# INDUCED MUTAGENESIS FOR EARLINESS IN GROUNDNUT (Arachis hypogaea L.)

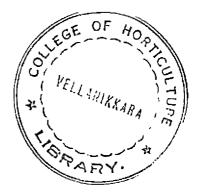
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## THESIS

submitted in partial fulfitment of the requirement for the degree MASTER OF SCIENCE IN AGRICULTURE Faculty of Agriculture Kerala Agricultural University

> Department of <u>Plant Breeding</u> COLLEGE OF AGRICULTURE Vellayani, Trivandrum



## DECLARATION

I hereby declare that this thesis entitled "Induced mutagenesis for earliness in groundnut (Arachis hypogaea L.)" is a bonafide record of research work done by me and that the thesis has not previously formed the basis for the award to me of any degree, diploma, associateship, fellowship or other similar title, of any other University or Society.

GIGI K.JOHN

Vellayani, 28-5-1987.

#### CERTIFICATE

Certified that this thesis, entitled "Induced mutagenesis for earliness in groundnut (<u>Arachis hypogaea</u> L.) is a record of research work done independently by Kumari.GIGI K.JOHN under my guidance and supervision and that it has not previously formed the basis for the award of any degree, fellowship or associateship to her.

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## INTRODUCTION

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## INTRODUCTION

Groundnut is an important oilseed crop of India and it occupies an area of 7.5 million hectares with a production of about 6.5 million tons (Anon., 1985). Besides oil (43-55%), groundnut kernels are rich in protein (25-28%) and vitamins B and E which makes a substantial contribution to human nutrition. In Kerala. groundnut is cultivated in an area of 11,824 hectares with an annual seed production of 11,768 tons (Anon., 1987) and is mostly grown in the drought prone area of Chittoor taluk in Palghat district. Nair (1978) pointed out that in Kerala there is considerable scope for cultivating groundnut in non-traditional areas as intercrop in coconut gardens, companion crop with tapioca and catch crop during third crop season (Summer) in double crop paddy fields, where there is a fallow period of nearly 90 days. He emphasised the necessity for evolving short duration varieties suited to the rice fallows. The results of crop sequence trials conducted at the Rice Research Station. Kayamkulam proved that groundnut can be grown profitably in the rice fallows (Anon., 1978). The cultivation of this crop is very limited in high rainfall regions of other districts during Kharif season because of the

non-availability of suitable high yielding varieties with seed dormancy and earliness. Trials conducted under the ICAR adhoc scheme at Mannuthy during Kharif seasons 1981, 1982 and Summer season 1982 (Anon., 1983) and in summer rice fallows of Onattukara and Kharif uplands of Vellayani (Radhika, 1984) revealed that the variety EC.119704 is high yielding with long (120 days) duration. In addition to the proven superiority in yield, this variety possesses fresh seed dormancy. The need for a variety with earliness and dormancy is badly felt for extending the cultivation of this crop during the Kharif season in the high rainfall regions of the State and in summer rice fallows.

Mutations are sudden heritable changes consisting of genetic alterations, ranging from single base substitution within the DNA to gross changes in chromosome structure. This can be used as a potent source for useful variants in the biological system either directly or after recombination. Mutation breeding is the best method for making simple corrections in an otherwise desirable variety. By induced mutagenesis, characters like earliness and resistance to certain diseases can be introduced in otherwise well adapted varieties

without significantly altering their own attributes (Sigurbjornsson and Micke, 1969).

According to Gregory (1968), modern techniques involving radiation treatments form an integral part of the genetic improvement of oil seed crops. In groundnut, induction of mutations has proved to be a good means for obtaining useful genetic diversity and the crop has an adequately balanced system of genetic units, capable of polydirectional changes (Gregory, 1956a). Ashri and Goldin (1965) stated that the advantages and disadvantages associated with polyploidy are felt in groundnut. The highly autogamous nature of the crop with delicate floral structures and the prevalence of genetic breakdown in recombination breeding favours the mutational approach in this crop. Misra (1980) suggested induced mutagenesis as a tool for breeding for earliness in groundnut. Therefore. a study was undertaken with the objective of induction and isolation of viable mutations with earliness, high yield and other desirable attributes in the dormant groundnut variety EC.119704 using gamma rays. The evolution of such a mutant variety will go a long way in increasing the production of groundnut in this State by cultivating them in kharif uplands and in the nontraditional but potential areas in summer rice fallows.

# **REVIEW OF LITERATURE**

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#### REVIEW OF LITERATURE

Induction of mutation is an additional tool for the improvement of crop plants, Mutation technique has been very useful in creating new variability which is an essential requirement for any crop improvement programme. The idea of inducing mutations with x-rays and utilising it in breeding new forms was proposed as early as 1909 by Devries. Subsequently the installation of cobalt and caesium sources made gamma rays available for use in the induction of mutations. The usefulness of artificially induced mutations in breeding programmes was recognised by many biologists even before Muller who established in 1927, that mutations could be induced in Drosophila. Stadler in 1928 discovered that x-rays could induce mutations in plants. Sigurbjornsson and Micke (1969) in a detailed analysis of the specific role of induced mutations have clearly projected out mutation breeding as one of the indispensable methods of plant breeding.

## 2.1 <u>Scope of induced mutagenesis in groundnut</u>

Induction of mutation has been proved to be a good means for obtaining useful genetic diversity in groundnut (Gregory, 1956a) and a potent source of genetic variability for certain specific characteristics (Norden, 1973). According to Hammons (1973) despite a long history of cultivation, broad subspecific variability and wide geographic distribution of the cultivated peanut, defects in its composition with respect to the requirements of man are widespread and for correcting many of these defects, no genetic resources are known to exist among the available varieties. Therefore, induced mutagenesis could be used as a potent genetic tool for developing new genes for various characters.

The groundnut plant is different from others due to the fact that it possesses a combination of pereniating habit and subterranean pod bearing nature which essentially promote survival than productivity as evidenced by its long evolutionary history (Prasad and Kaul, 1980a). The delicate floral structures, restricted recombination and hereditary instability are some of the factors frustrating the breeders efforts through conventional breeding methods. Added to these are the limited natural variability for the economic characters (Rathnaswamy, 1980). Prasad and Kaul (1980b) stated that a combination of breeding methods involving induced genetic variability and its further use by combination breeding would be more useful in groundnut improvement. Attempts for groundnut improvement by induced mutagenesis have been numerous (Gustaffsson and Gadd, 1965; Gregory, 1968; Norden, 1973).

#### 2.2 <u>Mutagens</u>

Both physical and chemical mutagens were employed by several groundnut mutation workers throughout the world, the most notable being the North Carolina group which include Gregory, the most eminent groundnut mutation breeder. Gamma irradiation was employed by Shivaraj and Rao (1963), Sanjeeviah (1967), Mouli and Patil (1976), Reddy et al. (1977), Sinha and Rahman (1979), Sivasubramonian (1979), Prasad and Kaul (1980b), Ratnaswamy (1980), Marghitu et al., (1982), Pushkaran (1983), Ramanathan (1983), Pathirana (1985) and Dutta et al. (1986) for the improvement of groundnut. Physical mutagens such as neutrons and x-rays were employed by Cooper and Gregory (1960), Bilquez and Martin (1961), Shivraj et al. (1962), Patil and Bora (1963), Patil (1966), Sanjeeviah (1967), Arzumanova (1970), Menon et al.(1970) and Sivasubramonian (1979).

Different chemical mutagens were used by various workers such as Ashri and Goldin (1965), Ashri and Levy (1974), Sivasubramoniam (1979), Prasad " and Kaul (1980b), Ramanathan (1983) and Sivaram et al. (1985a). However, the spectrum of mutagens utilised was very much limited when compared to the larger number of mutagens employed in cereals. The studies

designed to compare different kinds of mutagens are also few. Ashri and Goldin (1965) observed that diethyl sulphate is an efficient mutagen in peanut despite its amphidiploid nature. Ashri and Levy (1974) reported that gamma rays gave a higher mutation rate than EMS. Differential response among the chemical mutagens Ethyl methane sulphonate and diethyl sulphate was reported by Ashri and Herzog (1972).

Radiosensitivity in groundnut is greatly influenced by external conditions, particularly the relative humidity. A large variation in  $x_1$  effect from seed to seed results unless the environment of the dried groundnut seeds are well controlled. (Gregory 1956b). Among the various factors studied, moisture content, oxygen concentration and relative humidity were found to produce large variations in their effects on radiosensitivity in groundnut.

## 2.3 M<sub>1</sub> effects

Toxic effects produced by mutagen treatment on biological systems are expressed in the M<sub>1</sub> generation, the lower doses do not show any severe effects but higher doses produces gross visible disturbances. These disturbances will be expressed as reduction in germination, survival and fertility, growth inhibition,

chlorophyll chimeras and morphological variations.

2.3.1 Germination

The effect of mutagen on germination has been reported to be a reliable estimate of seedling lethality by several workers. Bilquez and Martin (1961) reported that in groundnut irradiation with x-rays at 8000 R stimulated germination but at 40,000 R, the germination was reduced. The germination percentage of groundnut seeds irradiated with 10 to 25 krad x-rays and gamma rays was found to decrease from 95 to 43% and 91.3 to 73.1% respectively as against 98% in the control (Sanjeeviah et al., 1967), Sivasubramonian (1979) found a progressive reduction in germination with increasing doses of gamma rays in the three varieties of groundnut studied viz. Pollachi-2, TMV-7 and TMV-11. Similar results were reported by Pushkaran (1983) in groundnut treated with gamma rays. Reduced germination was reported by Dorairaj (1979); Ratnaswamy (1980) and Ramanathan(1984a) following gamma irradiation in groundnut.

## 2.3.2 Survival

Survival count is a better estimate of lethality as it accounts for post germination lethality also. The relationship between doses of mutagens and survival of plants was studied by several workers. Gills and De Vinok (1959) studied the effect of x-rays on excised embryos as well as open seeds of groundnut presoaked in water for 48 hours. They have tried six doses from 1000 to 25,000 r and the LD 50 for survival was found to be between 2000 to 5000 r. Bilquez and Martin (1961) reported a high survival rate after the application of high doses of (40,000 R) x-rays in groundnut. Following gamma ray treatment survival reduction of groundnut plants and a linear dose effect relationship was noticed by Sanjeeviah (1967), Sivasubramonian (1979), Ratnaswamy (1980), Push@karan (1983) and Ramanathan (1984a).

## 2.3.3 Plant height

Rate of reduction in plant height has been used as an estimate of injury in several radiobiological experiments. Sanjeeviah et al.(1967) reported that both x-rays and gamma ray effectively reduced the plant height in groundnut with increasing doses. The average height, spread of plant and length and breadth of leaves were found to be a little higher in the dose rate of 25 kr gamma ray than its immediate lower doses. Injury of various types resulting from gamma irradiation were reported by many workers in groundnut, the most common being a reduction in plant height(Shivraj and

Rao, 1963; Sinha and Roy, 1969; Sivasubramoniam, 1979; Ratnaswamy, 1980; Pushkaran, 1983; and Ramanathan, 1984a).

#### 2.3.4 Fertility

Increased pollen sterility consequent to mutagenesis was reported to be mostly dose dependant (Gregory, 1968; Mouli and Patil, 1976; Sivasubramonian, 1979; Pushkaran, 1983 and Ramanathan, 1984a). Dutta et al. (1986) studied the effect of gamma rays (5 to 20 kr) on pollen sterility in groundnut. They have observed that pollen sterility increased almost linearly with increased dose.

