Studies on spindle and chromosome movement

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Introduction

The important factors in mitosis, and even the course of cell division, are not yet sufficiently known. Also little is known about the differences in mitosis in absolutely normal cells. In the same material the course of cell division differs and is dependant on the quantity of cytoplasm, the arrangement of chromosomes in the plate, the displacement of vacuoles, the length and the breadth of the cell and other factors.

In chromosome movement kinotechores play the main role, but also mobile chromosome ends are known, though the behaviour of chromosome arms in mitosis is not quite clear.

The spindle mechanism is much better known in animal than in plant material and our data concerning its behaviour in plant anaphase are not complete.

The purpose of this work was to throw light on some of these problems. The author was especially interested in the following questions:

1. The length of the spindle in anaphase.
2. The course of mitosis in very thin cells.
3. Movement of chromosome arms in mitosis.

Material and methods

Root tips of the following plants were used: *Allium cepa* (2n = 16), *Vicia faba* (2n = 12), *Tradescantia virginica* (2n = 24), *Triticum vulgare* (2n = 42), *Tinantia fugax* (2n = 64, 68), Darlington and Janaki Ammal 1945), *Agrostemma githago* (2n = 24, 48, Darlington and Janaki Ammal 1945), *Plantago lanceolata* (2n = 12, 24, Darlington and Janaki

A m m a l 1945) and *Brassica napus* (2n = 36). The fixatives were: N a v a s h i n fluid diluted with water (1 : 1) and F l e m m i n g - B e n d a (for a part of *Allium* root tips). Material was cut 10—20 μ. and the following methods of staining were used: H e i d e n h a i n iron hematoxylin, N e w t o n gentian violet, G r a m gentian violet counterstained with orange G (G e i t l e r 1942), N e w t o n gentian violet counterstained with 0,05% alizarin viridin in 45% acetic acid and F e u l g e n reaction counterstained with the same solution of alizarin viridin. The best results were obtained with the last method and with hematoxylin. A diluted solution of alizarin was quite satisfactory for staining the spindle even in thick cut material, while more concentrated solutions stain intensively the cytoplasm (Ö s t e r g r e n 1949). To study the fibers slides were stained overnight with acetocarmin (E h r e n b e r g and Ö s t e r g r e n 1942). It is difficult to obtain satisfactory slides with Newton gentian violet counterstained with alizarin viridin because alizarin bleaks in gentian solution and gentian violet in ailzarin. After tests however, slides may be obtained in which both spindle and chromosomes are well visible.

A L e i t z oil immersion lens 100x N. A. 1,30, a Z e i s s apochromate oil immersion lens 90x N. A. 1,30 and compensating eyepieces 10x and 15x were used. Measurements were made with A b b é's camera lucida of Z e i s s.

Spindle length and spindle mechanism.

On fixed cells the spindle length in metaphase, successive stages (I, II, III) of anphase and in early telophase were measured. Out of the material three groups (I, II, III) of cells in anaphase were chosen and exclusively these cells in which the distance between kinetochores differed only slightly were classified into one group, as a result there was no continuity between these stages of anaphase. The reason for this procedure was to establish whether in anaphase the length of the spindle changes and if so, at which moment.

In plants with large and not numerous chromosomes — i. e.: *Vicia faba, Allium cepa, Tradescantia virginica* — the metaphase spindle in some cells is cigar shaped while in other it is elipsoidal. Rarely is the spindle pointed at the ends and halfspindles usually resemble truncated cones, or are more or less semicircular. It is seldom that the poles of the spindle are well visible. In plants with

a higher chromosome number *(Triticum vulgare)* or with small and numerous chromosomes — i.e.: *Tinantia fugax, Agrostemma githago, Plantago lanceolata* and *Brassica napus* the spindle is very seldom pointed at the ends and in most cases is elipsoidal; in consequence it is difficult to point out the poles exactly. The fibrillar structure of the spindle in metaphase is better visible near the kinetochores, than at the poles.

