CHAPTER - 5

DISCUSSION

The present study reveals that seed germination in *Solanum integrifolium* decreases with the increasing doses of gamma irradiation. Patel and Shah (1974) in *Solanum melongena* and *Capsicum annuum*, Chopra and Singh (1978) in *Guizotia abyssinica*, Siddiqui et al. (1980) in *Luffa acutangula*, Kamla and Rao (1982) in *Brassica campestris* and *B. juncea* have also reported decreased seed germination as an effect of gamma-rays. Delayed germination due to gamma-irradiation, as observed in the present study has also been reported in the seeds of *Corchorus* sp. (Sen and Ghosh, 1968), *Phaseolus vulgaris* (Bajaj et al., 1970) and *Solanum melongena* and *Capsicum annuum* (Patel and Shah, 1974).

The causes responsible for the induction of inhibition in seed germination after gamma-irradiation may be chromosomal genic or physiological depending upon the mechanism involved. Ananthaswamy et al. (1971) recorded metabolic disturbances during the germination of wheat, irradiated with Co$^{60}$ gamma-rays, at 20-200 Kr dose levels. It has been suggested that chromosomes may be the primary site of damage in seeds and the damage may be at the metabolic level before it affects morphology or growth (Errera, 1955). However, Harber and Luippold (1959) reported that high doses of radiations probably destroy all germination
regulators in lettuce seeds. Sideris et al. (1971) reported destruction in the activity of gibberellic acid in vitro through irradiation.

Injurious effect of gamma-irradiation on the size and variations in the shape, arrangement and number of the cotyledonary leaves as observed in Solanum integrifolium has also been recorded in Carthamus tinctorius (Chauhan, 1969), Tectona grandis (Inamdar et al., 1977), Abelmoscus esculentus (Siddiqui et al., 1979) and Solanum melongena (Ahmad and Siddiqui, 1981). The factors responsible for the induction of abnormalities in cotyledonary leaves due to irradiation are not well known. However, Napp-Zinn (1955) suggested that anomaly in the proportion of growth hormones, which is so common under such circumstances, may to certain extent be responsible for abnormalities in cotyledonary leaves. The general disturbances in metabolic pathway due to irradiation may be one of the important factor responsible for such abnormalities (Devreux and Scarascia-Mugnozza, 1964).

The seedling growth in various treatments, as measured on 20th, 25th and 30th day after sowing, clearly indicates that their growth is lesser as compared to the growth in controls, and is inversely proportional to gamma-ray doses. Similar radiation induced inhibition in seedling growth was reported in Hordeum vulgare (Conger and Stevenson, 1969), Capsicum annuum (Iqbal, 1969), Sorghum vulgare (Goud et al., 1970), Guizotia abyssinica
(Chopra, 1972), in peanut (Hall and Silveira, 1976), Vicia faba (Roth, 1978), barley (Palomino, 1979) and Brassica campestris and B. juncea (Kamla and Rao, 1982).

Acute and chronic exposures to ionizing radiations in general, cause stunted growth in plants. This may possibly be due to destruction or inhibition of terminal meristems, destruction of auxin or of its synthesis, disturbance at nutritional level, failure of assimilation mechanism or inhibition of mitosis and/or chromosomal damage leading to secondary physiological changes.

The reduction in growth rate can be explained on the basis of differential killing of meristematic cells due to chromosomal biochemical injury. The cells most damaged will produce only a few cell-progenies so that the recurrence of growth will be only from those cells which are least damaged (Lea, 1955). Sparrow et al. (1961) reported that radiation-induced stunting growth is due to chromosomal damage. Conger and Stevenson (1969) irradiated barley seeds and found that retardation of seedling height and damage to chromosomes are closely correlated. Seedling height is not suppressed until 25 to 30 percent of the cells carry chromosomal anomalies.

Smith and Kersten (1942) found that in peas high X-ray doses (20 Kr) did not materially affect auxin production or transport, though there was gradual increase in destruction from apex to base. Gordon (1955) has shown that enzyme system involved in the conversion of tryptophan to indole-3-acetic acid (IAA) is highly radiosensitive. Gunckel and Sparrow (1961) in a comprehensive review, mentioned that the adverse effect on seedling growth in irradiated seeds might be due to specific effects on certain respiratory enzymes.

While summing up, it appears to be quite logical that chromosomes being hub of various life processes, when damaged due to radiation, lead to various metabolic anomalies which are expressed externally in various forms by the affected plants.

