RESULTS AND DISCUSSION

1. Germination studies :-

a] Germination percentage:-

The influence of aluminium toxicity, on seed germination performance in groundnut cultivars SB-11 and W-55 is recorded in **Table 9 and Plate 2.**

The inhibition in germination percentage for first 24 hrs was noticed in both cultivars and it is more significant in cv. SB-11 than W-55. However in cv. SB-11, the performance of seed germination improves with germination period than cv. W-55. It was recovered upto 120 hrs except the higher concentration i.e. 100 ppm Al treatment which reduced seed germination percentage in both theses cultivars.

Seed germination is the process which begins with intake of surrounding water by the process of imbibition which ultimately causes swelling of seeds. The number of seed technologist considered seed germination as emergence of radical through seed coat. In present investigation the swelling of seeds is considered for calculating germination percentage. The success of seed germination and further performance determined by availability of water in the soil. Although movement of water from the soil into seed is mediated by several factors, but water relation of the seed and of the soil are important of them, (Bewley and Black, 1994). According to them, continued availability of water to the seed depends on water potential of zones of the soil immediately surrounding the seed and on the rate at which water moves through soil. Water potential is particular value for each crop species below which seed can not germinate.

It has been observed by many workers that many heavy metals in low doses are essential for many plants but their higher concentration in soil may cause metabolic disorders as well as growth performance (Claire *et al.*, 1991). In most acidic soil, aluminium toxicity is recognized as a major growth limiting factor. In pigeonpea seeds pH of the medium is very important for the growth This important pulse grows better in acidic pH i.e. pH 5 rather than neutral or alkaline medium.

The Effect of aluminium on germination performance has been studied by some workers. Narayanan and Syamala, (1989) studied the effect of aluminium concentration ranging from 0-600 ppm, on germination response of pigeonpea cultivar LRG-30 and found that above 100 ppm Al level, germination percentage was reduced significantly. In soyabean, genotypic difference in aluminium tolerance, was noticed by Yang *et al.*, (2000). The aluminium resistant nature was shown by variety

Suzunari, while variety *Shisho* was found to aluminium sensitive. Neogy *et al.*, (2000) studied the effect of aluminium on growth of *Vigna radiata* seedling. Seed germination declined with increased Al concentration but lower concentration of Al promotes seed germination. However according to Rout *et al.*, (2001) the seed germination process is not affected by aluminum, they noticed that Al stimulate seed germination, development of new roots and seedling establishment.

Bhamburdekar, (2002) studied the effect of aluminium concentration on germination response of pigeonpea cultivar ICPL-87. He noticed the initial reduction in germination percentage recovers in later stages of germination by all different Al concentration ranging from 5 to 50 ppm. Jamal *et al.*, (2006) conducted an experiment for screening the effect of Al and Cr on germination in two *Vigna* species viz. *V. radiata* and *V. sinesis,* separately and together. They noticed that germination percentage of both *Vigna* species were not affected by aluminum and chromium.

In present investigation the initial decrease in germination percentage in both cultivars was recovered as germination hours increased especially with lower doses of Al concentrations than higher concentration. This data indicate the ability of both groundnut cultivars to germinate and grow under toxic effects of Aluminum.

b] Seedling growth

The effect of aluminum toxicity on groundnut seedling growth is shown in **Table. 10**

The emergence of radical is not evident upto 24 hrs of seed germination. But thereafter, both cultivars differ in response to Al treatment. In cv. SB-11, the increment in root length is significant with 10 ppm Al and 100 ppm Al treatment, but stimulation in shoot length was noticed with all Al concentration. In contrast to it, in cv. W-55 inhibition in root and shoot length is noticed by Al treatment and it is more significant with lower Al dose i.e. 10 ppm Al.

The inhibition of root growth was also reported by some workers such as Foy *et al.*, (1974); Rhue, (1979); Kochian, (1995). The primary response of Al on root was well documented by Kochian, in 1995. According to him the primary effect of Al was observed on meristem (Ryan *et al.*, 1993). It is localized to root apex (Sivaguru *et al.*, 1999) affects root membrane permeability. The swelling of root and inhibition of root growth in response to Al was noticed by Elison *et al.*, (1998). The accumulation of Al in the nucleus was reported by McLean and Gilbert, (1927). Wacker and Vallee,

Table. 9 Germination percentage

W-55

Treatment	Germination Percentage				
	24 h	48 h	72 h	96 h	120 h
Control	90	100	100	100	100
10 ppm	80	100	100	100	100
50 ppm	60	100	100	100	90
100 ppm	50	80	80	80	80

SB-11

Treatment	Germination Percentage				
	24 h	48 h	72 h	96 h	120 h
Control	50	100	100	100	100
10 ppm	40	100	100	100	100
50 ppm	40	80	80	90	90
100 ppm	40	80	80	90	90

final count should have been made on 10th day instead of fifth day.

Plate No. 2
Germinating seeds of cv W - 55 and SB-11
(after 24 hrs)

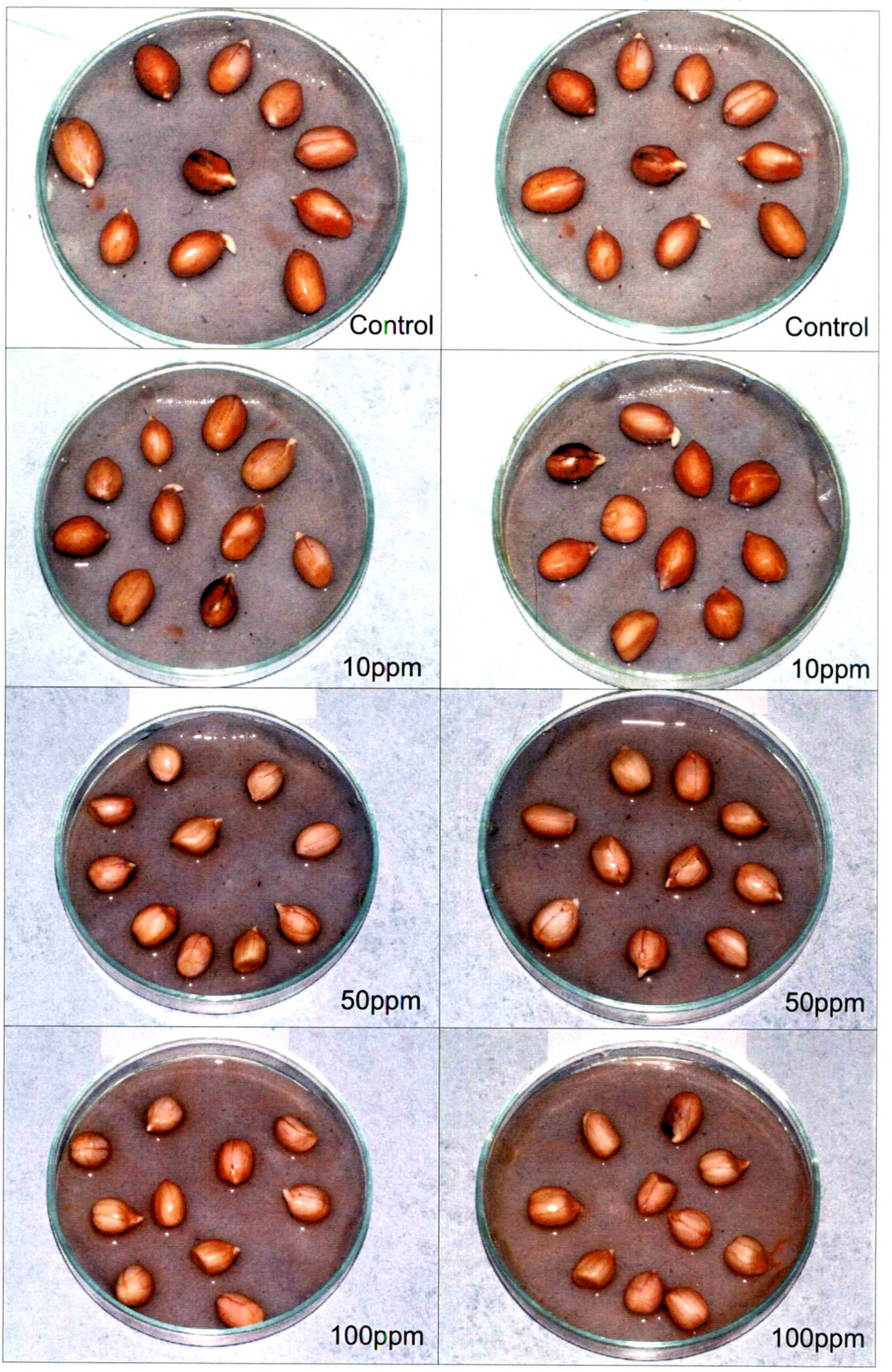
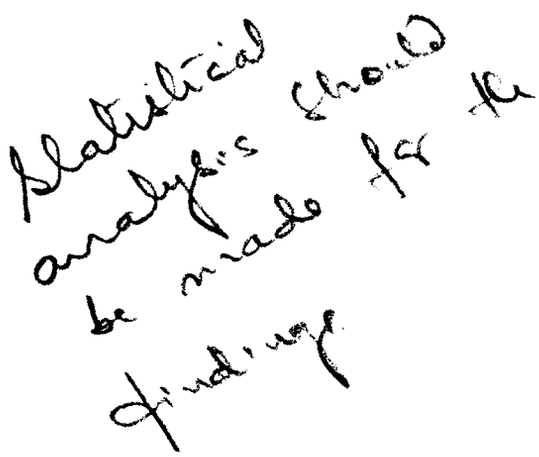


Table. 10 Effect of Al on growth parameters in groundnut (*Arachis hypogaea* L.) varieties.

		Germination Hours									
		cv. SB-11					cv. W-55				
Parameter	Treatment	24 h	48 h	72 h	96 h	120 h	24 h	48 h	72 h	96 h	120 h
Root length in cm	Control	-	0.41	0.5	0.6	0.7	-	1.08	2.2	3.1	3.5
	10 ppm	-	0.33	0.8	1.0	1.05	-	0.7	1.2	1.3	1.7
	50 ppm	-	0.5	0.4	0.6	1.0	-	0.5	1.3	1.4	2.0
	100 ppm	-	0.9	1.0	1.3	1.30	-	0.2	0.9	1.2	1.6
Shoot growth in cm	control	-	0.5	0.6	0.8	1.0	-	0.8	1.2	1.5	2.1
	10 ppm	-	0.46	0.70	1.02	1.07	-	0.66	0.8	0.9	1.1
	50 ppm	-	0.55	1.0	1.2	1.4	-	0.72	1.0	1.2	1.4
	100 ppm	-	0.6	1.0	1.3	1.4	-	0.27	0.7	1.1	1.3



(1959) detected the presence of Al with other trace elements in highly purified preparations of DNA and RNA, suggesting the possible role of Al in nucleic acid metabolism which interfere the cell division process ultimately affect the root tip and root growth.. Such roots may be pre dispersal to several soil fungi infection.

According to Asp *et al.,* (1988) shoot growth and seedling growth was mostly enhanced due to lower concentration of Al than higher concentration in all the species except in wheat seeds exposure to aluminum. According to Narayanan and Symala,(1989), the roots of pigeonpea were short, stubby, thickened, brittle and dark brown when germinating in aluminum solution. Hodson and Wilkins,(1991) found that aluminosilicate, by reacting silicate of cell wall with aluminum. Metal in bound form does not affect plant but it reduces growth when it was found in unbound form. According to Parker, (1995) roots of wheat cultivars that differ in aluminum tolerance acclimatized quite quickly after an initial severe decline in root elongation, which lasted from one to several hours after exposure to aluminum. Kidd and Proctor, (2000) studied the level of Al tolerance in different races of *Betula pendula* Roth. The Al at low concentrations (2 and 5 mg l^{-1}) enhanced growth in FM, and KP races but inhibition recorded in KP race of *Betula.* Similarly in Al sensitive race KP, there was a loss of apical dominance while increasing Al concentrations affects lateral and primary roots. Bhamburdekar, (2002) investigated the effect of Al on pigeonpea seed germination. He observed that root growth was inhibited with browning of root tips due to aluminum. Jamal *et al.*, (2006) studied the effect of aluminum and chromium on growth of two *Vigna* species viz. *V. radiata* and *V. sinesis.* In Al treated V*igna radiata*, root length was not inhibited. The length of root increases with increase in Al concentration, while in *V. sinensis*, Al concentration causes reduction in root length i.e. as the concentration of Al increases and root growth reduced, which confirm the observations of Foy *et al.,* (1978).

In present investigation, due to aluminum, root shows reduction in growth in both groundnut cultivars and it is more noticeable with cv. W-55 than cv.SB-11. The root growth inhibition was more significant than shoot. Similarly the browning of root tips was also noticed.

2. Moisture:-

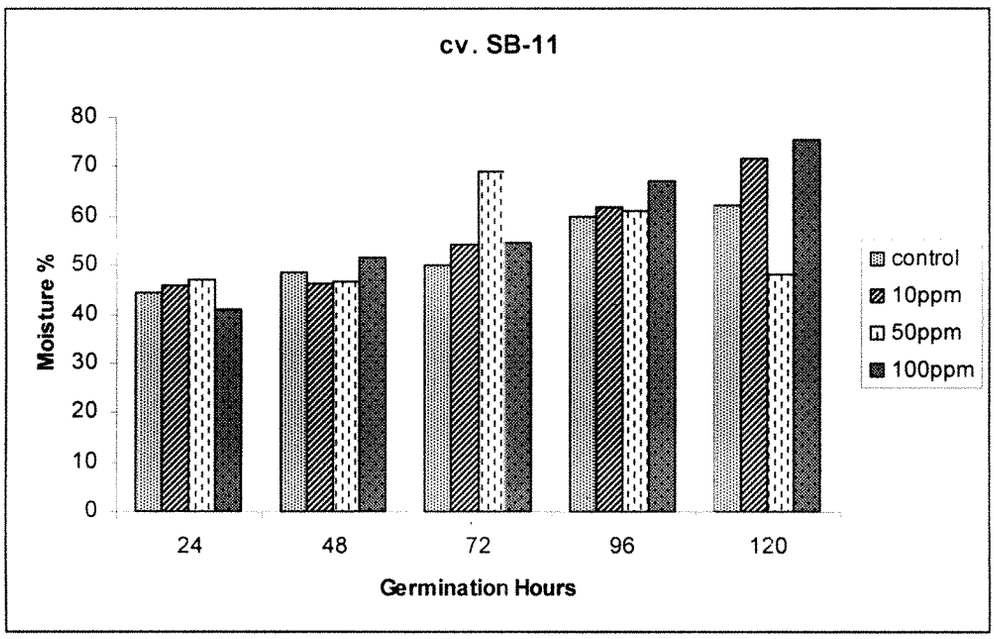
Effect of increasing concentration of Al toxicity on moisture percentage during groundnut seed germination is depicted in **Fig. 5**

In cv. W-55 the decrease in moisture percentage is noticed by treatment of Al during 24h and 120h of seed germination. However, between these hrs the pattern is not consistent with same cultivars. The increase in moisture percentage is evident by 10 and 100 ppm Al during major period of germination in cv. SB-11.

For all the living organisms water is important organic constituent, and about 90% of the total proportion of protoplasm for enzyme reactions which form the basic of biochemistry of plant. The cytoskeleton membrane has got water as the most important component. Thus water is essential solvent for translocation of metabolites and minerals within plants, which is essential for growth and enlargement.

Generally plant organs like seed consists of relatively small amount of water, are generally in dormant state. During germination of seeds number of functions played by water e.g. softening of seed coat, increased permeability of seed coat, activation of protoplasm of seed cells, medium for *de novo* synthesis of enzymes and other metabolic activities. For proper germination moisture content in seed should be more than 13% and if it is less than 5% it retards seed germination. However this percentage of moisture varies from plant to plant. According to Mixon, (1971) 35% of moisture was necessary for germination of groundnut seeds. He further experimentally demonstrates that about 55% to 60% of average seed moisture is necessary for emergence of radical in groundnut variety 'Early Runner'.

