CHAPTER 2

REVIEW OF LITERATURE
2.1 Potassium

2.1.1 Potassium consumption trend in India and the world during recent years

Global consumption of K has increased at an average rate of 4.4% per annum over the period 1999-2005 (IFA, 2005). Global K reserves are likely to be sufficient for hundreds or even thousands of years of agriculture (Sheldrick, 1985). The entire requirement of potash continued to be met through imports as we do not have commercially exploitable source of potash in our country. The quantum of import of MOP increased from 3.4 million tonnes during 2004-05 to 4.4 million tonnes during 2007-08. According to a report of Potash and Phosphate Institute, Canada (PPIC) (Hasan and Tiwari, 2002) on available potassium in Indian soils, out of 371 districts surveyed, soils were categorized to be of low K status in 76 districts, with medium K fertility in 190 districts and with high K fertility in 105 districts. A soil is generally categorized as low in K fertility if it contains less than 120 kg K (144 kg K$_2$O)/ha, which can be extracted by 1N ammonium acetate solution, and high in K fertility if the extracted K is more than 280 kg K (336 kg K$_2$O)/ha.

Historically the use of K fertilizers has been limited in many countries with extensive agricultural production, with crops and pastures relying on native soil reserves of K. Presently, many Asian countries indicated a negative K balance in farming system with many crops. Leaching of K, especially in sandy soils (Kayser and Isselstein, 2005), is also a significant contributor to poor K-use efficiency in farming systems. Root systems of annual crops generally occupy <1% of the soil volume (Barber, 1995), allowing roots to get in direct contact with only a tiny proportion of soil K. Therefore uptake by plants is highly dependent on the soil capacity to deliver K to the root surface by mass flow and diffusion, the later being the predominant process (Jungk, 2001; Jungk and Claassen, 1997; Liebersbach et al., 2004). The rates of diffusion are influenced by the plant capacity to take up K at the root surface and create a diffusive gradient from the bulk soil towards the root surface.

2.1.2 Importance of potassium in crops

Potassium (K) is essential for plant and animal life wherein it has many vital nutritional roles. In plants, potassium and nitrogen are the two elements required in
greatest amounts, while in animals and humans, potassium is the third most abundant element after calcium and phosphorus. Potash, the fertilizer trade term referring to fertilizer materials containing potassium, has become an increasingly important input for satisfying demands of an expanding population for food, fibre and other commodities. Potassium is the seventh most abundant element in earth’s crust (Sparks and Huang, 1985). In most plants, after carbon, hydrogen, oxygen and nitrogen, potassium is the most abundant element. The quantity of potassium absorbed by roots is second only to that of nitrogen for most cultivated plants, while the demand for potassium is higher for certain crops. Potassium promotes root water absorption and prevents unnecessary water loss by transpiration.

2.1.3 Availability of potassium in soils

Many soils lack sufficient quantities of available potassium for satisfactory yield and quality of crops. For this reason available soil potassium levels are commonly supplemented by potash fertilization to improve the potassium nutrition of plants, particularly for sustaining production of high yielding crop species and varieties in modern agricultural systems. Soil K can be assigned to four distinct pools differing in availability to plants: (1) soil solution K, (2) exchangeable K, (3) non exchangeable K (Positioned in interlayers of clay minerals, especially those of the 2:1:1 type) and (4) structural K (Moody and Bell, 2006). Plants take up K exclusively from the soil solution pool, which is in a dynamic equilibrium with the exchangeable and, to a lesser extent, the non exchangeable pools. Exchangeable K can be rapidly released from exchange sites on the surface of clay minerals and organic matter to replenish K depleted soil solution (Steingrobe and Classen, 2000). However, a release of non-exchangeable K from interlayer sites of clay minerals is a slow process and is mostly important in contributing to the replenishment of the soil solution and exchangeable pools in the long term (e.g. over successive crops; Pal et al., 2001a, 2002). The release of structural K into soil solution can be affected only by weathering of clay materials; hence it is an exceedingly slow process with no discernible effect during a single crop cycle (Pal et al., 2001b). Hence in soil with significant K clay fraction and therefore a potentially extensive non exchangeable K pool, the capacity of a genotype influence the dynamics of K release from that pool to the soil solution can influence the efficiency of K uptake (El-Dessougi et al., 2002; Trehan, 2005).
2.1.4 Potassium deficiency in crops

Potassium deficiency is recognized as an important limiting factor in crop production. At its early stages, potassium deficiency is reflected mostly in yield decreases. This early stage is called "hidden hunger" since no specific symptoms appear in the plant. As the intensity of the deficiency increases, significant symptoms appear: less strength of plant structures, less resistance to low water availability and less resistance to fungus diseases. Generally, optimal plant tissue K is in the range of 2-5% based on plant dry weight (Marshner, 1995). In addition, throughout the entire life cycle of most plants, this nutrient plays an important role in many physiological plant functions. Under severe potassium deficiency, lignifications of vascular bundles are reduced contributing to high lodging (Marshner, 1995).

One of the more visually obvious consequences on plant growth from insufficient levels of plant potassium is a reduction in plant stature (Cassman et al., 1989; Ebelhar and Varsa, 2000; Heckman and Kamprath, 1992; Mullins et al., 1994; Pettigrew and Meredith, 1997). This reduction in biomass because of K⁺ deficiency is often accompanied by a reduction in leaf area (Jordon-Meile and Pellerin, 2004; Kimbrough et al., 1971; Pettigrew and Meredith, 1997). Insufficient K⁺ levels reduced leaf area expansion leading to reduced leaf size in soybean (Huber, 1985) and maize (Jordon-Meile and Pellerin, 2004). Less leaf area may lead to an increased concentration of cellular component carbohydrates and/or nutrients over a given unit of leaf area compared with adequate K⁺ levels (Pettigrew, 2008). This combination of less leaf area, less solar radiation interception and reduced photosynthesis per unit leaf area under insufficient K⁺ levels leads to a reduction in the total photosynthetic assimilate pool produced in the plant's source tissue (leaves). Coupling this reduced photo-assimilate production with restricted assimilate transport from the leaves result in a smaller total assimilate supply available for the sink tissue for K⁺ deficient plants (Fig.1). This reduced assimilate supply under a K⁺ deficiency will ultimately diminish the yield and quality produced by those plants (Pettigrew, 2008).

2.1.5 Physiological role of potassium in plants

The total amount of potassium absorbed by the crop during the growing season depend upon the crop species being grown, the amount of native soil K⁺, the amount of fertilizer K⁺ applied, K availability in the soil, the environmental conditions during the growing season and the management practices employed (Eakin, 1972; Mengel and
Fig: Schematic diagram demonstrating the effect K has on various physiological processes and connecting those effected processes with yield and quality development (K-sufficient vs K deficient plants)
Kirkby, 1987; Mullins and Burmester, 1998). The actual amount removed from the field by a crop species depend upon the plant part or parts removed during harvest.

Once inside the plant, K⁺ is involved with many physiological processes. Potassium has been classified as the principal inorganic solute having a key role in osmoregulation (Hsiao and Lauchli, 1986). In addition, K is important in regulating more than 60 enzymes that catalyze a great number of metabolic activities (Suelter, 1970) maintaining charge balance by counterbalancing the negatively charged ions present in the cytoplasm, controlling phloem transport, increasing crop quality, increasing disease resistance and improving overall growth, development and yield (Marschner, 1995).

