PHYSIOLOGICAL RESPONSES OF COWPEA (*Vigna unguiculata* L. Walp) VARIETIES TO RHIZOBIA INOCULATION, PHOSPHORUS APPLICATION AND SEQUENTIAL CROPPING SYSTEM IN MINNA, NIGERIA

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PhD/SAAT/2013/522

DEPARTMENT OF CROP PRODUCTION
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MINNA

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A THESIS SUBMITTED TO THE POSTGRADUATE SCHOOL, FEDERAL UNIVERSITY OF TECHNOLOGY, MINNA, NIGERIA IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE AWARD OF THE DEGREE OF DOCTOR OF PHILOSOPHY (PhD) IN CROP PRODUCTION (CROP PHYSIOLOGY)

OCTOBER, 2019

DECLARATION

I hereby declare that this thesis titled: “Physiological responses of cowpea (V*igna unguiculata* L. Walp) varieties to rhizobia inoculation, phosphorus application and sequential cropping system in Minna, Nigeria” is a collection of my original research work and it has not been presented for any other qualification anywhere. Information from other sources (published or unpublished) have been duly acknowledged.

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MINNA, NIGERIA

SIGNATURE/DATE

CERTIFICATION

The thesis titled: "Physiological responses of cowpea (V*igna unguiculata* L. Walp) varieties to rhizobia inoculation, phosphorus application and sequential cropping system in Minna, Nigeria" by: ADEDIRAN, Olaotan Abimbola meets the regulations governing the award of the degree of Doctor of Philosophy (PhD) of the Federal University of Technology, Minna and it is approved for its contribution to scientific knowledge and literary presentation.

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DEDICATION

This work is dedicated to GOD ALMIGHTY who has been my pillar.

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ABSTRACT

Cowpea is the most important grain legume in Nigeria but the yield obtained on farmers' fields is far below the potential yield of the crop. This study aimed at exploiting rhizobia inoculation, phosphorus application, varietal differences and sequential cropping system to improve the productivity and profitability of cowpea per unit area in Minna, Nigeria. A glasshouse and two field experiments were conducted between 2015 and 2017. The glasshouse experiment was a factorial combination of four nitrogen sources (uninoculated, inoculated with USDA 3451 and USDA 3384 rhizobia strains and 90 kg N ha^{-1}), soils collected from 20 locations in the Nigerian savannas and two varieties of cowpea (IT93K-452-1 and IT99K-573-1-1) replicated three times and laid in a completely randomized design. The first of the field experiments conducted on three farmers' fields in Minna was a factorial combination of three nitrogen sources (uninoculated, inoculated with USDA 3451 and USDA 3384 rhizobia strains), three phosphorus rates (control, 20 kg P ha^{-1} and 40 kg P ha^{-1}) and three varieties (IT93K-452-1, IT99K-573-1-1 and TVX-3236) in the first year. In the second and third year of planting, the strains were replaced with BR 3262 and BR 3267 and 90 kg N ha^{-1} was added as part of the treatments. The treatments were laid in randomized complete block design. The second field experiment evaluated the performance of six cowpea varieties (IT93K-452-1, IT99K-573-1-1, TVX-3236, Kanannado, Oloyin and IT90K-76) in cowpea sequential cropping system. Data were collected on growth, yield, nodulation and physiological parameters. The results revealed that the cowpea varieties successfully formed symbiosis with the introduced rhizobia strains in all the locations. Rhizobia inoculation increased nodulation in 11 out of the 20 locations with percentage increase ranging from 4 to 43%. Plants fertilized with 90 kg N ha^{-1} had significantly higher biomass yield than the inoculated and uninoculated plants which had similar biomass yield. Phosphorus significantly ($P \leq 0.05$) increased the photosynthetic activities, nodulation, N-fixation, growth and yield of the cowpea varieties in the three years with application of 20 kg P ha^{-1} increasing grain yield by 49-95% over the control. Significant interactions exists between rhizobia inoculation, phosphorus and varieties on some growth and yield attributes. IT99K-573-1-1 maintained the highest productivity among the varieties however, TVX-3236 appeared to be more P efficient having significantly higher growth rate, nodule weight and grain yield at lower P rates. Number of pods per plant had the highest correlation coefficient with grain yield in the three years ($r=0.79$). Crop growth rate (CGR), leaf area index (LAI) and quantum yield of photosystem II (Phi 2) explained 67.29% of the variation in grain yield (R^2= 67.29%) and these three physiological parameters could significantly predict the grain yield. All the varieties were successfully planted in two sequence in each growing season except Kanannado and there was significant variation in the growth and yield attributes of the cowpea varieties in sequential cropping system. IT93K-452-1, IT99K-573-1-1, TVX-3236 and IT90K-76 all had significantly higher ($P \leq 0.05$) grain yield and profitability with gross margin ranging between ₦407,910 and ₦1,131, 967 than Oloyin and Kanannado varieties which had gross margin range of -₦123,065 and ₦88,931. Similarly, double cropping of these four varieties increased the gross margin by an average of 115% in 2016 and 140% in 2017 over the traditional system of planting once. It can be concluded from these results that N limits the productivity of cowpea in some soils of Nigerian savannas and rhizobia inoculation with highly effective strains than what was used in this study can overcome this limitation. CGR, LAI and Phi 2 could be exploited to increase the grain yield of cowpea through agronomic practices and plant breeding programs. Application of 20 kg P ha^{-1} is sufficient for the optimum performance of cowpea in the study area and double cropping of improved, early and medium maturing varieties of cowpea per season is capable of increasing the productivity and profitability of cowpea in the study area.

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CHAPTER ONE

1.0 INTRODUCTION

1.1 Background to the Study

Cowpea, *Vigna unguiculata* L. Walp, is an important grain legume. It is a food and fodder crop grown in the semi-arid tropics of Africa, Asia, Europe, United States, Central and South America (IITA, 2009). The grain contains about 25% protein (Ogbonnaya *et al*., 2003). Cowpea has been for many years a crop that combats food insecurity. It also provides cash income for subsistence farmers, retailers and food vendors in rural and urban areas. The haulm is quality fodder for livestock. In addition, the crop is capable of fixing nitrogen from the atmosphere for its growth demand (Asiwe *et al*., 2009), thereby reducing nitrogen fertilizer demand and cost of production. It is an important companion crop in most cereal-legume cropping systems because of the benefit from the residual nitrogen originating from the decay of its leaf litter, roots and root nodules (Okereke *et al*., 2006).

Inoculation is the practice of applying microorganisms to agricultural or chemical systems to bring about desirable transformations such as enhanced nitrogen (N_2) fixation, increased phosphorus uptake, more rapid biodegradation and improved disease resistance. The most important microorganism used as inoculant in legume crop production are rhizobia. Rhizobia inoculation is the process of introducing the commercially prepared rhizobia into the seed or soil to enhance nitrogen fixation. Cowpea establishes symbiotic association with rhizobia forming root nodules. It is inside these nodules that the nitrogenase enzyme in the rhizobium reduces N_2 into ammonia (NH_3) through the glutamate synthase/glutamine oxoglutarate aminotransferase (GS/GOGAT) pathway. This leads to exchange of nitrogenous solutes with host plant for photosynthate (Pule-Meulenberg, 2010) thereby making N available for the use of the host plant; but the amount of N_2

fixed is not in all cases enough due to the presence of ineffective or low numbers of indigenous rhizobia (Ulzen, 2014).

Nitrogen and phosphorus are the most important mineral nutrients limiting increased crop productivity in the Nigeria Savanna. Among these, phosphorus is the most critical nutrient needed by legumes for growth and optimum symbiotic relationship. It is important to cowpea performance as it increases seed yields, nodulation and nitrogen fixation (Magani and Kuchinda, 2009).

Sequential cropping refers to growing two or more crops in sequence on the same field per year; the succeeding crop is planted after the preceding crop has been harvested. This is achievable in some cowpea varieties if planted early. It is one of the alternatives of increasing the productivity of crop from a unit of land. Its adoption may substitute for expanding cropland acreage which may not always be available and sustainable due to increase in population and urbanization.

1.2 Statements of the Research Problem

Over the years, food requirements have increased while land availability has become less due to increasing population and urban development. Thus, the only way to increase agricultural production is to increase the yield of individual crop per unit area of land. Low crop productivity is a general problem facing most farming systems in sub-Saharan Africa. These low yields are pronounced in grain legumes and are often associated with declining soil fertility and reduced nitrogen fixation (Mfilinge *et al.*, 2014). Nitrogen fixation in cowpea is inferior to that of other grain legumes such as groundnut, soybean and pigeon pea, perhaps because of the inadequate effectiveness or efficacy of the indigenous rhizobia to supply the nitrogen required (Fening and Danso, 2001). The amount of N_2 fixed by cowpea is usually not adequate to meet the plant's demand for nitrogen thus affecting its yield (Ulzen, 2014).

Cowpea yields in Nigeria remain one of the lowest among all food legume crops with an average of 450 kg ha^{-1} (Omotosho, 2014). This is very low compared to the grain yield of over 2600 kg ha^{-1} reported in South Africa (Pule-Meulenberg *et al*., 2010). Low soil fertility is among the factors responsible for the low yield experienced in cowpea as most tropical soils are deficient in essential nutrients particularly N and P (Abayomi *et al*., 2008). Nigeria is the largest producer and consumer of cowpea worldwide (IITA, 2009). She accounts for about 45% of the global cowpea production and over 55% of the production in Africa. All the cowpea produced in the country is consumed and the shortfall is augmented with imports from neighbouring countries, mainly Niger Republic (Coulibaly and Lowenberg-DeBoer, 2013). Nigeria is projected to remain net-importer of cowpea through 2020 (Olufajo *et al*., 2014). The traditional cropping systems used in the production of cowpea in Nigeria in which cowpea is planted as a minor crop in intercrop or relay cropping is not delivering the potential yield of the crop.

1.3 Justification for the Study

Cowpea is an important grain legume in Nigeria and in many other parts of the world. It is estimated that cowpea supplies about 40% of the dietary protein requirement of most of the people in Nigeria (Kamai *et al*., 2014). The current demand for cowpea grains is nearly 3.2 million tons and this is expected to grow at 3.6% per year (Olufajo *et al.*, 2014). Therefore, there is need to work on improving the yield to meet the growing demand. Rhizobia inoculation has been reported to improve the yield of legumes and enhance soil fertility but there is a dearth of information on the responses of cowpea to rhizobia inoculation. Ulzen (2014) indicated that legumes grown without rhizobia inoculation may be retarded in growth with consequent low yield. Improvement in cowpea nodulation, nitrogen fixation and grain yield have been reported in few studies as a result of rhizobia inoculation and fertilizer application (Taiwo and Oladapo, 2000; Sarker *et al*., 2001; Fening and Danso, 2001; Abayomi *et al*., 2008; Nyoki, 2013; Omotosho, 2014) but very few have been documented in Nigeria. It is important to promote inoculant use in African agriculture, especially among resource-poor farmers who cannot afford expensive mineral fertilizers (Ndakidemi *et al*., 2006). Some African countries such as Rwanda, Malawi, Egypt and Zimbabwe have turned to efficient exploitation of biological nitrogen fixation (BNF) by legumes in their farming system in an attempt to cut down on fertilizer expenses (Otieno *et al*., 2007). Effective symbiotic relationship depends on the cultivar and the rhizobium strain (Fall *et al*., 2003). Hence, the need for the use of different varieties and different rhizobia strains. Use of improved crop cultivars and alteration in sowing dates of crops to accommodate more than one harvest in a growing season is an option to increase crop production from a unit of land, improve cash flow, spread risk and achieve more efficient use of equipment and greater return from investment (Prasa and Singh, 1997; Asante *et al*., 2001; Mahama *et al*., 2013). Many work have been done on cowpea-cereal in sequential cropping system but there is a dearth of information on cowpea in sequential cropping.

Increasing crop yield requires input from many areas of agriculture including physiology. The study of the processes that determine yield in crop (physiology) is very important for sustainable high yield realization in all crops. One of the most important approach of crop productivity improvement is effective evaluation of physiological traits. Hence, the need to study the physiological responses of selected varieties of cowpea to rhizobia inoculation, phosporus rates and cowpea sequential cropping system

1.4 Aim and Objectives

The aim of the study was to determine the physiological responses of cowpea varieties to rhizobia inoculation, phosphorus application and sequential cropping system in Minna, Nigeria.

The objectives of this study were to determine:

i. if cowpea growth in savanna soils is limited by Nitrogen (N)
ii. the effect of rhizobia inoculation on the photosynthetic activities, nodulation, N-fixation, growth and yield of selected cowpea varieties
iii. the effect of increasing phosphorus (P) rates on the photosynthetic activities, nodulation, N-fixation, growth and yield of selected cowpea varieties
iv. the inter-relationships between yield, growth and physiological characters of cowpea
v. the inter-relationship between environmental factors and photosynthetic efficiency of cowpea
vi. the performance of some selected cowpea varieties in cowpea sequential cropping system
vii. the profitability of the cowpea varieties in cowpea sequential cropping system
viii. and to identify how many times selected cowpea varieties can be grown in sequence within a cropping season in the southern guinea savanna of Nigeria.

CHAPTER TWO

2.0 LITERATURE REVIEW

2.1 General Description of Cowpea

Vigna unguiculata (L.) Walp is believed to have originated from Africa (Valenzuela and Smith, 2002). It belongs to the family Fabaceae. The common names include cowpea, black eye bean, black eye pea, *haricot dolique* (Fr), *makunde* (Po), *Mkunde* (Sw) (Madamba *et al.*, 2006). It is the most important pulse crop in the savanna regions of West and Central Africa. It is a climbing, trailing or more or less erect annual or perennial herb. It has well developed tap root with many lateral and adventitious roots. Some varieties are erect and bushy, while others have viney growth habit. There is a large morphological diversity found within the crop (Madamba *et al*., 2006). Cowpeas may reach a canopy height of 70–91cm, however, more determinate types was around 51–61 cm (FAO, 2006). Madamba *et al*. (2006) stated that the stem can be up to 4 m in length. The seed pods are borne above the leaf axil and is typically 7.5 to 15.2 cm long and has 6 to 13 seeds per pod Madamba *et al*. (2006). The 1000-seed weight is 150-300 g and between 4,180 to 8,800 seeds per kg (FAO, 2006)

Cowpea is a warm season crop well adapted to heat and drought conditions. Most cultivars are short-day plant or indeterminate in its flowering response although there are day neutral cultivars (FAO, 2006). The non-vining types tend to be more determinate (Ajeigbe *et al*., 2010). Generally speaking, cowpea is classified into two categories depending upon the time taken to reach maturity; these are early and late. Cowpea is tolerant to moderate shade and so grows with tall crops such as maize and sorghum (FAO, 2006). It grows best at day temperatures of 20-35^{o}C. It can be grown on a wide range of soil types with pH 5.5-8.3 provided the soil is well drained (Valenzuela and Smith, 2002). It is a low-altitude plant, but will grow quite well up to 1,500 m elevation (FAO, 2006). The seed coat can be either smooth or wrinkled and of various colors (white, cream, green, buff, red, brown, and black). Seed may also be speckled, mottled, or blotchy (Ajeigbe *et al*., 2010).

2.2 Uses and Importance of Cowpea

Cowpea is the most economically important indigenous African legume. It plays an important nutritional role in developing countries of the tropics and sub-tropics, especially in sub-Saharan Africa, Asia, Central and South America. The haulm is a quality fodder for livestock. Because of its high protein content (20-25%), it has been referred to as “poor man’s meat” (Fall *et al.*, 2003). Its young leaves, pods and peas are also rich in several vitamins and minerals (IITA, 2009) which have made its usage for human consumption and animal feeding more important. Mature grains contain 12 g water, 1407 kj energy, 23.5 g protein, 1.3 g fat, 60 g carbohydrate, 10.6 g fiber 110 mg Ca, 184 mg Mg, 424 mg P per 100 g (USDA, 2004). It’s high protein content, adaptability to different types of soil and intercropping systems, resistance to drought, and ability to improve soil fertility (through nitrogen fixation, decomposition of its remains or when used as green manure) and prevention of erosion (when grown as a cover crop) make it an important economic crop in many developing regions. The sale of the stems and leaves as animal feed during the dry season also provides a vital income for farmers (IITA, 2009).

The mature grains are cooked and eaten alone or together with vegetables. The immature seeds and pod as well as the leaves are also eaten as vegetable. The roots are sometimes eaten (for example in Ethiopia and Sudan). The decorticated seeds can as well be processed into bean cake (“*akara* or *moin moin”*). Small quantities of cowpea flour are processed into crackers, composite flour, baby food and coffee substitute (Madamba *et al*., 2006). Various medicinal uses of cowpea have been reported. The leaves and seeds are applied as a poultice to treat swellings and skin infection, leaves are chewed to treat tooth ailments, carbonated seeds are applied on insect bites, and the root is used as an antidote for snake bites and to treat epilepsy, chest pain, constipation and dysmenorrhea (Madamba *et al*., 2006). Their calcium and iron content are higher than that of meat, fish and egg and the iron content equates that of milk; the vitamins- thiamin, riboflavin, niacin content equal the levels found in lean meat and fish which make them very useful in blood cholesterol reduction (Agbogidi and Egho, 2012). Some researchers have shown that daily consumption of 100–135 g of dry beans reduces serum cholesterol level by 20% thereby, reducing the risk of coronary heart diseases by 40% (Adaji *et al*., 2007).

2.3 Physiological Measurements as a Crucial Tool for Crop Improvement

One of the most important approach to breeding for higher crop yield is effective evaluation of physiological traits in yield differentiation and recognition of the gene in control (Saki, 2012). Growth analysis are useful tools for quantitative analysis of growth in different subjects such as plant breeding, plant ecology and physiology (Ebrahim *et al.*, 2014). Growth indices such as crop growth rate (CGR), relative growth rate (RGR) and net assimilation rate (NAR), leaf area index (LAI), leaf area ratio, leaf area duration are indices which are often used for evaluation of plant productivity capability and environmental efficiency (De Sclaux *et al.,* 2000; Anzoua *et al.*, 2010). The crop growth rate measures the rate of dry matter production per unit time, the leaf area index is a reflection of the ability of the leaf canopy to intercept light energy, and thereby it can be used to predict photosynthetic capacity. The net assimilation rate is a measure of the photosynthetic efficiency of the leaf surface of the plant, the leaf area ratio is also is a measure of the efficiency of the leaf surface in producing dry matter. The leaf area duration measures the duration and extent of photosynthetic tissue of the crop canopy (Das, 2011).

Photosynthetic parameters are valuable descriptors for analyzing the sensitivity and recovery of plants under variable abiotic conditions, making it possible to assess acclimation and stress responses, determine the appropriate management practice to improve yield. It is also an important tool in predicting yield and ecological consequence in the face of climate change. Of recent, chlorophyll fluorescence has been used as an indicator of photosynthetic activities and measurement of stress in plants (Harb *et al*., 2018).

During photosynthesis, light energy is harvested by pigments found in antenna and photosystem cores. This energy is either used in photosynthesis (photochemical quenching), emitted as heat (non-photochemical quenching), or re-emmited as light (chlorophyll fluorescence). These three processes occur in competition such that the increase in one results in the decrease of the other. The fluorescence part can be measured and then used to calculate photochemical and non-photochemical quenching. This is done with a fluorometer. Common photochemical quenching measurements derived from chlorophyll fluorescence include: quantum yield of photosystem (PS) II (Φ PSII) which measures the proportion of light absorbed by chlorophyll associated with PSII that is used in photosynthesis. It is also called Phi 2 by some authors (Maxwell and Johnson, 2000), maximum quantum yield of PSII (FV/FM) which is the quantum efficiency of PSII if all the PSII reaction centers are open and photochemical quenching (q^p) which is the proportion of PSII reaction centers that are open. Quantum yield of photosystem is the most important measurements which measure the photosynthetic efficiency of the plant.

Non-photochemical quenching (NPQ) is another measurement made which is the measure of the amount of energy dissipated as heat. NPQ could be of three types the fastest, occurring in seconds to minute (qE) which involves the xanthophyll. The second type, taking minutes to hours (qT) occurs when the light harvesting complexes attached to photosystem II move to photosystem I in an attempt to balance light capture and electron flow through the two systems. The slowest takes hours to days and is known as qI, or photoinhibition. Photoinhibition occurs when the reaction centers start becoming damaged and must be broken down to be repaired. Other non-regulated non photochemical quenching Y(NO) or phi no can also be measured from chlorophyll florescence. It is a measure of the ratio of the energy that is lost through other means other than heat.

The advantage of this technique over many other techniques such as growth analysis is that it provides rapid and non-destructive measurement of plant physiological processes. Yield is a complex feature which depends on the function of physiological combined processes. The crucial role of physiological features in crop productivity improvement has been reported by a number of researchers (Esfahani *et al.*, 2006; Mahdavi *et al.*, 2006; Katsura, 2007).

2.4 Physiology of the Rhizobia - Legume Symbiosis

The atmosphere contains about 10^{15} tonnes of N_2 gas, which cannot be used in this form by most living organisms unless it is reduced to ammonia (Mabrouk and Belhadj, 2011). The symbiotic relationship between legume and rhizobia makes the nitrogen in the atmosphere available for plant use. This association is responsible for reducing 120 million tons of atmospheric nitrogen to ammonia each year (Ravikumar, 2012). The establishment of an effective and efficient symbiosis between rhizobia and the host legume is essential for viable legume production. Optimum growth of leguminous plants is usually dependent on symbiotic relationships with N_2-fixing bacteria (Xavier and Germida, 2003).

Rhizobia (species of the genera *Rhizobium*, *Bradyrhizobium*, *Sinorhizobium*, *Azorhizobium*, *Mesorhizobium* and *Allorhizobium*) are soil bacteria that fix nitrogen after becoming established inside root nodules of legumes (Mabrouk and Belhadj, 2011) which also serve as a protective enclosure around them. In general, they are gram-negative, aerobic, motile, non-sporulating rods. The symbiosis between legumes and rhizobia involves two stages which are nodulation and nitrogen fixation. The symbiotic relationship implies a signal exchange between the rhizobium and the legume that leads to mutual recognition and development of symbiotic structures. For the establishment of an effective symbiosis two main classes of bacterial symbiosis genes are needed: nodulation and nitrogen fixation genes. Nodulation genes (e.g. nodABC) encode enzymes responsible for the biosynthesis and secretion of Nod factors, which are host determinant lipochitooligosaccharides (LCOs) that interact with the plant flavonoids. Nitrogen fixation genes (nif and fix) include the structural genes for the nitro-genase (nifHDK), the enzyme responsible for atmospheric nitrogen fixation (Laranjo *et al.*, 2014).

Rhizobia live in the soil where they are able to sense flavonoids secreted by the roots of their host legume plant. The flavonoids trigger the secretion of nod factors, which in turn are recognized by the host plant and can lead to root hair deformation and several cellular responses, such as ion fluxes (Oldroyd, 2013). The best-known infection mechanism is called intracellular infection. In this case the rhizobia enter through a deformed root hair forming an intracellular tube called the infection thread. The infection triggers cell division in the cortex of the root where a new organ, the nodule, appears as a result of successive processes. A second mechanism is called "crack entry". In this case, no root hair deformation is observed and the bacteria penetrate between cells, through cracks produced by lateral root emergence. Later on, the bacteria become intracellular and an infection thread is formed. Infection threads grow to the nodule, infect its central tissue and release the rhizobia in these cells, where they differentiate morphologically into bacteroids and fix nitrogen from the atmospheric elemental N_2 into a plant-usable form, ammonium, using the enzyme nitrogenase (Oldroyd, 2013). In return, part of the legume photosynthate is used up by the rhizobium. In some cases, the benefit is not mutual such that the rhizobia are able to induce and infect nodules on their hosts without actually fixing nitrogen (Fening and Danso, 2002). Symbiotic effectiveness (ability to fix atmospheric nitrogen) of rhizobia population is one of the most important parameters for selecting strains for inoculant production (Fening and Danso, 2002).

The biological nitrogen fixed resultant from the rhizobia-legumes symbioses can benefit not only the host crop, but it may also have positive effects on subsequent crops. Yield increases of crops planted after harvesting legumes are often equivalent to those expected from application of 30 to 80 kg of fertilizer N ha^{-1} (Lupwayi *et al.*, 2004). Legume-rhizobia symbioses contribute at least 70 million tonnes of N per year, and approximately, half is derived from the cool and warm temperature zones, while the remainder is derived from the tropics (Mabrouk and Belhadj, 2011). Inputs of fixed N for alfalfa, red clover, pea, soybean, cowpea and vetch are estimated to be between 65 to 335 kg of N ha^{-1} year $-^{1}$ (Mabrouk and Belhadj, 2011). The major benefits of the legume-rhizobia symbiotic interaction are diminished nitrogen fertilizer requirements and improved plant growth and health (Giller, 2001). The result of the study conducted by Osunde *et al*. (2003) on soybean revealed that 27-50% nitrogen was derived from N_2 fixation. Zhang *et al.* (2003) reported that symbiosis of soybean and *Bradyrhizobium japonicum* can fix up to 200 kg N $ha\text{-}^{1}$ $yr\text{-}^{1}$ reducing the need for expensive and environmentally damaging nitrogen fertilizer. Even though cowpea is capable of deriving up to 99% of its N requirement from symbiotic fixation (Pule-Meulenberg *et al*., 2010), the quantity of N fixed by cowpea is usually low compared to some other legumes (Ulzen, 2014).

2.4.1 Factors affecting nitrogen fixation by rhizobia-legume symbiosis

In the Rhizobium-legume symbiosis, the process of N_2 fixation strongly depends on the physiological state of the host plant. Therefore, a competitive and persistent rhizobium strain is not expected to express its full N_2-fixation activity, if the vigor of the host legume is impaired (Mabrouk and Belhadj, 2011). Walley *et al.* (2006) observed that the amount of nitrogen supplied by biological fixation depends not only on the ability of the inoculant rhizobia to fix nitrogen, but also on the ability of the plant to provide energy to the rhizobia in the nodules. Thus, any factor or factors that influence either the rhizobia directly or the ability of the plant to provide energy to the nodules may have a negative impact on nitrogen fixation and ultimately, crop yield. The authors reported further that herbicides affect nitrogen fixation largely via indirect effects on plant growth and consequent availability of photosynthate to the root nodules.

Crop varieties vary in their genetic ability to fix nitrogen (Omondi *et al.,* 2014). The authors observed variation in the nitrogen fixing ability and grain yield of some soybean varieties. Likewise, Fall *et al*. (2003) observed variation in the nitrogen fixing ability of some cowpea varieties and it was reported that some genes could govern the higher nitrogen fixation character in those cowpea varieties that fixed higher nitrogen. Rhizobia differ in their ability to survive and nodulate legumes under adverse soil conditions (Fening and Danso, 2001). The authors reported that better response of cowpea to inoculation was obtained in an acidic soil due to the bradyrhizobium strain used which seemed tolerant to acidic condition and also observed that native strains were more efficient than the exotic strain (TAL 169) used in their experiment. Some rhizobium strain have been shown to grow under high salt conditions (Kucuk *et al.*, 2006).

Nitrogen fixation in a legume requires adequate nodulation (Giller and Wilson, 1993). Nodulated legumes have the potential to fulfill their demand for nitrogen by fixation and, as a result, can influence the nitrogen balance of the soil (Hardarson and Atkins, 2003).The formation of adequate numbers of nodules on legumes in any soil is dependent on the number of homologous rhizobia in the soil (Fening *et al.*, 2001). According to Theis *et al*. (1991), effective nodulation requires not less than 50 cells/g soil. Any factor that will affect the population of rhizobia in the soil will definitely affect nodulation, hence, nitrogen fixation. Fening and Danso (2002) reported that soil pH clearly influenced the numbers of indigenous rhizobia in the soil. Very low numbers were obtained in soil with average pH of 4.0. Sadal *et al.* (2013) reported that nitrogen fixation in soybean is more sensitive to drought than photosynthesis, and that the high concentrations of shoot ureide and N are associated with sensitivity of N_2 fixation to drought. Water deficiency and drought directly affect persistence and survival of rhizobia in soil, nodule activity and function and also limit nodulation through its effects on root-hair colonization and infection by rhizobia (Mabrouk and Belhadj, 2011). Krasova-Wade *et al*. (2006) also observed that the low moisture content of the semi- arid soils limits the biological nitrogen fixation.

Cultural practices have been reported to affect nitrogen fixation. In the experiment carried out by Matus *et al.* (1997) to determine the influence of tillage and crop rotation on nitrogen fixation in lentil and pea, it was reported that %N fixed was higher in zero/minimum tillage than conventional tillage. The same trend was observed in soybean by Omondi *et al.* (2014). Ferreira *et al*. (2000) further affirmed that rhizobia isolates from no tillage condition plots fixed more atmospheric nitrogen. Zhang *et al*. (2012) also reported that rhizobia population was high under no tillage condition. Minimal disturbance of soil in zero/minimal tillage encourages increase in rhizobia activity, hence, increase in nitrogen fixation (Omondi *et al*., 2014).

Cropping system has been reported to affect nitrogen fixation. Njira *et al.* (2012) reported that the total amount of nitrogen biologically fixed in intercrops (82.8 kg N $ha\text{-}^{1}$) pigeon pea/groundnuts doubled-up cropping (adding together the amount of nitrogen fixed by the component crops) was significantly higher by 33 and 35% than those of the sole groundnuts (55.8 kg N $ha^{-1)}$ and sole pigeon pea (54.1 kg N $ha\text{-}^{1}$), respectively. However, Ghosh *et al.* (2006) reported lower relative nitrogen and dry matter yield in pigeon pea that was intercropped with soybean than the sole cropped pigeon pea. Kouyat´e *et al.* (2012) also reported that cropping system significantly affected cowpea nodulation. The authors obtained low nodulation and grain yield in interhill intercropping with pearl millet.

Some research report have revealed that there is link between pesticide application and nodulation / nitrogen fixation. Fox *et al*. (2004) reported that some pesticides can mimic naturally occurring biochemicals and thereby interfere with various biochemical signaling processes between rhizobia and appropriate host plants. As a consequence, early nodulation events can be disrupted. However, according to their report, not all pesticides have a negative impact on nodulation and the extent to which nodulation was inhibited was dependent on pesticide concentrations. Anderson *et al.* (2004) observed that application of an herbicide (chlorsulfuron) at rates equivalent to two times field rates did not influence rhizobia growth but the subsequent ability of these rhizobia to form nodules was reduced. They further stated that when rhizobia were pre-exposed to relatively high levels of chlorsulfuron, subsequent nodule size and total nitrogen fixed was reduced. Walley *et al*. (2006) suggested that the impact of various herbicides on specific nodulation events may be highly dependent on specific environmental conditions, including different soil characteristics (pH, organic matter, moisture) and weather conditions.

2.4.2 Need for rhizobia inoculation

There is a growing worldwide demand for ecologically compatible, environmentally friendly techniques in agriculture, capable of providing adequate nourishment for the increasing human population and improving the quality and quantity of certain agricultural products. Hence, the application of beneficial microorganisms is an important alternative technique. A rhizobia inoculant is a formulation containing one or more beneficial bacterial strains in an easy-to-use and economical carrier material, which may be organic or inorganic or synthesized from defined molecules for application to leguminous seeds or soils to ensure effective nodulation of the host resulting in abundant supply of nitrogen for crop growth (IITA and N2Africa, 2014). It could be in powdered, liquid or solid form. The carrier could be peat or clay for powered and granular inoculant; water or mineral oil for liquid inoculant (IITA and N2Africa, 2014). Charcoal has also been described as an effective medium in room temperature to carry the rhizobia inoculum, having better physical and chemical characteristic than other carriers. The charcoal based Rhizobia inoculants have produced maximum nodulation and pod yield in *Cajanus cajan, Cajanus arietinum and V. sinensis* (Ravikumar, 2012). Inoculation with beneficial bacteria can be traced back to centuries. Farmers knew that when they mixed soil taken from an area previously grown to legume crops with soil in which non-legumes were to be grown, yields often improve. By the end of the 19th century, the practice of mixing naturally inoculated soil with seeds became a recommended method of legume inoculation in the USA. For almost 100 years, rhizobium inoculants have been produced around the world (IITA and N2Africa, 2014).

There is need for rhizobia inoculation when there are no indigenous rhizobia in the soil or the compactible indigenous rhizobia population in the soil is low i.e the native rhizobia are not numerous enough to stimulate biological nitrogen fixation. The native or the indigenous rhizobia are the rhizobia inhabiting the soils of an area. Rhizobia are not universally present in soils (IITA and N2Africa, 2014). In the survey conducted by Slattery and Pearce (2002), it was revealed that rhizobia strains are present on some soil types but absent in some other soil type, hence, no nodule was formed on the root of the legumes planted on such soils. In the study conducted in Ghana, it was reported that the abundance of cowpea bradyrhizobia was extremely variable among different soils and the trend of occurrence was Rainforest < Semi-deciduous forest < Guinea Savanna < Forest–Savanna transition < Coastal Savanna (Fening *et al.*, 2001). Acidity, salinity, soil temperature and moisture are among the soil factors that determine the availability and population of rhizobia present in a site. In the experiment conducted by Jida and Assefa (2014) to determine the effects of acidity on growth and symbiotic performance of *Rhizobium leguminosarum* bv. *viciae* strains isolated from faba bean producing areas of Ethiopia, it was reported that none of the tested strains was tolerant to pH 4.0 while two of them were found to be tolerant of pH 4.5. When tested at pH 5.0 only one isolate was sensitive. The authors reported further that all the acid tolerant strains were recovered from highly acidic soil (pH 4.8 – 5.2) and the acid sensitive strain was isolated from neutral soil. Hence, the need for inoculation with acid tolerant rhizobium strain in acidic soil to improve nodulation, nitrogen fixation and yield. There is need for inoculation under other stress conditions apart from soil acidity. Inoculation with stress tolerant strains of rhizobia may enhance the nodulation and nitrogen fixation ability of legumes under stressed conditions. For example, the ability of legumes to grow and survive in saline conditions is improved when they are inoculated with salt tolerant strains of rhizobia (Mabrouk and Belhadj, 2011).

Legume and rhizobia differ in the range of partners with which they can form symbioses. Some legumes are capable of freely associating with indigenous rhizobia while some are specific. A legume which nodulates with a restricted number of rhizobia strains (or species), or rhizobia strains nodulating a restricted range of host plants are termed specific. For example, pea is infected by *Rhizobum leguminosarum* biovar *viciae*, while common bean is infected by *Rhizobium leguminosarum* biovar *phaseoli* (Mfillinge *et al*., 2014). Conversely, promiscuity is the ability of a legume host to nodulate with a wide diversity of rhizobia strains, or the ability of a rhizobia strain to nodulate with a wide diversity of legume host plants. There is need for inoculation if the indigenous rhizobia are uninfective due to non-recognition of the host legume or the legume is not associating with the indigenous rhizobia because they are specific (Mfillinge *et al*., 2014). Depending on the cropping history of an area, symbiotically compatible rhizobia may or may not be present. The latter is common when a particular legume is newly introduced to an area. The specific bacteria to nodulate the introduced legume may not be present in the soil, especially if that legume had not been previously grown in the same field (CTAHR, 2014). In such case, there is need for introducing (inoculating with) a rhizobia strain that is compatible with the legume. Successful nodulation of a different legume, grown previously in the same field, does not ensure that the right rhizobia are present for the new crop (CTAHR, 2014).

Persistence of the existing rhizobia population is another factor to consider when in doubt of the need for inoculation. The persistence of rhizobia in the soil is affected by the soil type, pH, moisture and extreme temperature (IITA and N2Africa, 2014). Slattery and Pearce (2002) reported that poor rhizobia persistence was obtained in acidic soils indicating a need for re-inoculation of legumes when sowing into acidic soils. If the same legume was grown some years earlier with or without inoculation, the rhizobia population in the soil may no longer be large enough for good nodulation. Inoculation is recommended when the legume being planted has not been grown in that field in the past three years or with every planting of a high value crop (Pennstate Extension, 2015). CTAHR (2014) recommended that the best practice is simply to inoculate legume crops every season, particularly in climates with periods of drought or in acid or sandy soils conditions that can kill rhizobia between crop cycles.

Rhizobia strains differ in their ability to fix atmospheric nitrogen (Fall *et al*., 2003). There is need for inoculation when the indigenous rhizobia are ineffective (Bekunda *et al.,* 2010). A rhizobium strain can only be considered effective if it can fix nitrogen in the nodules of the host legume. The symbiotic effectiveness of a rhizobium strain is an estimation of host growth promotion and is usually based on the enhancement of plant shoot dry weight upon inoculation (Laranjo *et al*., 2014). Many soils are heavily infested with ineffective rhizobia capable of inducing nodulation without host benefit (Fening and Danso, 2002). Under such condition, inoculation with highly competitive and effective strain of rhizobia is needed to replace the ineffective native rhizobia. Inoculation is usually done to ensure the availability of the correct bacteria species and an effective strain of that species (CTAHR, 2014). Some rhizobia strains are capable of producing toxins which are harmful to the legume (Denison and Kiers, 2004). In such case, there is need to inoculate such legume or soil with more beneficial and highly competitive strain of rhizobium that could effectively compete for the nodule sites, enhance nitrogen fixation without adversely affecting the legume.

2.4.3 Benefits of rhizobia inoculation

Legume inoculation is a way of ensuring that the appropriate strain of rhizobium is present in the soil at the proper time and in numbers sufficient to assure a quick and effective infection and subsequent nitrogen fixation for the legume being planted (Mfillinge *et al.*, 2014). Increased nodulation as a result of rhizobia inoculation has been reported in several studies. Rudresh *et al.* (2005) reported increased nodulation in pea and chickpea as a result of rhizobia inoculation. Similarly, Huang and Erickson (2007) observed an increase in nodulation of inoculated pea and lentil as a result of rhizobia inoculation. Schweiger *et al.* (2012) reported that inoculation resulted in increased yield in soybean. Inoculation with effective rhizobia strains have also been reported to improve nitrogen fixation in many studies. Bambara and Ndakidemi (2010) reported that the amount of N-fixed per hectare were significantly increased with Rhizobia inoculation in *Phaseolus vulgaris*. Economic analysis by (Ndakidemi *et al*., 2006) on soybean and common bean in northern Tanzania showed that the increase in grain yield with inoculation translated into higher marginal rate of return and profitability for soybean and common bean. The authors stated further that with common bean, there was 66% increase in profit with inoculation in the Moshi district and 92% in the Rombo district relative to the control. With soybean, however, the increase in profit with inoculation was much larger, about 140% (Rombo) and 153% (Moshi).

The improved nodulation and nitrogen fixation in inoculated plant translated to improved growth and grain yield in many studies (Han and Lee, 2005; Zhou *et al.*, 2006; Elkoca *et al.*, 2007; Huang and Erickson, 2007; Bejandi *et al.*, 2011). Even where yield responses are not evident, inoculation may still have benefits by increasing seed N levels and N levels in plant residues (Vessey, 2004) thereby improving the quality of the crop and soil.

There are also reports on some rhizobia conferring increased resistance against plant pathogens (Avis *et al.*, 2008). Rhizobia may also act as non-symbiotic plant growth promoting bacteria (PGPB) as in the case of economically important non-legume crops such as rice or wheat, which are the best studied examples that benefit from rhizobia as endophytes (Biswas *et al.*, 2000; Chaintreuil *et al*., 2000). Inoculation with plant growth promoting rhizobia was reported to improve host plant growth and development in heavy metals contaminated soils by mitigating the toxic effects of heavy metals on plants (Zhuang *et al*., 2007). Han and Lee (2005) reported that rhizobia inoculation restricted sodium uptake by soya bean roots thereby suggesting that inoculation with rhizobia strains could alleviate salinity stress. Rhizobium has been reported to be key elements for plant establishment under drought and nutrient-unbalanced conditions (Requena, *et al*., 1996).

Inoculation with plant growth promoting rhizobia and arbuscular mycorrhizal fungi have also been used in several trials and have been reported to play significant roles in recycling of plant nutrients, maintenance of soil structure, detoxification of noxious chemicals and control of plant pests, reduction in the absorption of heavy metals by plant. (Filip, 2002; Horswell *et al*., 2006; Teng *et al*., 2010; Ismaiel *et al*., 2014).

2.4.4 Reasons for lack of response to rhizobia inoculation

To achieve a positive response to rhizobium inoculation, the introduced rhizobium must be infective, competitive and effective. Infectiveness is the ability of the rhizobia strain to form nodules with a particular legume while effectiveness is the ability of those nodules to fix nitrogen. The rhizobium must be able to compete with native rhizobia for the infection sites of the host legumes. Successful competition for nodule sites by native rhizobia is one of the reasons for the failure to achieve a response to inoculation with elite rhizobia strains (Theis *et al*., 1991). In the study carried out by Otieno *et al*. (2007), it was observed that inoculation with rhizobia increased number of nodulation and nodule weight but the increase in nodulation did not translate to increase in plant growth or grain yield in common bean, green gram, lima bean and lablab. The rhizobium strain used for the inoculation may be infective but not effective enough in bringing about the desired change.

Improper handling and failure to follow the recommended procedure for application are also reasons for failure in rhizobia inoculation. To record a positive response to rhizobia inoculation, the inoculum must be of good quality: prepared according to standard and viable. The bacteria in the carrier must be kept alive. The success of commercial inoculants is dependent on the number of viable bacteria available to participate in the infection process at the point of use (Catroux *et al*., 2001). To achieve this, the inoculant should be purchased from reputable dealers and properly handled. Proper handling of inoculant include keeping inoculant away from direct sunlight and heat. It should be kept cool but not frozen. The ideal storage temperature is between 4°and 26°C (CTAHR, 2014). Boonkerd (1991) reported that temperature was critical to the survival of soybean rhizobia in peat with substantial number at 10°C than at 30°C. Even if inoculant is used before the expiration date, it will not be effective if it has been improperly stored. However, Expired inoculant should not be used and inoculated seeds should not be stored for future use. Also, fertilizers or pesticides should not be mixed with inoculated seeds. Inoculated seeds should be planted almost immediately and furrows should be covered immediately after planting inoculated seeds to limit exposure to sunlight and heat as these reduce the effectiveness of inoculants (IITA and N2Africa, 2014).

Furthermore, to ensure positive response to rhizobia inoculation, the inoculant should be used in legume species for which the inoculant is effective and the inoculant should have the ability to multiply and survive in that soil type. Inoculants with multiple strains are usually effective over a wider range of legume species and soil conditions than inoculants with single rhizobia strain (CTAHR, 2014). The right quantity of inoculant per kg seed should be applied to ensure successful inoculation. The application rate is usually indicated on the label. However, it is recommended that at least, 5 g inoculant per kg seeds should be used (IITA and N2Africa, 2014). Theis *et al*. (1991) reported that inoculant rhizobia must be applied at a rate at least 1000 times greater than the estimated number of indigenous rhizobia.

Method of inoculation may enhance response to inoculation. Fening and Danso (2001) stated that method of rhizobia inoculation enhanced nodulation response and hence N fixation. The major methods of applying inoculants are seed inoculation (direct inoculation, where the inoculant is placed in direct contact with the seed) and soil inoculation (indirect inoculation, where by the inoculant is placed alongside or beneath the seed). Dusting, slurry and seed pelleting are the three major technique of applying inoculant to the seed (IITA and N2Africa, 2014).

