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MITOTIC ABERRATIONS INDUCED BY *CASSIA OCCIDENTALIS* L. IN *ALLIUM CEPA* L. ROOT TIP CELLS

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ABSTRACT

The effect of *Cassia occidentalis* was studied on mitotic activity of root tip cells of *Allium cepa* L. in order to view its phytochemicals potential. Onion root tips were treated with 5% (w/v) concentration of aqueous extracts of root, leaf, flower and stem of *C. occidentalis* prepared in distilled water for 18 hours. Mitotic indices were calculated and observed root squashes for different chromosomal abnormalities. Leaf and flower extracts inhibited mitosis completely whereas few stages of division were observed in root and stem extract treated samples. Various chromosomal aberrations included circular cells, balloon like disrupted cells, fragmentations, binuclear cells etc. The study concluded that leaf and flower extracts of *C. occidentalis* are potential inhibitors of cell division thereby inhibiting growth of neighbouring plants in nature.

Key Words: *Cassia occidentalis*, *Allium cepa*, Chromosomal Aberrations, Mitotic Index, Phytotoxicity

INTRODUCTION

C. occidentalis is a small annual shrub, 1.2 m tall; stems ribbed, glabrous; petiole with gland at base; leaves compound, usually having 4-6 pairs of elliptic or lanceolate acuminate glabrous leaflets each 3-7.5 cm long, the terminal pair largest; flowers in racemes, yellow, nearly 1 cm broad; 3 of the stamens infertile; pod linear, glabrous, 10-15 cm long, about 6 mm wide, the margins somewhat thickened, with about 20-30 flat, ovate, brown seeds each 3 mm long (Stone, 1970). It is a common roadside weed (Stone, 1970) and naturalizes primarily in dry, disturbed habitats such as pastures, pineapple or cane fields, river banks or sand dunes (Wagner *et al.*, 1999; Smith, 1985). This species is characterized by the fetid odour of crushed leaves, reddish purple stems, ovoid gland immediately above the thickened petiole base, its bright yellow flowers and the sickle shape, laterally compressed pods.

There are many studies related to allelopathic potential of *C. occidentalis* with wide reference of its inhibitory effect on seed germination and growth of other plant species (Swami, 2011) but the mechanism of inhibition has not been elucidated so far. Therefore, present study was planned with an aim to explore the mechanism underlying growth suppression of treated plants by *C.occidentalis*. *Allium cepa* has been selected as a test plant because it has been suggested as the best material for cytological tests (Fiskesjo, 1985).

MATERIALS AND METHODS

Sufficient plant material of *C. occidentalis* was collected, dried, powdered and stored in airtight jars till further use. Clean and healthy bulbs of *Allium cepa* were induced to root by placing them on 100 ml beakers filled with distilled water. In order to determine the effect of aqueous extracts of *C. occidentalis* on mitotic activity of root meristem, roots were raised from *A. cepa* bulbs, by immersing their bases in distilled water for three days in dark. On the fourth day, rooted onions were subjected to treatment with different extracts and distilled water as control. After 24 hours, roots were (1 cm long) separated and kept in fixative (Glacial Acetic acid: Absolute alcohol, 1:3) for another 24 h and finally transferred to 70% ethyl alcohol and kept at low temperature till used. For squash technique, some root tips were rinsed in distilled water and kept in acetocarmine (chromosomal stain) for 15-20 min. On a clean and dry slide, single root tip was placed and the tip portion (approximately 1 mm) squashed thoroughly with a fresh drop of acetocarmine. The mitotic stages in the root cells were observed under a binocular microscope

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(MIPS Integrated MAGNUS, India)). Five replicates were maintained for each sample and 10 areas (consisting of 100 cells each) per slide were examined under microscope. In addition, a visual microscopic observation of the root squashes for any type of abnormal mitotic arrangement of atypical cell disruption was also made.

RESULTS AND DISCUSSION

Roots of *A. cepa* clearly depicted the effect of various treatments on mitotic activity. In control roots, actively dividing cells at various stages of mitosis *i.e.* prophase, metaphase, anaphase and telophase with high mitotic index were observed $82 \pm 2\%$ (Figure 1). No chromosomal or mitotic aberrations were observed in any of control root tip. Root extract treated cells of *A. cepa* showed abnormalities *viz.* balloon like and disrupted cells with some dividing cells (Figure 2). However, in roots treated with 5% (w/v) of leaf extract, binucleated and elongated cells were observed (Figure 2, 40X). Some of the cells were balloon like showing eccentric nucleus. These cells were also observed in control roots but number increased with leaf extract treatment. In Figure 3, Flower extract treated root tips were observed to have abnormalities such as balloon shaped cells, irregular cells and binucleated cells. In stem extract treated cells (Figure 3), some of the dividing cells were present though the mitotic index was lower as compared to control *i.e.* 28 ± 1 . Some circular and binucleated cells were also present.

