DISCUSSION

Mutagenesis no doubt has been a convenient and adequately effective tool of induction of variability in crop plants. The variability so generated, is considerably large and quite helpful in selection procedure (Gregory, 1956 and Khan 1988). The study of biological damage in terms of lethality, seedling growth depression, survival at maturity, frequency of chimeric plants, pollen and seed sterility in $M_1$ generation chlorophyll and viable mutation frequency in $M_2$ generation are generally used to evaluate the mutagenic sensitivity of the biological system under study. The sensitivity of any biological system to a particular mutagenic treatment depends on various factors such as:

1) Chemical properties of the mutagen;
2) Properties of the biological system;
3) Temperature;
4) Duration of the treatment;
5) Hydrogen ion concentration;
6) Pre and post treatment condition.

In the present investigation, the sensitivity of plants to mutagenic agents in $M_1$, $M_2$ generations have been measured in terms of seed germination, seedling height, plant survival at maturity and pollen sterility
in the populations emerging from the treated seeds of all the three varieties of chillies.

During the mutagenic studies it was observed that EMS, MMS and SA bring about reduction in germination of seeds, seedling height, pollen fertility and survival at maturity. These reductions, with an exception of survival were found to be dose dependent. Earlier studies of Hsieh (1959), Sahu and Kumar (1978) and Chauhan (1982) have shown a linear-relationship between the dose of the mutagen applied and the parameters mentioned above.

Reduction in germination, seedling height, and growth was also reported in Gladiolus (Sax, 1955), in Saccharum (Vijaylaxami and Rao, 1960), in Allium cepa (Campos et al., 1960), in grapes (Shimatsuma, 1962; Dass and Mukherjee, 1968), in Chlorophytum tuberosum (Chowta, 1971), in Sorghum (Sreeramulu, 1972), in Brassica napus (Fowler and Stefansson, 1972), in Capsicum (Kamaluddin and Abraham, 1972), in Phaseolus vulgaris (Hussein and Disouki, 1976), in Pearl millet (Singh et al., 1978), in Turnera (Tarar and Dhyansagar, 1979 a; Choudhary and Dhyansagar, 1980). In legumes, the dose response in manifesting sensitivity to different mutagens as measured by germination, seedling
growth, survival, chromosomal aberrations and sterility was demonstrated by Gregory (1955), Blixt et al. (1960), Patil and Bora (1961), Sojodin (1962), Blixt (1964) and Santos (1965).

It has been observed that mutagens, besides reducing the percentage of seed germination also cause a remarkable delay in the initiation of seed germination. Delayed germination, as observed in the present investigation, has also been reported in other plants following mutagenic treatments (Pearson et al., 1975; Veleminsky et al., 1977; Rao, 1983). The germination delayed by more than two days in the lots treated with higher concentrations of mutagens may be attributed to inhibition of the mitotic proliferation in root and shoot meristems.

Seed germination in chilli (Capsicum annuum L.) decreased with the increasing concentrations of the mutagens but the extent of decrease in germination differed in different mutagenic treatments. Seed germination was found to be affected more adversely in different concentrations of sodium azide (SA) ranging from 0.01 to 0.04%. Sylvia and New Combe (1970), Rajput (1973), Dahiya (1973), Shakoor et al. (1978), Ganguli and Bhaduri (1980) and Subramanian (1980) observed a depressed germination after the mutagenic treatments in
mungbean where as Fuji and Matsumura (1958) found the germination percentage enhancing at certain dose levels. Sinha and Godward (1972) noticed stimulatory effect in germination percentage at the lowest dose level. Haringa (1964) showed that, in peas, germination capacity was greatly reduced after EMS treatment.

Several workers have attempted to explain the cause responsible for inhibition of seed germination. Griffiths and Johnson (1962) and Srivastava (1979) considered the reduction in germination percentage to be due to the weakening and disturbances of growth processes.

It has been observed that mutagens, besides reducing the percentage of seed germination, also cause a remarkable delay in the initiation of seed germination. The delayed germination as observed in the present investigations has also been reported in other plants following mutagenic treatments (Pearson et al., 1973; Veleminsky et al., 1977; Rao, 1983).

The survival of plants decreased appreciably in the mutagenic treatments in all the three varieties of Capsicum, however, there was no linear relationship between the concentrations of mutagens and the survival. Several workers have also reported the same relationship
in other plants. Swaminathan et al., 1962 while working with bread wheat, reported no change in survival percentage after EMS treatments, and so was done by Dubey (1973). However, a linear relationship between survival and the dose of mutagen was observed in rice (Siddiq and Swaminathan, 1968a, Michaelsen and Navaratna, 1968). Kalia (1984) observed significant reduction in survival percentage with different chemical mutagens.

The reduction in survival percentage of the treated population could be due to disturbed physiological and cytological processes or chromosome damage leading to mitotic arrest. Sojodin (1962) considered that the embryonal damage due to mutagen became apparent at lower stages of ontogenesis.

