CHAPTER-III

RESULTS AND DISCUSSION

A) Plant Performance :

The forty days old plants of groundnut cultivar JL-24 subjected to different treatments are shown in Figs.2 and 3. It is evidènt from the figure that there is marked collapse of control (raised from untreated seeds) plants as a result of 10 days water stress. On the other hand plants raised from seeds pretreated with CCC and kinetin are showing more or less appearance even under similar regimes of soil water deficit. In this respect the pretreatment in organic solvent acetone was found to be more effective rather than pretreatment with aqueous solution. It was noticed that leaves of water stressed plants of control were considerably wilted in contrast to those of plants raised from pretreated seeds. These findings are consistent with the observations of several workers that the pretreatments impart capacity of drought resistance in plant species.

B) Inorganic Constituents :

1) Phosphorus :

Influence of presowing soaking treatments with CCC and kinetin on phosphorus nutrition in groundnut grown under two water regimes is recorded in Fig.4. It is evident from the figure that the phosphorus content in control plants (regularly watered plants) is increased due to presowing soaking treatments with CCC (in water and acetone) and kinetin (in water)

Fig.2

Influence of presowing soaking treatment with CCC on groundnut (Arachis hypogaea L.) plants under water stress.

Fig.3

Influence of presowing soaking treatment with kinetin on groundnut (Arachis hypogaea L.) plants under water stress.


CONTROL
CCC (W)
CCC (A)


CONTROL
KINETIN (W)
KINETIN (A)

in case of leaf tissue. The pretreatments are highly effective in increasing phosphorus content in stem tissue of control plants in each case. The pretreatments with kinetin (in water and acetone) and CCC (in water) are effective in causing increase in the phosphorus content in the root tissue. It is clear from the figure that in the plants raised from untreated seeds, the phosphorus level is lowered due to water stress in the leaf and stem tissue, whereas in the root tissue a slight increase in phosphorus content is noticed. This pattern of phosphorus distribution under water stress conditions is not altered due to presowing soaking treatment with kinetin (in water and acetone) and CCC (in acetone) in the leaf and stem tissue. There is a marked decrease in phosphorus content in the stem tissue of water stressed plants raised from seeds pretreated with kinetin and CCC. The phosphorus content is slightly increased in the leaf tissue and decreased in the stem and root tissue of stressed plants raised from seeds pretreated with CCC (in water).

Phosphorus is one of the most essential components of large number of metabolites which play a key role in various life processes. It is a constituent of DNA and RNA, which form the chemical basis of life. It is a part and parcel of ATP which is biological energy currency. It is also incorporated in the phospholipids which form the building blocks of membrane. As a component of nucleotides and coenzymes

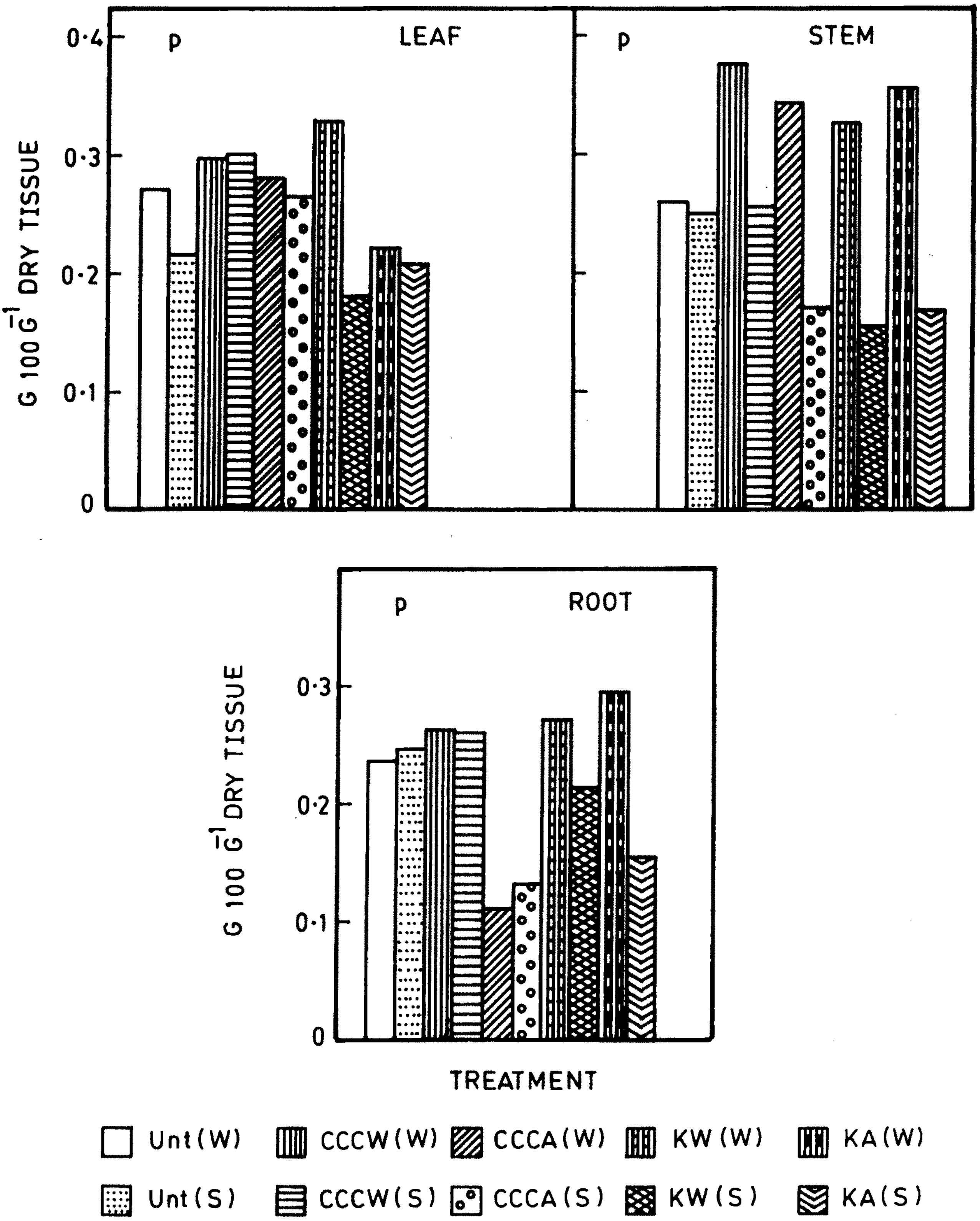

FIG. 4 EFFECT OF PRESOWING SOAKING TREATMENT WITH CCC AND KINETIN ON PHOSPHORUS CONTENT IN DIFFERENT PARTS OF GROUNDNUT UNDER WATER STRESS.

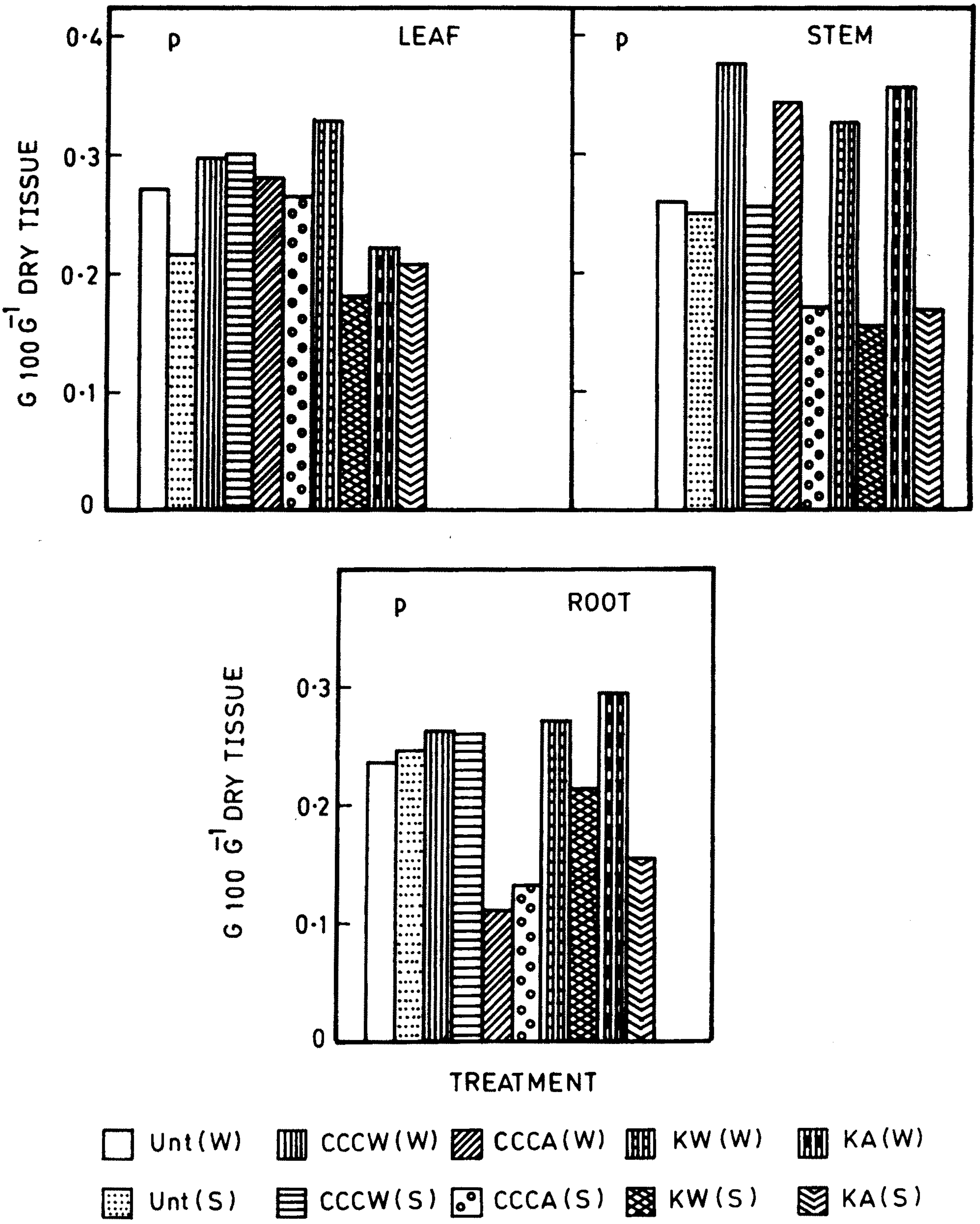

FIG. 4 EFFECT OF PRESOWING SOAKING TREATMENT WITH CCC AND KINETIN ON PHOSPHORUS CONTENT IN DIFFERENT PARTS OF GROUNDNUT UNDER WATER STRESS.

phosphorus plays very important role in various biochemical reactions. RuBP and PEP the two main CO_2 acceptors in plants are the compounds containing phosphorus. Phosphorus is also a constituent of various sugar phosphates such as fructose-1-6 diphosphate, phytic acid, the hexa phosphate ester of myo-inositol, or its calcium or magnesium salts (phytin), serves as a storage form of phosphate in seeds. Besides being component of such important metabolites, inorganic phosphate (Pi) plays a very important role in regulation of various pathways like photosynthesis and carbohydrate metabolism (Marschner, 1986). Phosphorus is absorbed in the main as the dihydrogen phosphate ion, $H_2PO_4^-$ from the soil solution.

Many workers have reported that water stress greatly hampers phosphorus uptake. Wilson *et al*. (1968) reported that phosphorus content in many legume forage crops was reduced due to water stress. Pande and Singh (1969) noticed that there was decrease in phosphorus uptake in rice under water stress conditions. Similar results were observed by Kongstrud (1969) in black currants and apple. Forde (1972) reported that drought reduced uptake of ^{32}p from roots of oil palm (*Elaeis quineensis* Jacq.). Eck *et al*. (1979) noticed reduction in phosphorus content in leaves of *Sorghum bicolor* under water stress. ✓Magdum (1984) reported that water stress caused adverse effect on phosphorus absorption in sunflower. Gujarathi (1984) however, noticed increase in phosphorus content of groundnut leaves under water stress conditions.

There are few attempts to understand the effect of growth regulators on phosphorus uptake. Darra and Saxena (1973) reported that there was increase in phosphorus uptake in maize, bajara and wheat due to pretreatment with IAA. Nimbalkar (1973) noticed that phosphorus content in sugarcane leaves was increased due to preplanting treatment with GA, IAA, kinetin and ascorbic acid. Chavan (1978) observed that pretreatment with IAA, ascorbic acid, kinetin and NaCl was effective in increasing phosphorus uptake in ragi plants. Naphade et al. (1986) reported that a seed pretreatment with a mixture of IAA and NAA was effective in increasing phosphorus uptake in sunflower plants.

Our observations indicate that in most of the cases the pretreatments with CCC and kinetin are effective in raising phosphorus status of the normally irrigated groundnut plants. However, under water stressed conditions these treatments are not found effective and the phosphorus nutrition is affected to almost similar degree in the untreated as well as pretreated plants due to water stress.

ii) Potassium :

The influence of presowing soaking treatments with CCC and kinetin on potassium status in groundnut plants under control conditions and under the conditions of drought is recorded in Fig.5. It is evident from the figure that in the

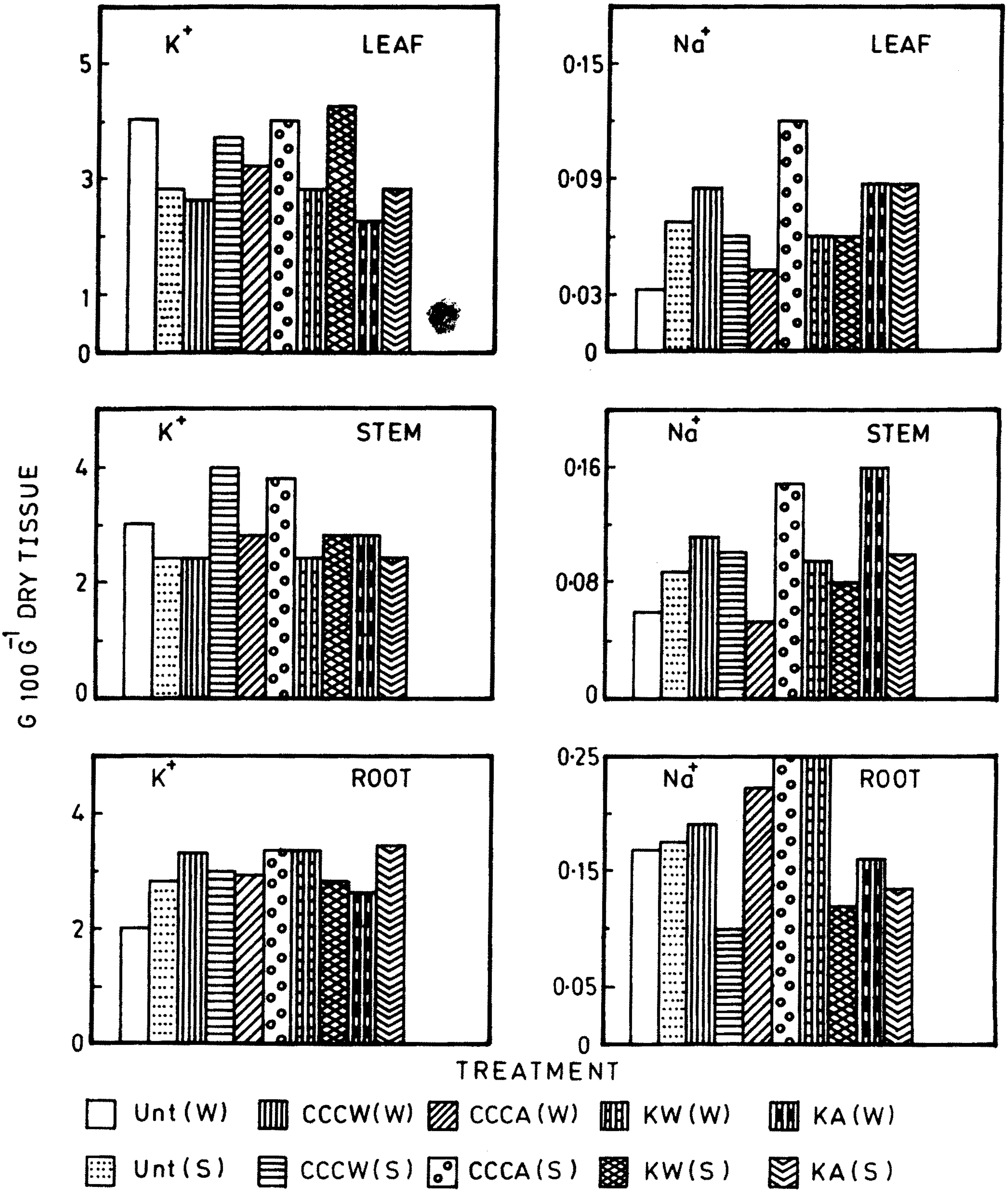

FIG. 5 EFFECT OF PRESOWING SOAKING TREATMENT WITH CCC AND KINETIN ON POTASSIUM AND SODIUM CONTENT IN DIFFERENT PARTS OF GROUNDNUT UNDER WATER STRESS.

control plants (raised from untreated seeds) the potassium content is in the following order Leaf > Stem > Root. This pattern of potassium distribution is changed due to pretreatments and more potassium accumulation in the root tissue is observed in case of CCC (in water) and kinetin (in water) pretreatments.

