Physiological Studies on Groundnut in Relation to Potassium Nutrition Under Drought Conditions

SHAHID UMAR

Master of Philosophy
IN
BOTANY

ALIGARH MUSLIM UNIVERSITY
ALIGARH
1986
ACKNOWLEDGEMENT

I wish to express, with a deep sense of gratitude, my indebtedness to Professor M.M.R.K. Afridi, Department of Botany, Aligarh Muslim University, Aligarh for his able guidance, keen interest and constant encouragement. I am also deeply thankful to Dr. Ram Snehi Dwivedi for suggesting the problem and for his able supervision, valuable criticism, keen interest and helpful suggestions throughout the course of the preparation of this dissertation.

I place on record my sincere thanks to Dr. Samiullah, Reader, Department of Botany, Aligarh Muslim University, Aligarh, for his advice, encouragement and constructive criticism during the preparation of the manuscript.

My sincere thanks are due to the Director, National Research Centre for Groundnut (ICAR), Junagadh (Gujarat) and to the Chairman, Department of Botany, Aligarh Muslim University, Aligarh, for providing the required facilities.

Thanks are also due to Mr. C.C. Joshi, Dr. C. Nautiyal, Dr. V. Ravindra, Dr. A.L. Singh, Dr. S.K. Suneja, Mr. V.S. Neqi, Mr. J.S. Dhapwali, Mr. A.N. Thakker and Mr. V.G. Koradia of Junagadh and to Dr. S.H. Afzal, Dr. Aqil Ahmad, Dr. Arif Iman, Dr. M. Aslam Darvaz, Dr. Firoz Mohammad, Dr. Ali Qaddar,
Dr. Masood Akhtar, Dr. M.M.A Khan, Dr. Shamim A. Ansari, Dr. Shadab A. Khan, Messrs Moinuddin, Nafees A. Khan, Faizan A. Khan, Mujahid A. Khan, Ikramul Haque, S.R.M. Ata and Miss Atiya K. Zaidi, Miss Nasreen Fatima and Miss S. Shama Muzaffar for their help and co-operation.

Special thanks are due to my parents, brothers, sister and my wife Shahin Umar for their help and encouragement at every step.

Finally, I am thankful to Dr. G.S. Sekhon, Director, Potash Research Institute of India, Gurgaon (Haryana), for granting Senior Research Fellowship.

(SHAHID UMAR)
## CONTENTS

<table>
<thead>
<tr>
<th>Section</th>
<th>Pages</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Introduction</td>
<td>1-4</td>
</tr>
<tr>
<td>2. Review of the Literature</td>
<td>5-47</td>
</tr>
<tr>
<td>3. Material and Methods</td>
<td>48-73</td>
</tr>
<tr>
<td>4. References</td>
<td>1-xxiv</td>
</tr>
</tbody>
</table>
CHAPTER I

INTRODUCTION
INTRODUCTION

Groundnut (syn. peanut, i.e. *Arachis hypogaea* L.), a member of Papilionaceae, belongs originally to South America. But easy adaptability of the groundnut helped it in spreading quickly in most parts of the world within a short span of about 150 years. It is the only cultivated plant that exhibits geotropic reproductive growth and geocarpic fruiting. Besides, the groundnut contains about 25% protein and 50% oil. The groundnut oil consists of mostly unsaturated fatty acids, particularly linoleic acid, which forms the bulk, linolenic acid and arachidonic acid, which are essential in human diet. These fatty acids make groundnut oil a balanced edible oil.

India is the largest producer of groundnut in the world. It accounts for about 39% and 37% of world groundnut cultivated area and production respectively. China and U.S.A. occupy second and third positions, contributing 15% and 10% respectively of the total world production. In addition, groundnut enjoys prominence among major edible oil yielding crops in India. It alone claims the largest share in area of cultivation (46%), production (67%) and edible oil production (59%).

Despite of continuous increase in cultivation area under groundnut, i.e. from 0.35 million hectares in 1910-11
to 7.4 million hectares in 1984-85, productivity of this crop has slumped from 13.4 g/ha (0.47 million tonnes) in 1910-11 to 9.5 g/ha (7.0 million tonnes) in 1984-85 which is far lower than 29.4 g/ha in U.S.A. This is mainly because of the fact that our groundnut is mostly cultivated under rainfed conditions. It is noted that failure in rainfall or its improper distribution, resulting in short to prolonged drought conditions, reduces pod yield to an extent of 25% to 100% and thus makes the groundnut an "unpredictable legume" (Misra, 1986).

This creates a paradoxical situation as the demand of edible oil far exceeds its production. The "population explosion" in India further disturbs the balance year after year. To bridge the gap between demand and production, India has no alternative but to import edible oil annually worth Rs. 1,000 crores, which drains away considerable foreign exchange and becomes a heavy burden on the economy. In this context, any major improvement in the production of groundnut, being cultivated on a large area with its contribution of the major share in edible oil production, is expected to provide an answer. As mentioned earlier groundnut is mostly grown under rainfed conditions where frequent droughts prevail. An understanding of its stress-agronomy coupled with judicious application of mineral nutrients may help improve the
productivity of the crop. Unfortunately, such studies have so far excluded groundnut.

It has been noted that, among various nutrients, potassium works efficiently in drought conditions and promotes a number of physiological activities in plants that tend to improve their drought tolerance. For example, maize roots penetrate 60 cm deeper when receiving potassium fertiliser, getting access to an extra 10 cm of water (Edward, 1981). Potassium activates a number of key enzyme systems (Evans and Sorger, 1966). It also lowers osmotic potential of root cells and increases water uptake (Mengel and Kirkby, 1980). It plays a specific role in most plant species in opening and closing of stomata and checks transpiration (Mengel and Kirkby, 1982). Besides, potassium has many biochemical and biophysical functions in photosynthesis such as partitioning and storage of assimilates affecting the "source" more than the "sink" (Beringer and Trolldenier, 1978). Accordingly, during vegetative growth when the productive system (leaf area) and the foundation for yield components and yield are built up, potassium requirement of plants becomes high and indispensable (Beringer and Haeder, 1981).

It is, therefore, assumed that if some of these ameliorating effects of potassium can be induced in groundnut
growing under drought conditions, it will not only help
utilise marginal land for groundnut cultivation but also
improve the productivity of the crop and this helps cater for
ever-increasing the needs of millions of people. Keeping
these facts in view, five field trials on groundnut are proposed
to be conducted in relation to different moisture regimes
and potassium nutrition at Junagadh (Gujarat). It may be
reiterated that Gujarat is one of the top contributing State
as far as the cultivation and production of groundnut is
concerned. The aims and objects of these trials will be
as follows:

1. To find out the optimum potassium requirements for
the growth and development of selected varieties of
groundnut under (i) rainfed and (ii) irrigated
conditions.

2. To determine the efficacy of split doses of potassium
applied at different stages of growth and development
of the crop.

3. To study the role of potassium in developing tolerance
in groundnut against drought stress.

4. To select the most efficient method of applying
potassium (basal and/or top dressing) so as to ensure
drought tolerance in groundnut leading to improved
productivity of the crop.
CHAPTER II

REVIEW OF THE LITERATURE
CONTENTS

2.1 Growth and development of groundnut

2.2 Physiological aspect of drought

2.3 Effect of drought on growth and yield components of groundnut

2.4 Physiological role of potassium

2.5 Water use efficiency in relation to potassium nutrition

2.6 Nitrate reductase activity

2.7 Proline content

2.8 Chlorophyll content

2.9 Energy harvest efficiency

2.10 Potassium nutrition of groundnut

2.11 Performance of other crops in relation to potassium nutrition

2.12 Conclusions
It is generally agreed that yielding ability of a crop depends on its vegetative and reproductive growth. The aim of all crop scientists is to maximise these factors through genetic variability and well understood agronomy. Of these, curtailment of the growing period of a crop to a manageable extent is of paramount importance. It is particularly true for groundnut as plant breeders have changed its habit from perennial to annual by rigorous genetic manipulation, so that groundnut can now be harvested within 3-6 months and is tetraploid (2n=46). The species "hypojaea" includes two sub-species described briefly below:

Sub species hypojaea: This includes Virginia, runner (spreading) and erect (bunch) types. These are characterised by the absence of inflorescence on main stem and bear fruiting body in alternate pairs on the n + 1 branches.

Sub species festigiata: This includes Valencia and Spanish types. They bear flowers both on main stem and branches. Branching is segmented (Gregory et al., 1951; Krapovikas, 1968).
The literature in relation to the performance of this crop, covering various agroclimatic conditions, is briefly reviewed below:

Ali et al. (1932) reported that there was no marked difference between growth pattern of the two types of cultivars during seedling stage. Conversely, bunch variety produced less leaves than spreading variety during the entire span of life, which ranged from 93-112 and 206-346, respectively. Similarly, vegetative growth period also varied for these varieties. The period of maximum growth was between 56-97 days in the case of bunch type and 70-125 days in case of spreading type.

Rao (1936) recorded periodical increase in the shoot weight of a bunch variety and two spreading varieties. The periodical gain in weight was higher for the variety 'Salmun' and low for the varieties 'local Mauritius' and 'Judiyathan Bunch.' Upto 49-54 days after sowing, the increase in plant weight was rapid, thereafter (particularly towards maturity) the increase gradually tapered off. The rate of increase in weight was more rapid during the first 30 and 35 days after sowing and there was a gradual decrease in the rate of increase as the plants grew older.
Prevot (1949) in his elaborate studies noted that there were two phases of growth in groundnut. He demarcated them (i) as slow growth at the time of flower-initiation and gynophore formation, under the influence of internal factors, and (ii) spurred growth accompanied by the increase intake of nutrition through gynophores.

Smith (1951) reported that the percentage of fertilised flowers was very low. It ranged from 4.9 to 50.0 in spreading and 21.9 to 67.5 in bunch types. Even after fertilisation only about 59.5% of the potential flowers elongated as peg. There was a general agreement that a greater per cent of early formed flowers developed into pods (Gregory et al. 1951, Shear and Miller, 1955).

Aunting and Anderson (1960) reported that experiments conducted at Kongwa Experimental Station, Tanjanjika, under rainfed conditions on red loam soil, indicated that dry matter production per plant in variety Natal Common (a bunch type) increased linearly from 0.23 g per plant on 8th day to 32.74 g per plant on 105th day. It was also observed that nearly 45-50 per cent of total dry matter produced in a plant was accumulated in seeds as against 16, 20, 11 and 1 per cent in shells, stem, leaves and roots respectively.