The seedlings raised from the treated seeds show a decreasing trend, from lower to higher concentrations of mutagen, in root and shoot length. Similar results were reported earlier in different crops plants such as barley (Konzok et al., 1961), wheat (Scarascias et al., 1961) and Brassica napus (Fowler and Stefansson, 1972). Scarascias et al., (1961) noted that rate of the seedling growth in wheat decreased with the increase in the dose of fast neutrons, thermal neutrons, X rays and in various concentrations of EMS, Roy et al., (1971)
observed inhibition of growth in *Cucumis sativus* after X-irradiation of seeds. Fowler and Stefansson (1972) observed in *Brassica napus* that EMS treatment markedly retarded the seedling growth. The extent of decrease in seedling height with increase in the concentration of mutagens was not uniform in the three varieties studied. Such a differential response has been reported earlier also (Bhatia, 1960; Kawai and Sato, 1965; Goud 1967; Reddy 1974; Khan, 1979; Khan and Siddiqui 1988).

Various workers have attempted to explain the phenomenon of the reduced seedling growth. Gray and Scholes (1951) and Lea (1955) suggested that it could be due to uneven damage to the meristematic cells as a consequence of genetic injury. The badly damaged cells would produce only a few cells progeny and growth will recur from those cells which are least damaged genetically. Thoday (1951) and Evans and Sparrow (1961) opine that the chromosomal damages and/or inhibition of cells division are the chief cause of reduced growth. Smith and Kersten (1942) reported that the decrease of seedling growth would be due to the destruction of auxin by ionizing irradiations.

Another group of workers believe that changes in auxin level, in a plant are basically responsible for the reduced growth. Halvey and Shoub (1965), Gupta and
Samata (1967), and Goud and Nayar (1968) demonstrated that seedling growth depression may be due to inhibition of auxin synthesis. Gunkel (1957) and Natrajan (1958) observed inhibition of mitosis in growth primordia and attributed it to auxin destruction. Skoog (1955), Haskins and Chapman (1956) and Cherry et al., (1962) suggested that changes occurring in the specific activity of several enzymes by treatments of different mutagens have an adverse effect on growth rate.

Palc and Howard (1956) have suggested that the possible interference of irradiation damage with the synthesis of new DNA may lead to inhibition of growth. Evans and Sparrow (1961) attributed the phenomenon to genetic loss due to chromosomal aberrations. According to Gunkel (1957) the possible influence of phytohormones and other physiological disturbances makes for stunted plant growth. Halvay and Shoub (1965) opined that the reduction may be due to inhibition of auxin synthesis. Gupta and Samata (1967) held that auxin had a rapid turnover rate in metabolically active tissues, and that the auxin biosynthesis is very sensitive to the ionizing irradiation. Mhaske (1971) has stated that the inhibition of growth at higher doses of γ-rays is due to physiological imbalances in plants caused by radiations. Tarar and Dhayansagar (1979a) attributed it to the reduction of auxin synthesis.
In the present investigation stunted growth has been noted to be a common feature at higher concentrations of EMS, MMS and SA. Whatever be the cause of the reduced seedling height, the facts remain that the chromosomes carrying various genes responsible for the life processes and expressions are one of the most sensitive element, and damage to any part of these vital and tiny element is bound to go a long way to bring about various physiological and metabolic disorders which in turn will bring about several morphological and growth abnormalities in the plant or plant organs. Generally the seedlings abnormalities like habit, branching, cotyledonary and vegetative leaves increased with the increasing concentrations of chemical mutagens.

Variations in the shape, size and number of the cotyledonary leaves as observed in chilli are the common effects of mutagens, confirming the results obtained in rice (Yamagata, 1966; Bose and Chowdhury, 1968), sorghum (Ramulu, 1970), maize (Chandrashekhar and Reddy, 1971), soybean (Lee and Halloran, 1975), lentil (Sharma and Kant, 1975), mungbean (Chaturvedi and Singh, 1981) and chilli (Khan, 1983).

The factors which are responsible for the induction of these cotyledonary abnormalities due to mutagenic agents are not well known. However, it was
known that anomaly in the proportion of growth hormones in treated materials may be responsible for these cotyledonary abnormalities (Napp-Zinn, 1955). According to Devreux and Mungnozza (1964) the general disturbance in metabolic pathway due to irradiation may be one of the important factors responsible for such abnormalities. Grover and Virk (1984) held a similar opinion for the occurrence of the chromosomal aberrations.

The presence of a single cotyledonary leaf in the some seedlings may be due either to cytochemical disturbances or to the acute chromosomal aberration leading to the death of leaf primordia or the embryonal cell responsible for leaf development. Extra cotyledonary leaf, on the other hand, may be due to formation and involvement of additional leaf primordia or the embryonal cell.

Development of abnormal leaves was among the most common abnormalities noticed in almost all treatments of EMS and MMS, and frequency of these increased according to the dose of the mutagens. Previously, the causes for these abnormalities were attributed to the mutagenic treatment to different crop plants such as *Brassica napus* (Fowler and Stefansson, 1972), *Vigna sinensis* (Ram Mohan, 1979; Sadashiva Reddy
et al., 1984). Excessive branching was attributed to the induced axillary buds (Johnson, 1936, 1948) and Gunkel and Sparrow (1954). The present study supports their views.

