

Cytomixis in Lamiaceae

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The record of cytomixis in plants dates back to the work of Gates (1911) in *Oenothera gigas*. Cytomixis has been used as a term for the phenomenon of chromatin migration from one microsporocyte to an adjacent one. It has also been reported to occur in meristematic tissues (Jacob 1941) and in the interface between somatic and meiotic cells (Cooper 1952). The artificial induction of this process has also been achieved by the application of Trifluralin (Bobak and Herich 1978). Kamra (1960) reported that it could be observed in normal diploid species, hybrids and apomictics. However, Salesses (1970) reported the occurrence of cytomixis only in natural and artificial triploid hybrids of *Prunus* but not in the diploid, tetraploid and hexaploids.

Despite the fact that cytomixis has been widely reported in several families of flowering plants, very little is known about the cause and evolutionary significance of this process. Most workers, however, accepted it to be a normal, though infrequent, cytological phenomenon and not an artifact of preparation as advocated by certain workers.

Heslop-Harrison (1966) demonstrated that cytoplasmic channels connecting microsporocytes through pores in the callose normally allow the exchange of small cell organelles but not nuclear material. He suggested that nuclear material transfer is induced *in vitro* and does not occur *in vivo*. Sarvella (1958) and Omara (1976), however, regarded the process as representing real migration of chromatin material during meiosis. This process has been considered as a possible cause of the production of aneuploid and polyploid gametes (Sarvella 1958, Bell 1964, Omara 1976).

In the present investigation cytomictic connections have been recorded in species of *Leonurus*, *Leucas* and *Mentha*. The present study has been conducted to elucidate the cause and significance of this process.

Materials and methods

The present paper deals with meiotic studies of different species and populations of three genera (viz. *Mentha* L., *Leucas* R. Br. and *Leonurus* L.) belonging to the family Lamiaceae.

Meiotic studies were made in fresh as well as fixed materials. Flower buds were fixed for 24 hrs. at room temperature in acetic ethanol mixture (1:3). Fuchsin staining and 2% acetocarmine staining were used for meiosis and pollen grain sterility studies.

To determine seed set by open pollination in *Mentha piperita*, two to four inflorescences of each plant were bagged after the flower had been pollinated and before the seeds matured. Seed germination tests were carried out in petri dishes in distilled water at 20°C.

Observations

Cytomixis, showing different stages in chromatin migration, has been observed in dif-

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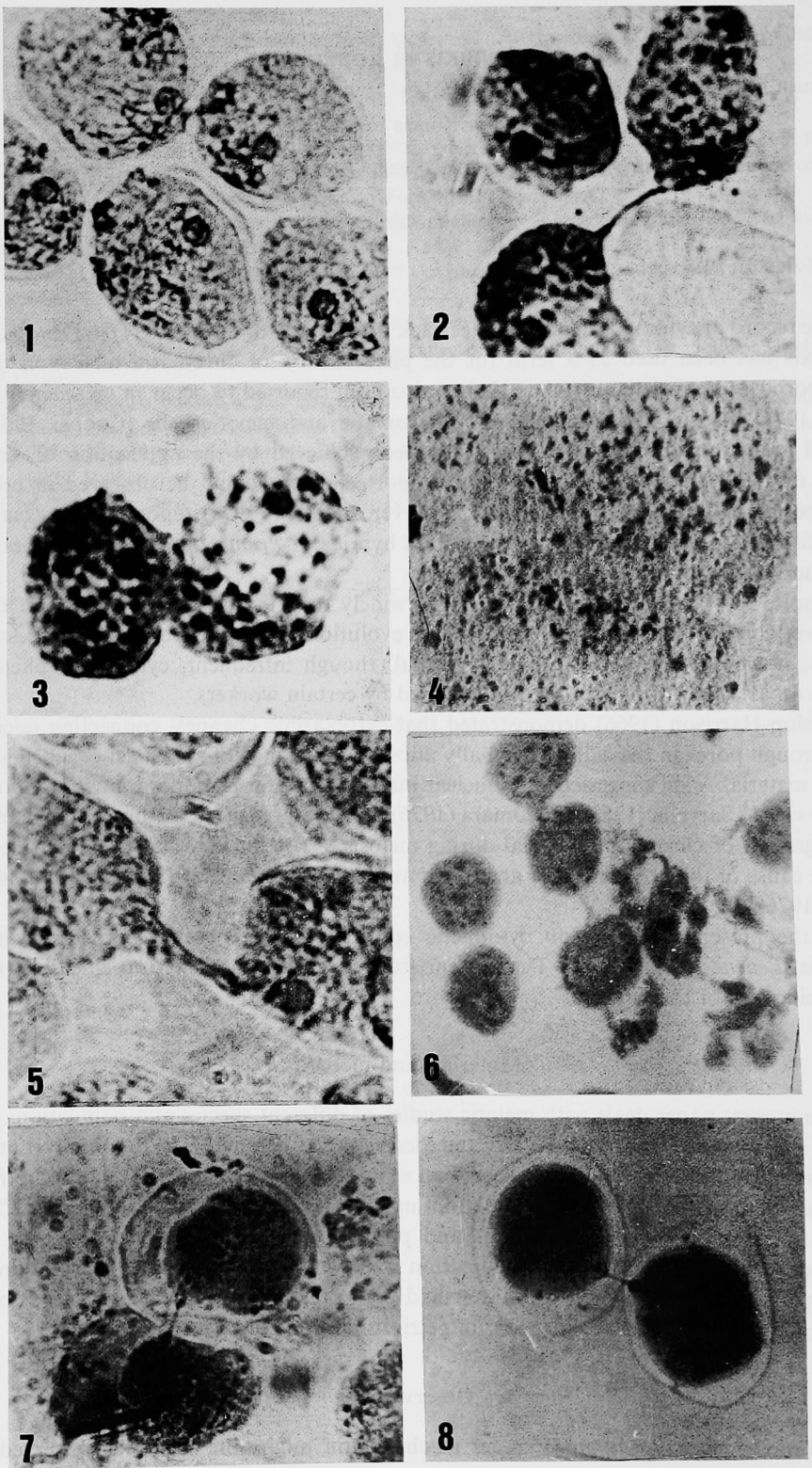


