

CHROMOSOME STUDY ON *CROCUS CANCELLATUS* SUBSP. *DAMASCENUS* FROM IRAN

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Meiosis behaviour in *Crocus cancellatus* subsp. *damascenus* (cytotype 8A) is reported for the first time. The results indicated that this subspecies (cytotype 8A) has chromosome complement of $2n=8$ and gametic number of $n=4$ chromosomes. Two bivalents were associated with nucleolus, that confirmed the presence of two pairs of satellite chromosomes in this taxon. One B-chromosome is reported here for the first time in both meiotic and mitotic stages in this cytotype.

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Key words. *Crocus cancellatus* subsp. *damascenus*, chromosome, B-chromosome, cytotype, Iran.

مطالعه کروموزومی روی زیر گونه *Crocus cancellatus* subsp. *damascenus* از ایران

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رفتار میوزی برای اولین بار در زیر گونه *Crocus cancellatus* subsp. *damascenus* (سیتوتیپ $2n=8A$) گزارش می‌شود. نتایج نشان می‌دهد که این زیر گونه (سیتوتیپ $2n=8A$) دارای مجموعه کروموزومی $2n=8$ و عدد گامتی $n=4$ می‌باشد. دو جفت کروموزوم به هستک متصل بود که تایید کننده حضور دو جفت کروموزوم قمردار در این زیر گونه می‌باشد. حضور یک عدد B کروموزوم در هر دو مرحله میوز و میتوز در این سیتوتیپ برای اولین بار گزارش می‌شود.

INTRODUCTION

Crocus cancellatus Herbert is a very widespread plant with a distribution from Yugoslavia to Iran (Mathew 1982). This species has a lot of variation in the general morphology and cytology and, in this case, ecology (Brighton 1977; Feinbrun & Shmida 1977; Mathew 1982; Heywood 1983). The *C. cancellatus* aggregate comprises of 5 subspecies (Mathew 1982), including: subsp. *cancellatus*, subsp. *mazziaricus* (Herbert) Mathew, subsp. *lycius* Mathew, subsp. *damascenus* (Herbert) Mathew and subsp. *pamphylicus* Mathew. Some of the subspecies are variable cytologically (Brighton 1977; Feinbrun & Shmida 1977). Fourteen different cytotypes occur in the *C. cancellatus* aggregate ranging from $2n=8$ to 18. *C. cancellatus* subsp. *damascenus* was first described by W. Herbert from hills near Damascus but other names have been bestowed upon it over the years including *C. edulis*, which refer to its use as a food. From the aggregate of *C. cancellatus* only subspecies *damascenus* is found in Iran. This subspecies distributed in W and SW of Iran.

Previous chromosome counts for this subspecies are $2n=8$ from Damaneh, $2n=10$ from Shahpur and Rezaieyeh (Brighton 1977), $2n=8$ from Golpayegan (Sanei et al. 2006), and $2n=8$ from Arak (Safid Khani) (Ebrahimzadeh et al. 1998). In this study meiosis behaviour of this cytotype ($2n=8A$) is reported for the first time.

MATERIAL AND METHODS

The corms of plants were collected from the southern areas of Arak: Sefid-Khani (Markazi province). Meiotic chromosomes were observed in pollen mother cells. Floral buds of appropriate size taken from the underground buds, and then fixed immediately in Piennr's fluid containing ethanol 96%, chloroform, propionic acid, 6:3:2 (v/v/v) for 24 hours. Anthers dissected out from the buds were squashed and stained with 2% acetocarmine. Permanent slides were made by the vanetian turpentine (Wilson 1945). Photographs of chromosomes were taken by Olympus photomicroscope at initial magnification of 330 x.

Voucher specimens were deposited in the Central Herbarium of Tehran University (TUH).

