

**“Effect of Ethyl Methane Sulfonate Mutagen in M₁ Generation
in Pigeonpea (*Cajanas Cajan L.Millasp*)”**



THESIS

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By

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2011

CERTIFICATE – I

*This is to certify that the thesis entitled “Effect of Ethyl Methane Sulfonate Mutagen in M₁ Generation in Pigeonpea (Cajanas Cajan L.Millasp)” submitted in partial fulfillment of the requirement for the degree of Master of science (Plant Breeding and Genetics) of Rajmata Vijayaraje Scindia Krishi Vishwa Vidyalaya, Gwalior is a record of the bonafied research work carried out by **Mr. Ashwani Rai** ID No RA/SH/1211/2009 under my guidance and supervision. The subject of the thesis has been approved by the Student’s Advisory Committee and the Director of Instruction.*

No part of the thesis has been submitted for any degree or diploma (Certificate awarded etc.) or has been published / published part has been fully acknowledged. All the assistance and help received during the course of the investigation has been acknowledged by him.

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CHAPTER - I

INTRODUCTION

The Pigeonpea (*Cajanus cajan* L. Millasp) is an annual crop, cultivated for its sweet tasting legumes, which is consumed as a popular staple diet in Asian countries and from an important role in predominantly vegetarian diet.. The genus *Cajanus* belongs to tribe Phaseoleae, subtribe *Cajainae*, family Leguminosae and sub-family Papilionoideae . Pigeonpea is mainly used in the form of Dal, as a source of protein in Indian diet. The dry stalks are used for fuel and wind breaks. The dried leaves and pod shells are used as animal fodder. The husk of seed, after milling, are used in animal feed.

This plant is termed with various names in different cultures such as Red gram, Congo pea, Gungo pea, No eye pea etc and are widely consumed throughout the world. Pigeonpea originated in Asian continent, a native to India, for as long as 3000 years. From India, it was taken to the eastern African region approximately a thousand years ago. The legumes of the Pigeonpea plant are not actually peas but contribute to one of the most famous pulse in the world. It occupies an area of around 0.36 million hectare with annual production of 0.24 million tones in Madhya Pradesh

It is grown throughout the tropical, sub-tropical and warmer temperature regions of the world. It is an annual crop as it overlaps the Kharif and Rabi season. The temperature, photoperiod and sowing conditions highly influence the flowering period and seed filling of pigeonpea. The optimum sowing date of the crop in Madhya Pradesh is from last week of June to second week of July under normal sown condition.

Mutations play a vital role in any crop improvement programme particularly which naturally occurring genetic variability has exhausted. Mutation may produce spontaneously or it may be brought about either by using chemical mutagen or by physical mutagen. Both these mutagens are equally important but, particularly, chemical mutagenesis is better than physical as chemical mutagens are easy to apply and handle. It also does not require large number of specific equipments and conditions.

Ethyl Methane Sulfonate (EMS) is a mutagenic, teratogenic, and possibly carcinogenic organic compound with formula $C_3H_8O_3S$. It produces random mutations in genetic material by nucleotide substitution; particularly by guanine alkylation. This typically produces only point mutations. It can induce mutations at a rate of 5×10^{-4} to 5×10^{-2} per gene without substantial killing. The ethyl group of EMS reacts with guanine in DNA, forming the abnormal base O-6-ethylguanine. During DNA replication, DNA polymerases that catalyze the process frequently place thymine, instead of cytosine, opposite O-6-ethylguanine. Following subsequent rounds of replication, the original G:C base pair can become an A:T pair.

EMS is often used in genetics as a mutagen. Mutations induced by EMS can then be studied in genetic screens or other assays. The main objective of mutation breeding is to create genetic variability and thereby increase the scope of selection. Looking to its prospects of development of new changes in pigeonpea, the study is aimed with the following objective

- 1 To assess the response of pigeonpea genotype to various doses of mutagen (EMS) in field and laboratory conditions.
- 2 To find out the deviation from normality of morphological traits in M_1 generation.
- 3 To determine the efficacy of mutagen on seed vigor and morphological traits in pigeonpea.

CHAPTER - II

REVIEW OF LITRATURE

The literature on importance of treatment in creation of variability and different aspects of the present study has been reviewed, summarized and presented as under.

Chaudhari and Patil (1953) observed a true-breeding mutant in pigeonpea with a prostrate growth. The form arose spontaneously in the F₃ of a cross between two normal varieties. The mutant is likely to be useful as cover crop, and in soil conservation and strip cropping. Pandiya *et.al.*(1954) observed a mutant bearing obovate leaflets with rounded base and apex and a dwarf mutant bearing very small leaflets have been observed. Pathak and Singh (1964) observed spontaneous mutant plant found in an early maturing line, no. 5, received from Malwa. The mutant was bushy and taller than the parental type. Four distinct types of branches were observed, three of which were sterile.

Kajjari (1956) observed a mutant with obcordate leaves, keel petals united at the top and two or three apocarpous ovaries. It bred true in two subsequent years. No cytological differences between this mutant and normal plants were found. The chromosome number for both being 2n-22. Joglekar and Deshmukh (1958) observed mutant with simple leaves and another with ovate-oblong trifoliolate leaves, respectively designated var. unifoliata and var. oval oblong trifoliolate.

Deshmukh (1959) had reported a non-branching single-stem spontaneous mutant associated with female sterility. Patil (1959) found a dwarf mutant differing in leaf and floral morphology from either parent from crossing *C. cajan* x *C. obcordifolio* and a genetic study of it was undertaken. Caldecott (1961) observed cultivar R-60 which was the least and in NP (WF)-15 the most, radiosensitive. An attempt has also been made to establish the correlation between male sterility in the m₁ and frequency of chlorophyll mutations in the M₂.

Jeswani and Deshpande (1962) reported a broad leaved variant in CP 32546, which had a large number of flowers with the standard covering the whole flower. The pollen from the mutant plants were used for crossing with six local strains and hybrid seed could be secured.

Jeswani and Deshpande (1962) studied in a sepaloid mutant; simple leaves replaced the normal trifoliolate ones and were associated with a sepaloid condition of

the flowers. A second mutant had simple leaves on the part of the plant and none on the upper part. Rudimentary floral organs, dwarf habit, and thin, straggling branches; a cleistogamous mutant possessed thick, puckered trifoliate leaves. In all the mutants the abnormal condition was recessive to the normal.

Joshi and Ramanujan (1963a) reported a simple-leaf mutant that showed close association with sepaldoidy, pointing to close linkage or pleiotrophy. Joshi and Ramanujan (1963b) reported the non flowering condition, found in a collection of CP-32 from Madhya Pradesh, is monogenically, recessive to flowering and does not appear to be linked to the pleiotropic locus controlling trifoliate vs. simple leaf and normal vs. sepaldoid flower. The multicarpellate condition of the pistil, isolated in an arhar culture from Uttar Pradesh, is monogenically recessive to the normal unicarpellate condition. This allele also controls the development of supernumerary petals, the development of stamens into petal.

Abrams and Velez-Fortund (1961) reported irradiation with gamma rays exceeding 16,000 R (roentgens) or exposure to neutrons for more than 2 hours impaired the viability of pigeonpea (variety kaki) seeds and reduced plant height in the X_1 generation. The X_2 generation was considerably more variable than the parent variety with respect to plant height and time of flowering.

Bhatnagar *et.al.* (1967) observed mutant of *Cajanus cajan* having purple coloration and curved stems; the branches are fused with the main stem at the point of emergence. Some plants showed fasciations of stem and branches and some of the main stem only. Selfing fascinated plants and crosses with normal D-419-2 and T-163 showed fascination to be recessive.

Singh and Akhauri (1969) studied on dry dormant seeds of *C. cajan* treated with gamma rays at 5 and 10 KR and with EMS of 0.5% and 1% strength. Gamma-rays produced more marked effects than EMS in reducing percentage of germination, increasing time taken for germination and retarding seedling growth. Gamma rays also produced more breaks in chromosomes than EMS. Shoot growth suffered more than root with both mutagens. Various cytological abnormalities were observed in roots grown from treated seeds. There appeared to be similarities in the trend and pattern of abnormalities caused by both the mutagens.

Khan *et.al.* (1973) studied the seeds of red gram subjected to x-rays, ethyl methane sulfonate (EMS) and diethyl sulfonate (DES). Strain SA-1 seems to be

more tolerant to the mutagens than Co-1. DES beyond 0.25% killed the seeds completely. There were genetic differences by their sensitivity to the mutagens. Singh (1973) irradiated pigeonpea seeds with Cobalt 60 gamma ray with the strength of 8, 10, 20 and 30 kR. Radiation- induced changes in the length of shoot, number of branches, number of pods per plant, 1000-grain weight, and grain yield per plant, indicated that length of shoot was negative correlated, whereas other characters were positively, correlated up to radiation dose of 10 kR. There was significant increase in grain yield at 10 KR irradiation dose over control and other dosages.

Khan and Veeraswamy (1974) observed that genetic variability has been induced through mutagenesis in a large number of crops, but the information available in pigeon pea (*Cajanus cajan* [L.] millsp.) is meager. Rao (1974) induced variability in pigeonpea for raceme length, pod numbers, seeds per pod, seed yield and earliness when gamma rays were used on presoaked seed at 2.5 kR, 5.0 kR, and 7.5 kR.

Sharma and Shrivastav (1974) observed that irradiation tetraploid pigeonpea seeds of T-21 148 seedlings were grown of which 30 have been reverted to the diploid. The mutant flowered and matured earlier than the parent, displayed high pollen fertility, and was characterized by pods streaked green at the base.

Shrivastava (1975) observed normal diploid and colchicine-induced tetraploid seeds treated with five doses of gamma rays and studied for germination percentage, survival of seeding, plant height and number of leaves. In diploids, increasing doses from 15 to 60 kR, resulted in a decrease for all these characters. In the tetraploids, 15 kR resulted in increased germination, plant height and number of leaves compared with untreated seeds. Survival in all cases was higher. It was greater at all doses except 60 kR, when it was the same as in the diploids and the numbers of leaves in all cases were higher than in the diploids. Jain (1976) reported induced mutations in pigeonpea for pod number, pod size, seed size and number of fruiting branches in pigeonpea due to ionizing irradiations.

Mohamed Sheriff and Veeraswamy (1977) observed fifteen red gram mutants (13 from gamma irradiation and two from EMS treatments of the strain Co-1) in M₅ generation. There were significant differences for all the characters. The

genetic advance was high for pod weight, number of pods per plant and plant height. The mutants showed a positive shift in their mean values.

Mohamed Sheriff *et.al.* (1977) worked in mutation breeding research in red gram (*Cajanus cajan* (L.) millsp). It has resulted in the development of a high yielding mutant S-18 (CO-3) suitable for cultivation under both rainfed and irrigated conditions. Its duration was 130 days. On an average, it recorded 1300 kg/ha and 1200 kg/ha under irrigated and rain fed conditions, or 9.8 and 9.1 kg/ha/day respectively. A special advantage of CO-3 is its resistance to root rot and tolerance to wilt and pod borers.

Chaturvedi and Sharma (1978) reported six male sterile mutants in red gram after treatment with 0.2 per cent EMS. Venkateswarlu *et. al* (1976) studied that the three types of flowers (mono, bi and tri carpellary) are found on the same plant with concomitant increase in the stamens and complete pollen and ovule sterility. Venkateswarlu *et. al* (1978) reported chlorophyll mutation with gamma rays and EMS in pigeonpea.

Wanjari *et.al.* (1978) observed a broad-leaved variant isolated from the F₃ of T-21 x EC-107656 in 1976-77(has been designated as a compact mutant. Dahiya and Sidhu (1979) reported a non branching single-stem spontaneous mutant reported by seems to be distinct from the parental stock in some characters such as leaf size, maturity, and plant height. Irradiating pigeonpea with gamma rays beyond 30 kR resulted in more than 50 per cent reduction in germination and survival percentage (Mehetre *et al.* 1983). Natarajan *et al.* (1983) reported induced mutation in pigeonpea in the M₂ with gamma irradiation and diethylsulfate (DES) .The maximum variability was recorded for number of pods per plant followed by plant height. Rao and Reddy (1983 and 1986) reported induced polygenic variability in pigeonpea due to gamma rays; diethyl sulphate, ethyl methane sunphate, and hydrazide hydrate in two varieties of pigeonpea (ICP 7439 and ICP 2836). The genotypic variance, heritability and genetic advance showed marked increase in the mutation induced population. Pawar *et al.* (1984) observed that a high yielding and large-seeded line in pigeonpea (T 6) was developed by irradiating a small seeded (T 21) variety.

Natarajan *et. al.* (1985) reported in pigeonpea the effect of gamma rays and DES of germination, survival of seedlings, plant height, pollen fertility, and seed

number in M_1 generation. The germination of seeds and survival of seedlings were gradually reduced by an increase in dose of mutagens, but the reduction was more with gamma rays than DES. Rao *et. al.* (1986) in green gram observed increased variability in various quantitative traits in EMS treated population. Heritability estimates for yield were lower of the treated population than control.

Chary and Bhalla (1988) isolated male sterile mutant from pigeonpea (*Cajanus cajan*) in M_2 after treating with 0.2% methyl methane sulphonate. The leaves of the mutant were long and narrow while the stem was slender compared to normal plants. Pollen grains of mutant were shrunken and distinct colpi whereas those of normal plants were distinctly tricolpate. Lgnacimuthu (1988) stated that in M_1 generation of black gram Pollen sterility increased with increase in dose or concentration of gamma rays and EMS either singly or in combination.

Li *et.al.* (1988) observed in soybean seeds cv. Chung-Hsing No.2 treated with ethyl methanesulphonate (EMS), sodium azide (SA) or a combination. They reported that in EMS treatments, seed germination rates were not influenced at the G_1 stage. However, germination ability was decreased when seeds treated at the S stage.

Sarker and Sharma (1988) treated seeds of lentil either gamma -irradiated or treated with ethyl methanesulfonate (EMS), N ethyl N nitrosourea (NEU) or sodium azide (SA). M_2 and M_3 plants were evaluated for yield components together with controls. The frequency of promising M_2 families from different treatments was in the order NEU >EMS > SA > gamma rays.

Bhatnagar *et. al.* (1989) treated seeds of soybean cv. Bragg with EMS [ethyl methanesulfonate]. It exceeded the parent in germinability by 15% and was 5 days earlier in maturity. It gave high yields in both the rainy and post-rainy seasons. The mutant has smaller seeds than Bragg.

Iqbal *et. al.* (1989) used various concentrations of EMS (0.1%,0.2%,0.3% and 0.4%) in *V. mungo*, which decreased the percentage germination, root and shoot length and fresh and dry weight of seedlings. Khorgade (1989) studied individual and combined effects of gamma rays and EMS in chickpea and found later to be more effective than former. Bhatnagar *et. al.* (1990-91) irradiated Barodra Dthakri local and L-550 of Kabuli with EMS 0.05%, 0.1%, 0.2% and MMS (0.001% and 0.01%) either singly or in combination. The resultant mutant MV 77-1 obtained from the 30 kR+0.1% EMS treatment produced highest yield (18.2g.) even under infested field condition.

Singh *et. al.* (1989) obtained albino, xantha and viridis mutants in Lentil following treatment of dry seeds with the mutagens. The 0.2% methyl methane sulphonate treatment gave a higher mutation frequency under higher concentrations. Tripathy (1990) treated seeds of mungbean var. Dhauri and Khurda Local subjected to 0.2, 0.4 and 0.6% ethyl methanesulfonate (EMS) in combination with gamma radiation (5, 10, 15, 20 and 30 kR). Combination treatments of low doses of EMS and gamma radiation are recommended for inducing mutations in yield components. Singh and Raghuvanshi (1991) treated dry seeds of black gram (*Vigna mungo*) cultivar T₉ were irradiated with a gamma ray source, followed by EMS [ethyl methanesulfonate] treatment. A bold seeded mutant was selected in the M₂ and subsequently evaluated in the M₃. It showed more vigorous growth and produced more leaves and pods, larger seeds and almost double the yield (16.8 vs. 9.0 g) compared with the control.

Singh and Yadav (1991) treated dry seeds of green gram cv. T44 and were exposed to gamma rays (5-40 kR), ethyl methane sulfonate (EMS, 0.01-0.05 M), combination treatments (5-40 kR gamma ray doses followed by 0.02M EMS) and recurrent doses of gamma rays (5-40 kR with further doses in the following generation). M₂ mutations were observed for the characters plant height, plant habit, branching pattern, leaf morphology, venation, pigmentation, peduncle length, pod characters, male sterility, maturity, seed colour and yield. Distinct mutants were advanced to the M₃ generation and data for certain quantitative characters, such as plant height, days to flowering, pods/plant and seed yield/plant, were recorded to assess their usefulness. The mutants VR Mra-13, VR Hy-30, VR Sh-16 and VR Bu-3 showed significant increases in pods and seed yield/plant. Tyagi and Gupta (1991) irradiated seeds of lentil (*Lens culinaris*) with gamma rays or treated with ethyl methane sulphonate (EMS). The 7 mutations obtained were stunted, dwarf and bushy dwarf habit, open flower, partially sterile, completely sterile and large leaved. Ignacimuthu and Babu (1992) studied EMS and gamma treated seeds of *Vigna sublobata*. Results showed high genetic variation, heritability and genetic advance for most of the yield contributing traits.

Kandagra and Shukla (1993) exposed seeds of *Vigna radiata* cv. K-851 to 15 kR and 20 kR doses of X-rays radiation and/or treated with ethyl methane sulphonate and diethyl sulfate. Variation for pod length seeds/pod and 100-seed weight was studied in M₁ and M₂ generations. All treatment had a deleterious effect

on pod length. Kansagra and Shukla (1993) exposed seeds of *Vigna radiata* cv. K-851 to 15 and 20 kR dose of X-rays radiation and/or treated with ethyl methane sulphonate and diethyl sulfate. Variation for pod length, seeds/pod and 100-seed weight was studied in M₁ and M₂ generations. All treatments had a deleterious effect on pod length.

Rao and Reddy (1993) treated seeds of *Cajanus cajan* varieties T21, ICP7409 and ICP283 with 5, 10, 20, 30 and 40 kR gamma rays at 438 rad/min or chemical mutagens (0.05, 0.1 and 0.15% DES [diethyl sulfate]; 0.1, 0.2, 0.3 and 0.4% EMS; and 0.02, 0.04, 0.06 and 0.08% HZ [hydrazine hydrate] for 3, 4 and 3 h, respectively). Var. T21 showed the highest sensitivity to mutagenic treatment and ICP 7409 the least. A 30 kR gamma ray dose induced the highest mutation frequency, whereas low concentrations of chemical mutagens were the most effective.

