DISCUSSION
Effluent Analysis:

The physico-chemical properties of the effluent (Table A) showed that it has alkaline pH and is rich in total and dissolved solids. The effluent was deficient in dissolved oxygen and possessed a high value of BOD and COD. The effluent contains appreciably large amounts of mineral nutrients and traces of heavy metals dissolved in it. The Indian Standard Institute (ISI) (1974), has recommended various standards for the disposal of industrial effluents. These standards comprise tolerance limits for the industrial effluents discharged into public sewers, inland surface waters, marine coastal areas and on land.

The ISI prescribed tolerance limits for discharge on land for pH, total solids, chlorides and BOD are 5.9 - 9.0, 2,100 mg/L, 600 mg/L, 500 mg/L respectively. It is evident from the effluent characteristic data (Table A) that the values of pH, total solids and chlorides exceeded the prescribed ISI limits. It is therefore obvious that some treatment is quite essential to minimize the pollution potential, before the chemical factory effluent is discharged. However, by diluting the effluent with large quantities of water, it satisfies the ISI limits and thus reduces the pollution load.

The larger amounts of essential plant nutrients like Ca, Mg, K, N and traces of heavy metals like Fe, Zn & Mn.
present in the effluent, indicates that it can be used as an additional source of liquid fertilizer in diluted form for various plants. The application of industrial wastes on agricultural soils are receiving increasing attention, as the effluent often contains plant nutrients which enhances the growth of the plants, Soon et al. (1980), Mohammad Ajmal and Ahsan Ullah Khan (1984 a,b & c), Mukherjee and Sahai (1988), Mishra (1987), Bishnoi et al. (1989), Brijesh Bahadur and Sharma (1989) and Subramaniyan et al (1990 b).

Germination:

The germination is controlled by various external factors like temperature, light, oxygen and external water supply. The first step, for the germination of seed is the hydration of endosperm (Cotyledon) and embryo axis. Initial uptake of water by germinating seed is determined by the balance of imbibitional force of the seed and the osmotic force of the nutrient medium. The process of germination starts with the absorption of a given medium. In the present findings, percentage germination (Fig. 1) was decreased with the corresponding increase of effluent concentrations. This is due to the presence of high amounts of various salts and metals in the effluent.
Which prevented or retarded the intake of water and caused toxicity to the embryo and endosperm. High osmotic pressure caused due to high salt concentration in the effluent might be the major cause for suppressed seed germination in higher concentrations. Decreased seed germination in various plants at higher concentrations due to the stress created by effluents and various salts were reported by Singh (1981), Bhatnagar et al. (1986) Prasad et al. (1988), Pramila and David Sen (1990) and Shitole and Dhavale (1990).

Along with the decreased germination percentage, a delay in seed germination was also observed in higher concentration. The seeds take up water during germination in order to hydrolyse the stored food material and to activate their enzymic system. An absorption takes place by osmosis, the salt content outside the seeds may act as a limiting factor, which might be responsible for the delay in germination (Troug, 1951). Adriano et al. (1973) and Mohammad Ajmal and Ahsan Ullah Khan (1985) also considered high salt content as a limiting factor for germination of seeds.

The plant nutrients in smaller quantities present in the diluted effluents are responsible for the enhancement
or germination percentage in lower effluent concentrations. Our present findings are in agreement with those of Sahai et al. (1979), Agrawal et al. (1980), Stehlik (1986) and Mishra (1988) who all reported an increased germination percentage in different plants due to low effluent concentrations.

The seedling growth increased in lower concentrations compared to control and it reduced considerably with increasing concentrations of effluent (Fig. 2 & 3). The inhibition of germination and subsequent seedling growth at higher concentrations is due to the high levels of total and dissolved solids in the effluent, which enrich the salinity and conductivity of the solute absorbed by seeds before germination. The growth retardation in plants due to excess quantities of micro nutrients, heavy metals, decomposition products as well as other toxic chemicals has been reported by Dolar et al. (1972), Amthor (1983), Rajaram et al (1988) and Karande (1990). These findings agrees with the present study. The increased seedling growth in lower concentrations, compared to control plants is due to the presence of growth promoting plant nutrients in the effluent, supporting the findings of Mohammad Ajmal and Ahsan Ullah Khan (1984 b), Somashekar