Dusting method involves the use of inoculant powder which is mixed directly with seed without using any water or other liquid. The dry inoculation method is often considered disadvantageous because the attachment of the inoculum to the seed is poor and much of it is lost prior to and during sowing. The rhizobia also have little protection when this method is used and therefore their survival is poor compared to the other methods (IITA and N2Africa, 2014). The slurry method involves initial mixing of the inoculant with water to form a uniform, pourable suspension. The slurry obtained is then added to the seeds and mixed in a bag or plastic bucket. In some instances sticker are used to ensure better adhesion of the bacteria to the seeds during the planting process. Popular sticker materials include gum arabic (40% in hot water), widely used in the Middle East and North Africa, carboxymethyl cellulose (4% in water), used most frequently in Australia, sugar (10% in water), corn syrup (10% in water), honey (10% in water), powdered milk (10% in water), evaporated milk (20% in water), mineral oil, or a vegetable oil such as peanut oil or soybean oil (CTAHR, 2014). Tests on soybean have shown that all of these can stick more than 100,000 live rhizobia to each legume seed, which is enough for good nodulation and nitrogen fixation (CTAHR, 2014). These stickers have better advantage over water because water is quickly absorbed by the seed and the inoculant can then blow away during planting, however, the sticker should not contain any substances that are harmful to the rhizobia or the seed (CTAHR, 2014).

In the seed pelleting methods, inoculants can be made into slurry and mixed with the seeds. The seeds are then coated with finely ground lime, clay, rock phosphate, charcoal, dolomite, calcium carbonate or talc, depending on soil conditions and plant needs. The method has several advantages, such as protection of rhizobia against low soil pH, desiccation, acidic fertilizers, fungicides or insecticides (IITA and N2Africa, 2014). In the experiment carried out by Afzal *et al*. (2012), it was reported that the method of inoculation affects colonization and performance of the inoculum strains in the phytoremediation of soil contaminated with diesel oil. Soil inoculation was reported to give the best result. Similarly, Fening and Danso (2001) reported that soil inoculation gave better result than seed inoculation in their experiment. The choice of methods for seed and soil inoculation depends on materials available, the climatic and soil conditions. Seed inoculation is an easy and convenient way of putting the rhizobia in the root zone of the developing seedling where infection of the root hairs can occur and nodules develop. Seed inoculation is cost effective than soil inoculation because the latter requires more inoculant. However, soil inoculation is usually recommended under the following conditions: when seeds are heavily pre coated with pesticides or herbicides, when planting in hot, dry soil (if legume seeds are planted in hot, dry soil and must wait for rain before they germinate, the rhizobia used to coat them are likely to die. Under these conditions, the rhizobia will survive better if the inoculant is placed in the soil below the seeds using granules), when seed inoculation has failed and when large numbers of rhizobia are needed (IITA and N2Africa, 2014).

The quality of the native rhizobia can affect plants response to inoculation (Date, 2001). A higher population of symbiotically competitive indigenous rhizobia will have an advantage over the introduced strains because it is already adapted to the conditions of the area. Theis *et al*. (1991) reported that the response to inoculation and competitive success of inoculant rhizobia are inversely related to number of indigenous rhizobia. Slattery and Pearce (2002) in their report stated that where there are low (<50 Rhizobium bacteria g/m soil) naturalized populations of rhizobia specific to a target legume, the introduction of new strains by seed inoculation is normally successful. On the other hand, inoculation into soils where naturalized rhizobia population is high (>10^3 Rhizobium bacteria g/m soil) introduction of new strains can be difficult and often unsuccessful.

2.5 Effect of Rhizobia Inoculation on Physiology, Growth and Yield of Legumes

Physiological processes in plants lead to growth and eventually yield. Leaf photosynthesis and rhizobia nitrogen fixation are the two main physiological processes of utmost importance to legume growth and development (Vollmann *et al*., 2011). The determination of chlorophyll content in plant is very important as this can be used to predict a lot of traits in physiological studies. Measuring leaf chlorophyll content can provide information on the nodulation and nitrogen fixation status of crop plants (Vollmann *et al.*, 2011). The chlorophyll content in leaves reflects photosynthetic activity and yield potential of plants. High chlorophyll content in different plant leaves was considered a favorable trait in crop production. Several studies have shown that leaf chlorophyll content is positively correlated with photosynthetic capacity and growth of plant (Teng *et al*., 2004; Rong-hua, 2006). Vollman *et al*. (2011) reported that chlorophyll content were correlated to 1000-seed weight, seed protein and seed oil content. Increase in plant chlorophyll and other physiological parameters as a result of rhizobia inoculation has been reported by a number of researchers. Bambara and Ndakidemi (2009) reported that rhizobia inoculation had significant effects on the leaf chlorophyll content, the photosynthesis, the intercellular CO_2 concentration and the transpiration rate of *Phaseolus vulgaris* L. Similarly, seed inoculation with *Bradyrhizobium japonicum* was reported to increase plant biomass and leaf chlorophyll content in soybean (Saadpanah *et al*., 1997). Zhou *et al*. (2006) obtained positive effects on plant photosynthesis and plant growth after rhizobia inoculation in soybean. Han and Lee (2005) obtained an increase in photosynthetic rate and nutrient uptake in soya bean as a result of inoculation with *Bradyrhizobium japonicum.*

The work of Vollman *et al*. (2011) revealed that nodulation affects chlorophyll content and that leaf size, plant height, number of pods per plant, 1000-seed weight, seed protein and oil content were also affected by nodulation. Bejandi *et al*. (2012) reported that seed inoculation with rhizobia had significant effects on nodulation, emergence percentage, time to maturity, chlorophyll content, plant height, seed protein, pods per plant, hollow pod and grain yield. The authors stated further that the inoculated plants had the highest values of the aforementioned traits (except time to maturity and hollow pods percentage) compared to plants that received fertilizer and the control. Namvar *et al.* (2011b) reported that inoculation with rhizobium improved growth indices including total dry matter, leaf area index, crop growth rate, relative growth rate, and net assimilation rate in chicken pea. Ismaiel *et al.* (2014) reported that inoculation of *Vicia faba* with Rhizobium and Arbuscular mycorrhyza (AM) fungi significantly enhanced the photosynthetic capacity by increasing the chlorophyll a and chlorophyll b contents of the plant over uninoculated control plants. The authors also observed that the highest total chlorophyll content was obtained at the flowering stage. In the experiment carried out by Ravikumar (2012) to find out the impact of artificial inoculation with *Rhizobium japonicum* in charcoal carrier on *Vigna mungo* and *Vigna radiata,* it was reported that the inoculated plants possessed higher height, fresh weight, number of roots, nodules, number of leaves, shoots, pods, length of pods, and seed weight, over the controls. Han and Lee (2005) obtained an increase in leaf area and total dry weight of inoculated soybean thereby leading to the increased plant growth observed in inoculated plants. Namvar *et al*. (2011a) reported that inoculation increased the plant height, number of primary and secondary branches, number of pods per plant and number of grains per plant in chickpea. In the experiment carried out by Huang and Erickson (2007) on pea and lentil, it was observed that shoot/root growth, seed yield of pea improved as a result of rhizobia inoculation and also, seedling height, nodule

mass and shoot biomass of lentil increased. The inoculation of seeds with rhizobium was reported to increase nitrogen accumulation and yield in soyabean (Sogut, 2006). Clayton *et al*. (2004) found *Rhizobium* inoculant to increase plant nitrogen content from 19 to 42 mg per plant in pea.
Increase in grain yield by 40% was reported by Osunde *et al*. (2003) in soya bean inoculated with rhizobium compared to the uninoculated. Arumugam *et al*. (2010) reported that plants inoculated with Rhizobium had significantly higher chlorophyll, dry weight of shoot and root when compared to the control. Taiwo and Oladapo (2000) also reported that single or dual inoculation of cowpea with bradyrhizobium increased percentage nitrogen and phosphorus in plant tissue as well as the percent N derived from the atmosphere when compared with uninoculated control plants. Recent research have shown that rhizobia may promote plant growth through mechanisms other than nitrogen fixation (Chernin and Glick, 2012). For example, the presence of 1-aminocyclopropane-1-carboxylic (ACC) deaminase activity in some rhizobia strains promotes plant growth through lowering of plant ethylene levels (Duan *et al.*, 2009).

2.5.1 Responses of cowpea to rhizobia inoculation

Nitrogen fixation has been acclaimed to be low in cowpea compared to some other legumes (Ulzen, 2014). In the experiment carried out by Fening *et al*. (2001) to assess the potential of improving N fixation in cowpea in Ghananian soils, it was reported that all the 45 cowpea cultivars used showed significant response to increasing N fertilizer application to a particular level, indicating that N fixation was not providing the plants with sufficient N for maximum growth and yield, hence indicating the need for inoculation in cowpea to improve the N fixed. The authors stated further that response of cowpea to N fertilization differed according to soil type.

Cowpea usually form symbiosis with rhizobia of the genus *Bradyrhizobium* (Vessey, 2004). Cowpea rhizobia were first classified in a heterogeneous group of slow-growing rhizobia nodulating promiscuous tropical and subtropical legume species known as 'cowpea cross-inoculation group. They were later transferred to the genus *Bradyrhizobium*. Currently this genus contains three named species, *B. japonicum* (groups I and Ia), *B. elkanii* and *B. liaoningense* (Krasova-Wade *et al*., 2006). There has been low response of cowpea to inoculation probably due to high incidence of cowpea bradyrhizobia in most tropical soils and the promiscuous nature of cowpea (Fening and Danso, 2002). Cowpea appears to be the most promiscuous legume which has been intensively studied, nodulating with a wide range of fast and slow growing rhizobia (Mpepereki *et al*., 2000).

Fening *et al*. (2001) reported that most soils in Ghana contained large population of indigenous bradyrhizobia capable of nodulating cowpea. The authors reported further that the 45 cowpea cultivars used in their study nodulated naturally in the various soil used. In the experiment conducted by Pule-Meulenberg *et al.* (2010) to examine the symbiotic functioning and biodiversity of cowpea bradyrhizobia in Africa, it was discovered that cowpea genotypes differed significantly in growth, N content and percentage nitrogen derived from the atmosphere (%NDFA) and that the 18 bradyrhizobia strains isolated differed significantly in their N_2 fixing efficiency. In a greenhouse experiment conducted by Aliyu *et al*. (2013) to determine the response of grain legumes to rhizobia inoculation in two savanna soils of Nigeria (Eutric Cambisols and Rhodic Nitisols), it was reported that cowpea did not respond to rhizobia inoculation. The authors attributed the poor response to the high rhizobia population density in the experimental soils and the quality of the inoculant used stating that the peat inoculants used in the trials might have been in storage for months and in an erratic temperature conditions which could have affected the bacterial number and hence viability. The authors added that low soil pH and other nutrient status could have aggravated the situation.

However, some positive responses to inoculation has been recorded in cowpea. In the experiment carried out by Fening and Danso (2001) to determine the response of cowpea to inoculation with indigenous bradyrhizobium strains, it was observed that cowpea responded positively to rhizobia inoculation with an increase in nodulation, shoot dry weight and nitrogen accretion in inoculated cowpea. Similarly, Nyoki and Ndakidemi (2014) reported that rhizobia inoculation in cowpea significantly improved the plant height in both screen house and field experiments relative to the control treatment. Arumugam *et al.* (2010) reported that cowpea plants inoculated with rhizobium had significantly higher shoot and root length, dry weight of shoot and root, total number of nodules and dry weight of nodules when compared to the control plants. Sarker *et al.* (2001) also obtained an increase in cowpea grain yield as a result of rhizobia inoculation.

Taiwo and Oladapo (2000) reported that single or dual inoculation of cowpea with bradyrhizobium (mixture of IRj2184A and IRc 256 strains) and Arbuscular Mycorrhiza increased plant height, number of nodules and weight, dry matter accumulation of shoot and %NDFA when compared with the uninoculated. The authors further reported that cowpea grain yield was enhanced by 20% due to dual inoculation in the first year. Improved nitrogen fixation and yield is obtainable in cowpea if it forms symbiosis with highly effective strains of rhizobium. Fening *et al*. (2001) stated that inoculation of cowpea with effective strains of bradyrhizobia species has considerable potential to improve nitrogen fixation in the crop. Hence, inoculation with good quality (viable), highly competitive and effective rhizobia strains is needed in cowpea.

2.6 Effect of Nutrient Management on the Physiology, Nodulation, N Fixation, Growth and Yield of Legumes

Mineral nutrients may influence N_2 fixation in legumes at various levels of the symbiotic interactions: infection and nodule development, nodule function, and host plant growth (O'Hara, 2001). Accumulation of sufficient N by legume and subsequent yield depend on the number, efficiency and compactibility of rhizobia and nutrient constraints that affect nodulation and nitrogen fixation (Aynabeba *et al*., 2001). Among all the essential nutrients needed for crop growth, nitrogen, phosphorus and potassium are needed in relatively large amount. The most important role of nitrogen in the plant is its presence in the structure of protein and nucleic acids, which are the most important building and information substances of every cell (Namvar *et al*., 2013). Nitrogen is a major part of the chlorophyll molecules and plays a necessary role in photosynthesis (Eutropia and Ndakemi, 2013). Nitrogen supply to a plant will influence the amount of protein, amino acids, protoplasm and chlorophyll formed. Moreover, it influences cell size, leaf area and photosynthetic activity (Namvar *et al.*, 2013). Hokmalipour and Darbandi (2011) reported that chlorophyll content increased as N application levels increased. There are a few report on positive effects of low nitrate concentrations on N_2 fixation in legume species. Abayomi *et al*. (2008) observed that though cowpea symbiotically fixes nitrogen, plant dependent on symbiotically fixed N alone may as well suffer from temporary N deficiency during the seedling growth once the cotyledonary reserves have been exhausted.

Namvar *et al.* (2011a) reported that application of suitable amounts of nitrogen fertilizer as a starter can be beneficial in improving growth, development and total yield of inoculated chickpea. Zhou *et al*. (2006) also reported that in soyabean plant treated with 5 mM $(NH_4)_2SO_4$, plant biomass, leaf area and chlorophyll content, net photosynthetic rate, stomatal conductance (gs), carboxylation efficiency (CE), maximum photochemical efficiency (Fv/Fm) of photosystem 2 (PS2), and quantum yield of PS2 (ΦPS2) were markedly improved as compared with the control plants. Namvar *et al.* (2011b) reported that total dry matter, leaf area index, crop growth rate, relative growth rate, net assimilation rate, plant height, number of primary and secondary branches, number of pods per chick pea plant and number of grains per pod were significantly affected by application of nitrogen fertilizer. The authors reported further that the highest grain yield in chicken pea was obtained in the inoculated plants that were treated with 75 kg urea ha^{-1}. Application of mineral N was reported to increase shoot dry matter yield of grain legumes (Moawad and Shamseldin, 2010). However, negative impact of nitrogen on nodulation has been reported by many researchers (Laws and Graves, 2005; Cheming'wa *et al.,* 2007; Otieno *et al.*, 2007). Alleviation of N problem through rhizobium inoculants is the best alternative in promoting legume productivity in Africa (Bambara and Ndakidemi, 2010).

Phosphorus is needed in relatively large amounts by legumes for growth and optimal symbiotic performance (Mfillinge *et al*., 2014). It functions as a constituent of nucleic acids and proteins. It is important in cell division and induces root growth, promotes seed formation and increases disease resistance and has also been reported to promote leaf area, biomass, yield, number of nodules and nodule mass in different legumes (Kamiti, 2011; Mfillinge *et al*., 2014). High phosphorus supply is needed for nodulation (Elkoca *et al*., 2007) as nodules themselves are strong sink for P; it was observed that the phosphorus content per unit dry weight is considerably higher in the nodules than in the roots and shoots, particularly at low external phosphorus supply (Weisany *et al.*, 2013). When legumes depend on symbiotic nitrogen and receive an inadequate supply of phosphorus, they may suffer from nitrogen deficiency (Weisany *et al*., 2013). Phosphorus is known to promote early root formation and the formation of lateral, fibrous and healthy roots, which play an important role in N_2 fixation, nutrient and water uptake (Bhuiyan *et al*., 2008; Niu *et al*., 2012).

Low soil P delays root emergence, lowers root hair numbers, affects root morphology and physiological characteristics that are important for N uptake (Kamiti, 2011). There are marked differences in rhizobia and plant requirements for P with the slow- growing rhizobia more tolerant to low P than the fast growing (Weisany *et al.,* 2013). Nitrogen fixing plants have an increased requirement for P over those receiving direct nitrogen fertilization, probably due to the need for nodule development and signal transduction, and phospholipids in the large number of bacterioids (Graham and Vance, 2000). Amjad *et al.* (2004) found that vine length, number of pods per plant, pod length, number of grains pod^{-1} and green pod yield of pea were significantly affected by the levels of P_20_5 and that number of grains pod^{-1} and green pod yield were maximum at the highest dose of P_20_5 (69 kg ha^{-1}). Phosphorus fertilizer was reported to have positive effects on N and P accumulation by cowpea lines (Kolawole *et al*., 2002). Magani and Kuchinda (2009) reported an increase in seed crude protein content with P application and therefore recommended application of 37.5 kg P $ha\text{-}^1$ in the Nigeria Northern Guinea Savanna. Ndakidemi *et al*. (2006) reported that grain yield as a result rhizobia inoculation in soybean and common bean was further improved with P supplementation. The authors reported further that with provision of supplemental P (26 kg P $ha\text{-}^1$), profit margins rose to 84% in Moshi and 102% in Rombo district of Tanzania. Ntare *et al*. (1993) reported that application of P increased seed yield by increasing crop growth rate. Jemo *et al*. (2006) reported that application of mineral P increased shoot dry matter yield of grain legumes subsequent maize grown on acid soils.

Potassium (K) is needed for the process of respiration as it encourages the plant stomata opening and energy production for photosynthesis process (Abdul-Hamid *et al*., 2009). Efficient cell development and growth of plant tissues, translocation and storage of assimilate and other internal functions which are based upon many physiological, biochemical and biophysical interaction require adequate K in the cell sap (Marschner, 2012). Imas and Magen (2007) studied the effect of K on soybean and concluded that plants growing at higher K level have better development of nodules and consequently higher N-fixation. In the study conducted by Kurdali *et al.* (2002) to determine the effect of soil moisture and potassium fertilizer on nodulation, dry matter production, and N_2 fixation by faba bean and chickpea, it was observed that the higher level of K fertilizer increased both dry matter production and total N_2 fixed in faba bean. Similarly, there was a corresponding increase in nitrogen fixation and its attributes with the increase in the potassium level in chick pea (Singh and Kataria, 2012).

Calcium deficiency was reported to limit nitrogen fixation in *Trifolium subterraneum Glycine Max* and *Medicago sativa* (Weisany *et al.*, 2013). Nutrient limitations in legume production result from deficiencies of not only major nutrients but also micronutrients such as molybdenum (Mo), zinc (Zn), boron (B) and iron (Fe) (Bejandi *et al*., 2012). Weisany *et al.* (2013) extensively reviewed the roles of micronutrients in rhizobia-legume symbiosis and reported that Mo, S, Cu, Fe and Co play significant roles in nitrogen fixation. Molybdenum is a micronutrient specifically for plants that form root nodules with nitrogen-fixing bacteria, though plants that do not form nodules also use trace amounts of it in nitrogen metabolism and uptake (Weisany *et al.*, 2013). It is important for chlorophyll synthesis in plant (Bambara and Ndakidemi, 2009). Its application was reported to have significant effects on the leaf chlorophyll content, the photosynthesis, the intercellular CO_2 concentration and the transpiration rate of *Phaseolus vulgaris* L. (Bambara and Ndakidemi, 2009). The nitrogen fixing enzyme, nitrogenase is composed of molybdenum and iron and without adequate quantities of these elements, nitrogen fixation can't occur (Weisany *et al.*, 2013). Zn is also an important micronutrient in legume production and its deficiency was reported to reduce the number and size of nodules (Desta *et al.*, 2013). The authors also reported that Zn and P application improved grain yield and grain uptake of N, P and Zn in faba bean as well as root distribution, nodule mass, concentration of N, P and Zn in nodule, root and leaf tissue at 50% flowering and grain yield. Bejandi *et al*. (2102) reported that the application of Fe, Zn, Mn and B enhanced the grain yield of chickpea and that the interaction between the micronutrients and inoculation yielded better result. Microbial inoculants constitute an important component of integrated nutrient management that leads to sustainable agriculture (Mfillinge *et al.*, 2014). Soils varying in soil fertility status will respond differently to rhizobia inoculation. Therefore research efforts on effective management of soil fertility variability are required to derive maximum benefits from inoculation (Aliyu *et al*., 2013).

Use of manure in legume production has also been documented. For example, Otieno *et al*. (2007) reported that manure application increased nodulation and grain yield during the short rains in common bean, green gram, lima bean and lablab. In the experiment carried out by Adeoye *et al.* (2011) to determine the effect of poultry and cattle manure on the growth and yield of TVX-3236 variety of cowpea, it was observed that the highest grain yield (854 kg ha^{-1}) was obtained from the plot treated with poultry manure and the lowest (310 kg ha^{-1}) from cattle dung and 334 kg ha^{-1} from the control. Omotosho (2014) also reported that application of organic manure and inorganic fertilizer increased the number of nodules, dry matter and number of pods per plant, number of seeds per pod and 100 seed weight in cowpea.

2.6.1 Nutrient management in cowpea

Cowpea is capable of performing well without fertilizer if the soil is fertile but most tropical soils are deficient in one or more major nutrients. In soils of low fertility, it responds to phosphorus and potash and often some nitrogen; up to 10 kg ha^{-1} of nitrogen and 40 to 70 kg ha^{-1} P_2O_5 and K_2O and calcium may be needed where the pH is low (FAO, 2006). Use of P in cowpea production has been well documented. Nkaa *et al*. (2014) reported that application of P significantly increased plant height, leaf area, number of leaves and number of branches, number and weight of nodules, total above ground dry matter, and seed yield in all cowpea varieties used. The authors recommended an application rate of 40 kg P ha-^{1}. Kamiti (2011) reported that cowpea growth can be improved by addition of inorganic P and N at a rate of 13-25 kg ha^{-1} and 10-15 kg ha^{-1} respectively. Sarker *et al.* (2001) stated that application of P_2O_5, K_2O, S fertilizer at the rate of 50, 30 and 20 kg ha^{-1} respectively improved cowpea seed yield and that the best performance of cowpea was obtained when the use of chemical fertilizer was combined with inoculants. Application of N P K fertilizer was reported to enhance early vegetative growth of cowpea (Abayomi *et al*., 2008). The authors reported that application of 30 kg N, 15 kg P_2O_5 and 15 K_2O ha-^{1} increased the plant height, number of leaves, reduced the number of days to flowering, nodule production, yield components and grain yield in cowpea.

Kolawole *et al.* (2002) reported that application of 30 kg P ha-^{1} had positive effects on shoot, grain, and root dry weights of cowpea varieties. The authors further reported that there were significant interactions between cowpea varieties and P levels on nodulation, shoot, root and grain weights and that variability among cowpea lines in shoot, grain and root dry weight response to P was more pronounced for shoots than roots.

2.7 Effect of Cropping System on the Productivity of Cowpea in Nigeria

Cropping system is an important management practice used by agriculturist to improve soil fertility, reduce pest and diseases and improve crop productivity. It is the management, pattern and sequence in which crops are arranged on a piece of land. The traditional cropping system used in cowpea production in Nigeria is not delivering the potential yield of the crop. The potential yield of cowpea is 2-3 t ha^{-1} but the yield obtained on farmers' field is averaged at 450 kg ha^{-1} which is 15-22% of the potential yield (Omotosho, 2014). Most of the farmers in Nigeria savannas plant cowpea in relay cropping with cereal or intercrop with cereals thereby limiting the availability of water, nutrient, space and light to cowpea and thus limiting the productivity of the crop. Cowpea is hardly planted as a major crop by the poor resource farmers who produce 90% of the cowpea in Nigeria. Researchers have reported that sole-cropping of cowpea produced significantly higher yield than when intercropped (Kouyat´e *et al*., 2012; Mbah, 2018). Kouyat´e *et al*. (2012) in his report stated that cropping system significantly affected nodulation and grain yield of cowpea with sole-crop of cowpea in rotation with millet producing the highest grain yield compared with the intercrop. The authors attributed this to shading effect of millet on cowpea as well as competition between the two plants for space, water, light, and nutrients when intercropped. Sequential cropping system otherwise known as double-cropping or triple cropping (depending on the number of sequence completed within a growing season) has numerous advantages in improving the yield of crop per unit area of land, increased profitability (Mbah, 2018), increased soil quality by preventing erosion, and more intensive use of land, labour and capital. Thus adopting Cowpea sequential cropping system (double cropping of cowpea) could be a strategy to increase the productivity of the crop per unit area to meet the growing demand in the face of growing population and urbanization.

However, when adopting sequential cropping system, the environmental factors such as temperature, rainfall, day length, wind, and non-climate factors such as pests, diseases, weeds, birds, economy of production must be such that could sustain the production of the crop in sequence hence it is important to select good crop varieties that will be able to complete as many life cycle as possible within the growing season considering all these factors.

CHAPTER THREE

3.0 MATERIALS AND METHODS

3.1 Experiment 1- Assessing the Need for Rhizobia Inoculation in Cowpea

3.1.1 Study location- This is a pot experiment carried out in the screen house of the School of Agriculture and Agricultural Technology, Federal University of Technology, Minna, Niger State.

3.1.2 Sources of seeds and rhizobia inoculants- Seeds of cowpea and rhizobia inoculants were obtained from the International Institute of Tropical Agriculture (IITA), Kano and Ibadan stations respectively.

3.1.3 Soil collection from different locations

Soils samples were collected from a total of 20 farmers' fields consisting of five fields each in Niger, Kaduna, Kano as well and the Federal Capital Territory, all in Nigerian Savanna (Figure 3.1). In each field, twenty soil samples were taken randomly from the depth of 0-20 cm and later bulked and mixed thoroughly to form a composite sample.

3.1.3.1 Soil analysis

The soil samples collected were subjected to routine physical and chemical analyses following the procedures of Okalebo *et al.* (1993). Soil particle size distribution was determined using the hydrometer method with calgon as a dispersing agent and the soils' textural classes were determined using the soil textural triangle. The soil pH in water and calcium chloride was determined using electrometric method as described by Okalebo *et al*. (1993).

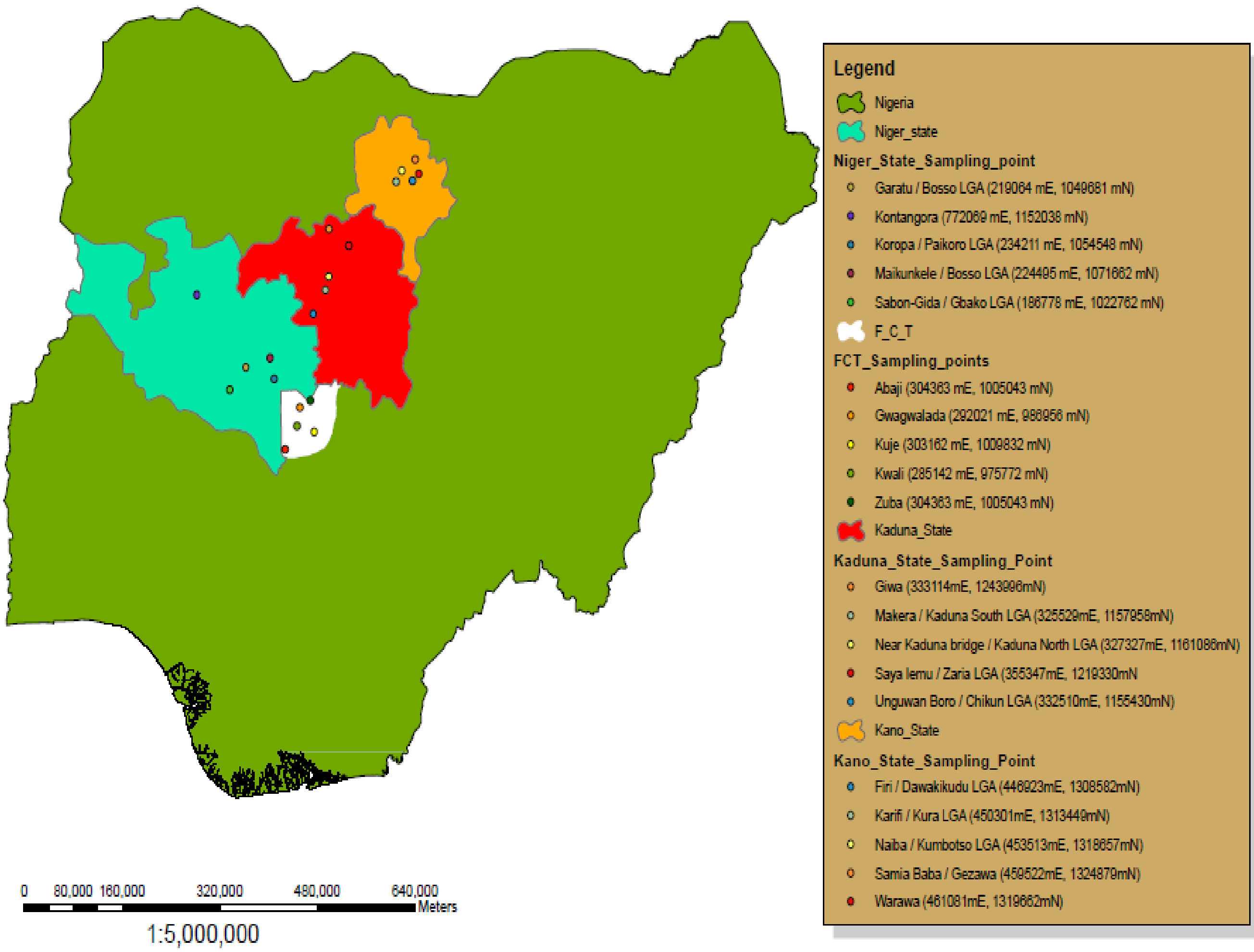


Figure 3.1: Map of Nigeria showing the soil sampling locations

Source: From this study

Organic carbon was determined by Walkley and Black's wet oxidation method. Total nitrogen was determined by the micro Kjeldahl's oxidation method involving digestion and distillation. The readily acid–soluble forms of phosphorus was extracted with hydrochloric acid and ammonium fluoride mixture (Bray No. 2 method). The phosphorus in the extract was determined by colorimetric method. Exchangeable cations were extracted with ammonium acetate solution. Potassium and sodium were read using flame photometer while exchangeable magnesium and calcium were read using atomic absorption spectrophotometer (Sparks, 1996).

3.1.4 Treatments and experimental design

This was a 20 x 2 x 4 factorial experiment consisting of soils collected from 20 farm locations, two varieties of cowpea and four N sources. The two cowpea varieties were: IT93K-452-1 (SAMPEA 8) (an extra early maturing variety, with tolerance to major insect pest) and IT99K-573-1-1 (SAMPEA 14) (an early maturing variety, with tolerance to drought, striga and alectra). The four N sources were Control (no rhizobia inoculation and no fertilizer), inoculation with USDA 3384 rhizobium strain, inoculation with USDA 3451 rhizobium strain and application of 90 kg Nha^{-1} (using urea as the source). These treatments were laid out in a completely randomized design with three replicates.

3.1.5 Pot filling

Perforated poly bags were filled with 2.5 kg top soil collected from each farm after they were thoroughly mixed and sieved using a 5 mm mesh. Stones and other debris were removed.

3.1.6 Inoculant and fertilizer application

The inoculants were applied at the rate of 5 g per kg seed using the slurry method which was done by coating the seeds with a sticker (solution of 85 ml water and 15 g sugar) before applying the inoculant to the coated seeds (IITA and N2Africa, 2014). All the pots received basal application of 60 kg P and 60 kg K ha^{-1} at planting using single super phosphate and muriate of potash as the source, respectively. The N was applied at the rate of 90 kg ha^{-1} to pots that received N treatments. Twenty five percent of the N was applied at planting while the remaining 75% was applied at three weeks after planting as described by Woomer *et al.* (2011).

3.1.7 Planting

Three seeds were sown per pot and the resultant seedlings were later thinned to two per pot at ten days after planting.

3.1.8 Data collection

Data were collected for the under listed parameters.

3.1.8.1 Number of nodules per plant and nodule weight (g/plant)

At 7 weeks after planting, soil was carefully washed off the plant roots and the nodules were removed from the root, counted and weighed on a Mettler balance. The average nodule weight and number per plant were determined by dividing the total values obtained by the number of plants per pot.

3.1.8.2 Dry weight

The harvested shoots were oven dried at 65 ^{o}C in paper bags until constant weight was attained.

3.1.9 Data analysis

All the data collected were subjected to analysis of variance using the statistical analysis system (SAS) version 9.1. Treatment means were separated using Waller-Duncan K-ratio T-test at 5% level of probability. Genotype plus genotype × environment interaction (GGE) analysis was carried out using the breeding management system (BMS, 2015) using the following model:

$\bar{y}_{ij} - \mu_j = \sum_{k=1}^{t} \lambda_k \alpha_{ik} \gamma_{jk} + \bar{\varepsilon}_{ij}$

Where $\bar{y}_{ij}$ is the cell mean of genotype (rhizobia strain) i in environment j; μj is the mean value in environment j; $i = 1, \cdots g$; $j = 1, \cdots e$, g and e being the numbers of genotypes and environments, respectively; and t is the number of principal components (PC) used or retained in the model, with $t \leq \min(e, g - 1)$. The model is subject to the constraint $\lambda_1 \geq \lambda_2 \geq \cdots \lambda_t \geq 0$ and to orthonormality constraints on the α_{ik} scores, with similar constraints on the γ_{jk} scores [defined by replacing symbols (i,g,α) with (j,e,γ)]. The ε_{ij} are assumed normally and independently distributed $(0, \sigma^2/r)$, where r is the number of replications within an environment.

3.2 Experiment 2- Physiological Responses of Cowpea Varieties to Phosphorus Application and Rhizobia Inoculation

3.2.1 Experimental site

The experiment was conducted on three farmers' plots in Minna, Niger State but was researcher managed. The coordinates of the farms are 09º 27.832´ N 006º 25.375´ E, 09º 31.203´N 006º 27.678´ E and 09º 28.026´ N 006º 25.325´ E in the Southern Guinea Savanna agro-ecological zone of Nigeria.

3.2.2 Soil sampling /analysis

Seven soil samples were taken randomly from each plot at a depth of 0-20 cm using an auger. The soil samples were bulked and thoroughly mixed to obtain composite samples. The samples were sieved with a 2 mm mesh sieve to remove stones and other debris. Soil physical and chemical analyses were carried out before planting following the procedures described in Experiment 1.

3.2.3 Treatments and experimental design

Three factors were evaluated viz: rhizobia inoculation, phosphorus (P) and cowpea varieties. Inoculation was at three levels in 2015 namely: control, inoculation with USDA 3384 and USDA 3451 rhizobia strains. Due to lack of response to the USDA strains, in 2016, the inoculation treatments were replaced with BR 3262, BR 3267 rhizobia strains and 90 kg N ha^{-1} was added which were repeated in 2017 with the addition of USDA 3451. In the three years, P was applied at three levels: control (no P application), 20 kg P ha^{-1} and 40 kg P ha^{-1} while IT93K-452-1, IT99K-573-1-1 and TVX-3236 cowpea varieties were used. Simultaneously, maize (the reference crop) was planted in between the cowpea plots and they equally received the various P treatments.

These treatments were factorially combined and arranged in a randomized complete block design.

IT93K-452-1 also known as SAMPEA 8 is an extra early maturing variety (55-60 days), semi-erect, white medium seeds with black hilum, with tolerance to major insect pests. IT99K-573-1-1 also known as SAMPEA 14 is an early maturing variety (60-65 days), semi-erect, white seeds with brown hilum, drought tolerant, striga and alectra resistant. TVX-3236 is a medium maturing (75-80 days) variety, semi-erect, cream with brown mottled seeds and with strip resistance. These varieties were developed by the International Institute for Tropical Agriculture, Ibadan, Nigeria.

3.2.4 Sources of seeds and inoculants

The same as in Experiment 1.

3.2.5 Land preparation and plot size

The plots were cleared and ridged at a spacing of 75 cm apart. Each plot was 4 m long and 3.75 m wide and there were 5 ridges per plot.

3.2.6 Inoculant application- The same as in experiment 1.

3.2.7 Planting

Three seeds were sown per hole on ridges at an intra-row spacing of 20 cm and the seedlings were later thinned to two per stand at ten days after planting.

3.2.8 Fertilizer application –Basal application of 40 kg K_2O ha^{-1} was done at planting using muriate of potash. Similarly, P was applied to the plots receiving P treatment (20 and 40 kg P ha^{-1}) at planting using single super phosphate and 20 kg N ha^{-1} was applied at 3 weeks after planting using urea.

3.2.9 Insecticide application- Magic force (15 g Lambda-cyhalothrine + 300 g Dimethoate) a contact and systemic insecticide with broad spectrum of action was applied at the rate of 0.3 kg ai (active ingredient) per ha to control insect pests.

3.3. Weeding- Weeding was done manually at 3 and 6 weeks after planting.

3.4 Data Collection

Six plants were randomly selected from the three inner ridges (net plot) and tagged for data collection. Untagged plants were used for dry matter determination. Data on the six plants were taken and the means were computed and recorded for the under-listed parameters:

3.4.1 Growth and physiological parameters

i. Vine length (cm) - This was done by the use of a meter rule to determine the length from the base of the plant to the tip of the longest vine.

ii. Number of leaves - Fully expanded leaves were counted on each tagged plant.

iii. Leaf area (cm^2) - Three fully expanded leaves per plant were selected at random for the determination of leaf area. The length and breadth of the leaflets were measured and the product was multiplied by a factor of 0.75 as described by Musa and Usman (2016).

iv. Stem diameter (cm) - The diameter of the main stem was measured at the base using a pair of Venier calipers.

v. Days to onset of flowering and 50% flowering – These were determined by observing the number of days to when the first flower was sighted in each plot and when 50% of the plant population flowered respectively.

vi. Number of nodules per plant and nodule weight (g/plant) – At 50% flowering, plants within 1 meter were carefully uprooted with a spade for nodule count and weight as described in Experiment 1.

vii. Crop growth rate (CGR) - This was calculated using the formula:

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

where W_1 and W_2 are total weight at time T_1 and T_2 respectively (Das, 2011).

viii. Net assimilation rate- This was calculated using the formula:

$$\text{NAR (g/}m^2\text{/day)} = \frac{W_2 - W_1}{T_2 - T_1} X \frac{\log_e L_2 - \log_e L_1}{L_2 - L_1}$$

where W_1, W_2 and T_1, T_2 are the same as in CGR above, L_1 and L_2 are leaf area at time T_1 and T_2 respectively (Das, 2011).

ix. Leaf area index- This was computed as the ratio of the leaf area to the area of ground covered using the formula: $\text{LAI} = \frac{\text{Leaf area of plant } (cm^2)}{\text{Land covered by plant } (cm^2)}$

x. Leaf area ratio (cm²/g) – This is the ratio of the total leaf area to the whole plant dry weight. It was computed using the formula: $\text{LAR} = \frac{\frac{\text{LA1}}{\text{W1}} + \frac{\text{LA2}}{\text{W2}}}{2}$

Where LA_1 and LA_2 are leaf areas and W_1 and W_2 are whole plant dry weights at the two sampling period (Das, 2011).

xi. Chlorophyll- At 50% flowering, six leaflets were picked at random from each tagged plant for chlorophyll determination. Chlorophyll a, b and total chlorophyll in leaves were extracted using 80% acetone. Their absorbance were read with a spectrophotometer at two wavelengths and the following calculations were made:

$$\text{Chlorophyll a } (\mu g/g \text{ tissue}) = 12.7\,(A663) - 2.69\,(A645) \times \frac{V}{1000 \times W}$$

$$\text{Chlorophyll b } (\mu g/g \text{ tissue}) = 22.9\,(A645) - 4.68\,(A663) \times \frac{V}{1000 \times W}$$

$$\text{Total chlorophyll } (\mu g/g \text{ tissue}) = 20.2\,(A645) + 8.02\,(A663) \times \frac{V}{1000 \times W}$$

Where A=Absorbance of specific wavelength, V=Final volume of chlorophyll extract in 80% acetone, W=Fresh weight of tissue extracted (Ibitoye, 2005).

xii. Quantum yield of photosystem II (Phi 2)

This was measured on three fully expanded topmost leaflets on each tagged plant using the multispec device.

xiii. Ratio of light energy that goes towards regulated non-photochemical quenching (Phi NPQ)

This was measured on three fully expanded topmost leaflets on each tagged plant using the multispec device.

xiv. Ratio of light energy that is lost through non-regulated processes (Phi No)

This was measured on three fully expanded topmost leaflet on each tagged plant using the multispec device.

xv. Leaf temperature (°C)

The difference between the leaf temperature and ambient temperature was measured on three fully expanded leaflet on each tagged plant using the multispec device.

xvi. Photosynthetically active radiation (PAR)

This is the amount of energy available for photosynthesis. It was measured using the multispec device.

xvii. Dry matter - The plant tissue was oven dried at 65°C in paper bags until constant weight was attained.

xviii. Biological N fixation (BNF) – The BNF was determined at 50% flowering using the total nitrogen difference method. This method compared the total N of N2 fixing plant and that of a non N2 fixing plant (usually known as a reference crop. Maize was used as the reference crop. The total amount of nitrogen in both the cowpea and the reference crop were determined and the amount of N fixed was calculated using the modified equations of Viera-Vargas *et al.* (1995).

$$\text{Total N in plants (g plant}^{-1}) = \frac{\text{shoot dry weight x } \% \text{ N in plant tissue}}{100}$$

Amount of N fixed = Total N in legume - Total N in reference crops

$$\%\text{NDFA} = \frac{\text{Total N in legume} - \text{Total N in reference crops}}{\text{Total N in legume}} \text{ x } 100$$

Where NDFA = nitrogen derived from the atmosphere.

xix. Total N determination – The dry shoots as well as the seeds of each treatment combination were milled after which N was determined in plant tissue using micro Kjedahl procedure as described by Ibitoye (2005). The total nitrogen in the plant sample was calculated using the formula:

Total N = V x 0.14. Where V is the volume of standard HCl used in the sample titration (Ibitoye, 2005).

xx Nodule effectiveness - Ten nodules were selected at random from each treatment combination and were dissected into halves. Nodules that appeared pinkish, reddish or wine in colour were considered effective as they contain leghemoglobin which is an important enzyme for nitrogen fixation in rhizobia (Tajima *et al.*, 2007). These were then expressed in percentage of the number of sampled nodules.

3.4.2 Yield parameters

i. Number of pods per plant – Total number of pods harvested from each tagged plant was recorded. The total for the six tagged plants was obtained and the average was recorded as number of pods per plant.

ii. Number of seeds per pod – Ten pods were randomly selected from the harvested pods and the average number of seeds per pod was computed.

iii. 100-seed weight (g) - Four replicates of 100 seeds each were counted from each treatment combination and weighed using a Mettler balance; the mean of the four replicates was recorded.

iv. Shell percentage (%) - The pods harvested from the net plot were weighed and shelled. The shell was equally weighed and expressed in percentage of the total pod weight

v. Grain yield (t ha^{-1}) – The grains harvested from the net plots were weighed and the total weight was used to calculate the yield in t ha^{-1}.

vi. Agronomic efficiency- This was calculated using the formula:

$$\text{Agronomic Efficiency (AE)} = \frac{\text{Yield of fertilized cowpea} - \text{Yield of control}}{\text{Quantity of nutrient applied}}$$

(Haruna and Usman, 2013).

vii. Grain harvest index- This was computed as the ratio of the grain yield to total dry matter yield (Zengeni and Giller, 2007)

3.4.3 Meterological data- Data on rainfall, temperature and relative humidity for the three growing seasons were obtained from the Department of Geography, Federal University of Technology, Minna.

3.4.4 Data analysis- All the data collected were subjected to analysis of variance using the statistical analysis system (SAS) version 9.1 and treatment means were separated using Wallller-Duncan K-ratio t-test. Pearson correlation analysis was carried out between the growth, physiological, environmental and photosynthetic efficiency with shoot and grain yield to determine their association. Multiple linear regression was used to determine the effect of physiological variables on grain yield and to determine the coefficients for the model:

$Y= b_0 + b_1x_1 + b_2x_2$ ----b_ix_i given that Y=grain yield (the dependent variable), b_0 is the predicted Y value if $x_1 - x_i =0$, $x_1 - x_i$ are the independent or predictor variables.