During the course of anaphase the halfspindles become more and more visible. In middle anaphase there are usually no difficulties to locate the poles of the spindle. In the course of anaphase the halfspindles become shorter and in later stages (end of anaphase) their length is 1—2 μ. In telophase they gradually disappear.

If mean values of spindle length are considered, it appears that the spindle in all studied plants elongates slightly in anaphase (I or II) cf. Table I. Such differences as appear in *Vicia,* where in anaphase I the mean value is 17,1 μ and in metaphase 17,3 μ, are surely accidental and may be not considered; this is the case also when the mean value in anaphase III is smaller than in anaphase II. In very early telophase in 7 out of 8 investigated plants, the distance between chromosome groups is longer than the spindle lenght in metaphase and only in *Tradescantia* it is shorter by 0,4 μ. The distance in early telophase may be considered — with little error — as the maximal separation between two groups of anaphase chromosomes; actually as in plant telophase due to contraction of chromosomes and formation of the phragmoplast, the distance between chromosome groups dimnishes, the maximal separation may be a little greater. In most cases the maximal separation is shorter then the length of the spindle in late anaphase.

According to R i s (1943) in animals there are two modifications of anaphase: 1. Chromosome separation is due to the shortening of chromosomal fibres and elongation of the spindle; these two processes may be separated in time. This type was found by R i s in: embryonic cells of *Tamalia,* primary spermatocytes of *Thelia, Protenor* and in other insects. 2. Only spindle elongation causes the movement of chromosomes; the length of the fibres is contant. R i s found this type in primary spermatocytes of the aphid *Tamalia.* He maintains (1943) that in plant anaphase chromosome movement is caused only by the shortening of chromosomal fibers. In some exceptional cases however in plants the spindle elogates in anaphase (S m i t h 1934). The measurements done on plants in the present

work indicate that R i s'es assumption is correct, as in plants the spindle elongation is very slight and cannot be compared to the spindle elongation in animals.

The hypothesis of traction fiber is undoubtedly the one best proved from all on chromosome movement, though we do not know exactly which fibers are responsible (C o r n m a n n 1944). The force moving the chromosomes is an elastic one (H u g h e s a n d S w a n n 1948, B a j e r and H r y n k i e w i c z 1950).

Il is not true however that the movement causes direct shortening of elastic fibers; this is indicated by measurements of the length of anaphase spindles, the maximal separation between chromosomes, and of the length of chromosomal fibers. In metaphase the fibers are fixed to kinetochores in steaks which become fainter as distance from kinetochores increases. In all my material they were never visible at the poles in metaphase. In the course of anaphase their new parts become distinctly visible and most so near the kinetochores. Later they become shorter and in late anaphase they are distinctly discernible as very short threads connecting the poles with kinotechores. As I was able to find in plants with large chromosomes, their thickness near the kinetochores is approximately the same both in metaphase and in anaphase. If the movement of the chromosomes were caused by direct shortening of these fibers it seems obvious that their thickness would increase in the course of anaphase. It appears therefore that the submicroscopical structural changes which are the basis of the movement (F r e y - W y s s l i n g 1946) must be of a very special kind. At the end of anaphase in plants with large chromosomes the length of the fibers is usually below or approximately 1 μ. As even fibers of such length may pull the chromosomes, and as they are best observable close to kinetochores, much better in anaphase than in metaphase, it seems probable that throughout the anaphase only the part of the fibers in close proximity to the kinetochore is active and causes most of the movement.

B ě l a ř (1929) suggests that there are two main anaphase factors: pulling (it would be the fiber action) and pushing (action of „Stemmkörper"). „Stemmkörper" originates from the spindle after the passing of the chromosomes through it on their way to the poles; its activity in animal division is evidant but may be explained also by other factors (cytoplasm streaming). As in polarised light the spindle is birefractive and „Stemmkörper" shows no birefraction (S c h m i d t 1937, 1939), halfspindles must have a different struc-

TABLE I.

Spindle length in mitosis and maximal separation of chromosomes (approximately early telophase) in μ. Mean values of 25 cells.