Dornaraj (1962) reported that flowering sequence showed a rapid increase immediately after commencement and reached
the peak in about a week, followed by a sudden decline. A lower rate of flower production was maintained subsequently for about 10 days. A second spell of increased flower production, of less intensity than the first then occurred and finally, there was a gradual decrease until cessation after 75 days.

Shashadri (1967) discussed details of varietal differences with regard to growth and development. He concluded that the attributes of different varieties were influenced by seasonal changes and other environmental factors.

According to Forestier (1963), various runner type varieties differed with regard to the rate of coverage of canopy. A very high co-relation was also found between the length of the cotyledonary laterals and coverage, showing the importance of these cotyledonary laterals in the skeleton of plant and canopy structure. In bunch types also, varietal differences were noted in leaf area index (LAI) in relation to the rate and time taken to reach the peak. The LAI values ranged from 3.2 to 5.0. A regular increase in leaf area and dry matter per plant from the 3rd leaf stage to peg formation was observed. The increase was not affected by onset of flowering. Maximum LAI was 4.0.

Maeda (1970) reported that in the varieties of groundnut studied by him, the total number of leaves per plant showed
an exponential increase from about 3 weeks after sowing which lasted till 90-100 after sowing. Similarly, the total leaf area per plant also varied. The leaves of the erect type were larger than those of spreading types.

Bhan (1973) noted faster growth rate on higher dry matter accumulation at early stage in erect varieties than in the spreading one, but growth continued for longer period in the spreading type. Forrestier (1973) concluded that for maximum yield the leaf area index at the 14th leaf stage should be more than 4, the total plant dry matter should be 500 g per m² and leaf dry matter should be 175 g per m².

ReCloud (1974) studied LAI and dry matter of Fordrunner variety under two levels of population, i.e. 7.5 and 10.6 plants per m². LAI calculated at 64th day was 3.0, whereas, at 87th day, when flowering was almost over it was 7.1. It decreased rapidly to 1.7 at harvest (137 days). It was also substantiated that at high population, there were 30 pods and 15 unfilled pods per plant at harvest, giving a potential yield of 6.3 t seeds/ha. He concluded that yield was not limited by the size of the photosynthetic sink, but probably by the low leaf area and less efficient leaves during the final filling period.

According to Banyi (1975), early cessation of dry matter accumulation in stem might be considered a desirable feature
for the crop. The accumulation of dry matter in leaf blade during the early stage also contributed towards the yield of the crop. Dry matter accumulation in the vegetative part and in the fruit occurred simultaneously and, since, the vegetative and reproductive phases overlapped each other, quick cessation of vegetative growth (increase in stem dry weight) might result in the availability of photosynthates for accumulation in the economically useful parts. He defined seed yield per given area of land in groundnut crops as the product of pod number, number of seeds per pod and size of their individual seed. He considered that the seed yield in groundnut also depended on the number of pegs formed and on the proportion of pegs that produced mature pods at harvest (pod to pod ratio).

William et al. (1975a) in Rhodesia, the early growth of cultivar Makalu Red, the alternate branching type, was slower than in the other three sequential types, which were similar to the former until about 12 weeks after sowing. After 12 weeks, Valencia R1 (sequential type) accumulated the highest dry matter and Natal Common, the lowest. The other sequential cultivar (59/66) accumulated similar amounts of dry matter as Makalu Red. The accumulation of stem mass in the three sequential cultivars was equal until about 14 weeks. Thereafter, cultivar 59/66 continued to grow rapidly while the rate
of stem growth in the other two cultivars was reduced. The differences with regard to the rate of accumulations of dry matter in these varieties continued till the end whereas growth of stem in Natal Common ceased rapidly.

William et al. (1975b) observed that shellednut yield was not related to leaf area or crop growth rate, but was more dependent on the number of seeds developed. Generally, the time taken for flower initiation was about 30 days from sowing. However, variation has also been observed. Within bunch types, the efficiency of flowers to produce pe 17 was inversely related to the number of flowers produced.

Andanigowda (1977) observed that the total number of flowers produced was 128-129 per plant in variety TMV-7. However, an increase in the number of flower production was not considered to be important for enhancing fruit setting efficiency as the number of flowers produced per plant was highly variable.

Varietal differences in five cultivars were considered partitioning of assimilates to be related with three physiological processes, namely between vegetative and reproductive parts, the length of filling period and the rate of fruit establishment. Of these, the partitioning of assimilates had the greatest effect on fruit yield. As such, this
characteristic seemed to be highly desirable as it ensured a high efficiency in converting available growth into the economically important part, i.e., pod (Ducan et al. 1978).

Studies conducted at Pindivanum (Tamil Nadu) showed that the rate of growth was very rapid in the first two fortnights of flowering. Peak growth was noted during the second fortnight in bunch types. In the spreading types, the maximum growth period continued till 2-3 fortnights after flowering. An alternate decrease and increase in growth rate also reported up to 6-7th week after commencement of flowering (Sastry, 1979).

Among various growth parameters, shoot length truly represented the genetic behaviour of any plant species. Virginia types grew taller than other cultivars, e.g., Spanish and Valencia. However, Spanish and Valencia grew and levelled off soon, but Virginia showed constant increase in height till flowering (Reddy et al. 1980).

Choudhari et al. (1985) reported that primary branches contributed 36% and 90% to the total number of pods per plant in the "Kharif" and "Summer" seasons respectively. The contribution of the first four nodes of primary branches, however, was 85% in both seasons. The number of mature seeds per plants was higher in "Summer" than in "Kharif" season.
Castray et al. (1985) reported that, in bunch type of groundnut, the total number of flowers per plant was not related to pod yield under field conditions. It was, therefore, suggested that low flower production was not a constraint on productivity. However, a positive significant relationship was observed between the number of flowers produced during the first two weeks after commencement of flowering and pod yield. Thus, the number of the expected pods could be predicted as early as 70 days after sowing.

Dwivedi (1986a) studied the yield performance of different genotypes of groundnut at Junagadh (Saurashtra). He observed the development of different attributes of the crop in various seasons giving emphasis to the reproductive phase. He reported that the crop sown during the month of March and November produced higher number of flowers, pegs, and pods as compared to other months. He observed that Virginia runner had a low flower pod ratio, indicating a higher percentage of flowers turning into pods than that the bunch type and suggested that emergence of peg close to the soil in Virginia runners might be the main reason for this observation.

To conclude, the work on yield performance of groundnut cited above indicates that a high-yielding variety should...
produce more pegs (flower to peg ratio), higher per cent conversion of pegs into mature pods (pod to peg ratio) and higher 100 seed weight.

2.3 Physiological aspect of drought

Water is the most important component of all living beings contributing more than half the weight of the body. It acts as a medium for all life processes. Therefore, scarcity of water creates several disorders in the body, plants being no exception. However, plants experience more fluctuations in the availability of water than animals as they can not move about to fulfill their water requirements. Further, they lose the bulk of water absorbed through transpiration. This could lead to alarming situation when transpiration exceeds the uptake in plants. It puts plants under severe strain and is called drought. There are many factors causing drought, e.g., low annual rainfall, late onset or early withdrawal of monsoons or a large gap between two successive rains. Drought curtails the productivity of plants considerably by impairing their physiological activities.

Drought decreases hydrostatic pressure and reduces water potential of the cell (Simonsgaard, 1976). This affects cell expansion and cell division adversely (Isaacs, 1973). On the other hand, Cleland (1967) noted a decrease in
cell wall synthesis, as measured by incorporation of labelled glucose, when plants experienced water deficit conditions. Similarly, Ben-Zioni et al. (1967) found that water deficit resulted in decreased incorporation of amino acids into proteins.

In addition, enzyme levels are also affected by low water availability. Of these, membrane bound enzymes, like ATPase, are likely to be influenced adversely by the drop in water potential (Zimmerman, 1978). Besides, nitrate reductase level in plants experiencing drought was decreased for which Bardzick et al. (1971) argued that suppression of protein synthesis could be the reason. Moreover, translocation of photosynthates is restricted which decreases the productivity of the crop. Lastly, accumulation of proline and abscisic acid is observed in the cell when drought prevails for a long time and proline is used as a pointer to assess the severity of drought.

2.3 Effect of drought on growth and yield components of groundnut

Plants encounter different types of stress during their life cycle. Stress may be due to low level of moisture, very low light intensity, severe cold, high temperature or high saline conditions in the soil. Occurrence of any of
these adverse conditions during crop growth will alter the
plant behaviour abnormally.

It is common knowledge that drought (inadequate
availability of water) reduces plant growth and development
by affecting various physiological processes adversely. The
effect of drought becomes visible when plants face it over
a period of several days. Baras and Klepper (1968) observed
that fairly large water deficits and low leaf water potential
could develop in an hour, when transpiration had been rapid.

Groundnut is grown largely under rainfed conditions
in our country. During monsoon seasons, plants experience
an intermittent moisture stress thus suffering from drought
periods once or twice during their life cycle. Unfortunately,
neither the magnitude of the stress nor its duration is
predictable. Hence, the type of stress tolerance required
in such situations must be of a general nature with emphasis
on regeneration in a normal soil moisture regime. Soil
moisture stress affects various aspects of plant growth right
from germination to grain filling.

Slatyer (1955) reported that the rate of dry matter
accumulation by groundnut was first reduced when the relative
turgidity of cells dropped below 90%. Decrease in dry matter
production of vegetative components with moisture stress was
observed by several workers (Fourrier and Prevot, 1959). Ilyiana (1959) reported that groundnut plants were less susceptible to moisture stress during pre-flowering stage.

Ochs and Werner (1959) reported that soil water deficit reduced the internode length more drastically than it reduced node number. Water deficit during several stages of growth was found to reduce the rate of daily leaf production.

Bilz and Ochs (1961) showed that the degree of partitioning of assimilates to leaves during the seed formation (90-120 days) of Spanish groundnut was inversely proportional to the number of fruits formed earlier. If pod formation was nearly complete prior to imposing water deficit, assimilate partitioning to fruits was increased by water deficit. However, water deficit during pod formation (50 to 80 days) reduced flowering, pod formation and final yield more than at any other stage. This treatment resulted in less partitioning to fruits (but more to leaves) during the subsequent period of seed filling (90-120 days) during which the plants were irrigated. It indicates that the influence of water deficit on partitioning of dry matter to leaves or fruit depends on the timing of moisture stress. Cheshadri (1962) observed that flower initiation was greatly affected by moisture stress in bunch type of groundnut.
Hiller and Clark (1970) observed that erect type of groundnut was susceptible to moisture stress during 50-80 days after sowing. Stress during this period caused more reduction in vegetative growth, flowering and pegging and decreased more yield than stress at any other growth period.