Factors responsible for the induction of leaf abnormalities are not very well known. Rao (1972) reported that the leaf abnormalities were due to several environmental factors such as fertility and availability of water and the degree of luxuriance during growth. Blixt (1972) stated that leaf aberrations seemed to be closely related to actual mutation process and the altered metabolism as a result of cellular damage.

In the present investigation, varying degree of pollen sterility has been observed in the different concentrations of the mutagen. Pollen fertility was very low in the first generation after mutagenic treatment but it gradually increased in subsequent generations in the present investigation. Varying degree of pollen sterility has been observed in different concentrations of the mutagen. The magnitude of sterility increased with the mutagen concentrations. Similar reports have been made earlier by Yamaguchi (1964), Majid (1969), Rajput (1973), Chandra et al., (1978), Khan (1979) and Ganguli and Bhaduri (1980). In the present investigation, the pollen sterility which was
proportional to concentrations, appears to be the result of cumulative effect of various meiotic abnormalities observed combined with some physiological and damage induced by chromosome breakage. The descending order of the amount of pollen sterility induced by the three mutagens was found to be EMS, MMS, SA. Fahmy and Fahmy (1957) demonstrated the high ability of alkylating agents to produce deficiencies of a cryptic nature. Das (1957) stressed that in addition to chromosomal aberrations, some genic and physiological changes might have been caused sterility.

In most cases, meiotic abnormalities are responsible for pollen sterility (Rana and Swaminathan 1964; Sinha and Godward, 1972; Ramanna; 1974), because meiosis is more prone to any conceivable type of disturbances (Darlington, 1937; Swanson, 1957). The percentage of pollen sterility was relatively much less in $M_2$ generation than $M_1$, indicating that some sort of recovery mechanism must be operating in the intervening period. Similar observations were made by Katiyar (1978) on Capsicum.

Pollen sterility caused by ionizing radiation has been reported by many investigators e.g. Patil and Bora (1961) in Arachis hypogea and Plantago with thermal neutrons and $Y$-rays by Dhanraj (1971) in Solanum
khasianum and by Singh and Roy (1971) in *Trigonella Foenum graceum*. Like wise chemical mutagens were found to induce pollen sterility (Moutschen-Dahmon, 1965; Sato and Gaul, 1967; Majid 1969; Tarar and Dhyansagar 1979b). Mikaelson *et al.* (1968) are of the opinion that EMS caused alkylation of DNA base will probably pair by chance with a wrong partner and give rise to so called transitions and this will result mutation.

In contrast to many reports (Lal, 1975, Sharma 1977; Sahu and Kumar, 1978; Khan 1979; Mallikarjunaradhya and Channabregowda, 1981) no appreciable change was noticed in the mean value for quantitative characters in *M*$_1$ generation. This was because in our study, macromutational variants were excluded from the assessment of means in *M*$_2$ and data were recorded on normal-looking plants only.

The chlorophyll mutation frequency is useful in assessing the potency of a mutagen. Hence, scoring of chlorophyll mutations has proved to be a much dependable index for evaluating the genetic effects of the mutagenic treatments. A comparison of chlorophyll mutations indicates that the frequency of chlorophyll mutations recorded in *M*$_2$ generation was concomitant with dose. Similar dose dependent increase in the chlorophyll mutations frequency was reported by Gaul (1964) in


In the present investigation revealed that EMS induced the highest frequency of chlorophyll mutants in all the three varieties of chilli. EMS induced chlorophyll mutations have been reported in peas and Lens esculenta (Wellensick, 1965 and Uhlik, 1972). Swaminathan et al., (1962) proposed that such a high frequency is due to the preferential action of EMS on chlorophyll development genes located near centromere. EMS is supposed to be specific to certain chromosommal regions (Goud, 1967) containing genes for chlorophyll
development and has been reported to induce high frequencies of chlorophyll mutations. (Natranjan and Upadhya 1964).

The frequency of chlorophyll mutations induced by SA much less as compared to EMS and MMS. The low chlorophyll deficient mutation frequency may be due to the inhibition of catalase and peroxidase and the increase in peroxide concentration in the cell (Kleinhofs et al., 1978). Nilan et al. (1973) observed that the greater effectiveness of azide in the acid form is probably due to better penetration of the cell membranes by the natural HN₃ molecules.

Blixt (1964), Heringa (1964), Wellensick (1965), Monti (1968) Siddiq and Swaminathan (1968a), Prasad (1972), Nerker (1977) and Sharma and Sharma (1979) studied the effectiveness and efficiency of various mutagens and concluded that alkylating agents are more effective and efficient in inducing mutations. Siddiq and Swaminathan, (1968b), Prasad (1972), Nerker (1977), Farook and Nizam (1978) reported that the lower doses of mutagens were more efficient and effective, compared to higher doses.