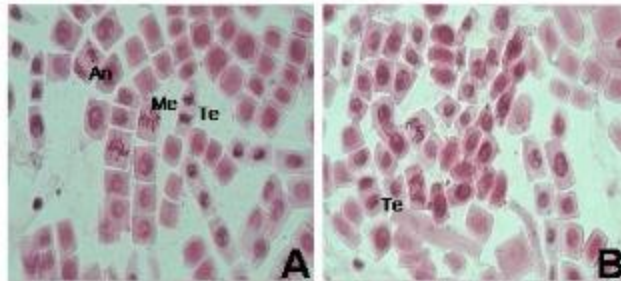


Figure 1: Control Root Tips of *Allium cepa* (Me-metaphase, An-anaphase, Te-telophase)

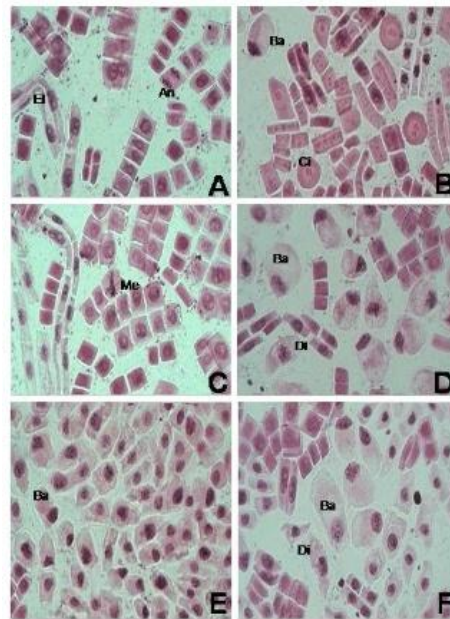


Figure 2: Root (A-D) and Leaf Extract (E-F) treated Root Tip cells of *Allium cepa* (El-elongated cell, Ci-circular cell, Ba-balloon cell, Me-metaphase cell, An-anaphase, Di-disrupted cell)

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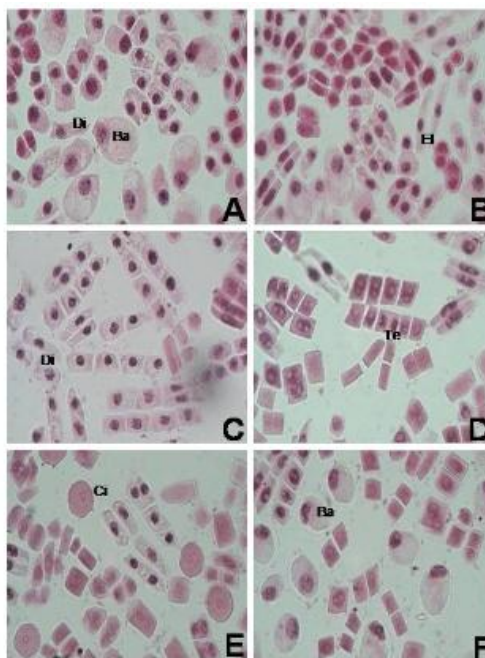


Figure 3: Flower (A-B) and Stem (C-F) Extract treated Root Tip cells of *Allium cepa* (El-elongated cell, Ci-circular cell, Ba-balloon cell, Te-telophase cell, Di-disrupted cell)

Thus most visible cytological changes were displaced nucleus, dispersed chromosomes, binuclear cells and balloon like cells. Such abnormalities are mainly related to incompletely developed microtubules (MTs) which are under control of Ca^{+2} ions (Vidakovic-Cifrek *et al.*, 2002; Hepler, 1992). A reduction in mitotic index can be due to blocking of DNA synthesis at G_1 or S phase (Schneiderman *et al.*, 1971; Sudhakar *et al.*, 2001) or blocking of G_2 phase (Van't, 1968).

Present study clearly elucidated the phytotoxic impact of *C. occidentalis* extracts. Many studies have revealed a reduction in growth of seedlings treated with *C. occidentalis* under laboratory conditions (Farooq *et al.*, 2008; Swami, 2011) and effect was more pronounced in radicle length as compared to plumule length. This study has further corroborated the effect of *C. occidentalis* extracts on mitotic activity of root tip meristem. Various aberrations are indicator of clastogenic impact of *C. occidentalis* which can be correlated with significant allelopathic potential of *C. occidentalis* to neighbouring plant species thereby acquiring an invasive habit. Flower and leaf extracts exhibited pronounced effect on mitotic activity of *A. cepa* cells which may be attributed to their rich phytochemistry as compared to stem and root.

ACKNOWLEDGEMENT

The financial support from University Grants Commission, New Delhi, India in the form of minor research project is greatly acknowledged.

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