It is evident from the present study that gamma-rays have an adverse effect on plant growth in *Solanum integrifolium*. The number of lateral branches also gets reduced by gamma-irradiation. The general growth inhibition, as an effect of radiation, has also been observed by Brock (1967) in *Arabidopsis thaliana*, Borogevic and Borogevic (1972) in wheat, Nath (1974) in *Sesamum*, Seetharam (1976) in linseed, Kumar and Das (1977) in *Brassica campestris*, Sinha and Sinha (1977) in *Coriandrum sativum*, Chauhan (1978) in *Solanum khasianum*, Sharon and Muridharan (1978) in *Sorghum vulgare* and Singh et al. (1980) in barley.
The ionizing radiations are known to cause inactivation of growth regulators in vivo and in vitro (Gordon and Weber, 1955; Sax, 1963). Sparrow et al. (1956) mentioned that although reduced stem growth is usually ascribed to reduced auxin levels, it may be an over simplification as nutritional levels and mechanism of assimilation may also be important factors. Adverse effects of ionizing radiations on photosynthesis and pigmentation are also well known (Gunckel and Sparrow, 1961; Kurganova and Anisimov, 1978; Singh et al., 1979; Gangwar et al., 1980). Such harmful effects of ionizing radiations lead to various forms of physical expression of damages such as abnormal and retarded plant growth, induction of mutation and death (Maherchandani, 1975).

Inactivation of enzymes, damage to chromosomes, nucleic acid, carbohydrates and other macromolecules are well known effects of radiation (Vant Hof and Sparrow, 1965; Casarett, 1968). The ionizing radiations are known to affect a wide range of physiological and biochemical activities in plants (Khanna and Maherchandani, 1980). On the whole it appears to be a complex biological phenomenon affected by a number of factors operating at cellular and subcellular levels.

Various kind of leaf abnormalities have been seen not only in different treatments but also in control although frequency of abnormal leaf production appears higher in the treated materials and is directly proportional to the gamma-ray doses. These abnormalities include reduction in size and varying degree of change in

The factors responsible for the formation of such type of abnormal leaves are not well understood. Hagen and Gunckel (1958) noted that the morphological changes which appeared under gamma-irradiation stress conditions in Nicotiana glauca and N. langsdorffii and in their interspecific hybrids have consistently shown changes in the free amino acid content. They found, where leaf abnormalities occurred in general, there was a concomitant increase in the free amino acid content of these leaves.

The present study on shoot apex organization in Solanum integrifolium reveals that the apical shoot meristem in this plant is highly sensitive to gamma-rays. Shoot apices, from control and from various treatments, exhibited a marked degree of organizational and behavioural differences. The topography of shoot apex did not remain static, but changed with the advancing age of the shoot apex and on the basis of topography a marked distinction could be made between vegetative, transitional and reproductive apices. For instance, in control apices, at an early transitional stage, the shape became hemispherical and remained as such during late transitional stage. This change was associated with the broadening of the shoot apex. At reproductive stage, it again
became dome shaped. However, in the irradiated apices, a definite tendency towards narrowing and flattening was observed. Reduction in the area of apical shoot meristem and its flattening was more pronounced at a late transitional stage in higher treatments. Virtually the pace and degree of flattening and the reduction in area were directly proportional to the irradiation. Ionizing radiations are known to induce flattening of the vegetative shoot apices in Pinus rigida (Bostrack and Sparrow, 1969), Cyamopsis tetragonoloba (Rai, 1971), Solanum melongena and Capsicum annuum (Patel and Shah, 1974), Sesamum indicum (Nath, 1974) and Carthamus tinctorius (Chauhan and Singh, 1975). The flattening of the shoot apices is probably due to restricted or partially ceased growth activity.

The tunica in control apices is two layered through all the phases of development but in irradiated apices, the inner tunica layer becomes gradually indistinct or discontinuous. Total degeneration or disruption of both the tunica layers has also been observed in higher doses of gamma-irradiation. Greater sensitivity of the inner tunica layer to gamma-irradiation has been observed earlier in many cases such as in Nicotiana tabacum and Coleus blumei (Crockett, 1957, 1968), concord grapes (Pratt, 1959), apple (Pratt et al., 1959), Lycopersicum pimpinellifolium (Kuehnert, 1962), Cyamopsis tetragonoloba (Rai, 1971), Guizotia abyssinica (Chopra, 1972), Sesamum indicum (Nath, 1974) and Carthamus tinctorius (Chauhan and Singh, 1975).
The outer tunica layer has been considered to be more radiation resistant than the inner in *Nicotiana tabacum* (Crocket, 1957), apple (Pratt *et al.*, 1959) and safflower (Chauhan, 1969). Pratt *et al.* (1959) believed that lower frequency of the cell division in the first tunica layer may be responsible for this differential radiation resistance. Varietal differences in radiation resistance of the outer tunica layer have also been reported by Pratt *et al.* (1959), for instance the cells of the first tunica layer in "McIntosh" variety of apple are more resistant than those in "Delicious".

Excessive vacuolation, disruption and disorganization of corpus cells in irradiated apices, as observed in the present study, have also been reported by many workers in other plants (Rai, 1971; Chopra, 1972; Nath, 1974; Chauhan and Singh, 1975). Virtually the enlargement of these cells was found to be directly proportional to the radiation doses. The idea that gamma-irradiation prevents mitotic division and induces cell elongation receives support from the interesting observations made by Schwartz and Bay (1956) and Sicard and Schwartz (1959) on wheat. They reported that in wheat grain receiving half a million roentgen of gamma-rays, mitosis completely ceased, but the seedlings developed by cell expansion only.