Water deficit causes dehydration of protoplasm (Levitt, 1956) which results in loss of turgor. Thus a reduction in moisture percentage is an obvious effect of water stress in almost all plant species. Ahmad *et al.*, (1979) noticed that water content in castor, bean and sunflower remained almost constant at lower as well as moderate level of salinity but it was decreased at higher level of salt concentration. Nigwekar in 1988 noticed in horsegram that moisture percentage negatively correlated with severity of drought. Salunkhe, (1990) observed in *Dodonaea viscosa* even under drought and saline condition higher level of moisture is maintained. Similarly, no significant difference in moisture percentage of *Brassica napus* and *B.carinata* under normal and saline condition (He and Cramer, 1993).



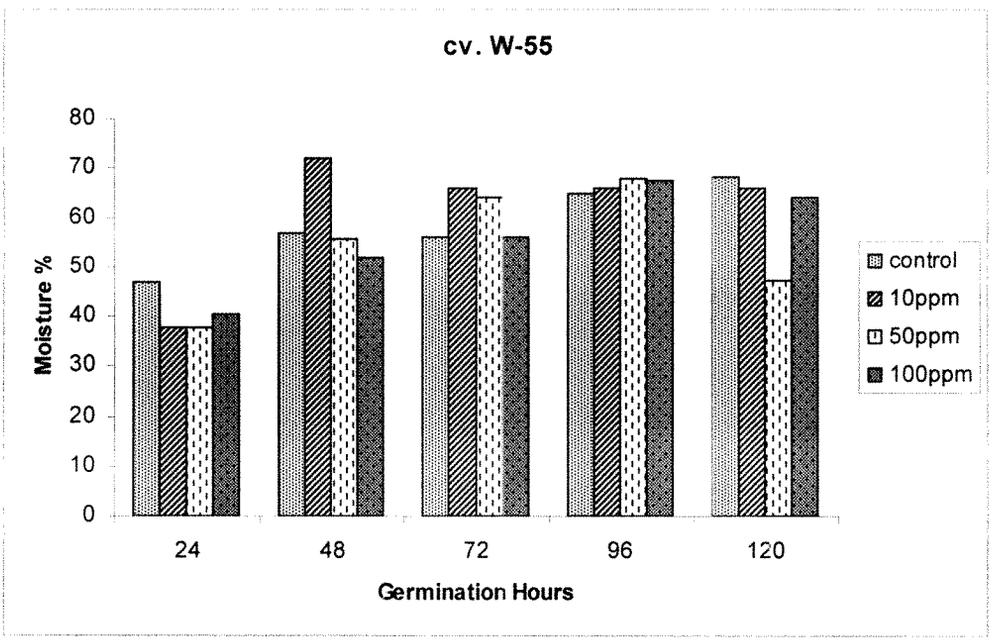


Fig. 5 Effect of Al toxicity on Moisture percentage during groundnut (*Arachis hypogaea* L.) seed germination.

The literature on the effect of Al on moisture content during seeds germination or growth stage is not available.

It has been observed by Ashraf and Orooj, (2005) that 10-15% moisture content suppresses seed germination capacity in *Trachyspermun* seeds.

In the present investigation the noticeable reduction in moisture percentage due to 50 ppm Al concentration at 120 hrs in both cultivars has no effect on germination performance. However increase in moisture percentage with all treatment after 48 hrs, stimulate seed germination in both cultivars, especially cv. SB-11 shows better performance to Al toxicity.

3. Qualititative determination of Al- tolerance :-

The visualization of Al tolerance with the help of hematoxylin stain in roots of Al treated groundnut seedlings is depicted in **Plate 4.**

Hematoxylin stain forms complexes with Al on roots and exhibit extensive dark purple staining. From the **Plate 4,** it is evident that, the development of purple color with hematoxylin was noticed with all Al concentrations in cv. W-55 and it is more distinct with 10 and 100 ppm Al treatment than 50 ppm where the entire portion of root is stained dark purple with hematoxylin all the studied cases. However in cv. SB-11, the formation of hematoxylin complex is evident only to higher dose of Al i.e. 100 ppm than other applied doses of Al.

Polle *et al.*, (1978) first time describe the qualitative difference in hematoxylin staining with Al stressed roots in Al–sensitive and an Al–tolerant wheat cultivars. Similar observation was also noticed by Wallace *et al.*, (1982). According to Rincon and Gonzales, (1992) and Tice *et al.*, (1992), the higher levels of Al was reported in root tips of Al sensitive wheat cultivars than to Al–tolerant ones.

The pattern of hematoxylin staining depends upon differential Al binding to uronic acid. i.e hematoxylin and reacting Al might be fixed in root tissue as $AlPO_4$. There are several reports of aluminium induced phosphate leakage viz. in *poplar* (McCormick and Bordon, 1972), wheat (Petterson and Strid, 1989) and sugarbeet (Lindberg, 1990). According to Luttge and Clarkson (1992), the presence of extracellular phosphate in Al sensitive might account for the differential staining of roots by hematoxylin. However the relationship of phosphate efflux to Al tolerance is not clear. In Al-sensitive cultivars of sugarbeet Al toxicity caused more accumulation

of extracellular $AlPO_4$, than in Al tolerant species, (Kesar *et al.,* 1977). Ownby, (1993) conduct an experiment to determine if the presence of extracellular phosphate in Al sensitive cultivars, and not in Al tolerant cultivars which may be due to differential staining of roots by hematoxylin. He further noticed that hematoxylin stain is prominent in cross wall and longitudinal walls of epidermis with idea that extracellular phosphate may immobilize Al in cell walls for reaction with hematoxylin.

It is evident from present investigation that the qualitative determination of Al tolerance might be observed with groundnut cv. SB-11 than cv. W-55.

4. Oil :-

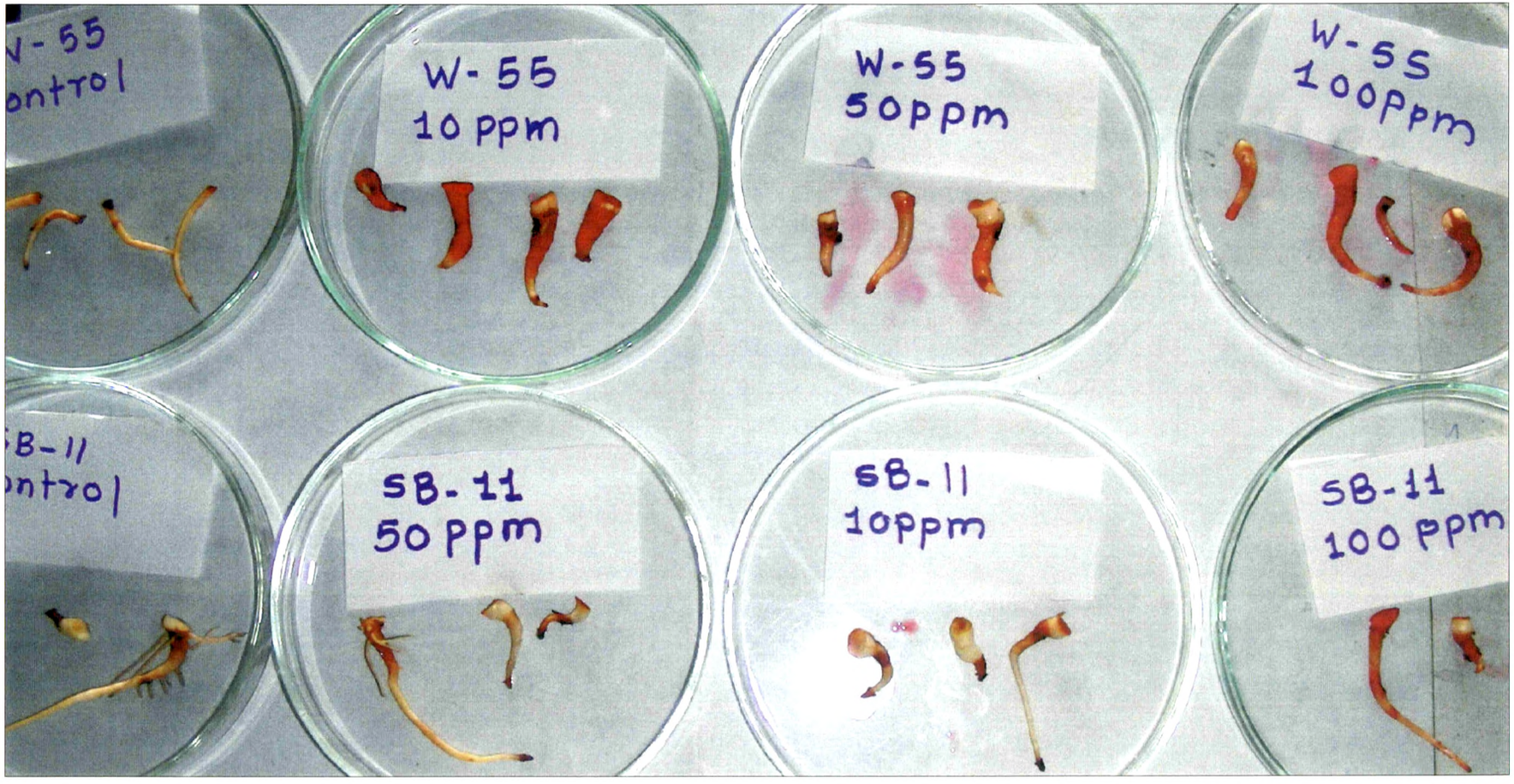
The effect of aluminium treatment on oil content during groundnut seed germination is depicted in **Fig. 6**.

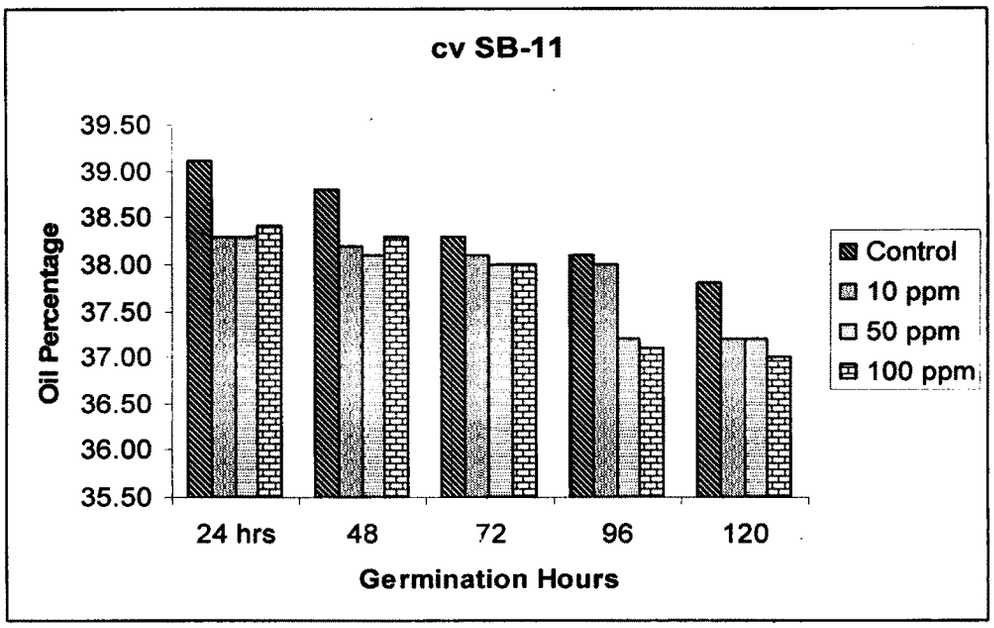
It is evident from our observation that oil content decreases with increase in germination period as well as with increase in aluminium concentration in both cultivars of groundnut. The pattern of decrease in oil content in more or less similar in both cultivars. The maximum reduction in oil content was noticed at 96 hrs by 100 ppm Al treatment in both cultivars.

Among the various oil seed crops groundnut is primarily used in the manufacture of vegetable oil (Vanaspati ghee). The most striking characteristic feature of groundnut is the manner of flowering and seed formation. The yellow flower born in the axils of leaves are self pollinated but after pollination, a gynophore developed from the base of ovary, which forces the ovary into the soil and fruit development occurs. The fruit is pod. The wall of the pod contains numerous seeds, which is composed of two cotyledons mainly contain oil and other food material. The stability of groundnut oil is called the shelf life. It is an important quality attribute. The ratio of saturated to unsaturated fatty acids actually determines oil quality especially the ratio of oleic acid to linoleic acid greater that 1.6 is an indicator shelf life without rancidity problem.

In oily seeds during germination oil stored are converted into fatty acids and glycerol. The fatty acids further breakdown into Acetyl Co-A by the process of β-oxidation. It is further converted into succinate by a series of reactions of glyoxylate cycle. **Fig. 7.** The succinate later on converted into carbohydrate is cytosol which is

Plate No. 4
Qualitative determination of Al tolerance
in groundnut





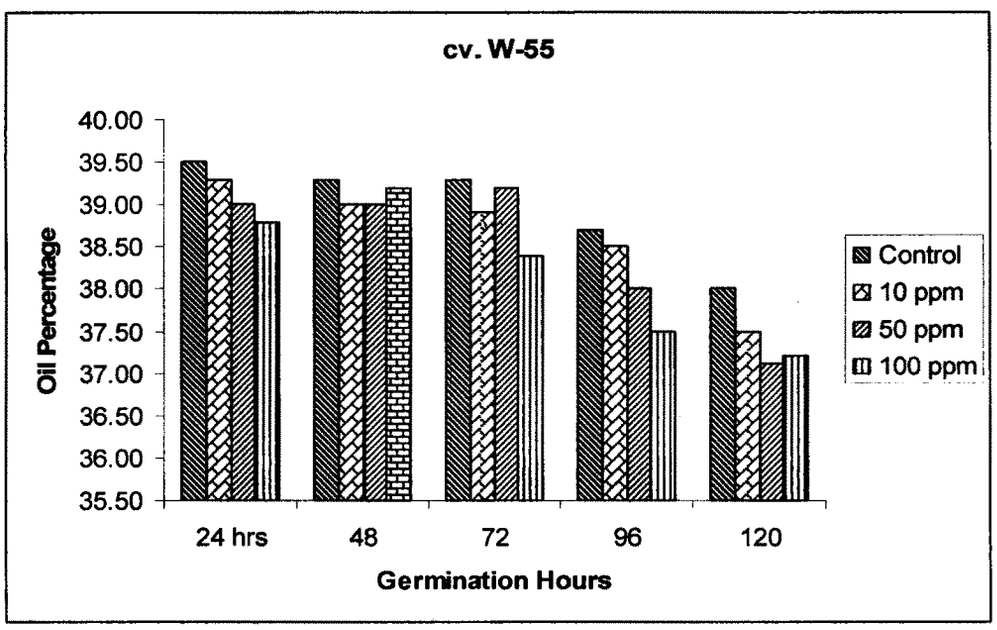


Fig. 6 The effect of Al toxicity on Oil content during groundnut (*Arachis hypogaea* L.) seed germination.

At 72 hours, oil content as well as Lipase activity is maximum - this should be made clear.