In the control plants the water stress causes a decline in potassium content in the leaf and stem tissue whereas there is slight increase in potassium status of the root of water stressed plants. It is evident from the figure that the presowing soaking treatments with both CCC and kinetin bring about increase in potassium content in the leaf tissue of the stressed plants in comparison to the corresponding control plants. This situation is also seen in case of stem tissue for all the pretreatments except kinetin pretreatment in acetone. Thus pretreatments are effective in elevating the potassium content in groundnut plants under the conditions of water stress.

Among various nutrients essential for the normal plant growth and metabolism, potassium is perhaps the most important one. An important role of potassium in activation of some key enzyme systems, carbohydrate metabolism, protein biosynthesis, assimilate translocation and stomatal movement is very well established. Evans and Sorger (1966) reported that there are more than 46 enzymes in living organisms (microorganisms, higher plants, animals) which are activated by the potassium. Recent studies reveal that potassium plays an important role in metabolic processes such as photosynthesis and respiration. Lauchli and Pfluger (1978) reported that potassium acts as counter ion for H^+ ion flux across the thylakoid membrane of chloroplast and mitochondrial membrane, thus taking part in the process of photosynthesis and respiration. Potassium is osmotically active and takes part in the turgor mediated plant processes such as stomatal movement and leaf movement (Mengel, 1977). It was observed that potassium deficiency caused decrease in the rate of translocation of photosynthates from the leaf to other plant parts in crops like sugarcane (Hartt, 1970). Though potassium plays an important role in plant metabolism we do not have clear idea about its mode of action, because potassium does not appear as a constituent of any plant metabolite and it is not incorporated in plant structures.

Potassium plays a vital role in growth and development of groundnut. Potassium deficiency symptoms appear in groundnut plants as yellowing of the margins of the older leaves. This is usually followed by interveinal chlorosis and finally by necrosis of the leaves, beginning at the margins and proceeding inwards untill the leaf fall off. Plants may be stunted and yields reduced before the visible deficiency symptoms occur. In potassium deficient plants reddish coloration of stem at the tips of branches are seen. Deficiency of potassium has been reported to reduce number of flower forming pegs as well as root growth. Potassium deficiency is evident when high proportion of pods produce only one kernel (Dwivedi, 1986). Basha and Rajeswara Rao (1980) studied the effect of phosphorus and potassium deficiency on CO_2 fixation and translocation in groundnut plants and reported that potassium deficiency directly influenced the translocation while phosphorus deficiency had an indirect effect on translocation by increasing the metabolic activity. Experiments conducted by Dwivedi (1986) revealed that application of potash in groundnut increased energy conservation in plant tissues as well as total energy harvest. The plant energy balancing factors such as transpiration, leaf temperature, stomatal resistance etc. were very much controlled due to potash. Pod yield and energy harvesting efficiency were also increased.

The uptake of potassium is highly selective and closely coupled to metabolic activity (Marschner, 1986). There are several reports which indicate that potassium nutrition is negatively influenced by soil water deficit. Richards and Wadleigh (1952) stated that water stress causes a definite decrease in potassium content in the plants. The experiments of Stewart and Hungate (1966) have demonstrated that potassium uptake is slightly reduced by water stress. A decrease in potassium content in the plants exposed to drought has been observed in number of experiments (Gilmore, 1971; Mengel and Boraunshweig, 1972; Singh and Prasad, 1979; Rao and Rammoorthy, 1981; Kawasaki et al., 1983; Ford, 1984). Brag (1972) noticed that the potassium status regulated the transpiration rates in wheat and pea plants. According to Mengel and Kirkby (1980) the mechanism of water uptake by roots and the ability of plants to exhaust soil water depends upon the potassium nutritional status of the plant. Low concentration of potassium results in the production of plants with high transpiration rate could be regulated by varied potassium concentrations. Transpiration is a process which is controlled by stomatal movements. Thus potassium might be helpful in controlling transpiration through its influence on stomatal behaviour. Reduction in the rate of transpiration is one of the criteria used to designate drought resistance in plants. Weimberg et al. (1982) proposed positive correlation between potassium content and proline accumulation in sorghum. According to Hubac et al. (1986) there exists positive correlation between the ability of a plant to resist water stress and the potassium content. Thus it is obvious that potassium status of leaf tissue is of great importance in evaluating drought resistance capacity.

The applications of growth regulators both as foliar spray and as presowing soaking treatments are reported to influence the potassium nutrition in many plant species. Nimbalkar (1973) from our laboratory noticed that the presowing soaking treatment with GA, IAA, AA and kinetin caused an increase in leaf potassium content. Chavan (1978) also noticed that the potassium content was increased in ragi leaves due to presowing soaking treatment of IAA, AA and kinetin. Naphade _et_ _al_. (1986) also reported a positive influence of seed pretreatment with ascorbic acid (AA) on potassium status in sunflower seeds and stalk. Shasidhar (1981) investigated influence of calcium chloride pretreatment on groundnut variety RS-218 and they noticed that the plants raised from pretreated seeds had higher degree of proline accumulation. According to them the better performance of this cultivar under stress conditions in fields following calcium chloride treatment is due to higher potassium content and proline accumulation. Our observations indicate that similar to calcium chloride pretreatment the presowing soaking treatment with CCC and kinetin are effective in maintaining optimum leaf potassium status in groundnut leaf and stem tissues under the conditions of water stress. The potassium content in all organs of groundnut plants raised from pretreated seeds with CCC (in acetone) in significantly increased. The increase in potassium content may have induced drought resistance in these plants.

iii) _Calcium_ :

The influence of presowing soaking treatment with CCC and kinetin on calcium status in groundnut plants under control

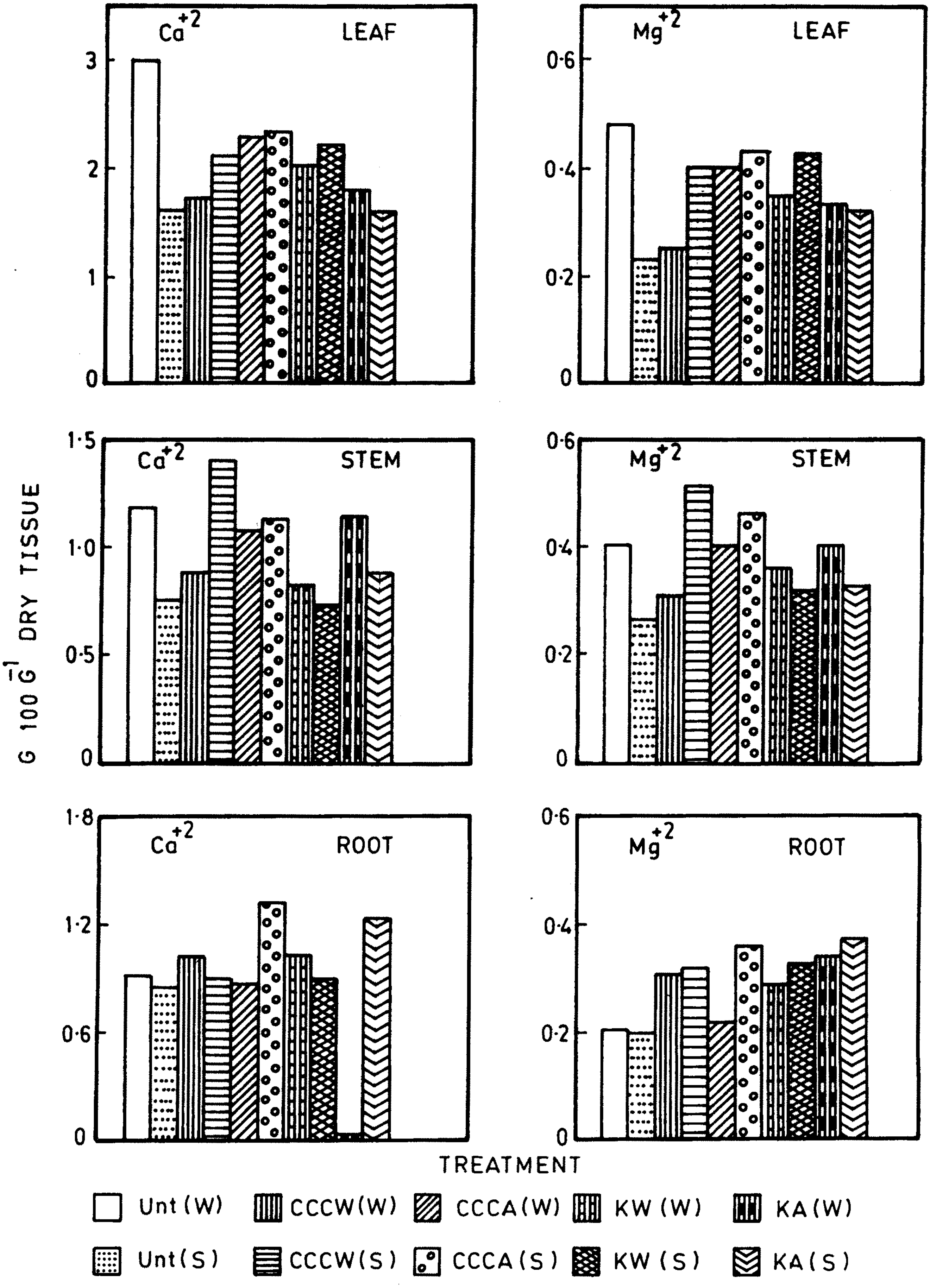

FIG. 6 EFFECT OF PRESOAING SOAKING TREATMENT WITH CCC AND KINETIN ON CALCIUM AND MAGNESIUM CONTENT IN DIFFERENT PARTS OF GROUNDNUT UNDER WATER STRESS.

conditions and under the conditions of drought is depicted in Fig.6. It is evident from the figure that in control plants (plants raised from untreated seeds) the calcium content is in the following order Leaf > Stem > Root. This pattern of calcium distribution in different plant organs is changed due to pretreatment. There is more accumulation of calcium in roots of plants raised from the seeds pretreated with CCC (in water) and kinetin (in water). However, there is decrease in calcium accumulation in the stem and leaf tissue of plants raised from the pretreated seeds.

In control plants (plants raised from untreated seeds) the water stress causes a decline in calcium content in all plant parts. The significant decrease in calcium content is observed in leaf and stem tissue of these plants. It is evident from the figure that the seed pre-treatment with CCC (in acetone and water) and kinetin (in water) brings about increase in calcium content of leaf tissue as compared to corresponding control plants. The stem tissues of plants raised from the seeds pretreated with CCC (in water and acetone) elevated content of calcium under water stress conditions. The stem tissue of plants raised from the seeds pretreated with CCC (in water) shows significant increase in calcium content. However, there is decrease in calcium content of stem tissue of plants raised from the seeds pretreated with kinetin (in acetone and water) and plants grown from untreated seeds under water stress conditions.

It can be seen from the figure that the water stress causes decline in calcium content of root tissue of plants raised from untreated seeds, pretreated seeds with CCC (in water) and kinetin (in water). However, the root tissue of plants raised from the seeds pretreated with CCC (in acetone) and kinetin (in acetone) show significant increase in Calcium content under water stress conditions. The maximum increase in calcium content is observed in root tissue of plants raised from the pretreated seeds with kinetin (in acetone) as compared to the corresponding control plants.

Calcium is a relatively large divalent cation with a hydrated ionic radius of 0.412 nm and a hydration energy of 1577 J/mol. In plants calcium exists in different forms. In plant tissues it may occur as free calcium, or in the form of calcium absorbed to indiffusible ions like carboxylic, phosphorylic and phenolic hydroxyl groups. The compounds of calcium usually occur as deposits in cell vacuoles. The seeds contain calcium mainly in the form of salt of phytic acid. Fifteen families of plants have been classified into three groups on the basis of calcium, magnesium and potassium contents by Horak and Kinzel (1971). These three groups are summarised as -

1. Oxalate : In these plants calcium is present as calcium oxalate.

2. Calciotrophic : These plants store large amount of calcium in soluble form.
3. Plants in which K/Ca ratio varies with soil samples.

There are many contravarsies regarding the functions of calcium in plant metabolism amongst the physiologists. However, the recent studies have helped much in defining the functions of calcium. The primary functions of calcium in plant metabolism are classified into three groups, viz. (i) Enzyme activation and stabilization (metabolism), (ii) Membrane formation activity and (iii) Cell wall structure.

Calcium plays an important role of enzyme activator in many enzyme systems. Davidson and Long (1958) observed that phospholipase activity in plants was at higher rates in presence of calcium. Dodds and Ellis (1966) reported that in plant cell wall an enzyme ATPase required Ca^{+2} or Mg^{+2} for its optimum activity. There are number of reports which suggest that calcium has inhibitory or stimulatory effects on many enzyme systems such as esterases, pectin esterases, lipoxygenases, nucleases, protein kinase, pyruvate kinase, polygalacturonic transaminase and glucose-6-phosphate dehydrogenase (Wyn Jones and Lunt, 1967; Clarkson and Hanson, 1980). Recently it is observed that calcium deficiency causes decrease in activities of enzymes nitrogenase and nitrate reductase (Gaudinova, 1983; Miller and Caude, 1983).

