Notes on the Histology of a Giant and an Ordinary Form of *Plantago*

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With 2 Text-Figures

Various cases of chromosome features in relation to a stature of plants have been studied by several authors. The gigantism among some forms of the same species or in the same genus was found in several cases to be closely related with the polyploidy of chromosomes, while in other cases it was shown that no such relation existed. *Plantago japonica*, a littoral species growing wild in Japan, has its inflorescence axis measuring sometimes more than 100 cm. long and is conspicuously larger in other parts of the body than *P. major*, var. *asiatica*. Prof. FUJII, to whom specimens of the former were handed over by Prof. NAGAOKA of the Physical Institute of our Imperial University, who collected them at the coast of Misaki, the Miura Peninsula, suggested to me to examine whether this giant species is a polyploid in comparison with the type species *Plantago major* or its allies. A cytological study has shown that this giant form, contrary to the expectation, has only half the number of chromosomes found in the above named other species *P. major*, var. *asiatica*. This notable fact led me to make a closer comparison between the two forms with regard to their histology.

Previous to the description of the aimed observation, some measurements on the three closely allied species will be given in the following table :

Table I

Plant Name	Spike length cm.	Leaf-blade		Petiole-length cm.	Number of veins in a leaf
		Length cm.	Width cm.		
<i>P. major</i>	31.5	12*	9.4*	13	7 Large veins** 2 Small ,,
<i>P. major</i> , <i>var. asiatica</i>	49.0	12.5	9	17.5	5 Large veins 2 Small ,,
<i>P. japonica</i>	93.0	21	13.5	21	7-9 Large veins† 1-2 Small ,,

Although a smaller number of specimens of each species were available, the magnitudes given above, which are average values of measurements on the herbaria of a few mature plants at my disposal, seem to represent the normal relations fairly well. From the above table it will be seen that *P. japonica* is a giant form. The seeds of *P. major* and of *P. japonica* show similar appearances, though they are smaller in size and higher in number than those of *P. major*, *var. asiatica*. *P. japonica* is edible and cultivated in certain districts in Japan, in which condition, according to Mr. MAKINO, the plants get to have a larger stature than in the wild. Thus there may be much larger individuals than those on which above measurements were based. In young petioles of *P. major*, *var. asiatica* and of *P. japonica*, I counted 5-6 larger bundles and about 5 smaller bundles in the former, and about 7 larger and 2-7 smaller in the latter. These numbers of larger bundles approximately correspond to those of large veins in the leaf of matured plants (see the table I).

When the investigation was begun, the mature plants of *P. japonica* were difficult to be obtained, so I raised the young plants from seeds in the green house during September 1924 to March 1925. The young *P. major*, *var. asiatica* was collected last winter in the Botanical Garden of the Imperial University and also kept in

* According to TENOPYR's measurements, his specimen of *P. major* gave 176.09 mm. and 110.06 mm. for the length and width of leaves.¹⁵⁾

** DE CANDOLLE¹⁾ mentioned 3-5-7 veins.

† 7-9 veins were counted by FRANCHET and SAVATIER.⁴⁾

the green house. This too was not mature. The roots used for the measurements of a cell and nuclear size were taken from these of the two species. As the type species *P. major* is not known growing wild in Japan, its seeds were not at my disposal, so no fresh material could be used for the comparison.

The size of nuclei and cells of *P. major*, var. *asiatica* and of *P. japonica* was measured respectively in one hundred and fifty cells of their root-tips. The used roots of *P. japonica* were somewhat larger, on the average, than the other. Both materials were fixed in the FLEMMING'S Bonn-solution which was added with some urea, and treated carefully in the same manner. The cells of dermatogen were chosen for measurements in a transitionary region from division tissue to permanent tissue. The microtome sections were cut both transversely and longitudinally. The results are shown in the next table (Table II).

All cells and nuclei were drawn in the same scale with the aid of a camera lucida, the magnification being about 1100 times. The measurements made on the drawings are shown on the table II in terms of millimeters, so that the real magnitudes in mm. are to be obtained approximately by dividing the numbers given on the table by 1100.

Table II.