Singh *et al.* (1993) treated seeds of mung bean with EMS and observed that mutagens at higher concentration and in combination with gamma rays were effective in modifying the seedling traits. Thakur and Sethi (1993) treated seeds of cv. Mash Kullu 1 with 5, 10, 15, 20, 25 and 30 kR gamma rays or 8, 10, 15, 20 and 25 mM (milli molar) EMS and observed that EMS induced a higher mutation frequency than gamma rays.

Patil and Guha (1993-1994) obtained three chickpea (*Cicer arietinum*) mutants (ACM 6, ACM 7 and ACM 18) from radiation with 10 kR gamma rays and treatment with .05 percent EMS (ethyl methane sulphonate). They reported that ACM 7 had 18.3% higher seed yield/plant than Warangal

Datta *et al.* (1994) observed relationship between biological effects in the M₁ and the sensitivity of two *Vigna radiata* cultivars to ethyl methanesulfonate (EMS). They investigated that mutagen treatment reduced germination, seedling height, mitotic index and survival to maturity. Kharakwal (1996) reported albina, chlorina and xantha chlorophyll mutations by treating chickpea with EMS of 0.1% and 0.2% concentrations.

Pandey *et al.* (1996) reported induced male sterility in long-duration pigeonpea [*Cajanus cajan*] by chemical mutation. The varieties, Bahar, DA11 and Pusa 9 were treated with streptomycin sulfate (SS) for 24 h and sodium azide (SA)

for 48 h. Four concentrations (0.1, 0.5, 1.0 and 1.5 M) of SS and SA (0.025, 0.05, 0.10 and 0.125%) were used for seed treatment, For both chemicals, germination percentage decreased with an increase in mutagenic dose.

Patil *et al.* (1996) obtained three chickpea (*Cicer arietinum*) mutants (ACM-6, ACM-7 and ACM-18) from irradiation with 10kR gamma rays and treatment with 0.05% EMS (ethyl methane sulphonate). They reported that ACM-7 had 18.3% higher seed yield/plant than warangal. Srivastava and Singh (1996) irradiated seed of pigeonpea (*Cajanus cajan*) with gamma rays or treated with EMS and MNH (N-methyl-N-nitrosurea). The dose of 0.2% EMS and 0.3% EMS had significant increase in total grain yield due to an increased numbers of pods per plant. The highest rate of high yielding mutants was obtained from 0.3% EMS treated in Pusa-85 and Pusa-60. Vanniarajan *et.al.*(1996) seeds of 2 *Vigna mungo* cultivars, ADT3 and Vamban 1, were gamma -irradiated at 20, 30, 40, 50, 60, 70, 80, 90 and 10 kR or soaked in 10, 20, 30, 40, 50, 60 and 70 mM ethyl methanesulfonate (EMS) for 6 hour. The highest level of variability after gamma-irradiation was shown by pod length and by pods/plant after EMS treatment.

Nanda *et al.*(1997) observed ethyl methane sulfonate (EMS) induced mutation resulted in decreases in survival, plant height, days to 50% flowering and days to first picking in the M₁ generation of *V. sesquipedalis* [*V. unguiculata* subsp. *sesquipedalis*]. Number of branches/plant, pods/plant, pod size, pod weight and yield/plant increased after mutation. In the M₂ generation, increased chlorophyll and foliar mutations occurred after high doses of EMS

Singh *et al.* (1997) studied on dry seeds of mung bean cv. PS16 were treated with various doses of gamma rays (20, 30 and 40 kR),ethyl methane sulfonate (EMS, 0.05-0.3%) and epichlorohydrin (ECH, 0.4%), and data on seed germination, seedling survival, plant fertility, mitotic index and seedling vigour were recorded in the M₁ generation. There was a linear relationship between doses of these mutagens and decrease in all these parameters.

Gautam and Mittal (1998) induced mutations in black gram (*Vigna mungo*) cv. T9 following seed treatment with gamma -rays (5, 10, 20, 30 and 40 kR), ethyl methane sulfonate (EMS; 10, 20, 30, 40 and 50 mM) and their combinations (EMS at 20 mM only) Few mutants recorded significantly higher grain yield as compared to the normal wild-type parent.

Waghmare and Mehra (1998) in a cultivar of *L. sativus*, (P 27) treated with gamma rays and ethyl methanesulfonate (EMS) and observed that EMS treatments produced a higher frequency of chimeras than the gamma ray treatments. Higher mutagenic doses proved to be more lethal

Wongpiyasatid *et al.* (1998) with a view to improving yield and resistance to *Cercospora* leaf spot and powdery mildew [*Erysiphe polygoni*] in mung bean (*Vigna radiata*), seeds of KPS1 and CN36 irradiated with gamma rays (doses of 500 Gy) or soaked in a 1% ethyl methanesulfonate (EMS) solution for 4 hour and observed that five promising mutants were derived from KPS1 via gamma irradiation: M5-8 with good pod setting; M5-19 with pods protruding up above the canopy; M5-21 and M5-22 with good plant type and high pod setting; and M5-24 with early flowering.

Sharma and Talukdar (1999) treated seeds of green gram (*Vigna radiata*) genotypes AAU34 and AAU39 were treated with variable doses of ethyl methanesulfonate (EMS) and gamma rays and observed that a tetrafoliate macromutant detected in the M₂ generation of AAU39 after treatment with 0.1% EMS + 10 kR gamma rays was also associated with higher yield performance. Tickoo and Chandra (1999) treated two varieties of mung bean with ethyl methanesulfonate (EMS) (0.1 and 0.2%) and observed that mean values showed a negative shift for most of the characters in the M₂.

Singh *et al.* (2000) in seeds of urd bean cultivars PDU1 and T9 were exposed to gamma radiation (10-40 kR), ethyl methanesulfonate (EMS; 0.01-0.04 M) or a combination of gamma radiation with 0.02 M EMS..They observed that positive and negative shifts in the mean were observed for all quantitative traits in both cultivars.

Singh *et al.* (2001) isolated superior mutant lines of urd improved yield and quality attributes. Urd bean cultivars Viz, PDU-land T9 were treated with gamma radiation, EMS, sodium azide, gamma radiation + EMS and gamma radiation + sodium azide. The variation in quantitative characters of induced mutant lines revealed that the values of GCV were moderate for days to flowering number of pods/plant. Low genetic variability was recorded for pod length, number of seeds/pod and 100seed weight.

Singh and Singh (2001) conducted study to isolate superior mutant lines of urd bean with improved yield and/or quality attributes. Urd bean cultivars PDU1 and T9 were treated with gamma radiation, ethyl methanesulfonate (EMS), sodium azide, gamma radiation + EMS, and gamma radiation + sodium azide. The variation in

quantitative characters of the induced mutant lines revealed that the values of genotypic coefficient of variation were moderate for days to flowering, number of pods/plant, plant height, and seed yield/plant.

Dhole *et al.* (2003) studied two soybean cultivars, JS-80-21 and Bragg treated with 0.2 and 0.4% ethyl methanesulfonate (EMS) Data were recorded for germination, mortality, pollen sterility, days to flowering, plant height, number of branches, number of pods, 100-seed weight, days to maturity and seed yield.

Kharkwal (2003) studied population of desi and kabuli chickpea mutanized through EMS 0.1% and 0.2% for 2 and 4 hours respectively. He evaluated M₃ generation for induction magnitude and directional changes, which clearly showed that mutagenic treatments succeeded in generating more favorable association among various components of yield.

Kumar *et al.* (2003) observed the effects of gamma radiation (20, 40, 60, 82 and 100 kR) and ethyl methanesulfonate (EMS; 10, 20, 30, 40 and 50 mM) on the germination, growth and survival of *P. lunatus* cv. LBS-1. Germination, survival and the frequency of chlorophyll mutants decreased, whereas the frequency of viable mutants increased with increasing rates of gamma radiation and EMS.

Gaikwad and Kothekar (2004) worked in some 250 dry and healthy seeds of uniform sizes of the lentil cultivars L-4611 and L-4639 were subjected to ethyl methanesulfonate (EMS, 0.05, 0.10 and 0.15%) for 4 h and sodium azide (SA, 0.01, 0.02 and 0.03%) for 6 hour. EMS and SA treatments dose-dependently decreased all biological parameters like germination, seedling height, pod fertility and plant survival in the M₁ generation.

Patil *et al.* (2004) studied in the Indian soybean cultivar, MACS-450, which has semi-determinate growth habit, medium maturity and high yield potential Observations on percentage germination, percentage leaf abnormalities, percentage survival, days to maturity, plant height, pods per plant, seed weight per plant and yield kg/ha were recorded in the M₁ generation. These characters were reduced as compare to control. Likewise, percentage reduction in various quantitative characters through various treatments was also recorded.

Samiullah and Wani (2004) in an experiment studied the comparative effects of two alkylating agents, ethyl methanesulfonate (EMS) and methyl methanesulfonate (MMS), on seed germination and seedling growth of mung bean (*V. radiata* cv. PDMA39). Both EMS and MMS brought about concentration-

dependent reduction in seed germination and seedling growth. The inhibition in seed germination ranged from 7.36 to 22.10% in EMS treatments, while it ranged from 12.63 to 26.31% in MMS treatments. The highest percentage injury in seedling height was recorded with 0.04% EMS (25.21%) and 0.04% MMS treatments (27.77%).

Wani (2004) studied that the effect of ethyl methane sulfonate (EMS) on the seed germination and pollen fertility of mung bean cultivars PS-10 and PS-16. Seeds were pre-soaked in distilled water for 9 hour prior to treatment with EMS at 0.01, 0.02, 0.03 and 0.04% for 6 hour. Seeds were either used to test for germination percentage or sown in the field for evaluation of pollen fertility. Seed germination and pollen fertility linearly decreased with increasing concentrations of EMS.

Khan and Wani (2005) observed in Seeds of mung bean cultivars Pant Moong-1, PS-10 and T-44 for 9 hour prior to treatment with 0.2% ethyl methanesulfonate (EMS), 0.02% methyl methanesulfonate (MMS) and 0.02% sodium azide (SA) for 6 hour. All chemical mutagens significantly decreased seed germination, pollen fertility and seedling height in populations emerging from treated seeds. The extent of decrease was not uniform in the cultivars studied.

Singh and Kole (2005) laboratory experiment was conducted to determine the effect of ethyl methane sulphonate (EMS) treatment on the germination frequency and seedling parameters (plumule length, radical length and percent of normal seedlings) and the detection of the lethal dose in mung bean cv. Pusa-9072. Presoaked (11 hour) Pusa-9072 seeds were treated with 0.1, 0.25, 0.5, 0.75 and 1.0 per cent EMS for 4 hour under dark. All the germination and seedling parameters were adversely affected due to the mutagenic treatment. Severe reduction in germination, frequency of normal seedlings, reduction in plumule and radical lengths and physiological injuries of radicals indicated effective mutagenesis.

Barshile *et al.* (2006) selected two chickpea cultivars (Vijay and Vishwas) used for the study of varietal differences in mutagenic sensitivity. The seeds were treated with three concentrations of sodium azide (SA; 2, 3 and 4 mM), ethyl methanesulfonate (EMS; 8, 12 and 16 mM) and three doses of gamma radiations (400, 500 and 600 Gy). There were significant decreases in germination and survival at maturity while seedling injury and pollen sterility increases with increases mutagen concentration.

Das *et al.* (2006) observed the efficiency of 15, 30, 45, 60 kR gamma radiation; 0.15, 0.30, 0.45 and 0.60% ethyl methanesulfonate (EMS) and 0.005, 0.010, 0.015 and 0.020 nitrosoguanidine (NG) for inducing mutations in green gram cv. Pusa Vishal. It was determined that germination, survival and plant growth were adversely affected by the mutagens, with the effects being more pronounced with increasing concentrations of the mutagens genetic variations in number of pods per plant, 100-seed weight and crop yield were induced by the mutagens, indicating the occurrence of micro-mutations.

Singh and Kole (2006) observed in seeds of mung bean cv. PUSA 9072 treated with ethyl methanesulfonate at 0.1, 0.25, 0.50, 0.75 and 1.0% for 4 hour under dark condition, field-grown mutagenized population with regard to 14 agronomic traits (days to germination, emergence of trifoliolate leaves, first flowering, 50% flowering, first pod setting, 50% pod setting, first maturity, plant height, number of primary branches, number of clusters per branch and per plant, number of pods per plant, pod length and number of seeds per pod) showed evidence for induction of variability in the M1 generation.

Singh *et al.* (2006) reported in pure and healthy seeds of lentil cv. K 75 were treated with ethyl methane sulfonate (at 0.02, 0.03, 0.04 and 0.05 M) and gamma-rays at different doses (10, 15, 20, 25 and 30 kR), alone or in combination. High doses of mutagenic treatments showed high reduction in grain yield per plant in combination treatments

Singh *et al.* (2007) mutagenic studies in lentil showed that higher doses of mutagens (gamma rays and EMS) either alone or in combination Frequency of viable mutations was high in EMS at lower doses. EMS 0.02 M was found more efficient to induce chlorophyll mutations than other mutagens.

Barshile and Apparao (2007) in two popular chickpea cultivars, Vijay and Vishwas, employing three well known mutagens, sodium azide (SA), ethyl methane sulphonate (EMS) and gamma radiation (GR) and observed that the 2 mM concentration of SA promoted plant spread in both the varieties, seed yield/plant in Vijay and 100-seed weight in Vishwas. Higher concentrations of SA and EMS as well as gamma radiation caused reduction in these traits. Further he reported that high heritability coupled with high genetic advance observed for traits such as plant height, number of pods per plant and number of seeds per plant, which offer wide scope for selection in breeding program.

Singh *et al.* (2007) in their mutagenic studies in lentil showed that higher doses of mutagens (gamma rays and EMS) either alone or in combination caused significant reduction in germination, seedling height, pollen fertility and plant survival as compared to their lower doses. Singh *et al.* (2007) in their mutagenic studies in lentil showed that higher doses of mutagens (gamma rays and EMS) either alone or in combination caused significant reduction in germination, seedling height, pollen fertility and plant survival as compared to their lower doses.

Singh and Singh (2007) taking two different cultivars of mung bean, namely T 44 and PDM 54 subjected to four doses each of gamma rays (10, 20, 30 and 40 kR) and ethyl methane sulphonate (0.01, 0.02, 0.03 and 0.04 M) as well as each dose of gamma rays with 0.02 M EMS in combinations. Besides pollen and ovule sterility, reduction in germination, seedling height and plant survival was recorded in mutagenic treatments.

Kumar *et al.* (2009) observed that among the spectrum of chlorophyll mutations consisted of albina, chlorina, viridish and xantha of these chlorophyll mutations, xantha was predominant in both the mutagenic treatments. The spectrum and frequency of chlorophyll mutations increases with increase in dose and concentrations of the mutagen EMS produced the highest frequency of mutations followed by gamma rays or their combination.

Okubara *et al.* (2009) studied that scarlet Rzl was derived from the allohexaploid wheat cultivar scarlet using EMS mutagenesis tolerant seedling displayed substantial root and shoot growth after 14 days in the 100-400 propagules per gram soil of *R. solani* AG-8 and *R. oryzae* in greenhouse assays. Senapati and Mishra (2010) studied on forty-five micro mutant line of black gram variety. Basant bahar (PDU-1) were developed by induced mutagenesis of gamma-rays, EMS, NG and MH singly and combination with gamma-rays. The characters like 100 seed weight, days to flowering and pod per plant were major contributors to genetic divergence. The magnitude of PCV, GCV, heritability and expected genetic advance exhibited high in different traits would be effective in isolation of different lines with improvement in these traits.

Senapati and Mishra (2010) studied on forty-five micro mutant line of black gram variety Basant bahar (PDU-1) were developed by induced mutagenesis of gamma-rays, EMS, NG and MH singly and combination with gamma-rays. The

characters 100 seed weight, days to flowering and pod per plant were major contributors to genetic divergence. the magnitude of PCV,GCV, heritability and expected genetic advance exhibited high in different traits would be effective in isolation of different lines with improvement in these traits.

CHAPTER III

MATERIALS AND METHODS

The present investigation was carried out during the Kharif season of at 2010-11 in the research field of RAK College of Agriculture, Sehore (M.P.). The experimental material used and the method applied during the course of present investigation has been described below.

3.1. Experimental site:

The experiment was conducted in the experimental area of All India Coordinated Research Project on Pigeonpea (AICRP) at RAK College of Agriculture, Sehore; M.P. the field was fairly uniform with gentle slope, adequate drainage and normal fertility status.

3.2. Experimental soil:

The soil of the field is clay loam Vertisol with 52% clay, 41.3% silt and 6.6% sand with pH ranging from 7.2 to 7.8. The soil was low in available nitrogen, medium in available phosphorus and high in available potassium.

3.3. Climate and season:

Sehore is located at the latitude of 23.12° N, longitude of 77.05° E and an altitude of 498.7 meter above the mean sea level. It lies in the western track of Vindhyan plateau agro climatic zone of Madhya Pradesh and enjoy sub tropical climate. The annual rainfall varies from 1000-1250mm with major precipitation in the month of July and August.

The weekly meteorological observations recorded during crop season (July 2010 to January 2011) have been presented in Table 3.1. The range of minimum and maximum temperature recorded was between 15.5°C and 37°C, respectively. Relative humidity varied from 64% to 100 % during the crop period. (Table 3.1)

Table 3.1: Meteorological data during the crop season (July, 2010 to January, 2011)

Standard Week No.	Temperature °C		RH (%)	Rainfall mm
	Max.	Min.		
28	32.18	24.54	88.14	18.00
29	32.01	24.66	93.71	59.00
30	28.66	23.78	94.14	21.00
31	28.66	23.36	96.28	62.00
32	27.86	23.40	96.86	08.50
33	29.40	23.34	98.14	123.00
34	29.88	29.10	93.71	36.00
35	29.70	23.56	98.42	73.00
36	28.08	23.35	98.71	48.00
37	29.45	22.58	66.14	29.50
38	29.66	20.67	53.71	03.50
39	28.33	17.80	93.75	00.00
40	30.80	18.14	86.71	00.00
41	32.98	19.07	81.00	00.00
42	27.12	21.51	83.73	09.00
43	31.50	19.60	86.70	11.00
44	30.10	15.10	76.60	00.00
45	30.10	16.30	81.30	00.00
46	29.50	19.70	98.40	14.00
47	26.50	17.50	90.80	03.00
48	27.00	17.40	96.60	04.50
49	22.40	07.80	91.70	05.00
50	23.10	04.80	88.70	00.00
51	23.30	10.50	84.40	00.00
52	28.80	12.00	91.40	00.00
01	20.30	04.60	81.40	00.00

Source: AAS, RAK College of Agriculture, Sehore, (M.P.).