#### 2.3.5 Chimeras

Gregory (1968) reported chimeric plants with both variegated and normal branches. Habib et al.(1980) observed a male sterile mutant in groundnut. They have also noticed that one branch of this mutant was bearing normal flowers having 9.6% pollen sterility only, indicating that the mutant was a sectorial chimera. In the  $M_1$  generation of gamma irradiated groundnut, plants with chlorophyll deficient sectors were observed by several workers.(Sanjeeviah (1967), Sivasubramonian; (1979), Dorairaj(1979), Pushkaran(1983) and Ramanathan (1984a).

#### 2.3.6 Morphological variations

Ashri and Goldin (1965) reported the occurrence of nonheritable morphological changes in groundnut in the M<sub>1</sub> generation following diethyl sulphate treatment. Changes in growth habit, stem thickness and other morphological characteristics following irradiation of groundnut seeds with x-rays and gamma rays were reported by a number of investigators (Gregory 1968; Arzumanova, 1970 and Pushkaran, 1983).

## 2.4 Mutations in the M2 generation

## 2.4.1 Chlorophyll mutations

## i) Frequency

Chlorophyll mutation frequency can be estimated as mutations per 100  $M_1$  plants, mutations per 100  $M_2$ inflorescence and mutations per 100  $M_2$  plants. Among these, mutations per 100  $M_2$  plants is considered as the best index since it bears a proportionate relationship to the initial rate of mutation. Moreover it is independent of the variations in progeny size and the size of the mutated sector (Gaul, 1960). Levy and Ashri (1975) noticed that ethyl methane sulphonate induced chlorophyllmutations in groundnut were most frequently followed by plant size mutations. Sivasubramaniam (1979) reported that chlorophyll mutation frequency

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in groundnut showed an increase with the increasing dose of gamma rays. Further, the frequency of chlorophyll mutations were found to vary significantly among the different varieties. Ramanathan and Rathinam (1983b) observed a low frequency of chlorophyll mutation in groundnut following gamma irradiation. The chlorophyll mutation frequency estimate on  $M_1$  plant basis and  $M_2$ plant basis in gamma irradiated groundnut varieties Spanish Improved, TG3 and TG 14 revealed that the chlorophyll mutation frequency increased with increase in the dose of mutagen (Pushkaran, 1983).

#### ii) <u>Spectrum</u>

Considerable variation in the spectrum of chlorophyll mutants were observed by several investigators. Patil and Bora (1963) described xantha and virescent mutants in groundnut after x-irradiation. Gregory (1968) recorded chlorophyll mutants including 'light green' and 'albino' following x-ray treatment in groundnut. Sinha and Roy (1969) isolated a mutant named (Virescent) from the  $M_2$  generation of TMV-1 variety of groundnut treated with 30 kr gamma rays. Patil (1973) reported 'chlorina' as a new chlorophyll deficient character in groundnut. Pushkaran (1983) observed different types of chlorophyll mutants, such as albino, xantha, chlorina

and viridis in the M<sub>2</sub> generation of gamma irradiated groundnut varieties. Ramanathan and Rathinam (1983b) reported that the spectrum of chlorophyll mutations following mutagen treatments vary according to the dose of mutagen and variety.

#### iii) <u>Segregation ratio</u>

Studies on Patil and Bora (1963) on segregation ratio could reveal the nature of genes controlling chlorophyll development. They have noticed the occurrence of one xantha and one virescent mutant after x-irradiation in groundnut. The plant progenies in the x<sub>5</sub> generation segregated for normal and virescent types in ratios ranging from 1:1 and 15:1 indicating that the development of chlorophyll in groundnut is possibly controlled by more than one locus. Sinha and Roy (1969) reported a phenotypic segregation ratio of 28 virescent : 17 normal making it difficult to assess the exact genetic nature of virescent mutation. Srivastava (1970) observed two spontaneous mutants with mottled leaf in the groundnut variety C.501. The progeny of both the mottled plants were found to segregate into 15 mottled: 3 normal (1st plant) and 21 mottled: 12 normal (2nd plant), the total being 36 mottled: 15 normal which approximates to a 3:1 ratio

denoting monogenic inheritance and the dominance of mottled leaf to normal one.

#### 2.4.2 Viable mutations

Quite a large number of viable mutations have been reported by groundnut mutation breeders. Gregory (1956a) x-ray irradiated about one lakh seeds and in the  $R_2$ , a large number of mutants were observed. Economically viable mutants in groundnut including a non-dormant mutant worthy of utilisation was observed by Prasad and Kaul (1980b) following gamma irradiation. Mutations affecting more than one character of the same plant was reported in several investigations. These changes are reported to be inherited as a single unit of recombination. The Israeli breeders, Ashri and Goldin (1965) found major deviations in growth habit affecting more than one character following diethyl sulphate treatment. They observed mutations involving two or three characters at a time and suggested that this may be a pleiotropic mutant with a syndrome effect. Pushkaran (1983) has reported a variety of viable mutants affecting one or a constellation of morphological characters. Two mutants with semispreading growth habit and short stature having bold seeds, more number of pods.

high seed yield and shelling percentage were obtained by gamma irradiation of the TMV.9 variety of groundnut (Ramanathan and Rathinam, 1983a). Tiwari and Khanorkar (1984) reported two miniature mutants which were shorter producing few pods, small seeds and leaflets in comparison to the parent plants.

Patil (1966) reported tall mutants having a height of 120-130 cm as compared to 70-90 cm of the control. The increased height was found to be due to greater length of internodes (6 cm compared to 3.5 cm in control). Short mutants having short internode length was also noticed. Sanjeeviah et al.(1967) observed dwarf plants in varying frequencies under different doses of gamma radiations in groundnut. Such mutants were more frequent at higher doses of gamma irradiation than with x-rays indicating that the locus concerned was more sensitive to gamma rays.

A number of mutations affecting branching habit such as non-branching, less branching, non-secondary branching and tertiary branching were reported by Patil (1966). Gregory (1968) reported three mutants in groundnut affecting plant habit in the  $x_2$  generation. They are stixt (s) in which lateral branches arise from 15-30<sup>o</sup> angle with the main axis, runner (R) in which lateral

branches arise from approximately  $90^{\circ}$  angle with the main axis and runner bunch in which the lateral branches arise from  $60^{\circ}$ -  $75^{\circ}$  angle with the main axis. Mouli and Patil (1976) isolated a suppressed branching mutant in groundnut variety TG-2 following 40 kR gamma ray treatment. Patil and Mouli (1978) observed a bunchy top mutant in the M<sub>3</sub> progeny of Spanish Improved variety of groundnut treated with 50 kR gamma rays. The studies on induced mutants by Prasad et al. (1984) indicated that genetic restructuring of peanut plant to combine compact canopy frame and higher pod yield is possible in the case of virginia genotypes, while it is not possible in the Spanish types because reduction in vegetative growth will result in decreased pod production.

Early maturing mutants were reported by several workers. An early maturing groundnut mutant from Pollachi-1 worthy of exploitation was recorded by Dorairaj (1979) following gamma irradiation. Sin pa detha I a mutant maturing 10-15 days earlier was obtained from Spanish type Mague-10 following gamma irradiation at 40 kR. (Anon., 1982). Mutant lines which outyielded the parents by over 15% along with earliness were isolated from groundnut variety T54 by Marghitu et al.(1982).

Patil (1966) reported several mutations in groundnut affecting leaf characters such as lethery leaf mutant, cup leaf mutant, imparipinnate leaf mutant and mutants with fused leaflets. Gregory (1968) also isolated leaf mutants in groundnut viz. Lupinus, cup, Flop, multiple leaflets and corduroy. Mouli and Patil (1979) isolated two mutants having modified stipules after x-ray treatment. Bifurcated leaflets in groundnut was observed by Mouli et al.(1984) in a cross between Gujarath Dwarf and a recombinant of two induced mutants (sl) small leaf and (v) virescent of Spanish Improved. A narrow leaved virginia type mutant  $(TMV_2 NLM)$  from Spanish  $TMV_2$  with improved dry matter production, high pod number and resistance to <u>Cercospora</u> leaf spot was reported by Prasad et al.(1984).

Patil (1966) isolated a multicarpellary groundnut mutant having two carpels. Double vexillum mutant was isolated by Sinha and Roy (1969) from the M<sub>2</sub> progeny of the groundnut variety 41-C in 45 kR treatments. This mutant had an extra interpolated posterior petal so that there were two vexillum petals instead of normal one. Groundnut mutants with large, long and small pods were reported by Patil (1966). TG-1a bold seeded variety of groundnut suitable for the Indian export market was isolated by Patil (1974) after x-irradiation. A gamma ray induced Spanish bunch mutant with large pod was reported by Mouli and Kale (1982).

## 2.5 Achievements

Gregory from his intensive work has established beyond doubts that groundnut is eminently suitable for mutation breeding. The first achievement in groudnut through mutation breeding was the evolution of the mutant variety 'NC-4x' by x-ray irradiation of the variety NC-4. This mutant variety was high yielding with better pod and seed quality than the parent (Gregory, 1957). Cooper and Gregory (1960) produced  $x_2$  and  $x_3$  progenies with increased resistance to leafspot, (<u>Cercospora arachidicola</u> and <u>C@rcospora</u> <u>personata</u>) from peanut seeds irradiated with 10,000 to 18,500 r x-rays.

Mutation research leading to the development of improved TG (Trombay Groundnut) varieties at the Biology and Agriculture Division of BARC, India is an illustration of research benefits to the nation. Quite a number of mutations were induced using x-ray irradiation in the variety Spanish Improved (Patil, 1966). Subsequent screening of true breeding mutants and their sibs for high yielding ability and other desirable attributes, resulted in the isolation of twenty lines which were later released as mutant varieties TG-1 to TG-20 (Patil and Mouli 1979).

TG-1 a large poded mutant isolated had large kernels weighing 85 g per 100 kernels as against 50 g of the parent.

In addition to TG-1 other mutants having early maturity (TG-2), increased pod setting (TG-3) and reduced plant height (TG-4, TG-5 and TG-6) were isolated and released.

Hybridization among mutants resulted in the selection of TG-7 to TG-13 varieties having dark green leaves, few branches and pods with less prominent venation and more oil content than the parent variety Spanish Improved. Improved varieties TG-14 to TG-20 were developed after inter-crossing the mutants. TG-14 selected in a cross, darker green x virescent, was high yielding especially for rabi cultivation. TG-16 with large pods derived after crossing TG-1 and Virescent had earliness and better recovery of kernels. TG-17 had good plant type, less number of primary branches, more flowering nodes and increased harvest index than Spanish Improved. TG-18 and 19 having large and extra large kernels respectively along with increased kernel recovery were also developed from different crosses.

Mutants Bp1 (early) and Bp2 (mid early) in maturity evolved by Sinha and Rahman (1979) through gamma irradiation had compact habit, large kernels and pods. Bp1 and Bp2 were released in Bihar as Sonya Bold 1 and Sonya Bold 2 respectively for cultivation.

CO.2 a new high yielding mutant variety of groundnut was evolved by Sivaram et al.(1985b) by treating the seeds of Pol-1 with 0.2% EMS.

Sharma et al.(1981) reported that the essential amino acid content per seed was higher in the Trombay groundnut (TG) mutants than in Spanish Improved. Large seeded cultures with increased sucrose content of 5.6-8% as compared to 3.6% in Spanish Improved were also noticed. Sharma et al. (1984) reported that $\tau$ G-8, TG-9, TG-17 and TG-18 had improved quality of oil with a lower content of linoleic acid and higher content of oleic acid.

# MATERIALS AND METHODS

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#### MATERIALS AND METHODS

The research programme was carried out during 1986 at the Department of Plant Breeding, College of Agriculture, Vellayani, Trivandrum.

3.1 <u>Materials</u>

The groundnut variety EC.119704 available at the Department of Plant Breeding was used for the study. It is a high yielding, semi open variety having medium plant stature, dark green leaves and large pods with 120 days duration. It also possesses fresh seed dormancy of about 45-50 days.