Pioneering work by Fischer (1968) and Fischer and Hsiao (1968) demonstrated how the reversible K⁺ flux into and out of stomatal guard cells controlled stomatal aperture by affecting osmotic potential of the guard cells, with malate and Cl⁻ serving as major counter ions (Talbott et al., 1998). Guard cell uptake is mediated by K⁺ specific uptake channels and is coupled with proton extrusion into the apoplast (Hoth et al., 1997). Stomatal opening during the course of the day is thought to be a two phase process with K⁺ promoting opening early in the day and then giving way to sucrose as the principle driving osmotic force around mid day (Tabott and Zeiger, 1996). Because of this close coordination between K⁺ guard cell concentration and stomatal aperture, insufficient leaf levels of K⁺ can lead to decreased stomatal conductance (Bednarz et al., 1998; Longstreth and Nobel, 1980). As expected with this decrease in stomatal conductance, insufficient K⁺ levels also lead to decreased photosynthesis per unit leaf area (Bednarz et al., 1998; Longstreth and Nobel, 1980; Pier and Berkowitz, 1987; Wolf et al., 1976). However, this decreased stomatal conductance only partially accounts for the photosynthetic decline observed with lower K⁺ levels. Non stomatal factors also contribute to this reduction in photosynthesis, particularly when the deficiency becomes extreme (Basile et al., 2003; Bednarz et al., 1998; Huber, 1985; Tester and Blatt, 1989). Bednarz et al. (1998) reported that the onset of a developing potassium deficiency was the principal factor limiting photosynthesis, whereas when the potassium deficiency became more extreme, non stomatal or biochemical factors became the overriding reason for the decreased photosynthesis.

In addition to the reduced stomatal conductance and photosynthesis observed under K⁺ deficient conditions, the transport of photosynthetic assimilates away from source tissue via the phloem is also restricted (Ashley and Goodson, 1972; Mengel and Haeder, 1977; Mengel and Viro, 1974). This restriction on the transport of
photosynthesized can lead to accumulation of sugars in the leaf tissue of K⁺ deficient plants (Bednarz and Oosterhuis, 1999; Huber, 1985; Pettigrew, 1999). This accumulation of sugars undoubtedly contributes small proportion to the increased specific leaf weights observed when cotton plants were grown under conditions of low soil potassium (Pettigrew, 1999; Pettigrew and Meredith, 1997).

Potassium is also involved directly or indirectly in plant protein metabolism (Blevins, 1985). This involvement can begin with the stimulation of NO₃⁻ uptake and transport within the plant, as K⁺ serves as the accompanying counter cation (Blevins et al., 1978a, 1978b). Furthermore Mengel (1980) also demonstrated that the transport of amino acids is enhanced by higher K⁺ levels, especially the transport of amino acids to developing seeds. Potassium involvement is crucial for most of the steps of protein synthesis process, beginning with enzyme activation and continuing through ribosome synthesis and mRNA turnover (Blevins, 1985; Evans and Wildes, 1971). Reinforcing the connection between K⁺ levels and protein is the observation that crops with high seed protein concentrations also tend to have high K⁺ indices (amount of K⁺ harvest in grain/total K⁺ in grain and stover (Blevins, 1985).

Potassium is known to be taken up by plant roots at high rates and is quickly transported to upper plant parts. The downward transport of K⁺ from tops to roots is also a rapid process, so that once K⁺ absorbed, it is rapidly distributed throughout the entire plant. Long distance transport of K⁺ has a direct impact on water transport and is also related to the movement of assimilates in the phloem. This long distance transport is essentially dependent on K⁺ transport across plant membranes and it is also necessary for various physiological processes. Therefore, potassium plays an important role in the transport of assimilates and nutrients. The photosynthesis products (photosynthates) must be transported from the leaves (sources) to the site of their use or storage (sinks). Potassium promotes phloem transport of photosynthates mainly sucrose and amino acids to the physiological sinks (fruits, roots, tubers, seeds and grains) (Mengel, 1997). K plays a positive role in phloem loading with sucrose, in increasing the transport rate of phloem sap solutes and in phloem unloading (Herlihy, 1989). This role of K is related to its contribution to the osmotic potential in the sieve tubes and to its function in ATP synthesis which provides the energy for the loading of photosynthates. In plants well supplied with K, the concentration of potassium, the osmotic potential of the phloem sap and the volume flow rate, are all higher than in plants supplied with a lower K level. As a result, sucrose concentration in the phloem sap is increased (Marschner, 1995).
Potassium not only promotes the translocation of newly synthesized photosynthates but also has a beneficial effect on the mobilization of stored material (Mengel and Kirkby, 1987). Potassium plays also an important role as counter ion for nitrate transport in the xylem. After nitrate reduction in shoot, charge balance has to be maintained by corresponding net increase in organic acid anions. Part of these organic anions (mainly malate) can be re-translocated with K as the accompanying cation through the phloem to the roots (Marschner, 1995).

2.1.6 Effect of potassium on yield attributes

Integrating the effect of K⁺ on the above said physiological processes signifies that the K⁺ level can have profound effect on crop growth and development. The production of yield is sequential process. First the vegetative plant is produced (the factory to produce assimilate via photosynthesis), then the reproductive sink (yield container) is established during flowering and seed set and finally (Dennis and Bruening, 2003), seed number is primary yield component because it is determined first in the sequence. Its determination provides the first opportunity for the crop to adjust its potential yield to the level set by the environment. Seed size is determined later, during seed filling, after the initial adjustment in seed number, so its variation is naturally less important. There is critical period during crop development when seed number is determined, before and after the critical period; seed number is not directly affected by environmental conditions (Adams, 1967; Egli, 1998). Seed number in the most crops is a function of the rate of photosynthesis during this critical period. It seems clear that assimilate supply during the critical period for seed set is an important determinant of seed number. High levels of photosynthesis and large numbers of seed are characteristics of high yield environments, while the converse is true in low yield.

To achieve or maintain maximal maize yields, supplemental K⁺ fertilization is often required, particularly on soils testing low for native available soil K⁺. Many researchers have reported maize yield increases in response to K fertilization (Ebelhar and Varsa, 2000; Heckman and Kamprath, 1992; Mallarino et al., 1999). However, Burns and Ebelhar (2006) did not find improvement in grain yield on K fertilization, although they reported increased K⁺ tissue concentrations on K application. Soybean plants have also been found to respond positively to potassium fertilization. These potassium fertilization responses can also be achieved under a number of management regimes when the soil tests low for available K⁺. Soybean yield increases can be
obtained with K fertilization when grown under conventional tillage (Casanova, 2000; Heckman Kamprath, 1995; Jones et al., 1977), when the fertilizer was banded or broadcast (Borges and Mallarino, 2000; Buah et al., 2000; Yin and Vyn, 2002a, 2003, 2004), and sometimes when the K is applied in a foliar application (Hag and Mallarino, 2005; Nelson et al., 2005). The positive yield response to K can be attributed to increase in most of the yield components. The number of pods per plant (Bharati et al., 1986; Jones et al., 1977; Nelson et al., 1945) and the weight of individual seeds (Bharati et al., 1986) increased in response to K fertilization. Coale and Grove (1990) found that increased soybean yield under high K affinity was because of increased production of both total and main stem pods per plant and more seeds per pod. They did not find an increase in seed size in response to K as Bharati et al. (1986) reported. There are also reports of wheat responding to K fertilization when soil tested high for exchangeable K (Fixen et al., 1986; Sweeney et al., 2000).