RESULTS AND DISCUSSION

Different cytotypes were designated by Brighton (1977) for *C. cancellatus* subsp. *damascenus* as follows: 1- Karyotype with four pairs acrocentric chromosomes ($2n=8A$). 2- Karyotype with one or more pairs of metacentric chromosomes ($2n=8M$). 3- karyotype with five pairs of chromosomes ($2n=10A$, $10M$, $10H$ see Brighton 1976). According to Mathew (1982), flora of Iran has only one subspecies of aggregate of *C. cancellatus* (*C. cancellatus* subsp. *damascenus*). Previous chromosome counts for this subspecies are $2n=10$ from Oromiyeh (Rezaiyeh) and Salmas (Shahpur) with highly heteromorphic karyotypes ($10H$) and $2n=8$ with four acrocentric chromosomes ($8A$) (Brighton 1977). Meiosis has been examined only in the *C. cancellatus* subsp. *damascenus* from Lebanon ($2n=8M$) and Turkey ($2n=10A$) by Hywood (1983), but up to the present not in $2n=8A$ cytotype. Our sample showed chromosome complement of $2n=8$ with four pairs of acrocentric chromosomes in parashima cells around the anthers (Fig.1.), which agree with the previous report by Ebrahimzadeh et al. (1998) and Sanei et al. (2006). Also, four bivalents at first metaphase and diakinesis were observed (Figs. 2, 3). Chiasma frequency in 20 cells gave a mean of 1.90 per bivalent. Chiasmata were terminal and interstitial location at metaphase I (Fig. 4). Chromosome segregation at anaphase one was (4-4) (Fig. 5). Ebrahimzadeh et al. (1998) reported that *C. cancellatus* subsp. *damascenus* has $2n=8A$ with one pairs of satellite chromosome, whereas, Sanei et al (2006) showed two pairs of satellite chromosomes in this taxon. Our results indicated that the later report is correct. Because, we found two bivalents of chromosomes which were associated with nucleolus (Fig. 3), that confirm the presence of two satellite chromosomes in this taxon. Occasionally in some cells, one B-chromosome in both division of mitotic and meiotic were observed (Fig. 6, 7). This B-chromosome showed a tendency to lag at first anaphase (Fig. 8). Brighton (1977) believed that the Robertsonian translocation have an important role in the cytological evolution in the *C. cancellatus*. Cytogenetic study on our sample not showed occurrence of interchanges and inversion phenomenon in the trend of microspogenesis. In a few cells tetravalents at metaphase were observed (Fig.9), but was not clear to occurrence of translocation at prophase stage. The results of this paper and previous meiotic and mitotic