Gamma ray irradiation was done at the School of Genetics, Tamil Nadu Agricultural University, Coimbatore, utilising the Cobalt-60 gamma chamber. The source was operated at an intensity of 0.4237 million Roentgens per hour.

3.2 Methods

3.2.1 Irradiation with gamma rays

Kernels of uniform size and maturity were selected and dried uniformly. Five samples of 250 kernels each were exposed to gamma radiation at doses of 10, 20, 30, 40 and 50 krads.

## 3.2.2 M<sub>1</sub> generation

## 3.2.2.1 Laboratory studies

Fifty kernels from each of the five treatments along with on untreated standard were kept in petri-dishes lined with moist filter paper at 10 kernels each in 5 replications for the study of germination and initial root and shoot growth. Germinated kernels were counted everyday from the third day onwards upto the seventh day and the total count was expressed as percentage of germination. On the 7th day, the length of shoot and root was measured from the point of their differentiation to the growing apex in all the germinated seeds.

After the seventh day, the germinated kernels in each treatment and replication were transplanted separately in galvanized iron trays filled with soil. The height of the seedlings was measured from the soil surface to the tip of the terminal bud at weekly intervals.

## 3.2.2.2 Field experiment

The field experiment was laid out in Randomised Block Design with 6 treatments and 4 replications. The treatments included the 5 doses of gamma rays and one unirradiated control. In each plot, fifty kernels were sown at a spacing of 30 x 20 cm. The following observations were recorded from the field trials.

## i) Germination of seeds

Germination counts were taken from the seventh day onwards and continued upto the 20th day after sowing and expressed as percentage.

### ii) <u>Survival</u>

Counts of surviving seedlings were taken on the 15th, 30th and 45th days after sowing and expressed as percentage of the total number of kernels sown in each treatment.

## iii) Plant height

Measured on the 30th and 60th days from 20 plants selected at random from each of the treatments and control in each replication. The height was measured in cm from the soil surface to the tip of the terminal bud and the mean plant height was expressed as percentage with respect to the control.

## iv) Pollen fertility

Forty plants (10 plants from each replication) were selected at random from each of the treatments and the control for the estimation of pollen fertility. Pollen from mature flower buds were mounted over the slides using glycerine-acetocarmine stain. The unstained, undersized, partially stained and shrivelled pollen grains were scored as sterile and the uniformly stained, well filled pollen as fertile. Ten microscopic fields were observed on each slide and fertile and sterile pollen were counted. Fertility of each plant was estimated as percentage of the number of fertile pollen grains to the total number of pollen grains scored. The mean fertility of each dose was estimated and expressed as a percentage of the control.

#### v) Chlorophyll chimeras

The plants were observed regularly for chlorophyll deficient sectors on leaves and the plants showing such patches were scored as chimeras.

## vi) Morphological variations

Periodical observation of the plants was made and plants which differed in stature such as dwarfs, talls and plants showing variations in number, size and shape of leaves were recorded.

All the  $M_1$  plants were harvested, mature pods collected and dried individually. The untreated control was also harvested and pods collected. The number of pods obtained from each of the  $M_1$  plant was recorded.

# 3.2.3 M2 generation

 $M_2$  generation was raised as  $M_1$  plant progenies. These progenies were sown in rows at a spacing of 20 cm within rows and 30 cm between rows. Untreated control progenies were also sown after every 10 treated progeny rows for comparison.

### 3.2.3.1 Chlorophyll mutations

The  $M_2$  seedlings were observed in the early morning from the tenth day of sowing onwards up to the 20th day and the chlorophyll mutations were scored. The progenies segregating for mutations were counted first and the chlorophyll mutation frequency on  $M_1$  plant progeny basis was estimated as the number of plant progenies segregating per 100  $M_1$  plants.

The number of mutants in each segregating progeny was counted separately. The number of normal plants in each of the segregating and non-segregating progeny were also counted. These data were utilised for the estimation of mutation frequency on  $M_2$  plant basis ie., the number of mutants per 100  $M_2$  plants. The different types of chlorophyll mutants were identified and scored for calculating the spectrum of the relative percentage of different types of mutants. The spectrum of chlorophyll mutants was classi-

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fled as follows:

Viridis	-	Plants with light green leaves
Chiorina	-	Plants with yellow green leaves
Maculata	-	Plants showing irregular patches of
		chlorophyll deficient spots on the
		leaves

The number of mutants and number of normal plants were counted to calculate the segregation-ratio as percentage of mutants to the total number of plants in the segregating  $M_1$  progenies.

### 3.2.3.2 Viable mutations

The progenies segregating for viable mutations were scored. The days to first blooming in all the plants was noted. Viable mutants were scored at fortnightly intervals on the basis of canopy characters and various morphological features and were described with respect to the deviation from normal plants. Mutations affecting pod characters were scored and described at the time of harvest. The progenies segregating for mutations were counted first and the viable mutation frequency on  $M_1$ plant progeny basis was estimated as the number of plant progenies segregating per 100  $M_1$  plant progenies. The number of mutants in each segregating progeny was counted separately. The number of normal plants in each of the segregating and non-segregating progeny were also counted. These data were utilised for the estimation of mutation frequency on  $M_2$  plant basis. The different types of viable mutants were identified and scored separately for calculating the spectrum of mutants.

### 3.2.3.3 Total mutations

The scores for chlorophyll and viable mutations were used for estimating the total mutation frequency. A progeny was counted as segregating if it segregated for a chlorophyll and/or a viable mutation. Total mutation frequency was estimated as number of mutations per 100  $M_1$ plant progeny.

## 3.2.4 Mutagenic effectiveness and efficiency

The formulae suggested by Konzak et al.(1965) was adopted for estimating the effectiveness and efficiency of the mutagen at different doses.

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Mutagenic effectiveness = 
krad
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$$Mutagenic efficiency = \frac{M \times 100}{L}; \frac{M \times 100}{I}; \frac{M \times 100}{S}$$

Where,

M - Mutation frequency on M<sub>1</sub> plant progeny basis,
 krad - Dose of radiation in kilorad,

- L Percentage of lethality on the basis of survival reduction of seedlings at 30 days,
- I Percentage of injury ie., height reduction of seedlings at 30 days and
- S Percentage of pollen fertility reduction.

The M<sub>2</sub> plants were harvested on the 90th day after sowing. Viable mutants located were harvested and pods collected separately. The remaining plants were harvested progeny wise. Plants with lesser number of immature pods were selected as early types. Their pods were collected separately, cleaned and dried.

### 3.2.5 Economic mutants

Economic mutants were selected from the harvested M<sub>2</sub> plants. Plants with less than 30 per cent immature pods and ten or more mature pods per plant were selected.

Observations on the following characters were recorded in the selected mutants.

- Days to first blooming; The number of days from sowing to opening of the first flower.
- ii) Number of pods per plant: The number of mature and immature pods per plant were counted.
- iii) Percentage of immature pods: The percentage of immature pods was calculated as

- iv) Pod yield per plant: Weight of mature pods collected immediately after harvest.
- v) Haulms yield per plant: The fresh weight of green matter after removing the pods.
- vi) Fresh seed dormancy: Half the number of mature pods per plant up to a maximum of five from each selected mutant was kept for germination immediately after harvest.

The shelling percentage and mean kernel weight was estimated using the pods shelled for dormancy studies.

vii) Shelling percentage: Estimated as the ratio of the weight of kernels to the weight of shell and expressed as percentage. viii) Kernel weight: Mean kernel weight was estimated.

ix) Colour of testa: The colour of testa was noted from the mature kernels kept for germination studies.

# RESULTS

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#### RESULTS

The groundnut variety EC.119704 was used for the study. It is a high yielding variety with large pods and kernels and fresh seed dormancy for about 45 to 50 days. Uniform sized kernels of the variety EC.119704 were exposed to gamma irradiation at a dose range of 10 to 50 krad for inducing earliness and isolating desirable types. The effects of gamma rays in the  $M_1$  were studied. From the  $M_2$ , 16 economic mutants with early maturity, high pod yield and fresh seed dormancy were isolated. The results of the study are presented.

## 4.1 Effects of gamma rays in the M. generation

# 4.1.1 Germination of seeds

The data on mean days to germination and the percentage germination of kernels in the  $M_1$  generation are presented in Table 1.

The germination percentage did not show any significant difference between the various doses under laboratory conditions, except for a slight reduction at 20 and 30 krad dose. Under field conditions there was a slight increase in germination at the lowest dose (10 krad) and a steady decline in germination with increasing doses. Early germination was observed at the lowest dose (10 krad) while at the higher doses there was a slight delay in germination.

Labora	Laboratory condition				
Germina- tion (per- centage of control)	Mean days to germina- tion	Percen- tage of control	Germination (percentage of control)		
100.00	2.09	100.0	100.0		
100.00	2.06	98.6	107.0		
87.6	2.34	111.9	97.6		
89.6	2.35	112.4	96.2		
100.0	2.39	114.4	86.2		
100.0	2.40	114.8	78.6		
	Germina- tion (per- centage of control) 100.00 100.00 87.6 89.6 100.0	Germina- tion (per- centage of control)       Mean days to germina- tion         100.00       2.09         100.00       2.06         87.6       2.34         89.6       2.35         100.00       2.39	Germina- tion (per- centage of control)Mean days to germina- tionPercen- tage of control100.002.09100.0100.002.0698.687.62.34111.989.62.35112.4100.02.39114.4		

Table 1. Days to germination and germination percentage of kernels in the M<sub>1</sub> generation

### 4.1.2 Survival of plants

The data on survival of plants in the M<sub>1</sub> generation are presented in Table 2. The survival of plants under field conditions is illustrated in Figure 1.

At all the three stages ie. 15th, 30th and 45th day after sowing the survival decreased with increasing dose except for a slight increase at the lowest dose of 10 krad. A comparison between germination and survival percentages indicate that part of the germinated seeds failed to survive and establish during the post germination period at all the doses above 10 krad and this lethality persisted even upto the 45th day. By the 45th day the lethality was 82.2 per cent at the highest dose.

### 4.1.3 Plant growth

Data on plant growth in the M<sub>1</sub> generation under laboratory conditions is presented in Table 3. The effect of gamma rays on shoot and root growth is illustrated by Figure 2a and 2b.

