REVIEW OF LITERATURE
Ever since the dawn of the civilization, it has been the major task of man to engage in a continuous endeavor to improve his living conditions. One of the main tasks in which human beings have been engaged in securing relief from hunger a basic human need. Today India is engaged in the gigantic task of feeding over 1000 million people and a huge cattle population on which the poor farmer is dependant for his livelihood. Secondly the control of insects, weeds, fungi and other pests of economic public health are of utmost importance to our government. The task could have been impossible but for the green revolution of 1960, which has given reasonable hope for the country being not only self
sufficient in the production of adequate food and fodder for feeding its
teeining human and animal population but has become the largest
producer of some important commodities, on the other hand pesticides
have given rise to serious problems (Gupta, 1989). Pesticides necessitate
close scrutiny, not only because of the large annual volume and varieties
of chemicals used, but also because of their high stress index
(Khilchevskaya, 1980). In addition they are only the chemicals to which
the greatest varieties of organisms are exposed directly and indirectly,
sooner or later. Furthermore the humans that come into contact with
pesticide during their manufacture and application must be very high and
in fact, immeasurable. Besides being toxic to the targets as per design,
pesticides have other effects on these as well as non target systems. These
effects are the genetic effects recognized for their long term value. The
genetic purity built across thousands of years and therefore needing
careful preservation is currently under threat by the not so careful usage
of pesticides. The genetic effects may be genic (i.e. mutagenesity),
chromosomal (i.e. clastogenisity) or genomic (aneugenic) to various
agroecosystems, including man. These three terms together are referred to
as genotoxicity and the later two as karyotoxicity.
Pesticides are thus known to cause heritable defects, cancers and birth defects in man and contribute to other health factors such as heart ailments and aging. All these obviously threaten the genetic health of current and future generations. The effects on the flora and fauna and on the ecosystems are also well documented. The effects of pesticides on plants and other organism were studied extensively by previous worker. The brief account of the use and effects of pesticides is given in this part of this study.

Plant bioassays have gained in popularity since the 1991 St. Petersburg conference. Even before the publication of the results of the conference in 1994, more and more genotoxicity studies using plant bioassay appeared in the literature. These assays have provided a practical application of genetic toxicology to real world population problems. It seems that the scientific community at the international level also recognizes the value of these systems. Ma et al. (1978) were compiled more than 200 papers on the Plant bioassays in own review literature. The popularity of the plant bioassay was due again when it was included in the International Program on Chemical Safety (IPCS) which is under the World Health Organization (WHO) and United Nations Environment Program (UNEP). Eleven countries participated in
validation project using four major Plant bioassays i.e. *Vicia* root chromosome aberration, *Arabidopsis* embryo mutation, *Tradescantia* stamen hair mutation and *Tradescantia* micronucleus tests in the mid 1980s (Ma 1999). Nearly 2 years of collaborative effort of eleven countries completed the validation project.

In particular *Tradescantia*, micronucleus bioassay was selected as one of the assays for this IPCS collaborative study owing to its recognized potential monitoring capabilities, as well as its broad data base (Hopke *et al.* 1982; Lo 1985; Ruiz *et al.* 1992; Sandhu *et al.* 1989;). *Tradescantia* micronucleus assay has been used in the monitoring of air (Ma 1990; Ma *et al.* 1984) waste water (Grant *et al.* 1989, 1992; Ma 1992) contaminated soil (Kong and Ma 1998) insecticide (Ma *et al.* 1983) and drinking water (Ma *et al.* 1985) as well as with the testing of many mutagens or carcinogens (Ma 1979; Fang 1981; Sandhu *et al.* 1989; Knassamuller *et al.* 1992).

*Tradescantia* stamen hair mutation assay was adapted for the detection of mutagenic airborne agents volatile organic compounds (Sparrow and Schairer, 1971) and still later for the evaluation of chemical agents in liquid form (Schirer *et al.* 1982). Furthermore the *Tradescantia* stamen hair mutation assay has been used to test a wide range of
chemicals such as pesticides (Ahmed and Grant, 1972) sewage sludge (Hopke et al., 1982) soil samples in the vicinity of lead smelter (Lower et al., 1983) and engine exhaust fumes (Ma et al., 1983). Recently the Tradescantia stamen hair mutation assay has been used in the in situ monitoring of mutagenic chemicals waterways and aquatic effluents (Grant et al., 1992; Ma et al., 1994).