3.5 Experiment 3-Physiological Responses of Cowpea Varieties to Sequential Cropping System

3.5.1 Experimental site - This experiment was conducted on three farmers' plot in Minna, Niger State but was researcher managed. The coordinates of the farms are 09^{o} 27.832´ N 006^{o} 25.375´ E, 09^{o} 31. ´ N 006^{o} 27.678´ E, 09^{o} 28.026´ N 006^{o} 25.325´ E in the Southern Guinea Savanna agro-ecology of Nigeria.

3.5.2 Soil sampling and analysis -Soil sampling and procedures for soil analyses were the same as in Experiments 1 and 2.

3.5.3 Treatments and experimental design- The treatments consisted of six varieties (IT93K-452-1, IT99K-573-1-1 and TVX-3236, Kanannado, Oloyin and IT90K-76 of cowpea planted sequentially in 2016 and 2017. The first planting was done on 21st of May in 2016 and 19th of May in 2017. In each year, the second planting was done after the harvest of the first crop. The treatments were arranged in a randomized complete block design with three replicates. Each farmer's field served as a replicate. Treatment plot size was the same as in Experiment 2.

Kanannado is a trailing variety with big white seeds and gray hilum. It is a land race and it is highly cherished by people in the study area mainly because of its big seeds. It is late maturing (90-120 days). Oloyin (honey beans) is equally a land race with medium sized light brown seeds. It is semi-erect, medium maturing (75-80 days) and highly relished by the South-western people of Nigeria for its sweetness. IT90K-76 is an improved variety bred by the International Institute for Tropical Agriculture. It has erect growth habit and is extra early (55-60 days) with deep brown seeds. IT93K-452-1, IT99K-5733-1-1 and TVX-3236 were as described in Section 3.2.3 of Experiment 1.

3.5.4 Cultural practices

N, P and K fertilizers were applied at the rate of 20, 40, 40 kg ha^{-1} respectively, using urea, single super phosphate and muriate of potash respectively while P and K were applied at planting. Land preparation, pest control and all other cultural practices were the same as in Experiment 2.

3.5.5 Data collection

Parameters measured were the same as in Experiment 2 with the exception of nodulation and BNF which were not measured in Experiment 3.

3.5.6 Data analysis- All the data collected were subjected to analysis of variance using the statistical analysis system (SAS) version 9.1 and treatment means were separated using the Waller-Duncan K-ratio T-test at 5% level of probability

3.5.7 Profitability analysis- The profitability of the various treatments was calculated following the procedures of Ayalew *et al.* (2018). The net financial income (NFI) was obtained using the equation:

NFI=GM-TFC

where GM is the gross margin and TFC is the total fixed cost.

GM is given as:

GM= TR-TVC

where TR is the total revenue and TVC is the total variable cost.

The Profit / cost ratio (cost-benefit ratio) was calculated using the formula:

$$\text{Profit / cost ratio} = \frac{NFI}{TC}$$

where NFI is same as above and TC is the Total cost

The total revenue was obtained by multiplying the yield with the current market price at three months after harvest. The total cost of production was calculated considering all operational expenses from acquisition of land up to storage of the produce before sale.

CHAPTER FOUR

4.0 RESULTS AND DISCUSSION

4.1 Results

4.1.1 Experiment 1: Assessing the need for rhizobia inoculation of cowpea in Nigerian savannas

4.1.1.1 Soil properties

The results of the soil analysis of the 20 locations are presented in Table 4.1. The soils were generally very low in nitrogen and organic carbon, low in potassium (except Dawakin Kudu and Zaria soils which had moderate K values). Furthermore, phosphorus content was moderate except Kwali soil which had low available P according to the ratings of Chude *et al.* (2012).

4.1.1.2 Nodulation

i. Number of nodule per plant

No significant difference was observed between inoculated and uninoculated plants in respect of number of nodule (Table 4.2). However, plants that received 90 kg N ha^{-1} produced significantly lighter nodules than inoculated and uninoculated plants. Plants sown on Sabongida soil produced significantly higher number of nodules (47) than the remaining locations. The least (9) was obtained in plants sown on Kumbotso, Dawakin Kudu and Kontagora soils. Variety IT93K-452-1 produced significantly higher number of nodules (22) than IT99K-573-1-1 (19). There was a significant interaction between inoculation and location as well as location and variety on this trait. Figure 4.1 shows that inoculated plants produced more nodules than the uninoculated in 11 out of the 20 locations viz: Gwagwalada, Giwa, Kura, Kwali, Dawakin Kudu, Sabongida, Kaduna-north,

Table 4.1: The physical and chemical properties of the soils obtained from different locations

Locations	pH (H_2O)	Organic C (g kg $^{-1}$)	Total N (g kg $^{-1}$)	Available P (mg kg^{-1})	Ca	Mg	K	Na	ECEC	Sand (g kg $^{-1}$)	Silt (g kg $^{-1}$)	Clay (g kg $^{-1}$)	Textural
					Exchangeable cations (cmol kg^{-1})								
SB	6.79	2.75	0.26	21.6	1.15	1.5	0.12	0.23	3.22	782.1	113.6	104.3	Sandy
GA	4.93	0.79	0.32	18.10	1.25	0.9	0.12	0.20	2.57	792.8	92.1	115.1	Sandy
DA	4.27	1.00	0.13	21.03	0.9	0.4	0.48	0.31	2.22	795.9	99.3	104.8	Sandy
GW	5.38	0.48	0.10	20.06	0.75	0.85	0.13	0.21	2.1	778.7	123.6	97.7	Sandy
KU	6.95	1.43	0.13	31.2	1.45	2.00	0.28	0.35	4.16	727.9	133.1	139	Sandy
KUR	4.52	2.87	0.13	28.15	1.1	0.15	0.21	0.32	1.86	728.9	132.4	138.7	Sandy
ZU	5.9	3.07	0.10	18.2	0.75	1.05	0.25	0.22	2.44	807	103.8	89.2	Loamy
KOT	5.48	1.88	0.26	22.06	1.00	1.25	0.12	0.23	2.75	844	67.1	88.9	Loamy
GE	6.5	1.27	0.10	18.50	0.70	1.8	0.16	0.23	2.97	798.7	88.3	113	Sandy
KO	4.31	1.87	0.15	23.13	0.80	1.85	0.12	0.24	3.11	698	145	157	Sandy
WA	5.38	2.67	0.13	21.26	0.60	1.45	0.11	0.16	2.38	789.6	93	117.4	Sandy
MAB	6.56	0.52	0.21	17.50	1.15	1.05	0.13	0.20	2.67	802.3	82	115.7	Sandy
KW	5.96	1.88	0.21	3.01	0.75	1.15	0.14	0.21	2.32	806.2	84.6	109.2	Sandy
KUJ	5.93	2.67	0.29	28.42	1.25	0.5	0.13	0.2	2.22	728.7	119	152.3	Sandy
AB	6.76	1.48	1.26	23.31	0.90	0.2	0.12	0.17	1.51	782.2	80.3	137.5	Sandy
ZA	6.1	2.19	0.17	26.42	0.75	0.45	0.34	0.26	1.98	801.3	96.2	102.5	Sandy
KS	6.81	3.59	0.25	18.22	0.95	4.3	0.15	0.8	5.71	725.4	110.9	163.7	Sandy
GI	6.96	1.24	0.10	16.51	0.15	1.1	0.13	0.23	2.78	701	122	177	Sandy
KN	6.03	2.67	0.29	18.36	0.95	1.5	0.14	0.21	2.9	794	99	107	Sandy
CH	6.96	2.33	0.32	19.20	1.10	1.55	0.11	0.16	3.04	847.7	92.5	59.8	Loamy

SB- Sabon-gida, GA- Garatu, DA-Dawakin Kudu, AB-Abaji, GW-Gwagwalada, KU-Kumbotso, KUR- Kura, ZU-Zuba, KOT-Kontagora, GE- Gezawa, KO-Koropa, WA- Warawa, MAB- Maikunkele, KW-Kwali, KUJ- Kuje, AB- Abaji, ZA-Zaria, KS-Kaduna -south, GI-Giwa, KN- Kaduna-north, CH-Chikun

Table 4.2: Responses of two cowpea varieties to rhizobia inoculation on soils obtained from different locations

Treatments	Number of nodules per plant	Nodule dry weight (mg/plant)	Shoot biomass (g/plant)
Inoculation (I)			
Uninoculated	25 a	143.67a	4.25b
USDA 3451	24a	150.78a	4.36b
USDA 3384	24a	133.00a	4.48b
90 kg Nha^{-1}	11b	52.92b	4.78a
MSD	3	19.00	0.27
Locations (L)			
Sabongida	47a	225.00a	5.86a
Abaji	39b	223.33a	4.5defg
Zaria	36bc	148.75bc	5.18bc
Maikunele	30cd	127.92bcd	3.31h
Zuba	29d	167.08b	5.66ab
Warawa	21e	144.17bc	4.6cdefg
Giwa	20e	137.08bc	5.08bcd
Kaduna-south	20e	131.67bc	5.09bcd
Kwali	19e	87.50defg	4.16fg
Kaduna-north	19ef	123.75cd	4.51defg
Garatu	17ef	110.83cdef	4.72cdef
Gwagwalada	17ef	115.83cde	4.87cde
Kura	17ef	108.33defg	4.84cde
Kuje	16efg	139.58bc	5.96a
Gezawa	15efg	117.08cde	4.29efg
Koropa	13efg	66.25gh	4.63cdefg
Chikun	11fg	69.17fgh	3.01h
Kumbotso	9g	40.13h	1.61i
Dawakin Kudu	9g	75.00efgh	3.43h
Kontagora	9g	43.33h	4.06g
MSD	7	42.5	0.61
Variety (V)			
IT93K-452-1	22a	115.46a	4.24b
IT99K-573-1-1	19b	124.72a	4.70a
MSD	2	13.4	0.19
Interactions			
I x L	*	*	NS
I x V	NS	*	*
L x V	*	NS	*
I x L x V	NS	NS	NS

Means followed by dissimilar alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan k-ratio t test, MSD-Minimum significant difference, *-Significant ($P \leq 0.05$), NS- Not significant ($P>0.05$)

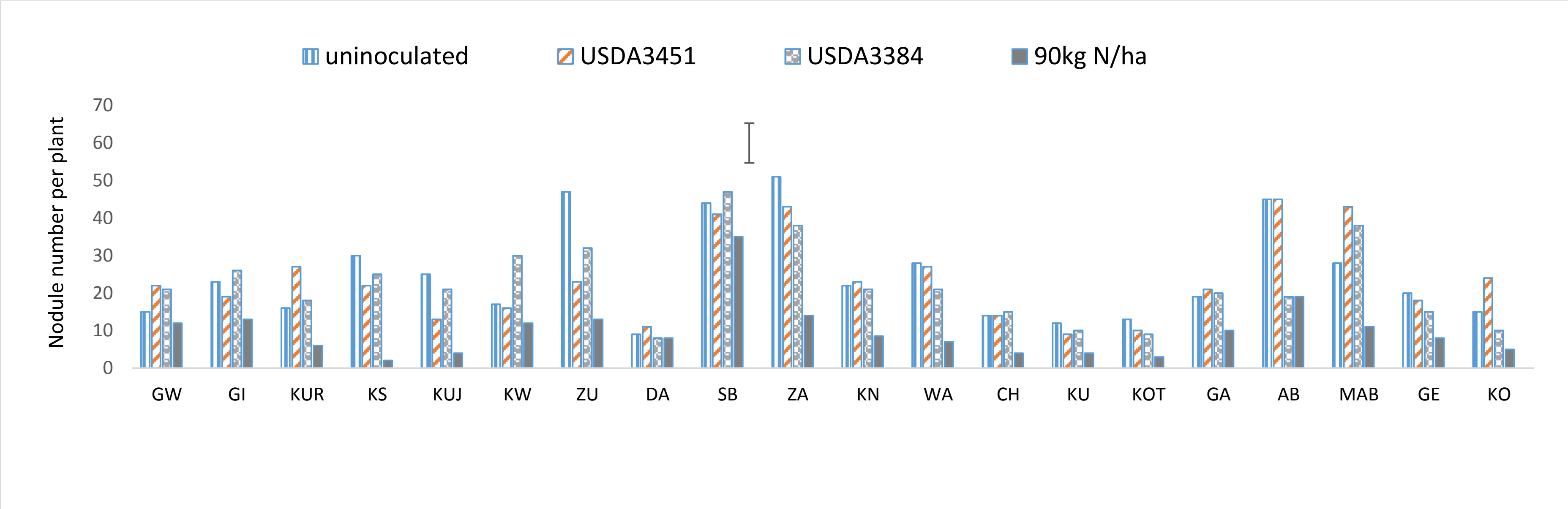


Figure 4.1: Interaction between location and inoculation on number of nodules of cowpea

I- LSD at P =0.05

SB- Sabon-gida, GA- Garatu, DA-Dawakin Kudu, AB-Abaji, GW-Gwagwalada, KU-Kumbotso, KUR- Kura, ZU-Zuba, KOT-Kontagora, GE- Gezawa, KO-Koropa, WA- Warawa, MAB- Maikunkele, KW-Kwali, KUJ- Kuje, AB- Abaji, ZA-Zaria, KS-Kaduna -south, GI-Giwa, KN- Kaduna-north, CH-Chikun

Chikun, Garatu, Maikunkele and Koropa though the differences were only significant in Kwali and Maikunkele soils. Inoculation with USDA 3451 and USDA 3384 increased number of nodules by 32 and 29% respectively over the control plants in Gwagwalada, 41 and 11% respectively in Kura, 10 and 5% respectively in Garatu and 35 and 26% respectively in Maikunkele. In Dawakin Kudu, Kaduna-north and Koropa, only USDA 3451 increased number of nodules over the control plants by 18, 4 and 38% respectively. In Giwa, Kwali, Sabongida and Chikun, only USDA 3384 increased number of nodules by 12, 43, 6, 7% respectively over the control plants. Although uninoculated plants produced higher number of nodules than the inoculated plants in some of the locations namely: Zuba, Kaduna-south (Makera), Kuje, Zaria, Warawa, Kumbotso, Kontagora and Gezawa, the difference was only significant in Zuba. Plants that received 90 kg N ha^{-1} produced the least number of nodules in all the locations

In most locations, there was no significant difference in the response of number of nodules produced among the two varieties. However, in Sabongida and Zaria, variety IT93K-452-1 produced significantly higher number of nodules than IT99K-573-1-1 and the reverse was the case in Maikunkele (Figure 4.2).

ii Nodule dry weight

The inoculated and uninoculated plants produced similar nodule weight which were significantly heavier than the nodules produced by the 90 kg N ha^{-1} fertilized plants. Plants grown on Koropa, Dawakin Kudu, Chikun, Kumbotso and Kontagora soil samples produced significantly lighter nodules. The nodule dry weight of the two varieties were similar (Table 4.2). Both inoculation x location and inoculation x variety had significant effect on nodule dry weight per plant**.** Inoculation increased nodule dry weight in 11 out of the 20 locations namely, Gwagwalada, Giwa, Kura, Zuba,

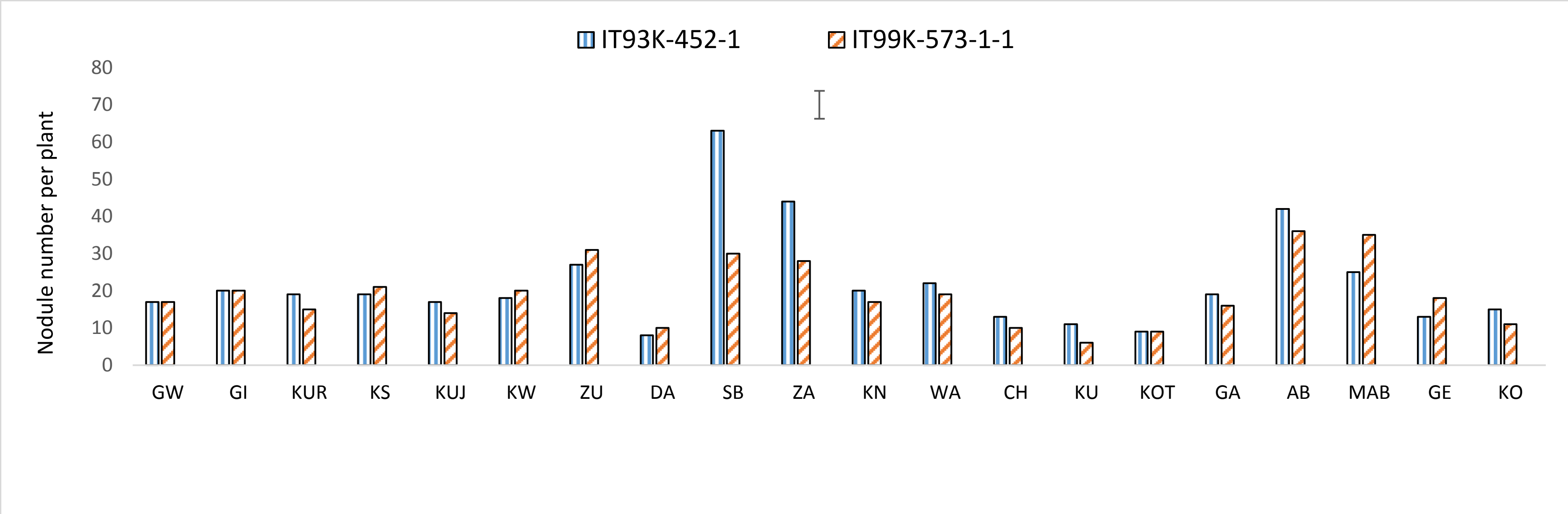


Figure 4.2: Interaction between location and variety on number of nodules of cowpea

I- LSD at P =0.05

SB- Sabon-gida, GA- Garatu, DA-Dawakin Kudu, AB-Abaji, GW-Gwagwalada, KU-Kumbotso, KUR- Kura, ZU-Zuba, KOT-Kontagora, GE- Gezawa, KO-Koropa, WA- Warawa, MAB- Maikunkele, KW-Kwali, KUJ- Kuje, AB- Abaji, ZA-Zaria, KS-Kaduna -south, GI-Giwa, KN- Kaduna-north, CH-Chikun

Dawakin Kudu, Sabongida, Zaria, Kaduna-north, Garatu, Maikunkele and Koropa though the differences were not significant in most of these locations. Plants inoculated with USDA 3451 produced heavier nodules than those inoculated with USDA 3384 in eight out of the eleven locations and the difference was significant in Zuba, Sabongida and Koropa. However, in soils of Kaduna-south and Zaria, USDA 3384 produced significantly heavier nodules than USDA 3451 inoculated plants. Plants that received 90 kg N ha^{-1} produced significantly lighter nodules than the other treatments in all the locations except in Sabongida where plants that received 90 kg N ha^{-1} produced heavier nodules than those inoculated with USDA 3384 (Figure 4.3).

The interaction effects of rhizobia inoculation and variety on nodule weight revealed that IT99K-573-1-1 plants inoculated with USDA 3384 rhizobia strain produced significantly heavier nodules than IT93K-452-1 variety inoculated with the same strain. There was no significant difference between the weight of nodules produced in the two varieties in uninoculated plants and those inoculated with USDA 3451 (Figure 4.4).

4.1.1.3 Shoot biomass yield

The inoculated and uninoculated plants produced similar but significantly lower shoot biomass yield than plants that received 90 kg N ha^{-1} (Table 4.2). Plants that received 90 kg N ha-^{1} had the mean and median values of 4.62 and 4.58 g/plant respectively. The interquartile range was 3.56 - 5.34 g/plant indicating that fifty percent of the plants weighed between 3.56 and 5.34 g/plant. Next in performance were plants inoculated with USDA 3384 with the mean and median values of 4.48 and 4.47 g/plant respectively. The interquartile range was 3.82 -5.75 g/plant (Figure 4.5).

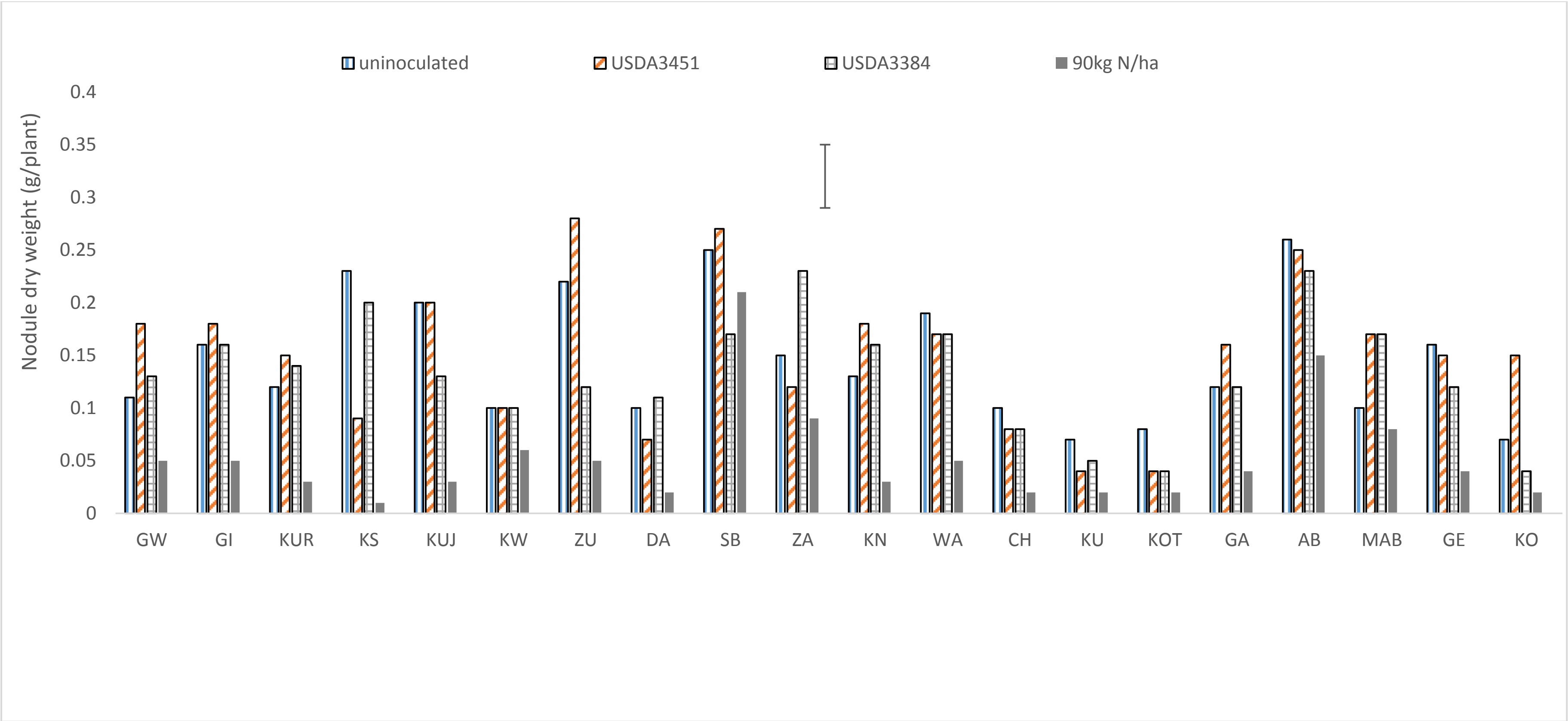


Figure 4.3: Interaction between location and inoculation on nodule dry weight of cowpea

I- LSD at P =0.05

SB- Sabon-gida, GA- Garatu, DA-Dawakin Kudu, AB-Abaji, GW-Gwagwalada, KU-Kumbotso, KUR- Kura, ZU-Zuba, KOT-Kontagora, GE- Gezawa, KO-Koropa, WA- Warawa, MAB- Maikunkele, KW-Kwali, KUJ- Kuje, AB- Abaji, ZA-Zaria, KS-Kaduna -south, GI-Giwa, KN- Kaduna-north, CH-Chikun

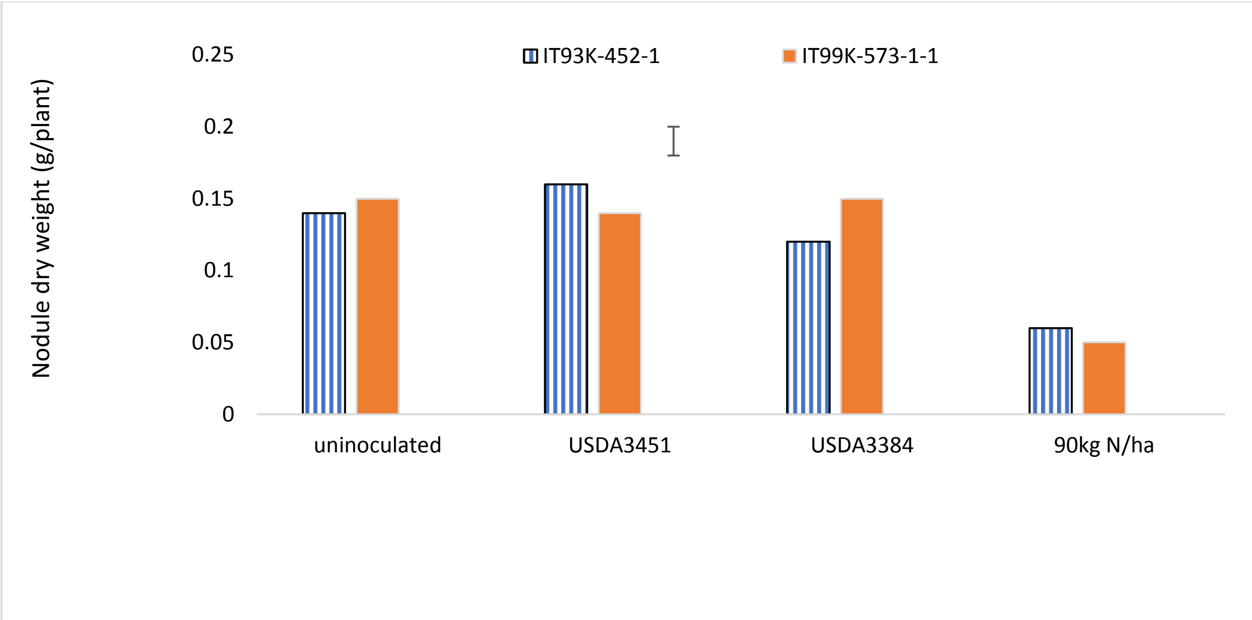


Figure 4.4: Interaction between inoculation and varieties on nodule dry weight of cowpea

I- LSD at P =0.05

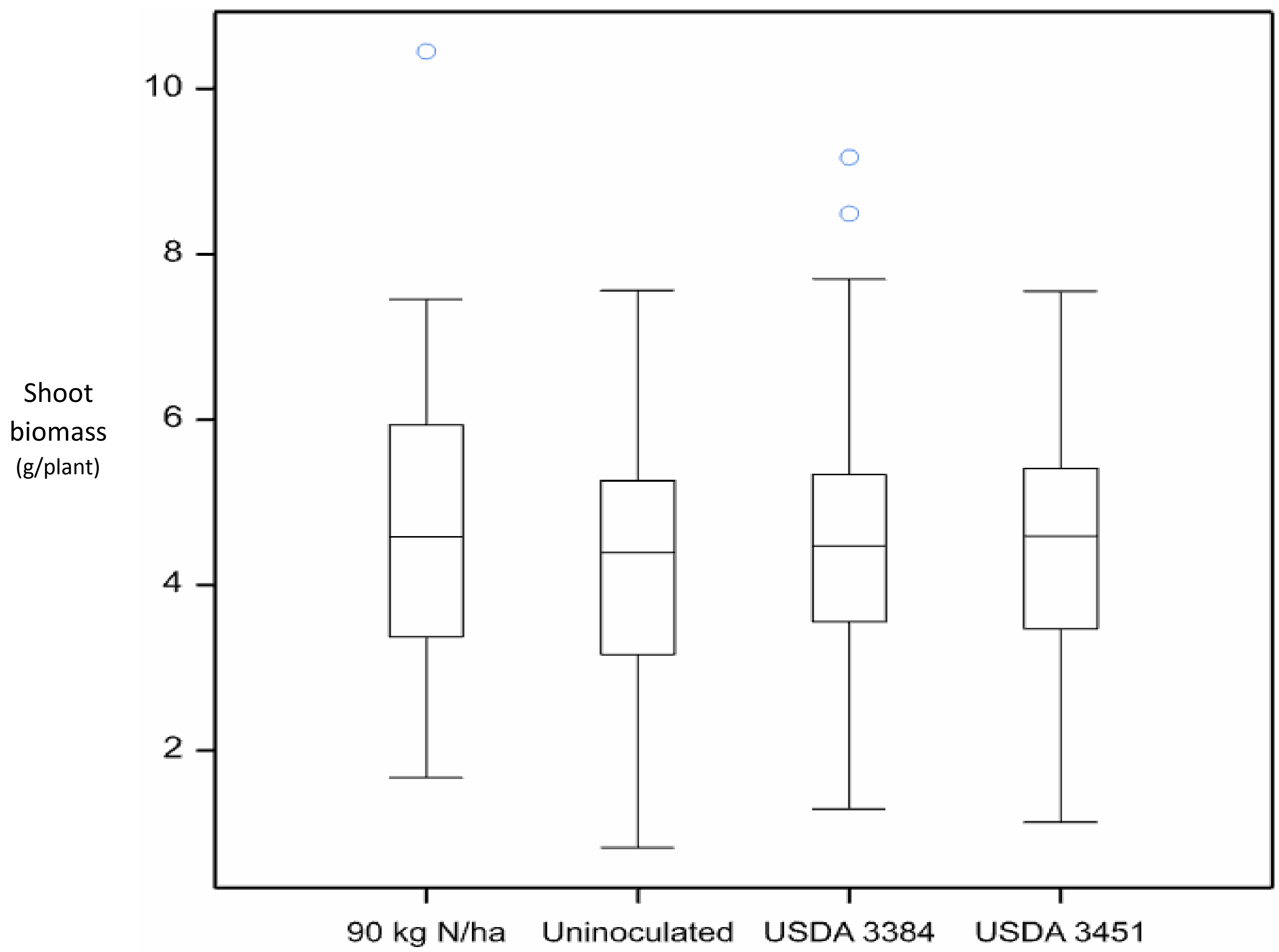


Figure 4.5: Boxplot showing the shoot biomass yield of the inoculation treatments

Next in performance were plants inoculated with USDA 3451 with the mean and median values of 4.36 and 4.59 g/plant respectively. The interquartile range of the plants was 3.72 - 5.79 g/plant. The uninoculated plants produced the least biomass with the mean and median values of 4.25 and 4.39 g/plant respectively. The interquartile range of the plant was 3.16 - 5.26 g/plant.

Significant variation exists between the biomass yield of the plants sown in soils of the different locations (Table 4.2). Plants grown in Kuje soil had the highest mean performance (5.96 g/plant). The upper and lower quartile values were 6.88 and 5.25 g/plant respectively indicating that 50% of the plant in Kuje weighed between 5.25 and 6.88 g/plant. This was followed by Sabongida soil which had the mean value of 5.86 g/plant. However, the interquartile range of the plants was 5.28 - 6.71 g/plant. The least performance was recorded in plants grown in Kumbotso soil. The mean and median values were 1.61 and 1.55 g/plant respectively. The upper and lower quartile values were 1.83 and 1.39 g/plant respectively (Figure 4.6).

IT99K-573-1-1 produced significantly higher biomass than IT93K-452-1 (Table 4.2). IT99K-573-1-1 produced shoots with the mean and median values of 4.67 and 4.80 g/plant respectively. The interquartile range was 3.78 - 5.83 g/plant. The mean and median biomass weight recorded in IT93K-452-1 were 4.19 and 4.12 g/plants respectively and the interquartile range of the biomass was 3.11 - 5.20 g/plant (Figure 4.7).

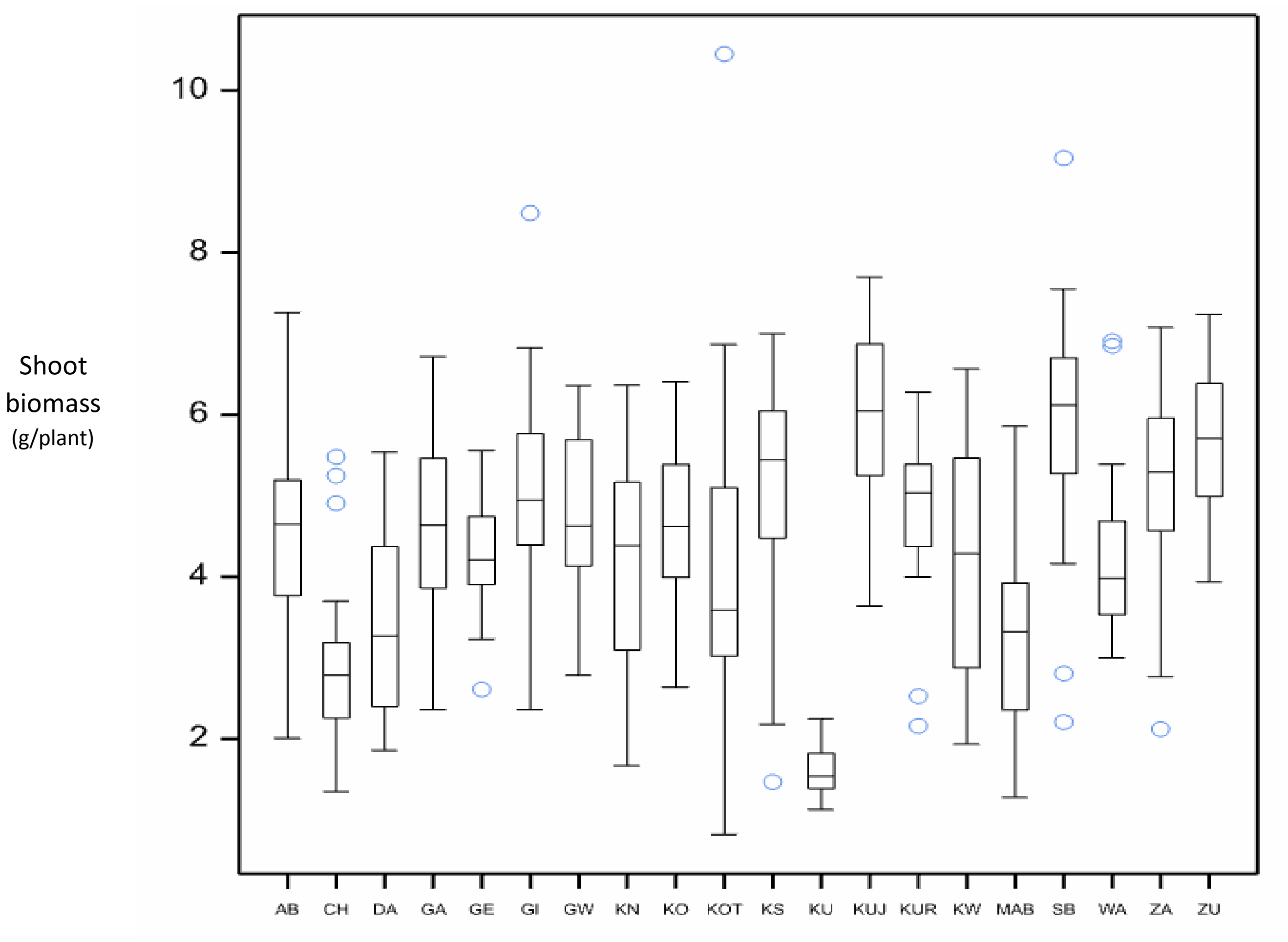


Figure 4.6: Boxplot showing the shoot biomass yield across the locations

SB- Sabon-gida, GA- Garatu, DA-Dawakin Kudu, AB-Abaji, GW-Gwagwalada, KU-Kumbotso, KUR- Kura, ZU-Zuba, KOT-Kontagora, GE- Gezawa, KO-Koropa, WA- Warawa, MAB-Maikunkele, KW-Kwali, KUJ- Kuje, AB- Abaji, ZA-Zaria, KS-Kaduna -south, GI-Giwa, KN-Kaduna-north, CH-Chikun

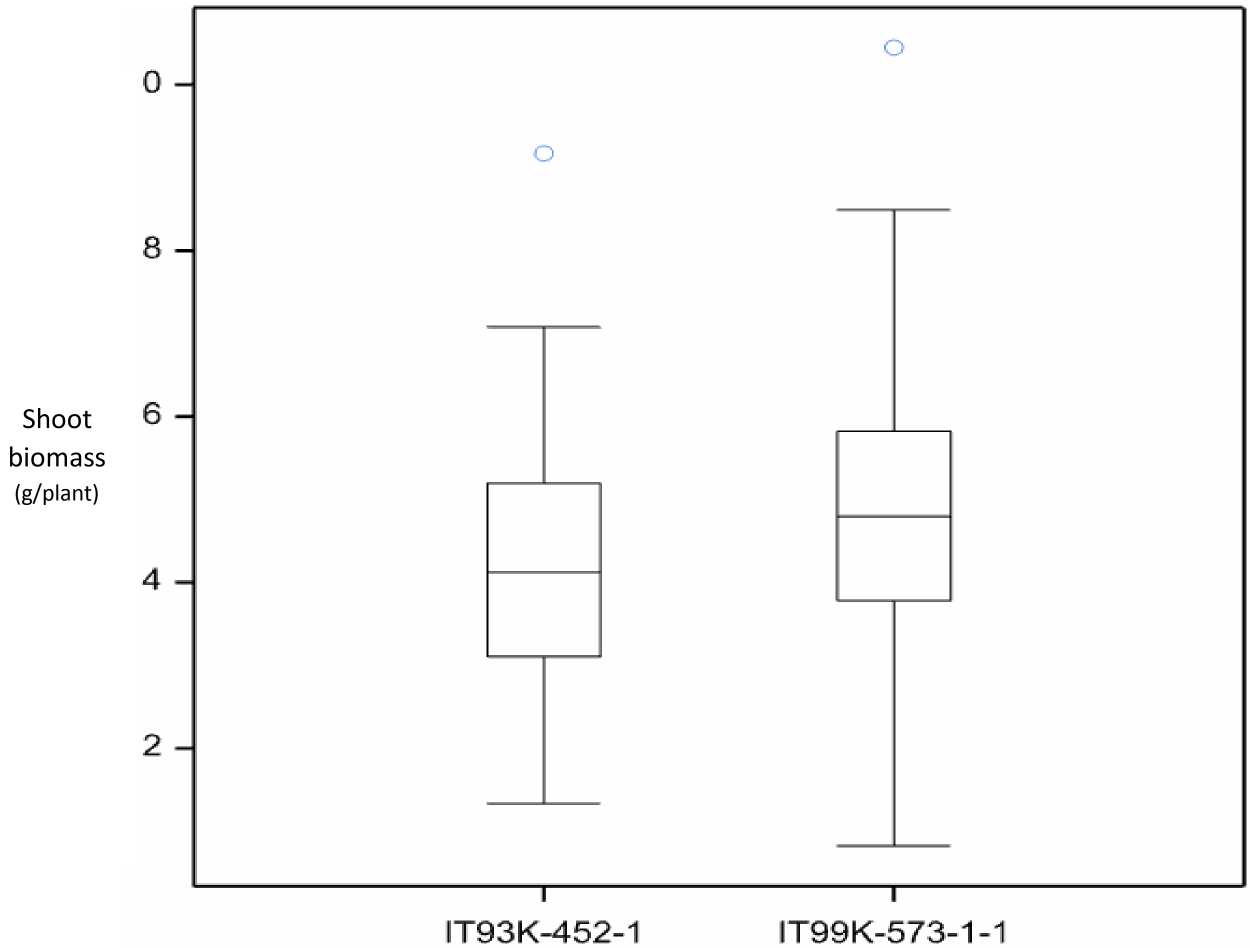
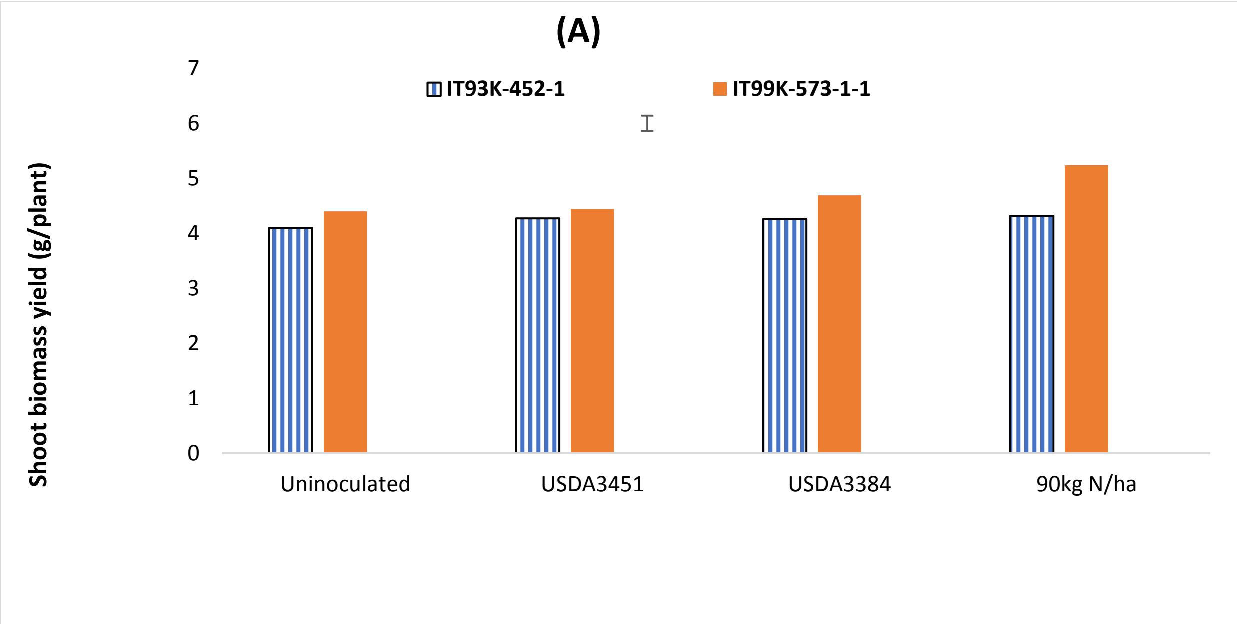


Figure 4.7: Boxplot showing the shoot biomass yield of cowpea varieties

Significant interaction exists between inoculation and variety as well as location and variety on shoot biomass. Figure 4.8A shows that IT99K-573-1-1 produced a higher shoot biomass than IT93K-452-1 across the inoculation treatment. The difference between the two varieties was however only significant in plants that received 90 kg N ha $^{-1}$. IT99K-573-1-1 plants that received 90 kg N ha $^{-1}$ produced significantly higher biomass than all the other treatment combinations. There was no significant difference between the biomass produced by IT93K-452-1 plants that received 90 kg N ha $^{-1}$ and other inoculation treatments. IT99K-573-1-1 produced higher shoot biomass than IT93K-452-1 in 80% of the locations though the differences were not significant in most of the locations except in Giwa and Kaduna-south (Figure 4.8B).