As the proportion of total plant weight that is economic yield, harvest index quantifies the capacity of genotype to allocate resources, including K to the harvested organs. A high harvest index is fundamental to efficient utilization of all resources taken up by the plant and is therefore of significant interest to breeders. The dramatic yield improvements for many crops that have occurred during the 20th century have been largely because of improvements in harvest index (Hay, 1995).

The importance of high harvest index as a mechanism of K utilization efficiency has been widely documented for a number of species including wheat (Damon and Rengel, 2007; Woodend and Glass, 1993; Zhang et al., 1999), rice (Yang et al., 2004), sweet potato (George et al., 2002) and common bean (Fageria et al., 2001).

Harvest index has such a dominant influence on K efficiency for some species so that vegetative measures for K efficiency can be poorly correlated with K efficiency for grain yield. Potassium utilization efficiency of sweet potato genotypes was positively related to harvest index (George et al., 2002). Similarly in rice, K-efficient rice genotypes had higher harvest indices than inefficient ones when grown at low K (Yang et al., 2004). Harvest index was positively correlated with grain yield and biomass per unit time of K taken up. Grain filling rate was seven times lower during the second week of grain filling (but similar in the first week) for K inefficient rice genotypes compared with two K-efficient rice genotypes in K-deficient soil (Yang et al., 2004).
2.1.7 Effect of potassium on quality attributes

Potassium is often referred as the *quality element* for crop production (Usherwood, 1985). Potassium has been widely proven to have a crucial role in many crop quality parameters. Parameters for measuring the influence of K on crop quality reflect the specific functions of K in plant growth. For example, the influence of K on improved protein quality and quality in crop production is a reflection of the role of K in protein synthesis and N use efficiency. Supporting this high grain yield concept are the findings of Yang et al. (2004), that when maize was grown without manure in China, a greater grain protein content was produced using NPK fertilizers rather than using a fertilizer composed of only N and P. Quality seed and improved plant sustainability reflect the role of K in disease resistance and substrate translocation. Potassium also enhances storage and shipping quality of bananas, tomatoes, potatoes, onions and many other crops, and also extends their shelf life (Usherwood, 1985; Mengel, 1997).

2.2 Lentil

2.2.1 Growth habit of lentil

Lentil is an important cool season food legume. Its seeds provide high levels of protein and, when consumed in combination with cereals, they provide adequate amounts of essential amino acids for the human diet. Lentil production is beneficial for producers, as lentil has a high tolerance for extreme environmental conditions such as drought and hot temperatures, and can be grown in semi arid regions without irrigation. Moreover, the crop can be grown in rotation with cereal crops to reduce soil erosion, improve disease and weed control, and reduce demand for nitrogen fertilizers. Lentil is a self pollinating, quantitative long day or day neutral diploid, annual grain legume suitable for cultivation in warm temperate, subtropical and high altitude tropical regions of the world (Muehlbauer et al., 1995). It can grow on a wide range of soil types and soil pH and is more sensitive to water logging and soil pH<6.5 in comparison to a range of other legumes including pea, faba bean and chickpea (Tang and Thomson, 1996). Lentil is usually grown alone but can be intercropped with a range of species including wheat, barley, rice sugarcane, mustard etc. However, in comparison to other legumes, it is considered to be drought and low temperature tolerant (McKenzie and Hill, 1990).
2.2.2 Lentil production in India and the world

In 2004, total global lentil production was approximately $4 \times 10^6$ metric tonnes (FAOSTAT, 2007). Lentil is grown in many countries but production is dominated by India, Canada and Turkey with annual production of 61.4% of world production in 2006. A positive trend in lentil production is due to increased area rather than increasing yield. Lentil production in India has always been important as it is one of the most important *rabi* crops in the country. India has been producing lentil since 1st century AD and has always been an important producer of the crop. In fact, India was the largest producer of the lentil crop in the world until recently Canada took over the lead leaving India at the second place. Indian production of this crop hovers around 10 lakh metric tons per year that is cultivated on about 14 lakh hectares of land. India consumes almost 90-95% of its domestic produce to fulfill its domestic consumption demand as one of the most important constituent in the country’s staple diet. India also indulges in the exports of the left over lentil seeds and stands at fourth position in the major lentil exporting countries list with an export figure of 136,921 metric tons in the year 2004.

Lentil crop is grown in India in the winter season in various states. Uttar Pradesh accounts for the maximum production in the country contributing to around 45% of the country’s production as well as for the maximum area under lentil cultivation. Besides Bundelkhand region of U.P and M.P., Sagar, Jabalpur, and Bhopal in Madhya Pradesh, Kanpur in Uttar Pradesh and Kota in Rajasthan are the main districts where lentil is cultivated primarily (Tickoo et al., 2005).

<table>
<thead>
<tr>
<th>Year</th>
<th>Area (ha)</th>
<th>Production (tonnes)</th>
<th>Productivity (kg/ha)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1970-71</td>
<td>750 000</td>
<td>370 000</td>
<td>497</td>
</tr>
<tr>
<td>1980-81</td>
<td>930 000</td>
<td>470 000</td>
<td>607</td>
</tr>
<tr>
<td>1990-91</td>
<td>1190 000</td>
<td>850 000</td>
<td>717</td>
</tr>
<tr>
<td>2000-01</td>
<td>1480 000</td>
<td>920 000</td>
<td>619</td>
</tr>
<tr>
<td>2003-04</td>
<td>1450 000</td>
<td>1100 000</td>
<td>759</td>
</tr>
</tbody>
</table>

(Source: Tickoo et al., 2005)
Table 2.2: Area, production and productivity of lentil in major lentil growing states of India in 2001-2002.

<table>
<thead>
<tr>
<th>States</th>
<th>Area (ha)</th>
<th>Production (tonnes)</th>
<th>Yield (kg/ha)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Uttar Pradesh</td>
<td>580 000</td>
<td>460 000</td>
<td>793</td>
</tr>
<tr>
<td>Madhya Pradesh</td>
<td>480 000</td>
<td>230 000</td>
<td>479</td>
</tr>
<tr>
<td>Bihar</td>
<td>170 000</td>
<td>140 000</td>
<td>824</td>
</tr>
<tr>
<td>West Bengal</td>
<td>70 000</td>
<td>40 000</td>
<td>571</td>
</tr>
<tr>
<td>Rajasthan</td>
<td>20 000</td>
<td>20 000</td>
<td>1000</td>
</tr>
<tr>
<td>Assam</td>
<td>20 000</td>
<td>10 000</td>
<td>500</td>
</tr>
<tr>
<td>Others</td>
<td>130 000</td>
<td>70 000</td>
<td>538</td>
</tr>
<tr>
<td>All India</td>
<td>1470 000</td>
<td>970 000</td>
<td>664</td>
</tr>
</tbody>
</table>

(Source: Tickoo et al., 2005)

2.3 Nitrogen assimilation by plants

For most of the crop species, the plant life cycle can be roughly divided into: (i) the vegetative growth phase, when young developing roots and leaves behave as sink organs that efficiently absorb and assimilate inorganic nitrogen for amino acid and protein synthesis, and (ii) the remobilization phase when senescing tissues start to behave as source organs translocating organic molecules to ensure the formation of new developing and/or storage organs (Hirel and Lemaire, 2005). Therefore, during these two phases of plant growth and development, a better understanding of the metabolic and genetic control of nitrate uptake, nitrate partitioning between roots and shoots, nitrate reduction and its subsequent assimilation and transfer into organic molecules is required. A better knowledge of the regulatory mechanisms controlling these different metabolic processes and their relationships will allow selection of target plant criteria and physiological traits which can then be used as markers, both for breeding new genotypes or new cultivars exhibiting a better nitrogen use efficiency (NUE), and for rationalization of crop fertilization.

