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Cytogenetic Studies of Some Tetraploids in Allium

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Allium屬內 數種의 四倍體에 對한 細胞遺傳學的 硏究

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ABSTRACT

The present paper was carried out to clear up the polyploidal constitution and the banding patterns of three species in tetraploidal *Allium*(X=8) through the studies of meiosis, karyotype and G-bands. From the meiotic data and G-banding patterns obtained under this study, it is confirmed that *A. tuberosum* and *A. chinense* are autotetraploids, and *A. senescens* is allotetraploid. Some aneuploids out of the employed species were found; *A. senescens* is 2n=34, and *A. chinense* is 2n=33. The chromosome types of these species are meta- and submetacentrics except the sat-chromosomes and the f-chromosomes.

G-bands of these species are generally located in the end of each arm. *A. senescens* is similar in the quantity of heterochromatin with *A. chinense*, but *A. tuberosum* has a little than the other species. The quantity of heterochromatin is higher in small groups of chromosome than large ones, and higher in short arms than long arms.

INTRODUCTION

All the species of the genus *Allium* comprise a distinct odor in bulb and have many common characters. In chromosome constitution, these are three basic number 7, 8 and 9 with ploployidy up to 13x(Ownbey and Asse, 1955; Asse, 1959), and include the chromosome complement composed of metacentric and submetacentric elements. Recently cytotaxonomic and cytogenetic studies in a related species were by far improved by the banding technique which could be identify with greater precision each chromosome pair of the species complement(Schnedl, 1972; Yosida and Sakai, 1973; Marks and Schweizer, 1973; Vosa, 1973).

Ved Brat(1965) reported that *A. tuberosum*, *A. senescens* and *A. chinense* have the same flowering season, and the chromosomes of these three species have basic number of 8.

The present paper is a cytogenetic studies through meiosis, karyotype and G-banding patterns using Giemsa staining method; especially the research of polyploidy by means of banding pattern is concentrated upon the subject.

MATERIALS AND METHODS

A. tuberosum in materials used in this study was obtained from a local farm in the suburbs of Daegu city, *A. senescens* was from mountaneous land of Seongju and *A. chinense* was from Mt. Chili. Each species was replanted in a flowerpot in order to investigate the further studies including the crossing between species and varieties. For meiotic study, the pollen mother cells were squashed in

aceto-orcein being fixed in 1 : 3 acetic-alcohol to which a little ferricchloride was added.

The root-tips pretreated in aquous 0.05% colchicine at 10°C for 6 hours were fixed in 1 : 3 acetic-alcohol for 30 minutes. The materials were hydrolyzed in 1 N HCl at 60°C for 20 seconds and left overnight in room temperature after squash. Coverglass was artificially prized off with pin from slide instead of dry ice method.

For the production of chromosomal band, the preparations were immersed in 8% barium hydroxide at room temperature for 30 minutes and rinsed in distilled water, and then incubated in 4x SSC at 60°C for 30minutes. After the preparations were stained with Giemsa for 1 hour, rinsed and air dried. Giemsa stain and photomicrograph were made by the technique used for routine chromosome analysis in our laboratory (Seo and Kim, 1975). The size of H—segment was estimated with micrometer in order to draw in greater precision the diagrammatic representation of banding patterns.

RESULTS AND DISCUSSION

1. *A. tuberosum*

So far as has been shown, *A. tuberosum* shows polyploid having 8 as the basic number (Katayama, 1936). In meiotic division different configurations ranging from 8 tetravalents to 32 univalents were observed(see in Fig. 1); not so much as to octavalent clumps. Lagging chromosomes and fragments were observed in anaphase I as well. It is assumed that abnormal figures are due to the irregular division that occurred at meicsis. As shown in Table 1, complete 8 tetravalents were observed in 83(44%) out of 290 cells examined.

Fig. 3 shows the mitotic metaphase plates of *A. tuberosum*. Satellite is apparently characterized by heterochromatic region revealed by means of Giemsa stain(b of Fig. 2). The quantity of heterochromatin is generally little in comparision with the