The mitotic root meristem of Allium cepa and Vicia faba have been the pioneer cytogenetic materials for clastogenicity studies of physical and chemical agents since the early 1930s. Based upon the review papers published under the US EPA Gene-Tox Program (Grant 1982a, Ma 1982) chromosome aberration frequencies in the root tips of these plants were utilized as the indicators of clastogenicity in most studies prior to 1980's. In recent years, chromatid aberrations (Cortes and Mateos, 1991), sister chromatid exchanges (Cortes et al. 1987; Kuglik and Slotova, 1991) and micronuclei (Arora et al., 1969; Degrassi and Rozzoni, 1982; Dash et al 1988; panda et al., 1980; Chandra et al., 2002, 2004, 2005; Chuhan et al. 1999, 2004, 2005; Rank et al., 2002; Saxena et al 2004, 2005; Yuzbasioglu, 2003) have been used more frequently as parameters of clastogenicity. Both of these systems have a large data base with an extensive list of chemical agents which have bee tested (Grant 1982).
Several other higher plants provide unique and valuable systems for detecting and analyzing the effects of chemical mutagens. (Nillan and Vig 1976). Such plants include Allium cepa, Allium sativum, Hordeum vulgare, Zea mays, Lycopersicon esculentum, Arabidopsis thaliana, Glycin max, Crepis capillaris, Lillium, Pisum sativum and Nicotiana tabacum.

As a group these plants offer systems for the analysis of almost all known genic and chromosomal aberrations which have been induced in eukaryotes by chemical or physical mutagens. Some of the advantages in utilizing plant systems have been reviewed by several workers (Nillan and Vig 1976, Kihlman 1975, Grant, 1978). These are (1) the chromosome organization of plants is similar to that of human; (2) many plants are easy to grow; (3) some have short generation time; (4) the cost, handling and space requirements are relatively small; (5) the cost and time of training technician to handle a variety of end points following mutagen treatment is relatively small; (6) mutagenic effects can be studied under a wide range of environmental conditions such as large difference in pH, water content, temperature and metabolic rates; (7) most of the plant systems have been in use for many years and are reliable systems which have been adapted for newer techniques such as
chromosome banding, sister chromatid exchange studies. Perhaps the most serious disadvantage of plant system for the detection of genetic risks to man is the lack of similarity between vegetative and mammalian metabolism. Nevertheless, the positive correlation which has been noted between aberrations induced by same chemicals in plant root tip cells and in cultured mammalian cells indicates that a plant root system must be recognized as an appropriate first tier assay system. The ease and accuracy of detecting and quantifying chromosomal aberrations have largely depend on the progress in the development of cytological techniques over the years. Most of the important concepts on the spontaneous and induced chromosomal aberrations came from the study of plant tissues such as root meristem (e.g. Vicia faba, Allium cepa, Hordeum vulgare), pollen mother cells and pollen grains (Tradescantia) which were amenable for simple smear and squash techniques using stains such as aceto-carmine, aceto-orecin and feulgen. Low number and large sized chromosomes made these plants species ideal for chromosome studies (Natrajian 2005).

Considerable emphasis has been placed internationally in studying the genetic hazards of pesticides. Epstein and Legator (1971) detailed the mutagenic nature of various pesticides, their potential public health
hazards, methods and program for mutagenicity testing. Durham and Williams (1972) studied the mutagenic, teratogenic and carcinogenic properties of several pesticides. Fleck and Hollander (1982) also studied the genetic toxicology of the chemicals used in the agricultural perspectives. The carbamate pesticides including barban, benomyl (Spasojevic 1974), carbaryl (Amer and Farah, 1968), chlorpropham (Sawamura, 1965) propham (Doxey, 1949 and Drenne 1953) and diallate were reported as C-mitotic chemicals.

It is more than twelve decades since the existence of chromosomes and their divisions during cell proliferation were demonstrated. Since then, advances in our knowledge with regard to the fine structure of chromosomes, their replication during cell division, and localization of genes in chromosomes, transcription and expression of genes at chromosomal level have been considerable.

Chromosome aberrations have been used as a measure of reproductive success in plants for many years and have been correlated with morphological and taxonomical changes, fertility-sterility relationships, mutations and other characteristics. The first observation of correlation between reduction in fertility and cytological abnormalities by Kostoff in 1931 when he observed that the seed set of tobacco plants
greatly reduced after the pesticides treatment. He found several chromosomal abnormalities which he considered as the cause of partial sterility of plants.