Lenka and Misra (1973) reported that water deficit delayed flowering by 1-2 days and reduced the total number of flowers. Irrigation at 25% depletion produced more flowers, seeds/pod and seed weight was higher per pod than obtained in absolute moisture stress. Reproductive efficiency of flowers was also maximum with irrigation at 25% depletion. In most cases, soil moisture deficit during pegging and pod development primarily reduced pod number while it rarely affected weight per pod (Varnell et al., 1976; Vivekanandan and Gunasekara, 1976).

The studies of Andanigowda (1977) revealed that erect type of groundnut plant was not very susceptible to moisture stress, even when the level of moisture was very low. The work of Pallas et al. (1977) on groundnut revealed that soil water tension of - 1.5 MPa during the growth seasons resulted in a 7 per cent loss of sound seeds and 5 per cent decrease in germination in the most drought resistant genotype. During drought, newly formed pegs were also found not to penetrate dry soil.
Pandey et al. (1984) studied groundnut growth in semi-arid tropical regions by subjecting it to different moisture gradient in the field. Plant growth analyses were computed from samples taken at frequent intervals. Increasing water stress resulted in progressively less leaf area duration, crop growth rate and shoot dry matter production. They concluded that the growth of the crop was often limited by variation in the amount and duration of rainfall.

Bennett et al. (1984) studied the effect of 28 days drying period, achieved by withholding irrigation on various mid-day physiological parameters and soil water content. It was noted that as soil drying progressed, the reduction in soil water content caused larger reduction in leaf water potential and consequent decrease in leaf moisture content. This resulted in stomatal closure and higher leaf temperature than air temperature. They concluded that water seemed to maintain plant body temperature besides, participating in various metabolic pathways.

Ong (1984) investigated the partitioning of dry matter to stem, leaves and pods under water stress condition. He reported that mild water stress promoted pod and pod production, because reproductive growth was less affected than the growth of leaves and stem. Ong et al. (1995) also noted that the lowest soil moisture content reduced the LAI, leaf number per
plant and leaf size were decreased as soil water deficit increased.

It may, therefore, be concluded on the basis of the effects of moisture stress on yield characteristics of groundnut cited above that drought reduces yield primarily by decreasing the pod number. It causes a decline in the quality of groundnut at harvest and in its shelling per cent as drought delays the onset of rapid fruit growth and thus causes late fruit maturation.

2.4 Physiological role of potassium

Potassium, an essential mobile macronutrient, is most abundantly distributed in all plants but does not appear to be a constituent of the plant body. The function of this element in plant metabolism is biophysical, involving ion fluxes as it maintains turgor pressure of the cell via osmoregulation and activates about 40-53 enzymes, categorised in three groups, namely enzymes transferring phosphoryl groups, enzymes catalyzing eliminating processes and unclassified enzymes (e.g. starch synthase). Some of the common enzymes activated by potassium are pyruvate kinase, 6 phosphofructokinase NAD synthetase, fructose-1,6-biphosphate aldolase etc., covering a wide spectrum of plant metabolism (Evans and Sorger, 1966).
Nitsos and Evans, 1969; Sauter, 1970). Thus, potassium takes active part in opening and closing of stomata and helps in the synthesis of proteins and ATP (Webster, 1956; Beringer, 1982). It is also involved in the translocation of photosynthates. These physiological roles account for its greater demand in maintaining high crop productivity (Beringer, 1982). The function of potassium becomes indispensable in water stress condition where it facilitates water uptake and prevents excessive water loss through transpiration. Therefore, water use efficiency of plants increases in its presence. This improves the performance of crops in drought condition (Linsner and Harwig, 1968; Arag, 1972; Mengel, 1977). Besides, potassium helps nodulation in legumes and thus enhances dinitrogen fixation (Marxhner, 1983).

2.5 Water use efficiency in relation to potassium nutrition

Rogali (1958) grew wheat, barley, maize and clover in water culture and in soil in pot experiments. He found that added potassium increased the permeability of root cells to water. Whereas plants lacking potassium transpired more water, the addition of a small amount of potassium greatly reduced transpiration.

Achitov (1961) suggested that potassium supply reduced transpiration and increased water uptake. It thus improved
water use efficiency. He concluded that treatments which could reduce transpirational loss would be valuable under conditions of moisture stress. Blanchet et al. (1962) and Linser and Herwig (1968) concluded that the effect of potassium is of particular importance in crop production, since it reduces water loss through transpiration and increases organic matter production in crop well supplied with potassium.

Fischer (1969) studied stomatal behaviour in relation to potassium nutrition. He observed that stomatal resistance depends, on the number of stomata per unit leaf area, as well as on the geometry of the stomatal pores. Thus for a given plant species, stomatal resistance (diffusive resistance) is related to the degree of the stomatal opening. The variation in stomatal aperture is achieved by turgor changes of guard cells. Because of their non-uniform wall thickness, the guard cells are bent as their turgor (water content) increases, and this results in the opening of the pore. The reverse occurs when guard cells lose water. The fluctuation in guard cells water content which bring about the change in their turgor are caused by means of an active potassium pump. Potassium ions are pumped into the guard cells from the surrounding subsidiary or epidermal cells, when stomata are about to open. Such an influx of ions decreases the solute potential of the
guard cell in comparison with that of the epidermal cells, thus causing net water influx to the guard cells. On the other hand, the pump is inactivated when stomata are about to close, which causes a positive movement of $K^+$ out of the guard cell. The importance of $K^+$ in opening and closing of stomata has been investigated and discussed by many workers (Fisher and Hsiao, 1969; Humble and Raschke, 1971; Trolldenier, 1971). However, it has been suggested that the efficiency of water taken up from the soil and its transport upwards are more important than stomatal conductance in determining drought resistance by sorghum and cotton (Ackerson and Krieg, 1977).

According to Mengel (1977), potassium is the most important inorganic solute in the plant that is osmotically active. The osmotic role of potassium causes this solute to be an outstanding factor in plant water relations, that is, in the absorption, translocation and loss of water. It is well established that plants supplied with sufficient potassium shows less water loss because of reduced rate of transpiration.

Barinier and Trolldenier (1978) concluded that potassium nutrition may support both tolerance and avoidance of drought. Plants adequately supplied with potassium respond almost immediately to water stress (induced by hot winds) by
reducing their transpiration, while plants with moderate or severe potassium deficiency are obviously unable to close their stomata efficiently. Thus, with adequate potassium nutrition, water utilisation is improved, less being transpired per unit of dry matter produced.

2.6 Nitrate reductase activity

Nitrate reductase is the first enzyme in a sequence of reactions leading to assimilation of nitrate into amino acids and thence into cell organic nitrogen compounds. Incorporation of this inorganic nitrogen to organic form requires it to be reduced to NH$_3$$^+$. This consists basically of two steps: the reduction of NO$_3^-$ to NO$_2^-$ and the further reduction of NO$_2^-$ to NH$_3$$^+$. Reduction of NO$_3^-$ to NO$_2^-$ is catalysed by nitrate reductase and takes place in the cytoplasm. This enzyme was found to be a sulphahydryl metalloflavoprotein (metallo FAD/protein) containing molybdenum. It was initially isolated from Neurospora and was later characterised in higher plants from the leaves of Glycine max (Nason and Evans, 1953; Nicholas and Nason, 1955). It is an inducible enzyme and is synthesised only when nitrate is present in the medium (Hewitt and Afridi, 1959; Afridi and Hewitt, 1964; Brevers and Hagman, 1969).
Nitrate accumulation in plants is due to several factors, among which drought is possibly very important one. The response of nitrate reductase to water stress was studied by Salasubramaniam et al. (1974), to determine the stability of this enzyme in various crops. In irrigated crops, nitrate accumulation normally did not occur unless nitrogen application was excessive. It has also been shown that accumulation of nitrate in various plants could be responsible for nitrate poisoning of livestock.

The response of nitrate reductase to water stress over a 24 h cycle was studied in wheat, along with relative water content, nitrate and proline content of leaf. All the variables showed changes over the 24 h cycle. The change in leaf nitrate content followed the change in relative water content. Moreover, the proline content in the unirrigated plants was maximum when the relative water content was lowest which coincided with reduced nitrate reductase activity (Najyopai et al. 1977).

Suczek and Buzynski (1979), reported that presence of NH\textsubscript{4}\textsuperscript{+} in nutrient solution containing NH\textsubscript{4}\textsuperscript{+}NO\textsubscript{3} with K\textsuperscript{+} removed, inhibited nitrate reductase activity in cucumber leaves. The absence of K\textsuperscript{+} in NaNO\textsubscript{3} medium also decreased NRA. The addition of K\textsuperscript{+} enhanced NRA in the leaves of plants growing in solution containing NaNO\textsubscript{3}. 
Khanna et al. (1980) studied the effect of potassium (0, 50, 100, and 200, mg/pot) on the growth characteristics and nitrate reductase activity in maize seedlings during water stress and subsequent recovery. Nitrate reductase activity (NRA) rose in irrigated plants at 24 h after potassium application. Subsequently, as water stress developed, potassium helped in maintaining NRA for the first two days.

Sinha and Nicholas (1981) suggested that plants with higher NRA under drought condition may have better potentiality for water stress resistance.

2.7 Proline content

Proline is one of the major constituents of biological proteins and is synthesised from glutamic acid (Voegel and Davis 1952). During a period of water deficit, a range of amino acids accumulate to a greater or lesser degree in different organisms, but the most frequent and extensive response is an increase in the concentration of proline. Accumulation of proline upon dehydration due to water deficit has been recorded in bacteria (Tempest et al. 1970; Measures, 1975) and in higher plants (Palfi et al. 1973).

Proline acts as a compatible solute to regulate osmotic potential, reducing water loss from the cell during
water deficit. Proline is oxidised readily in turbid tissue glutamate and amino group. Thus, it becomes a ready source of energy which is released when glutamate passes through Kreb cycle. Secondly, amino group becomes available where required. During stress periods proline accumulation is the process of energy and amino group conservation. It accumulates in young leaves and shoots under water stress and is non-toxic. It also serves as sink for soluble nitrogen and protects enzymes from the effect of toxic compounds. As nitrogen uptake is curtailed and NRA reduced under water stress, nitrogen accumulation in compounds like ammonia could be toxic to plants while accumulation as free proline is not. Good corelation with proline accumulation and drought resistance have been worked out in different crops (Singh et al., 1972). This may serve as attribute for drought resistance screening.

