

**EVALUATION OF PIGEONPEA
(*Cajanus cajan* L. Millsp.) GENOTYPES FOR
PHOSPHORUS UPTAKE EFFICIENCY**

**BY
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2004BS163M**

**Thesis submitted to the Chaudhary Charan Singh
Haryana Agricultural University in the partial
fulfilment of the requirements for the degree of**

**MASTER OF SCIENCE
IN
PLANT PHYSIOLOGY**



**COLLEGE OF BASIC SCIENCES AND HUMANITIES
CCS HARYANA AGRICULTURAL UNIVERSITY
HISAR-125004 (HARYANA)**

2008

**Dedicated
To
My Little Angel**

CERTIFICATE-I

This is to certify that this thesis entitled, "**Evaluation of pigeonpea (*Cajanus cajan* L. Millsp.) genotypes for phosphorus uptake efficiency**", submitted for the degree of **Master of Science** in the subject of **Plant Physiology** of the Chaudhary Charan Singh Haryana Agricultural University, Hisar, and is a bonafide research work carried out by **Ishu Lamba, Adm. No. 2004BS163M** under my supervision and that no part of this thesis has been submitted for any other degree.

The assistance and help received during the course of investigation have been fully acknowledged.

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Acknowledgement

I express my deep sense of gratitude with reverence to my Major Advisor Dr. J.K.Sandooja, professor, Plant Physiology, Deptt. of Horticulture CCS Haryana Agricultural University, Hisar and for his kind initiation, over enlivening and emboldening posture, incessant forbearance, continuous encouragement, inspiring guidance, constructive criticism, pleasant discussion and friendly behaviour throughout the investigation and preparation of this manuscript.

But for the patience and understanding of Dr. Sunita Sheokand, Professor, Department of Botany & Plant Physiology, without whose efforts this work would not have been completed. His day to day interest in my project, critical evaluation of results and encouragement will be remembered.

I wish to express my heartfelt gratitude toward other members of my Advisory Committee Dr. Sarla Malothera, Professor, Department of Chemistry & Bio- Chemistry; Dr. Deepak Grover, Professor, Department of Mathematics and Statistics and Dr. A.C.Malik, Sr.Scientist, Department of Agronomy CCS Haryana Agricultural University, Hisar for their constant help, indispensable advice and valuable suggestions offered during the entire spell of investigation and preparation of manuscript.

I feel indebted to faculty members of the Department of Botany & Plant Physiology especially to Dr. Sawraj Kumari, Dr. C.L. Goswami, Dr. S.K. Goyal, and Dr. K.D.Sharma for their moral boosting and soothing words.

I wish to accord my special thanks to Dr. R.K.Sharma and Dr. S.S. Dudeja Department of Microbiology for providing the facilities.

The help and cooperation extended by office staff members is gratefully acknowledged. I express my heartiest thanks to

Sh. Surajbhan, Sh. Ratan Kumar and other laboratory staff for their constant help.

My vocabulary utterly fails in expression my sincere gratitude to my father and mother-in-law Dr. R.S.Singal and Smt. Raj Rani and also my father Sh. S.S.Lamba, mother Smt. Raj Bala who always provide support, sustain spirit and Endeavour at every critical juncture of my life. Their pain and sacrifice surpass all the materialistic achievements.

I would also remember the sacrifices made by my husband Mr. Vikramjeet Singal, and his enduring support, infinite patience, untiring constant inspiration and moral support throughout my study. Above all, he always stood by me in the hours of crisis and toughest times of my life.

My sister Manisha (Motu), brother-in-law Satyajeet (chhotu) and loving baby Kiyati recalled for their naughty, sweet and melodious actions which relieve me from the outrageous work.

I would never be able to repay back the kind consideration, support and affectionate help from my beloved friends Dr. Anita Kumari, Sukham, Ashwani, Dr. Vinita, Dr.Champa Dr. Sunder and heartliest thanks to my neighbours Mrs. Savi & Mr.Virender I wish to acknowledge them for their timely guidelines and selfless help.

In the last but not the least I wish to accord my special thanks to Professor and Head, Dr. H.R. Dhingra for providing me each and every facility during my whole course of work.

“He choose me; I am glad he showed me the path”. The almighty is over truly honoured; he led and guided me throughout the endeavour.

I express sincere thanks to Arjun Negi who help me in framing the structure of this manuscript with a technical hand in typing.

Place: Hisar

(ISHU LAMBA)

Date: December, 2008

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ABBREVIATIONS

P	:	Phosphorus
N	:	Nitrogen
CO ₂	:	Carbon-di-oxide
C ₂ H ₄	:	Ethylene
g	:	Gram
cm	:	Centimeter
Min ⁻¹	:	Per minute
ARA	:	Acetylene Reductase Activity
h ⁻¹	:	Per hour
mg	:	Milligram
μ mole	:	Micro mole
μg	:	Microgram
kg	:	Kilogram
wt.	:	Weight
ml.	:	Milliliter
C.D.	:	Critical difference
C.R.D	:	Completely Randomized Design
<i>et al.</i>	:	<i>et alia</i> = and others
i.e.	:	That is
<i>Viz.,</i>	:	Namely
cv.	:	Cultivar
%	:	Per cent
N.S.	:	Non significant
Etc.	:	Etcetera

CHAPTER-I

Introduction

Pigeonpea (*Cajanus cajan* L. Millsp.) is commonly known as Arhar or red gram. It accounts for about 11.8 percent of the total pulse area and 17 percent of total pulse production of India. Pigeonpea is grown throughout the tropical countries of the world especially in the more arid regions.

Phosphorus is second only to N as the most limiting element for plant growth. (Bielecki, 1973; Vance et. al., 2000). The amount of P in plants ranges from 0.05% to 0.30% of total dry weight. Crop yield is reduced to 40% of the total yield due to limited phosphorus availability. The availability of phosphorus to the plants is very low in soils, as it is easily locked up by the presence of Fe, Al and Ca ions in the soil. phosphorus use of efficiency is defined as the ability of a plant genotype to incorporate or utilize in the production of total plant biomass (Balir, 1993). To distinguish efficiency of genotypes from their genetic yield potential, their response to phosphorus deficiency stress must be studied. Genotypes having the same yield potential may behave differently with respect to their phosphorus deficiency. On the other hand, phosphorus responsiveness can be defined as the capacity of the genotypes to increase uptake or yield when supply of nutrients to the roots is increased.

Adu-Gyamfi et al., 1989 reported the variation in phosphorus utilization efficiency among different cultivars of pigeonpea. When

phosphorus availability was limited, efficient cultivars obtained reasonably high yield through an effective translocation of the absorbed phosphorus to the leaf. Dry matter production and nitrogen fixation are strongly controlled by phosphorus absorption ability rather than phosphorus utilization efficiency. High phosphorus conc. in the nodules is advantageous to the plant in terms of phosphorus distribution under the phosphorus deficient conditions. Use of undue additional phosphorus fertilizer in the soil leads to a problem of environmental pollution. So there is a dire need to minimize the phosphorus use as per need.

Intra-specific variations of phosphorus efficiency of different crops has also been reported by Ahmad *et al.* (2001). He has reported that by using the crops with high phosphorus efficiency will enhance phosphate utilization and will also protect the environmental pollution in developed countries where the phosphorus fertilizer is being used in excess.

Fageria & da Costa (2000) also reported that the genotypes which produce more dry matter in a soil with a low phosphorus level respond well to added phosphorus. Adu-Gyamfi *et al.* (1989) studied the different pigeonpea genotypes to various levels of phosphorus absorption & its utilization efficiency & reported that, increasing the phosphorus application rate significantly increased nitrogen fixation in all the cultivars. Similar results of variation in phosphorus use efficiency in different genotypes of pigeonpea was reported by Fujita *et al.* (2004). He also reported that a low supply of phosphorus significantly decreased the plant biomass, average leaf area & photosynthetic activity of all genotypes. This decrease was smallest in ICPH-8 and largest in UPS120. ICPH-8 retained more plant biomass at low phosphorus supply due to higher

absorption capacity rather than the high phosphorus utilization efficiency. Hence they concluded that phosphorus deficiency effects primary sink activity by restricting cellulose biosynthetic in the meristematic sink cells.

Gahoonia & Nielsen (2004) tried to find out the traits for efficient absorption of phosphorus & reported that root morphology, root hairs, mycorrhizal symbiosis, uptake kinetics, root induced changes in the rhizosphere were mainly related to phosphorus uptake efficiency. Root soil contact is an important factor for uptake of a less mobile nutrient such as P by crop plant (Schjorring and Nielsen, 1987).

Gilbert et. al. (1999) studied the acid phosphatase deficient white lupin roots. They found that white lupin developed proteoid roots (short, lateral, density clustered roots) when grown in phosphorus deficient conditions. The acid phosphatase activity was detected in root exudates in the presence of organic phosphorus sources. It was found that under phosphorus deficient conditions, a unique isoform of acid phosphatase was induced.

Keeping these considerations in view the investigations were carried out in six pigeonpea genotypes for phosphate uptake efficiency with the following objective:

- To identify genotypes which are efficient in phosphorus uptake and utilization.
- To identify characters associated with efficient phosphate uptake/utilization.
- To evaluate genotypes for nodulation & nitrogen fixation efficiency under varying phosphorus regimes.

CHAPTER-II

Review of Literature

Growth & development in the plants depends upon various minerals, environmental conditions, growth hormones and important physiological processes going on in the plant. Plant requires sixteen essential minerals for its proper growth. Out of these nine i.e. C, H, O, N, P, K, S, Mg, Ca are called as macronutrients as their requirement is more than 1mg/g of fresh weight of plant. Whereas other seven nutrients are called as micronutrients. Among the macronutrients C, H & O are taken up by the plants from the soil and atmosphere in the form of CO₂ and H₂O. Whereas NPK are most critical element which have got major role to play & are called as critical elements. These are taken up by plants in large amount from the soil. As a result of which there is a deficiency of these nutrients in the soil. These nutrients alter the various physiological processes and have got the main role in growth and development of the plants. N is generally applied by the farmers in the soil in the form of urea, Kisan khad etc. P is applied by the farmers but being immobile. It is not fully absorbed by the plants. It has the major role to play in the plant metabolism. N is also supplemented to a great extent among the pulses with the help of symbiotic nitrogen fixer i.e. *Rhizobium sp.* which are growing in association with roots of the plant & helps in formation of nodules. So, in case of the crops of family leguminosae, N deficiency is rarely observed. So, P is one

of critical element for the members of leguminosaeae. Pigeonpea is one of member of leguminosaeae.

Phosphorus is one of the key components in ATP, phospholipids, nucleic acids & also plays a pivotal role in energy conservation and metabolic regulation (Marschner 1995, Raghothama 1995). Plant response to N, P and K limitations differs (de Groot *et al.* 2003 a, de Groot *et al.* 2003 b, Epstein and Bloom 2005); this may be due to the different functions of these nutrients in plants. Limitation of phosphate leads to both molecular and developmental adaptations (Raghothama 1995, Schachtman *et al.* 1998, Lopez Bucio *et al.* 2002).

Pigeonpea (*cajanus cajan* L. Millsp.) is an important protein crop used extensively as a substitute for extensive animal protein in developing countries. Pigeonpea is a legume crop widely cultivated as an intercrop in semi-arid regions, it is generally observed to yield better than other pulse crops in low phosphorus soils. Possible reasons for better efficiency in phosphorus include (i) an extensive rooting habits for better absorption (ii) strong mycorrhizal development and (iii) the ability of pigeonpea to extract more soil phosphorus at low level normally unavailable to other crop plants. So, the crop is tolerant to low phosphorus supply and grows well in phosphorus deficient soils of the tropical environment. Variations in P utilization efficiency also exist among the different cultivars of pigeonpea. The efficient cultivars absorb more P from the soil under limiting conditions and obtain high yield through an effective translocation of absorbed P to the leaves for photosynthesis. (Adu-Gyamfi *et al.*,1989). Phosphorus is an important plant

macronutrient, making up about 0.2% of a plant's dry weight. Plants can't grow without a reliable supply of this nutrient (Schachtman *et al.* 1998). It is second only to N as the most limiting element for plant growth (Bielecki, 1973; Vance *et al.*, 2000). The amount of P in plants ranges from 0.05% to 0.30% of total dry weight. The plant availability of P is very low in soils, as it is easily locked up by the presence of iron (Fe), aluminium (Al) and calcium (Ca) ions. Phosphorus efficiency is defined as the ability of a plant genotype to acquire P from the rhizosphere solution and to incorporate or utilize it in the production of total above or below ground level plant biomass or utilizable plant materials such as seed, grain, fruits, forage etc. (Blair, 1993). Thus it can be assumed that efficiency of genotypes is in fact their response to P-deficiency stress and genotypes having the same yield may behave differently with respect to their P-deficiency stress tolerance.

It is estimated that over 50% of common bean production in tropical soils is limited by P-deficiency (CIAT, 1992). P fertilization is frequently not economically feasible for resource limited farmers in developing world and is often inefficient in tropical soils due to fixation of P in insoluble forms by iron and aluminium oxides. (Sanchez & Uehara, 1980). Therefore fertilization is not an adequate solution in itself. Use of P fertilizers in the soil leads to problem of environmental pollution. So, there is a dire need to minimize the P as per need. Ahmad *et al.* (2001) reported that by using the crop with high efficiency will enhance the phosphorus utilization & will also protect the environmental pollution in developed countries where the P fertilizers are being used in excess. So to minimize the P use, we have to evaluate such bean genotypes which are capable

of producing economic yields at low levels of native or added P. Such genotypes might be superior due to a greater ability to recover phosphorus from fertilizer and/or native fractions of soil P, or due to greater productivity per unit of P absorbed. For present purposes, efficient genotypes will be defined as those that are capable of producing superior yields under P-limiting conditions, irrespective of the mechanism involved (Beebs *et al.*, 1997)

Fresh and Dry weight of different plant parts:

Fresh and Dry weight of plants contribute toward the biomass of the plant. However, dry weight is considered as a better parameter for biomass production. As fresh weight of any plant can vary because of differential condition of the water whereas dry weight include the result of all metabolic processes which the plant has undergone. Better dry weight production per unit time per unit area will indicate the better efficiency of the plant. Fresh and dry weight of the plant is correlated with the application of various nutrients which are the raw material for the production of biomass.

Dixit *et al.* (1983) reported that phosphorus increased plant growth of Bengal gram. Yahiya *et al.* (1995) observed that phosphorus application increased the shoot dry weight. Dry matter yield was also increased significantly by an increased level of phosphorus (Parihar and Tripathi, 1989). Similar results were obtained by Adu-Gyamfi *et al.* (1989).

Fresh and dry weight of sorghum increased with increasing quantity of residual phosphorus and P application. Phosphorus use efficiency was maximum with 25g P ha⁻¹ and minimum with 50kg P

ha⁻¹ in loam soil (Munir *et al.*, 1999). Different quantities of residual P in different soils significantly improved fresh yield (t ha⁻¹) and quality traits like dry matter content. In sorghum fodder (Rashid and Ranjha *et al.*, 2007). Shoot dry matter yield and root dry matter weight increased with increasing P application levels upto 80kg P ha⁻¹ (Osman *et al.* 2002).

Tsvetkova and Georgiev (2003) studied that phosphorus deficiency treatments decreased the whole plant fresh and dry mass. Low P treatment (0.03 ppm) significantly reduced shoot weight, root length & root surface area in differ cultivars compared to control (1.0 ppm) reported by Joseph *et al.* (1991). Application of KH₂ PO₄ @ 200mg Kg⁻¹ to legumes increased fresh root weight, dry matter accumulation & P uptake over the control (Tamboli *et al.*, 2001).

Fujita *et al.* (2004) also reported that a low phosphorus supply significantly decreased the plant biomass & P uptake in all the genotypes studied. In all the pigeonpea cultivars, the maximum whole plant dry weight was obtained in a P-deficient soil at 100kg P ha⁻¹ application. Fageria and da Costa (2000) also reported that, genotypes which produce a lot of dry matter in a soil with a low P level and respond well to added P are the most desirable because they are able to express their high yield potential to a wide range of P environments.

Increasing fresh and dry weight with P application was also observed in soyabean (cassman *et al.* 1981; Israel, 1987), clover (Powell, 1977; Robson *et al.*, 1981), common bean (Pereira and

Bliss, 1987, 1989), pigeonpea (Itoh, 1987), and cowpea (Cassman *et al.*, 1981).

Leaf area, photosynthetic rate and photosynthetic pigment (chlorophyll a, b & carotenoids):

Crop productivity and yield depend on many physiological processes and environmental factors. But among them main physiological process is photosynthesis which contributes about 90% of total dry matter. A plant must process an efficient photosynthetic mechanism to be highly productive biologically. Photosynthetic productivity depends upon leaf area development, leaf age, growth stage. Photosynthesis also varies time to time during day periods depending on the dynamics of light intensity, air temperature and relative humidity of the day. (Biswas *et al.* 2001).

Genotypic variability in photosynthetic rate has been identified within several crop species (Mahon, 1990; Babu *et al.*, 1985). Some have reported a negative correlation between leaf area and photosynthetic rate (Bhayasari and Brown, 1986) while other showed that such relationship was consistent (Watanable and Tabuchi, 1972). Hesketh *et al.* (1981) demonstrated positive correlation between leaf photosynthetic rate and chlorophyll content.

Chlorophyll content is an index of organic matter production and plant growth (Mescht *et al.*, 1999; Lahai *et al.*, 2003). This is because increased photosynthesis has been linked to increased chlorophyll content in plants (Chowdhury and Kohri, 2003).

As a result, chlorophyll content is a measurement of physiological activities in plants. Dudeja and Chaudhary (2005) reported that changes in chlorophyll (chl.) content can occur as a result of nutrient deficiency, exposure to environmental stresses and certain herbicides, and differences in irradiance during plant growth. Thereby chl. content can be used to manage nutrient optimization programmes that both improve crop yield and help to protect the environment (Cassman *et al.*, 1993, Peng *et al.* 1996). Relationships between the primary reactions of photosynthesis and chlorophyll a fluorescence is also very useful and provides information about the photosynthetic capability and vitality of plants. (Liehtenthaler,1988; Genty *et al.*, 1989; Schreiber *et al.*,1995; Owens,1996; Strasser *et al.* 1997).

Dry matter production and N₂ fixation in a crop may depend on photosynthesis by leaves and subsequent translocation of photosynthates to the growing plants and nodules. Photosynthesis is influenced by P supply (Mahon, 1983) suggesting that the ability of a cultivar to absorb & translocate an appreciable amount of P to the leaf under the low P-condition should increase its dry matter production.

It has been proposed that P deficiency may reduce crop growth by inhibiting leaf expansion and photosynthesis. Leaf area development was very sensitive to P supply. Phosphorus deficiency primarily reduced leaf area by diminishing the number of leaves through the effects on number of nodes, branching and relative leaf appearance rate and secondarily by reducing leaf expansion (Lynch

et al., 1991). Yahiya *et al.* (1995) observed that phosphorus application increased leaf area. Similar findings were reported by Idris *et al.* (1989) in an experiment on chickpea.

Nodule weight, Leghaemoglobin content and nitrogenase activity:

Biological N₂ fixation is the process that changes inert N₂ to biologically useful NH₃. This process is mediated in nature only by bacteria. In legumes and a few other plants, the bacteria live in small growths on the roots called nodules. Within these nodules nitrogen fixation is done by the bacteria, and the NH₃ produced is a partnership between a bacterium and a plant.