Table 1. Cytomixis in the three genera of Lamiaceae

Name of taxa	Occurrence	Nature	Meiotic abnormalities
<i>Mentha piperita</i> Linn. Cytotype I	Cytomixis has been observed in almost every stage, starting from prophase I (Fig. 1) to metaphase II (Fig. 4). It occurs more commonly in first meiotic prophase and metaphase. Sometimes adjacent cytomictic microsporocytes have been observed to represent different meiotic stages (Fig. 4).	It occurs both as cytoplasmic bridges (Fig. 2) and as direct cytoplasmic contact (Fig. 3).	Anaphase I abnormalities includes early separation and late separation of bivalents. Micronuclei, ranging between 1 to 14, are also present in almost all of the dyads. Unequal distribution of chromosomes at telophase II is also observed.
<i>Mentha arvensis</i> L. var. <i>javanica</i> (Bl.) HK. f.	Cytomixis has been observed only in first meiotic prophase.	Occurs only as cytoplasmic bridges (Fig. 5).	Microsporocytes have been shown not to undergo any division beyond prophase I, which is followed by disintegration.
<i>Leonurus sibiricus</i> L. Populn. V.	Cytomixis has been observed only in second meiotic metaphase (Fig. 6).	Occurs only as cytoplasmic bridges (Figs. 6, 7).	No meiotic abnormalities have been recorded.
<i>Leonurus sibiricus</i> L. Populn. VII	Cytomixis has been observed only in second meiotic telophase (Fig. 7).	Occurs only as direct cytoplasmic contact.	No meiotic irregularities have been recorded.
<i>Leucas aspera</i> Spreng.	Cytomictic connections have been noted only in second meiotic telophase (Fig. 8)	Occurs only as cytoplasmic bridges.	No meiotic abnormalities have been noted.

Table 2. Pollen sterility in the three genera of Lamiaceae

Name of taxa	Population	Pollen sterility (%)
<i>Leonurus sibiricus</i> L.	I	17.64
" "	II	14.94
" "	III	26.20
" "	IV	10.86
" "	V	30.52
" "	VI	16.27
" "	VII	26.58
" "	VIII	14.94
" "	IX	12.35
<i>Leucas aspera</i> Spreng.	—	10.57
<i>Mentha piperita</i> L. Cytotype I	—	51.77
<i>Mentha arvensis</i> L. var. <i>javanica</i> (Bl.) HK. f.	—	No pollen grains are formed.

Figs. 1–8. 1–4: *Mentha piperita*. Cytotype I—PMCs showing cytomixis at different stages of meiosis. 1, cytomixis involving microsporocytes at prophase I stage. $\times 1500$ approx. 2, cytomictic connections in the form of a bridge involving microsporocytes at metaphase I stage. $\times 1500$ approx. 3, PMCs showing direct fusion during first meiotic metaphase. $\times 1500$ approx. 4, PMCs showing direct fusion during second meiotic division. One microsporocyte is at prophase II and another at metaphase II. $\times 1500$ approx. 5, *Mentha arvensis* L. var. *javanica*. Cytomictic connection in the form of a bridge involving microsporocytes at first meiotic prophase. $\times 1500$ approx. 6, *Leonurus sibiricus* Population V. Cytomictic connection in a series of PMCs. $\times 800$ approx. 7, *Leonurus sibiricus* Population VII. Cytomictic connection involving microsporocytes at telophase II. $\times 1500$ approx. 8, *Leucas aspera*. Cytomictic connection involving microsporocytes at telophase II. $\times 1500$ approx.

ferent meiotic stages. It has been observed in both fixed and fresh flower buds. In some taxa rows of cells have been shown to be connected by cytomictic connections (Fig. 6). Cytomixis is particularly common in first meiotic division although it has been observed as late as second meiotic telophase (Figs. 7, 8). Cytomictic connections between microsporocytes have been noticed both as cytoplasmic bridges (Figs. 2, 5) and as direct cytoplasmic contact between adjacent cells with their common walls dissolved (Figs. 3, 4).