	Metaphase			Anaphase I			Anaphase II			Anaphase III			Maximal separation (early telophase)		
	mean	min.	max.	mean	min.	max.	mean	min.	max.	mean	min.	max.	mean	min.	max.
Allium cepa	*18,8*	15	21,5	*19,1*	17	22,5	*20,6*	16,5	23	*22,4*	17	25	*18,2*	16	24
Vicia faba	*17,3*	15	21,5	*17,2*	13	23,5	*19,8*	16	24	*21,9*	17	28	*20,6*	16,5	24,5
Tradescantia virginica	*22,6*	17	28	*21,3*	15	27	*24,6*	21	30	*26,5*	22	30	*22,2*	15	27,5
Triticum vulgare	*15,3*	12	20	*15,8*	13	19,5	*17,5*	14	19,5	*17,3*	12	19	*17,5*	15	19,5
Tinantia fugax	*11,3*	9	13	*13*	9,5	17	*13,8*	9	20	*13,8*	10	21	*12,5*	10	21
Agrostemma githago	*9,6*	7	10,5	*10,6*	8	15	*10,5*	8,5	13,5	*10,8*	8,5	14,5	*10,8*	7	17,5
Plantago lanceolata	*8*	6,5	9,5	*8,8*	7,5	10	*9,3*	7,5	10,5	*9,7*	8	11	*8,4*	7	10
Brassica napus	*7,8*	6	10	*8,6*	6,5	10	*8,6*	6,5	11,5	*8,7*	6,5	11	*8,3*	6,5	9,5

TABLE II.

Spindle length in very thin cells of *Tinantia fugax* (Cells not included into Table I)

	Spindle length in μ										Mean value
Metaphase	18	19	20	20	20	20	21	22	22,5	23	21,6
Anaphase	17	18	20	20	20	22	23	23,5	24	25	22,3
Early telophase	17	18	18	19,5	20	20	20	21	22	25	21,1

ture from „Stemmkörper". All elements in cell division (chromosomes, spindle, „Stemmkörper"), must be considered as a separate system and it seems probable that submicroscopical changes in „Stemmkörper" may cause greater or lesser movement in different objects. In some cases this seems to be the action of „Stemmkörper" in plants. In this way may be explained the facts observed in staminal hairs of *Tradescantia virginica* in cells 5—6 from the top of the hair (B a j e r 1950). Two large vacuoles which are found usually at the ends of such cells become concave in late anaphase — which suggests that a very weak force acts from the equator of the cell.

Perhaps it is allowable to assume that the lack of spindle elongation in plants and the visible action of „Stemmkörper" is caused by mechanical factors (cell walls). If this was correct such an elongation would be found in long cells, as the distance between two groups of chromosomes in telophase is very considerable in such cells, and in other cells in telephase, chromosomes are often very close to the walls. In practice in very long and usually thin cells the anaphase separation does not meet obstacles any longer. Observations in all material and measurements of spindle length in cells longer than 50μ were done in *Tinantia fugax* (Table II). As the long cells are very thin it is very difficult to discern whether there is late or early anaphase and so such stages were not distinguished. Measurements prove that in such cells there is also no spindle elongation. The distance between two groups in telophase in these cells is much longer (of Table I) which is caused by the length of the spindle and oblique metaphase plate. The more oblique the plate, the longer is the separation of chromosomes, though the distance travelled does not differ much from that in cells with normal dimensions. The explanation is in Fig. 1. The distance between two groups is longer, because the contraction in telophase is towards the poles. This means that the spindle does not elongaie in long cells and there is no special action of „Stemmkörper" which would be comparable to that of animals.

When we compare the distance between two chromosome groups in late telophase in root tips and *Tradescantia* staminal hair cells it is obvious that in the first case the contraction is smaller and begins later.

Conclusions

Slight elongation of the spindle in plant anaphase may be caused by one or two main factors:

1. The formation of the very ends of the spindle in the first part of anaphase. It is indicated by the fact that at the poles the spin-

dle is not distinctly visible in metaphase and its shape at the poles is variable in different cells. In most cases the pointed spindle is discernible in anaphase.