Amer and Farah (1968) and Swamura (1965) were reported the induction of bi-nucleate and multinucleate conditions including tri and tetrapolar anaphases following the treatment of certain pesticides. Chromosome contraction has been observed following treatment of Tradescantia root tips with mercury compounds and some carbamates (Ahmad and Grant 1972, Storey and Jordan 1968). Klasterka et al. (1976) and McGill et al. (1974) suggested that chromosome stickiness arises from improper folding of chromosome fiber into single chromatids and chromosomes. As a result there is intermingling of the fibers and the chromosomes become attached to each other by means of subchromatid bridges. Amer and Ali (1969) reported that pentachlorophenol induced fragmentation of both mitotic and meiotic chromosomes of Vicia faba. Wuu and Grant (1966) have also reported the fragmentation in Hordeum by the treatment of linuron.

The most common abnormalities recorded in these categories are chromosome and chromatid break, acentric fragments, chromatid and subchromatid exchanges, chromatid gaps and heterochromatic regions
and sister chromatid exchanges at metaphase, chromatid and chromosome bridges and fragments at anaphase. A very detailed classification system has been proposed by Savage (1975). Chromosome breaks, fragments, chromatid exchanges and dicentric chromosomes are generally considered unstable aberrations; deletions inversions, duplications and translocation considered stable aberrations. Evans (1977) considered that chromosome breakage involve the DNA molecule responsible for the linear continuity of the chromosome. Such aberrations are result of unfinished repair of mispair of DNA. Price and Schank (1973) reported that the growth retarding chemical maleic hydrazide induces chromosome breakage largely in heterochromatic regions.

Singh et al. (1979) studied the effects of several insecticides on the morphological and cytological effects of insecticides on *Hordeum vulgare*. The study revealed that insecticides treatments were reduced the seed germination, seedling height, pollen fertility and chiasma frequency. Different chromosomal aberrations viz. chromosome fragments at mitotic metaphase, chromatid bridges, fragments laggards, bridges with fragments were observed in the somatic cells of barley. Such somatic chromosome bridges were also observed by Sakamura (1920), Masukodani (1948), Swaminathan and Natrajan (1956) and Srivastava...
(1966). Univalents, precaucious separation and non-orientation of bivalents at meiotic metaphase I; and laggards bridges, bridges with laggards, tripolarity and unequal separation at anaphase I were observed in meiotic cells of the treated populations in frequencies significantly higher than those in the control. Singh et al. (1980) have been reported the induction of chlorophyll mutation by seed treatment with insecticides in *Hordeum vulgare*. All the insecticides induced mutations when applied in the G1 and S phase of the cell cycle.

During the course of cytogenetical investigations using Para Dichloro Benzene (PDB) as prefixing agent for the proper spreading of somatic chromosomes when the root tips of *Lens esculenta* were treated beyond 6 hrs. various types of abnormalities in the morphology of chromosomes and their behavior during mitosis were noticed in many cells (Sarbhoy 1980). Carey and McDonough (1943), Sharma and Bhattacharya (1956), Srivastava (1966) made extensive studies on the influence of PDB in altering the chromosomal morphology of several plants. Simonet and Guinochet (1939) successfully employed PDB in producing heteroploidy in *Linum usitatissimum*. Carey and Mc Donough (1943) working with *Allium* obtained polyploid cells during metaphase stage.
The genotoxicity of Malathion was evaluated with *Tradescantia* micronucleus test (Ma et al. 1983). In this study they adapted four different treatment procedure like absorption of Malathion and water mixture through stem, spraying of Malathion and water by spraying onto plant cuttings, spraying of the mixture on open population of plants in green house and adsorption of Malathion fumes through the leaves and buds.

Kabir and Alam (1986) was studied to undertaken the occurrence of possible irregularities in M1 and M2 barley plants by the treatments of insecticides Carbicron-100 EC and Vapona -50 investigation was made to see the meiotic changes in M1 and M2 barley plants due to treatment of Carbicron and Vapona, applying three methods (seed treatment, seedling spraying and seed treatment plus seedling spray) in three different concentrations. The effects of insecticides in pollen mother cells showed various kinds of abnormalities such as laggards, bridges, fragments and micronuclei. The abnormal PMC’s were found to increase with an increase of concentration of insecticides. Considering the secondary effects of insecticides as observed in M1 and M2 generation of barley plants in the present investigation, it is suggested that the farmers should not use insecticides indiscriminately on any crop plants but should choose
specific insecticide for a particular crop which has less harmful effect to the plant and at the same time effective in controlling the harmful organisms.

