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On the Spiral Structure of Chromosomes in Some Higher Plants

By

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With Plate XVII

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In recent years, the spiral structure of chromosomes has become increasingly discussed, though this structure formerly received little attention from most cytologists.

The aim of the present observations was to see how widely the structure is visible in higher plants rather than to look into its real nature.

OBSERVATION

Observations were made with fresh pollen mother cells of any plants placed at my disposal, by staining them with acetocarmine with which Fujii (1926) first succeeded in demonstrating very clearly this structure in pollen mother cells of *Tradescantia virginica*. With this method of staining, chromosomes are stained deeply and present some varieties of structure which appears differently according to the size of the chromosomes. When the chromosomes are of relatively large size, the structure is mostly of a spiral nature, more or less distinct. The plants in which the spiral or zig-zag structure was observed with more or less distinctness are :—

Cryptomeria japonica; Najas major; Sagittaria Aginashi; Secale cereale; Tradescantia virginica; Rhoeo discolor; Zebrina pendula;

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Chlorophytum comosum; Hosta Sieboldiana; H. Sieboldiana, var. longipes; Hemerocallis longituba; H. fulva var. Kwanso(?); Kniphofia sp.; Allium fistulosum; A. Cepa; A. odorum; Tulipa sp.; Lilium tigrinum; L. auratum; L. elegans; L. avenaceum; L. concolor var. parthenion; L. speciosum; L. davuricum; L. japonicum; L. longiflorum; Muscari botryoides; Tricyrtis hirta; Aloë variegata; Amaryllis sp.; Crinum asianticum var. japonicum; Narcissus poeticus (?); Iris Kaempferi; Belamcanda punctata; Phajus grandifolius (?); Lathyrus odoratus; L. maritimus; Solanum glaucum.

When the chromosomes are of small size, this structure is obscure, the chromosomes appearing as homogeneous solid masses. The plants carrying these chromosomes are :—

Triticum sativum (?); Pollia japonica; Asparagus officinalis; Yucca sp.; Deutzia gracilis; Aristolochia debilis; Rumex acetosa; Fagopyrum esculentum; Papaver somniferum; Passiflora caerulea; Lythrum Salicaria vax. vulgare; Primula japonica; P. malacoides; Datura sp.; Veronica sp.; Antirrhinum majus; Digitalis purpurea; Luffa cylindrica; Cucurbita pepo; Platycodon grandiflorum; Oenothera sp.; etc.

Heterotype division

The fine threads in the early prophase are too slender for their structure to be clearly traced with this method of staining, though there are some exceptional cases such as *Lilium* and *Amaryllis* in which a fine zig-zag structure is observable. Thicker threads which have been spun out of the synizetic knot sometimes present relatively distinct zig-zag aspects. This zig-zag appearance of the threads is especially clearly observed in the young pollen mother cells of *Amaryllis, Tricyrtis, Hemerocallis* and *Aloë*. As the condensation of the threads proceeds, the zig-zag or spiral aspect becomes distinct. In *Najas, Hosta, Tricyrtis, Hemerocallis, Amaryllis* and *Tulipa*, it is recogniable in each strand of the twisted bivalent threads.

In diakinesis, contrary to expectations, most chromosomes do not

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present the coiling appearance, but often show, in many plants, a meshlike structure. Only in some few cases, such as in *Lilium*, *Hosta*, *Amaryllis* and *Narcissus*, each component thread of gemini is observed to be composed of two interlaced or parallel-running chromatic spirals, though they are not so distinct as in the other stages.

In heterotype metaphase, the chromosomes become massive and the pitch of the turn of their spiral is so short that it is often very difficult to trace from one turn to the other, so that the chromosomes appear as if they consisted of many discontinuous discs. This reminds us of the description by BELLING (1926) of the chromosome structure. A clear manifestation of the spiral can, however, be attained artificially by a slight pressure upon the cover glass. It can be clearly demonstrated in this way, especially in Tradescantia, Rhoeo, Zebrina, Hosta and Lilium. In Figs. 1 to 5, such chromosomes from various plants are shown. In Tradescantia, Rhoeo and Aloë et al., each univalent chromosome contains in it a somewhat large spiral which seems to be formed of a coiling band (Fig. 5). While this thick band has been found by KAUFMANN (1926) in Tradescantia pilosa, by KUWADA and SUGIMOTO (1926) in Tradescantia virginica to be double, Fujii (1926) regards it as a secondary spiral which consists of a more slender primary spiral. Often the peripheral regions of this band stain far more deeply than the middle region, and it presents an appearance of a double spiral. This double aspect of the spiral is visible in several regions as indicated by an arrow in Fig. 5. Figs. 1 to 6 show moreover that the chromatic portion of the chromosome forms a continuous spiral or zig-zag thread instead of consisting of a row of discontinuous chromatic bodies or discs as reported by SANDS (1923) and BELLING (1926). (Compare SANDS' figures in Plate XXIX and Belling's Fig. 3 with Figs. 3 and 5 in the present paper.) A more clear evidence for the continuous nature of the chromatic portion in question is afforded by the drawn out portion of chromosomes separating from each other to the poles (Fig. 7).

It is noted here that in Fig. 7, there can be seen two chromatic

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bodies each of which is connected by a long drawn-out spiral thread to the distal end of the separating chromosome. This seems to suggest that in a certain region of the chromosome the spiral is liable to be unravelled, while in the bulk of it, it has a strong tendency to retain its coiling nature. If such a region is found near the end of the chromosome, the end region will thus appear like a "satellite".