3.4. Experimental material:

The experimental material under study consisted of 2 genotypes. JA 4 and ICPL 87 .JA 4 is medium (175days) and *ICPL 87* (135days) is early maturing variety. JA 4 is a stable performing variety released from Sehore center like ICPL 87 is a determinate type early duration variety released from ICRISAT.

The material was sown in single (environment) conditions i.e. rainfed in a randomized block design with 3 replications in field and 3 replication in laboratory. Each plots consisted of 4rows of 5 meter row length. Row to row distance was 60 cm and plant-to-plant distance was 20 cm. Fertilizer dose of 20: 50: 20 kg NPK/ha was uniformly applied as basal dose. Plant protection measures were taken as and when required. Five randomly selected competitive plants were tagged for recording various observations from each replication. .

3.5. Experimental details

The solution of EMS was prepared with following concentrations:

Doses:	EMS concentration
	0.1%
	0.2%
	0.3%
Time intervals for seed treatment:	4hour
	6hour
	8hour

So that, there are 10 treatment combinations including with control. Description regarding various treatments is presented in Table 3.2.

Table 3.2. : Description regarding various treatments

S. No	Treatment Combination	Description	S. No	Treatment Combination	Description
1	T ₁	CONTROL	6	T ₆	0.2% at 6 hour
2	T ₂	0.1% at 4hour	7	T ₇	0.2% at 8 hour
3	T ₃	0.1% at 6 hour	8	T ₈	0.3% at 4hour
4	T ₄	0.1% at 8 hour	9	T ₉	0.3% at 6hour
5	T ₅	0.2% at 4 hour	10	T ₁₀	0.3% at 8 hour

All seeds were water soaked for 4 hours before treatment. EMS solutions were prepared with the help of 1 ml pipette. For 0.1% solution 1 ml EMS was added in 1 lt. of water. Similarly for 0.2% solution 2 ml in 1 lt. of water and for 0.3% solution 3ml EMS in 1 lt. of water was added. Three containers for field and three for laboratory controls have been separated. As per treatment combinations the seeds were kept in different concentrations for four, six and eight hours. After treatment, the seeds were washed in running tap water for half an hour. Control treatments are only water drained.

The treated seeds were subjected to field sowing and other set of treated seed was used for laboratory evaluation. These treated seeds along with control was planted under randomized block design (RBD) in three replications during kharif season 2010.

Random selection of five plants in each plot was made and the observations were recorded on each selected plant. The means for each character under study were computed on the basis of five plants for each genotype.

3.5.1. Observations recorded:

Laboratory observation:

These are recorded in three interval of 7 days which is 7 days, 14 days and 21 days from germination onward.

1. Germination percent:

The seeds showing well grown plumule and radical were counted as germinated seeds. Counted seeds are expressed in percent germination.

2. Plumule length

It is that part of seedling which is responsible for shoot growth. It is measured in cm It was recorded 7 days after treatment of seeds in laboratories. This observation was recorded at every three interval of 7 days

3 Radical length:

It is that part of seedling which is responsible for root growth. It is measured in cm It was recorded 7 days after treatment of seeds in laboratories. This observation was recorded at every three interval of 7 days

Field observations

1. Days to 50% flowering:

It was recorded as number of days taken from the date of sowing to 50% percent flowering (anthesis) .

2. Days to maturity:

It was recorded as number of days taken from the date of sowing till 80 percent maturity of the pods of the plants.

3. Primary branches per plant:

The number of primary branches per plant was recorded as total number of main branches arising from main stem.

4. Plant height (cm):

The height of the plant was recorded in centimeter from the ground level to tip of main stem at the time of maturity.

5. Number of pods per plants:

The number of effective pods per plant was counted at the time of maturity for each tagged plant.

6. Biological yield per plant (g):

The dry weight of plant along with pods was recorded after cutting root portion for each genotype (grams).

7. Seeds per plant:

The number of effective seeds per plant was counted from randomly selected five plants of each genotype and the average is taken as seeds per plant.

8. Seed yield per plant (g):

The seed yield per plant was recorded after harvest.

9. Harvest index (%):

The value of harvest index was obtained by dividing the seed yield per plant by biological yield per plant and expressed in percentage.

$$\text{H.I. (\%)} = \frac{\text{Seed yield per plant (g)}}{\text{Biological yield per plant (g)}} \times 100$$

10. Hundred Seed weight (g):

After threshing and proper drying, randomly drawn 100 seeds from the produce of the sample plants were recorded in grams.

3.6. Statistical analysis:

The model for experimental design used in randomized block design can be expressed as follows.

$$P_{ij} = \mu + g_i + r_j + e_{ij}$$

Where, P_{ij} = phenotypic effect of i th genotype in the j th replication.

μ = general population mean

g_i = effect of i th genotype.

r_j = effect of j th replication.

e_{ij} = error associated with the experiment.

The skeleton for analysis of variance for randomized block design is given below.

Source of variation	d. f.	Sum of squares	Mean sum of squares
Replication	(r-1)	SSr	$\sigma^2 r$
Genotypes	(g-1)	SSg	$\sigma^2 e + r \sigma^2 g$

Error	(r-1)(g-1)	SSe	$\sigma^2 e$
Total	(rg-1)		

Where,

r = number of replications

g = number of genotypes.

$\sigma^2 g$ = genotypic variance (=MSt-MSe/r)

$\sigma^2 e$ = error variance

3.6.1. Test of significance:

The mean sum of squares for genotypes and replications were tested against the error mean sum of squares for calculating F values which were compared with tabulated F value at 5 and 1 percent level of significance.

3.6.2. Standard error of mean.

It was calculated as formula given below.

$$SEm \pm = \sqrt{(MSe/r)}$$

Where,

SE m \pm = standard error of mean

MSe = mean sum of square due to error

r = number of replication.

3.6.3. Standard error of differences.

It was calculated as formula given below

$$SE d \pm = \sqrt{(2MSe/r)}$$

Where,

SEd \pm = standard error of differences.

Mse = mean sum of square due to error.

r = number of replications.

3.6.4. Critical difference:

It was measured as formula mentioned below.

CD = SEd X t value at 5% level of significance.

Where,

CD = critical difference

SEd = standard error of difference

t = table value of 5% probability level of error df.

3.7. Genetic analysis:

The mean, range, components of variance, genotypic and phenotypic coefficient of variation and heritability in broad sense, genetic advance and expected genetic advance were calculated as per procedure mentioned below.

1. Mean:

Mean was calculated using following conventional formula

$$\bar{X} = \frac{\Sigma X}{N}$$

Where,

\bar{X} = simple mean

ΣX = summation of all the observation.

N = number of observation

2. Range:

It is the range of lowest and highest values of each trait taken in the observations.

3.8. Correlation coefficient:

The correlation coefficient values(r) were calculated at genotypic, phenotypic and environmental levels according to the formula given by Johnson *et al* (1955a) and described by Singh and Choudhary (1985) and presented below.

$$r(xy) = \frac{\text{COV.}(xy)}{\sqrt{\text{Var.}(x) \text{Var.}(y)}}$$

Where,

$r(xy)$ = correlation coefficient between character x and y.

$\text{COV}(xy)$ = covariance between character x and y.

$V(x)$ = variance of x character.

$V(y)$ = variance of y character.

CHAPTER - IV

RESULTS

This chapter deals with the outcome of the investigation for various parameters recorded for different characters observed in the study. The M_1 generation had been subjected to statistical analysis as per the RBD calculated. The results thus obtained have been presented in this section under following heads:

4.1 Analysis of variance in M_1 generation.

4.2 Mean performance of traits in M_1 generation.

4.1 Analysis of variance in M_1 generation

The mean sum of squares based on ANOVA for different characters have been presented in Table 4.1 and Table 4.11. The F value suggested that differences among the treatments were highly significant for all the characters.

I. Laboratory study

4.1.1 Germination percent (first week)

For germination in first week genotype, concentration, time and interaction, between genotype x concentration, genotype x time, time x concentration and genotype x time x concentration were highly significant (Table 4.1). There was a significant decrease in the germination of variety ICPL 87(78.63%). The decrease was 5.76% than variety JA 4(83.47%). (Table 4.2.1)

There was a significant decrease of 18.67% in 0.3% concentration of EMS (73.22%) than the control (90.00%). (Table 4.2.1)

Significant decrease in germination was observed in 8 hours treatment of EMS (79.04%). The decrease was 12.17% than control (90%). (Table 4.2.1)

Genotype x Concentration interaction was highly significant for first week of germination. Percent reduction of germination (17.77%) was highest in 0.3% EMS concentration of genotype ICPL 87 as compared to control (86.00%) while it was 16.23% in JA 4 for 0.3% EMS treatment (75.44%) as compared to control (94.00%)(Table 4.2.1).

Genotype x Time interaction was also found significant. In 8 hours treatment of EMS expressed significantly decrease of germination was found among variety

tested .It was more (77.75%) in ICPL 87 than JA 4 (80.33%).. However in overall, time duration has affected the germination at first week.(Table 4.2.1)

Table 4.2.1: Effect of EMS concentration and time duration on germination percentage in first week in pigeonpea genotype.

Germination percentage in first week				
Concentration	Genotypes		Mean	CD at 5% (Conc.)
	ICPL 87	JA 4		
0.1%	80.11	85.44	82.78	0.99
0.2%	77.11	79.00	78.10	
0.3%	71.00	75.44	73.20	
Control	86.00	94.00	90.00	
Mean	78.55	83.36	81.03	
CD at 5% (Genotype)	0.70			
CD at 5% (G x Con.)= 1.40				
Time duration				CD at 5% (Time duration)
4 hours	79.91	85.91	82.91	0.86
6 hours	78.25	84.16	81.20	
8 hours	77.75	80.33	79.04	
Mean	77.97	83.46	81.05	
CD at 5% (G x Time duration) = 1.21				

Concentration x Time interaction was significant. As compared to control There was 23.33% reduction in germination in 0.3% EMS concentration at 8 hours treatment (69.50%).(Table 4.2.2)

Table 4.2.2: Effect of EMS concentration and time duration on germination percentage in first week in pigeonpea genotype.

Germination percentage in first week				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	86.83	82.17	79.33	82.77

0.2%	79.33	77.67	77.17	78.05
0.3%	75.33	74.83	69.50	73.22
Control	90.17	90.17	90.17	90.17
Mean	82.92	81.21	79.05	81.06
CD at 5% (Con. x Time duration)= 1.72				

Genotype x Concentration x Time interaction was also found significant. Genotype ICPL 87 in 0.3% EMS concentration at 8 hours treatment showed lowest germination of 67.60% which was significantly lower than other combinations. (Table 4.2.3)

Table 4.2.3: Effect of EMS concentration and time duration on germination percentage in first week in pigeonpea genotype.

Germination percentage in first week					
Genotype	Concentration (%)	Time durations			
		4 hours	6 hours	8 hours	Mean
ICPL 87	0.10%	82.00	79.67	78.67	80.11
	0.20%	77.00	76.00	78.33	77.11
	0.30%	74.33	71.00	67.67	71.00
	Control	86.33	86.33	86.33	86.33
JA 4	0.10%	91.67	84.67	80.00	85.47
	0.20%	81.67	79.33	76.00	79.00
	0.30%	76.33	78.67	71.33	75.44
	Control	94.00	94.00	94.00	94.00
Grand Mean = 82.81%					
CD at 5% (G x Con. x Time duration)= 2.44					

4.1.2 Germination per cent (second week)

For germination in second week genotype, concentration, time and interaction, between genotype x concentration, genotype x time, time x concentration and genotype x time x concentration were highly significant (Table 4.1). There was a significant decrease in the germination of variety ICPL 87(84.58%). The decrease was 2% than variety JA 4(86.39%) (Table 4.3.1)

There was a significant decrease of 17% in 0.3% concentration of EMS (77.83%) than the control (94.00%).

Significant decrease in germination was observed in 8 hours treatment of EMS (83.17%). The decrease was 11.52% than control (94%).(Table 4.3.1)

Table 4.3.1: Effect of EMS concentration and time duration on germination percentage in second week in pigeonpea genotype.

Germination percentage in second week				
Concentration	Genotypes		Mean	CD at 5% (Conc.)
	ICPL 87	JA 4		
0.1%	87.55	87.22	87.38	0.70
0.2%	83.77	81.67	82.72	
0.3%	75.33	80.33	77.83	
Control	91.66	96.33	94.00	
Mean	84.58	86.38	85.73	
CD at 5% (Genotype)	0.49			
CD at 5% (G x Con.) = 1				
Time duration				CD at 5% (Time duration)
4 hours	85.67	87.75	87.71	0.61
6 hours	84.17	87.00	85.59	
8 hours	83.91	82.42	83.17	
Mean	84.58	86.39	85.48	
CD at 5% (G x Time duration) = 0.86				

Genotype x Concentration interaction was highly significant for second week of germination. Percent reduction of germination (17.82%) was highest in 0.3% EMS concentration of genotype ICPL 87 (75.33%) as compared to control (91.66%). It was 15.29% in JA 4 for 0.3% EMS treatment (80.33%) as compared to control (96.33%).(Table 4.3.1)

Genotype x Time interaction was also found significant. In 6 hours treatment of EMS expressed significantly decrease of germination among variety tested .It was more reduction in germination in genotype ICPL 87(84.17%) than JA 4 (87.00%). However in overall, time duration has affected the germination at second week. (Table 4.3.1)

Concentration x Time interaction was significant. As compared to control (94.00%) there was 21.62 % reduction in germination in 0.3% EMS concentration at 8 hours treatment (73.67%).(Table 4.3.2)

Table 4.3.2: Effect of EMS concentration and time duration on germination percentage in second week in pigeonpea genotype.

Germination percentage in second week				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	91.83	87.00	83.33	87.38
0.2%	83.67	82.83	81.67	82.72
0.3%	81.33	78.50	73.67	77.83
Control	94.00	94.00	94.00	94.00
Mean	87.71	85.59	83.16	85.48
CD at 5% (Con. x Time duration)= 1.21				

Genotype x Concentration x Time interaction was also found significant. Genotype ICPL 87 in 0.3% EMS concentrations at 8 hours treatment showed lowest germination of 71.67% which was significantly lower than other combinations (Table 4.3.3)

Table 4.3.3: Effect of EMS concentration and time duration on germination percentage in second week in pigeonpea genotype.

Germination percentage in second week				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	89.66	86.00	87.00
	0.20%	82.33	83.66	85.33
	0.30%	79.00	75.33	71.67
	Control	91.66	91.66	91.66
JA 4	0.10%	94.00	88.00	79.66
	0.20%	85.00	82.00	78.00
	0.30%	83.66	81.66	75.66
	Control	96.33	96.33	96.33
Grand Mean =85.48				
CD at 5% (G x Con. x Time duration)= 1.72				

4.1.3 Germination percentage (third week)

For germination in third week genotype, concentration, time and interactions, between genotype x concentration, genotype x time, time x concentration and genotype x time x concentration were highly significant (Table 4.1). Genotype varied significantly. ICPL 87 has significantly (1.38%) lower germination (86.91%) than JA 4 (88.13%) (Table 4.4.1).

There was a Significant 14% decrease in 0.3% concentration of EMS (80.77%) was observed than the control (94.33%). (Table 4.4.1)

Significant decrease in germination was observed in 8 hours treatment of EMS (84.75%). The decrease was 10.15% than control (94.33%). (Table 4.4.1)

Genotype x Concentration interaction was highly significant for third week of germination. Percent reduction of germination was highest in 0.3% EMS concentration (13.00%) of genotype ICPL 87 as compared to control (91.67%) while it was 15.70% in JA 4 for 0.3% EMS treatment (81.78%) as compared to control (97.00%). (Table 4.4.1)

Genotype x Time interaction was also found significant. It was found 6 hours treatment of EMS expressed significantly difference of germination among variety tested .It was more reduction in ICPL 87 (85.67%) than JA 4 (88.58%). The time duration of 4 hours and 8 hours did not differ in genotypes. (Table 4.4.1)

Table 4.4.1: Effect of EMS concentration and time duration on germination percentage in third week in pigeonpea genotype.

Germination percentage in third week				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	90.22	89.44	89.83	0.82
0.2%	86.00	84.33	85.17	
0.3%	79.78	81.78	80.78	
Control	91.67	97.00	94.34	
Mean	86.91	88.13	87.52	
CD at 5% (Genotype) = 0.58				
CD at 5% (G x Con.) = 1.16				

Time duration				CD at 5% (Time duration)
4 hours	90.25	91.17	90.71	0.71
6 hours	85.67	88.58	87.13	
8 hours	84.83	84.67	84.75	
Mean	86.92	88.14	87.53	
CD at 5% (G x Time duration) = 1				

Concentration x Time interaction was significant. As compared to control (94.00%) there was 21.62 % reduction in germination in 0.3% EMS concentration at 8 hours treatment (75.83%). (Table 4.4.2)

Table 4.4.2: Effect of EMS concentration and time duration on germination percentage in third week in pigeonpea genotype.

II Germination percentage in third week				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	93.33	90.33	85.83	83.90
0.2%	89.00	83.50	83.00	85.17
0.3%	86.17	80.33	75.83	80.78
Control	94.33	94.33	94.33	94.33
Mean	90.70	87.12	84.74	86.04
CD at 5% (Con. x Time duration)= 1.42				
CD at 5% (G x Con. x Time duration)= 2				

Genotype x Concentration x Time interaction was also found significant. Genotype ICPL 87 in 0.3% EMS concentration at 8 hours treatment showed lowest germination of 74.67% which was significantly lower than other combinations. (Table 4.4.3)

Table 4.4.3: Effect of EMS concentration and time duration on germination percentage in third week in pigeonpea genotype.

Germination percentage in third week				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	91.66	90.33	88.66
	0.20%	90.33	83.33	84.33
	0.30%	87.33	77.33	74.66
	Control	91.66	91.66	91.66

JA 4	0.10%	95.00	90.33	83.00
	0.20%	87.66	83.66	81.66
	0.30%	85.00	83.33	77.00
	Control	97.00	97.00	97.00
Grand Mean = 86.04%				
CD at 5% (G x Con. x Time duration)= 2.00				

4.1.4 Plumule length (first week)

For plumule length (cm) in its first week genotype, concentration time and interaction between genotype x concentration, genotype x time, time x concentration and genotype x time x concentration were highly significant. There was a significant decrease in the plumule length of variety JA 4 (1.76cm). The decrease was 12% than variety ICPL 87 (2.01cm). (Table 4.5.1)

There was a significant decrease of 66% in 0.1% concentration of EMS (1cm) than the control (2.97cm) (Table 4.5.1).