Plant Name	Number of nuclei and cells	Average size of nuclei		r=Average radius of nuclei	Average size of cells		Product of the length and width of cells
		Length	Width		Length	Width	
<i>P. major</i> , var. <i>asiatica</i>	50 nuclei and 50 cells in longitudinal sections	8.22	6.67	$r=3.72$ $r^3=51.48$	18.82	16.32	307.14
	99 nuclei and 100 cells in transverse sections	8.35	6.18	$r=3.63$ $r^3=47.84$	20.84	10.63	221.53
<i>P. japonica</i>	50 nuclei and 50 cells in longitudinal sections	7.02	5.28	$r=3.08$ $r^3=29.23$	19.72	15.73	310.2
	97 nuclei and 100 cells in transverse sections	6.98	5.18	$r=3.04$ $r^3=28.09$	22.96	10.66	244.75

Considering the data of the table II, we see that the average volume of nuclei in *P. major*, var. *asiatica* is much larger than in *P.*

japonica, while the products of the length and width of cells in both species are nearly the same. The volume ratio of nuclei in the two species is about 1.7:1. The shape of nuclei is nearly spherical, while the cells are irregular cubes. Moreover the three dimensions can not be measured in one section, consequently a highly reliable measure of the volume of each cell is difficult to be obtained. For getting some notion of cell volumes, however, I used the average width of cells in a longitudinal section as one dimension, viz. the height of cells in a transverse section. The volume ratio of cells thus obtained was 3440.19:3578.35, or 1:1.04. It will be noticed that the average volume of nuclei in *P. major*, var. *asiatica* is approximately twice as that in *P. japonica*, while the average cell volumes of both species are almost the same.

Then to get some idea of the chief cause of the giantness of *P. japonica*, further measurements on other somatic characters were made, the result of which is given in the table III.

Table III

Plant Name	Average size of 20 stomata of a leaf, magnified ca. $\times 1100$		Average number of stomata of a leaf, in a definite area (in a field of ZEISS K. 12 $\times$ DD)	Average number of upper epidermal cells in a definite area (in a field of ZEISS K. 12 $\times$ DD)	Average number of palisade cells in a definite linear distance (in a diameter of a field of ZEISS K. 12 $\times$ DD)
	Length cm.	Width cm.			
<i>P. major</i> var. <i>asiatica</i>	36.6	21.5	5.4	15	9.6
<i>P. japonica</i>	31.0	22.0	10.0	31	10.0

The measurements shown in the table III were made on the very individuals of *P. japonica* from which the material was taken before for the measurements of the dimensions of cells and nuclei. They have been cultivated outdoors for about two months since the end of March. They had still no spike, and their leaves were seemingly not

full grown; still some leaves though of a small size, were nearly in the mature state of development, as far as the development of their stomata showed. A number of mature individuals of *P. major*, *var. asiatica* were collected in the beginning of June in the Botanic Garden, and their leaves and spikes were used for the measurements given in the table III. For the comparison of the somatic characters, the leaves of both species in about the same developmental stage were selected, the tissues being chosen from the corresponding part of the organs.

In the table III we may see that the size of cells in *P. japonica* is generally smaller than that of the other species. According to JONES¹⁰⁾, though he gave no measurements, the leaves of *Digitalis purpurea* were larger and broader than those of *D. grandiflora*, while the cells and stomata of the latter were larger than those of the former. This seems to be a parallel case with that of *Plantago*. At any rate, as far as my measurements on the tissues used are

Average number of cells in a linear row of $\frac{1}{8}$ cm. length of a petiole		Average number of cells in a linear row of $\frac{1}{8}$ cm. length of a spike	
At the region above 3-3 $\frac{1}{2}$ cm. from the base	At the region above 10-12 cm. from the base	At the region above 10-15 cm. from the base	At the region above 20 cm. from the base in <i>P. major</i> , <i>var. asiatica</i> and 50 cm. in <i>P. japonica</i>
33.4	22.0	21.5	25.0
33.6 31.0*	32.4 34.0*	18.0*	28.0*

concerned, the giantness of *P. japonica* is to be ascribed to the high number of cells in the corresponding parts or organs of this plant.

The examination of the chromosome number and the nuclear size in both species shows that the one with larger nuclei has the double number of chromosomes when compared with the other with

smaller nuclei; viz. in *P. major*, *var. asiatica*, the haploid chromosome

* These measurements were obtained from one of the largest dried specimens of the plant.

number is 12^* and the diploid 24^* (Fig. 1), while the nucleus of *P. japonica* has 12 chromosomes in the root-tip cell (Fig. 2).

The shape and size of somatic chromosomes in both species are generally alike, though the chromosomes shown in Fig. 2 seem to be

Fig. 1

Fig. 2

somewhat smaller than those of Fig. 1. EKSTRAND⁹⁾ reported the chromosome numbers of five species of *Plantago*. According to him, *P. major* has the 6 haploid and 12 diploid chromosomes. However, owing to the lack of any description and figures about the size of somatic chromosomes of this species, I can not make any comparison between this and the two other species I have examined.