The effect of herbicide (Garlon – 4) has been studied on the root mitosis of *Allium cepa* (El-Khodery *et al.* 1989). Garlon affects the relative duration of each mitotic stage as compared with control. It also caused reduction in mitotic index, indicating mitotic inhibition and increase in the frequency of abnormal mitosis, the percentages of which are highly significant. Star metaphase was one of the chromosomal abnormalities observed. Such type of abnormality was also observed after treatment of *Vicia faba* root tips with “ROGOR” (Amer and Farah, 1974) and considered as being a fore step of complete disturbance of the spindle (Amer 1966). The effect of Garlon-4 on root mitosis stimulates that of colchicines in the type of abnormal meta and anaphase and the induction of polyploid cells as well as accumulation of metaphases.

Kumar and Sinha (1989) determine the threshold dose (dose of pesticides on which they retain their pesticidal property but have little or no cytotoxic/ genotoxic effect) of a few commonly used pesticides lindane, malathion and metacid in a well known test system.
Sinha et al. (1989) studied the cytological effects of pesticide phosalone on root meristem cells of *Allium cepa*. The study revealed that phosalone had a detrimental effect on the test material, there was inverse relation between the mitotic index and the dosage and time of treatment, and the direct relation between the mitotic index and the dosage and the time of treatment of percentage of abnormalities, treatments not only brought down the frequency of dividing cells, but also produced a good number of anomalies in the mitotic cells. There was a marked decrease in the mitotic index and gradually increase in the percentage of chromosomal abnormalities as the concentration of the experimental solution and the time of treatment increased. Diplochromatids, chromosome erosion, pycnotic cells and highly disintegrated nuclei were some of the other anomalies that were observed in varying degrees after the treatment with pesticide phosalone.

Some information related to the cytological effects of two benzoyl phenyl urea namely XRD473 and IKL 7899 on root tip meristems of *Vicia faba* and *Hordeum vulgare* were provided by the Abdel Rahem and Ragab (1989). The used agents had the pronounced cytological effect on *Vicia faba* and *Hordeum vulgare* which cause a higher proportion of mitotic anomalies. The frequency of aberrant cells tends to increase by
increasing the concentration of the used agents. A considerable percentage of multinucleated cells were observed. It has been shown that such cells could arise from multipolar ana-telophase cells or anaphases with lagging chromosomes.

The cytotoxic effects of two organophosphorus pesticides viz. methyl parathion and phosphomidon were assessed employing chromosomal aberration bioassay in root tip cells of okra, onion, bean and guar (Patil and Shirashyad, 1991). Different cytological aberrations viz. non-orientation and mis-orientation of chromosomes and stickiness, bridges, precocious movement, laggards and fragments, diagonal spindle formation, clumped nuclei, micronuclei and bi and trinucleated cells were observed in increased frequency with increasing concentrations of pesticides.

Carbendazim has been found to be clastogenic both during meiosis and mitosis in pearl millet and sunflower (Harichand et al. 1991). The study revealed that the chromosomal aberrations were more serious in pollen mother cells as compared to somatic cells. The number of somatic cells with chromosome aberrations generally showed an increase with increase in concentration of Carbendazim. The chromosomal aberrations in pollen mother cells of pearl millet also increased with increase in
concentration of the fungicide. However reverse was true in case of sunflower which is unexplainable by them. Severe distortions observed in the pollen mother cells of pearl millet. Majority of the pollen mother cells of sunflower were characterized by lagging chromosomes. Choudhary and Sajid (1986) while using Carbendazim as spray on *Pisum sativum* observed effects on chiasma frequencies upto M$_2$ generations.

Topaktas and Rencuzogullari (1991) studied the cytological effects of commonly used herbicides gesagard and igran in the region of turkey in barley. Gesagard and igran are isomer of each other. In this study, it has been found out that although gesagard and igran are the isomers of each other; their effects on the cells with in the same concentration and same treatment duration are different. They concluded from the present study that herbicides affecting the tubulin proteins destroy the spindle fibres of the cells and cause C-mitosis. They affect the histone proteins and chromosome contraction. They were also affecting the DNA; they increase the frequency of chromosome aberration. Where as Savkoic *et al.* (1984) has been reported that gesagard has not increased chromosomal aberrations in mouse.

Akhter *et al.* (1992) reported the effects of age on wheat and barley seeds. Germaination percentage of different years old wheat and barley
seeds were found to decrease gradually with an increase of the storage times. Mitotic index and chromosomal irregularities from root tip cells were also studied. Most of the irregularities were characterized by the precocious separation of chromosomes and inactivation of spindle mechanism, chromosome fragments and laggards, bridge condensed and sticky chromosome, ring chromosome etc. in both the materials the frequencies of dividing cells were found to decrease with the increase of age of seeds.