Fresh weight and dry weight of the seedlings increased with the advancement of the germination indicating activation of physical processes leading to growth. The decreased fresh and dry weights of the seedlings at higher concentrations (Figs. 4, 5, 6 & 7) is mainly due to the retarded seedling growth in those concentration. The higher amounts of various salts in the effluent, decreases the uptake of water. In general, various salts and metals will interfere the absorption of moisture by seedlings. The reduction in fresh weight is on this account. Sahai et al. (1983), Mohammad Ajmal and Ahsan Ullah Khan (1983) and Deepakumari et al. (1989) have also reported decreased fresh and dry weights in different seedlings due to higher effluent concentrations. The enhanced seedling growth in lower concentration simultaneously increases the seedling fresh and dry weights. This shows the growth promoting effect of effluent in lower concentration. Soon et al. (1980), Singh et al. (1985) and Mishra (1988) also observed increased seedling growth and its fresh and dry weights in different seedlings grown in diluted effluents. The above findings
are in agreement with the present study.

**Growth and Development**

Growth itself is an expression of cell enlargement brought about by the action of water (Vaadia et al. 1961). Considering height as a growth parameter; it was observed that, height of the plant increased with time irrespective of treatments (Fig. 8, Plate 5 A & B). However, in plants grown after the presoaking treatments of seeds in higher effluent concentration, the height decreased considerably. It shows that the initial growth retardation of seedlings due to excess salt contents in the effluent is prolonged till the end of plant growth. Khosla (1980), Clapp et al. (1983), Mohammad Ajmal et al. (1984b) and Sundaramoorthy and Lakshmanacharya (1989) have also reported similar results in different plant species due to various effluents. Bernstein and Hayward (1958) have pointed out, that increased accumulation of soluble salts and its increased osmotic pressure is related to the degree of growth inhibition. Eaton (1942) and Hayward and Wadleigh (1949) have also supported the view, that reduced water availability induced by high osmotic pressure of
the root medium was the factor restricting the growth of the plants.

The increased plant growth at lower concentrations might be due to the beneficial effects of presoaking treatments given to the seeds before germination with effluent containing optimum levels of mineral nutrients. Kidd and West (1918, '19) studied the seed pretreatment with chemicals and suggested that the factors influencing the plant, during early stages of development may also profoundly influence its subsequent life history. An enhanced plant growth by presoaking treatments of seeds with various industrial effluents at lower concentration were reported by Singh (1981), Bhatnagar et al. (1986) and Rajaram et al. (1988) in peas, paddy and black gram respectively. Our present study is in conformity with those of the above work.

As given in the figures (9 & 11), higher concentration of effluents decreased the dry weight of root, stem and leaf, whereas lower concentrations increased the dry weight. The retarded plant growth in higher concentration is the main cause for the reduction of dry weights. The reduced plant dry weights with the advancement of plant growth in higher concentrations is attributed mainly to Na⁺ and Cl⁻.
toxicity. Phytotoxicity due to excess uptake of Na+ and Cl- have been reported by Gates et al. (1966), Wilson (1967), Behera et al. (1980) and Balaguru and Khannan (1982).

The pod number and its dry weight increased in lower concentration compared to control as shown in the Figs. 12, 13 & 21. These values were reduced in higher concentration. The enhanced plant growth in lower concentrations simultaneously increased the number of pods. The retarded plant growth due to the stress created by effluent in higher concentrations decreases the pod production considerably. Sahai et al. (1985) and Singh and Mishra (1987) reported an increased crop production in Phaseolus radiatus L. and in Zea mays L and Oryza sativa L. respectively when they irrigated the plants with diluted effluents.

Growth Indices:

Relative growth rate in guar was positively correlated with NAR (Table - 2). This finding agrees with the earlier findings of Bhaskar (1983) and Skariah (1988). The leaf weight ratio was higher and NAR was less (Figs. 15 & 16) in plants grown in higher concentration. This may be due to the slow growth of plants affected by
effluents. LWR decreases as the growth proceeds, suggesting the greater mobilization of photosynthates from leaves (Friend 1966, '69) as indicated by higher NAR. Further supply of energy for substrates is entirely depended on photosynthesis and the NAR is the net balance after respiration. Thus higher rate of photosynthesis in lower concentration of effluent pretreated plants results in higher NAR and dry matter production of plants. This is clearly evidenced by higher total dry weights (Fig. 9) in plants grown in lower concentration, indicating higher RGR (Fig. 14). Effluent pretreatment in lower concentration of seeds, appear to accelerate metabolic activities resulting in production of more plastic materials which are mobilized to the apical region at faster rates.