Significant decrease in plumule length (cm) was observed in 8 hours treatment (1.76cm). The decrease was 40.74% of EMS treatment than control (2.1cm). (Table 4.5.1)

Table 4.5.1: Effect of EMS concentration and time duration on plumule length (cm) in first week in pigeonpea genotype.

Plumule length (cm) in first week				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	1.43	0.56	1.00	0.71
0.2%	1.68	1.70	1.69	
0.3%	1.81	1.92	1.87	
Control	3.10	2.80	2.95	
Mean	2.01	1.75	2.13	
CD at 5% (Genotype)	0.05			
CD at 5% (G x Con.) = 0.1				

Time duration				
4 hours	1.87	1.74	1.81	0.06
6 hours	2.21	1.98	2.10	
8 hours	1.95	1.57	1.76	
Mean	2.01	1.76	1.89	
CD at 5% (G x Time duration) = 0.087				

Genotype x Concentration interaction was highly significant for first week of plumule length. Percent reduction of plumule length (66.45%) was highest in 0.1% EMS concentration (1.43cm) as compared to control (3.13cm) of genotype ICPL 87. It was 73.64% in JA 4 (0.56cm) for 0.1% EMS treatment as compared to control (2.69cm). (Table 4.5.1).

Genotype x Time interaction was also found significant. In 8 hours treatment of EMS expressed significantly difference of plumule length (cm) was found among variety tested .It was more reduction in JA 4 (1.57cm) than ICPL 87 (1.95cm). However in overall time duration has affected the plumule length at first week. (Table 4.5.1)

Concentration x Time interaction was significant. As compared to control (2.96cm) there was 66.45% reduction in germination in 0.1% EMS concentration 8 hours treatment (1.16cm) and 0.2% for 4 hours (1.16cm) and at par with 0.1% for 6 hours (1.22cm). (Table 4.5.2)

Table 4.5.2: Effect of EMS concentration and time duration on plumule length (cm) in first week in pigeonpea genotype.

Plumule length (cm) in first week				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	1.16	1.13	0.62	0.97
0.2%	1.21	2.52	1.45	1.73
0.3%	1.88	1.69	2.02	1.86
Control	2.97	2.97	2.97	2.97
Mean	1.81	2.08	1.77	1.89
CD at 5% (Con. x Time duration)= 0.12				

Genotype x Concentration x Time interaction was also found significant. Genotype JA 4 in 0.1% EMS concentrations at 8 hours treatment showed lowest plumule length of 0.24cm which was significantly lower than other combinations. (Table 4.5.3)

Table 4.5.3: Effect of EMS concentration and time duration on plumule length (cm) in first week in pigeonpea genotype.

Plumule length (cm) in first week				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	1.66	1.63	1.00
	0.20%	1.07	2.76	1.20
	0.30%	1.60	1.33	2.50
	Control	3.13	3.13	3.13
JA 4	0.10%	0.65	0.80	0.24
	0.20%	1.33	2.26	1.70
	0.30%	2.16	2.05	1.54
	Control	2.80	2.80	2.80
	Mean	1.80	2.09	1.76
Grand mean=1.89				
CD at 5% (G x Con. x Time duration)= 0.17				

4.1.5 Plumule length (second week)

For plumule length (cm) in its second week genotype, concentration time and interaction between genotype x concentration, genotype x time, time x concentration and genotype x time x concentration were highly significant while time and its interaction with genotype was not significant (Table 4.1). There was a significant decrease in the plumule length of variety JA 4 (2.43cm). The decrease were 16% than variety ICPL 87 (2.90cm).(Table 4.6.1)

There was a significant decrease of 43% in 0.1%concentration of EMS (1.97cm) than the control (3.5cm). (Table 4.6.1)

Genotype x Concentration interaction was highly significant for second week of plumule length. Percent reduction of plumule length (50%) was highest in 0.2% EMS concentration (2.17cm) as compared to control (4.31cm) of genotype ICPL 87. It was 38.30% in JA 4 (1.73cm) for 0.1% EMS treatment as compared to control (2.80cm). (Table 4.6.1)

Concentration x Time interaction was significant. As compared to control (3.5cm) there was 64% reduction in germination in 0.1% EMS concentration at 8 hours treatment (1.25cm) at par with 0.2% for 4 hours (1.63cm) (Table 4.6.2).

Table 4.6.1: Effect of EMS concentration and time duration plumule length (cm) in second week in pigeonpea genotype.

Plumule length (cm) in second week				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	2.23	1.70	1.97	0.27
0.2%	2.17	2.70	2.44	
0.3%	2.91	2.50	2.71	
Control	4.31	2.69	3.50	
Mean	2.91	2.40	2.65	
CD at 5% (Genotype)	0.2			
CD at 5% (G x Con.) = 0.39				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	2.70	2.00	1.25	1.98
0.2%	1.64	3.04	2.73	2.47
0.3%	3.10	2.40	2.67	2.72
Control	3.50	3.50	3.50	3.50
Mean	2.74	2.74	2.38	2.67
CD at 5% (Con. x Time duration) = 0.48				

Genotype x Concentration x Time interaction was also found significant. Genotype JA 4 in 0.1% EMS concentration at 8 hours treatment showed lowest

plumule length (cm) of 1.40cm which was significantly lower than other combinations at par with 0.2% at 4 hours (1.66cm) (Table 4.6.2).

Table 4.6.2 Effect of EMS concentration and time duration on plumule length (cm) in second week in pigeonpea genotype.

Plumule length (cm) in second week				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	2.76	2.53	1.40
	0.20%	1.66	2.83	2.00
	0.30%	3.10	2.06	3.56
	Control	4.31	4.31	4.31
JA 4	0.10%	2.64	1.46	1.10
	0.20%	1.61	3.23	3.45
	0.30%	3.10	2.74	1.78
	Control	2.69	2.69	2.69
Grand Mean = 2.66				
CD at 5% (G x Con. x Time duration)= 0.68				

4.1.6 Plumule length (third week)

For plumule length (cm) in its third week genotype, concentration time and interaction between genotype x concentration, genotype x time, time x concentration and genotype x time x concentration were highly significant. There was a significant decrease in the plumule length of variety JA 4 (3.31cm). The decrease was 9.50% than variety ICPL 87 (3.67cm). (Table 4.7.1)

There was a significant decrease of 42% in 0.1%concentration of EMS 2.59cm then the control (4.48cm). (Table 4.7.1)

Significant decrease in plumule length (cm) was observed in 8 hours treatment (3.28cm). The decrease was 26.78% of EMS treatment than control (4.48cm). (Table 4.7.1)

Genotype x Concentration interaction was highly significant for third week of plumule length. Percentage reduction of plumule length (47%) was highest in 0.2% EMS concentration (2.68cm) as compared to control (5.10cm) of genotype

ICPL 87. It was 35% in JA 4 (2.36cm) for 0.1% EMS treatment as compared to control (3.87cm). (Table 4.7.1)

Genotype x Time interaction was highly significant for third week of plumule length (cm). Percentage reduction (31%) of plumule length (cm) was highest in 8 hours EMS treatment (3.53cm) as compared to control (5.1cm) of genotype ICPL 87 while it was 21.70% reduction in JA 4 for 8 hours EMS treatment (3.03cm) as compared to control (3.87cm) in JA 4 (Table 4.7.1).

Table 4.7.1: Effect of EMS concentration and time duration plumule length (cm) in third week in pigeonpea genotype.

Plumule length in third week				
Concentration	Genotypes		Mean	CD at 5% (Conc.)
	ICPL 87	JA 4		
0.1%	2.82	2.35	2.59	0.11
0.2%	2.68	3.72	3.20	
0.3%	4.04	3.31	3.68	
Control	5.10	3.87	4.49	
Mean	3.66	3.31	3.49	
CD at 5% (Genotype) =0.08				
CD at 5% (G x Con.)= 0.16				
Time duration				CD at 5% (Time duration)
4 hours	3.69	3.36	3.52	0.10
6 hours	3.77	3.54	3.66	
8 hours	3.52	3.03	3.28	
Mean	3.66	3.34	3.49	
CD at 5% (G x Time duration) = 0.14				

Concentration x Time interaction was significant. As compared to control (4.48cm) There was 60% reduction plumule length (cm) in 0.1% EMS concentration 8 hours treatment (1.77cm).(Table 4.7.2)

Table 4.7.2: Effect of EMS concentration and time duration plumule length (cm) in third week in pigeonpea genotype.

Plumule length in third week				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	3.27	2.72	1.77	2.59
0.2%	2.20	3.81	3.60	3.20
0.3%	4.14	3.62	3.27	3.67
Control	4.49	4.49	4.49	4.49
Mean	3.52	3.66	3.28	3.49
CD at 5% (Con. x Time duration)= 0.20				

Genotype x Concentration x Time interaction was also found significant. Genotype JA 4 in 0.1% EMS concentration at 8 hours treatment showed the lowest plumule length (cm) 1.65cm which was significantly lower than other combinations at par with 0.1% EMS concentration at 8 hours treatment (1.80cm) of ICPL 87. (Table 4.7.3)

Table 4.7.3: Effect of EMS concentration and time duration plumule length (cm) in third week in pigeonpea genotype.

III Plumule length in third week				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	3.36	3.20	1.88
	0.20%	2.13	3.22	2.70
	0.30%	4.16	3.55	4.41
	Control	5.10	5.10	5.10
JA 4	0.10%	3.10	2.23	1.65
	0.20%	2.27	4.40	4.50
	0.30%	4.12	3.68	2.12
	Control	3.87	3.87	3.87
	Grand mean	3.48		
CD at 5% (G x Con. x Time duration)= 0.28				

4.1.7 Radical length (first week)

For radical length (cm) in its first week genotype, concentration and interaction between genotype x concentration, genotype x time, time x concentration and genotype x time x concentration were highly significant while time was not differ significant (Table 4.1). There was a significant decrease in the radical length of variety JA 4 (2.09cm). The decrease was 16% than variety ICPL 87 (2.49cm).(Table 4.8.1)

There was a significant decrease of 57.18 % in 0.1% concentration of EMS 2.35cm than the control (3.31cm). (Table 4.8.1)

Table 4.8.1: Effect of EMS concentration and time duration radical length (cm) in first week in pigeonpea genotype.

Radical length (cm) in first week				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	2.33	2.37	2.35	0.11
0.2%	2.40	2.71	2.56	
0.3%	2.32	3.55	2.95	
Control	2.90	2.70	3.30	
Mean	2.49	2.09	2.79	
CD at 5% (Genotype)	0.081			
CD at 5% (G x Con.)= 0.16				
Time duration				CD at 5% (Time duration)
4 hours	2.10	3.69	1.30	0.1
6 hours	2.21	3.67	2.94	
8 hours	2.27	2.56	2.41	
Mean	2.19	3.10	2.65	
CD at 5% (G x Time duration) = 0.14				

Genotype x Concentration interaction was highly significant for first week of radical length. Percent reduction of radical length (19.65%) was highest in 0.3% EMS concentration (2.33cm) as compared to control (2.90cm) of genotype ICPL 87. It was 12.23% in JA 4 (2.37cm) for 0.1% EMS treatment as compared to control (3.73cm). (Table 4.8.1)

Genotype x Concentration interaction was highly significant for third week of radical length (cm). This interaction created a vice-versa changes. Percent reduction of radical length (27%) was highest in 4 hours EMS concentration (2.10cm) of genotype ICPL 87 as compared to control (2.90cm) while it was 36% increase in JA

4 for 4 hours EMS treatment (3.69cm)) as compared to control (2.70cm). (Table 4.8.1)

The interaction between time duration and concentrations was significant. Application of 0.1% for 8 hours (1.46cm) recorded the lowest radical length which was significantly 56% than control (3.31cm) (Table 4.8.2)

Table 4.8.2: Effect of EMS concentration and time duration radical length (cm) in first week in pigeonpea genotype.

II Radical length (cm) in first week				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	3.41	2.18	1.46	2.35
0.2%	2.27	3.70	1.70	2.56
0.3%	3.16	2.53	3.17	2.95
Control	3.31	3.31	3.31	3.31
Mean	1.30	2.94	2.41	2.79
CD at 5% (Con. x Time duration)= 0.20				

Genotype x Concentration x Time interaction was also found significant. Genotype JA 4 in 0.1% EMS concentration at 8 hours treatment showed lowest radical length (1.22cm) which was significantly lower than other combinations (Table 4.8.3).

Table 4.8.3: Effect of EMS concentration and time duration radical length (cm) in first week in pigeonpea genotype.

III Radical length (cm) in first week				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	3.50	1.80	1.70
	0.20%	2.81	2.50	1.90
	0.30%	2.76	1.63	2.56
	Control	2.90	2.90	2.90
	0.10%	3.32	2.57	1.22

JA 4	0.20%	1.73	4.9	1.51
	0.30%	3.55	3.43	3.76
	Control	3.73	3.73	3.73
Grand Mean =2.79				
CD at 5% (G x Con. x Time duration)= 0.28				

4.1.8 Radical length (second week)

For radical length (cm) in its second week genotype, concentration time and interaction between genotype x concentration, genotype x time, time x concentration and genotype x time x concentration were highly significant (Table 4.1). There was a significant decrease in the radical length of variety ICPL 87 (3.51cm). The decrease was 5.62% than variety JA 4 (3.72cm). (Table 4.9.1)

There was a significant decrease of 17.43% in 0.1%concentration of EMS 3.22cm than the control (3.90cm). (Table 4.9.1)

There was a significant decrease of 19.74% in 8 hours of EMS 3.13cm than control (3.90cm). (Table 4.9.1)

The concentration of EMS solution created a vice versa changes for genotype x concentration. Percent increase of radical length (16%) was highest in 0.2% EMS concentration (3.94cm) of genotype ICPL 87 as compared to control (3.40cm) while in JA 4 it was 27.10% reduction for 0.1% EMS concentration (3.20cm) as compared to control (4.39cm)(Table 4.9.1).

Genotype x Time interaction was highly significant for second week of radical length (cm). It was found 8 hours EMS treatment (3.06cm) express significant reduction in ICPL 87 as compared to its control (3.40cm) while same treatment of EMS (3.02cm) express significant reduction in JA 4 as compared to its control (4.39cm). Percent reduction rate was higher in JA 4 (71%) as compared to ICPL 87 (10%) with respect to there control. (Table 4.9.1)

Table 4.9.1: Effect of EMS concentration and time duration on radical length on second week (cm) in pigeonpea genotype.

I Radical length (cm) in second week				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	3.29	3.20	3.22	0.33
0.2%	3.94	3.29	3.62	
0.3%	3.41	3.92	3.67	
Control	3.40	4.39	3.90	
Mean	3.51	3.70	3.60	
CD at 5% (Genotype) =0.23				
CD at 5% (G x Con.)=0.47				
Time duration				CD at 5% (Time duration)
4 hours	3.43	3.67	3.55	0.28
6 hours	3.30	3.28	3.79	
8 hours	3.06	3.02	3.13	
Mean	3.26	3.72	3.49	
CD at 5% (G x Time duration) = 0.17				

The interaction between time duration and concentrations was significant. Application of 0.2% for 8 hours (2.23cm) recorded the lowest radical length which was significantly 45.25% lower than control (4.00cm). (Table 4.9.2)

Table 4.9.2: Effect of EMS concentration and time duration on radical length on second week (cm) in pigeonpea genotype.

Radical length (cm) in second week				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	4.07	3.29	2.49	3.28
0.2%	2.87	4.27	2.23	3.12
0.3%	3.58	3.60	3.82	3.67
Control	4.00	4.00	4.00	3.80
Mean	3.55	3.79	3.14	3.49
CD at 5% (Con. x Time duration)= 0.57				
CD at 5% (G x Con. x Time duration)= 0.80				

Genotype x Concentration x Time interaction was also found significant. Genotype JA 4 in 0.2% EMS concentration at 8 hours treatment showed lowest

radical length (1.75 cm) which was significantly lower than other combinations (Table 4.9.3).

Table 4.9.3 Effect of EMS concentration and time duration on radical length on second week (cm) in pigeonpea genotype.

III Radical length (cm) in second week				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	4.29	3.11	2.47
	0.20%	3.10	3.03	2.71
	0.30%	3.35	3.44	3.44
	Control	3.61	3.61	3.61
JA 4	0.10%	3.83	3.45	2.51
	0.20%	2.64	5.50	1.75
	0.30%	3.79	3.76	4.20
	Control	4.40	4.40	4.39
	Mean	3.54	3.79	3.14
	Grand Mean	3.49		
	CD at 5% (G x Con. x Time duration)= 2.44			

4.1.9 Radical length (third week)

For radical length (cm) in its third week genotype, concentration and interaction between genotype x concentration, genotype x time, time x concentration and genotype x time x concentration were highly significant (Table 4.1). There was a significant decrease in the radical length of variety ICPL 87 (3.80cm). The decrease was 10% than variety JA 4 (4.24cm). (Table 4.10.1)

There was a significant decrease of 14.74% in 0.1% concentration of EMS 3.70cm at par with 0.2% (3.76cm) than control (4.27cm). (Table 4.10.1)

There was a significant decrease of 16.67% in 8 hours of EMS treatment (3.66 cm) than control (4.27cm). (Table 4.10.1)

Table 4.10.1 Effect of EMS concentration and time duration on radical length on third week (cm) in pigeonpea genotype..

I Radical length (cm) in third week				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	3.53	3.88	3.70	0.15
0.2%	3.39	4.14	3.77	
0.3%	4.40	4.28	4.32	
Control	4.27	4.64	4.27	
Mean	3.80	4.24	4.03	
CD at 5% (Genotype) = 0.1				
CD at 5% (G x Con.) = 0.20				
Time duration				CD at 5% (Time duration)
4 hours	4.26	4.15	4.21	0.13
6 hours	3.62	4.77	4.20	
8 hours	3.55	3.78	3.66	
Mean	3.81	4.23	4.02	
CD at 5% (G x Time duration) = 0.18				

Genotype x concentration interaction was highly significant for third week of radical length (cm). It was 20.60% decrease in ICPL 87 for 0.2% EMS concentration (3.39cm) as compared to control (4.27cm) while in JA 4 0.1% EMS concentration (3.88cm) show lowest radical length in third week which was 16.37% lower than control (4.64cm). (Table 4.10.1)

Genotype x Time interaction was also found significant. It was found 8 hours treatment of EMS (3.55cm) expressed significantly 17% decrease of radical length in ICPL 87 while in JA 4 it was 18.53% reduction of radical length at 8 hours (3.78cm) as compared to control (4.64 cm). The time duration of 6 hours and 8 hours did not differ in genotypes..(Table 4.10.1)

The interaction between time duration and concentrations was significant. Application of 0.2% for 8 hours (2.71cm) recorded the lowest radical length which was significantly 30.08% lower than control (4.27cm). (Table 4.10.2)

Table 4.10.2: Effect of EMS concentration and time duration on radical length on third week (cm) in pigeonpea genotype.