Mutagenic effectiveness is an index of the response of a genotype to the increasing dose of the mutagen. The order of mutagenic effectiveness as
determined on the basis of mutated plant progenies was MMS, SA and EMS. All the three chemical mutagens were found to be effective at lower concentrations. The decline in the mutagenic effectiveness recorded at higher doses shows that the increase in mutation rate was not proportional to the increase in the doses of various mutagens. Similar results were obtained by Gupta and Yashvir (1975) for *Setaria italic*, by Nerker (1977) for *Lathyrus sativus*, by Singh and Chaturvedi (1980) for *Vigna radiata* and Gukasyan and Akopyan (1974) in chilli.

The mutagenic efficiency indicates the extent of genetic damage recorded in $M_2$ generation in relation to the biological damage caused in $M_1$. As reported by Sharma (1977), Ramulu (1970), Nerker (1977) the mutagenic efficiency decreased with increasing dose of all the mutagens in our study also. The greater efficiency at lower doses is because the biological damage generally increased with the enhancement in the dose at a higher rate than the mutations yielded in $M_2$ at the same dose (Konzak *et al.*, 1965). This can be taken as an established fact for almost all situations.

The lowest efficiency was recorded in SA and the highest in EMS, MMS being intermediate. The results indicate that the efficiency calculated on the basis of
sterility was higher as compared with that based on seedling injury. This trend was obtained in all the three varieties of chilli. Gaul et al. (1962) reported the mutagenic efficiency to be related to sterility and further observed that the lower the dose, the higher the efficiency. This is because the lower dose may cause relatively less damage, enabling the organism to express the induced point mutations successfully. The sterility induced by chemical mutagens, more particularly, by alkylating agents was not found in many cases to be associated with chromosomal abnormalities (Bansal and Natrajan, 1965). Prasad (1968), observed that NMU induced minimum visible mitotic changes in Triticum durum, but resulted in a very high degree of sterility. It appears that gene mutations may be responsible for such sterility, although the cytologically undetectable cryptic structural changes may also contribute to some extent.

Efficiency of the mutagens differed in the three varieties. Chandola (1968) and Goud et al. (1970) reported that the genetic architecture of the organism is a potent factor in determining its response to mutagens. Varietal differences in relation to mutagenic efficiency have also been reported by Gupta and Yashvir (1975) in foxtail millet.
Besides chlorophyll mutations, several morphological mutants, exhibiting changes in their morphological features, were isolated on the screening of M\textsuperscript{2} populations. These mutants differ from control and also among themselves in heights, growth, and flowering habits. The frequency of morphological mutants differed in different mutagenic treatments and also among the varieties. The range of such mutants was relatively wide with EMS treatment followed by MMS and SA. The segregation pattern and breeding behaviour of these mutants were not tested in M\textsuperscript{3} generation. Various investigators suggested that such mutants might be either a result of pleiotropic effects of mutated genes or a cryptic chromosome deletion. Many such morphological mutants have been extensively studied in different crop plants such as *Lycopersicon* (Khankar, 1974), *Capsicum* (Subhash et al, 1981), *Cajanus cajan* (Chary and Bhalla, 1988) and *Phaseolus mungo* (Jana, 1962).

As a result of mutagenic treatment of seeds with chemical mutagens, the plants showed varying degree of meiotic irregularities like reduction in chiasma frequency, multivalents univalents, disturbed bivalent associations, stickiness laggards, unequal division, bridges, fragments and the formation of micronuclei at telophase II.
Generally the meiotic irregularities increased with an increase in the concentrations of chemical mutagens. The frequency of abnormalities induced by present series of treatments was mostly concentration dependent and was higher in meiosis I than in meiosis II.

The chiasma frequency following mutagenic treatments has been found to increase in some cases (Mather, 1934, Darlington and LaCour, 1953, while in others no change (Darlington and La Cour, 1953; Sybenga, 1960) or decrease (Ramulu, 1971 a; Al-Allaf and Godward 1979). Sadanandam and Subhash (1984); Nagalla and Nagalla (1977) in Capsicum. Prasad and Godward (1969) reported in Phalaris that there was no significant correlation of the distribution of chiasmata between bivalents of the nuclei, though a slight reduction in number of chiasmata per pollen mother cell was observed following X-irradiation of dry seeds.

Lawrence (1961) working with Lolium and Tradescantia concluded that the decrease in chiasma frequency following mutagenic treatments might possibly occur at two stages, viz., during DNA synthesis and sensitive period at/or slightly before the stages of chiasma formation. In the former case the decrease in the frequency of chiasmata may be due to disturbance in chromosome coiling, failure or restricted pairing at
pachytene and the delay in DNA synthesis, while in the latter it may be affecting the process leading to chiasma formation. The chemical mutagens also in general, caused a higher reduction in chiasma frequency. Jain and Basak (1965) working with Delphinium reported that the mutagen treatments induced univalent formation through cryptic structural changes in some of the chromosomes which restrict pairing and thus reduce chiasma formation. According to Al-Allaf and Godward (1979) cryptic structural changes such as minute delations or small inversions in some of the chromosome in the embryonic initials or initial which had survived the elimination process to take part in the formation of the inflorescence, may be responsible for the failure of these the segments to form chiasmata and hence reduce the frequency.

