POSSIBLE BENEFITS OF RHIZOBIAL INOCULATION AND PHOSPHORUS SUPPLEMENTATION ON NUTRITION, GROWTH AND ECONOMIC SUSTAINABILITY IN GRAIN LEGUMES

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Abstract

It is documented that majority of soils in Africa have low levels of nitrogen and phosphorous and hence the capability to support plant growth such as leguminous crops is limited. The supply of these mineral nutrients is vital in enhancing legume growth and development. The potential role(s) of nitrogen from rhizobial inoculants and phosphorous in legumes growth with respect to growth, yield, economic benefits, photosynthesis and plant nutrition is given special attention in this review.

Key words: Biological Nitrogen Fixation, Chlorophyll, Nutrients, Soybean, Yields.

{**Citation:** Eutropia V. Tairo, Patrick A. Ndakidemi. Possible benefits of rhizobial inoculation and phosphorus supplementation on nutrition, growth and economic sustainability in grain legumes. American Journal of Research Communication, 2013, 1(12): 532-556} www.usa-journals.com, ISSN: 2325-4076.

Introduction

Soybean (*Glycine max* L.) is a legume native to East Asia perhaps in North and Central China (Laswai *et al*., 2005) and it is grown for its edible bean, oil and protein around the world. They are eaten in fresh green state and dry beans. Soybean is found in Family *Fabacea* and Species *Glycine max* (Shurtleff *et al*., 2007). The plant can also be used as forage for animals and soil fertility improvements if used as soil cover crop. Soybean can be grown as a sole crop, intercropped, or mixed with important cereals such as maize, sorghum and millets (Mmbaga *et al*., 2003). Soybean has been recognized as one of the premier agricultural crops today, thus it is the best source of protein and oil and has now been recognized as a potential supplementary source of nutritious food (Wilcox and Shibles, 2001). It has been found to substitute other sources of good quality protein such as milk, meat and fish, therefore has become very suitable to some areas where other protein sources are scarce or too expensive to afford (Anwar *et al*., 2010). Soybean contains a good quality protein of around 42% and 19.5% oil (Wilcox and Shibles, 2001). Different protein of soybean is considered complete, because it supplies sufficient amounts of the types of amino acids that are required by the body for building and repair of tissues (Jinze, 2010). Essential amino acids found in soybean are methionine, isoleucine, lysine, cystine, phenylalanine, tyrosine, threonine, tryphophan as well as valine (Laswai *et al*., 2005). Amino acids are used in the formation of protoplasm, the site for cell division and therefore facilitate plant growth and development. In soybean seed, there is a protein, which contains amino acids required for human nutrition and livestock (Zarei *et al*., 2012). Soybean has been found to have different uses; for example in food industry, soybean is used for flour, oil, cookies, candy, milk, vegetable cheese, leathin and many other products (Coskan and Dogan, 2011).

The symbiotic relationship between the soybean root and rhizobial root colonies and subsequent symbiotic nitrogen fixation is one of the most important physiological processes, which occurs in the growth, and development of the soybean plant. A research done by Bambara and Ndakidemi, (2009) concluded that *Rhizobium* inoculation in legumes stimulated growth and is an alternative source to the expensive commercial nitrogen fertilizers. In biological processes, nitrogen combines with C, H, O, and S to create amino acids, which are the building blocks of proteins (Uchida, 2000). Nitrogen is highly needed for all enzymatic reactions in a plant, also is a major part of the chlorophyll molecules and plays a necessary role in photosynthesis and is a major component of several vitamins (Uchida, 2000). In legumes and other leafy vegetables, nitrogen improves the quality and quantity of dry matter and protein (Uchida, 2000). For soybean and indeed other nitrogen fixing legumes, nitrogen requirements in the field are met by either soil mineral nitrogen acquisition or symbiotic N_2 fixation. To achieve the maximum yield of soybean at lower cost, it is necessary to use N_2 fixation by root nodules (Harper, 1974). Report by Burias and Planchon, (1990) indicate that high nodulation and high N_2- fixation rates increase soybean yields. Besides, seed protein content increased when specific *Bradyrhizobium* species was used to inoculate soybean (Egamberdiyeva *et al*., 2004). Yet, the ability of soybean to fix atmospheric nitrogen is not always adequate for yield maximization due to different factors like temperature, soil physical and chemical characteristics such as pH of the soil, low phosphorus, presence of inefficient native rhizobia in the soil that compatible to the legume planted (Wesley *et al*., 1998) as well as other biotic agents like insects, weeds and diseases (Serraj and Adu-Gymfl, 2004). Inoculation of soybean with specific *Bradyrhizobium* strains improves the plant dry matter, nitrogen concentration, nitrogen accumulation, and grain yield (Javaid *et al*., 2010).Nitrogen (N) is the most limiting nutrient for crop yields, and nitrogen fertilizers is an expensive input in agriculture costing more than US$45 billion per year globally (Gyaneshwar *et al*., 2002).

After nitrogen, phosphorus (P) is another plant growth-limiting nutrient despite being abundant in soils in both inorganic and organic forms. However, many soils throughout the world are phosphorus -deficient because the free phosphorus concentration (the form available to plants) even in fertile soils is generally not sufficient (Gyaneshwar *et al*., 2002). Root improvement, stalk and stem vigor, flower and seed formation, crop production, crop maturity and resistance to plant pests and diseases are the attributes associated with phosphorus availability. Phosphorus is needed in relatively large amounts by legumes for growth and has been reported to promote leaf area, biomass, yield, nodule number and nodule mass in different legumes (Berg and Lynd, 1985: Pacovsky *et al*., 1986: Kasturikrishna and Ahlawat, 1999). Furthermore, phosphorus has important effects on photosynthesis, root development, fruiting and improvement of crop quality (Sara *et al*., 2013). Large amount of phosphorus applied as fertilizer enters in to the immobile pools through precipitation reaction with highly reactive Aluminium (Al^+) and Iron ($Fe3^+$) in acidic, and Calcium (Ca^{2+}) in calcareous or normal soils (Gyaneshwar *et al*., 2002: Hao *et al*., 2002). Efficiency of P fertilizer throughout the world is around 10 - 25 % (Isherword, 1998), and concentration of bioavailable phosphorus in soil is very low reaching the level of 1.0 mg kg^{-I} soil (Goldstein, 1994). Microbial community influences' soil fertility through soil processes such as decomposition, mineralization, storage and release of nutrients. Microorganisms enhance the phosphorus availability to plants by mineralizing organic phosphorus in soil and by solubilizing precipitated phosphates (Chen *et al*., 2006: Kang *et al*., 2002: Pradhan and Sukla, 2005).

In soybean production, phosphorus and inoculation with the appropriate *Rhizobium* strains have quite prominent effects on nodulation, growth and yield parameters (Shahid *et al*., 2009: Kumaga and Ofori, 2004).The factors which control the amount of nitrogen fixed include available soil nitrogen, genetic determinants of compatibility in both symbiotic partners and lack of other yield-limiting factors like edaphic factors associated with phosphorus deficiency, soil acidity, mineral elements nitrogen and other various microelements like Cu, Mo, Co, B which are necessary for N_2 fixation (Harold *et al*., 1992).The absence of the required rhizobia species and optimal phosphorus levels limit legume production in different parts of the world. Inoculation with compatible and suitable rhizobia with optimum phosphorus levels may be essential where a low population of native rhizobial strains prevail and is one of the key components of which grain legume farmers can use to optimize yields.