The relationship between nitrate reductase and proline accumulation was examined in several crop species. There was a sharp decline in enzyme activity in response to water stress in wheat, barley, sorghum, maize, *brassica* and safflower and a rapid and considerable simultaneous accumulation of proline in all these species. In barley and wheat, when was fed to plants stressed with polyethylene glycol, the loss of NRA was reduced. This suggested that exogenous (and possibly endogenous) proline protects the enzyme from inactivation during water stress (Sinha and Rajagopal, 1975).
Hakki et al. (1977) surveyed 10 groundnut cultivars for free proline accumulation in leaves and found that, under stress conditions, the drought tolerant cultivars accumulated more proline and fixed more $^{14}$CO$_2$ than the other cultivars. However, proline content was not specifically correlated with relative water content as high or low temperature and salinity also induced proline accumulation. Marcana (1991) reported that potassium significantly increased free proline accumulation in beans.

2.8 Chlorophyll content

Boger (1964) observed that potassium influenced the photosynthetic processes by controlling chlorophyll synthesis. When potassium is withheld from the growing medium of Chlorella vulgaris suspensions a structural protein in the chloroplast is not filled entirely with chlorophyll but, after potassium is added to the cells, chlorophyll is formed even in dark.

Molotkovsky and Dzybenko (1970) and Coneva (1971) showed that potassium increased the photo-chemical activity of chloroplast in maize. The experiments in nutrient solution showed that the effect of potassium in increasing chlorophyll
content was accompanied by increased activity of chlorophyllase. Froster (1976) demonstrated the dependence of chlorophyll content in the flag leaf of spring wheat on leaf potassium content. The chlorophyll concentration was increased by 88% at the highest potassium level.

Khanne et al. (1980) reported that potassium increased leaf area expansion leading to increased leaf area per plant with decreased chlorophyll content under drought condition. During recovery from water stress, potassium helped in maintaining higher leaf area expansion rate and chlorophyll content. Chlorophyll content has an important influence on the rate of photosynthesis.

Reddi et al. (1980) studied the chlorophyll content of leaves in groundnut varieties under drought condition. Chlorophyll reduction was noticed in all the cultivars due to moisture stress.

Bark and Chau (1983) reported that groundnut cultivar grown in pots given 270 mg K/kg soil, lost iron deficiency symptoms. Potassium as KCl was better than as KNO$_3$ or $K_2$HPO$_4$ and it increased the chlorophyll content by 73% to reach 90%. Potassium fertilisation, at the rate of 135 to 405 mg K/kg soil, ameliorated iron chlorosis in groundnut grown in an
extremely calcareous soil (63%, CaCO₃). Such treatment
doubled and even tripped the chlorophyll content. Potassium
availability in soil is important in achieving this effect.
K₂SO₄ was found to be more effective than KCl. These results
are attributed to the cation-anion balance and consequent
rhizosphere acidity.

2.9 Energy harvest efficiency

The basic process of energy metabolism is the conversion
of radiation energy into chemical energy. Being the energy
source, light intensity and light quality are the most
important environmental factors influencing assimilation.
The energy efficiency of crops generally does not exceed
1-2% of photosynthetically active radiation (PAR). An
improvement in the efficiency of solar energy utilisation of
PAR up to 3-5% is feasible for enhancing plant productivity and
intensification in agriculture. The rate of energy harvest
by a crop on a unit land area is totally dependent upon leaf
area index, geometry of canopy and per unit leaf area energy
harvest. At early stage of crop growth, the canopy development
is slow as a result of which energy interception is low even
when all the conditions are favourable (Dwivedi 1986 c).

It should be emphasised that crop production is one of
the few production processes with a positive energy balance.
It now seems likely that in order to meet future energy needs, this acquisition of energy by plants will play an increasingly important role. Hall (1977) cites 5 plant species, eucalyptus trees, hibiscus shrubs, Napier grass (a tropical fodder grass), sugarcane, and cassava, which are considered to be suitable for 'sun energy harvesting.' Recently species of Euphorbiaceae have also been considered as possible 'energy crops.' The advantage of these species is that they have a low water requirement and can grow in rather arid regions. Some workers have found relationship between energy harvest and potassium nutrition of plant. The available literature on this aspect is briefly described below:

According to Stoy (1962) photosynthesis in Bastaar's (1959, 1959, 1962) experiments increased up to light intensities between about 40,000 and 60,000 lux in maize, wheat, beet, red clover and sugarcane. The plants were unable to utilise higher light intensity.

Amberger (1969) reviewed the function of potassium in carbohydrate metabolism and elucidated the connection that has for a long time been known between potassium nutrition and light utilisation in photosynthesis. The rate of photosynthesis per unit leaf area depended upon light
intensity and also on a number of physiological and morphological factors. Potassium has an important role in these metabolic processes.

Kicke (1973) worked with oats in pot culture and found that potassium uptake was increased by increasing light intensity and that its increased uptake led to better utilisation of light energy. Haeder and Mengel (1975) and Mengel and Haeder (1976), grew oats in solution culture. They reduced light intensity during the generative phase to half the normal intensity and found that enhanced potassium supply during the change from vegetative to reproductive development largely compensated for the reduction in light. Increasing potassium increased yield significantly at the mature stage in normal daylight.

Smid and Peaslee (1977) grew maize in the open in sand culture at densities of 33,000, 99,800 and 1,19,000 per ha and light intensity varied by artificial shading. Potassium was applied in solution at 15, 45, 135 and 400 μg/cm². Assimilation rate depended upon the point of insertion of the leaf. Assimilation rate fell as light intensity was reduced slightly between 7.5 and 3.2 lumen/cm² and more strongly thereafter. Potassium concentration in the plant increased with increasing
potassium content of the nutrient solution at all light intensities. The assimilation rate increased with increase in potassium content and this increase was more marked at high intensity. At 1.6 lumen/cm² potassium supply had much less effect on assimilation rate. Thus potassium was claimed to improve the efficiency of light utilisation.

Steineck and Hæder (1970) reported that potassium promotes water storage in the cell, turgor of the cytoplasm and enzyme proteins, thus providing favourable conditions for photosynthesis and the succeeding steps in metabolism. Turgor has an important influence on leaf alignment and affects light interception.

The specific function of potassium in energy conversion process is not yet completely understood. However, it is recognised that potassium is involved in metabolic reactions including those of ATP synthesis and energy transfer (Pfluger and Mengel 1972; Lauchi and Pfluger, 1973; Sereinger 1982).

Dhivedi et al. (1985) reported that energy conservation in the phycobiomass of groundnut is significantly higher than that of wheat and Cynodon dactylon, mainly because of the fact that groundnut accumulates energy-rich organic substances such as oil and protein. Thus, groundnut harvest
conserves more solar energy than other crops. However, it is desirable that research into improving plant type of groundnut for higher dry matter productivity and partitioning in seeds intensified since it has a much higher potential for harvesting solar energy.

In the end, the excellent as yet unpublished review of Dwivedi may be cited wherein he observed that potassium augments solar energy harvesting efficiency of groundnut during both "Kharif" (low light intensity) and "summer" (high light intensity) seasons in bunch and spreading genotypes of groundnut (Dwivedi, 1985).

2.10 **Potassium nutrition of groundnut**

Generally, groundnut is grown in soils that are high in potassium. The level of exchangeable potassium in groundnut growing soils of the world in general ranges between 40 kg/ha and 1,700 kg/ha. However, in practice, soils having less than 150 kg/ha, 150-250 kg/ha and more than 250 kg/ha potassium are considered as low, medium and high potassium soils respectively for groundnut. Except the soils of Kerala, Bihar and Orissa, most soils of other states of India, where groundnut is grown, (including Gujarat) are rich in potassium. The stunted growth of groundnut and drying up and necrosis of
leaf margins are manifested under potash deficiency. Reddish colour of stem at the tips of branches is also observed.
Deficiency of potassium has been reported to reduce the number of flower forming pegs. Sufficient quantity of potash is required for sound growth of pegs. Root growth is drastically reduced due to potash deficiency. The deficiency of this nutrient is also evident when high proportion of pods are produced but with only one seed each (Owivedi, 1986 b).

Harris and Bledsoe (1951) reported that groundnut plant is quite erratic in its response to fertiliser application. Groundnut is sensitive to an unbalanced nutrient supply and this is considered responsible for conflicting results. Black (1968) observed that high levels of potassium hastened the flowering in groundnut. This might be due to the better growth and vigour in early stages of the plants as a result of applied potassium. The nutrient increased the number of pegs formed per plant. Appreciable delay in maturity due to potassium deficiency was also reported. Anon (1972) found that for every one tonne of unshelled nuts and two tonnes of hay, groundnut crop removed about 38 kg potassium.

Badnaur (1976) conducted field experiment at Bangalore (Karnataka) to study the response of various crops to potassium fertiliser application. Groundnut and cowpea gave maximum
yield with 30 kg K₂O/ha, but maize and ragi responded maximally to 60 kg K₂O/ha. Applications of 120 kg K₂O/ha and 140 kg K₂O/ha did not increase the yield significantly.

Gopalaswamy et al. (1976) worked out the economical optimum dose for irrigated bunch groundnut. It was extrapolated to be 75 kg K/ha which was 22 kg K/ha more than earlier recommendation, i.e., 53 kg K/ha for the same groundnut. Gopalaswamy (1977) conducted an experiment for three years. His results revealed that the rainfed bunch groundnut did not respond to the application of N and P₂O₅. However, there was a significant interaction between N and K at N₀ level application of 40 kg K significantly increased the yield. The economical optimum dose was computed to be 45.2 kg K/ha for the sandy loam soil with low potassium content.

Naddi et al. (1977) conducted the experiment under the agroclimatic condition of Royal, seema region of Andhra Pradesh for rainfed bunch groundnut in red sandy loam. The yield maximisation level of potassium was found to be 100.2 kg K₂O/ha whereas profit maximisation level was 70.7 kg K₂O/ha with a net return of Rs. 1.20 per rupee spent on potassium.

The results of two years study revealed that maximum production of pod occurred with the applied potash at the rate
of 80.5 kg/ha but the economical optimum dose was computed to be 77.2 kg/ha, which was expected to yield 2,676 kg/ha groundnut pod with input cost and output price ratio as 0.5 : 1. When the cost of potash remained constant the economical optimum dose of potash increased with an increase in the price of pods. On the other hand, an increase in the cost of potash reduced the economical optimum dose irrespective of price of pods. The study emphasised the need to limit potash application according to input cost and anticipated output price in irrigated groundnut (Gopalswamy et al. 1978).