2. It is not improbable that change in length is due to „Stemmkörper" action which is indicated by some observations. However in plants the action of this last factor is not proved. Especially in the case of plants with large chromosomes *(Vicia, Allium, Tradescantia)*, the first factor seems to be more probable. The elongation of the spindle is so slight that approximately it may be considered as non existant.

Cell walls do not cause the slight elongation of the spindle. This is proved by two facts:

1. Chromosome groups at the end of anaphase often stop at some distance from cell walls.

2. Observations in long cells where the spindle has the possibility of elongation without meeting obstacles. In this case a slight elongation only was found; the mechanism of very long separation of the chromosomes in such cells is explained above.

Mitosis in thin cells

In most root tips near the center very thin and long cells with elongated nuclei are found. Cells of this shape were found in all studied plants, most frequently however in plants with large chromosomes *(Vicia, Allium, Tradescantia, Triticum)* and also in *Tinantia* and *Agrostemma*. The course of mitosis was studied in such cells. During prophase the nuclei do not change their shape and after the desappearing of nuclear membrane the chromosomes are scattered over a large space. Before metakinesis the chromosome are not near each other as — is the case in most meristematic cells where orientation of chromosomes in prophase is the same as in last telophase (B ě l a ř 1929, M a k a r o w 1948) — but are scattered. Metakinesis similarly to the further stages appears to be at first much more irregular than it is in reality. The very irregular and probably longer lasting movement at this stage is caused by the lack of room: the breadth of the cell is not large enough to allow the chromosomes to form the metaphase plate perpendicularly to the long axis of the cell. The formation of the plate perpendicularly to the long axis of the cell would be possible if the chromosomes were at least half as numerous as is the case. Such plate would perhaps be formed, if the chromosomes were close to each other, and it seems strange that this does not happen.

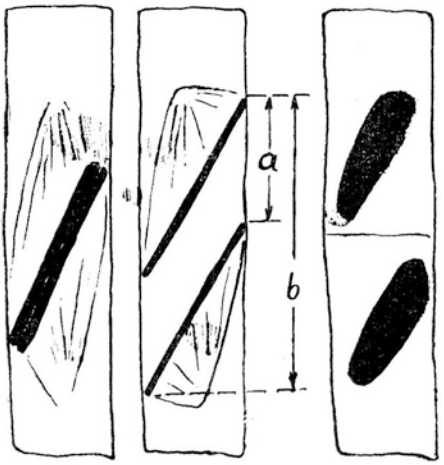

Fig. 1. Diagram of cell division in thin cells. Oblique metaphase plate; two groups of chromosomes moving parallely to the position of the plate. The distance travelled by the two groups of chromosomes = a (approximately); contraction in telophase is usually in direction of the poles and causes the distance between nuclei in telophase (b) to be greater than the maximal separation.

As a result of metakinesis a very oblique metaphase plate is formed and its angle to the long axis of the cell is often less than 20°. The arrangement of the chromosomes in the metaphase plate is well visible only from one side of the plate; it is only when the plane, in which the kinetochores lie, is parallel to the optical axis of the microscope. In this case — though usually with some difficulties — the spindle is also discernible. From approximately 50 metaphases obserdle is also discernible. From approximately 50 metaphases observed in *Allium* I was not able to establish the plane of metaphase plate only in less than 10 cases (cf. Fig. 2b). If such great mechanical difficulties in the formation of metaphase plate are considered, the plate will be found very regular. The analysis of the plate is difficult and not possible in all cells. Where it was possible it was found that the kinetochores lie in one plane and only few of them are out of the plane, which is normally found in most metaphases in cells with average dimensions. In metaphase both the plate and the individual chromosomes oscillate irregularly (Hughes and Swann 1948, Molé-Bajer 1951) and therefore the kinetochores which are not in plane of the plate are probably pulled to one of the poles. In cells of normal size, kinetochores of two chromosome groups do not begin to move simultaneously and