	Survival							
Doses	15t	h day	30th d	ау	45th da	y .		
	Percen tage of control	Letha- lity percent- age	Percent age of control	Letha- lity percen- tage	Percent age of control	Letha- lity percen- tage		
Control	100,00	0.0	100.00	0.0	100.00	0.0		
10 krad	105.8	-5.8	105.7	-5.7	106.6	-6.6		
20 krad	96.8	3,2	91.2	8.8	87.5	12,5		
30 krad	98.1	1.9	69.2	30.8	57.9	42.1		
40 krad	87.8	12.2	55.9	44.1	51.3	48.7		
50 krad	80.1	19.9	23.3	76.7	17.8	82.2		

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Table 2. Survival of plants in the M<sub>1</sub> generation

Figure 1. Survival of plants under field condition

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т <sub>1</sub>	-	Cor	ntrol
т <sub>2</sub>	-	10	krad
т <sub>з</sub>	-	20	krad
т <sub>4</sub>	-	30	krad
Т <sub>5</sub>		40	krad
т <sub>6</sub>	-	50	krad

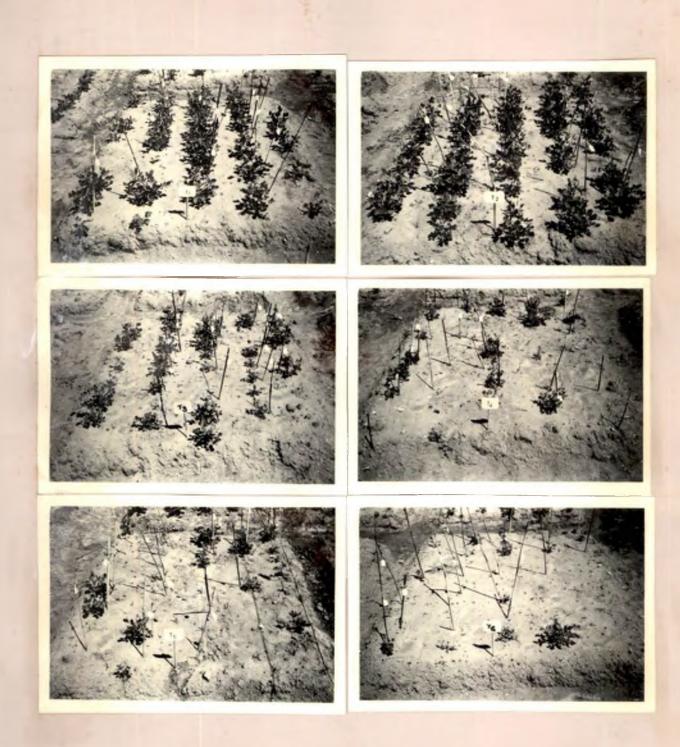


Figure-1

Shoot	Percen-		Percen-	Shoot/	Plant height				
Doses	length (cm)	tage of control	length (cm)	tage of control	root ratio		14th day	21	lst day
	- <u> </u>			(percen- tage of control)	( cm)	Percent- age of control	(cm)	Percent- age of control	
Control	7.9	100.0	7.3	100.0	100.0	5.4	100.0	7.2	100.0
10 krad	7.6	96.2	6.8	93.2	103.7	5,5	101.9	6.8	94 <b>.4</b>
20 krad	5,5	69.6	2.6	35.6	196.3	2.4	44 <b>.4</b>	4.4	61.1
30 krad	5.4	68.4	2.5	34.3	200.0	1.7	31.5	3.2	44.4
40 krad	5.7	72.2	2.9	39.7	182.9	1.6	29.6	2.8	38.9
50 krad	5.0	63.3	1,9	26.0	243.5	0.8	14.8	1.0	13.9

Table 3. Plant growth in M<sub>1</sub> generation under laboratory conditions

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Figure 2. Shoot and root growth under laboratory condition.

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Control
 10 krad
 - 10 krad
 - 20 krad
 - 30 krad
 - 40 krad
 - 50 krad

. .





Figure-2

The length of shoot and root decreased with increasing dose. Root growth was inhibited to a greater extent than shoot growth, consequently, the shoot/root ratio percentage was more than 100 in all the treatments. The height of plants transplanted into galvanized iron trays also showed a decreasing trend with increasing doses (Figure 3).

The plant height in the M<sub>1</sub> generation under field conditions are presented in Table 4.

The control recorded maximum height (6.9 cm) on the 30th day and 50 krad treatment the minimum (2.8 cm). There was a reduction in plant height with increasing dose. The magnitude of this height reduction or injury percentage was more on the 30th day as compared to that on the 60th day. A partial recovery of seedlings from injury was noticed after the 30th day.

# 4.1.4 Fertility

The pollen fertility in the  $M_1$  generation is presented in Table 5.

The percentage of pollen sterility increased with increasing dose. However, even in the highest dose of 50 krad the pollen sterility was only 46 per cent. Figure 3. Plant height under laboratory conditions

1- Control
 2 - 10 krad
 3 - 20 krad
 4 - 30 krad

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- 5 **\_** 40 krad
- 6 50 krad



Figure-3

	Plant height					
Doses		30th day			60th day	
	(cm)	Percentage of control	Injury percentage	( cm )	Percentage of control	Injury percent- age
Control	6.9	100.0	-	14.1	100.0	-
10 krad	6.6	95 <b>.7</b>	4.3	14.3	101.4	-1.4
20 krad	5.1	73.9	26.1	12.5	88.7	11.3
30 krad	4.2	60.9	39.1	11.4	80 <b>.9</b>	19.1
40 krad	3.9	56,5	43,5	10.7	75.9	24.1
50 krad	2.8	40.6	59.4	8.7	61.7	38.3

# Table 4. Plant height in the $M_1$ generation under field conditions

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Doses	Pollen ferti- lity(percent- age)	Percentage of con- trol	Pollen steri- lity(percent- age of control)
Control	96.9	100.0	0,0
10 krad	94.5	<b>97.</b> 5	2,5
20 <b>kr</b> ad	85.4	88.1	11.9
30 krad	78.3	80 <b>.8</b>	19.2
40 krad	66.9	69.0	31.0
50 krad	52.3	54.0	46.0
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Table 5. Pollen fertility in the  $M_1$  generation

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The effect of gamma rays in the M<sub>1</sub> generation under field condition is graphically represented in Figure 4.

### 4.1.5 Chlorophyll chimeras

A number of plants with chlorophyll deficient patches on leaves was observed in the  $M_1$  generation. Figure 5 represents the chlorophyll chimeras in the  $M_1$  generation.

One plant with completely yellow coloured leaves on a branch was observed in 20 krad treatment. All the leaves on this branch retained the yellow colour till maturity. Plants with chlorophyll deficient chimeras such as light green sectors on leaves, yellow and green patches on the leaves and chlorophyll deficient patches were observed in 20, 30 and 50 krads respectively. But majority of these plants produced normal green leaves during later growth period.

### 4.1.6 Morphological abnormalities

A brief description of the radiation induced morphological abnormalities in the M<sub>1</sub> generation are presented below:-

### 4.1.6.1 Laboratory conditions

Seedling abnormalities in the M<sub>1</sub> under laboratory

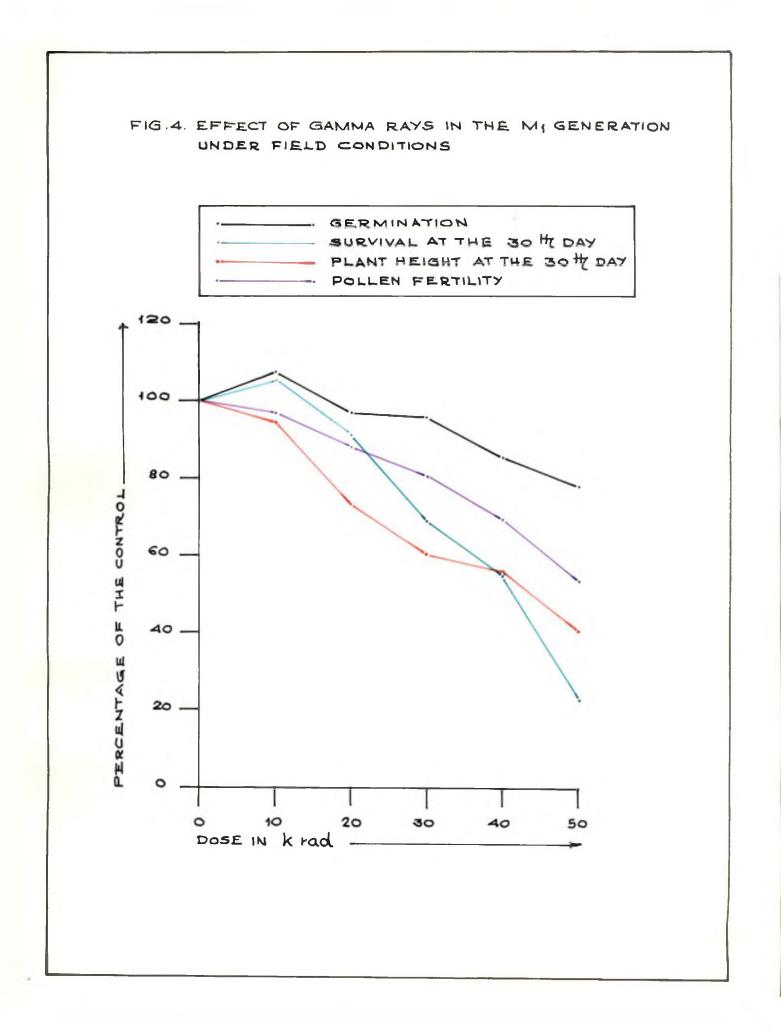


Figure 5. Chlorophyll chimeras in the  $M_1$ 

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1 - Control		
2 - Light green	)	
3 - Yellow	)	20 krad
4 - Yellow with 7 leaflets	>	
5 - Light green	}	<b>3</b> 0 krad
6 - Yellow	)	
7 - Yellow	-	50 krad

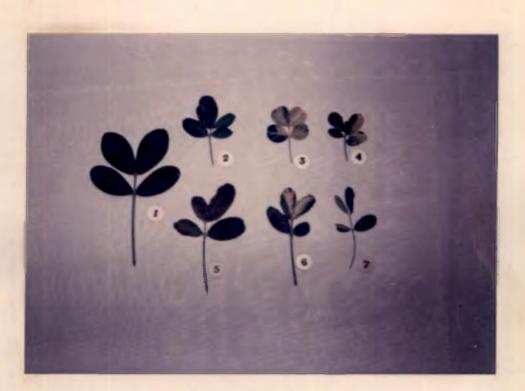


Figure-5

conditions are illustrated in Figure 6.

Three seedlings with apical bud inhibition were observed at 10 and 20 krad treatments. Normal branches developed on both the sides in two of them, while in the third one branching was noted only on one side. In 10 and 20 krad treatments, seedlings with enlarged hypocotyl, reduced internode length and clumped appearance were observed. However, these seedlings did not survive. One seedling with complete inhibition of shoot growth was observed at 30 krad treatment. But it failed to survive. A seedling with small narrow sickle shaped leaf-lets was observed at 30 krad treatment. The leaflet abnormality was noticed only in the first formed leaves and subsequent leaf development was normal. A seedling without hypocotyl development was also observed at 30 krad treatment. Another seedling with complete inhibition of root and shoot, but with normal hypocotyl development was observed at 40 krad treatment. In 50 krad treatment seedlings with very large and thick cotyledons having anthocyanin pigmentation and complete shoot and root growth inhibition were noticed.

### 4.1.6.2 Field conditions

The morphological variations observed in the field are described below.

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Figure 6. Seedlings abnormalities in M<sub>1</sub> under laboratory conditions.

> - Control 1

4

5

6

7 8

- 2 - 10 krad - Apical bud inhibition
- 3 - 30 krad - Abnormal leaf
  - 20 krad Clumping of seedling 11
  - 10 krad -
  - 20 krad Apical bud inhibition
  - 30 krad Inhibition of shoot growth
  - 50 krad Inhibition of shoot growth and pigmented cotyledons.
- 9 - 30 krad - Inhibition of hypocotyl.
- 10 - 40 krad - Root and shoot growth inhibition
- 11 - 10 krad - Apical bud inhibition.



Figure-6

Tall plants with thick stem and large leaves having anthocyanin pigmentation were observed in 30 krad treatment. In 40 krad treatment a plant with stunted growth and small leaves was observed. This plant later produced a branch with long internodes and 8-9 leaflets. Dwarf plants with small leaves were observed in 50 krad treatment also.

Some of the leaf variations observed in the M<sub>1</sub> under field conditions are presented in Figure 7.

Plants bearing bifurcated leaflets and 5 to 6 leaflets were observed in 20 krad treatment, while plants bearing 3 small leaflets and with forked petioles were observed in 40 krad treatment. Plants with broad petiole bearing 2 leaflets and 8 leaflets were observed in 50 krad treatment.

### 4.2 Effects in the M, generation

4.2.1 Chlorophyll mutation

### 4.2.1.1 Frequency

Frequency of chlorophyll mutations in the  $M_2$  generation are presented in Table 6.