Lakshminarasinhan and Kurandran (1978) conducted a field experiment on groundnut variety M.13 (suitable for table purpose) to study the effect of split application of potash given at 1:2, 1:2 and 2:1 ratios (soil : foliar) on the quality of the seed. Among the various ratios tried, 60 kg K₂O/ha the 2:1 ratio recorded higher nitrogen and potassium uptake. Reducing sugars increased to 4.2% with this level of potash application. Increase in total and reducing sugars with application of potash at 90 kg K₂O/ha in 1:1 and 2:1 ratio was observed. The oil content was not influenced by the treatments.

Jutstein (1979) studied the effect of 0, 150, 300 or 450 kg K/ha on the yield response a Valencia type groundnut
culvated on alluvial grumso soil. Potassium application prolonged the vegetative period and delayed reproductive development. Treated plants yielded more than control plants of 300 kg/ha receiving the optimum potassium rate and improved their reproductive efficiency.

Rao et al. (1980) conducted field experiments in two seasons (summer and rainy season of 1978) at Tirupati (Andhra Pradesh) to study the individual and combined effect of potassium, calcium and magnesium on irrigated TMV-2 groundnut. There were 16 treatments in summer and 18 treatments in rainy season with various ratios of K:Ca:Mg with two controls in which the source of phosphorus was either single superphosphate or diammonium phosphate. A common fertilizer dose of 30 kg N and 17 kg P/ha was also applied in the form of urea and diammonium phosphate respectively. It was found that 30 kg K/ha in summer and 40 kg K/ha in rainy season gave maximum number of filled pod per plants. Addition of 20 kg Ca/ha and 30 kg K/ha significantly increased pod number. 30 kg K/ha proved optimum for pod weight in both seasons. Maximum pod weight was found with the combination of 80 kg K + 40 kg Ca/ha in summer and 120 kg K + 40 kg Ca/ha in the rainy season. Test weight was significantly increased with 40 and 30 kg K/ha in both the seasons. Higher test weight was obtained in 30 kg K/ha + 40 kg Ca/ha in summer and 120 kg K + 40 kg Ca/ha
In the rainy season, with every increase in the level of potassium, the shelling per cent was increased in both the seasons. For pod yield, 40 kg K/ha proved optimum. Yield increase with 40 kg K/ha was 90.3% over the control. The effect of calcium and magnesium was found to be significant in some of the parameters at different levels of potassium.

Hair et al. (1981) conducted a field trial in red loam soils at Vellarajani (Kerala). They noted that potassium at higher levels up to 50 kg K₂O/ha significantly increased the height of plant and number of leaves per plant. Potassium at higher levels up to 75 kg K₂O/ha decreased the time taken for flowering and increased the number of pegs formed per plant. The test weight of pods and hundred pod weight were increased significantly by potassium application up to 50 kg K₂O/ha. Higher level of K₂O increased the yield of pods and haulm. Optimum and economic levels of potassium were 124 and 116 kg K₂O/ha respectively.

Reddy and Reddi (1981) performed a field experiment to study the response of potassium on two groundnut varieties (A-11, 1192 and TW-2) in sandy loam soil of Tirupati (Andhra Pradesh). Both varieties gave maximum average yield of 322 kg/ha at the level of 79 kg K/ha during the rainy season. In the second year, 40 kg K/ha gave 1,663 kg pods/ha
which was at par with the yield (1,774 kg/ha) obtained with 30 kg K/ha.

Dwivedi (1985) reported that application of potassium increased oil and protein content and energy value of seed. It was suggested that potassium application augmented solar energy harvesting efficiency of groundnut during both "Khari" (low light intensity) and "Rabi" season (high light intensity). Increase in stomatal resistance and decline in transpiration rate was noted due to potassium application in GAUG-1 (bunch) and GAUG-10 (spreading) genotypes of groundnut.

Kanseria (1986) studied potash depletion rate and uptake of potash by crops including groundnut in medium black calerrious soils of Junagadh and concluded that potash application enhanced yield. He recommended that potash fertilisation should be considered for intensive cropping of groundnut.

2.11 Performance of other crops in relation to potassium nutrition

Potassium was first recognised as an essential element for plant growth following the work of Hone in 1762. He carried out an interesting experiment by growing barley in sandy soil. In one treatment, Hone added potassium sulphate to the soil and this resulted in increased growth of barley. It showed
that potassium or sulphate has a beneficial effect on plant growth. Later researchers such as de Saussure and Sprengel recognised that potash was present in plant ash obtained from different plant species (Mangel and Kirkby, 1930).

Liebig (1941) proposed after reviewing the analytical data of his time that potassium was in some ways involved in plant metabolism. The experience of farmers around Giessen, the German University town in which he worked, indicated the beneficial influence of manuring crops with plant ash. He recognised that potash was the essential growth factor in the ash. Furthermore, Liebig was also aware of the fact that the clay fraction of the soil provided a source of potassium for plant growth. In his book, "Die Organische Chemie in Thier Anwendung auf Agrikulture Und Physidogie" (Organic Chemistry in Relation to Agriculture and Physiology) he wrote "Their must be a component in clay which has an influence on plant life and which directly participates in plant development. This component is the ever present potash or sodium."

Van Der Pasuw (1958) compiled a report on fertiliser response of potato, wheat, grass and bean over the 15 years from 1935 to 1949. Crops differed in their response to potassium and the optimum level depended upon the crop. If there were more than 46 rainless days after planting, potato
yield declined in proportion to the number of rainless days in the (control treatment) but was maintained if the potassium supply was good. Potassium uptake was reduced in dry years. On the other hand, wheat, being much less responsive than potatoes showed an increase in yield vis-à-vis an increased in potassium supply in dry years. Such effects were not evident on grasses and beans.

Barber (1971) found that response of soybeans to potassium depended on rainfall in the 12th weeks following planting. If rainfall was below 380 mm, response was nearly linear.

Single grain weight depended much on the potassium status of the plant at flowering stage. Late application of potassium had little effect on grain development. Further rate of potassium uptake by cereals after flowering was probably very low. However, the potassium status of leaves and culms at the grain filling period had a substantial impact on photosynthesis and on translocation of photosynthates from these organs towards the ears (Mengel and Forster, 1971).

Diffusion is the major transport mechanism for potassium in the soil and soil moisture content plays an important role for nutrient availability to plants. Many irrigation experiments have demonstrated positive interactions between
irrigation and fertiliser application. Several findings have shown that diffusive flux increases with soil moisture (Mengel and Von Braunshweig, 1972).

Wichens (1975) found that *Lolium perenne* could take up very large quantities of non-exchangeable potassium when soil moisture was satisfactory. However, under drier conditions, release of non-exchangeable potassium was much restricted. Thus, under moist soil conditions, only occasional response to potassium fertiliser was recorded, but consistent response was obtained when conditions were dry. This finding was of practical importance showing that even plant species with a high potassium exploiting power might be inefficient to extract soil potassium under dry conditions and sub-optimal potassium supply might result in much reduced yield.

Haulay (1976) found that drought resistance in vines was increased by heavy preplanting application of potassium. Forster (1976) made the observation that under optimum potassium nutrition, the senescence of the flag leaf was delayed in wheat. This resulted in a prolonged 'leaf area duration' which, according to Evans et al. (1975), was important for grain development.

Ralph (1976), conducting field experiment on clay soils in England, found that grain yield of winter wheat
was increased by improving the single grain weight as a consequence of potassium application. The treatment often increased the number of grain per ear. He observed that potassium especially promoted the development of the proximal, central, and distal spikelets.

Schon et al. (1976) conducted field trials for 20 years on a loess soil containing much non-exchangeable potassium. They showed that *Vicia faba* and a grass clover mixture responded most favourably to potassium fertiliser. It indicated that legumes were not very effective in exploiting soil potassium. Potassium markedly increased the yield of potatoes, but affected the grain yield of cereals slightly. Hernando and Orihuel (1977) noted tomatoes to be more responsive to potassium under drier watering schedule.

Nour and Weibel (1978) reported that diffusion of nutrients to the root surface was restricted and root was forced to grow towards regions which still contain available nutrients. Crops like legumes or row crops, had a less dense root system than drought resistant species or the small grain cereals. Therefore, they were obviously less capable of absorbing non-exchangeable inter layer potassium. Barber (1978) also reported that legumes required higher doses of potassium.
Secer (1978) observed that, shortly after pollination, a substantial amount of nitrogen was still stored in the culms and leaves of wheat in the form of protein. These proteins were mobilized at the stage of most rapid growth of the grain and were used for the synthesis of grain proteins. Plants with high potassium status were found to be more efficient in mobilizing the stored leaf proteins and in translocating the resulting amino acids towards the grain. From Secer's results it is also clear that the beneficial effect of potassium on grain filling was not related to the potassium content of the grains but resulted exclusively from the influence of potassium on the translocation of assimilates from the vegetative plant parts towards the ear. On soils of medium potassium availability in Ohio, response of maize in dry years to potassium was as much as 2,700 kg/ha compared with only 198 kg/ha in years with optimum rainfall (Johnson, 1979). Glass (1980) found that potassium requirement was likely to be higher in the tropics than in the temperate climate.

Optimal potassium requirement of crops is dependent on the potassium availability in the soil and on its requirement
by the respective crop. Its availability is determined by the potassium concentration of the soil solution and the rate at which it is buffered by the potassium reserves. Diffusion is the major mechanism of potassium transport to the roots. As the diffusion paths are short, the roots have to grow towards the nutrients. Plant species having only a sparse root system, are, therefore, at a disadvantage and need higher doses of fertiliser. Similarly, modern high yielding varieties have higher potassium requirements, especially during their vegetative growth (Beringer, 1982).

Saxena (1985) in his excellent review on "the role of potassium in drought tolerance" emphasised the importance of potassium in water uptake and in the regulation of water loss through stomata under both controlled as well as field conditions and established that the water relations of plants and water use efficiency could be improved by the application of potassium that in turn would affect the final yield under water stress conditions.

12 Conclusions

The literature reviewed above includes studies on physiological analysis of growth, yield, enzymes like nitrate reductase, chlorophyll and proline content in various plants with respect to the application of potash under normal and
water stress conditions. It appears from the review that there are very few reports concerning the effect of potash fertilisation on various physiological processes including productivity of groundnut. The work on groundnut is invariably aimed at increasing the final pod yield without much understanding of the physiological processes leading to it. It is considered logical to study those factors that limit the growth and yield of the crop and may help understand these processes to ensure enhanced productivity of better quality groundnut.