II Radical length (cm) in third week				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	4.38	3.49	3.25	3.71
0.2%	3.75	4.84	2.71	3.77
0.3%	4.43	4.19	4.43	4.35
Control	4.27	4.27	4.27	4.27
Mean	4.21	4.20	3.67	4.03
CD at 5% (Con. x Time duration) = 0.25				

Genotype x Concentration x Time interaction was also found significant. Genotype JA 4 in 0.2% EMS concentration at 8 hours treatment showed lowest radical length of (2.36cm) which was significantly lower than other combinations. (Table 4.10.3)

Table 4.10.3 Effect of EMS concentration and time duration on radical length on third week (cm) in pigeonpea genotype..

III Radical length (cm) in third week				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	4.58	3.08	2.93
	0.20%	3.83	3.28	3.05
	0.30%	4.70	4.20	4.31
	Control	3.90	3.90	3.90
JA 4	0.10%	4.16	3.90	3.56
	0.20%	3.66	6.38	2.36
	0.30%	4.15	4.16	4.54
	Control	4.64	4.64	4.64
	Grand mean	4.023		
CD at 5% (G x Con. x Time duration)= 0.35				

II. Field study

The varieties tested are different maturity group. ICPL 87 is an early flowering and early maturity while JA 4 has medium flowering and maturity.

4.2.1 Days to 50% flowering

For the characteristics days to 50% flowering, there was a vice versa effect in variety under study. An increase was found in early maturing variety ICPL 87 while a decrease was observed in medium maturity variety JA 4 variety these trait there was increase of 7 to 8 days in 50% flowering of ICPL 87 in 0.2 % EMS treatment which was significantly higher over its control. The decrease in flowering of 3-4 days in JA 4 was also observed in 0.2% EMS treatment (Table 4.12.1).

The time of treatment also affected 50% flowering which showed increase of about 3-4 days in ICPL 87 from 4 hours to 8 hours while in JA 4 there was a decrease about 6-7 days from 4 hours to 8 hours EMS treatment. (Table 4.12.1)

Table 4.12.1: Effect of EMS concentration and time duration on days to 50% flowering in pigeonpea genotype.

Concentration	Days to 50% flowering		Mean	CD at 5% (Concentration)
	Genotypes			
	ICPL 87	JA 4		
0.1%	107.00	121.67	114.34	1.70
0.2%	107.67	117.78	112.73	
0.3%	105.56	117.00	111.28	
Control	100.00	120.00	110.00	
Mean	105.06	119.11	112.01	
CD at 5% (Genotype) =		0.49		
CD at 5% (G x Con.) =		2.40		
Time duration				CD at 5% (Time duration)
4 hours	103.67	122.00	112.84	1.4
6 hours	104.75	118.83	111.79	
8 hours	106.75	116.50	111.63	
Mean	105.06	119.11	111.86	
CD at 5% (G x T) =		2.08		

The interaction of genotype x concentration x time was significant for this trait , There was an increase of 12 days in 0.2% concentration of 8 hours treatment in

ICPL 87 while a decrease of 9 days was observed in 0.2% EMS of 8 hours treatment in JA 4. It clearly shows that genetic constitution of early & medium maturing genotype respond in opposite direction for 50% flowering. (Table 4.12.2)

Table 4.12.2: Effect of EMS concentration and time duration on days to 50% flowering in pigeonpea genotype.

II Days to 50% flowering				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	106.00	106.00	109.00
	0.20%	102.00	109.00	112.00
	0.30%	106.66	104.00	106.00
	Control	100.00	100.00	100.00
JA 4	0.10%	124.00	122.00	119.00
	0.20%	126.00	116.33	111.00
	0.30%	118.00	117.00	116.00
	Control	120.00	120.00	120.00
Grand mean = 112.08 days				
CD at 5% (G x Con. x Time duration)= 4.16				

4.2.2 Days to maturity

Concentration of EMS significantly affected of early maturing variety ICPL 87. In ICPL 87 there was an increase in maturity from 138 days (control) to 146 days in 0.1% EMS treatment. (Table 4.13.1)

Table 4.13.I: Effect of EMS concentration and time duration on days to maturity in pigeonpea genotype.

I Days to maturity				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	146.00	170.67	158.34	0.70
0.2%	145.67	168.00	156.83	
0.3%	142.11	167.56	154.84	
Control	138.00	169.00	153.50	
Mean	142.95	158.81	150.88	
CD at 5% (Genotype) = 0.50				

CD at 5% (Genotype x Concentration) = 1				
Time duration				CD at 5% (Time duration)
4 hours	141.25	170.75	156.00	0.61
6 hours	143.50	168.75	156.13	
8 hours	144.08	166.92	155.50	
Mean	142.94	168.89	155.87	
CD at 5% (G x T) =0.86				

The interaction of concentration x time was significant for this trait. There was delay of 6-7 days shown by 0.1% concentration at 6 hours EMS treatment (159days) than control (153.50days).(Table 4.13.2).

Table 4.13.II: Effect of EMS concentration and time duration on days to maturity in pigeonpea genotype.

II Days to maturity				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	157.50	159.50	158.00	158.33
0.2%	157.50	157.00	156.00	156.83
0.3%	155.50	154.50	154.50	154.83
Control	153.50	153.50	153.50	153.5
Mean	156.00	156.12	155.50	155.87
CD at 5% (Con. x Time duration) = 1.22				

Highest increase in maturity from 138 to 149 days was noticed in 0.2% EMS concentration of 8 hours treatment was observed in variety ICPL 87 which was found significantly different from other treatments. (Table 4.13.1)

However when all the interaction combination were taken into consideration an increase of 5- 6 days (175 days) as compared to control (169 days) was observed in 0.2% EMS treatment of 4 hours treatment in JA 4 also. (Table 4.13.3)

Table 4.13.3: Effect of EMS concentration and time duration on days to maturity in pigeonpea genotype.

III Days to maturity				
		Time durations		
Genotype	Concentration (%)	4 hours	6 hours	8 hours
	0.10%	145.00	146.00	147.00
ICPL 87	0.20%	140.00	148.00	149.00
	0.30%	142.00	142.00	142.33
	Control	138.00	138.00	138.00
	0.10%	170.00	173.00	169.00
JA 4	0.20%	175.00	166.00	163.00
	0.30%	169.00	167.00	166.66
	Control	169.00	169.00	169.00
Grand Mean=155.87				
CD at 5% (G x Con. X Time duration)= 1.73				

In general it was noticed that enhancement in days to maturity was noticed in both the variety.

4.2.3 Primary branches per plant

The interaction of concentration x time of EMS significantly affected primary branches per plant of early maturing variety ICPL 87. In ICPL 87 there was an increase in no. of primary branches per plant from 8.9 (0.3% EMS concentration at 4 hours) to 13.70 (0.3% EMS concentration at 8 hours treatment). (Table 4.14.1)

Table 4.14.1: Effect of EMS concentration and time duration on primary branches per plant in pigeonpea genotype.

Primary branches per plant				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	11.20	8.90	11.6	10.57
0.2%	12.50	10.40	10.00	10.97
0.3%	8.90	10.70	13.70	11.10
Control	9.50	9.50	9.50	9.50
Mean	10.53	9.88	11.20	10.53
CD at 5% (Con. X Time duration) = 2.31				

However when all the interaction combination were taken into consideration an increase of primary branches per plant from 4 to 5 as compared to control (8.80) was observed in 0.1% EMS concentration at 8 hours treatment in ICPL 87 while it was 4 to 5 as compared to control (14.80) was observed in 0.2% EMS treatment of 4 hours treatment (14.8) in JA 4 also.(Table 4.14.2)

Table 4.14.2: Effect of EMS concentration and time duration on primary branches per plant in pigeonpea genotype.

II Primary branches per plant				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	10.80	8.60	13.40
	0.20%	10.20	12.40	10.40
	0.30%	9.60	10.20	12.80
	Control	8.80	8.80	8.80
JA 4	0.10%	11.60	9.20	9.80
	0.20%	14.80	8.40	9.60
	0.30%	8.20	11.20	14.60
	Control	10.20	10.20	10.20
	Mean	10.53	9.88	11.20
Grand Mean= 13.20				
CD at 5% (G x Con. X Time duration)= 3.27				

4.2.4 Plant height (cm)

For the characteristics plant height (cm) there was a vice -versa effect in variety under study. An increase was found in early maturing variety ICPL 87 while a decrease was observed in medium maturity variety JA 4 variety for these traits. There was increase of 17 to 18cm plant height (cm) of ICPL 87 in 0.2 % EMS treatments (109.33cm) which was significantly higher over its control (91.60cm). The decrease in plant height (cm) of 7-8cm in JA 4 was also observed in 0.3% EMS treatment (131.27cm) as compared to control (138.80cm)(Table 4.15.1).

The time of treatment also affected plant height (cm) which showed increase of about 10cm in ICPL 87 for 8 hours treatment (102.48cm) as compared to control (91.60cm) while in JA 4 there was an decrease about 6.8cm for 8 hours EMS treatment (131.85cm) as compared to control (138.80cm). (Table 4.15.1).

Table 4.15.1: Effect of EMS concentration and time duration on plant height (cm) in pigeonpea genotype.

I Plant height (cm)				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	106.60	141.47	124.30	3.12
0.2%	109.33	132.33	120.83	
0.3%	99.99	131.27	115.59	
Control	91.60	138.80	115.20	
Mean	101.86	125.96	113.91	
CD at 5% (Genotype) = 2.20				
CD at 5% (Genotype x concentration) = 4.41				
Time duration				CD at 5% (Time duration)
4 hours	102.75	139.55	121.15	2.70
6 hours	100.35	136.5	118.42	
8 hours	102.48	131.85	117.16	
Mean	101.86	135.97	152.9	
CD at 5% (G x T) =3.83				

The interaction of concentration x time was significant for this trait. There was increase of 15-16cm shown by 0.1% concentration at 4 hours EMS treatment (130.60cm) which was 13.36% higher than control (115.20cm).(Table 4.15.2).

Table 4.15.2: Effect of EMS concentration and time duration on plant height (cm) in pigeonpea genotype.

Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	130.60	117.80	123.70	124.03
0.2%	127.80	125.10	109.60	120.83
0.3%	111.00	115.60	120.17	115.59
Control	115.20	115.20	115.20	115.20
Mean	121.15	118.43	117.14	118.90
CD at 5% (Concentration x Time duration) =5.40				

The interaction of genotype x concentration x time was significant for this traits. There was an increase of 21cm in 0.2% concentration of 4 hours treatment in ICPL 87 while an increase of 10-11cm was observed in 0.1% EMS of 4 hours treatment (149.40cm) in JA 4 it variety show that genetic concentration of early & medium maturing genotype respond in same direction for plant height (cm) (Table 4.15.3).

Table 4.15.3: Effect of EMS concentration and time duration on plant height (cm) in pigeonpea genotype.

Plant height (cm)				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	111.80	99.60	108.40
	0.20%	112.40	115.00	100.00
	0.30%	95.20	95.20	109.33
	Control	91.60	91.60	91.60
JA 4	0.10%	149.40	136.00	139.00
	0.20%	143.20	135.20	118.60
	0.30%	126.80	136.00	131.00
	Control	138.80	138.80	138.80
	Mean	121.15	118.42	117.17
CD at 5% (G x Con. x Time duration)= 7.65				

4.2.5 Number of pods per plant

For the characteristics of number of pods per plant there was a vice versa effect in varieties under study. An increase was found in early maturing variety ICPL 87 while a decrease was observed in medium maturity variety JA 4 for these traits. There was increase of 20-21 numbers of pods per plant of ICPL 87 in 0.1% EMS treatments (48.20) which was significantly higher over its control (27.40). The decrease in number of pods per plant of 18 in JA 4 was also observed in 0.2% EMS treatment (18) as compared to control (58) .(Table 4.16.1)

The time of treatment also affected number of pods per plant which showed increase of about 22 in ICPL 87 for 8 hours (49.92) while in JA 4 there was an decrease about 13.60 for 8 hours EMS treatment (44.40) as compared to control (58). (Table 4.16.1)

Table 4.16.1: Effect of EMS concentration and time duration on no. of pods per plant in pigeonpea genotype.

I No. of pods per plant				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	48.20	59.53	53.87	0.78
0.2%	47.22	40.00	43.62	
0.3%	45.00	56.07	50.53	
Control	27.40	58.00	42.7	
Mean	41.96	53.40	47.5	
CD at 5% (genotype) = 0.55				
CD at 5% (Genotype x concentration) =1.10				
Time duration				CD at 5% (Time duration)
4 hours	37.30	62.50	49.90	0.68
6 hours	39.37	53.30	46.33	
8 hours	49.22	44.40	46.81	
Mean	41.86	53.40	47.68	
CD at 5% (G x T) =0.96				

The interaction of concentration x time was significant for this trait. There was increase of 14 no. of pods per plant shown by 0.1% concentration at 4 hours EMS treatment (56.70) which was 32.78% higher than control (42.70). (Table 4.16.2)

Table 4.16.2: Effect of EMS concentration and time duration on no. of pods per plant in pigeonpea genotype.

No. of pods per plant				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	56.7	49.1	55.8	53.86
0.2%	47.1	44.9	39.27	43.62
0.3%	53.10	49.03	49.47	50.53
Control	42.7	42.7	42.7	42.7
Mean	49.9	76.3	46.81	57.67
CD at 5% (Con. x Time duration) = 1.35				

The interaction of genotype x concentration x time was significant for this trait. There was an increase of 19-20 in 0.1% concentration of 4 hours treatment in ICPL

87 while it was treatment increase of 13-14 number of pods per plant was observed in 0.1% EMS of 4 hours treatment in JA 4 and 36-37 number of pods per plant decrease for 0.2% EMS concentration at 8 hours treatment (21.40) as compared to control (58). It clearly shows that genetic constitution of early & medium maturing genotype respond for number of pods per plant. (Table 4.16.3)

Table 4.16.3: Effect of EMS concentration and time duration on no. of pods per plant in pigeonpea genotype.

No. of pods per plant				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	46	37.8	60.8
	0.20%	33.2	51.4	57.13
	0.30%	42.6	40.86	51.53
	Control	27.4	27.4	27.4
JA 4	0.10%	67.4	60.4	50.8
	0.20%	61	37.6	21.4
	0.30%	63.6	57.2	47.4
	Control	58	58	58
Grand Mean =		48.43		
CD at 5% (G x Con. x Time duration)= 1.91				

4.2.6 Biological yield per plant (gm)

For the characteristics biological yield per plant (gram) there was a vice- versa effect in variety under study. An increase was found in early maturing variety ICPL 87 while a decrease was observed in medium maturity variety JA 4 variety for these traits. There was increase of 26gm to 27gm in biological yield per plant (gram) of ICPL 87 in 0.2 % EMS treatments which was significantly higher over its control (33.70gm). The decrease in biological yield per plant (54.60gram) in JA 4 was observed in 0.2% EMS treatment (70.6gm) as compared to control (125.20gm) (Table 4.17.1).

The time of treatment also affected biological yield per plant (gram) which showed increase of about 12.96 gm in ICPL 87 for 8 hours (46.66gm) while in JA 4

there was an decrease about 48.70gm of 8 hours EMS treatment (76.50gm) as compare to there control (125.20gm). (Table 4.17.1)

Table 4.17.1: Effect of EMS concentration and time duration on biological yield (gm) per plant in pigeonpea genotype.

Biological yield (gm) per plant				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	59.93	103.93	81.93	0.59
0.2%	37.91	70.60	54.25	
0.3%	31.33	80.60	55.97	
Control	33.70	125.20	79.45	
Mean	32.29	95.08	63.68	
CD at 5% (Genotype) = 0.41				
CD at 5% (Genotype x Concentration) = 0.83				
Time duration				CD at 5% (Time duration)
4 hours	34.43	102.50	68.47	0.51
6 hours	41.08	106.25	73.67	
8 hours	46.66	76.50	61.58	
Mean	40.72	95.08	67.90	
CD at 5% (G x T) = 0.72				

The interaction of concentration x time was significant for this trait. There was a non uniform effect shown by this interaction. There was a decrease of 63.17% of biological yield per plant shown by 0.3% concentration at 4 hours EMS treatment (50.20gm) while it was increase 58.28% for 0.1% EMS concentration of 6 hours treatment (98gm) than control (79.46gm). (Table 4.17.2)

Table 4.17.2: Effect of EMS concentration and time duration on biological yield (gm) per plant in pigeonpea genotype.

Biological yield (gm) per plant				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	82.5	98	65.3	81.93
0.2%	61.70	52.60	48.47	54.25
0.3%	50.2	64.6	53.10	55.97

Control	79.46	79.45	79.45	79.45
Mean	68.46	73.66	61.58	67.9
CD at 5% (Con. x Time duration)	= 1.02			

The interaction of genotype x concentration x time was significant but non uniform for this trait. There was an increase of 30-31gm in 0.1% concentration of 8 hours treatment (63.80gm) while a decrease of 10gm was observed in 0.2% EMS concentration at 4 hours treatment (23.80gm) as compared to control (33.70gm) in ICPL 87. In JA 4 it was increase 9gm in 0.1% concentration of 8 hours (134.20gm) while it was decrease of 78.60gm for 0.2% EMS concentration at 8 hours. It clearly shows that genetic constitution of early & medium maturing genotype respond was distinct for different treatment combinations for biological yield per plant (gram). (Table 4.17.3)

Table 4.17.3: Effect of EMS concentration and time duration on biological yield (gram) per plant in pigeonpea genotype.

Biological yield (gram) per plant				
Genotype	Concentration (%)	Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	54.20	61.80	63.80
	0.20%	23.80	39.60	50.33
	0.30%	26.00	29.20	38.80
	Control	33.70	33.70	33.70
JA 4	0.10%	110.80	134.20	66.80
	0.20%	99.60	65.60	46.60
	0.30%	74.40	100.00	67.40
	Control	125.20	125.20	125.20
Grand Mean =67.897				
CD at 5%(G x Con. x Time duration) = 1.44				

4.2.7 Number of seeds per plant

For the characteristics number of seeds per plant, there was a same effect in variety under study. An increase was found in early maturing variety ICPL 87. There was an increase of 19-40 in ICPL 87 from 0.3% to 0.1% EMS concentration. There was a non uniform effect shown by JA 4 in respect to concentrations. It increase 13 in no. for 0.1% EMS concentration while no. of seeds per plant decreases from 19-27 for 0.2% and 0.3%EMS concentration (Table 4.18.1).