The recent concept of calmodulin is a new aspect of study of role of calcium in plant metabolism (Cheung, 1980; Cormier _et_ _al_., 1982). Calmodulin is a calcium binding protein which binds to calcium in such a way that calcium protein complex stimulates many enzymes and physiological process at very low concentrations. There are several reports which indicates that an important function of calcium is to stabilize membrane (Clarkson and Hanson, 1980).

Calcium is very important for groundnut specially for the development of gynophore ~~and~~ pod setting. Groundnut absorbs calcium through root system and also through peg and pod to meet the immediate needs of calcium. Under calcium deficient conditions there is no proper growth of pods and pods remain unfilled which are known as "pops". Calcium deficiency symptoms are significant when calcium concentration in tissue declines below 0.7%. Calcium deficiency is characterized by the development of localized pitted area on lower surface of leaves. Later on large necrotic spots are found on both the surfaces of leaflets which lead to bronz coloration. The youngest foliage presents a distorted appearance. Cracking of basal stem and die back of shoot at later stages of growth has also been reported. Pitted condition of leaves associated with the collapse of large hypodermal water storage cells. Poor root growth and marked reduction in flower and peg formation are noticed. Pal and Lalorya (1973) reported that there was accumulation of amino acids and amides in root and leaves of groundnut under low calcium levels.

Effect of water stress on calcium status in few plants has been investigated. Richards and Wadleigh (1952) reported that water stress caused variable effects on calcium content in plants. Takeshi (1966) reported that there was increase in calcium content of leaves of Brassica rapa and Vigna sinensis under water stress conditions. Kongstrud (1969) carried out experiments on black currants and apples to study the effect of water stress and reported that there was increase in calcium content in black currants but not in apples. Rahman et al. (1971) reported that water stress caused increase in Ca content in plant species such as Chloris cayana, Panicum turgidum, Panicum antiodotale, Panicum maximum, Panicum coloratum, Oryzopsis miliacea, Medicago sativa and Crotalaris aegyptica. Samuels (1972) observed increase in calcium content in sugarcane under water stress conditions. Ford (1984) studied 14 different species of leguminous plants. He noticed that out of 14 species only six species showed increase in calcium content in plant tissues under water stress conditions and two species showed decrease in calcium content and remaining six species showed no significant change. Gujarathi (1984) reported that under water stress two groundnut varieties JL-24 and SB-11 showed varietal difference in calcium contents in leaf tissue. He observed that calcium uptake in variety JL-24 was decreased due to 6 days water stress, however, due to 9 days water stress there was increase in calcium content. On the contrary, variety SB-11 showed opposite pattern of calcium accumulation to that of JL-24 variety. In variety SB-11 calcium content increased due to 6 days water stress and decreased due to 9 days water stress. Malathichari et al., (1986) reported that high calcium content of tissue decreased rate of water loss, maintained higher water status and induced membrane stability in groundnut and cowpea leaves under water stress conditions. Thus calcium accumulation under stress conditions may be an adaptive feature to resist drought conditions.

There are very few attempts to understand the influence of growth regulators on calcium uptake. Darra and Saxena (1973) reported that presowing soaking treatment of maize seeds with IAA increased calcium uptake. Nimbalkar (1973) noticed that calcium content of sugarcane leaves was increased due to pre-planting treatment with GA, IAA and ascorbic acid while it was reduced due to kinetin pretreatment. Our findings indicate that the pretreatments with CCC (in water and acetone) and kinetin (in water) are effective in increasing calcium status of leaf tissue under the conditions of drought. Calcium status of stem tissue is increased due to pretreatment with CCC and kinetin (in acetone). The calcium content of root tissue is increased under stress conditions in plants raised from seeds pretreated with CCC (in acetone) and kinetin. Thus there is elevation in calcium level due to presowing soaking treatments in groundnut. The elevation in calcium content in water stressed plants will be beneficial for preventing water stress induced membrane damage.

iv) <u>Magnesium</u> :

The influence of presowing soaking treatment with CCC and kinetin on magnesium status in groundnut plants under irrigated conditions and under the conditions of water stress is recorded in the Fig.6. It is evident from the figure that in irrigated control plants (plants raised from untreated seeds) the magnesium content is in the following order Leaf > Stem > Root. This pattern of magnesium distribution in different plant parts is changed due to pretreatment. There is more accumulation of magnesium in roots of plants raised from the seeds pretreated with CCC and kinetin. While there is decrease in magnesium content of stem and leaf tissue of plants grown from pretreated seeds with kinetin and CCC in acetone and water.

In control plants (plants raised from untreated seeds) the water stress causes a decline in magnesium content in all plant parts. The significant decrease in magnesium content is observed in leaf and stem tissues in this case. It is evident from the figure that the seed pretreatment with CCC (in water and acetone) and kinetin (in water) brings about increase in magnesium content of leaf tissue of the water stressed plants as compared to corresponding control plants. The stem tissue of plants raised from the seeds pretreated with CCC (in water and acetone) shows elevated magnesium content under drought conditions. There is significant increase in magnesium status of stem tissues of plants raised from the seeds pretreated with CCC (in water). A decrease is seen in magnesium content of stem tissue of control plants and plants raised from the seeds pretreated with kinetin (in acetone and water) under water stress conditions.

It is evident from the figure that the water stress causes very little decline in magnesium content of root tissue of plants raised from untreated seeds. At the same time, the plants raised from the seeds treated with CCC as well as kinetin show increases in magnesium content in root tissue under water stress conditions.

Magnesium occupies prominent place in plant mineral nutrition. It occurs in the form of a small and strongly electropositive cation in soil solution. Magnesium plays vital role in plants metabolism. In higher plants average value of magnesium ranges from 50 mg to 700 mg per 100 g dry tissue (Ferry and Wards, 1959). Cammarano et al. (1972) reported that magnesium functions as bridging element for joining the subunits of ribosomes. Under magnesium deficiency conditions or in presence of excessive levels of potassium, the subunits of ribosomes dissociate and protein synthesis ceases (Sperrazza and Spermalli, 1983). Mg^{2+} is required for the synthesis and assembly of solar energy harvesting molecules, chlorophylls. Magnesium is also required for photosynthesis. High concentrations of Mg^{2+} and K^+ are required in the chloroplasts and cytoplasm to maintain a high pH between 6.5 and 7.5 compared to much lower vacuolar pH of 5 to 6. The enzyme activity is regulated by pH which is maintained by Mg^{+2}, K^+ and upto certain extent by Ca^{+2} (Smith and Raven, 1979). Magnesium is also required for the RNA polymerase and hence for the formation of RNA in the nucleus. This role of magnesium might be related to both joining between individual DNA strains and neutralization of the acid proteins of the nuclear matrix (Wunderlich, 1978).

There are several enzyme systems which require magnesium as activator. Most of these enzymes are involved in transfer of phosphate (e.g. phosphatase, ATPase) and carboxyl groups (e.g. carboxylase). It is reported by Leonard and Hotchkiss (1976) that for maximal activity of membrane bound ATPase in corn roots there is requirement of Mg^{+2} and K^+. The activity of an enzyme RuBP carboxylase is highly dependent on both Mg^{+2} and pH. In presence of Mg^{+2} the activity of this enzyme increases (Sugiyama <u>et</u> <u>al</u>., 1968). Magnesium also increases an activity of enzyme glutamate synthetase (O'Neal and Joy, 1974).

In groundnut magnesium deficiency is directly related with tikka disease. Mg^{+2} deficiency symptoms in groundnut have been described by Harris (1949), Bledose and Harris (1950) and Reid and York (1958). The first symptom of magnesium deficiency is interveinal chlorosis of the terminal leaves and stunting of the plant. In severe cases, plants completely lose their green colour and die. Magnesium deficiency occurs in groundnut when tissue concentration drops below 0.3%. The groundnut plants require more magnesium after 30 days of plant growth.

There are few attempts to study the effect of drought on magnesium nutrition. Jenne <u>et</u> <u>al</u>. (1958) reported that there was increase in magnesium content of non-irrigated maize plants. Rahman <u>et</u> <u>al</u>. (1971) noticed that there was increase in magnesium content in leaves of eight species of forage crops under water stress conditions. Kawasaki <u>et</u> <u>al</u>. (1983) observed increase in magnesium uptake in sorghum and maize under water stress conditions. On the other hand decrease in magnesium content was evident in bean plants under water stress conditions. These observations indicate that plant species differ with respect of Mg^{+2} nutrition under drought conditions. Gujarathi (1984) reported that water stress causes rise in magnesium level in groundnut leaves.

There are very few attempts to understand the influence of growth regulators on magnesium uptake. Nimbalkar (1973) noticed that magnesium content of sugarcane leaves was decreased due to preplanting treatment with GA, IAA, kinetin and ascorbic acid. In contrast to sugarcane, in case of groundnut we can notice marked changes in magnesium content in the different plant parts in response to water stress and pretreatments. In general there is elevation of leaf magnesium content as compared to control (untreated) plants due to pretreatment with CCC and kinetin under water stress conditions. There is elevation in magnesium content of stem tissue in plants raised from the seed pretreated with CCC and kinetin. Similarly there is elevation in magnesium content of root tissue of plants grown from seeds pretreated with both CCC and kinetin as compared to control (untreated) plants under water stress conditions. Maintenance of optimum level of Mg^{2+} in leaf tissue under the conditions of water deficit is of great importance in view of the key role played by this element in various metabolic reactions. It is also important for maintaining the stability of chlorophylls in general and photosynthetic apparatus in particular. According to Pitman (1981) the plants of arid zone have adapted to low water potential by acquiring ability to make use of magnesium as osmotica or to store sugars in the vacuoles and other organic solutes in the cytoplasm. Thus accumulation of magnesium in leaf, stem, root tissue may be regarded as adaptive feature acquired by groundnut plants through pretreatment to overcome drought conditions.

v) Manganese :

Figure 7 records the influence of presowing soaking treatment with CCC and kinetin on manganese contents in different plant parts of irrigated and water stressed groundnut plants. It is evident from the figure that manganese content is maximum in the leaf tissue and minimum in the stem tissue of control (untreated) plants. This pattern of manganese distribution in different plant parts is changed due to some of the pretreatments. The plants grown from seeds treated with CCC (in water) show maximum manganese content in root tissue and minimum in stem tissue. The plants raised from the seeds pretreated with kinetin (in water) show maximum manganese content in roots which is followed by leaf and stem. It is evident from the figure that presowing soaking treatment with kinetin (in acetone) causes elevation in manganese content of root tissue which is followed by stem and leaf tissue.

It can be seen from the figure that the water stress causes an increase in manganese content of root and stem tissue of control plants (raised from untreated seeds). At the same time a decrease in manganese content is seen in leaf tissue in control plants under water stress conditions. The pretreatment with CCC (in acetone and water) has caused elevation in manganese content of leaf, stem and root tissue of groundnut plants under water stress conditions. There is significant increase in manganese content in leaf tissue of water

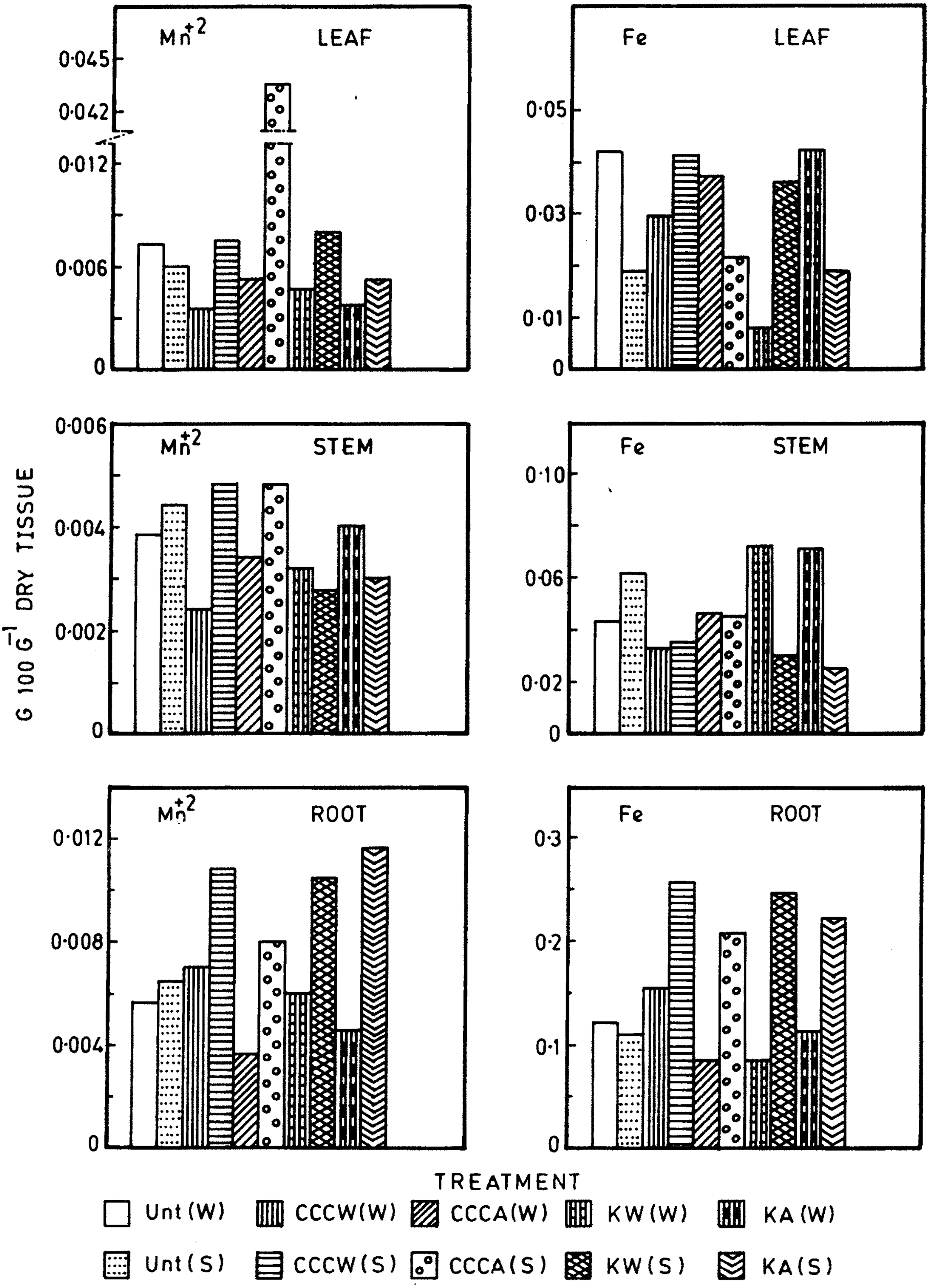

FIG. 7 EFFECT OF PRESOWING SOAKING TREATMENT WITH CCC AND KINETIN ON MANGANESE AND IRON CONTENT IN DIFFERENT PARTS OF GROUNDNUT UNDER WATER STRESS.

stressed plants raised from the seeds pretreated with CCC (in acetone). Kinetin pretreatment causes increase in manganese content in root and leaf tissue of groundnut plants subjected to water stress, however, under this treatment decrease in manganese content in stem tissue is noticeable.