Effects of *Bradyrhizobium japonicum* inoculation and Phosphorus supplementation on Biological Nitrogen fixation (BNF) in legumes

Biological nitrogen fixation (BNF) is the process in which nitrogen gas (N_2) from the atmosphere is incorporated into the tissue of legume plants, with the help of soil microorganisms. It is the symbiosis relationship that results in the formation of a specialized structure called nodule. The bacterial micro symbiont fixes nitrogen for macrosymbiont which is the legume plant in return for reduced carbon (C) from the plant host to the microsymbiont which is the rhizobia bacteria (Gyaneshwar *et al*., 2002). For the symbiosis to occur, it is associated with complex mechanisms starting with the mutual exchange of diffusible signal molecules (Ndakidemi and Dakora, 2003). Furthermore, seeds and root molecules mainly flavonoids are secreted by legume plants of which can be sensed by some specific species of rhizobia (Makoi and Ndakidemi, 2010). In return, a lipochito-oligosaccharides known as Nod factors are secreted by rhizobia, which stand as recognizable receptors such as kinase of the legumes. The nodule then appears on the roots and facilitates the nitrogen-fixing bacteria (Santos *et al*., 2013).

There is huge reservoir of atmospheric nitrogen (78%) in the atmosphere and unfortunately, it is not available for organisms like plants and animals. For it to be utilized it must be broken down to reactive compounds that can be easily metabolized e.g. NO_3^- or NH_4^+. Besides, nitrogen atoms must be bonded chemically with oxygen and hydrogen through the N_2 fixation process and carbon through nitrogen assimilation process (Vitousek *et al*., 2002). The most common forms of fixed N_2 are NH_4^+ and NO_3^-. Rees *et al*.(2005) reported that 3×10^{14} g of nitrogen is fixed annually. Stability of N_2 molecules with triple bond between the nitrogen atoms requires large amount of energy to break and thus make it difficult for animals and plants to access for their growth. The amounts of energy required to break the triple, double and the single bonds of N_2 molecule are 225, 100, and 39 Kcal mol^{-1}, respectively (Howard and Rees, 2005). In biogeochemical nitrogen cycle, the N_2 fixation is the process of converting atmospheric N_2 into NH_4^+ (Fisher and Newton, 2002). Reduction of N_2 into NH_4^+ requires high amount of activation energy. To produce NH_3 by Haber-Bosch reaction, temperature of 300-500^{o}C and the pressure over 300 atmospheres in the presence of Fe based catalysts is needed.

Leguminous plants are able to fix atmospheric N_2 through the association with rhizobia. The legume plant supplies the carbohydrate for bacterial growth while the bacteria fix atmospheric N_2 into NH_4^+, to be converted into amino acids that can be used by the plant to synthesize proteins for its growth and development (Russell, 2008). Symbiotic association is a highly specified relationship between the host plant and the bacteria. *Rhizobium*-legume symbiosis involves the interaction between the plant and the bacteria leading to initiation and development of the root nodules. Soil bacteria like *Rhizobium* live in nodules as nitrogen fixing bacteroids. A single rhizobial cell that infects a root hair can increase the progeny by 10^{10} within few weeks. The efficiency of symbiotic BNF is markedly dependent on the mutual compatibility of both partners, and is influenced by a number of environmental factors which are macrosymbiont which comprises of variety, nodulin, photosynthate availability and tolerance of stress; microsymbiont which comprises of ineffectiveness, effectiveness, competitive ability and saprophytic competence and environmental factors which comprises of combined nitrogen, light, temperature, water, aeration, salinity and biotic agent (Vincent, 1980).

Nitrogen is a primary (macro) nutrients which plays a most important roles in legumes for the formation of amino acids which is the building blocks of protein. It is also important for cell division and vital for plant growth (Uchida, 2000). Nitrogen is a chlorophyll component it promote vegetative growth and green colouration of foliage. It is also directly involved in photosynthesis and is a necessary component of vitamins and aids in production and use of carbohydrates and influence energy reactions in plants as well (Sara *et al*., 2013). Nitrogen is the most commonly deficient nutrient among macro and micronutrients, besides considerable amount of nitrogen is removed from soil when protein-rich grain or hay is harvested (Uchida, 2000). However, nitrogen is the key component of healthy growing; all plants other than legumes should be fertilized by nitrogenous fertilizer.

Legume plants particularly soybean are unique for their ability to fix nitrogen from atmosphere by symbiotic relationship with *Rhizobium* bacteria (Coskan and Dogan, 2011). Rhizobia require a plant host; therefore, they cannot independently fix nitrogen. These bacteria are located around root hair and fixes atmospheric nitrogen using particular enzyme called nitrogenase. When this mutualistic symbiosis established, rhizobia use plant resources for their own reproduction whereas fixed atmospheric nitrogen is used to meet nitrogen requirement of both itself and the host plants. Supply of nitrogen through biological nitrogen fixation has ecological and economic benefits (Ndakidemi *et al*., 2006).

$$N2 + 8H2 + 16ATP \xrightarrow{\text{Nitrogenase enzyme}} 2NH3 + 2H2 + 16ADP + 16Pi$$

The ability of soybean to fix N_2 and its ability to produce nodules together with good percentage of protein, has brought about its important and uniqueness (Wilcox and Shibles, 2001). During formation and emergence of root hairs, N_2 fixation is affected by many factors such as the presence and density of nodulating bacteria in the root zone, the physical and chemical properties such as humidity, temperature, salt concentration in the soil, pH levels and deficiencies of several mineral nutrients (Abdul-Jabbar and Saud, 2012). It is a well-established fact that, when legumes are grown in soils high in available nitrogen, the nitrogen fixation rate is reduced (Solomon *et al*., 2012).

Phosphorus is among 17 essential nutrients for plant growth. Its functions cannot be performed by any other nutrient, and an adequate supply of phosphorus is required for optimum growth and reproduction (Uchida, 2000). Phosphorus is classified as a major nutrient, meaning that it is required by crops in relatively large amounts. Despite the considerable amount of total phosphorus in tropical soils, phosphorus deficiency is one of the most important fertility problems in tropical agriculture (Haru and Ethiopia, 2012). The importance of phosphorus in biological nitrogen fixation is well known, as it is an energy driven process (Haru and Ethiopia, 2012). Phosphorus is involved in several key plant functions, including energy transfer, photosynthesis, transformation of sugars and starches, nutrient movement within the plant and transfer of genetic characteristics from one generation to the next (Uchida, 2000). Generally, phosphorus is vital to plant growth and is found in every living plant cell. *Rhizobium* bacteria use phosphorus as an essential ingredient in converting atmospheric N_2 to ammonium (NH_4), a form useable by plants (Dakora and Keya, 1997). *Rhizobium* is able to synthesize the enzyme nitrogenase, which catalyzes the conversion of N_2 to two molecules of ammonia (NH_3) (Tsvetkova *et al*., 2003). Nodule formed in leguminous plants shows a typical healthy and effectiveness by showing a pink or red colour and is brought about by formation of protein called leghemoglobin. This protein contains both iron (Fe) and molybdenum (Mo) which is responsible for binding oxygen. Besides, it creates a low oxygen environment within the nodule, which allows *Rhizobium* bacteria to live and fix N_2 easily, and in a comfortable environment (Lindermann *et al*., 2003; Chowdhury *et al*., 1998).

Phosphorus influences nodule development through its basic functions in plants as an energy source. Phosphorus plays a vital function in increasing plant tip and root growth, decreasing the time needed for developing nodules to become active and of benefit to the host legume. Furthermore, P increases the number and size of nodules and the amount of nitrogen assimilated per unit weight of nodules, increasing the percent and total amount of nitrogen in the harvested portion of the host legume and improving the density of Rhizobia bacteria in the soil surrounding the root (Bashir *et al*., 2011). Phosphorus brings about the ability of catalyzing stress in the symbiotic relation between root bacteria and legume plants (Tsvetkova *et al*., 2003). Inadequate P restricts root growth, the process of photosynthesis, translocation of sugars, and other such functions, which directly or indirectly influence nitrogen fixation by legume plants (Olivera *et al*., 2004).