It is generally recognised that groundnut is mostly cultivated in those parts of the world where frequent drought prevails and regulates the productivity of the crop. The role of potassium in relation to drought condition has been well established in various crops other than groundnut. Keeping in view the stimulating effect of potassium on these crops under drought conditions, it is proposed to undertake the present investigation on groundnut in order to explore the possibility of augmenting its per capita productivity for catering to the need of the ever-increasing population of our country.
CHAPTER III

MATERIAL AND METHODS
<table>
<thead>
<tr>
<th>Section</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>3.1 Proposed study</td>
<td>48</td>
</tr>
<tr>
<td>3.2 Field preparation</td>
<td>48</td>
</tr>
<tr>
<td>3.3 Soil characteristics</td>
<td>49</td>
</tr>
<tr>
<td>3.4 Seeds</td>
<td>50</td>
</tr>
<tr>
<td>3.5 Creation of drought</td>
<td>50</td>
</tr>
<tr>
<td>3.6 &quot;Kharif&quot; trials</td>
<td>50</td>
</tr>
<tr>
<td>3.6.1 Experiment 1</td>
<td>51</td>
</tr>
<tr>
<td>3.6.2 Experiment 2</td>
<td>53</td>
</tr>
<tr>
<td>3.7 &quot;Rabi&quot; trials</td>
<td>55</td>
</tr>
<tr>
<td>3.7.1 Experiment 3</td>
<td>55</td>
</tr>
<tr>
<td>3.7.2 Experiment 4</td>
<td>55</td>
</tr>
<tr>
<td>3.8 Summer trial</td>
<td>55</td>
</tr>
<tr>
<td>3.8.1 Experiment 5</td>
<td>58</td>
</tr>
<tr>
<td>3.9 Sampling technique</td>
<td>58</td>
</tr>
<tr>
<td>3.10 Growth and yield parameters</td>
<td>60</td>
</tr>
<tr>
<td>3.10.1 Plant height</td>
<td>60</td>
</tr>
<tr>
<td>3.10.2 Leaf number</td>
<td>60</td>
</tr>
<tr>
<td>3.10.3 Leaf area</td>
<td>60</td>
</tr>
<tr>
<td>3.10.4 Leaf area index (LAI)</td>
<td>60</td>
</tr>
<tr>
<td>3.10.5 Leaf area duration (LAD)</td>
<td>60</td>
</tr>
<tr>
<td>3.10.6 Crop growth rate (CGR)</td>
<td>61</td>
</tr>
<tr>
<td>3.10.7 Biomass production</td>
<td>61</td>
</tr>
<tr>
<td>3.10.8 Energy harvest efficiency</td>
<td>61</td>
</tr>
<tr>
<td>Section</td>
<td>Description</td>
</tr>
<tr>
<td>---------</td>
<td>-------------</td>
</tr>
<tr>
<td>3.10.9</td>
<td>Yield analysis</td>
</tr>
<tr>
<td>3.10.10</td>
<td>Harvest index (HI)</td>
</tr>
<tr>
<td>3.11</td>
<td>Physiological, chemical and biochemical parameters</td>
</tr>
<tr>
<td>3.11.1</td>
<td>Relative water content (RWC)</td>
</tr>
<tr>
<td>3.11.2</td>
<td>Leaf temperature, diffusive resistance and transpiration</td>
</tr>
<tr>
<td>3.11.3</td>
<td>Solar radiation (SI &amp; PAR) perception by leaves</td>
</tr>
<tr>
<td>3.11.4</td>
<td>Nitrate reductase activity (NRA)</td>
</tr>
<tr>
<td>3.11.5</td>
<td>Proline content</td>
</tr>
<tr>
<td>3.11.6</td>
<td>Estimation of chlorophyll</td>
</tr>
<tr>
<td>3.11.7</td>
<td>Plant analysis for NP and K</td>
</tr>
<tr>
<td>3.11.8</td>
<td>Digestion of samples</td>
</tr>
<tr>
<td>3.11.9</td>
<td>Estimation of Nitrogen</td>
</tr>
<tr>
<td>3.11.10</td>
<td>Estimation of phosphorus</td>
</tr>
<tr>
<td>3.11.11</td>
<td>Estimation of potassium</td>
</tr>
<tr>
<td>3.11.12</td>
<td>Estimation of crude protein</td>
</tr>
<tr>
<td>3.11.13</td>
<td>Soluble sugars</td>
</tr>
<tr>
<td>3.11.14</td>
<td>Oil content</td>
</tr>
<tr>
<td>3.12</td>
<td>Statistical analysis</td>
</tr>
</tbody>
</table>
CHAPTER III

MATERIAL AND METHODS

3.1 Proposed study

It is proposed to conduct five experiments during "Kharif", "Rabi" and "Zaid" (summer) season, under rainfed (natural drought) and irrigated conditions (induced drought) at the farm of the National Research Centre for Groundnut (ICAR), Junagadh, Gujarat.

3.2 Field preparation

Before sowing, the field will be thoroughly ploughed to ensure maximum soil aeration. It will also help to eliminate weeds. The standard farm practices required for groundnut cultivation will be undertaken. The plot size will be sixteen sq. m. A gap of 3 m between two plots will be left so as to avoid seepage of water from one plot into another in induced drought experiments.

3.3 Soil characteristics

Before sowing, soil samples from various places in the experimental field will be collected from a depth of about 0-15 cm and 15-30 cm. These samples from the two depths will be mixed separately and will be analysed for various physico-chemical characteristics of the soil. The details of this soil analyses will be recorded for each experiment as per the proforma given in Table 1.
Table 1. Formats for recording soil analysis of the field for various physico-chemical properties (Experiments 1-5)

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Experiment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
</tr>
<tr>
<td>(1) Texture</td>
<td></td>
</tr>
<tr>
<td>(2) Particle size</td>
<td></td>
</tr>
<tr>
<td>distribution.</td>
<td></td>
</tr>
<tr>
<td>Sand %</td>
<td></td>
</tr>
<tr>
<td>Silt %</td>
<td></td>
</tr>
<tr>
<td>Clay %</td>
<td></td>
</tr>
<tr>
<td>(3) pH (1:2)</td>
<td></td>
</tr>
<tr>
<td>(4) Conductivity</td>
<td></td>
</tr>
<tr>
<td>i.e. E.C. 1:2</td>
<td></td>
</tr>
<tr>
<td>(mhos/cm)</td>
<td></td>
</tr>
<tr>
<td>(5) Available nitrogen</td>
<td></td>
</tr>
<tr>
<td>(kg N/ha)</td>
<td></td>
</tr>
<tr>
<td>(6) Available phosphorus</td>
<td></td>
</tr>
<tr>
<td>(kg P₂O₅/ha)</td>
<td></td>
</tr>
<tr>
<td>(7) Available potassium</td>
<td></td>
</tr>
<tr>
<td>(kg K₂O/ha)</td>
<td></td>
</tr>
<tr>
<td>(8) Calcium carbonate</td>
<td></td>
</tr>
</tbody>
</table>
3.4 **Seeds**

Authentic seeds of groundnut will be obtained from Gujarat Beej Nijam, Junagadh. In these studies two varieties will be included.

1. **GAUG - 1** - Small seeded bunch type maturing in 105-110 days.
2. **GAUG - 10** - Bold seeded spreading type maturing in 130-135 days.

The five experiments will be conducted in three seasons two in "Kharif", two in "Rabi" and one in summer. In all experiments, varieties, levels of nitrogen and phosphorus, sources of fertilizers, number of replicates, size of plots and seed rate will be kept uniform.

3.5 **Creation of drought**

Under induced drought conditions, irrigation will be given at 20 days intervals (Experiments 3 and 4) and also after 15 days (Experiment 5).

3.6 **"Kharif" trials**

Two experiments will be conducted in this season. One experiment in one year and the other in the consecutive year. The crop will be completely rainfed (natural drought)
in these trials. The design of each experiment will be factorial randomised block.

3.6.1 **Experiment 1**

The experiment will be laid out according to factorial randomised block design. The object of this experiment will be to find out the optimum potassium requirement of two varieties of groundnut (GAUQ - 1 and GAUQ - 10) under rainfed conditions (natural drought 1). Potassium will be applied at the rate of 0, 26, 52 and 78 kg K\(_2\)O/ha. In addition, a uniform basal doses of 12 kg N and 25 kg P\(_2\)O\(_5\)/ha will be applied to all plots at the time of sowing as per the recommendation of Gujarat Agricultural University, Junagadh. The sources of potassium, nitrogen and phosphorus will be muriate of potash, urea and superphosphate respectively.

The size of each plot will be sixteen sq m. The seed rate for GAUQ - 1 will be 90 kg/ha and for GAUQ - 10, 50 kg/ha. Each treatment will be replicated three times. The summary of the experiment is given in Table 2.
Table 2. Summary of proposed treatment (Experiment 1)

<table>
<thead>
<tr>
<th>Treatments (kg K₂O/ha)</th>
<th>Varieties</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>V₁</td>
<td>V₂</td>
</tr>
<tr>
<td>K₀</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>K₂₆</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>K₅₂</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>K₇₈</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

The following parameters will be studied in all experiments at 30, 60 and 90 days after sowing.

**Morphological parameters**
1. Plant height
2. Leaf number
3. Leaf area

**Physiological parameters**
1. Crop growth rate (CGR)
2. Leaf area index (LAI)
3. Leaf area duration (LAD)
4. Leaf temperature (LT)
5. Diffusive resistance (DR)
6. Transpiration rate (TR)
7. Relative water content (RWC)
8. Solar radiation interception (SI & EIR)
Chemical and biochemical parameters

1. Chlorophyll content
2. Proline content
3. Nitrate reductase activity
4. NPK contents (in leaf, stem, root, shell and seed)

Yield characteristics

1. Number of pegs per plant
2. Number of pods per plant mature and immature
3. Peg/pod ratio
4. Mature/im mature pod ratio
5. Pod yield g/ha
6. Seed yield g/ha
7. Shelling per cent
8. 100 seed weight
9. Harvest index
10. Oil content
11. Protein content
12. Sugar content
13. Oil yield q/ha

3.6.2 Experiment 2

This experiment will be conducted in the next consecutive year in "Kharif" season.
The aim of the experiment will be to find out the growth stage of groundnut varieties at which potassium requirement is maximum. Potassium will be applied at the rate of 0, 26, 52 and 78 kg K$_2$O/ha in split doses, half at the time of sowing and half at pod development stage. The summary of the experiment is given in the Table 3. All the attributes mentioned in Experiment 1 will be observed.