Of various instances hitherto reported on the relation between the chromosomes and the stature of plants, a provisional classification may be given: I) chromosome number same, size of chromosome as well as of nucleus alike, cell size smaller (*Hedera helix conglomerata*, a dwarf form: ** *H. helix typica*¹²⁾), II) chromosome number same, cell size alike¹⁵⁾ (*Plantago major*²⁾: *P. lanceolata*¹¹⁾), III) Chromosome number same, chromosome and cell size larger (*Primula sinensis*, var. *gigas*: *P. sinensis*⁶⁾; *Phragmites communis*, var. *Pseudodonax*, a giant form: *P. communis*¹⁶⁾), IV) chromosome number same, body size dwarf (*Oenothera nanella*: *O. Lamarckiana*, etc.), V) chromosome number larger, nuclear or cell size larger (gigas-, hero-races; *Oenothera gigas*: *O. Lamarckiana*⁵⁾; gigas-form in *Solanum*²⁰⁾; *Hedera helix hibernica*: *H. helix typica*¹²⁾; some forms in *Bryum*, *Mnium* and *Amblystegium****; varieties of *Athyrium Filix-foemina*³⁾; *Taraxacum albidum*: *T. platycarpum*¹³⁾; triploid forms in *Canna*¹⁸⁾ and *Morus*¹⁴⁾; multiploid forms in *Chrysanthemum*¹⁹⁾), VI) chromosome number larger (double); a) chromosome size alike, nuclear size larger (?) (*Erigeron unalaskensis*, a dwarf form: *E. eriocephalus*⁸⁾), b) chromosome size alike, nuclear

* These chromosome numbers I have determined agree with MIYAJI's determination.⁹⁾

** The symbol: used hereafter, whenever put in brackets, means "in comparison with."

*** MARCHAL, EL. and EM. 1906-11 (after TISCHLER¹⁷⁾)

size larger, cell size larger or alike (*Plantago major*, var. *asiatica**, a smaller form: *P. japonica*), VII) chromosome number larger, chromosome size smaller, body size smaller (*Rumex acetosella*** : *R. acetosa*; some species of *Carex*⁷⁾). There are also exceptions which are not included in this classification.

Although the results of my measurements in the present case of *Plantago* may not be conclusive, it may be with a certain confidence concluded that the size of somatic chromosomes in the two forms of *Plantago* is alike, and the one, a smaller form possessing double the number of chromosomes found in the larger form, has larger nuclei, and that the cell size in the larger form is generally smaller than that in the other, or in certain cases it is nearly alike in both forms, as far as the measurements on the tissues employed are concerned. Accordingly the giantness of *P. japonica* is due to the larger number of cells in the corresponding part or organ of this form. Thus the present case of *Plantago* forms a subclass of the class VI) of the above classification.

In concluding I desire to express my sincere thanks to Prof. K. FUJII for his valuable advice and criticisms during the course of this work.

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* *P. major*, var. *asiatica* has double the number of chromosomes of *P. major*, but the size relation of chromosomes as well as of nuclei and cells cannot be made out, as the data regarding the latter plant are wanting as mentioned before.

** ROTH, H. 1906 (after HOLMGREN⁸⁾)

BIBLIOGRAPHY

- 1 DE CANDOLLE, A. (1852) Prodr. **13, 1**
 - 2 EKSTRAND, H. (1918) Sv. bot. Tidskr. **12**
 - 3 FARMER, J. B. and DIGBY, L. (1907) Ann. of Bot. **21**
 - 4 FRANCHET, A. et SAVATIER, L. (1879) Enum. Pl. Jap. **2**
 - 5 GATES, R. R. (1909) Arch. f. Zellf. **3**
 - 6 GREGORY, R. P. (1909) Proc. Cambridge Phil. Soc. **15**
 - 7 HEILBORN, O. (1924) Hereditas **5**
 - 8 HOLMGREN, J. (1919) K. Sv. Vet. Akad. Handl. **59**
 - 9 ISHIKAWA, M. (1916) Bot. Mag. Tokyo, **30**
 - 10 JONES, W. N. (1912) Journ. Genet. **2**
 - 11 NĚMEC, B. (1910) Das Problem d. Befruchtungsvorgänge usw.
 - 12 OEHM, G. (1924) Beih. z. bot. Centralbl. **40**
 - 13 OSAWA, I. (1913) Arch. f. Zellf. **10**
 - 14 OSAWA, I. (1916) Bull. Imp. Sericult. Exp. Stat. Japan, **1**
 - 15 TENOPYR, L. A. (1918) Bull. Tor. Bot. Cl. **45**
 - 16 TISCHLER, G. (1918) Ber. d. d. bot. Ges. **36**
 - 17 TISCHLER, G. (1920-1) Allgemeine Pflanzenkaryologie
 - 18 TOKUGAWA, Y. and KUWADA, Y. (1924) Jap. Journ. Bot. **2**
 - 19 TAHARA, M. (1921) Journ. Coll. Sci. Imp. Univ. Tokyo, **43**
 - 20 WINKLER, H. (1916) Zeitschr. f. Bot. **8**
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