The polygon view of the genotype plus genotype by environment interaction (GGE) biplot showing the best rhizobia strain in each location is presented in Figure 4.9. The principal component one (PC1) and two (PC2) explained 82.60% (61.76 and 20.84% respectively) of the GGE variation. The location at the vertex of each quadrant had the best performance in that quadrant. Plants that received 90 kg N ha-^{1} performed best in Kontagora, Garatu, Abaji, Kaduna-south, Kuje, Kumbotso, Gezawa, Gwagwalada, Dawakin Kudu, Kaduna-north and Sabongida soils representing 55% of the location. The uninoculated plants performed best in Chikun, Kwali, Maikunkele, Zuba Warawa and Kura soils. Plants inoculated with USDA 3451 and USDA 3384 performed best in Giwa, Zaria, Kwali and Kura soils. None of the inoculation treatment performed well in Koropa soil.



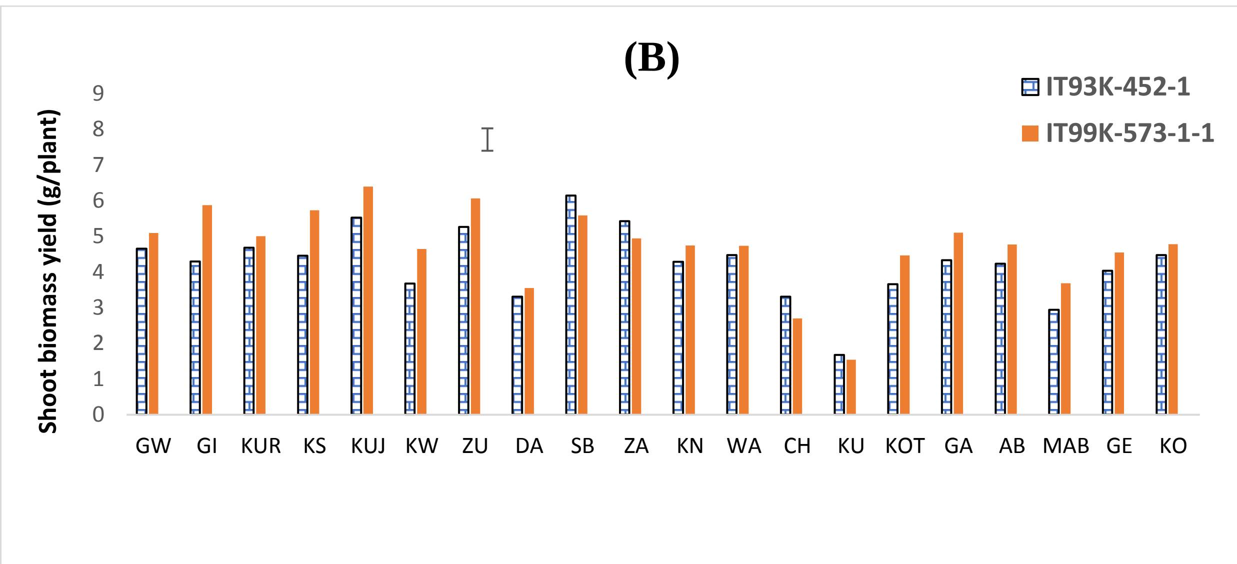


Figure 4.8 (A): Interaction between inoculation and variety on shoot biomass yield of cowpea (B): Interaction between location and variety on shoot biomass yield of cowpea

I- LSD at P =0.05

SB- Sabon-gida, GA- Garatu, DA-Dawakin Kudu, AB-Abaji, GW-Gwagwalada, KU-Kumbotso, KUR- Kura, ZU-Zuba, KOT-Kontagora, GE- Gezawa, KO-Koropa, WA- Warawa, MAB-Maikunkele, KW-Kwali, KUJ- Kuje, AB- Abaji, ZA-Zaria, KS-Kaduna -south, GI-Giwa, KN-Kaduna-north, CH-Chikun

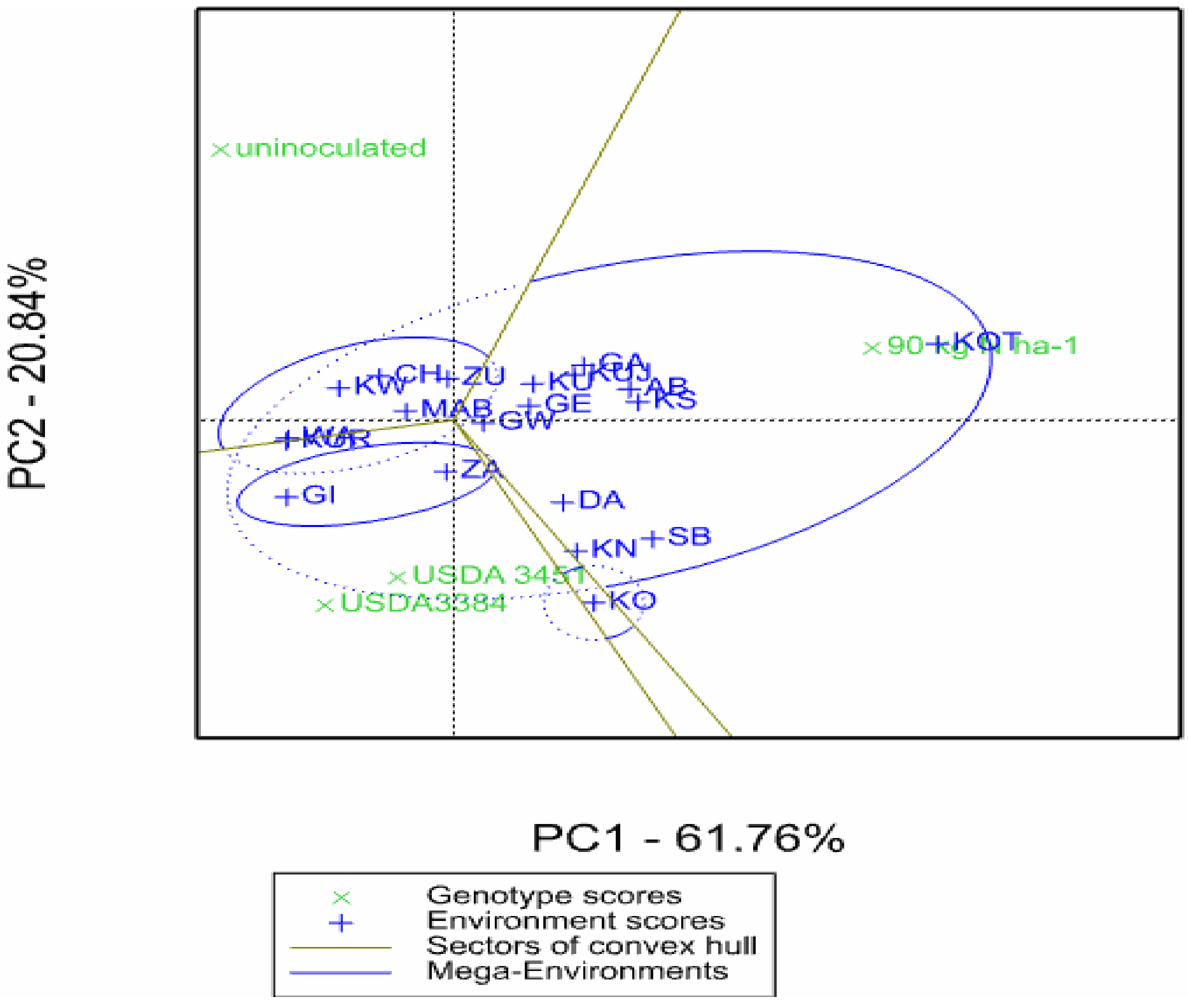


Figure 4.9: Genotype (rhizobia strain) x environment biplot for shoot biomass yield.

4.1.2 Experiment 2: physiological responses of cowpea varieties to rhizobia inoculation and phosphorus application

4.1.2.1 Growth attributes

i. Leaf area

The leaf area of cowpea varieties in response to rhizobia inoculation and phosphorus application is presented in Table 4.3. The uninoculated plants produced significantly larger leaves than those from plants inoculated with USDA 3451 and USDA 3384 which produced similar leaf sizes in 2015. In 2016, plants inoculated with BR 3262 produced significantly larger leaves than plants inoculated with BR 3267 while in 2017, the uninoculated plants and those inoculated with BR 3267 produced significantly larger leaves than those inoculated with BR 3267.

In 2015 and 2016, the leaf area increased significantly as the phosphorus rates increased with the least and highest values recorded at 0 and 40 kg P ha^{-1} respectively. In 2017 however, plants that received 20 and 40 kg P ha^{-1} produced leaves of similar sizes of 120.88 and 119.33 cm^2 respectively and both were significantly larger than the value (103.25 cm^2) recorded in the unfertilized plants. There was no significant difference between the sizes of leaves produced by IT99K-573-1-1 and IT93K-452-1 but they were significantly greater than that of TVX-3236 leaves in 2015 and 2017. In 2016 the greatest leaf area of 186.55 cm^2 recorded for IT99K-573-1-1 was similar to that of IT93K-452-1 but significantly greater than the value (155.66 cm^2) recorded in TVX-3236 plants.

The interaction between rhizobia inoculation and phosphorus on leaf area was significant in the three years ($P \leq 0.01$ in 2015 and 2016 and $P \leq 0.05$ in 2017). In 2015, the leaf area of the uninoculated plants increased significantly with increase in the phosphorus rates with the highest value (238.00 cm^2) recorded in those that received 40 kg P ha^{-1} (Table 4.4). In plants inoculated with USDA 3451 however, there was no significant difference between the leaf area of plants that

Table 4.3: Effects of rhizobia inoculation and phosphorus application on leaf area (cm^2) of cowpea varieties

	Cropping seasons		
	2015	**2016**	**2017**
Treatments			
Inoculation (I)			
Uninoculated	192.30a	173.17ab	118.57a
USDA 3451	171.90b		114.65ab
USDA 3384	167.24b		
BR 3262		185.11a	118.10a
BR 3267		155.65b	105.63b
90 kg N ha^{-1}		173.34ab	115.49ab
M S D	11.02	22.40	11.14
Phosphorus (P)			
0 kg ha^{-1}	150.93c	129.08c	103.25b
20 kg ha^{-1}	177.61b	179.20b	120.88a
40 kg ha^{-1}	202.99a	207.18a	119.33a
M S D	11.02	19.40	8.63
Variety (V)			
IT93K-452-1	191.53a	172.85ab	125.09a
IT99K-573-1-1	191.08a	186.95a	131.69a
TVX-3236	148.83b	155.66b	86.68b
M S D	11.02	19.40	8.63
Interactions			
I × P	**	**	*
I × V	NS	**	*
P × V	*	NS	NS
I × P × V	NS	*	*

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test. MSD-Minimum significant difference, *, **- significant at P=0.05 and 0.01 respectively, NS- not significant at P=0.05

Table 4.4: Interaction effects of rhizobia inoculation and phosphorus on leaf area (cm^2) of cowpea in 2015 – 2017

	Phosphorus (kg ha^{-1})		
	0	20	40
Inoculation			
		2015	
Uninoculated	146.0de	192.82b	238.00a
USDA 3451	142.95e	180.81bc	191.94b
USDA 3384	159.19de	163.75cd	178.78bc
SE±		6.73	
		2016	
Uninoculated	120.93fg	175.76cde	222.83ab
90 kg N ha^{-1}	142.66ef	172.92cde	239.76a
BR 3262	96.00g	195.81bc	175.15cde
BR 3267	156.74def	172.30cde	190.99bcd
SE±		7.01	
		2017	
Uninoculated	118.37 a-d	119.05a-d	118.28a-d
90 kg N ha^{-1}	124.35ab	124.35ab	135.34a
BR 3262	90.37e	124.78ab	101.74de
BR 3267	103.90cde	116.91a-d	123.14abc
USDA 3451	109.00b-e	119.30a-d	118.28a-d
SE±		6.87	

Means followed by different alphabets are significantly different at P=0.05 using Waller-Duncan K-ratio t test, SE- standard error of the mean received 20 and 40 kg P ha^{-1} but both were significantly larger than the leaves of the unfertilized plants. In plants inoculated with USDA 3384 significant difference in leaf area was only recorded between 40 kg P ha^{-1} (178.78 cm^2) and unfertilized plants (159.19 cm^2).

In 2016, the leaf area of the uninoculated plants significantly increased with increase in phosphorus rates with the smallest and largest leaves obtained at 0 and 40 kg P ha^{-1} respectively (Table 4.4). When plants were fertilized with 90 kg N ha^{-1} there was no significant difference between the control plants and those to which 20 kg P ha^{-1} was applied. In plants inoculated with BR 3262 application of 20 kg P ha^{-1} resulted in significant increase in leaf area compared to the control but no further significant increase was recorded at 40 kg ha^{-1}. In BR 3267 inoculated plants, leaf area did not differ significantly among the different P rates applied. Overall, plants fertilized with a combination of 90 kg N ha^{-1} and 40 kg P ha^{-1} had significantly larger leaves than all other treatment combination except those of the uninoculated plants that equally received 40 kg P ha^{-1}. Also, the leaf area (96.00cm^2) recorded in unfertilized BR 3262 was significantly lower than in all other treatment combination except those of unfertilized uninoculated plants. In 2017, there were no significant differences in the leaf areas among all the P treatments under each inoculation treatments of the fertilized and unfertilized plants except in BR 3262 plants in which the leaf area of plants fertilized with 20 kg P ha^{-1} produced significantly larger leaves than the unfertilized plants and those fertilized with 40 kg P ha^{-1} (Table 4.4).

The interaction between rhizobia inoculation and variety on leaf area was significant in 2016 and 2017. In 2016, there were no significant differences among the leaf area values of inoculated and uninoculated IT93K-452-1 as well as those that received 90 kg N ha^{-1} treatment (Figure 4.10). In IT99K-573-1-1 plants, the greatest leaf area of 218.62 cm^2 recorded in uninoculated plants was significantly higher than those of other treatments which were all similar. In TVX-3236 variety, plants that received 90 kg N ha^{-1} had significantly larger leaves than those observed in the inoculated and uninoculated plants which were similar. Uninoculated plants of IT99K-573-1-1 had the largest leaves among all the treatment combinations but the value was statistically similar to those of TVX-3236 variety that received 90 kg N ha^{-1} and IT93K-452-1 plants inoculated with BR 3267. In 2017, the uninoculated IT93K-452-1 plants produced the largest leaves statistically similar to the leaves of plants inoculated with BR 3262 and USDA 3451. The uninoculated IT99K-573-1-1 plants equally produced the largest leaves similar to those recorded for all inoculation treatments except BR 3267. In TVX-3236 variety however, plants inoculated with BR 3262 produced significantly larger leaves than all the other inoculation treatments which were similar. Figure 4.11 shows that the leaf area of both IT93K-452-1 and IT99K-573-1-1 increased significantly as the phosphorus level increased from 0 to 20 and then to 40 kg P ha^{-1}. In TVX-3236 however, significant increase in the leaf area was recorded between 0 and 20 kg P ha^{-1} ; the value obtained at 40 kg P ha^{-1} was not significantly different from those at 0 and 20 kg P ha^{-1}.

The interaction between rhizobia inoculation, phosphorus and variety on leaf area in 2016 is presented in Figure 4.12. Leaf area increased with increase in phosphorus rates in the uninoculated IT93K-452-1 and IT99K-573-1-1 plants with the maximum and minimum values observed in plants that received 0 and 40 kg P ha^{-1} respectively. In both varieties however, there was no significant difference between the leaf area values recorded for plants to which 0 and 20 kg P ha^{-1} was applied. In uninoculated TVX-3236, maximum leaf area was recorded at 20 kg P ha^{-1} which was significantly greater than that at 0 kg P ha^{-1} but similar to the value recorded at 40 kg P ha^{-1} application.

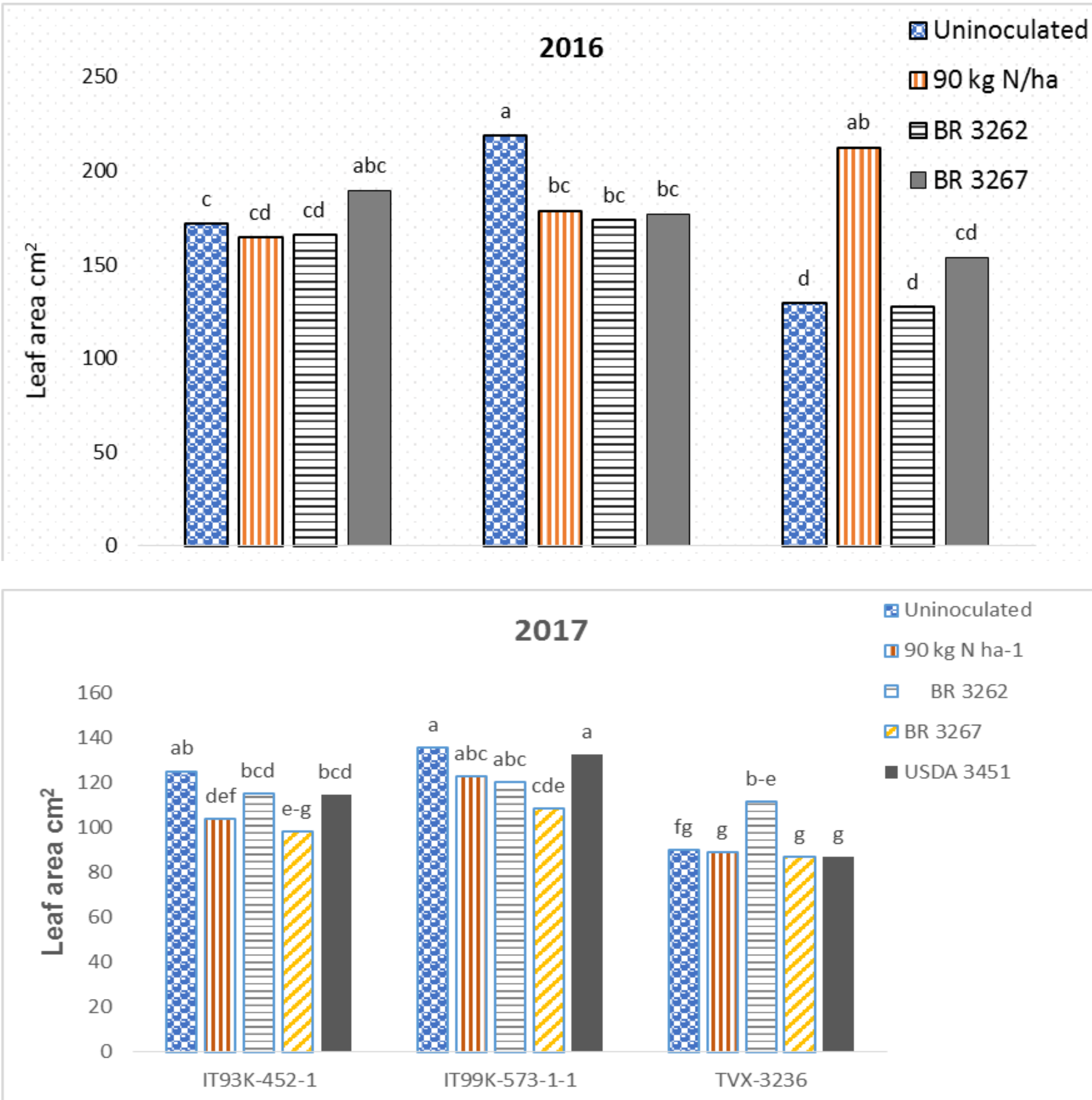


Figure 4.10: Interaction effects of rhizobia inoculation and variety on leaf area of cowpea in 2016 and 2017

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

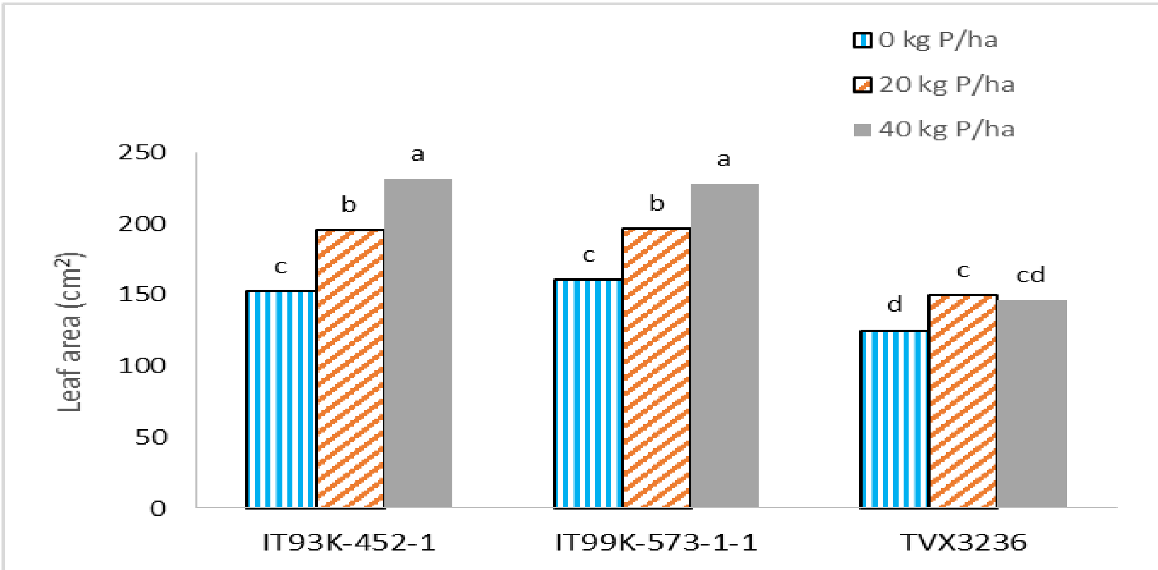


Figure 4.11: Interaction effects of phosphorus and variety on leaf area of cowpea in 2015

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

In plants that received 90 kg N ha^{-1} leaf area increased with increase in phosphorus rates in the three varieties. However in IT93K-452-1 variety, the leaf area of plants that received 40 kg P ha^{-1} was only significantly higher than the unfertilized. In TVX-3236 there was no significant difference between the fertilized and unfertilized plants.

Application of 20 kg P ha^{-1} to plants of both IT93K-452-1 and IT99K-573-1-1 inoculated with BR 3262 resulted in the highest leaf area which was significantly greater than the value recorded at 0 kg P ha^{-1} but was similar to the values obtained at 40 kg P ha^{-1}. In TVX-3236 variety inoculated with BR 3262 however, leaf area was greatest at 40 kg P ha^{-1} and was significantly higher than 0 kg P ha^{-1} but similar to the value obtained at 20 kg P ha^{-1}.

In plants inoculated with BR 3267 rhizobia strain, unfertilized IT93K-452-1 plants had similar leaf area with plants that received 40 kg P ha^{-1}. In IT99K-573-1-1 plants, leaf area increased with increase in phosphorus rates but the value for the plants that received 40 kg P ha^{-1} was only significantly higher than that of the unfertilized plants. No significant differences were recorded in the leaf area values of the fertilized and unfertilized TVX- 3236 plants (Figure 4.12).

The interaction between rhizobia inoculation, phosphorus and variety on leaf area was significant in 2017. The optimum inoculation and phosphors combination for enhanced leaf area varied with the variety planted. The inoculation of IT93K-452-1 with BR 3262 combined with the application of P at 20 kg ha^{-1} resulted in the production of the largest leaves (138.59cm^2) which was however similar to most of the other inoculation phosphorus combination except combination of 90 kg N ha^{-1} and 0 and 40 kg P ha^{-1}, BR 3262 and 0 kg P ha-^{1}, BR 3267 and 0 and 20 kg P ha^{-1}. Furthermore, uninoculated IT99K-573-1-1 plants to which 20 kg P ha^{-1} was applied produced leaves of the

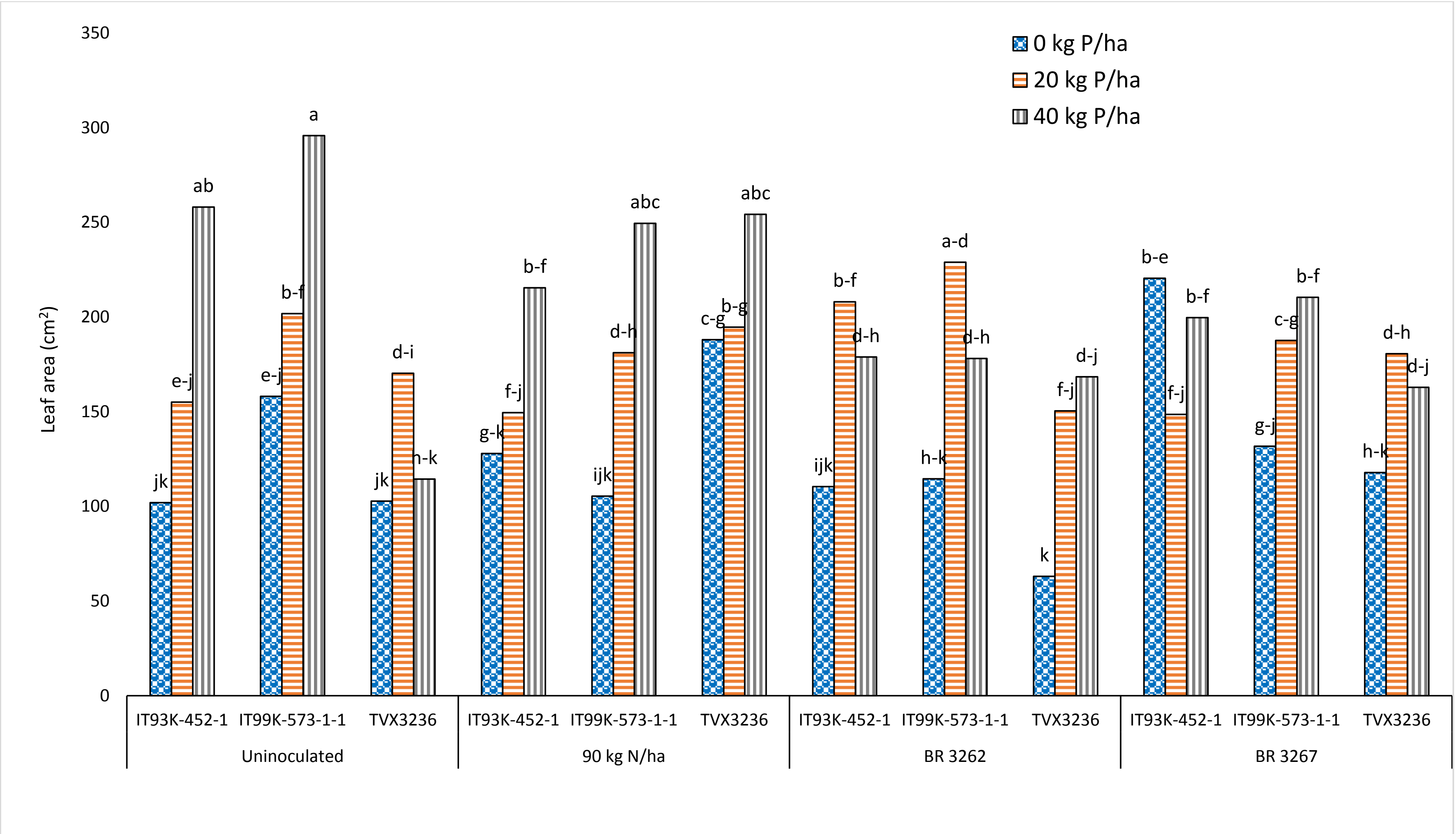


Figure 4.12:. Interaction effects of rhizobia inoculation, phosphorus and variety on leaf area of cowpea in 2016

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

greatest area (151.00 cm^2). The value was however statistically similar to those of the other inoculation + phosphorus combination except 90 kg N ha^{-1} and 0 kg P ha^{-1}, BR 3262 and 0 and 20 kg P ha^{-1}, and BR 3267 in combination with all the P treatments. In TVX-3236, the combination of BR 3262 and 20 kg P ha^{-1} gave the greatest area of 133.62 cm^2 which was significantly greater than the values recorded for all other combination (Table 4.5).

ii. Number of leaves per plant

The number of leaves produced by the different cowpea varieties in response to rhizobia inoculation and phosphorus application is presented in Table 4.6. There was no significant difference between the number of leaves produced by inoculated and uninoculated plants in 2015. In 2016, the highest number of leaves was obtained in plants fertilized with 90 kg N ha^{-1} which was however only significantly higher than the values obtained in plants inoculated with BR 3262. In 2017, the uninoculated plants and those inoculated with BR 3262 and USDA 3451 produced significantly higher number of leaves than plants inoculated with BR 3267.

In 2015, application of 20 kg ha^{-1} resulted in the production of the highest mean number of leaves (*ca* 50) and the value was significantly higher than the number of leaves (*ca* 46) produced by plants that received 40 kg P ha^{-1}. In 2016, plants to which 40 kg P ha^{-1} was applied produced significantly higher number of leaves (*ca* 53) than those that received 20 kg P ha^{-1}. In 2017, application of 20 and 40 kg P ha^{-1} resulted in the production of similar number of leaves that were significantly higher than the number of leaves produced by the unfertilized plants. The least number of leaves was recorded in the unfertilized plants in the three years.

Table 4.5: Interaction effects of variety, inoculation and phosphorus on leaf area (cm^2) of cowpea in 2017

Variety	Inoculation	Phosphorus ($kg\ ha^{-1}$)	Leaf area (cm^2)
IT93K-452-1	Uninoculated	0	116.771b-k
IT93K-452-1	Uninoculated	20	133.31a-d
IT93K-452-1	Uninoculated	40	125.45a-i
IT93K-452-1	$90kgNha^{-1}$	0	78.75o-q
IT93K-452-1	$90kgNha^{-1}$	20	132.07a-d
IT93K-452-1	$90kgNha^{-1}$	40	101.22h-q
IT93K-452-1	BR 3262	0	93.18j-q
IT93K-452-1	BR 3262	20	138.59ab
IT93K-452-1	BR 3262	40	114.32b-m
IT93K-452-1	BR 3267	0	101.98g-p
IT93K-452-1	BR 3267	20	83.57n-q
IT93K-452-1	BR 3267	40	109.47c-n
IT93K-452-1	USDA 3451	0	98.81i-q
IT93K-452-1	USDA 3451	20	129.96a-f
IT93K-452-1	USDA 3451	40	114.90b-l
IT99K-573-1-1	Uninoculated	0	126.41a-i
IT99K-573-1-1	Uninoculated	20	151.00a
IT99K-573-1-1	Uninoculated	40	129.83a-g
IT99K-573-1-1	$90kgNha^{-1}$	0	102.23f-o
IT99K-573-1-1	$90kgNha^{-1}$	20	132.07a-d
IT99K-573-1-1	$90kgNha^{-1}$	40	134.94a-c
IT99K-573-1-1	BR 3262	0	102.05f-p
IT99K-573-1-1	BR 3262	20	118.92b-j
IT99K-573-1-1	BR 3262	40	140.41ab
IT99K-573-1-1	BR 3267	0	113.41b-m
IT99K-573-1-1	BR 3267	20	95.81j-q
IT99K-573-1-1	BR 3267	40	115.87b-l
IT99K-573-1-1	USDA 3451	0	131.41a-e
IT99K-573-1-1	USDA 3451	20	137.47ab
IT99K-573-1-1	USDA 3451	40	128.98a-h
TVX-3236	Uninoculated	0	79.86opq
TVX-3236	Uninoculated	20	97.15j-q
TVX-3236	Uninoculated	40	93.57j-q
TVX-3236	$90kgNha^{-1}$	0	73.61q
TVX-3236	$90kgNha^{-1}$	20	90.45k-q
TVX-3236	$90kgNha^{-1}$	40	103.53e-o
TVX-3236	BR 3262	0	105.36d-o
TVX-3236	BR 3262	20	133.62a-c
TVX-3236	BR 3262	40	95.61j-q
TVX-3236	BR 3267	0	80.03opq
TVX-3236	BR 3267	20	87.94l-q
TVX-3236	BR 3267	40	93.45j-q
TVX-3236	USDA 3451	0	74.21pq
TVX-3236	USDA 3451	20	86.39m-q
TVX-3236	USDA 3451	40	100.23i-q
SE $\pm$		9.95	

Means followed by different alphabets are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, SE- standard error of the mean

Table 4.6: Effects of rhizobia inoculation and phosphorus application on number of leaves of cowpea varieties

	Cropping seasons		
	2015	2016	2017
Treatments			
Inoculation (I)			
Uninoculated	39.89a	45.32ab	20.42a
USDA 3451	42.19a		16.98b
USDA 3384	42.41a		
BR 3262		47.33a	18.09ab
BR 3267		40.73b	20.43a
90 kg N ha^{-1}		45.36ab	19.82a
M S D	2.61	5.33	2.42
Phosphorus (P)			
0 kg ha^{-1}	29.11c	33.78c	14.55b
20 kg ha^{-1}	49.63a	46.90b	20.82a
40 kg ha^{-1}	45.74b	53.38a	22.07a
M S D	2.61	4.62	1.87
Variety (V)			
IT93K-452-1	43.56b	45.23a	18.79b
IT99K-573-1-1	34.44c	48.92a	17.51b
TVX-3236	46.48a	39.90b	21.14a
M S D	2.61	4.62	1.87
Interactions			
I × P	*	NS	NS
I × V	NS	**	NS
P × V	NS	*	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, *, **- significant at P=0.05 and 0.01 respectively, NS- not significant at P=0.05

The number of leaves produced per plant varied significantly among the varieties and the order of performance was TVX-3236 > IT93K-452-1 > IT99K-573-1-1 in 2015. In 2016, the number of leaves produced by IT99K-573-1-1 was similar to that of IT93K-452-1 and both produced statistically more leaves than the value recorded in TVX-3236 plants. In 2017 however, TVX-3236 produced significantly higher number of leaves than both IT93K-452-1 and IT99K-573-1-1 plants which produced similar number of leaves.

The interaction between rhizobia inoculation and phosphorus application on number of leaves was significant in 2015 ($P \leq 0.05$). Application of P at 20 and 40 kg P ha^{-1} significantly increased the number of leaves in uninoculated and uninoculated plants compared to the values recorded at 0 kg P ha^{-1} (Figure 4.13A). Significantly fewer leaves were produced when uninoculated and those inoculated with USDA 3451 were fertilized with 40 kg P ha^{-1} than when fertilized with 20 kg P ha^{-1}. Contrary to the above trend, the number of leaves obtained at 20 kg P ha^{-1} was not significantly different from that recorded 40 kg P ha^{-1} in plants inoculated with USDA 3384.

The interaction between rhizobia inoculation and variety on number of leaves in 2016 is presented in Figure 4.13B. There was no significant difference between the number of leaves of inoculated and uninoculated IT93K-452-1 plants. In IT99K-573-1-1 variety, uninoculated plants produced significantly greater number of leaves than those obtained in the other inoculation treatments which were similar. TVX-3236 plants fertilized with 90 kg P ha^{-1} produced significantly higher number of leaves than those produced by both the uninoculated and inoculated plants which were similar in number.

The interaction between phosphorus and variety on number of leaves in 2016 was significant and is presented in Figure 4.13C. Number of leaves increased significantly with each increase in P rate in IT99K-573-1-1. There was no significant difference between the values recorded for IT93K-452-1 at 0 and 20 kg P ha^{-1}. Application of P at 40 kg P ha^{-1} resulted in greater number of leaves than at 20 kg P ha^{-1}. TVX-3236 plants to which 20 and 40 kg P ha^{-1} were applied produced similar number of leaves which were significantly greater than those of the unfertilized plants.

iii. Vine length

The vine length of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2015 to 2017 is presented in Table 4.7. There was no significant difference among the vine length of the inoculated and uninoculated plants in 2015.nd 2016. However in 2016, the longest vine was recorded in plants inoculated with BR 3262 but the value was only significantly higher than those recorded in plants inoculated with BR 3267. In 2017, plants inoculated with BR 3262 produced significantly longer vines than the other inoculation treatments which produced similar vine length.

In 2015, plants to which 40 kg P ha^{-1} were applied produced significantly longer vines (239.37 cm) than the unfertilized plants (128.50 cm); there was no significant difference between the vine length of the plants that received 20 and 40 kg P ha^{-1}. In 2016, vine length increased significantly with increase in P rate with the highest and lowest values recorded in plants that received 40 kg P ha^{-1} and 0 kg P ha^{-1} respectively. In 2017, application of 20 and 40 kg P ha^{-1} resulted in the production of similar vine length but the values were significantly greater than that of the unfertilized plants.

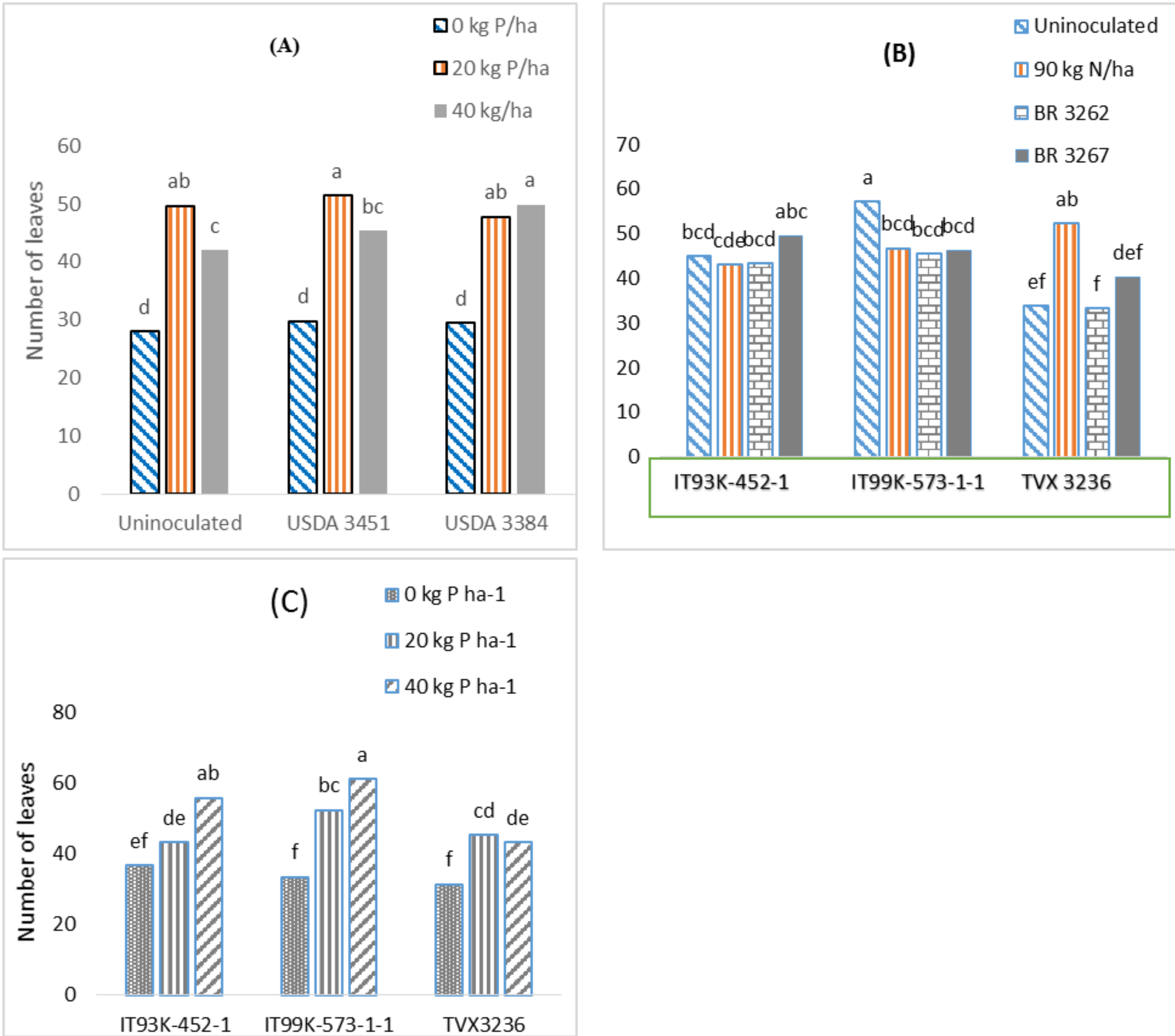


Fig 4.13: Interaction effects of (A): rhizobia inoculation and phosphorus on number of leaves of cowpea in 2015, (B): rhizobia inoculation and variety on number of leaves of cowpea in 2016, (C): Phosphorus and variety on number of leaves of cowpea in 2016

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

Table 4.7: Effects of rhizobia inoculation and phosphorus application on vine length (cm) of cowpea varieties

	Cropping seasons		
	2015	**2016**	**2017**
Treatments			
Inoculation (I)			
Uninoculated	160.89a	106.41ab	60.53b
USDA 3451	150.67a		57.37b
USDA 3384	225.07a		
BR 3262		112.16a	68.07a
BR 3267		95.40b	57.80b
90 kg N ha^{-1}		106.34ab	59.52b
M S D	99.31	13.00	5.59
Phosphorus (P)			
0 kg ha^{-1}	128.5b	79.11c	52.31b
20 kg ha^{-1}	168.70ab	109.83b	64.38a
40 kg ha^{-1}	239.37a	126.01a	65.28a
M S D	99.31	11.26	4.33
Variety (V)			
IT93K-452-1	213.56a	105.94a	66.44a
IT99K-573-1-1	180.15ab	114.58a	65.74a
TVX-3236	142.93b	94.43b	49.79b
M S D	69.31	11.26	4.33
Interactions			
I × P	NS	**	NS
I × V	NS	**	NS
P × V	NS	*	NS
I × P × V	NS	*	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, *, **-significant at P=0.05 and 0.01 respectively, NS- not significant at P=0.05

In 2015, IT99K-573-1-1 had the longest vine which was however similar in value to that of IT93K-452-1 but significantly greater than the vine length of TVX-3236. In 2016 and 2017, IT93K-452-1 and IT99K-573-1-1 produced similar vine length and both were significantly longer than the vine length of TVX-3236 plants.

Inoculation x phosphorus, inoculation x variety, phosphorus x variety and inoculation x phosphorus x variety were all significant in 2016 ($P \leq 0.01$; 0.05). Vine length increased significantly with increase in P rates in the uninoculated plants with the longest vine obtained at 40 kg P ha^{-1} and the shortest at 0 kg P ha^{-1} (Figure 4.14A). Similar trend was observed in plants to which 90 kg N ha^{-1} was applied but the plants that received 20 kg P ha^{-1} and the unfertilized plants produced similar vine length. The vine length of plants inoculated with BR 3262 increased significantly with P application with a peak at 20 kg P ha^{-1} beyond which a non-significant decline at 40 kg P ha^{-1}. Though P increased the vine length in plants inoculated with BR 3267, the increase was not significant (Figure 4.14A).

The interaction between rhizobia inoculation and variety on vine length at 7 WAS is presented in Figure 4.14B. IT93K-452-1 plants responded to all the inoculation treatments similarly in respect of vine length as there was no significant difference between the vine length of inoculated and uninoculated plants as well as plants that received 90 kg N ha^{-1}. Uninoculated IT99K-573-1-1 plants produced significantly longer vines than the inoculated plants and those that received 90 kg N ha^{-1} which were similar. TVX-3236 plants that received 90 kg N ha^{-1} had significantly longer vines than the uninoculated and inoculated plants which produced similar vine lengths.

The interaction between phosphorus and variety on vine length revealed that the vine length of IT93K-573-1-1 plants increased with increase in P rates with the longest and shortest vines recorded at 40 kg P ha^{-1} and unfertilized plants respectively (Figure 4.14C). The same trend was observed in IT93K-452-1 plants, however, there was no significant difference between the vine length of the unfertilized plants and those that received 20 kg P ha^{-1}. P application significantly increased the vine length of TVX-3236 plants but the longest vine was obtained at 20 kg P ha^{-1} application.