Manganese is one of the essential micronutrients. It is absorbed mainly as Mn^{2+} and is translocated predominantly as the free divalent cation in the xylem from the roots to the shoot (Graham, 1979). Manganese acts as an activator of many enzymes. So far only one mangano protein, manganin, has been isolated from seeds of groundnut by Dickert and Rozacky (1969). It is now established that manganese is required in both lower and higher plants for the Hill reaction (Cheniae and Martin, 1968). Arginase is specifically activated by manganese. Manganese is prominent as an activator of enzymes mediating of the reactions of the Krebs cycle. Manganese activates RNA polymerase (Ness and Woolhouse, 1980). Manganese is a structural constituent of ribosomes (Lyttleton, 1960). Manganese has the lowest complex stability constants and thus it forms weakest bonds therefore, it can replace magnesium in many reactions such as phosphokinases and phosphotransferases (Clarkson and Hanson, 1980). Sevilla <u>et</u> <u>al</u>. (1980) isolated an enzyme superoxide distmutase with one atom of manganese from leaf extracts of pea. Superoxide dismutases (SOD) are present in all aerobic organisms and play an essential role in survival of these organisms in the presence of oxygen (Halliwell, 1978). SOD protect tissue from the destructive effects of the oxygen free radical O_2^- (super oxide). Manganese is required for the activities of decarboxylating enzymes of C_4 pathway. The enzyme NADP-malic enzyme, PEP carboxylase and NAD-malic enzyme are manganese specific enzymes. Different crop species show variation in their manganes requirement. Stout (1961) reported that 0.005% of manganes is adequate for the growth and metabolism of multicellular plants. Reid and Cox (1973) reported that for groundnut plants the adequate manganese concentration of leaf tissue ranges between 20 and 40 ppm. The availability of all the micronutrients except molybdenum tends to decrease with increasing pH. Under very acid conditions manganese is highly available and toxicity may occur whereas at high pH the availability is often decreased to the point of deficiency. Martin (1959) has noted manganese toxicity in groundnuts grown on very acid soils. Leveque and Beley (1959) studied the effect of high levels of manganese in sand culture experiments. They found that accumulation of manganese in leaf margin, delayed flowering and maturing and impaired fruit development. Under toxic conditions the tissue concentrations of N, P and Mg increased while that of K and Ca decreased. Mn deficiency leads to interveinal chlorosis and necrotic spots. However, veins tend to remain green, Bussler (1960) termed the deficiency as 'Marsh Spots'. Young leaves are mostly affected by Mn deficiency.

There are very few attempts to study the fate of micronutrients like manganese under the conditions of water stress. Sakanoue and Iguchi (1968 a,b,c) reported that water stress increased the absorption of manganese in rice. In contrast to these observation Pande and Singh (1969) noticed that there was reduction in manganese content of rice leaves under water stress conditions. Gujarathi (1984) reported that there was increase in manganese content of groundnut leaves under water stress.

Almost no attention has been paid to the influence of growth regulators on mangenese nutrition in plants. Chavan (1978) observed reduction in manganese content of ragi leaves due to pretreatment with IAA, AA, kinetin and NaCl. It is observed in the present investigation that the presowing soaking treatments with CCC and kinetin have positive effect on manganese level in groundnut plants. Increase in manganese content in leaf and root tissue of groundnut due to pretreatments under the conditions of water stress may help the plants in maintaining normal metabolic activity.

vi) Iron :

The effect of presowing soaking treatment with CCC and kinetin on iron status in groundnut plants under irrigated conditions and under the conditions of water stress is recorded in Fig.7. It is evident from the figure that in control plants

(plants raised from untreated seeds) the iron content is maximum in root tissue. In stem tissue iron content is slightly more than that of the leaf tissue. The iron content in different plant parts in control plants is in the order root > stem > leaf. This pattern of iron distribution is not changed due to seed pretreatment. If the iron content in different parts of watered plants in each treatment is compared, it can be seen that in the leaf and root tissue the iron content is reduced due to presowing soaking treatments with kinetin and CCC.

In control plants (plants raised from untreated seeds), water stress causes increase in iron content of stem tissue and decrease in iron content of leaf and root tissue. The pretreatment with CCC (in water) and kinetin (in water) bring about elevation in iron content of water stressed leaves as compared to those of corresponding control plants. There is decrease in iron content of stem tissue of plants raised from the seeds pretreated with CCC (in acetone) and kinetin (in water and acetone) as compared to corresponding control under drought conditions. The water stress causes decrease in iron content of root tissue of plants grown from untreated seeds. However, all the four seed pretreatments cause elevation in iron content of root tissue under water stress conditions. The maximum accumulation of iron takes place in root tissue of plants grown from seeds pretreated with CCC (in water) under water stress conditions which is followed by kinetin (in water), kinetin (in acetone) and CCC (in acetone) pretreatments. It is evident from the figure that all the four pretreatments cause relatively more pronounced accumulation of iron in root tissue than stem and leaf tissues.

The most important function of iron in plant metabolism is that it forms prosthetic group of many enzyme systems. The most intensively studied iron containing prosthetic groups are iron porphyrins (hemes). Such well known heme proteins are the cytochromes which contain a heme-iron porphyrin complex as prosthetic group. Cytochromes are the constituents of redox systems in mitochondria and chloroplast which participate in electron transport in cyclic and non-cyclic reactions of photophosphorylation as well as in respiratory chain. The iron sulphur proteins also play important role in respiratory electron transport. In addition to these enzymes, catalase and peroxidase are also heme enzymes. The leghaemoglobin an iron containing protein participates in the mechanism of nitrogen fixation in root nodules of leguminous plants. Though iron plays important role in plant metabolism, the heme pigments constitute only about 0.1% of the total iron in plant leaves (Dekock _et al._, 1960). The remaining iron is stored in the form of ferric phosphoprotein known as phytoferritin. It was reported by Hyde _et al._ (1963) that reserve stock of phytoferritin in leaf tissue might be used for photosynthetic needs by developing plastids. In green leaves about 80% of the iron is localized in the chloroplast regardless of the iron nutritional status (Marschner, 1986).

It was noticed in few experiments that water stress negatively influences the rate of iron uptake in plants (Basiouny and Briggs, 1971; Rahman et al., 1971). Ivanov and Karkash (1965) observed increase in soluble iron content in root tissue under drought conditions.

There are only few attempts to study the effect of growth regulators on iron uptake. Darra and Saxena (1973) observed that IAA pretreatment reduced iron content in maize leaves. Similar observations were made by Chavan (1978) in leaves of ragi plants raised from seeds pretreated with 50 ppm solutions of GA, IAA and AA.

In groundnut we can notice marked changes in iron content in the different plant parts in response to water stress and pretreatments. In general there is elevation of root iron content as compared to control (untreated) plants due to pretreatment with both kinetin and CCC. The accumulation of iron in root tissue may be due to decrease in rate of iron translocation under water stress conditions. As iron is an essential element, decrease in iron content in leaves of groundnut under water stress may lead to metabolic disorders. This situation is seen in the control plants of groundnut (raised from untreated seeds).and plants raised from seeds pretreated with both CCC and kinetin (in acetone). However, the presowing soaking treatment with aqueous solutions of both these growth regulators cause increase in iron content in the leaf tissue corresponding to respective control plants. Although iron increase in roots may not be of any metabolic advantage to the plants, the elevation of this element in leaf tissue will be of great help under stress conditions.

vii) Sodium :

Figure 5 records the influence of presowing soaking treatment with CCC and kinetin on sodium contents in different plant parts of irrigated and water stressed groundnut plants. It is evident from the figure that sodium content is maximum in the roots and minimum in the leaf tissue of control (untreated) plants. This pattern of sodium distribution is more or less similar in case of pretreatment with CCC (in water and acetone) and pretreatment with kinetin (in water). It is evident from the figure that presowing soaking treatments cause increase in sodium content in the leaf tissue. The sodium content shows an increase in leaf, stem and roots of control (untreated) groundnut plants exposed to water stress. The presowing soaking treatment with CCC in water and kinetin also cause elevation of sodium level in the stem tissue. The sodium content in the roots of watered plants is increased due to CCC pretreatment and kinetin (in water) treatment.

It can be seen from the figure that the water stress causes an increase in sodium content in the leaf, stem and root of control (untreated) groundnut plants. This pattern is also shown by plants raised from seeds pretreated with CCC (in acetone). However, the pretreatment of seeds with CCC (in water) causes a decrease in sodium content in all plant parts. The kinetin pretreatment does not bring about any marked change in sodium content in the water stressed leaf tissue as compared to corresponding control. In the stem and roots of water stressed plants there is decrease in sodium content due to kinetin pretreatment.

Sodium is considered as one of the non-essential mineral nutrients by several plant physiologists because they are unable to find any important role of sodium in plant metabolism. It is argued by Rains(1972) that sodium helps in maintaining favourable water balance in the plants. This argument is based on the observations that the treatment with sodium chloride many times increases leaf succulence. There are also few observations which indicate that the salt treated plants become more drought resistant (Karadge and Chavan, 1980). Sodium has been regarded as an essential micronutrient in case of C_4 species (Brownell and Crossland, 1972). However, in C_3 species like groundnut the role of sodium is not very well established. Sodium has been reported to act as an activator for some enzyme systems (Evans and Sorger, 1966). Sodium attains a real prominence in saline soils where it is a dominant cation. Sodium chloride and sodium sulphate salinity have been found to cause several metabolic disorders in groundnut cultivars TMV-10 and SB-11 (Karadge and Chavan, 1980).

There are few attempts to study the effect of drought on sodium nutrition. In the leaves of *Brassica rapa* and *Vigna sinensis* drought was found to have very little influence on sodium content (Takeshi, 1966). Rahman *et al.* (1971) reported that water stress caused increase in sodium content in plant species such as *Chloris cayana*, *Panicum turgidum*, *Panicum coloratum*, *Oryzopsis miliaceae*, *Medicago sativa* and *Crotalaris aegyptica*. Ford (1984) studied the accumulation of low molecular weight solutes in water stressed tropical legumes such as *Cicer arietinum* CV Tyson, *Cajanus cajana* ICP 7035, *Lablab purpureus* CV Highworth, *Vigna radiata* CV Berken, *V.mungo* CV Regur, *V.unguiculata* CPI 28215, *Glycine max* CPI 26671 and pasture species like *Stylosanthes hamata* CV Verano, *S.scalora* CV Seca. *Psoralea eriantha*, *Rhycosis minima*, *Macroptilium atropurpureum* CV Siratra, *Glycine tomentella* and *Centrospermuma* sp. aff. pubescens CV Belalto. He noticed that in most of the legumes leaf sodium status is very low. While there was slight increase in sodium content of leaf tissues in *Stylosanthes scabra* CV seca and *Centrosperma* spp. aff. pubscens CV Belalto under water stress conditions. By taking into consideration the above observations he concluded that sodium is of little osmotic importance in legume plants under drought conditions. Our observations also indicate that due to water stress there is slight increase in sodium content, particularly in the leaf tissue. However, this increase is not sufficient to bring about any marked change in ionic balance in water stressed plants. Gujarathi (1984) also noticed slight increase in sodium content of groundnut leaves under water stress.

There are very few attempts to understand the influence of growth regulators on sodium uptake. Darra and Saxena (1973) reported that presowing soaking treatment of maize seeds with IAA reduced sodium uptake. Nimbalkar (1973) observed that the leaf sodium content of sugarcane was slightly reduced due to IAA pretreatment and slightly elevated due to ascorbic acid pretreatment. He observed no change in sodium content under the conditions of kinetin and GA pretreatment. In contrast to sugarcane, in case of groundnut we can notice marked changes in sodium content in the different plant parts in response to seed pretreatments. In general there is elevation of leaf sodium content as compared to the control (untreated) plants due to pretreatment with both kinetin and CCC. It is suggested by Lawlor and Milford (1973) that sodium can increase the drought resistance of water stressed sugarbeet by altering the leaf water balance. Similar situation may prevail to some extent in groundnut plants raised seeds, which have undergone presowing soaking treatment with CCC and kinetin.

C) Organic Constituents :

i) Moisture percentage :

The influence of presowing soaking treatment with CCC and kinetin in acetone and water on leaf moisture content in groundnut leaves under water stressed conditions is recorded in Table 1. The leaves of groundnut plants grown from untreated seeds were almost collapsed (Fig.1), dry and rolled and their moisture percentage was declined to 9%. However, the pretreatment with CCC in water brings about retention of moisture percentage upto 51.62% in groundnut leaves under water stress condition. Seed pretreatments with CCC (in water) and kinetin (in acetone and water) were also able to maintain the leaf tissue water content higher than that of untreated plants under drought conditions. Drought causes water deficit in most of the plant parts. Gujarathi (1984) observed decrease in leaf moisture content of groundnut leaves under drought conditions. He also observed that this reduction in moisture percentage differed with variety. In variety JL-24 there was more reduction in moisture percentage than variety SB-11 under drought conditions.

Levitt (1956) reported that water deficits cause dehydration of protoplasm which causes loss in turgor. According to May and Milthorpe (1962) there is decrease in relative turgidity below 90%. Levitt (1972) reported that a severe dehydration leads to pronounced decrease in respiration

Table [illegible] : Influence of presowing soaking seed treatment with CCC and kinetin on Stomatal behaviour, moisture percentage and relative water content in groundnut (<u>Arachis</u> <u>hypogaea</u> L.) under water stress.

Sr. No.	Treatment	Surface	Relative humidity %	Light intensity Quantum	Diffusive resistance for H_2O Sec Cm^{-1}	Transpiration μg Cm^{-2} Sec^{-1}	Flow rate Cm^3 Sec^{-1}	Diffusive conductance for CO_2 Sec Cm^{-1}	Diffusive resistance for CO_2 Sec Cm^{-1}	Moisture Percentage %	Relative water content
1	Untreated (coat level)	L	26.533	394.75	7.08	3.15	0.489	0.4388	2.2785	79.033	63.9252
		U	27.066	387.5	7.59	2.74	0.418				
2	Untreated (stressed)	L	44	340	NR	NR	NR	NR	NR	9.00	28.06
		U	44	340	NR	NR	NR				
3	CCCW(Stressed)	L	47.8	689	19.6	0.83	0.068	0.1443	6.9268	26.69	44.47
		U	47.6	614.5	25.8	0.64	0.050				
4	CCCA (Stressed)	L	42.06	486.66	7.71	2.58	0.2386	0.2947	3.3928	51.62	54.98
		U	45.46	509.66	18.64	1.74	0.1603				
5	KW(Stressed)	L	45.6	390	NR	NR	NR	NR	NR	17.60	26.92
		U	45.6	490	NR	NR	NR				
6	KA(Stressed)	L	47.6	470	53.43	1.1866	0.093	0.03906	25.5959	21.25	35.16
		U	47.2	496	179.2	0.114	0.00933				

NR : Not Recordable

Values are mean of three determinations.

rate. According to Iljin (1923) water deficit in leaves causes partial or complete closure of stomata. Such a stomatal closure adversely affect the movement of CO_2 in assimilatory leaves which adversely affect the rate of photosynthesis. A reduction in water content in leaf tissue also causes reduction in the rate of translocation. Bayles _et_ _al_. (1937) reported that there exist correlations between drought hardiness and water retention of leaves and other plants parts under conditions of drought.

There are few attempts to understand influence of growth regulators on moisture percentage of leaves. According to Chinoy _et_ _al_. (1984), the greatest advantage of seed pretreatment was in the ability of the pretreated seeds to extract sufficient moisture from the soil when its moisture content was too low for quick germination and finally due to accelerated growth, higher yields were obtained. These workers observed that seed pretreatment with AA resulted in more efficient water absorbing capacity in number of crops such as wheat, oat, barley, sesamum, ragi and maize. Giri _et_ _al_. (1987) reported that foliar application of CCC was effective in reducing moisture loss in groundnut leaves in semiarid rainfed conditions. They also reported that priming treatment of groundnut seeds with CCC was effective in increasing water potential in leaf tissue. Moisture retention under water stress conditions by plant tissues is one of criteria used to designate drought resistance capacity of that plant.