On  $M_1$  plant basis, the 30 krad treatment produced the highest mutation frequency (64.4) and the lowest

Figure 7.	Leaf variations	in M <sub>1</sub>	under	field
	conditions.	•		

- 1 Control
- 2 50 krad
- 3 = 40 krad
- 4 20 krad
- 5 20 krad
- 6 20 krad
- 7 = 40 krad
- 8 **-** 50 krad

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Figure-7

		of M <sub>1</sub> plant	Mutation	Number o	Mutations	
Doses	Scored	genies Segregating	per 100 - M <sub>1</sub> plants	Scored	Chlorophyll mutants	per 100 M <sub>2</sub> plants
10 krad	153	87	56.9	1233	282	22.9
20 krad	89	40	44.9	53 <b>7</b>	107	19.9
30 krad	59	38	64.4	377	80	21,2
40 krad	44	21	47.7	262	44	16.8
50 krad	11	4	36 <b>.</b> 4	28	6	21.4

# Table 6. Frequency of chlorophyll mutations in the $M_2$ generation

frequency (36.4) by the 50 krad treatment. The mutation frequencies estimated on  $M_2$  plant basis were comparatively lower than that estimated on  $M_1$  plant basis. The highest mutation frequency estimated on  $M_2$  plant basis was at 10 krad (22.9) and the lowest frequency (16.8) at 40 krad treatment.

### 4.2.1.2 Spectrum

The spectrum of chlorophyll mutations in the  $M_2$  generation are presented in Table 7.

Chlorina, viridis and maculata are the 3 types of chlorophyll mutants observed. The frequency of chlorina was highest 10 krad treatment (31.2) and lowest at 40 krad (13.6). Maximum frequency of viridis was observed at 20 krad (24.3) and the minimum at 30 krad (7.5) while in the case of maculata the maximum frequency was observed at 30 krad (70.0) and the minimum at 10 krad (49.3). Among the three types of chlorophyll mutations relative percentage of maculata was comparatively higher at all the treatments.

### 4.2.1.3 <u>Segregation ratio</u>

The segregation ratio $\mathfrak{A}$ chlorophyll mutations in the  $M_2$  generation are presented in Table 8.

	Total	Relative percentage(Spectrum				
Doses	number of mutants	Chlorina	Viridis	Maculata		
10 krad	282	31.2	19.5	49.3		
20 kra <b>d</b>	107	15.9	24.3	59.8		
30 krad	80	22.5	7.5	70.0		
40 krad	44	13.6	18.2	68.2		
50 krad	6	16,7	16.7	66.6		
Total	519	25,1	18.5	56,4		

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Table 7. Spectrum of chlorophyll mutations in the  ${\rm M}_2$  generation

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Doses	Number of plants in segregating M <sub>1</sub> progenies	Number of mutants	Segregation ratio(percent- age of mutants)
10 krad	921	282	30.6
20 krad	356	107	30.1
30 krad	283	80	28.3
40 krad	203	44	21 <b>.7</b>
50 krad	16	б	37.5

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Table 8. Segregation ratio of chlorophyll mutants in  $M_2$  generation

The segregation ratio was higher than the expected value at all the doses probably due to the low population in the  $M_2$  progeny rows. The highest segregation ratio of 37.5 was observed at 50 krad and the lowest ratio of 21.7 at 40 krad treatment.

### 4.2.2 Viable mutations

All mutations affecting the morphology of different plant parts were classified as viable mutations. Individual viable mutations were scored in the  $M_2$  generation by visual observations. The changes induced by the mutagen affected either one or a combination of plant characters.

### 4.2.2.1 Frequency

The frequency of viable mutations in the  $M_2$  generation is presented in Table 9.

The frequency of viable mutations were estimated as number of mutation per 100  $M_1$  plant progenies and per 100  $M_2$  plants. The frequencies were found to decrease with increasing doses except for a slight increase at 20 krad treatment. The frequency was higher on  $M_1$  plant basis than on  $M_2$  plant basis. Among the five doses maximum frequency of 60.7 and 18.2 respectively on  $M_1$  and  $M_2$  plant basis were recorded at 20 krad and the minimum frequency (18.2 and 8.0 respectively) at 50 krad treatment.

	Doses	Numl prog	ber of M <sub>l</sub> plant genies	Mutations per 100	Number o	Mutants per 100		
		Scored	Segregating	M <sub>1</sub> plants	Scored	Viable mutants	M <sub>2</sub> plants	
10	krad	153	74	48.4	1233	155	12,6	
20	krad	89	54	60.7	548	100	18.2	
30	krad	59	<b>2</b> 9	49.2	403	48	11.9	
40	krad	44	18	40,9	26 <b>2</b>	30 .	11.5	
50	krad	11	2	18.2	25	2	8.0	

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Table 9. Frequency of viable mutations in the  ${\rm M}_2$  generation

# 4.2.2.2 Spectrum

Changing the spectrum of mutations in a predictable manner by induced mutagenesis is an important goal of current mutation research. A wide spectrum of mutations affecting various morphological characters such as height, duration, leaf, pod and kernel characters were isolated. The spectrum of viable mutations in the M<sub>2</sub> generation are presented in Table 10.

Among the relative percentage of different types of mutations estimated, mutations affecting plant height, such as tall and dwarfs, plant types such as compact and spreading, branching and non-branching, size and shape of leaves and pod and kernel characters were observed. In the spectrum, relative percentage of dwarf types were maximum (39.1) while the percentage of non-branching types were minimum (3.28). Among the mutants isolated for plant height dwarf types were common. The relative percentage of tall mutants was only 4.78. Plant type mutants with spreading habit were less when compared to compact mutants. The relative percentage of compact mutants was 18.2 and that of spreading types was 3.58. A large number of leaf type

-	Total number		Relative percentages (Spectrum)							
Doses	of mutants	Tall	Dwarf	Compact	Spread- ing	Non- branch- ing	Leaf types	Early	Pod and kernel mutants	
10 krad	155	6.45	29.68	18.71	1.94	2.58	25.81	7.74	7.06	
20 krad	100	2.00	44.00	20,00	5.00	4,00	18.00	3.00	4.00	
30 krad	<b>4</b> 8	4.16	50,00	10,42	-	6.25	29.17	-	-	
40 krad	30	6.67	56 <b>.</b> 67	20.00	13.33	-	3.33		-	
50 krad	2	-	-	50,00	-	-	50.00	-	-	
Total	335	4.78	39 <b>.1</b> 0	18,20	3.58	3.28	22.09	4.48	4.48	

Table 10. Spectrum of viable mutants in the  $M_2$  generation

mutants were also observed. The relative percentage of leaf type mutants was 22.09. Early as well as pod and kernel mutants were observed only at 10 and 20 krad treatments. Since all the M<sub>2</sub> plants were harvested at 90 days for isolating the early maturing mutants, the late maturing types could not be detected. Variability in almost every character was observed in the viable mutants indicating the possibility for altering any character in groundnut through induced mutagenesis. A brief description of the viable mutants detected are presented below.

## i) Early maturing mutant

Twelve early maturing mutants with desirable canopy, pod characters and high pod yield were isolated from 10 and 20 krad treatments.

#### ii) Tall mutant

Tall plants (Figure 8b) with long internodes and large leaves as compared to normal plants (Figure 8a) were observed at 10 and 30 krad treatments.

#### iii) Dwarf mutants

Dwarf plants with narrow leaves and less number of branches (Figure 9b) and some with small leaves (Figure 9a) were observed in 10 and 20 krad treatments. A dwarf mutant with extremely reduced leaflets and suppressed Figure 8a. A normal plant of var.EC.119704

Figure 8b. Tall mutant in 30 krad.

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Figure-8b

Figure-8a

Figure 9a. - Dwarf mutant with small leaves in 20 krad

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Figure 9b. - Dwarf mutant with narrow leaves and less branches in 10 krad.

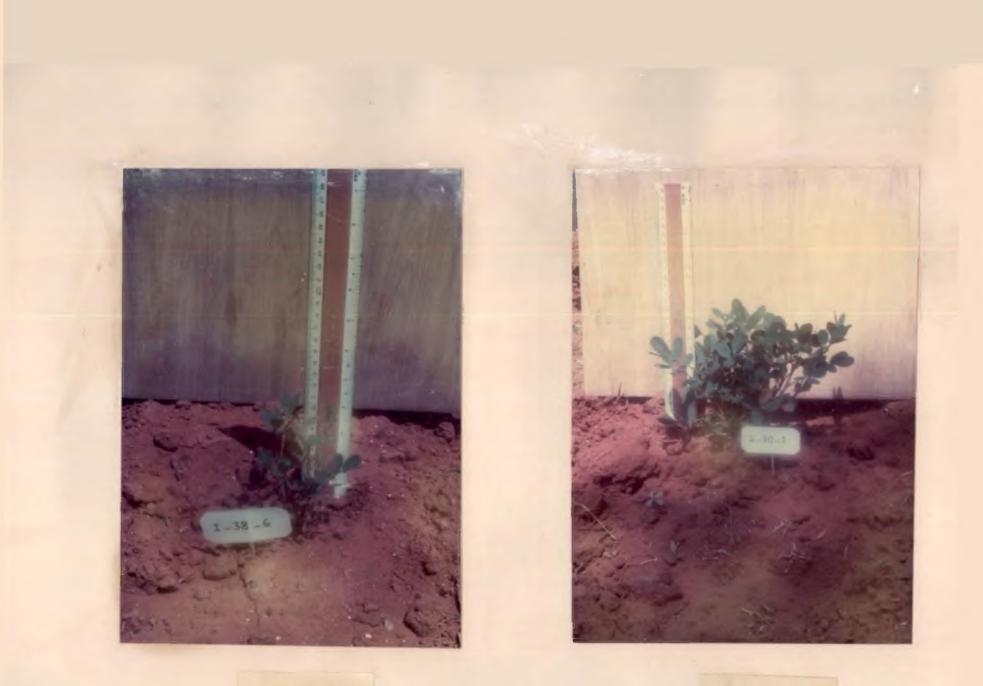


Figure-9b

Figure-9a

growth was observed. The extremely stunted growth of stem and branches with minute leaflets gives a "bunchy top" appearance to the mutant. Normal flowering and pod setting was not seen in this mutant. Dwarf mutants were observed in all the treatments.

#### iv) Erect mutant

In this mutant the main axis was short and erect and the lateral branches were produced slightly oblique to the main axis, but almost parallel to it. (Figure 10a). This mutant was observed at 10 krad treatment.

#### v) Compact mutant

Mutants with compact plant type were observed (Figure 10b) in 10 and 40 krad treatments, with an angle of branching less than that of control.

## vi) Spreading mutant

Mutants with spreading growth habit were observed at 10,20 and 40 krad treatments.

#### vii) Non-branching mutant

Mutants with the complete absence of vegetative branches were observed at 10 and 20 krad treatments (Figure 11a and 11b). Figure 10a \_ Erect mutant in 10 krad

Figure 10b - Compact mutant in 10 krad

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Figure-10b

Figure-10a

Figure 11a. Non-branching mutant in 10 krad

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Figure 11b - Non-branching mutant in 20 krad.

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Figure-11b

Figure-11a

## viii) Mutant with flattened stem

A plant with stunted growth and flattened main axis which was split into 4 at the tip was observed at 20 krad treatment. Flowering and pod setting was not observed in this mutant.

#### ix) Yellow leaf mutant

A short yellow leaf mutant with multiple leaflet was observed at 20 krad treatment (Figure 12).

It was sparsely flowering and was non-productive. Another yellow leaf mutant observed at 30 krad treatment had light yellow leaflets with green mid rib and veins. The leaf colour of this mutant changed subsequently.

#### x) Curly leaf mutant

A late flowering mutant with curly leaves and few immature pods was observed in 50 krad treatment(Figure 13a).