The control shoot apices, almost throughout the course of development and irradiated apices, during early vegetative
phase, exhibited cytohistological zonation. However, cytohistological zonation, was not observed in 60 Kr dose in the seedlings even on the 15th day after germination. Injury to the cytohistological zonation was reflected in the vacuolated cytoplasm and uniformly poor staining capacity of the meristem. The apices, which survived initial shock of radiation injury, showed clear signs of recovery during their reproductive phase when the cytohistological zones could be easily distinguished. Such observations are in accordance with those of Nath (1974) in Sesamum indicum and Chauhan and Singh (1975) in Carthamus tinctorius.

If responses of the shoot apices are a key to the growth reaction of the plant, as visualised by Gunckel and Sparrow (1961), the far reaching effects of the formative responses of the apical shoot meristem, in the irradiated Solanum integrifolium, can very well be imagined.

The experimental results of the present study indicate that radiation has an injurious effect on the epidermal tissues of both cotyledonary and vegetative leaves in Solanum integrifolium. There is a gradual decrease in the number of stomata per unit area, in index, in size of the guard cells and pore width, with the increase of dose, whereas the number of degenerated guard cells and of trichomes borne on the vegetative leaves per unit area increases with the increase in the intensity of gamma-ray irradiation. Similar type of observations were also reported by

Certain abnormalities in the development of stomata such as contiguous and twin stomata, unequal guard cells, stoma without pore, stoma with single guard cell, stoma with single subsidiary cell and degenerated guard cells were observed in the present investigation. These features were recorded earlier by Siddiqui et al. (1980) in *Luffa acutangula* and Ahmad and Siddiqui (1981) in *Solanum melongena* L. cv. Pusa Purple Long.

However, the number of spines borne on the vegetative leaves remained almost constant.

A size reduction proportional to that of vessel elements and fibres in plants raised from the irradiated seeds of *Solanum integrifolium* has been observed in the present study. This reduction in length and width of vessel elements is likely to affect water conduction to some extent and therefore it is more likely that this decreasing trend and the increase in number of trichomes per unit area, observed in the present study, might be adoptive features to meet the new situation for reducing the transpiration rate.

A normal flower of *Solanum integrifolium* consists of 5 fused sepals. However, a good deal of variations in the number
of fused and free sepals has been observed in plants at different irradiation levels. Though such a variation was not dose specific, Nath (1974) in Sesamum indicum observed marked variation in the number of floral parts and their shape in the floral bud from gamma-irradiated plants. He observed reduction, as well as, enhancement in the number of floral parts, while in the present study only increase in number has been observed. According to Heslop-Harrison (1967), the development of a plant organ is a two-phase process. The initial phase comprises initiation of primordium, whereas the second phase includes determination and subsequent differentiation in an appropriate manner. Both the phases are highly sensitive to external manipulations, such as application of growth substances, chemical mutagens, ionizing radiations, etc. In the present case, both the phases seem to have been affected by irradiation thus resulting in varying number of fused and free sepals.

The development of anther wall layers in Solanum integrifolium conforms to the dicotyledonous type of Davis (1966) in control as well as in plants raised from the irradiated seeds. The wall of the normal anther comprises an outer layer, the epidermis, followed by 3-4 layers of endothecium, two middle layers and an inner most layer, of the tapetum. The cells of the endothecium develop fibrous thickening only at apical region to make anther dehiscent. Dehiscence is porous. However, identical results have also observed in plants raised from the irradiated seeds and control.
Tapetum is glandular when mature and its cells are mostly 2-3 nucleate in control and in plants raised from irradiated seeds. However, tapetum too, exhibited various structural and behavioural abnormalities in treated materials. Its longevity was also affected in most of the treatments. Tapetal cells are known to play a vital role in the nutrition of developing microspores (Goebel, 1905; Juel, 1915). The chief function of the tapetum is said to be the production and transport of enzymes, hormones and nutritive materials which are utilized in microsporogenesis, apart from the recent, but renewed emphasis on its role in exine formation and in maintaining a pool of nucleosides and nucleotides for DNA synthesis. The exact synchronization of meiosis in anther locules is chemically controlled through the tapetal secretion (Linskens, 1958). According to Singh (1965) development of normal and healthy microspores always follows degeneration of tapetal cells. Several workers who worked on male sterility such as Erichsen and Ross (1963), on *Sorghum*, Kaul and Singh (1966), on *Manihot usitatissimum*, also lend support to this view. These workers observed persistent nature and abnormal behaviour of the tapetum to be the cause of microspore-degeneration. Chauhan (1968) in *Chenopodium album* and Nath (1974) in *Sesamum indicum* have also reported persistent nature and abnormal behaviour of the tapetal cells produced as a result of gamma-irradiation.
Solanum integrifolium shows varying degree of meiotic irregularities as a result of the treatment with the physical mutagen, gamma-rays. Some of the abnormalities are reduction in chiasma frequency, occurrence of univalents, disturbed bivalent associations, stickiness, laggards, precocious, unequal separation of chromosomes and bridges, etc. Generally the frequency of these abnormalities increases with the increase in gamma-ray doses and is mostly dose-dependent.