The symbiotic association of leguminous plant with (Brady) rhizobium results in the formation of root nodules where *Rhizobium sp.* fix N₂ as a natural process under field conditions. Mature nodules are rich source of soluble plant proteins (Boland *et al.*, 1978), the major nodule protein being leghaemoglobin. It provides anaerobic conditions to bacteria as *Rhizobium* acts in anaerobic conditions. It is O₂ scavenger like haemoglobin. So, Leghaemoglobin content of nodules was estimated as a index of nodule functioning (Basavraj, 2002).

A range of environment disturbances can inhibit N₂ fixation of legume nodules and negative effect of environmental stresses on N₂-fixation may be divided into: (a) effects on plants infection by Rhizobia; (b) effects on nodule growth and development; (c) direct effect on nodule formation (Gonzalez *et al.*, 2001).

Nitrogenase is a key enzyme which is helpful to convert the N_2 of the atmosphere into NH_3 . This enzyme is capable of converting N_2 into NH_3 in three steps with the addition of three molecules of H_2 . Nitrogenase activity is an indicator for the functioning of rhizobium in all species. Higher nitrogenase activity will lead down to more protein synthesis.

The Brady/Rhizobium-legume symbiosis is based on the exchange of nutrients between symbionts (Kahn *et al.*, 1985). Of the studies on mineral requirements of symbiosis (reviewed in Robson, 1983), phosphate (P) has received considerable attention due to dramatic effect to nodulated legumes in low P soils. Studies with soyabean (Cassman *et al.*, 1981; Israel, 1987), clever (Powell, 1977; Robson *et al.*, 1981), common bean (Pereisa and Bills, 1987, 1989), pigeonpea (Itoh, 1987), and cowpea (Casshan *et al.* 1981) have consistently shown a positive response to P fertilization; whole plant N concentration, nodule number, nodule mass, nitrogenase activity were found to increase as a result of P fertilization. Increased nodule specific nitrogen activity resulting from the addition of P implies more efficient nitrogen fixation but reasons for this P response is not understood. Israel (1987) report that external application of N_2 reduces the requirement of the plant for P as less P is taken up by the plant. Reason for above explanation is not clear.

Nodulated plants were grown in N-free nutrient solution at increasing levels of P concentrations in the solution (0.2-5 μMP).

Increased P supply improved plant growth and nodule functioning. When nitrogenase activity (H_2 -evolution) was measured at the 1 and 5 μ MP of P, the activity was more than doubled at the 5 μ MP supply than 1 μ MP. Coinciding with the improved nitrogenase activity, nodule number and plant P and N content improved with higher P-supply. (Sulieman *et al.* 2008). As reported by Adu-Gyamfi *et al.* (1989) that the high P concentrations in the nodules of all the cultivar of pigeonpea suggested that nodules have advantages over host plant in terms of P distribution under P deficient conditions. The results suggest that P plays an important role in dinitrogen fixation through an effective translocation of P to the leaf. Thus when P supply is limited, efficient cultivars obtained reasonably high yield through an effective translocation of the absorbed P to the leaf.

Graham and Rosas (1979), Robson *et al.* (1981) and Jakobsen (1985) have observed that P increases symbiotic dinitrogen fixation by stimulating host plant growth rather than exerting a direct effect on nodule initiation, growth, development & functions.

On the other hand, Gates and Wilson (1974), Casshan *et al.* (1981) and Bonetti *et al.*, (1984) have demonstrated the effects of P on rhizobium growth, nodule growth & function. Israel (1987) found that P has specific roles in nodule initiation, growth and function in addition to its involvement in host plant growth process.

Significant increase in number, dry weight and N content of nodules due to phosphorus application was also observed in chickpea by many scientists (Parihar and Tripathi, 1989 Kumpawat and Manohar, 1994; Kumpawat and Manohar 1994 b). These results were confirmed by Singh and Bajpai (1990) on soyabean.

Tsvetkova and Georgiev (2003) reported that phosphorus deficiency treatments decreased nodule weight, number & functioning. Under conditions of phosphorus oversupply the decrease in plant growth, nodulation and acetylene reduction was stronger.

The influence of P on N₂ fixation and dry matter production of young pea (*Pisum L.* cv. Bodil) plants grown in a soil sand mixture was investigated in growth cabinet experiments by Iver Jakobsen (2008). He observed that nodule dry weight, specific C₂H₂ reduction and P concentration in shoot responded to P addition before any growth response could be observed.

The P concentration in nodules responded only lightly to P addition. The effect of phosphorus and zinc on nodulation and nitrogen fixation was studied by Shukla and Yadav (1982) in chickpea. They conducted an experiment on the effect of P and Zn nodulation & N₂ fixation in chickpea (*cicer arietinum L.*) by using treatment combinations of five levels of P (0, 25, 50, 100 & 250 ppm) & six levels of Zn (0, 5, 10, 20, 40 & 100 ppm.) The number, dry matter, leghaemoglobin content of nodules and amount of N fixed generally increased with Zn alone upto 19ppm and P alone upto 50ppm and decreased with higher levels. The results showed that balanced P & Zn nutrition is essential not only for plant growth but also for maximum activity of Rhizobium for N₂ fixation.

Root hairs, root length & root-shoot ratio:

Roots are the most important organs meant for absorption of various nutrients from soil. The various parameters of roots such as length of root, primary and secondary branches of root and number

and length of root hairs are most important contributing characters which helps in absorption. Majority of water and the nutrients are absorbed by epidermal cells and root hair of the roots. Root hairs are tubular shaped outgrowths of epidermal cells that plays an important role in acquisition of water and nutrients, especially phosphate.

Among the traits for P efficiency root hairs are believed to be very important for P acquisition by increasing the soil volume explored by the plant (Clarkson, 1985; Peterson and Farquhar, 1996). Studies with many species showed a significant correlation between radius, density and length of root hairs and P content in plants, indicating a close relationship between root hairs and P uptake from soils (e.g., Foshe *et al.*, 1991 ; Hottmann and Jung, 1995). In barely and wheat, genotypic variation in root-hair length could significantly affect P uptake from the soil. (Gahoonia *et al.*, 1979; Gahoonia & Nielsen, 1997).

Root growth, leading to increase root-soil contact, is a important factor determining the uptake of less mobile nutrients such as phosphorus in soils (Schjerring and Nielsen, 1987). Root hairs increase root-soil contact at low carbon cost. (Clarkson, 1991). Selection for root hairs may be a feasible option for improving P acquisition of crop plants (Caradus, 1994). Root hairs are believed to absorb nutrients (Fohse *et al.*, 1991 ; Itoh and Barber 1983a) and water (Cailloux, 1972). Root hairs can exploit soil in the immediate vicinity of root surface more effectively due to their geometrical arrangement on roots, and ability to increase root surface area (Classen, 1990). Previous studies (Fohse *et al.* 1991;

Itoh and Barber, 1983 a) have shown the importance of root hairs in P uptake differences between plant species. The extent of P depletion zone close to roots can be extended by root hairs (Bhat *et al.*, 1976; Lewis and Quirk 1967).

Plants typically respond to P deficiency by allocating more carbon to roots, thus increasing their root to shoot ratio. In addition, low P availability can dramatically alter the spatial configuration of the root system by increasing root hairs and promoting lateral root formation. In *Lupinus albus*, P deficiency promotes the formation of cluster roots, also termed as *proteoid* roots (Johnson *et al.*, 1996). Increased root branching in response to P limitation has also been reported for *Arabidopsis* (*Arabidopsis thaliana*). Growth of *Arabidopsis* in moderate to limiting supply of P results in a redistribution of root growth from primary to lateral roots (Williamson *et al.*, 2001). Reduced primary root elongation in low P conditions was accompanied by increased root branching, perhaps concentrating root biomass near the soil surface for more efficient nutrient foraging. (Lopez-Bucio *et al.* 2002, 2003).

Root proliferation and root elongation as well as the formation of mycorrhiza are further hallmarks of plant responses to low P availability (Smith & Read, 1997). The root systems of plants show highly plastic movements. One important parameter is the availability of phosphate, which is highly immobile in soils such that the arrangement of roots within the soil will profoundly affect the ability of the plant to acquire this essential nutrient. In *Arabidopsis* low phosphate availability favored lateral root growth over primary root growth, through increased lateral root density and length and

reduced primary root growth mediated by reduced cell elongation (Williamson *et al.*, 2001).

When exposed to phosphorus (P) deficiency white lupin (*Lupinus albus* L.) exudes large amounts of citric acid and malic acid from specialized clusters of closely spaced lateral rootlets (proteoid roots) which have limited growth and density covered with root hairs. As a first visible symptom of P deficiency, cluster root formation started along the oldest lateral roots, approximately 14 days after sowing. (Neumann *et al.*, 1998).

Fohse *et al.* (1981) have shown that P influx per unit root length in seven plant species grown in a low -P soil varied from 0.6×10^{-14} to 4.8×10^{-14} mol cm⁻¹ s⁻¹. The large contribution of root hairs to P uptake was partly due to their surface area, which was similar to that of root cylinder.

In field plots of *Beta vulgaris*, the total root length production over the entire growing season was 3-4 times the size of the living root system at harvest (120km m⁻²) in high P plots, and five times (200km m⁻²) in low-P plots. (Steingrobe, 2001). The author calculated a 25% increase in Pi uptake at low P supply as a result of this enhanced root length production compared with that at the root production of high P-plants. A similarly enhanced root length production, at a low P supply has been observed for *Hordeum vulgare* (Steingrobe *et al.* 2001). Increased root production, without a proportional increase in living-root biomass i.e. enhanced root turnover, allows greater amounts of uptake of immobile soil resources, such as P. (Shane and Lambers, 2005 a).

Root hairs are a fairly common root structure, and increased root-hair length and number are considered to be an adaptation that enhances P_i acquisition and a plants competitive advantage when soil P_i is limiting for growth (Bates and Lynch, 2001). Species that develop more or longer root hairs e.g. *Lotium perenne*, are for more efficient at accessing P_i from soils, and thus show less of a growth response in P-fertilized soils than do species that lack these traits., e.g. *Podocarpus totara* (Clarkson, 1981). This point was elegantly demonstrated in a comparison of genotypes of *Hordeum vulgare*; genotypes with a capacity to form longer root hairs (about 1mm) took up more P, and tended to yield better when P_i was limiting crop growth compared with genotypes having root hairs half the length about (0.5 mm) (Gahoonia and Nielsen, 2004). Root-hair abundance and length is enhanced by P deficiency (Schmidt, 2001).

Legumes such as common bean (*Phaseolus vulgaris* L.) and pigeonpea (*Cajanus cajan* L.) have shown genotypic difference in their ability to obtain P (Boner *et al.* 1996; Subbarao *et al.* 1997). Due to very low mobility of soil P in relation to plant demand, it appears reasonable to look for differences among cowpea genotypes in their volume of soil exploited by roots & root hairs. If root induced processes differ among the genotypes, it is expected that the quantity of P absorbed by the soil volume exploited by roots & root hairs would differ. The lengths of roots and root hairs and the extent of root induced processes affect phosphorus uptake efficiency by plants. (Kresinikott *et al.*, 2003)

Large differences in root morphology and distribution exist between genotypes of plant species (Atkinson, 1991; Nielsen, 1983;

O' Toole and Bland 1987; Romer *et al.* 1988). Leon and Schwang (1992) found that yield stability of oats and barley cultivars was related to their total root length. Root hairs enhance the effectiveness of roots to exploit rhizosphere soil for P; because of their geometrical arrangement on root surface and their ability to increase effective root surface area many times (Fohse *et al.* 1991; Gahoonia *et al.* 1997). The role of root hairs in P uptake is now well documented (Bates and Lynch, 2000; Gahoonia and Nielsen, 1998). A considerable intraspecific variation in root hair formation has been found in genotypes of white clover (Caradus, 1979), common bean (Yan *et al.* 1945), barley & wheat (Gahoonia *et al.*, 1997, 1999) and cowpea (Krasilnikott *et al.*, 2001).

Gahoonia *et al.* (1997) studied the root hair of wheat & barley cultivars in relation to P depletion from rhizosphere in three soils of different P levels. Their results showed that variation in root hairs of cereal cultivars was considerable and it play a significantly role in P acquisition, especially in low P soils.

Gahoonia and Neilson (2004) found out the traits for efficient absorption of P and reported that root morphology, root hairs, mycorrhizal symbiosis, uptake Kinetics, root induced changes in the rhizosphere were mainly related to P uptake efficiency. Root length and root hair length respectively differed among the eight genotypes of cowpea. The length of roots varied between 89 and 365 m pot⁻¹ and the root hairs varied between 0.23 and 0.38 mm as reported by Krasilnikott *et.al.*, (2003). Increase in soil P level reduced the length of root hairs. The variation in root hairs between the cultivars was largest in no-P soils. When 10kg P ha⁻¹ was

applied the root hairs lengths did not differ between the cultivars. Barley cultivars with longer root hairs absorbed more phosphorus from the rhizosphere soil and also absorbed more phosphorus in field. (Hans Lanbers *et al.*, 2006).

Acid and alkaline phosphatase activity:

Soils contain nearly half of the total phosphorus in organic forms, mostly as complex organic esters, in which (P) phosphorus is bound to carbon via oxygen. The importance of soil organic P as a source of plant availability P depends on its rate of solubilization and release as inorganic P in soil solution. Phosphatases may play a specific role in hydrolysis of organic P compounds (Brown and Tabatabai, 1978; Eivazi and Tabatabai, 1977; Juma and Tabatabai, 1988; Sharply, 1985). Soil contains always considerable amount of total phosphatases which is protected by different soil components (Burns, 1982), whereas the activity of soluble enzymes in bulk soil are mostly very low (Asmar, 1992; Asmar *et al.*, 1992). Phosphatases are thus present in soils but their activities in rhizosphere soil may increase due to direct release of extracellular phosphatases from the roots of growing plants and due to the release by the growing microbial biomass utilizing the root exudates (Barber and Lynch, 1976).

Increased phosphatase activity in the rhizosphere soil seems to increase the hydrolysis of soil phosphate esters (Bielski and Johnson, 1972), releasing inorganic P for plant uptake (Tarafder and Jung K. 1987). However, if plants continue to grow poorly in P deficient soils, despite marked increase in root exocellular phosphatase activity (Alexander and Hardy, 1981), it may be

argued that P mobilized through the increased phosphatase activity is not able to compensate for an inadequate supply of inorganic P.

Phosphatases have been traditionally classified as being alkaline or acid phosphatases according to whether their optimal pH for catalysis is above or below pH 7.0 (Vincent *et al.* 1992). Acid phosphatase differs from alkaline phosphatase by displaying optional activity below pH 7.0 and generally having low substrate specificity. Plant alkaline phosphatases generally display absolute substrate specificity. While acid phosphatases do not normally exhibit an absolute substrate specificity.

Acid phosphatase play a pivotal role in plant cell metabolism by regulating inorganic phosphate level as the enzyme causes hydrolysis of p-nitrophenal phosphate into p-nitrophenol and inorganic phosphate (iP). This iP indirectly controls the carbohydrate metabolism of plants. Although many acid phosphatases have been studied, the functions of these enzymes are still unclear because of the difficulties associated with determining the *in vivo* substrates for the various isoenzymes of acid phosphatase present in the plant tissue (Dutt *et al.* 1944).

Both extra and intracellular acid phosphatases appears to be ubiquitous in plants. Intracellular forms occur in all plant tissues in both vacuolar & cytoplasmic locations. While extra cellular acid phosphatase is typically located at the root apical meristem on the outer surface of root cells.

White lupin (*Lupinus albus* L.) develops proteoid roots when grown in phosphorus (P) deficient conditions. These short, lateral,

densely clustered roots are adapted to increase P availability. Roots forms P-deficient (-P) plants had significantly greater acid phosphatase activity in both root exudates and extracts than comparable samples from P sufficient (+P) plants. The increase in activity in -P plants was found most pronounced in the proteoid regions. Phosphorus acquisition by lupin may involve increased secretion of acid phosphatase under P-deficient conditions. Phosphorus deficient lupin secreted twenty times more acid phosphatases activity from the roots than P-sufficient controls and had the greatest levels of acid phosphatase activity of nine crop species tested. (Tadano & Sakai 1991).

When exposed to phosphorus (P) deficiency, white lupin (*Lupinus albus* L.) exudes large amount of citric and malic acid from specialized clusters of closely spaced lateral rootlets (proteoid roots), which have limited growth and are densely covered with root hairs. (Gardner *et al.* 1983; Dinkelaker *et al.* 1989; Gerk *et al.* (1994).

Yield and attributing characters:

Dixit and Patil (1984) studied a set of ninety two varieties for yield contributing characters in soyabean. Yield has been observed to be influenced directly and indirectly by number of pods, number of seeds and number of branches. Seventy-nine varieties of pigeonpea were used in a study on yield per plant was found to be highly and positively correlated with pods per plant and number of secondary branches per plant by More *et al.* (1984). Twenty-nine genotypes of pigeonpea were characterized for a number of traits by Balyan and Sudhakar (1985). They found that yield per plant,

number of secondary branches, number of pods per plant 100-seeds weight showed maximum genotypic variability. Most of the characters were positively and significantly correlated with yield per plant.

Balanced use of nitrogen and phosphorus increased yield by 15-30 per cent compared with nitrogen alone (National Fertilizer Development Centre 1990). Dixit *et al.* (1983) found that application of phosphorus gave a higher yield compared with no fertilizer. Borhohain and Agarwal (1986) reported that seed yield of chickpea increased with increasing levels of phosphorus. 80kg P_2O_5 ha⁻¹ gave the highest yield of 1.33 tonnes ha⁻¹ compared with 0.79 tonnes ha⁻¹ (control). Similar observations were recorded by Tomar *et al.* (1988). They reported that chickpea yields with 25, 50 and 75 kg P_2O_5 ha⁻¹ were 2.07, 2.26 and 2.18 tonnes ha⁻¹ compared with 1.89 tonnes ha⁻¹ without phosphorus (control). These results were confirmed by Shanim and Naimat (1987), Parithar and Tripathi (1989), Nadeem (1999) and Takankhar *et al.*, (1998).

Dry matter yield was also increased significantly by an increased level of phosphorus (Parihar and Tripathi, 1989). Yahiya *et al.* (1995) reported that phosphorus application increased photosynthetic efficiency and seed yield in chickpea. Phosphorus application increased the seed yield in red chickpea (Reddy *et al.* 1993). Similar results have been reported by others (Tomar *et al.* 1988); Karet *et al.* 1989; Kumpawat *et al.* 1990; Jat and Mali 1992; Gawad *et al.* 1993; Verma and Yadav, 1993; Verma, 1994; Kasole

et al., 1995; Mishra, 1995; Sarkar *et al.*, 1995; Singh, 1995; Raghuwanshi *et al.*, 1997; Yadav and Shrivastava, 1997; Gupta *et al.*, 1998. In a trial on chickpea, Takankhar *et al.* (1998) reported that application of 76kg P₂O₅ ha⁻¹ produced the highest seed yield of 1.25 tonnes ha⁻¹.

CHAPTER-III

Materials & Methods

The present investigations were carried out on six genotypes of Pigeon pea (*Cajuns Cajon* L. Mills.) viz. H2001-37, Paras, H2000-37, H2001-4, ICPL-88039 and H2000-47 in the green house of Department of Botany and Plant physiology in College of Basic Sciences & Humanities during. Seeds of these pigeonpea genotypes were obtained from Department of Plant Breeding, College of Agriculture, CCS HAU, Hisar.