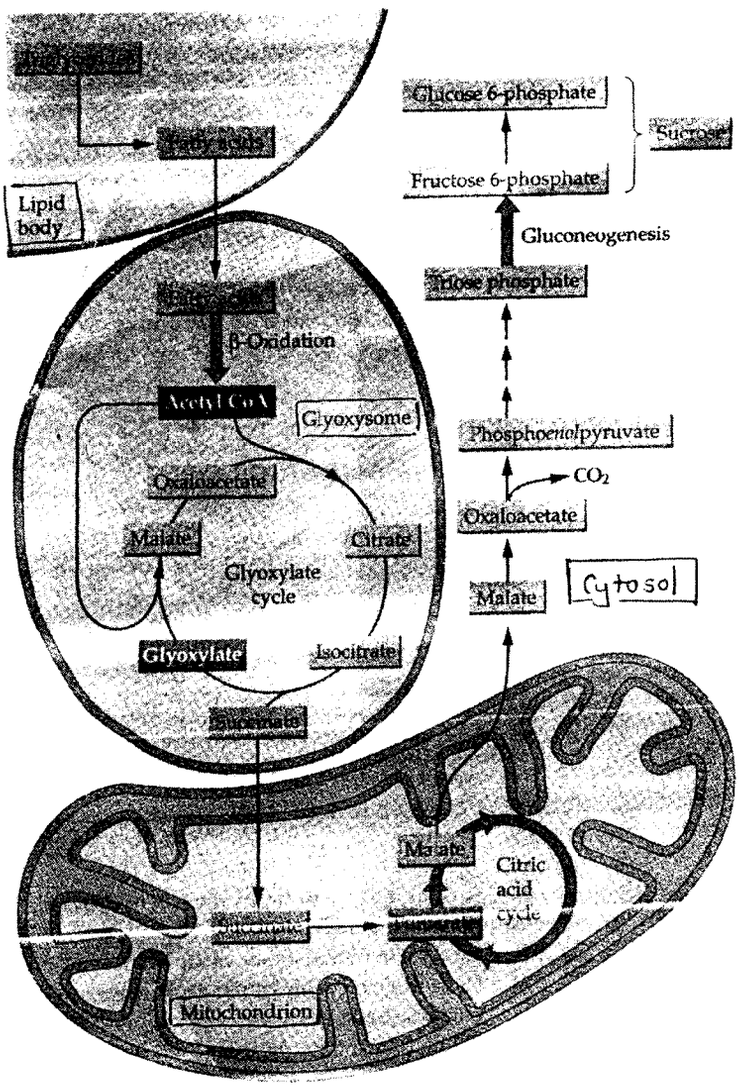


Fig. 7. Gluconeogenesis in fat storing seedling.

Plate No. 3
Germinating seeds of cv W - 55 and SB-11
(after 120hrs)

used in different metabolic activities. Thus presence of several glyoxysomes in close contact with oil bodies in germinating oil seeds noticed by many physiologist.

It has been noticed by Appelqvist, (1977) and Abdin *et al.*, (2003) that mineral elements, play important role in oil accumulation and its composition in oil seed crops. They further emphasis the role of Sulphur in oil accumulation. Sulphur is an important mineral which is an constituent of amino acids like Cystein, Methonine, Thioredoxins, Sulpholipids as well as coenzymes. The oil seeds crops require 'S' in large quantities and their role in oil biosynthesis was reported by many workers as Ahmad and Abdin (2000). Abdin *et al.*, (2003) and Fazil *et al.*, (2005).

However earlier workers like Schug and Hanneklau's, (2000) recorded the effect of balanced and combined application of 'S' and 'N' in increase in oil content.

The literature on the influence of aluminium on oil content during seed germination is not available. However the effect of various stresses including moisture stress, salt stress as well as insecticides, on reduction in oil content in Indian mustard seeds was investigated by Munshi *et al.*, (1986, 1987) Sukhija *et al.*, (1983) recorded, the decrease in fatty acid biosynthesis by the influence of various nutrients in higher plants. Similar reports were also noticed by Fazil *et al.*, (2005) in developing seeds of oil seeds crops, *Brassica compestris* land *Eruca sativa* Mill.

Asharf and Orooj, (2005) studied effect of salt stress on seed oil concentration in a traditional medicinal plant 'ajwan' (*Trachyspermum ammi* L.) and they reported little alteration in seed oil concentration with increasing salt level.

According to Peter, (2007) during germination of *Arabidopsis* seeds, storage oil breakdown to supply carbon skeleton, energy for early seedling growth and massive amount of H_2O_2 within the peroxisomes as a by product of fatty acid. Caterin *et al.*, (2007) reported, that in oil yielding capacity significantly decreased under salt stress in two sunflower hybrids. Increase in groundnut oil content due to application of phenolic compounds such as 8 amino 3-6 disulphate naphol and 1, 3 dihydroxy benzene was reported by Singh *et al.*, (1991).

In oily seeds, oil either stored in endosperm (castor, bean) or in cotyledon (Groundnut, sunflower). However, during seed germination of these seeds shows different response. In Seeds which stored oil endosperm, which shrivels as the fat is metabolized and carbohydrate products are translocated outside endosperm. But in groundnut, during first few days of germination the enzymes of glyoxylate cycle

increases rapidly in cotyledons glyoxysomes, (Buchhan, *et al.*, 2000). Thus yellowing of cotyledons is noticed due to development of rudimentary thylakoid formation.

In present investigation, the yellowing of cotyledon is noticed with decreasing oil content in cv. SB-11, due to aluminium treatment **Plate. 3.** However a very little effect was noticed in alteration of oil content due to aluminium toxicity in both cultivars of groundnut.

5. Carbohydrates :-

The effect of aluminium toxicity on carbohydrate content during groundnut seed germination is depicted **Fig. 8-13.**

The influence of aluminum toxicity (Al. 10 ppm. 50 ppm, 100 ppm) on carbohydrate content from 24 hrs to 120 hrs of seed germination in two groundnut (*Arachis hypogaea* L.) cv. SB-11 and cv. W-55 was different. In cv. SB-11 upto 72 hrs, the reducing sugars was increased but then decreased significantly with further germination of seeds. However opposite trend was notices in cv. W-55 with respect to increasing concentration of Al. The non-reducing sugars in cv. SB-11 decreased upto 72 hrs but then increased in later stages of germination. However it is increased throughout the course of germination in cultivar W-55. The starch content was decreased in both groundnut cultivars throughout the course of seed germination due to Al treatment.

The carbohydrates are the most abundant class of biomolecules distributed in plants and animals. They are indispensable for all living organisms. It mainly serving as skeletal structures in plants found as reserve food in various storage organs of plants such as seeds. Carbohydrate metabolism plays a key role during seed germination especially in seeds which stores starch as a major food reserve. In protein rich seeds especially in legumes, amino acids like glutamate, aspartate etc. are liberated due to proteolytic activities. These amino acids serve as precursors for sugar biosynthesis. Similarly in oily seeds, synthesis of sugars from fats noticed due to the process of gluconeogenesis. Thus during seed germination process, there is marked alteration in carbohydrate fractions including reducing, non reducing sugars and starch content. Such alterations determine the overall respiratory turnover as well as availability of carbon skeletons required for various metabolic processes.

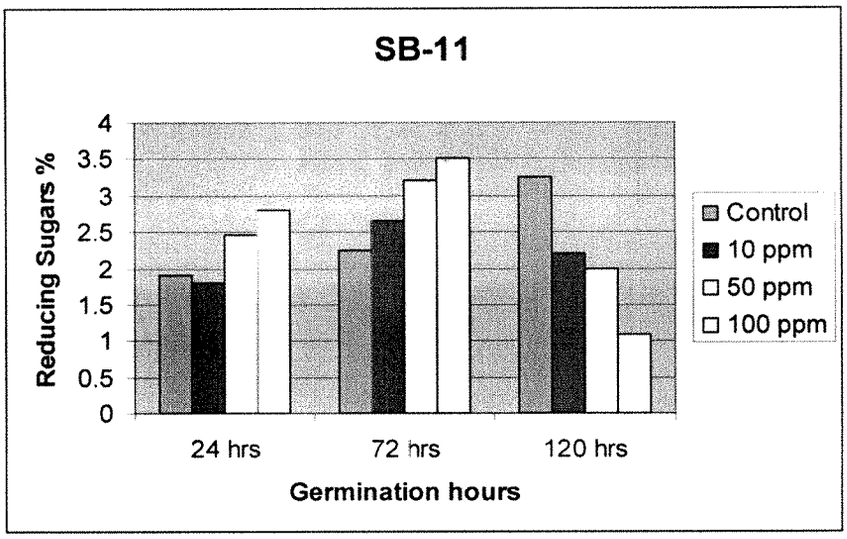


Fig. 8 Effect of aluminum toxicity in reducing sugar content during groundnut (cv. SB-11) seed germination

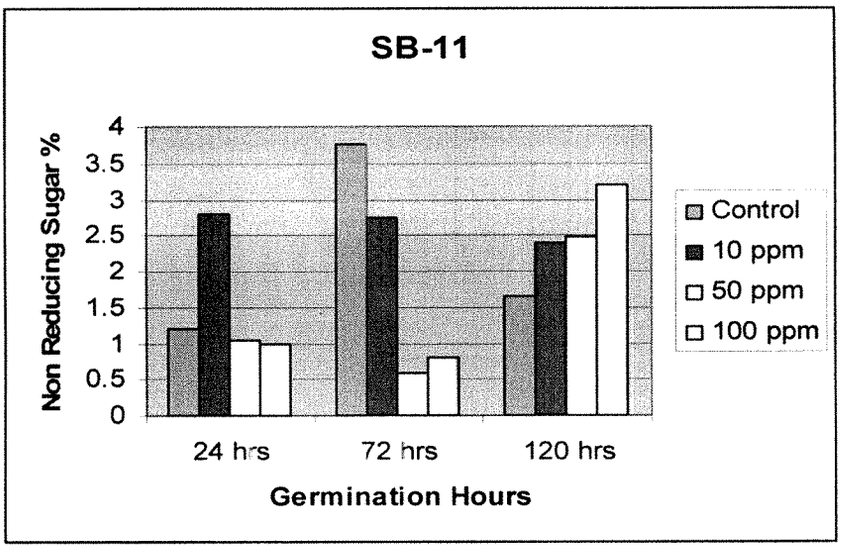


Fig. 9 Effect of aluminum toxicity in non-reducing sugar content during groundnut (cv. SB-11) seed germination

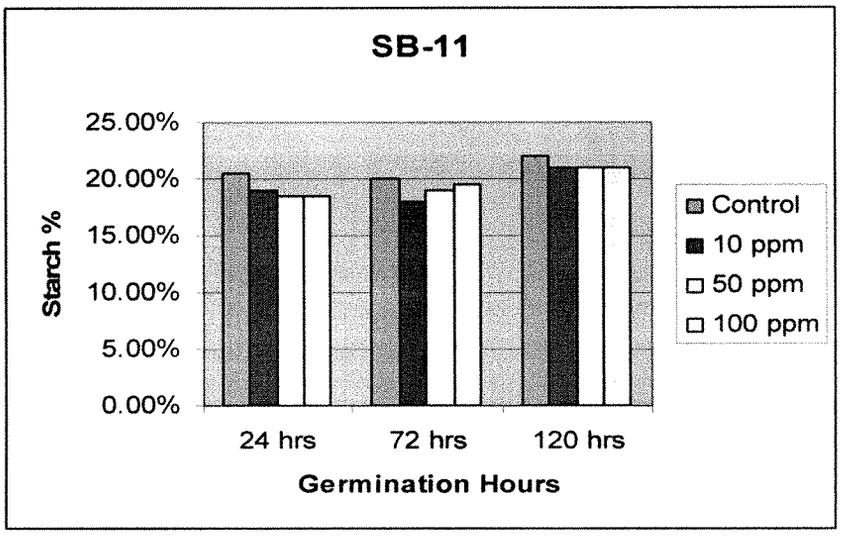


Fig. 10 Effect of aluminum toxicity in starch content during groundnut (cv. SB-11) seed germination

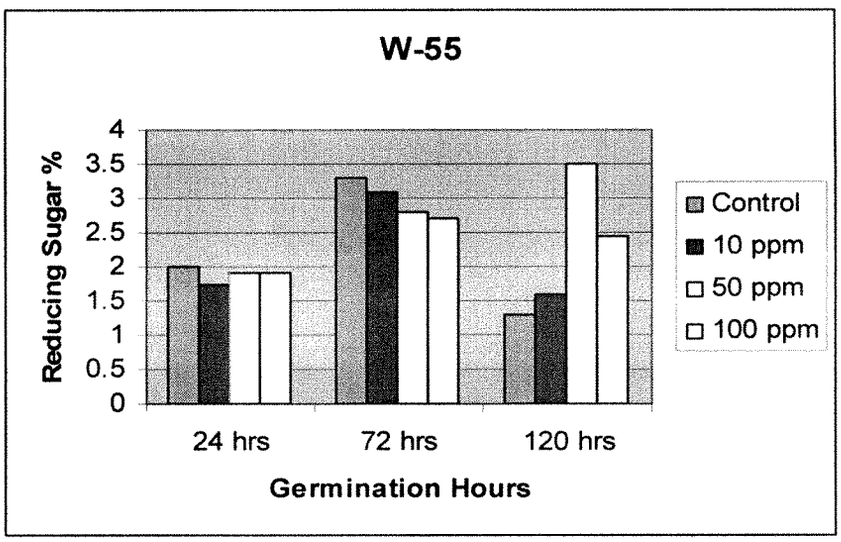


Fig. 11 Effect of aluminum toxicity in reducing sugar content during groundnut (cv. W-55) seed germination

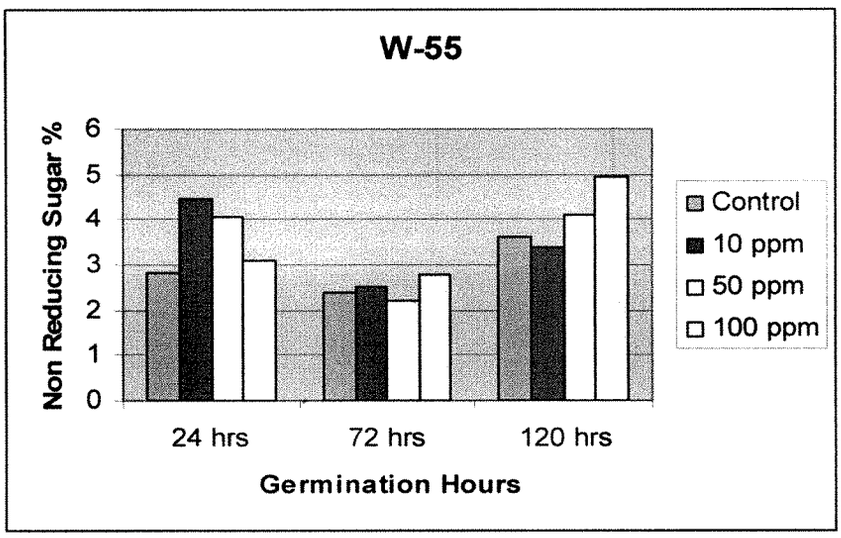


Fig. 12 Effect of aluminum toxicity in non-reducing sugar content during groundnut (cv. W-55) seed germination

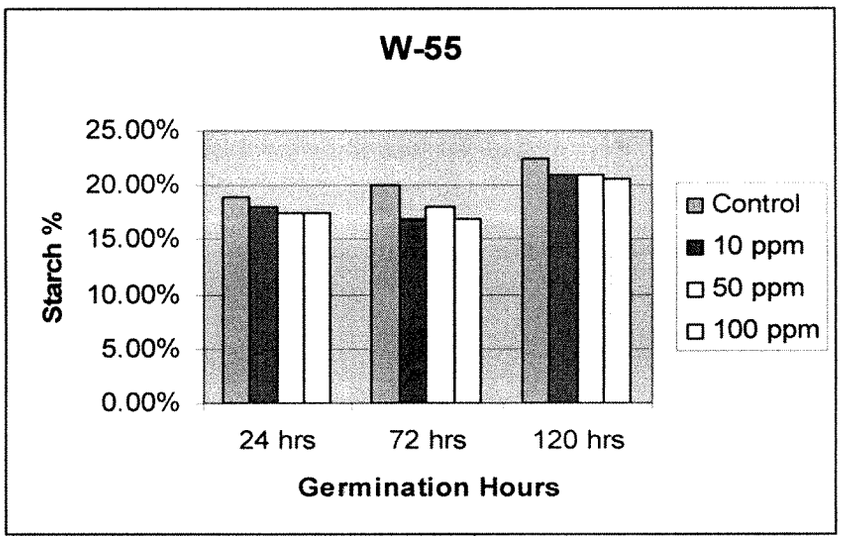


Fig. 13 Effect of aluminum toxicity in starch content during groundnut (cv. W-55) seed germination

The knowledge of influence of aluminium toxicity on carbohydrate metabolism is very scanty. There are only few attempts to study the influence of Aluminium on carbohydrate metabolism in plants (Schnable, 1970; Sarkunan *et al.*, 1984; Tabuchi 2004). Shestakov, (1940,) first time reported the influence of carbohydrate metabolism in the nodules of *Pisum* roots. The distribution of starch in the Al affected nodules was not restricted to starch sheath, but it is distributed throughout the tissue.