The chiasmata frequency has been found to increase in some cases due to mutagenic treatments (Mathur, 1934; Darlington and Lacour, 1953), while in other cases it decreases (Sree Ramulu, 1971a; Al-Allaf and Godward, 1979). Lawrence (1961) reported, in Lolium and Tradescantia, that the decrease in chiasmata frequency might possibly occur at two stages, viz. during DNA synthesis and at/or slightly before the stage of chiasmata formation. In the former case the decrease in the frequency of chiasmata may be due to the disturbance in chromosome coiling, failure of or restricted pairing at pachytene and the delay in DNA synthesis, while in the latter case it may be affecting the process leading to chiasma formation. Jain and Basak (1965) reported that structural changes in some of the chromosomes restrict pairing and thus reduce chiasmata formation. Similarly univalents are formed due to cryptic structural changes and restricted pairing (Jain and Basak, 1965; Al-Allaf and Godward, 1979). Univalents appear either due to precocious separation, at equatorial plane or are seen as
laggards. Thus it seems that the occurrence of univalents is the result of desynapsis. Such a phenomenon has also been reported by Li et al. (1945) and Bozzini and Maitini (1971) in wheat, and Krishnaswamy and Meenakshi (1957) in Pennisetum typhoides. Presence of ring bivalents even at metaphase I is probably due to inhibited or incomplete terminalization.

Sticky chromosomes are reported as a result of partial dissociation and altered pattern of organization of nucleo-proteins (Evans, 1962; Katiyar and Roy, 1976). They affected the disjunction adversely. Sometimes they, perhaps, caused clumping of the chromosomes (Prasad, 1965; Shaikh and Godward, 1972; Srivastava, 1979). According to Darlington (1953) stickiness may be due to depolymerization of DNA on the surface of the chromosomes, which is rendered more fluid by the treatment. As in S. integrifolium, Tarar and Dnyansagar (1980) observed stickiness at anaphase also and attributed it to a change in the cytoplasm viscosity. Bose and Saha (1970) concluded that the univalents separating precociously seemed to be a result of desynapsis. According to Roy et al. (1971) precocious separation of bivalents at metaphase I in Cucumis sativus was due to the failure of chiasmata formation in bivalents. The occurrence of bridges at anaphase I has also been reported by several workers and attributed to crossing over between relatively inverted segments (Mc Clintosh, 1931) or to the union of chromatids during meiotic prophase (Rees and Thomson, 1955; Lewis and John, 1966; Newmann, 1966). It is also possible that bridges may result
due to stickiness of chromosomal ends (Sawamura, 1965; Sudhakaran, 1971). Thomas (1961) observed that in some cells interstitial chiasmata in the translocated chromosomes failed to complete terminalization and this results in the formation of a bridge during anaphase. The bridges with and without fragments have also been observed by Ghatnekar (1964), Akhund-Zade (1968), Bose and Saha (1970), Chowdhary et al. (1971) and Tarar and Dnyansagar (1980).

Formation of laggards has been reported by Rangaswami (1935), Ammini (1968), Bose and Saha (1970), Bose and Maiti (1971), Chowdhary et al. (1971) and George (1971) produced as a result of treatment with physical/chemical mutagens. According to Rangaswami (1935) and Magoon et al. (1958) the lagging nature of chromosomes seems to be due to some trouble in terminalization of chiasmata, resulting from changes in homology of the paired chromosomes.

The cells showing unequal division may arise when a bivalent fails to disjoin and moves as a whole to one of the poles. Unequal division of chromosomes has also been reported in Pennisetum typhoides (Krishnaswami and Rangaswami-Ayyangar, 1941) as a result of the use of X-rays, in tomato (Bose and Saha, 1970) by using DES and X-rays and in Rhoeo discolor (Ammini, 1968) by the use of maleic hydrazide.

Precocious separation increases significantly in higher doses in S. integrifolium. It has also been reported in Arachis hypogea raised from X-ray irradiated seeds (Patil and Bora, 1961)
in tomato by X-rays and DES treatments (Bose and Saha, 1970) and in *Turnera ulmifolia var. angustifolia* by gamma-rays and EMS treatments (Tarar and Dnyansagar, 1980).

Unsynchronized movement of chromosomes has also been observed at anaphase I. Similar results have been reported in *Turnera ulmifolia* (Tarar and Dnyansagar, 1980) treated with gamma-rays and EMS. According to them unsynchronized bivalents or laggards might appear due to discrepancies in spindle formation. Sometimes telophase II resulted in 5-groups of chromosomes, most probably due to the disturbed spindle mechanism and non-orientation of chromosomes which resulted in the grouping of chromosomes at more than four poles. Later, these formed many micronuclei in a single microspore mother cell.