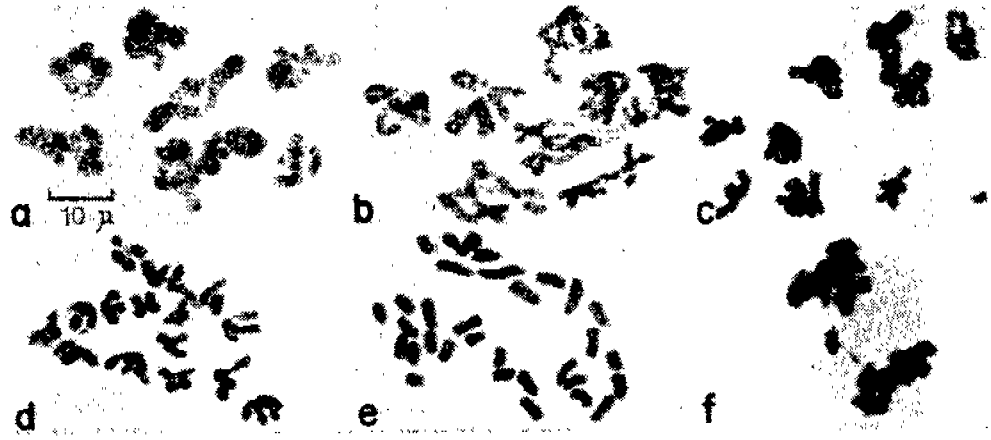

Fig. 1. Chromosome configuration at metaphase and anaphase in *Allium tuberosum*. a; 8_{IV}, b; $7_{IV}+2_{II}$, c; $6_{IV}+3_{II}+2_I$, d; 16_{II}, e; 32_I, f; lagging chromosome.

Table 1. Association of chromosomes at the first metaphase in *A. tuberosum*

Configuration	Figures having various types						
	8_{IV}	$7_{IV}+2_{II}$	$6_{IV}+4_{II}$	$5_{IV}+6_{II}$	16_{II}	32_I	Rest
No. of figures	83	29	18	12	16	6	26
Percentage	44	15	9	6	8	4	14

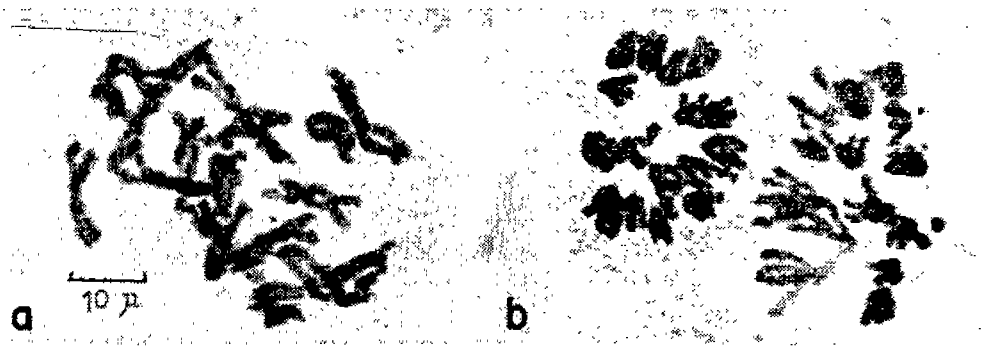

Fig. 2. Chromosome configuration at metaphase and anaphase in *A. senescens*. a; 16, b; 1st anaphase.

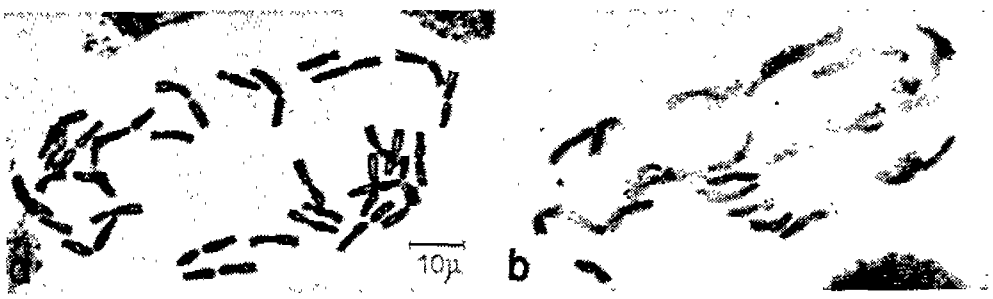

Fig. 3. Photomicrographs of general(a) and G-banding(b) metaphase in *A. tuberosum*.

other two species. Chromosome constitution of *A. tuberosum* consists of metacentric except the sat-chromosome(I in Fig. 6). It is confirmed that this species is autotetraploid owing to the behaviours of chromosome pairing in meiosis and the distribution of Giemsa band.

All ten plants under study are generally unique in the distribution of their band. In chromosome G and H the bands appeared in the end of both arms. A,B,E and F chromosomes have a only thin band in the end of short arm, while C chromosome has in the end of long arm. D chromosome has minor bands in the end of short arm and near the submetacentric region of long arm.