The reduction in reducing sugars due to increasing Al supply was noticed by Sarkunan *et al.*, (1984) in rice plant. This investigation was reported by measuring respiration rate. Similar report was also noticed with other toxic elements such as 'Cd' (Greger *et al.*, 2006). The opposite trend was observed in Jawar by Cambaria *et al.*, (1983). They noticed increase in reducing sugars with low concentration of Al treatment i.e. 2 ppm only. In present investigation, in cv. W-55 increase in reducing sugars at 120 hrs with increasing Al concentration was noticed. However similar report was also reported in cv. SB-11 only upto 72 hrs. This increase in reducing sugars may be due to breakdown of non reducing sugars by elevation in activity of hydrolic enzymes such as invertase. These finding correlate with finding of Pillay (2006), who reported similar increase in reducing sugar content in *Hyptis saveolens* L. at higher concentration of Al (500 ppm and 1000 ppm).

In 1990, Pahlsson studied the influence of various concentrations of Al on various fractions of carbohydrate in beech plant. He observed increase in non reducing sugars, total sugars and starch content in response to Al treatment. The non reducing sugars in cv. SB-11 was decreased successively with upto 72 hrs with higher concentration of Aluminium i.e. 50 ppm and 100 ppm, while in cv. W-55 the non reducing sugars are increased throughout course of germination. Schnable (1970), reported a stimulating effect of Al on starch synthesis which was more intense in the lower epidermis having more number of stomata, he further thinks that Al is able by this mean to regulate stomatal movements.

However, decrease in starch content, during seed germination of both cultivars due to Al treatment is evident in the present investigation. The product of this starch degradation useful for growth and development of groundnut.

6. Enzymes :-

a] Lipase :-

Effect of increasing concentration of aluminum treatment on lipase activity during groundnut seed germination, is depicted in **Fig. 14, 15.**

In groundnut cv. W-55, aluminum stimulates the enzyme activity throughout the course of germination except 96 hrs. The highest enzyme activity was noticed by 100 ppm concentration of Al at 48 hrs and 72 hrs of germination. Similar pattern is also reported in cv. SB-11 i.e higher Al-concentration stimulate lipase activity upto 72 hrs, but 10 ppm Al promotes enzyme activity at 120 hrs.

The storage lipids is the respiratory fuel for seedling growth in oil seeds. Break down of these fats to fatty acids, which are then oxidized by the enzymes of the β-oxidation and the glyoxylate cycle inside the glyoxysomes (Beevers, 1969; Hutton and stumpf, 1969). In fatty seeds during seed germination, lipid is converted into sucrose. The enzyme Lipase is mainly responsible for degradation of this lipids.

Acid lipase and Alkaline lipase are the two groups of lipases found in seeds. According to Huang *et al.,* (1978) the storage tissues of peanut seeds (*Arachis hypogaea* L.) contained only alkaline lipase activity, which acts on mono acyl glycerol rather than triglycerol, and the activity of enzyme lipase decreases during the course of seed germination.

In germinating oily seeds, lipase activity is a prerequisite for the metabolism of storage lipid. Hormones control mobilization of reserve materials in seeds and major role played by embryo. However in castor bean lipid degradation takes place in the endosperm (Bewley and Black, 1978). According to Huang and Moreau, (1978) during germination, triglycerides and starch, stored in the cotyledons of the groundnut are mobilized. About 60% of lipase activity associated with the glyoxysomes, 15% with mitochondria and 25% with a membrane fraction at density of 1.12 gcm^{-3}. Lipids in glyoxysome was associated with the membrane organelle and hydrolyzed only monoglycerides where as mitochondria and membrane fraction enzymes hydrolyzed mono-, di-, and triglycerides equally.

Although aluminium toxicity is recognized as major factor limiting growth in plants in most acidic soil, there is not much data available on effect of aluminium on lipase activity during seed germination. Younis *et al.,* (1987) shows that inducing germination of seed of flax, cotton, castor bean which grown in salt concentration on

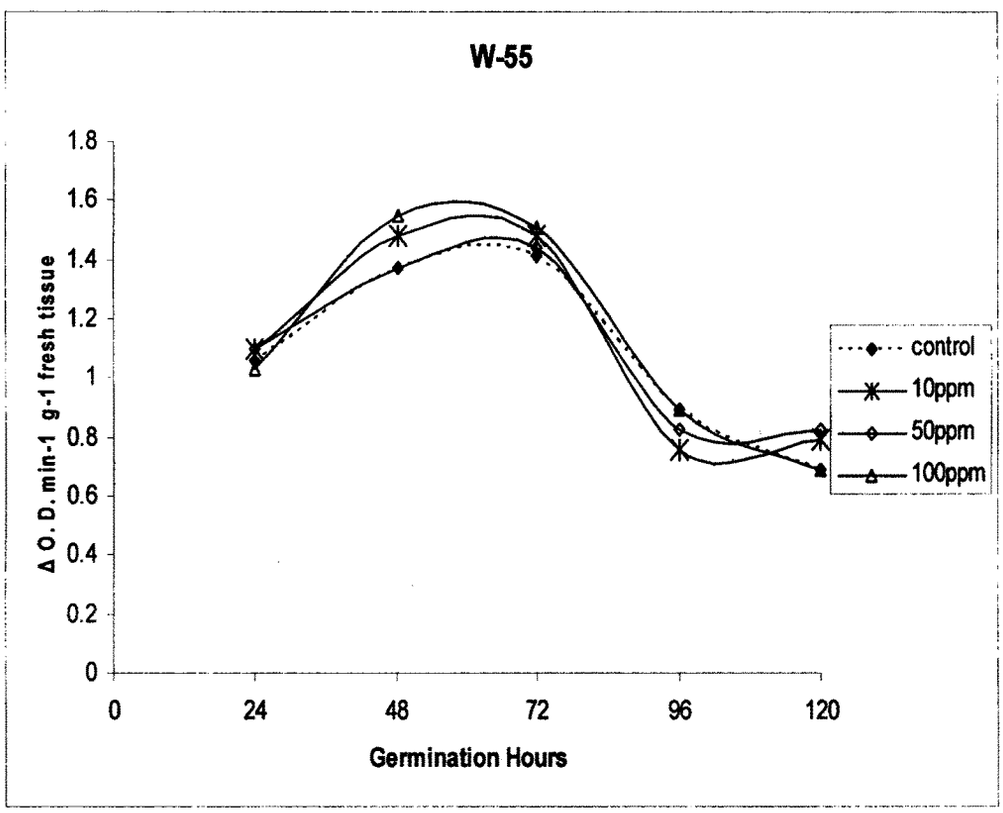


Fig. 14 Effect of Al on Lipase enzyme during groundnut (*Arachis hypogaea* L.) Seed germination.

Correlation between lipase activity and oil content needs to be clarified.

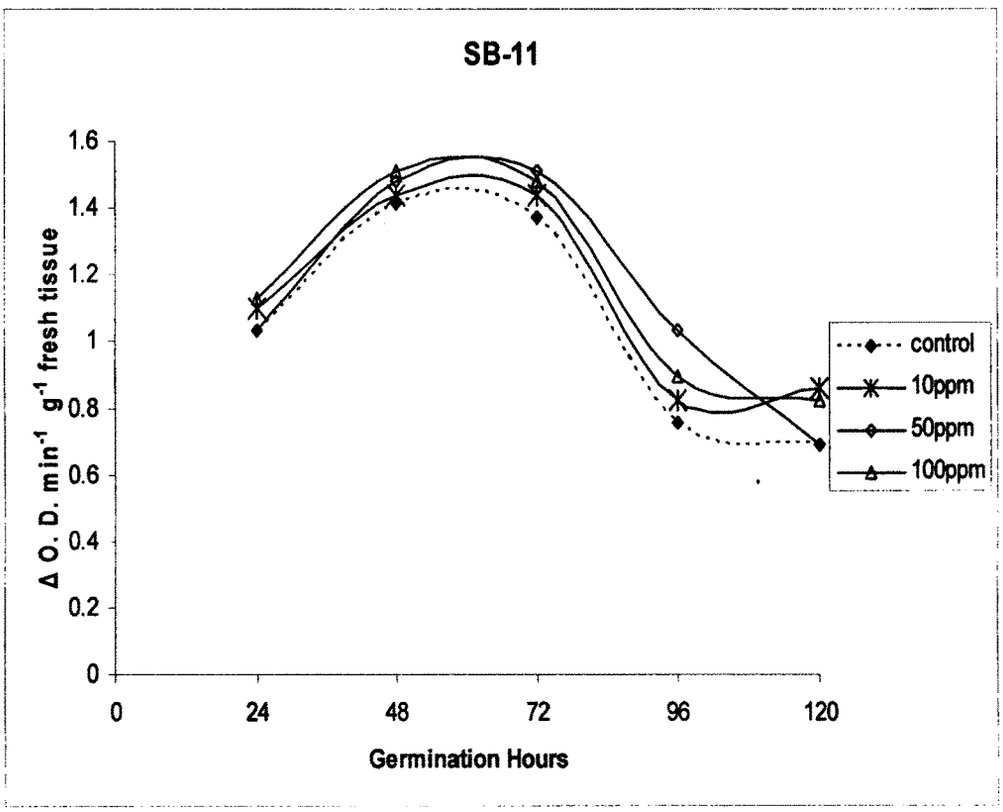


Fig. 15 Effect of Al on Lipase enzyme during groundnut (*Arachis hypogaea* L.) Seed germination.

the whole two salt concentrations (0.5% and 1% NaCl) causes marked decrease in lipase activity. However in cotton and castor bean, at lower salt concentration lipase activity showed a significant increase.

According to Miled *et al.*, (2000), worked on NaCl effects on lipase activity in germinating rape seed, shows that lipase activities from subcellular fractions were found to decrease under increasing salt concentration.

Shaha *et al.*, (2004) worked on lipase activity in germination oil seed and ionic effect on mobilization of seed storage and shows that there was delay in degradation of triacylglycerol (TAG), which is major form of food reverse, as salt concentration (100 mM) and buffer (150 mM) concentration increased while in control cotyledons TAG molecules were quickly hydrolyzed. In cotyledons of germinated *Brassica* delay in TAG degradation caused by salts and buffer stress is may be due to an inhibition of lipase involved in TAG hydrolysis. *In vivo,* TAG degradation was inhibited NaCl at elevated concentrations at 150 mM salt solution, this activity was totally inhibited and TAG level equal to dry seeds. Many reports showed that most lipase activity occurred in the microsomal fraction (1000009 pellet), (Hills *et al.*, 1988; Marphy, 1989).

Slawomir Borek *et al.*, (2006), shows in germination of yellow lupine seeds, low level of soluble sugars, accelerates the mobilization of storage materials. While transfer of carbon from liquid into amino acids can be stimulated by high level of sugar.

The stimulation of enzyme lipase activity due to Al in both groundnut cultivar is evident from our investigation .The maximum activity of enzyme reported during 48 to 72 hrs of seed germination might be due to high demand of respiratory fuel for growing groundnut seed.

b] Peroxidase :-

Fig. 16, 17 shows the effect of aluminium concentration, on the activity of enzyme peroxidase during groundnut seed germination.

The initial reduction in enzyme activity upto 48 hrs of seed germination was noticed in both cultivars of groundnut. But there after stimulation of enzyme peroxidase activity was recorded by all concentrations of Al treatments in cv. SB-11. However in cv. W-55, the only elevation of enzyme activity by 50 ppm Al treatment was noticed at 96 hrs.

Peroxidase enzymes contains heme and consist of three classes such as class I, consist enzymes from mitochondria, chloroplast, bacteria. Class II from fungi and class III are true plant peroxidase (Welinder, 1992). The detail structure of enzyme peroxidase, in *Arachis hypogaea* was studied by Schuller *et al.*, (1996). According to them, peroxidases are formed from a colorless glycoproteins combined to a brown red ferroporphyrin. It is present in various subcellular components, cell membranes. Enzyme Peroxidase is one of the most extensively studied oxidative enzyme system in plants (Gasper *et al.*, 1982). The intensification of peroxidase in legumes was noticed by Ioana, (1969). Peroxidase is also involved in the wasteful respiration process in plants, (Baba *et al.*, 1965). The peroxidase play important role in IAA oxidation (Fox *et al.*, 1965) in lignin synthesis (Stafford, 1965) and in regulation of cell growth and differentiation. Ivanova *et al.*, (1967), reported that mitochondrial peroxidase may be associated with electron transfer system from $NADH_2$ to cytochrome. It catalyze the reduction of nitrate, in presence of some specific electron donors (Pelvi *et al.*, 1972).

Gopalachari, (1963) reported that in roots and cotyledons of *Phaseolus mungo,* peroxidase activity increases during seed germination and in seedling growth. By studying four varieties of soyabean Komboj and Nainawatte, (1978) conclude the possible correlation between peroxidase activity and germination capacity. According to Zairo, (1983) during wheat seed germination, both quantitative and qualitative changes takes place in peroxidase activity during germination and active sites may differ in germinating seeds.

The effect of Al on the activity of enzyme peroxidase is very scanty. However the activity of this enzyme is inducible due to oxidative stress was reported by some workers as Foyer *et al.*, (1994) Sreedevi *et al.*, (2008). The toxic effect of several metals including 'Cd' leads to production of relative oxygen species (Dixit *et al.*, 2001). The antioxidant enzyme like peroxidase help in scavenging ROS and thus protect plant species from cellular damage. The effect of Al on the activity of enzyme peroxidase was carried out by some workers. According McElroy and Nason, (1954) Al is specific activator of enzyme ascorbate oxidase. It may be postulated that Al by adversely affecting many physiological stress reaction which enhanced peroxidase activity, (Peters *et al.*, 1989). Cackmark and Horst, (1991) reported that, in response to aluminium, peroxidase activity is increased in soyabean root tips. Similarly in rice and French beans seed germination, increasing Al concentration enhances peroxidase activity was noticed by Subramanyam, (1998). This may be related with lipid

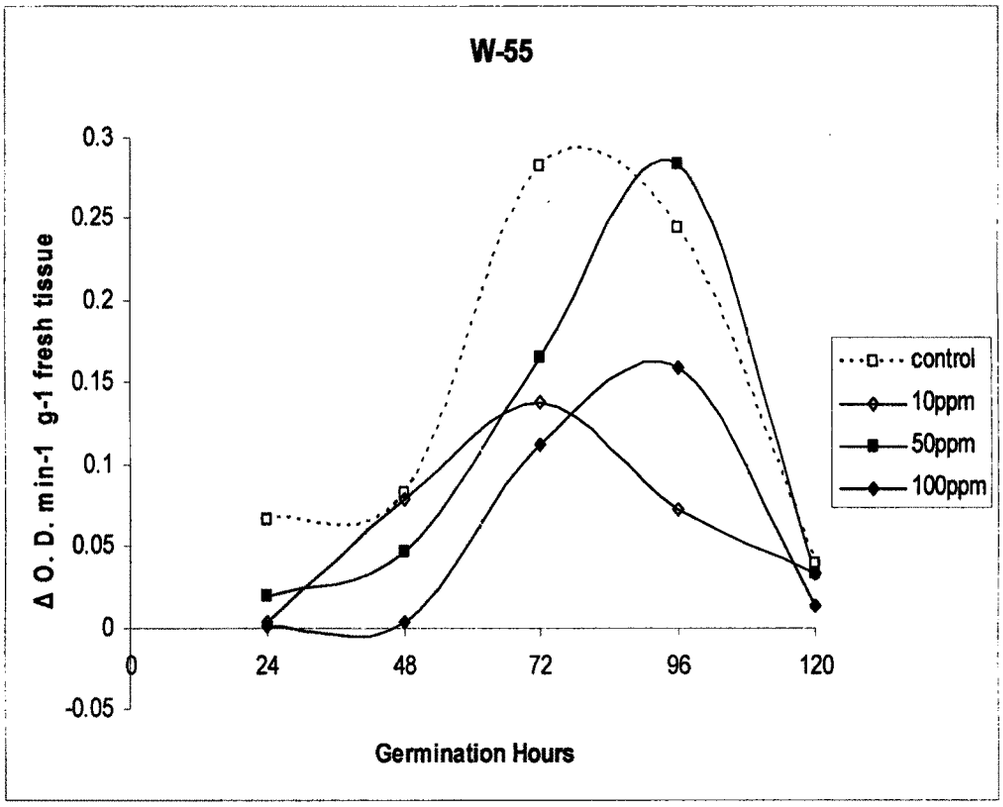


Fig. 16 Effect of Al on Peroxidase enzyme during groundnut (*Arachis hypogaea* L.) seed germination