#### xi) Dark green mutant

Mutants with dark green leaves (Figure 13b) were noticed in all the treatments. Some of the mutants were stunted in growth.

A mutant with margins of the leaflets slightly curved in the form of a cup was observed at 30 krad treatment. This cup shaped leaf mutant produced only Figure 12. Yellow leaf mutant in 20 krad.

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Figure-12

Figure 13a. Curly leaf mutant in 50 krad.

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Figure 13b. Dark green mutant in 40 krad.

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Figure-13b

Figure-13a

a few flowers and pods. Mutant plants with five to eight leaflets instead of the four leaflets in the control plants were observed at 20 and 40 krad treatments. Two plants observed in 10 krad treatment produced large pods and kernels than the control. Mutants with deeply constricted pods were noticed in 10 krad treatment as compared to the shallow constricted pods in the control. Mutants with light brown and brown coloured testa were obtained in 10 and 20 krad treatments, instead of the light pink coloured testa of the control.

#### 4.2.3 Total mutations

The frequency of total mutations in the  $M_2$  generation is presented in Table 11.

The mutation frequencies per 100  $M_{f_1}$  plants shows that the maximum frequency was recorded at 30 krad (84.8) and the minimum at 50 krad (54.5). However, the total mutation frequency did not show any definite dose relationship.

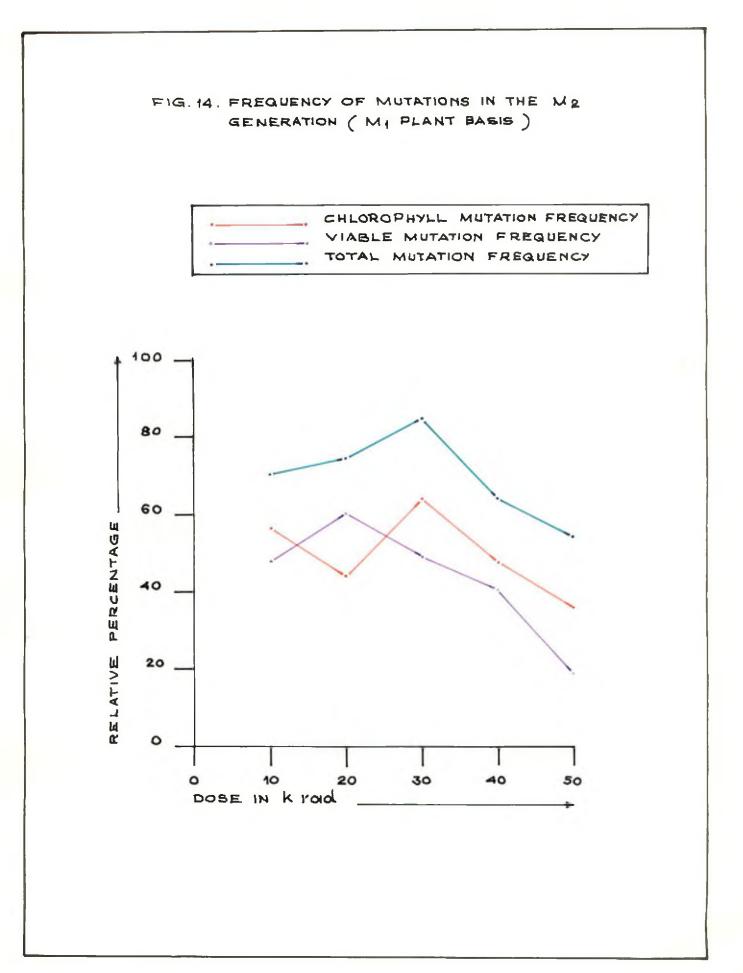
The frequency of mutation in the M<sub>2</sub> generation is graphically presented in Figure 14.

Doses	Number progen:	of M <sub>1</sub> plant ies	Mutations per 100		
	Scored	Segregating	- M <sub>1</sub> plants		
10 krad	153	110	71,9		
20 krad	<b>8</b> 9	66	74.2		
30 krad	59	50	84.8		
40 krad	44	28	63 <b>.6</b>		
50 krad	11	6	54 <b>.5</b>		

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Table 11. Frequency of total mutations in the  $M_2$  generation



#### 4.3 Mutagenic effectiveness and efficiency

Mutagenic effectiveness and efficiency of gamma rays in inducing chlorophyll and viable mutants are presented in Tables 12 and 13 respectively.

The effectiveness of inducing chlorophyll and viable mutations decreased with increasing dose of gamma rays. This shows that the increase in frequency of mutation was not proportional to the increase in the dose of the mutagen. It was also observed that the lower doses were more effective than higher doses in inducing chlorophyll and viable mutations. The maximum value for effectiveness was recorded for chlorophyll (569) and viable mutations (484) in 10 krad treatment and the minimum value recorded for chlorophyll (73) and viable mutations (36) in 50 krad treatments.

Mutagenic efficiency was also found to decrease with increase in the dose of gamma ray irrespective of the criteria adopted for estimation. The lower doses were more efficient in inducing chlorophyll and viable mutations in all the cases. The increase in lethality, injury and sterility with increasing doses, at rates faster than those for mutations resulted in the lower efficiency of higher doses.

	Percentage	Percentage	Percentage	Mutations	Effe <b>cti-</b>	Efficiency			
Doses	survival reduction at 30 days (Lethality) (L)	height reduction at 30 days (Injury) (I)	pollen fertility reduction (Sterility) (S)	per 100 M <sub>1</sub> plants (M)	veness <u>M x 100</u> krad	<u>M x 100</u> L	<u>M x 100</u> I	<u>M x 100</u> S	
10 krad	-5.7 <sup>*</sup>	4.3	2.6	56.9	569	<u>مل</u>	1323	2188	
20 krad	8.8.	26.1	11.9	44.9	225	510	172	37 <b>7</b>	
30 krad	30.8	39.1	19.2	64.4	215	209	165	335	
40 krad	44.0	43.5	30.9	47.7	119	108	110	154	
50 krad	76.7	59.4	46.0	36.4	73	47	61	79	

Table 12. Mutagenic effectiveness and efficiency (Chlorophyll mutations)

\* Negative value taken as zero for estimation of efficiency.

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	age 1 on áys ity)	ag vys	tion days trage ity lity)	در	Effectiveness	Efficiency			
Do <b>s</b> es	Percenta survival reduction at 30 da (Lethali	Percentage height reduction at 30 days (Injury) Percentage Pollen fertility (Sterility		s Effectiveness offul M x 100 truck trad trad krad		M x 100 L	M x 100 I	M × 100 S	
	(L)	(I)	(S)	(M)					
10 krad	-5 <b>.7</b> *	4,3	2,6	48,4	484	ঝ	1126	1861	
20 krad	8.8	26.1	11,9	60 <b>.7</b>	303	68 <b>9</b>	233	<sup>、</sup> 510	
30 krad	30.8	39.1	19.2	<b>4</b> 9.2	164	160	126	256	
40 krad	44.0	43.5	30.9	40.9	102	93	94	132	
50 krad	76.7	59.4	46.0	18.2	36	24	31	39	

# Table 13. Mutagenic effectiveness and efficiency (viable mutations)

\* Negative value taken as zero for estimation of efficiency.

#### 4.4 Economic mutants

The selected economic mutants and their characteristics are presented in Table 14.

Plants with less than 30 per cent immature pods and ten or more mature pods were selected as economic mutants. The following characters were studied in the selected mutants.

#### i) Days to first blooming

The control flowered 29 days after sowing. Flowering was found to be 1 or 2 days ahead in five of the selected mutants. But in six others it was delayed by 3 to 4 days. The maximum days to blooming recorded was 33.

# ii) Number of pods per plant (Mature and Immature)

The number of mature pods were higher compared to the control in 13 mutants, while in three it was same as in the control. The maximum number of mature pods recorded was 17 as against 8 in the control. The percentage of immature pods were much lower in the mutants indicating earliness in maturity.

S1.	Plant	Days to first	Number	of pods	Percen-	Pod	Haulm	Shell-	Mean	Colour	Fresh seed
No.	Number	bloom- ing	Mature	Immature	tage of immature pods	yield/ plant (g)	yield/ plant (g)	ing percen- tage	kernel weight (g)	testa	dormancy
1.	Control	29	8	12.0	60,0	11.5	95.0	68.0	0.46	Light	Present
2.	.1-8-3	29	8	1	11.1	10.0	50.0	55.0	0.27	pink "	n
з.	1_13_7	27	16	6	27.3	30.0	87.0	43.5	0.29	Brown	57
4.	1-16-8	27	10	З	23.0	22.5	85.0	71.4	0.50	11	त
5.	<b>1–</b> 17–3	33	10	5	33.0	12.0	92.0	50.0	0,20	Light pink	81
6.	1-19-1	31	14	10	41.7	25.0	120.0	65.4	0.43		49
7.	1-19-2	28	10	2	16 <b>.7</b>	20.0	<b>7</b> 5.0	53.3	0.40	11	n
8.	1-20-3	29	10	3	23.0	17.0	100.0	75.0	0,30	n	n
9.	1-20-9	28	17	5	22.7	27.0	90.0	65.4	0.34	1	Ħ
10.	1-22-11	29	14	4	22.2	27.0	65.0	60.0	0.47	11	11
11.	1-23-1	29	14	З	17.6	25.0	95.0	66 <b>.7</b>	0.40	11	11
12.	1-25-3	33	15	8	34.8	35.0	200.0	36.4	0.20	11	ti
13.	1-26-1	29	8	1	11.1	10.0	100.0	5 <b>7.1</b>	0,25	11	ព
14.	1-102-1	27	14	4	22.2	30.0	120.0	60.0	0.33	11	11
15.	2-1-3	33	11	6	35.3	20,0	67.0	53.5	0,33	17	11
16.	2-6-1	33	10	4	28.6	12.0	105.0	57.5	0.26	42	R
17.	2 <b>–38–1</b>	33	8	2	20.0	10.0	62.0	43.5	0.17	43	17

Table 14. Selected economic mutants and their characteristics

#### iii) Pod yield per plant

All the selected mutants except three recorded higher pod yield per plant than the control. The pod yield per plant in the control was 11.5 g. Two selected mutants had 12g, and eleven mutants more than 20 g. The maximum pod yield recorded was 35 g. Three mutants recorded lower pod yield than the control(10g).

#### iv) <u>Haulms yield per plant</u>

Haulms yield per plant showed wide variation among the selected mutants the maximum being 200 g and the minimum 50 g. The haulm yield recorded by the control was 95 g. Six selected mutants recorded higher yield than the control.

#### v) Shelling percentage

The shelling percentage was low in majority of the selected mutants. The control had a value of 68. Only two mutants recorded higher values (71.4 and 75) than the control.

#### vi) Mean\_kernel\_weight

All the selected mutants except two recorded low mean kernel weight than that of the control (0.46g) indicating that control had larger kernels when compared to the selected mutants.

# vii) Colour of testa

Two of the selected mutants produced brown coloured testa as against the light pink colour in the control.

# viii) Seed dormancy

The mutants were tested for the presence of fresh seed dormancy and it was observed that in all the selected mutants this did persist. ł

# DISCUSSION

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#### DISCUSSION

Mutation breeding is generally accepted as a complementary method of crop improvement. It is probably the best method where the objective is to effect a specific change in an otherwise acceptable genotype. Groundnut variety EC.119704 was found to be adapted and promising with stable performance and yield potential. Rice fallows in Kerala are the non-traditional but potential areas for large scale cultivation of groundnut. As pointed out by Nair (1978), the lack of adapted and promising varieties maturing in 90 days or less is the limiting factor for commercial production of groundnut in summer rice fallows. The long duration of EC.119704 limits its cultivation in summer rice fallows. Therefore, a research programme was undertaken with the objective of evolving an early maturing groundnut variety by induced mutagenesis in the promising variety EC.119704. Uniform kernels of the variety were exposed to five doses of gamma rays viz. 10, 20, 30, 40 and 50 krads. The  $M_1$  and  $M_2$  generations were evaluated and the results of the study are discussed.