In the present investigation, it is noticed that the moisture content is retained by plants grown from seeds pretreated with CCC in acetone to maximum extent. The remaining pretreatments are also effective in maintaining leaf moisture content upto some extent. Our findings in this respect support the observations recorded by Giri _et al_. (1987), and indicate that the presowing soaking treatments with CCC and kinetin may be causing more efficient water absorption under drought conditions and more effective retention of the absorbed water.

ii) Relative Water Content (R.W.C.) :

Influence of presowing soaking treatment with CCC and kinetin on the relative water content (R.W.C.) in the leaf tissue of groundnut plants exposed to drought conditions is recorded in Table 1. It can be seen from the table that R.W.C. in the leaves is considerably reduced due to water stress in the control plants raised from untreated seeds. The pretreatment with CCC in acetone and water is effective in raising the R.W.C. in the plants subjected to water stress as compared to control plants. There is not much difference in the relative water content of the leaves of water stressed plants raised from untreated seeds and seeds pretreated with kinetin.

Relative water content is the water content of a tissue expressed as a percent of the water content of the fully turgid tissue. The relative water content is a function, dependent on the physical and chemical properties of the tissue of the components of the chemical potential of the water. Singh and Prasad (1980) observed decrease in relative water content of groundnut leaves under water stress condition. Mukherjee and Choudhari (1983) noticed decrease in relative water content of *Vigna* leaf tissue under water stress conditions. Kuhad and Sheoran (1986) reported that water deficit caused decline in relative water content of cluster bean (*Cyamopsis* *tetragonoloba* L.) leaves. Similar observations were also recorded by Ravindra *et* *al*. (1988) in case of groundnut leaves. There are few attempts to understand the influence of growth regulators on relative water content of plant leaf tissue. Gill and Singh (1978) reported that application of CCC as foliar sprays was effective in increasing relative water content of wheat leaf tissue under water stress conditions. Giri *et* *al*. (1987) noticed that foliar application and seed pretreatment of CCC was effective in increasing relative water content in groundnut leaves.

Relative water content has been recognized as one of the most reliable criteria for assessing the water status of the tissue and maintenance of higher relative water content is no doubt helpful in maintaining optimum water balance in the leaf tissue. This is greately achieved by CCC pretreatment. Our findings in this respect support the observations

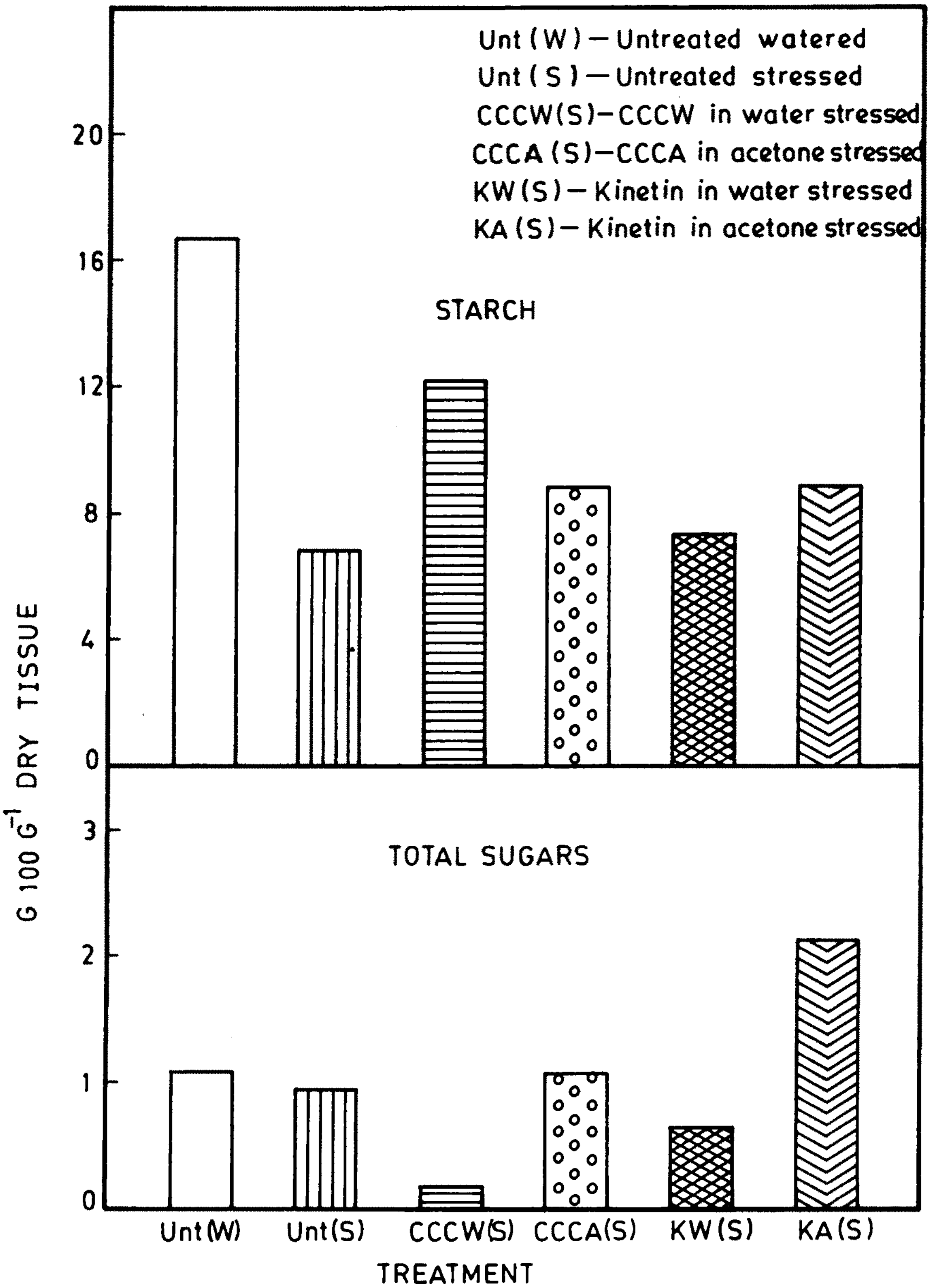

FIG. 8 EFFECT OF PRESOWING SOAKING TREATMENT WITH CCC AND KINETIN ON STARCH AND TOTAL SUGARS CONTENT IN LEAVES OF GROUNDNUT UNDER WATER STRESS.

recorded by Giri et al. (1987). It is further interesting to note here that the growth retardant is highly effective in this respect while the growth promoter kinetin is totally ineffective.

iii) Carbohydrates :

The influence of presowing soaking treatment on the level of carbohydrate fractions (starch and total sugars) in the leaves of groundnut under water stress conditions is recorded in figure 8. It is evident from the figure that water stress causes a marked decline in leaf starch content in the control plants which are raised from untreated seeds. In case of the pretreated plants also the leaf starch content is less compared to regularly watered plants. However, if the starch level in the water stressed plants in different treatments is compared it can be seen that as compared to control plants (raised from untreated seeds), the starch level in the pretreated plants is relatively high; the difference being particularly in the plant leaves raised from seeds soaked with aqueous solution of CCC.

The total sugar content is reduced due to water stress in the leaves of control plants raised from untreated seeds. Similar pattern is also seen in water stressed plants raised from seeds pretreated with aqueous solutions of CCC and kinetin. The pretreatment with kinetin in acetone is effective in raising the level of soluble sugars in the leaf tissue. If the total carbohydrate content in the leaf tissue in the water stressed plants in different treatments is taken into consideration, it can be seen that the total carbohydrate level is increased due to presowing soaking treatment in water stressed groundnut plants.

Carbohydrates represent one of the most important categories of organic compounds as they form a link between photosynthesis and respiration. The starch and sucrose are the major end products of photosynthetic carbon metabolism while glucose is a substrate of respiration. The four carbon sugar erythrose is a precursor of aromatic pathway. The pentose sugar ribose is a component of nucleic acid. The complex polysaccharide cellulose plays a structural role. Besides being involved so intensively in the metabolism, sugars also play an osmoregulatory role because of their osmotically active nature.

There are several reports on metabolism of carbohydrates under water stress conditions. As early as 1948, Eaton and Ergle observed conversion of starch into sugars under water stress conditions in two cotton varieties. Fukutotku and Yamada (1982) reported that there was accumulation of carbohydrates in water stressed soybean. They also noticed increase in soluble sugars (like sucrose) under water stress conditions.

Sharma and Singh (1985) studied the effect of water stress on carbohydrate metabolism in groundnut variety M-37. They observed marked decrease in non-reducing sugars under water deficit conditions. Vu and Yelenosky (1989) reported that water deficit caused increase in reducing sugars and decrease in leaf starch content of Valencia orange. They also reported that this decrease may be due to inhibition of photosynthetic carbon dioxide assimilation and reduction in RuBP case activity due to water stress. It is evident from our observations that there is slight lowering of total sugars (Reducing sugars and non-reducing sugars) due to water stress in leaves of groundnut cultivar JL-24. Slatyer (1969) suggested that the carbohydrate metabolism is affected by water deficit through direct and indirect effects on photosynthesis. Lee _et al_. (1974) observed decrease in reducing sugars, sucrose and starch accumulation in drought tolerant as well as drought susceptible varieties of pea under water stress conditions. Chang and Ryan (1987) noticed decrease in leaf starch content of cotton plants under drought conditions. They also reported that starch decrease may be due to increase in α - amylase activity and inhibition of an enzyme sucrose synthetase which caused accumulation of sucrose and orthophosphate in the chloroplasts. They suggested that orthophosphate inhibits ADP-glucose pyrophosphorylase activity which is responsible for starch synthesis in the chloroplast. In the present investigation also a decrease in starch content in groundnut leaves under stress conditions is seen and this situation prevails also in the plants raised from seeds pretreated with kinetin and CCC.

Vora et al. (1974) opined that accumulation of sugars in Sorghum vulgare under water stress conditions may play protective role. According to Ackerson (1981) increase in concentration of leaf carbohydrates help in osmotic adjustment during water stress in cotton plants. Jones et al. (1981) reported that sugars play an important role in the mechanism of drought resistance. They suggested that sugars may be accumulated in cells of expanded leaves in vacuoles and thus maintain turgour or the sugars may be translocated to the root to maintain the growth of new roots. These new roots may be helpful in absorbing more water from the soil. According to Stewart (1978) accumulated carbohydrates may contribute carbon for synthesis of amino acids like proline.

There are several attempts to understand the influence of growth regulators on carbohydrate metabolism. Agakishiev and Nikitina (1972) reported that foliar application of CCC was effective in increasing carbohydrate level. Asmaeva and Avundzhyan (1973) noticed increase in carbohydrate content in wheat when seeds were pretreated with CCC. Chinoy et al., (1968) observed no increase in the sugar content of groundnut plants when treated with CCC (2000 ppm). Bhandari and Sen

(1975) noticed that CCC treatment caused decrease in sugar content of citrus seedlings. Litvinova and Yuldashev (1971) reported that CCC was effective in causing decrease in total carbohydrate content in stem and leaves of cotton variety G.460. Bhattacharjee and Gupta (1981) observed reduction in insoluble carbohydrate content in sunflower plants. On the other hand Bhadre (1983) reported that CCC pretreatment was effective in increasing carbohydrate production in leaves of cotton variety Laxmi and H-4. In this respect they observed more increase in sugar content than starch content. Upretty *et al*. (1985) observed that CCC treatment caused increase in total sugars, non-reducing sugars and starch content in *Avena sativa* CV Kent.

Our observations indicate that the level of sugars in groundnut leaves is not elevated due to CCC pretreatment as well as aqueous pretreatment of kinetin. Thus the pretreatments are not effective in enhancing the role of sugars in osmotic adjustment in water stressed leaves. At the same time the starch content in the water stressed leaves is raised (as compared to water stressed plants raised from untreated seeds) due to pretreatment both with CCC and kinetin. Thus it is probable that the starch degradation which prevails in leaves during drought is slightly lowered due to seed pretreatment.

Table 2 : Influence of presowing soaking treatment with CCC and kinetin on RNA, -SH content and Nitrate reductase activity in groundnut (*Arachis hypogaea* L.) leaves under water stress.

Sr.No.	Treatment	RNA $g\ 100^{-1} g$ dry tissue	- SH Co M	Nitrate Reductase activity $\mu g\ NO_2$ lib $g^{-1} h^{-1}$
1	Unt(W)	0.06837	2.2884×10^{-4} M	163.3864
2	Unt(S)	0.09673	1.0411×10^{-4} M	77.8485
3	CCCW(S)	0.1602	1.3652×10^{-4} M	118.6122
4	CCCA(S)	0.2407	2.7197×10^{-4} M	119.1241
5	KW(S)	0.06483	1.7492×10^{-4} M	95.9331
6	KA(S)	0.02030	3.3750×10^{-4} M	110.2137

Values are mean of three determinations.

iv) Ribose nucleic acid (RNA) :

Influence of presowing soaking treatment with CCC and kinetin on RNA content of groundnut plants is recorded in Table No.2. It is evident from the table that due to water stress the RNA content is slightly elevated in control plants of groundnut. The presowing soaking treatment with CCC (in acetone and water) and kinetin (in acetone) bring about marked increase in RNA content in the leaves of water stressed plant.

RNA, ribose nucleic acid, is a polymer of nucleslides. The nucleslides are made up of one of the nitrogen bases (adenine, cytosine, uracil, guanine), a pentose sugar ribose and a phosphate group. There are different types of RNA molecules such as m-RNA, t-RNA, r-RNA which are necessary for the process of protein synthesis. Usually RNA is a single stranded molecule but RNA molecules like t-RNA show much folded structure like clover leaf or hair pin.

Since RNA represents an important macromolecule associated with protein biosynthesis, the changes in total RNA pool are many times investigated to assess the metabolic state of the tissue. A decline in RNA content under the conditions of leaf senescence is very well documented. Such a decline would undoubtedly lead to an inhibition of protein synthesis. In view of Gates (1964) protein synthesis is interrupted in stressed plants. As early as 1940 Sisakian reported reduced

\- protein synthesizing activity as dessication occured in drought resistant plants while drought sensitive species showed no net synthesis. Thus it is clear that if the level of RNA which is one of the major components of protein synthesizing machinery is properly maintained in stressed plants then there is some possibility of avoiding the damage to protein synthesizing machinery. Kessler and Frank-Tishel (1962) also opined that increase in RNA content of leaf tissue during dehydration might induce drought resistance. Accumulation of nucleic acids, particularly RNA has been marked as an index for physiological resistance against environmental stress (Singh and Gill, 1972).

There are few reports which indicate that the presowing soaking treatments can increase the RNA level in plants. Thus Chinoy _et_ _al_. (1984) reported that presowing soaking treatment with ascorbic acid caused elevation of RNA content in many crop species. Gill and Singh (1978) reported that CCC treated wheat plants maintained higher levels of nucleic acids and proteins under water stress conditions suggesting that degradative processes are more in favour of untreated plants than that of CCC pretreated plants under water stress conditions. Amer _et_ _al_. (1988) observed increase in RNA content in maize under water stress conditions when seeds were pretreated with CCC. They also observed that there was not such increase under irrigated conditions.