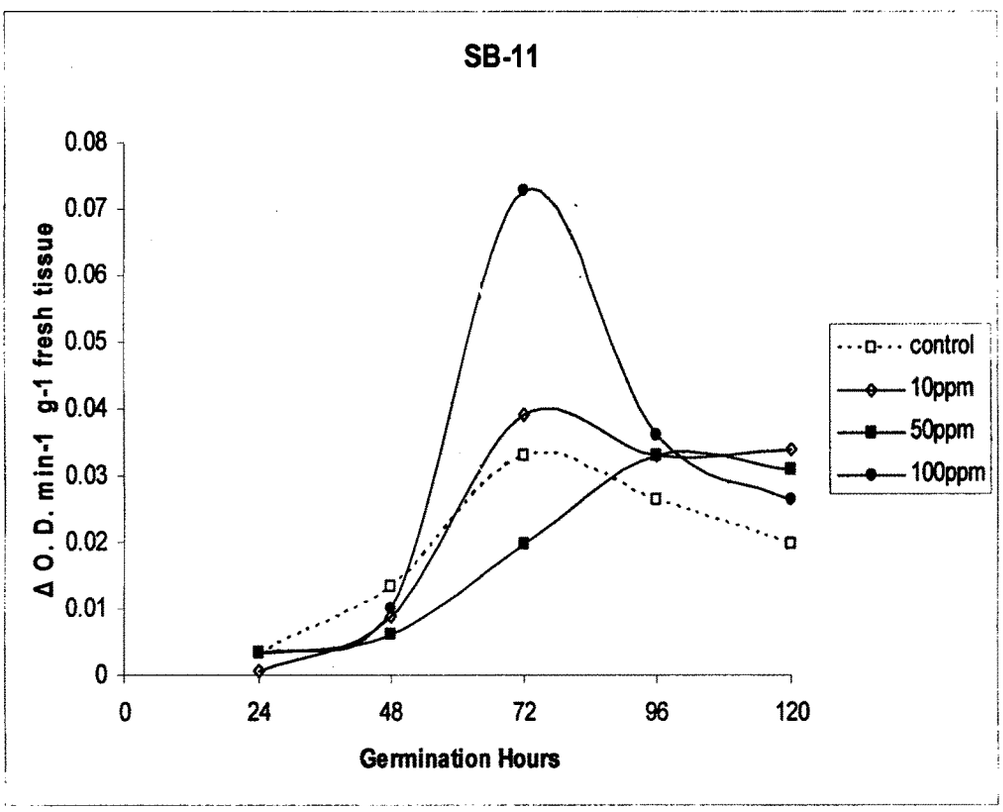


Fig. 17 Effect of Al on Peroxidase enzyme during groundnut (*Arachis hypogaea* L.) seed germination.

peroxidation and increment in Al treatment. Bhamburdekar, (2002) reported in pigeonpea seed germination that low doses of Al treatment inhibitory for peroxidase while enzyme activity is enhanced by higher concentration. Recently Panda and Khan, (2004) suggested the detoxification of H_2O_2 produced in *Lemna* under Al^{3+} treatment. There was 6-9 fold increase in ascorbate peroxidase activity due to aluminium.

It is evident from our investigation that Al treatment modified the activity of peroxidase in both groundnut cultivars may be through disturbance of Calcium homeostasis by aluminum as Calcium is component of peroxidase (Van Huystee, 1969). The stimulation in enzyme activity in cv. SB-11 may reflect a common strategy to overcome the stresses due to Al.

7. Inorganic minerals :-

Minerals elements play important role in plant growth and development by acting as a cofactor for number of enzymes. But their deficiency (Nason *et al.,* 1951), adversely affect the overall yield as well as quality of oil in different oil seed crops like groundnut (Appelqvist, 1977; Abdin *et al.,* 2003). The high requirement of different macro and micro nutrients for the growth and development of groundnut was investigated by different workers such as (Cox *et al.,* 1970; Hartzog and Adams 1980; Singh, 2004 and Singh and Choudhari, 2006).

However being a comparatively drought tolerant crop with low transpiration, the groundnut is susceptible to various nutritional disorders due to insufficient supply of minerals (Beringer and Taha, 1976). Hence it worth while to study the influence of Al toxicity on mineral uptake in groundnut.

a. Nitrogen :-

It is clear from **Fig. 18 that,** Nitrogen (N) level due to Al treatment is found to be increased in cotyledons than embryo axis in cv. SB-11 and it is significant by higher concentration (100 ppm) Al treatment. Similar pattern is also noticed in embryo axis of cv. W-55, opposite trend was recorded in cotyledons of same cultivar.

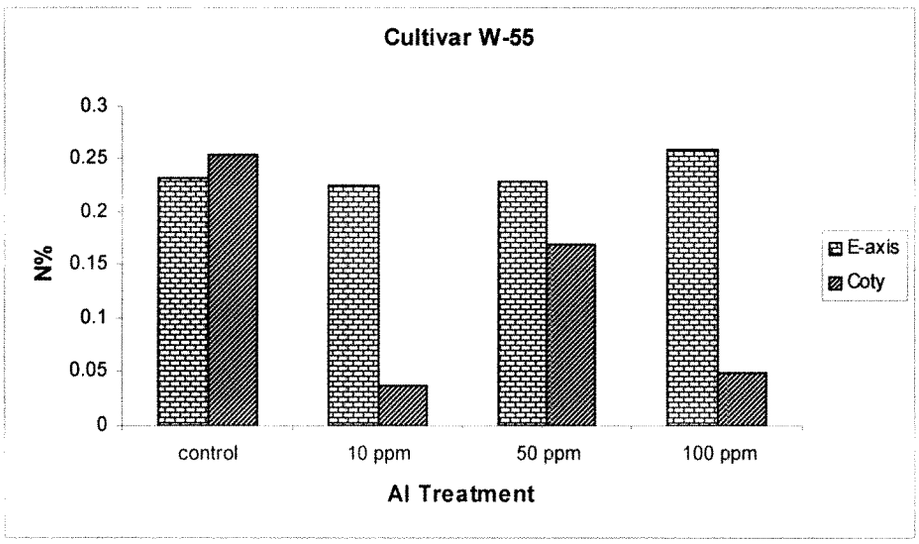
Inspite of the large quantities of molecular nitrogen present in the atmosphere, soil is the chief source of nitrogen to most of the plants. Soil consist of 0.1 to 1% of nitrogen.

In plants, nitrogen performs several functions. It is an essential component of proteins, hormones chlorophylls, vitamins etc. It is supposed to be very essential mineral element, found in purines and pyrimidines of nucleic acid, RNA and DNA thus it is essential for protein synthesis (Kumar and Purohit, 2003). Nitrogen is found in the structure of porphyrin molecule which is essential for synthesis of chlorophyll and cytochromes.

The mineral nutrition of groundnut was well documented by Singh in 1999. According to him Nitrogen (N) is very important element in groundnut as it is required for the vegetative and reproductive growth, nutrient absorption, photosynthesis and production of assimilates for developing pods, (Singh, 1999). The Nitrogen (N) requirement of groundnut is much higher than cereals because of it's high protein content and most of it absorbed through nitrogen fixation. Groundnut is capable of fixing 'N' from both symbiotic N fixation (60-80%) and by root nodules and soil 'N' (20-40%). The effect of Al on nitrogen and nitrogen containing compounds initiated with work of Klimashevsky *et al.,* (1970). According to Nambiar and Anjaiah, (1989) the toxic effect of Al were more on 'N' fixation than plant growth. They demonstrated that the amino acid composition of proteins from the roots modified due to Al treatment. These changes leads to Al tolerance capacity of plant. According to Durieux *et al.,* (1995) the primary response of Al addition is inhibition of NO_3^- uptake. Lidon *et al.,* (1998) reported that aluminium toxicity modulates nitrate to ammonia reduction.

While Lidon *et al.,* (1999) noticed that in *Zea mays* cv. XL-72.3, shoot nitrogen (N) decreased significantly with increasing Al concentration, at different rates. He also conclude that observed Al mostly retained in root tissue, but the visible effects are seen both in root and shoot and hence, N, P, Fe contents significantly decreased in shoot, similar results were also noticed by Munns (1965a, 1965b), Maclean and Chiasson, (1966); McLead and Jackson, (1967); Clark *et al.*, (1981) and Furlani and Clark, (1981).

In contrast to earlier investigation increase in 'N' in cv. SB-11 and embryo axis of cv. W-55 is an adaptive nature for maintenance of nitrogen level. However significant decrease in 'N' level in cotyledon of cv. W-55 may be due to transport of it to the developing embryo.



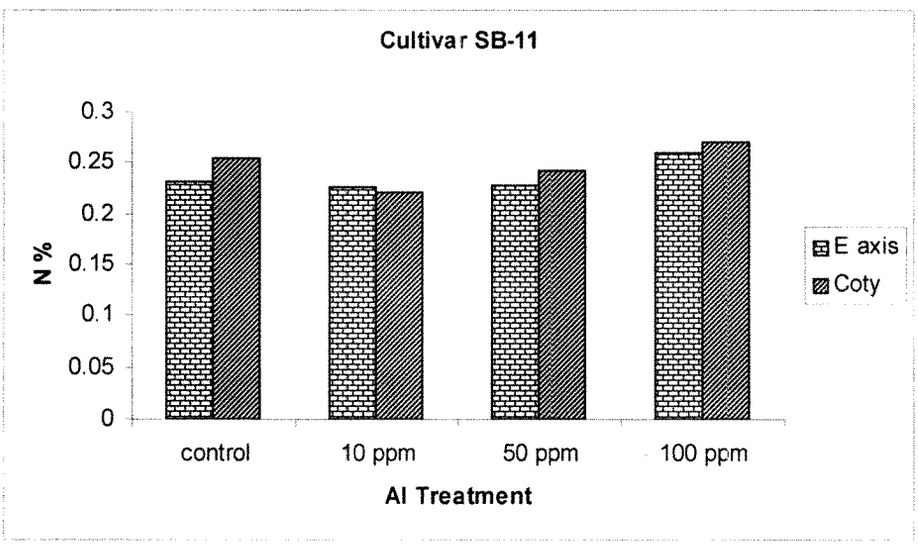


Fig. 18 Effect of Al-treatment on Nitrogen content during groundnut (*Arachis hypogaea* L.) seed germination.

b. Phosphorus :-

The changes in uptake of phosphorus due to aluminium treatment is recorded in **Fig. 19.**

Both the groundnut cultivar responses similar to Al treatment. The slight increase in phosphorus content was noticed by Al treatment in both cotyledons and embryo axis of cv. W-55 and cv. SB-11, except the marginal reduction in phosphorus content in cotyledons of both varieties by 10 ppm Al treatment.

In soil, Phosphorus is present in both organic and inorganic forms. It is absorbed primarily as $H_2PO_4^-$, a monovalent inorganic phosphate. The organic forms like nucleic acid, phospholipids and inositol phosphates are eventually decomposed. It is converted into an inorganic form which is readily absorbed by plant root system.

The availability of P is controlled by soil pH solution. The dissolved iron and aluminium which precipitate out phosphate as unabsorbable phosphates of iron and aluminium.

The phosphorus ('P') is an important constituent of nucleic acid, phospholipids and nucleotides. It is most important constituent of ATP and other high energy compounds and coenzymes NADP involved in respiration, photosynthesis, nitrogen metabolism, carbohydrate metabolism and fatty acid synthesis.

Phosphorus plays vital role in growth and development of groundnut. The shelling percentage, oil yield and nodulation in groundnut has been increased by phosphorus. On global level 'P' is the most deficient element. The role of Phosphorus is very critical at flowering and pod formation stages of groundnut crop (Singh *et al.*, 1991). The low 'P' availability decreases nodulation and nitrogen fixation rate. The 'P' deficient groundnut plant shows significant decrease in root-shoot ratio (Anuradha *et al.*, 1995).

It has been reported earlier by Rondal and Vose (1963) that 'P' uptake enhanced by Al in sugarcane. In response to aluminium treatment, there is disturbance in phosphorus metabolism, which was brought about by marked decrease in sugar phosphorylation, probably through inhibition of hexokinase (Clarkson and Hanson, 1980). The drastic reduction attributed to presence of positively charged hydrated Al oxides on the cell surfaces that were responsible for absorption, precipitation and further fixation of P, which was not available for plant uptake. (Fageria and Carvalho, 1982).

As early as in 1995 Lorenc-plucinska, determined the free inorganic phosphate level (Pi) and phosphorylation potential (pp) in scots pine seedling roots grown under various concentration of Al (0.5 and 1.0 mM). He observed that, this lower concentration of Al had no effect on both these phosphorus fractions in 3 weeks as compared with higher Al concentration, while after 9 weeks, under 40 mM Al concentration, both Pi level and pp content decreased significantly. It was revealed that inactivation of 'P' in roots by precipitation of aluminium phosphate in and on the roots (Tanaka and Navasero, 1966). Similar finding reported earlier by Fageria and Carvalho, (1982) where 40 and 60 ppm Al in nutrient solution resulted in drastic reduction of 'P' concentration and contents of both stems and roots of rice. Kidd and Proctor (2000). reported slight variation in 'P' uptake in different races of *Betula pendula* growing in Al nutrient solution. Lidon *et al.,* (1999, 2000) and Patel *et al.,* (2002) also reported that excess aluminium in nutrient solution significantly decreased the 'P' uptake in various plant species. However Shkolink, (1980), reported that aluminium did not affect, phosphorylation and amount of sugar esters and nucleotides. Similarly Horst and Goppel, (1986) found no influence of aluminium on phosphorus metabolism.

Recently to Ramachandran *et al.*, (2004) noticed that due to higher concentrations of Al (160 ppm and 320 ppm), the concentration of 'P' in both shoot and root of rice seedlings was significantly reduced. After Ramchandran (2004), according to Macklon and Sim, (2006), the uptake of 'P' affected by concentration of Al much about 100 m mol m^{-3} were toxic. The 'P' and Al taken up into the root, stored in vacuoles of cortex cell. The Al absorbed confined to root while 'p' transported to the shoot.