Nitrogen is often considered to be one of the most important factors limiting plant growth in natural ecosystems and in most agricultural soils. In modern agricultural systems, where plants rely on fertilizers to meet their demand for nitrogen, inadequate practices still cause environmental problems, mainly linked to nitrate loss in the
environment (Lawlor et al., 2001). Most plant species are able to take up and assimilate nitrate, ammonium, urea, and amino acids, but the response to a particular form of nitrogen varies from species to species (Forde and Clarkson, 1999). The major source of inorganic nitrogen available to plants is a mixture of nitrate and ammonium, with nitrate being the predominant form in a well-aerated soil as a consequence of bacterial nitrification (Marquez et al., 2005). The nitrate present in the soil solution is then taken up by roots and assimilated within plant cells by three sequential steps: (i) Transport across the plasma membranes, using a variety of high-affinity or low-affinity transport systems. (ii) Reduction of nitrate to nitrite and nitrite to ammonium through the consecutive action of nitrate reductase (NR) and nitrite reductase (NiR), and (iii) Assimilation of ammonium into organic nitrogen, yielding glutamine and glutamate as the primary organic nitrogen compounds that distribute nitrogen to all other N-containing metabolites and macromolecules (Marquez et al., 2005).

2.4 Nitrogen fixation

2.4.1 N₂ fixation by legumes

One of the most important characteristics of legumes is their ability, in symbiosis with *Rhizobium* bacteria, to form nodules on the root system and to fix atmospheric nitrogen for growth. Legumes having effective biological nitrogen fixation (BNF) can therefore be grown without nitrogen fertilizer application. Having these characteristics, they are particularly important in developing countries due to the often-high cost and/or restricted availability of nitrogen fertilizer. Biological nitrogen fixation is an important and integral component of sustainable agricultural systems. Furthermore, biological nitrogen fixation from legumes offers more flexible management than fertilizer nitrogen because the pool of organic nitrogen becomes slowly available to non-legume species (Peoples et al., 1995).

Ledgard and Giller (1995) reviewed factors that can affect N fixation, their work can be summarised as follows:

1. An effective symbiosis is required for N fixation to occur.
2. Poor soil nutrition can result in poor nodulation or N fixation.
3. Low soil pH can limit N fixation.
4. High soil N levels can inhibit nodulation and N fixation.
Grain legumes, including lentil, can take up and assimilate N from the soil prior to nodulation, to an extent that affects growth (Andrews et al., 1992). Lentil can fix adequate N for their own requirements in most situations. However, generally the legume/rhizobium symbiosis can be affected by many factors and as a consequence is variable in productivity. In lentil, Turay et al. (1991) showed that both in the glasshouse and field, early N increased leaf area and shoot and root dry weight. However, three weeks after nodules were first visible; nodule dry weight of lentil was decreased by up to 92% with increased early N. This decline in nodule dry weight was likely to be due to increased uptake of \( \text{NO}_3^- \) as legume species which are less affected by early N such as field beans take up lower amounts of \( \text{NO}_3^- \) (Andrews et al., 1992).

For proper management and a full realization of the benefits of this plant-microbial association, it is necessary to know the amount of \( \text{N}_2 \) fixed by legumes under different field conditions. Generally, the level of \( \text{N}_2 \) fixation in legumes depends on plant species (Senaratne and Ratnasinghe, 1993), host genotypes (Kumaga et al., 1994; Vessey, 1992), soil N status and rhizobial strains \( \text{N}_2 \) fixation ability (Hungria and Neves, 1987), drought stress (Badarneh and Ghawi, 1994; Kurdali, 1996), environment and their interactions.

The concentration of fertilizer N to total N in crops varied between 1-13% in legumes and between 4-44% in non-legumes, indicating that non-legumes crops were mainly dependant on soil N and in case of legumes on soil N and symbiotic N fixation. The amounts of soil N in plants were in the range of 11-26% of total plant N in legumes and 56-96% in non-legumes. These values varied during the growing season and over season (Asfary and Charanek, 1995). Few studies conducted in semi-arid areas on lentil indicated that the amount of \( \text{N}_2 \) fixation varied from 10 to 129 kg N ha\(^{-1} \) depending on location and rainfall (ICARDA, 1989; Badarneh and Ghawi, 1994). Similar amount of \( \text{N}_2 \) was fixed by lentil under different climatic conditions in Canada (Van Kessel, 1994). Kurdali et al. (1997) reported that lentil crop derived 58-68% of N\(_2\) from fixation and the actual amounts of fixed N\(_2\) were from 111 to 154 kg N ha\(^{-1}\). The reported rate of N\(_2\) fixation was 3.4 kg ha\(^{-1}\) day\(^{-1}\) for soybean (Zapata et al., 1987a), 4 kg ha\(^{-1}\) day\(^{-1}\) for faba beans (Zapata et al., 1987b) and 4.4 kg ha\(^{-1}\) day\(^{-1}\) for irrigated lentil (Van Kessel, 1994). Giller and Wilson (1991) reported that estimates of N\(_2\) fixation ranging from 23 to 250 kg N ha\(^{-1}\) with a median of 110 kg N ha\(^{-1}\) for annual legumes with a growth periods of 100-150 days. Grain legumes although obtain N from the atmosphere, may have negative N balances due to significant N removal in the grain (Karlen et al., 1994).
2.4.2 Rhizobium and N$_2$ fixation

Rhizobia encompass a range of bacterial genera, including *Rhizobium*, *Bradyrhizobium*, *Sinorhizobium*, *Mesorhizobium*, *Altorhizobium*, and *Azorhizobium*, which are able to establish a symbiosis with leguminous plants. Rhizobium species are found worldwide; they interact with a large diversity of plants even outside the Leguminosae and have been found as endophytes. They elicit the formation of specialized organs, called nodules, on roots or stems of their hosts, in which they reduce atmospheric nitrogen and make it available to the plant. Symbiotic nitrogen fixation is an important source of nitrogen, and the various legume crops and pasture species often fix as much as 200-300 kg N ha$^{-1}$ (Peoples et al., 1995). Globally, symbiotic nitrogen fixation has been estimated to be at least 70 million metric tons of nitrogen per year (Brockwell et al., 1995).

Nodules, the sites for symbiotic nitrogen fixation are formed as a result of series of interactions between rhizobia and leguminous plants. However, there are number of factors which affect the nodulation on legume roots including host micro-symbiont compatibility, physicochemical conditions of the soil and presence of both known and unknown bio-molecules such as flavonoids, polysaccharides and hormones (Antoun et al., 1978; Tisdale et al., 1990). It is a molecular dialogue between the host plant and a compatible strain of rhizobium, which serves as an initiator for the development of nodules (Murray et al., 2007). The rhizobial infection begins, when the rhizobium enters the root in a host-controlled manner (Limpens et al., 2003). A critical aspect of rhizobium-legume association is the fact that it is often manipulated under nitrogen limiting field conditions in such a way that crop production could be enhanced easily and inexpensively (Hubbell et al., 1979; Freiberg et al., 1997). The amount of nitrogen fixed, depends upon the presence of effective nodules on the host roots which are a prerequisite for the potential gain of nitrogen from the system. Although, the rhizobia commonly occur in soils but often fail to cause nodulation, either because of some unspecified type of antagonism that prevents root colonization by the rhizobial strain (Jadhav et al., 1994). Inoculation is a management practice by which the rhizobium-legume symbiosis is exploited through overcoming nodulation failure or ineffective nodulation. These affect survival and growth of the rhizobia in the soil, infection and nodulation of the host root and functioning of nitrogen fixation reactions within the nodule. It is important to recognize that both types of mineral elements and the
concentration required for these additional functions may not be the same as those required for normal growth of the plant itself.