Multivalents formation by chemical mutagens as described here has also been reported by many workers. Heiner et al., (1960), Konzak et al.,(1961) and Sree Ramulu (1971 b) working with gamma rays and DES reported that the chemical mutagens produce chromosomal aberrations less frequently than radiations, Chaghtai and Hasan (1979) recorded multivalents with increasing dosage of mutagens of EMS, MES and MMS in Lens esculenta and suggested translocations may have been produced due
to terminal affinities of chromosomes. Jain and Raut (1966) working with tomato (Lycopersicon esculentum) and See Ramulu (1973) working with Sorghum, also obtained multivalents by chemical mutagens. Wolff (1960) found that restitution or rejoining of broken ends involves repair enzymes and series of chemical reactions.

Burton and Powell (1966) in Pennisetum typhoides observed that the frequency of multivalents induced by EMS was less than that of thermal neutron treatments. According to Akhund-Zade and Khvostova (1966) alkylating agents induced mainly isochromatid deletions which lead to the formation of chromatid bridges at anaphase. The bridges are broken and accentric fragments are lost which lead to the disturbance of the chromosome balance and ultimately to cell death.

Konzak et al., (1961) reported that the chromosome bridges are most likely due to inversions induced by chemicals. The aberrant behaviour of chromosomes which lag in anaphase I may be due to delayed terminalization or stickiness of the ends of the chromosomes. Mann (1977) suggested that Alkylating agents reacts with several nucleophilic centres in a cell, with DNA being the primary site of alkylation. The alkylated DNA separates and leave the DNA depurinated; as resulting in gaps in the DNA molecule. These gaps may
initiate some exchange processes at this stage, giving rise to various types of observed aberrations. Katiyar (1978) found the induced pollen sterility was dependent on and invariably higher than the concurrent meiotic abnormalities. Bridges were noticed in anaphase I, sometimes accompanied by lagging chromosomes. The lagging and unoriented chromosomes were mostly univalents; these resulted in the formation of micronuclei of different sizes and numbers varying from cell to cell. As a result of these meiotic irregularities the treated plants exhibited a high level of pollen sterility.

The occurrence of bridges at anaphase I, in the present material has also been observed by several workers and has been attributed to crossing over between relatively inverted segments (MC Clintock, 1931) or the reunion of chromatids during meiotic prophase (Rees and Thomson, 1955; Lewis and John, 1966; Newmann, 1966). Jones (1968) described contrasting modes of origin of bridges in simple terms, "One depends on crossing over between relatively inverted segments, the other depends on inverted crossing over between non-oriented segments". It is also possible that bridges may result due to stickiness of chromosome's ends (Carlson, 1954; Sawamura, 1965; Sudhakaran, 1971). Thomas (1961) observed that in some cells interstitial chiasmata in the translocated chromosomes failed to complete
terminalization and during anaphase this results in a bridge. Bose and Saha (1970) supporting Rees (1952) said that a single bridge without fragment could result from the failure of division of end genes brought about by nucleic acid upset. Bora et al. (1961) observed that absence of fragments from bridges may be due to smallness of fragments which disappeared in earlier divisions or originated from a chromatic portion.

The bridges have also been observed by Ghatneker (1964), Gostimskii and Hvostova (1965), Akhund-Zade (1968), Bose and Saha (1970), Nawar et al. (1971) Chowdhary et al. (1971), Bose and Maiti (1971), Sree Ramulu (1971 b) and Tarar and Dhyansagar (1980). Unsynchronized movement occurred significantly in mutagens treated population. Similar results of unsynchronization of meiotic stages were observed in Turnera ulmifolia (Tarar and Dhyansagar, 1980) treated with gamma rays and EMS. According to Tarar and Dhyansagar (1980) unsynchronized bivalents or laggards might be due to discrepancies in spindle formation.

Frequency of univalents was mostly dose dependent. Most of the univalents in the present investigation showed either precocious separation before the initiation of anaphase in other bivalents at equatorial plate or were seen as laggards at anaphase.
Thus it seems that the occurrence of univalents is the result of desynapsis. Such a phenomenon was also supported by Li et al. (1945) and Bozzini and Maitini (1971) in wheat. Krishnaswamy et al. (1941) in Pennisetum typhoides and Krishnaswamy and Meenakshi (1957) in Solanum. Krishnaswamy and Meenakshi (1957) studied the inheritance of desynaptic mutant and found it to be simple Mendelian recessives.

Some of the chromosomes pairs exhibited weak synopsis resulting in the formation of univalents. The occurrence of high degree of univalents and an increase in frequency of univalents at metaphase I, and their random distribution on spindle, non disjunction and the formation of micronuclei seem to be the outcome of some disturbances during pairing of homologous chromosomes.

Precocious separation of chromosomes increased significantly in all the mutagens. The precocious separation was also observed in Arachis hypogea (Patil and Bora, 1961) raised from X-ray irradiated seeds and in tomato (Bose and Saha, 1970) by X-rays and DES treatments. Bose and Saha concluded that the univalents separating precociously seemed to be as a result of desynapsis. According to Roy et al. (1971) precocious separation of bivalents at metaphase I in
Cucumis sativus was attributed to the failure of chiasma-formation in pairs. In Turnera ulmifolia var. angustifolia (Tarar and Dhyansagar, 1980) precocious separation was also observed by gamma rays and EMS treatments.