The interaction between rhizobia inoculation, phosphorus and variety on vine length in 2016 is presented in Figure 4.15. The optimum P and inoculation treatment combination depended on the cowpea variety. In IT93K-452-1, uninoculated + 40 kg P ha^{-1} combination had the longest vine, the value of which was however similar to those of 90 kg N + 40 kg P ha^{-1}, BR 3262 + 20 kg P ha^{-1} and BR 3267 + 40 kg P ha^{-1} combination. In IT99K-573, uninoculated + 40 kg P ha^{-1} combination again had the longest vine, the value of which was however significantly greater than those of other treatment combinations except that of 90 kg N + 40 kg P ha^{-1} . The longest TVX-3236 vine resulted from a combination of 90 kg N + 40 kg P ha^{-1}. The value was however similar to those recorded in 90 kg N + 20 kg P ha^{-1}.

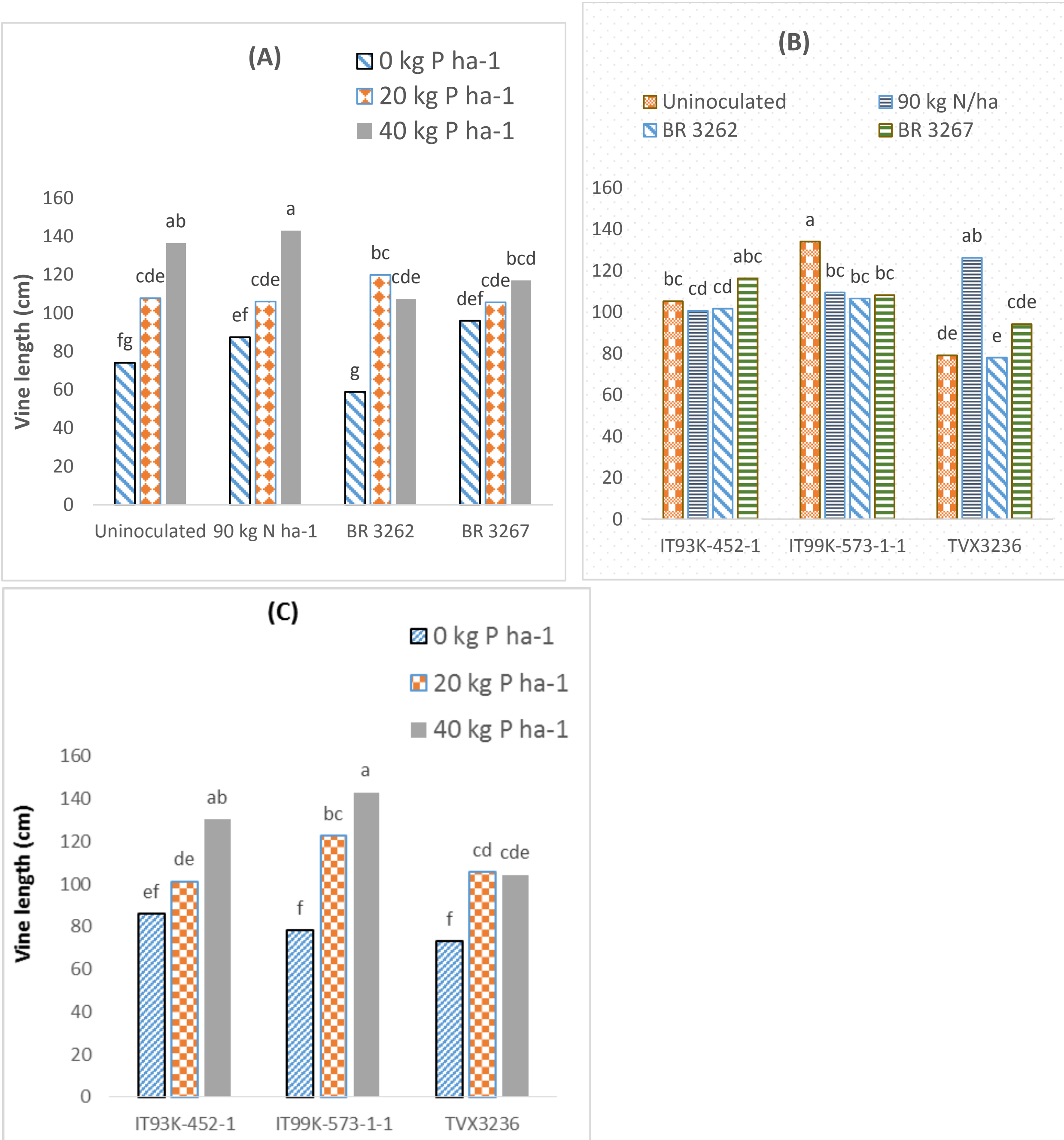


Figure 4.14: Interaction effects of (A) rhizobia inoculation and phosphorus (B) rhizobia inoculation and variety (C) phosphorus and variety on vine length of cowpea in 2016

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

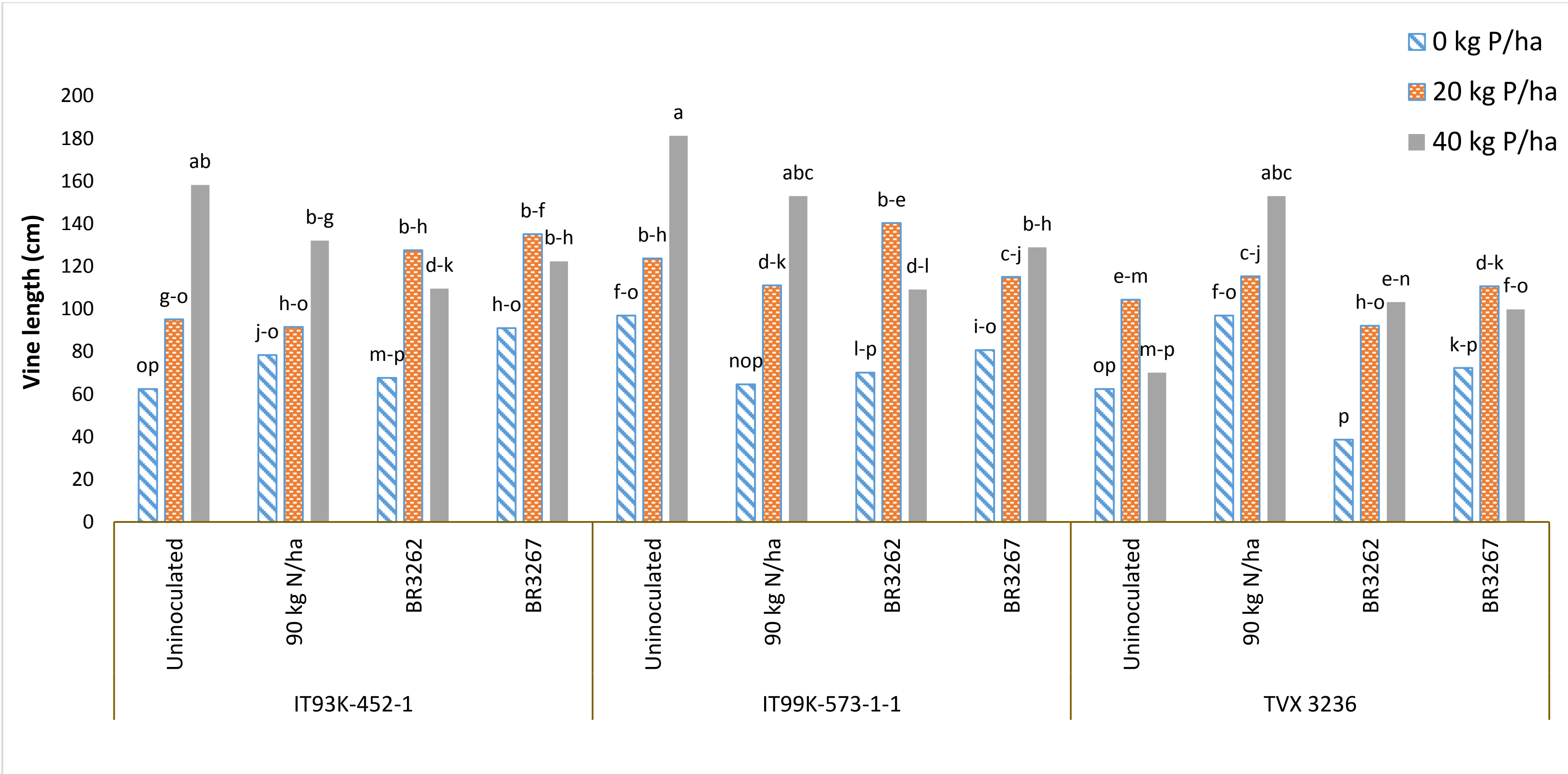


Figure 4.15: Interaction effects of rhizobia inoculation, phosphorus and variety on vine length of cowpea in 2016

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

iv. Stem diameter

The stem diameter of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2015-2017 is presented in Table 4.8. There was no significant difference between the stem diameter of the inoculated and uninoculated plants in 2015. In 2016, plants inoculated with BR 3262 rhizobia strain produced the biggest stem similar to the values obtained in plants fertilized with 90 kg N ha^{-1}. Plants inoculated with BR 3267 strain produced the thinnest stem and the value was similar to the stem diameter recorded in the uninoculated plants. In 2017, plants inoculated with BR 3262 rhizobia strain produced the biggest stem but the value was similar to the value recorded in the uninoculated plants.

Plants that received 20 and 40 kg P ha^{-1} produced similar stem diameter which were significantly larger than the values recorded in unfertilized plants in 2015 and 2016 but in 2017, the order of performance in respect of stem diameter was 40 kg P ha^{-1} > 20 kg P ha^{-1} > 0 kg P ha^{-1}.

The order of performance in respect of the stem diameter among the varieties was IT93K-452-1 > IT99K-573-1-1 > TVX-3236 however in 2016 and 2017, the difference between IT99K-573-1-1 and IT93K-452-1 was not statistically significant.

The interaction between rhizobia inoculation and variety on vine length was significant in 2015. There was no significant difference between the stem diameter of inoculated and uninoculated IT93K-452-1 and IT99K-573-1-1 plants. Significantly larger stems were produced when TVX-3236 plants were inoculated with USDA 3451 strain, than when uninoculated (Figure 4.16A). The stem diameter recorded when plants were inoculated with USDA 3384 was similar to that for USDA 3451 and the uninoculated.

Table 4.8: Effects of rhizobia inoculation and phosphorus application on stem diameter (cm) of cowpea varieties

	Cropping seasons		
	2015	**2016**	**2017**
Treatments			
Inoculation (I)			
Uninoculated	0.94a	1.14bc	0.50ab
USDA 3451	0.93a		0.42c
USDA 3384	0.93a		
BR 3262		1.28a	0.53a
BR 3267		1.11c	0.48b
90 kg N ha^{-1}		1.25ab	0.49b
M S D	0.05	0.13	0.04
Phosphorus (P)			
0 kg ha^{-1}	0.82b	0.93b	0.42c
20 kg ha^{-1}	0.98a	1.28a	0.50b
40 kg ha^{-1}	1.00a	1.37a	0.53a
M S D	0.05	0.11	0.03
Variety (V)			
IT93K-452-1	1.01a	1.22a	0.49a
IT99K-573-1-1	0.85c	1.26a	0.51a
TVX-3236	0.93b	1.10b	0.44b
M S D	0.05	0.11	0.03
Interactions			
I × P	NS	**	NS
I × V	*	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test.MSD-Minimum significant difference, *, **-significant at P=0.05 and 0.01 respectively, NS- not significant at P=0.05

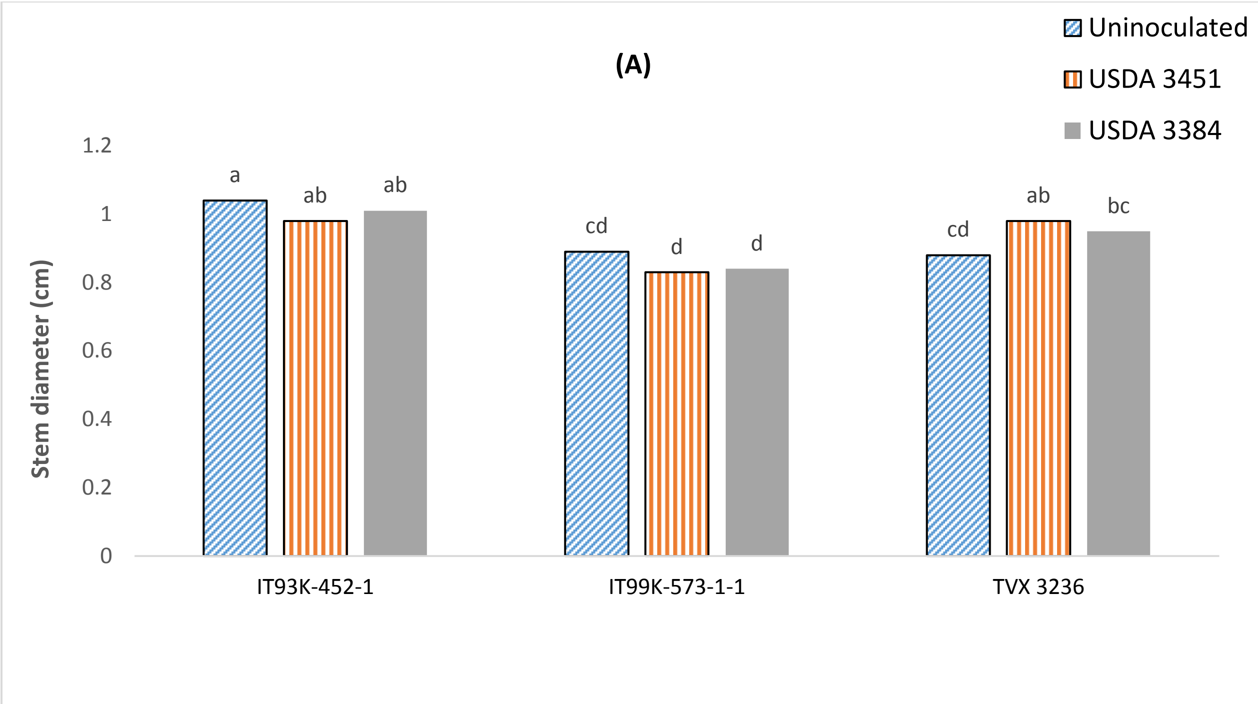
The interaction between rhizobia inoculation and phosphorus on stem diameter in 2016 is presented in Figure 4.16B. In uninoculated plants and those inoculated with BR 3262, there was no significant difference between the plants that received 20 kg P ha^{-1} and 40 kg P ha^{-1} but both produced significantly larger stems than the unfertilized plants. Plants that received 90 kg N ha^{-1} produced significantly larger stem when fertilized with 40 kg P ha^{-1} than when fertilized with 20 and 0 kg P ha^{-1}. Plants inoculated with BR 3267 rhizobia strain had significantly larger stem when fertilized with 40 kg P ha^{-1} than 0 kg P ha^{-1}.

4.1.2.2 Physiological attributes

i. Chlorophyll content

The chlorophyll content of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2015 is presented in Table 4.9. Leaves from plants inoculated with USDA 3451 had significantly higher concentration of chlorophyll a than those from plants inoculated with USDA 3384 and the uninoculated plant which had similar concentration. However, there was no significant difference between the inoculated and uninoculated plants in respect of chlorophyll b and total chlorophyll content.

Chlorophyll a, b and total chlorophyll concentrations increased significantly with increase in P application with the highest and lowest concentration recorded in plants that received 40 kg P ha^{-1} and 0 kg P ha^{-1} respectively. IT99K-573-1-1 and TVX-3236 plants had similar leaf chlorophyll concentration which were significantly higher than what was recorded in IT93K-452-1 leaves. The interaction between rhizobia inoculation and phosphorus on chlorophyll a content is presented in Figure 4.17. The chlorophyll a content of uninoculated plants and those inoculated with USDA 3451 increased significantly with increase in P application. In plants inoculated with USDA 3384,



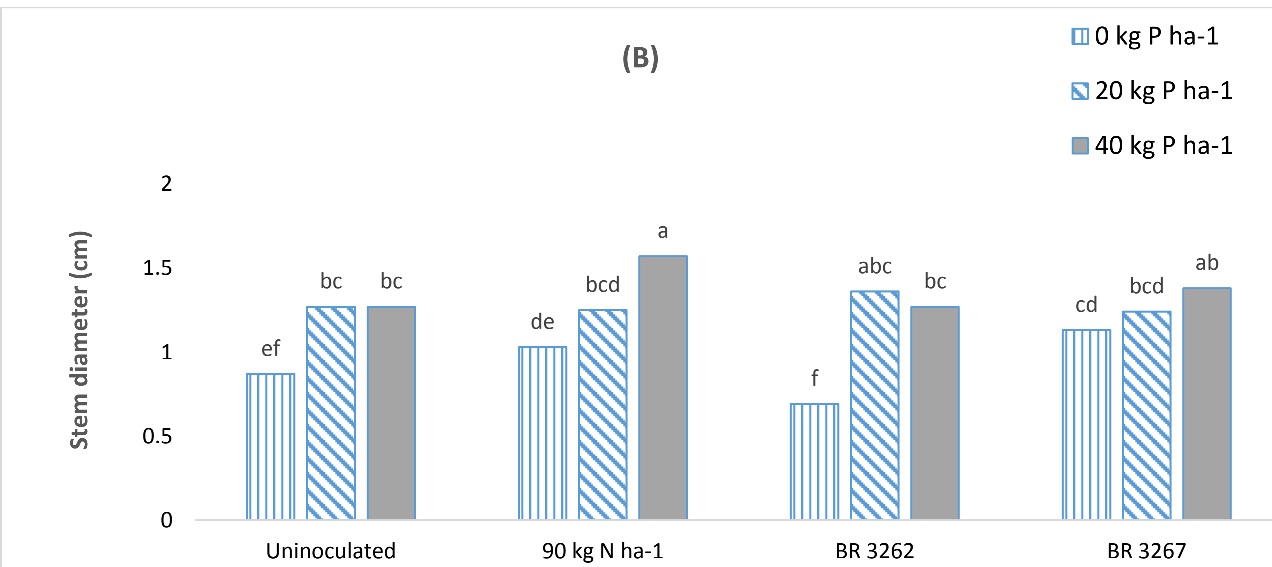


Figure 4.16: Interaction effects of (A): rhizobia inoculation and variety on stem diameter of cowpea in 2015 (B): rhizobia inoculation and phosphorus on stem diameter of cowpea in 2016

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

Table 4.9: Effects of rhizobia inoculation and phosphorus application on leaf chlorophyll content of cowpea varieties in 2015

Treatments	Chlorophyll a (µg/g)	Chlorophyll b (µg/g)	Total chlorophyll (µg/g)
Inoculation (I)			
Uninoculated	11.55b	6.74a	18.29a
USDA 3451	11.93a	6.68a	18.61a
USDA 3384	11.61b	6.79a	18.40a
M S D	0.19	0.28	0.44
Phosphorus (P)			
0 kg ha^{-1}	11.02c	6.21c	17.23c
20 kg ha^{-1}	11.92b	6.84b	18.76b
40 kg ha^{-1}	12.15a	7.16a	19.31a
M S D	0.19	0.28	0.44
Variety (V)			
IT93K-452-1	11.10b	6.49b	17.59b
IT99K-573-1-1	12.03a	6.87a	18.84a
TVX-3236	11.96a	6.85a	18.87a
M S D	0.19	0.28	0.44
Interactions			
I × P	**	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets are significantly different at P=0.05 using Waller-Duncan K-ratio T-test , MSD-Minimum significant difference , **significant at P=0.01, NS- not significant at P=0.05

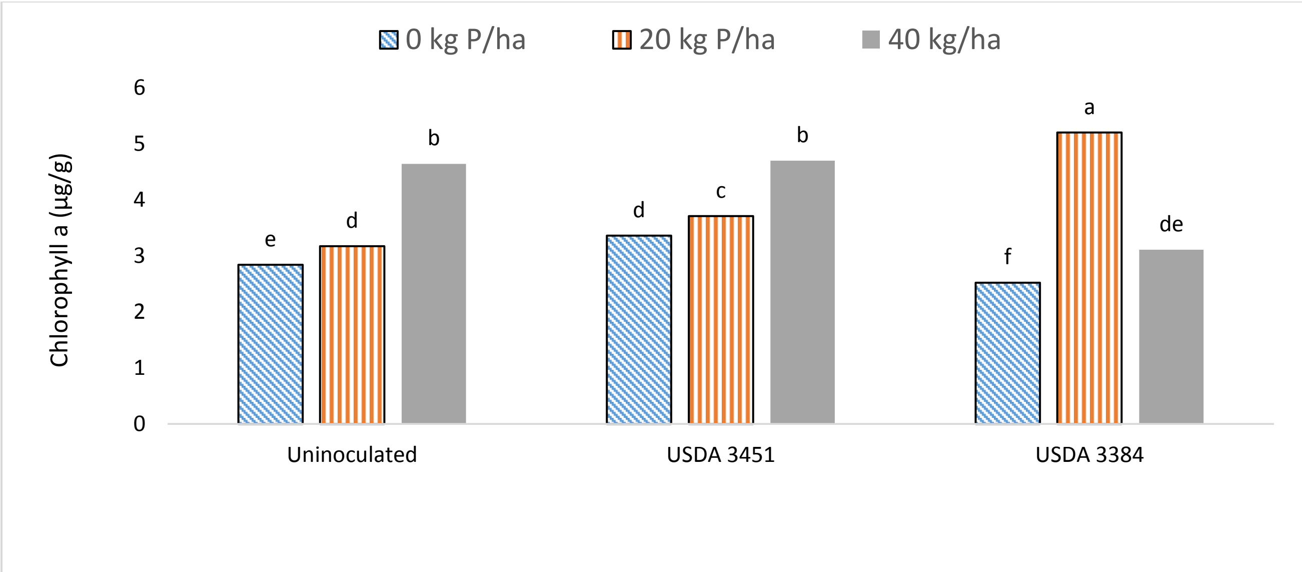


Figure 4.17: Interaction effects of rhizobia inoculation and phosphorus application on chlorophyll a concentration of cowpea leaves in 2015

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

plants that received 20 kg P ha^{-1} had significantly higher chlorophyll a concentration than 0 and 40 kg P ha^{-1}.

The chlorophyll content of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2016 is presented in Table 4.10. Plants fertilized with 90 kg N ha^{-1} had significantly higher chlorophyll a content in their leaves than the uninoculated plants. There was however no significant difference between the chlorophyll a content of the inoculated plants and plants that received 90 kg N ha^{-1}. The uninoculated plants however had significantly higher chlorophyll b content than the inoculated plants but similar to what was recorded in plants fertilized with 90 kg N ha^{-1}. Plants fertilized with 90 kg N ha^{-1} had significantly higher total chlorophyll content than what was recorded in all the inoculated plants but the value was similar to what was recorded in the uninoculated plants.

Plants fertilized with 20 and 40 kg P ha^{-1} had similar chlorophyll a content which were significantly higher than the concentration found in the unfertilized plants. Plants fertilized with 40 kg P ha^{-1} had significantly higher chlorophyll b concentrations than what was obtained in the unfertilized plants but similar to the concentration obtained in plants fertilized with 20 kg P ha^{-1}. There was also no significant difference between the chlorophyll b concentration obtained in unfertilized plants and those that received 20 kg P ha^{-1}. The order of performance in respect of total chlorophyll was 40 kg P ha^{-1} > 20 kg P ha^{-1} > 0 kg P ha^{-1}

There was no significant difference between the chlorophyll a concentration observed in the three varieties. However, IT99K-573-1-1 plants had significantly higher chlorophyll b concentration than IT93K-452-1 and TVX-3236. IT99K-573-1-1 leaves had significantly higher total chlorophyll content than the value recorded in IT93K-452-1 plants however, this was similar to what was recorded in TVX-3236 plants.

Table 4.10: Effects of rhizobia inoculation and phosphorus application on leaf chlorophyll content of cowpea varieties in 2016

Treatments	Chlorophyll a µg/g	Chlorophyll b µg/g	Total chlorophyll µg/g
Inoculation (I)			
Uninoculated	17.34b	9.70a	27.04ab
90 kg ha^{-1}	21.46a	8.18a	29.64a
BR 3262	19.41ab	5.10b	24.51b
BR 3267	19.89a	4.26b	24.15b
M S D (0.05)	2.26	1.97	2.90
Phosphorus (P)			
0 kg ha^{-1}	16.95b	5.80b	22.76c
20 kg ha^{-1}	19.99a	6.51ab	26.50b
40 kg ha^{-1}	21.64a	8.12a	29.76a
MSD	1.96	1.71	2.51
Variety (V)			
IT93K-452-1	18.84a	5.38b	24.22b
IT99K-573-1-1	20.40a	7.94a	28.34a
TVX-3236	19.34a	7.11b	26.45ab
MS D			
Interactions			
I × P	NS	NS	NS
I × V	**	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, **significant at P=0.01, Ns- not significant at P=0.05

The interaction between rhizobia inoculation and variety on chlorophyll a is presented in Figure 4.18. In IT93K-452-1 variety, plants inoculated with BR 3262 strain had the highest chlorophyll a concentration but the value was similar to what was obtained in plants fertilized with 90 kg N ha^{-1}. The uninoculated plants had the significantly lowest chlorophyll a concentration among the IT93K-452-1 plants. For IT99K-573-1-1 plants however, there was no significant difference between the chlorophyll a concentration of the inoculated, uninoculated and plants that received 90 kg N ha^{-1}. In TVX-3236 variety, the uninoculated plants had the highest chlorophyll a concentration followed by plants that received 90 kg N ha^{-1} which was similar to the value recorded in plants inoculated with BR 3267. The least concentration was recorded in plants inoculated with BR 3262.

The chlorophyll content of the leaves of the different cowpea varieties in response to rhizobia inoculation and phosphorus application in 2017 is presented in Table 4.11. Plants inoculated with BR 3262 had the highest chlorophyll a and total chlorophyll contents which were however only significantly greater than those of the uninoculated plants and there was no significant differences among the concentrations found in uninoculated plants, plants fertilized with 90 kg N ha^{-1} and plants inoculated with BR 3267 and USDA 3451. Plants fertilized with 90 kg N ha^{-1} had significantly higher chlorophyll b content than uninoculated and USDA 3451 plants but the value was similar to what was recorded in plants inoculated with BR 3262 and BR 3267. The uninoculated plants and those inoculated with BR 3451 had the lowest chlorophyll b content which were significantly lower than the value recorded in plants that received 90 kg N ha^{-1} but similar to the value recorded in plants inoculated with BR 3262 and BR 3267.

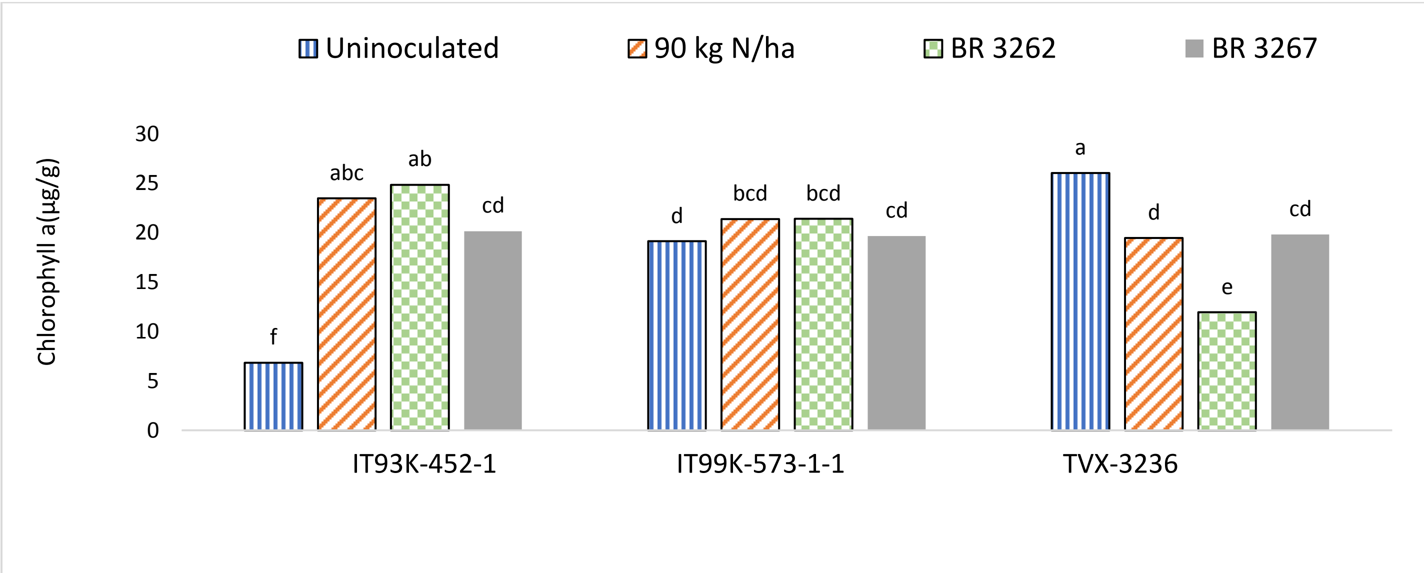


Figure 4.18: Interaction effects of rhizobia inoculation and variety on chlorophyll a concentration of cowpea leaves in 2016

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

Table 4.11: Effects of rhizobia inoculation and phosphorus application on leaf chlorophyll content of cowpea varieties in 2017

Treatments	Chlorophyll a (µg/g)	Chlorophyll b (µg/g)	Total chlorophyll (µg/g)
Inoculation (I)			
Uninoculated	11.96b	7.97b	19.93b
90 kg N ha^{-1}	12.26ab	8.60a	20.86ab
BR 3262	12.90a	8.17ab	21.07a
BR 3267	12.21ab	8.14ab	20.36ab
USDA 3451	12.12b	8.08b	20.20b
MSD	0.73	0.49	1.22
Phosphorus (P)			
0 kg ha^{-1}	11.14b	7.43b	18.56b
20 kg ha^{-1}	13.03a	8.68a	21.71a
40 kg ha^{-1}	12.71a	8.47a	21.18a
MSD	0.57	0.38	0.95
Variety (V)			
IT93K-452-1	12.06a	8.04a	20.09a
IT99K-573-1-1	12.30a	8.20a	20.50a
TVX-3236	12.52a	8.34a	20.86a
MSD	0.57	0.38	0.95
Interactions			
I × P	NS	NS	NS
I × V	**	NS	NS
P × V	NS	NS	NS
I × V × P	NS	NS	NS

Means followed by different alphabets within a column are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference. **significant at P=0.01, Ns- not significant at P=0.05

Plants that received 20 and 40 kg P ha^{-1} had similar chlorophyll a, b and total chlorophyll content which were significantly higher than the value recorded in the unfertilized plants. There was no significant difference between the chlorophyll a, b and total chlorophyll content of the three varieties.

The interaction between rhizobia inoculation and variety on chlorophyll a concentration in 2017 is presented in Figure 4.19. The chlorophyll a concentration of uninoculated IT93K-452-1 plants as well as those fertilized with 90 kg N ha^{-1} and plants inoculated with BR 3262 and USDA 3451 were similar but significantly higher than the concentration recorded in plants inoculated with BR 3267. In IT99K-573-1-1 variety, plants inoculated with BR 3262 had significantly higher chlorophyll a concentration than the remaining treatment combinations except BR 3267 inoculated plants. The uninoculated plants had the lowest chlorophyll a concentration but the value was similar to what was recorded in plants inoculated with USDA 3451. In TVX-3236 variety, plants that received 90 kg N ha^{-1} had significantly higher chlorophyll a concentration than plants inoculated with USDA 3451.

ii. Crop growth rate

The crop growth rate of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2015 is presented in Table 4.12. There was no significant difference between the crop growth rate of inoculated and uninoculated plants at the different sampling periods. Plants fertilized with 20 and 40 kg P ha^{-1} had similar growth rates that were significantly higher than the growth rate of the unfertilized plants throughout the growth period. At the early growth stage (3-5 WAS), IT93K-452-1 had a significantly higher growth rate than IT99K-573-1-1 and TVX-3236

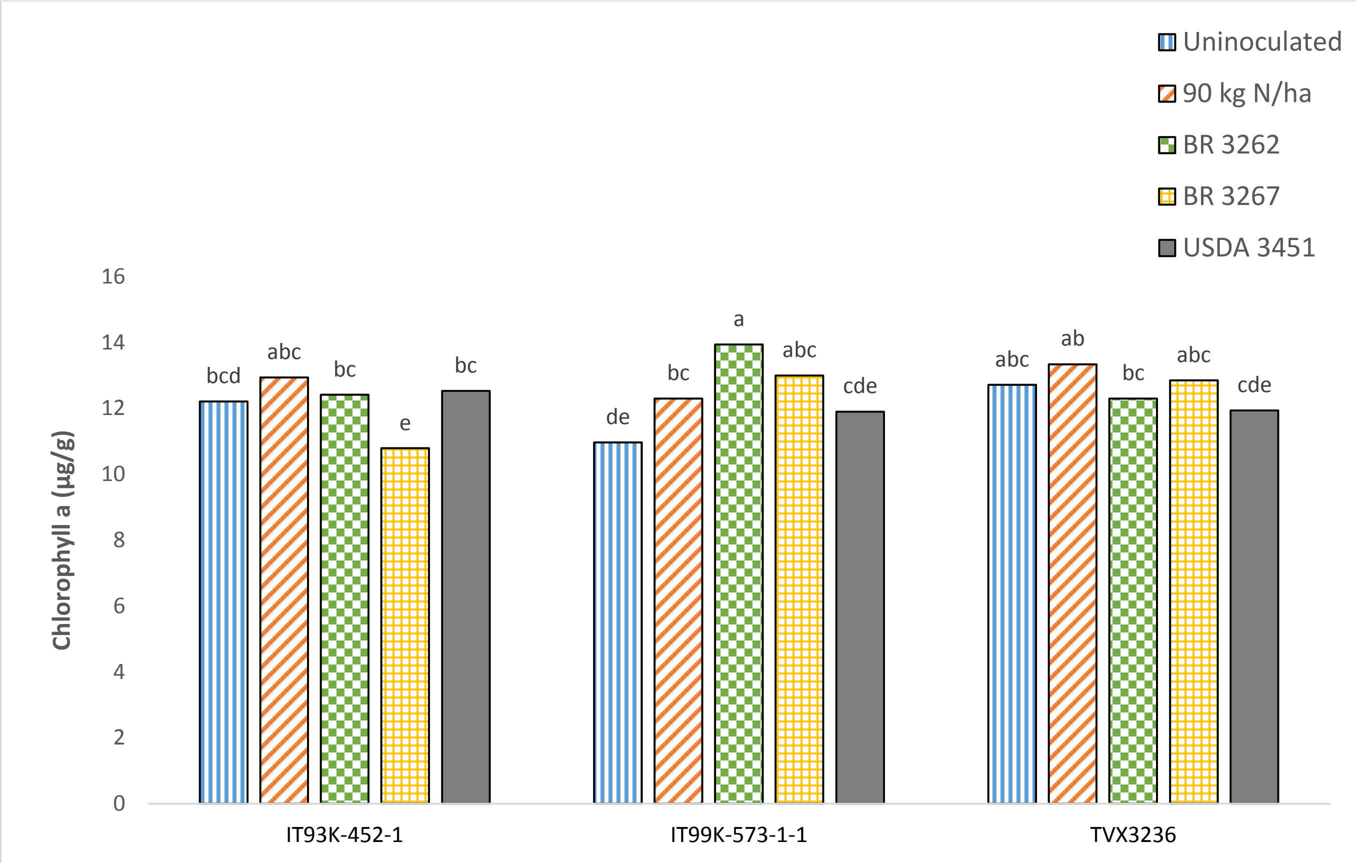


Figure 4.19: Interaction effects of rhizobia inoculation and variety on chlorophyll a concentration of cowpea leaves in 2017

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

Table 4.12: Effects of rhizobia inoculation and phosphorus application on crop growth rate (g/m^2/day) of cowpea varieties in 2015

	Growth stages			
Treatments	Early	Mid	Late	Mean
Inoculation (I)				
Uninoculated	5.10a	9.79a	4.11a	6.33a
USDA 3451	3.75a	10.37a	2.00a	5.37a
USDA 3384	4.53a	10.60a	2.44a	5.86a
MSD	1.92	1.78	1.53	1.09
Phosphorus (P)				
0 kg ha^{-1}	2.65b	5.90b	1.19b	3.25b
20 kg ha^{-1}	4.92a	11.80a	3.99a	6.90a
40 kg ha^{-1}	5.80a	13.04a	3.36a	7.40a
MSD	1.92	1.78	1.53	1.09
Variety (V)				
IT93K-452-1	4.58a	11.49a	3.44a	6.51a
IT99K-573-1-1	4.34a	9.67b	2.48a	5.50a
TVX-3236	4.45a	9.58b	2.62a	5.55a
MSD	1.92	1.78	1.53	1.09
Interactions				
I × P	NS	NS	NS	NS
I × V	NS	NS	NS	NS
P × V	NS	NS	NS	NS
I × V × P	NS	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, NS- not significant at P=0.05

plants which were similar. At later stage however, there was no significant difference between the growth rate of the three varieties.

The crop growth rate of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2016 is presented in Table 4.13. There was no significant difference between the crop growth rate of inoculated and uninoculated plants at the different sampling periods. At the early growth stage (3-5 WAS) plants fertilized with 20 and 40 kg P ha^{-1} had similar growth rates that were significantly higher than the growth rate of the unfertilized plants. At mid growth stage (5-7 WAS) however, the order of performance in respect of crop growth rate was 40 kg P ha^{-1}> 20 kg P ha^{-1} > 0 kg P ha $^{-1}$. The same order was recorded at late growth stage (7-9 WAS) but there was no significant difference between the growth rate of plants that received 20 and 40 kg P ha^{-1} as well as between the unfertilized plants and plants fertilized with 20 kg P ha^{-1}. Across the growth stages (mean crop growth rate), the growth rate of plants fertilized with 20 and 40 kg P ha^{-1} were similar but significantly higher than the growth rate of the unfertilized plants.

IT93K-452-1 and IT99K-573-1-1 had similar growth rates that were significantly higher than the growth rate of TVX-3236 plants at the early growth stage in 2016. At mid-growth stage however, there was no significant difference between the growth rates of the three varieties. At late growth stage, the growth rate of IT93K-452-1 and IT99K-573-1-1 plants were similar and significantly lower than the growth rate of TVX-3236 plants. Across the growth stages, the growth rate of TVX-3236 plants were significantly higher than the growth rate of IT93K-452-1 and IT99K-573-1-1 which were similar. Generally, the growth rate increased up to mid-growth stage after which there was a decline.

Table 4.13: Effects of rhizobia inoculation and phosphorus application on crop growth rate (g/m^2/day) of cowpea varieties in 2016

	Growth stages			
Treatments	Early	Mid	Late	Mean
Inoculation (I)				
Uninoculated	3.33a	6.02a	4.97a	4.78a
90 kg N ha^{-1}	3.49a	6.64a	4.83a	4.99a
BR 3262	2.94a	5.39a	4.36a	4.23a
BR 3267	3.25a	6.08a	4.02a	4.45a
MSD	0.78	1.39	3.55	1.18
Phosphorus (P)				
0 kg ha^{-1}	2.63b	4.27c	2.56b	3.15b
20 kg ha^{-1}	3.59a	6.10b	5.40ab	5.03a
40 kg ha^{-1}	3.54a	7.74a	5.68a	5.65a
MSD	0.68	1.21	3.07	1.02
Variety (V)				
IT93K-452-1	3.31a	6.00a	1.68b	3.66b
IT99K-573-1-1	3.97a	5.97a	3.10b	4.35b
TVX-3236	2.48b	6.13a	8.86a	5.82a
MSD	0.68	1.21	3.07	1.02
Interactions				
I × P	NS	NS	NS	NS
I × V	NS	**	NS	NS
P × V	NS	*	NS	NS
I × V × P	NS	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, *, **- significant at P=0.05 and 0.01 respectively, NS- not significant at P=0.05, The interaction between rhizobia inoculation and variety on crop growth rate at mid-growth stage in 2016 is presented in Figure 4.20A. There was no significant effect of inoculation on growth rate of IT93K-452-1 plants in the mid-growth stage. Furthermore, there was no significant difference between the growth rate of inoculated and 90 kg N ha^{-1} fertilized IT99K-573-1-1 plants. However, when IT99K-573-1-1 plants were uninoculated, the highest growth rate was recorded but the value was similar to the growth rate of plants fertilized with 90 kg N ha^{-1}. In TVX-3236 variety however, plants fertilized with 90 kg N ha^{-1} had significantly higher growth rate than the inoculated and uninoculated plants which were similar.

The interaction between phosphorus and variety on crop growth rate at mid-growth stage was significant. The result revealed that IT93K-452-1 plants that received 40 kg P ha^{-1} had significantly higher growth rate than those that received 20 kg P ha^{-1} and the unfertilized plants which were similar (Figure 4.20B). In IT93K-573-1-1 variety however, plants that received 20 and 40 kg P ha^{-1} had similar growth rate that were significantly higher than the growth rate of the unfertilized plants. There was no significant difference between the growth rate of the fertilized and unfertilized TVX-3236 plants.

The crop growth rate of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2017 is presented in Table 4.14. There was no significant difference between the growth rate of the uninoculated, inoculated and 90 kg N ha^{-1} fertilized plants at early-mid growth stage. At late growth stage however, the uninoculated plants and 90 kg N ha^{-1} fertilized plants had significantly higher growth rate (7.77 and 7.66 g/m^2/day respectively) than plants inoculated with BR 3267 and USDA 3451 but similar to the value recorded in plants inoculated with BR 3262.

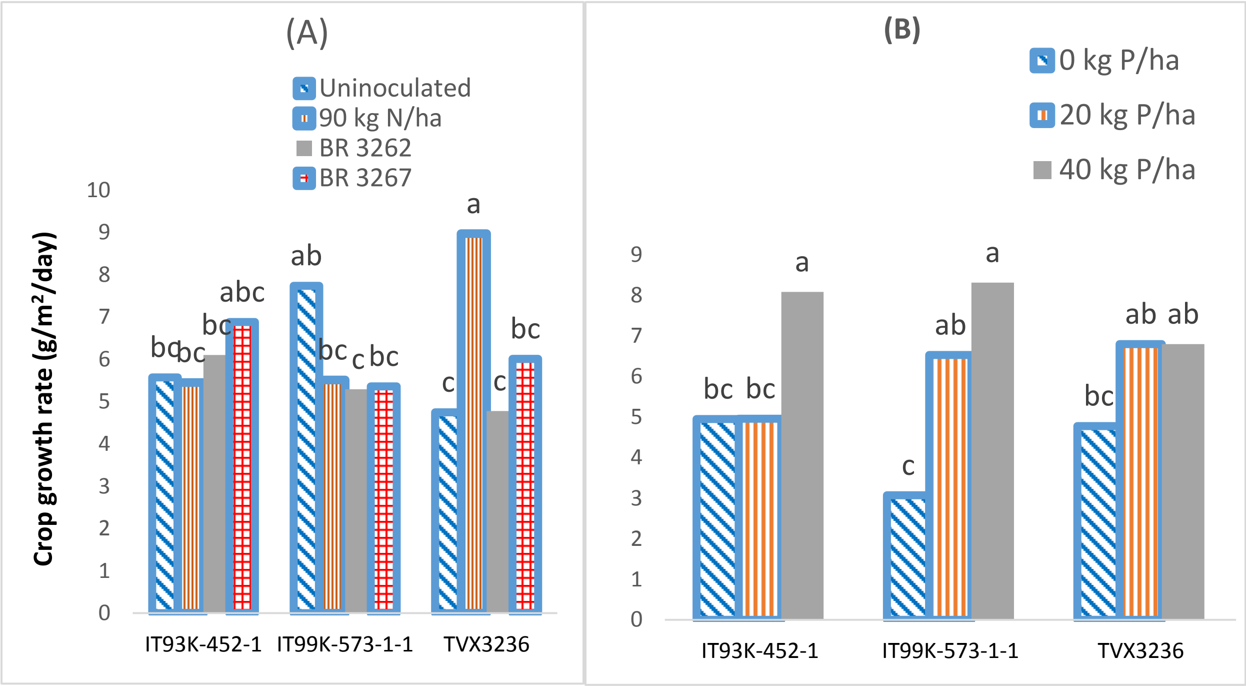


Figure 4.20: Interaction effects of (A) rhizobia inoculation and variety (B) phosphorus and variety on crop growth rate at mid-growth stage in 2016

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

Plants inoculated with BR 3267 had the lowest growth rate (4.74 $g/m^2/day$) but the value was similar to the growth rate of plants inoculated with USDA 3451 (6.01 $g/m^2/day$). Across the growth stages (mean crop growth rate), there was no significant difference between the growth rate of the uninoculated, inoculated and 90 kg N ha^{-1} fertilized plants.