2. *A. senescens*

Ved Brat(1965) reported that this species is tetraploid having basic number of 8, and the first meta phase has 16 bivalents in PMC. The author obtained such the results as Ved Brat during the observation of meiotic division(Fig. 2). Fig. 4 shows the mitotic metaphase plates of *A. senescens*. All 50 plants were employed. Among them 47 plants were 2n=32, and 3 were aneuploids, 2n=34, which a pair of small subtelocentric is added. Fig. 5 shows the idiogram and diagram of this species, and these chromosomes consist of meta- and submetacentric except the sat-chromosome and f-chromosome. It is clearly supposed that this species is allotetraploid from banding distribution and meiotic observation. The quantity of heterochromatin is higher in small chromosome groups than large ones, and in short arms than long arms. Small chromosome (f) hasn't any band. The distribution of G-band in this species are roughly as follows. In C and D chromosomes no band appeared. A, B, D, E, F and M chromosomes have only one band either in the end of short arm or long arm. In the rest chromosomes the bands are located in the end of both arms, and also additionally in intercalary region. Especially K chromosome is characterized with three large bands in the short arm.

3. *A. chinense*

Chromosome number of this species was reported as 2n=16(Darlington, 1956), but as shown in Fig.5, this species is observed as 2n=32 mostly. 2 out of 45 plants under study were found 2n=33. Among

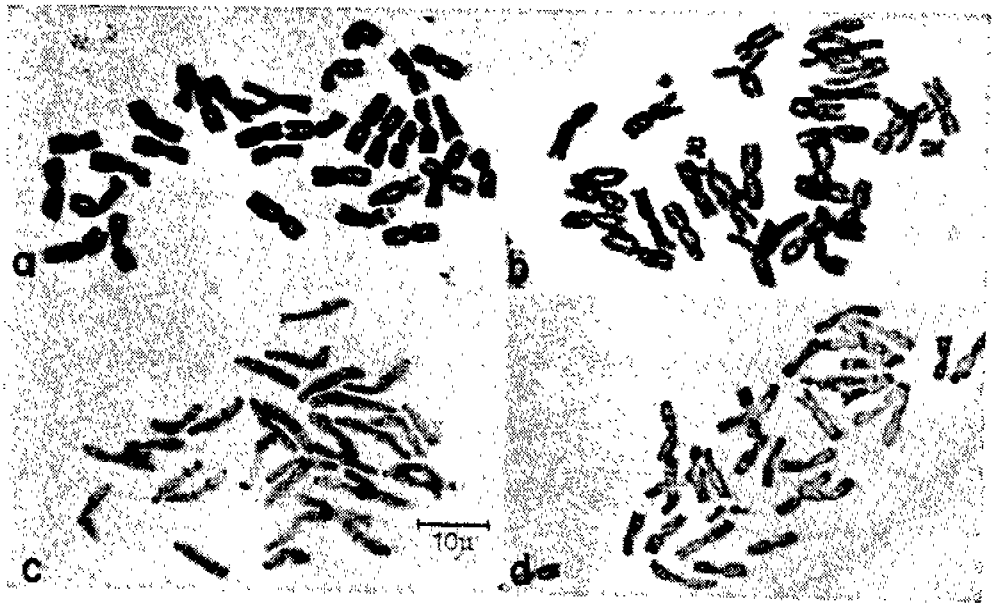

Fig. 4. Photomicrographs of general (a and b) and G-banding (c and d) mataphase plates in *A. senescens*. a and c; 2n=32, b and d; 2n=34. x1000.