In contrast to earlier reports, the slight increase in 'P' uptake due to Al treatment might be benefited for nodulation as well as nitrogen fixation in groundnut.

c. Potassium :-

The effect of various concentration of aluminium (10 ppm, 50 ppm and 100 ppm) on uptake of potassium in groundnut seedling germination cultivars SB-11 and W-55 is depicted in **Fig. 20.**

The reduction of 'K' uptake due to 10 and 50 ppm of Al treatment is noticed in embryo axis and cotyledons of cv. W-55. But it is slightly elevated in cotyledon due to higher Al concentration. The similar response was also observed in cv. SB-11,

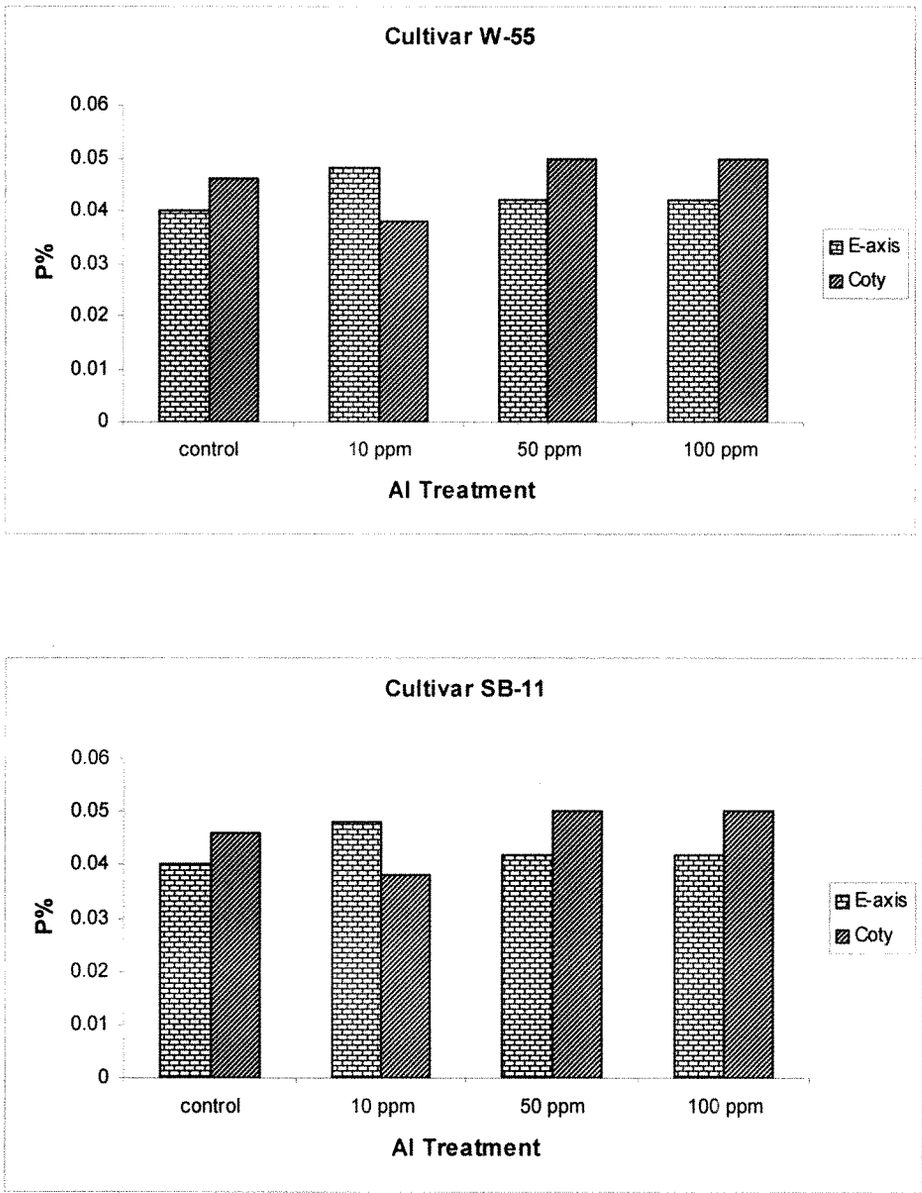
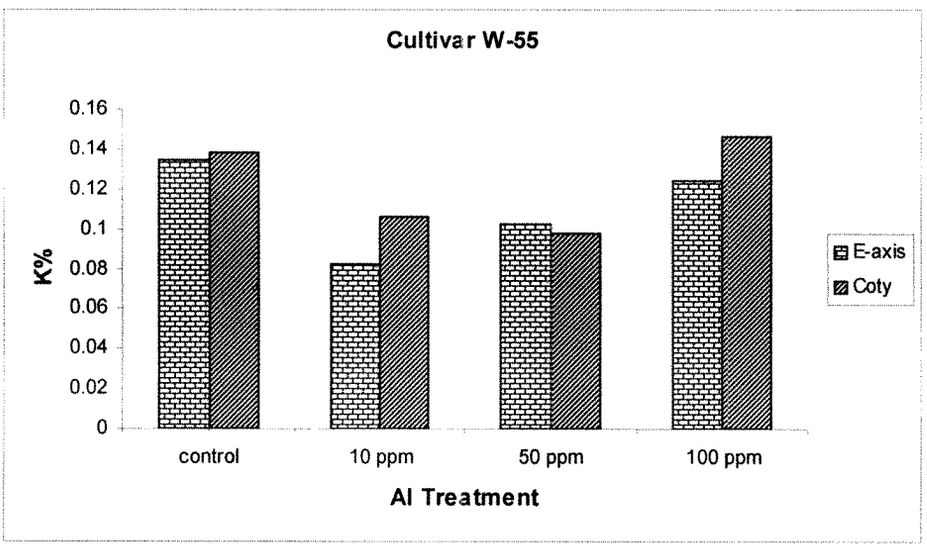


Fig. 19 Effect of Al-treatment on Phosphorus content during groundnut (*Arachis hypogaea* L.) seed germination.



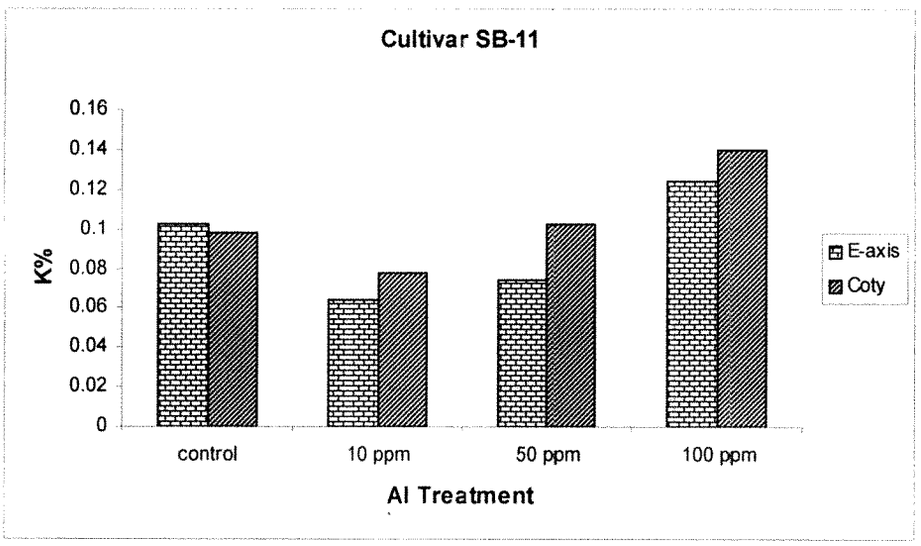


Fig. 20 Effect of Al-treatment on Potassium content during groundnut (*Arachis hypogaea* L.) seed germination.

the only difference is in increase in 'K' uptake due to higher Al treatment in both cotyledon and embryo axis.

Potassium is the most important nutrient, among various nutrients essential for the normal plant growth and metabolism. Mode of action of potassium is not known clearly. It is an activator in protein metabolism. Potassium taking part in the process of photosynthesis and respiration, by acting as counter ion for H^+ ion flux across the thylakoid membrane of chloroplast and mitochondrial membrane (Lauchli and Pfluger, 1978). Lindhauer, (1989) explained the role of K^+ in growth, cell extention and storage of assimilates.

Soils are usually most deficient in potassium. For which commercial mixed fertilizers are practical in crop cultivation. Most of the potassium content of the soil is non exchangeable (fixed) thus it is unavailable to plant. Generally K present in soil in different forms such as,

Soluble $\rightleftharpoons$ Exchangeable $\rightleftharpoons$ Fixed

'K' 'K' 'K'

When 'K' salts are added in soil, the bound ions are released and are replaced by the newly added 'K' ions. The absorption of 'K' by the plants release of exchangeable 'K' which in turn causes the release of bound 'K'.

In groundnut, potassium is essential for growth and development. According to Basha and Rajeswara Rao, (1980) K^+ deficiency influenced rate of translocation, CO_2 fixation, as well as maintenance of water status, stomatal movement of plant. The uptake of potassium is highly selective and closely coupled to metabolic activity (Marschner, 1986). According to Singh (1999) the low level of 'K' may be balance by elevating Ca, Mg level in groundnut.

The effect of aluminium toxicity on K^+ uptake is still not clear. Andrew *et al.*, (1973) showed that aluminium treatment caused increase in potassium content in the eleven legume species namely *Desmodium uncintum, stylosantles humilis, Glycine Wightii, Medicago sativa, M. Scutellata, M. truncatula, Trifolium rueppelliannum, I. Semipilosum, T.repens, Lotononis bainesii, Macroptillium lathyroides.* Also Lee and Pritchard, (1884) noticed that in *Trifolium repens,* Al treatment stimulates K^+ uptake. Jan (1991) reported increase in K^+ uptake due to Al treatment in three rice cultivars. Lidon *et al.,* (1999, 2000) have reported, in maize plant, an enhancement in the K^+ concentration, with 9 to 81 mg L^{-1} of solution Al concentration. Similar finding noticed in wheat seedling grown in Al-treated nutrient solution Patel *et al.,* (2002).

Fageria and Carvalho, (1982) and Tan and Keltjens, (1990) who reported an inhibitory effect of Al on K^+ concentration of rice and sorghum cultivars.There are some reports which indicates that reduction in uptake of K^+ due to Al treatment. Balakumar *et al.*, (1992) observed that in the presence of Al, the uptake of Ca, K, Mg, Mn, P and Fe was highest in rice cv. co-37 and co-31 and in cv. ADT-36, total nutrient uptake was most inhibited. In rice, for root absorption site Al competes with K^+ and reduces the K^+ uptake (Alam, 1983)., Which is correlate with work of Sivasubrannian and Talibudeen, (1971).During pigeonpea seedling germination aluminium treatment reduces the K^+ uptake was noticed by Bhamburdekar, (2002).

According to Ramachandran *et al.*, (2004) the K^+ uptake depends on concentration of Al upto 80 ppm Al concentration, uptake of K^+ is reduced and at 320 ppm Al significantly enhanced the K^+ concentration in rice root, while in shoot as Al concentration increases, K^+ uptake.

It is evident from our results that only higher Al treatment (100 ppm) enhanced 'K' uptake but lower Al concentration adversely affect in 'K' uptake may be balanced by more absorption of Mg.

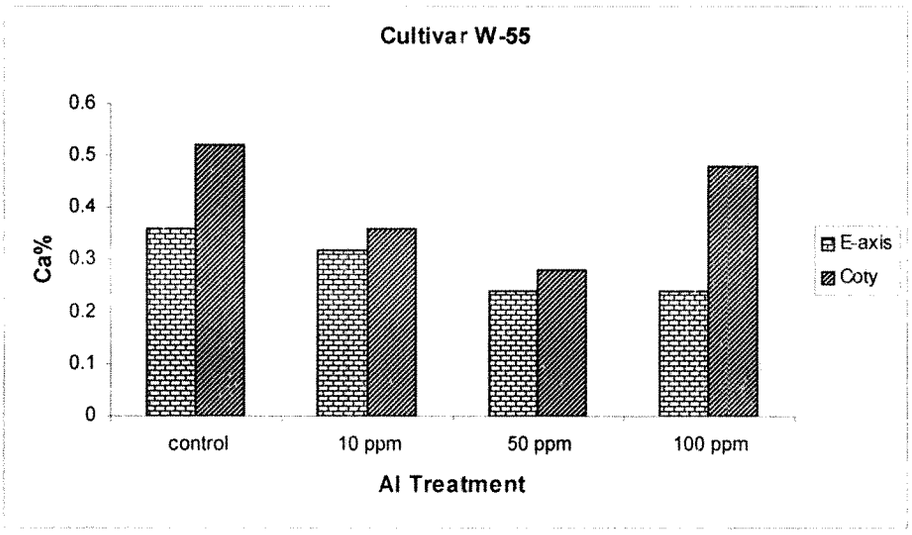
d. Calcium :-

The influence of various concentration of Al on uptake of calcium in groundnut seedlings is depicted in **Fig. 21.**

In cv. W-55, decrease in uptake of calcium with increase in Al concentration is noticed in both cotyledon and embryo axis. However in cv. SB-11, eventhough lower concentration of Al (10 and 50 ppm) decrease Ca uptake both in cotyledon and embryo axis but higher concentration i.e. 100 ppm Al elevated Ca uptake in cotyledons.

Calcium is the most abundant element present in soil. The non exchangeable form of calcium in soil is anorthite ($CaAl_2SiO_3$). Calcium carbonate and insoluble calcium phosphate also found in the soils. It is absorbed as divalent Ca^{2+}

Marschner, in 1986, explain the role of calcium in plants, It is a relatively large divalent mineral element significant for detoxifying higher concentrations of other mineral elements. Almost all plants require Ca, It plays important role such as structural component, membrane and ion regulation, physiological and cytological functions. The Ca^{2+}, calmodulin regulates various enzymes like β–glucans synthetase, NAD Kinase, Ca^{2+} and H^+-ATPase, NAD-oxido-reductase, protein kinase. It



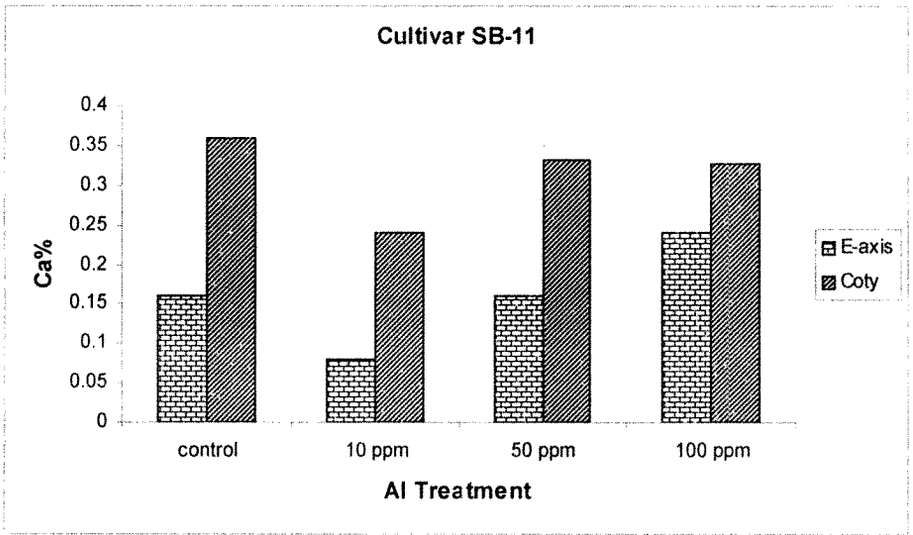


Fig. 21 Effect of Al-treatment on Calcium content during groundnut (*Arachis hypogaea* L.) seed germination.

maintains structural and functional integrity of plant membrane. (Epstein, 1972). Ca affects chromatin organization, enzyme conformation and through calmodulin regulation of many metabolic processes (Clarkon and Hanson, 1980) Calcium increases net absorption of potassium. The important Ca^{2+} modulated protein is calmodulin Ortiz *et al.,* (1994).

According to Singh, (1999) calcium is very important mineral element for groundnut. Often lack of Ca can reduce the yield and quality more than any other element. For gynophore development and pod filling calcium requirement is very high. Calcium and potassium level in the fruiting zone affects seed quality (Cox *et al.,* 1982; NRCG, 1996; Zharare *et al.,* 1997).

The tissue concentration less than 0.7% shows deficiency symptoms of calcium (Dwivedi,1986). Calcium is taken up directly from the soil by pods and inadequate supply results in pods without seeds called pop (Cox and Reid, 1964). Adam *et al.,* (1993) reported the minimum seed Ca is required for maximum seed germination. They further reported that seed Ca correlated with germination of seedling vigour than soil Ca level. The seed Ca concentration required also increases with seed size of groundnut (Cox *et al.,* 1976).

The inhibition of calcium uptake in some legume species such as *Desmodium uncitum, stylosanties humilis, Glycine wightii, Medicago sativa, M. Scutella, M. truncatula, Trifolium rueppellianum, T. semipilosum, T. repens, Lotononis, bainesii, Macroptillium lathyroides* was reported by Andrew *et al.,* (1973). Similar inhibition of Ca uptake was noticed by using kinetic analysis by Clarkson and Sanderson (1971), Guerrier, (1982), Foy (1974). Ownby and Dees, (1985) reported that soil amended with $AlCl_3$ showed complete cessation of growth in groundnut which inhibit uptake and distribution of Ca rather than Al in plant tissue. Due to Al toxicity the foliar symptoms like Ca deficiency was documented by Kumar and Purohit, (2003), it includes curling or rolling of young leaves and collapse of growing points and petioles due to toxic effect of Al.

However stimulatory effect of Ca accumulation due to Al also recorded by Jan (1991) and Lidon *et al.,* (1999). Recently Ramachandran *et. al.,* (2004). noticed increasing solution Al concentration significantly reduced concentration of Ca both root and shoot.