Experiment 1: Screening of genotypes for Phosphorus uptake efficiency.

The crop was raised in earthen pots with 5kg of dune sand in screen house under natural conditions. The seed of uniform size were selected and sterilized with 1 percent of sodium hypochlorite solution for 2-3 minutes, then washed 4-5 times with distilled water and dried on filter paper. The sterilized seeds were treated with rhizobium culture provided by Department of Microbiology, HAU, Hisar. Finally five seeds were sown in each pot at approximately uniform depth (2cm) and distance (5cm). One week after germination thinning was done to leave three plants of comparable growth in each pot. A starting dose of 5µm NH₄NO₃ was given in the beginning, only once which helps in nodule formation.

Nutrient Solution:

Distilled water was used for the preparation of nitrogen free nutrient solution (Wilson and Reisenauer, 1963).

For different P treatments, the P concentration in the Wilson and Reisenauer (1963) will be considered as 100% (as given Table-a). The modified nutrient solution will be:-

Table – a:

	Compound	Concentration (M)	Weight
A.	Macronutrients		
(i)	(a) KH_2PO_4 (100%)	2.0×10^{-3}	54.4g/2L
	(b) KH_2PO_4 (60%)	2.0×10^{-3}	32.04g/2L
	(c) KH_2PO_4 (30%)	2.0×10^{-3}	16.32g/2L
	(d) KH_2PO_4 (15%)	2.0×10^{-3}	8.160g/2L
(ii)	KH_2PO_4	2.0×10^{-3}	69.6g/2L
(iii)	CaSO_4	3.0×10^{-3}	81.6g/2L
(iv)	MgSO_4	1.0×10^{-3}	49.2g/2L
B.	Micronutrients		
(i)	$\text{MnCl}_2 \cdot 4\text{H}_2\text{O}$	5.0×10^{-5}	0.196g/2L
(ii)	$\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$	2.0×10^{-6}	0.116g/2L
(iii)	H_3BO_3	2.5×10^{-5}	0.308g/2L
(iv)	$\text{C}_4\text{SO}_4 \cdot 5\text{H}_2\text{O}$	5.0×10^{-7}	0.248g/2L
(v)	Na_2MoO_4	5.0×10^{-7}	0.242g/2L
(vi)	$\text{CoCl}_2 \cdot 6\text{H}_2\text{O}$	1.7×10^{-7}	0.80g/2L
(vii)	CaCO_3	2g/2L	(At the time of mixing)

(viii)	Ferric Citrate	2mg/2L	Freshly Prepared
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The plants were supplied with 250ml nutrients solution per pot at weekly intervals. The treated plants were supplied with modified nutrient solution.

Sampling:-

Sampling was done at three stages.

- (i) Vegetative stage (Approx. 40-50 days after sowing)
- (ii) Reproductive stage (Approx. 70-80 days after sowing)
- (iii) Pod filling stage (Approx. 100-120 days after sowing)

Three plants were selected for sampling from each treatment at each stage.

Statistical Analysis: Factorial Complete Randomized Design (Factorial-CRD) was used for all parameters for statistical analysis.

Observation: Following observation were recorded at three sampling stages:

1. Fresh weight of root (g/plant)
2. Fresh weight of shoot (g/plant)
3. Dry weight of root (g/plant)
4. Dry weight of shoot (g/plant)
5. Leaf area (cm²)
6. Total chlorophyll content (mg/g fresh weight)

7. Photosynthetic rate ($\text{mg CO}_2 \text{ fixed min}^{-1} \text{ cm}^{-2}$)
8. Phosphorus content of root (mg/plant)
9. Phosphorus content of shoot (mg/plant)
10. Nitrogen content of root (mg/plant)
11. Nitrogen content of shoot (mg/plant)
12. Fresh weight of nodules (g/plant)
13. Leghaemoglobin content (arbitrary fluorescence units)
14. Nitrogenase activity ($\mu\text{mole C}_2\text{H}_4 \text{ produced h}^{-1}\text{g}^{-1}$ fresh weight of nodules)
15. Yield and its attributing characters (g/plant)

Methods:

- **Fresh weight:** The plants were uprooted from the soil and their components i.e. leaves, stem, root and nodules were separated and their fresh weights were taken at desired sampling stages i.e. 45, 75, 110 days after sowing.
- **Dry weight:** One plant from each replication was uprooted, washed & its root, stem, leaves were separated and wrapped in a piece of paper and was kept at room temperature for one day. Then dried in oven at $70\text{-}80^\circ\text{C}$ till constant weight was obtained. The dry weight of different organs of the plant of different treatment at different sampling stages was noted.

- **Photosynthetic rate:** For measuring photosynthetic rate, third leaf from the top of the plant was taken at every sampling stage. Photosynthesis was measured by portable Infra Red Gas Analyser (IRGA) (CIRAS-1, PP systems, England) between 9.00 to 10.00 AM at the desired sampling stages i.e. 45, 75, 110 days after sowing.
- **Total Chlorophyll:** For the purpose of chlorophyll content third leaf from the top at each sampling stage was taken from the plant and chlorophyll a, chlorophyll b and total chlorophyll were estimated by Dimethyl Sulphoxide (DMSO) method of Hiscox and Israelstom (1979) at all sampling stages from fresh leaf of the plant. 100mg of leaf material was suspended in a test tube containing 5ml DMSO. The test tube were then placed in an oven at 60°C for about 2hr to facilitate the extraction of pigment. After 2 hr, the absorbance was read at 454, 645 and 665 nm on spectrophotometer. DMSO was used as blank. Calculations for different pigments were made according to following equations:

$$\text{Chlorophyll a } \mu\text{g ml}^{-1} = (12.19A_{665} - 3.45A_{645})$$

$$\text{Chlorophyll b } \mu\text{g ml}^{-1} = (12.19A_{645} - 5.32A_{665})$$

$$\text{Carotenoids } \mu\text{g ml}^{-1} = [1000 A_{454} - 2.86 \times \text{chl (a)} - 129.9 \times \text{chl (b)}] / 221$$

$$\text{Chlorophyll 'a' content} = (\mu\text{mol g}^{-1})$$

Chlorophyll 'b' content =
($\mu\text{mol g}^{-1}$)

Carotenoid content =
($\mu\text{mol g}^{-1}$)

Total chlorophyll = chl 'a' + chl 'b'

Estimation of Nitrogen and phosphorous content

Digestion of sample

0.5 gm of dry samples of leaf, stem and root were taken in a 50 ml conical flask. 10 ml of diacid mixture of HNO_3 and HClO_4 was added which was mixed (in a ratio of 4:1) and kept for over- night. Next day conical flasks were kept on a hot plate and heated gently at first. Then heated more vigorously until a clear colorless solution resulted or till white fumes cease to come out. Do not take it to dryness. Heating was discontinued, when the volume was reduced to 3-4 ml. Solution was cooled and volume was made upto 50ml by adding distilled water. Then it was filtered through Whatman No. 1 filter paper and used for further analysis.

Total P in plant samples was determined by (Vanadomolybdophosphoric yellow color method as Jackson (1973)).

(A) Reagents

- Vanadate-molybdate reagent: Prepare solution 'A' by dissolving 25 g of ammonium molybdate in about 400 ml of

warm water. Prepare solution 'B' separately by dissolving 1.25 g of ammonium metavanadate in about 300 ml of warm (70°C) water, cool at room temperature and add 250 ml of concentrated HNO_3 and cool again at room temperature. Now add solution 'A' to solution 'B' slowly with continuous stirring and dilute to one litre with distilled water.

- 6N HCl solution: Dilute 517 ml of concentrated HCl to one litre.
- 2,4 di-nitrophenol indicator, 2.5%: Dissolve 2.5 g of 2,4 di-nitrophenol in 100 ml of 95 per cent ethanol.
- Ammonia solution (%).
- Preparation of standard curve: Dissolve 0.2195 g of dried KH_2PO_4 in water, acidifying with 25 ml of 7N H_2SO_4 and dilute to 1 litre. This solution contains 50 ppm P. From 50 ppm standard P solution. Take appropriate volumes to prepare working solutions of 0.05, 0.1, 0.2, 0.3, 0.4, 0.8 and 1.0 ppm. Develop the color in identical manner. Prepare standard curve by plotting P concentrations on X-axis and per cent transmittance on Y-axis on a semi-log graph paper.

Procedure:

Color development for phosphorous estimation

1. 1ml of aliquot (digested plant material) was taken in a 25 ml volumetric flask.
2. 2-3 drops of 2,4 di-nitrophenol indicator were added to it.
3. Ammonia solution was added till yellow color appears and now add 6N HCl (drop wise) till it become colorless.

4. Then 5 ml of vanadomolybdate solution was added and diluted to 25 ml with distilled water.
5. Mix content thoroughly and read the intensity of yellow color on spectrophotometer (Spectronic 20) by using blue filter at 440 nm wavelength.
6. Run a blank without P solution simultaneously.
7. Calculate P content of sample using the standard curve.

(B) Calculations

$$\begin{aligned}\text{Total P (ppm) in plant sample} &= A \times 250 \\ \text{Total P (\%) in plant sample} &= (A \times 250)/10000 \\ &= A \times 0.25\end{aligned}$$

where ,

$$\begin{aligned}\text{Transmittance (\%)} \text{ of the test solution} &= T \\ \text{Concentration of P as read from standard} &= A \text{ ppm} \\ \text{curve against transmittance}\end{aligned}$$

Total Nitrogen in plant samples was determined by Colorimetric method (Nessler's reagent) (Lindner, 1944).

(A) Reagents

- 10% sodium hydroxide solution: 100 g of AR grade NaOH was dissolved in 1 litre distilled water.
- 10% sodium silicate solution: 100 g of AR grade sodium silicate was dissolved in 1 litre of distilled water.
- Nessler's reagent: 45.5 g of mercuric iodide (HgI_2) was weighed and placed it in 1 litre volumetric flask. 35 g of KI in about 400 ml of distilled water was dissolved in a 500 ml beaker. KI solution was dissolved to the flask containing HgI_2

slowly by rotating the flask, so that all HgI_2 is dissolved. Take all-other 500 ml beaker and dissolve 112 g of KOH in about 400 ml distilled water. Cool it and add to HgI_2 solution and make the volume up to 1 litre with distilled water. Keep it for overnight, because some precipitation is there which gets settled down. Next day, use the supernatant liquid in analysis. The solution is called Nessler's reagent.

- Preparation of standard solution and standard curve: Stock solution of 50 ppm N from any nitrogenous compound viz. EDTA was prepared. From the stock solution, appropriate quantity was taken to prepare the working standard solutions of 0.5, 1, 1.5, 2, 2.5 and 3 ppm N. Add all the reagents except NaOH (because here, there is no acidity). Prepare standard curve by plotting % transmittance (T) on Y-axis and concentration on X-axis on a semi-log graph paper.

Procedure:

Color development for nitrogen estimation

1. 1 ml of digested plant material (depending upon the N content in plant) was taken in a 25 ml of volumetric flask and about 5 ml of distilled water was added.
2. 1ml of 10% NaOH was added to neutralize the acidity of solution. Add 1 ml of 10% sodium silicate (to avoid the turbidity) and then make the volume to about 20 ml with distilled water.
4. 2 ml of Nessler's reagent was added, it will give orange color

complex slowly and make the volume up to file mark with the help of distilled water.

5. Read the intensity of color on spectrophotometer (Spectronic 20) by using blue filter at 440 nm wavelength.
6. Calculate N content of sample using the standard curve.

(B) Calculations:

$$\text{Total, N (ppm) in plant sample} = A \times 12500$$

$$\begin{aligned} \text{Total N (\%) in plant sample} &= (A \times 125.00)/10,000 \\ &= A \times 1.25 \end{aligned}$$

where,

$$\text{Transmittance (\%) of the test solution} = T$$

$$\text{Concentration of N as read from standard} = A \text{ ppm}$$

curve against transmittance T

Nodule leghaemoglobin

The nodule leghaemoglobin was determined by using saturated oxalic acid solution. 100 mg of pea nodules were ground in 1 ml of potassium phosphate buffer (pH 7.0, 0.1M) and 10% PVP. The ground mixture was centrifuged at 4⁰ C for 10 minutes. Supernatant was used for further estimations. To 0.2 ml aliquot of supernatant, 2 ml of saturated solution of oxalic acid was added and the mixture was autoclaved at 120°C for an hr. To again 0.2 ml aliquot of supernatant, 2 ml of saturated solution of oxalic acid was added and the mixture was not autoclaved. It was cooled in ice water and the crystals of oxalic acid were removed by centrifugation. Fluorescence was measured at 403 nm excitation

and 602 nm emission wavelength by Spectrofluorometer (Elico SL-174). Leghaemoglobin was measured in arbitrary units by taking difference of unautoclaved samples and autoclaved samples (Becana and Klucas, 1992).

Determination of ARA of root nodule system

- Uproot legume plant and shake gently to remove adhering soil.
- Detach the root system along with nodules.
- Transfer the whole nodulated root system to assay vials or assay flasks.
- Insert serum stopper and replace 10% of air with equal volume of freshly prepared acetylene.
- Incubate the assay vials for 30-60 min at 28°C.
- Measure the ethylene formed using Gas Liquid Chromatograph (GLC). Using porpack column with oven temperature of 90°C and temperature of injector and detector was kept at 120°C. Standard ethylene graph was prepared and calculations were done with a standard ethylene graph prepared separately.
- Detach the nodules from root. Take fresh weight or dry them in oven at 80°C for 24 hour and take dry weight.
- Determine the volume of air space in the assay vials/assay flasks.
- Calculate C_2H_4 formed by using standard formula.

Calculation for ARA:

Standard ethylene gas is supplied as compressed gas in air tight cylinder. Standard ethylene comes as mixture with inert gases like argon. The volume of ethylene in gas mixture is mentioned on the cylinder and can be 110 vpm or 1050 vpm (volume per million).

To calculate C_2H_4 concentration, let us consider a standard ethylene cylinder with a volume of 110 vpm.

A. According to Avogadro's law
 22,400 ml of any gas at NTP = 1 mole (M)

1 ml of any gas having 110 vpm ethylene = $1 \times 110 \times 10^3 / 22400$

= 4.91 nmoles

B. 1.0 ml of standard ethylene at X 8
 attenuation of GLC showed = 54 chart units

(C.U.)

Thus 1.0 ml of standard ethylene = 54 C.U. = 4.91
 nmoles

1 chart unit = $4.91 / 54$ nmoles

Suppose nodule sample or culture
 growth in a tube showed attenuation = BC.U. at X 64

The units of C_2H_4 formed after using the formula will be nmoles of C_2H_4 produced $h^{-1}mg^{-1}$ protein in free living cultures. In nodules, the unit of ARA will be umoles of C_2H_4 produced $h^{-1}g^{-1}$ fresh/dry weight of nodules.

Experiment 2: Identification of characters associated with phosphate uptake efficiency.

Sterilized seeds were germinated in petriplates and transferred to culture tubes containing 0.3% agar-agar medium with different phosphorus concentrations i.e. 15%, 30%, 60% and 100%.

Observations: Following observations were recorded at 10 DAS:

1. Root length (cm)
2. Number of root hairs.
3. Root-shoot ratio.
4. Acid phosphatase activity of roots (μmol of p-nitrophenol released $\text{min}^{-1}\text{mg}^{-1}$ protein)
5. Alkaline phosphatase activity of roots (μmol of p-nitrophenol released $\text{min}^{-1}\text{mg}^{-1}$ protein)

Root Length:

Average root length was measured with the help of a scale. Average of root length was expressed.

Root Hairs:

Root hairs were counted after staining with a temporary stain and placing it under the microscope. The average numbers of root

hairs were expressed.

Estimation of Phosphatase Enzyme:

Preparation of crude extract for enzyme studies: Roots were homogenized in a chilled glass mortar with a pestle using acid washed sand as abrasive. The extraction medium (pH 7.4) containing 0.05 M tris-HCl buffer (pH 7.4); 0.4M sucrose; 0.1 M KCl; 10 mM MgCl₂; 0.01 M EDTA (Ethylene diamine tetra acetic acid); 0.01 M mercapto-ethanol and 2.5% PVP (Polyvinyl pyrrolidone) (Copper and Beever, 1969). The homogenate was centrifuged at 16000 rpm for 30 minutes at 0-4° C. The supernatant was used for estimating the activities of alkaline phosphatase.

Alkaline phosphatase activity:

Assay method of Jones (1969) was employed using

Pnitrophenyl phosphate as substrate. The pH optima for alkaline phosphatase was determined and was found to be 7.8 for alkaline phosphatase.

Substrate solution:

P-nitrophenyl phosphate (5 mg/10 mg)	10 ml
0.1 M MgCl ₂	1 ml
0.1 M tris-HCl buffer (pH 7.8)	30 ml
Total	41 ml

Procedure:

The incubation mixture consisting of 1 ml of enzyme extract and 1 ml of substrate solution was incubated at 37°C for 20 minutes. Reaction was stopped with 5 ml of 0.1 N NaOH and the p-nitrophenol liberated was determined by measuring colour

intensity at 405 nm. A standard curve (0.05 K mole-0.25 p mole) was also prepared by using pure p-nitrophenol. The activity of enzyme was expressed as moles of p- nitrophenol/hour under specific experimental conditions.

Acid phosphatase:

It play a pivotal role in plant cell metabolism by regulating inorganic phosphate level as the enzyme causes hydrolysis of p-nitrophenol phosphate into p-nitrophenol and inorganic phosphate (iP). This iP indirectly control the carbohydrate metabolism of plants. Assay of acid phosphatase was carried out by method of Jones (1969) using p-nitrophenyl phosphate as a substrate.

Reagents:

- (a) p-nitrophenyl phosphate – 0.5 mg/ml
- (b) 0.1 M $MgCl_2$
- (c) 0.1 M acetate buffer (pH 4.8)
- (d) Substrate solution : Reagents a, b and c were mixed in the ratio of 10:1:30.
- (e) 0.1 N NaOH

Procedure:

Mixture of 50 μ l of enzyme extract and 1 ml of substrate solution was incubated at 37°C for 20 min. The solutions were then made alkaline with 5 ml of 0.1 N NaOH to stop the reaction and O.D. of yellow coloured product i.e. p-nitrophenol was recorded at 410 nm. A standard curve was prepared using graded concentrations of p-nitrophenol. Enzyme activity was expressed as pmol of p-nitrophenol released per min per mg of protein.

CHAPTER-IV

Experimental Results

Present investigations were carried out under two experiments and the results obtained are as under:

EXPERIMENT-I

1. Fresh weight of root (g/plant):

Fresh weight of root in different genotypes of pigeonpea increased with increasing level of phosphorus. On 45th day after sowing, 15% P showed 3.05 g/plant whereas in 100% P application this fresh weight increased to 6.90 g/plant, when considered irrespective of variety. Similarly different genotypes also show differential behaviour. Minimum fresh weight was observed in H2001-4 i.e. 2.70 g/plant followed by ICPL-88039 i.e. 3.54 g/plant. The maximum fresh weight was observed in H2000-47 i.e. 5.82 g/plant followed by Paras i.e. 5.46 g/plant, when considered irrespective of phosphorus level. Among the interactions minimum fresh weight was observed in H2001-4 with 15% phosphorus i.e. 1.89 g/plant and maximum was in H2000-37 at 100% phosphorus i.e. 8.44 g/plant.