The stickiness of chromosomes due to chemical mutagens was recorded by Sato and Gaul (1967) in barley, and Tarar (1980) in Turnera ulmifolia. Darlington and LaCour (1945) observed this feature in irradiated plants of Allium and Trillium and suggested there was a reduction of correctly polymerized nucleic acid on the chromosomes producing characteristic errors of spiralization which combined with superimposed excess of nonpolymerized nucleic acid to cause surface stickiness. Kaufmann et al., emphasized (see Evans 1962) that radiation induced chromosomes stickiness was not due to the depolymerized acid, but due to partial diassociation of nucleo-proteins and alteration in their pattern of organization. In the present investigation the stickiness could be due to the disturbances in cytochemically balanced reactions by the effect of alkylating agents (Rao and Lakshmi 1980).

Occurrence of laggards in all the treatments as observed in the present investigation has also been observed by Rangaswami (1935), Ammini (1968), Bose and

At Anaphase I, instead of the expected 12:12 segregation of chromosomes, unequal disjunction of chromosomes was observed.

The cells showing unequal division may arise when one chromosome of a quadrivalent goes to one pole and the remaining three to the other. It is also possible that a bivalent may fail to disjoin and move as a whole to one of the poles. These possibilities seem to exist because in the present study the cells with unequal distribution of chromosomes were observed in those treated plants in which quadrivalents were not noticed. Unequal distribution was also observed in Pennisetum typhoides (Krishnaswamy and Rangaswami-Ayyangar, 1941) by X-ray, tomato (Bose and Saha, 1970) by DES and X-rays and Rhoeo discolor (Ammini, 1968) by Maleic hydrazide.

Stretching of chromosomes was also observed in almost all the treatments with the significant
Some abnormalities were observed at second meiotic stages also but frequency was significantly lower and sometimes negligible. More than 4 groups of chromosomes were also observed at telophase II, most probably due to disturbed spindle mechanism forming micronuclei. The micronuclei in the treated plants might also have resulted due to non-orientation of chromosomes, laggards, or chromosome fragments.

Some of the mutagens have given high rates of mutation as compared to others. For a number of gene loci or group of gene loci, EMS, MMS most effective. SA also exhibited consisted effect in most of the cases even in $M_2$.

Improvement of the cultivated plants largely depends on the extent of genetic variability available within the species. Variability observed in the existing germplasm collection owes its origin to such initial spontaneous mutations that were able to survive under specific environmental conditions and their subsequent recombinations. Mutagenesis has proved to be a handy tool to enhance the natural mutational rate and thereby enlarging the genetic variability and increasing the scope for obtaining the desired selections. Particularly, induction of micromutations in the
polygenic system, controlling the quantitative characters is important for crop improvement. The best use of micromutation breeding comes from the extensive work of Gregory (1965) and Gaul et al. (1969). A substantial improvement in yield following selection in irradiated populations was demonstrated by these workers. Some optimistic conclusions regarding the potential value of induced micro-mutations for crop improvement were drawn by other workers (Gustafsson, 1947; Rawlings et al., 1958; Koo, 1962; Griffiths and Johnson, 1962; Scossiroli, 1965; Brock, 1963; Goud, 1967; Swaminathan, 1969; Rajput, 1974; Kaul, 1980; Kaul and Kumar, 1983; Khan, 1986; and Venkatram and Subhash 1984.

The degree of success in the genetic improvement of particular traits in crop plants depends on the magnitude of the genetic parameters and the breeding methodology adopted. Since most of the economically important characters are influenced by environment.

In the present study, data on seven quantitative characters namely, days to flowering, plant height, days to maturity, number of fertile branches, number of fruits, 1000 seed weight, and total plant yield were analysed quantitatively to assess the extent of induced variability in M₂ and M₃ generations of the three varieties of chilli.
Opinions differ regarding the direction of mutations. Gaul (1965) and Aastveit (1966) hold that the induced polygenic mutations do not follow any particular direction but occur at random. According to Bateman (1959), Brock (1965) and Goud (1967), the polygenic mutation always follow a particular direction opposite to the previous history of selections. The characters selected previously for an intermediate mean would be expected to respond to the mutagenic treatment without subsequent selections with an increase in the variance but no change in the mean.

The variation observed for all the seven quantitative characters due to mutagenic treatments indicates the potential usefulness of mutation breeding for the improvement of this crop. The extent of variability produced by the three chemical mutagens differed in their action. Enlargement in range of variability for plant yield and its attributes such as number of fertile branches, number of fruits and also 1000 seed weight for the three varieties of chilli in M2 and M3 generation of the treated populations is indicative of the wider scope for selection. Although mean shifted on either side of the control mean, most of it went towards the positive side in the case of number of fertile branches, number of fruits, 1000 seed weight
and plant yield. During mutagenesis, it mutations occur at random for the quantitative characters, no significant change is expected in mean values. The above observations on yield components give us the idea that more positive mutations had occurred for these characters resulting in the shift in mean values in the positive direction.