In our experiment CCC and kinetin (in acetone) pretreatments are effective in increasing RNA content in leaf tissue of groundnut leaves under water stress conditions. Thus in groundnut the presowing soaking treatment with CCC (in both acetone and water) is highly effective in maintaining higher level of RNA which may be of great significance in stabilizing protein synthetic machinery under drought conditions. The analysis of various RNA fractions will probably throw more light on this aspect.

v) Free proline :

Influence of presowing soaking treatment with CCC and kinetin (in acetone and water) on the level of free proline in different plant parts is recorded in the Fig.9 and 10. It is evident from the figure that in the watered plants the free proline level in the different plant parts is very low. But when the plants are subjected to water stress the proline content gets elevated in all parts. It is interesting to note here that the initial level of proline in different parts of groundnut plants raised from pretreated seeds is relatively low as compared to initial proline level in groundnut plants raised from untreated seeds. However, once the water stress is set in the accumulation of proline is more rapid in the pretreated plants. In this respect the pretreatment of groundnut seeds with kinetin is highly effective. The presowing

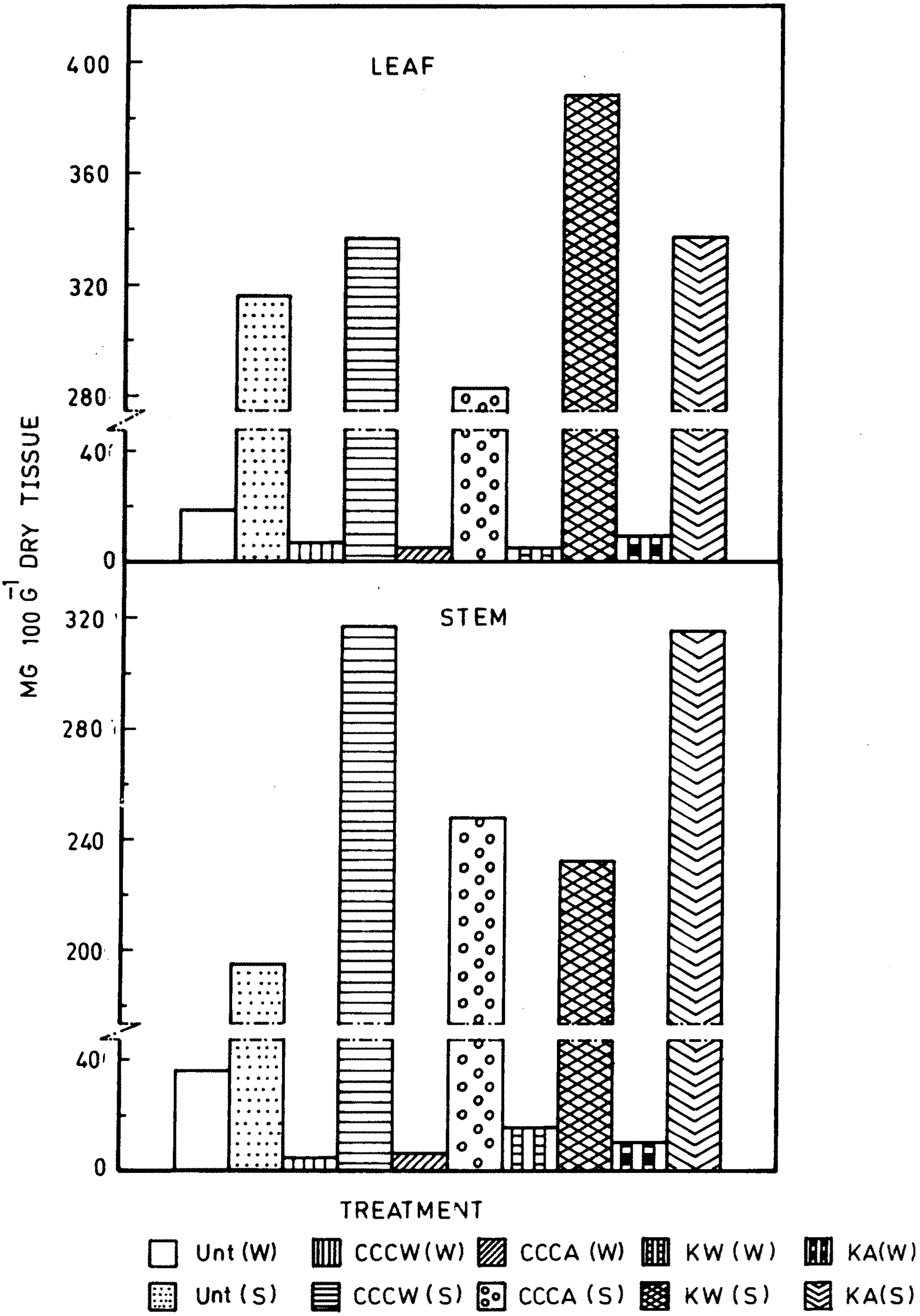

FIG. 9 EFFECT OF PRESOWING SOAKING TREATMENT WITH CCC AND KINETIN ON PROLINE CONTENT IN DIFFERENT PARTS OF GROUNDNUT UNDER WATER STRESS.

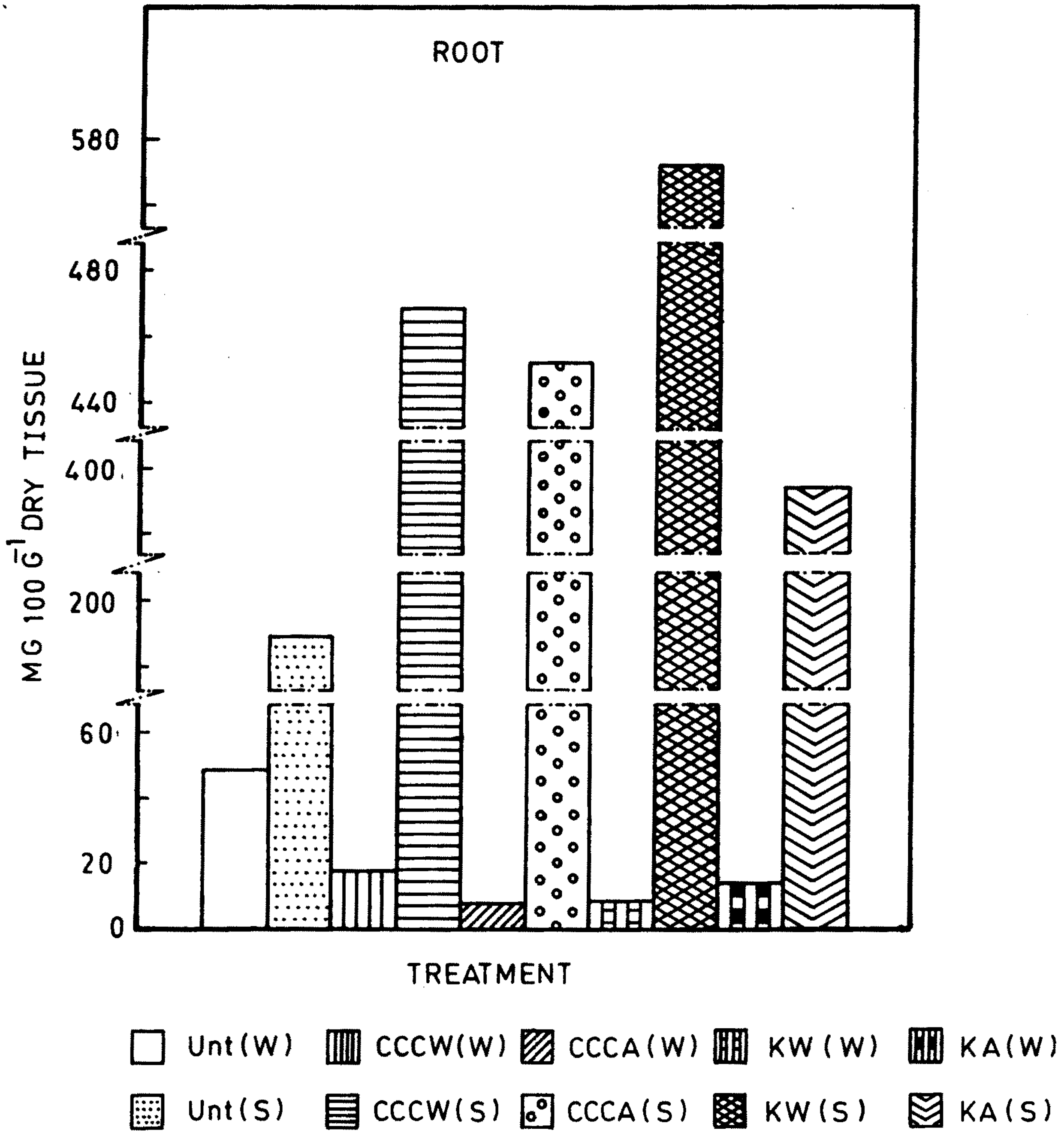

FIG. 10 EFFECT OF PRESOWING SOAKING TREATMENT WITH CCC AND KINETIN ON PROLINE CONTENT OF GROUNDNUT ROOT UNDER WATER STRESS.

soaking treatment promote marked accumulation of free proline in root tissue of groundnut.

Proline a cyclic amino acid belonging to glutamate family, is found to play several important roles in the drought resistance processes. Paleg and Aspinall (1981) are of opinion that proline accumulated under water stress conditions, acts as compatible solute regulating and reducing water loss from the cell. According to Itai <u>et</u> <u>al</u>. (1988) elevated levels of proline due to stress may play an important role in regulation of stomatal movement. Proline also serves as a source of carbon and nitrogen for biosynthesis of variety of compounds during a post stress recovery. Sinha and Rajgopal (1975) reported that when proline was fed to barley and wheat plant stressed with PEG, the loss of nitrate reductase activity was reduced. Schobert and Tachesche (1978) observed that proline plays an important role in protecting bovine albumin from denaturation by ammonium sulfate or ethanol. There are several reports of extensive accumulation of free proline in various plant species in response to environmental stress (Palfi <u>et</u> <u>al</u>., 1973; Singh <u>et</u> <u>al</u>., 1973). Mukherjee <u>et</u> <u>al</u>. (1982) reported different trends in proline accumulation in plants of different ecological group under water stress conditions. They found that there was more proline accumulation in <u>Vigna</u> (mesophyte) which is followed by <u>Ipomea</u> (Semiaquatic plant), Trapa (a and [illegible] (1987) in Phaseolus aconitifolius Jacq

floating species) and negligible in Vallisneria (a submerged species). Sivaram Krishnan et al. (1988) reported that there was extensive proline accumulation in leaves of sorghum plants under drought conditions. Ganesan et al. (1988) observed increase in proline content of cotton leaves under water stress conditions. Although there are several reports which indicate that leaves are the sites of proline accumulation in some plants like cotton accumulation of proline in stem is evident (Janagoudar et al., 1983). The accumulation of proline under stress conditions has been attributed to protein breakdown, lesser incorporation of proline in the protein and stimulation of biosynthesis of proline from the precursors glutamate and arginine.

There are few reports regarding the influence of growth regulators on tendency of plants to accumulate proline. Tretyakov and Gomer (1985) reported that CCC application to alfalfa plants was effective in causing increase in an amino acid proline in root neck zone. Amer et al. (1988) observed increase in proline content of leaves of maize plants raised from seeds pretreated with CCC under water deficit conditions. Thus accumulation of free proline is clearly an adaptive response of the plants towards drought. Our observations demonstrate that the presowing soaking treatment of groundnut seeds with CCC and kinetin increase the intensity of this response. The extensive accumulation of free proline in the roots of water stressed plants raised from the pretreated seeds may be of great help in restoring plant metabolism during post stress period.

vi) Titratable acidity status (TAN) :

The influence of presowing soaking treatment with CCC and kinetin on titratable acidity status in leaves of groundnut subjected to water stress is depicted in Fig.11. It is evident from the figure that water stress causes increase in acid status of leaves of groundmut plants raised from untreated seeds, seeds pretreated with CCC (in acetone) and kinetin (in acetone). However, there is slight decrease in acid status of leaves of plants raised from the seeds pretreated with aqueous solutions of CCC and kinetin.

The organic acids represent an important class of metabolites in all living cells. The tricarboxylic acid cycle (TCA cycle) which is considered as metabolic pathway of prime importance in both photosynthetic and non-photosynthetic tissues represents a major source of majority of organic acids found in higher plants. These include malate, citrate, cisaconitate, isocitrate, fumarate, and succinate. Besides above organic acids, some other organic acids like tartarate, oxalate, glycolate are heavily synthesized and extensively

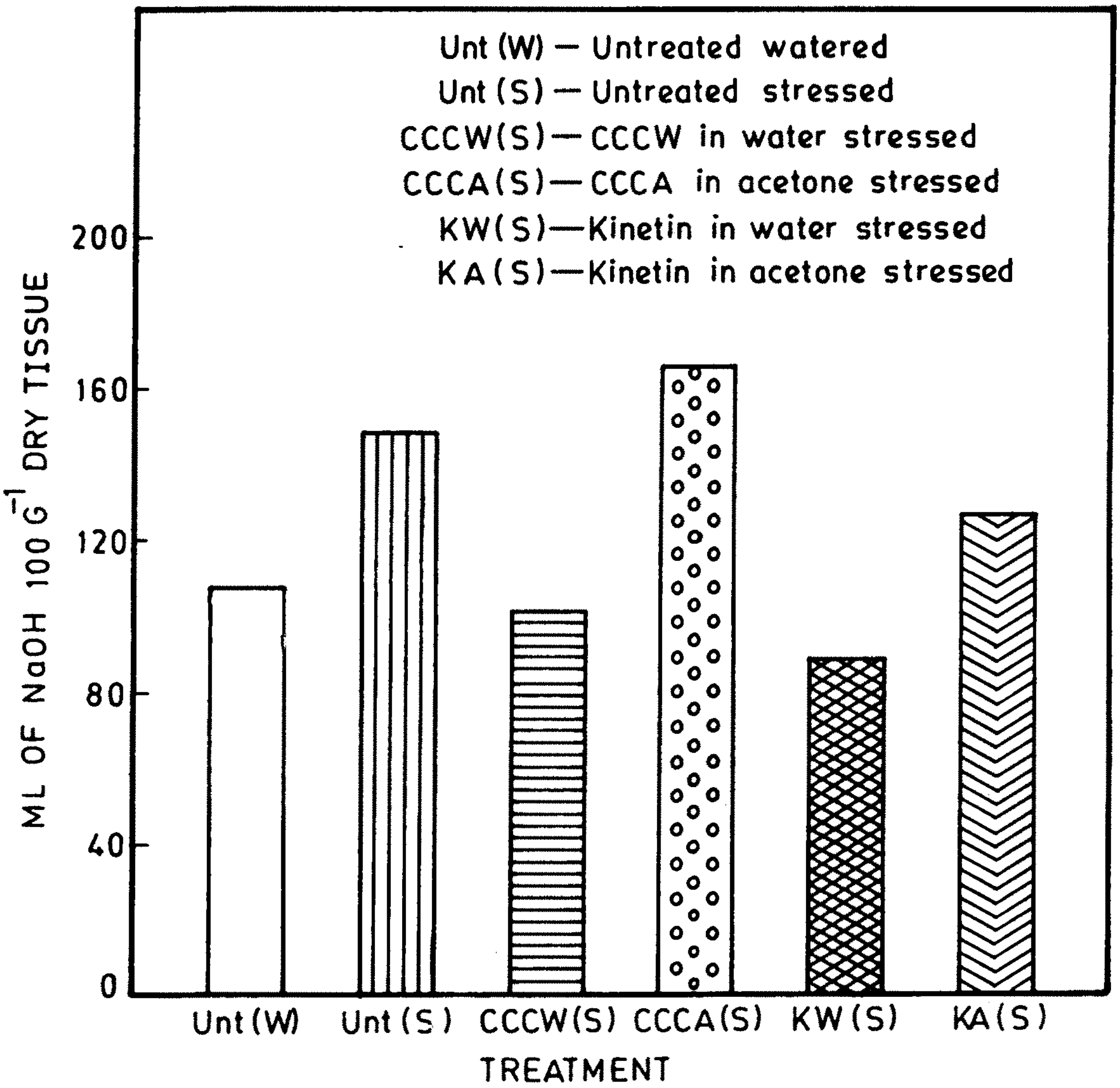

FIG. 11 EFFECT OF PRESOWING SOAKING TREATMENT WITH CCC AND KINETIN ON TITRATABLE ACID NUMBER (TAN) IN LEAVES OF GROUNDNUT UNDER WATER STRESS.

accumulated in some plant tissues. Malic acid is also synthesized during photosynthesis in C_4 species whereas glycolic acid is an important intermediate of photorespiration. The organic acids provide carbon skeletons for biosynthesis of variety of metabolites. They also play important role in maintenance of ionic balance in plant cell. The organic acid like malic acid plays an important role in stomatal movements. Jones _et al_. (1979) has indicated that organic acids play an important role in osmotic adjustment during stress conditions.