There was an increase of 19-40 in ICPL 87 from 0.3% to 0.1% EMS concentration. In JA 4 it was an increase of 14 number of seeds for 0.1% EMS

concentration (128) while it decrease 27 no. of seeds for 0.3% EMS concentration (87.33). (Table 4.18.1)

Table 4.18.1: Effect of EMS concentration and time duration on no. of seeds per plant in pigeonpea genotype.

No. of seeds per plant				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	91.11	128.00	109.39	2.75
0.2%	81.67	95.67	88.67	
0.3%	69.67	87.33	78.5	
Control	51.00	114.00	82.5	
Mean	73.36	106.16	89.76	
CD at 5% (Genotype) = 1.94				
CD at 5% (Genotype x concentration) = 3.89				
Time duration				CD at 5% (Time duration)
4 hours	58.33	130.5	94.4	
6 hours	51.25	93.75	92.5	
8 hours	70.5	94.25	82.37	
Mean	73.36	106.16	89.76	
CD at 5% (G x T) = 3.37				

The time of treatment also affected number of seeds per plant which showed increase of about 7-20 in ICPL 87 from 4 hours to 8 hours while in JA 4 there was a distinct effect for time durations. It increases 16 for 4 hours treatment while no. of seeds per plant decreases in 19-20 for 6hours and 8 hours EMS treatment.(Table 4.18.1)

The interaction of concentration x time was significant for this trait. Highest no. of seeds per plant was shown by 0.1% concentration at 4 hours EMS treatment (126) which was 52.31% higher than control (83) while 0.2% EMS concentration of 8 hours treatment (72.50) lowest no. of seeds per plant which was 12% lower than control (83). (Table 4.18.2)

Table 4.18.2: Effect of EMS concentration and time duration on no. of seeds per plant in pigeonpea genotype.

II No. of seeds per plant				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	125.67	102.5	100	109.39
0.2%	88.50	105.00	72.50	88.67
0.3%	81.00	80.00	74.50	78.50
Control	82.50	82.50	82.50	82.50
Mean	94.41	92.50	82.38	89.76
CD at 5% (Con. x Time duration) =4.77				

The interaction of genotype x concentration x time was significant for this trait. There was a non uniform result was observed for different combinations. There was 25 no. of seeds decrease in 0.2% concentration of 8 hours treatment while it was 101no. of seeds increase in 0.2% EMS concentration at 6 hours treatment in ICPL 87. There was an increase of 48 in 0.1% EMS concentration of 4 hours treatment and it decrease of 56 in 0.2% EMS concentration of 6 hours treatment in JA 4. It show that genetic constitution of early & medium maturing genotype respond in asymmetric ways for various combinations. .(Table 4.18.3)

Table 4.18.3: Effect of EMS concentration and time duration on no. of seeds per plant in pigeonpea genotype.

No. of seeds per plant				
Genotype	Concentration (%)	III Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	89.33	91	93
	0.20%	26	152	67
	0.30%	67	71	71
	Control	51	51	51
JA 4	0.10%	162	114	107

	0.20%	151	58	78
	0.30%	95	89	78
	Control	114	114	114
Grand Mean = 89.69				
CD at 5%(G x Con. x Time duration) = 6.74				

4.2.8 Seed yield per plant (gram)

For the characteristics seed yield per plant (gm) there was a vice versa effect in variety under study. An increase was found in early maturing variety ICPL 87 while a decrease was observed in medium maturity variety JA 4 for this trait. There was increase of 5-6 gm in seed yield per plant (gm) of ICPL 87 in 0.2 % EMS treatments which was significantly higher over its control (4.56gm). The decrease in 3-4 gm seeds per plant in JA 4 was also observed in 0.2% EMS treatment as compared to control (11.40gm) (Table 4.19.1).

The time of treatment also affected seed yield per plant (gm) which showed increase of about 5-6gm in ICPL 87 for 8 hours while in JA 4 there was an decrease about 2-3 gm for 8 hours EMS treatment. (Table 4.19.1).

Table 4.19.1: Effect of EMS concentration and time duration on seed yield (gm) per plant in pigeonpea genotype.

Seeds yield (gram) per plant				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	7.96	12.65	10.31	1.24
0.2%	9.92	8.28	9.10	
0.3%	5.98	8.70	7.34	
Control	4.56	11.40	7.98	
Mean	7.10	10.25	9.68	
CD at 5% (Genotype) = 0.88				

CD at 5% (Genotype x concentration) = 1.77				
Time duration				CD at 5% (Time duration)
4 hours	5.76	12.83	9.33	1.08
6 hours	7.26	9.36	8.31	
8 hours	8.30	8.59	8.43	
Mean	7.11	10.26	8.68	
CD at 5% (G x T) = 1.53				

The interaction of concentration x time was significant for this trait. Highest seed yield (gm) per plant was shown by 0.1% concentration at 4 hours EMS treatment (10.94gm) which was 37.09% higher than control (7.98gm). (Table 4.19.2)

Table 4.19.2: Effect of EMS concentration and time duration on seed yield (gram) per plant in pigeonpea genotype.

Seeds yield (gram) per plant				
Concentrations	Time durations			
	4 hours	6 hours	8 hours	Mean
0.1%	10.94	10.06	9.92	10.3
0.2%	10.04	8.02	9.24	9.1
0.3%	8.22	7.18	6.64	7.35
Control	7.98	7.98	7.98	7.98
Mean	9.3	8.31	8.45	8.69
CD at 5% (Con. x Time duration) =2.16				

The interaction of genotype x concentration x time was significant for this trait , There was an increase of 9.44gm in 0.2% concentration of 8 hours treatment in ICPL 87 while a decrease of 6- 7 gm seeds per plant was observed in 0.2% EMS of 8 hours treatment in JA 4. It clearly shows that genetic constitution of early & medium maturing genotype respond in opposite direction for seed yield per plant (gm). (Table 4.19.3)

Table 4.19.3: Effect of EMS concentration and time duration on seed yield (gram) per plant in pigeonpea genotype.

Seeds yield (gram) per plant				
Genotype	Concentration (%)	III Time durations		
		4 hours	6 hours	8 hours
ICPL 87	0.10%	7.28	8.20	8.40
	0.20%	5.28	10.48	14.00
	0.30%	5.92	5.80	6.24
	Control	4.56	4.56	4.56
JA 4	0.10%	14.60	11.92	11.44
	0.20%	14.80	5.56	4.48
	0.30%	10.52	8.56	7.04
	Control	11.40	11.40	11.40
Grand Mean =8.68				
CD at 5% (G x Con. x Time duration)= 3.05				

4.2.9 Harvest Index (%)

For the characteristics harvest index (%) there was a same effect in variety under study . An increase was found in early maturing variety ICPL 87 as well as medium maturity variety JA 4 variety for this trait. There was increase of 12-13% in harvest index (%) of ICPL 87 in 0.2 % EMS treatments which was significantly higher over its control (13.42%) while genotype JA 4 did not differ significantly. (Table 4.20.1).

The interaction of genotype x concentration was significant for ICPL 87 while JA 4 did not differ significantly. There was increase of 90.16% in 0.2% concentration of 8 hours (25.52%) as compared to control (13.42%) in ICPL 87. (Table 4.20.1)

Table 4.20.1: Effect of EMS concentration and time duration on harvest index (%) in pigeonpea genotype.

Harvest Index (%)				
Concentration	Genotypes		Mean	CD at 5% (Concentration)
	ICPL 87	JA 4		
0.1%	13.30	13.05	6.65	1.24
0.2%	25.52	11.00	12.76	
0.3%	19.62	11.05	9.81	
Control	13.42	9.12	6.71	
Mean	17.95	11.05		
CD at 5% (Genotype) = 0.88				
CD at 5% (Genotype x concentration) = 1.77				

CHAPTER - V

DISCUSSION

The production of pulses in India is very low which results in low per capital availability of consumption. There is a possibility of importing significant quantity of pulses to fulfill the need, as India itself is the second largest producer and consumer of pulses in the world. Pigeonpea is grown mostly by marginal and small farmers with limited capacity to invest on costly inputs because of this the genetic structure of pigeonpea has been determined more by natural selection than the human selection. In other words, the existing varieties of pigeonpea have been selected for low level of management. The genetic structure of most of the varieties is not good enough to respond positively to better levels of management.

In India, 43 varieties of cereal crops, 38 of grain legumes, 23 of oilseeds, 13 of fiber crops and 10 of millets developed by mutagenesis have been released/approved for cultivation (Kharakwal, 1996). The polygenic characters, viz, high grain yield, early maturity, plant type, quality characters, grain characters abiotic resistance have been improved by mutagenesis (kharwal,1996). These findings provide enough evidence that mutation is a potential tool to be employed for the improvement of qualitative characters of crop plants, provided large populations are raised and vigorous selection procedures are followed. In the backdrop of this, the present investigation entitled "Effect of Ethyl Methane Sulphonate Mutagen in M₁ Generation in Pigeonpea (*Cajanus cajan* L. Millasp)" has been undertaken. The experimental material comprised of pigeonpea genotypes popularly known as JA-4 and ICPL-87. The experiment was conducted by treating seeds of JA-4 and ICPL-87 with 3 concentrations of EMS i.e. 0.1%, 0.2% ,0.3% under the physiological conditions of seeds(i.e. presoaking of seed for 4 hours) for the duration of 4,6 and 8 hours. In M₁ generation observations in field viz, days to flowering, days to maturity, plant height (cm), number of primary branches per plant, number of pods per plant, seed yield (g/plant), biological yield/plant, no. of seeds/plant, 100-seed weight and harvest index were recorded and in laboratory viz. germination percent, plumule length and radical length in three succeeding week. The M₁ generation had been subjected to statistical analysis as per the RBD .The results thus obtained have been discussed in this section under following heads.

Analysis of variance in M₁ generation

Mean performance of M₁ generation

5.1 Analysis of variance in M₁ generation

The highly significant mean sum of squares obtained for all the characters indicated generation of marked genetic variability through different treatments using various concentrations and duration of EMS.

Mean performance of M₁ generation

5.2.1 Germination per cent

The germination per cent, plumule length and radical length was found to be drastically reduced due to mutagenic treatments. The effects of EMS were found to be dose dependent. The maximum reduction was reported in higher dose of treatment. Minimum germinated seeds, plumule length and radical length with the highest treatment dose of 0.3% EMS. These results are in agreement with those of Khan *et.al.*(1994), Singh (1997) *et. al.* and Kumar and Mishra(1999) in green gram and Gaikwad and Kothekar (2004)and Mohd. Rafiq *et al.* (2004) in lentil. The reduced germination due to mutagenic treatment may be attributed to physiological disturbance, particularly due to imbalance and phosphate, amylase, ribonuclease and nucleotidase (Shrivatav, 1973). Delay in the synthesis of DNA and reduced activity of amylase such as catalyses and lipase and disrupt of metabolic pathway concerned with process of germination (Singh, 1974) resulting into killing of cells.

In ICPL 87 in first week genotype ICPL 87 in 0.3% EMS concentration at 8 hours treatment showed lowest germination of 67.60% which was significantly 21.61% lower than control (86.33%). Genotype ICPL 87 in second week 0.3% EMS concentration at 8 hours treatment showed lowest germination of 71.67% which was significantly 21.80% lower than other control (91.66%). Genotype ICPL 87 in third week 0.3% EMS concentration at 8 hours treatment showed lowest germination of 74.66% which was significantly 18.54% lower than control (91.66%).

Data regarding germination per cent has been show that germination per cent in M₁ generation in first week genotype JA 4 in 0.3% EMS concentration at 8 hours treatment showed lowest germination of 71.33% which was significantly 24.11% lower than control (94%). In second week 0.3% EMS concentration at 8 hours treatment showed lowest germination of 75.66% which was significantly 21.45% lower than other control (96.33%). In third week 0.3% EMS concentration at 8 hours

treatment showed lowest germination of 77% which was significantly 20.61% lower than control (97%).

5.2.2 Plumule length (cm)

Data regarding radical length show that genotype ICPL 87 in 0.1% EMS concentration at 8 hours treatment showed lowest plumule length of 1cm which was significantly 2.13cm lower than control (3.13cm). In second week 0.1% EMS concentration at 8 hours treatment showed lowest plumule length of 1.40 cm which was significantly 2.9cm lower than control (4.31cm). In third week 0.1% EMS concentration at 8 hours treatment showed lowest plumule length of 1.88 cm which was significantly 3.22cm lower than control (5.10cm).

Data regarding plumule length show that genotype JA 4 in 0.1% EMS concentration at 8 hours treatment showed lowest plumule length of 0.24cm which was significantly 2.56cm lower than control (3.13cm). In second week 0.1% EMS concentration at 8 hours treatment showed lowest plumule length of 1.10 cm which was significantly 1.59cm lower than control (2.69cm). In third week 0.1% EMS concentration at 8 hours treatment showed lowest plumule length of 1.65 cm which was significantly 2.22cm lower than control (3.87cm).

Singh *et. al.* (2007) studied on the seeds of Indian mustard cultivars Varuna and Pusa Bold treated with gamma radiation (5, 10, 20, 30, 40, 50 and 100 kR) and ethyl methanesulfonate (EMS at 0.01, 0.05, 0.1 and 0.2%) for 6 hrs after 12 hrs presoaking in double distilled water. Callus initiation from cotyledon and plumule explants was observed on MS media containing various concentrations of growth regulators after 11-18 days of incubation from cut ends of the various explants in both the cultivars. Pusa Bold showed early callus initiation compared to Varuna.

5.2.3 Radical length (cm)

Data regarding radical length (cm) had show that Genotype ICPL 87 in 0.1% EMS concentration at 8 hours treatment showed lowest radical length (1.70cm) which was significantly 1.20cm lower than control (2.90cm). In second week 0.1% EMS concentration at 8 hours treatment showed lowest radical length (2.47cm) which was not significantly different than control. In third week 0.1% EMS

concentration at 8 hours treatment showed lowest radical length (2.93cm) which was significantly 0.97cm lower than control (3.9cm).

Genotype JA 4 in first week 0.1% EMS concentration at 8 hours treatment showed lowest radical length (1.22cm) which was significantly 2.51cm lower than control (3.73cm). In second week 0.2% EMS concentration at 8 hours treatment showed lowest radical length (1.75cm) which was significantly 2.65cm lower than control (4.39cm). In third week 0.2% EMS concentration at 8 hours treatment showed lowest radical length (2.36cm) which was significantly 2.28cm lower than control (4.64cm).

Singh and Kole (2005) observed that all the germination and seedling parameters were adversely affected due to the mutagenic treatment. Severe reduction in germination, frequency of normal seedlings, reduction in plumule and radicle lengths and physiological injuries of radicals indicated effective mutagenesis in mung bean.

5.3.1 Days to 50% flowering

Days to 50% flowering is an important parameter for days to maturity of crop added with yield factor. It is required for assessing mutagenic efficiency as early flowering is one of the aims of mutagenic treatment.

Data pertaining to days to 50% flowering in JA 4 have shown that the maximum days to reach 50% flowering (126days) has been exhibited by treatment 0.2% EMS concentration at 4 hours treatment which was 6 days delaying as compared to control (120days).

In ICPL-87 days to 50% flowering treatment 0.3% EMS concentration at 8 hours induced delaying by 6 days as compared to control (120days).

These findings are found to be dose dependent as increase in dose cause delayed flowering. In M_1 generation it may be due to pro-se and persistent vegetative growth or due to mitotic arrest in the flower primordial following mutagenesis. It may be due to inhibition in synthesis of acid or acute damage of chromosome at physiological level of cells According to Kumar and Mishra (1999) in green gram, it is due to mutagen in physiological status of cell along with its rapid metabolic activity. It is also recorded by Kumar and Singh (1982b) in black gram, Navinchandra (1988) in *Gloriosa superben* and Khan *et al.* (1994) in *Vigna radiata*.

5.3.2 Days to Maturity

This character is important as it governs by all previous character and mutagenic efficiency on days to maturity concludes for yield which is basic requirement of mutation breeding.

These experimental found to be dose dependent in case of M₁ generation, which show shift by delay in maturity. In 0.2% EMS concentration at 8 hours treatment show significantly 11 days delay as compared to control (138days) in ICPL 87. In JA 4, it was 6 days late maturity shown by 0.2% EMS concentration at 4 hours treatment (175 days) while 6 days earliness shown by 0.2% EMS concentration at 8 hours treatment (163days) as compared to control(169days).

Change in specific activity of enzymes and inhibition of DNA synthesis may be the probable causes for delay maturity in M₁ generation in present finding. The results are in accordance with finding of Khan (1982) in bean, Srinivas *et al.* (1999) and Gautam and Mittal (1998) in urd and mung bean, Singh *et al.* (1997) and Bhadra (1982) in various pulse crops.

5.3.3 Number of primary branches/plant

The Number of primary branches/plant was recorded at the time of maturity. There was increase in the number of primary branches with respect to control, when all the interaction combination was taken into consideration. An increase of primary branches per plant from 4 to 5 as compared to control (8.80) was observed in 0.1% EMS concentration at 8 hours treatment in ICPL 87 while it was 4 to 5 as compared to control (14.80) was observed in 0.2% EMS treatment of 4 hours treatment (14.8) in JA 4 also.

The decrease in the number of primary branches in present investigation was found to be dose dependent. It was concluded that decreased number may be due to the hazardous effect of the mutagenic treatment causing further reduction in the content of DNA and decreasing the metabolic activity. Such decrease in the number of primary branches in higher doses was reported by Muldhure (1990) in *Vigna mungo* and Mohammad Anis *et al.* (1999) by gamma rays treatment and Kumar and Mishra (1990) by EMS treatment in green gram.

5.3.4. Plant height

The effect of different dose of chemical mutagen (EMS) on plant height was critically examined at maturity. As a result of treatment the plant height there was an increase of 21cm in 0.2% concentration of 4 hours treatment in ICPL 87 while an

increase of 10-11cm was observed in 0.1% EMS of 4 hours treatment (149.40cm) in JA 4 variety. It shows that genetic concentration of early & medium maturing genotype respond in same direction for plant height.

The reduction in the height as a result of high concentration of mutagen as reported in present investigation has been observed by Naveen Chandra (1988) in *Gloriosa superba* through EMS and DES treated material. Similar results were obtained by Tarar and Dnyansagar (1979b) in *Turnera ulmifolia*. In *Vicia faba* and Muldhure (1990) in *Vigna mungo*. Mohammand Anis *et al.* (1999) in *Vigna mungo* observed reduction in height at higher exposure to gamma rays.

The reduction in plant height due to exposure to higher concentration of chemical mutagen may be ascribed many factors resulting in imbalance of hormonal activities and their synthesis.