Most tropical soils are deficient in appropriate effective strains capable of fixing nitrogen and available phosphorus. There is a need to find appropriate rhizobia strain which will enhances nitrogen fixation attributes and yield of soybean under different phosphorus levels.

Bradyrhizobium japonicum Inoculation and Phosphorus (P) Supplementation on Growth and Chlorophyll Accumulation in Soybean (_Glycine max_ L.)

Nitrogen is the most limiting nutrients in agricultural production, despite its abundance in gaseous form in earth's atmosphere (Sanginga *et al*., 1997). Planting leguminous plants like beans, nuts and others in between growing season and season to season cropping systems may benefits the plants and soil by yielding nitrogen (Solomon *et al*., 2012). Most nitrogen is naturally present in the soil as organic content (Dashora, 2012). However, N deficiency in plants are due to erosion, run off and leaching of nitrate in non-producing soil horizon (Zarei *et al*., 2011).Some of the most common symptoms of nitrogen deficiency in plants include the yellowing, dropping of leaves and poor growth, furthermore delaying flowering and fruiting may also be present.

Nitrogen is a crucial element for both leguminous and non-leguminous crops, has constructive impacts on growth, and yields in legumes (Wood *et al*., 1993). It is a major constituent of chlorophyll, the most essential pigment needed for photosynthesis and amino acids, the building blocks of proteins. It is also found in other bio molecules such as ATP and nucleic acids (Wagner, 2012). Nitrogen is a factor in many biological compounds that plays a major role in photosynthetic activity. Besides, is part of the enzymes associated with chlorophyll synthesis, which reflect relative crop nitrogen status and yield level in plants (Hokmalipour *et al*., 2011). Its deficiency impairs growth and it constitutes one of the major yield limiting factors for crop production decline. Nitrogen is highly needed for all enzymatic reactions in a plant, also is a major part of the chlorophyll molecules and plays a necessary role in photosynthesis and is a major component of several vitamins. Furthermore, in legumes and other leafy vegetables, N improves the quality and quantity of dry matter and protein (Uchida, 2000). Green colour in the leaf is vanished due to nitrogen deficiency and this may cause the decrease in leaf area and intensity of photosynthesis as well (Chu *et al*., 2005). Nitrogen supply has large effect on leaf growth because it increases the leaf area of plants and, on that way, it influences on photosynthesis functional (Bojovi ć *et al*., 2009).

Root hairs, root tips and the outermost layers of root cells are the most pathways of phosphorus entering the plants (Better crops, 1999: Rotaru, 2010). Once phosphorus is inside the plant roots, phosphorus may be stored in the root or transported to the upper part of the plants (Singh and Sale, 2000). During various chemical reactions, it is integrated into organic compounds, including nucleic acids (DNA and RNA), phosphoproteins, phospholipids; sugar phosphate compounds like adenosine triphosphate (ATP) (Bashir *et al*., 2011). When ADP and ATP transfer the high-energy phosphate to other molecules, by phosphorylation, the stage is set for many essential plant processes to occur. The ATP is then available as an energy source for many other reactions that occur within the plant, and the sugars are used as building blocks to produce other cell structural and storage components. Phosphorus is an essential part of the process of carrying the genetic code from one generation to the next, giving the blueprint for all characteristic of plant growth and development (Longstreth, 1980).When P is limiting, the most prominent effects are a reduction in leaf expansion, leaf surface area and the number of leaves (Bekere *et al*., 2012). Shoot growth is more susceptible than root growth, which leads to a decline in the shoot-root dry weight ratio (Better crops, 1999). However, root growth is also reduced by P deficiency, leading to fewer roots mass to attain water and nutrients (Uchida, 2000). Commonly, inadequate phosphorus slows the processes of carbohydrate utilization, development of a dark green leaf color or plants leaves develop a purple color (Samavat *et al*., 2012). Since phosphorus is readily mobilized in the plant, when a deficiency occurs the phosphorus is translocated from older tissues to active meristematic tissues, resulting in foliar deficiency symptoms emerging on the lower part of the plant (Weisany *et al*., 2013). Other property of P deficiency on plant growth consists of delayed maturity, reduced quality seeds, fruit and decreased disease resistance. Symbiotic legumes have a high requirement for phosphorus (Israel, 1987). This huge required amount are essential in stimulating root and shoot growth in plants and influences the efficiency of the *Rhizobium*-legume symbiosis through facilitation of energy transfer reactions which involve ATP in nitrogenase activity (Leidi *et al*., 2000). It is believed that phosphorus is effectively translocated into grain at high rates, since phosphorus is necessary for the production of protein, phospholipids and phytin in soybean grain. Phosphorus (P) is among the important elements needed for crop growth and production in many tropical soils. However, many tropical soils are phosphorus -deficient (Buerkert *et al*., 2001; Nekesa *et al*., 2007). Phosphorus deficiency can limit nodulation by legumes soybean, not only that but also the soybean cannot grow, produce or tolerate stresses but, P- supplementation can overcome the deficiency (Kamara *et al*., 2010). A soil, which is depleted with nitrogen as limiting nutrient, has found to have enhanced symbiotic N_2 fixation (Ndakidemi *et al*., 2006). Plants need phosphorus for growth throughout their life cycle, especially during the early stages of growth and development for proper and well-built roots. The primary role of phosphorus compounds in plants is to store and transfer energy produced by photosynthesis to be used for growth and reproduction (Leidi *et al*., 2000). Legumes such as soybean need phosphorus for adequate growth and nitrogen fixation. Sufficient phosphorus levels are also required to enhance different plant organs growth and promote nodulation and early maturity (Kamara *et al*., 2010). Studies done by Ndakidemi *et al*. (2006) and Shahid *et al*. (2009) provide a proof that increased phosphorus application enhances plant growth significantly. Supplementing legumes with nutrients, especially phosphorus has great potential for increasing yields, as it not only promotes plant growth but also enhances symbiotic establishment for increased N_2 fixation (Gangasuresh *et al*., 2010). In soybeans, the demand for phosphorus is greatest during pod and seed development where more than 60% of phosphorus ends up in the pods and seeds (Kumar and Chandra, 2008) and (Shahid, Sleem *et al*, 2009). Phosphorus is a crucial elements in crop production which plays important role for many characteristics of plant growth such as sugar and starch utilization, photosynthesis use, cell division and organization, nodule formation, root development, flower initiation and seed and fruit development (Gangasuresh *et al*., 2010). Phosphorus being required in large quantities in young cells, particularly shoots and root tips of soybean, where metabolism is high and cell division is rapid, highest concentration of P is required in seeds of the mature soybean plants (Sanginga *et al*., 1997).

Nitrogen and Phosphorus are the major component of the leaf chlorophyll, which influences the leguminous plant to manufacture its own food through photosynthesis process, which ultimately increases yields and uptake of important nutrients in different soybean plant tissues (Imsande, 1989). Phosphorus plays a very important function in almost every plant process that involves energy transfer. High-energy phosphate, detained as a part of the chemical structures of adenosine diphosphate (ADP) and adenosine triphosphate (ATP), is the source of energy that drives the huge number of chemical reactions within the plant. The most important chemical reaction in nature is photosynthesis. It utilizes light energy in the presence of chlorophyll to combine carbon dioxide and water into simple sugars, with the energy being captured in ATP (Montanaro *et al*., 2007). Studies shows that at the early vegetative growth stage of soybean plant, a leaf chlorophyll content increased following inoculation while at the late pod filling stage shows just a significant effect on the leaf chlorophyll content following inoculation in comparison with un inoculated soybean plant (Katundala, 2011). Usually, legume inoculation increases the leaf chlorophyll content and plant biomass, besides, the leaf chlorophyll content of nodulated leguminous plants remained at high levels until the pod filling stage, and diminish at the flowering stage (Koutroubas *et al*., 1998). Important macronutrients such as N and P and other micronutrients may affect the metabolic reactions in photosynthesis (Marschner, 1995). Insufficient levels of this mineral nutrient in the growth of plants may lower the chlorophyll accumulation, which limits photosynthesis due to their involvement in carbohydrate synthesis (Lambers *et al*., 2006). Studies by Sara *et al*. (2013) showed that the content of chlorophyll found in leaves is a good sign of how the leguminous plant has fixing nitrogen. Some macro- and micronutrients are important for the normal growth processes of plants. The presence of nitrogen and phosphorus, the essential macronutrients which plays a very important role in plants, might significantly affect the chlorophyll formation in plants once plentifully available in the growth media.

