Cytomixis in Pollen Mother Cells of *Polygonum tomentosum* Schrank

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Cytomixis refers to the migration of chromatic material from one cell to an adjacent cell through cytoplasmic connections. It was first recorded by Kronicke (1901) in pollen mother cells (PMS) of *Crocus sativus*, and occurs in several families of flowering plants (Bell 1964, Cheng *et al.* 1975, Kamara 1960, Narain 1979, Omara 1976, Sirvastav and Rain 1980). Cytomixis is generally reported during microsporogenesis in genetically unbalanced plants such as hybrids (de Nettancourt and Grant 1964), mutants (Gottschalk 1970, Datta and Biswas 1984), triploids (Salesses 1970), apomicts (Mantu and Sharma 1983), plants affected by triflurine (Bobak and Harich 1978) and even in some normal diploids (Sapre 1978, Chauhan 1981).

Cytomixis has also been recognised in somatic cells (Bowes 1973, Jacob 1941, Brown 1947, Sarvella 1958, Tarkowska 1960, 1965), at the interface between somatic and meiotic cells (Cooper 1952) and in the epidermal cells of scales and leaves (Tarkowska 1960, 1965).

Very little is known about the occurrence, cause or significance of cytomixis. Heslop-Harrison (1966) reported that cytomixis normally allows the exchange of cell organelles between cells. However Omara (1976) suggested that this process can represent a real migration of chromatic material between cells during meiosis. Sarvella (1958), Bell (1964) and Sapre and Dayarani (1987) have stated that cytomixis may be a cause of many polyploid and aneuploid forms in many species of plant.

During a programme of cytological studies on Egyptian members of the Polygonaceae, cytomixis was observed only in *Polygonum tomentosum*. This distinctive plant is considered by Akeroyd (1993) to fall within the widespread species *P. lapathifolium* L. Previous counts have shown it to have $2n=22$ (Bolkhovskikh *et al.* 1969). It is an annual ruderal of wet mud.

Materials and methods

Preanthesis flower buds were collected from five localities near Rashid City, Behera governate, Egypt between 8 a.m. and 10 a.m. Buds were fixed in a freshly prepared solution of absolute ethanol and glacial acetic acid (3/1, v/v) for 24 hr. Buds were later stored in 70% ethanol at 4°C. Both fixed and freshly collected anthers were squashed in 2% acetocarmine using an iron needle. Pollen grains were also stained in acetocarmine to study pollen stainability. Photographs were taken from freshly prepared slides using a Richter photostar research microscope with automatic camera.

Results and discussion

Many authors suggest that cytomixis is an artifact of fixation (Linnert 1955, Takats 1959, Ahadi and Sharma 1988). However, I found that it occurs at an equal frequency in fixed and freshly stained material.

In *P. tomentosum*, cytomixis is represented by a variety of conditions ranging from empty

cytoplasmic connections (Figs. 3, 7) to complete cell fusion (Fig. 2), sometimes rows of up to four cells are connected together (Fig. 1). In some cases, cells showed multiple connections, as was also reported by Ahadi and Sharma (1988) and Basavaiah and Murthy (1987). In both meiosis I and meiosis II, cell fusions formed about 17% of cytotoxic events (Table 1). The total proportion of PMC's showing cytotoxicity was 20%, and this did not differ significantly between populations.

In general, cytotoxicity was less commonly observed in meiosis II than in meiosis I, as reported by Bahl and Tyagi (1988) and Ahadi and Sharma (1988). Cytotoxic connections observed during metaphase and prophase of meiosis I, II (Figs. 3, 7) typically contained univalents and disoriented chromosomes (Fig. 3).

Figs. 1-7. 1. A series of PMC's showing cytoplasmic connections. 2. PMC's showing direct fusion during meiosis. 3. PMC's showing two cytoplasmic connections during cytotoxicity at metaphase I. 4, 5, 6. Abnormal anaphase I, telophase I and tetrad stages showing univalents and lagging chromosomes. 7. PMC's showing two cytoplasmic connections during cytotoxicity at prophase II.

Table 1. Number and percentage of cytotoxic and abnormal PMC's and pollen fertility in *Polygonum tomentosum* L.

F.C.	No. of obser. cells		No. of cytotoxic cells				% of cyto. cells	% of abn. cells		% of pollen fertility
			MI		MII					
	MI	II	C.C.	D.F.	C.C.	D.F.		MI	II	
1	230	200	45	11	23	3	19.06	33.2	22.9	70.4
2	310	250	60	12	26	4	18.21	35.0	24.1	66.7
3	200	170	45	12	23	3	22.43	30.6	20.7	66.6
4	180	160	42	11	19	2	21.7	28.1	21.3	67.7
5	210	180	43	10	22	3	20.0	28.9	19.5	69.7
G.M	266	192	47	11.2	22.6	3	20.29	31.2	21.7	68.6
S.E	20.1	14.3	2.54	0.33	1.09	0.28	0.71	1.16	0.73	0.69

C.C.=cytoplasmic connection, D.F.=direct fusion, MI=meiosis I, G.M.=grand mean, cyto.=cytotoxicity, MII=meiosis II, abn.=abnormal, F.C.=field collection, S.E.=standard error.

A high proportion of meiotic cells (overall 31.2 in MI and 21.7 in MII), both those with and without cytomictic connections, displayed irregularities in the form of laggard chromosomes and univalents (Figs. 4, 5). This proportion also did not differ significantly between populations (Table 1). It seems probable that a combination of cytomixis and disturbed meiosis leads to a high proportion of aneuploid pollen grains. Micronuclei are frequently observed in tetrads (Fig. 6). This suggestion is confirmed by mitotic chromosome counts reported by Nassar (1987) for this species in the same population, recording $2n=54$, which represents an aneuploid form of the pentaploid number ($5X=55$, where $X=11$).

Not surprisingly, the disturbed and aneuploid cytology of pollen grains leads to the abortion of many, so that the average levels of pollen stainability are only 68.2%. Once again, this figure does not differ significantly between populations (Table 1).

The factors responsible for cytomixis are not yet precisely understood. Various factors have been invoked, such as pathological conditions (Morrisset 1979), mechanical injury (Sarvella 1958), and a genetic control system (Mantu and Sharma 1983). In the present study seems very unlikely that cytomictic phenomena are caused by fixation artifacts, as was suggested by Jacob (1941), Takats (1959), or Gottschalk (1970). However in the present study, it is notable that meiosis are very disturbed, suggesting incomplete genomic homology, but it is less clear whether this is a primary or a secondary (postcytomictic) phenomenon. Another possibility that these populations at Rashid are hybridogenous between two members of the *P. lapathifolium* aggregate which show substantial levels of genomic differentiation, and this certainly needs further investigation.

Summary

Cytomixis, representing the migration of chromatic material from one cell to another, was observed in PMC's of *Polygonum tomentosum* Schrank. Cytomixis was observed at all stages of meiosis, but became less frequent in the later stages of meiosis. Direct nuclear fusion and cytoplasmic connections between cells were noted, so that a series of PMC's were sometimes connected together.

Cytomictic cells were often accompanied by other meiotic aberrations such as univalents, laggard and disoriented chromosomes. Levels of pollen stainability are low, indicating a high of male sterility in these populations.

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