It is well established that the auxin has rapid turn over rate in metabolically active tissue and that auxin biosynthesis is very sensitive to the mutagens. Tarar and Dnyansagar (1979a) and Gupta and Samata (1967) stated that stunted growth due to chemical mutagens may be attributed to the inhibition of phytochromes responsible for the normal growth at the DNA, RNA level.

5.3.5 Number of seeds/plant

There was an increase in the number of seeds with respect to control to be depending on these all treatment and their combinations. The distinct result was observed for different combinations. There was 25 no. of seeds decrease in 0.2% concentration of 8 hours treatment while it was 101 no. of seeds increased in 0.2% EMS of 6 hours treatment in ICPL 87. There was an increase of 48 seeds per plant in 0.1% EMS concentration of 4 hours treatment and decrease of 56 in 0.2% EMS concentration of 6 hours treatment in JA 4. It showed that genetic constitution of early & medium maturing genotype respond in asymmetric ways for various combinations.

The decrease in number of seeds per plant in present investigation was found to be dose dependent. It was concluded that decreased number may be due to the hazardous in the metabolic activity.

In higher doses, it was reported by Muldhure (1990) in *Vigna mungo* and Mohammad Anis *et al.* (1999) by gamma rays treatment and Kumar and Mishra (1990) by EMS treatment in green gram.

5.3.6 Number of pods per plant

Decrease in number of pods per plant was found to be dose dependent in M₁ generation. There was an increase of 19-20 in 0.1% concentration of 4 hours treatment in ICPL 87 while it increased by 13-14 number of pods per plant in 0.1% EMS of 4 hours treatment in JA 4. It clearly showed that genetic constitution of early & medium maturing genotype respond in same direction for number of pods per plant.

5.3.7 Biological yield per plant (gm)

The polygenic parameter is significant important. There was a vice- versa effect in variety under study.

There was an increase of 30-31gm in 0.1% concentration of 8 hours treatment (63.80gm) while a decrease of 10gm was observed in 0.2% EMS concentration at 4 hours treatment (23.80gm) as compared to control (33.70gm) in ICPL 87. In JA 4 it was increase of 9gm in 0.1% concentration of 8 hours (134.20gm) while it was decrease of 78.60gm for 0.2% EMS concentration at 8 hours. It clearly showed that genetic constitution of early & medium maturing genotype respond was distinct for different treatment combinations for biological yield per plant (gram).

5.3.8 Seed yield per plant (gm)

Critical examination of seed yield had been done after maturity for different doses of chemical mutagen (EMS). This polygenic parameter is significantly important. It is concluding character of all other quantitative characters. There was an increase of 9.44gm in 0.2% concentration of 8 hours treatment in ICPL 87 while a decrease of 6- 7 gm seeds per plant was observed in 0.2% EMS of 8 hours treatment in JA 4. It clearly showed that genetic constitution of early & medium maturing genotype respond in opposite direction for seed yield per plant.

Increased crop yield (cereals grains and several other crops) following mutagen treatments to seeds have reported by Chauhan and Sharma (1974). Khan (1988) in mung bean observed increase the mean values of all three character (number of pods, 100 seed weight and seed yield) after treatment of seeds with gamma rays and EMS used singly or in combination. Singh *et al.* (2001) reported

increase in the pods per plant, seed per pod, 100 seed weight and yield after mutations by gamma rays, EMS and epichlorohydrin in mung bean.

Reduction in yield at higher dose of irradiation and chemical mutagens has been reported by several workers. Kundu and Singh (1982b) in Blackgram, Naveen Chanra (1988) in *Gloriosa superba* with EMS and DES treatments, Mohammad Anis (1999) in *Vigna mungo*, Khan *et al.* (1994) in *Vigna radiata*.

Similar reports had been observed by Premsekhar and Appadural (1981) in pigeonpea who observed stimulation in seed weight in combinations of treatment. These findings are in agreement with Kansgra and Shukla (1993). Hepziba and Subramian (1993) and Patil and Guha (1993-34) in various pulses.

5.3.9 Harvest index

It is an important parameter for assessment of mutagenic effectiveness and efficiency among different yield contributing character in present investigation.

Application of concentration levels and its interaction with genotype ICPL 87 show significant effect on harvest index while JA 4 did not differ for these traits under investigation.

The interaction of concentration x time was significant for this trait. There was increase of 7.25% in 0.2% concentration of 8 hours as compared to 0.1% EMS concentration for 4 hours treatment (72.50).

These findings are in agreement with Kansgra and Shukla (1993). Hepziba and Subramian (1993) and Patil and Guha (1993-34) in various pulses.

Chapter-VI

Summary, conclusion and suggestions for further work

The present investigation on study on chemical mutagenesis through ethyl methane sulphonate in two varieties of pigeonpea viz. JA 4 and ICPL 87 was conducted to assess the mutagenic effects of chemical mutagen on these varieties. The pure healthy and uniform seeds of popular genotype (JA 4 and ICPL 87) of pigeonpea were taken and presoaked for 4 hours and treated with different concentration with 4 hours, 6 hours and 8 hours treatment respectively. Thus a complete package of 10 treatments of 3 concentrations (0.1%, 0.2%, and 0.3%) of each variety with control was planted in 3 replication in petri plate in laboratory conditions and three replicated in field for M₁ generation during Kharif seasons of 2010-11, respectively at the research field of R.A.K. College of Agriculture, Sehore (M.P.) Study on chemo sensitivity for various morphological and yield attributing characters and observations on mutational range were observed in the present investigations. The various observation and results obtained in the investigation have been enumerated here under:

6.1 Summary

- Large amount of genetic variability has been generated through chemical mutagenesis induced by EMS, which may be of immense value for improvement in pigeonpea.
- Germination per cent varied in first week in JA 4 ranged from 71.33 (0.3% EMS concentration at 8 hours) to 94% (control) in JA 4 with mean value of 81.36%, in second week ranged from 75.66(0.3% EMS concentration at 8 hours) - 96.3% (control) with mean value of 84.40% and in third week ranged from 77%(0.3% EMS concentration at 8 hours)-97% (control) with mean value of 86.37% .
- In ICPL 87 in first week ranged from 67.66%(0.3% EMS concentration at 8 hours) to 86.33%(control) with mean value of 77.10%, in second week ranged from 71.66(0.3% EMS concentration at 8 hours)-91.66%(control) with mean value of 83.17% and in third week ranged from 74.66(0.3% EMS concentration at 8 hours)-91.66%(control) with mean value of 85% seeds .

- Plumule length in first week in JA 4 varied from 0.65 cm(0.1% EMS concentration at 4 hours) to 2.8cm(control) while in ICPL 87 it ranged from 1.16cm to 3.31cm in first week. Plumule length in second week varied from 1.10cm(0.1% EMS concentration at 8 hour treatment) - 3.35cm(control) In JA 4 while in ICPL 87 it ranged from 1.40cm(0.1% EMS concentration at 8 hours) - 4.31cm(control) In third week plumule length varied from 1.65cm(0.1% EMS concentration at 8 hour treatment)-4.5 cm(0.2% EMS concentration at 8 hours) in JA 4 while in ICPL 87 it ranged from 1.88cm(0.1% EMS concentration at 8 hours)-5.1cm(control) in third week plumule length .
- Radical length varied from 1.70cm (0.1% EMS concentration at 8 hour treatment) to 3.5cm(0.1% EMS concentration at 4 hour treatment) in ICPL 87 while in JA 4 it ranged from 1.22cm(0.1% EMS concentration at 8 hours) to 4.90cm(0.2% EMS concentration at 6 hours) in first week. Radical length in second week varied from 2.38cm(0.1% EMS concentration at 6 hour treatment) – 4.36cm(0.1% EMS concentration at 4 hour treatment) in ICPL 87 while in JA 4 it ranged from 1.75cm(0.2% EMS concentration at 8 hour treatment) - 4.36cm(control). In third week radical length varied from 2.93 cm (0.1% EMS concentration at 8 hour treatment) - 4.7 cm(0.3% EMS concentration at 4 hour treatment) in ICPL 87 while in JA 4 it ranged from 2.36cm(0.2% EMS concentration at 8 hour treatment) – 6.39cm (0.2% EMS concentration at 6 hour treatment) in third week.
- In JA 4 was 40.47 days to 50% flowering ranging from 111(0.2% EMS concentration at 8 hour treatment) to 126(0.2% EMS concentration at 4 hour treatment) whereas treatment 0.2% EMS concentration at 8 hour treatment showed an earliness of 9 days for 50% flowering as compare to control (120 days).
- In ICPL 87 days to 50% flowering ranging from 100 (control) to 109(0.1% EMS concentration at 8 hour treatment) days whereas treatment 0.1% EMS treatment 8 hours showed an earliness of 9 days for 50% flowering.
- Days to maturity in JA 4 ranged from 163days (0.2% EMS concentration at 8 hour treatment) -175 days (0.2% EMS concentration at 4 hour treatment).

- In ICPL 87 ranged from 138 days (control) -149 days (0.2% EMS concentration at 8 hour treatment) while it was interesting to note that the highest dose of treatment could induce earliness.
- Number of primary branches per plant ranged from 8.20 (0.2% EMS concentration at 8 hour treatment) to 14.80 (0.2% EMS concentration at 4 hour treatment) in ICPL 87 while in JA 4 its ranged from 8.60 (0.1% EMS concentration at 6 hour treatment) to 13.40 (0.2% EMS concentration at 4 hour treatment) in M₁ generation in JA 4 which was found to be does dependent.
- Plant height in JA 4 ranged from 118.60cm (0.2% EMS concentration at 8 hour treatment) to 149.40cm(0.1% EMS concentration at 4 hour treatment) and in ICPL ranged from 112.40cm (0.2% EMS concentration at 4 hour treatment) to 91.60cm(control) which was found to be does dependent.
- Seed index ranged in ICPL 87 9gm to 11 gm while the highest seed index of 11 g was reported with 0.3% EMS concentration at 4 hour treatment.
- In JA 4 from 8.2 to 9.2 g while the highest seed index of 9.2 g was reported with 0.2% EMS concentration at 8 hour treatment and 0.3% EMS concentration at 6 hour treatment, although seed index did not differ under consideration.
- No. of pods per plant ranged from 61 (0.1% EMS concentration at 8 hour treatment) to 27 (control) in ICPL87 while it was 67 (0.1% EMS concentration at 4 hour treatment) to 21(0.2% EMS concentration at 8 hour treatment) in JA 4.
- Biological yield ranged from 23.80 gm (0.2% EMS concentration at 4 hour treatment) to 63.80gm (0.1% EMS concentration at 8 hour treatment) in ICPL 87 while it was 46.60gm (0.2% EMS concentration at 8 hour treatment) to 134.20 gm (0.1% EMS concentration at 6 hour treatment).
- Number of seeds per plant ranged from 26 (0.2% EMS concentration at 4 hour treatment) to 152 (0.2% EMS concentration at 6 hour treatment) in ICPL 87 while it was vice - versa for JA 4. It was 58 (0.2% EMS concentration at 6 hour treatment) to 151(0.2% EMS concentration at 4 hour treatment).
- Seed yield per plant ranged from 4.56 gm (control) to 14 gm (0.2% EMS concentration at 6 hour treatment) while it was 4.48 gm(0.2% EMS

concentration at 6 hour treatment) to 14.8 gm (0.2% EMS concentration at 4 hour treatment).

- The interaction of genotype x concentration was significant for harvest index. JA 4 did not differ significantly. There was increase of 90.16% in 0.2% concentration of 8 hours (25.13%) as compared to control (13.42%) in ICPL 87.

6.2 Conclusion

The chemical mutagenesis through EMS has generated large amount of genetic variability, which can be exploited through further screening and evaluation.

- The concentration of 0.3% EMS is not suitable for pigeonpea as the treated seeds a few germinated.
- The mutagen EMS is capable of producing both types of mutants namely viable and lethal, which is an indication is significant effect on pigeonpea.
- The majority of mutagen treatments induced negative soft in mean away from control for yield and yield components in m_1 generation treatment viz, pre-soaked treated seeds with 0.2% regard to induction of mutation it induced the earliness in maturity by 10 days, increased seed index of to the control along with the yield improvement of 17% in American double dollar variety of pigeonpea.

6.3 suggestions for further work

On the basis of present investigation, suggestions have been made for further work:

- The two types of promising seeds induced by control₂ be evaluates further to develop as variety.
- The EMS mutagen with higher concentration may be tried.
- Other chemical and physical mutagens singly or in combination may be tried to generate more useful variability
- The 0.2% treatment of EMS induced increase in seed no. per plant which is a prime yield attributing character for pigeonpea genotype. The work can be further conducted with various combinations of doses and time.

- The 0.2% treatment of EMS induced increase in seed yield in both genotypes. It shows that lower doses at time combination can induce for addition effect of gene for seed yield. More genotype of population can be affected and evaluated for improvement in pigeonpea
- Some other combination can also be used to manipulate and creation of variability in pigeonpea for improvement of this crop.

REFERENCE

- Abrams, R. and Velez Fortund (1961). Radiation research with pigeonpea (*Cajanus cajan*) results on x_1 and x_2 generations. *J. Agric. Univ. P. Rico.* **45** (4): 197-204.
- Abrams R. (1967) Studies on natural cross pollination in pigeonpea (*Cajanus cajan*). *J. Agric. Univ. P. Rico.* **51** (1): 1-21.
- Abrams, R. and Velez-Fortund (1962). Radiation research with pigeonpea (*Cajanus cajan*): results on x_1 and x_4 generations. *J. Agric. Univ. P. Rico.* **45** (4): 197-204.
- Anonymous (1935) . The gungo or pigeonpea. *Jamaica Agric. Soc.* **39**-330.
- Anonymous (1986). List of varieties. *Mutation Breeding Newsletter* **28**:19-23.
- Barshile, J.D. and B.J. Apparao (2007). Induced variability in chickpea through EMS, SA and gamma radiation. *Journal of Food Legumes.* **20** (1): 38-40.
- Barshile, J.D.; Auti , S.G.; Dalve, , S.C. and B.J. Apparao (2006). Mutagenic sensitivity studies in chickpea employing SA, EMS and gamma rays. *Indian Journal of Pulses Research.* **19** (1): 43-46.
- Bhadra, S. K. (1982). Studies on genetic improvement of black gram through induced mutation. Ph.D. Thesis, IARI. New Delhi.
- Bhatia, C.R. (1977). Mutation breeding of groundnut, rice, and pigeonpea. *Mut. Breed Newsletter.* **9**: 6-7.
- Bhatnagar, C.P.; Handa, D.K. and Alka Mishra (1988). Kabuli chickpea mutants resistant to root-knot nematode, *Meloidogyne javanica* *International Chickpea Newsletter.* (19): 16-17.
- Bhatnagar, P.S.; Sengupta, P.K.; Gangwar; L.C. ; Saxena J.K. and V. Kumar (1967). A fascinated mutant in pigeonpea. *Sci. Cult.* **33**: 120-121.
- Bhatnagar, P.S.; Prabhakar, S.; Tiwari S.P. and J.S. Sandhu (1989). Improvement of soybean variety 'Bragg' through mutagenesis. *Mutation Breeding Newsletter.* (33): 15-16.
- Campaion, B.; Srarvoli, F.; Doria, E. ; Tagliabue, I.; Galasso, I.; Fileppi, M; Bollini R. and E. Nilesonn (2009). Isolation and characterization of an Ipa (low phytic acid) mutant in common bean (*Phaseolous vulgaris* L.). *Theor Appl Gent.* **118**: 1211-1221.

- Chary, S.N. and J.K. Bhalla (1988). EMS induces male sterile mutants in pigeonpea (*Cajanus cajan* L.). *Indian Journal of Genetics*. **48** (3): 303-304.
- Chaturvedi, S.N. and R.P. Sharma (1978). EMS Induced sterile mutants in red gram. *Cur. Sci.* **47**:173-174.
- Chaudhari, B.B. and J.A. Patil. (1953). 'Creeping', A Mutant In Mill sp. *Curr. Sci.* **22**:153.
- Dahiya, B.S. and P.S. Sidhu (1979). No branching (single stem) mutant in pigeonpea (*Canjanus cajan* [L.] millsp.). *Tropical Grain Legume Bulletin*. **15**: 24-25.
- Das, T.R.; Misra, R.C. and P.K. Sahu (2006). Efficiency of mutagenic treatments in expression of macro- and micro-mutations in M₂ generation in greengram and its early predictability on basis of M₁ parameters. *Environment and Ecology*. **24** (2): 283-288.
- Datta, B.K.; Bhattacharya, G.N. and Sudhendu Mandal (1994). Differential susceptibility of two varieties of *Vigna radiata* in response to ethyl methane sulphonate. Environmental pollution: impact of technology on quality of life Proceedings, Santiniketan, India, 21-23 February, 1992: 25-30.
- Deshmukh, N.Y. (1959). Sterile mutants in Tur (*Cajanus cajan*) *Nagpur Agricultural Collage Magazine*. **33**: 20-21.
- Dhole, V.J.; Maheshwari, J.J. and S. Patil (2003). Studies on mutations induced by EMS in soybean (*Glycine max* (L) Merrill). *Agricultural Science Digest*. **23** (3): 226-228.
- Fisher, R.A. (1921). Statistical method for research workers. *Oliver and Boyd. Edinburg*. **26**(2):171-173
- Gaikwad, N. B. and V.S. Kothekar (2004). Mutagenic effectiveness and efficiency of ethyl methane sulphonate and sodium azide in lentil (*Lentil culinaris* Medik.). *Indian Journal of Genetics and Plant Breeding*. **64** (1): 73-74.
- Gautam, A.S. and R.K. Mittal (1998). Induced mutations in blackgram (*Vigna mungo* (L.) Hepper). *Crop Research Hisar*. **16**(3): 344-348.
- Gupta , M.N. and Y. Samta (1967). The relationship between developmental stages of floral buds and somatic mutation induced by acute X rays and chronic gamma irradiation in *Cosmos bipinnatus*. *Radial Bot.* **7**:225-240.
- Hakur, H. L. and G.S. Sethi (1993). Characterization and segregation pattern of some macromutations induced in black gram (*Vigna mungo* L. Hepper). *Indian Journal of Genetics and Plant Breeding*. **53** (2): 168-173.