Low level of phosphorus and nitrogen may impair growth of most legumes. Furthermore, inadequate compatible rhizobial strain to a particular legume plants may result into poor plant growth including less chlorophyll formation and photosynthesis. There is a need of supplying legumes with appropriate rhizobial inoculants with phosphorus to bring about optimum productivity in legumes.

Nutrients uptake in legumes as affected by *Rhizobium* inoculation and Phosphorus supplements

An ever-increasing world population requires the need to produce more food in a land, which is steadily shrinking, and loose its properties in each year. This situation creates significant pressure on suitable land already in production and yet requires continuous expansion of food producing ecosystems into less fertile areas (Rengel, 2008). Continuous cultivation without field fallowing shows a severe deficiency of most of the major nutrients especially nitrogen (Abbasi *et al*., 2008). The demand for nitrogen in a deficient soil is normally achieved by the use of chemical fertilizers. However, the high cost of mineral nitrogen fertilizers and their un-availability at the time of requirement are the two major constraints responsible for low fertilizer nitrogen inputs. This emphasizes the importance of developing an alternative means to meet the demand of nutrients in plants through the use of beneficial bacteria in the ecosystem that are sustainable agronomically, environmentally friendly and affordable. Plants require essential elements of which carbon, hydrogen, and oxygen are derived from the atmosphere and soil water (Uchida, 2000). Nitrogen is the most essential mineral elements that are required by plants in great quantities and the availability of this element in soils influences growth and crop yields (Shiri-Janagard *et al*., 2012). Other essential elements are phosphorus, potassium, calcium, magnesium, sulfur, iron, zinc, manganese, copper, boron, molybdenum, and chlorine, which are supplied either from soil minerals and soil organic matter or by organic or inorganic fertilizers (Roy *et al*., 2006).

Efficient capture of nutrients from soil by roots is a critical issue for plants given that in many environments, nutrients have poor availability and may be deficient for optimal growth (Buerkert *et al*., 2001). Whilst nutrient supply in soil is often improved by the application of fertilizers, a wide range of physico-chemical parameters, environmental and seasonal factors and biological interactions (Salvagiotti *et al*., 2008) governs the availability of nutrients. The rate of root growth and the plasticity of root architecture, through either root growth or extension of root hairs, are clearly important for effective exploration of soil and interception of nutrients (Richardson *et al*., 2009). However, the importance of different root traits is dependent on the nutrient in question and other factors that include plant species and soil type. For example, for nutrients present at low concentrations in soil solution and/or with poor diffusivity (e.g. P as either HPO_4^{2-} or $H_2PO_4^{-}$, and micronutrients, such as Fe and Zn), root growth and proliferation into new regions of soil and release of root exudates are of particular importance (Richardson, *et al*., 2009). In contrast, nutrients present in either higher concentrations (e.g. K^+, NH_4^+), or with greater diffusion coefficients (e.g. NO_3^-, SO_4^- and Ca^{2+}), are able to move more freely toward the root through mass flow, where root distribution and architectural characteristics that facilitate water uptake are of greater relative significance (Richardson *et al*., 2009).

Total nutrient uptake by leguminous plants depends on yield obtained, which may vary with season, variety, soil, and cultural practices (Rogers, 1997). Grain legumes take up relatively small amounts of nutrients early in the season, but as they grow and develop, the daily rate of nutrient uptake increases (Weisany *et al*., 2013). Soybeans need an adequate supply of nutrients at each developmental stage for optimum growth. High-yielding soybeans remove substantial nutrients from the soil, and this should be taken into account in an overall nutrient management plan (Rogers, 1997). The use of rhizobial inoculants and phosphorus supplement on legumes may play a great practical importance in cropping system in developing countries, which can increase soil fertility and plant productivity (Abbasi *et al*., 2008), thereby increasing the farm income of the farmers. Among legumes, soybean (*Glycine max* L. Merril) is important N_2-fixing crop, cultivated throughout the world. Soybean obtains nitrogen directly from the soil and indirectly from symbiotic fixation when nodulated with effective strains of *Bradyrhizobium japonicum*. In soils not previously cropped with soybean or soils in nontraditional areas of soybean production seldom, contain sufficient population of native *Bradyrhizobium japonicum* to ensure satisfactory nodulation (Mohammadi *et al*., 2012). It has been reported that soybean could reduce the fertilizer nitrogen requirement of a following crop if substantial amount of nutrient (nitrogen) remain in the soil (Abbasi *et al*., 2008).

In food-producing system, crops like legumes must be provided with sufficient nutrients for energetic growth and high outputs, positioning an emphasis on understanding interactions of the soil-plant microbe governing nutrient attainment by plants (Ndakidemi *et al*., 2011).Total nutrient uptake by leguminous plants depends on yield obtained, which will vary with season, variety, soil, and cultural practices (Rogers, 1997). It has been reported that *Rhizobium* inoculation significantly increases the uptake of Phosphorus (P), Potassium (K), Calcium (Ca), Magnesium (Mg) in different plant organs (Makoi *et al*., 2013).

Phosphorus (P) is one of the major plant growth-limiting nutrients although it is abundant in soils in both inorganic and organic forms. Phosphorus is vital for plant cell division, growth, root lengthening, seed and fruit development, and early ripening as well. It is a part of several compounds including oils and amino acids in plants (Uchida, 2000). Phosphorus -supplementation can enhance plant growth by increasing the efficiency of biological nitrogen fixation, enhancing the availability of other trace elements such as Fe, Zn etc. Phosphorus also plays a major role in energy storage and transfer as ADP and ATP (adenosine di- and triphosphate) and DPN and TPN (di- and triphosphopyridine nucleotide). Phosphorus is part of the RNA and DNA structures, which are the major components of genetic information. Phosphorus also aids the plants in root development and it increases seed yields (Uchida, 2000), besides it promote nodulation as well. It has been reported that phosphorus application influences the content of others nutrients in leaves and seeds e.g. Zn, Mn, Fe, B, Mo etc (Singh *et al*., 2011).

Microorganisms such as *Bradyrhizobium* inoculants may significantly have an effect on the chemistry of nutrients in soils by enhancing nutrients uptake by plants. To-date, most *Bradyrhizobium* inoculants have been developed and are primarily used for supplying N_2 to plants. Little is known about their effect on supplying nutrients in legumes. However, Ndakidemi *et al*. (2011) and Makoi *et al*., 2012 reported *Bradyrhizobium* inoculation enhancing the uptake of P, K, Ca, Mg, S, Mn, Fe, Cu, Zn, B and Mo in leguminous plants. Studies on the influence of P and inoculation for the successive uptake of macronutrients and micronutrients in different plant organs are not enough documented. There is a need of establishing how beneficial bacteria and P application can affect the uptake of nutrients in leguminous crops.

Yields and yields component of soybean (*Glycine max* L.) as affected by *Bradyrhizobium japonicum* inoculation and phosphorus supplementation

In agricultural systems, a biotic stresses like nitrogen and phosphorus deficiency results into significant contribution in reduction of crop productivity and yields. Phosphorus (P) and Nitrogen are major limiting soil nutrients in most tropical soils of which its low level could limit growth, dinitrogen fixation and yields of legumes (Olufajo, 1990). Among various factors that can contribute to soybean success, phosphorus and rhizobial inoculation had quite prominent effects on nodulation, growth and yield parameters (Shahid *et al*., 2009).