Plants fertilized with 20 and 40 kg P ha^{-1} had similar growth rate that was significantly higher than the growth rate of the unfertilized plants at early growth stage. At mid-growth stage, plants fertilized with 40 kg P ha^{-1} had the highest growth rate (12 $g/m^2/day$) which was significantly higher than the growth rate of plants fertilized with 20 kg P ha^{-1} and the unfertilized plants (8.55 and 7.74 $g/m^2/day$ respectively). At late growth stage however, the order of growth rate was 20 kg P ha^{-1} > 40 kg P ha^{-1} > 0 kg P ha^{-1}. Across the different stages, the order of growth rate was 40 kg P ha^{-1} > 20 kg P ha^{-1} > 0 kg P ha^{-1}.

At early growth stage, IT93K-452-1 and IT99K-573-1-1 plants had similar growth rates that were significantly higher than the growth rate of TVX-3236 plants. At mid-growth stage however, IT99K-573-1-1 and TVX-3236 had similar growth rates plants that were significantly higher than the growth rate of IT93K-452-1 plants. At late growth stage, the growth rate of IT99K-573-1-1 plants was the highest and it was similar to the growth rate of TVX-3236 plants but significantly higher than the growth rate of IT93K-452-1. Across the growth stages, IT93K-452-1 plants and TVX-3236 plants had similar growth rate that was significantly lower than the growth rate of IT99K-373-1-1 plants. Generally, the growth rate increased from 3-7 WAS after which there was a decline.

Table 4.14: Effects of rhizobia inoculation and phosphorus application on crop growth rate (g/m^2/day) of cowpea varieties in 2017

	Growth stages			
Treatments	Early	Mid	Late	Mean
Inoculation (I)				
Uninoculated	3.96a	8.10a	7.77a	6.61a
90 kg N ha^{-1}	4.26a	9.04a	7.66a	6.99a
BR 3262	3.52a	10.2a	6.90ab	6.87a
BR 3267	4.33a	9.93a	4.74c	6.33a
USDA 3451	4.21a	10.66a	6.01bc	6.96a
MSD	0.92	2.80	1.57	1.05
Phosphorus (P)				
0 kg ha^{-1}	2.80b	7.74bc	4.49c	5.01c
20 kg ha^{-1}	4.46a	8.55b	8.52a	7.18b
40 kg ha^{-1}	4.90a	12.46a	6.83b	8.06a
MSD	0.72	2.18	1.21	0.81
Variety (V)				
IT93K-452-1	4.40a	7.67b	5.58b	6.26b
IT99K-573-1-1	4.35a	11.20a	7.56a	7.70a
TVX-3236	3.42b	9.88a	6.70ab	6.29b
MSD	0.72	2.18	1.21	0.81
Interactions				
I × P	NS	NS	**	NS
I × V	NS	NS	NS	NS
P × V	NS	NS	NS	NS
I × V × P	NS	NS	**	NS

Means followed by different alphabets within a column are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, **- significant at P= 0.01, NS- not significant at P=0.05

The interaction effects between rhizobia inoculation and phosphorus application on crop growth rate at mid-growth stage in 2017 is presented in Figure 4.21. The result revealed that the uninoculated plants, 90 kg N ha^{-1} fertilized plants and those inoculated with USDA 3451 had their highest growth rate at 20 kg P ha^{-1} application. In plants inoculated with BR 3262, the growth rate of plants fertilized with 20 and 40 kg P ha^{-1} were similar but significantly higher than the growth rate of the unfertilized plants. In plants inoculated with BR 3267, there was no significant difference between the growth rate of the P fertilized and unfertilized plants.

The interaction between rhizobia inoculation, phosphorus application and variety on crop growth rate at late growth stage is presented in Figure 4.22. All the treatment combination had the highest growth rate at 20 kg P ha^{-1} application except IT93K-452-1-plants inoculated with BR 3267 and TVX-3236 inoculated plants in which the highest growth rate was recorded at 40 kg P ha^{-1} application.

iii. Net assimilation rate

The net assimilation rate of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2015 is presented in Table 4.15. The net assimilation rate of the inoculated plants were significantly higher than the uninoculated plants at vegetative stage. However, at reproductive stage, the uninoculated plants had the highest net assimilation rates but it was not significantly higher than the rate recorded in the inoculated plants. Plants inoculated with USDA 3384 had significantly higher mean net assimilation rate than plants inoculated with USDA 3451 and the uninoculated plants.

There was no significant difference between the net assimilation rate of P fertilized and unfertilized plants throughout the sampling period. Likewise, there was no significant difference between the net assimilation rate of the three varieties throughout the sampling period.

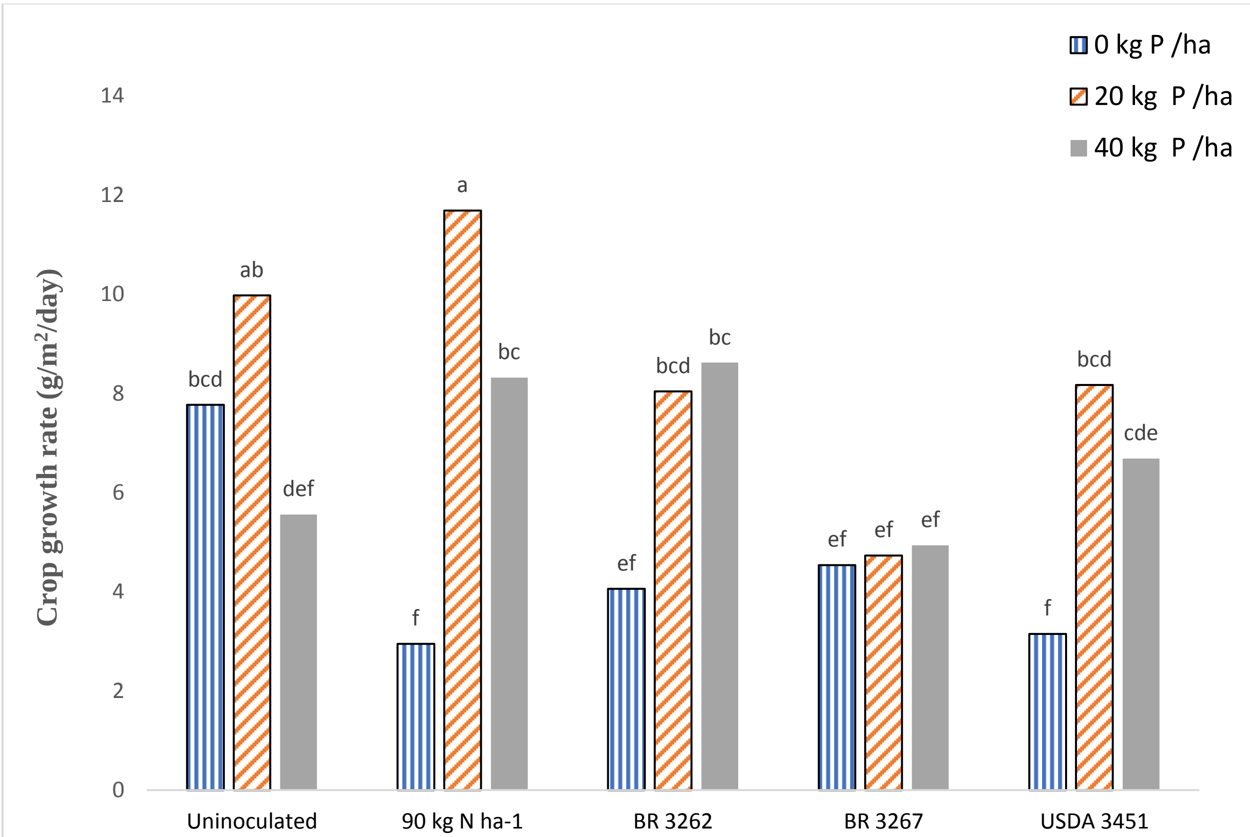


Figure 4.21: Interaction effects of rhizobia inoculation and phosphorus application on crop growth rate of cowpea at mid-growth stage in 2017

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

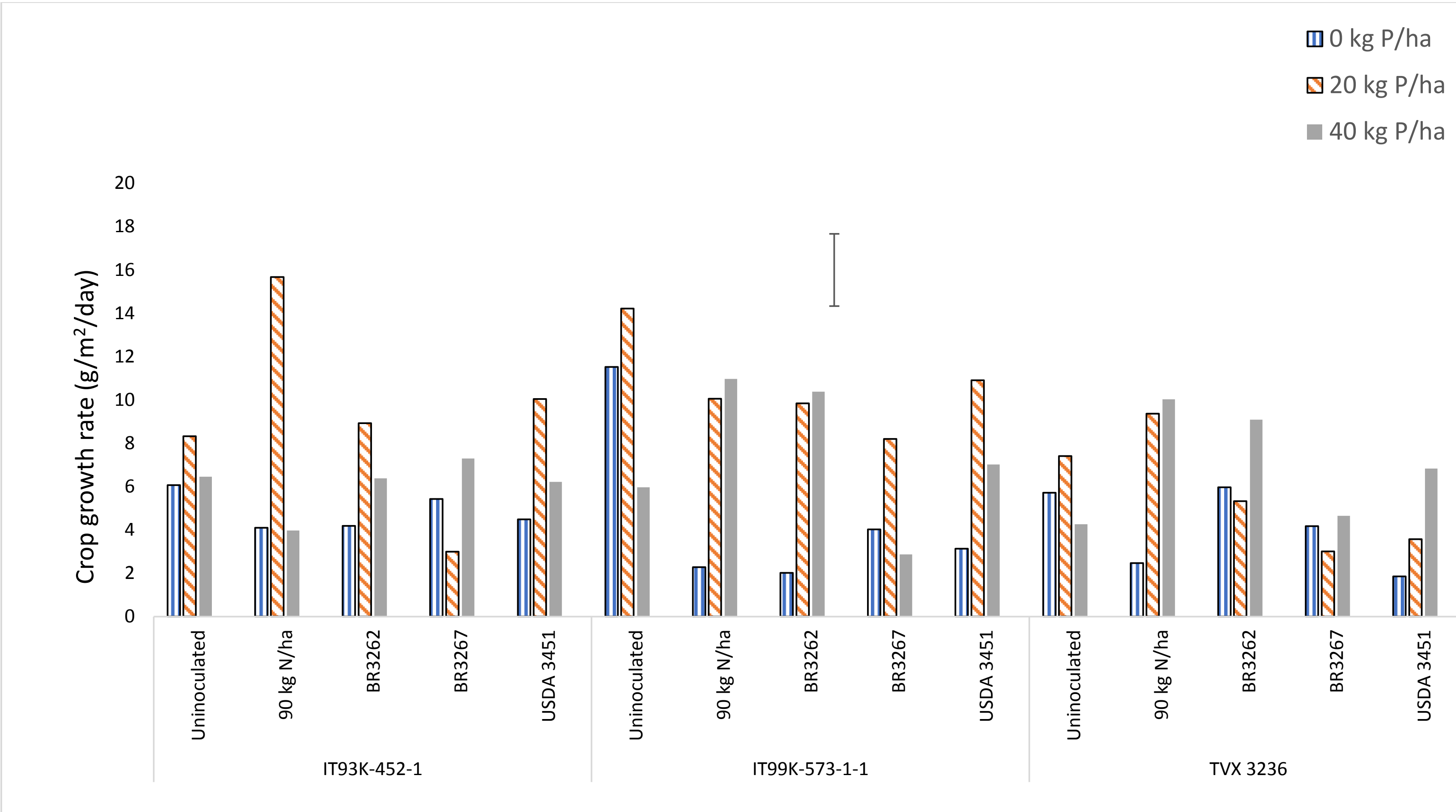


Figure 4.22: Interaction effects of rhizobia inoculation, phosphorus application and variety on crop growth rate of cowpea at mid-growth stage in 2017

I- LSD at P =0.05

Table 4.15: Effects of rhizobia inoculation and phosphorus application on net assimilation rates (g/m^2(leaf area)/day) of cowpea varieties in 2015

	Developmental stages		
Treatments	**Vegetative**	**Reproductive**	**Mean**
Inoculation (I)			
Uninoculated	4.1 x10^{-3}b	9.2 x10^{-4}a	2.5 x10^{-3}b
USDA 3451	4.8 x10^{-3}a	6.5 x10^{-4}a	2.7 x10^{-3}b
USDA 3384	5.1 x10^{-3}a	8.7 x10^{-4}a	3.0 x10^{-3}a
MSD	6.0 x10^{-4}	4.0 x10^{-4}	3.0 x10^{-3}
Phosphorus (P)			
0 kg ha^{-1}	4.6 x10^{-3}a	8.7 x10^{-4} a	2.7 x10^{-3}a
20 kg ha^{-1}	4.7 x10^{-3}a	7.5 x10^{-4} a	2.7 x10^{-3}a
40 kg ha^{-1}	4.8 x10^{-3}a	8.2 x10^{-4} a	2.8 x10^{-3}a
MSD	6.0 x10^{-4}	4.0 x10^{-4}	3.0 x10^{-3}
Variety (V)			
IT93K-452-1	4.8 x10^{-3}a	7.4 x10^{-4}a	2.7 x10^{-3}a
IT99K-573-1-1	4.5 x10^{-3}a	8.7 x10^{-4}a	2.7 x10^{-3}a
TVX-3236	4.7 x10^{-3}a	8.6 x10^{-4}a	2.8 x10^{-3}a
MSD	6.0 x10^{-4}	4.0 x10^{-4}	3.0 x10^{-3}
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × V × P	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05

The net assimilation rate of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2016 is presented in Table 4.16. There was no significant difference between the net assimilation rate of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants throughout the sampling period. There was equally no significant difference between the net assimilation rate of P fertilized and unfertilized plants throughout the sampling period. The net assimilation rate of TVX-3236 plants was only significantly higher than IT93K-452-1 and IT99K-573-1-1 plants which were similar at reproductive stage. In general, the net assimilation rate reduced with age of the plants

The net assimilation rate of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2017 is presented in Table 4.17. There was no significant difference between the net assimilation rate of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants at vegetative stage. At reproductive stage however, the uninoculated plants had significantly higher net assimilation rate than plants inoculated with BR 3267 and USDA 3451. There was no significant difference between the net assimilation rate of inoculated plants and those fertilized with 90 kg N ha^{-1}. Across the developmental stages, there was no significant difference between the net assimilation rate of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants.

There was no significant difference between the net assimilation rates of P fertilized and unfertilized plants at vegetative stage. At reproductive stage however, plants fertilized with 20 kg P ha^{-1} had significantly higher net assimilation rate than plants that received 0 and 40 kg P ha^{-1}. Plants fertilized with 20 kg P ha^{-1} had the highest mean net assimilation rate which was similar to the value recorded in plants that received 40 kg P ha^{-1} but significantly higher than the value recorded in the unfertilized plants.

Table 4.16: Effects of rhizobia inoculation and phosphorus application on net assimilation rates (g/m^2(leaf area)/day) of cowpea varieties in 2016

	Developmental Stages		
Treatments	**Vegetative**	**Reproductive**	**Mean**
Inoculation (I)			
Uninoculated	5.2×10^{-3}a	1.9×10^{-3}a	3.5×10^{-3}a
90 kg N ha^{-1}	5.5×10^{-3}a	1.9×10^{-3}a	3.7×10^{-3}a
BR 3262	4.6×10^{-3}a	2.0×10^{-3}a	3.3×10^{-3}a
BR 3267	4.4×10^{-3}a	1.9×10^{-3}a	3.1×10^{-3}a
MSD	1.3×10^{-3}a	5.0×10^{-4}a	7.0×10^{-4}
Phosphorus (P)			
0 kg ha^{-1}	4.8×10^{-3}a	2.1×10^{-3}a	3.4×10^{-3}a
20 kg ha^{-1}	5.0×10^{-3}a	1.8×10^{-3}a	3.4×10^{-3}a
40 kg ha^{-1}	5.0×10^{-3}a	1.9×10^{-3}a	3.4×10^{-3}a
MSD	1.2×10^{-3}a	4.0×10^{-4}a	6.0×10^{-4}
Variety (V)			
IT93K-452-1	4.7×10^{-3}a	1.8×10^{-3}b	3.2×10^{-3}a
IT99K-573-1-1	4.7×10^{-3}a	1.5×10^{-3}b	3.1×10^{-3}a
TVX-3236	4.7×10^{-3}a	2.5×10^{-3}a	3.9×10^{-3}a
MSD	1.2×10^{-3}	4.0×10^{-4}	6.0×10^{-4}
Interaction			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × V × P	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05

Table 4.17: Effects of rhizobia inoculation and phosphorus application on net assimilation rates (g/m²(leaf area)/day) of cowpea varieties in 2017

	Developmental Stages		
Treatments	**Vegetative**	**Reproductive**	**Mean**
Inoculation (I)			
Uninoculated	6.8×10^{-3} a	4.7×10^{-3} a	5.7×10^{-3} a
90 kg N ha^{-1}	6.4×10^{-3} a	4.4×10^{-3} ab	5.4×10^{-3} a
BR 3262	6.0×10^{-3} a	4.2×10^{-3} ab	5.1×10^{-3} a
BR 3267	7.8×10^{-3} a	3.4×10^{-3} b	5.6×10^{-3} a
USDA 3451	7.2×10^{-3} a	3.4×10^{-3} b	5.3×10^{-3} a
MSD	2.0×10^{-3}	1.2×10^{-3}	1.3×10^{-3}
Phosphorus (P)			
0 kg ha^{-1}	6.2×10^{-3} a	3.7×10^{-3} b	4.9×10^{-3} b
20 kg ha^{-1}	7.1×10^{-3} a	4.7×10^{-3} a	5.9×10^{-3} a
40 kg ha^{-1}	7.2×10^{-3} a	3.62×10^{-3} b	5.4×10^{-3} ab
MSD	1.6×10^{-3}	9.0×10^{-4}	1.0×10^{-3}
Variety (V)			
IT93K-452-1	6.4×10^{-3} b	3.9×10^{-3} a	5.1×10^{-3} b
IT99K-573-1-1	6.1×10^{-3} b	4.0×10^{-3} a	5.1×10^{-3} b
TVX-3236	8.0×10^{-3} a	4.1×10^{-3} a	6.1×10^{-3} a
MSD	1.6×10^{-3}	9.0×10^{-4}	1.0×10^{-3}
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × V × P	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05

At vegetative stage, TVX-3236 plants had significantly higher net assimilation rate than IT93K -452-1 and IT99K-573-1-1 plants which were similar. At reproductive stage however, the net assimilation rate of the three varieties were similar. Across the developmental stages, the net assimilation rate of IT93K -452-1 and IT99K-573-1-1 were similar and significantly lower than the assimilation rate of TVX-3236 plants. In general, the net assimilation rate reduced as the plant aged.

iv. Leaf area ratio

The leaf area ratio of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2015 is presented in Table 4.18. There was no significant difference between the leaf area ratio of the inoculated and uninoculated plants at the vegetative stage. At the reproductive stage, the uninoculated plants had the highest leaf area ratio which was similar to plants inoculated with USDA 3451. Plants inoculated with USDA 3384 had the least leaf area ratio but it was similar to the leaf area ratio of plants inoculated with USDA 3451. Across the developmental stages, the leaf area ratio of the uninoculated plants and USDA 3451 inoculated plants were similar and significantly lower than the leaf area ratio of USDA 3384 inoculated plant.

There was no significant difference between the leaf area ratio of P fertilized and unfertilized plants throughout the sampling period. Likewise, there was no significant difference between the leaf area ratio of the three varieties at the vegetative stage. At the flowering stage however, IT93K-452-1 plants had significantly higher leaf area ratio than IT99K-573-1-1 and TVX-3236 plants which were similar. Across the developmental stages, there was no significant difference between leaf area ratio of the three varieties.

Table 4.18: Effects of rhizobia inoculation and phosphorus application on leaf area ratio (cm^2/g) of cowpea varieties in 2015

Treatments	Vegetative stage	Reproductive stage	Mean
Inoculation (I)			
Uninoculated	398.86a	438.84a	418.85a
USDA 3451	401.03a	426.72ab	413.88a
USDA 3384	363.19a	384.43b	373.81b
MS D	42.83	44.17	34.48
Phosphorus (P)			
0 kg ha^{-1}	365.26a	405.64a	385.45a
20 kg ha^{-1}	392.46a	434.15a	413.31a
40 kg ha^{-1}	405.36a	410.2a	407.78a
M S D	42.83	44.17	34.48
Variety (V)			
IT93K-452-1	394.76a	439.38a	417.07a
IT99K-573-1-1	391.68a	392.37b	392.03a
TVX-3236	376.65a	418.24b	397.44a
LMS D	42.83	44.17	34.48
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05

The leaf area ratio of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2016 is presented in Table 4.19. There was no significant difference between the leaf area ratio of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants at the vegetative stage. At the reproductive phase also, there was no significant difference between the leaf area ratio of the inoculated and uninoculated plants. However, plants fertilized with 90 kg N ha^{-1} had the highest leaf area ratio but the value was similar to the other inoculation treatment except plants inoculated BR 3262 which had the least leaf area ratio. Across the developmental stages, the leaf area ratio of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants were similar. There was no significant difference between the leaf area ratio of the P fertilized and unfertilized plants at the vegetative phase. At the reproductive phase however, plants fertilized with 40 kg P ha^{-1} had the highest leaf area ratio which was similar to the value obtained in plants fertilized with 20 kg Pha^{-1} but significantly higher than the value recorded in the unfertilized plants. Across the developmental stages, the leaf area ratio of the P fertilized and unfertilized plants were similar and there was no significant difference between the leaf area ratio of the three varieties throughout the sampling period.

The interaction between rhizobia inoculation and variety on LAR at vegetative and flowering stage as well as on the mean LAR is presented in Figure 4.23A, B and C respectively. In IT93K-452-1 variety, plants fertilized with 90 kg N ha^{-1} had the highest LAR and the value was significantly higher than what was recorded in the inoculated and uninoculated plants. In IT99K-573-1-1 variety, though the uninoculated plants had the highest LAR, there was no significant difference between the LAR of the inoculated and uninoculated plants. In TVX-3236 variety, plants inoculated with BR 3267 had the highest LAR. The value was similar to the LAR of the uninoculated plants but significantly higher than the LAR of 90 kg N ha^{-1} fertilized and BR 3262

Table 4.19: Effects of rhizobia inoculation and phosphorus application on leaf area ratio (cm^2/g) of cowpea varieties in 2016

	Developmental stages		
Treatments	**Vegetative stage**	**Reproductive stage**	**Mean**
Inoculation (I)			
Uninoculated	339.90a	468.72ab	404.31a
90 kg N ha^{-1}	378.18a	518.91a	448.55a
BR 3262	330.48a	444.61b	387.55a
BR 3267	361.86a	485.03ab	423.44a
MSD	67.2	61.18	61.26
Phosphorus (P)			
0 kg ha^{-1}	359.23a	431.59b	395.41a
20 kg ha^{-1}	345.44a	480.34ab	412.89a
40 kg ha^{-1}	353.14a	526.02a	439.58a
MSD	58.20	52.98	53.05
Variety (V)			
IT93K-452-1	372.35a	488.33a	430.34a
IT99K-573-1-1	336.42a	490.91a	413.67a
TVX-3236	349.04a	458.71a	403.88a
MSD	58.20	52.98	53.05
Interactions			
I × P	NS	NS	NS
I × V	*	*	**
P × V	NS	*	NS
I × V × P	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, *, **-significant at P=0.05 and 0.01 respectively NS- not significant at P=0.05

inoculated plants (Figure 4.23A). Similar trend was observed in IT93K-452-1 and IT99K-573-1-1 plants at the flowering stage (Figure 4.23B). But in TVX-3236 plants, the LAR of 90 kg N ha^{-1} fertilized plant and those inoculated with BR 3267 were similar. Across the developmental stages (mean LAR), IT93K-452-1 plants fertilized with 90 kg N ha^{-1} had significantly higher LAR than the uninoculated and inoculated plants which had similar LAR and there was no significant difference between the LAR of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants of IT99K-573-1-1. In TVX-3236 plants, plants inoculated with BR 3267 strain had the highest LAR but the value was similar to the LAR of 90 kg N ha^{-1} fertilized plants (Figure 4.23C).

The interaction between phosphorus and variety on LAR was significant at the flowering stage. There was no significant difference between the LAR of P fertilized and unfertilized plants of IT93K-452-1 and TVX-3236 plants but in IT99K-573-1-1 variety, LAR increased significantly with increase in P rates with the peak recorded at 40 kg P ha^{-1} application (Figure 4.23D).

The leaf area ratio of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2017 is presented in Table 4.20. There was no significant difference between the leaf area ratio of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants at the vegetative stage. At the reproductive phase however, plants inoculated with BR 3262 rhizobia strain had the highest LAR but the value was similar to what was recorded in the other inoculation treatments except plants inoculated with BR 3267 rhizobia strain. Averagely, there was no significant difference between the leaf area ratio of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants. Furthermore, there was no significant difference between the leaf area ratio of the P fertilized and unfertilized plants as well as the three varieties throughout the sampling period.

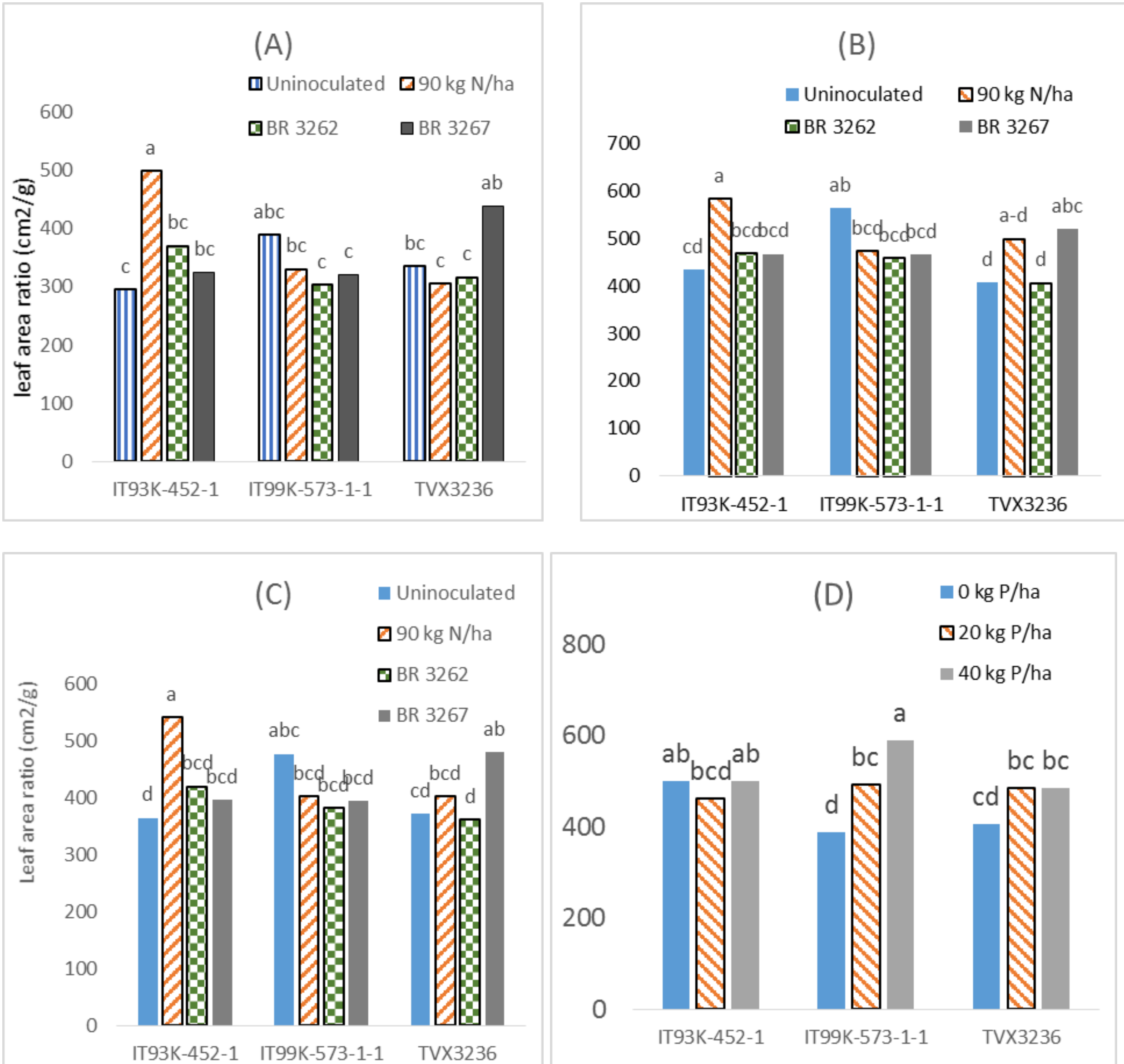


Figure 4.23: Interaction effects of (A) rhizobia inoculation and varieties on leaf area ratio of cowpea at vegetative phase, (B) rhizobia inoculation and varieties on leaf area ratio of cowpea at reproductive phase, (C) rhizobia inoculation and varieties on mean leaf area ratio, (D) phosphorus and variety on leaf area ratio of cowpea at reproductive phase in 2016

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

Table 4.20: Effects of rhizobia inoculation and phosphorus application on leaf area ratio (cm^2/g) of cowpea varieties in 2017

Treatments	Developmental Stages		
	Vegetative stage	Reproductive stage	Mean
Inoculation (I)			
Uninoculated	210.41a	223.88a	217.15a
90 kg N ha^{-1}	201.55a	198.36ab	199.95a
BR 3262	213.68a	224.33a	219.00a
BR 3267	200.45a	192.15b	196.30a
USDA 3451	212.66a	221.50ab	217.08a
MSD	38.28	30.27	32.40
Phosphorus (P)			
0 kg ha^{-1}	203.65a	210.22a	206.94a
20 kg ha^{-1}	201.83a	206.01a	203.92a
40 kg ha^{-1}	217.77a	219.9a	218.84a
MSD	30.23	22.63	25.09
Variety (V)			
IT93K-452-1	219.89a	212.63a	216.26a
IT99K-573-1-1	219.13a	215.28a	217.21a
TVX-3236	184.23b	208.22a	196.22a
MSD	30.23	22.63	25.09
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × V × P	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference , NS- not significant at P=0.05

v. Leaf area index

The leaf area index (LAI) of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2015 is presented in Table 4.21. There was no significant difference between the leaf area index of uninoculated and inoculated plants in 2015. Averagely, the leaf area index of the uninoculated plant was the highest and it was similar to the value recorded in plants inoculated with USDA 3451 but significantly higher than the value recorded in plants inoculated with USDA 3384.

The leaf area index of plants fertilized with 20 and 40 kg P ha^{-1} were similar and significantly higher than the LAI of the unfertilized plants which had the least LAI throughout the sampling period. There was no significant difference between the LAI of the three varieties at 3 WAS. At 5 and 7 WAS however, the LAI of IT93K-452-1 plants were significantly higher than the value recorded in IT99K-573-1-1 and TVX-3236 plants which were similar. The mean LAI followed the same trend.

The interaction between rhizobia inoculation and phosphorus on LAI at 5 and 7 WAS and mean LAI is presented in Figure 4.24 A, B and C respectively. At 5 WAS, the LAI of uninoculated and USDA 3451 inoculated plants increased with increase in P rates with the maximum LAI obtained at 40 kg P ha^{-1} (Figure 4. 24A). However, in the uninoculated plants, the increase in LAI between 20 and 40 kg P ha^{-1} was not significant. In plants inoculated with USDA 3384, the increase in LAI as a result of P application peaked at 20 kg P ha^{-1} application after which there was a decline at 40 kg P ha^{-1} application. Similar trend was maintained at 7 WAS (Figure 4.24B) and the trend observed for mean LAI was similar (Figure 4.24C).

The interaction between rhizobia inoculation and variety on LAI at 7 WAS is presented on Figure 4.24D. Uninoculated plants of IT93K-452-1 had the highest LAI and the value was significantly higher than the LAI of plants inoculated with USDA 3451 but similar to the LAI of plants inoculated with USDA 3384. In IT99K-573-1-1 variety, plants inoculated with USDA 3451 had significantly higher LAI than plants inoculated with USDA 3384 but the value was similar to what was recorded in the uninoculated plants. There was no significant difference between the LAI of inoculated and uninoculated plants of TVX-3236 variety.

The interaction between phosphorus and variety on LAI in 2015 was significant at 7 WAS. The LAI of IT99K-573-1-1 and TVX-3236 plants increased with increase in P rates up to 40 kg P ha^{-1} (Figure 4.25). However, there was no significant difference between the LAI of plants that received 20 and 40 kg P ha^{-1}. In IT93K-452-1 plants, P application increased LAI up to 20 kg P ha^{-1} application after which there was a decline at 40 kg P ha^{-1} application.

The interaction between rhizobia inoculation, phosphorus application and variety on LAI was significant at 5 WAS and is presented in Figure 4.26. The uninoculated plants of IT93K-452-1 and IT99K-573-1-1 increased with increase in P rates up to 40 kg P ha^{-1}. However, the difference between 20 and 40 kg P ha^{-1} was not significant in IT99K-573-1-1. In uninoculated TVX-3236 variety however, P application increased LAI up to 20 kg P ha^{-1} application after which there was a decline at 40 kg P ha^{-1} application. In plants inoculated with USDA 3451, P application increased LAI up to 20 kg P ha^{-1} application after which there was a decline at 40 kg P ha^{-1} application in the three varieties. In plants inoculated with USDA 3384, LAI increased with increase in P rates up to 40 kg P ha^{-1} in the three varieties however, there was no significant difference between the values recorded at 20 and 40 kg P ha^{-1} except in IT99K-573-1-1 variety.

Table 4.21: Effects of rhizobia inoculation and phosphorus application on leaf area index of cowpea varieties in 2015

	Sampling period			
Treatments	3 WAS	5 WAS	7 WAS	Mean
Inoculation (I)				
Uninoculated	0.56a	3.79a	5.17a	3.18a
USDA 3451	0.52a	3.62a	4.90a	3.01ab
USDA 3384	0.51a	3.38a	4.73a	2.87b
MSD	0.10	0.44	0.45	0.28
Phosphorus (P)				
0 kg ha^{-1}	0.34b	2.02b	2.90b	1.75b
20 kg ha^{-1}	0.59a	4.26a	5.90a	3.59a
40 kg ha^{-1}	0.65a	4.51a	6.08a	3.72a
MSD	0.10	0.44	0.45	0.28
Variety (V)				
IT93K-452-1	0.56a	4.14a	5.61a	3.44a
IT99K-573-1-1	0.59a	3.27b	4.51b	2.79b
TVX-3236	0.44a	3.38b	4.68b	2.84b
MSD	0.10	0.44	0.45	0.28
Interactions				
I × P	NS	*	**	**
I × V	NS	NS	*	NS
P × V	NS	NS	*	NS
I × V × P	NS	**	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, WAS – weeks after sowing, MSD-Minimum significant difference, *, **- significant at P=0.05 and 0.01 respectively NS- not significant at P=0.05

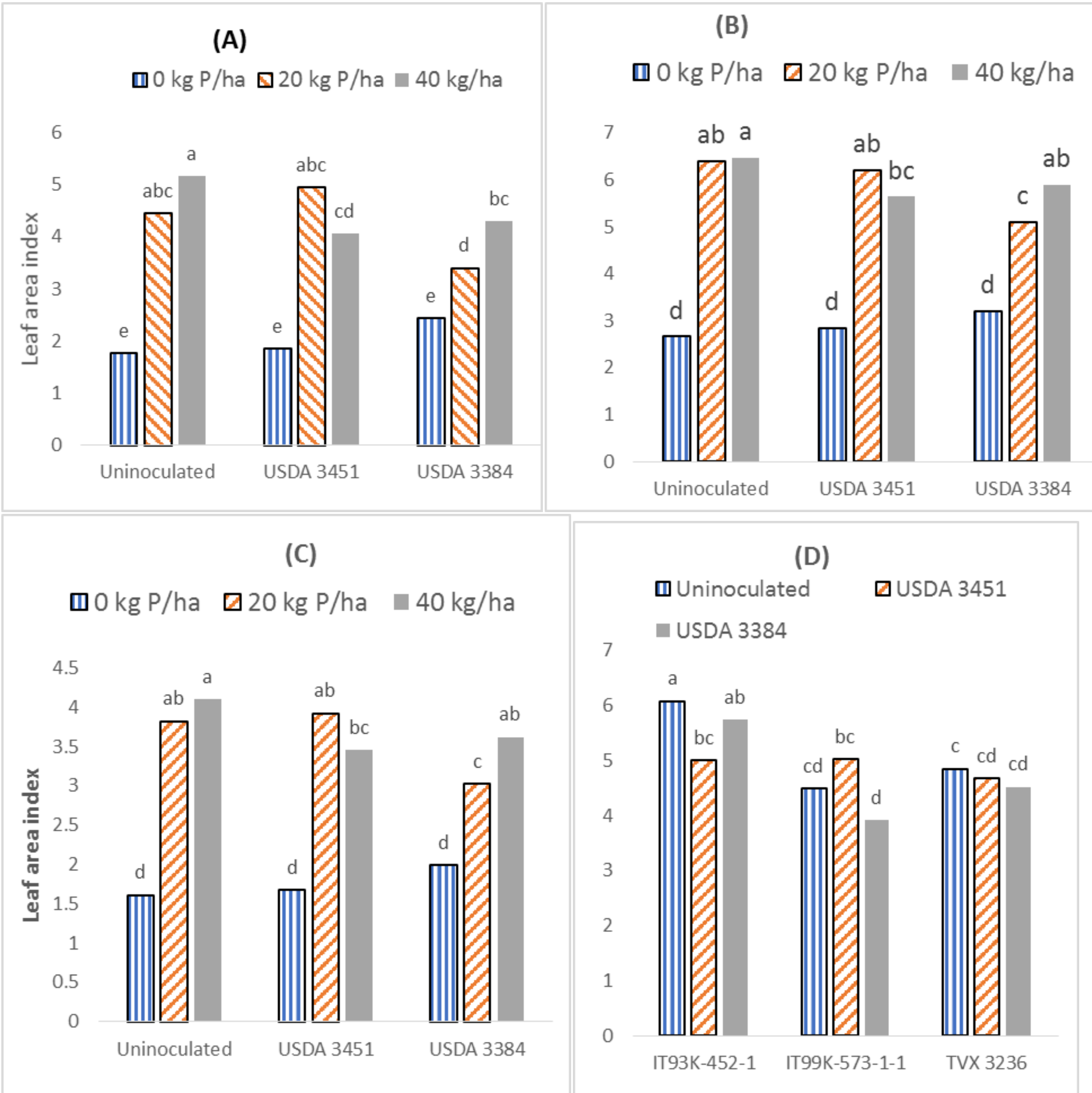


Figure 4.24: Interaction effects of (A) rhizobia inoculation and phosphorus on leaf area index of cowpea at 5 weeks after sowing, (B) rhizobia inoculation and phosphorus on leaf area index of cowpea at 7 weeks after sowing, (C) rhizobia inoculation and phosphorus on mean leaf area index, (D) rhizobia inoculation and variety on leaf area index of cowpea at 7 weeks after sowing in 2015

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

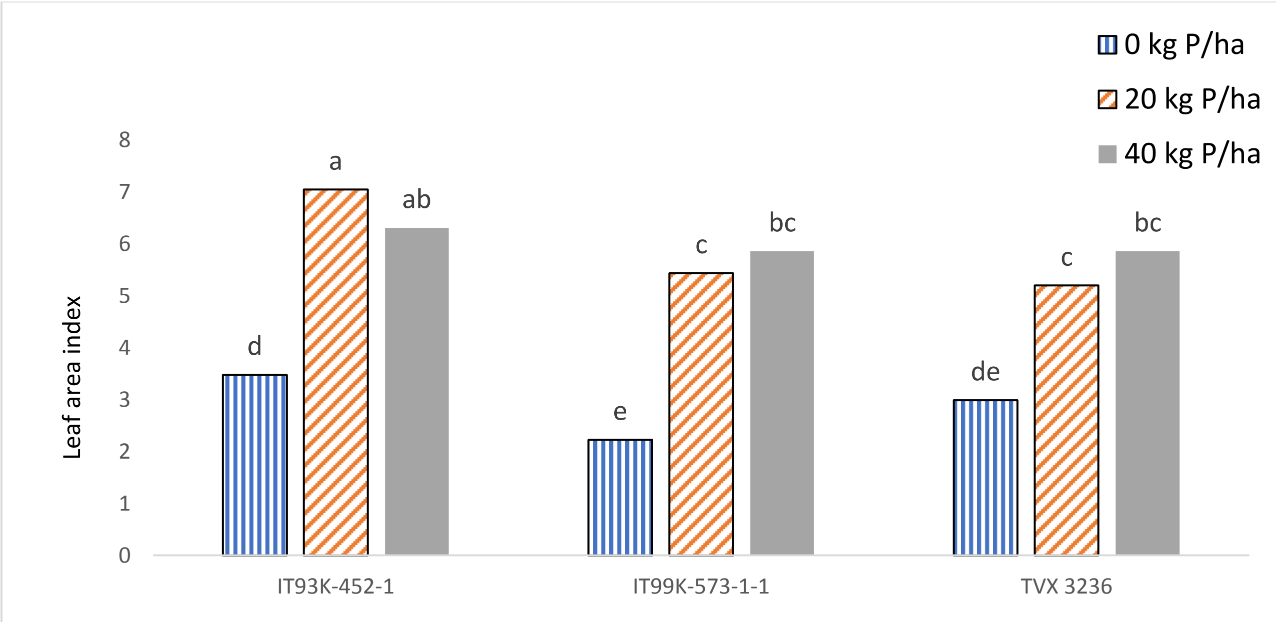


Figure 4.25: Interaction effects of phosphorus and varieties on leaf area index of cowpea at 7 weeks after sowing in 2015

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

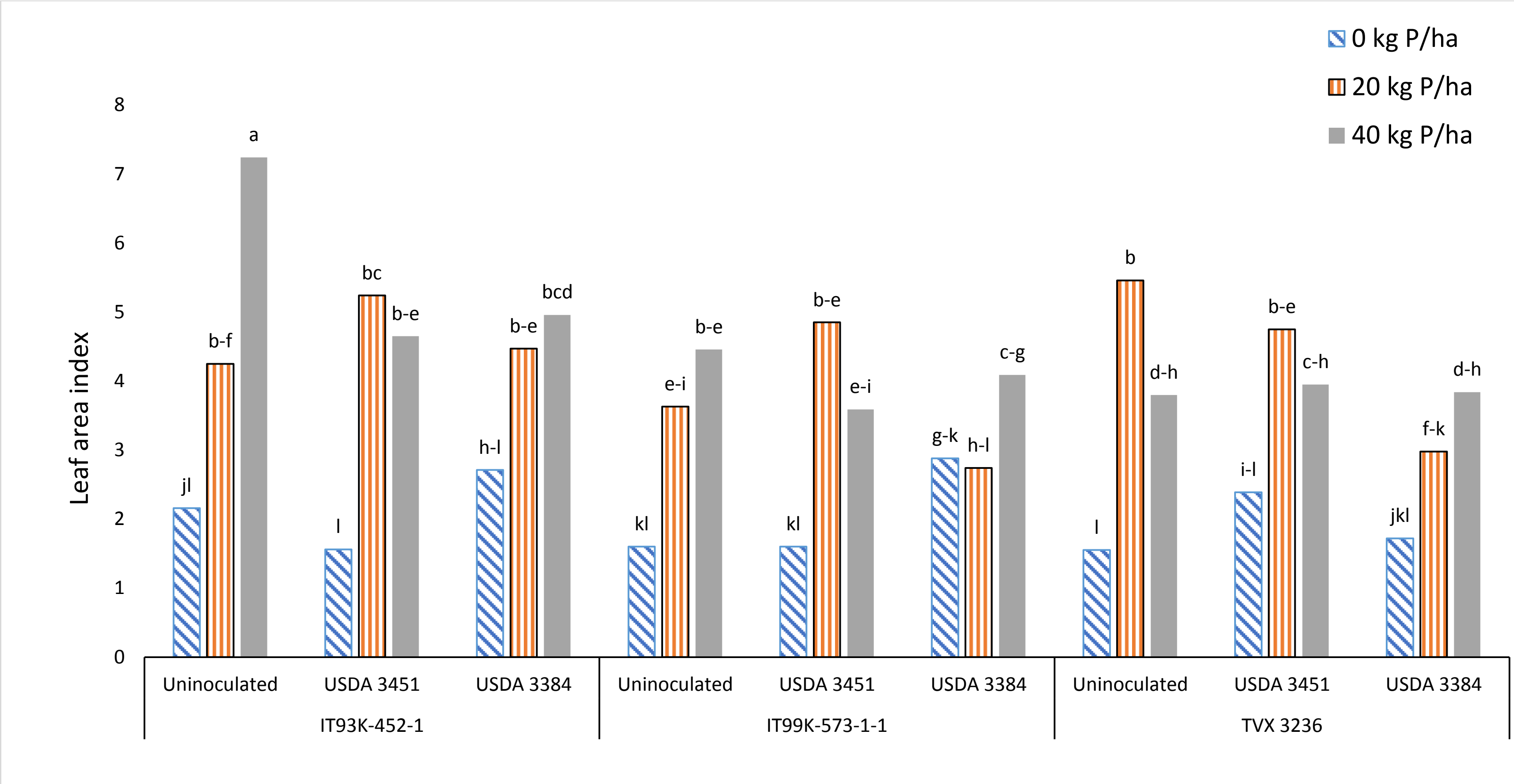


Figure 4.26: Interaction effects of rhizobia inoculation, phosphorus application and varieties on leaf area index of cowpea at 5 weeks after sowing in 2015

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

The leaf area index (LAI) of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2016 is presented in Table 4.22. There was no significant difference between the leaf area index of the uninoculated, inoculated and 90 kg N ha^{-1} fertilized plants throughout the sampling period in 2016. Averagely however, plants fertilized with 90 kg N ha^{-1} had the highest LAI but the value was similar to what was recorded in the other inoculation treatment except in plants inoculated with BR 3262 which had the least LAI.