On 75th day after sowing, 15% phosphorus showed 7.5 g/plant whereas in 100% phosphorus this fresh weight increased to 22.89 g/plant, when considered in terms of phosphorus level application. Similarly different genotypes also show differential

behaviour. Minimum fresh weight was observed in H2001-37 i.e. 11.44 g/plant followed by H2000-37 i.e. 12.81 g/plant. The maximum fresh weight was observed in ICPL-88039 i.e. 16.16 g/plant followed by H2001-4 i.e. 15.86 g/plant, when considered irrespective of phosphorus level. Among interactions minimum fresh weight was observed in genotype H2000-47 with 15% of phosphorus application i.e. 5.85 g/plant and maximum was observed in ICPL-88039 i.e. 28.81 g/plant with 100% of phosphorus application.

Similarly on 110th day after sowing fresh weight of root increased with increasing level of phosphorus. 15% phosphorus application showed 8.45 g/plant where as 100% of phosphorus application fresh weight increased upto 23.45 g/plant. Different genotypes also showed differential behaviour. Minimum fresh weight was observed in genotype H2000-47 i.e. 12.85 g/plant followed by H2001-37 i.e. 12.93 g/plant. The maximum fresh weight was recorded in ICPL-88039 i.e. 16.80 g/plant followed by H2001-4 i.e. 16.73 g/plant, when considered in terms of genotypes. Among the interactions, maximum fresh weight of root was observed in H2000-37 with 100% of phosphorus application (28.88 g/plant) while minimum fresh weight was recorded in ICPL-88039 with 15% of phosphorus application (i.e. 12.51 g/plant).

In general in all genotypes the fresh weight of roots increased with increasing levels of phosphorus and all genotypes with 100% of phosphorus application resulted in maximum fresh weight while minimum fresh weight was observed in genotypes with 15% of

phosphorus application. Genotype H2000-47 and H2001-4 showed maximum fresh weight of root even with 15% of phosphorus application. There was not much reduction in fresh weight of root at low phosphorus application. Hence, on the basis of fresh weight H2001-4 and H2000-47 genotypes are most efficient pigeonpea genotypes with low level of phosphorus.

Table :1. Effect of different levels of phosphorus application on fresh weight of root (g)/plant at different stages in different genotypes of pigeonpea (*Cajanus Cajan* L. Millsp.) at 45, 75 & 110 days after sowing.

45 days						75 days					110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mea n	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H2001-37	3.57	4.16	4.99	6.96	4.92	7.64	11.30	11.66	15.15	11.44	9.31	12.46	13.98	15.98	12.93
PARAS	3.17	4.86	6.00	7.82	5.46	6.53	11.64	15.12	20.81	13.52	6.83	13.12	16.95	26.19	15.77
H2000-37	3.11	4.25	4.26	8.44	5.02	6.58	10.00	13.87	20.80	12.81	9.13	12.19	16.87	24.24	15.61
H2001-4	1.89	1.96	2.17	4.78	2.70	9.89	11.93	16.47	25.17	15.86	10.51	14.28	16.78	25.34	16.73
ICPL-88039	2.67	2.77	2.87	5.85	3.54	9.04	11.43	15.35	28.81	16.16	9.02	12.87	16.68	28.62	16.80
H2000-47	3.90	5.73	6.12	7.54	5.82	5.85	12.42	16.10	22.62	15.24	5.90	8.85	16.29	20.36	12.85
Mean	3.05	3.96	4.40	6.90		7.59	11.45	14.76	22.89		8.45	12.29	16.26	23.45	

C.D. at 5%

Variety	0.35
Treatment	0.29
Variety x Treatment	0.71

C.D. at 5%

Variety	0.48
Treatment	0.39
Variety x Treatment	0.97

C.D. at 5%

Variety	0.88
Treatment	0.72
Variety x Treatment	1.75

2. Fresh weight of shoot (g/plant):

Fresh weight of shoot in different genotypes of pigeonpea increased with increasing level of phosphorus. On 45th day after sowing, fresh weight with 15% phosphorus application was 4.59 g/plant whereas in 100% phosphorus application this fresh weight increased to 7.62 g/plant, when considered irrespective of genotypes. Similarly different genotypes also show differential behaviour in terms of fresh weight of shoot. Maximum fresh weight was recorded in ICPL-88039 i.e. 6.53 g/plant followed by H2001-37 i.e. 6.33 g/plant. The minimum fresh weight was observed in H2000-47 i.e. (5.25g) g/plant followed by H2001-4 (5.32g/plant), when considered in terms of genotypes. Among the interactions minimum fresh weight was observed in Paras at 100% of phosphorus i.e. 8.95g/plant and minimum fresh weight was recorded in H2000-37 with 15% of phosphorus application i.e. 4.34 g/plant.

On 75th day of sampling after sowing maximum fresh weight of shoot was observed with 100% of phosphorus application i.e. 26.90 g/plant while minimum fresh weight was recorded at 15% phosphorus i.e. 20.05 g/plant, when considered as mean basis irrespective of genotypes. Different genotypes also showed differential response in terms of fresh weight of shoot. Maximum fresh weight was observed in H2000-37 (24.87 g/plant) followed by H2001-37 (23.74 g/plant). Minimum fresh weight was recorded in Paras (21.90 g/plant) followed by ICPL-88039 (22.15 g/plant),

when considered irrespective of phosphorus levels. Among the interactions, maximum fresh weight of shoot was observed in H2000-37 with 100% of phosphorus (i.e. 29.34 g/plant) while minimum was observed in ICPI-88039 with 15% of phosphorus (i.e. 18.63 g/plant).

Similarly on 110th day after sowing maximum fresh weight of shoot was recorded in 100% of phosphorus application i.e. 26.18 g/plant followed by 60%, 30%, and 15% of phosphorus application i.e. 20.03, 15.37 and 13.45 g/plant respectively, when considered irrespective of genotype. Among genotypes maximum fresh weight was observed in H2001-4 i.e. 20.59 g/plant followed by H2000-37 i.e. 19.44 g/plant. Minimum fresh weight was recorded in Paras i.e. 17.05 g/plant followed by ICPL-88039 i.e. 17.31 g/plant. Among the interactions maximum fresh weight of shoot was observed in H2000-37 at 100% of phosphorus application i.e. 28.88 g/plant while minimum fresh weight was recorded in ICPL-86039 at 15% of phosphorus application i.e. 12.51 g/plant.

So, on the basis of fresh weight of shoot H2000-37 and H2001-37 are most efficient pigeonpea genotypes at low phosphorus levels (15%) in terms of shoot fresh weight.

Table :2. Effect of different levels of phosphorus application on fresh weight of shoot (g)/plant at different stages in different genotypes of pigeonpea (*Cajanus Cajan* L. Millsp.) at 45, 75 & 110 days after sowing.

45 days						75 days					110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H 2001-37	5.61	5.70	6.17	7.84	6.33	20.83	22.41	24.23	27.49	23.74	14.36	16.71	18.09	27.66	19.21
PARAS	4.35	4.97	4.14	8.95	5.60	19.16	20.85	21.97	25.60	21.90	13.28	14.72	18.13	22.05	17.05
H2000-37	4.34	5.55	7.23	7.72	6.21	22.57	23.92	23.64	29.34	24.87	13.54	15.92	19.40	28.88	19.44
H2001-4	4.38	4.66	6.10	6.15	5.32	20.16	21.49	25.15	26.65	23.36	13.69	15.25	24.97	28.45	20.59
ICPL-88039	4.41	5.60	7.40	8.69	6.53	18.63	20.60	23.11	26.24	22.15	12.51	14.74	18.84	23.13	17.31
H2000-47	4.43	4.98	5.22	6.38	5.25	18.96	22.28	24.70	26.06	23.00	13.33	14.87	20.77	26.90	18.97
Mean	4.59	5.24	6.04	7.62		20.05	21.93	23.80	26.90		13.45	15.37	20.03	26.18	

CD at 5%

Vareity 0.55
Treatment 0.45
Variety x Treatment 1.11

CD at 5%

Vareity 0.82
Treatment 0.62
Variety x Treatment 1.64

CD at 5%

Vareity 0.24
Treatment 0.19
Variety x Treatment 0.48

3. Dry weight of root (g/plant):

Dry weight of root of pigeonpea increased with increasing levels of phosphorus. On 45th day after sowing minimum dry weight was observed with 15% of phosphorus application i.e. 0.62 g/plant. It increased with increasing phosphorus application. Similarly different genotypes also showed differential behaviour in terms of dry weight of root. Maximum dry weight was recorded in Paras i.e. 1.01 g/plant followed by H2001-37 i.e. 0.91 g/plant. While minimum dry weight of root was observed in H2000-37 and H2000-47 i.e. 0.64 g/plant, when considered in terms of genotypes. Among the interactions, maximum dry weight was observed in Paras with 100% of phosphorus application i.e. 1.26 g/plant and minimum dry weight was observed in H2000-37 with 15% of phosphorus i.e. 0.29 g/plant.

On 75th day after sowing, maximum dry weight of root was recorded in 100% of phosphorus application i.e. 4.73 g/plant while minimum dry weight was observed at 15% of phosphorus i.e. 0.62 g/plant, when considered irrespective of genotype.

Different genotypes also showed differential response in terms of dry weight of root. Maximum dry weight was recorded in plants of genotype H2000-47 (4.17 g/plant) while minimum dry weight was observed in H2001-4 (i.e. 2.97 g/plant), when considered irrespective of phosphorus application levels. Among interactions, maximum dry weight of root was recorded in H2000-47 at 100% of phosphorus applications i.e. 6.07 g/plant followed by ICPL-88039 at 100% of phosphorus i.e. 5.96 g/plant. While minimum dry weight

was recorded in H2001-37 at 15% of phosphorus application i.e. 1.92 g/plant.

Similarly on 110th day after sowing, when considered in terms of phosphorus application levels same trend of dry weight of root was observed as that of previous sampling stages i.e. dry weight of root increased from 15% of phosphorus to 100% of phosphorus application. Maximum dry weight was recorded in 100% phosphorus application i.e. 5.44 g/plant while minimum dry weight was at 15% of phosphorus application. Different genotypes of pigeonpea also showed differential behaviour. Maximum dry weight was observed in H2000-47 i.e. 4.65 while minimum dry weight was recorded in H2001-37 i.e. 3.42 g/plant.

Among the interaction between genotypes and phosphorus application levels, maximum dry weight of root was observed in H2000-47 at 100% of phosphorus application i.e. 7.00 g/plant while minimum dry weight was recorded in ICPL-88039 i.e. 2.23 g/plant.

In terms of dry weight of root H2001-37 and H2000-37 are the most efficient genotypes of Pigeonpea as they showed maximum dry weight of root even with 15% of phosphorus application.

Table :3. Effect of different levels of phosphorus application on dry weight of root (g)/plant at different stages in different genotypes of pigeonpea (*Cajanus Cajan* L. Millsp.) at 45, 75 & 110 days after sowing.

45 Days						75 Days					110 Days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H 2001-37	0.74	0.91	0.98	0.99	0.91	1.92	2.94	3.51	4.42	3.20	2.25	2.92	3.73	4.77	3.42
PARAS	0.71	1.03	1.04	1.26	1.01	2.41	2.70	2.80	4.31	3.06	2.42	3.29	3.50	5.16	3.59
H2000-37	0.29	0.57	0.82	0.88	0.64	2.23	2.56	3.67	3.96	3.11	3.24	3.47	3.71	4.67	3.77
H2001-4	0.71	0.72	0.79	1.13	0.84	2.52	2.58	3.11	3.67	2.97	2.63	3.83	4.12	5.00	3.90
ICPL-88039	0.71	0.81	0.90	1.09	0.88	2.28	2.79	3.70	5.96	3.68	2.23	2.68	3.62	6.05	3.65
H2000-47	0.56	0.59	0.60	0.81	0.64	2.21	4.18	4.23	6.07	4.17	2.58	4.23	4.78	7.00	4.65
Mean	0.62	0.77	0.86	1.03		2.26	2.96	3.50	4.73		2.56	3.40	3.91	5.44	

CD at 5%

Variety	0.05
Treatment	0.04
Variety x Treatment	0.10

CD at 5%

Vareity	0.05
Treatment	0.04
Variety x Treatment	0.10

CD at 5%

Variety	0.08
Treatment	0.06
Variety x Treatment	0.16

4. Dry weight of shoot (g/plant):

Dry weight of shoot in different genotypes of pigeonpea at different stages increased with increase in different levels of phosphorus application. At 15%, 30%, 60% and 100% of phosphorus application the dry weight of shoot was 1.37, 1.69, 1.95 and 2.34 g/plant respectively on 45th day after sowing, when considered irrespective of genotypes. Genotypic variation was also found in different pigeonpea genotypes. Maximum dry weight of shoot was recorded in Paras i.e. 2.04 g/plant followed by H2001-37 and ICPL-88039 i.e. 2.00 g/plant. While minimum dry weight was observed in H2000-47 and H2001-37 i.e. 1.55 g/plant and 1.70 g/plant respectively. Among the interactions, maximum dry weight of shoot was recorded in Paras at 100% of phosphorus application i.e. 2.97 g/plant while minimum was found in H2000-37 at 15% of phosphorus application i.e. 1.10 g/plant.

On 75th day after sowing, when considered irrespective of genotypes, dry weight of shoot increased from 5.88 g/plant (at 15% of phosphorus application) to 9.92 g/plant (at 100% of phosphorus application). Among the genotypes, H2000-37 showed maximum dry weight of shoot (i.e. 9.56 g/plant) followed by H2000-47 (i.e. 8.54 g/plant) when considered irrespective of phosphorus application levels. Among the interaction, H2000-47 showed maximum dry weight of shoot at 100% of phosphorus application i.e. 11.17 g/plant while minimum dry weight was recorded in H2001-4 at 15% of phosphorus application i.e. 4.74 g/plant.

Similarly on 110th day after sowing, when considered in terms of phosphorus application increase in dry weight of shoot was observed from 15% of phosphorus application to 100% to phosphorus application. Maximum dry weight was recorded at 100% of phosphorus application (i.e. 5.28 g/plant) while minimum dry weight was observed at 15% of phosphorus application i.e. 8.85 g/plant. Genotypic variations were found between different pigeonpea genotypes. H2000-37 has maximum dry weight of shoot i.e. 7.89 g/plant followed by ICPL-88039 i.e. 7.80 g/plant while H2000-47 showed minimum dry weight of shoot i.e. 6.45 g/plant. Among the interactions between genotypes and phosphorus application level H2000-37 was found to have maximum dry weight of shoot at 100% of phosphorus application (9.53 g/plant) while H2001-4 has minimum dry weight at 15% of phosphorus application (4.55 g/plant).

In terms of dry weight of shoot H2001-37 and H2000-37 are efficient genotypes because both the genotypes have maximum dry weight (i.e. 1.65 and 7.97) of shoot with 15% of P application also.

Table : 4. Effect of different levels of phosphorus application on dry weight of shoot (g)/plant at different stages in different genotypes of pigeonpea (*Cajanus Cajan* L. Millsp.) at 45, 75, 110 days after sowing.

45 days					75 days						110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mea n	15%	30%	60%	100%	Mean
H 2001-37	1.65	1.84	2.05	2.48	2.00	5.17	5.82	8.12	9.76	7.22	5.68	6.34	8.13	8.96	7.28
PARAS	1.31	1.51	2.38	2.97	2.04	5.02	5.58	8.18	8.53	6.83	5.11	6.42	7.64	8.79	6.99
H2000-37	1.10	1.70	1.91	2.11	1.70	7.97	9.53	9.86	10.90	9.56	6.02	7.18	8.83	9.53	7.89
H2001-4	1.41	1.73	1.64	2.13	1.73	4.74	6.66	9.13	9.69	7.55	4.55	5.91	7.65	8.58	6.67
ICPL-88039	1.46	1.89	2.14	2.52	2.00	6.76	7.63	8.92	9.44	8.19	5.61	7.48	8.59	9.51	7.80
H2000-47	1.31	1.47	1.59	1.84	1.55	5.63	7.53	9.83	11.17	8.54	4.73	5.78	7.54	7.75	6.45
Mean	1.37	1.69	1.95	2.34		5.88	7.12	9.01	9.92		5.28	6.52	8.06	8.85	

CD at 5%

Variety	0.13
Treatment	0.11
Variety x Treatment	0.27

CD at 5%

Variety	0.25
Treatment	0.20
Variety x Treatment	0.50

CD at 5%

Variety	0.19
Treatment	0.15
Variety x Treatment	0.38

5. Leaf Area (cm²):

Leaf area in different genotypes of pigeonpea at different stages of sampling increased with increase in different levels of phosphorus. On 45th day after sowing irrespective of genotypes, when considered, maximum leaf area was recorded at 60% of phosphorus application i.e. 104.36 cm² followed by 30% of phosphorus application i.e. 95.40 cm². Minimum leaf area was recorded at 15% of phosphorus application i.e. 87.57 cm².

Various pigeonpea genotypes also showed differential behaviour in terms of leaf area. H2000-37 has showed maximum leaf area i.e. 109.58 cm² followed by Paras i.e. 99.24 cm² on mean basis irrespective of phosphorus application levels. Minimum leaf area was observed in ICPL-88039 i.e. 81.89 cm². On the basis of interactions between genotypes and phosphorus application levels, maximum leaf area was recorded in H2000-37 at 100% of phosphorus application (113.41 cm²) while minimum leaf area was observed in ICPL-88039 at 15% of phosphorus application (77.13cm²).

On 75th day of sampling stage when considered irrespective of genotypes, similar trend was observed as on 45th day i.e. increase from 15% of phosphorus application to 100% of phosphorus application. Leaf area increased from 141.99cm² (at 15% of phosphorus) to 192.92cm² (at 100% of phosphorus

application). Different genotypes of pigeonpea also showed differential response in leaf area. Maximum leaf area was recorded in H2000-37 (214.38cm^2) followed by Paras and H2001-4 i.e.

158.68cm^2 and 158.37 respectively, while H2000-47 was found to have minimum area i.e. 152.57 cm^2 . Among the interactions H2000-37 has maximum leaf area at 100% of phosphorus application i.e. 236.34 cm^2 while minimum leaf area was recorded in H2001-37 at 15% of phosphorus application i.e. 113.48 cm^2 .

On 110th day after sowing, leaf area of the plant decreased in all the treatments as compared to 45th and 75th day of sampling stage. So on 110th day of sampling leaf area was minimum among all sampling stages in all the treatments. Among varieties minimum leaf area was observed in ICPL-88039 (i.e. 88.38 cm^2) whereas maximum leaf area was observed in H2000-37 (i.e. 109.33 cm^2). Among the different phosphorous levels the plants applied with low phosphorous i.e. 15% had minimum leaf area and maximum leaf area was observed in 100% of phosphorous application.

Hence, on the leaf area basis H2000-37 genotype was found to have maximum leaf area with 15% of phosphorus application. It is most efficient genotypes of pigeonpea in terms of leaf area at low phosphorus conditions.

Table :5. Effect of different levels of phosphorus application on leaf area (cm²) at different stages in different genotypes of pigeonpea (*Cajanus Cajan L. Millsp.*) at 45, 75 & 110 days after sowing.