Seed yields in legumes are highly attributed to nutrients availability (Hussain *et al*., 2011). In legume, nitrogen is more useful because it is the main component of amino acid as well as protein. Legumes can obtain nitrogen through atmospheric fixation in their root nodules in symbiosis with soil rhizobia and as a result have a potential to fit in nitrogen deficit soils. To reduce the production cost with mineral fertilizers and provide protection to the environment, more legume production could be achieved through seed inoculation with beneficial *Rhizobium* bacteria (Hussain *et al*., 2011), which are known to influence nodulation, symbiotic nitrogen fixation, growth and yield of legumes.

Phosphorus is the second most vital plant nutrient but for legumes, it presumes primary significance, which plays important role in root proliferation and thereby atmospheric nitrogen fixation. Singh *et al*. (2008) reported that the yield and nutritional quality of legumes is greatly influenced by application of phosphorus and biofertilizers. Phosphorus is crucial in the production of protein, phospholipids and phytin in legume grains (Rahman *et al*., 2008). Its application also plays a vital role in increasing legume yield through its effect on the plant itself and also on the fixation process by *Rhizobium*. For example, it is widely reported that phosphorus stress may lead to reduced growth, and yield in field crops including legumes such as soybean. P stress reduces nitrogen fixation due to decreased nodule formation and reduced nodule sizes and finally affecting the yield and grain quality and quantity (Sadeghipour and Abbasi, 2012).

Ability of legumes crops to fix atmospheric N_2 through symbiosis with soil bacteria has position the crops belonging to the leguminousae family as important and valuable worldwide. Legumes are able to assimilate atmospheric N_2, convert it into useful nutritional products, and contribute both agronomically and economically in many cropping systems in agriculture (Belkheir *et al*., 2001). The plant crop yield is a dependent variable, relies upon all other growth and yield contributing character (Achakzai and Bangulzai, 2006). The maximum yields of a legume crop depends upon its yield components, such as the number of branches per plant, pods per plants, seeds per pod and seed weight, however density of the plant is an important agent that affect yield and yield components of legumes (Dahmardeh *et al*., 2010).

Shahid *et al*. (2009) reported that seed production in soybean can increase by 70-75% when the proper bacterial strains were used to inoculate soybean seeds. The higher nodulation due to inoculation resulted in higher nitrogen fixation by *Rhizobium* and eventually the number of pods per plant which bring about higher grain yields as a whole (Onduru *et al*., 2008; Singh *et al*., 2011). In other studies, Ibrahim *et al*. (2011) reported increased yield and yield component of soybean by inoculating the seeds with specific strain of rhizobia. Studies done in some parts of African countries shows that soybeans which is not inoculated, need 24-39 kg P ha^{-1} so that maximum yields can be attained. Furthermore, legumes particularly soybean which uses nitrogen through dinitrogen fixation require more P than those using mineral nitrogen (Olufajo, 1990). However, scientists found that, legume seed inoculation with proper *Rhizobium* strain together with minor amounts of phosphorus at early growth stage could stimulate the root nodulation and increase biological nitrogen fixation eventually higher yields to at most 78% (Morad *et al*., 2013).

Biofertilizers seem to be an attractive and cost effective source of nitrogen for legume cultivation and perhaps it requires little technical expertise. Biological nitrogen fixation and grain yields of legumes are normally increased when inoculated with effective and efficient strain of *Rhizobium* (Okereke *et al*., 2001). It has also been reported that nodule number, dry weight and soybean shoot yield increased when seeds inoculated with *Rhizobium* (Egamberdiyeva *et al*., 2004). Benefits of rhizobia inoculation is not well pronounced especially in developing countries, perhaps technology transfer lagged behind in regard to that knowledge in most African farmers (Okereke *et al*., 2001).

With the worldwide emphasis on sustainable agricultural systems, increase in grain legume production such as soybean will come mostly from supplementing the crops with phosphorus (in deficient environments) and through the use of rhizobial inoculants rather than the use of inorganic nitrogen fertilizer. This is due to the ability of the soybean to fix large quantity of atmospheric nitrogen and make it available for plant growth and increased yields.

Economic benefits of *Rhizobium* inoculants and phosphorus-supplementation in legumes

Production of grain-legumes is increasing significantly due to their vast use in different situations including human food, animal feed as well as industrial demands. Considering the increasing needs for human consumption of plant products and the economic constraints of applying fertilizer in legumes, there is a greater role for grain legumes in cropping systems, especially in regions where affordability of fertilizer is difficult (Ndakidemi *et al*., 2006).

Grain legumes such as soybean, cowpea and common bean have many potential uses and are grown in different agro-ecological zones (Yagoub *et al*., 2012). They are economically important crops used in a wide range of products (Tahir *et al*., 2009). They play a significant role in sustainability of agricultural systems. Biological Nitrogen fixation is becoming more attractive and economically viable nitrogen inputs, substitute of inorganic fertilizers for resource poor farmers, and is an environmentally friendly agricultural inputs (Bekere *et al*., 2012). For economically viable and environmentally sensible farming practices, nitrogen inputs should be managed successful through symbiotic nitrogen fixation (Sharma *et al*., 2011).

Most tropical soils experience low nitrogen, which is the major constraints in crop production (Yakubu *et al*., 2010). Small-scale agriculture which is practiced in most sub-Saharan Africa, cover majority of the people, of which, chemical fertilizers are unaffordable because of increasing prices in each year (Yakubu *et al*., 2010). Through different cropping systems like intercropping of cereals and legumes and crop rotation has found to be an alternative source and means of improving fertility of the soil and boost productivity and income of the farmers (Ndakidemi *et al*., 2006). Several studies have shown that through biological Nitrogen fixation, which is enhanced by inoculation to the compatible host legume leave residual nitrogen in the soil which add organic matter and also become source of cheap nutrients for the next cropping season to cereal crops and other legumes as well (Zahran, 1999). Biological Nitrogen Fixation is therefore considered to have ecological and economic benefits (Ndakidemi *et al*., 2006).

The nutrient supply in crop production is one of the key components to higher yields (Gehl, *et al*., 2005). Increased crop yields due to mineral nutrient supplementation in the developed world are widely documented (Giller *et al*., 1998). However, Africa is reported to have the lowest use of fertilizer in the world. The per capita consumption of fertilizer in Tanzania is standing at 8 kg ha $^{-1}$ as compared with 52 kg ha $^{-1}$ for South Africa and Zimbabwe and 27 kg ha $^{-1}$ for Malawi (Walter, 2007). Nitrogen (N) is the most limiting nutrient for crop yields, and nitrogen fertilizers is an expensive input in agriculture costing more than US$45 billion per year globally (Gyaneshwar *et al*., 2002). Biological nitrogen fixation can reduced the need for N fertilizers, resulting in an economy estimated in US dollar 3 billion per crop season (Nicolas *et al*., 2006). Ndakidemi *et al*.(2006) reported that the combined application of bacterial inoculants and P fertilizer to field legume plants of soybean and common bean significantly increased biomass production and grain yield compared with the single use of nitrogen and P or rhizobial strains alone. From the economic analysis, the increase in grain yield with inoculation translated into a significantly higher marginal rate of return and dollar profit for soybean and common bean farmers in Tanzania (Ndakidemi *et al*., 2006). In view of increasing price of fertilizers, it seems the cost of nutrients will be increasing in most cropping systems. Evidently, legumes will remain the component of the farming system in remote areas comprised of poor farmers due to their capacity to fix nitrogen. Research efforts should be directed in assessing the optimum combinations between organic and inorganic fertilizers that will offer immediate economic returns to the resource poor farmers who cannot afford the full package of inorganic fertilizers.