Table 3. Summary of the proposed treatments (Experiment 2)

<table>
<thead>
<tr>
<th>Treatments (kg K$_2$O/ha)</th>
<th>Varieties</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>K$_0$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>K$_{B13 + T13}$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>K$_{B26 + T26}$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>K$_{B39 + T39}$</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>
3.7 *Rabi* trials

The *Rabi* trials will include two experiments (Experiment 3 and 4), one following Experiment 1 of *Kharif* season and the other one (Experiment 4) following the Experiment 2 of *Kharif* season. These experiments will be conducted under induced drought conditions by irrigating the crop at two sets of intervals i.e. 10 days interval (normal irrigation) and 20 days interval (50% cut in irrigation). These schedules of irrigation will be followed in both experiments. The design of the experiments will be split plot.

3.7.1 **Experiment 3**

The aim of this experiment will be to study the effect of same levels of potassium (Experiment 1) under induced drought conditions. The summary of the experiment is given in Table 4.

3.7.2 **Experiment 4**

This experiment will be conducted in the *Rabi* season in the next year as Experiment 2. The aim of this experiment will be to find out the growth stage of *groundnut* at which the potassium requirement is maximum under induced drought condition. Potassium will be applied in split doses as in
Table 4: Summary of the proposed treatments (Experiment 3)

<table>
<thead>
<tr>
<th>Sub-plot</th>
<th>Main plot</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>$K_0I_1$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_0I_2$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_1I_1$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_1I_2$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_2I_1$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_2I_2$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_3I_1$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_3I_2$</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>

Experiment 2. All the attributes of the Experiment 1 will be observed. The summary of the experiment is given in Table 5.
Table 5: Summary of the proposed treatment (Experiment 4)

<table>
<thead>
<tr>
<th>Sub plot</th>
<th>Main plot</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>$K_0I_1$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_0I_2$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_{213} + T_{13}I_1$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_{213} + T_{13}I_2$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_{226} + T_{26}I_1$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_{226} + T_{26}I_2$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_{339} + T_{39}I_1$</td>
<td>+</td>
<td>+</td>
</tr>
<tr>
<td>$K_{339} + T_{39}I_2$</td>
<td>+</td>
<td>+</td>
</tr>
</tbody>
</table>
3.8 **Summer trial**

In this season only one experiment will be conducted. The main aim of this experiment will be to study the effect of potassium nutrition at different irrigation intervals during the season. In this study only one variety GAUC - 1 (bunch type) will be taken.

3.8.1 **Experiment 5**

The experiment will be laid out according to factorial randomised block design. Potassium will be applied at the rate of 0, 26 or 52 kg K₂O/ha at the time of sowing. Water stress will be created by irrigating the plot at an interval of 15 days (25% depletion) or 20 days (50% depletion). The control will be irrigated every 10 days (normal). All the parameters of Experiment 1 will be observed in this study. The summary of the experiment is given in Table 6.

3.9 **Sampling technique**

Random plant samples will be taken in triplicate from each plot at all experiments, 30, 60 and 70 days after sowing and at harvest.
Table 6: Summary of the proposed treatments (Experiment 3)

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Variety $V_1$</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>$K_0I_1$</td>
<td>+</td>
<td>No potash and 10 days irrigation</td>
</tr>
<tr>
<td>$K_0I_2$</td>
<td>+</td>
<td>No potash and 15 days irrigation</td>
</tr>
<tr>
<td>$K_0I_3$</td>
<td>+</td>
<td>No potash and 20 days irrigation</td>
</tr>
<tr>
<td>$K_1I_1$</td>
<td>+</td>
<td>26 kg $K_2O$/ha and 10 days irrigation</td>
</tr>
<tr>
<td>$K_1I_2$</td>
<td>+</td>
<td>26 kg $K_2O$/ha and 15 days irrigation</td>
</tr>
<tr>
<td>$K_1I_3$</td>
<td>+</td>
<td>26 kg $K_2O$/ha and 20 days irrigation</td>
</tr>
<tr>
<td>$K_2I_1$</td>
<td>+</td>
<td>52 kg $K_2O$/ha and 10 days irrigation</td>
</tr>
<tr>
<td>$K_2I_2$</td>
<td>+</td>
<td>52 kg $K_2O$/ha and 15 days irrigation</td>
</tr>
<tr>
<td>$K_2I_3$</td>
<td>+</td>
<td>52 kg $K_2O$/ha and 20 days irrigation</td>
</tr>
</tbody>
</table>
3.10 Growth and yield parameters

The following growth characters will be studied in each sample:

3.10.1 Plant height

Plant height will be measured from the base to the tip of the uppermost leaf.

3.10.2 Leaf number

Number of leaves per plant will be counted.

3.10.3 Leaf area

Leaf area will be measured by using LI-3100 AREA METER and the total area of all the leaves of a single plant will be obtained by adding the readings for individual leaves.

3.10.4 Leaf area index (LAI)

Leaf area index will be determined by using the following formula suggested by Watson (1947):

\[
\text{LAI} = \frac{\text{Leaf area per plant}}{\text{Area occupied per plant}}
\]

3.10.5 Leaf area duration (LAD)

LAD will be determined by using another formula also suggested by Watson (1947):
$\text{LAD} = \frac{(L_1 + L_2)(T_2 - T_1)}{2}$

Where: $L_1 = \text{LAI at time } T_1$
$L_2 = \text{LAI at time } T_2$

### 3.10.6 Crop growth rate (CGR)

CGR will be calculated by using the formula:

$$\text{CGR} = \frac{W_2 - W_1}{T_2 - T_1} \quad (\text{g m}^2 \text{ day}^{-1})$$

Where, $W_1$ and $W_2$ are the biomass per sq m, at time $T_1$ and $T_2$ respectively.

### 3.10.7 Biomass production

The total biomass production of each cultivar will be the cumulative effect of biomass of leaf, stem, petiole, root, peg and pod and will be computed accordingly.

### 3.10.8 Energy harvest efficiency

Energy conservation will be determined after harvesting the plants and subjecting them to energy estimation. Dried powdered samples will be pressed into pellets. The combustible value of each part of the plant in terms of energy, will be determined in triplicate samples with the help of a "PAR-OXYGEN 3042 CALORIMETER". The energy values will be expressed as Cal g$^{-1}$ dry matter. To determine energy conservation, the energy values of different part will
be multiplied by total dry weight of the respective parts. For simplicity all the recorded values of SI in the form of \( w \text{ m}^2 \) and PAR in the form of \( K \text{ m}^2 \) for a period of 170 days will be converted into K Cal \( \text{m}^2 \) 120 days. The energy conserving efficiency will be calculated on total PAR and SI basis.

The energy conserved (K Cal \( \text{m}^2 \)) by the plant will be divided by PAR or SI (K Cal \( \text{m}^2 \)) and the result will be multiplied by 100 to get energy efficiency. Thus,

\[
\text{Efficiency} \% = \frac{\text{Energy conserved by plant (area/time)}}{\text{Solar radiation (Area/time)}} \times 100
\]

3.10.9 Yield analysis Plants will be harvested when the majority of the pods have matured. The per plot yield will be recorded. The weight of air dried mature pods will be recorded. The pods will be shelled by hand and the kernels will be separated into fully filled (mature) and shrivelled (immature) kernels. They will be counted and weighed.

3.10.10 Harvest index (HI) = \( \frac{\text{Biomass of pods}}{\text{Total biomass}} \times 100 \)

3.11 Physiological, chemical and biochemical parameters

The following parameters will be studied using standard procedure briefly described for each.
3.11.1 Relative water content (RWC)  
RWC will be determined by the method of Barras and Weatherly (1962). Turgid weight will be determined by soaking the leaf discs in petridishes containing water for 3 hours and will be calculated by the following formula:

\[
RWC = \frac{W_f - W_d}{W_t - W_d} \times 100
\]

Where:  
\( W_f \) = Fresh weight  
\( W_d \) = Oven dried weight  
\( W_t \) = Fully turgid weight.

3.11.2 Leaf temperature, diffusive resistance and transpiration

Leaf temperature, stomatal resistance and transpiration rate will be measured by using the LI-1600 steady state porometer.

3.11.3 Solar radiation (SI & PAR) perception by leaves

Integrated photosynthetically-active solar radiation (PAR) and total solar irradiation (SI) for a day, and thereby for the whole growth period, will be measured with the help of LICOR SOLAR MONITOR (Spectroradiometer). These observations will be recorded from early seedling stage till the maturity of crop.
3.11.4 Nitrate reductase activity (NRA)

NRA will be estimated by the intact tissue assay method of Jaworski (1971), which is based on the reduction of nitrate to nitrite. This nitrite will be determined colorimetrically by the Griess Illsosvay method (Snell and Snell, 1949). The following reagents will be used:

Reagents

A. 0.1 M phosphate buffer: This will be prepared by dissolving 27.2g/l KH₂PO₄ and 45.63 g/l K₂HPO₄·7H₂O. To achieve pH 7.4, 16 ml of KH₂PO₄ and 84 ml of K₂HPO₄ solution will be taken and diluted to 200 ml with distilled water.

B. 0.2 M KNO₃: It will be made by dissolving 20.2g/potassium nitrate in one litre of water.

C. 5% Isopropanol: 5 ml isopropanol will be diluted with distilled water to 100 ml.

D. 0.5% Chloramphenicol: 50 mg chloramphenicol will be dissolved in a little quantity of distilled water and the final volume made upto 100 ml with distilled water.

E. 1% Sulphanilamide in 3N HCl: 1g sulphanilamide will be dissolved in 3N HCl (1 ml HCl + 4 ml water).

F. 0.02% N-1 Naphthyl ethylene diamine di-hydrochloride: 20mg N-1 Naphthyl ethylene diamine di-hydrochloride will be dissolved in water and the final volume made upto 100 ml.
Procedure: 250 mg of leaf punches will be suspended in screw capped vials containing 2.5 ml of A, 0.5 ml of B, 2.5 ml of C and 2 drops of D. After sealing, the vials will be incubated at 30°C in the dark for about 7 hours. NRA in the medium will be determined by taking 0.4 ml incubated solution and 0.3 ml of each of reagents E and F. After 20 minutes, the solution will be diluted with 4 ml of water to make the volume upto 5 ml and optical density will be measured at 540 nm. If nitrite concentration found to be too high, it will be further diluted.

Standard for NRA: Keeping in mind that 1.5 mg NaNO₃/100 ml gives 10 μg NO₂/ml, 0, 2.5, 5, 7.5, 10 ..... 20.0 μg NO₂ will be taken and 0.3 ml each of reagents E and F as above will be added. After 20 minutes, the solutions will be diluted with distilled water to make the volume upto 5 ml and their optical density measured at 540 nm and plotted on a graph paper to obtain a standard curve. The amount of nitrite produced by the activity of nitrate reductase in the assay will be estimated with the help of this standard curve. The amount of nitrite formed as a result/reduction of nitrate will be used as an index of the activity of the enzyme.