45 days						75 days					110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H 2001-37	84.46	95.87	108.95	101.84	97.78	113.48	140.77	170.86	201.71	156.71	83.59	84.97	99.33	103.47	92.84
PARAS	91.75	97.29	104.86	103.05	99.24	127.01	120.12	181.94	205.52	158.65	92.60	98.69	109.42	108.06	102.19
H2000-37	103.55	105.01	116.35	113.41	109.58	190.74	212.04	217.99	236.34	214.28	104.99	107.96	109.04	115.34	109.33
H2001-4	82.35	94.81	98.25	65.27	85.17	144.63	159.33	160.15	169.34	158.37	85.45	98.10	101.53	106.36	97.86
ICPL-88039	77.13	82.22	85.18	83.04	81.89	139.58	142.71	168.98	169.15	155.11	83.19	85.23	89.26	95.84	88.38
H2000-47	86.19	97.21	112.57	102.62	99.65	136.49	141.11	157.23	175.47	152.57	86.19	99.74	112.57	104.44	100.74
Mean	87.57	95.40	104.36	94.87		141.99	152.68	176.19	192.92		89.34	95.78	103.53	105.59	

CD at 5%

Variety	7.25
Treatment	4.25
Variety x Treatment	18.50

CD at 5%

Variety	5.02
Treatment	4.10
Variety x Treatment	N.S.

CD at 5%

Variety	0.08
Treatment	0.06
Variety x Treatment	0.16

6. Photosynthetic rate (mg CO₂ fixed min⁻¹ cm⁻²)

Photosynthetic rate of different pigeonpea genotypes at different stages increased with increasing levels of phosphorus application. On 45th day after sowing, 15%, 30%, 60% and 100% P application, the photosynthetic rate recorded was 9.98, 10.61, 11.29 and 13.28 mg CO₂ min⁻¹ cm⁻². Hence maximum photosynthetic rate was recorded at 100% of phosphorus application while minimum was observed at 15% of phosphorus application. Genotypic variations were observed in different pigeonpea genotypes. Maximum photosynthetic rate was observed in Paras i.e. 12.82 mg CO₂ min⁻¹ cm⁻² followed by ICPL-88039 i.e. 11.78 mg CO₂ min⁻¹ cm⁻². While minimum photosynthetic rate was found in genotype H2001-37 i.e. 10.22 mg CO₂ min⁻¹ cm⁻². Among the interaction between genotypes and phosphorus level application, Paras was found to have maximum photosynthetic rate at 100% of phosphorus application i.e. 14.07 mg CO₂ min⁻¹ cm⁻² while minimum photosynthetic rate was recorded in H2001-37 at 15% of phosphorus application i.e. 8.77 mg CO₂ min⁻¹ cm⁻².

Similarly, on 75th day after sowing, when considered irrespective of genotypes in terms of photosynthetic rate, same trend of increase in photosynthetic rate was observed from 15% to 100% of phosphorus application. Maximum photosynthetic rate was

recorded in 100% of phosphorus application i.e. 13.96 mg

$\text{CO}_2 \text{ min}^{-1} \text{ cm}^{-2}$ while 15% of phosphorus application has minimum photosynthetic rate. i.e. $10.25 \text{ mg CO}_2 \text{ min}^{-1} \text{ cm}^{-2}$. Among the various genotypes of pigeonpea, Paras showed maximum photosynthetic rate (i.e. $12.70 \text{ mg CO}_2 \text{ min}^{-1} \text{ cm}^{-2}$) followed by ICPL-88039 i.e. $12.38 \text{ mg CO}_2 \text{ min}^{-1} \text{ cm}^{-2}$, when considered irrespective of phosphorus application levels. Minimum photosynthetic rate was observed in H2001-4 genotypes i.e. $11.30 \text{ mg CO}_2 \text{ min}^{-1} \text{ cm}^{-2}$, when considered irrespective of phosphorus application levels. Minimum photosynthetic rate was observed in H2001-4 genotypes i.e. $11.30 \text{ mg CO}_2 \text{ min}^{-1} \text{ cm}^{-2}$. Among the interaction also Paras genotype showed maximum photosynthetic rate at 100% of phosphorus application i.e. $14.90 \text{ mg CO}_2 \text{ min}^{-1} \text{ cm}^{-2}$ while minimum photosynthetic rate was recorded in H2001-4 genotype at 15% of phosphorus application.

On 110th day after sowing, 100% of phosphorus application has maximum photosynthetic rate i.e. $11.63 \text{ mg CO}_2 \text{ min}^{-1} \text{ cm}^{-2}$ whereas 15% of phosphorus application has minimum photosynthetic rate i.e. $9.06 \text{ mg CO}_2 \text{ min}^{-1} \text{ cm}^{-2}$, when considered irrespective of genotypes. Various genotypes of pigeonpea also showed difference response in terms of photosynthetic rate.

ICPL-88039 found to have maximum photosynthetic rate i.e. 10.77

mg CO₂ min⁻¹ cm⁻² followed by H2000-47 i.e. 10.69mg
CO₂ min⁻¹ cm⁻².

Hence, on the basis of photosynthetic rate Paras and ICPL-88039 are the most efficient genotypes because there was not much reduction in photosynthetic rate of these genotypes even at 15% of phosphorus application. Both the genotypes showed maximum photosynthetic rate at low phosphorus levels (i.e. 15%).

Table :6. Effect of different levels of phosphorus application on photosynthetic rate (mg CO₂ fixed min⁻¹ cm⁻²) at different stages in different genotypes of pigeonpea (*Cajanus Cajan* L. Millsp.) at 45, 75 & 110 days after sowing .

45 days						75 days					110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mea n	15%	30%	60%	100%	Mean
H 2001-37	8.77	9.37	10.00	12.73	10.22	10.80	12.20	12.60	13.80	12.35	8.83	9.43	10.27	11.23	9.94
PARAS	12.00	12.07	13.13	14.07	12.82	10.30	11.50	14.10	14.90	12.70	9.47	10.27	10.53	12.23	10.63
H2000-37	9.30	9.34	10.70	13.70	10.76	9.60	10.10	12.07	14.03	11.45	8.37	9.47	10.27	11.63	9.93
H2001-4	9.27	10.83	12.47	12.67	11.31	9.40	10.40	11.77	13.63	11.30	8.67	9.57	10.20	10.77	9.80
ICPL-88039	11.47	11.50	11.20	12.93	11.78	11.70	11.53	12.60	13.70	12.38	9.77	10.17	11.57	11.57	10.77
H2000-47	9.07	10.52	10.27	13.60	10.86	9.70	10.20	12.70	13.67	11.57	9.27	9.93	11.20	12.37	10.69
Mean	9.98	10.61	11.29	13.28		10.25	10.99	12.64	13.96		9.06	9.80	10.67	11.63	

CD at 5%

Variety	1.22
Treatment	1.00
Variety x Treatment	N.S.

CD at 5%

Variety	0.50
Treatment	0.40
Variety x Treatment	0.99

CD at 5%

Variety	0.21
Treatment	0.17
Variety x Treatment	0.42

7. Total Chlorophyll content (mg/g fresh weight):

Total chlorophyll content in the different pigeonpea genotypes varied with different levels of phosphorus application. On 45th day after sowing, with different levels of phosphorus application i.e. 15%, 30% 60% and 100% the chlorophyll content was 1.49, 1.62, 1.42 and 1.34 mg/g fresh weight respectively. The highest chlorophyll content was recorded at 30% of phosphorus application, when considered irrespective of genotypes. Whereas lowest value of total chlorophyll content was observed at 100% (i.e. 1.34 mg/g fresh weight) of phosphorus application followed by 60% of phosphorus application i.e. 1.42 mg/g fresh weight. Various pigeonpea genotypes also showed differential response, when considered irrespective of phosphorus application levels. Maximum total chlorophyll content was recorded in H2001-37 i.e. 1.71 mg/g fresh weight followed by Paras genotype i.e. 1.62 mg/g fresh weight. Whereas ICPL-88039 was found to have minimum total chlorophyll content (i.e. 0.91 mg/g fresh weight). Among interactions, 60% phosphorus application in genotype H2001-37 has highest chlorophyll content i.e. 1.81 mg/g fresh weight while ICPL-88039 genotype has lowest total chlorophyll content in 100% of phosphorus application i.e. 0.47 mg/g fresh weight.

Similarly, on 75th day after sowing, when considered irrespective of phosphorus application same genotypes i.e. Paras and H2001-37 has highest chlorophyll content. Paras has highest value of chlorophyll content i.e. 1.47 mg/g fresh weight. While lowest chlorophyll content was recorded in H2001-4 genotype with

15% of phosphorus application (i.e. 0.71 mg/g fresh weight), when considered irrespective of phosphorus applications. In terms of phosphorus application levels highest chlorophyll content was recorded with 100% phosphorus application i.e. 1.26 mg/g fresh weight, while 15% of phosphorus application has lowest chlorophyll content, i.e. 1.20 mg/g fresh weight. Among the interactions, ICPL-88039 genotype with 100% of phosphorus application has the highest chlorophyll content i.e. 1.47 mg/g fresh weight, while minimum chlorophyll content was recorded in the same genotype with 15% of phosphorus application i.e. 0.62 mg/g fresh weight.

On 110th day after sowing, when considered irrespective of genotypes, chlorophyll content increased with increasing levels of phosphorus applications. At 15% of phosphorus application, the chlorophyll content was 0.34 mg/g fresh weight, while with 100% phosphorus application it increased upto 0.38 mg/g fresh weight. Among the various genotypes of pigeonpea. H2001-37 genotype was found to have maximum chlorophyll content i.e. 0.57 mg/g fresh weight, when considered irrespective of phosphorus applications. Whereas, H2000-37 genotypes has minimum value of chlorophyll content (i.e. 0.23 mg/g fresh weight). Among the interaction between pigeonpea genotypes and phosphorus application levels, highest chlorophyll value was recorded in H2001-37 genotype with 60% of phosphorus application while minimum chlorophyll was found in H2000-37 genotype with 15% of phosphorus application.

So, H2001-37 and H2000-37 genotypes are the most efficient genotypes in terms of chlorophyll content as both genotypes showed maximum value of chlorophyll content even with 15% of phosphorus application.

Table :7. Effect of different levels of phosphorus application on total chlorophyll (mg/g fresh weight) at different stages in different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 45, 75 & 110 days after sowing.

45 days						75 days					110 days				
Level of Phosphorus															
	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H 2001-37	1.57	1.79	1.81	1.69	1.71	1.39	1.44	1.34	1.30	1.37	0.56	0.52	0.62	0.58	0.57
PARAS	1.56	1.69	1.62	1.59	1.62	1.42	1.58	1.48	1.40	1.47	0.54	0.26	0.33	0.30	0.36
H2000-37	1.70	1.73	1.66	1.62	1.68	1.81	1.01	0.87	1.01	1.17	0.18	0.25	0.22	0.27	0.23
H2001-4	1.53	1.44	1.18	1.21	1.34	0.71	1.03	1.21	1.44	1.10	0.25	0.24	0.22	0.45	0.29
ICPL-88039	1.05	1.36	0.76	0.47	0.91	0.62	0.87	1.35	1.47	1.08	0.29	0.33	0.48	0.47	0.39
H2000-47	1.51	1.70	1.48	1.45	1.54	1.23	1.17	1.02	0.96	1.09	0.22	0.27	0.25	0.23	0.24
Mean	1.49	1.62	1.42	1.34		1.20	1.18	1.21	1.26		0.34	0.31	0.35	0.38	

CD at 5%

Variety	0.34
Treatment	0.17
Variety x Treatment	0.50

CD at 5%

Variety	0.24
Treatment	0.20
Variety x Treatment	0.40

CD at 5%

Variety	0.28
Treatment	0.18
Variety x Treatment	0.30

8. Total phosphorus content of root (mg/plant):

Total phosphorus content of root of different pigeonpea genotypes increased with increasing levels of phosphorus application. On 45 days after sowing the maximum phosphorus content of root was found in 100% of phosphorus application i.e. 0.50 mg/plant followed by 60%, 30% and 15% of phosphorus application i.e. 0.33, 0.22 and 0.12 mg/plant respectively, when considered irrespective of genotypes. Among genotypes, H2001-37 genotype was found to have maximum total root phosphorus content i.e. 0.36 mg/plant followed by Paras genotype i.e. 0.33 mg/plant. Whereas minimum phosphorus content of root was recorded in H2000-37 (i.e. 0.023 mg/plant), when considered irrespective of phosphorus application levels. Among the interaction, Paras genotype with 100% of phosphorus application has maximum root phosphorus content i.e. 0.67 mg/plant. While H2000-37 genotype with 15% of phosphorus application was found to have minimum root phosphorus content (i.e. 0.06 mg/plant).

On 75 days after sowing, when considered irrespective of genotypes, the total root phosphorus content increased with increasing level of phosphorus application. The maximum root phosphorus content was recorded at 100% of phosphorus application i.e. 2.36 mg/plant while minimum root phosphorus content was recorded with 15% of phosphorus application i.e. 0.55 mg/plant. When considered irrespective of phosphorus application levels on the mean basis, H2000-47 genotype was found to have

maximum root phosphorus content i.e. 1.76 mg/plant whereas minimum root phosphorus content was recorded in H2001-4 genotype i.e. 1.09 mg/plant. Among the interactions, maximum root phosphorus content was recorded in H2000-47 genotype with 100% of phosphorus application (i.e. 3.34 mg/plant) and minimum root content was found in H2001-37 genotype with 15% of phosphorus applications (i.e. 0.40 mg/plant).

Similarly at 110 days after sowing, the maximum root phosphorus content was recorded in 100% of phosphorus application i.e. 2.38 mg/plant followed by 60%, 30% and 15% of phosphorus applications i.e. 1.37, 0.99 and 0.61 mg/plant respectively, when considered irrespective of genotypes. Various pigeonpea genotypes also showed differential behaviour when considered irrespective of phosphorus application levels. H2000-47 genotype has the maximum root phosphorus content i.e. 1.80 mg/plant followed by ICPL-88039 genotype i.e. 1.30 mg/plant. Whereas minimum root phosphorus content was observed in H2001-4 genotype. Among the interactions, H2000-47 genotype with 100% of phosphorus application was found to have maximum root phosphorus content (i.e. 3.27 mg/plant). While minimum root phosphorus content was recorded in H2001-37 genotype with 15% of phosphorus application (i.e. 0.4 mg/plant).

Hence, on the basis of phosphorus content of root H2000-37 and H2001-37 are the most efficient genotype of pigeonpea which showed highest root phosphorus content at low phosphorus levels.

Table :8 Effect of different levels of phosphorus application on total phosphorus content of root (mg/plant) at different stages in different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 45, 75 & 110 days after sowing.

45 days					75 days						110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mea n	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mea n
H 2001-37	0.19	0.36	0.43	0.46	0.36	0.40	1.03	1.37	2.25	1.26	0.41	1.07	1.28	2.25	1.25
PARAS	0.11	0.22	0.34	0.67	0.33	0.60	0.84	1.12	2.37	1.23	0.61	0.93	1.26	2.32	1.28
H2000-37	0.06	0.17	0.30	0.40	0.23	0.65	0.77	1.29	1.78	1.12	0.75	0.91	1.24	1.91	1.20
H2001-4	0.09	0.13	0.34	0.52	0.27	0.54	0.90	1.20	1.72	1.09	0.55	0.91	1.43	1.84	1.18
ICPL-88039	0.11	0.20	0.27	0.58	0.29	0.57	0.81	1.11	2.68	1.29	0.67	0.82	1.04	2.68	1.30
H2000-47	0.18	0.25	0.27	0.41	0.28	0.55	1.25	1.90	3.34	1.76	0.67	1.29	1.97	3.27	1.80
Mean	0.12	0.22	0.33	0.50		0.55	0.93	1.33	2.36		0.61	0.99	1.37	2.38	

CD at 5%

Variety	0.03
Treatment	0.02
Variety x Treatment	0.05

CD at 5%

Variety	0.04
Treatment	0.03
Variety x Treatment	N.S.

CD at 5%

Variety	0.02
Treatment	0.02
Variety x Treatment	0.04

9. Total phosphorus content of shoot (mg/plant):

Total phosphorus content of shoot in various pigeonpea genotypes increased with increasing levels of phosphorus. On 45 days after sowing, 100% of phosphorus application has the maximum shoot phosphorus content i.e. 1.82 mg/plant followed by 60%, 30% and 15% of phosphorus applications i.e. 1.16, 0.73 and 0.45 mg/plant respectively when considered irrespective of genotypes. Among the genotypes, Paras has the maximum total shoot phosphorus content i.e. 1.19 mg/plant followed by ICPL-88039 i.e. 1.13 mg/plant, when considered irrespective of phosphorus application levels. Minimum shoot phosphorus content was recorded in H2001-4 genotype i.e. 0.92 mg/plant. Among the interactions between genotypes of pigeonpea and phosphorus application levels, Paras genotype with 100% of phosphorus application level was found to have maximum shoot phosphorus content (i.e. 2.50 mg/plant) whereas 15% phosphorus application of the same genotype has minimum shoot phosphorus content i.e. 0.35 mg/plant.

On 75 days after sowing, when considered irrespective of genotypes, the different phosphorus levels showed same trend in shoot phosphorus content i.e. increased with increasing phosphorus applications 100% of P application has maximum shoot phosphorus content i.e. 7.72 mg/plant followed by 60%, 30% and 15% i.e. 5.58, 3.55 and 2.41 mg/plant.

Among the genotypes, H2001-37 (i.e. 5.42 mg/plant) genotype was found to have maximum shoot phosphorus content,

when considered irrespective of phosphorus application levels. Minimum shoot phosphorus content was recorded in H2001-4 genotype (i.e. 4.50mg/plant). Among the interactions, H2001-37 genotype with 100% of phosphorus application levels has maximum shoot phosphorus content (i.e. 9.18mg/plant). Minimum shoot phosphorus content was recorded in H2001-4 genotype with 15% of phosphorus application i.e. 1.94 mg/plant.

At 110 days after sowing, when considered irrespective of genotypes, the total shoot phosphorus content increased with increasing phosphorus levels. Maximum shoot phosphorus content was recorded with 100% phosphorus application i.e. 7.10 mg/plant and minimum shoot phosphorus content was recorded in 15% of phosphorus application i.e. 2.43 mg/plant. Among the genotypes, H2001-37 genotype was found to have maximum shoot phosphorus content i.e. 5.00 mg/plant, when considered irrespective of phosphorus applications. Minimum shoot phosphorus content was observed in H2001-4 genotype i.e. 4.36 mg/plant. Among the interactions, H2001-37 genotypes with 100% of phosphorus application has the highest value of total shoot phosphorus content i.e. 7.92 mg/plant, whereas the minimum value of shoot phosphorus content was recorded in the same genotype with 15% of phosphorus application i.e. 2.38 mg/plant.

So, on the basis of total shoot phosphorus content H2000-37 and H2000-47 genotypes are the most efficient genotypes of pigeonpea as both genotypes showed highest value of total shoot phosphorus content even with lowest phosphorus application i.e. 15% of phosphorus application.

Table :9. Effect of different levels of phosphorus application on total phosphorus content of shoot (mg/plant) at different stages in different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 45, 75 &110 days of after sowing.