The LAI of plants fertilized with 20 and 40 kg P ha^{-1} were similar and both were significantly higher than the LAI of the unfertilized plants throughout the sampling period except at 3 WAS where the LAI of the unfertilized plants were similar to those that received 20 kg P ha^{-1}. IT99K-573-1-1 variety had the highest LAI among the varieties throughout the sampling period but its value was similar to what was recorded in IT93K-452-1 but significantly higher than the LAI recorded in TVX-3236 plants which had the least LAI throughout the sampling period. At 5 and 7 WAS, there was no significant difference between the LAI of TVX-3236 and IT93K-452-1 plants. Averagely, the order of LAI among the varieties was IT99K-573-1-1 > IT93K-452-1 > TVX-3236 but the LAI of IT93K-452-1 and TVX-3236 were statistically similar.

The interaction between rhizobia inoculation and variety on LAI at 7 WAS in 2016 is presented in Figure 4.27A. There was no significant difference between the LAI of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants of IT93K-452-1 and IT99K-573-1-1 varieties. In TVX-3236 variety however, plants fertilized with 90 kg N ha^{-1} had the highest LAI than the inoculated and uninoculated plants which had similar LAI. Similar trend was observed for the mean LAI (Figure 4.27 B).

Table 4.22: Effects of rhizobia inoculation and phosphorus application on leaf area index of cowpea varieties in 2016

	Sampling Period			
Treatments	3 WAS	5 WAS	7 WAS	Mean
Inoculation (I)				
Uninoculated	0.22a	1.69a	4.77a	2.23 ab
90 kg N ha^{-1}	0.18a	1.87a	5.34a	2.46a
BR 3262	0.19a	1.59a	4.40a	2.06b
BR 3267	0.21a	1.84a	5.23a	2.43a
MSD	0.06	0.45	0.95	0.36
Phosphorus (P)				
0 kg ha^{-1}	0.16b	1.46b	3.10b	1.58b
20 kg ha^{-1}	0.21ab	1.87a	5.50a	2.53a
40 kg ha^{-1}	0.23a	1.91a	6.21a	2.78a
MSD	0.05	0.39	0.82	0.31
Variety (V)				
IT93K-452-1	0.23a	1.75ab	4.87ab	2.29b
IT99K-573-1-1	0.25a	2.09a	5.55a	2.63a
TVX-3236	0.13b	1.41b	4.39b	1.97b
MSD	0.05	0.39	0.82	0.31
Interactions				
I × P	NS	NS	NS	NS
I × V	NS	NS	**	*
P × V	NS	NS	NS	NS
I × P × V	NS	NS	**	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test,. WAS – weeks after sowing, MSD-Minimum significant difference. *, **- significant at P=0.05 and 0.01 respectively NS- not significant at P=0.05

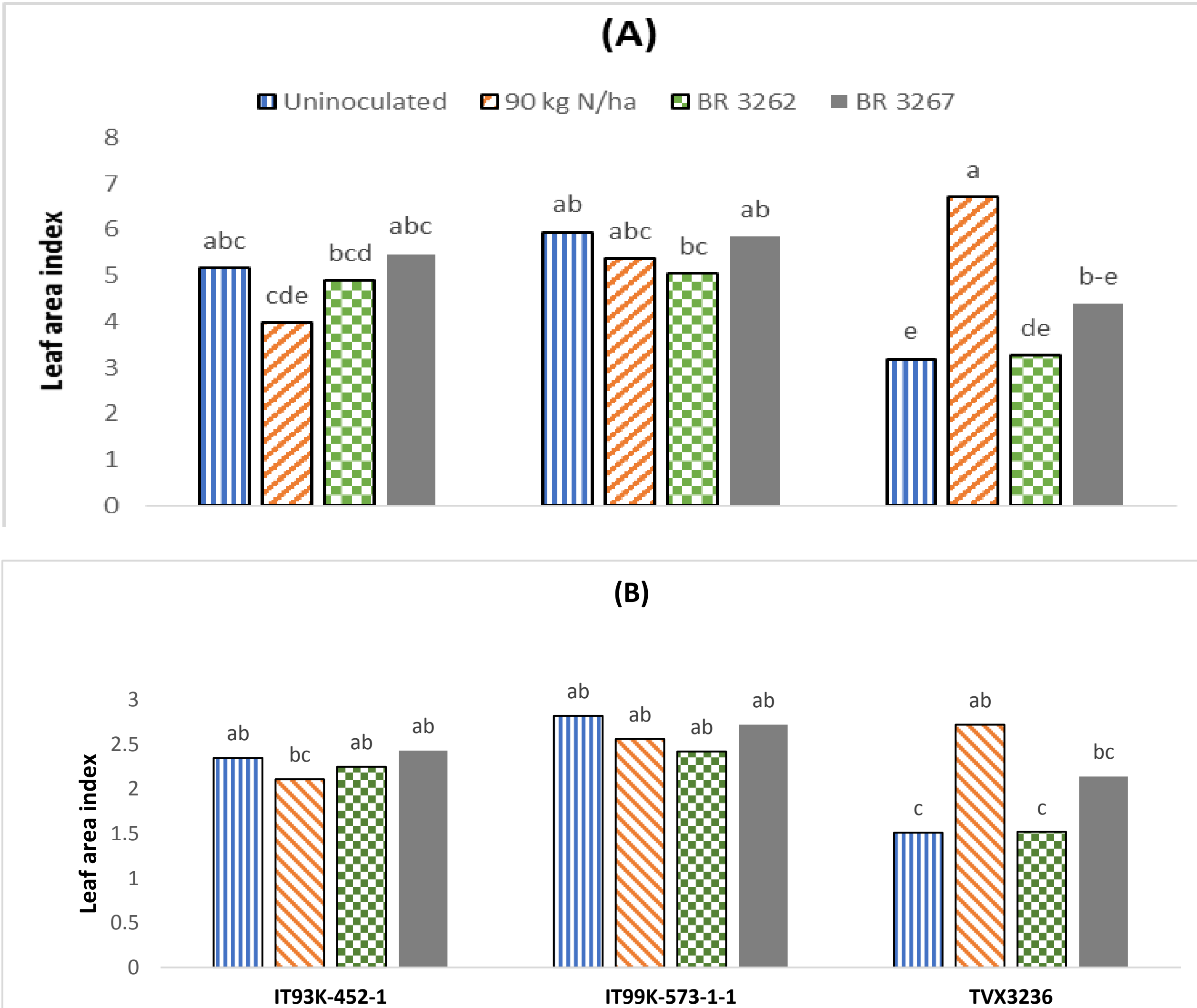


Figure 4.27: Interaction effects of rhizobia inoculation and varieties on (A) leaf area index of cowpea at 7 weeks after sowing in 2016 (B) mean leaf area index of cowpea in 2016

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

Figure 4.28 shows the interaction between rhizobia inoculation, phosphorus application and variety on LAI at 7 WAS. The uninoculated plants of IT93K-452-1 and IT99K-573-1-1 had their highest LAI at 40 kg P ha^{-1}. The uninoculated TVX-3236 plants however, had their highest LAI at 20 kg P ha $^{-1}$ application and there was a significant decline at 40 kg P ha^{-1}. Plants fertilized with 90 kg N ha^{-1} had their highest LAI at 40 kg P ha^{-1} application in the three varieties. However, there was no significant difference between the LAI of plants that received 20 and 40 g P ha^{-1} in the three varieties. IT93K-452-1 and IT99K-573-1-1 plants inoculated with BR 3262 had the highest LAI at 20 kg P ha^{-1} but TVX-3236 plants inoculated with the same strain had its highest LAI at 40 kg P ha^{-1} application. However, there was no significant difference between the LAI at 20 and 40 kg was not significant. In IT93K-452-1 plants, those inoculated with BR 3267 strain, there was no significant difference between the LAI of the fertilized and unfertilized plants. IT99K-573-1-1 inoculated with the same strain had its highest LAI at 40 kg P ha^{-1} but it was similar to the value recorded at 20 kg P ha^{-1} application. TVX-3236 plants inoculated with the same strain had the highest LAI at 20 kg P ha^{-1}. Generally, the unfertilized plants had the least LAI among all the treatment combination except in IT93K-452-1 plants inoculated with BR 3267.

The leaf area index (LAI) of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2017 is presented in Table 4.23. There was no significant difference between the leaf area index of the uninoculated and inoculated plants at 3 and 5 WAS. At 7 WAS however, the uninoculated plants had significantly higher LAI than plants inoculated with BR 3267 and 90 kg N ha^{-1} fertilized plants. In average, the LAI of the uninoculated plants, 90 kg N ha^{-1} fertilized plants and BR 3262 and USDA 3451 inoculated plants were similar but the LAI of plants inoculated with BR 3267 was significantly lower than that of the uninoculated plants and those inoculated with BR 3262 and USDA 3451.

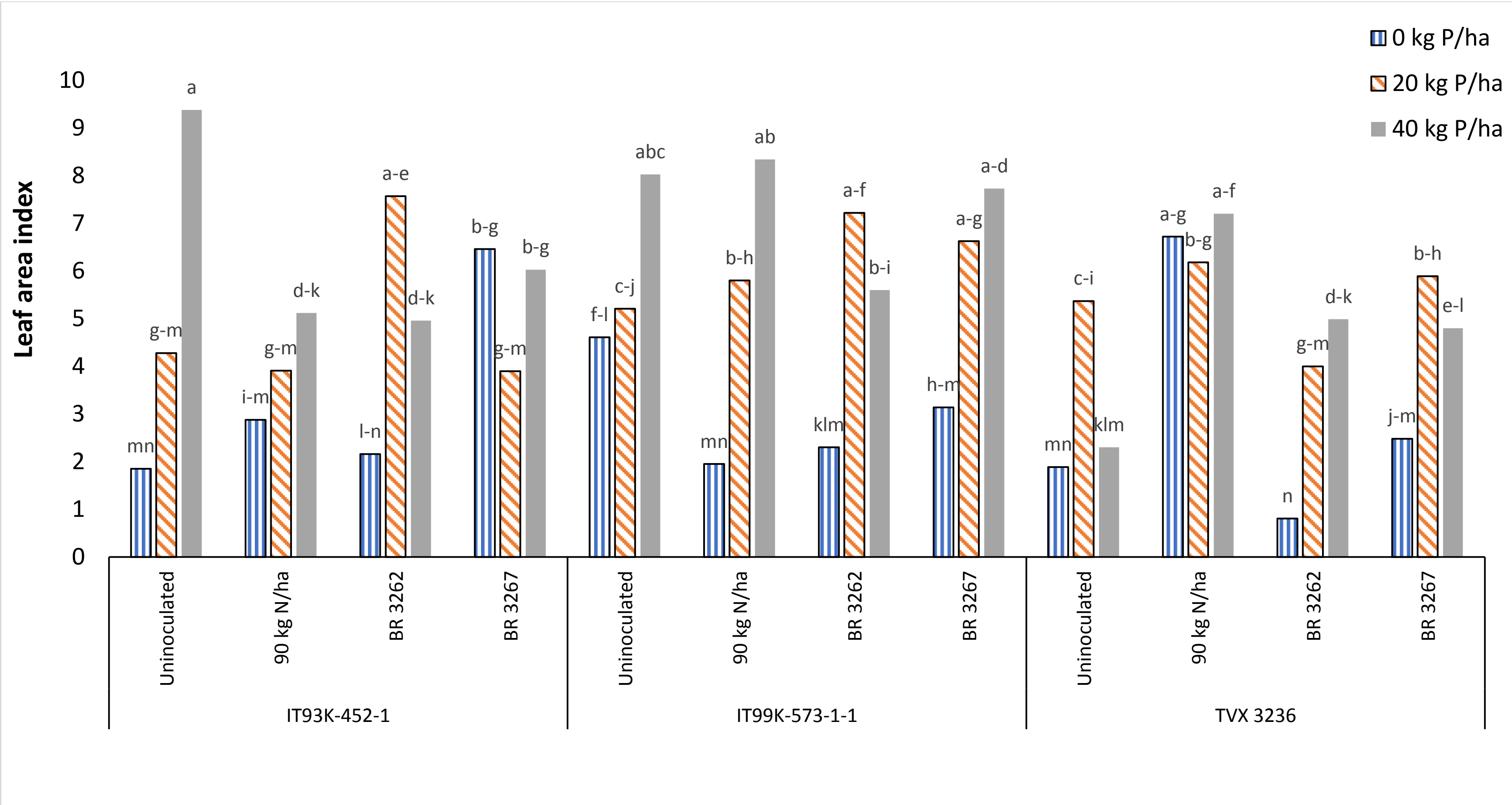


Figure 4.28: Interaction effects of rhizobia inoculation, phosphorus application and varieties on leaf area index of cowpea at 7 weeks after sowing in 2016

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

Table 4.23: Effects of rhizobia inoculation and phosphorus application on leaf area index of cowpea varieties in 2017

Treatments	Sampling period			Mean LAI
	3 WAS	5 WAS	7 WAS	
Inoculation (I)				
Uninoculated	0.16a	0.97a	1.62a	0.92a
90 kg N ha^{-1}	0.17a	1.06a	1.31bc	0.84ab
BR 3262	0.17a	0.88a	1.60a	0.88a
BR 3267	0.16a	0.90a	1.11c	0.73b
USDA 3451	0.16a	1.01a	1.50ab	0.89a
MSD	0.04	0.27	0.23	0.12
Phosphorus (P)				
0 kg ha^{-1}	0.15b	0.72b	1.03b	0.64c
20 kg ha^{-1}	0.17ab	1.00a	1.58a	0.92b
40 kg ha^{-1}	0.18a	1.17a	1.68a	1.01a
MSD	0.03	0.21	0.18	0.09
Variety (V)				
IT93K-452-1	0.18a	1.02a	1.44a	0.89a
IT99K-573-1-1	0.19a	1.13a	1.49a	0.94a
TVX-3236	0.12b	0.74b	1.35a	0.74b
MSD	0.03	0.21	0.18	0.09
Interactions				
I × P	NS	NS	NS	NS
I × V	NS	NS	NS	NS
P × V	NS	NS	NS	NS
I × V × P	NS	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test,. WAS – weeks after sowing, MSD-Minimum significant difference, NS- not significant at P=0.05

The LAI of plants fertilized with 20 and 40 kg P ha^{-1} were similar and both were significantly higher than the LAI of the unfertilized plants throughout the sampling period except at 3 WAS in which there was no significant difference between the LAI of the unfertilized plants and those fertilized with 20 kg P ha^{-1}. The LAI of IT93K-452-1 and IT99K-573-1 plants were similar and were significantly higher than the LAI of TVX-3236 plants throughout the sampling period except at 7 WAS in which though TVX-3236 had the lowest LAI value but, it was not significantly different from the value recorded in the other two varieties

vi. Quantum yield of photosystem II (Phi 2)

The quantum yield of photosystem II of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2017 is presented in Table 4.24. There was no significant difference between the quantum yield of photosystem II of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plant at the vegetative phase. At the reproductive phase however, the uninoculated plants had the highest quantum yield. The value was significantly higher than the value recorded in plants inoculated with BR 3262 and BR 3267 but similar to the quantum yield of plants fertilized with 90 kg N ha^{-1} and plants inoculated with USDA 3451.

At the vegetative phase, the quantum yield of plants that received 20 and 40 kg P ha^{-1} were similar and they were significantly higher than the value recorded in the unfertilized plants. The same trend was observed at the reproductive stage but there was no significant difference between the quantum yield of the unfertilized plants and those that received 20 kg P ha^{-1}. At the vegetative stage, IT99K-573-1-1 plants had the highest quantum yield but the value was similar to what was recorded in IT93K-452-1. TVX-3236 plants had the least quantum yield but the value was similar to the quantum yield of IT93K-452-1 plants. The same trend was observed at the reproductive stage. The quantum yield recorded at the reproductive stage were generally lower than those recorded at the vegetative phase.

Figure 4.29 shows the interaction effects between rhizobia inoculation and variety on Phi 2 of cowpea at the reproductive stage. In IT93K-452-1 variety, plants fertilized with 90 kg N ha^{-1} had the highest quantum yield values. This was however similar to the values recorded in the uninoculated plants and those inoculated with BR 3267. Plants inoculated with BR 3262 had the least values but it was not significantly different from what was recorded in other inoculated plants.

In IT99K-573-1-1 variety, the uninoculated plants had significantly higher quantum yield values than all the inoculated plants and 90 kg N ha^{-1} fertilized plants which were similar. In TVX-3236 variety, the uninoculated plants equally had the highest quantum yield value but the value was similar to what was recorded in other inoculation treatments except plants inoculated with BR 3262 which had the least value.

vii. Ratio of light that goes towards non-photochemical quenching

The ratio of light that goes towards non-photochemical quenching (phi npq) of cowpea varieties as affected by rhizobia inoculation and phosphorus application in 2017 is presented in Table 4.24. There was no significant difference between the phi npq of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plant at the vegetative phase. At the reproductive phase however, the uninoculated plants had significantly lower phi npq value than the inoculated and 90 kg N ha^{-1} fertilized plants. At the vegetative phase, the phi npq of plants that received 20 and 40 kg P ha^{-1} were similar and they were significantly lower than the value recorded in the unfertilized plants. At the reproductive stage however, the phi npq of unfertilized plants and those that received 20 kg P ha^{-1} were similar and significantly higher than the value recorded in plants that received 40 kg P ha^{-1}. There was no significant difference between the phi npq recorded in the three varieties at the vegetative phase.

At the reproductive stage, the value recorded in IT93K-452-1 and IT99K-573-1-1 plants were similar and significantly lower than the value recorded in TVX-3236 plants.

viii. Ratio of light that is lost through non-regulated processes

The ratio of light that is lost through non-regulated processes (phi no) of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2017 is presented in Table 4.24. There was no significant difference between the phi no of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plant at the vegetative stage. At the reproductive stage, there was equally no significant difference between the phi no of the inoculated and uninoculated plants. However, the uninoculated plants had significantly higher phi no value than plants fertilized with 90 kg N ha^{-1}. Plants fertilized with 20 and 40 kg P ha^{-1} had similar phi no values that were significantly higher than the values recorded in the unfertilized plants at the vegetative and reproductive stage.

At the vegetative stage, there was no significant difference between the phi no of the three varieties. At the reproductive stage however, IT99K-573-1-1 plants had the highest Phi no value but it was similar to what was recorded in IT93K-452-1 plants and significantly higher than the value recorded in TVX-3236 plants.

ix. Leaf temperature difference

The difference between the leaf temperature and the ambient temperature of cowpea varieties as affected by rhizobia inoculation and phosphorus application in 2017 is presented in Table 4.24. There was no significant difference between the leaf temperature of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plant at the vegetative phase and the reproductive stage. Likewise, there was no significant difference between the leaf temperatures of the P fertilized and unfertilized plants at the vegetative and reproductive stages. Furthermore, the leaf temperature of the three varieties were not significantly different at the two stages.

Table 4.24: Effects of rhizobia inoculation and phosphorus application on photosynthetic activities of cowpea varieties at vegetative and reproductive stage in 2017

Treatments	Phi 2 stage I	Phi 2 stage II	Phinpq stage I	Phinpq stage II	Phi no stage 1	Phi no stage II	Leaf temp. diff stage I	Leaf temp. diff stage II
	(mol e^- mol^{-1} quanta)						(°C)	
Inoculation (I)								
Uninoculated	0.52a	0.46a	0.20a	0.26b	0.28a	0.28a	-3.92a	-3.34a
90 kg N ha-^{1}	0.50a	0.43ab	0.22a	0.32a	0.28a	0.26b	-3.85a	-3.83a
BR 3262	0.50a	0.39c	0.19a	0.35a	0.29a	0.27ab	-3.76a	-3.63a
BR 3267	0.52a	0.40bc	0.19a	0.34a	0.28a	0.26ab	-3.60a	-4.14a
USDA 3451	0.52a	0.43ab	0.21a	0.31a	0.28a	0.27ab	-3.79a	-3.46a
MSD	0.03	0.03	0.04	0.04	0.02	0.02	0.93	0.99
Phosphorus (P)								
0 kg ha^{-1}	0.49b	0.40b	0.24a	0.32a	0.27b	0.25b	-3.76a	-3.97a
20 kg ha^{-1}	0.53a	0.42ab	0.19b	0.33a	0.29a	0.28a	-3.93a	-3.81a
40 kg ha^{-1}	0.53a	0.44a	0.18b	0.20b	0.29a	0.27a	-3.66a	-3.25a
MSD	0.02	0.02	0.03	0.03	0.01	0.02	0.72	0.76
Variety (V)								
IT93K-452-1	0.52ab	0.43a	0.20a	0.31b	0.28a	0.27ab	-3.88a	-3.69a
IT99K-573-1-	0.53a	0.43a	0.19a	0.29b	0.28a	0.28a	-3.96a	-3.69a
TVX-3236	0.50b	0.40b	0.21a	0.34a	0.29a	0.25b	-3.52a	-3.66a
MSD	0.02	0.02	0.01	0.03	0.01	0.02	0.72	0.76
Interactions								
I × P	NS	NS	NS	NS	NS	NS	NS	NS
I × V	NS	*	NS	NS	NS	NS	NS	NS
P × V	NS	NS	NS	NS	NS	NS	NS	NS
I × P × V	NS	NS	NS	NS	NS	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, stage 1- vegetative stage, stage II- reproductive stage, MSD-Minimum significant difference, * - significant at P=0.05, NS- not significant at P=0.05

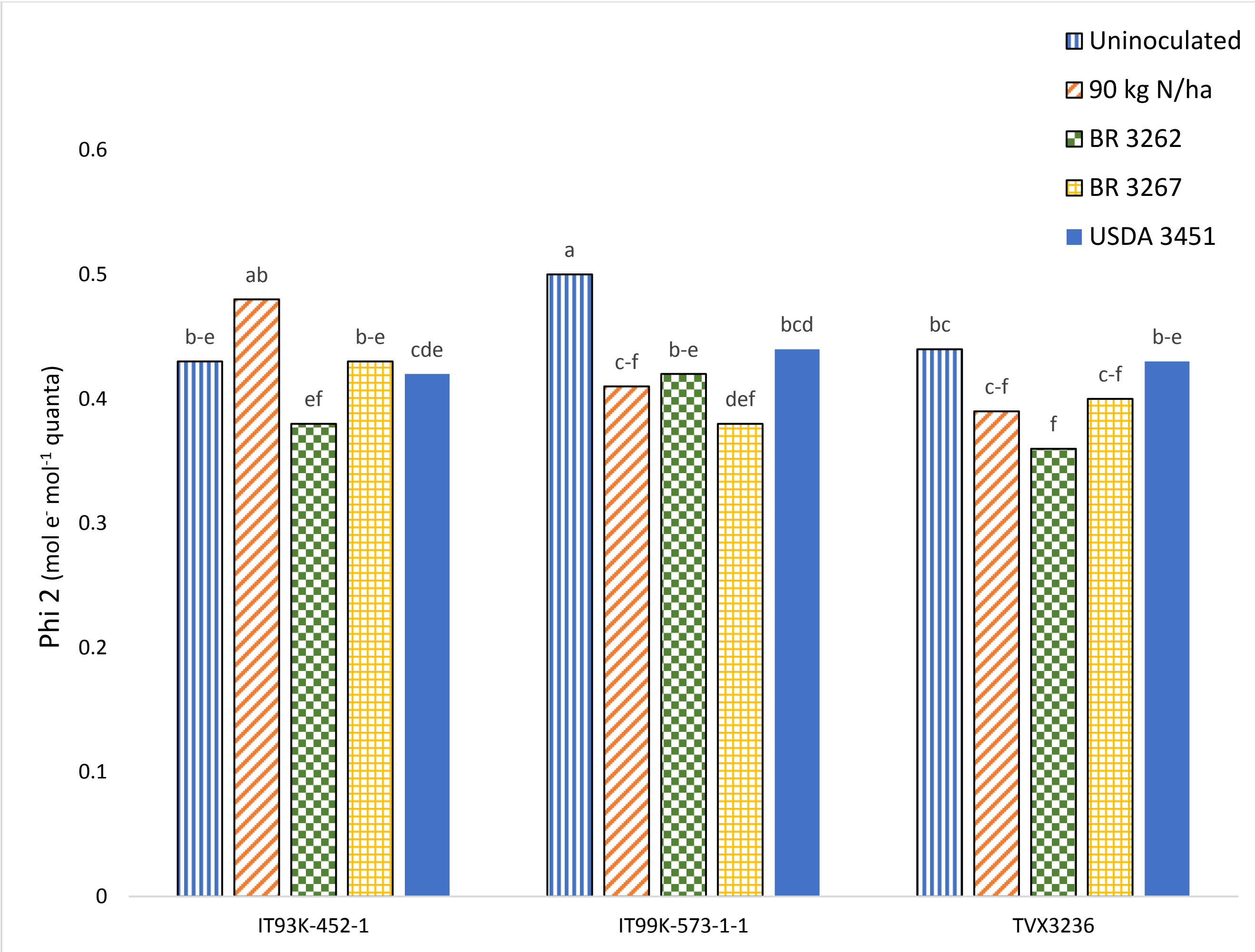


Figure 4.29: Interaction effects of rhizobia inoculation and varieties on quantum yield of photosystem II (Phi 2) of cowpea at pod filling stage in 2017

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

4.1.2.3 Nodulation and nitrogen fixation attributes

i. Nodulation

The nodulation of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2015 is presented in Table 4.25. There was no significant difference between the nodules dry

weight of the inoculated and uninoculated plants. Plants inoculated with USDA 3384 however had significantly higher number of nodules than the uninoculated plants but it was similar to the value obtained in plants inoculated with USDA 3451.

There was no significant difference between the percentage of infective nodules of inoculated and uninoculated plants. Plants fertilized with 20 and 40 kg P ha^{-1} had similar number of nodules, nodule dry weight and percentage of infective nodules that were significantly higher than the values recorded in the unfertilized plants. IT93K-452-1 and IT99K-573-1-1 plants had similar nodule weight that were significantly higher than the weight recorded in TVX-3236 plants. IT93K-452-1 and TVX-3236 plants however produced similar number of nodules that were significantly higher than the number recorded in IT99K-573-1-1 plants. There was no significant difference between the percentage of infective nodules of the three varieties.

The interaction between rhizobia inoculation and phosphorus application on nodule dry weight and number of nodules is presented in Table 4.26. The nodule dry weight of the uninoculated plants increased significantly with increase in P rates with the maximum value recorded at the maximum P application rate (40 kg P ha^{-1}). Similar trend was recorded in plants inoculated with USDA 3451 but the increase in the nodule dry weight as a result of additional 20 kg P ha^{-1} application was not significant. In plants inoculated with USDA 3384, plants that received 20 kg P ha^{-1} produced significantly heavier nodules than those that received 40 kg P ha^{-1}. Among all the

Table 4.25: Effects of rhizobia inoculation and phosphorus application on nodule dry weight, number and effectiveness of cowpea varieties in 2015

Treatments	Nodules Dry weight (g/plant)	Nodule no	Nodule Effectiveness (%)
Inoculation (I)			
Uninoculated	0.10a	15.06b	71.89a
USDA 3451	0.11a	17.11ab	67.64a
USDA 3384	0.11a	19.92a	65.76a
M S D	0.02	4.52	10.23
Phosphorus (P)			
0 kg ha^{-1}	0.01b	4.49b	45.43b
20 kg ha^{-1}	0.15a	23.21a	80-89a
40 kg ha^{-1}	0.16a	24.39a	79.29a
M S D	0.02	4.52	10.23
Variety (V)			
IT93K-452-1	0.11a	22.27a	70.12a
IT99K-573-1-1	0.12a	11.77b	69.91a
TVX-3236	0.07b	18.06a	65.26a
M S D	3.40	0.02	10.23
Interactions			
I × P	**	**	NS
I × V	**	NS	NS
P × V	**	*	NS
I × P × V	**	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, *, ** - significant at P=0.05 and 0.01 respectively, NS- not significant at P=0.05

Table 4.26: Interaction effects of rhizobia inoculation and phosphorus on nodule dry weight and number of nodules in 2015

	Phosphorus (kg ha^{-1})		
	0	20	40
	Nodule dry weight (g/plant)		
Inoculation			
Uninoculated	0.004d	0.09c	0.21a
USDA 3451	0.02d	0.15abc	0.17ab
USDA 3384	0.01d	0.2a	0.1bc
SE+		0.01	
	Number of nodules per plant		
Uninoculated	5.14c	11.23c	28.82ab
USDA 3451	4.36c	24.89b	22.06b
USDA 3384	3.96c	33.51a	22.29b
SE±		2.76	

Means followed by different alphabets are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, SE- standard error of the mean treatment combinations, plants without P fertilization consistently produced the lightest nodules. Similar trend was observed for number of nodules.

The interaction between rhizobia inoculation and variety on nodule dry weight in 2015 is presented in Table 4.27. When IT93K-452-1 plants were uninoculated, the heaviest nodules were produced. The value was however similar to the weight recorded in the same variety inoculated with USDA 3451 but significantly higher than those recorded in plants inoculated with USDA 3384. There was no significant difference between the nodule weight of the inoculated and uninoculated plants of IT99K-573-1-1 and TVX-3236 varieties.

The interaction between phosphorus and variety on nodule dry weight and number of nodules is presented in Table 4.27. The nodule dry weight of IT93K-452-1 plants increased significantly with increase in P rates with the maximum obtained at 40 kg P ha^{-1}. In IT99K-573-1-1 plants however, P significantly increased nodule dry weight up to 20 kg P ha^{-1}. In TVX-3236 plants, the maximum nodule dry weight was obtained at 20 kg P ha^{-1} and application up to 40 kg P ha^{-1} reduced the nodule dry weight. The number of nodules of IT93K-452-1 and IT99K-573-1-1 plants increased with increase in P rates with the maximum obtained at 40 kg P ha^{-1} but there was no significant difference between the number recorded at 20 and 40 kg P ha^{-1}. Similar trend was observed in TVX-3236 plants. The interaction between rhizobia inoculation, phosphous application and variety on the nodule dry weight is presented in Figure 4.30. The result revealed that the nodule weight of the uninoculated IT93K-452-1 plants increased significantly with increase in P application with the maximum obtained at 40 kg P ha^{-1}. In uninoculated IT99K-573-1-1 and TVX-3236 plants however, the weight of the nodules produced at 20 and 40 kg P ha^{-1} application were similar. In IT93K-452-1 and IT99K-573-1-1 plants inoculated with USDA 3451 strain, plants that

Table 4.27: Interaction effects of rhizobia inoculation and variety on nodule dry weight, phosphorus and variety on nodule dry weight and number of nodules of cowpea in 2015

	Varieties		
	IT93K-452-1	IT99K-573-1-1	TVX-3236
	Nodule dry weight (g/plant)		
Inoculation			
Uninoculated	0.14ab	0.09abc	0.07bc
USDA 3451	0.13abc	0.14a	0.07c
USDA 3384	0.07c	0.13abc	0.12abc
SE±		0.01	
	Nodule dry weight (g/plant)		
Phosphorus			
0 kg P ha^{-1}	0.01d	0.02d	0.01d
20 kg P ha^{-1}	0.12bc	0.17ab	0.16ab
40 kg P ha^{-1}	0.21a	0.17ab	0.10c
SE ±		0.01	
	Number of nodules per plant		
Phosphorus			
0 kg P ha^{-1}	3.88e	3.86e	5.73e
20 kg P ha^{-1}	29.04ab	15.44d	25.15b
40 kg P ha^{-1}	33.88a	16.00cd	23.30bc
SE ±		2.76	

Means followed by different alphabets are significantly different at P=0.05 using Waller-Duncan K-ratio T-test. SE- standard error of the mean

received 40 kg P ha^{-1} had the heaviest nodules but the values recorded at 20 and 40 kg P ha^{-1} were similar.

In TVX-3236 plants inoculated with the same strain, plants that received 20 kg P ha^{-1} had the heaviest nodules. Plants inoculated with USDA 3384 produced the heaviest nodules at 20 kg P ha^{-1} application in the three varieties. Plants without P fertilization consistently produced the lightest nodules in all the treatment combinations.

The nodulation of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2016 is presented in Table 4.28. Plants inoculated with BR 3262 and the uninoculated plants had the heaviest nodules which were significantly heavier than the nodules of plants inoculated with BR 3262 strain and 90 kg N ha^{-1} fertilized plants which had the lightest nodules. Similar trend was observed for the number of nodules. The percentage of infected nodules of the inoculated and uninoculated plants were similar and significantly higher than the percentage recorded in plants fertilized with 90 kg N ha^{-1}. The nodule dry weight of plants fertilized with 20 and 0 kg P ha^{-1} were similar likewise the percentage of infected nodules and both were significantly higher than the values obtained in the unfertilized plants. The order of performance in respect of number of nodules was 40 kg P ha^{-1} > 20 kg P ha^{-1} > 0 kg P ha^{-1}. There was no significant difference between the three varieties in respect of nodule dry weight, number and percentage of infected nodules in 2016.

The nodulation of cowpea varieties in response to rhizobia inoculation and phosphorus application in 2017 is presented in Table 4.29. The nodule dry weight of the inoculated and uninoculated plants were similar and significantly higher than the value recorded in plants fertilized with 90 kg N ha^{-1}. The number of nodules recorded in the uninoculated plants and those inoculated with the two BR strains were similar. The number of nodules produced by plants inoculated with BR 3267 strain were significantly higher than those produced in USDA 3451 inoculated plants.

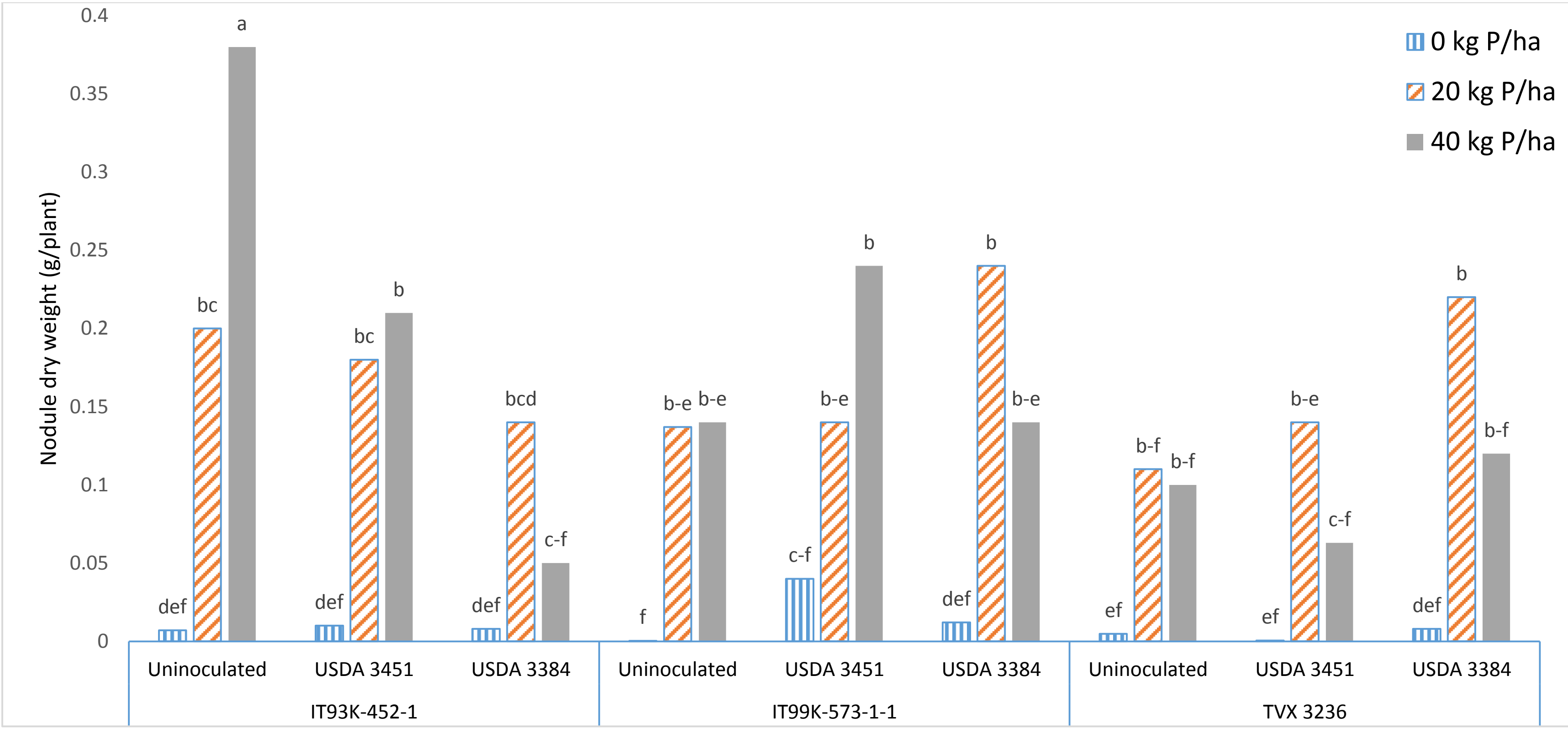


Figure 4.30: Interaction effects of inoculation, phosphorus and variety on nodule dry weight of cowpea in 2015

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

Table 4.28: Effects of rhizobia inoculation and phosphorus application on nodule dry weight, number and effectiveness of cowpea varieties in 2016

Treatments	Nodule dry weight (g/plant)	Number of nodules	Nodule Effectiveness (%)
Inoculation (I)			
Uninoculated	0.05a	8.46a	57.00a
90 kg N ha^{-1}	0.02b	4.69b	37.00b
BR 3262	0.05a	9.49a	60.00a
BR 3267	0.03b	4.12b	64.00a
MSD	0.02	2.82	13.48
Phosphorus (P)			
0 kg ha^{-1}	0.02b	4.13c	37.00b
20 kg ha^{-1}	0.04a	6.83b	61.00a
40 kg ha^{-1}	0.05a	9.11a	68.00a
MSD	0.02	2.44	11.68
Variety (V)			
IT93K-452-1	0.04a	6.58a	51.00a
IT99K-573-1-1	0.04a	7.79a	54.00a
TVX-3236	0.03a	5.70a	60.00a
MSD	0.02	2.44	11.68
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × V × P	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05

Plants fertilized with 90 kg N ha^{-1} produced the significantly lowest number of nodules. Plants inoculated with BR 3267 strain produced significantly lesser percentage of infected nodules than the uninoculated plants and those inoculated with BR 3262 strain. Plants fertilized with 90 kg N ha^{-1} had the least percentage of infected nodules and the values were significantly lower than what was recorded in the inoculated and uninoculated plants. Plants fertilized with 20 and 40 kg P ha^{-1} had similar number of nodules, nodule dry weight and percentage of infective nodules that were significantly higher than the values recorded in the unfertilized plants.

The nodule dry weight of IT93K-452-1 and IT99K-573-1-1 plants were similar and significantly higher than the dry weight of TVX-3236 plants. Similar trend was observed for number of nodules but there was no significant difference between the number of nodules produced by IT93K-452-1 and TVX-3236 plants. There was no significant difference between the percentage of infected nodules of the three varieties.

Table 4.30 shows the interaction between rhizobia inoculation and phosphorus application on number of nodules in 2017. The number of nodules of uninoculated plants, those fertilized with 90 kg N ha^{-1} and those inoculated with BR 3262 increased with increase in P rates with the maximum value obtained at 40 kg P ha^{-1}. However, the increase at 40 kg P ha^{-1} application was only significant in plants inoculated with BR 3262. Plants inoculated with BR 3267 and USDA 3451 produced the highest number of nodules at 20 kg P ha^{-1}.

The interaction between rhizobia inoculation and variety on number of nodules in 2017 is presented in Table 4.30. IT93K-452-1 plants produced the highest number of nodules when inoculated with BR 3267.