In groundnut, Ca requirements is greater during flowering and pod filling period. Hence marginal reduction in Ca uptake due to Al treatment during seed germination cannot impose a major limitation for growth and development.

e. Magnesium :-

The effect of Aluminium stress on Mg content is depicted in **Fig. 22.**

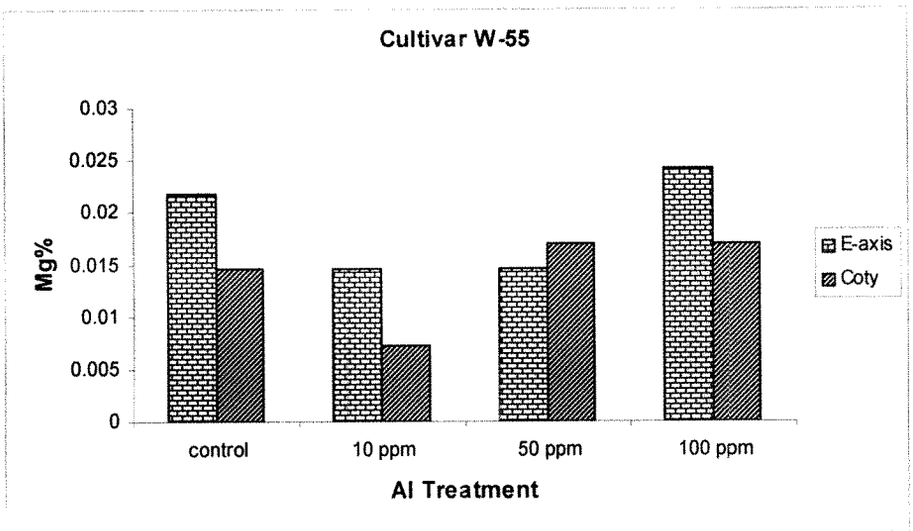
In the cotyledons of cv. SB-11, the reduction in Magnesium uptake was recorded by concentration of Al and it is more significant with 50 and 100 ppm Al, while in Embryo axis, Mg uptake is increased by 10 ppm of Al. Magnesium uptake is reduced in embryo axis of cv. W-55 due to lower Al concentration, (10 ppm) treatment, but it is increased by higher concentration of Al (100 ppm). However in cotyledons, initial reduction in Mg uptake due to low Al treatment recover by 50 and 100 ppm concentration of Al treatment.

Mg is most important mobile strongly electropositive and exchangeable divalent cation. It is present abundantly in soil in the form of magnesium silicate. It is an unavailable form becomes available after weathering to the plants.

Magnesium is a component of chlorophyll and it serves as a cofactor in most of the enzymes that activate phosphorylation process as a bridge between pyrophosphate structures of ATP or ADP and the enzymes molecules. Several enzymes involved in carbohydrate metabolism require Magnesium as an activator in which ATP is involved. However various factors such as Mn^{2+}, K^{+}, NH_4^{+} and even low pH affects the uptake of Mg. The activity of CO_2 fixing enzymes ,RuBP carboxylase, and PEP carboxylase requires Mg^{2+}. Magnesium also increases activity of enzyme glutamate synthetase, O' Neal and joy, (1974).

Groundnut plant require high amount of Magnesium after 30 days of plant growth. According to Harris, (1949), the first symptom of Magnesium deficiency is interveinal chlorosis at the terminal leaves and stunting plant. Plants completely lose their green colour and die. (Bledose and Harris 1950, and Reid and York, 1958).

Videl and Broyer, (1962) reported reduction in Mg uptake. in Maize due to toxicity of Al. Alam (1983) noticed in rice plant parts, that as Al level increasing from 1-10 ppm reduced uptake and concentration of Mg. Thrornton *et. al.*, (1986) reported that lower concentration of Al elevated Mg content while higher level reduced Mg level in *Acer*. The possible explanation for reduction of Mg due to Al toxicity was given by Shkolink, (1984) according to him, Al absorbed by cell walls may strongly



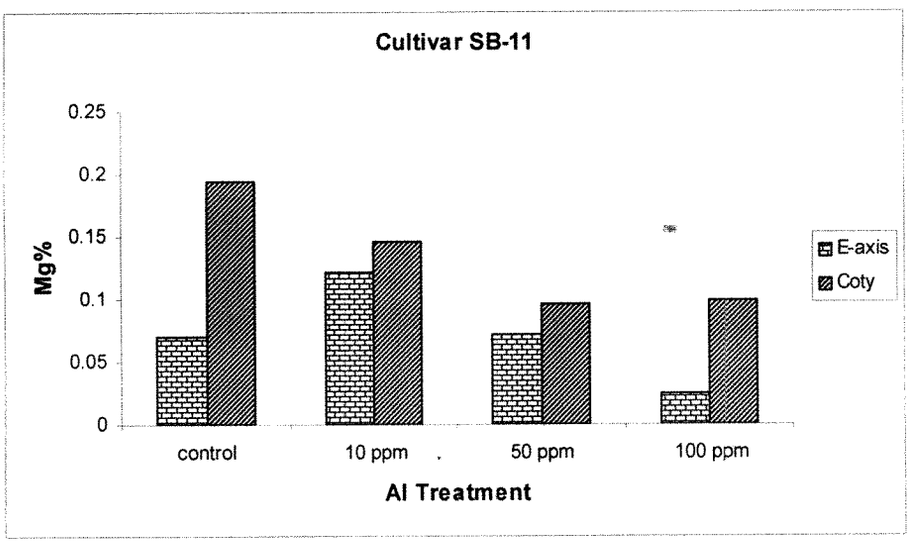


Fig. 22 Effect of Al-treatment on Magnesium content during groundnut (*Arachis hypogaea* L.) seed germination.

inhibit the active sites of enzyme situated in the cell walls, thereby significantly impairing the uptake of nutrients and active transport. Another possible explanation for reduction in uptake of Mg due to Al was given by Fageria and Carvalho, (1982). According to them, there is competition of Al with Ca and Mg for common binding sites at or near the root surface which is the reason for reduction in uptake of these elements. Similar reducing trend was also noticed by Bhamburdekar, (2002), in pigeonpea seeds. Recently Ramachadran *et. al.*, (2004) noticed that the increasing Al concentrations, causes continuous gradual and significant reduction in Mg uptake in rice root.

In agreement with these findings ,in the present investigation, Mg uptake is reduced due to Al treatment in cotyledons of both cultivars.

f. Iron :-

The effect of aluminium treatment on uptake of Iron in two cultivars of groundnut during seed germination is depicted in **Fig. 23.**

The concentration of Fe in the embryo axis of cv. W-55, increased with respect to 50 ppm Al treatment only, but the Al has no effect on Fe uptake in cotyledons. However Fe uptake it of Al treatment in both cotyledons and embryo axis of cv. SB-11.

Generally soils are not deficient in iron, but they may be deficient in soluble forms of iron. The availability of iron to the plant is controlled by soil pH. Generally acidic soils favors availability of soluble iron forms than other types of soils such as neutral or alkaline soil.

In plant metabolism iron performs a number of important functions in the overall plant metabolism. It is very important because it forms prosthetic group of many enzyme system. Iron porphyrin (hemes) are most studied iron containing prospthetic group. Iron is constitutent of hemeproteins like cytochrome and plays major role in photosynthesis and respiration through electron transfer system. The number of reactions which are carried out by enzymes like catalase, peroxidase, cytochrome oxidase are activated by Fe content. Cell division of plants is also affected by Iron.

In the mechanism of nitrogen fixation in root nodules of leguminous plants, the leghemoglobin iron containing protein is involved. Heme pigments constitute only 0.1% of total iron in plant leaves (Dekock *et al.*, 1960) while remaining Iron is

localized in the chloroplast of green leaves, (Marschner, 1986). In biosynthesis of chlorophyll, Iron plays important role.

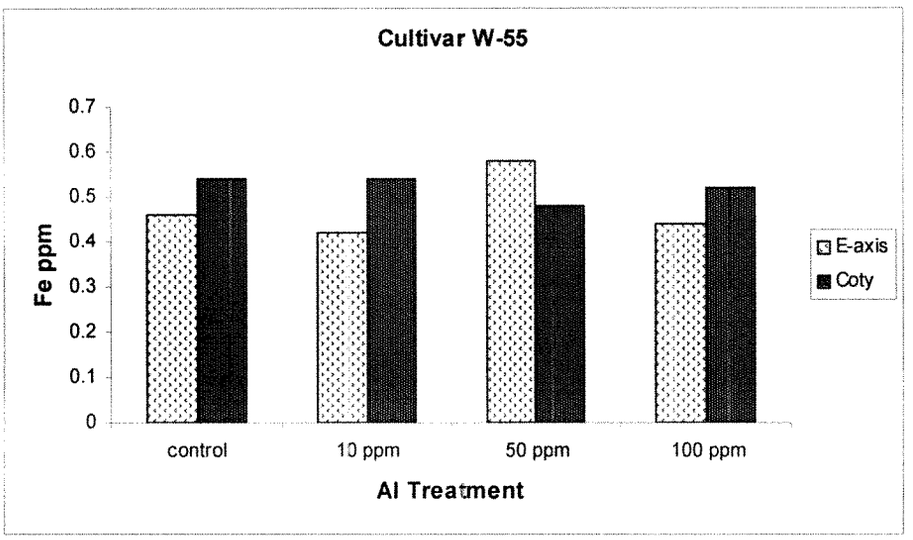
The high free $CaCO_3$, HCO_3, moisture, heavy metals, pH and available P, poor aeration, heavy manuring and low organic matter content in acid soils and damage root enhance iron deficiency in groundnut (Singh 1994a., Singh *et al,*. 1991a,b., Wallace *et al.*,1976).

The iron deficiency is visible in groundnut. The chlorosis of young rapidly expanding leaves, interveinal chlorosis, and in severe deficiency leaves become white papery. (Singh, 1994a., Singh *et al.,* 1991a,b, Singh and Dayal, 1992). The root peroxidase activity was identified as indicator of iron deficiency which found to be decreased in their root stem and leaves (Singh and Chaudhari, 1992). The Fe deficiency also limits nodule development in groundnut grown in calcarious soil (O' Hara *et al.,* 1988).

Alam, (1983) reported the accumulation of iron in roots of rice with increasing Al concentration upto 10 ppm, but further addition of even 2 ppm Al to nutrient medium decrease Fe content. The reduction in 'Fe' content in leaves and roots of *Zea mays* under various Al treatments was noticed by Gerezabek and Edelbaucer, (1986).

The solubility and availability of Fe increases through hydrolysis of Al in calcarious soils was noticed by Foy, (1984). Roy *et al.,* (1988) reported that, Al interfaces with reduction of Fe^{3+} and Fe^{2+} which is vital reaction of iron metabolism in oat plant. Jan, (1991) reported, that there is indefinite correlation between Al treatment and Fe content. The inhibition of Fe uptake due to Al treatment was noticed by Patel *et al.,* (2002). Lidon *et al.,* (1999) noticed in Maize shoots, that concentration of N, P, and Fe decreased significantly between the O and the 81 Mg L^{-1} Al treatment. Similar observation also by Ramachandran *et. al,*. (2004), in rice root, the concentration of Al increased and uptake of iron decrease. This reduction in Fe content attributed to the reduction of cellure respiration in plants by Al causing an inhibition in the uptake of micronutrients.

In our investigation no definite trend is observed. However enhancement of Fe content in embryo axis of both cultivars due to 50 ppm Al treatment might be an adaptive feature for groundnut metabolism, as according to Singh, (1999) the application of Fe reduces incidence of tikka and rust diseases.



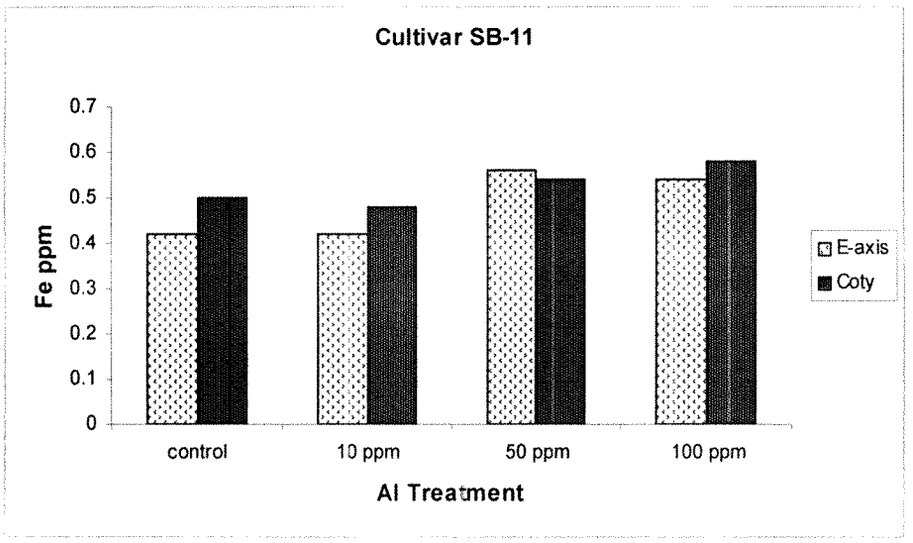


Fig. 23 Effect of Al-treatment on Iron content during groundnut (*Arachis hypogaea* L.) seed germination.

g. Manganese :-

The effect of aluminium treatment on uptake of manganese (Mn) during groundnut seed germination is depicted in **Fig. 24.**

In cv. SB-11, the marginal decrease in manganese uptake in cotyledons is recorded by 10ppm and 50ppm of Al treatment. But 100 ppm Al, uptake of Mn slightly enhanced. However in embryo axis, increased concentration of Al stimulates Manganese uptake. In embryo axis of cv.W-55, Mn uptake is reduced except at 50 ppm Al treatment,but continuous reduction in Mn content is noticed in cotyledons with all Al concentrations employed.

Manganese is one of the essential micronutrients exist in various forms in soil , But in soil it is present in insoluble compounds in tri and tetravalent forms and thus it is largely unavailable to the plant. In poorly aerated acid soils,this tri or tetravalent forms are reduced to form bivalent form,Thus it is available to plants. It is mainly absored as bivalent and exists in various forms in soil as bivalent ion Mn^{2+}. It is translocated predominantly as the free divalent cation in the xylem from the roots to the shoot (Graham, 1979).

For many enzymes Manganese acts as an activator. In all plants for the Hill reaction Manganese is required (Cheniae and Martin, 1986). Manganese is activator of many enzymes of Kreb cycle. Ness and Wollhouse, in 1980 reported that Manganese activates RNA polymerase. It is structural constituent of ribosomes (Lyttleton, 1960). Because of lowest complex stability constant, it forms weakest bond. Due to which it can replace magnesium in many reactions such as phosphokinase, and Phosphotransferases. (Clarkson and Hanson, 1980). The enzyme NADP – malic enzyme, PEP carboxylase, and NAD-malic enzyme are manganese specific enzyme. There is variation in different crop species in their Manganese requirement .According to Stout(1961),0.005% of Manganese is essential for growth and metabolism of multicellular plants.

Martin, (1959) has noted Manganese toxicity in groundnut grown on acid soils. The availability of Mn increases to a toxic level at low soil pH i.e. in acid laterite soil. Dickert and Rozacky, (1969), isolated, Manganin–Mangano protein, from groundnut seeds. Reid and Cox, (1973) reported that for groundnut plants the adequate manganese concentration of leaf tissue ranges between 20 to 40 ppm. While above 200 ppm, Mn toxicity is observed. Mn deficiency leads to interveinal chlorosis and necrotic spots. However, vein tend to remain green.

