f*.

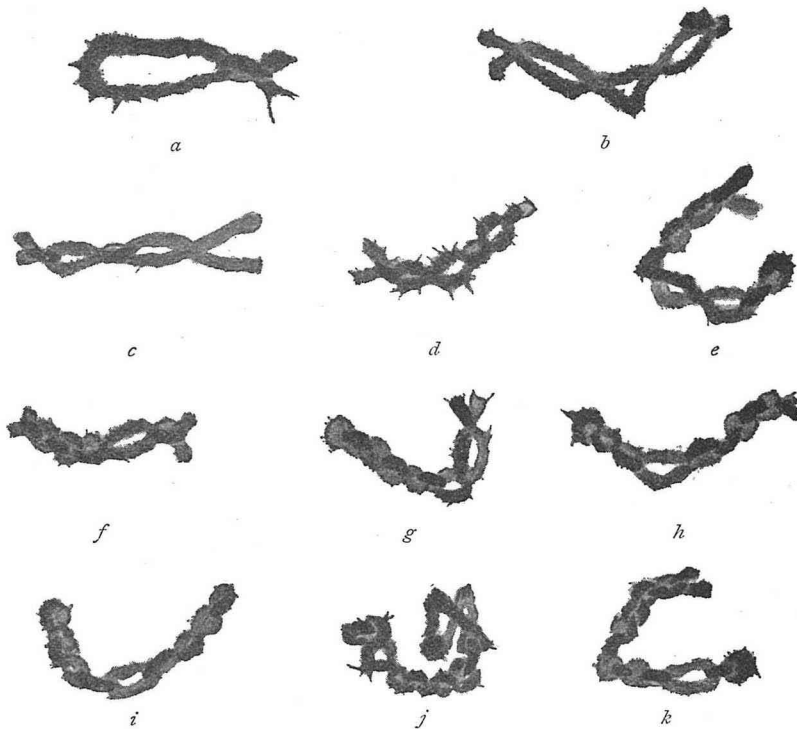


Fig. 10. Examples of configurations taken up by the M-gemini in the diakinesis. The number of points of intersection or attachment is 3 in *a*, 4 in *b*, 5 in *c*, 6 in *d*, 7 in *e*, 8 in *f*, 9 in *g*, 10 in *h*, 11 in *i*, 12 in *j* and 13 in *l*.

To obtain some knowledge about the quantitative aspect of the variation in the number of points of intersection or attachment a statistical investigation was attempted. In carrying out the investigation, only the chromosomes which were in the heterotype metaphase and only those which were found in an orientation convenient for observation were examined, and they were very carefully drawn by the aid of ABBE's camera lucida. The drawings were checked by repeated re-examination of the preparation so as to make the observational error as small as possible. The results obtained are shown in Table I and Table II.

TABLE I  
The case of m-gemini

Preparation No.	Number of points of intersection or attachment						Total
	0-1	2	3	4	5	6	
C <sub>3</sub> -B (3)	2	21	104	106	42	6	281
C <sub>3</sub> -B (1)	1	14	50	88	44	7	204
C <sub>1</sub> -B (1)	5	30	86	77	19	4	221
C <sub>2</sub> -B (1)	0	21	47	52	23	7	150
C <sub>3</sub> -N (4)	3	49	122	60	10	0	244
Total (%)	11 (1.00)	135 (12.27)	409 (37.18)	383 (34.82)	138 (12.55)	24 (2.18)	1100 (100)

As is seen from Tables I and II the variations range from the no-intersection-point class to the 6-intersection-point class in the case of the m-gemini, and from the 3-intersection-point class to the 13-intersection-point class in the case of the M-gemini. The mode is found in the former in the 3-intersection-point class and in the latter in the 8-intersection-point class. It is noted here that, while the curve obtainable from Table II is fairly symmetrical, that from Table I is asymmetrical.