The NAR was found higher in lower concentrations compared to control. As the NAR was higher, the utilization of assimilates was faster, obviously leaf weight was lesser. This indicates that translocation of photosynthates occurred from leaves to the growing region. Similar results were reported by Bhaskar (1983), Sahai et al. (1985) and Mary and Vivekanandan (1990). Increase in NAR was interpreted as increase in net photosynthesis by
Throne and Evans (1964) and Sweet and Wearing (1966). Negative correlation of LWR with NAR in the present study agrees with the earlier findings of Vora (1977) and Friend (1966).

Inadequate utilization of photosynthates allowing accumulation of dry matter in higher concentration resulted into the retarded growth as evidenced by less production of leaf (Fig. 10). This may be the reason for lowest value of NAR and increased LWR in higher concentration. Under effluent stress, the assimilates produced were being utilized simultaneously, but since the supply (assimilates) was greater than utilization, there was accumulation of assimilates in the leaves. LWR was therefore increased in higher concentration. Low rate of net assimilation in higher concentrations resulted in higher leaf dry matter accumulation and less pod weight at harvest time. Net assimilation rate being higher during pod development time indicates that pods are also actively taking part in photosynthesis. Inananga et al. (1979a, 1979b) and Skariah (1988) reported the case of pod photosynthesis in rape and radish respectively.

Yield components

The yield in terms of number of pod per plant,
number of seeds per pod, dry weight of 100 pods and dry weight of 1000 seeds were improved in plants treated with lower effluent concentration compared to control (Table 2a). These values decreased in higher concentration. Chinoy (1968) pointed out the importance of pretreatment of seeds with various chemicals and concluded that, the pretreated seeds has an ability to extract sufficient moisture for quick and better germination even under condition of water stress. Accelerated germination helps the quicker establishment of the stand of the crop. Consequently, the roots penetrate to deeper layers quickly and absorb more water for a quicker growth of the crop. All this would result finally in an increased crop yield.

In our studies, the pretreated seeds in lower effluent concentrations accelerated the seed germination and attained quicker growth over higher concentrations. This accelerated growth in lower concentrations helped the plants for a better yield. The pod production processes in lower concentrations are relatively more active and beneficially prolonged as evidenced from the higher values of pod number. Again these functions are further substantiated by higher values of NAR, in case of plants raised from the pretreated seeds in lower effluent concentration. However, the presence of larger quantities
of various salts in higher concentration of effluent, delayed the seed germination and seedling growth in these concentrations. The retarded plant growth reduces the crop productivity and finally the yield components in higher concentrations.

The yield in plants irrigated with diluted effluents increased compared to control (Table - 3). The enhanced plant growth due to the presence of growth promoting plant nutrients in the effluents simultaneously increase the crop yield. Harvest data (Table - 3) confirms the beneficial effects of the effluent in diluted form, mentioned above by the increase in the growth characters like dry weight of shoot, leaf, plant, number of pods and 1000 grain weights. A favourable yield responses to low effluent concentration has been noted earlier for number of crops e.g. yield of kidney bean and pearl millet were improved by low concentration of diary processing effluent (Ajmal et al. 1984 c) of rice and tomato by paper factory effluent (Rajanna and Obisami, 1979) of Kidney bean and lady's finger by textile factory effluent (Mohammed Ajmal and Khan, 1985) of Phaseolus radiatus by distillery effluent (Sahai et al., 1985) of maize and paddy by fertilizer factory effluent (Singh and Mishra, 1987) and of guar by
Chemical factory effluent (Ramesh Kumar et al., 1990). Our studies support the findings of the above workers, who reported positive results through effluent application.

**Photosynthetic pigments:**

**Seedlings:**

Chlorophyll pigments like total chlorophyll, chlorophyll-a and chlorophyll-b were increased in lower effluent concentrations compared to control (Fig. 22). In seedlings, treated with high concentration of effluents, all the pigment contents were reduced considerably. The increased pigment content in seedlings treated with the lower concentrations of effluent, may be due to the favourable effects of Ca, Mg, K, Na and Cl on pigment synthesis (Cottenie, 1973; Singh and Mishra, 1987 and Sahai et al., 1985). The reduction in photosynthetic pigments observed in seedlings treated with high concentrations of effluent may be attributed to salinity induced by excessive soluble salts in the effluent. The loss in total content of chlorophyll pigments is the usual symptom of salt stress in higher plants. Ponomavera (1971) and Divate and Pandey (1981). Agrawal and Mehrotra (1978) reported a reduced chlorophyll pigment in rice seedlings due to high salt
content in the irrigated water. Sahai and Srivastava (1986), Bhatnagar et al. (1986) and Deepa Kumari et al. (1989) reported an increased pigment content along with the enhanced seedling growth in different plant species due to lower effluent concentrations. They also observed a reduced photosynthetic pigments along with the retarded growth of seedlings in higher effluent concentrations. The rapid growth of seedling and increased amount of pigment contents in lower concentration and the retarded seedling growth along with decreased pigment contents in higher concentrations, observed by us are in agreement with the above findings.