# 5.1 M1 effects

#### 5.1.1 Germination and survival

Germination of irradiated kernels remained unaffected under laboratory conditions except for slight reduction at 20 and 30 krads. Germination under field conditions was found to decrease with increasing doses with a slight increase at the lowest dose. Increase in the dose of gamma rays resulted in a corresponding delay in germination but the lowest dose of 10 krad hastened the germina-This is in agreement to the report of Bilquez and tion. Martin (1961) that x-rays at 8000 R stimulated germination in groundnut while at 40,000 R the germination was reduced. The increase in percentage of germination at lower doses was attributed to the activity of enzymes involved in the synthesis of auxins ( Casarett: 1968). Reduced germination percentage at moderate and high doses can be due to varied response of irradiation on the chromosome compliments or due to adverse physiological effects (Pushkaran, 1983).

The relationship between percentage of survival and the dose of radiation was inverse ie. an increase in the dose of radiation resulted in a decrease in the percentage of survival except at 10 krad. Reduction in survival consequent to gamma irradiation was reported in

groundnut by Sivasubramanian (1979), Ratnaswamy (1980), Pushkaran (1983) and Ramanathan (1984a) in conformity to the present findings. The reduction in survival is an index of post-germination mortality in treated plants due to cytological and physiological disturbances by the effect of radiation. The cytological abnormalities caused by irradiation may lead to structural changes in the chromosomes. This interferes with the normal growth and development of organs which might have led to the reduction in survival with irradiation (Sreekumari Amma, 1985).

# 5.1.2 Plant growth

A reduction in growth of shoot and primary root was observed in the present investigation. The study of shoot and root ratios in different treatments indicated that the inhibition for root growth was more than that for shoot growth. The plant height recorded on 15th, 30th and 45th days indicated that there was reduction in plant height with increasing doses at all the stages. These observations are in accordance with the findings of Gregory (1957), Shivraj and Rao (1963), Patil (1966), Sinha and Roy (1969), Sivasubramoniam (1979), Ratnaswamy, (1980), Pushkaran (1983) and Ramanathan (1984a) in groundnut. The inhibition of growth at higher doses of

the mutagen can be interpreted in physiological, biochemical and anatomical view points, such as inhibition in the rate of assimilation and consequent changes in the nutrient level of plants (Ehrenberg, 1955) and inactivation of vital enzymes especially those concerned with respiration (Casarett, 1968), Sanjeeviah et al.(1967) reported stimulation of growth at lower doses, the stimulatory effect on plant growth at lower doses may be due to the destruction of inhibitory substances and an increase in physiologically active substances like auxin. giberellin etc. The reduction in plant height on the 30th day was more drastic than that on the 60th day. This indicates an apparent recovery of M, plants from injury at the later stages of growth. The recovery might be due to the growth of uninjured meristematic cells which replaced the injured ones as growth proceeded.

#### 5.1.3 Fertility

Pollen fertility indicates the percentage of viable pollen grains which in turn reflects the normal functioning of reproductive system. According to Kivi (1962), reduction in pollen fertility in  $M_{\underline{f}}$  plants is a reliable parameter indicating the effectiveness of mutagenic treatment. An inverse relationship between pollen fertility and radiation dose was observed in this study.

Similar results were recorded by Gregory (1968), Prasad (1972), Mouli and Patil (1979), Pushkaran (1983) and Ramanathan (1984a) in groundnut. According to Gaul et al.(1966) the cryptic structural changes in the chromosomes and chromosomal aberrations are the causes for  $M_1$  sterility following radiation treatments. As the treatment doses enhanced, the deleterious effects of irradiation were more marked in the chromosome compliment which were conclusively proved by cytological observations. Thus meiotic abnormalities including bridges, laggards, fragments, univalents etc. interrupt the normal development of fertile microspores thereby increasing the sterility with increase in the doses (Nair, 1975).

#### 5.1.4 Chlorophyll chimeras

Plants with chlorophyll deficient sectors were found in 20,30 and 50 krad treatments. In groundnut such chimeric plants were reported earlier by Bilquez et al. (1965), Patil (1966), Sinha and Roy (1969) and Pushkaran (1983).

According to Erikson and Lindgren (1970), the M<sub>1</sub> plant following seed irradiation will carry an induced mutation in certain part of the shoot, the other parts being normal or differently mutated. The part containing

the mutation is frequently referred to as a mutated sector of the plant, but the plant may be as well as sectorial or a periclinal or mericlinal chimera. In mutation research it is of great interest to reveal mutated sectors in the  $M_1$  plants. In the present study, plants with chlorophyll deficient patches on the leaves were observed. A plant with one chimeric branch and other normal branches was also observed. This may be due to the fact that only a part of the embryo was affected producing chimeric regions in one branch. Thus the induction of leaf chimeras by the action of radiations may be attributed to changes in the genetic material by induction of chromosomal aberrations and structural disturbances in the grana of the chloroplast in leaves.

#### 5.1.5 Morphological abnormalities

Morphological variations in the M<sub>1</sub> plants depend on the dose and duration of exposure, age, physiological condition of plants and environmental condition during and after the exposure. In the present study, several morphological variations such as enlarged cotyledons, leaf variations, inhibition of root and shoot growth and forking of the main stem were observed in the seedlings. The specific changes which lead to the initiation of such changes are

still unknown. It is probable that physiological disturbances have played a major role in this phenomenon Gigas, stunted and dwarf plants and plants with variations in leaf size and number were also observed. Abnormal plant types were reported in groundnut by many workers like Sinha and Roy (1969); Arzumanova (1970) and Pushkaran(1983). Though the exact genetic reasons for the occurrence of leaf abnormalities following mutagen treatments were not known, Gunckel and Sparrow (1961) attributed these to chromosome breakage, disruption in auxin synthesis and transport, normal metabolism and accumulation of free amino acids.

## 5.2 Mutations in the Mo generation

5.2.1 Chlorophyll mutations

i) Frequency

Chlorophyll mutations have been widely employed for assessing the effectiveness of mutagenic treatments in higher plants. (Gaul, 1964, Nilan et al. 1965, Kawai, 1969). Gaul (1964) stressed the reliability of chlorophyll mutations as the basis for the assessment of effectiveness and stated that

- (1) they are the most frequent gene mutations,
- (2) they can be clearly recognised and classified.
- (3) they can be studied in a small space under semicontrolled green house conditions and

### (4) they provide rapid information as only seedlings need be grown.

In the present study, the chlorophyll mutation frequencies estimated as the number of mutations per 100  $M_1$  plants gave higher values than the frequencies estimated on  $M_2$  plant basis. This evidently was an over estimation of the mutation event in consideration of the differentiated nature of the embryo. Mutation frequencies calculated on  $M_1$  plant basis and  $M_2$  plant basis, followed no definite relationship with the dose. Similar result was observed by Nair and Nair (1977) in sesamum. In general, mutation frequencies increased with the increase in dose. In groundnut, this type of dose frequency relationship was reported earlier by Arzumanova (1970), Sivasubramonian (1979), Pushkaran (1983) and Ramanathan and Rathinam (1983b).

#### ii) <u>Spectrum</u>

Chlorina, viridis and maculata were the spectrum of chlorophyll mutants observed and the relative percentages of these mutants varied in the different treatments. A wide spectrum of chlorophyll mutations were recorded in groundnut by Patil and Bora (1963), Gregory (1968), Sinha and Roy(1969), Patil (1973), Tai et al.(1977), Sivasubramonian(1979) and Ramanathan and Rathinam (1983b). In this study, maculata

was the most frequent type and viridis was the least. Patil (1973) has reported that there are at least three major genes  $V_4$ ,  $Cl_1$  and  $Cl_2$  controlling the development of chlorophyll in groundnut. They are non-allelic and do not show linkage. The chlorophyll deficiency resulting from homozygous recessive condition of all the three loci, results in seedling lethality.

#### iii) Segregation ratio

The segregation ratios of various types of chlorophyll mutants of groundnut in the  $M_2$  generation were studied by Patil and Bora (1963), Sinha and Roy (1969) and Srivastava (1970). It was observed in this investigation that the segregation ratios of chlorophyll mutations were higher than the expected value and there was no dose dependence. The higher segregation ratio indicates severe elimination of initial cells in the seed primordia following irradiation (Sur, 1969). The higher segregation ratios indicates the possibility of obtaining higher frequencies of mutants in  $M_2$  generation. The increase in segregation ratio with an increase in dose will be of great value in mutation breeding.

#### 5.2.2 Viable mutations

The viable mutations observed were plants with differences in growth habit such as dwarf, tall, compact,

spreading and non-branching types. Early mutants and mutants with altered pod and kernel characters and mutants with leaf variations were also observed. Some of the stunted and dwarf mutants had profuse branching giving them a bushy appearance. In such plants the growth and differentiation of the main shoot apex stops very early in the ontogeny while the axillary buds continue further growth. Sanjeeviah (1967) observed in groundnut, varying frequencies of dwarf mutants under different doses of radiations. He has reported that such mutants were more frequent with gamma rays than with x-rays and suggested that the locus or loci concerned are more sensitive to gamma rays than x-rays. In accordance with the present results, Patil (1966): Sanjeeviah et al. (1967), Dorairaj (1979) and Pushkaran (1983) have also reported dwarf mutants in groundnut treated with x-rays and gamma rays. The production of dwarf mutations was attributed to the reduced auxin levels by Gunckel and Sparrow (1967). According Csillery (1980) the number of nodes on the main axis is not altered in dwarf mutants but the reduced internode length is responsible for their short height. Bunchy top mutants similar to those reported by Patil and Mouli (1978) in groundnut were also observed in the present study.

Tall mutants having increased height over the control along with long internodes were isolated in this study. Nayar and George (1969) obtained tall mutants in sesame with increased height, number of internodes and length of internode following gamma irradiation. They concluded that height was controlled by a single gene. It is possible that the gene in the dominant condition blocks the release of some growth promoting hormones in the system and in the recessive condition there is no such blocking. Tall and semi-tall mutants induced by gamma rays and x-rays in groundnut were reported by Patil (1966). These were found to be due to greater length of internodes. The compact mutant observed during this investigation had dark green leaves but failed to set pod. The very compact nature of the mutant was found to be due to the retarded growth of the main shoot. In agreement to the present observations, compact mutants were reported in  $M_2$  of a virginia variety of groundnut by Prasad et al. (1984). It was also observed that the frequency of mutants for canopy characters was much higher in the case of virginia than in the case of spanish varieties. They have suggested that a genetic restructering of peanut plant with a combination of compact canopy and high pod number could be possible in the virginia types. Mouli and Patil (1976) had isolated a mutant in groundnut with suppressed growth

of primary branches and low pod setting. Spreading mutants have been reported by several workers like Levy and Ashri (1975), Rao (1979) and Pushkaran (1983). The non-branching mutants observed during this study were rare in groundnut. However, Pushkaran (1983) has also reported non-branching mutants.

Viable mutants with altered leaf-size, shape, colour and number of leaflets such as curly leaf mutant, cup leaf mutant, narrow leaf mutant, small leaf mutant and mutants with 5 to 7 leaflets were noticed in the present work. Leaf mutants of various types were already reported in groundnut by Patil (1966), Gregory (1968), Srivastava (1970), Mouli and Patil (1979), Mouli et al.(1984) and Prasad et al. (1984). Csillery (1980) identified certain genes responsible for the leaf shape and size although they are pleiotropic in effect. In this study the variations in leaf size and shape may probably be due to the long lasting effects of the chromosomal damage bringing about alterations in the genetic material. The mutability of the gene controlling shape and size of the leaf and with pleiotropic effects may be visualised as a complex phenomenon which can possibly be the ultimate result of mutagen genotype interaction.