In addition to (better) nodulation, certain rhizobial cultures also produced considerable increase in shoot length and shoot weight and root length and root weight in lentil seedlings as compared to the control. The reason behind might be the production of some of the phytohormones, additional to the positive responses on the seedlings growth exhibited by the nitrogen-fixing interactions (Sevilla et al., 2001). As far as the increase in shoot fresh weight and root fresh weight is concerned, similar work was conducted by Chanway et al. (1989). They isolated certain strains of rhizobia from lentil seedlings and obtained the similar results. The similar data were reported by Yanni (1992). He observed the response of lentil, chickpea and pea. Pal et al. (2000) also reported the same trend in the results in case of root dry weight and shoot dry weight. They stated that inoculation with rhizobium increased the growth, yield and nutrient uptake significantly in pot and field experiments. Comparable results were reported by Sindhu et al. (1999). From the above cited literature, it is clear that growth of lentil seedlings can be enhanced by inoculating the seeds with the effective rhizobial strains. The rhizobial infection starts, when the bacterium enters the root in a host-controlled manner. In the legumes, the rhizobium becomes trapped in a cavity formed by the curling of root hair. The root hair plasma membrane invaginates and a tube like structure is formed by which the rhizobium enters the plant and reaches the base of the root hair. Consequently, the infection thread reaches a nodule primordium in the cortex of the root that develops into a nodule, upon release of the rhizobium (Limpens et al., 2003).

2.4.3 Potassium and nitrogen fixation

The benefit of K on N₂-fixation process stems from the multiple role of K in the plant metabolism:

- Potassium activates more than 60 enzyme systems, including the nitrogenase enzyme which is essential for N₂-fixation.
- Potassium enhances the synthesis of the carbohydrates in leaves and their further translocation to the roots. Once in the root system, the carbohydrates stimulate growth of new root hairs as well as nodule development and function.
- Potassium contributes to good root growth providing a proper “home” for the nodules in which N is fixed.
Studies on the effect of K on the N$_2$-fixation by root nodules of another leguminous crop (*Vicia faba*) show that the better carbohydrate supply of nodules, by plants well supplied with potassium, results in a higher carbohydrate turnover in the nodules and thus the provision of ATP and reducing electrons required by the nitrogenase is enhanced (Mengel et al., 1974). Plants growing at higher K level have better development of nodules and consequently higher N$_2$-fixation.

Direct measurements of the effects of K fertilization on legume N$_2$ fixation over the entire growing season have not been made. Such measurement would require the use of the $^{15}$N dilution technique or the difference method. Without such data, it is difficult to ascertain the overall impact of K fertilization on legume N$_2$ fixation. Most studies have measured parameters that are positively associated with legume N$_2$ fixation (e.g. nodulation, nodule mass and acetylene reduction) at one or a few points in the growth cycle.

The influence of potassium supply on nodulation was studied on soybean grown in nutrient solution. The dry matter yield, nodule parameters (nodule number and fresh weight of nodule per plant, average weight of nodule) and total nitrogen accumulation in the plant increased with increasing K supply (Premaratne and Oertli, 1994). In another study on beans, nitrogenase activity increased in proportion to K fertilization especially during the early stages of growth. The cost of the production of N fertilizer coupled with declining world fossil fuel reserve has led to great interest in biological nitrogen fixation of atmospheric dinitrogen "N$_2$" (Wittwer, 1978). The protein rich legume crops have been of special interest, largely because many of them appear to have the capacity to acquire sufficient N for maximum or nearly maximum yields through their symbioses with rhizobia (Minchin et al., 1981).

### 2.4.4 Potassium and *Rhizobium*

Although legume-rhizobium symbioses are highly productive under optimal growing conditions, soil deficiencies of essential elements drastically decrease legume nodulation, N fixation, N accumulation and yields (Robson, 1978). Of the various elements, K is especially important for enhancing the productivity of legume-rhizobial symbioses.

Numerous studies have demonstrated that K is required in greater quantities than any other soil mineral for total N accumulation in legumes and that dry matter and total N accumulation in leguminous crops are extraordinarily responsive to K fertilization. It
has been suggested that K may have beneficial effect on symbiotic nitrogen fixation by affecting parameters such as number, fresh weight and size of nodules (Premaratne and Oertli, 1994) or by affecting the productivity of nodule, that is the amount of nitrogen fixed per unit time and unit mass of the nodule and qualitative requirement for K has been demonstrated for some rhizobia. (Premaratne and Oertli, 1994).

**2.4.5 Nitrogen metabolising enzymes and nitrogen fixation**

An effective nitrogen fixing symbiosis requires an appropriate interaction between the plant and bacteria (Verma and Long 1983). Plants require N from soil or from atmosphere, by symbiotic N$_2$ fixation. In root nodules, nitrogen fixation generates ammonium, which is processed via the GS/GOGAT cycle after being excreted into the plant cytosol; the first step is catalysed by glutamine synthetase which uses ATP and forms glutamine, then glutamine is converted to glutamate by glutamate synthase (NADH-GOGAT) (Cullimore and Bennett, 1988). This process and the subsequent transformation into amino acids and ureides, which occurs in the plant-cell fraction of nodules, are both energy-consuming process.

A close relationship between nitrogen fixation and GS and GOGAT activities has been found within legumes. Nitrogenase, GS and GOGAT activities increase during nodule development (Egli et al., 1989; Reynolds et al., 1982), decrease with harvest of shoots and increase again with shoot re-growth (Crale and Heichel, 1981; Groat and Vance, 1981). The efficiency of the assimilation of the fixed nitrogen by the plant enzymes GS and GOGAT could play an important role in plant productivity.

Even if the GS/GOGAT pathway is considered to be the major pathway for ammonium assimilation under normal growth conditions, the role of glutamate dehydrogenase (GDH) under some environmental conditions cannot be dismissed (Karina et al., 2006). This enzyme may play an alternative role to the GS/GOGAT cycle in ammonium assimilation under specific physiological conditions that make the ammonium concentration increase (Aubert et al., 2001). GDH catalyses two reversible reactions, the first one is implied in the nitrogen metabolism (the incorporation of ammonium on 2 oxoglutarate to produce glutamate) and the second one is involved in carbon metabolism (regeneration of the C skeletons by the production of 2 oxoglutarate) (Temple et al., 1998; Kwinta et al., 2001). In C and energy limiting conditions the function of GDH related to the replenishment of tricarboxylic acid (TCA) cycle intermediates via its oxidation to 2-oxoglutarate (Robinson et al., 1991). Sufficient
genotypic variability has been measured for GOGAT activity in alfalfa (Jessen et al., 1988) and for GS activity in *Phaseolus* (Hungria et al., 1991) and this suggests that these enzymes could be used as possible complementary selection criteria in breeding programmes.