A standard curve will be plotted by taking various concentrations of potassium nitrite. The optical density
of the samples will be compared with this calibrated curve and NRA expressed as n mol NO$_2^-$/h g fresh leaf tissue.

3.11.5 Proline content: It will be determined in leaves according to the method of Bates et al. (1973). The following reagents will be used:

Reagents
Acid ninhydrin: It will be prepared by warming 1.75 g ninhydrin in 30 ml glacial acetic acid and 20 ml 6M orthophosphoric acid (407 ml/l) with agitation until dissolved. It will be stored at 4°C, being stable for 24 hr.

Procedure:
(1) 100 mg to 200 mg (depending on availability) of plant material will be homogenised in 10 ml of 3% sulphosalicylic acid and the homogenate will be filtered through a Whatman No.1 filter paper.

(2) 5 ml filtrate will be reacted with 2 ml acid ninhydrin and 2 ml glacial acetic acid in a test tube for 1 hour at 100°C in a water bath and the reaction will be terminated in ice box.

(3) The reaction mixture will be extracted in 5-10 ml or more of toluene after mixing vigorously with a test tube stirrer for 15-20 second.
(4) The chromatophore containing toluene will be aspirated from the aqueous phase. The absorbance will be read at 520 nm at room temperature, using toluene that has reacted with the reagents without the sample for blank.

(5) The proline concentration will determined from a standard curve prepared by taking graded concentrations (0, 10, 20, 30, 40, 50, 60, 70 and 80 ug), of proline and it will be calculated on fresh or dry weight basis in leaves.

3.11.6 Estimation of chlorophyll

For estimation of chlorophyll, the method of Arnon (1949) will be followed.

Procedure:

100 mg of freshly cut leaf material (from the uppermost 3 leaf blades) will be grinded in a mortar and pestle with 80% acetone. The extract will be filtered through Whatman No.1 filter paper. The washing of the extract will be done using 80% acetone. The solution will be made upto 50 ml. Optical density readings will be taken at 645 nm and 663 in a spectrophotometer.
The amounts of chlorophyll a, chlorophyll b, and total chlorophyll will be calculated using the following formulae:

\[
\text{Chl. } a = 12.7 \times (D_{663}) - 2.67 \times (D_{645}) \text{ mg Chl/litre.}
\]

\[
\text{Chl. } b = 22.9 \times (D_{645}) - 4.68 \times (D_{663}) \text{ mg Chl/litre.}
\]

\[
\text{Total chlorophyll} = 20.2 \times (D_{645}) + 0.02 \times (D_{663}) \text{ mg Chl/litre}
\]

Where \( D \) represents the optical density reading. The chlorophyll content will be finally expressed as mg chlorophyll per gram fresh weight.

3.11.7 **Plant analysis for N, P, and K**

Random plant samples will be taken in triplicate. Different plant parts will be separated from each other and will be dried in an oven for 24 hours. The samples of leaf, stem, root, kernel, and shell will be made into fine powder, using a 72 mesh screen. The powder thus obtained will be kept at 70°C overnight before digestion and analysis. The nitrogen, phosphorus, and potassium content will be estimated using standard methods.

3.11.8 **Digestion of samples**

100 mg of dry leaf powder will be taken in a 50 ml kjeldahl flask. 2 ml of chemically pure sulphuric acid will be added and the flask heated for about two hours to dissolve
the powder. This heating with acid will turn the contents black. After cooling the flask for about 15 minutes, 0.5 ml of chemically pure 30 per cent hydrogen peroxide will be added dropwise. The solution will now be heated again for about 15 minutes till the colour turns light yellow. It will be cooled and again 3--4 drops of hydrogen peroxide will be added, followed by heating for about 15 minutes to get clear extracts. Excess of hydrogen peroxide will be avoided as it would otherwise oxidise the ammonia in the absence of organic matter. The peroxide-digested material will be transferred to a 100 ml volumetric flask with three or four washings with double distilled water and the volume will be made up to the mark. This will serve as a stock solution for the estimation of nitrogen, phosphorus and potassium.

3.11.9 Estimation of Nitrogen

The estimation of nitrogen in the sample will be made according to the method given by Lindner (1944).

A 10 ml aliquot of the peroxide digested material will be transferred to a 50 ml volumetric flask. 7 ml of 2.5 N sodium hydroxide will be added to neutralise the excess of the acid partially. To prevent turbidity, 1 ml of 10 per cent sodium silicate will be added to the flask and the
volume will be made up to the mark. In a 10 ml graduated test tube, a 5 ml aliquot of this solution will be taken and 0.5 ml of Nessler's reagent added and mixed thoroughly. The final volume will be made up with double distilled water and the tube kept for about 5 minutes for maximum colour development. This solution will be taken in a colorimetric tube and its optical density measured at 525 nm. A blank will also be run simultaneously. A standard curve of known dilutions of ammonium sulphate solution will be plotted. The reading of each sample will be compared with this calibration curve to determine the quantity of nitrogen present in each sample.

3.11.10 Estimation of phosphorus

Phosphorus will be estimated by the method of Fiske and Subba Rao (1925). In a 10 ml graduated tube, a 5 ml aliquot will be taken and 1 ml of molybdate reagent (2.5% ammonium molybdate in 10 NH₂SO₄) will be added carefully, followed by addition of 0.4 ml 1:2:4 - amino-nephthol sulphonic acid. This will turn the contents blue. The volume will be made up and the solution will be allowed to stand for about 5 minutes for maximum colour development. Then, it will be transferred to a colorimetric tube and the optical density read at 620 nm. A blank will be run for each
determination. A calibration curve will be prepared by using known dilutions of a standard monobasic potassium phosphate solution.

3.11.11 Estimation of potassium

Potassium will be estimated using a flame photometer. A blank will be run side by side. The readings will be compared with a calibration curve plotted for different dilutions of a standard potassium sulphate solution.

3.11.12 Estimation of crude protein

The protein content will be estimated by multiplying the nitrogen content with 6.25 for leaf, stem and other plant parts and with 5.46 for seeds which is the protein factor for groundnut (Jones, 1931).

3.11.13 Soluble sugars

Soluble sugars will be extracted as per the method of Dubois et al. (1956) and estimated by the procedure given in AOAC (1965).

Reagents: Anthrone reagent

A. 0.2 per cent anthrone in concentrated $\text{H}_2\text{SO}_4$.

B. Standard glucose solution (50 $\mu$g/ml).
**Procedure**

For extraction 100 mg of finely powdered material will be taken in a 100 ml conical flask containing 40 ml of double distilled water. The contents will be shaken and autoclaved at 15 psi for 4 hrs. After cooling, the supernatant will be filtered in a 100 ml volumetric flask through non-absorbent cotton with 4 washings with distilled water and volume made up to 100 ml.

To estimate the quantity of soluble sugars in each sample, a suitable aliquot of the respective extract will be taken and final volume made up to 1.0 ml with distilled water. To it, 5 ml of anthrone reagent will be added and heated in a boiling water bath for 10 minutes. After cooling, absorbance will be read at 620 nm. Sugar content of the samples will be determined by using a standard curve prepared for glucose (10-50 ug) and results will be expressed as mg/gm dry wt. of tissue.

3.11.14

**Oil content**  
Oil in the plant material will be extracted with petroleum ether (40-60°C) and estimated gravimetrically (Nicholas,1968).

**Procedure** (a) Extraction: 10 grams of the plant tissue will be taken in tube and the lipids will be extracted using
a soxhlet apparatus at 50-60°C temperature for 8-10 hours till all the oil is extracted from the groundnut powder. After the complete evaporation of petroleum ether, the lipid or oil extract will immediately be used for further study.

(b) Total oil content: The amount of oil present in the tissue will be determined gravimetrically. The oil extract will be taken in pre-weighted porcelain crucible and dried in a vacuum oven at 35-40°C to a constant weight and oil content will be expressed on per cent basis.

3.12 Statistical analysis

All data will be analysed statistically according to the design of the experiment. The results of the experiments will be discussed in the light of relevant literature published by other workers. Correlations will be worked out to establish the association of various physio-morphological parameters with seed yield, oil yield and protein content and yield in each groundnut variety. If established, these correlation studies could be utilised to predict yield and quality of the seed of this crop. In case of adverse predictions, this information could help in taking timely corrective measures at an early stage as suggested for other crops (Mitra and Wasiuddin, 1979; Akhtar et al., 1984; Afzal et al., 1985; Ansari et al., 1985, Saniuallah et al., 1985).
REFERENCES
REFERENCES


PIKKE, C.H., and SUBBA ROW, Y. 1925. The colorimetric deter-

FORESTIER, J. 1969. Development of the erect groundnut in
the forest region. Catiers Caborston, Biologic, 9: 33-63.


und chlorophyllgehalt des Fahnblattes und auf die
Kornentragkomponenten von Sommer - weizen -
Untersuchungen an fun versdhiedenen sommerweizensorten
Zeitschrift fur Ackerund Pflanzenbau 143, 169-179.
(Cited from Steineck, and Haeder, 1975).

"Brunisite, de la tume e minerale et de trempeage
des grains. Oleagineux 13: 395-407 (Cited from
Sastry, 1979).

GAASTRA, P. 1958. Light energy conversion in field crops in
comparison with the photosynthetic efficiency under
laboratory conditions. Vrediel, Landbouwhoogsch.
Wageningen 58: 1-12.


GUTSTEIN, Y. 1979. Response of groundnuts to K fertilisation 
and its absorption pattern from the soil profiles. 
Soil and fertilizers 42 : 5.

HAEDER, H.E. and MENGEN, K. 1975. Einfluss der Lichtintensitat 
bei varietter kaliumernahrung auf CO₂ 
Assimilation und Ertragshildung bei sommerweizen. 
Zeitschrift fur Pflanzen ernahrung und Bodenkunde 
133 : 573-582 (Cited from Steineck and Haeder, 
1979).

HALL, D.O. 1977. Solar energy and biology for fuel food and 

HARRIS, C. and BLEDSOE, R.W. 1951. Nutrition and physiology 

HERNANDO, V., GRIMUEL, B. 1977. Importance of interaction of 
K X Irrigation on tomato crops. Proceedings of the 
international seminar on soil environment and 
 fertility management in intensive agriculture. 


SASTRY K.S. 1979. Scheme for drought tolerance studies on groundnut castor and safflower. Published by Department of Crop. Physiology, University of Agricultural Sciences, Bangalore.


STOY, V. 1962. Dry matter production as an objective in plant breeding. 3rd congress of the Association for research on plant breeding Eucarpia, Paris, pp. 32-47.