Conclusion

Rhizobial inoculation and phosphorus supplements are effective in improving growth, yield, economic benefits, and photosynthesis and plant nutrition in legumes. It is strongly recommended to adopt these technologies in the cultivation of legumes in Africa.

Acknowledgements

This study was funded by Tanzania Commission for Science and Technology (COSTECH) and The Nelson Mandela African Institution of Science and Technology (NM-AIST).

References

Abbasi M, Majeed KA, Sadiq A, Khan SR (2008). Application of *Bradyrhizobium japonicum* and phosphorus fertilization improved growth, yield and nodulation of soybean in the sub-humid hilly region of Azad Jammu and Kashmir, Pakistan. Plant Production Science 11(3): 368-376.

Abdul-Jabbar, BKA and Saud, HM (2012). Effects of Phosphorus on biological nitrogen fixation in soybean under irrigation using saline water. Global Journal of Science Frontier Research Agriculture & Biology. 12: (1): 65-72

Achakzai AKK, Bangulzai MI (2006). Effect of various levels of nitrogen fertilizer on the yield and yield attributes of pea (*Pisum sativum* L.) cultivars. Pakistan. Journal of Botany, 38(2): 331 – 340.

Anwar AHMN, Podder AK, Hasem MA, Bala P, Islam MA (2010). Effect of *Bradyrhizobium* inoculants on the growth and yield of soybean varieties PB-1 and G-2. Journal of Soil Nature 4(1), 39-48.

Ayoola OT (2010). Yield performance of crops and soil chemical changes under fertilizer treatments in a mixed cropping system. African Journal of Biotechnology. 9(26): 4018-4021.

Bashir K, Ali S, Umair A (2011). Effect of different phosphorus levels on xylem sap components and their correlation with growth variables of mash bean. Sarhad Journal of Agriculture, Vol. 27, No. 4.

Bekere W, Hailemariam A (2012). Influences of Inoculation Methods and Phosphorus levels on Nitrogen Fixation Attributes and Yield of Soybean (*Glycine max* L.). At Haru, Western Ethiopia. American Journal of Plant Nutrition and Fertilization Technology 2(2): 45-55.

Belkheir AM, Zhou X, Smith DL (2001). Variability in yield and yield component responses to genistein pre-incubated *Bradyrhizobium japonicum* by soybean [*Glycine max* (L.)] cultivars. Plant and Soil 229: 41 – 46.

Berg RK and Lynd JQ (1985). Soil fertility effects on growth, yield, nodulation and nitrogenase activity of Australian winter pea. Journal of Plant Nutrition. 8:131-145.

Bojovi ć B, Markovi ć A (2009).Correlation between nitrogen and chlorophyll content in wheat (*Triticum aestivum* L.).Kragujevac Journal of Science 31 (2009) 69-74.

Buerkert A, Bationo A, Piepho HP (2001). Efficient phosphorus application strategies for increased crop production in sub-Saharan West Africa. Field Crops Research 72(1): 1-15.

Burias N, Planchon C (1990). Increasing soybean productivity through selection for nitrogen fixation. *Agronomy Journal*. 92(6): 1031-1034.

Chowdhury MU, Ullah MH, Afzal MA, Khanam D, Nabi SM (1998). Growth, nodulation and yield of cowpea as affected by *Rhizobium* inoculation and micronutrients in the hilly region. Bangladesh Journal of Agricultural Research, 23(2): 195-203.

Coskan A, Dogan K (2011). Symbiotic Nitrogen Fixation in Soybean. Soybean Physiology and Biochemistry, Edited by Hany A. El-Shemy, ISBN 978-953-307-534-1.

Dahmardeh M, Ramroodi M, Valizadeh J (2010). Effect of plant density and cultivars on growth, yield and yield components of faba bean (*Vicia faba* L.). African Journal of Biotechnology. Vol. 9 (50), pp. 8643 – 8647.

Dakora FD, Keya SO (1997).Contribution of legume nitrogen fixation to sustainable agriculture in sub-Saharan African. Soil Biology and Biochemistry, 29, 809–817. doi: 10.1016/S0038-0717(96)00225-8.

Dashora K (2011). Nitrogen Yielding Plants: The Pioneers of Agriculture with a Multipurpose. American-Eurasian Journal of Agronomy, 4 (2): 34-37.

Egamberdiyeva D, Qarshieva D, Davranov K (2004). Growth and yield of soybean varieties inoculated with *Bradyrhizobium spp* in N-deficient calcareous soils. Institute of Microbiology, Uzbek Academy of Sciences, A. Kadiriy str. 7B, Tashkent, 700128, Uzbekistan.

Fisher K, Newton WE (2000).Nitrogen fixation general overview. In Leigh, G. J. (Ed.), Nitrogen fixation at the millennium. Elsevier, Amsterdam. Pp 1-34.

Gachimbi L, Onduru D, Muchena F, Jager De A (2004). Impact of *Rhizobium* and Phosphate Fertilizers in enhancing legume production in Mbeere District in Kenya using farmers' field school approach.

Gangasuresh P, Muthuselvi V, Muthulakshmi E, Muthumari S, Maniammal G (2010). Synergistic Efficiency of Phosphate solubilizer associated with Nitrogen fixer on the Growth of Soybean (*Glycine max*). International Journal of Biological Technology (2010) 1(2): 124-130.

Gehl RJ, Schmitt JP, Maddux LD, BW Gordon (2005). Corn yield response to Nitrogen rate and Timing in sandy Irrigated soils. Site visited on November 14, 2009.

Giller KE, Amijee F, Brodrick SJ, Edje OT (1998).Environmental constraints to nodulation and nitrogen fixation of *Phaseolus vulgaris* L. in Tanzania II. Response to N and P fertilisers and inoculation with *Rhizobium.* African Crop Science Journal, l6, 171–178.

Gyaneshwar P, Kumar GN, Parekh LJ, Poole PS (2002).Role of soil microorganisms in improving P nutrition of plants. Plant and Soil. 245: 83-93.

Hao X, Cho CM, Racz GJ and Chang C (2002). Chemical retardation of phosphate diffusion in an acid soil as affected by liming. Nutrient Cycling in Agro ecosystems, 64:213-224.

Harold H, Keyser, Li F (1992). Potential for increasing biological nitrogen fixation in soybean. Plant and Soil. 141: 119-135.

Harper JE (1974). Soil and symbiotic nitrogen requirements for optimum soybean. Crop Science. Vol. 14 No. 2 p. 255-260.

Haru A, Ethiopia W (2012). Influences of Inoculation Methods and Phosphorus Levels on Nitrogen Fixation Attributes and Yield of Soybean (*Glycine max* L.).American Journal of Plant Nutrition and Fertilization Technology 2(2): 45-55.

Hayat R, Ali S, Khan FS (2004). Effect of Nitrogen and *Rhizobium* Inoculation on Yield, N uptake and Economics of Mungbean. International Journal of Agriculture & Biology 1560-8530/2004/06-3-547-551.

Hokmalipour S, Darbandi MH (2011). Effects of Nitrogen Fertilizer on Chlorophyll Content and Other Leaf Indicate in Three Cultivars of Maize (*Zea mays* L.). World Applied Sciences Journal 15(12): 1780- 1785.

Hussain N, Mehdi M, Kant RH (2011).Response of Nitrogen and Phosphorus on Growth and Yield Attributes of Black Gram (*Vigna mungo*). Research Journal of Agricultural Sciences 2011, 2(2): 334-336.

Ibrahim KA, Elsheikh EAE, Naim AMEI, Mohamed EA (2011). Effect of *Bradyrhizobium* Inoculation on Yield and Yield's Components of Soybean (*Glycine max* (L.)) Grown in Sudan. Australian Journal of Basic and Applied Sciences, 5(7): 793 – 799.

Imsande J (1989). Rapid dinitrogen fixation during soybean pod fill enhances net photosynthetic output and seed yield: A new perspective. Agronomy Journal. 81: 549-556.