2.5 Potassium and nitrogen metabolism

The interaction of K with the other macronutrients has been extensively studied and it is an important aspect of improving crop yields. Potassium plays several roles in plant metabolism, and performs these roles positively; it should interact positively with other nutrients. Positive interactions of K with N and P have been reported (Dibb and Thompson, 1985). They reported that the increased K allowed for rapid assimilation of absorbed NH$_4^+$ ions in the plant, maintaining a low, non toxic level of NH$_3$. Increased yield of crops with the addition of N and P requires higher level of K in the soil (Dibb and Thompson, 1985; Fageria et al., 1997). The relationship of potassium with nitrogen metabolism is well documented (Ruan et al., 1999; Pettigrew and Meredith, 1997). The stimulating effect of K$^+$ on nitrate metabolism is due to an increase of nitrate export in xylem (Rufty et al., 1981). Casadesus et al. (1995) observed a coupled loading of potassium and nitrate into the xylem since K is proposed as a transporter of photo assimilates from the source to the sink and acts fundamentally as a nitrate co-transporter in its uptake, through xylem transport, and its recycling through phloem transport (Blevins, 1985). NO$_3^-$ is taken up by plant roots via an active process (Streeter and Batra, 1984), therefore, the uptake may be affected through the influence of K on the translocation of photosynthetic assimilates needed to support this active uptake process (Ashley and Goodson, 1972). Ruiz and Romero (2002) have reported that by increasing the rates of K$^+$ application, the uptake and transport of nitrate towards the aerial parts of the plants may be increased. As the plants grown under the deficient K supply fail to transport nitrate efficiently into shoots, the excess nitrogen supply in the soil leads to the leaching of nitrate, thus causing the surface and ground water pollution. Withdrawal of K from the soil affects NO$_3^-$ assimilation since nitrate is assimilated largely by roots (Siebrecht and Tischner, 1999). The studies by Krauss (2004) clearly demonstrated that K-deficient plants have a repressed activity of enzyme nitrate reductase. Thus the plants receiving inadequate supply of K would absorb small amount of nitrate from the soil and therefore there would be less conversion into proteins. Both nitrate and protein are important for the crop quality.
2.6 Carbon and nitrogen accumulation and partitioning

Plants respond to environmental factors that limit the acquisition of above-ground resources relative to below-ground resources (e.g. nutrients or water) by shifting their partitioning to tissues and processes associated with gaining less resource (Gedroc et al., 1996). Husain et al. (1990) also reported that plants respond to water shortage by allocating more carbon to roots and, if this occurs for a prolonged period, enhanced root growth may result. Partitioning of dry matter in plants may be influenced by many environmental factors, including drought and nutrient deficiency (Gregory, 1989; Wardlaw, 1990). Fredeen et al. (1989) noted that much more dry matter was partitioned to roots than shoots in phosphorus and nitrogen-deficient plants of soybean. Recently, Sangakkara et al. (2000) found that the legume species mungbean and cowpea allocated more carbon to roots under a low soil moisture regime. Water stress during vegetative growth retards nodulation and decreased nitrogen fixation (Zablutowicz et al., 1981; Abdel-Ghafar et al., 1982; Saito et al., 1984). Sangakkara et al. (1996) found that under water stress the number of nodules reduced in common bean and faba bean. Nutrient addition decreased the root fraction and increased the leaf fraction, indicating that nutrient deficiencies limit shoot growth more than root growth. These findings are in accordance with several reports showing that more dry mass was partitioned to roots than shoots in plants grown at a low nutrient level, i.e. greater amounts of photosynthates were allocated for root growth. This phenomenon was observed in *Phaseolus vulgaris* (Cakmak et al., 1994; Al-Karaki et al., 1995) and other plants (Fredeen et al., 1989; Aloni et al., 1991). An important reason for the increasing allocation of carbon to the roots during the vegetative stage is the need to establish an adequate root system before the onset of competition for assimilates from the increasing sink activity of the fruits at pod filling (Gregory et al., 1997). During vegetative growth, the uptake of nutrient by plants with a high root fraction could potentially be higher than the requirement for shoot growth. Provided that any excess of uptake can be accumulated in the shoot, this will become available to supply part of the requirement of the developing pods and seeds during the reproductive phase. This should result in delayed leaf senescence, prolong the period of photosynthesis and lead to higher yields.

Efficiency of N uptake and use relative to the production of grain requires that processes associated with absorption, translocation, assimilation, and redistribution of N operate effectively (Moll et al., 1982). Among these processes, efficient uptake of N by
plants and its translocation to grain are crucial for increasing yields. Plant N accumulation and grain yield generally exhibit positive quadratic relationships (Cassman et al., 2002; Fageria and Baligar, 2001). Plant tissue analysis is the determination of elemental concentration in samples from particular organs or portions of a crop sampled at certain times or stages of morphological development (Fageria et al., 2003). Concentration is usually expressed on dry weight basis. Nutrient concentrations are influenced by plant age, plant part analyzed (leaves, shoots, or whole top), cultivar, water and nutrient availability in the soil, and control of pests. Although nutrient concentrations are influenced by several soil, plant, climatic factors, plant tissue concentrations are still relatively more stable and useful parameters compared with soil analysis for identifying nutritional status of crop plants. Concentrations of most nutrients are restricted to fairly narrow ranges (Fageria and Baligar, 2005).

Concentrations of N in most tissues of crop plants decrease with advancement of plant age. It may be because with increasing plant age, more dry matter is produced that dilutes the concentration of nutrients accumulated (Fageria et al., 1997). Maman et al. (1999) reported that N concentrations were high during vegetative stages of growth and decreased with plant age in pearl millet. Nitrogen concentrations were higher when leaves were analyzed rather than whole aboveground plant. Nitrogen concentration in plant leaves has been associated with grain yield. Higher N concentrations later in the later stages of plant growth are useful because more N may be translocated to grain ultimately giving higher yields. High N mobilization into grain also results in a high grain harvest index (Boote et al., 2003).

The N content/unit biomass (N% in dry matter) is frequently used as an indication of how adequate N supply is for crop growth (Lawlor et al., 2001). However, N% is not a fixed value and decreases as the crop develops and its structure and biochemical composition change. Leaves, with a large content of proteins etc., are formed in early growth, and supporting stems, with more cellulose and lignin and little protein, later. So, N-content (N%) decreases with age and it also depends on environment (Lawlor, 2002).

When dry matter or grain yield is multiplied by nutrient's concentration, the results are a measure of nutrient uptake and expressed in accumulation or uptake units. The nutrient uptake values are considered as useful indicators of soil fertility depletion,
and related to crop yield levels. Nutrient accumulation in crop plants follows dry matter accumulation patterns (Fageria and Baligar, 2005). The amount of N accumulated generally parallels dry matter accumulation and increases with plant age (Ntamatungiro et al., 1999). Uptake values vary from crop species to species, and are higher in grain than in straw, indicating that grains are greater sinks for N accumulation compared with other parts of the tops. Generally, N uptake in grain has positive significant associations with grain yield (Fageria and Baligar, 2001; Lopez-Bellido et al., 2003). Hence, improving N uptake in grain may lead to improved grain yield (Fageria and Baligar, 2005).