Leaf:

The chlorophyll pigments in leaf increased maximum at the time of flowering and fruit setting stages in all the plants irrespective of the treatments. (Fig. 23). The pigment contents increased, slightly over control in plants treated with lower effluent concentration and these values reduced in higher concentrations. The enhanced plant growth in lower concentration, simultaneously increases the pigment contents. Jha and Gupta (1988) and Senhal et al. (1988) reported an increased chlorophyll pigments in rice
and sugar cane grown in diluted effluents. The retarded plant growth reduced the pigment content in higher concentrations. Similar results were reported by Saxena and Jabeen (1989) and Deepa Kumari et al., (1989).

**Histochemical changes**

Terminal portions of a plant viz, the root and shoot apices carry the potency for proliferation and organogenesis throughout its life span, Nougrede (1967). Structural organization and chemical characteristics of these regions are liable to modify under altered environmental conditions. Growing apices of a young seedling get their nourishment through the translocation of food from the storage region by the action of various enzymes. Thus the manifested performance of the apical region of a young plant is largely dependent on the metabolic processes taking place in the apical and storage region as well as its interactions.

There are a number of external factors which adversely affect the metabolic processes and development of a plant. Dissolved salts in the growing medium can bring about a potential difference in the cell, which may affect the qualitative and quantitative properties of the macromolecules. Through in situ localization and
quantification of certain macromolecules i.e. RNA, insoluble polysaccharides and total proteins, an attempt was made to evaluate the action of effluent at different dilutions on the cytochemical characteristics of the growing regions and storage part of the developing seedling.

The cell area of the shoot apices and primary leaves in 10% pretreated seedlings increased over control (Table - 10, Fig. 26). In 60%, the cell area was found reduced both in shoot apices and primary leaves. Whereas the cotyledonary cell area increased in 60% pretreated seedlings over control. In 10%, the cotyledonary cell size decreased compared to control (Table - 10). Growth of the apical meristem is sensitive to a small decrease in plant water potential, Barlow et al. (1977). Our present results supports this view; the larger amounts of various salts in the effluent medium at 60% pretreated seeds prevented the absorption of water for the growing embryo through the seed coat by creating an osmotic stress. The reduced water absorption might have affected the metabolic processes, thereby reducing the cell elongation and cell division. The reduced rate of cell division in these seedlings also decreases the length of shoot apex (Fig. 26, Plate- 7 a, b &
The enhanced seedling growth in 10% due to the favourable effects of pretreatment might have contributed to the increased cell proliferation and cell size compared to control seedlings.

Both RNA and protein contents were increased in all the three organs of 10% pretreated seedlings compared to control (Table - 10, Plate - 6). In 60% pretreated seedlings, the RNA and protein contents in shoot apices, primary leaves and cotyledons decreased when compared to 10% and control seedlings. However, the insoluble polysaccharides increased in all the three regions of 60% pretreated seedlings over 10% and control (Plate - 7 d, e & f). Maximum amount of polysaccharides was observed in cotyledons compared to primary leaves and shoot apices.

It is well known that RNA synthesis is essential for effective cell enlargement (Key, 1964). In the present study, the RNA content in all the three regions reduced along with the retarded seedling growth in 60%. The reduced RNA synthesis affected the protein metabolisms directly and it decreased the protein synthesis. The increased level of RNA content in 10% effluent pretreated seedlings also enhances the protein synthesis as evidenced by the increased amount of protein contents in shoot.
apices, primary leaves and cotyledons. The biochemical estimation also revealed an increased amount of protein and RNA in both embryo axis and cotyledons (Figs. 32 & 38). The increased levels of protein content in the shoot apices and its reduction in the cotyledons is due to the faster mobilization of food reserves from the storage tissue to the growing regions. The increased levels of insoluble polysaccharide in the shoot apices, leaves and cotyledons of 60% pretreated seedlings may be due to their accumulation in larger quantities resulting from less degradation of these macromolecules. In 10% and control seedlings, the amount of polysaccharides reduced in all the regions. Here the polysaccharides might have hydrolysed and used for more energy liberation through increased respiration, to result in enhanced growth of the seedlings. The remarkable increase of reducing sugars in the embryo axis and cotyledons of 60% pretreated seedlings (Figs. 28 & 29) proved by biochemical analysis could be related to the above mentioned results of polysaccharide status revealed in the histochemical study.