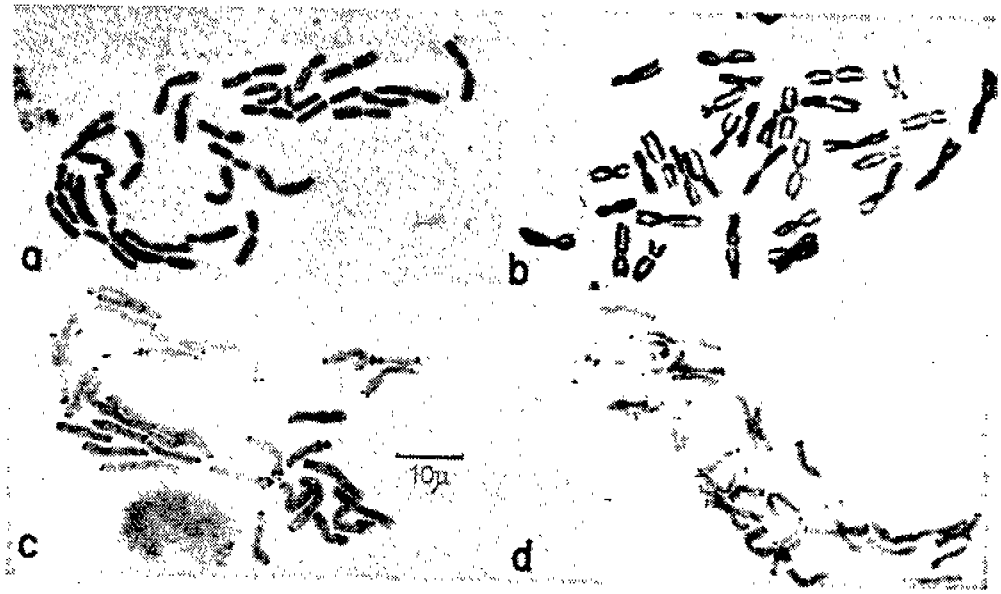

Fig. 5. Photomicrographs of general (a and b)and banding (c and d) metaphase plates in *A. chinense*. a and c; 2n=32, b and d; 2n=33.

4 sat-chromosomes, one small sat-chromosome was observed sometimes. From banding point of view, A and D chromosomes have a band in the end of short arm while B chromosome hasn't any band. In chromosome C the bands appear in the end of both arms. E, G and H chromosomes have bands in the end of both arms and sometimes the another band near the subtelocentric region of the short arm. In sat-chromosome the minor band appears in the end of long arm, major band in the end of short arm, and satellite stained deeply. From the standpoint of banding pattern, it is confirmed that

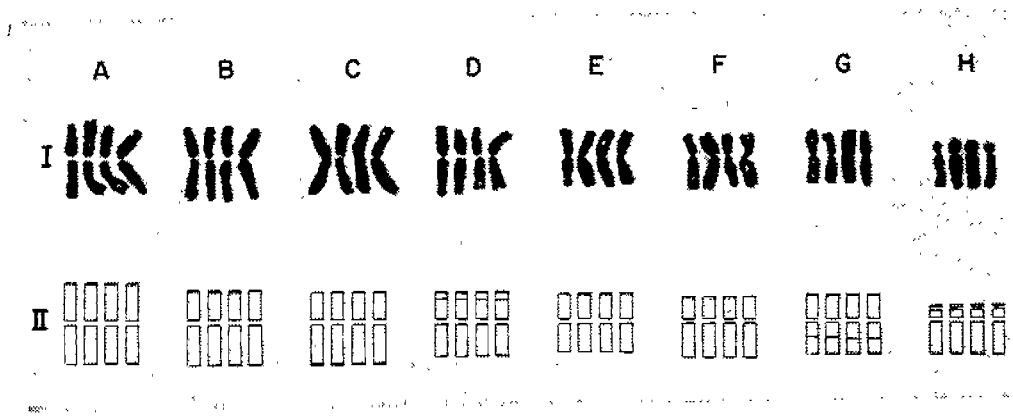

Fig. 6. Idiogram(I) and diagrammatic representation of G-band(II) in *A. tuberosum*.

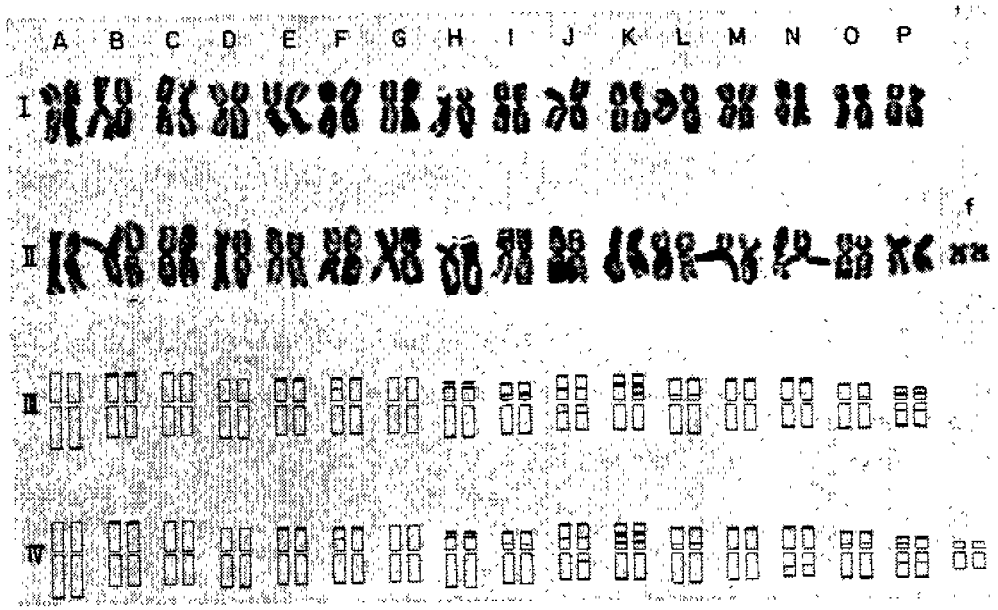

Fig. 7. Idiogram(I and II) and diagrammatic representations of G-banding (III and IV) in *A. senescens*. I and III; 2n=32, II and IV; 2n =34.

this species is autotetraploid.