Determining the total N or soluble protein content of a leaf appears to be an interesting alternative for monitoring the N status of the plant during the grain filling period (Hirel et al., 2005). The progressive decrease in leaf or stem N observed during grain-filling period corresponds to the degradation of leaf proteins, mostly represented by the photosynthetic enzyme ribulose 1-5-bisphosphate carboxylase (Peoples et al., 1980; Wittenbach, 1979). Larger amounts of protein are present in the topmost section of the plants (Kichey et al., 2006). It indicates that plant invests more N in the photosynthetic apparatus located in the upper part of the canopy (mostly represented in wheat by the flag leaf), to capture as much of the incident light as possible and thus maximize carbon assimilation as a function of N availability (Gastal and Lemaire, 2002).

2.6.1 Coordination between nitrogen and carbon metabolisms

Carbon and nitrogen metabolisms are two major metabolic pathways, which have been intensively studied and correlated in plants. Both are interdependent but closely linked in numerous biochemical pathways. Any change favourable or otherwise to nitrogen assimilation brings about repercussions on carbon assimilation and vice-versa. These two processes must remain in equilibrium, which is ensured by fine and complex regulatory mechanisms that are well co-ordinated to enable optimal growth and development of plants even under varying environmental conditions (Ferrario et al., 2001). However, the ratio of carbon to N assimilated is not rigidly fixed, for if the supply of nitrate increases relative to CO₂ then more amino acids and proteins accumulate relative to the rate of CO₂ assimilation and to the carbohydrate content which may decrease as growth is stimulated (Lawlor, 2002). When ample nitrate is available, a large proportion of N is stored (i.e. metabolically inactive) as protein and to a smaller
extent, as amino acids and nitrate in short or long-term pools until their capacity is saturated. With inadequate N, the amino acid content of leaves falls, less protein is synthesized and growth is decreased generally more than photosynthesis, so carbohydrates accumulate and the N/C ratio is low.

CO$_2$ fixation during photosynthesis provides sugars that are important metabolites in plants necessary for intermediary metabolism. Sugars are used as fuel for respiratory metabolism and thereby serve as the energy supply for plants. Furthermore, sugars are substrates for the synthesis of storage and structural polysaccharides such as starch and cell wall compounds. N, on the other hand, is quantitatively the most important inorganic nutrient for plants. Nitrate is the main available N source for many plants and serves through its reduction for the synthesis of amino acids and other N compounds necessary for plant's growth (Krapp and Truong, 2005). In addition to their crucial role in plant metabolism, N and C compounds have profound effects on plant developmental processes from germination to flowering.

There is a close interaction in the very early phases of N and C metabolisms, both using the light energy, with some 10% of the electron flux in photosynthesizing leaves used in nitrate reduction (Foyer et al., 2001). C skeletons are essential for the incorporation of inorganic N into biologically relevant molecules, as amino acids, proteins, nucleic acids and a vast number of secondary metabolites. During nitrate assimilation, 55% of the photosynthetic fixed C is converted via glycolysis into phosphoenolpyruvate (PEP) and further invested into the synthesis of organic acids. For the GS/GOGAT cycle to work, there is a strict requirement for N metabolism to interact with C metabolism since GS activity requires energy in the form of ATP and the GOGAT uses C-skeletons and reductant in the form of 2-oxoglutarate and reduced ferredoxin or NADH, respectively. The GS/GOGAT pathway transfers ammonium to glutamate to form glutamine, then the fixed N is transferred to 2-oxoglutarate, thus leading to the biosynthesis of two molecules of glutamate. The key organic acid, 2-oxoglutarate, is mainly derived from sugar respiration and amino acid transamination reactions. However, the exact enzymatic origin of this compound is still unknown (Hodges, 2002).

Conversely, the dependence of photosynthesis on N assimilation is obvious particularly with respect to the proteins (N products) involved in the immense photosynthetic process. The key enzyme for CO$_2$ fixation, Rubisco, is the main N
containing component of the cell. Indeed, the amount of Rubisco in a leaf cell may reach 50% of the total protein. Furthermore, in addition to Rubisco, a large number of thylakoid proteins, some of which display very high turnover rates and a large number of other catalytic activities, are needed for the primary and secondary photosynthetic reactions (Krapp and Truong, 2005).

Therefore, the C to N ratio has to be carefully regulated to ensure proper functioning of the huge metabolic network. In order to maintain a viable C/N status under a large range of growth conditions, plants have evolved complex mechanisms to regulate the delicate network of these two major assimilatory pathways (Krapp and Truong, 2005).

**2.6.2 C/N ratio of plants**

Significant and positive correlations between C/N ratio and biomass NutE and C/N ratio and yield NutE were found to be present in cultivars elsewhere. It may be due to the improvement of plant biomass and grain yield via enhanced assimilation of carbohydrates and their translocation to grain, without a corresponding improvement in remobilization of nitrogen to the developing grains. It further indicates that carbon economy of a plant is closely connected with its nitrogen economy. Therefore, as nitrogen function in plant growth and nutrition is closely connected to carbon, the C/N ratio controls nitrogen availability and potentially affects interactions through the processes of organic residue decomposition and soil organic matter formation in soil and biomass production (photosynthesis - respiration) in plants (Wilkinson et al., 2000). Moreover, the carbon economy of a plant is closely connected with its nitrogen economy through the distribution of nitrogen between different plant parts with consequences for various metabolic processes. Nitrogen uptake and allocation also depend on carbon allocation between different organs and nitrogen composition of these organs; however, a higher nitrogen concentration in the plant is likely to lead to a higher rate of photosynthesis, but also results in an increased rate of respiration (Chapin et al., 1987). Therefore, differences in allocation of nitrogen and carbon between organs or utilization in different types of compounds may decisively affect a plant’s efficiency with respect to the use of nitrogen as observed in the present study.
Though, a lot of research has been arranged on role of K in translocation of photosynthates. However, the relationships between carbon and nitrogen partitioning in relation to K nutrition in legumes plants have not been studied in detail.

2.7 Genotypic variation in lentil under potassium application

Although, the yield improvement response to K fertilization on low K testing soils is a fairly uniform response, there may be genotypic variation for this response. This phenomenon has been demonstrated in cotton (Pettigrew, 2008). Considerable variation in efficiency of K uptake and utilization has been identified among existing genotypes for a variety of crop species including lucerne (James et al., 1995), beans (Shea et al., 1968), soybean (Sale and Campbell, 1987). Furthermore, the more potassium responsive genotypes in the study by Cassman et al. (1989) were subsequently shown to produce a more extensive root system than the less K efficient genotype (Brouder and Cassman, 1990). Therefore, the genotypic differences in K response may probably be because of the fact that more K responsive genotypes were able to take up K at a greater rate or more efficiently because of bigger root system. Clement-Bailey and Gwathmey (2007) also reported that K fertilization was more critical for early maturing cotton varieties rather than later maturing varieties. Genotypic differences for K uptake and use efficiencies have also been detected in other crops.

It is reasonable to assume that genotypic variation in K utilization efficiency exists within all the major crop species. Future increases in the yield of grain crops will depend upon the understanding of the physiological basis of yield production. Past improvements in crop yields, that helped increase food production to sustain growing world production, were not always based on physiological principles. Increasing yield in the future will probably be more difficult as the increase must build on a higher base level, and as modifications of plants and crop production system would face new restrictions imposed by the environment, economics and social policy.