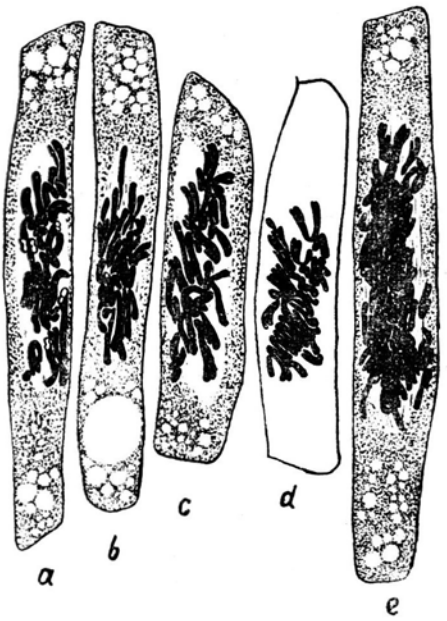

Fig. 2. Metakinesis and metaphase in thin cells. a—c. *Allium cepa*, d — *Triticum vulgare*, e — *Tinantia fugax*. a and e metakinesis, chromosomes scattered over large space (cf. Fig. 3a); b — metaphase or late metakinesis — it is not possible to establish the position of kinetochore plane; c — metaphase-kinotochore plane not exactly perpendicular to the axis of the microscope d — metaphase a—c and *e Feulgen* reaction and alizarin virdin, d — *Newton* gentian violet. a—d — 1000×, e — 2000×.

gradually chromosomes move to the poles, one by one. In the case of small and numerous chromosomes this gradual movement is much more distinct, anaphase begins often on one side of the plate and slowly all the plate is affected. At the beginning of the movement the differences in distance travelled are small and usually less than 1μ. Often however all chromosomes in the plate begin their movement simultaneously. Also at first it that the anaphase in very thin cells is much more irregular than it proves to be after exact analysis. Often the movement of all chromosomes in anaphase does not begin exactly at the same moment and the mechanical conditions make the anaphase more difficult. Chromosomes placed at the edges of the plate have no difficulty to move to the poles. On the other hand those placed near or at the center of the plate, move usually with difficulty or are not able to move at all. In the latter case these chromosomes begin the anaphase movement a little later. If all chromo-

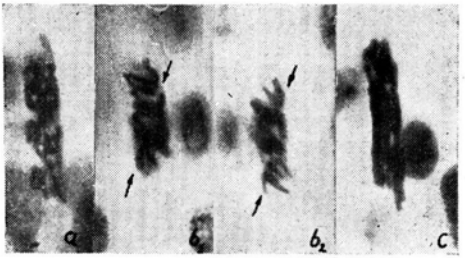

Fig. 3. Metakinesis and metaphase in thin cells. a — *Allium cepa* — metakinesis cf. Fig. 2a; b_1—b_2 *Triticum vulgare* the same cell — metaphase — arrows indicate the plane of kinotochore plane cf. Fig. 2d; c — metaphase in *Allium cepa* where the kinetochore plane is not perpendicular to the microscope axis. a, c. — *Feulgen* reaction and alizarin virdin. b. — *Newton* gentian violet.

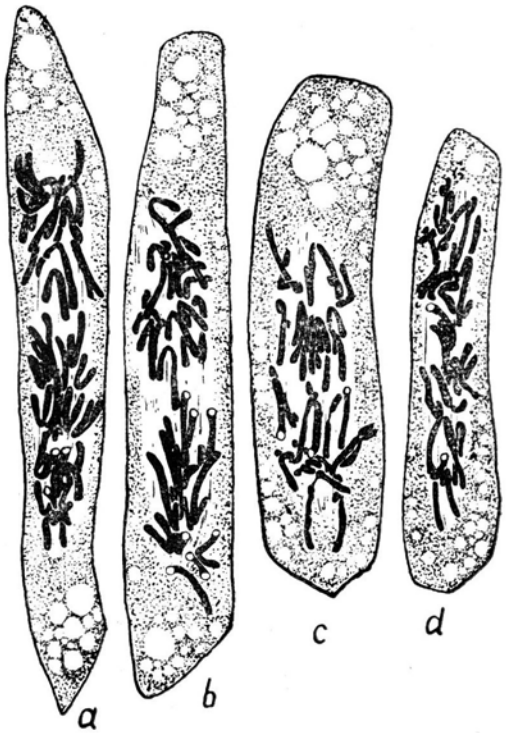

Fig. 4. Anaphase in thin cells. a—b *Allium cepa* cf. Figs. $5a_1$—a_2 and 5b; c—d *Vicia faba*. Different orientation of chromosomes moving to the poles. Anaphase seems to be very irregular due to oblique position of metaphase plate. *Feulgen* reaction and alizarin virdin. 750×.