**Mineral constituents:**

All green plants require the same basic set of mineral nutrients and the various elements are used by different
plants for essentially similar ends. Therefore all plants possess mineral uptake mechanism. The plant species differ markedly in accumulation and requirement for different metals, Hodgson (1970), Leeper (1972) and Page (1973).

The potassium content in seedlings treated with higher concentration of effluent were found decreased (Table - 11). In lower concentrations, the amount of K increased compared to control. Ehrler (1960) and Gates et al. (1970) reported a decreased amount of potassium content in rice and glycine due to an increase in NaCl in the root medium. Mohammad Ajmal and Ahsan Ullah Khan (1985) observed an increased amount of potassium in kidney bean and in lady's finger irrigated with diluted effluents. Our present work is in conformity with the above results.

Calcium has long been known to make an ameliorating effect on growth of plants under saline condition, Epstein (1972) and Vora and Patel (1975). The amount of calcium decreased in higher concentration and it increased in lower concentrations of effluent pretreated seedlings over control (Table 11). An appreciably larger amounts of NaCl in higher concentrations of effluent, decreases the uptake of calcium by plants. Low levels of Ca\(^+\) increased the
membrane permeability (Lakaye and Epstein, 1971) leading to an increase of Cl\textsuperscript{-} and Na\textsuperscript{+} in the salt treated plants. High concentration of Na\textsuperscript{+} and Cl\textsuperscript{-} observed in the 60% effluent treated seedlings agrees with the above statement.

Magnesium content decreased in higher concentrations and increased over control in 10 and 30% (Table - 11). Decreased amount of Mg due to high concentration of NaCl were reported by Divate and Pandey (1981) and Reddy (1984) in grape and bajara respectively. Mohammad Ajmal and Ahsan Ullah Khan (1985) observed an increased amount of Ca, Mg & K in Kidney bean and Lady's finger irrigated with diluted effluents. They observed an increased plant growth along with the increased minerals in lower concentrations. The increased seedling growth and larger amounts of Ca, Mg and K observed in lower concentrations in the present study are in agreement with the above findings.

The sodium and chloride in all the concentrations increased with the increase in effluent concentrations. In control, the Na\textsuperscript{+} and Cl\textsuperscript{-} were slightly reduced compared to 10%. The presence of larger amounts of Na\textsuperscript{+} and Cl\textsuperscript{-} in the effluent at higher concentrations reduces the uptake of
other elements through the root medium. Of all the minerals observed in the seedlings of Guar, Na+ and Cl- were accumulated maximum. Malakondiah and Rao (1979) working with peanut and salinity, reported more accumulation of Na+ and Cl- than K+ and Ca++. Greenway (1965) and Gates et al. (1970) reported the similar results. An increased amount of Na+ ions were observed by Mohammad Ajmal and Ahsan Ullah Khan (1985) in Kidney bean and lady's finger treated with higher concentration of effluent. The above findings agrees with the present study.

**Carbohydrate Metabolism**

Both total and reducing sugars showed the same trend in embryo axis and cotyledon (Figs. 28 & 29). Total and reducing sugars increased with the advancement of germination. Total and reducing sugar content in embryoaxis and cotyledon at higher concentrations increased considerably. As the germination advanced, there was a metabolic turnover of sugars, e.g. the respiratory activity increased by which sugars are oxidized and their level was decreased in lower concentrations; conversion of sugars into other carbohydrates can also occur. However, since the sugars were utilized only for osmotic adjustments,
interconversion into other cellular polysaccharide could not occur; thus, there was a lesser availability of those cellular constituents probably for cellulose, which results into retardation of extension or elongation of root and shoot as observed by Kaufmann (1968). The presence of high amounts of metallic salts in the effluent enhanced the total sugar content to a considerable extent in higher concentrations and accumulated in the embryo axis due to effluent stress. This supports the theory that sugars accumulate, wherever there is stress (Parker, 1972; Maranville and Paulsen, 1970). The higher concentration of total and reducing sugars in the embryo axis might be due to the translocation of sugars from cotyledon to embryo axis in order to overcome the stress condition.

A number of workers have ascribed the protective role of sugars against water deficit. Our results confirm the findings of Wadkar et al. (1984) who reported an increased amount of sugar content in the leaves of *Cynodon dactylon* grown in polluted water. Antony (1980) and Shitole and Dhavale (1990) also observed an increased sugar content due to salinity in bajara and green gram and Safflower respectively. During water stress, sugars help in maintaining turgidity and protoplasmic constituents.
(Maranville and Paulsen, 1972) by replacing water crystal lattice on the protein (Parker, 1968). Thus an increase or accumulation of sugars during salt stress has a protective role.