The genotoxicity of two widely used agrochemicals in Pakistan, methyl parathion (insecticide) and tri-miltox (fungicide) have been studied to evaluate their ability to induce chromosomal aberrations in the meristematic root tips of *Allium cepa* by Ahmad and Yasmin (1992). Comparing the three treatments applied, seed treatment produced maximum chromosomal aberrations, and the root treatment with recovery period produced the minimum chromosomal aberrations with the root treatment occupying the intermediate position.

The study on the genotoxicity of organophosphorus pesticide, malathion on *Allium* root meristems in vivo was carried out by the Priya *et al.* (1996). Malathion is a widely used organophosphorus pesticide with a wide spectrum of activity and diverse applications in agriculture.
Malathion treated cells showed a reduction in the mitotic index during all dose and durations. The pesticidal compound induced structural aberrations of chromosome such as metaphasic clumps, fragments, vagrant chromosomes, and anaphasic bridges. The chromosomal aberrations observed in the treated cells were found to be of higher frequency during longer exposures. Malathion also induced the mitotoxicity in human lymphocyte cultures. (Nicholas, A. H., Michele, V., Berche, H. V. D. 1979 Mutat. Res. 31: 9).

Chromotoxic effects of one fungicide (Dithane M-45) and two insecticides (Aldrex-30 and Metacid-50) were studied by Pandey et al. (1994) on the basis of chromosomal aberrations during root tip mitosis. A wide range of chromosomal abnormalities were recorded in the treated roots. All the three chemicals had severe effect on cell division. Dithane M-45, Aldrex-30 and Metacid-50 are used frequently at home and kitchen garden in addition to agricultural field has positive chromotoxic effects. This study warrants indiscriminate spraying of these chemicals for chemotherapy and plant protective measures at home garden and kitchen garden.

Azadirachtin, based upon its computer automated structure evaluation (Rosenkranz and Klopman, 1995a), is supposed to act as a
genotoxic carcinogen due to the presence of a furan moiety having the biophore – O–CH= which incidentally occurs in many known genotoxic carcinogens including aflatoxins. Moreover, the calculated electronegativity of azadirachtin is of the same order of magnitude as that for DNA reactive molecules (Rosenkranz and Klopman, 1995b). Azadirachtin thus can be considered as a potential genotoxic carcinogen on account of its electrophilicity as well as its putative DNA reactivity.

The genotoxic effect actually observed in murine spermatocytes is therefore in conformation with the predicted genotoxicity of azadirachtin. Azadirachtin has also been predicted to cause cellular toxicity (Rosenkranz and Klopman, 1995a).

Higher incidence of synaptic disturbances and numerical variations in chromosomes confirms the cytotoxicity of the neem extract which is in full agreement with the observations made in mitotic chromosomes (Awasthy et al., 1995, 1999; Khan and Awasthy, 2001).

The morphological and cytological effects of atrazine and propazine on grain sorghum were evaluated by Randall et al. (1996). Atrazine and propazine at various rates were incorporated into soil as preemergent treatments. Morphological injuries to sorghum and chromosome aberrations were monitored and recorded as a guide for future uses of
these herbicides for weed control during seed production and cytological field studies. Both atrazine and propazine reduced plant height but propazine caused less injury than atrazine. The majority of the chromosomal aberrations were multiple nucleoli detected in diplotene and early diakinesis cells and asynchronization of chromosome movement was also reported.

Induction of adaptive response by conditioning dose of paraquat and hydrogen peroxide in embryonic shoot cells of *Hordeum vulgare* and root meristem cells of *Allium cepa* was tested against the genotoxicity of challenge doses of methyl mercuric chloride, maleic hydrazide or ethyl methane sulphonate (Patra *et al.* 1997). The results provide clearcut evidence that whereas $\text{H}_2\text{O}_2$ induced adaptive response for the chromosome damage caused by methyl mercuric chloride and maleic hydrazide, paraquat induced the same for methyl mercuric chloride and ethyl methane sulphonate but not for damaged caused by maleic hydrazide.

Dursban, an insecticide of the organophosphorus group of pesticides was studied for genotoxicity in plant cells (*Crepis capillaries*) with regard to its ability to induce structural chromosomal aberrations and micronuclei (Dimitrov and Gadeva 1997). It was observed that at
concentrations comparable to those applied in agricultural practices, dursban did not induce chromosomal aberrations as estimated by anaphase and metaphase analysis. Laggards were observed in the anaphase.