According to Muller (1954) mutational process is predominantly chromosomal and not genic in origin, but Westergaard (1957) suggested that while a majority of induced mutations may arise from structural changes in chromosome, but a few point mutations also do occur.

Varying degree of pollen sterility has been observed at different irradiation levels. More than 8 percent pollen grains were found sterile in control populations also but the magnitude of sterility increased with the increasing doses of gamma-irradiation. Such an impact of ionizing radiations on pollen viability has also been reported in large number of cases by Bora et al.

Pollen sterility is an end result for which there could be varied causes. According to Sato and Goul (1967) the induced sterility may be classified into chromosomal, genic or physiological depending upon the mechanism involved. However, boundary lines between these categories are not always very clear. In majority of the cases there are meiotic abnormalities that are responsible for pollen sterility (Sinha and Goodward, 1972; Nath, 1974; Katiyar and Roy, 1974, 1975, 1976; Sinha and Sinha, 1977; Katiyar, 1978; Kumar and Das, 1978). This is simply because of the fact that meiosis is prone to all types of disturbing, influences, (Swanson, 1957). The meiotic abnormalities bear a linear relationship with the radiation dose and that the frequencies of the aberrations in $R_1$ were always higher than those in $R_2$ (Katiyar, 1978). In the present study also the percentage of pollen sterility was relatively much less in $R_2$-populations than in $R_1$. These differences indicate that some sort of recovery mechanism might be operating in the intervening period.
Katiyar (1978) has reported that the induced pollen sterility is dose-dependent, but it is always higher than the detectable meiotic abnormalities. It can, therefore, be assumed that besides the observable meiotic abnormalities some undetectable changes also play important role in the induction of pollen sterility. Das (1955, 1957), while working on X-rayed barley, stressed that in addition to chromosomal aberrations, some genic and physiological changes might also cause sterility.

The differences in tapetal behaviour during microsporogenesis in fertile and cytoplasmic male-sterile lines have been reported in many species (Laser and Larsten, 1972; Overman and Wormke, 1972; Horner, 1977; Warmke and Lee, 1977; Albertsen and Palmer, 1979; Lee et al., 1979). Differences in tapetal behaviour have often been proposed as primary causes of male sterility (Lee et al., 1979).

Therefore, it may be assumed that in the irradiated populations of *S. integrifolium* also, the cytogenetical, physiological and morphological causes were responsible for the sterility of pollen grains.

The megasporangium, megasporogenesis and development of female gametophyte, in control and in plants raised from the irradiated seeds of *S. integrifolium*, basically follow the same course as described earlier by Khan and Siddiqui (1983). The embryo sac development is of polygonum type. However, certain variations
in the arrangement and position of antipodals were observed in the control as well as in the irradiated population.

The regular occurrence of polygonum type of embryo sac development in the ovules of irradiated populations also confirms the stability of embryo sac type observed earlier in Ricinus communis (Singh and Gunckel, 1965), Sesamum indicum and Martynia dyandra (Nath, 1974). Parthasarthy (1938) also observed in Oryza sativa that the male gametophyte was more sensitive to radiation than the female one. Swanson (1957) further observed that the aneuploid female gametophyte could survive the effect of irradiation to a greater extent, than the aneuploid pollen. However, total immunity of female gametophyte from ionizing radiations has not been reported in any plant.

It is evident from the present study that the development of the female gametophyte may be arrested at any stage depending upon the existing cytomorphological and physiological conditions. Barrit and EutoN (1966) found that apple clones produced from gamma-irradiated buds showed retarded and irregular embryo sacs. This was considered by them to be the major cause of reduced seed set. Ohba (1964) reported in Cryptomeria and Alnus that their reproductive organs have the highest degree of radiation-sensitiveness during the initial stages of their differentiation. In the present study also the megaspore mother cell and the functional megaspore stages appear to be most
radiation-sensitive. The cytogenetical and most of the physiological causes responsible for degeneration of pollen mother cells and microspores may hold good in this case also.

The gamma-ray induced degeneration of functional megaspore or female gametophyte results in poor seed setting in $R_1$ generation. Many workers such as Ehrenberg (1955), Mikaelson and Aastveit (1957), Choudhri and Dasgupta (1961) and Kivi (1962) have reported poor seed set in $R_1$-generation as a result of irradiation. However, they could not ascertain the basic cause of it or of the failure of megasporogenesis. Nath (1974) made good attempt to trace back the factors responsible for ovule sterility in *Sesamum indicum*. The opinion of Cave and Brown (1954) may be considered regarding the effects of pollen from an irradiated plant on the embryo development and its subsequent abortion. According to them the male gametes from abnormal or normal but irradiated pollens may affect the embryo development in the following three manners: (i) genetic imbalance, (ii) mechanical difficulties and (iii) toxicity of irradiated chromatin.