The evidence of the present study shows that the homologous chromosomes within and between each individuals and population may be represent polymorphic from standpoint of banding patterns, and the size and number of bands among these species was somewhat differed each other.

The quantity of heterochromatin revealed on the chromosome of *A. tuberosum* is by far little than the other two species, but banding pattern and its quantity of *A. senescens* are more or less similar with those of *A. chinense*.

The treatment forming band was reported so far the various methods by many authors. It was found that the treating time and concentration of barium hydroxide are the most important process during the banding treatment of these three species. For the sake of detailed minor band, the degree of stain is the key point: In deeply stained preparations it was observed nothing but the major band, but light blue colour could be well observed both the major and minor band.

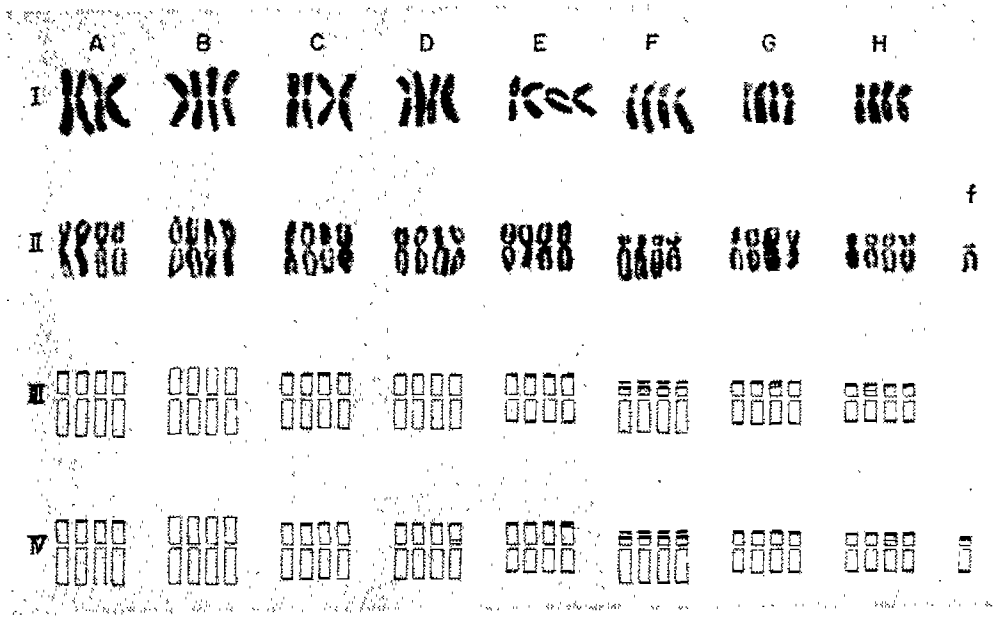

Fig. 8. Idiogram (I and II) and diagrammatic representations of G-banding patterns (III and IV) in *A. chinense*. I and III; 2n=32, II and IV; 2n=33.

摘 要

Allium屬 中 부추(*A. tuberosum*), 두메부추(*A. senescens*), 산부추(*A. chinense*)의 3種에 對하여 減數分裂狀況과 Giemsa band法을 通하여 核型을 調査한 바 그 結果는 다음과 같다.

1. 부추 및 산부추는 同質四倍體이고 두메부추는 異質四倍體인 것으로 밝혀졌다.

2. 本實驗에서 異數體도 發見되었는데 두메부추에 있어서 50個體中 3個體가 2n=34, 산부추에 있어서는 45個體中 2個體가 2n=33이었다.

3. 3種을 通하여 band의 所在는 染色體兩腕의 末端에 主로 位置하고 있으며 異質染色質의 量은 短腕이 長腕보다, 또 적은 染色體가 큰 染色體보다 一般的으로 많다. 種別로 보면 부추가 그 量이 가장 적고 他種은 서로 비슷한데 부추보다는 많다.

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