45 days						75 days					110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mea n	15%	30%	60%	100%	Mea n
H 2001-37	0.51	0.73	1.02	1.61	0.97	2.38	3.78	6.34	9.18	5.42	2.38	3.81	5.87	7.92	5.00
PARAS	0.35	0.61	1.31	2.50	1.19	2.34	3.63	6.54	7.67	5.05	2.53	3.64	5.99	7.56	4.93
H2000-37	0.45	0.80	1.37	1.73	1.09	2.87	3.52	5.42	6.95	4.69	2.85	3.25	5.31	6.17	4.40
H2001-4	0.35	0.77	0.89	1.68	0.92	1.94	3.33	5.75	6.98	4.50	1.91	3.38	5.38	6.75	4.36
ICPL-88039	0.48	0.84	1.31	1.89	1.13	2.57	3.66	5.44	6.61	4.57	2.78	3.79	4.98	7.04	4.65
H2000-47	0.55	0.64	1.06	1.53	0.94	2.36	3.39	5.41	8.94	5.02	2.15	3.26	5.10	7.15	4.42
Mean	0.45	0.73	1.16	1.82		2.41	3.55	5.82	7.72		2.43	3.52	5.44	7.10	

CD at 5%

Variety	0.03
Treatment	0.03
Variety x Treatment	0.07

CD at 5%

Variety	0.06
Treatment	0.06
Variety x Treatment	N.S.

CD at 5%

Variety	0.03
Treatment	0.03
Variety x Treatment	0.06

10. Nitrogen content of root (mg/plant):

Nitrogen content of different genotype increased with increasing levels of phosphorus applications. On 45 days after sowing N content of root increased from 15% to 100%. At 100% of phosphorus application the value of N content of root was highest i.e. 0.77 mg/plant. It decreased with 15% of phosphorus application and was found to be the lowest i.e. 0.23 mg/plant. Among the genotypes of pigeonpea, H2001-37 genotype was found to have maximum N content of root i.e. 0.72 mg/plant, when considered irrespective of phosphorus application. The minimum N content of root was recorded in H2001-4 genotype i.e. 0.34 mg/plant. Among the interactions between genotypes of pigeonpea and phosphorus applications, Paras genotype with 100% of phosphorus application has the maximum N content of root while minimum N content was recorded in H2000-47 genotype with 15% of phosphorus application i.e. 0.10 mg/plant.

On 75 days after sowing, when considered in terms of phosphorus application the N content of root increased with increasing levels of phosphorus. At 100% to phosphorus application the value of N content recorded was 7.09 mg/plant followed by 60%, 30% and 15% which were 4.37, 2.28 and 1.17 mg/plant respectively. Different genotypes of pigeonpea also respond differently irrespective of phosphorus application levels in terms of nitrogen content of root. ICPL-88039 genotype was found to have maximum N content of root (i.e. 5.05 mg/plant) followed by H2001-37 i.e. 4.75 mg/plant. Whereas minimum N content of root

was recorded in Paras genotype i.e. 2.48 mg/plant. Among the interactions ICPL-88039 genotype was found to have the highest value of nitrogen content of root with 100% of phosphorus application levels i.e. 9.83 mg/plant. While lowest value of N content was recorded in H2000-47 genotype with 15% of phosphorus application i.e. 0.49 mg/plant.

Similarly, on 110 days after sowing, when considered irrespective of genotypes, 100% of phosphorus applications have the maximum N content of root i.e. 5.54 mg/plant followed by 60%, 30% and 15% of phosphorus application which have 2.42, 1.46 and 0.81 mg/plant respectively. Among the genotypes of pigeonpea H2001-37 genotype was found to have maximum N content of root i.e. 2.85 mg/plant, when considered irrespective of phosphorus application levels. Minimum N content of root was observed in H2000-47 genotype i.e. 2.23 mg/plant. Among the interactions, H2001-4 genotype with 100% of phosphorus application has the maximum N content of root while H2000-47 genotype with 15% of phosphorus application (i.e. 0.52 mg/plant) has the minimum value of N content of root.

So, in terms of N content of root H2001-37 genotype of pigeonpea is the most efficient genotype in phosphate uptake efficiency because only H2001-37 genotype showed highest N content of root even with lowest phosphorus application i.e. 15% of phosphorus application.

Table : 10. Effect of different levels of phosphorus application on total nitrogen content of root (mg/plant) at different stages in different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 45, 75 & 110 days after sowing.

45 days						75 days					110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H 2001-37	0.50	0.61	0.82	0.95	0.72	1.74	4.20	5.55	7.51	4.75	1.49	2.08	2.96	4.87	2.85
PARAS	0.23	0.44	0.74	0.98	0.60	1.21	1.79	2.46	4.44	2.48	0.99	1.61	2.77	5.06	2.61
H2000-37	0.12	0.18	0.60	0.55	0.36	1.30	1.87	4.52	5.79	3.37	0.62	1.42	2.19	6.07	2.57
H2001-4	0.16	0.18	0.34	0.70	0.34	0.81	1.03	3.82	5.37	2.76	0.55	1.22	2.14	6.55	2.62
ICPL-88039	0.31	0.38	0.45	0.71	0.46	1.48	3.43	5.44	9.83	5.05	0.69	1.13	2.14	5.93	2.47
H2000-47	0.10	0.17	0.47	0.73	0.36	0.49	1.34	4.44	9.60	3.96	0.52	1.31	2.34	4.76	2.23
Mean	0.23	0.33	0.57	0.77		1.17	2.28	4.37	7.09		0.81	1.46	2.42	5.54	

CD at 5%

Vareity	0.07
Treatment	0.06
Variety x Treatment	0.14

CD at 5%

Vareity	0.12
Treatment	0.10
Variety x Treatment	0.23

CD at 5%

Vareity	0.03
Treatment	0.03
Variety x Treatment	0.07

11. Total N content of shoot (mg/plant):

Total N content of shoot of different pigeon pea genotypes increased with increasing levels of phosphorus applications. When considered irrespective of genotypes, 100% of phosphorus application treatment was found to have highest N content of shoot i.e. 4.45 mg/plant followed by 60%, 30% and 15% of phosphorus applications i.e. 3.13, 2.11 and 1.35 mg/plant. Different genotypes of pigeonpea also showed differential response when considered irrespective of phosphorus application levels. H2001-37 genotype of pigeonpea was found to have maximum N content of shoot i.e. 3.24 mg/plant followed by ICPL-88039 genotype i.e. 3.18 mg/plant. Minimum N content of shoot was recorded in H2000-37 genotype i.e. 2.26 mg/plant followed by H2001-4 genotype i.e. 2.44 mg/plant. Among the interactions, Paras genotype with 100% of phosphorus application has highest N content of shoot (i.e. 5.35 mg/plant) while lowest value was recorded in H2000-37 with 15% of phosphorus application.

On 75 days after sowing, when considered in terms of phosphorus application levels, the N content of shoot increased with increasing levels of phosphorus application. Highest value of N content of shoot was recorded at 100% of phosphorus application i.e. 13.41 mg/plant while lowest value was observed at 15% of phosphorus applications i.e. 3.61 mg/plant. Various pigeonpea genotypes also showed differential behaviour, when considered irrespective of phosphorus application level. H2000-37 genotype has the maximum N content of shoot i.e. 9.30 mg/plant followed by

Paras genotype i.e. 8.03 mg/plant, whereas minimum N content of shoot was recorded in H2001-4 genotype i.e. 6.74 mg/plant. Among the interactions, H2001-4 genotype has the maximum N content of shoot with 100% of phosphorus application i.e. 15.87 mg/plant while the same genotype was found to have the minimum N content value at 15% of phosphorus application i.e. 1.64 mg/plant.

Similarly on 110 days after sowing, 100% of phosphorus application has the maximum value of N content of shoot i.e. 18.02 mg/plant followed by 60%, 30% and 15% of phosphorus treatments i.e. 11.19, 7.11 and 4.26 mg/plant, when considered irrespective of genotypes. Among genotypes of Pigeonpea, H2000-47 genotype was found to have maximum N content of shoot followed by H2001-37 genotype i.e. 12.24 mg/plant when considered irrespective of phosphorus application levels. Minimum N content of shoot was recorded in Paras genotype of pigeonpea i.e. 5.62 mg/plant. Among the interactions, H2000-47 genotype has the maximum N content of shoot with 100% of phosphorus application i.e. 25.15mg/plant while minimum N content was observed in H2001-4 genotype with 15% of phosphorus application level i.e. 2.03 mg/plant.

Hence, in terms of nitrogen content of shoot, H2000-37 and H2001-37 were the most efficient genotypes of pigeonpea as both genotypes found to have highest N content of shoot even with 15% of phosphorus application. On the basis of all the sampling stages, phosphorous use efficiency with low phosphorous application for nitrogen uptake was maximum in H2000-37 genotype (5.24 and

6.20 mg/plant at 75th and 110th day after sowing respectively) was found to be most efficient genotype. However, this was followed by genotype H2001-37 (3.64 and 4.81 mg/plant 45th and 75th day after sowing respectively) and H2000-47 (3.8 and 4.98 mg/plant at 45th and 110th day after sowing respectively) which were almost at par with each other with low level of phosphorous application.

Table : 11. Effect of different levels of phosphorus application on total nitrogen content of shoot (mg/plant) at different stages in different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 45, 75 & 110 days after sowing.

45 days						75 days					110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H 2001-37	1.86	2.70	3.48	4.91	3.24	3.64	4.74	8.05	14.05	7.62	4.81	6.88	11.68	25.58	12.24
PARAS	1.02	1.90	3.87	5.35	3.03	3.73	6.17	9.24	12.98	8.03	3.06	4.09	7.02	8.32	5.62
H2000-37	0.90	1.72	2.95	3.48	2.26	5.24	7.33	11.65	12.99	9.30	6.20	9.81	14.09	18.11	12.05
H2001-4	1.28	2.02	2.42	4.03	2.44	1.64	3.03	6.43	15.87	6.74	2.03	5.36	9.76	16.28	8.36
ICPL-88039	1.58	2.35	3.73	5.07	3.18	3.52	5.93	9.36	12.30	7.78	4.50	7.08	10.81	14.72	9.28
H2000-47	1.47	1.97	2.31	3.90	2.41	3.88	5.80	8.52	12.30	7.62	4.98	9.48	13.79	25.15	13.35
Mean	1.35	2.11	3.13	4.45		3.61	5.50	8.87	13.41		4.26	7.11	11.19	18.02	

CD at 5%

Variety 0.10
Treatment 0.08
Variety x Treatment 0.20

CD at 5%

Variety 0.05
Treatment 0.04
Variety x Treatment 0.09

CD at 5%

Variety 0.04
Treatment 0.03
Variety x Treatment 0.08

12. Fresh weight of Nodules (g/plant):

Fresh weight of root nodules of different genotypes of pigeonpea increased with increasing levels of phosphorus application. When considered irrespective of genotypes, 100% of phosphorus application has maximum fresh weight of nodules i.e. 0.469 g/plant followed by 60%, 30% and 15% of phosphorus application i.e. 0.37, 0.30 and 0.24 g/plant on 45 days after sowing. Different genotypes of pigeonpea also showed differential behaviour when considered irrespective of phosphorus treatment levels. H2000-37 genotype has the maximum fresh weight of nodules i.e. 0.39 g/plant followed by H2000-47 i.e. 0.37 g/plant. Whereas minimum fresh weight of nodules was observed in Paras genotype i.e. 0.29 g/plant followed by H2001-4 and ICPL-88039 i.e. 0.33 g/plant. Among the interactions between genotype and phosphorus application levels, H2000-37 genotype with 100% of phosphorus application was found to have the maximum fresh weight of nodules i.e. 0.57 g/plant while minimum nodule fresh weight was recorded in H2001-4 and ICPL-88039 genotypes with 15% of phosphorus application i.e. 0.22 g/plant.

Similarly on 75 days after sowing, when considered irrespective of genotypes, nodule fresh weight increased with increasing levels of phosphorus application. At 15% of phosphorus application the nodule fresh weight was 0.35 g/plant. It increased upto 1.15 g/plant with 100% of phosphorus application level. Among the different genotypes of pigeonpea, H2001-37 genotypes followed by Paras has the maximum nodule fresh weight i.e. 0.87

g/plant and 0.73 g/plant respectively. Minimum nodule fresh weight was recorded in ICPL 88039 genotype i.e. 0.58 g/plant, when considered irrespective of phosphorus application levels. Among the interactions between genotypes and phosphorus treatments, H2001-37 genotype with 100% of phosphorus application was found to have maximum nodule fresh weight (i.e. 1.54 g/plant) while H2001-4 genotype with 15% of phosphorus application has the minimum fresh weight of nodule i.e. 0.2 g/plant.

On 110 days after sowing, when considered irrespective of genotypes, 100% of phosphorus application treatment has the maximum nodule fresh weight i.e. 0.90 g/plant followed by 60%, 30% & 15% of phosphorus applications i.e. 0.51, 0.35 & 0.28 g/plant respectively. Among the genotypes of pigeonpea Paras genotype followed by H2001-37 genotype has the highest nodule fresh weight i.e. 0.59 and 0.58 g/plant respectively. Minimum nodule fresh weight was recorded in H2001-4 genotype followed by H2000-47 i.e. 0.43 and 0.47g/plant respectively, when considered irrespective of phosphorus application levels. Among the interactions, H2001-37 with 100% of phosphorus application was found to have the maximum nodule fresh weight while H2000-37 genotype 15% of phosphorus application has the minimum value of nodule fresh weight.

Hence in terms of nodule fresh weight H2000-37 and H2001-37 are the most efficient genotypes as both the genotypes showed highest value of fresh weight of nodules even with 15% of phosphorus application.

Table : 12). Effect of different levels of phosphorus application on fresh weight of nodules (g)/plant at different stages in different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 45, 75 & 110 days after sowing.

45 days						75 days					110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H 2001-37	0.25	0.27	0.33	0.50	0.34	0.60	0.41	0.94	1.54	0.87	0.33	0.37	0.61	1.00	0.58
PARAS	0.23	0.28	0.33	0.34	0.29	0.33	0.69	0.83	1.09	0.73	0.29	0.42	0.72	0.94	0.59
H2000-37	0.27	0.30	0.43	0.57	0.39	0.32	0.45	0.84	1.12	0.68	0.24	0.31	0.41	0.99	0.49
H2001-4	0.22	0.33	0.37	0.37	0.33	0.28	0.50	0.80	1.08	0.67	0.25	0.33	0.41	0.75	0.43
ICPL-88039	0.22	0.28	0.34	0.50	0.33	0.30	0.40	0.65	0.96	0.58	0.25	0.33	0.48	0.94	0.50
H2000-47	0.24	0.37	0.43	0.46	0.37	0.29	0.61	0.65	1.13	0.67	0.30	0.33	0.44	0.81	0.47
Mean	0.24	0.30	0.37	0.46		0.35	0.51	0.79	1.15		0.28	0.35	0.51	0.90	

CD at 5%

Variety	0.10
Treatment	0.08
Variety x Treatment	0.21

CD at 5%

Variety	0.10
Treatment	0.08
Variety x Treatment	0.19

CD at 5%

Variety	0.09
Treatment	0.07
Variety x Treatment	0.17

13. Leghaemoglobin content (arbitrary Fluorescence units)

The Leghaemoglobin content of different pigeonpea genotypes increased with increased levels of phosphorus application. On 45 days after sowing, when considered irrespective of genotypes, 100% of phosphorus application showed maximum Leghaemoglobin content i.e. 21.78 followed by 60%, 30% and 15% of phosphorus application i.e. 15.50, 13.83 and 10.89. Among the various genotypes of pigeonpea, H2001-37 genotype has the maximum Leghaemoglobin content (i.e. 21.00) followed by Paras genotype i.e. 15.67, when considered irrespective of phosphorus application treatment. Whereas minimum Leghaemoglobin content was observed in ICPL-88039 i.e. 12.33 followed by H2000-37 i.e. 14.00. Among the interactions, H2001-37 genotype with 100% of phosphorus application was found to have maximum Leghaemoglobin content i.e. 31.00, while minimum Leghaemoglobin content was recorded in H2000-37 genotype with 15% of P application i.e. 7.00.

At 75 days after sowing, when considered irrespective of genotypes, 100% of phosphorus application has highest Leghaemoglobin i.e. 45.17 whereas lowest Leghaemoglobin content was recorded in 15% of phosphorus application i.e. 25.17. Various pigeonpea genotypes also showed differential behaviour when considered irrespective of phosphorus treatments. Paras genotypes was found to have highest Leghaemoglobin content i.e. 38.50 followed by both H2001-37 and H2000-37 genotypes i.e. 38.00. While minimum Leghaemoglobin content was recorded in H2001-4

i.e. 32.25. Among the interactions between genotypes and P application treatments, H2001-37 genotype with 100% of phosphorus application treatments has the highest Leghaemoglobin content while H2000-47 genotypes with 15% of phosphorus application was found to have minimum Leghaemoglobin content.

Similarly, on 110 days after sowing, Leghaemoglobin content increased with increasing levels of phosphorus application when considered irrespective of genotypes. At 100% of P application the value of Leghaemoglobin content was maximum i.e. 25.17. It decreased upto 12.83 with 15% of phosphorus application. Among the genotypes, Paras followed by H2001-37 has the maximum Leghaemoglobin content i.e. 22.50 and 21.25 respectively. Whereas minimum Leghaemoglobin content was recorded in H2000-47 genotype i.e. 14.75. Among interactions, Paras genotype with 100% of phosphorus application has highest Leghaemoglobin content i.e. 30.00 while lowest Leghaemoglobin content was recorded in H2001-4 genotype with 15% of phosphorus application i.e. 9.00.

Hence on the basis of Leghaemoglobin content H2000-37 and H2001-37 are the most efficient pigeonpea genotypes because both genotypes showed highest leghaemoglobin content even with lowest phosphorus application level. The leghaemoglobin content in all the treatments increased from 45th to 75th day after sowing and decreased on 110th day after sowing.

Table : 13. Effect of different levels of phosphorus application on legheamoglobin (arbitrary fluorescence units) Content at different stages in different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 45, 75 & 110 days after sowing.

45 days						75 days					110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H 2001-37	15.00	20.00	21.00	31.00	21.75	25.00	38.00	40.00	49.00	38.00	18.00	20.00	22.00	25.00	21.25
PARAS	12.00	15.00	17.00	18.67	15.67	29.00	35.00	42.00	48.00	38.50	15.00	21.00	24.00	30.00	22.50
H2000-37	7.00	12.00	12.00	25.00	14.00	30.00	35.00	42.00	45.00	38.00	12.00	18.00	20.00	26.00	19.00
H2001-4	12.00	14.00	17.00	17.00	15.00	25.00	30.00	34.00	40.00	32.25	12.00	16.00	21.00	28.00	19.25
ICPL-88039	9.33	10.00	11.00	19.00	12.33	22.00	32.00	43.00	47.00	36.00	11.00	15.00	17.00	20.00	15.75
H2000-47	10.00	12.00	15.00	20.00	14.25	20.00	33.00	39.67	42.00	33.67	9.00	13.00	15.00	22.00	14.75
Mean	10.89	13.83	15.50	21.78		25.17	33.83	40.11	45.17		12.83	17.17	19.83	25.17	

CD at 5%

Variety	2.16
Treatment	1.76
Variety x Treatment	4.31

CD at 5%

Variety	4.25
Treatment	3.47
Variety x Treatment	N.S.

CD at 5%

Variety	3.85
Treatment	3.14
Variety x Treatment	N.S.