The sperm nuclei resulting from irregular mitosis in irradiated pollen would contain not only variable amounts of chromatin but would also be expected to be damaged in a variety of ways. This genetic imbalance and mechanical difficulties produced by the introduction of broken chromosome of the sperm nuclei are bound to lead to ultimate abortion of the developing embryo. Cave
and Brown (1954) have actually observed such mechanical difficulties in the lethal ovules of Lilium.

In contrast to control, the irradiated generation has exhibited higher percentage of short styled flowers and poor fruit setting. A similar observation regarding the short styled flowers has been reported in the case of Solanum melongena in the recent past (Ahmad and Siddiqui, 1981).

Cellular development is most common in Solanaceae (Mohan Ram and Kamini, 1964; Saxena and Singh, 1969a) and the gamma irradiation has not changed the basic developmental pattern of endosperm in S. integrifolium.

The first division in the primary endosperm is transverse in Solanum integrifolium as it occurs in tetraploid Solanum nigrum (Saxena and Singh, 1969a). However, on the other hand, Bhaduri (1936) in Lycopersicon esculentum and Petunia nyctaginiflora and Dnyansagar and Cooper (1960) in Solanum phureja have reported the first division to be vertical. The two cells of the endosperm divide vertically and subsequent divisions occur in various planes resulting in a multicellular endosperm. Considerable amount of endosperm persists around the embryo in a mature seed.

Pattern of embryo development in Solanum integrifolium is of chenopodiad type (Myosotis variation) in control as well as in plants raised from the irradiated seeds. No alteration in the
pattern of embryogeny has been observed in the present study as a result of irradiation treatments. Even the highest dose of gamma rays used in the present investigation, could not alter the basic pattern of embryogeny of this species. These investigation broadly tally with earlier. Studies on the other taxa of Solanaceae (Soueges, 1920,1922a,b, 1936; Bhaduri, 1936; Crete, 1954, 1961a,b,c,d; Jos and Singh 1968; Saxena and Singh, 1969b; Singh and Raghuvanshi, 1977; Ahmad and Siddiqui, 1981).

Several workers have studied embryogeny in relation to gamma irradiation (Eunus, 1955; Mericle and Mericle, 1957, 1960; Devreux and Scarascia-Mugnozza, 1964). Mericle and Mericle (1957) have observed various anomalies in the embryo of barley, such as lack of organization, vacuolation in the cells, collapsed cells, disruption and proliferation in the course of development. Mericle and Mericle (1961) have taken several criteria to measure radio sensitivity in barley. They have observed that earlier stages of proembryo are more radiosensitive than later stages, although irradiation in these may result in the growth inhibition in roots and shoots during subsequent development. According to Devreux and Scarascia-Mugnozza (1964), growth rate of embryo may increase or decrease depending upon the stage and time of irradiation in *Nicotiana tabacum*.

Certain variations in the shape and number of cotyledons have been observed in different treatments. Induction of abnormal cotyledons as an effect of radiation has also been recorded in
Eranthis hemimalis (Hacclius and Reinholz, 1953), in Nicotiana tabacum (Devreux and Scarscia-Mugnozza, 1964) and in Carthamus tinctorius (Nath, 1974). The factors responsible for the induction of more than two cotyledons due to irradiation are not fully understood. However, Napp-Zinn (1955) suggested that anomaly in the proportion of growth hormones, which is so common under such circumstances, may to certain extent, be responsible for the production of abnormal cotyledons in developing embryo.

Seed coat development under various gamma-ray doses was found to be the least affected and the anatomical pattern of seed coat was similar to that of control seeds. Seed coat structure in relation to gamma-irradiation has also been studied by Chauhan (1972) in Carthamus tinctorius, Nath (1974) in Sesamum indicum and Chopra and Singh (1976) in Guizotia abyssinica and these investigators also did not find any variation in the anatomical structure of the seed coat. Thus, it appears that irradiation given at seed stage does not alter the anatomical features of the seed coat in seeds produced on plants grown from the irradiated seeds.

The injurious effect of gamma-rays on fruit set, fruit weight, length and yield as observed in the present study has also been reported earlier in Abelmoschus esculentus (Yashvir, 1977). Reduction in berry size with the increase of gamma-rays doses has been reported by Chauhan et al. (1975) in Solanum khasianum and Ravindran and Chauhan (1981) in Solanum indicum. Indira and
Abraham (1977) observed total sterility in the fruits due to irradiation in tetraploid *Capsicum annuum*. It is very likely that poor seed set in higher doses adversely affected the normal physico-chemical processes which ultimately led to the formation of smaller berries.

It has been observed in the present study that seed set decreased with the increase of dose rate. Mackey (1959) and Bhaskaran and Swaminathan (1962) have reported poor seed set with the increase of ploidy in wheat. Indira and Abraham (1977) have reported poor seed set in tetraploid *Capsicum annuum* due to irradiation.