2.8 Foliar K fertilization

The foliar application of mineral nutrients offers a method of supplying nutrients to higher plants more rapidly than methods involving root application. Soybean response to foliar fertilization has been extensively examined by researchers starting in the 1970’s (Garcia and Hanway, 1976; Parker and Boswell, 1980; Vasilas et al., 1980; Haq and
Mallarino, 1998, 2000). Foliar fertilization with solutions containing macronutrients, such as N, P, K, and S, has shown inconsistent soybean grain yield response (Boote et al., 1978; Parker and Boswell, 1980; Haq and Mallarino, 1998, 2000). Garcia and Hanway (1976) reported large yield increases from foliar application of NPKS fertilizer mixtures during reproductive stages. Some researchers also investigated growth response to foliar fertilization of macronutrients at early vegetative stages (Haq and Mallarino, 1998, 2000; Mallarino et al., 2001). They theorized that foliar application of nutrients to plants when they are rapidly growing would complement the nutrients the plant would be obtaining from the soil and, consequently, increase yield.

Several studies have been conducted on foliar fertilization in soils with initially high soil test levels (Parker and Boswell, 1980; Haq and Mallarino, 1998) and most of soybean yield responses to foliar fertilizer were observed when soil test K and other nutrients were below the optimal level or when plants suffered from water stress. A recent study by Nelson et al. (2005) showed significant soybean yield response to foliar K fertilizer (K₂SO₄) applied at the rate of 36 kg K ha⁻¹. However, most of the reported growth responses to foliar fertilizer applications have been variable and inconsistent (Boote et al., 1978; Parker and Boswell, 1980; Haq and Mallarino, 1998, 2000). It was established that foliar fertilizer applied at early vegetative stages increased yields and enhanced quality in soybean (Mallarino and UI-Hag, 1998) and garden bean (Stancheva et al., 2004). Plant response is dependent on species, fertilizer form, concentration, and frequency of application, as well as the stage of plant growth (Faulkner, 1999; Eddy, 2000). Quality parameters like protein and oil content of seed was significantly improved by foliar K application in groundnut. However, the response was better with potassium sulphate (K₂SO₄), probably due to the positive effect of sulphate in enhancing the protein and oil content in crops (Umar et al., 1999). Potassium application has also been reported to improve quality of groundnut (Umar and Bansal, 1997) and soybean (Magen, 1997). Foliar K application in particular, leads to considerable yield and quality response in cotton and tomato crops in Egypt and Israel, respectively (Eid et al., 1997; Williams and Kafkafi, 1997).

Potassium requirement rise many folds when maximum vegetative growth takes place and pods are set on plants. Additionally developing pods have high K requirements; therefore it is critical to provide an adequate potassium level to the plants when the pegs start to form (Umar et al., 1999). This K requirement can only be met by pre-plant soil K application. Hence, higher yields were obtained with basal K
application than with the foliar alone K treatments (Umar et al., 1999). Pod and dry matter yields of groundnut were significantly higher when K₂O ha⁻¹ was basal applied to the soil as compared to the foliar alone K treatments irrespective of the K source used for foliar application, crop variety and growth season.

2.9 Effect of legume fixed N on wheat yield through intercropping

The most widespread and consistent effect of legumes is to improve the N economy of soil through N₂-fixation. This does not mean that legumes always make net contributions of N to soils in which they grow. What it does mean is that the N balance of a legume-cereal sequence is, in most cases, more positive than that of a cereal-cereal sequence in the same soil. Thus, the inclusion of legumes in cropping systems can arrest the decline of soil N fertility that invariably accompanies intensive agriculture and, at least, reduce the requirements for inputs of fertilizer N. The latter is of particular relevance to less developed countries where N fertilizers are used sparingly because of their high costs, the low per capita income and limited credit facilities of most farmers, and lack of effective infrastructure for production and distribution.

Intercropping of cereals and grain legumes is a neglected theme in agricultural science and practice in both conventional and organic (Dahlmann and von Fragstein, 2006). Cereal-legume intercropping plays an important role in subsistence food production in both developing and developing countries, especially in situations of limited water resources (Tsubo et al., 2005). Intercropping improves the utilisation of available resources and cause yield advantages and increased yield stability (Ofori and Stern, 1987; Vandermeer, 1989; Willey, 1979). Yield advantages occur when intercrop components compete only partly for the same plant growth resources. Intercropping is practiced in many parts of the world as a result of its ecological benefits (Francis, 1986). However, it is particularly important and has continued to be widely employed not only in the tropics but also in the temperate regions (Vandermeer, 1989). Intercropping contributes to high grain yield in both tropical, subtropical and temperate areas; it can be environmentally benign by reducing accumulation of NO₃-N in soil profile (Stuelpnagel, 1993). Several investigations clearly show that it has a significant influence on yield increase (Fortin et al., 1994; Lesoing and Francis, 1999). Lentil could derive two-third of its nitrogen needs from the atmosphere (Badarneh, 2005). At maturity, the level of symbiotically fixed nitrogen in two years has been reported to be
154 and 117 kg N/ha in mono-cropped lentil and 95 and 41 kg N/ha in lentil intercropped with barley (Schmidtke et al., 2004). Use of bio-inoculants *Rhizobium* *spp.* and *Azospirillum brasilense* helps in enhancing nodulation in lentil (Tiwari and Misra, 2000; Kumar and Chandra, 2005) and thereby nitrogen fixation. Therefore, these inoculants may need to be used to ensure greater inputs of nitrogen fixation in lentil based intercropping systems. Legume/cereal intercropping systems occupy a prominent place in agricultural systems of the tropics (Ofori, 1987). Studies done by (Gunasekera and Senaratne, 1990) have shown that application of appropriate level of K (i.e., 80 kg ha\(^{-1}\)) on maize/mungbean intercropping system alleviated the competitive depression of mungbean in dual stand. Hegazy and Genaidy (1995) reported that the application of potassium sulphate improved the growth of soybean when either mono- or intercropped on clayey soil. The relative K fertilizer efficiencies were reduced in intercropping compared to mono-cropping, thereby, increasing the economic optimum K fertilizer rates.

2.10 Critical appraisal of the review of literature

The literature reviewed above includes studies on the factors that influence physiological analysis of various growth parameters, photosynthetic and biochemical traits and legume-grain yield under various environments of potassium application. It appears from the review that research efforts to enhancement of symbiotic nitrogen fixation to K nutrition are minimal. Since, the legumes grown under potassium-deficient conditions suffer from nitrogen shortage and produce lesser yield and because the information available about the effect of potassium fertilization on nodulation and N\(_2\) fixing ability of rhizobium in lentil is quite meagre.

Therefore it is rational to study the effect of potassium fertilization on various aspects of nodulation by lentil.

Since, it is well recognized that plants respond to environmental factors that limit the acquisition of above-ground resources relative to below-ground resources by shifting their partitioning to tissues and processes associated with gaining less resource. Therefore, the C to N ratio has to be carefully regulated to ensure proper functioning of the huge metabolic network. In order to maintain a viable C/N status under a large range of growth conditions, plants have evolved complex mechanisms to regulate the delicate network of these two major assimilatory pathways.
Though, a lot of research has been arranged on role of K in translocation of photosynthates. However, the relationships between carbon and nitrogen partitioning in relation to K nutrition in legumes plants have not been studied in detail. Moreover, the role of potassium in relation to nitrogen metabolism is well established and studies report on the effect of K on NRA and nitrate. However, there are no reports regarding the effect of K and rhizobium application on other nitrogen metabolizing enzymes.

Keeping these facts in view, studies have also been undertaken to determine the relationships between potassium fertilization and nitrogen fixation, using various cultivars of lentil and strains of rhizobium.