Chauhan et al. (1999) studied the cytogenetic effects of two commercially formulated alpha cyano pyrethroid insecticides viz. cypermethrin and fenvalerate in the root meristem cells of Allium cepa. Both the compounds significantly inhibited the mitotic index and induced chromosome and mitotic aberrations. The types of aberrations induced by both the compounds were quite similar and both the compounds induced high percentage of mitotic aberrations. Puig et al. (1989) also reported the genotoxic potential of these compounds in human lymphocytes and Agrawal et al. (1994) studied the genotoxicity of these chemicals in mouse bone marrow.

El-Shazly and El-Sheikh (2000) demonstrated the dose dependant effects of aflatoxin B2 treatments on the components of the mitotic cell cycle in Vicia faba root meristem cells. The most evident effect appears to be the accumulation of cells in the Go/G1 phase at the expense of other phases of cell cycle. These results indicate that this toxin acts as an inhibitor of cell cycle progression at the G1 transition point. The capacity
of aphlatoxin to induce clastogenic aberrations may be regarded as an indication of its genotoxic potential.

Rencuzogullari et al. (2001) investigated the cytogenetic effects of sodium metabisulphite, a food preservative in root tip cells of Allium cepa. Sodium meta bisulphate significantly reduce the mitotic index at all concentrations and treatments periods, it also increased the mitotic abnormalities dose dependently. Njagi and Gopalan (1982) also studied the cytogenetic effects of another food preservative sodium benzoate and sodium sulphite earlier and reported the formation of anaphase bridges, premature chromosme condensation heading to pycnotic nuclei and chromatin erosion in interphase nuclei in Vicia faba root meristems. The food preservatives sorbic acids and its potassium and sodium salts induce SCE’s CA’s and micronuclei in mice and also induce the frequency of 6-thioguanine resistant mutations in Chinese hamster cells (Abe and Sasaki 1977, Hasegawa et al. 1984, Mukherjee et al. 1988).

The genotoxic effects of carbosulfan, a carbamate pesticide were evaluated by using chromosome aberrations, bone marrow micronucleus and sperm abnormality assay in mice (Giri et al. 2002). All the three acute doses of carbosulfan induced significant dose dependant increase in the frequency of chromosomal aberrations micro nucleated polychromatic
erythrocyte and sperm head abnormalities. It suggested that carbosulfan is a potent genotoxic agent and may be regarded as a potential germ cell mutagen also. The findings of the present study was in line of the previous work have been done by Chauhan et al. 2000. Chauhan et al. 2000 also reported the induction of chromosome aberrations, micronucleus formation and sperm abnormalities in mouse following carbofuran exposure.

Yuzbasioglu (2003) was investigated the cytogenetic effects of fungicide afugan on the meristematic cells of *Allium cepa* in his study. He evaluated the value of EC 50 for afugan using root growth inhibition test and then the roots of *A. cepa* were treated with various concentrations for different periods and found that it significantly decrease the mitotic index and induce significant percentage of chromosomal aberrations in the root meristem cells of *A. cepa*. As a result he concluded that afugan has mitodepressive, clastogenic and turbogenic effects in *A. cepa* and should be regarded as a potential mutagen for non-target organisms.

Inceer et al. (2003) has been stated that copper chloride has harmful effects on the root tip cells of *Helianthus annus*. In addition to these the increase concentration of heavy metals in soil and water can lead to certain irreversible cytogenetic effects in plants and even in higher
organisms. They also reported the various chromosomal abnormalities due to the treatment with various concentrations of copper chloride. Histological studies of radish seedlings exposed to varying concentrations of cadmium chloride showed the structural alterations in roots, stems and leaves (Vitoria et al. 2003). In roots, the alterations in cambial cells with a loss of organization in the cambial region were observed. An increased number of root hairs were observed in the treated plants compared to control.

Chlorophyllin, a salt derivative of chlorophyll, has been demonstrated to be a potent agent against the deleterious action of different classes of mutagens. The toxic, cytotoxic and genotoxic effects of chlorophyllin were examine in Allium cepa. Chlorophyllin was not shown to have genotoxic effects and it acts as antigenotoxic agent at lower concentration. Since the antigenotoxic effect of chlorophyllin was maintained at higher concentrations. It can not be ruled out that this substance somehow interferes with cell metabolism, hindering effective recovery from mitomycin-C induced damage.