Bora et al. (1961) have studied that low seed set is due to pollen and ovular abnormalities caused by abnormal chromosomal behaviour during microsporogenesis. The poor seed set observed during the present study can be explained on cytological ground as there were greater meiotic abnormalities leading to pollen and ovule sterility. Poor seed set may also be due to damages caused by ionizing radiation in male and female gametophytes.

Thus, it may be concluded that different doses of gamma irradiation affect adversely the process of seed germination, seedling growth, plant growth, epidermal tissues, xylem elements, microsporogenesis, pollen viability, flower and fruit set and yield, but they do not affect the basic developmental patterns of microsporangium, megasporangium, megasporogenesis, female gametophyte, endosperm, embryo and seed in *Solanum integrifolium*. 
The seedling growth (root and shoot length) was measured on 20th, 25th and 30th day from the date of sowing in controls and plants raised from seed irradiated with different doses of gamma-rays. The seedling growth at different intervals was maximum in control and showed decreasing trend from lower to higher doses of gamma-irradiation. Percent injury in the seedling growth was maximum in 60 Kr and showed an increasing trend from lower to higher doses. The seedlings, treated with 60 Kr, could not survive for more than 30 days after being sown.

Injurious effect of different doses of gamma-irradiation on the vegetative growth such as height of the main shoot, number of lateral branches and vegetative leaves was observed. These showed maximum values in controls and a decreasing trend from lower to higher doses. Percentage of injury for vegetative growth was maximum in 50 Kr and showed an increasing trend from lower to higher doses. Differences in shape, size, margin and apex of the leaves have been observed between control plants and plants raised from the treated seeds. However, production of these abnormal leaves was found very high in irradiated populations.

Effect of various doses of gamma-rays was studied with respect to shoot apex organization, on 15th, 30th and 45th day after sowing and was compared with that of control. The shoot apices in controls and irradiated materials were hemispherical on 15th day, slightly convex on 30th day and dome shaped on 45th day. The surface outline of the shoot apices were more or less flat on 30th day in 40-60 Kr doses.
The two layered tunica was observed in the shoot apices of control and treated material on 15th day. The tunica layers were either indistinct or poorly distinct and showed increased vacuolation on 30th day in 40-60 Kr. However, vacuolation in the cells of tunica layers was more pronounced in 50 and 60 Kr treatments on 30th day after sowing.

The corpus zone in the control apices was composed of smaller cells, rich in cytoplasm both on 15th and 30th, a feature also observed in 2.5-30 Kr treatments. The cells of the corpus on the other hand were much enlarged and depleted in cytoplasm on 30th day in 40-60 Kr.

Epidermal cells on both the surfaces in the cotyledonary and vegetative leaves were variable in shape and size. The lower epidermal cells were sinuous or slightly sinuous while they were straight walled in the upper epidermis in controls as well as in plants raised from irradiated seeds. The leaves were amphistomatic and stomata were of anomocytic type. Occasionally, occurrence of paracytic type of stomata was also observed both in control and treated materials. Stomatal abnormalities, like contiguous and twin stomata, unequal guard cells, stomata without pores, stomata with single guard cell, stomata with single subsidiary cell and degenerated guard cells were also observed. The presence of degenerated guard cells was more common in material exposed to higher doses of gamma-irradiation. The number of stomata per unit area, stomatal index, size of the guard cells and pore was maximum
in controls and decreased gradually from lower to higher doses of gamma-rays. Stellate and capitâte glandular types of trichomes were observed both in controls and in plants raised from treated seeds. The frequency of stellate type of trichomes showed an increasing trend from lower to higher irradiation doses.

The adverse effect of gamma-irradiation on the size of vessel elements and on fibre length has been observed. These showed maximum values in controls and decreased gradually from lower to higher doses of irradiation. The percent reduction in vessel element size and fibre length also exhibited an increasing trend from lower to higher doses in plants raised from irradiated seeds.

Floral organogeny takes place in acropetal succession in controls as well as in plants raised from irradiated seeds. Variations in the number and arrangement of floral parts were observed in treated materials.

Microsporangium is tetrasporangiate and the development of anther wall layers conforms to the dicotyledonous type in control as well as in plants raised from treated seeds. The anther wall layers consist of an epidermis, 3-4 layered endothecium, 2-middle layers and a glandular tapetum, the cells of which may be 1-3 nucleate. The cells of endothecium are non-fibrous except at the tip region. The middle layers and tapetum degenerate during pollen maturity. The longivity of the tapetum was affected in
different treatments and it persisted for quite long time in 40 and 50 Kr treatments. Dehiscence of anther is porous.