Invertase activity of the embryo axis and cotyledon decreased with germination hours (Fig. 27). The higher concentration of effluent treatment decreased the enzyme activity both in embryo axis and cotyledons. Depressed invertase activity resulted in a greater accumulation of sugars observed in the present study is in agreement with that of Sandhya Giri (1981). The increased sugar content may be considered to be an adaptation of tissue, by which an osmotic adjustment is maintained by the cells. An inhibited invertase activity in bajara and green gram due to various salts were reported by Antony (1980). The increased levels of proline in higher concentrations (Fig. 35) lead to conclude that, in order to overcome the adverse conditions created by the excess amounts of various salts in the effluent, the seedlings might have utilized their carbohydrate resources to synthesise osmoregulatory substances.
Protein Metabolism

Protein metabolism is one of the fundamental events of the cell. The protein content decreased with time in all the seedlings irrespective of the treatments. Decreased protein content with increasing concentration of effluent observed in our studies (Fig. 32) supports the results of earlier workers (Sheoran and Garg, 1978; Bisht and Agrawala, 1980 and Sahai, et al., 1985).

The protein content of embryo axis and cotyledon decreased in high effluent concentration and it slightly increased in 10% compared to control. The decrease in protein content can be correlated with the decreased growth of seedlings as measured by fresh weight, dry weight and seedling growth. Behera and Misra (1983) reported a reduced rate of protein synthesis in rice seedlings along with the increase of effluent concentration. Our findings agrees with the above work. Presence of high concentration of various cations and anions in higher effluent concentrations suggests, a close similarity for the decrease in proteins induced by the effluent stress.

The stunted growth of seedlings (Figs. 2 & 3) may be due to the decreased protein synthesis, as protein are
major constituents of protoplasm. The protein contents in the embryo axis was found more than that of the cotyledons. Effluent pretreatment favoured the faster mobilization of protein from the cotyledons as evidenced by corresponding transient increase in embryonic axis and subsequent decrease might indicate their utilization, inorder to provide nutrition for the growing embryo axis.

The decreased protein content was evidenced by the enhanced protease activity (Fig. 30) which was, in accordance with the findings of Todd et al. (1970), Sheoran and Garg (1978) and Karande (1990). The reduced hydrolytic activity of protease in the pretreated seedlings at lower effluent concentrations, leaves no room for doubt whatsoever that hydrolysis of proteins is going on at a faster rate in lower concentrations compared to higher effluent concentrations. Stimulation of protease activity might have resulted into formation or synthesis of a new type of protein. It is known that plants acquire resistance by the synthesis of new proteins (Genkel, 1970). Mane and Shitole (1988) reported, an increased protein content in methi leaves compared to control plants, when they irrigated the field with paper mill effluent.
Soluble protein content increased in higher concentrations, both in embryo axis and cotyledons (Fig. 31) whereas, the amino acid content decreased considerably (Fig. 33). The increased protease activity could be attributed to the increased soluble protein content and less accumulation of amino acids and protein. The increased amino acid content in the embryo axis and its reduction in the cotyledons found in the present study is due to the translocation of amino acids to the growing embryo axis (Fig. 33). Decreased protein content, free amino acids and increased protease activity and soluble protein found in the present study in higher concentration (Figs. 30, 31, 32 & 33) would lead to conclude that inhibition of protein synthesis occurred due to the stress created by various salts and metals in the effluent. Similar results were reported by Reddy (1984) and Mehta and Bharati (1983) in bajara and gram seedlings by various salts.

Accumulation of free proline in adverse environment is the most striking feature of plants. Figs. 34 & 35 shows that, both in seedlings and leaf more proline contents were accumulated in higher concentration. It slightly decreased in 10% effluent treatment compared to control. Chu et al. (1976 a) reported that proline plays a protective role.
for plants in adverse conditions. The accumulation of larger quantities of Na+ and Cl- found in the seedlings (Table - 11) treated with higher concentrations of effluent reduced the seedling growth by creating stress condition. The enhanced seedling growth at lower concentration reduced the proline content. Stewart et al. (1966) and Singh et al. (1973) have suggested that proline acts as a storage compound and might be the major source of energy and nitrogen required by the plants for rapid recovery from adverse conditions. Treichel (1975) showed a close relationship between proline accumulation and osmotic potential of the irrigated solution. This suggests that the increase of free proline is a water stress effect rather than salt. Stewart and Lee (1974), Shanta and Karadge (1989) and Shitole and Dhavale (1990) reported an increased free proline content in plants due to various salts and water stress.