The cytotoxic effects of synthetic insecticide Malathion and leaf extract of Pongamia pinnata (L.) on seed germination of Vicia faba has been evaluated by the Chandra and Kumar (2004).
Effects of *Pongamia pinnata*, an indigenous plant used in Ayurvedic Medicine in India on the temporal variations of circulatory lipid peroxidation products and antioxidants in ammonium chloride-(AC)-induced hyperammonemic rats has been studied. These alterations clearly indicate that temporal redox status could be modulated by extract of *Pongamia pinnata* during hyperammonemic conditions, which may also play a crucial role in disease development.

Saxena *et al.* (2004) were tested the genotoxic effects of Diuron, a persistent substituted urea herbicide, in root meristem cells of *Allium sativum* and compare the sensitivity with *Allium cepa*. Microscopic analysis revealed significant and dose dependant induction of mitotic as well as chromosomal breaks. The frequency of mitotic aberrations was every time found much higher than that of chromosomal aberrations. Based on the data of valence charge densities on the atoms of the herbicide molecule and spectroscopic studies, a possible mechanism of interaction of diuron with DNA molecule for chromosomal aberrations has been proposed. The electron microscopic study of Chauhan *et al.* (1998) revealed that the treatment of diuron reduces the number of cortical microtubules in interphase cells and disturb the parallel array of microtubules at metaphase.
To validate the use of *Allium sativum* as a sensitive test model for genotoxicity, the cytogenetic effects of a commercially formulation of the pyrethroid insecticide, cypermethrin, were evaluated in the root meristem cells of *A. sativum*. Ultraviolet and Fourier transform infrared (FTIR) spectral measurements were also carried out to understand the interaction of cypermethrin with DNA. Cells analyzed immediately after the exposure had a significant, dose dependant inhibition of mitotic index and induction of mitotic and chromosomal aberrations. A bathochromic shifts observed in UV absorption spectra reveals that cypermethrin binds with DNA. Based on spectroscopic data and structural properties a possible mechanism has been proposed for the interaction of cypermethrin with DNA resulting in chromosomal aberrations. Surrales *et al.* (1995) previously reported the micronucleus induction property of five synthetic pyrethroid insecticides in whole blood and isolated human lymphocytes cultures.

Chandra and Kumar (2005) investigated cytotoxic effects of insecticide Malathion and extract of *Pongamia pinnata* L. Pierre on seed germination of *Vicia faba*.

Relative genotoxic effects of three synthetic pyrethroid insecticides cypermethrin, Alphamethrin and fenvalerate were evaluated by Rao *et al.*
(2005) in root meristems of *Allium cepa*. Cytological analysis revealed that the test compounds elicited varying degrees of cytotoxic turbogenic and clastogenic effects. In the ultimate analysis, cypermethrin and Alphamethrin have more turbogenic and weak clastogenic activity whereas fenvalerate have relatively strong clastogenic activity in vitro.

Yi *et al.* (2005) investigated the effects of sulphur di oxide hydrates exposure on cell cycle, sister chromatid exchange and micronuclei in barley (*Hordeum vulgare* L.). It induced the sister chromatid exchange and micronucleus in barley root cells with different effective concentrations and with different trends as treatment concentration increased. These remarkable effects in causing DNA damage and consequent chromosome damage suggest that SO$_2$ is genotoxic agent and its genotoxicity may influence the mitotic activity and plant growth under SO$_2$ stress. Cytogenetic damage of SO$_2$ and its hydrates in human lymphocytes and other mammalian cells has been reported earlier (Meng and Zhang 1992, Meng *et al.* 2002). Previous results indicated that SO$_2$ inhibited plant growth, caused visible foliar damage such as chlorosis and necrosis, influenced the activities of enzymes for scavenging reactive oxygen species in plant cells (Noji *et al.* 2001) and decreased photosynthesis.
Karabay and Öğuz (2005) examined the cytogenetic and genotoxic effects of the neonicotinoid insecticide imidacloprid and the organophosphate insecticide methamidophos, when administered alone or in combination. These insecticides were tested with the bone marrow chromosome aberration assay and micronucleus test in rats and by the bacterial mutation assay. All the concentrations of insecticides induced a dose-related increase in the micronucleus frequency. Dose-related increase in the number of revertants was observed with the two Salmonella strain.

Cytogenetic effects of dinocap were investigated in root tip cells of Allium cepa by Celik et al. (2005). Dinocap has also induced significant increase of chromosomal aberrations at metaphase stage in all concentrations and treatment periods. As a result of this it can be concluded that, dinocap has the possibility of mutagenicity to non-target organisms since it has exerted clastogenic as well as turbogenic effects in A. cepa similar to many other pesticides. They concluded that the concentration used in the field is very high and should be very harmful to many organisms. So, dinocap should be used very carefully.