If we calculate the mean number of the points of intersection by the formula  $M = \frac{\sum fV}{n}$ , we obtain  $M_m = 3.52$  for the m-gemini and

TABLE II  
The case of the M-geminus<sup>1)</sup>

Preparation No.	Number of points of intersection or attachment											Total
	3	4	5	6	7	8	9	10	11	12	13	
C <sub>1</sub> -B (3)	1	3	7	13	33	39	41	28	8	8	0	181
C <sub>2</sub> -B (1)	0	2	11	18	37	46	35	17	10	4	0	180
C <sub>1</sub> -B (1)	2	2	3	12	15	15	15	1	1	0	0	66
C <sub>2</sub> -B (1)	2	4	2	11	17	26	22	13	6	6	0	109
Na <sub>5</sub> (2)	1	0	3	6	11	24	22	24	18	7	4	120
C <sub>2</sub> -N (4)	1	0	6	26	28	28	11	5	2	0	0	107
C <sub>2</sub> -N (8)	0	2	5	16	35	34	17	7	3	2	0	121
C <sub>15</sub> -N (1)	0	1	4	8	22	21	24	8	6	4	0	98
C <sub>1</sub> -L <sub>5</sub> (3)	0	1	0	2	19	12	21	9	8	1	2	75
Total (%)	7 (0.66)	15 (1.42)	41 (3.87)	112 (10.60)	217 (20.53)	245 (23.18)	208 (19.68)	112 (10.60)	62 (5.86)	32 (3.03)	6 (0.57)	1075 (100)

$M_M=8.10$  for the M-gemini. This shows that the M-geminus can have as many intersection points as 2.3 times those of the m-gemini. It has been reported by SHARP (1914) that in the somatic cells of *Vicia*

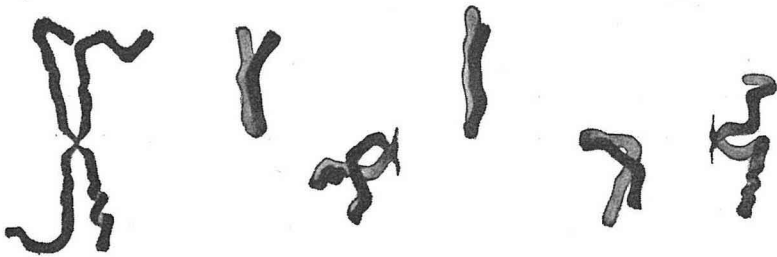


Fig. 11. Six pairs of chromosomes from a homotype spindle.

*fab*a two of the twelve chromosomes are about twice as long as the other ten. Fig. 11, which represents a group of 6 chromosome pairs

<sup>1)</sup> In Fig. 8a an M-geminus with two points of intersection or attachment is reproduced. This was drawn from a preparation which was not used for this statistical observation, and hence this case is excluded from this table.

in the stage of homotype metaphase in the variety *megarosperma*, shows that the M-chromosome is still longer than twice the length of the m-chromosome. Thus we see that the number of points of intersection between the component univalents is nearly proportional to the length of the chromosomes, and are led to the conclusion that the number of intersections or attachments is determined simply by chance.

### SUMMARY

In the pollen mother cells of *Vicia faba* there is found a wide range of variation in the configuration of gemini, which is due to the number of points of intersection or attachment of the component univalents in a period in the heterotype prophase.

The number of these points varies from 1 to 6 in the short m-chromosomes and from 2 to 13 in the long M-chromosome. The statistical results are shown in Tables I and II.

The ratio between the mean numbers of intersection points of the M-chromosome and m-chromosome is nearly equal to the ratio between the lengths of these two kinds of chromosomes, a result which suggests that the variation in the number of these points is merely a matter of chance.

In conclusion the author wishes to express his cordial thanks to Prof. Y. KUWADA of the Botanical Institute, College of Science, Kyoto Imperial University, for his kind advice and criticism, and also to Mr. S. NAGAO of the same institute for his kind help on the statistical side of this investigation.

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