Increase in acidity status of leaf tissue under drought indicate stimulation of organic acid synthesis due to water stress. Lawlor and Fock (1977) observed increase in content of succinate, fumarate, and aconitate in water stressed sunflower leaves. Singh and Prasad (1980) noticed increase in organic acid contents (malic acid, malonic acid, succinic acid and fumaric acid) of groundnut leaves under water stress conditions. Timpa _et al_. (1986) observed increase in organic acid content of cotton leaves under drought conditions. They observed more accumulation of citric acid in cotton leaves under water stress conditions. Chavan (1978) reported that water stress caused increase in titratable acidity status of ragi leaves. Nalwade (1983) observed increase in titratable acidity status of niger leaves under water stress conditions.

There are only few attempts to understand influence of growth regulators on organic acid content of plant tissue. Gill and Singh (1978) reported that spraying of CCC (500 ppm) on wheat caused increase in acidity status of wheat leaves under drought conditions.

It is evident from the present investigations that there is no marked influence of presowing soaking treatments on the level of organic acids in the water stressed plants. In the control plants the water stress has caused elevation of titratable acidity status and this trend is also seen in case of pretreatments with CCC and kinetin (in acetone). However, the relative increase in case of CCC treatment is very small and in case of kinetin pretreatment there is slight decrease in titratable acidity status values as compared to water stressed control plants. It is interesting to note that the presowing soaking treatments with aqueous solution of both CCC and kinetin have caused lowering of organic acid level as compared to irrigated plants. The organic acid increase is many times correlated with increase in hydrolytic activities and increase in respiration rate. In this content the decrease in titratable acidity status due to pretreatments with aqueous solutions of kinetin and CCC appears to be of importance.

vii) Ascorbic acid content :

Influence of presowing soaking treatment on ascorbic acid content in groundnut leaves under drought conditions is recorded in Fig.12. It is evident from the figure that ascorbic acid content is increased in leaf tissue of groundnut raised from untreated seeds, seeds pretreated with CCC (in acetone) and kinetin (in acetone) in response to water stress. However, there is decrease in ascorbic acid content due to pretreatment with CCC (in water) and kinetin (in water) under drought conditions.

Excellent work of Chinoy and coworkers (1969) has highlighted the key role of ascorbic acid (Vitamin C) as a plant growth regulator in number of physiological processes such as germination, flowering and stress tolerance. Ascorbic acid stimulates amylase, protease and RNAase activities and RNA content in various plants (Chinoy *et al*., 1969; Chinoy and Saxena, 1972). Ascorbic acid plays an important role in oxidative phosphorylation (Abraham *et al*., 1968). Arnon *et al*. (1954, 1956, 1957) reported that ascorbic acid plays an important role of electron transporting agent during photosynthetic phosphorylation in illuminated chloroplast or as a factor stabilizing the activity of chloroplast. Joshi and Hegde (1978) reported that ascorbic acid stimulated chlorophyll synthesis and rate of carbon utilization in rice and

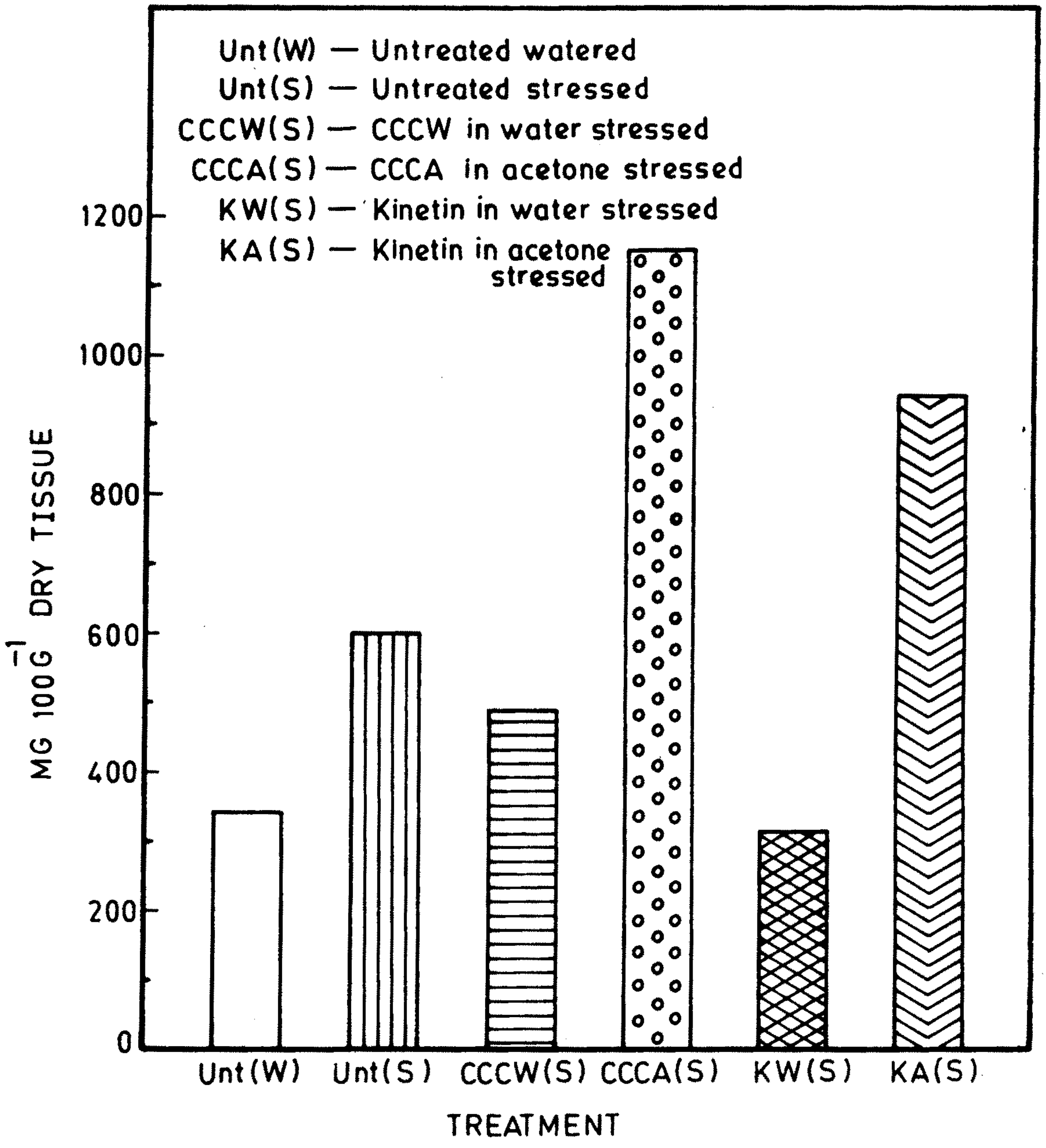

FIG.12 EFFECT OF PRESOWING SOAKING TREATMENT WITH CCC AND KINETIN ON ASCORBIC ACID CONTENT IN LEAVES OF GROUNDNUT UNDER WATER STRESS.

sugarcane. They also observed that ascorbic acid induced resistance to salt stress.

There are number of attempts made by various workers which show that water stress causes increase in ascorbic acid content in plant tissue. As early as 1944 Eisele observed increase in ascorbic acid content of oat seedlings under moderate water stress. Hunter <u>et</u> <u>al</u>. (1950) reported increase in ascorbic acid content of upper leaves of <u>Brassica</u> <u>rapa</u> under water stress conditions. Farkas and Rajhathy (1955) noticed increase in ascorbic acid content in tomato plants under drought conditions. Increase in ascorbic acid content in plant tissues during period of extreme drought was evident in many other studies (; Ivanov and Karakash, 1965; Luckicheva, 1968). Garg and Singh (1971) noticed more ascorbic acid and ascorbinogen content in leaves of drought resistant rice variety Taichung as compared to variety IR 8 under water stress. Vora <u>et</u> <u>al</u>. (1975) reported that water stress caused increase in ascorbic acid, ascorbigen and net ascorbic acid content in sesamum leaves.

There are few reports describing influence of seed pretreatment on ascorbic acid content in plants. Startseva (1963) reported that presowing soaking treatment of seeds with micro-elements increased ascorbic acid content in plant tissues under the conditions of drought. Kamynina (1965) noticed increase in ascorbic acid content of pea plants at the times of blossoming when seeds were pretreated with aqueous solutions of salts of microelements such as Mn, Cu, Co, Mo and Vn. Vyas et al. (1965) reported that presowing soaking seed treatment with maleic hydrazide or sulphanilamide caused increase in ascorbic acid content in groundnut leaves. Dogra and Sinha (1983) reported that applications of 50 ppm and 100 ppm cycocel as foliar sprays were more effective in increasing ascorbic acid content in Phyllanthus urinaria than higher concentrations (250 and 500 ppm).

According to Chinoy et al. (1984) rapid turnover of ascorbic acid helps in maintaining normal metabolic activity in the growing plants under water stress conditions. On the basis of the extensive data on ascorbic acid turnover, AA-FR-peroxidase activity, AA-MM complexing and free radical content as well as taking into consideration enhanced growth, earlier development and increased yield of crop after restoration of the normal water regime, Chinoy et al. (1984) postulated that a period of a short duration during the early stage of plant growth stimulated the redox environment of the cell towards the reductive side thus protecting enzymic activity of the cell. The accumulation and binding of ascorbic acid and ascorbigen to macromolecules protects them against oxidation (Vora et al., 1975). Further, the decrease in the free water content of the cell due to wilting brought about the molecules of ascorbic acid and macromolecules like DNA and RNA closer to each other thus facilitating the formation of a charge transfer complex (Chinoy, 1969;). Enhancement in the activity of AA-FR peroxidase during the period of revival produced the free radical of ascorbic acid (Chinoy, 1969,). On restoring the normal water regime, both these processes enhanced electron energy which was utilized for faster synthesis of cell constituents leading to rapid growth, earlier development and better yield of plants which were wilted during the juvenile phase for a short duration and subsequently returned to normal water regime.

Our findings revealed that there is more accumulation of ascorbic acid in groundnut leaves raised from untreated seeds, seeds pretreated with CCC and kinetin (in acetone) under drought conditions. The significant increase is observed in leaves of plants raised from seeds pretreated with CCC (in acetone). Thus accumulation of ascorbic acid in leaf tissue may be an adaptive feature to overcome drought conditions as indicated by Chinoy and co-workers.

D) Enzymes and SH content :

i) Nitrate reductase activity :

The influence of presowing soaking treatment with CCC and kinetin on nitrate reductase activity of groundnut leaf tissue in Table 2. It is evident from the table that in irrigated control plants there is highest activity of an enzyme nitrate reductase. In case of control (untreated) plants and pretreated plants, the leaf tissue shows decrease in activity of nitrate reductase under water stress conditions. However, the activity of nitrate reductase is more in pretreated plants as compared to untreated (control) plants under water stress conditions.

Nitrate is the major source of nitrogen available to most plant species. Enzyme nitrate reductase reduces nitrate to nitrite with the help of NADH in cytoplasm. The activity of NR is stimulated by CO_2, light and nitrate. The half life of NR is short. Its synthesis is stimulated by light. Hageman _et_ _al_. (1961) reported that light intensity and nitrate availability affect the activity of nitrate reductase in corn. They reported that under low light intensity there was accumulation of nitrate. It is now very well established that nitrate reductase activity is a rate limiting step of nitrogen assimilation in most of the higher plants where nitrate is the major source of nitrogen available. A positive correlation between leaf NR activity and growth rate has been noticed by some workers (Reilley, 1976; Austin _et_ _al_., 1978). In legume plants like groundnut also NR is found to play an important role in nitrogen metabolism (Chavan, 1987). A decline in nitrate reductase activity under drought conditions is evident in number of experiments. Huffaker _et al_. (1970) noticed reduction in the activity of NR in barley under water stress conditions. Becker and Fock (1986) noticed reduction in NR activity of maize leaves under drought conditions. Rajgopal _et al_. (1977) observed the responses of NR, water content and nitrate content to water stress. They noticed change in leaf nitrate content following the changes in relative water content. They reported that R.W.C. and N.R. activity was lowest in water stressed plants. Goyal _et al_. (1985) reported that under drought conditions the NR activity was reduced. Reddy and Veeranjaneyulu (1988) noticed decline in NR activity in 7 day old horsegram seedlings when stress was induced by PEG-6000. Deshmukh and Srivastava (1987) reported that there was marked reduction in NR activity during water stress conditions in sunflower. Shaner and Boyer (1976 a,b) opined that nitrate flux regulated the activity of NR. Under water stress conditions the reduction in nitrate flux may cause reduction in NR activity. Sinha and Nicholas (1981) reported that the enzyme may be regulated through (i) the availability of nitrate, (ii) an inhibition of protein synthesis consequent upon the reduction in polyribosome level during stress and the inherent high turnover rate of the enzyme, or (iii) a reduction in the availability of NADH through effects on photosynthesis or respiration.