Polyamines also reported as the inducer of the chromosomal abnormalities. It also induced binucleated cells and micronucleated cells
as well (Gomurgen et al. 2005). Polyamines are essential component of living organisms. Polyamines affected the duration of each mitotic stage. A strong reduction of mitotic index in the meristematic zone and inhibition in the elongation zone of maize has been reported by the Agazo et al. (1995) followed by the treatment of spermidine.

Chauhan and Gupta (2005) recommended that the coexistence of herbicides (diuron) and pyrethroid insecticide (deltamethrin) in plants synergies the toxicity inducing drastic ultrastructural alteration which are different from its independent effects. Present findings indicate that the coexistence of substituted urea herbicides and synthetic pyrethroid insecticide can cause irreversible damage in plants. The interaction of these two groups of chemicals seems to change the mode of action, resulting in the induction of typical ultrastructural alterations that are not induced when the cells are exposed to individual compound. Several studies, however have reported antagonistic effects of diuron on the toxicity of different herbicides and even metals (Geoffroy et al. 2002). Nicolaus et al. (1989) observed the antagonistic interaction between diuron and oxyfluorfen and emphasized that the presence of diuron decreases the production of H$_2$O$_2$. Though herbicides and insecticides are
generally not used simultaneously in the field, yet a possibility of their interaction can arise due to long residual life of urea herbicides.

The genotoxicity of herbicides Roundup (glyphosate), Stomp (pendimethalin) and Reglon (diquat) were compared in plant *Crepis capillaris* and mouse bone marrow test system using chromosomal aberrations and micronuclei (Dimitrov *et al.* 2006). Roundup did not induce chromosomal aberration or micronuclei in either test system. Reglon also did not induce chromosomal aberration in either test system; however it increased micronucleus frequency in both plant cells and mouse bone marrow polychromatic erythrocytes. The responses of the two test systems to stomp were quite different. Stomp did not induce chromosomal aberrations in plant cells, but increased their incidence in plant cells, but increased the incidence in mouse cells. Stomp increased the frequency of micronuclei in both test systems. The induction of micronuclei in plant cells may have been due to the spindle destroying effect of the herbicide, since all concentration of stomp produce C-mitosis. Results indicate that both similarities and differences for the genotoxicity of the herbicides roundup, stomp and reglon in plant cells and mouse bone marrow. It also indicates that the plant test system may be more susceptible to spindle poison effects of these agents.
Kumar and Rai (2007) make the investigation to evaluate the influence of cadmium and mercury concentrations on somatic cells as well as gametic cells of soyabean, since the most pronounced effect of heavy metals on plant development is growth inhibition, which is inseparably connected with cell division. Both the metals elicited similar type of chromosomal aberrations but the percentage of these abnormalities and total abnormalities induced differed between the two treatments. The result showed that a close colinearity existed between the concentration of metal treatment and percentage of chromosomal aberrations.

_Pisum sativum_ showed high sensitivity against cadmium (Fusconi et al. 2007). The reduction in root growth at lower concentration and the significant effect at higher concentration confirm the sensitivity. Fusconi _et al._ analyzed the effects of cadmium in root apices of _P. sativum_ with cell viability, cell proliferation, and microtubule pattern.

Fernandes _et al._ (2007) studied the mechanism of micronuclei formation in polyploidized cells of _Allium cepa_ after exposure of trifluralin herbicide, an agent that promotes a cellular damage due to its direct action on the microtubules. It showed the evidences that the exceeding genetic material of polyploidized cells tends to be eliminated from the
nucleus in the form of micronucleus. It was proved by the fact by the presence of various carrying micronucleus after exposition of the *Allium cepa* root tips tested with several concentration of trifluarlin herbicide.

Cytogenetic effects of commercially formulated herbicide atrazine were investigated on the somatic cells of *Allium cepa* and *Vicia faba* (Srivastava and Mishra; 2009). Significant inhibition of mitotic index and increase in the frequencies of chromosome aberrations were observed. Results of the study indicate that both the plant bioassay found to be sensitive indicators for the genotoxicity assessment as the outcome of majority of *in vivo* / *in vitro* mammalian test system.

Srivastava and Singh (2009) examined the effects of insecticide Profenophos on germination, early growth, meiotic behavior and chlorophyll mutation of barley (*Hordeum vulgare* L.).