The details concerning the cytomictic connections in the different taxa have been presented in Table 1. Pollen sterility on the basis of stainability reaction with aceto-carmin was recorded for all the investigated taxa (Table 2).

Seed set by open pollination on the five plants (in *Mentha piperita*) was about 15 per cent. Barring a few, all of them failed to respond to the germination test.

Discussion

The origin and evolutionary implications of cytomixis are not precisely understood. Several suggestions have been put forward to explain the probable origin of cytomixis. These include the effect of fixation (Linnert 1955, Takats 1959, Heslop-Harrison 1966), abnormal pathological conditions (Bobak and Herich 1978, Morisset 1978), physiologically controlled behaviour (Sarvella 1958, Bell 1964), nutritional deficiency (Milajajev 1967) as well as genetic reasons (Brown and Bertke 1974, Omara 1976).

In *Mentha piperita* Linn. cytotype I ($2n=120$), the process of cytomixis probably occurs as a natural phenomenon and not as an artifact as proposed in several other genera. Owing to the persistent occurrence of cytomictic connections in almost all the stages of meiotic division both in fixed as well as in fresh material, it must be considered as a regular cytological phenomenon in this cytotype. Gottschalk (1970) has proposed that this process is limited to genetically unbalanced plants such as haploids, triploids and other hybrids. The regular formation of sixty bivalents during first meiotic metaphase is encountered in this investigated cytotype of *M. piperita*. Exclusive bivalent formation confirms its allopolyploid origin (Kundu and Sharma 1985). The frequent occurrence of several meiotic irregularities is also indicative of its genetically disturbed nature. This cytotype of *M. piperita* therefore seems to substantiate Gottschalk's aforementioned claim. But the presence of cytomixis in *Leonurus* and *Leucas*, as recorded in the present investigation, would seem to negate this claim, as they are cytologically quite stable diploid genera.

Aneuploid plants are a common feature of polyploid population. Along with the intraspecific polyploid series in *M. piperita* several aneuploid cytotypes of this species have also been noted (Morton 1956, Sharma and Bhattacharya 1959, Belyaeva *et al.* 1970). That cytomixis is specifically involved in the formation of aneuploid gametes has been adequately documented in the literature (Sarvella 1958, Bell 1964, Salesses 1970). Maréchal (1963) also noted that cytomixis contribute to the production of aneuploid gametes. He, however, did not attribute any evolutionary significance for those aneuploid gametes because of their non-viable nature. Salesses (1970) speculated a direct correlation between the production of aneuploid as well as unreduced male gametes and the cytomictic connections in the genus *Prunus*.

The occurrence of cytomictic connection in this cytotype of *Mentha* is important in that, it is associated with high pollen sterility (51.77%) and no viable seed formation. However, mere cytomixis cannot account entirely for such a high percentage of pollen sterility recorded here. Obviously, other factors including meiotic irregularities such as laggards, early separation of bivalents, micronuclei formation are also involved. Nevertheless, it is difficult to state precisely the other causes of infertility. It is probable that cytomixis is responsible for the production of aneuploid gametes in this cytotype. From an evolutionary point of view,

however, this is of limited significance because of the absence of viable seed setting.

Cytomixis has been noted within two out of the nine populations of *Leonurus sibiricus* studied. These two populations also possess a high percentage of sterile pollen grains in comparison to others (Table 2). The relatively high percentage of pollen sterility noted in populations V and VII may presumably be attributed, at least partly, to such cytotoxic connections.

In *Mentha arvensis* L. var. *javanica* (Bl.) HK. f., the cytotoxic connections involving microsporocytes at first meiotic prophase may be one of the factors leading to the subsequent disintegration of the microsporocytes.

Abstract

Cytomixis has been observed in the pollen mother cells of *Leucas aspera*, two species of *Mentha* and two populations of *Leonurus sibiricus*. In *Mentha piperita* L. ($2n=120$), it has been noted to be of common occurrence as it occurs in almost all the stages of meiotic division. The probable correlation between the aneuploid gamete production, pollen sterility and the cytotoxic connections in all the investigated taxa has been indicated.

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