somes may move at the first stage of anaphase, kinetochores of two chromosome groups would move approximately parallely to the position in the metaphase plate. The orientation of the chromosomes in anaphase is different and it is more or lesss regular. As the chromosomes are crowded their arms move often before the kinetochores and usually their orientation is very strange. In Figs. 3—5 some anaphases are assembled and represent the course of such anaphases. In most cases the anaphase movement is not so irregular as it seems to be, and two nuclei are formed. No efforts were made however to examine the numbers of the two chromosome groups in late anaphase.

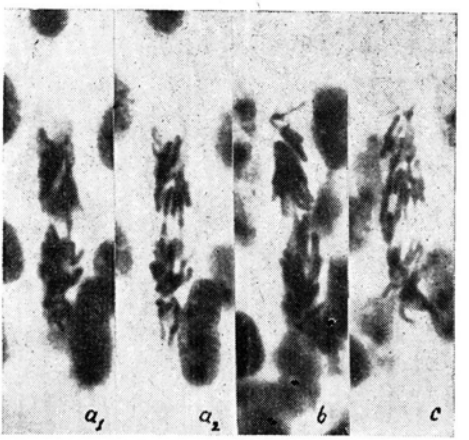

Fig. 5. Anaphase in thin cells in *Allium cepa*. a_1—a_2 the same cell cf. Fig. 4a; b cf. Fig. 4b; explanation in text. F e u l g e n reaction and alizarin viridin.

In some cases the separation of the two chromosome groups is not considerable enough and restitution nuclei are formed in telophase.

In telophase very thin and long nuclei are always formed.

C o n c l u s i o n s

Observations of mitosis in thin cells give further informations concerning metaphase and anaphase. The formation of metaphase plate, which in such difficult mechanical conditions must be considered as quite regular, proves that the metaphase stage, with the plate in which most kinetochores are exactly in one plane, is necessary in normal cell division. Its role in the course of mitosis is not known however.

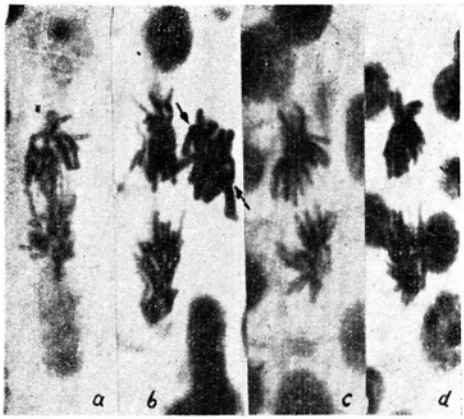

Fig. 6. Anaphase in thin cells in *Allium cepa*. a earlier stages, b—d later than in Fig. 5. b — near one group of anaphase chromosomes a slightly oblique metaphase plate; arrows indicate the position of kinetochore plane; c—d late anaphases most regular from all represented and forming transition to normal anaphase with numerous chromosome arms preceding kinotechores in their movement (cf. text). F e u l g e n reaction and alizarin viridin.

