Mitotic Abnormalities Induced by Silk Dyeing Industry Effluents in the Cells of Allium cepa

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Summary  The silk dyeing industry effluents were examined for the induction of mitotic abnormalities in Allium cepa root system. Cytotoxicity of the effluent was studied by treating Allium cepa roots with different concentrations of effluent (25, 50, 75, 100%) for different durations (6, 12, 24, 48 h) along with control. The effluent inhibited cell division and a strong dosage effect was obvious from a decline in the mitotic index with the increase in effluent concentration and duration of treatment. The effluent also induced a wide range of mitotic abnormalities. The abnormalities observed included stickiness of chromosomes, fragments, bridges, laggards, binucleate cells and vacuolated nuclei. The results showed that silk dyeing industry effluents act as potential mutagens.

Key words  Dye effluent, Chromosomal abnormalities, Stickiness, Bridges, Allium cepa.

Ever since prehistoric times, man was fascinated to colour the objects of daily use by employing inorganic salts or natural pigments of vegetative or animal origin. But today more than 100,000 synthetic dyes are known of which majority are used to dye textile fibers. Many of these synthetic dyes are found to be toxic in nature and cause havoc in life systems (Khanna and Das 1991, Anupam Kaur et al. 1993, Sudhakar et al. 2000). Except for a few handful of direct exporters none of the small units who primarily undertake contracts have effluent treatment plants. Dye wastewater contributes a number of contaminants including acids or caustic liquors, dissolved solids and colour. The dyestuff molecules are normally biologically resistant and thus many a times the conventional biological methods of effluent treatment are impractical. Studies have shown that the untreated dyeing effluents contain substances that could endanger the aquatic life (Sivaram Prasad et al. 1995, Senthilnathan and Azeez 1999). The wastewater from different industries is also known to induce chromosomal abnormalities in plant cells (Ravindran and Ravindran 1978, Shanthamurthy and Rangaswamy 1979, Malabika Ray and Barman 1987, Somashekar 1987, Thangapandian et al. 1995). Though the silk dyeing units are present in large numbers, their effluents have not come in for much genotoxic testing. Therefore it was considered desirable to test the mutagenic potential of the silk dyeing effluents on the mitotic cells of Allium cepa, as this information will be helpful in understanding the mechanism of cytological damage as well as its implication on environmental pollution.

Materials and methods

Silk industry dye effluents were collected from the dyeing units located in and around Bangalore City. Standard methods (APHA 1995) were employed during the collection, preservation and analysis of the dye effluent. Suitable concentrations of the effluent were made with distilled water. Germinating onion bulbs with roots 2–2.5 cm long were immersed in 25, 50, 75, 100% effluent for 6, 12, 24, 48 h. A control with distilled water was maintained in all the cases. Cytological prepara-


tions were made from the control and treated root tips of *Allium cepa* following the methods recommended by Sharma and Sharma (1980). Observations and photomicrographs were made with a Leitz Orthoplan microscope.

**Results**

The average values of physico-chemical characteristics of the effluents are given in Table 1. The effluents were acidic and contained high amounts of total solids and chlorides.

The effluent inhibited cell division in the treated root tips of *Allium cepa*. A marked lowering of the mitotic index with increasing concentration and duration of treatment of the effluent was noted (Table 2).

Screening of mitotic root tip divisions revealed a wide range of chromosomal abnormalities in all the concentrations and duration of treatment. Some of the abnormalities are presented in the Figs. 1–9. The percentage of abnormalities was dose and duration dependent. Abnormalities increased with higher concentration and longer duration. The abnormalities observed were sticky anaphase, precocious movement, lagging fragments, C-metaphase, multiple anaphase bridges and chromatid gaps. Acentric fragments produced due to the breaks were also abundantly found. One of the peculiar abnormalities observed was the banded nature of the chromosomes. The cells and nuclei were not an exception to the effluent treatment. Binucleated cell and vacuolated nuclei were common in the higher concentrations.

**Discussion**

The observations of the present study is a clear indication of the mitoclastic and clastogenic property of the effluent, which is evident from the lowering of the mitotic index and manifestation of spindle abnormalities. The lowering of mitotic index might have been achieved by the inhibition of DNA synthesis at S-phase.

Stickiness of chromosomes observed in the treated cells reveal the depolymerisation effect of effluent on the nucleic acid of the chromosomes. Soheir *et al.* (1989) reported a high level of chromosomal stickiness in metaphase stage after herbicide treatment. Patil and Bhat (1992) reported that stickiness is a type of physical adhesion involving mainly the proteinacious matrix of chromatin material. The formation of small fragments can be attributed to the chromosomal breakage due to the effect of the effluents.

The formation of bridges could be attributed to chromosomal stickiness (El-Khodary *et al.*
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1990) and to chromosome breakage and reunion (Haliem 1990). Induction of bridges and breaks may lead to loss of genetic material (Salam et al. 1993). Binucleate cells that were frequently found at higher concentrations may be due to the inhibition of cell wall development at telophase. Several cyclic hydrocarbons, methylated oxypurines and plant extracts were known to inhibit cell plate formation (Amer and Farah 1971, Abraham and Cherian 1976). The banded nature of the chromosomes might be due to the enzymatic action of the microorganisms present in the effluent. Combined effect of the pretreatment of cells with colchicine and the effluent may be responsible for the induction of C-metaphase.

Vacuolated nuclei observed in the treated cells reveal the severe cytological effect of the efflu-
The precocious movement of the chromosomes might have been caused by the early terminalisation, stickiness of chromosome and/or because of the movement of the chromosome ahead of the rest during anaphase (Permjit and Grover 1985).

From the above observation it is evident that silk dyeing effluents are potential genotoxic agents in the environment. The abnormalities manifested may be due to the action of organic and inorganic traces present in the effluent and also might be exerted by the microorganisms whose secretions cause cytological damage (Rangaswamy et al. 1982). Hence extensive pollution control measures are to be taken to prevent the lethal damage caused by the flow of untreated pollutants. Further the use of eco-friendly dyes should be encouraged in view of the safety of the future generation.

### References


### Table 3. Types and percentages of mitotic abnormalities induced by the different concentrations of silk dyeing industry effluent in *Allium cepa* root tips after different treatment durations

<table>
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<tr>
<th>Treatment</th>
<th>Conc. (%)</th>
<th>Percentage of different types of abnormalities</th>
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* Include chromatid gaps, binucleate cells, vacuolated nuclei etc.