There are few attempts to understand the influence of growth regulators on NR activity. Dev (1970) showed that presowing soaking treatment with CCC solution led to increase in the activity of NR in the leaves, at the three leaf stage and decrease of the same in the young cotton roots. Bhadre (1983) observed increase in NR activity in leaves of cotton varieties Varalaxmi and NCT-19 when seeds were pretreated with CCC. In our findings the NR activity has been increased due to pretreatments with CCC and kinetin under water stress conditions in groundnut leaves as compared to water stressed control plants. The significant increase in NR activity is observed in plants raised from seeds pretreated with CCC (in acetone and water). Sinha and Rajgopal (1975) observed reduction in loss of NR activity of PEG stressed wheat and barley plants when they were fed with proline. The more accumulation of proline in pretreated groundnut plants (Fig. 9 and 10) may be related with increased NR activity. The increase in NR activity under stress conditions may help in providing nitrogen pool to metabolic reactions which lead to the synthesis of various amino acids and proteins.

ii) Sulfhydryl groups :

The influence of presowing soaking treatment with CCC and kinetin on SH content of groundnut leaves under stress conditions is depicted in Table 2 . It is evident from the table that the water stress causes decrease of SH content in groundnut leaves of plants raised from untreated seeds under stress conditions. However, the pretreatments with CCC and kinetin are effective in causing increase in SH content of groundnut leaves under water stress conditions. The marked increase is observed in leaves of plants pretreated with kinetin in acetone.

The -SH group occurs in plant protoplasm mainly as a component of proteins, an essential amino acid (cysteine or CSH) and a peptide (glutathione or GSH). Levitt (1962) proposed SH hypothesis or sulfhydryl - disulfide (SH $\rightleftharpoons$ SS) hypothesis of freezing injury and resistance. The injury was assumed to be from the following series of three steps.

i) Proteins molecules approach each other as a protoplasm is progressively freeze-dehydrated.

ii) When they are sufficiently close, chemical combination occurs between S atoms of adjacent protein molecules, a) by oxidation of 2 SH groups or b) by SH $\rightleftharpoons$ SS interchange as follows :

$$2\ RSH + \frac{1}{2} O_2 \rightleftharpoons RSSR + H_2O.$$

iii) The protein molecules undergo a conformational change due to this aggregation leading to injury or death.

The conformational change occurs due to freeze dehydration. Freezing tolerance would be due to prevention of intermolecular SS bonding.

Since drought injury is due to cell dehydration it may be explained on the basis of freezing injury. It is believed that the drought injury may be due to the aggregation of proteins due to intermolecular bonding of the dehydrated and therefore, more closely packed protein molecules. Kaloyereas (1958) obtained correlation between SH content and drought resistance. It is probable that mechanical stress subjects the proteins to unfolding or denaturation. Gaff (1966) suggested that in membrane proteins there arise SS bonding, when cell undergoes dehydration causing drought injury. This type of protein aggregation may be induced under water stress conditions (Gunning et al., 1968). Thus the formation of disulfide bond between two adjacent cysteine residues in polypeptide chains is of fundamental importance for the tertiary structure and thus the function of enzyme proteins. This bond may form a permanent (covalent) cross-like link between polypeptide chains or a reversible dipeptide bridge. During dehydration, the number of disulfide bonds in proteins increase at the expense of -SH groups, and this shift is associated with protein aggregation and denaturation (Tomati and Galli, 1979). The protection of -SH groups in proteins from the formation of disulfide bridges is considered to of great importance for providing cellular resistance to dehydration caused by drought and heat (Levitt, 1980). Thus SH accumulation is one of the criteria to assess drought resistance. Tombesi (1951, 1952) found that there was decrease in glutathione content under drought conditions in plants of Pelargonium zonale. Chinoy et al. (1969) have noticed an increase in -SH contents due to ascorbic acid pretreatment in many crop species. In our experiments, pretreatment with CCC and kinetin are found to be effective in increasing SH content in leaf tissue which may be an adaptive feature to overcome drought damage.

E) Photosynthetic pigments :

Influence of presowing soaking treatment with CCC and kinetin on leaf chlorophyll content is shown in Fig.13. It is evident from the Figure that presowing soaking treatment with CCC and kinetin (in acetone) has caused increase in the levels of both chlorophylls a and b in the leaves of water stressed plants. Among various treatments pretreatment with CCC in acetone is highly effective in bringing about this effect. The level of accessory pigments carotenes in the leaves of water stressed plants of groundnut raised from different treatments is also shown in the figure. It is evident from the figure that there is no marked difference

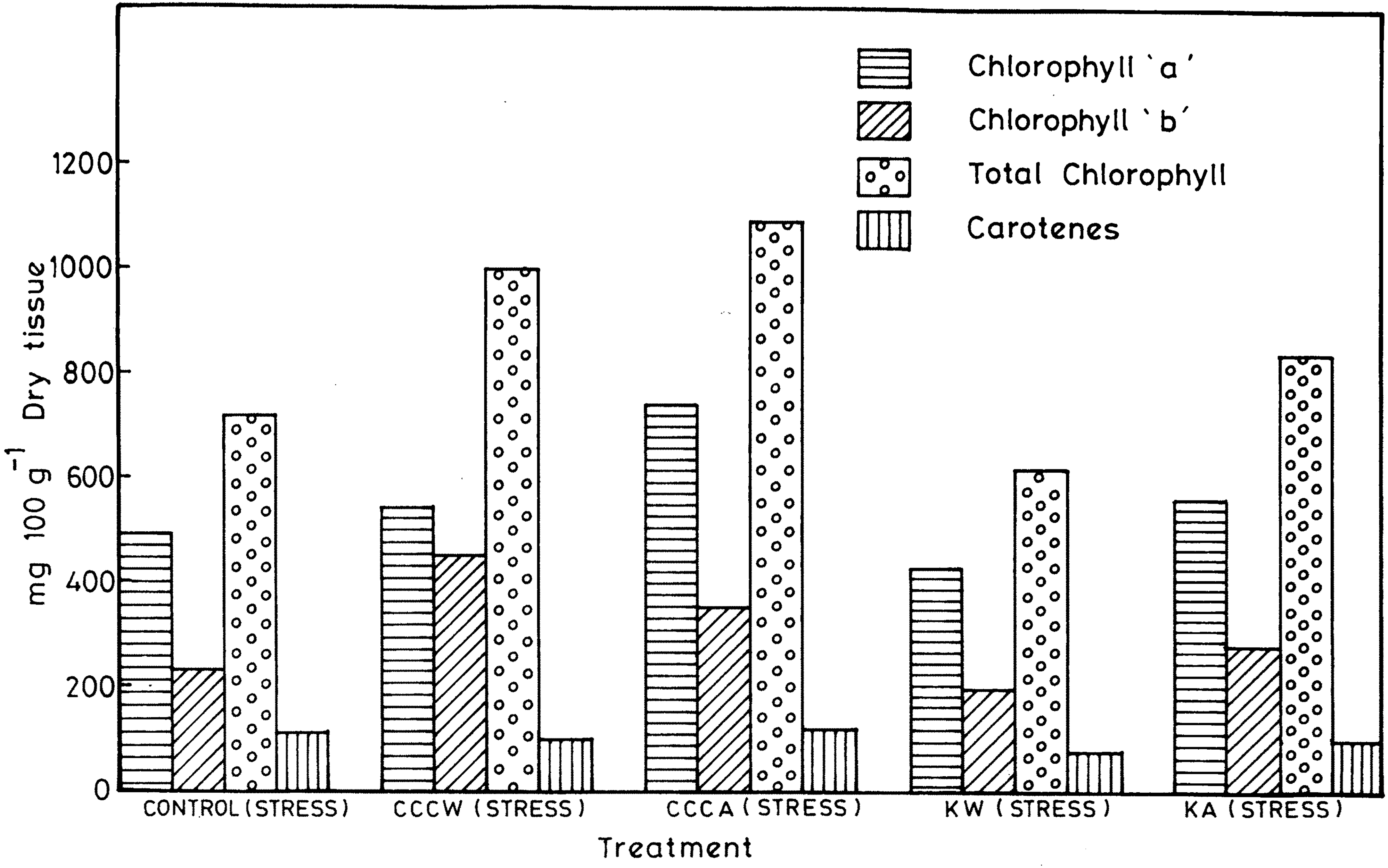

Fig. 13 EFFECT OF PRESOWING SOAKING TREATMENT WITH CCC AND KINETIN ON LEAF PIGMENT CONTENT OF GROUNDNUT (*Arachis hypogaea* L.) CV JL-24 UNDER WATER STRESS.

in the level of carotenoids in the leaves of water stressed groundnut plants raised from seeds subjected to different treatments and the untreated seeds.

Chlorophylls are main light harvesting molecules usually present in the chloroplast. In higher plants chlorophyll a and b are present. Chlorophylls are magnesium chelates of closed tetrapyrrole rings. Chlorophyll a is present in all green plants, however, Chl. b is present in higher green plants, euglenophyta and chlorophyta. The chlorophyll molecules play an important role in harvesting solar energy and thereby taking part in light reactions of photosynthesis. Therefore, chlorophyll content and their stability have direct bearing on photosynthetic efficiency.

Carotenoids are the accessory pigments which take part in the harvesting of solar energy and help the main photosynthetic pigments (chlorophylls) in trapping and transfer of solar energy. They probably act as antena pigments and accept radiant energy and transmit it with some losses to chlorophyll a molecule. The possible function assigned to carotenoids is photoprotection of chlorophyll against bleaching.

Virgin (1965) reported that water stress caused inhibition of chlorophyll 'a' formation. There are number of reports which indicate that water stress causes reduction in chlorophyll content (Reddy et al., 1980; Mayoral et al., 1981; Gujarathi [illegible] 1984; Biswas and Choudhari, 1986). Mohanty and Boyer (1976) observed reduction in quantum yield under water stress conditions. Gomez et al. (1981) observed that carotenoid content along with chlorophylls were high in tolerant varieties of sugarcane under low relative humidity than those of the susceptible variety. They also reported that the formation of carotenoids is a part of protective mechanism against unfavourable conditions like water stress.

There are few reports on the influence of growth regulators on chlorophyll content in plant leaves. Appleby et al. (1966) observed an increase in chlorophyll content in wheat plants when seeds were pretreated with CCC. Humphries (1963) found that chlorophyll content in the leaves was increased in tobacco plants in culture solution containing CCC. Yadav et al. (1978) reported that foliar application of CCC was effective in increasing chlorophyll and carotenoid contents in leaves of *Trifolium alaxandrum* (Berseem) plants. There are also several other reports which indicate that CCC influences chlorophyll content positively in various plants (Kanvel, 1969 in tomato leaves; Halevy and Shilo, 1970 in barley leaf discs; Gaber et al., 1981 in cotton leaves; Bhadre, 1983 in cotton varieties Varalaxmi and H-4). Gill and Singh (1978) reported that spraying of CCC (500 ppm) on wheat CV Kalyan Sona-227 was effective in maintaining higher levels of chlorophylls in leaves under water stress conditions. Zayed et al. (1985) observed increase in carotenoid content in leaves of Okra plants when they were sprayed with CCC (500, 750 mg/1). Lischchuck et al. (1985) reported that water stress caused accumulation of carotenoids in leaves of sweet cherry plants when treated with CCC.

The stability of chlorophylls under the conditions of water stress has been regarded as one of the important criteria for assessment of drought resistance (Kushinirenko et al., 1971; Tadzhiveva, 1980). It is apparent from the present investigation that the presowing soaking treatment with both CCC and kinetin are helpful in increasing the stability of chlorophylls in groundnut leaves under drought conditions. The increase in chlorophyll content caused by presowing soaking treatment would undoubtedly help the plants in maintaining photosynthetic activity under the conditions of water stress. At the same time it must be mentioned here that the presowing soaking treatments are not effective in raising the level of accessory and protective pigments carotenoids under the conditions of drought.

F) Stomatal behaviour :

Influence of presowing soaking treatment on stomatal behaviour in leaves of groundnut is recorded in Table 1. It is evident from the table that the water stress induces a complete stomatal closure in the leaves of control plants. This is reflected in eliminating transpiration and marked reduction in diffusive conductance for carbon dioxide. In the plants raised from seeds treated with CCC and kinetin (in acetone) the transpiration rate is slightly maintained so also the diffusive conductance to carbon dioxide. In this respect the pretreatment with CCC in acetone is highly effective. The presowing soaking treatment of groundnut seed with kinetin solution in acetone is more effective in maintenance of normal stomatal behaviour while the treatment with aqueous solution of kinetin is totally ineffective. These observations indicate that the seed pretreatment with solution of growth regulator in organic solvent acetone is more effective than that in water.

An exchange of gases between a living plant and its surroundings occurs constantly. In respiration oxygen is absorbed from the atmosphere and carbon dioxide is released into the atmosphere. In photosynthesis carbon dioxide is taken from the surroundings and oxygen is given out into the atmosphere. Transpiration is a process in which water vapour is released into atmosphere. In addition to gases like carbon dioxide, oxygen, water vapour, trace amounts of other volatile substances are exchanged between higher plants and the external atmosphere. For example ethylene, a plant growth hormone, may diffuse into external atmosphere while the synthetic gases of automobile exhaust may diffuse into plant tissues. In leaves of land plants gaseous exchange mainly takes place through stomatal pores. Stomata are present not only in the epidermis of leaves but also in certain fruits like banana, citrus, cucumber. The epidermis of leaf can be considered to be a multiperforated septum, through the pores of which outward diffusion of water vapour occurs. The path of outward diffusion starts at the sites of evaporation on wet mesophyll cell walls which border on substomatal cavities, continuous in stomatal pores, proceeds through a thin layer of water vapour which lies next to the surface of the leaf and ends in the bulk air beyond the boundary layer. The pathway of diffusion can be considered to be a system of resistances in series. These can be grouped into two categories - 1) the internal resistance and 2) the resistance external to leaf. The internal resistance to outward diffusion of water vapour from a leaf is associated with stomata and can be called the stomatal resistance. The stomatal resistance depends upon several factors such as shape and size of substomatal cavity, stomatal pore and size of stomatal pore. The size of stomatal pore is more important. The smaller the stomatal openings, the greater the resistance to outward diffusion of water vapour. The stomatal pore size varies with the turgidity of guard cells and therefore, it is actually a variable resistance. The leaf resistance is considered as a dynamic quantity which for a given leaf may change continuously with environmental conditions. It has been realized that leaf resistance is a function of relative humidity, temperature of the leaf, osmotic potential of the stomataes and other factors. The fall in photosynthesis at low water potential is accompanied by antiparallel changes in stomatal resistance in many species.

Water supply to a leaf plays a critical role in determining the size of the stomatal aperture, especially during periods of water shortage. When leaf water deficits begin to develop, guard cells become less turgid and the size of the stomatal operture decreases. If water shortage contimues and if leaf water stress becomes severe stomata usually will close completely and will remain closed. When the water content of a leaf drops below a critical value, stomata will close partially or completely even other factors required for opening of stomata are favourable. The stomatal closure is regarded as one of the major factors which limit photosynthetic carbon dioxide assimilation in higher plants. Such situation prevails in water stressed plants. The stomatal opening is prevented due to abscissic acid the level of which rises tremendously during water stress and this effect may persist several days beyond the return of the plant to a higher water potential, (Beadle _et al_., 1985). It is argued that the roots send the message to the leaves which induces stomatal closure as there proceeds a process of drying of the soil. According to Davis _et al_. (1986) this message is a change in the cytokinin supply to the leaves.

It is evident from the present investigation that the presowing soaking treatment especially with CCC are effective in maintaining some stomatal openings in groundnut leaves under the conditions of drought. This may be possibly due to maintenance of good water status in the leaves and thus keeping the turgor in guard cells. It has been reported by Nawata _et al_. (1985) that CCC causes increase in the cytokinin level in the root tissue. Thus the pretreatments with CCC and kinetin (in acetone) might also be influencing the hormonal balance which controls stomatal behaviour.