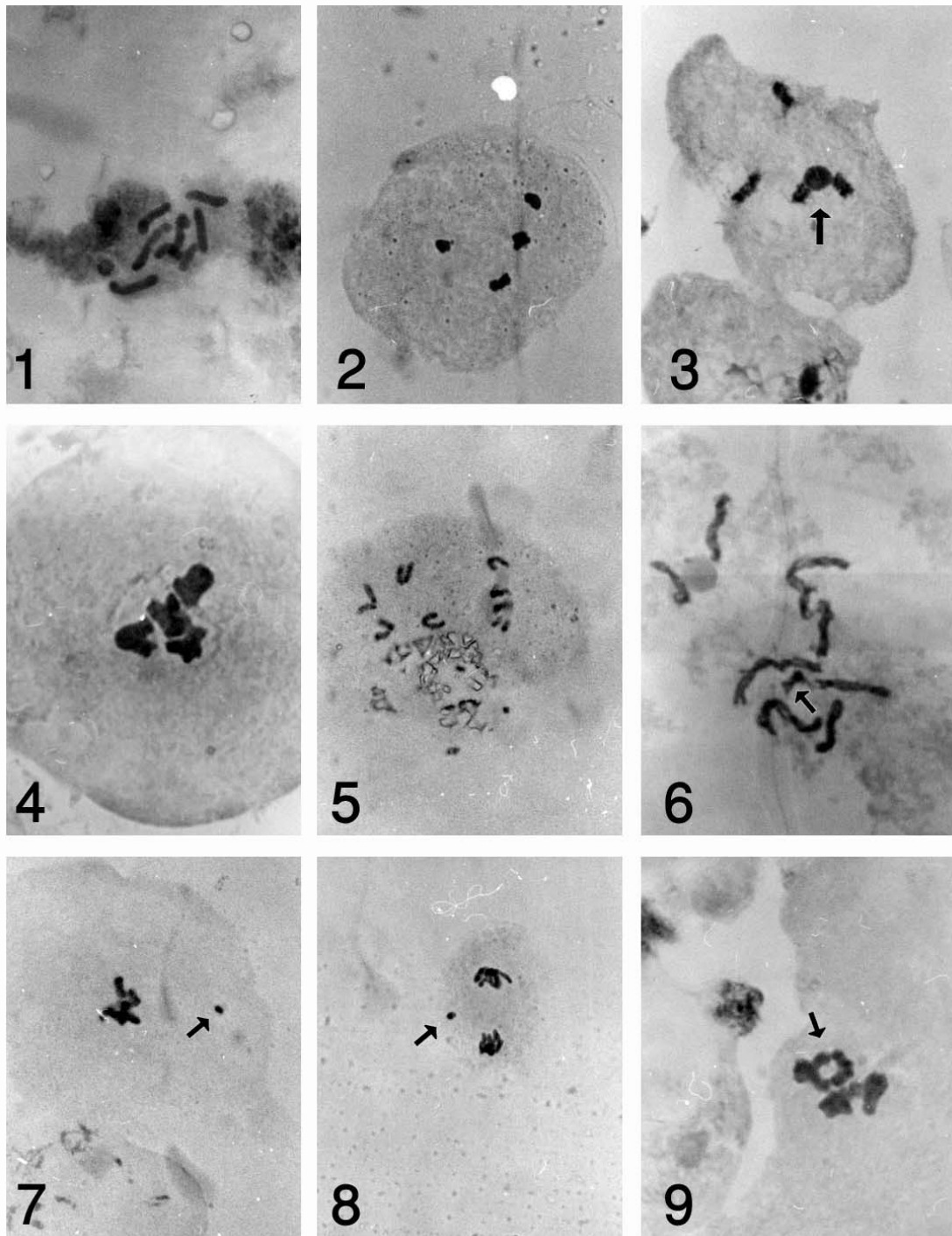
studies (Feinbrun 1957, Sopova 1972, Lovka et al. 1971, Brighton 1976, Brighton 1977, Heywood 1983, Ebrahimzadeh et al. 1998, Sanei et al. 2006) show that the morphological and cytological variation in different populations of *C. cancellatus* need to be critically reconsidered. There are cytological variations including length and types of chromosomes (Brighton 1976), which can not be interpreted by any possible translocation. The occurrence of cytotypes such as $10A$, $8A$, $10M$, $10H$ suggest possible occurrence of cytologically different species in *C. cancellatus* s. l. Cytologically it is expected that $8A$ compliments resulted from segregation of $4M$, or the cytotype $10M$ would give a $20A$ compliment. Such cytotypes have not yet been found in *C. cancellatus*.

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Figs.1-9. Meiosis and mitosis. Fig.1- Somatic metaphase, $2n=8$. Fig.2- First metaphase, showing four bivalents. Fig.3- Diakinesis, showing two attached bivalents with nucleolus (arrow). Fig.4- Metaphase I, showing four bivalents with terminal and interstitial chiasmata. Fig.5- Anaphase I, showing (4-4) segregation. Fig.6- Somatic metaphase chromosomes ($2n=8 + 1B$), B-chromosome indicated by arrow. Fig.7- Metaphase I, showing one B-chromosome (arrow). Fig.8- Anaphase I, showing laggard B-chromosome (arrow). Fig.9- Metaphase I, tetraivalent indicated by arrow. Initial magnification X340.