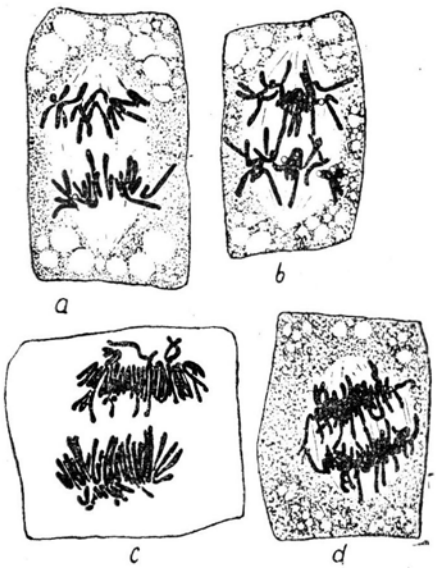

Fig. 7. Movements of ends of chromosome arms. a—b — *Allium cepa*, c — *Triticum vulgare*, d — *Tinantia fugax;* a — only some arms precede kinetochores; b — numerous arms precede kinetochores cf. Fig. 8b; c — late anaphase chromosome arms bended by cell walls cf. Fig. 8c. a, b and d *Feulgen* reaction and alizarin viridin. c — *Newton* gentian violet. a—c 725×, d 1450×.

Observations of chromosome movements in anaphase show some of their characteristic features. Usually chromosomes in the middle of the plate cannot move in the first stages of anaphase and „await" some free space. This means that their ablility to move lasts some time, which confirms the statement according to which the chromosomes move, so as if every one of them had its own movement mechanism and each one moved independantly of the other.

Movement of chromosome arms

The observation of anaphase in root tips shows that the course of anaphase is not the same in all cells, though usually the differences are in detailes only. Differences may be caused by mechanical factors as is the case with thin cells. In cells with mean dimensions (though in large and older cells vacuoles may cause some difficulties in division), of which the cross section is rectangular, and containing much cytoplasm, the cell division is most regular. In the case of *Allium*, in which anaphase was analysed most exactly, typical anaphase, in which kinetochores of all chromosomes were orientated towards the poles and chromosome arms had V shaped arrangement (kinotechores in *Allium* have medial or submedial position), was found in less than 20% of cells. Such orientations found more often in late than in middle anaphase. Early stages of anaphase cannot be considered because at this moment the chromosomes change their orientation.

The duration of changes in orientation differs, but there is no doubts that late stages of anaphase are stabilised. In this stage chromosomes move to the poles without changing their orientation, and it is visible in numerous cells that the ends of some chromosemes

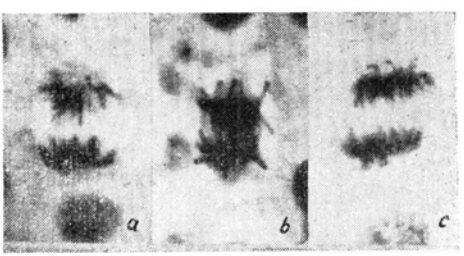

Fig. 8. Movements of chromosome arms. a—b *Allium cepa*, c — *Triticum vulgare*. a — only some, b — numerous arms precede kinetochores (b cf. Fig. 7b). c — chromosome arms bended by cell walls (cf. Fig. 7c). a—b Feulgen reaction and alizarin viridin, c — Newton gentian violet.

precede kinetochores in their movement to the poles. Some chromosomes have a V shaped arrangement with arms and not kinetochores pointing to the poles. Very often one arm of the chromosome is the first to move and seems to pull the kinetochore and the second arm; in such cells two arms may form a stright line. In cells with similar shape the number of chromosomes in which the ends precede kinotechores differs. I two groups of chromosomes in anaphase the number of such ends preceding kinetochores is usually different but the differences are not great and often the following numbers were found: 6—5, 5—3, 4—3, 4—2, 4—1 and so on. The dependence between the thickness of the cell and the number of such ends was found: the thiner the cell the more chromosome arms precede kinetochores in anaphase. It was ascertained that a continuous transition between anaphase in thin cells and anaphase in cells with normal dimensions exists as was previously described.

Throughout anaphase the orientation of such chromosome arms usually does not change and their movement is approximately parallel to the spindle. In many cases their orientation may change, and in late anaphase they may be placed across the spindle. The arm ends however may stretch into the cytoplasm. In telophase, when maximal separation of the chromosomes is approximately equal the cell length, the arm ends touch the cell walls and are bended.