14. Acetylene reductase activity (ARA):

ARA of different genotypes of pigeonpea increased with increasing levels of phosphorus application. On 45 days after sowing when considered irrespective of genotypes, at 100% of phosphorus application the value of ARA was 144.91 ($\mu\text{mole C}_2\text{H}_4$ produced $\text{h}^{-1} \text{g}^{-1}$ fresh weight of nodule) followed by 60%, 30% and 15% of phosphorus application with the values 128.55, 108.37 and 96.06 $\mu\text{mole C}_2\text{H}_4$ produced $\text{h}^{-1} \text{g}^{-1}$ fresh weight of nodule respectively. Among the genotypes when considered irrespective of phosphorus application level, Paras genotypes was found to have maximum ARA i.e. 205.92 $\mu\text{mole C}_2\text{H}_4$ produced $\text{h}^{-1} \text{g}^{-1}$ fresh weight nodules. Whereas minimum ARA was recorded in H2000-47 genotype i.e. 71.17 $\mu\text{mole C}_2\text{H}_4$ produced $\text{h}^{-1} \text{g}^{-1}$ fresh weight of nodules. Among the interactions, Paras genotype with 100% of phosphorus application was found to have the maximum ARA i.e. 257.13 $\mu\text{mole C}_2\text{H}_4$ produced $\text{h}^{-1} \text{g}^{-1}$ weight of nodule. While minimum ARA was recorded in H2001-4 genotype with 15% of phosphorus application i.e. 62.33 $\mu\text{mole C}_2\text{H}_4$ produced $\text{h}^{-1}\text{g}^{-1}$ fresh weight of nodules.

On 75 days after sowing, when considered irrespective of genotype, the ARA increase with increasing level of phosphorus. With 100% of phosphorus application the value of ARA was 200.48 $\mu\text{mole C}_2\text{H}_4$ produced $\text{h}^{-1}\text{g}^{-1}$ fresh weight of nodules followed by 60%, 30% and 15% of phosphorus application i.e. 169.28, 147.84 and 123.51 $\mu\text{mole C}_2\text{H}_4$ produced $\text{h}^{-1}\text{g}^{-1}$ fresh weight of nodules.

Various pigeonpea genotypes also showed differential behaviour when considered irrespective of phosphorus application levels. Paras genotype was found to have maximum ARA i.e. 236.70 μ mole C_2H_4 produced $h^{-1}g^{-1}$ fresh weight of nodules. Among the interactions, Paras genotype with 100% of phosphorus application has the maximum ARA i.e. 304.45 μ mole C_2H_4 produced $h^{-1}g^{-1}$ fresh weight of nodules while H2000-47 genotype with 15% of phosphorus application was found to have ARA i.e. 91.10 μ mole C_2H_4 produced $h^{-1}g^{-1}$ fresh weight of nodules.

Similarly, on 110 days after sowing, ARA increased with increasing levels of phosphorus application. When considered irrespective of genotypes, 100% of phosphorus application was found to have maximum ARA i.e. 193.34 μ mole C_2H_4 produced $h^{-1}g^{-1}$ fresh weight of nodules followed by 60%, 30% and 15% of phosphorus application i.e. 164.20, 139.08 and 116.79 μ mole C_2H_4 produced $h^{-1}g^{-1}$ fresh weight of nodules. Among the genotypes, Paras genotype followed by H2001-37 has the highest value of ARA i.e. 231.52 and 165.42 μ mole C_2H_4 produced $h^{-1}g^{-1}$ fresh weight of nodules respectively. When considered irrespective of phosphorus application levels. H2000-47 genotype was found to have minimum ARA i.e. 85.72 μ mole C_2H_4 produced $h^{-1}g^{-1}$ fresh weight of nodules. Among the interaction, Paras genotype with 100% of phosphorus application has the maximum ARA i.e. 303.28 μ mole C_2H_4 produced $h^{-1}g^{-1}$ fresh weight of nodules while H2000-47

genotype with 15% of phosphorus application was found to have minimum ARA i.e. $85.72 \mu \text{ mole C}_2\text{H}_4 \text{ produced h}^{-1}\text{g}^{-1}$ fresh weight of nodule.

Hence in term of ARA, Paras is the only most efficient genotype of pigeonpea which showed highest value of ARA even with lowest phosphorus supply (i.e. 15% of phosphorus application).

Table : 14. Effect of different levels of phosphorus application on acetylene reductase activity ($\mu\text{mole C}_2\text{H}_4$ produced $\text{h}^{-1} \text{g}^{-1}$ fresh weight of nodules) at different stages in different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 45, 75 & 110 days after sowing.

45 days						75 days					110 days				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H 2001-37	86.79	95.71	114.61	125.07	105.54	135.45	158.41	194.12	208.44	174.10	133.82	148.68	177.26	201.92	165.42
PARAS	138.58	197.30	230.66	257.13	205.92	155.07	223.13	264.15	304.45	236.70	158.64	200.15	264.01	303.28	231.52
H2000-37	111.82	124.27	135.67	138.12	127.47	142.12	159.72	161.75	190.07	163.42	133.29	152.75	159.60	177.35	155.75
H2001-4	62.33	79.11	96.06	113.39	87.72	93.18	117.41	117.76	147.83	119.05	73.97	102.00	111.94	136.58	106.12
ICPL-88039	84.33	109.07	137.29	145.45	119.03	124.10	130.52	176.11	221.24	162.99	115.31	132.90	172.08	207.98	157.07
H2000-47	92.53	44.79	57.03	90.31	71.17	91.10	97.86	101.79	130.83	105.40	85.72	98.00	100.28	132.96	104.24
Mean	96.06	108.37	128.55	144.91		123.51	147.84	169.28	200.48		116.79	139.08	164.20	193.34	

CD at 5%

Variety
9.71
Treatment
8.00
Variety x Treatment
17.42

CD at 5%

Variety
10.78
Treatment
12.96
Variety x Treatment
20.56

CD at 5%

Variety
4.91
Treatment
4.01
Variety x Treatment
9.83

15. Yield and attributing characters:

Yield and its attributing characters i.e. pods/plant and 100 seed weight increased with increasing levels of phosphorus in all pigeonpea genotypes. When considered irrespective of genotypes yield/plant at 100% of phosphorus application was found 15.17 g/plant followed by 60%, 30% & 15% of phosphorus application i.e. 13.54 g/plant, 10.76 g/plant and 8.89 g/plant respectively. Among the different pigeonpea genotypes, H2000-37 was found to have maximum yield i.e. 15.07 g/plant followed by H2001-37 genotype i.e. 13.74 g/plant followed by H2001-37 genotype i.e. 13.74 g/plant, when yield was observed in H2000-47 genotype i.e. 9.72 g/plant. Among the interaction between genotypes and phosphorus application levels, H2000-37 genotype with 100% of phosphorus application was found to have maximum yield i.e. 20.25 g/plant while minimum yield was obtained by H2000-47 genotype with 15% of phosphorus application i.e. 7.10 g/plant.

Similarly pods per plant also increased with increasing level of phosphorus applications. When considered irrespective of genotype at 100% of phosphorus application the value of pods/plant was 18.04 pods/plants followed by 60%, 30% and 15% of phosphorus application i.e. 16.88, 16.29 and 15.29 pods/plant. Different genotypes of pigeonpea also show differential behaviour in terms of pods per plant. When H2000-37 genotype was found to have maximum pods/plant i.e. 22.00 pods/plant followed by H2001-4 genotype i.e. 18.38 pods/plant. Minimum pods/plant were obtained by H2000-47 genotype i.e. 12.31 pods/plant. Among interaction,

H2000-37 genotype with 100% of phosphorus application was found to have maximum pods i.e. 23.50 pods/plant while minimum pods were obtained by H2000-47 genotype i.e. 10.75 pods/plant.

100 seed weight also increased with increasing levels of phosphorus application. When considered irrespective of genotypes, 100 seed weight at 100% of phosphorus application was recorded maximum i.e. 6.66 g followed by 60%, 30% & 15% of phosphorus application i.e. 6.58, 6.56 and 6.50 g respectively. Various pigeonpea genotypes also showed differential behavior in term of 100 seed weight. H2001-37 genotype was found to have maximum 100 seed weight i.e. 6.89 g followed by H2000-37 i.e. 6.66g. Minimum 100 seed weight was recorded in ICPL-88039 genotype with 15% of phosphorus application i.e. 6.31 g.

Hence in terms of yield & its attributing character H2000-37 and H2001-37 are most efficient genotypes as both genotypes has highest yield, 100 seed weight and pods/plant even with 15% of phosphorus application.

Table : 15. Effect of different levels of phosphorus application on yield/plant (g) and its attributing characters of different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) after harvesting.

Yield/Plant						Pods/Plant					100 seed weight				
Level of Phosphorus															
Variety	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean	15%	30%	60%	100%	Mean
H 2001-37	11.2 0	13.0 6	15.02	15.69	13.74	15.25	16.00	16.75	18.25	16.56	6.73	6.86	6.87	7.09	6.89
PARAS	8.40	9.33	11.44	12.04	10.30	13.00	14.25	15.00	16.25	14.63	6.54	6.57	6.60	6.63	6.59
H2000-37	9.95	11.2 3	18.85	20.25	15.07	20.50	21.75	22.25	23.50	22.00	6.60	6.64	6.68	6.71	6.66
H2001-4	9.79	13.5 4	13.64	17.67	13.66	17.25	18.00	18.50	19.75	18.38	6.39	6.43	6.47	6.51	6.45
ICPL-88039	6.90	9.08	10.70	13.50	10.05	15.00	15.75	16.00	16.75	15.88	6.31	6.36	6.37	6.40	6.36
H2000-47	7.10	8.30	11.60	11.87	9.72	10.75	12.00	12.75	13.75	12.31	6.43	6.47	6.49	6.62	6.50
	8.89	10.7 6	13.54	15.17		15.29	16.29	16.88	18.04		6.50	6.56	6.58	6.66	

CD at 5%

Variety 0.11
Treatment 0.13
Variety x Treatment 0.22

CD at 5%

Variety 1.02
Treatment 1.26
Variety x Treatment 2.37

CD at 5%

Variety N.S.
Treatment N.S.
Variety x Treatment N.S.

EXPERIMENT-II

1. Root Length (cm):

Primary root length of different genotypes of pigeonpea increased with increasing levels of phosphorus applications. When considered irrespective of genotypes, maximum primary root length was observed with 100% of phosphorus application i.e. 13.8 cm followed by 60%, 30% and 15% i.e. 12.2, 11.9 and 11.1 cm respectively. Among the genotypes, H2000-37 genotype was found to have the maximum primary root length i.e. 13.3 cm followed by ICPL-88039 i.e. 12.5 cm, when considered irrespective of phosphorus application levels. Minimum primary root length was recorded in Paras genotype i.e. 11.5 cm. Among interactions, H2000-37 genotype was found to have maximum primary root length with 100% of phosphorus application i.e. 14.7 cm. Minimum primary root length was recorded in Paras genotype with 15% of phosphorus applications.

2. Root Hairs:

Number of root hairs of different genotype of pigeonpea decreased with increasing levels of phosphorus applications. When considered irrespective of genotypes, maximum no. of root hairs were recorded with 15% of phosphorus application i.e. 109 which decreased upto 53 with 100% of phosphorus application i.e. 86 and 73 respectively. Among genotypes, H2001-37 genotype has maximum number of root hairs i.e. 127 followed by Paras genotype i.e. 68, when considered irrespective of phosphorus application levels. Minimum no. of root hair was recorded in ICPL-88039 (i.e. 62) followed by H2000-37 (i.e. 64). Among interaction, H2001-37 genotype with 15% of phosphorus applications has maximum no. of root hairs i.e. 179. Where as minimum no. of root hairs recorded in ICPL-88039 genotype with 100% of phosphorus applications.

Table: 16. Effect of different levels of phosphorus application on root length (cm) of different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 10 days after sowing.

Level of Phosphorus					
Variety	15%	30%	60%	100%	Mean
H 2001-37	10.5	11.4	11.5	13.3	11.7
PARAS	10.4	11.3	11.6	12.7	11.5
H2000-37	12.1	13.0	13.4	14.7	13.3
ICPL-88039	11.5	11.7	12.4	14.5	12.5
Mean	11.1	11.9	12.2	13.8	

CD at 5%

Variety	0.40
Treatment	0.62
Variety x Treatment	1.00

Table: 17. Effect of different levels of phosphorus application on root hairs of different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 10 days after sowing.

Level of Phosphorus					
Variety	15%	30%	60%	100%	Mean
H 2001-37	179	136	109	82	127
PARAS	85	77	69	43	68
H2000-37	79	69	64	43	64
ICPL-88039	93	62	51	42	62
Mean	109	86	73	53	

CD at 5%

Variety	20
Treatment	10
Variety x Treatment	40

3. Root-shoot ratio:

Root shoot ratio in different pigeonpea genotypes increased with increasing levels of phosphorus. When considered irrespective of genotypes, 60% of phosphorus application has highest root-shoot ratio i.e. 2.26 followed by 30% of phosphorus application i.e. 1.61. 15% and 100% phosphorus applications has the lowest root-shoot ratio i.e. 0.74 and 0.89 respectively. Different pigeonpea genotypes showed differential behaviour. Maximum root-shoot ratio was recorded in Paras genotype i.e. 1.83 followed by H2000-37 genotype i.e. 1.53. Minimum root-shoot was observed in H2001-37 genotype i.e. 1.03. Among interactions Paras genotype with 60% of phosphorus application was found to have highest root-shoot ratio while minimum root-shoot was recorded in H2001-37 and Paras genotype i.e. 0.09 with 100% of phosphorus applications.

4. Acid phosphatase activity (μmol of p-nitrophenol released $\text{min}^{-1}\text{mg}^{-1}$ protein):

Acid phosphatase activity decreased with increase in phosphorus application levels in different pigeonpea genotypes, 15% of phosphorus application treatment has the highest acid phosphatase activity i.e. $0.70 \mu\text{mol}$ of p-nitrophenol rel. $\text{min}^{-1}\text{mg}^{-1}$ of protein followed by 30%, 60% and 100% of phosphorus applications i.e. 0.40, 0.29 and $0.13 \mu\text{mol}$ of p-nitrophenol rel. $\text{min}^{-1} \text{mg}^{-1}$ of protein. Among genotypes, H2001-4 genotype was found to have the highest acid phosphatase activity i.e. $0.65 \mu\text{mol}$ p-nitrophenol rel. $\text{min}^{-1}\text{mg}^{-1}$ of protein followed by H2000-37 i.e. $0.58 \mu\text{mol}$ p-nitrophenol rel. $\text{min}^{-1}\text{mg}^{-1}$ of protein, when considered irrespective of phosphorus application levels. Minimum acid phosphatase activity was recorded in H2000-47 genotype i.e. $0.10 \mu\text{mol}$ p-nitrophenol rel. $\text{min}^{-1}\text{mg}^{-1}$ of protein. Among interaction, H2001-4 genotype with 15% phosphorus application levels has the highest acid phosphatase activity i.e. $1.41 \mu\text{mol}$ p-nitrophenol rel. $\text{min}^{-1}\text{mg}^{-1}$ of protein, whereas minimum acid phosphatase activity was recorded in H2000-47 genotype with

100% of phosphorus application i.e. 0.01 μmol p-nitrophenol $\text{rel. min}^{-1} \text{mg}^{-1}$ of protein.

Table: 18. Effect of different levels of phosphorus application on root/shoot ratio of different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 10 days after sowing.

Level of Phosphorus					
Variety	15%	30%	60%	100%	Mean
H 2001-37	0.91	1.00	2.13	0.09	1.03
PARAS	1.01	3.21	3.00	0.09	1.83
H2000-37	1.01	2.02	2.01	1.08	1.53
ICPL-88039	0.03	0.22	1.91	2.31	1.12
Mean	0.74	1.61	2.26	0.89	

CD at 5%

Variety	0.03
Treatment	
t	0.03
Variety x Treatment	0.06

Table: 19. Effect of different levels of phosphorus application on acid phosphatase activity (μmol of p - nitrophenol released $\text{min}^{-1} \text{mg}^{-1}$ of protein) of different genotypes of pigeonpea (*Cajanus cajan* L. Millsp.) at 10 days after sowing.

Level of Phosphorus					
Variety	15%	30%	60%	100%	Mean
H 2001-37	0.58	0.39	0.33	0.22	0.38
PARAS	0.47	0.35	0.30	0.19	0.33
H2000-37	0.97	0.66	0.48	0.20	0.58
H2001-4	1.41	0.65	0.44	0.11	0.65
ICPL-88039	0.55	0.24	0.11	0.07	0.24
H2000-47	0.20	0.11	0.07	0.01	0.10

Mean	0.70	0.40	0.29	0.13	
CD at 5%					
Variety			0.05		
Treatment			0.04		
Variety x Treatment			0.11		

CHAPTER-V

Discussion

Phosphorus is the second most critical element after nitrogen which is required for the proper growth of the plants. Availability of phosphorus to the plants is very low as it is easily locked up by the presence of Fe, Al ions etc. in soil. Legumes are found to be efficient in P utilization. Pigeonpea is one of the important legume crops which is mainly effective for P utilization. However, this response varies in different genotypes of pigeonpea. In the present experiment, six pigeonpea genotypes were screened in order to find out the genotype which is efficient in P uptake & utilization.

EXPERIMENT –I

First experiment was conducted for screening of six genotypes for phosphorus uptake efficiency which are Paras, ICPL-88039, H2000-37, H2000-47, H2001-37 & H2001-4. Phosphorus was applied @ 15%, 30%, 60% of recommended dose.

Fresh weight of root:

Fresh weight of the root increased with increasing levels of phosphorus in all the genotypes at all levels of sampling i.e. on 45, 75 and 110 days after sowing. This appears obvious because increasing level of P increased the various metabolic activities going on inside the plant and thus more fresh weight was produced. These results are in confirmation with the results of Tsvetkova & Georgiev

(2003) which reported that phosphorus deficiency treatments decreased the whole plant fresh and dry mass in soyabean (*Glycine max* L. Merr.) However genotype H2000-47 and H2001-4 showed maximum fresh weight of root (table-1) even with 15% of P application. This indicates that these genotypes are more efficient because they are capable of producing maximum fresh weight even with low level of P application. Such variations in the genotypes are because of different gene setup in different genotypes. However, H2001-4 genotype could not give the higher efficiency at the initial sampling stage i.e. 45 days after sowing. However, at later stages i.e. 75 & 110 days after sowing, this was found to be most efficient at low level of P. This indicates that perhaps this genotype might have higher efficiency at the later stages i.e. 75 & 110 days after sowing. Similar variations are also very well documented in soyabean (Cassman *et al.* 1981 ; Israel, 1987), clover (Powell, 1977; Robson *et al.*, 1981), common bean (Pereira and Bliss, 1987, 1989), pigeonpea (Itoh, 1987) and cowpea (Cassman *et al.*, 1981).

Fresh weight of Shoot:

Fresh weight of shoot also increased with increasing levels of P application in all genotypes of pigeonpea at all sampling stages i.e. 45, 75 & 110 days after sowing.

This increase in fresh weight of shoot (table-2) is obvious because increasing level of P increased the various metabolic activities inside the plant and thus more fresh weight was produced. These results are correlated with the results of Tamboli *et al.* (2001) which stated that application of KH_2PO_4 @ 200mg kg^{-1} to legumes

increased fresh weight, dry matter accumulation and P uptake over the control. However genotypes H2001-37 and H2000-37 showed maximum fresh weight of shoot even with low P application levels i.e. 15% of P level. This indicates that these genotypes are most efficient because they gave highest fresh weight and are capable of producing maximum fresh weight even with 15% of P application. Such variations in the genotypes are because of different gene make up in different genotypes. However genotype H2000-37 was found to be the most efficient at low P levels at 75 days after sowing. This indicates that perhaps this genotype might have higher efficiency at the reproductive stage only i.e. at 75 days after sowing only. Munir *et al.* (1999) reported that residual phosphorus when applied to wheat @ 0, 25, 50, 75, 100, 150 kg P₂O₅ P ha⁻¹ increased fresh and dry weight of sorghum with increasing quantity of residual phosphorus. Phosphorus use efficiency was maximum with 25 kg and minimum with 50 kg P ha⁻¹ in loam soil. In case of clay loam soil, it was maximum at 50 kg and minimum with 25kg P ha⁻¹. Effect of residual phosphorus was maximum when 150kg P ha⁻¹ was applied to the wheat crop.