The mean flowering time increased or decreased significantly after the mutagenic treatments. Flowering was early by 4 days with 0.2% of EMS treatment for the var. NRH in M₃ generation. The early flowering was also reported by Ramkanth et al. (1977). Bhattacharya and Bairagi (1982) and Khan (1984a). Kaul (1980a) suggested that the mutation of the two dominant genes to their recessive forms makes for an early flowering in peas. Oka et al. (1958) reported that the average number of days to flowering was not altered much in some of the treatments indicating that the mutations in major genes had been in both the directions i.e. for
earliness as well as for lateness. Sharma and Saini (1969) reported that the direction of mutation depends upon the genotype, character under the study and the dose applied. The decrease in mean values, is presumably due to the predominant incidence of micromutations for early flowering.

The adverse effect of the mutagens on plant height was clear in all the three varieties of chilli. The treatments of SA at various concentrations gave the maximum reduction in plant height in all the three varieties in both generations except the variety Jwala in the M₃ generation where it was slightly increased as compared to control.

The extent of reduction in growth is related to the mechanism of action of a given mutagen. As a respiratory inhibitor, azide may inhibit energy system resulting in the inhibition of mitosis which can be associated with seedling growth depression.

The reduction in the mean plant height was reported by Goud et al. (1969) in ragi, by Bajaj et al. (1970) in Phaseolus vulgaris, by Rajput (1974) in mungbean, Shah et al., in chilli.

The data obtained on days to maturity resulted in a significant gain in reducing the maturity period by
approximately 4-days with 0.2% EMS and 0.02 MMS treatments in M3 generation. The mean days to maturity was 80.50 and 79.87 in EMS and MMS treatments, respectively, whereas in control, it was 84.40 for the var. NRH. Similar gain of reducing days to maturity is notice with the treatment of 0.2% EMS in the var. Jwala, giving a mean value of 78.20 days in contrast to 82.67 days in control. The EMS treatments seems to be more effective in reducing the maturity period in chilli. Khan (1983) reported similar findings in mungbean.

The mean number of fertile branches and fruits per plant increased spontaneously in M2 and M3 generations resulting from the treatments with mutagens. It can be safely concluded that the number of fertile branches are correlated with the number of fruits per plant. The treatment of EMS with 0.2% concentration gave the mean number of fertile branches to the tone of 11.57 compared with 8.50 in the control. Also, the mean number of fruits per plant increased to 67.20, whereas the mean of the control was only 54.83. This was obtained in M3 generation of the var.5 Black. The var. Jwala with a similar treatment gave the highest mean values of 11.87 for the number of fertile branches and 77.33 for the number of fruits per plant in contrast to control which produced 7.10 as the mean for the branches and 58.67 as
the mean values for the fruits per plant. The var. NRH behaved somewhat differently.

The author is of the view that the increase in the number of fruits per plant in the present investigation is obviously due to an increase in the number of flowers. Flower shedding was not noticed in the three varieties studied. The period from the start of flower bud formation to the production of fruits took two weeks. As the fruits started forming, there was no fresh forming. It can also be said without any doubt that the number of fruit sets were higher in the lines which produced large number of flowers. All these three characters viz., number of fruits per plant, number of fertile branches and number of flowers seems to be highly correlated. An increased number of fruits was also reported in *Phaseolus vulgaris* (Mujeeb, 1970) *Cicer arietinum* (Bhatti et al., 1970), *Capsicum annuum* (Shah et al., 1986) and *Vigna radiata* (Dahiya, 1973; Rajput, 1974; Khan 1983).

The character of 1000 seed weight is a reliable source of measuring yielding ability is chilli. Contrary to the findings of many workers such as Ghafoor et al. (1968) in barley, Miah et al. (1971) and Jana and Roy (1971) in rice and Abidi and Haq. (1971) in *Brassica*
campestris and Khuspe and Ugale (1977) in Capsicum annuum. In the present investigations, 1000 seed weight has shown a very significant increase from the control with most of the treatments in the three varieties of chilli. The increase in the mean values is due to the predominant incidence of favourable mutations in the treated populations (Khan, 1986). This character has been reported to be governed by a relatively smaller number of genes, unlike other polygenic traits (Syakudo and Kobori, 1958; Ghose et al, 1960).

Earlier studies on mutation breeding in crop plants such as soyabean (Papa et al., 1961), oats (Griffiths and Johnson, 1962), wheat (Swaminathan, 1963; Scossiroli, 1964; Borojevic, 1969), barley (Gaul, 1963, 1967), rice (Jana and Roy, 1973; Chakrabarti, 1975) and Rama Rao et al., (1991) in chilli brought out a reduction in mean values for plant yield when compared to the control, although Tickoo and Jain (1973), Chaturvedi and Singh (1980) Khan (1986) found positive mean shifts in mungbean. The mean plant yield, in the present investigation, has shown significant positive shifts with almost all the mutagenic treatments in the three varieties.

The efficiency of the chemical mutagens could be
because the seeds treated with these mutagens are first soaked in water for 6 hours. The growth initials in the embryo in seed have already started their mitotic activity and may be in a better position to respond to the mutagenic effect.