Lee, (1971) reported in potato roots that due to Aluminium, Mn is accumulated. Alam, (1983) noticed , decline in Mn content in all parts except stem in barely which were treated with aluminium. Same observations were also recorded in rice. The decline in Mn uptake due to Al concentration in Maize, was noticed by Mathan, (1980) Cambraia, *et. al.*, (1983) reported, that a very low Al concentration can also shows reduction in Mn uptake in roots and tops in Jawar. Lidon *et. al.*. (1999) noticed, Manganese concentrations significantly decrease upto 79% due to 0 and 9 Mg L^{-1} of Al treatment in maize shoot. Patel *et al.*, (2002), has been reported, inhibitory effect of Al on Cu, Mn, Fe, Zn content in wheat seedlings. Recently Ramchandran *et. al.*, (2004) observed reduction in uptake of Mn with increasing concentration of Al in rice roots, could be related to the reduction of cellular respiration in plants. These evidences reported the role of Mn influences the level of auxin in plant tissue.

In present investigation cv. SB-11 show better response to Al treatment by increasing uptake of Mn for catalyzing many metabolic processes at germination stage than cv. W-55.

h. Zinc :-

The effect of aluminium toxicity on uptake of Zinc during groundnut seed germination is depicted in **Fig. 25.**

It can be seen that in cv. SB-11, zinc uptake is marginally decreased due to Al toxicity in embryo axis. But opposite trend was noticed in cotyledon. While cv. W-55, higher concentration of Al decreased uptake of zinc in both embryo axis and cotyledons, in contrast to slight increase in Zn uptake by 10 ppm Al in embryo axis.

Zinc is micronutrient. The sources of Zn in the soil are ferromagnesium minerals, magnetite and biotite. Zn is released from these minerals and it is absored as divalent Zn^{2+}. Soil pH in the main factor for availability of zinc. It acts as an activator of several enzymes including alcoholic dehydrogenase pyridine nucleotide dehydrogenase, and carbonic anhydrase. The enzyme carbonic anhydrase is a acts as an activator for some phosphate transferring enzymes like phosphodiestrase (Kumar and Purohit, 2001).

The Zn deficiency in groundnut occurs mainly in the uppear leaves showing chlorosis, reduction in size of young leaves. Under severe deficiency, the entire leaflet become chlorotic.

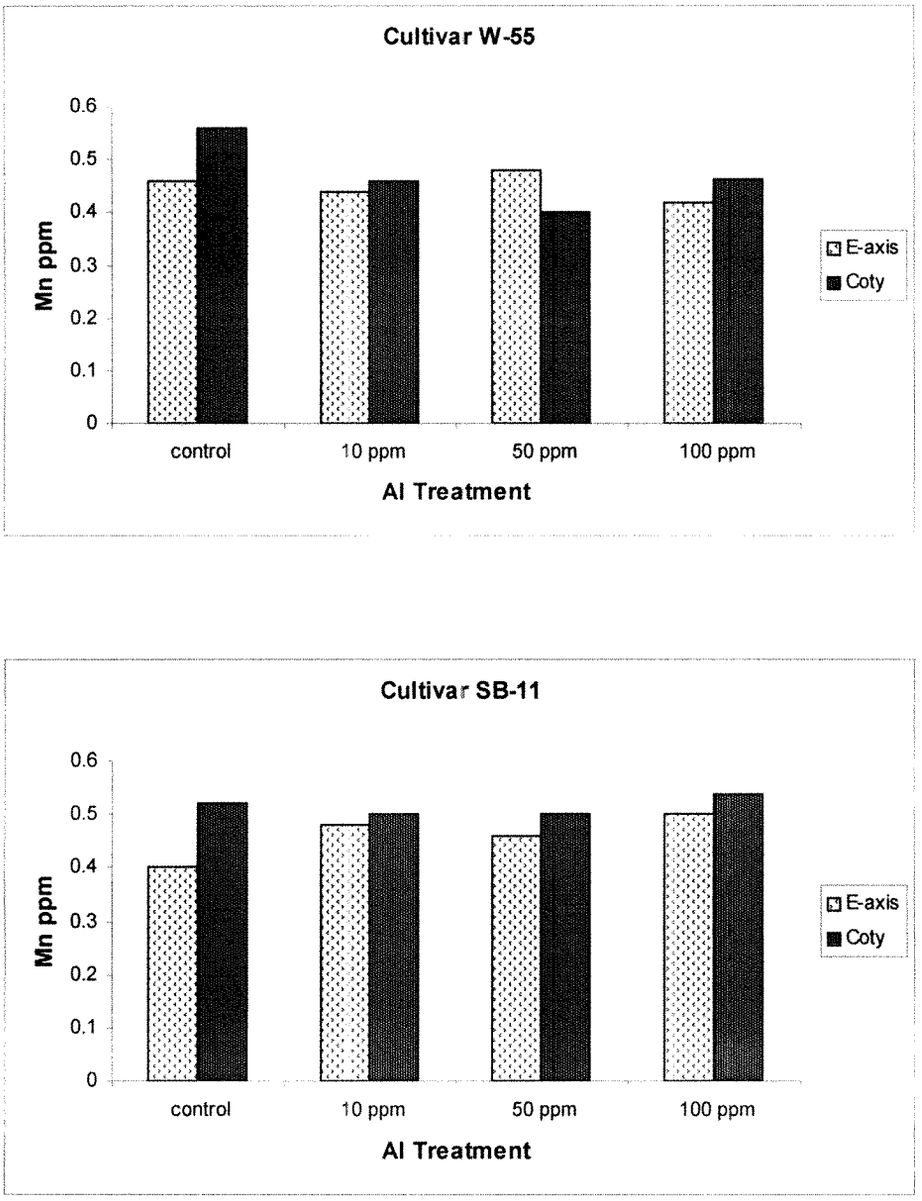


Fig. 24 Effect of Al-treatment on Manganese content during groundnut (*Arachis hypogaea* L.) seed germination.

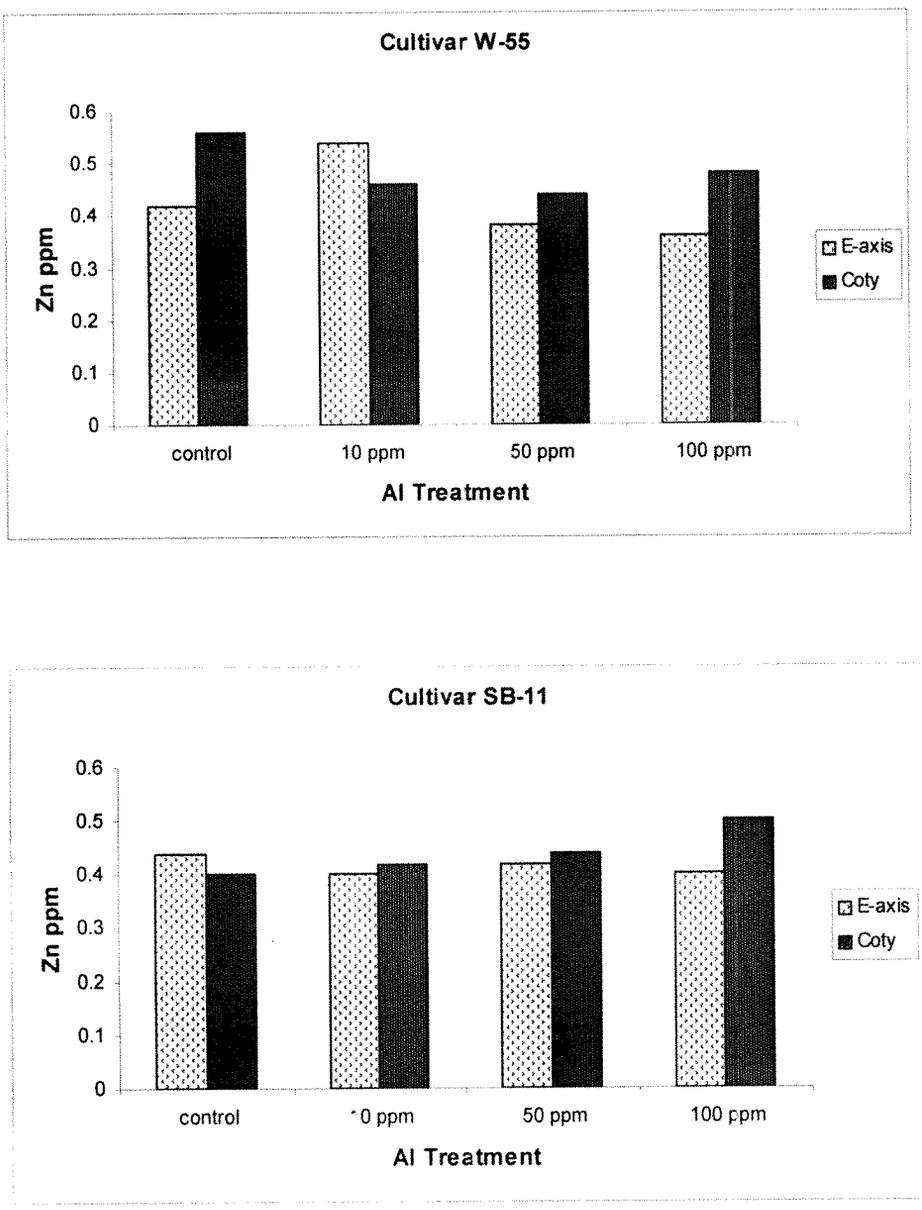


Fig. 25 Effect of Al-treatment on Zinc content during groundnut (*Arachis hypogaea* L.) seed germination.

According to Dwivedi, (1986); Singh, (1994b), the critical level of Zn in groundnut reported to be 0.7 ppm. The leaf tissue less than 20ppm show zinc deficiency. However Zn toxicity recorded above 200 ppm affects growth.

Simon *et al.*,(1994) shows in plants of tomato green house cultivators *Mountain pride* and *Florameric,* that Al treatment reduces zinc in root, stem, leaf content. However according to Lidon *et al.,* (1999) a clear tendancy was not observed concering the Zn content in *maize* shoot, when the concentrations of aluminium increases from, 0 to 81 mg L^{-1} Al. Ramachandran *et al.,* (2004) reported the concentration of Zn in rice root was significantly reduced with increasing Al concentration of nutrient solution. The observed inhibitory effect of Al on different plant micronutrient including Zn has been recorded in wheat seedlings by Patel *et al.*, (2002).

In present investigation, decreasing uptake of zinc with increasing concentration of Al is noticed in embryo axis of cv. SB-11 and cotyledons of cv. W-55. could be attributed the reduction of cellular respiration in plants by Al causing an inhibition in uptake of micronutrients (Fageria and Carvalho, 1982).

i. Molybdenum :-

The effect of aluminium toxicity on uptake of Mo during groundnut seed germination is recorded in **Fig. 26.**

In both cv. SB-11 and W-55, it has been seen, that Al toxicity inhibits the Mo content in embryo axis. This inhibition is maximum with lower concentration of Al than higher Al (100 ppm) concentration. However inhibition on Mo content in the cotyledons both cultivars restricted to only lower doses of Al, but higher (100 ppm) Al concentration, stimulate Mo uptake and it is noticeable with cv. SB-11.

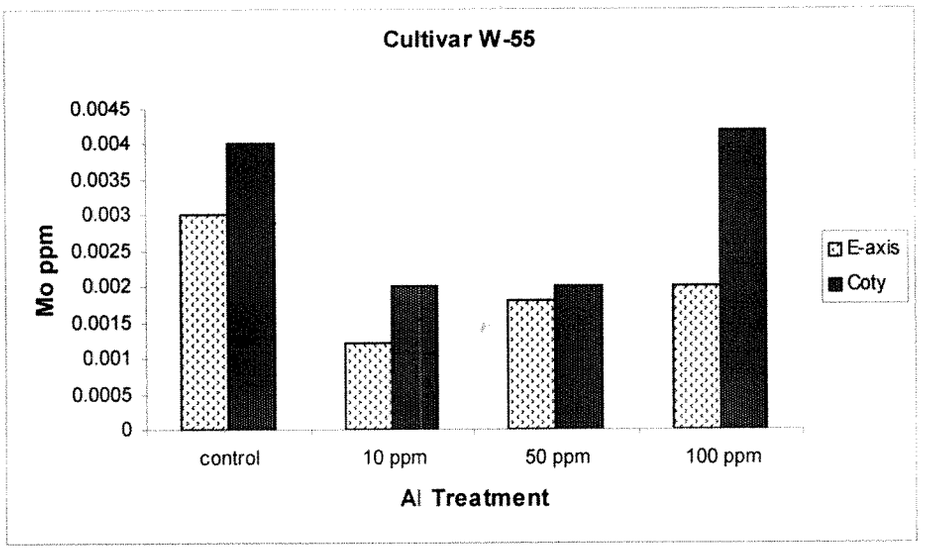
Molybdenum is present as dissolve ions forms in soil. It is an exchangeable from absorbed to soil. It is an exchangeable from absorbed to soil particles while in plants absorbed it as an non-exchangeable form as MoO_4^{2-}. However Mo is soil incentive to the positive effect of that pH elevation. Molybdenum is required for the functioning of several complex enzyme systems involved in nitrogen metabolism in plants. In nodulated legumes, Mo is necessary for the reduction of atmospheric nitrogen (N_2) to ammonia by nitrogenase. The critical limits of Mo was found to be 0.04 ppm in soil and 0.2 ppm in leaves (Singh, 1994b). However, the Mo concentration above 1 ppm in soil was toxic to groundnut plant. In acid soils Mo

deficiency has been reported by Singh, (1999). The adequacy of Mo for plant growth is determined by a number of soil and plant factors. Acid soils containing appreciable amounts of non-crystalline oxides and hydroxide of Fe and Al frequently retain Mo in non-plant available state.

In groundnut, Molybdenum play essential role, nitrogen fixation and involved in several enzyme system. The disturbance in nitrogen metabolism is the symptom of Mo deficiency. first symptoms appears in older leaves and progress towards the younger until plant die. The reduction in chlorophyll content due to which leaves becomes bright, yellow, green inter veinal chlorotic mottling and at later stages leaf margin curled and it collapse completely. The reduction in number of nodules in Mo–deficient plant was reported by Singh and Choudhari, (1992, 1993b)

The literature on the influence of aluminium treatment on Mo content is very scanty. Simon *et al.*, (1994) when working with tomato plant, they reported that by addition of Al, the uptake of Mo is reduced in both, tomato cultivars viz. *Mountain pride* and *Florámerica*, grown in green house they further noticed that low concentration of Al promotes accumulation of Mo in roots and inhibited its transport it into stem and leaves.

In present investigation, both cultivars of groundnut differ in their response of Mo uptake with Al treatment. The increase in Mo in cotyledons by higher Al treatment may be correlated with increasing nitrogen demand by elevating nitrogen fixation activity of growing groundnut seeds.



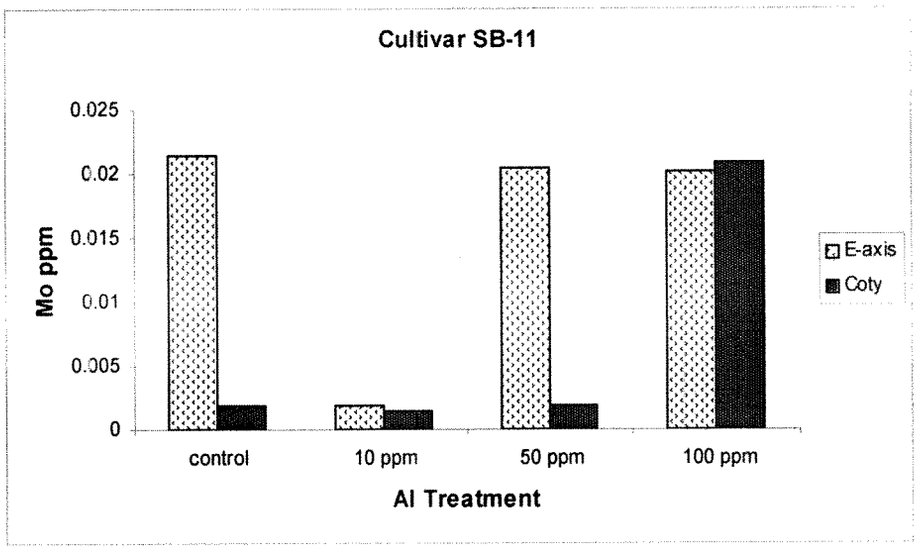


Fig. 26 Effect of Al-treatment on Molybdenum content during groundnut (*Arachis hypogaea* L.) seed germination.