Dry weight of root:

Total biomass (dry weight) is the best indicator rather than the fresh weight because fresh weight may be because of higher water content whereas dry weight is mostly because of biomass production. So this gives an indication for higher metabolic activities in the plant. Dry weight of root (table-3) increased with increasing levels of P application in all pigeonpea genotypes at all the sampling

stages i.e. 45, 75 and 110 days after sowing. This is because increased level of P increased various metabolic activities inside the plant and thus more dry weight was produced. These results are in confirmation with the results of Osman *et al.* (2002) which reported that in native Texas legumes shoot dry matter yield and root dry matter weight increased with increasing P application level upto 80kg P ha⁻¹. Genotypes H2001-37, H2000-37 and H2001-4 showed maximum dry weight of root even with low P supply i.e. 15% of P application level. This indicates that these genotypes are more efficient at low P level because they are capable of producing maximum biomass even with low level of P application. Such variation in the genotypes is because of different genetic make up in different genotypes. Variability of traits associated with P efficiency in wild and cultivated genotypes of common bean (*Phaseolus vulgaris* L.) were studied by Araujo *et al.* (1998). However H2001-37 was efficient at the initial stage (45 days after sowing) at low P levels. This indicates that perhaps this genotype has higher efficiency at initial stages only i.e. at 45 days after sowing. Genotype H2000-37 which was having higher fresh weight had not a higher dry weight. This may be because of the reason that this genotype might have increased the uptake of water without much biomass production, which resulted in increased fresh weight and could not affect the dry weight.

Dry weight of Shoot:

Dry weight of shoot also increased with increasing levels of P application in all pigeonpea genotypes at all the sampling stages i.e. 45, 75 and 110 days after sowing. This is because dry matter is

better indicator for higher metabolic activities as explained in previous table that increased P application increased the metabolic processes inside the plant & thus more dry weight was produced. Genotypes H2001-37 and H2000-37 showed maximum dry weight of shoot even with low P supply i.e. 15% of P application (table-4). This indicates that these genotypes are more efficient because they are capable of producing more biomass even with low P application level. However, H2001-37 could not give the higher efficiency during later stages i.e. at 75 & 110 days after sowing while at initial stage i.e. at 45 days after sowing, this genotype is found to have maximum dry weight of shoot even with 15% of P application. This indicates that this genotype might have higher efficiency only at initial stage i.e. at 45 days after sowing. Shoot dry weight at low phosphorus supply has been considered as a screening parameter for phosphorus efficiency in different genotype (Cottman *et al.* 1986; Fageria *et al.* 1998). Genotypic variations in pigeonpea and cowpea genotypes in terms of dry matter production were also confirmed by Vesterager *et al.* (2006).

Leaf Area:

Leaf area indicates the total photosynthetic area of the plants. Higher leaf area might lead to more photosynthates. However this also depends upon the amount of various pigments & rate of photosynthesis of plant. Leaf area of different pigeonpea genotypes increased with increasing levels of P applications in all three sampling stages i.e. 45, 75 & 110 days after sowing (table-5). Total leaf area of H2000-37 was found to be higher at lower levels of P (i.e. 15%) at all sampling stages i.e. 45, 75 and 110 days after sowing. H2000-37 performed well in terms of biomass production

also. Low P supply significantly decreased leaf area and photosynthetic activity of all pigeonpea genotypes as reported by Fujita *et al.* (2004). Phosphorus deficiency primarily reduced leaf area per plant by diminishing the number of leaves through the effects on number of nodes, branching and relative leaf appearances rate and secondarily by reducing leaf expansion as reported by Lynch *et al.* (1991). Yahiya *et al.* (1995) observed that phosphorus application increased the leaf area. Similar finding were reported by Idris *et al.* (1989) in an experiment on chickpea. The minimum leaf area on 110th day may be because of senescence of the leaf at this stage. As a result of which, the senescent leaf abscised & total leaf area of the plant decreased thereby.

Total chlorophyll content:

Total chlorophyll content varied with increasing levels of phosphorus. It was found highest with 30% of P application during initial stage i.e. at 45 days after sowing while at later stages total chlorophyll content increased with increasing levels of P application (table-6). At 45 and 75 days after sowing H2000-37 genotype has maximum chlorophyll content even with low P levels i.e. 15% whereas during later stage i.e. 110 days after sowing H2001-37 genotype was found to have highest chlorophyll content. Chlorophyll content is a measurement of physiological activities in plants as reported by Dudeja and Chaudhary (2005).

Photosynthetic Rate

Photosynthetic rate increased with increasing levels of P application in all pigeonpea genotypes at all sampling stages i.e. 45, 75 and 110 days after sowing (table-7). Chowdhary and Kohri

(2003) also confirmed same in betel-vine (*Piper bettle* L.). As far as photosynthetic rate is concerned, on 45 days after sowing Paras genotype showed maximum photosynthetic rate followed by ICPL-88039. Whereas on later stage i.e. 75 and 110 days after sowing ICPL-88039 proved better with higher rate of photosynthesis. However, genotype ICPL-88039 could not produce higher biomass in term of dry weight. This may be due to reason that the photosynthates produced could not be directly utilized in dry mass production and might have been utilized in catabolic activities. Genotypic variability in photosynthetic rate has been identified within several crop species (Mahon,1990; Babu *et al.*,1985).

Phosphorus content of root:

Phosphorus content of roots and shoots increased with increasing levels of phosphorus application at all sampling stages in all pigeonpea genotypes. Maximum phosphorus was taken up by the roots of H2001-37 genotypes on 45th day whereas on 75 and 110 days after sowing (table-8) maximum phosphorus was taken up by the roots of H2000-37. This indicates that these roots are better adopted for uptake of phosphorus even at low levels in the soil. These results are in confirmation with Israel, 1993 in soyabean (*Glycine max* L.Merr.).

Phosphorus content of shoot:

Total phosphorus content of shoot also increased with increasing phosphorus levels in all pigeonpea genotypes at all three sampling stages i.e. 45, 75 and 110 days after sowing (table-9) H2000-47 showed maximum shoot phosphorus content even with

15% of phosphorus application only at initial stage i.e. 45 days after sowing while during later stages, H2000-37 showed maximum shoot phosphorus content even with low phosphorus application (i.e. 15%). Olivera and Tegera *et al.* (2004) studied effect of phosphorus in common bean (*Phaseolus vulgaris*). Plants inoculated with *Rhizobium tropici* CIAT1899 were grown in Leonard jars under controlled conditions, with P-deficient, (0 and 1mM), P-medium (0.5, 1 and 1.5 mM) and P-high (2mM) condition in a N-free nutrient solution. The P application, increased shoots and root phosphorus content (4 and 6-fold, respectively) in plant harvested at the onset of flowering. Genotypic variations in P utilization of soyabean (*Glycine max* L. Murr.) grown in various sparingly soluble P sources were studied by Tang *et al.* (2007).

Nitrogen content of Root:

The nitrogen content of root was also affected by different level of phosphorus application in different genotypes of pigeonpea. However, at lower level of phosphorus application H2001-37 had the highest content of nitrogen as compared to all other genotypes at all stages of growth (table-10). This indicates that the genotype H2001-37 has a higher efficiency for nitrogen fixation even at low level of phosphorus. These results are in confirmation with Israel, (1993) that decreased N concentration in plant organs was main physiological response to onset of P deficiency.

Nitrogen content of Shoot:

The nitrogen content of shoot was affected by different levels of phosphorus application in different genotypes of pigeonpea. However at lower level of phosphorus application H2001-37 had

highest content of nitrogen as compared to all other genotypes at the initial stage i.e. 45 days after sowing (table-11). Whereas, at later stages H2000-37 genotype was found to have maximum shoot nitrogen content even at low phosphorus level. This indicates that the genotype H2000-37 has a higher efficiency for nitrogen fixation even at low levels of phosphorus as observed in table 14 for acetylene reductase activity. Israel (1993) studied phosphorus

and nitrogen status of plant tissue in soyabean (*Glycine max* L. Merr.). Phosphorus deficiency decreased nitrogen concentration in plant organs and hence decreased plant growth. These results are in confirmation with previously published data (Sa and Israel, 1992), that the total response of symbiotic dinitrogen fixation in soyabean plants to altered phosphorus supply function of both indirect effect on host plant growth and more direct effects on the metabolic function of nodules.

Fresh weight of nodules:

Fresh weight of nodules increased with increasing phosphorus application levels in all pigeonpea genotypes at all sampling stages i.e. 45, 75 and 110 days after sowing (table-12). Genotype H2000-37 showed maximum fresh weight of nodules at initial stage i.e. 45 days after sowing, whereas during later growth stages i.e. 75 and 110 days after sowing H2001-37 genotype showed maximum fresh weight of nodules with lowest phosphorus supply i.e. 15% of phosphorus application. This may be because of more nitrogen uptake by the genotype H2000-37 as shown in table 10. The higher nitrogen content as fixed by *Rhizobium* sp. in nodules lead to more size of nodule and thus higher fresh weight of nodules. Nodules are

a strong sink for P, reaching concentration 3-fold higher than in other organs with little effect from P deficiency (Vadez *et al.*, 1999) although nodule dry weight is greatly reduced by such deficiency (Ribet and Drevon 1995). Genotypic variations for P uptake and dinitrogen fixation in cowpea on low P soils were studied by Jemo *et al.* (2006).

Leghaemoglobin Content:

The higher leghaemoglobin content in the nodules is directly correlated with higher nitrogen fixation activity of nodules as shown in table 10 and 12. However on 75th day highest leghaemoglobin content was observed in H2000-37 genotype which was at par in H2001-37 (table-13). As a result of which it fixed more amount of nitrogen and thus higher dry weight. Similar results were obtained by Gates and Wilson (1974) who studied the effect of P on *rhizobium* growth, nodule weight and function in *stylosanthes humilis* HBK (*Townsville stylo*).

Acetylene Reduction Activity (ARA):

Acetylene reduction activity increased with increasing levels of phosphorus application in all genotypes at all sampling stages. Acetylene reduction activity was highest in genotype Paras at all stages of growth. Paras also recorded high photosynthetic rate at low P conditions (table-14). Thamir (1998) studied that nodule number, nodule mass and niterogenase activity was found to increase as a result of P fertilization. Nodulated plants of *Medicago truncatula* were grown in N-free nutrient solution at increasing levels of P concentration in the solution (0.2-5 μ M P treatment).

When nitrogenase activity was measured at the 1 and 5 μ M P level, the activity doubled at the 5 μ M P treatment supply (Sulieman *et al.* 2008).

Yield/Plant:

Yield/plant at low application P i.e. 15% was maximum in H2001-37. This may be due to the reason that various yield contributing characters i.e. dry weight of root and shoot, phosphorus content, chlorophyll content, nitrogen content of root and shoot was higher in genotype H2001-37. However, number of pods/plant was highest in H2000-37. Higher number of pods/plant could not be correlated with the yield because number of pods alone do not contribute for the yield. It is number of grains/pod and 100 seed weight which also effect the yield (table-15). 100 seed weight was again maximum in H2001-37 at low level of phosphorus i.e. 15%. This confirms that genotype H2001-37 was most efficient in production of total biomass and this yield/plant at low levels of phosphorus. Genotypic variations in pod number of groundnut with P treatments (*Arachis hypogaea* L.) were studied by Wissuwa and Ae (1999).

EXPERIMENT-II

Second Experiment was conducted to identify the characters associated with efficient P uptake. The seeds of pigeonpea were sown in laboratory condition in agar-agar medium and four genotypes were taken which were found to be effective in experiment-I.

Root Length:

At low level of P application (15%) root length as observed on 10th day after sowing was maximum in H2000-37 (table-16). This shows that this genotype was most effective for the maximum root length at low levels of P application. Root growth is a key trait for phosphorus efficiency. (Lynch, 1995 and Manashe *et al.* 2000).

Root Hairs:

Root soil contact is an important factor for uptake of a less mobile nutrient such as phosphorus. Root hairs are important structures for the absorption of water & mineral by the root. Maximum number of root hairs was observed in H2001-37 (table-17). This indicates that this genotype might prove successful in the absorption of nutrients & water and thus may contribute toward the higher production. Schmidt, (2001) and Fohse *et al.*, (1991) found that in *Phaseolus vulgaris* var. nanus. cv.Loma the large contribution of root hairs in P uptake was due to their increased surface area, small radius and perpendicular growth into the soil from the root axis. In low P available soil the plant species having more root hairs were able to satisfy a higher proportion of their P demand required for maximum growth (Gahoonia and Neilsen, 2004). Genetic variations in genotypes of cowpea (*Vigna unguiculata*) in terms of root and root hairs under P uptake

efficiency were studied by Krasilnikoff *et al.* (2003). Decrease in number of root hairs with increase phosphorus application in all the genotypes at all sampling stages might be due to the reason that at higher phosphorus level a plants require less root hairs to absorb it. So the number of root hairs decreased with higher phosphorus application.

Root and Shoot Ratio:

Root/shoot ratio indicates, a proportionate utilization of various minerals during the metabolism. Genotype Paras & H2000-37 had higher root/shoot ratio at ten days after germination

(table-18). Johnson *et al.* (1996) resulted that plants typically respond to P deficiency by allocating more carbon to roots, thus increasing their root to shoot ratio Increased in root-shoot ratio is a common response to phosphorus deficiency (Lynch *et al.*, 1991 and Moller & Pettersin, 1999).

Acid Phosphatase Activity:

Acid phosphatase activity also increased with increasing P levels (table-19). Genotype H2000-37 had highest acid phosphatase activity at low P (15%). The genotype H2000-37 had highest phosphorus even in the shoot. Perhaps this might be the reason for highest acid phosphatase activity in this genotype. Increased acid phosphatase activity under phosphorus deficiency has been reported by Vance *et al.* (2003) and Tomscha *et al.* (2004). Genetic variation for acid phosphatase activity in roots were more related to severity of P deficiency in the tissue of a given genotype rather than superior P adaptation (CIAT, 1986; McLachlan, 1980; Yan, 1992).

CHAPTER-VI

Summary and Conclusion

The present investigation entitled, "Evaluation of pigeon pea (*Cajanus cajan* L. millsp.) genotypes for phosphorus uptake efficiency " was carried out in the Department of Plant Physiology, CCS Haryana Agricultural University, Hisar with the objective to evaluate pigeonpea genotype for phosphorus uptake efficiency and nodulation and nitrogen fixation efficiency under different phosphorus regimes.

Experiment-I:

All parameters studied showed increasing trend with increasing phosphorus level in the media from 15% to 100% in all genotypes tested.

- Maximum fresh weight of root was observed in ICPL-88039 genotype followed by H2001-4 whereas minimum fresh weight observed in H2001-37 followed by H2000-47 genotype.
- Fresh weight of shoot was maximum in H2000-37 genotype followed by H2001-37 genotype whereas minimum fresh weight was observed in Paras genotype.
- Dry weight of root was maximum in H2000-47 and minimum in H2001-37 genotype.
- Dry weight of shoot was maximum in H2000-37 genotype and minimum in Paras genotype. However, fresh and dry weight

showed variation in different genotypes at different sampling stages.

- Photosynthetic rate, total chlorophyll content and leaf area was maximum in Paras followed by H2000-37. However, at low level of phosphorus, both Paras and H2000-37 performed equally well.
- Phosphorus content at low phosphorus application was better in case of H2000-37 genotype both in root and shoot. H2001-37 genotype had got highest content of nitrogen with lower phosphorus application.
- Nodule weight, acetylene reduction activity, leghaemoglobin content was higher in genotypes Paras and H20001-37. At low P levels H2001-37 and H2000-37 performed well, which helped these genotypes to fix more nitrogen.

Experiment-II:

- At low P levels, root length was maximum in genotype H2000-37, while root hairs were highest in H2001-37 genotype.
- Acid and alkaline phosphatase activity was highest in genotype H2001-4 at low phosphorus application.

On the basis of above two experiments, it seems that with lower level of P application H2000-37 and H2001-37 showed the better performance in terms of various parameters and yield contributing characters. These two genotypes are followed by Paras and ICPL-88039. So it can be concluded on the basis of above study, out of six genotypes studied four genotypes of pigeonpea

namely Paras, ICPL-88039, H2000-37 and H2001-37 can performed well even with low P application.

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ABSTRACT

Title of Dissertation	Evaluation of Pigeonpea (<i>Cajanus cajan</i> L. Millsp.) genotypes for phosphorus uptake efficiency
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Admission No.	2004BS163M
Title of Degree	Master of Science in Plant Physiology
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Year of Award of Degree	2008
Major Subject	Plant Physiology
Total No. of Pages in the Thesis	103(Chapters)+ xvi = 119
No. of words in the abstracts	Approx. 169
Key words-	Pigeonpea, phosphorus efficiency, fresh and dry weight of root and shoot, leaf area, photosynthetic rate, total chlorophyll content, total phosphorus content, total nitrogen content, fresh weight of nodules, leghaemoglobin content, acetylene reductase activity

An experiment was conducted with six pigeonpea genotypes namely Paras, ICPL-88039, H2000-37, H2000-47, H2001-4, and H2001-37, at different levels of phosphorus i.e. 15%, 30%, 60% and 100% in pot culture house. Various parameters like fresh and dry weight of root and shoot, leaf area, photosynthetic rate, total chlorophyll content, total phosphorus content of root and shoot, total nitrogen content of root and shoot, fresh weight of nodules, leghaemoglobin content, acetylene reduction activity, yield and its attributing characters were analysed at three sampling stages i.e. 45, 75 & 110 days after sowing. Another experiment was conducted in laboratory with

four pigeonpea genotypes i.e. H2001-37, Paras, H2000-37 and ICPL-88039 with same treatments of phosphorus. However, the parameters studied were root length, number of root hairs, root-shoot ratio and alkaline phosphatase activity only 10 days after sowing

- Maximum fresh wt. of the root was observed in ICPL-88039 and minimum fresh weight was observed in H2001-37, whereas maximum fresh weight of shoot was observed in H2000-37 genotype and minimum was in Paras genotype.
- Dry weight of root was maximum in H2001-27 whereas dry weight of shoot was maximum in H2000-37. Minimum root was observed in H2001-37 genotype whereas minimum dry weight of shoot was observed in Paras genotype.
- Photosynthetic rate, chlorophyll content, leaf area was maximum in Paras genotype (Check variety). However, total phosphorus and nitrogen content at low phosphorus application was better in H2000-37 genotype.
- Nodule weight, acetylene reductase activity, leghaemoglobin content was high in Paras & H2001-37 genotype. However at lower P application H2001-37 performed better.

In second experiment, at low phosphorus application maximum root length was observed in H2000-37 where number of root hairs were highest in H2001-37 genotype. Phosphatase activity was highest in H2001-37 genotype at low phosphorus application.

Signature of the Student

Major Advisor

Head of the Department