Mutants with altered pod characters were also isolated. These include small pod mutant, deeply pod constricted mutant and long pod mutants. Such mutants have been reported earlier by Patil (1974), Mouli and Kale (1982) and Ramanathan and Rathinam (1983b) in groundnut.

In some of the viable mutants isolated, several characters have simultaneously changed. Hammons (1953a) recorded a cup mutant characterised by a complex of morphological features which were ascribed to pleiotropic effects of a single gene. The ability of groundnut to multimutate and yet maintain the phenotypic stability has been indicated and related to the genetic background effects by Gregory (1957, 1961). In groundnut, Ashri and Goldin (1965) observed mutations involving two or three characters at a time and suggested that this may be a pleiotropic mutant with syndrome effect. The loss of a chromosome segment may also result in such effects as stated by Patil (1966).

#### 5.2.3 Mutagenic effectiveness and efficiency

According to Konzak et al.(1965) the term effectiveness is a measure of mutations in relation to dose and efficiency is an estimate of mutation rate in relation to other induced biological effects such as lethality, injury and sterility.

Gaul and Frimmel (1972) were of the opinion that the effectiveness of a mutagen is of theoretical importance but does not have any immediate practical implication, while for practical purposes the aim is to get high efficiency. They defined efficiency as the ratio of chlorophyll mutations to biological change. To obtain high efficiency, the mutagenic effect must greatly surpass other effects in the cell such as chromosomal aberrations and toxic effects which generally lead to The effectiveness and efficiency of mutagens damage. depend on the nature and characteristics of the organism as a whole, as well as on the specific properties of the mutagen. Groundnut has been proved to be an ideal material for genetic improvement through induced mutagenesis and gamma irradiation was successfully employed by many workers like Shivaraj and Rao (1963), Sanjeeviah (1967), Menon et al. (1970), Sivasubramonian (1979), Prasad and Kaul (1980b), Pushkaran (1983) and Ramanathan (1984b). The chlorophyll mutations are taken as a basis for effectiveness and efficiency estimates on the assumption that the other types of mutations are induced with frequencies parallel to those of chlorophyll mutations (Kawai, 1969).

The effectiveness with regard to chlorophyll and viable mutations in this study were the highest at the lowest dose employed and decreased with increasing doses. This inverse relationship may probably be due to the failure of mutation frequency to increase proportionately with increase in dose of gamma rays. Mutagenic efficiency estimated on the basis of lethality, injury and sterility were also found to be the maximum at the lowest dose employed and decreased with increasing doses. According to Konzak et al.(1965), the greater efficiency of lower doses of mutagen may be because of the fact that lethality, injury and sterility increased with mutagen dose at rates faster than those for mutations.

#### 5.2.4 Economic mutants

Plants with less than 30 per cent immature pods and more than 10 mature pods per plant at 90 days were isolated. These mutants included plants which flowered 1 to 2 days earlier than the control. Patil (1972) has reported that days to first blooming can be considered as an index of maturity, since early flowering by a day would make groundnut mature earlier by about 7-10 days. However, no such relationship was observed between days to flowering and days to maturity in this investigation. Early mutants in groundnut were also obtained by Patil and Thakare (1969), Sinha and

Rahman (1979) Mouli et al. (1979), Pushkaran (1983) and Marghitu et al. (1984).

High yielding mutants were isolated based on the number of mature pods per plant. Seven of them had almost double the number of mature pods when compared to the control. Pod yield per plant was about three times that of the control in some of the mutants. This is in agreement to the report of Ramanathan (1984b) that number of mature pods contributed to higher yield in the mutants. The mean kernel weight was less than that of control in most of the selected mutants. High yielding mutants in groundnut were reported earlier by several workers (Dorairaj (1979), Ratnaswamy (1980), Ramanathan (1984b) and Sivaram et al. (1985a). All the selected mutants had fresh seed dormancy indicating that gamma irradiation at doses of 10 to 50 krad did not break seed dormancy in groundnut.

The economic mutants isolated with a combination of desirable attributes like earliness, short and compact canopy, more number of pods, dark green leaves and seed dormancy could be used directly or in cross breeding programmes. The early mutants isolated can be cultivated in the rice fallows. The dwarf and compact mutants isolated can be ideal for companion cropping with tapioca and needs further evaluation for confirmatory results. The practical utility of these selected mutants have to be fully assessed by evaluating them in  $M_3$  and  $M_4$  generations. At least a few of them will be useful in increasing the production of groundnut in the State by fitting well into the rice based cropping system and also as an intercrop of tapioca in uplands during Kharif<sup>2</sup>.

In the present investigation on induced mutagenesis in the groundnut variety EC.119704, a wide spectrum of both academically and economically significant viable mutants have been isolated. The results indicate that groundnut is highly sitable for genetic improvement through induced mutagenesis as suggested by Gregory (1956b) and Norden (1973). The suggestion of Misra (1980) that induced mutagenesis is a tool for breeding for earliness in groundnut has proved to be a reality since early mutants with more number of mature pods could be isolated in this study.

## SUMMARY

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#### SUMMARY

Kernels of the groundnut variety EC.119704 were subjected to irradiation with gamma rays at doses of 10, 20, 30, 40 and 50 krads employing the  $^{60}$ Co gamma source at the School of Genetics, TNAU, Coimbatore. The effect of radiation in the M<sub>1</sub> and M<sub>2</sub> generations are summarised below.

The  $M_1$  effects such as germination (percentage and period), survival ( percentage on 15th, 30th and 45th day), plant height ( on 30th and 60th day) and pollen fertility reduction, chlorophyll chimeras and morphological abnormalities were studied. In the Ma generation, the frequency of chlorophyll and viable mutations were estimated on  $M_1$  plant progeny and  $M_2$ plant basis. The different types of chlorophyll and viable mutants were identified and scored separately and the relative percentages of different types of mutants (spectrum) were calculated. The segregation ratios of chlorophyll mutants were estimated as percentage of mutants to the total number of plants in the segregating progenies. Total mutation frequency was estimated as number of mutations per 100 M<sub>1</sub> plants. Mutagenic effectiveness and efficiency were estimated

using the formulae suggested by Konzak et al. (1965).

Germination of irradiated kernels remained unaffected under laboratory conditions except for a slight reduction at 20 and 30 krads. Under field conditions germination percentage was found to decrease with increasing doses with a slight increase at the lowest dose. Increase in the doses of gamma rays resulted in a corresponding delay in germination but the lowest dose of 10 krad hastened the germination. An increase in the dose of radiation resulted in a decrease in the percentage of survival except at 10 krad. A reduction in growth of shoot and primary root was also observed. The study of shoot/root ratios in different treatments indicated that the inhibition for root growth was more than that for shoot growth. The plant height recorded on 15th, 30th and 45th days indicated a reduction with increasing doses at all the stages. An inverse relationship between pollen fertility and radiation dose was also observed. Plants with chlorophyll deficient sectors were found in 20, 30 and 50 krad treatments. Several morphological variations such as enlarged cotyledons. leaf variations, inhibition of root and shoot growth and forking of the main stem were observed in the seedlings. i,

The chlorophyll mutation frequencies estimated as the number of mutations per 100  $M_1$  plants gave higher values than the frequencies estimated on  $M_2$  plant basis but no definite dose relationship was observed. Chlorina, viridis and maculata were the spectrum of chlorophyll mutants observed and their relative percentages varied in the different treatments.

The viable mutations observed were plants with differences in growth habit such as dwarf, tall, compact, spreading and non-branching types. Early mutants, pod and kernel mutants and mutants with leaf variations were also observed. Tall mutants having enhanced height over the control along with long internode were isolated in this study. Viable mutants with altered leaf-size, shape, colour and number of leaflets such as curly leaf mutant, cup leaf mutant, narrow leaf mutant, small leaf mutant and mutants with 5 to 7 leaflets were noticed. Mutants with altered pod characters such as small poded mutant, deeply pod constricted mutant and long pod mutants were also isolated. In some of the viable mutants isolated several characters had changed simultaneously.

The effectiveness with regard to chlorophyll and viable mutations were the highest at the lowest dose employed and decreased with increasing doses. Mutagenic efficiency estimated on the basis of lethality, injury and sterility were also found to be the maximum at the lowest dose employed and decreased with increasing doses. Among the five doses employed 10 krad was found to be the most effective and efficient.

The M<sub>2</sub> plants were harvested on the 90th day to isolate early mutants. The economic mutants isolated include mutants which flowered 1 to 2 days earlier than the control. However, no relationship was observed between days to flowering and days to maturity. High yielding mutants were isolated based on number of mature pods per plant at 90 days. Seven of them had almost double the number of mature pods when compared to the control. Pod yield per plant was about three times that of the control in some of the mutants. The mean kernel weight was less than that of control in most of the selected mutants. All the selected mutants had fresh seed dormancy indicating that gamma irradiation at doses of 10 to 50 krad did not affect seed dormancy in groundnut. The practical utility of these mutants are to be

confirmed by raising the  $M_3$  and  $M_4$  generations.

The results obtained thus indicate that induced mutagenesis holds promise in the genetic improvement of groundnut. The early mutants isolated can be further evaluated and cultivated in the summer rice fallows which constitute the non-traditional but potential area for the groundnut production in the State. The economic mutants isolated with a combination of desirable attributes like earliness, short and compact canopy large number of pods, dark green leaves and seed dormancy can be used directly or in cross breeding programmes for developing desirable types.

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\* Originals not seen.

# INDUCED MUTAGENESIS FOR EARLINESS IN GROUNDNUT (Arachis hypogaea L.)

By GIGI K. JOHN

### ABSTRACT OF A THESIS submitted in partial fulfilment of the requirement for the degree MASTER OF SCIENCE IN AGRICULTURE Faculty of Agriculture Kerala Agricultural University

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#### ABSTRACT

A research programme was carried out to induce earliness in the promising groundnut variety EC.119704 by irradiating uniformly dried kernels with gamma rays at five doses viz., 10, 20, 30, 40 and 50 krads. In the  $M_1$  generation, germination, survival, plant height and pollen fertility were estimated. In the  $M_2$  generation, studies on chlorophyll mutations (frequency, spectrum and segregation ratio), viable mutations (frequency and spectrum) and economic mutants were carried out. Mutagenic effectiveness and efficiency were also estimated.

Germination and survival in the M<sub>1</sub> generation were found to decrease with increasing doses. But the lowest dose of 10 krad stimulated germination and the higher doses retarded germination. The length of root and shoot decreased with increasing dose. The inhibition of root was more pronounced. Plant height recorded on 15th, 30th and 45th days was found to decrease with increasing doses. Pollen fertility showed an inverse relationship with the dose of gamma rays. Chlorophyll chimeras and morphological abnormalities were also observed.

The chlorophyll mutation frequency estimated on M<sub>1</sub> progeny as well as on M<sub>2</sub> plant basis showed no definite dose relationship. Chlorina, viridis and maculata constituted the spectrum of chlorophyll mutants observed. The segregation ratio also did not show any dose dependence. The viable mutation frequencies were dose independent. A wide spectrum of mutations affecting various morphological characters such as growth habit, duration, size and shape of leaves and pod and kernel characters were isolated, Effectiveness and efficiency decreased with increasing doses of gamma rays. Among the five doses employed, 10 krad was found to be the most effective and efficient. The early maturing mutants isolated with a combination of other desirable attributes like short and compact canopy, dark green leaves, large number of pods and fresh seed dormancy can be cultivated in the summer rice fallows of our State or it can be used in cross breeding programmes for developing desirable types.