Higher concentration of proline in the seedlings and leaves observed in our studies may be either due to the breakdown of protein or due to denovo synthesis. However, there are evidence that the accumulation of proline during stress conditions is due to denovo synthesis (Thompson et al. 1966; Barnett and Naylor, 1966

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and Bogers et al., 1976) rather than break down of protein.

Breyhan et al. (1959) and Rena and Splittstoesser (1974) suggested that proline could serve as a precursor for chlorophyll via conversion to glutamate and it could be incorporated in chlorophylls. Such an incorporation might take place during recovery from stress. Consequently proline could serve as a precursor pool for chlorophyll synthesis immediately following the alleviation of stress (Jager and Mayer, 1978). In our study, the data of proline and chlorophyll (Figs. 23 & 35) obtained suggests that accumulated proline is utilized for chlorophyll synthesis when adverse conditions are relieved.

Phenols:

A major role has been assigned to phenolic acids as regulators of plant growth and development. Vendring and Buffel (1961) and Wain and Taylor (1965) found analogous nature of phenols to growth regulators. The phenolic contents showed fluctuations as germination hours advanced (Fig. 34). In control and lower concentration of effluent treated seedlings, the phenolic contents decreased with time whereas in high concentrations it increased with
germination hours. High levels of phenols and decreased plant growth in higher concentrations observed in the present study are supporting the results of Kefeli et al. (1969) and Kefeli and Kutacek (1977). They correlated reduced plant growth with high levels of phenols. Phenols are secondary metabolites which affect growth in different manner by lowering the activity of hormones like IAA, GA and cytokinin. Phenols cause depression in the biosynthesis of the IAA precursor L-tryptophan, Kefeli and Kutacek (1977).

Gradual decrease in phenolic contents with the advancement of growth in lower concentrations explains that phenols were utilized for regulation of plant growth. However, high levels of phenols in higher concentrations disturbed the plant growth. A perusal of literature gives any reference on phenolics changes due to industrial effluents.

Ribonucleic acid metabolism:

The RNA content in embryo axis increased with time in all the concentration irrespective of the treatments (Fig 38). In control and lower concentration the RNA content increased markedly. In the seedlings treated with higher effluent concentration these macromolecule showed a continuous decline. A continuous decrease in RNA content
in rice seedlings. simultaneously with an increase in effluent concentrations and its increase in lower concentration were reported by Behera and Misra (1983). The above findings is in agreement with the present study. The decreased RNA content also influenced the protein content and retarded growth of the embryo axis as the protein synthesis was inhibited. The inhibition of protein and nucleic acid synthesis under salt stress is well known, Rauser and Hanson (1966) and Pathak et al (1974).

A continuous increase of RNase activity (Fig. 36) with time, in higher concentrations decreased the RNA content in both embryo axis and cotyledons. It is obvious that decreased RNA content is due to the process of hydrolysis of RNA by stimulated RNase activity under effluent stress. During stress, most hydrolysing enzymes are stimulated as they are released by the breaking of lysosomes and lipoprotein membrane as the cells are shrinked due to water stress as remarked by Vieira-da-silva (1976). Sandhya Giri (1981) reported an increased RNase activity and decreased amounts of RNA content in gram and tur seedlings due to NaCl, MgCl₂ and CaCl₂ salts.
In recent years, lot of attention is given to peroxidase, due to its implication in large number of biochemical and physiological processes, its occurrence as multiple organ specific isoenzymes which may change quantities and qualities during growth, Yip (1964) and Shannon (1969).

The peroxidase activity of embryo axis decreased with time, whereas in cotyledon it increased as the growth advanced (Fig. 41). The peroxidase activity increased in embryo axis of higher concentrations and it decreased in lower concentration compared to control. The increased peroxidase activity in higher concentrations could possibly be due to denovo synthesis of enzyme proteins. Farkas et al. (1964) reported that the increase in activity of enzyme under stress is associated with destruction of cellular membrane and the concomitant release of previous immobilized protein. Behera and Misra (1985) reported a decreased peroxidase activity in rice seedling with an increase in effluent concentration and the effluent at 133.
lower concentrations did not effect the peroxidase activity.