The movement of chromosome ends was confirmed in plants with large chromosomes *(Vicia, Allium, Tradescantia, Triticum)* and in these with small but long chromosomes*(Tinantia, Agrostemma).*

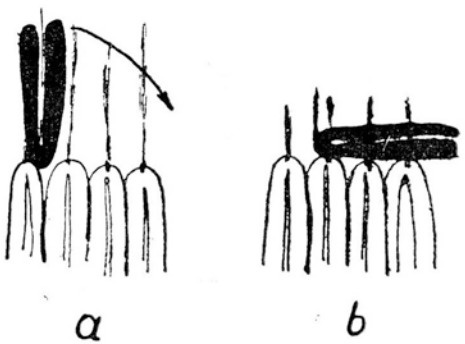

Fig. 9. Arrangement of some chromosomes in early (a) and late (b) anaphase which shows the possibility of transverse movement across the spindle; only in such a case the change of posi.ion from a to b — may be explained

In plants with small chromosomes such as *Tinantia* it is difficult though possible to examine the orientation and arrangement of chromosomes in anaphase, in other plants however no efforts of analysis were made.

The number of such moving chromosome arms increases in dependence of the chromosome number, and in *Tinantia* often surpasses 20 ends. Similarly in *Triticum* more mobile ends are observed than in *Tradescantia* and in *Allium*. Such ends are least numerous in *Vicia faba*.

After staining fibers (acetocarmine slides) connected with kinetochores no fibers attached to chromosome ends were noticed.

Conclusions

In metaphase the chromosomes are differently orientated in the plate and are more or less crowded. Although chromosome arms stretch in all directions, kinetochores are in one plane. If chromosomes are crowded and their arms are out of the kinetochore plane, arms of some chromosomes in anaphase may be pushed before kinetochores. In the course of anaphase the breadth of chromosome groups dimnishes and as a result the orientation of chromosome arms in telophase is often similar to that of early anaphase or even metaphase. The movement of chromosome arms is caused by the orientation of metaphase chromosomes and is dependant on chromosome crowding. This movement is not active, as no fibers connecting the ends were noticed. The movement is of a different kind than in meiosis, where it is caused by fiber action and the force acting on the ends is of the same order as the one acting on kinetochores (Östergren and Prakken 1946).

The movement of chromosome ends in anaphase and the bending of chromosomes observed in short cells in telophase indicate that the chromosomes are rather stiff.

Observations of chromosome arm movements seem to indicate on the possibility of a transverse movement across the spindle (Östergren 1949, Gajewski 1949, Bajer 1951). If this were possible the expectetd change in the orientation of chromosomes with a particular arrangement in anaphase, would develop as in Fig. 9. Such cases were obserwed, and this supports this assumption.

SUMMARY

The course of mitosis was studied in root tips of plants with large *(Allium cepa, Vicia faba, Tradescantia virginica, Triticum vulgare)* and small *(Tinantia fugax, Agrostemma githago, Plantago lanceolata, Brassica napus)* chromosomes and the following results were obtained:

1. In anaphase as compared to metaphase, spindle length slightly increases (by several microns). It may be caused by two factors: a) formation of the very ends of the spindle just in anaphase, b) action of „Stemmkörper". The action of the later factor cannot be considered as being proved in plants.

2. As in long cells the spindle does not elongate the lack of this elongation cannot be caused by mechanical factors (cell walls etc.).

3. Chromosomes in anaphase do not begin their movement simultaneously, though the differences are slight.

4. The course of mitosis in very thin and long cells appears to be irregular, though after exact analysis it usually proves to be normal which is proved by the following facts:

a. The metaphase plate is formed normally and most kinetochores lie exactly in one plane.

b. The orientation of anaphase chromosomes is caused by the oblique position of metaphase plate.

c. In most cases two nuclei are formed and though it seems strange, only very rarely restitution nuclei are formed.

5. The ability of motion of chromosomes lasts some time and they move as if each of them had its own movement mechanism.

6. Often in anaphase ends of chromosome arms precede kinetochores in their movement to the poles, which is caused by the orientation of metaphase chromosomes and most probably not by fiber action.

7. The orientation changes of some chromosomes seem to indicate that there is a possibility of transverse movement across the spindle .

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