Israel DW (1987).Investigation of the role of phosphorus in symbiotic dinitrogen fixation. Plant Physiology. 84, 835–840.

Jabbar BKA, Saud HM (2012). Effects of Phosphorus on biological nitrogen fixation in soybean under irrigation using saline water. Global Journal of Science Frontier Research Agriculture & Biology. Volume 12 Issue 1Version 1.0.

Javaid A, Mahmood N (2010). Growth, nodulation, and yield response of soybean to biofertilizers and organic manures. Plant Journal of Botany. 42 (2): 863-871.

Jessop RS, Hetherington SJ, Hoult EH (1984). The effect of soil nitrate on the growth, nodulation and nitrogen fixation of chick peas (*Cicer arietinum*).Plant and Soil. 82, 205 – 214.

Jinze X (2010).Tanzania soybean development strategy (tsds) 2010 to 2020.

Kamara AY, Kwari J, Ekeleme F, Omoigui L, Abaidoo R (2010). Effect of phosphorus application and soybean cultivar on grain and dry matter yield of subsequent maize in the tropical savannas of north-eastern Nigeria. African Journal of Biotechnology 7(15).

Kashturikrishna S and Ahlawat PS (1999). Growth and yield response of pea (*Pisum sativum*) to moisture stress, phosphorus, sulphur and zink fertilizers. Indian Journal of Agronomy. 44:588-596.

Katundala P (2011). Symbiotic Nitrogen fixation and seed development of genetically modified soybean in relation to *Bradyrhizobium i*noculation and nitrogen use under saline dykeland soil conditions. P.132

Koutroubas SD, Papakosta DK, Gagianas AA, Papanikolaou EP (1998). Estimation and partitioning of nitrogen Fixed by soybean in Mediterranean climates. Journal of Agronomy Crop Science. 181: 137-144.

Kumar R, Chandra R (2008). Influence of PGPR and PSB on *Rhizobium leguminosarum* Bv. Viciae strain competition and symbiotic performance in lentil. World Journal of Agricultural Sciences. 4 (3): 297-301.

Lambers H, Shane MW, Cramer MD, Pearse SJ, Veneklaas EJ (2006). Root structure and functioning for efficient acquisition of phosphorus: matching morphological and physiological traits. Annual Botanical. 98: 693-713.

Laswai HS, Mpagalile JJ, Silayo VCK, Ballegu WR (2005) (a).Use of soybeans in food formulation in Tanzania. In: Myaka FA, Kirenga G, Malema B (eds) 2006. Proceedings of the First National Soybean Stakeholders Workshop, 10th-11th November 2005, Morogoro- Tanzania. Pp. 52–59.

Leidi EO and Rodriguez-Navarro DN (2000). Nitrogen and Phosphorus availability limit N2 fixation in bean. New Phytologist. (2000), 147, 337-346.

Lindemann WC, Glover CR (2003). Nitrogen fixation by legumes.

Makoi, JHR and Ndakidemi, PA 2007. Biological, ecological and agronomic significance of plant phenolic compounds in rhizosphere of the symbiotic legumes. *African Journal of Biotechnology*, 6: (12)1358-1368.

Makoi JHJR, Bambara S, Ndakidemi PA (2013). *Rhizobium* inoculation and the supply of molybdenum and lime affect the uptake of macroelements in common bean (*P. vulgaris* L.) plants. American Journal of Crop Science. 7(6):784-793

Marschner H (1995). Mineral Nutrition of higher plants. Academic Press, San Diego.

Mmbaga ET, Friesen D (2003). Adoptable maize/legume systems for improved maize production in northern Tanzania. African Crop Science Conference Proceedings, Vol. 6.649-654.

Mohammadi K, Sohrabi Y, Heidari G, Khalesro S, Majidi M (2012). Effective factors on biological nitrogen fixation. African Journal of Agricultural Research Vol. 7(12), pp.1782-1788.

Montanaro G, Dichio B, Xiloyannis C, Celano G (2005). Light influences transpiration and calcium accumulation in fruit of kiwifruit plants (*Actinidiadeliciosavardeliciosa*). Plant Science, 170: 520-527.

Morad M, Sara S, Alireza E, Reza CM, Mohammad D (2013). Effects of seed inoculation by *Rhizobium* strains on yield and yield components in common bean cultivars (*Phaseolus vulgaris* L.). International Journal of Biosciences, Vol. 3, p. 134-141, 2013.

Ndakidemi PA, Bambara S, Makoi JHJR (2011). Micronutrient uptake in common bean (*Phaseolus vulgaris* L.) as affected by *Rhizobium* inoculation, and the supply of molybdenum and lime. Plant Omics Journal. 4(1):40-52.

Ndakidemi PA, Dakora FD, Nkonya EM, Ringo D, Mansoor H (2006). Yield and economic benefits of common bean (*Phaseolus vulgaris*) and soybean (*Glycine max*) inoculation in northern Tanzania. Australian Journal of Experimental Agriculture. 2006, 46, 571- 577.

Ndakidemi, PA and Dakora, FD (2003). Legume seed flavonoids and nitrogenous metabolites as signals and protectants in early seedling development. Functional Plant Biology.30: 729 -745.

Nekesa A, Ruto O, Thuita EC, Ndungu MN, Kifuko KW, Bationo A (2007). The potential of underutilized phosphate rocks for soil fertility replenishment in Africa: case studies in Western Kenya.

Nicolas P, Chiara P, Karine M, Gilles I, Alexandre J (2006). Reactive oxygen and nitrogen species and glutathione: key players in the legume – *Rhizobium* symbiosis. Journal of Experimental Botany, Vol. 57, No. 8, pp. 1769 – 1776.

Olivera M, Tejera N, Iribarne C, Ocana A, LIuch C (2004).Growth, nitrogen fixation and ammonium assimilation in common bean (*Phaseolus vulgaris*): effect of phosphorus. Physiologia Plantarum. 121: 498-505.

Olufajo OO (1990). Response of promiscuously nodulating soybean to N and P fertilization and *Bradyrhizobium* inoculation in ferruginous tropical soils (Haphustalf). Fertilizer Research. 25: 93-100, 1990.

Pacovsky RS, Bethlenfalvay GJ and Paul EA (1986).Comparisons between P-fertilized and mycorrhizal plants. Crop Sciences. 26:151-156.

Rahman MM, Bhuiyan MMH, Sutradhar GNC, Paul AK (2008). Effect of Phosphorus, Molybdenum and *Rhizobium* Inoculation on yield and yield attributes of mungbean. International Journal of Sustainable Crop Production. 3(6): 26-33 (October 2008).

Rengel Z (2008). Bioavailability of Phosphorus and Micronutrients in the soil-Plant-Microbe Continuum. 5th International Symposium 15 MOM 2008-November 24th-28th, 2008-Pucon, Chile

Richardson AE, Barea JM (2009).Acquisition of phosphorus and nitrogen in the rhizosphere and plant growth promotion by microorganisms. Plant Soil (2009) 321:305–339.

Rogers, D (1997). Soybean production handbook. Publication Volume C 449.

Rotaru V (2010). The effects of phosphorus application on soybean plants under suboptimal moisture conditions. LucrăriŞtiinţifice-vol. 53, Nr. 2/2010, Seria Agronomie.

Roy RN, Finck A, Blair GJ, Tandon HLS (2006). Plant nutrition for food security. A guide for integrated nutrient management. FAO, 2006.

Russel SJE (2008). Soil conditions and plants growth, Daya Books.

Sadeghipour O, Abbasi S (2012). Soybean response to drought and seed inoculation. World Applied Sciences Journal 17(1): 55 – 60.

Salvagiotti F, Cassman KG, Specht JE, Walters DT, Weiss A, Dobermanni A (2008). Nitrogen uptake, fixation and response to fertilizer N in soybeans: A reviewAgronomy and Horticulture Department 4-29-2008.