Study of various chromosome associations and aberration in the treated population were studied together with microsporogenesis. The characters selected for study were frequency of chiasmata and occurrence of multivalents, univalents, laggards, bridges, stickiness, precocious and unequal separation of chromosomes etc. The chiasmata frequency decreased with the increasing doses of gamma-rays. Number of ring bivalents and univalents were related with the number of rod bivalents because as the frequency of rod bivalents decreased with higher irradiation doses, the number of ring bivalents and univalents increased. Increase in the frequency of the occurrence of unequal division and laggards was significant at 5% level in 30-50 Kr and 40-50 Kr respectively. Although the bridges, stickiness, unsynchronized movement of chromosomes and precocious separations have also been observed to increase with the increasing doses, but their frequencies were insignificant stages of second meiotic division also showed some chromosomal abnormalities, but their frequencies were insignificant as compared to controls.

In M₂-generation the chromosomal abnormalities were generally lower than those of M₁-generation, showing a tendency of recovery. General increase in chiasmata frequency and in the occurrence of rod bivalents was associated with the decreased frequency of the occurrence of ring bivalents, univalents and other
allied abnormalities. The occurrence of bridges, laggards and unequal divisions was high in $M_2$-generation but it was comparatively lower than that of $M_1$-generation.

The microspore tetrads were usually tetrahedral and rarely decussate in controls. Besides these, isobilateral type of microspore tetrad were also observed in treated materials. The mature pollen grains are spherical in outline with a smooth exine. Generally pollen grains are tricolporate, rarely these may be tetracolporate in 50 Kr treatments. Germination of pollen grains is generally monosiphonous occasionally it may be polysiphonous in materials treated with 30-50 Kr doses. The average diameter of thirty pollen grains measured was 16.84 um.

Effect of different doses of gamma-irradiation on pollen viability has been observed in two successive $R_1$- and $R_2$-generation. Percent pollen viability in $R_1$ and $R_2$-generations was maximum in control. There was a significant gradual decrease in percent pollen viability in $R_1$-generation with the increase in irradiation doses. The plants of $R_2$-generation did not show any remarkable decrease in their percentage pollen viability.

Ovary is bicarpellary, syncarpous and bilocular with axile placentation. The ovules are campylotropous, unitegmic, tenuinucellate. The innermost layer of the integument differentiates as integumentary tapetum. The female archesporium is usually single-celled, occasionally two celled, in control as well as in plants raised from treated seeds. The archesporial cell
directly functions as megaspore mother cell, undergoes meiosis and produces a linear megaspore tetrad. Generally chalazal megaspore is functional but rarely the two chalazal megaspores remain healthy and the other two degenerate. The development of female gametophyte is of monosporic, 8-nucleate and polygonum type. Rarely a tendency from monosporic to bisporic development of female gametophyte has also been observed in 50 Kr. Variations in the arrangement of the cells of egg-apparatus and antipodals have also been observed. Occurrence of twin embryo sacs and degeneration of embryo sacs have also been observed in controls and in treated materials but their occurrence are more frequent with higher doses of irradiations.

The adverse effect of gamma-irradiation on flower set was also observed. Heterostyly is a common feature in control and in plants raised from irradiated seeds. The number of long styled flowers was maximum in controls and showed a decreasing trend with lower to higher doses of irradiation, whereas, short styled flowers showed an increasing trend from lower to higher doses. The medium styled flowers did not show any remarkable decrease.

Pollination is anemophilous. Double fertilization has been observed. The development of endosperm is ab initio cellular in controls as well as in plants raised from irradiated seeds. The first division in the primary endosperm cell is transverse forming primary micropylar chamber which divides longitudinally
and primary chalazal chamber which divides transversely forming four cells arranged in inverted T-shaped manner. Variations in the plane and sequence of cell division in the endosperm development have also been observed in controls as well as in treated plants.

The embryogeny is myosotis variation of chenopodiad type. However, variation in the curvature and in size and number of cotyledons present in the mature embryo, have also been observed in controls as well as treated materials.

The mature seeds are smooth, bilaterally, flattened, kidney shaped, endospermic with curved dicotyledonous embryo and cream coloured. The cells of endothelium and middle layers are totally consumed and lignified epidermis alone constitutes the seed coat.

Inhibitory effect of gamma-irradiation on fruit set, length, diameter, weight and yield of fruits has been also observed. They were maximum in control and showed decreasing trend from lower to higher doses. The percent injury for fruit set, length, diameter, weight and yield was maximum in 50 Kr treatment and showed increasing trend with the increase in the intensity of gamma-rays.

Injurious effect of gamma-irradiation on seed set per fruit and weight of the seeds obtained from the control as well as treated populations was also observed. The number of seed per fruit and weight of 100 seeds were maximum in controls and showed
decreasing trend with lower to higher doses. The percent injury for seed set and weight was maximum in 50 Kr and showed increasing trend from lower to higher doses.

On the basis of above findings it is concluded that different doses of gamma-irradiation affect adversely the process of seed germination, growth, epidermal tissues, xylem components, microsporogenesis, pollen viability, flower and fruit set and yield in *Solanum integrifolium*, whereas, the basic developmental pattern of microsporangium, megasporangium, male and female gametophytes, endosperm, embryo and seed do not exhibit any change.