Peroxidase activity increased in plant cells under various stress conditions such as influence of toxic gases or mechanical injuries to plants. Increased peroxidase activity has been cited as an indicator of physiological stress, Levitt (1972). Gardiner and Cleland (1974) observed an increase in peroxidase activity and the cessation of cell elongation occur simultaneously indicating that peroxidase might control the process of cell elongation. The retarded seedling growth in higher effluent concentration is attributed to the increased peroxidase activity in the present study. Halevy (1962) reported a significant inverse correlation between peroxidase activity and growth rate in cucumber seedlings. From the above works, it can be inferred that inhibition of growth increases the peroxidase activity.

Leaf:

The leaf peroxidase activity fluctuated with the advancement of growth. In control and lower concentrations, the peroxidase activity decreased compared to higher concentration (Fig. 42). The retarded plant growth in
higher concentration may be the reason for increased peroxidase activity. Reduced rate of photosynthesis may also effect the peroxidase activity in plants, Robert (1984). The enhanced plant growth and increased amount of chlorophyll pigments observed in lower concentrations (Figs. 9 & 23) may also show decreased peroxidase activity. Depressed catalase activity is also associated with the increased peroxidase activity, Nikolayevskii (1968) and Judel (1972). Decreased catalase activity and increased rates of peroxidase activity observed in the leaves of higher concentrations of effluent pretreated plants agrees our results with the above findings.

Catalase activity

Considering the effluent effect on oxidizing enzymes, it is clear that catalase activity of embryo axis and cotyledons was decreased in seedlings treated with high concentrations of effluent (Fig. 39). In lower concentrations, catalase activity increased compared to control. The pollutants may alter the catabolic activities either damaging the tissue or by disturbing the metabolic pool. Catalase directly affect the oxidation and reduction
of Cytochrome 'C' oxidase system in mitochondrial respiration, Yokoyama (1956).

The early rapid growth and vigorous cell division resulted in the higher rate of respiration by which large amounts of $H_2 O_2$ is produced inside the cell. To eliminate this toxic substances in the cells, the catalase activity was found more in seedlings of lower effluent concentrations, where a considerable increase in seedling growth was observed. The retarded growth of seedlings treated with higher concentration of effluent, decreased the production of $H_2 O_2$ and as a result, the catalase activity was found reduced. The decreased catalase activity could be correlated with the decreased growth due to the stress created by effluent in higher concentrations. The catalase activity was inhibited due to the lack of substrate as the rate of respiration was less. Depressed catalase activity by salts in gram seedlings was reported by Bharadwaj (1964). A marked decreased in specific activity of catalase in the embryo axis and cotyledons due to excess supply of heavy metals were reported by Ashok Kumar and Bisht (1986). The above works are in conformity with the present findings.
Leaf

Like embryo axis and cotyledons, the leaf catalase activity (Fig. 39) was also reduced in plants raised after the presoaking treatment in higher effluent concentrations. In lower concentrations, these values increased compared to control. At the time of flowering and pod setting stages the leaves of all the plants, irrespective of the treatments showed a sharp increase in catalase activity and the enzyme activity decreased considerably at the time of harvest. The increased growth in lower concentration may be attributed to the enhanced activity of enzyme catalase. The sharp decrease in catalase activity nearing to harvest in all the plants irrespective of the treatments may be due to the beginning of senescence. From the above results it is clearly evident that a positive correlation is existing between the plant growth and catalase activity. The decreased proline content, peroxidase activity and increased rate of photosynthetic pigments found in plants treated with lower effluent concentration may also indicate that the increased plant growth leads to high rate of catalase activity. A depressed catalase activity was reported by Agrawala and Mehrotra (1978) in rice leaves grown in irrigated water containing excess amounts of
salts. Agrawala et al. (1977) observed a decreased catalase activity in corn, soyabean and in barley leaves supplied with excess amounts of heavy metals in the root medium.

The effluent used for the present study altered the plant metabolism in many ways. The stress created by the effluents at higher concentrations produces retardation in seedling growth and arrests the metabolic rates. Presence of high concentration of various cations and anions and the interaction among different cellular salts deposited in the seeds during the presoaking treatments suggests that effluent stress induced conformational changes in enzyme molecules. All the enzymes studied in the present investigation assume importance in the plant metabolism. The increased peroxidase activity, sugars, decreased catalase activity, loss of cellular macromolecules like protein, ribonucleic acid and photosynthetic pigments observed in higher effluent concentrations appears that the metabolism in general is retarded due to effluent stress. Of all the macromolecule components of guar seedlings, RNA seemed to be the most susceptible to effluent stress. However, the stimulated seedling growth, due to the presence of growth promoting plant nutrients
in the lower effluent concentrations induces the metabolic rates to such an extent, that there was no metabolic arrests in these concentrations.