Sanginga N, Dashiell K, Okogun JA, Thottappilly G (1997). Nitrogen fixation and N contribution by promiscuous nodulating soybeans in the Southern Guinea savanna of Nigeria. Plant and Soil. 195: 257-266.

Santos MA, Gerald IO, Garcia AAF, Bortolatti N, Schiavon A, Hungria M (2013). Mapping of QTLs associated with biological nitrogen fixation traits in soybean. Hereditas 000: 001 – 009.

Sara S, Morad M, Reza CM (2013). Effects of seed inoculation by *Rhizobium* strains on Chlorophyll content and protein percentage in common bean cultivars (*Phaseolus vulgaris L*.). International Journal of Biosciences .Vol 3, No. 3, p. 1-8.

Shahid MQ, Saleem MF, Khan HZ, Anjum SA (2009). Performance of soybean (*Glycine max* L.) under different phosphorus levels and inoculation. Pakistan Journal of Agricultural Sciences. 46(4): 237-241.

Sharma MK, Kumawat DM (2011). Co-inoculation study of *Bradyrhizobium japonicum* and *Aspergillus Niger* in Soybean for Nitrogen fixation. Journal of Microbiology, Biotechnology, 2011/121(3) 383 – 394 Food Sciences.

Shiri-Janagard M, Raei Y, Gasemi-Golezani G, Aliasgarzad N (2012). Influence of *Bradyrhizobium japonicum* and phosphate solubilizing bacteria on soybean yield at different levels of nitrogen and phosphorus. International journal of Agronomy and Plant Production, Vol., 3(11), 544-549, 2012.

Shurtleff W, Aoyagi A, (2007). History of Green Vegetable Soybeans and Vegetable Type Soybeans. Lafayate California.

Sinclaor TR, Vadez V (www.icrisat.org).The Future of Grain Legumes in Cropping Systems. http://dx.doi.org/10.1071/CP12128.

Singh A, Baoule A, Ahmed HG, Dikko AU, Aliyu U, Sokoto MB, Alhassan J, Musa M, Haliru B (2011). Influence of phosphorus on the performance of cowpea (*Vigna unguiculata* (L) Walp.) varieties in the Sudan savanna of Nigeria. Agricultural Sciences 2(3): 313-317.

Singh RP, Gupta SC and Yadav AS 2008. Effect of levels and sources of phosphorus and PSB on growth and yield of blackgram (*Vigna mungo* L. Hepper.). Legume Research. 31(2): 139-141.

Smith DL and Hume DJ (1987). Comparison of 1: tSsay methods for N_2 fixation utilizing white bean and soybean. Canadian Journal of Plant Science. 67: 11-19.

Solomon T, Pant LM, Angaw T (2012). Effects of Inoculation by *Bradyrhizobium japonicum* Strains on Nodulation, Nitrogen Fixation and Yield of Soybean (*Glycine max* L. Merill) Varieties on Nitisols of Bako, Western Ethiopia. International Scholarly Research Network ISRN Agronomy Volume 012, Article ID 261475, 8 pages doi:10.5402/2012/261475.

Tien HH, Hien TM, Son MT, Herrridge D (2002). *Rhizobial* Inoculation and N_2 Fixation of Soybean and Mungbean in Eastern Region of South Vietnam. ACIAR Proceedings 109e.

Tsvetkova GE, Georgiev GI (2003). Effects of phosphorus nutrition on the nodulation, nitrogen fixation and nutrient use efficiency of *Bradyrhizobium japonicum*-Soybean (*Glycine max* L. Merr.) Symbiosis. Bulgarian Journal of Plant Physiology. Special Issue 2003, 331-335.

Uchida, R (2000). Essential nutrients for plant growth: Nutrient functions and deficiency symptoms. Plant nutrient management in Hawaii's soils. College of Tropical Agriculture and Human Resources, University of Hawaii at Manoa: 31-55.

Vincent JM (1980). Factors controlling the legume-*Rhizobium* symbiosis.In Nitrogen Fixation.Vol.2.Eds. W E and W H OrmeJonhson. Pp 103-129. University Park Press. Baltmore, MD.

Vitousek PM, Cassman K, Cleveland C, Crews T, Field CB, Grimm NB, Howarth RW, Marino R, Martinelli L, Rastetter EB, Sprent JI(2002). Towards an ecological understanding of biological nitrogen fixation.

Walter, D (2007). Tanzania: The challenge of moving from Subsistence to profit. Business for Development. OECD publication for development.

Weisany W, Raei Y, Allahverdipoor KH (2013). Role of Some of Mineral Nutrients in Biological Nitrogen Fixation. Bulletin of Environment, Pharmacology and Life Science. Vol 2 (4) March 2013: 77-84.

Wesley TL, Lamond RE, Martin VL, Duncan SR (1998). Effects of late season white bean and soybean. Canadian Journal of Plant Science. 67: 11-19.

Wilcox JR, Shibles RM, (2001). Interrelationships among seed quality attributes in Soybean. Crop Sciences. 41(1):11-14.

Wood CW, Torbert HA, Weaver DB (1993). Nitrogen fertilizer effects on Soybean Growth, Yield and Seed Composition. Journal of Production Agriculture. Vol.6, no.3.

Yagoub SO, Ahmed WMA, Mariod AA (2012). Effect of Urea, NPK and Compost on Growth and Yield of Soybean (*Glycine max* L.), in Semi-Arid Region of Sudan. ISRN Agronomy Volume 2012(2012), Article ID 678124, 6 pages.

Yakubu H, Kwari JD, Sandabe MK (2010). Effect of Phosphorus Fertilizer on Nitrogen Fixation by some Grain legume Varieties in Sudano-Sahelian Zone of North Eastern Nigeria. Nigerian Journal of Basic and Applied Science (2010), 18(1): 19-26.

Zahran HH (1999). *Rhizobium*-legume Symbiosis and Nitrogen Fixation under severe Conditions and in an Arid Climate. Microbiology and Molecular Biology Reviews. 1999 December, 64(4): 968-989.

Zarei I, Sohrabi Y, Heidari GR, Jalilian A, Mohammadi K (2012). Effects of biofertilizers on grain yield and protein content of two soybean (*Glycine max* L.) cultivars. African Journal of Biotechnology 11(27): 7028-7037.

Legumes play a significant role in sustainable agriculture through their ability to improve soil fertility and health. Legumes, with a mutual symbiotic relationship with some bacteria in soil, can...Â Besides, P also plays a significant role to root development, nutrient uptake, and growth of legume crops. But most of the agricultural soils have inadequate amounts of P to support efficient BNF as it exists in stable chemical compounds which are least available to plants. The deficiency of P causes significant yield reduction in leguminous crops. The mineral P sources are nonrenewable, unlike N. So there is a need to enhance P use efficiency (PUE) for better legume productivity and soil sustainability. Benefits of phosphorus include healthy bone formation, improved digestion, regulated excretion, protein formation, hormonal balance, and improved energy extraction.Â It is an essential part of our diet, particularly as children, when the most growth and development occurs. Sometimes phosphorus is confused with phosphate. In reality, they are two different things. Effects of rhizobial inoculation and applied nitrogen on the utilization efficiency of N, P and K were studied in relation to the yield advantage in additive maize/mungbean intercrops at Los BaÃ±os, Philippines in 1988. Inoculation increased grain yield of both maize (Zea mays L.) and mungbean (Vigna radiata (L.) Wilczek). Yield of maize increased by 60% in the sole crop and 71% in the intercrop as the N application rate was increased from 0 to 90 kg/ha, with a corresponding decrease of 29â€“35% in the yield of the associated mungbean.Â Radiation interception and growth in an intercrop of pearl millet/groundnut. Field Crops Research 7, 141â€“160. Natarajan, M. & Willey, R. W. (1980).