In the late anaphase when all the chromosomes have just arrived at the poles, it is possible to trace distinct spiral threads in each half of the longitudinally divided chromosomes in *Lilium*, *Hosta*, *Najas*, *Tricyrtis*, *Secale* and *Tulipa*. Figs. 6 and 8 show the chromosomes of *Lilium* and *Hosta* in this stage.

In telophase, although all the chromosomes fuse together and the boundary of each chromosome becomes obscure, there is still a certain indication of zig-zag or spiral features in the chromosomes. One of the daughter chromosome groups in *Hosta* is reproduced in Fig. 9.

Interkinesis

Observations of some recent workers in the meiotic division have confirmed the conspicuous spiral appearance of chromosome threads in the interkinetic nucleus in certain plants; for instance, in Fritillaria by NEWTON (1926), in Lathyrus by MAEDA (1928), in Crepis by KATÔ (unpublished) with fixed materials. In *Tradescantia*, Fujii (1926) has also demonstrated fine coiled threads with acetocarmine and regards these coils as the primary spirals. In the present observations of Zebrina, Tradescantia, Lilium, Tricyrtis, Rhoco and Sagittaria, we find certain differences among them in the behaviour of their chromosomes in interkinesis. While in Sagittaria, the chromosomes remain in a less unravelled state, in the other plants, they are unravelled to a more or less extent and run sinuously, though the acetocarmine preparation may not allow us to draw any definite conclusion. Fig. 10 shows slender chromosomes in Zebrina of a far finer spiral structure than in the heterotype division. In *Tradescantia*, this fine spiral becomes unravelled to a certain extent and assumes a sinuous aspect in the

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mid-interkinesis.

Homotype division

In the homotype metaphase or a slightly earlier stage where the chromosomes become contracted again in length, we can easily discriminate very distinct coils or spirals in the chromosomes (Figs. 11 to 13). They are much more slender and the number of their turns appears to be more numerous than in the heterotype division. While in *Lilium, Rhoco, Allium, Tricyrtis, Najas* and *Hosta*, the spiral is such as just described, in *Sagittaria* it consists of such a thick band that it reminds us of the spiral in the heterotype division of *Tradescantia* (compare Fig. 12 with Fig. 14).

In telophase, the chromosomes are contracted further and the spiral is still clearly observable, especially in *Allium*, *Zebrina*, *Tradescantua* and *Lilium*. In *Najas*, the zig-zag feature of the chromosomes is recognized also in the nuclei in the interphase before the first division in pollen grains.

Pollen grain

In the first division in pollen grains, the spiral aspect can also be observed satisfactorily in *Najas*, *Phajus* and *Tradescantia*. Each slender chromosome in meta- and anaphase presents a fine coiling appearance such as we have seen in the corresponding stages in the homotype division.

SUMMARY

In many higher plants, the spiral structure of chromosomes was demonstrated with the acetocarmine method of staining.

In those plants that carry chromosomes of relatively large size, it was traced in various stages during the meiotic divisions in pollen mother cells.

A certain unusual feature of chromosomes in the homotype division

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was found in Sagittaria, which is left for future investigation.

In conclusion, I wish to express my sincere gratitude to Prof. Y. KUWADA for his kind guidance throughout the investigation and also to Prof. N. TAKAMINE, of the Eighth High School, who kindly placed some of his materials at my disposal.

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EXPLANATION OF PLATE XVII

All the figures are microphotographs taken from acetocarmine preparations with ZEISS' apochr. imm. 2mm. and comp. oc. 15 except Fig. 3 which was taken with a comp. oc. 18.

Fig. 1 and 2; *Lilium tigrinum*. Chromosomes in heterotype metaphase. Continuous thread nature of the spiral is seen in places.

Fig. 3: Zebrina pendula. Heterotype metaphase. 12 bivalent chromosomes of various shapes are shown.

Fig. 4; Narcissus boeticus (?). A bivalent chromosome in the early stage of segregation in heterotype metaphase.

Fig. 5; *Tradescania virginica*. A chromosome set io meta-or early anaphase of heterotype division. Some of the chromosomes appear as if they consisted of numerous discs, while the others present distinctly a spiral character in places. The arrow indicates the

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doubleness of the spiral.

Fig. 6; Lilium longiflorum. Chromosomes at the end of heterotype anaphase.

Fig. 7; *Hosta Sieboldiana*. Chromosome groups in heterotype telophase, showing the so-called "chromosome bridge" which presents clearly the spiral aspect. In the drawn out portion of the spiral thread, two chromatic bodies are seen.

Fig. 8; Ditto. An oblique view of a daughter chromosome group in heterotype telophase.

Fig. 9. Ditto. A daughter nucleus in late telophase or early interkinesis. The spiral aspect is somewhat obscure, though there are still recognizable some indications of coiling nature in places.

Fig. 10; Zebrina pendula. A nucleus in interkinesis. Fine spiral threads are seen in the slender chromosomes.

Fig. 11; *Lilium tigrinum*. Chromosomes in late interkinesis or early prophase of the homotype division, showing the spiral structure.

Eig. 12: Tradescantia virginica. Homotype anaphase in side view. All the chromosomes show their fine spiral structure.

Fig. 13; *Lilium tigrinum*. Chromosomes in homotype metaphase, dislocated from the original positions by a slight pressure on the cover glass. The spirals are drawn out in places by the pressure.

Fig. 14: Sagittaria Aginashi. A chromosome in the same stage. The pitches of the spiral are so short that the chromosome seems as if it consisted of some number of discontinuous discs.

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