- Iepziba, S.J. and M. Subramanian(1993). Variability in M₁₃ and M₄ mutants in urd bean . *Indian J. Pulses Res.* **6** (2): 197.
- Iqbal, M; Khan, A. and N. Rohatgi (1989). Germination and growth responses of black gram (*Vigna mungo* (L.) Hepper) to ethyl methane sulphonate (EMS). *Indian Journal of Applied and Pure Biology.* **4** (2): 127-129.
- Jain, H.K. (1976). Induced mutations and improved plant types in pulses .Evaluation of seed protein alterations by mutation breeding . Part 3. Vienna ,Austria : *International Atomic Energy Agency* . pp209
- Jaswani, L.M. and R.B. Deshpande, (1962a). Inheritance studies of some sterile mutants in pigeon pea. *Indian Journal of Genetics.* **22**(3): 236-240.
- Jeswani, L.M and R.B. Deshpande (1962b). Inheritance studies on some sterile mutants in pigeonpea. *Indian Journal of Genetics and Plant Breeding.* **22** (3): 236: 240.
- Joglekar, R.G. and N.Y. Deshmukh (1958). Mutation in pigeonpea (*Cajanus cajan*). *Nagpur Agric. Coll. Mag.* **32**: 23-29.
- Joshi, B.C. and S. Ramanujan (1963 a). Genetics of two mutant in pigeonpea. *Indian J. Genet. Pl Breed.* **23**: 64-66.
- Joshi, B.C. and S. Ramanujam (1963 b). Genetics of two mutants in pigeon pea. *Indian Journal of Genetics.* **23** (1): 64-66.
- Kajjari, N.B. (1956). A new mutation in *Cajanus cajan* Millsp. *Curr. Sci.* **25**: 333.
- Kansagra, U.M. and P.T. Shukla (1993). Variability studies in induced mutants in mung bean. *Madras Agric. J.* **80**(4): 180-184.
- Khan W. M. A .; Sivaswamy, N. and K.R. Ramaswamy (1973). Sensitivity of the red gram (*Cajanus cajan* (L.) Millsp.) Strains to different mutagens. *Madras Agric. J.* **60** (6): 406-407.
- Khan, M. A. W.; Sivaswamy, N. and K. Ramaswamy (1973). Sensitivity of two red gram strains to differant mutagens. *Madras Agric. J.* **60**: 406-407.
- Khan, I. A.(1982). Mutation studies in mung bean (*Vigna radiata* L. Wilczek) variability and genetic advance after selection of quantitative characters. *Bangladesh J.Biol. Sci.* **40**: 11-13.
- Khan, I. A.(1988). Mutation studies in mung bean (*Vigna radiata* L.Wilczek) IX Estimates of genetic variability . *Legumes Res.* **11** (2):89-93.

- Khan, M. A.W. and R. Veeraswamy (1974). Mutations induced in red gram (*Cajanus cajan* [L.] Millsp.) by gamma radiation EMS. *Radiation Botany*. **14**: 237-242.
- Khan, S. and M.R. Wani (2005). Comparison on the effect of chemical mutagens on mung bean. *Advances in Plant Sci*. **18** (2): 533-535.
- Khan, S. and M.R. Wani (2004). Chemomutagenic effect of two alkylating agents on mung bean (*Vigna radiata*) *Bionotes*. **6** (3): 82-83.
- Khan, S.; Siddiqui, A.; and M. Nadeem (1994). Variation in quantitative characters of mung bean after seed treatment with DES. *Advances in Plant Sci*. **7** (1): 41-45.
- Kharakwal, M.C. (2003). Performance and comparison of micro mutational selections of green gram in advance mutation generation for yield and physiological attributes. *Indian J. Genet.*, **53** (4): 445-447.
- Kharawal, M.C (1996). Mutational improvement or plant type in chickpea. *Pulse Crops Newsletter*. **1** (3): 3-4.
- Khorgorde, P.W. (1989). Effect of individual and combined treatment of gamma rays and ethyl methane sulphonate on chickpea (*Cicer arietinum* L) unpublished Ph.D thesis. *Punjabrao Krishi Vidyapeeth, Akola*.
- Kulkarini, G.B.(1973). Pigeonpea (*Cajanus cajan* (L.) Druce), a useful crop for florida. *Proce. Soil Crop Sci. Soc. Fla.* **28**: 162-167.
- Kumar, A; Pramahansh P. and R. Prasad (2009). Induced chlorophyll and morphological mutations in mungbean (*Vigna radiate*:Wilezek) *Legumes Res*. **32** (1): 4145.
- Kundu, S.K. and D.P. Singh (1982 a). Note on gamma rays induced variability for flowering and chlorophyll mutants in black gram. *Indian J. Agric Sci.* **52** (3): 190-191.
- Kundu, S.K. and D.P. Singh (1982b).Gamma rays induced variability for quantitative characters in black gram. *Madras. Agric. J.* **69** (10): 644-648.
- Li, W.C. (1988). Mutagenic effects of ethyl methanesulphonate, sodium azide and combination of both on soybean seeds. *Journal of Agriculture and Forestry*. **37**(2): 107-112.
- Malic, I.A.(1988). High yielding and early maturing mutant in mung bean ((*Vigna radiate* L.Wikzek). *Mutation Breeding Newsletter*. **32**: 7-8.

- Mehetre, S.S., Desmukh, R.B., and R.G. Rodga, (1983). Effect of gamma-rays on different growth and economic characters in pigeonpea. *Indian Journal of Heredity*. **13**: 19-23.
- Mohamad , A. ; Sharma P. K. and Abbasi Nagma (1999). Gamma rays induced variability in urd bean (*Vigna mungo* (L) Hepper) M generation *J. Indian Bot. Soc.* **78**: 111-113.
- Mohamad R.; Samiullah, W. and Kouser Parveen (2004). Mutagenic effects of EMS in lentil (*Lens culinaris* Medik) *Madras Agric. J.* **64** (9) : 61-64.
- Mohamed S. N. and R. Veeraswamy.(1977). Genotypic and phenotypic variability of mutants in red gram (*Cajanus cajan*(L.) Millsp). *Madras Agric.J.* **64** (1): 44-45.
- Mohamed S. N.; Khan M.A.W.; and R.S. Annappan (1977). Red gram Co.3-an economic mutant strain for Tamil Nadu. *Madras Agric. J.* **64** (9): 561-564.
- Morita, R.; Kusaba, M.; Tide, S.; Nishion, T. and M. Nishimura (2009) Development of PCR marker to detect the *glb1* and *lgc1* mutation for the low easy to digest protein rice varieties. *Theor Appl Genet.* **119**: 125-130.
- Muldhure, B.D. (1990). Cytogenetic studies in (*Vigna mungo* (L) Hepper) Ph. D. thesis, Submitted to Nagpur University, Nagpur.
- Nanda, S.N.; Sahu , A. ; Panda J.M. and N. Senapati (1997). Effect of ethyl methane sulphonate (EMS) on asparagus bean (*Vigna sesquipedalis*). *ACIAR Food Legume Newsletter.* (25): 6-8.
- Natarajan, N.; Ramalingam, S.R., and M. Sivaswamy, (1983). Induced variation in quantitative characters in red gram. *Madras Agric J.* **70**: 219 – 222.
- Naveen Chandra (1988). Cytological and mutational studies on *Gloriosa superba* Linn Ph.D. thesis submitted to Nagpur University, Nagpur.
- Okubara, P.A.; Steber, C.M. ; SeMecom, V.L.; Walter, N.I.; Paulitz T.C.and K.K. Kidwell (2009). Scarlet Rz 1, an EMS- generated hexaploid wheat with tolerance to the soil borne necrotropic pathogens. *Rhizoctonia solani* AG-8 and *Rizoclonia oryzae*. *Theor Appl Genet.* **119**: 293-303.
- Pandey, N.; Ojha, C.B.; Jha V.B. and N.B. Singh(1996). Effect of chemical mutagens on the rate of germination, seedling mortality, and induced sterility in pigeonpea. *International Chickpea and Pigeonpea Newsletter.* (3): 65-67.

- Pandya, P.S., Pati, J.A. and B.B. Chaudhary (1954). Round and tiny leaf mutant in *Cajanus cajan* Millsp. *Puna Agric. Coll. Mag.* **45**: 18.
- Panase, V.G. and P.V. Sukhatme (1967). Statistical method for agricultural workers. ICAR New Delhi, pp 67-69.
- Pathak G.N. and K.P. Singh (1964). A new type of mutant in pigeonpea (*Cajanus cajan* (L.) Millsp.). *Sci Cult.* **30**: 397-398.
- Patil A.; Taware S.P. and V.M. Raut (2004). Induced variation in quantitative traits due to physical (gamma rays), chemical (EMS) and combined mutagen treatments in soybean [*Glycine max* (L.) Merrill]. *Soybean Genetics Newsletter.* (31): 1-6.
- Patil, B.N. and V.Y. Guha (1993-94). Growth attributes in some induced mutants of chickpea (*Cicer aritinum* L.). *PKV Res. J.* **15**(2) : 97-101.
- Patil, J.A. (1959). A mutation in *Cajanus cajan* (L.) Millsp. *Puna Agric. Col. Mag.* **49**: 264.
- Premshakar, S. and P. Appadura (1981). Effect of doses of Gamma rays and induced mutation in pigeonpea. *Indian J. Agric. Sci.* **51**: 381-384.
- Rakshit; S. and V.P. Singh (2001). Chemosensitivity studies in mung bean and urdbean. *Indian Journal of Pulses Research.* **14**(2): 112-115.
- Pawar, S.E.; Thakare, R.G.; Mitra, R.; Krishna, T.G. and C.R. Bhatia (1984). Induced mutations for the genetic improvement of pulse crops. Pages 361-367 in Pulse production - constraints and opportunities (Srivastava, S.C., B. Bhaskaran, S., Menon, K.K.G., Ramanujam, S. and Rao, M.V., eds). New Delhi, India: Oxford and IBH Publishing Co.
- Rao, C.H. (1974). Studies on induced variability in pigeonpea and chickpea. Ph. D. Thesis, *Indian Agricultural Research Institute, New Delhi, India.*
- Rao, D.M. and Reddy, T.P. (1983). Response of pigeonpea cultivars to mutagens. *International Pigeonpea Newsletter.* **2**: 15-16.
- Rao, D.M., and Reddy, T.P. (1986). Induced polygenic variability in *Cajanus cajan* L. *All India Congress of Cytology and Genetics.* (5): 303-309.
- Rao, D.M. and T.P. Reddy (1993). Induction and evaluation of certain beneficial mutants in *Cajanus cajan* L. *Advances in Plant Sci.* **6** (2): 246-259.
- Rao, V.S.; Rao, S. V.; Singh B.S. and C.A. Jagadish (1986) Heritability studies on EMS treated green gram (*Vigna radiata* (L.) Wilczek). *Journal of Research APAU.* **14** (2): 141-143.

- Rao, V.S., Singh B.S. and C.A. Jagdish (1986). Recurrent EMS treatment of green gram (*Vigna radiata* L.) *J. Res APAU*. **14** (2): 129-133.
- Sarker, A. and B. Sharma (1988). Efficiency of early generation selection for induced polygenic mutations in lentil (*Lens culinaris* Medik). *Indian Journal of Genetics and Plant Breeding*. **48**(2): 155-159.
- Senapati, N. and R.C. Mishra (2010). Genetic Divergence and variability studies among micro mutants in black gram (*Vigna mungo*) Hepper). *Legume Res.* **33** (2): 108-113.
- Sharma, A. and P. Talukdar (1999). Sensitivity of green gram (*Vigna radiata* L. Wilczek) to physical and chemical mutagen. *Journal of Interacademia*. **3** (1): 11-18.
- Sharma, D. and M.P. Shrivastava (1974). An induced useful mutant *Cajanus cajan* (L.) Millsp. *JNKVV Res. J.* **8** (3-4): 263-266.
- Sharma, R.P. and H.C. Bansal, (1970). Influence of radiation and chemically induced sterility in mutation frequency and spectrum in barley. *Indian J. of Genet.* **30**: 544-500.
- Shrivastava, L. S.; Chand H. and S. Kumar (1973). Dose response studies on EMS and MMS treated gram. *Sci. and Cul.* **39** (8): 346-347.
- Shrivastava, M.P. (1975). Effect of gamma irradiation on diploid and tetraploid seeds of *Cajanus cajan* (L.) Millsp. *Curr.Sci.* **44** (5):167-168.
- Singh G.; Sareen P.K. and R.P. Saharan (1997) Mutation studies in mungbean (*Vigna radiata* L. Wilczek). *Journal of Nuclear Agriculture and Biology*. **26** (4): 227-231.
- Singh R. and C.R. Kole (2006). Delineation of EMS-induced genetic variability in some agronomic traits in mungbean [*Vigna radiata* (L.) Wilczek]. *Crop Research, Hisar*. **32** (1): 94-96.
- Singh, A.K. and A.K. Singh (2007). Biological influence of gamma rays and ethyl methane sulphonate and their synergistic effects in mungbean *Journal of Food Legumes*; **20** (2): 153-155.
- Singh, A.K.; Diwakar, M.C.; Singh J.K. and J. Singh (1993). Mutagenic responses of mungbean (*Vigna radiata* L. Wilczek). *Journal of Applied Biology*. **3** (1/2): 75-79.
- Singh, B.B.(1973). Effect of gamma-irradiation on growth and yield of pigeonpea (*Cajanus cajan*). *Hort. J. Sci.* **2** (3-4):83-88.

- Singh, B.B.; Gupta S.C. and B.D. Singh (1974). Note on 'UPAS-120' an early maturing mutant of pigeonpea . *Indian J. Agric. Sci.* **54** (4): 233-234.
- Singh, M. and V.P. Singh (2001). Genetic analysis of certain mutant lines of urdbean for yield and quality traits in M4 generation. *Indian Journal of Pulses Research*; **14** (1): 60-62.
- Singh, R. and C.R. Kole (2005). Effect of mutagenic treatment with EMS on germination and some seedling parameters in mung bean. *Crop Research*, Hisar. **30** (2): 236-240.
- Singh, R. R.; Prasad ,B.K. and H.B. Singh (2007). Development of Alternaria blight tolerant lines through somaclonal variants in Indian mustard (Brassica juncea (L.) Czern and Coss). *Brassica* **9** (1/4): 21-27.
- Singh, R.H. and S.S.Raghuvanshi (1991). Bold seeded mutant in black gram. *Mutation Breeding Newsletter*. **38**: 5-6.
- Singh, S.P.; Singh,.;N.K; Singh R.P. and J.P. Prasad (2007). Effect of mutagens on mutation frequency and spectrum in lentil. *Journal of Food Legumes*. **20** (1): 33-37.
- Singh, S.P.; Singh, N.K.; Singh R.P. and J.P. Prasad (2006). Mutagenic effect of gamma rays and EMS on nodulation, yield and yield traits on lentil. *Indian Journal of Pulses Research*. **19** (1): 53-55.
- Singh, S.N. and S.B. Akhauri (1969). Effect of gamma radiation and E.M.S. on *Cajanus cajan*. *Proc. Indian Sci. Cong. Assoc.* **56** (3): 348-349.
- Singh, V. P.; Singh, M. and J.P.Lal (2001). Gamma ray and EMS induced genetic variability for quantitative traits in urdbean (*Vigna mungo* L. Hepper). *Indian Journal of Genetics and Plant Breeding*. **60** (1): 89-96.
- Singh, V.P. and R.D.S. Yadav (1991). Induced mutations for qualitative and quantitative traits in green gram (*Vigna radiata* L. Wilczek) cv. T 44. *Journal of Genetics and Breeding*; **45** (1): 1-5.
- Srinivas, P.; N. Hual alai; S. Saengehot; S. Nyamponsal; Peerasak Srinivas; Napopon Hual-alai; Saengehot Supavinee Nyamponsal; Sumana; K. Oono (ed.) and S. Miyazakji (1999). The use of wild relatives and gamma radiation in mung bean and black gram breeding. The seventh Ministry of Agriculture, Forestry and Fisheries (MAFF), Japan International Workshop on Genetic Resources, Ibaraki, Japan, 13-13 Oct., Part I, *Wild Legumes*, 205-218.

- Srivastava, A. and V.P. Singh (1996). Induced high yielding pigeonpea mutants. *Mutation Breeding Newsletter*. **42** (8-9).
- Tarar, J.L. and V.R. Dnyansagar (1979 a). Meiotic studies in *Tumera ulmifolia* Linn. Var. angusifolia wild . *The J. Ind. Bot. Soc.* **58** (2): 167-175.
- Tarar, J.L. and V.R. Dnyansagar (1979 b). Studies of ethyl methane sulphonate and gamma rays induced sterility *Tumera ulmifolia* L. *J. Cytol. Genet.* **14**: 124-130.
- Tickoo, J.L. and N. Chandra(1999). Mutagen induced polygenic variability in mungbean (*Vigna radiata* Wilczek L.). *Indian Journal of Genetics and Plan Breeding.* **59** (2): 193-201.
- Tripathy, S.K. (1990). Effect of mutagenic treatments on micro mutations in mungbean (*Vigna radiata* (L.) Wilczek.). *Orissa Journal of Agricultural Research.* **3** (3-4): 284-287.
- Tyagi, B.S. and P.K. Gupta (1991). Induced macro mutation in lentil. *Lens.* **18** (1-2): 3-7.
- Vanniarajan, C.; Vivekanandan P. and J. Ramalingam (1996). Induced variability for quantitative characters in black gram. *Crop Research Hisar*, **11** (3): 341-346.
- Venkateswarlu, S., Singh, R.M. and R.B.Singh (1976). EMS induced multi acarpelate condition in *Cajanus cajan*. *Curr. Sci.* **45** (2): 773-774.
- Venkteswarlu, S.; Singh, R.M.; Singh, R.B. and B.D. Singh (1978). Radio sensitivity and frequency of chlorophyll mutations in pigeonpea. *Indian Journal of Genetics.* **38**: 90-95.
- Waghmare, V. N. and R.B. Mehra (1998) Mutagenic sensitivity of gamma rays and ethyl methane sulfonate in *Lathyrus sativus* L. *FABIS-Newsletter.* (41): 8-12.
- Wani,M.R.(2004). Effect of EMS on seed germination and pollen fertility in mung bean. *Bionotes.* **6** (2): 56.
- Wanjari, .B., Khadilkar, B.T., and D.R. Kutarekar, (1978). Spontaneous variants in tur. *Nagpur Agriculture Collage Magazine* **50**: 41-42.
- Wilhelm E.P.; Turner A.S. and D.A. Lauric (2009). Photoperiod insensitive Ppd_Ala mutations in tetraploip wheat (*Triticum durum* desf.) *Theor Apl Genet.* **118**: 285-294.

Wongpiyasatid, A.; Chotechuen, S.; Hormchan, P.; Ngampongsai, S.; Lamseejan S. and S. Pichitporn (1998). Mutant mung bean lines from radiation and chemical induction . *Journal Natural Sciences*. **32** (2): 203-212.

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