In the present investigation, increase in the mean seed yield per plant may be due to the selection of the normal looking plants in M₂ generation which could lead to elimination of the aberrant plants, and also to the genetic nature of the changes induced after the mutagenic treatment.

The mean plant yield increased in M₂ generation except at higher concentrations of the mutagens, and in M₃ generation there was a complete positive trend in the mean values of the seed yield per plant in all the treatments given to the three varieties of chilli. A better performance for yield and yield components in M₃ generation may be because quite a number of aberrant plants are eliminated in M₂ generation itself. Oka et al. (1958); Kao et al. (1960) and Gaul (1964) have suggested that the selection process should be delayed until M₃ or later generations following the mutagenic treatments. Yoshida et al. (1969) reported that M₃ and M₄ selections are by no means inferior to M₂ selections. In
the present investigation, the performance of M₃ appears to be superior to M₂ generation.

The data reveal that mutation affecting the quantitative, character are not at random in all the varieties studied. This may be because all the three varieties had not reached its peak in performance and still retained the capacity for effectively utilizing mutagenic changes in the positive way for yield and its components. This may be connected with the history of selection of these varieties which consisted of only hybridization to the best of our knowledge.

In general, the lower concentrations of the mutagens used in the present investigation were found to be more effective and efficient in increasing mean values for yield and yield components, compared to the higher concentrations. The present investigation also revealed that mutagenesis could be employed to induce the quantitative changes in the genetic architecture. Thus, the induced genetic variability can effectively be exploited for the improvement of chilli.

**Induction of Polyploidy:**

In the present study the shoot treatment of *C. frutescens* L. var. suryamukhi has given encouraging results in the induction of autotetraploidy in contrast to complete failure reported by Pal *et al.* (1941).
As regards the morphological characters the polyploids exhibited characters like stunted growth, coarse and dark green leaves, such characters have also been reported in induced polyploids by many other workers (Pal and Ramanujam, 1939; Pal et al. 1941, Palfi et al. 1961, Murthy et al. 1968; Tapadar, 1963; Biswas and Bhattacharya 1971, 1972). The leaves and the floral parts were all larger than their diploids counterparts as indicated in Table 67... Pal et al. (1941) obtained fertile polyploids, with large fruits in the colchicine induced polyploids of Capsicum annuum L. but in the present investigation polyploids were completely sterile and there was no fruit setting; while large fruits were produced in the diploids which turned to attractive red when ripe. The polyploids were better in most of respects than the diploids, being more hardy. The pollen exhibits 58.38% of fertility in the polyploids and pollen grains of different size were met with thereby indicating different chromosome composition. The polyploids show univalents, multivalents, multiple spindles, multipolar spindles, unequal segregation and breakdown of spindle mechanism. On the whole these various meiotic anomalies all lead to the production of gametes with unbalanced number of chromosomes such gametes may be sterile. But this does not explain the complete sterility of polyploids, 58.38% of
pollen is fertile which is sufficient to bring about fertilization but style disintegrates before the bud opens.

Dobzhansky (1941) has classified sterility as genic and chromosomal. In genic sterility are included all types which are produced by failure of sex organs to develop up to the point where meiosis can take place or certain genically controlled anomalies of spindle behaviour and genically controlled asynapsis or desynapsis (Clarke 1940, Li, Pao and Li 1945). The chromosomal sterility on the other hand is marked by lack of homology between the chromosomes of the individual which results in higher percentage of univalent chromosomes. Thus sterility in polyploids is both chromosomal as well as genic- the genic-sterility causes disintegration of style before fertilization can be effected. The multiple spindles and abnormal behaviour of spindle mechanism may lead to production of sterile gametes this is also genic sterility. The failure of chromosomes to pair and other related anomalies are instances of chromosomal sterility.

Beasley (1940) reported almost complete sterility in the autotetraploids of *Gossypium herbaceum*. Einset (1944, 1947a) found that only a small part of high sterility in lettuce autotetraploids accounted for the abortion of pollen grains and ovules. The main cause was
the failure of pollen grains to complete growth down the style and the inhibition of fertilization.

The meiotic instability may be looked upon as a source of variation from an evolutionary point of view. Complement fractionation provides a method of decreasing the level of ploidy while somatic doubling and unreduced gametes tends to increase the ploidy level, thereby clearly indicating the reversible nature of polyploids.

In *Capsicum frutescens* gametes are produced which have a wide range of chromosome complements resulting from complement fractionation and non disjunction. Recombinations of the these gametes could yield progeny with different chromosome number. These can be perpetuated as distinct variations with different level of ploidy provided they prove to contain favourable gene combinations. Gametes with variable chromosome numbers can be produced by increasing the meiotic instability. Thus a reservoir of variability may be maintained and from it, depending on the requirement, various types of gametes having different gene combination can be obtained. The importance of plants having the capacity to produce such variable gametes from evolutionary point of view is clear and also they are of great value to plant breeders. The gametes in such cases provide a ready tool to produce new types suitable economically to meet varying requirements.
But unfortunately these plants have proved to be totally sterile and no success has been achieved till now to bring about fruit setting.