Table 4.29: Effects of rhizobia inoculation and phosphorus application on nodule dry weight, number and effectiveness of cowpea varieties in 2017

Treatments	Nodule dry weight (g/plant)	Number of nodules	Effectiveness (%)
Inoculation (I)			
Uninoculated	0.13a	10.64ab	70.37a
90 kg N ha^{-1}	0.02b	2.68c	35.93c
BR 3262	0.12a	11.28ab	71.11a
BR 3267	0.12a	11.97a	61.11b
USDA 3451	0.12a	9.88b	67.41ab
MSD	0.03	2.04	8.36
Phosphorus (P)			
0 kg ha^{-1}	0.05b	5.65b	52.00b
20 kg ha^{-1}	0.13a	10.61a	62.89a
40 kg ha^{-1}	0.13a	11.60a	68.67a
MSD	0.03	1.57	6.48
Variety (V)			
IT93K-452-1	0.12a	9.26ab	58.22a
IT99K-573-1-1	0.12a	10.31a	64.00a
TVX-3236	0.08b	8.29b	61.33a
MSD	0.03	1.57	6.48
Interactions			
I × P	NS	**	NS
I × V	NS	**	NS
P × V	NS	NS	NS
I × V × P	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05, **- significant at P=0.01, NS- not significant at P=0.05

Table 4.30: Interaction effects of rhizobia inoculation and phosphorus, rhizobia inoculation and varieties on nodule number of cowpea in 2017

		Phosphorus (kg ha^{-1})	
	0	20	40
Inoculation			
Uninoculated	7.29efg	11.70cd	12.93bc
90 kg N ha^{-1}	0.32i	3.74hi	3.96gh
BR 3262	8.12ef	8.34def	17.37a
BR 3267	6.33fgh	16.07ab	14.50bc
USDA 3451	6.22fgh	13.21bc	10.22cde
SE +		1.25	
		Varieties	
	IT93K-452-1	IT99K-573-1-1	TVX-3236
Inoculation			
Uninoculated	11.88abc	11.57abc	8.46cd
90 kg N ha^{-1}	2.51f	2.05f	3.47ef
BR 3262	8.43cd	11.81abc	13.59a
BR 3267	14.06a	12.56ab	9.27bcd
USDA 3451	9.44bcd	13.54a	6.66de
SE$\pm$		1.25	

Means followed by different alphabets are significantly different at P=0.05 using Waller-Duncan K-ratio T-test. SE- standard error of the mean

The value was however similar to the numbers produced in the uninoculated plants. In IT99K-573-1-1 variety, there was no significant difference between the number of nodules of the inoculated and uninoculated plants. The uninoculated TVX-3236 plants produced the highest number of nodules and the value was significantly higher than the number produced in uninoculated and other inoculated plants of the same variety which were similar. Among all the treatment combinations, plants fertilized with 90 kg N ha^{-1} produced the least nodules in the three varieties.

ii. Nitrogen uptake

The N uptake of cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.31. There was no significant difference between the quantity of N taken up by the inoculated and uninoculated plants in 2015. Similarly in 2016, there was no significant difference between the N-uptake of inoculated and uninoculated plants. However, plants inoculated with BR 3262 had significantly higher N-uptake than plants fertilized with 90 kg N ha^{-1} which equally had similar N-uptake with the uninoculated and plants inoculated with BR 3267. In 2017, plants fertilized with 90 kg N ha^{-1} had the highest N-uptake. This was similar to the value recorded in the uninoculated plants and both were significantly higher than the values recorded in plants inoculated with BR 3262 and BR 3267 strains. The values obtained in plants inoculated with USDA 3451 and uninoculated plants were similar.

The order of N uptake was 40 kg P ha^{-1} > 20 kg P ha^{-1} > 0 kg P ha^{-1} in 2015. In 2016 and 2017, the N uptake of plants fertilized with 20 and 40 kg P ha^{-1} were similar and significantly higher than the N-uptake of the unfertilized plants. In 2015, the N-uptake of IT93K-452-1 plants was the highest and similar to the value recorded in IT99K-573-1-1 plants. The value recorded in TVX-

3236 plants were however the lowest and significantly lower than that of IT93K-452-1 plants. In 2016 and 2017, the N-uptake of IT93K-452-1 and IT99K-573-1-1 plants were similar and significantly higher than the value obtained in TVX-3236 plants.

The interaction between phosphorus and variety on N-uptake in 2016 revealed that the N-uptake of IT93K-452-1 and IT99K-573-1-1 plants increased with increase in P rates up to 40 kg P ha^{-1} but in IT93K-452-1, there was no significant difference between the value recorded at 20 and 40 kg P ha^{-1} application (Table 4.32). In TVX-3236 variety, the highest N–uptake was obtained at 20 kg P ha^{-1}. The unfertilized plants consistently had the least N-uptake values in the three varieties and the highest was obtained in IT99K-573-1-1 plants that received 40 kg P ha^{-1}.

The interaction between rhizobia inoculation and phosphorus application on N-uptake in 2017 is presented in Table 4.32. The uninoculated plants, 90 kg N ha^{-1} fertilized plants and plants inoculated with USDA 3451 had their highest N uptake when fertilized with 20 kg P ha^{-1}. Plants inoculated with BR 3262 and BR 3267 rhizobia strains however had their highest N uptake values at 40 kg P ha^{-1}. There was however no significant difference between the values recorded at 20 and 40 kg P ha^{-1} in plants inoculated with BR 3262 strain.

iii. Nitrogen fixation

The N fixation of the three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.33. There was no significant difference between the quantity of N fixed by the inoculated and uninoculated plants in 2015 and 2016. In 2017, the uninoculated plants fixed significantly higher quantity of N than the inoculated plants. In 2015 and 2017, the quantity of N fixed by plants that received 20 and 40 kg P ha^{-1} were similar and significantly higher than the quantity fixed by the unfertilized plants. In 2016 however, 40 kg P ha^{-1} only fixed significantly higher N quantity than the unfertilized plants.

Table 4.31: Effects of rhizobia inoculation and phosphorus application on N-uptake (g/plant) of cowpea varieties

	Cropping seasons		
	2015	2016	2017
Treatments			
Inoculation (I)			
Uninoculated	0.81a	0.47ab	0.53ab
USDA 3451	0.77a		0.45bc
USDA 3384	0.76a		
BR 3262		0.54a	0.43c
BR 3267		0.43ab	0.43c
90 kg N ha^{-1}		0.41b	0.57a
M S D	0.13	0.12	0.08
Phosphorus (P)			
0 kg ha^{-1}	0.39c	0.30b	0.31b
20 kg ha^{-1}	0.88b	0.50a	0.57a
40 kg ha^{-1}	1.07a	0.59a	0.56a
M S D	0.13	0.11	0.07
Variety (V)	0.88a	0.53a	0.51a
IT93K-452-1	0.75ab	0.52a	0.52a
IT99K-573-1-1	0.69b	0.34b	0.42b
TVX-3236	0.13	0.11	
M S D			
Interactions			
I × P	NS	NS	**
I × V	NS	NS	NS
P × V	NS	*	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test. MSD-Minimum significant difference, *, **- significant at P=0.05 and 0.01 respectively, NS- not significant at P=0.05

Table 4.32: Interaction effects of phosphorus and variety, rhizobia inoculation and phosphorus application on N-uptake (g/plant) of cowpea in 2016 and 2017 respectively.

	Phosphorus (kg ha^{-1})		
	0	20	40
Variety		**2016**	
IT93K-452-1	0.37de	0.56bc	0.64ab
IT99K-573-1-1	0.32de	0.48bcd	0.77a
TVX-3236	0.21e	0.45cd	0.35de
SE±		0.07	
Inoculation		**2017**	
Uninoculated	0.35e	0.75a	0.48bcd
90 kg N ha^{-1}	0.27e	0.72a	0.71a
BR 3262	0.27e	0.49bcd	0.53b
BR 3267	0.36de	0.37cde	0.55b
USDA 3451	0.31e	0.54b	0.51bc
SE±		0.05	

Means followed by different alphabets are significantly different at P=0.05 using Waller-Duncan K-ratio T-test. SE- standard error of the mean

quantity of N fixed by plants that received 20 and 40 kg P ha^{-1} were similar but plants that received IT93K-452-1 plants fixed significantly higher N than the remaining two varieties which fixed similar N quantity in 2015. In 2016 IT93K-452-1 and IT99K-573-1-1 fixed similar N quantity and the value recorded in IT99K-573-1-1 plants were significantly higher than that of TVX 326 plants. In 2017, IT93K-452-1 and IT99K-573-1-1 plants fixed similar N quantity that were significantly higher than the value recorded in TVX-3236 which fixed the least quantity of N in the three years.

iv. Percentage nitrogen derived from atmosphere (NDFA)

Percentage of N derived from the atmosphere of the three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.34.There was no significant difference between the quantity of N the uninoculated and inoculated plants derived from the atmosphere in 2015 and 2016. In 2017, there was no significant difference between the NDFA of the inoculated and uninoculated plants except that plants inoculated with BR 3267 derived significantly lower N from the atmosphere than the uninoculated plants.

Plants that received 20 and 40 kg P ha^{-1} derived similar percentage of N from the atmosphere in 2015 and 2017 and the value was significantly higher than what was obtained in the unfertilized plants. In 2016, there was no significant difference between the percentage NDFA of the fertilized and unfertilized plants.

IT99K-573-1-1 plants derived the highest N from the atmosphere in 2015 and the value was significantly higher than the NDFA of IT93K-452-1 and TVX-3236 plants which were similar. In 2016, IT99K-573-1-1 plants had the highest NDFA which was similar to the values recorded in IT93K-452-1 plants but significantly higher than the value recorded in TVX-3236 plants. In 2017, IT93K-452-1 and IT99K-573-1-1 plants had similar NDFA values which were significantly higher

Table 4.33: Effects of rhizobia inoculation and phosphorus application on nitrogen fixation (kg ha^{-1}) of cowpea varieties

	Cropping seasons		
	2015	**2016**	**2017**
Treatments			
Inoculation (I)			
Uninoculated	57.33a	37.15a	36.85a
USDA 3451	52.00a		27.08b
USDA 3384	52.00a		
BR 3262		31.70a	24.43b
BR 3267		29.16a	23.62b
M S D	17.33	16.03	9.25
Phosphorus (P)			
0 kg ha^{-1}	24.00b	22.27b	18.40b
20 kg ha^{-1}	62.67a	33.62ab	34.68a
40 kg ha^{-1}	66.67a	44.13a	30.68a
M S D	17.33	16.03	8.01
Variety (V)			
IT93K-452-1	68.00a	34.59ab	34.64a
IT99K-573-1-1	50.67b	44.32a	31.22a
TVX-3236	42.67b	19.11b	18.13b
M S D	17.33	16.03	8.01
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, NS- not significant at P=0.05

Table 4.34: Effects of rhizobia inoculation and phosphorus application on percentage of nitrogen derived from the atmosphere (NDFA) (%) of cowpea varieties

		Cropping seasons	
	2015	**2016**	**2017**
Treatments			
Inoculation (I)			
Uninoculated	46.86a	48.89a	49.98a
USDA 3451	46.75a		36.43ab
USDA 3384	49.03a		
BR 3262		45.36a	35.06ab
BR 3267		48.11a	32.03b
M S D	4.52	12.87	10.69
Phosphorus (P)			
0 kg ha^{-1}	43.23b	47.81b	29.97b
20 kg ha^{-1}	48.72a	42.58b	39.50a
40 kg ha^{-1}	50.69a	61.98a	41.16a
M S D	4.52	12.87	9.25
Variety (V)	52.71a	49.75ab	44.00a
IT93K-452-1	45.44b	65.50a	42.03a
IT99K-573-1-1	44.49b	37.12b	24.61b
TVX-3236	4.52	12.87	9.25
M S D			
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, NS- not significant at P=0.05

than the value recorded in TVX-3236 plants which derived the least percentage of N from the atmosphere in the three years.

4.1.2.4 Number of days to flowering

The number of days to flowering of the three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.35. The uninoculated plants flowered significantly earlier than the inoculated plants in 2015. Similarly, the uninoculated plants were the earliest to reach 50% flowering. This was followed by plants inoculated with USDA 3384 and plants inoculated with USDA 3451 were the latest to attain 50% flowering. In 2016 and 2017, there was no significant difference between the number of days to 1st flower at sight and 50% flowering of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants.

In 2015, plants fertilized with 20 and 40 kg P ha^{-1} started flowering at similar time but they flowered significantly earlier than the unfertilized plants. Plants fertilized with 40 kg P ha^{-1} were the earliest to reach 50% flowering and they were similar to plants fertilized with 20 kg P ha^{-1} but significantly earlier than the unfertilized plants. In 2016 and 2017, there was no significant difference between the number of days to 1st flower sighting in the fertilized and unfertilized plants. Plants that received 20 and 40 kg P ha^{-1} reached 50% flowering at similar time but it was significantly earlier than the number observed in the unfertilized plants in 2016. In 2017, there was no significant difference between the days to 50% flowering in fertilized and unfertilized plants.

IT93K-452-1 plants were the earliest to flower and earliest to reach 50% flowering. This was followed by IT99K-573-1-1 plants and the latest to flower and reach 50% flowering was TVX-3236 plants in the three years. In 2016 however, there was no significant difference between the number of days to 1st flower at sight in IT93K-452-1 and IT99K-573-1-1 variety and in 2017, the number of days to reach 50% flowering in these two varieties were not significantly different.

Table 4.35: Effects of rhizobia inoculation and phosphorus application on number of days to first flower at sight and 50% flowering of cowpea varieties

	2015		2016		2017	
	Days to 1st flower at sight	**Days to 50% flowering**	**Days to 1st flower at sight**	**Days to 50% flowering**	**Days to 1st flower at sight**	**Days to 50% flowering**
Treatments						
Inoculation (I)						
Uninoculated	44.740b	51.63c	39.59a	50.07a	40.44a	48.30a
USDA 3451	46.07a	62.96a			39.96a	47.85a
USDA 3384	45.820a	52.67b				
BR 3262			40.89a	59.70a	40.41a	47.56a
BR 3267			40.00a	49.41a	40.04a	47.85a
90 kg N ha^{-1}			40.04a	50.63a	40.30a	47.44a
MSD	0.64	0.83	1.53	1.54	1.46	1.62
Phosphorus(P)						
0 kg ha^{-1}	46.19a	53.11a	40.78a	51.61a	40.62a	48.27a
20 kg ha^{-1}	45.52b	52.41ab	39.50a	49.47b	39.91a	47.62a
40 kg ha^{-1}	44.93b	51.74b	40.11a	49.53b	40.16a	47.51a
MSD	0.64	0.83	1.32	1.33	1.13	1.26
Variety(V)						
IT93K-452-1	41.04c	47.74c	37.47b	46.44c	38.16c	46.16b
IT99K-573-1-1	43.15b	50.07b	38.19b	48.42b	39.29b	46.40b
TVX-3236	52.44a	59.44a	4.72a	55.75a	43.24a	50.84a
MSD			1.32	1.33	1.13	1.26
Interactions						
I × P	NS	NS	NS	NS	NS	NS
I × V	NS	NS	NS	NS	NS	NS
P × V	NS	NS	NS	NS	NS	NS
I × P × V	NS	NS	NS	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, NS- not significant at P=0.05

4.1.2.5 Yield attributes

i. Pod length

The pod length of three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.36. There was no significant difference between the pod lengths of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants in the three years. Though plants that received 20 and 40 kg P ha^{-1} had longer pods than the unfertilized plants in the three years, the difference was not significant in 2015. The order of performance in respect of pod length in the three years was IT99K-573-1-1 > IT93K-452-1 > TVX-3236.

ii. Number of seed per pod

The number of seeds per pod of the three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.37. There was no significant difference between the number of seed per pod of the inoculated, uninoculated and 90 kg Nha^{-1} fertilized plants in the three years. In 2015 and 2016, there was no significant difference between the number of seeds per pod of the P fertilized and unfertilized plants but in 2017, plants fertilized with 40 kg P ha^{-1} had significantly higher number of seeds per pod than the 20 kg P ha^{-1} fertilized and unfertilized plants which had similar number of seeds per pod. In 2015 and 2017, IT93K-452-1 and TVX-3236 plants produced similar number of seeds per pod which were significantly higher than the number of seeds produced by IT99K-573-1-1 plants. In 2016, IT93K-452-1 and IT99K-573-1-1 plants had similar number of seeds per pod which were significantly lower than the number recorded in TVX-3236 plants.

Table 4.36: Pod length (cm) of three cowpea varieties as affected by rhizobia inoculation and phosphorus application

	Cropping seasons		
	2015	2016	2017
Treatments			
Inoculation (I)			
Uninoculated	16.06a	15.07a	15.36a
USDA 3451	16.64a		14.66a
USDA 3384	15.95a		
BR 3262		15.31a	15.29a
BR 3267		15.08a	14.53a
90 kg N ha^{-1}		15.79a	14.54a
M S D	0.99	1.03	0.89
Phosphorus (P)			
0 kg ha^{-1}	15.85a	14.41b	14.35b
20 kg ha^{-1}	16.63a	15.68a	14.99ab
40 kg ha^{-1}	16.17a	15.84a	15.29a
M S D	0.99	0.89	0.69
Variety (V)			
IT93K-452-1	15.23b	15.61b	15.0b
IT99K-573-1-1	19.21a	17.13a	16.60a
TVX-3236	14.21c	13.19c	13.02c
M S D	0.99	0.89	0.69
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05

Table 4.37: Number of seed per pod of three cowpea varieties as affected by rhizobia inoculation and phosphorus application

	Cropping seasons		
	2015	2016	2017
Treatments			
Inoculation (I)			
Uninoculated	11.98a	11.32a	10.45a
USDA 3451	11.83a		10.01a
USDA 3384	11.96a		
BR 3262		11.41a	10.04a
BR 3267		11.88a	10.63a
90 kg N ha^{-1}		11.68a	10.63a
M S D	0.88	1.12	0.73
Phosphorus (P)			
0 kg ha^{-1}	11.76a	11.22a	9.83b
20 kg ha^{-1}	11.65a	11.73a	10.16b
40 kg ha^{-1}	12.35a	11.77a	10.97a
M S D	0.89	0.96	0.56
Variety (V)	12.06a	11.09b	10.63a
IT93K-452-1	10.96b	11.21b	9.56b
IT99K-573-1-1	12.74a	12.41a	10.82a
TVX-3236	0.89	0.96	0.56
M S D			
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test. MSD-Minimum significant difference, NS- not significant at P=0.05

iii. One hundred seed weight

The one hundred seed weight of the three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.38.There was no significant difference between the seed weight of the inoculated and uninoculated plants in 2015. In 2016, plants fertilized with 90 kg N ha^{-1} had significantly heavier seeds than the inoculated and uninoculated plants which had similar seed weight. In 2017, there was no significant difference between the seed weight of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants.

Plants fertilized with 20 and 40 g P ha^{-1} had similar heavier seeds than the unfertilized plants in 2015 and 2016. In 2017 however, there was no significant difference between the seed weight of P fertilized and unfertilized plants. The order of performance of the varieties in respect of weight of seeds was IT99K-573-1-1 > IT93K-452-1 > TVX-3236 in the three years

iv. Number of pods per plant

The number of pod produced by the three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.39. There was no significant difference between the number of pods produced per plant in the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants in the three years. In 2015 and 2016, the number of pods produced by plants fertilized with 20 and 40 kg P ha^{-1} were similar and were significantly higher than the number of pods produced by the unfertilized plants. In 2017, the order of performance in respect of number of pods per plants was 40 kg P ha^{-1} > 20 kg P ha^{-1} > 0 kg P ha^{-1}.

There was no significant difference between the number of pods produced by the three varieties in 2015. In 2016, the quantity of pods produced by TVX-3236 were significantly higher than the quantity produced by IT93K-452-1 and IT99K-573-1-1 which produced similar number of pods.

Table 4.38: One hundred seed weight of three cowpea varieties as affected by rhizobia inoculation and phosphorus application

	Cropping seasons		
	2015	2016	2017
Treatments			
Inoculation (I)			
Uninoculated	15.96a	13.16b	13.34a
USDA 3451	16.07a		13.09a
USDA 3384	15.03a		
BR 3262		13.78b	13.45a
BR 3267		13.46b	13.38a
90 kg N ha^{-1}		14.56a	13.53a
M S D	1.31	0.73	0.74
Phosphorus (P)			
0 kg ha^{-1}	14.45b	13.02b	13.13a
20 kg ha^{-1}	16.30a	14.03a	13.49a
40 kg ha^{-1}	16.31a	14.17a	13.45a
M S D	1.31	0.64	0.58
Variety (V)	15.84b	14.28b	13.70b
IT93K-452-1	18.93a	15.90a	16.60a
IT99K-573-1-1	12.29c	11.04c	9.78c
TVX-3236	1.31	0.64	0.58
M S D			
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05

In 2017, TVX-3236 produced significantly higher number of pods than the IT93K-452-1 but there was no significant difference between the quantity produced by TVX-3236 and IT99K-573-1-1 plants as well as IT99K-573-1-1 and IT93K-452-1 plants.

v. Shell percentage

The shell percentage of the three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.40. There was no significant difference between the shell percentage of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants in the three years. Similarly, there was no significant difference between the shell percentage of the P fertilized and unfertilized plants in 2015 and 2017. In 2016, the unfertilized plants had significantly higher shell percentage than the 20 and 40 kg P fertilized plants which had similar shell percentage. There was no significant difference between the shell percentage of IT93K-452-1 and IT99K-573-1-1 plants but both had significantly higher shell percentage than TVX-3236 plants in 2015. In 2016, IT93K-452-1 and IT99K-573-1-1 had similar shell percentage but the shell percentage of TVX-3236 was only significantly lower than that of IT93K-452-1 plants. In 2017, there was no significant difference among the shell percentage of the three varieties.

Table 4.39: Number of pods per plant of three cowpea varieties as affected by rhizobia inoculation and phosphorus application

	Cropping seasons		
	2015	2016	2017
Treatments			
Inoculation (I)			
Uninoculated	8.23a	7.20a	7.73a
USDA 3451	8.60a		7.71a
USDA 3384	8.39a		
BR 3262		5.93a	7.94a
BR 3267		6.78a	7.43a
90 kg N ha^{-1}		7.20a	8.03a
M S D	1.65	1.41	1.15
Phosphorus (P)			
0 kg ha^{-1}	6.71b	5.57b	5.70c
20 kg ha^{-1}	8.84a	7.09a	8.30b
40 kg ha^{-1}	9.68a	7.66a	9.30a
M S D	1.65	1.22	0.89
Variety (V)			
IT93K-452-1	7.96a	6.13b	7.14b
IT99K-573-1-1	8.36a	6.28b	7.72ab
TVX-3236	8.90a	7.90a	8.45a
M S D	1.65	1.22	0.89
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05

Table 4.40: Shell percentage (%) of three cowpea varieties as affected by rhizobia inoculation and phosphorus application

	Cropping seasons		
	2015	**2016**	**2017**
Treatments			
Inoculation (I)			
Uninoculated	22.64a	23.75a	26.52a
USDA 3451	24.20a		26.92a
USDA 3384	23.32a		
BR 3262		22.15b	28.80a
BR 3267		22.62ab	27.86a
90 kg N ha^{-1}		23.71a	28.26a
M S D	2.24	1.56	3.2
Phosphorus (P)			
0 kg ha^{-1}	23.73a	25.92a	27.97a
20 kg ha^{-1}	23.77a	21.98b	28.00a
40 kg ha^{-1}	22.66a	21.27b	27.04a
M S D	2.24	1.35	2.48
Variety (V)			
IT93K-452-1	24.31a	24.34a	27.14a
IT99K-573-1-1	24.73a	23.04ab	28.09a
TVX-3236	21.14b	21.79b	27.78a
M S D	2.24	1.35	2.48
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05

vi. Shoot biomass yield

The shoot biomass yield of the three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.41. In 2015, the uninoculated plants had the highest shoot biomass yield which was significantly higher than the shoot biomass yield of plants inoculated with USDA 3451 but similar to the shoot biomass yield of plants inoculated with USDA 3384. In 2016 and 2017, plants fertilized with 90 kg N ha^{-1} had the highest shoot biomass yield which was similar to the value recorded in the uninoculated plants and the least shoot biomass yield was obtained in plants inoculated with BR 3267 in the two years. Plants fertilized with 20 and 40 kg P ha^{-1} had similar shoot biomass yield in 2015 and 2016 which were significantly higher than the shoot biomass yield of the unfertilized plants. In 2017, the order of performance in respect of shoot biomass yield was 40 kg P ha^{-1} > 20 kg P ha^{-1} > 0 kg P ha^{-1}. In 2015, there was no significant difference between the shoot biomass yield of the three varieties. In 2016 and 2017 however, IT93K-452-1 and IT99K-573-1-1 had similar shoot biomass yield which were significantly higher than the shoot biomass yield of TVX-3236 plants.

The interaction between rhizobia inoculation and phosphorus application on shoot biomass yield in 2017 is presented in Table 4.42. Phosphorus application increased the shoot biomass yield up to 20 kg P ha^{-1} application after which there was a decline in all the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants except in plants inoculated with BR 3267 which had the highest shoot biomass yield at 40 kg P ha^{-1}. However, there was no significant difference between the value recorded at 20 and 40 kg P ha^{-1} application.

Table 4.41: Shoot biomass yield (t ha^{-1}) of three cowpea varieties as affected by rhizobia inoculation and phosphorus application

	Cropping seasons		
	2015	**2016**	**2017**
Treatments			
Inoculation (I)			
Uninoculated	2.77a	2.00ab	1.85ab
USDA 3451	2.31b		1.64c
USDA 3384	2.54ab		
BR 3262		1.71ab	1.66bc
BR 3267		1.67b	1.46c
90 kg N ha^{-1}		2.11a	1.90a
M S D	0.45	0.43	0.20
Phosphorus (P)			
0 kg ha^{-1}	1.39b	1.22b	1.22c
20 kg ha^{-1}	3.00a	2.11a	2.03a
40 kg ha^{-1}	3.22a	2.28a	1.86b
M S D	0.45	0.37	0.16
Variety (V)			
IT93K-452-1	2.81a	2.01a	1.76a
IT99K-573-1-1	2.43a	2.24a	1.91a
TVX-3236	2.37a	1.38b	1.44b
M S D	0.45	0.37	0.16
Interactions			
I × P	NS	NS	**
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD-Minimum significant difference, NS- not significant at P=0.05

Table 4.42: Interaction effects of rhizobia inoculation and phosphorus application on shoot biomass yield of cowpea in 2017

	Phosphorus (kg ha^{-1})		
	0	20	40
Inoculation			
Uninoculated	1.69cde	2.31ab	1.55def
90 kg N ha^{-1}	0.89h	2.52a	2.29ab
BR 3262	1.09gh	1.97bc	1.92c
BR 3267	1.31fg	1.42efg	1.65c-f
USDA 3451	1.12gh	1.93c	1.87cd
SE±		0.12	

Means followed by different alphabets are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, SE-standard error of the mean

vii. Pod yield

The pod yield of the three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.43. In 2015, plants inoculated with USDA 3451, had the highest pod yield (1.89 t ha^{-1}). This was however similar to the value recorded in the uninoculated plants (1.77 t ha^{-1}). Plants inoculated with USDA 3384 had the lowest pod yield (1.67 t ha^{-1}) which was significantly lower than the pod yield of USDA 3451 plants. In 2016, plants fertilized with 90 kg N ha^{-1} had the highest pod yield (1.99 t ha^{-1}) which was significantly higher than the pod yield of BR 3262 plants which had the lowest pod yield (1.58 t ha^{-1}). There was however no significant difference between the pod yield of the inoculated and uninoculated plants. In 2017, the pod yield of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants were similar.

Plants fertilized with 20 and 40 kg P ha^{-1} had similar pod yield in 2015 and 2016 which were significantly higher than the pod yield of the unfertilized plants. In 2017, the order of performance in respect of pod yield was 40 kg P ha^{-1} > 20 kg P ha^{-1} > 0 kg P ha^{-1}. In 2015 and 2017, IT99K-573-1-1 produced significantly higher pod yield than IT93K-452-1 and TVX-3236 plants which produced similar pod yield. In 2016 however, there was no significant difference between the pod yield of the three varieties.

Table 4.43: Pod yield of three cowpea varieties as affected by rhizobia inoculation and phosphorus application

	Cropping seasons		
	2015	**2016**	**2017**
Treatments			
Inoculation (I)			
Uninoculated	1.77ab	1.76ab	1.78a
USDA 3451	1.89a		1.79a
USDA 3384	1.67b		
BR 3262		1.58b	1.85a
BR 3267		1.72ab	1.72a
90 kg N ha^{-1}		1.994a	1.85a
M S D	0.21	0.38	0.27
Phosphorus (P)			
0 kg ha^{-1}	1.06b	1.20b	1.30c
20 kg ha^{-1}	2.08a	1.97a	1.93b
40 kg ha^{-1}	2.19a	2.13a	2.16a
M S D	0.21	0.331	0.21
Variety (V)			
IT93K-452-1	1.67b	1.630a	1.68b
IT99K-573-1-1	2.15a	1.850a	2.03a
TVX-3236	1.51b	1.81a	1.68b
M S D	0.21	0.33	0.21
Interactions			
I × P	**	NS	*
I × V	**	NS	NS
P × V	**	NS	NS
I × P × V	**	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, *, **- significant at P= 0.05 and 0.01 respectively, NS- not significant at P=0.05

The interaction between rhizobia inoculation and phosphorus application on pod yield in 2015 and 2017 is presented in Figure 4.31 A and B respectively. In 2015, P application increased the pod yield of uninoculated plants and plants inoculated with USDA 3451 up to 20 kg P ha^{-1} application after which there was a decline at 40 kg P ha^{-1}.

In plants inoculated with USDA 3384 however, pod yield increased significantly with increase in P application up to 40 kg P ha^{-1}. In 2017, P application increased the pod yield up to 40 kg P ha^{-1} application in all the inoculated plants and plants fertilized with 90 kg N ha^{-1}. However, the difference between 20 and 40 kg ha^{-1} was only significant in plants fertilized with 90 kg N ha^{-1}. Among all the treatment combination, the highest pod yield (2.51 t ha^{-1}) was obtained in 90 kg N ha^{-1} fertilized plants that received 40 kg Pha^{-1} and the least (1.00 t ha^{-1}) was obtained in the same 90 kg N ha^{-1} fertilized plants that did not receive P fertilizer.

The interaction between rhizobia inoculation and variety on pod yield in 2015 is presented in Figure 4. 31C. When IT93K-452-1 plants were inoculated with USDA 3451, significantly higher pod yield was recorded than when uninoculated. However, uninoculated IT99K-573-1-1 plants produced significantly higher pod yield than when the same variety was inoculated with USDA 3384 strain. In TVX-3236 variety, there was no significant difference between the pod yield of the inoculated and uninoculated plants.

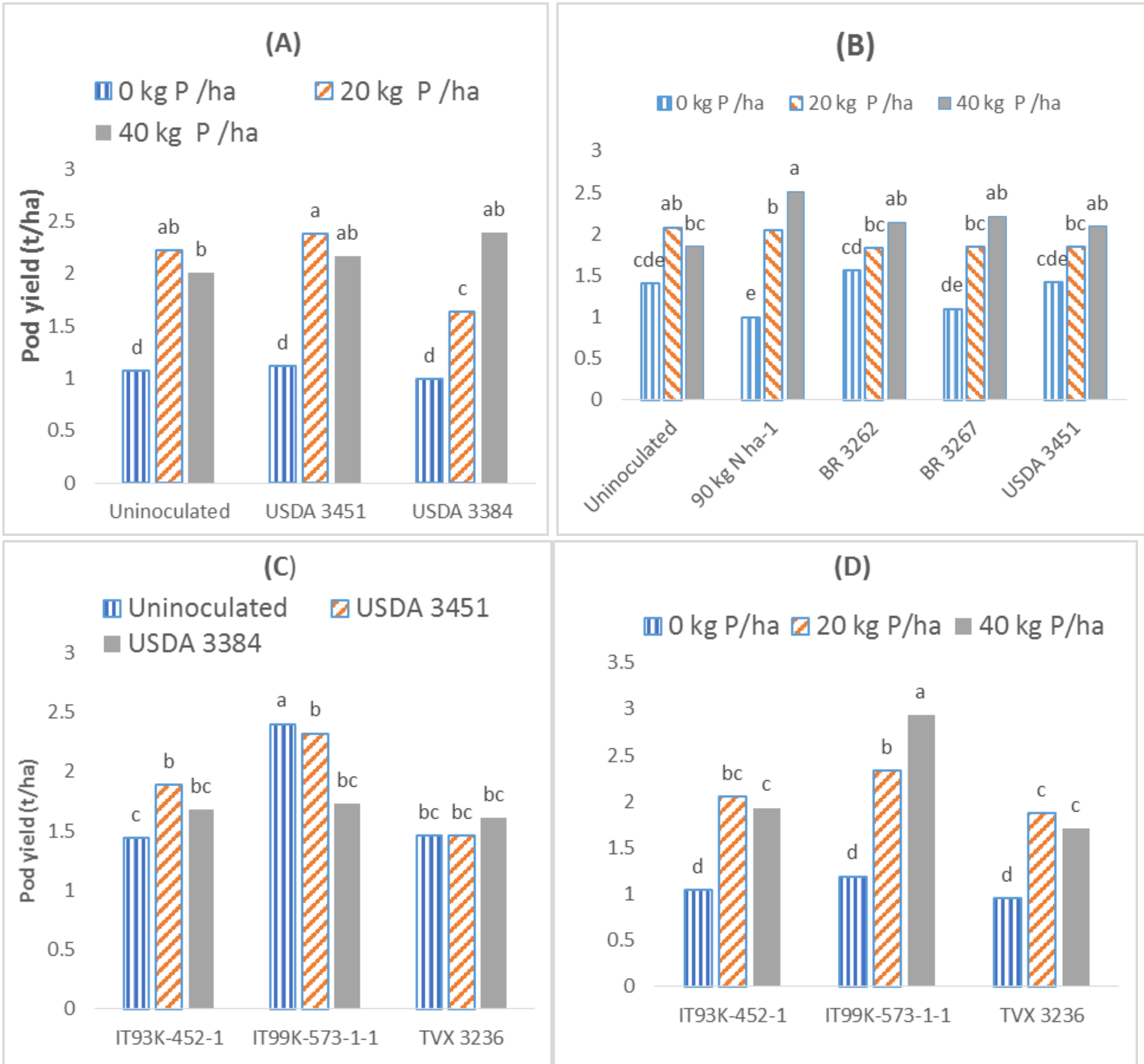


Figure 4.31: Interaction effects of (A) rhizobia inoculation and phosphorus on pod yield of cowpea in 2015 (B) rhizobia inoculation and phosphorus application on pod yield of cowpea in 2017 (C) rhizobia inoculation and varieties on pod yield of cowpea in 2015 (D) phosphorus and varieties on pod yield of cowpea in 2015

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

The interaction between phosphorus and variety on pod yield in 2015 is presented in Figure 4.31D. IT93K-452-1 plants and TVX-3236 plants produced the highest pod yield when fertilized with 20 kg P ha^{-1}. IT99K-573-1-1 plants however had its highest pod yield when fertilized with 40 kg P ha^{-1}.

The interaction between rhizobia inoculation, phosphorus application and variety on pod yield is presented in Figure 4.32. All the uninoculated plants of the three varieties produced their highest grain yield when fertilized with 20 kg P ha^{-1}. IT93K-452-1 and TVX-3236 plants inoculated with USDA 3451 had their highest pod yield when fertilized with 20 kg P ha^{-1}. IT99K-573-1-1 plants inoculated with the same strain produced the highest pod yield at 40 kg P ha^{-1} application. In IT99K-573-1-1 and TVX-3236 plants inoculated with USDA 3451, the significantly highest pod yield was obtained at 40 kg P ha^{-1}. In IT9K-452-1 plants inoculated with the same strain, the pod yield obtained at 20 and 40 kg P ha^{-1} was similar. The unfertilized plants generally had the least pod yield in all the treatment combination. Among the unfertilized plants, uninoculated and USDA 3451 inoculated IT99K-573-1-1 plants and USDA 3384 inoculated IT93K-452-1 plants had pod yield above 1 t ha^{-1}. Among all the treatment combination, 40 kg P ha^{-1} fertilized IT99K-573-1-1 plants inoculated with USDA 3451 strain had the highest pod yield (3.04 t ha^{-1}) but similar value (2.98 t ha^{-1}) was obtained in 20 kg P ha^{-1} fertilized uninoculated IT99K-573-1-1 plants. The least pod yield (0.59 t ha^{-1}) was recorded in unfertilized IT99K-573-1-1 plants inoculated with USDA 3384.

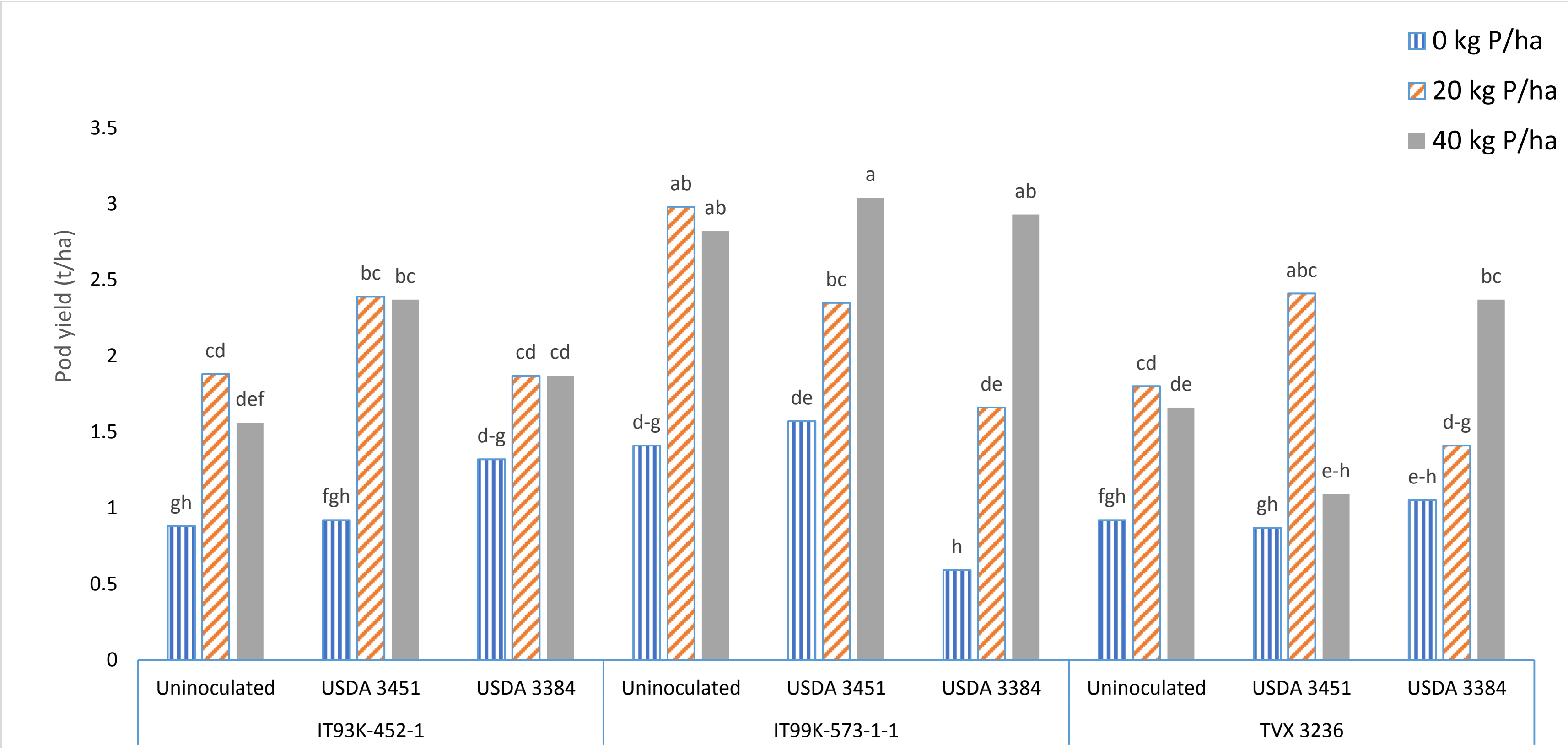


Figure 4.32: Interaction effects of rhizobia inoculation, phosphorus application and varieties on pod yield of cowpea in 2015

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

viii. Grain yield

The grain yield of the three cowpea varieties in response to rhizobia inoculation and phosphorus application in the three years is presented in Table 4.44. In 2015, plants inoculated with USDA 3451 had the highest mean grain yield (1.43 t ha^{-1}) and the median value of 1.47 t ha^{-1}. The lower and upper quartile range of 0.93 and 1.94 t ha^{-1} respectively were recorded in plants inoculated with this strain. Next in performance were the uninoculated plants with the mean and median values of 1.36 and 1.29 t ha^{-1} respectively. The lower and upper quartile grain yield values of the uninoculated plants were 1.03 and 1.59 t ha^{-1} respectively. Plants inoculated with USDA 3384 had the least mean grain yield (1.36 t ha^{-1}) and the median value of 1.29 t ha^{-1}. The lower and upper quartile grain yield values of the plants inoculated with this strain were 0.89 and 1.67 t ha^{-1} respectively (Figure 4.33A).

In 2016, plants fertilized with 90 kg N ha^{-1} had significantly higher grain yield than plants inoculated with BR 3262. There was however no significant difference between the grain yield of the inoculated and uninoculated plants (Table 4.44). Plants that received 90 kg N ha^{-1} had mean grain yield value of 1.54 t ha^{-1} and median value of 1.34 t ha^{-1}. The interquartile range of 1.01 - 1.90 t ha^{-1} was obtained in these plants. Next in performance were the uninoculated plants with the mean and median values of 1.35 and 1.23 t ha^{-1} respectively. The lower and upper quartile grain yield values of the uninoculated plants were 0.93 and 1.61 t ha^{-1} respectively. This was followed by BR 3267 inoculated plants with the mean grain yield value of 1.34 t ha^{-1} and the median value of 1.27 t ha^{-1}. The lower and upper quartile grain yield values of the uninoculated plants were 1.05 and 1.70 t ha^{-1} respectively. Plants inoculated with BR 3262 had the least grain yield with the mean and median values of 1.25 and 1.07 t ha^{-1} respectively and interquartile range of 0.90 - 1.47 t ha^{-1} (Figure 4.33B).

Table 4.44: Grain yield of three cowpea varieties as affected by rhizobia inoculation and phosphorus application

	Cropping seasons		
	2015	2016	2017
Treatments			
Inoculation (I)			
Uninoculated	1.36a	1.35ab	1.32a
USDA 3451	1.43a		1.32a
USDA 3384	1.28a		
BR 3262		1.18b	1.32a
BR 3267		1.34ab	1.24a
90 kg N ha^{-1}		1.540a	1.34a
M S D	0.16	0.31	0.22
Phosphorus (P)			
0 kg ha^{-1}	0.81b	0.89b	0.94c
20 kg ha^{-1}	1.58a	1.53a	1.40b
40 kg ha^{-1}	1.67a	1.64a	1.58a
M S D	0.16	0.27	0.17
Variety (V)			
IT93K-452-1	1.26b	1.20a	1.23b
IT99K-573-1-1	1.61a	1.44a	1.46a
TVX-3236	1.19b	1.42a	1.23b
M S D	0.16	0.27	0.17
Interactions			
I × P	**	NS	NS
I × V	**	NS	NS
P × V	**	NS	NS
I × P × V	**	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, **- significant at P= 0.01, NS- not significant at P=0.05

Similarly in 2017, plants fertilized with 90 kg N ha^{-1} had the highest grain yield with the mean, median and interquartile range of 1.34, 1.40 and 0.85 - 1.71 t ha^{-1} respectively. Next in performance were the uninoculated plants with the mean, median and interquartile range of 1.32, 1.27 and 0.86 - 1.66 t ha^{-1} respectively. Plants inoculated with BR 3262 had the mean and median values of 1.32 and 1.44 t ha^{-1} respectively and interquartile range of 0.97 - 1.61 t ha^{-1}. Plants inoculated with USDA 3451 had similar mean grain yield value (1.32 t ha^{-1}) with plants inoculated with BR 3262. The median value was 1.41 t ha^{-1} and it had the interquartile range of 1.04 - 1.60 t ha^{-1}. Plants inoculated with BR 3267 had the least mean grain yield in 2017 with mean and median values of 1.24 and 1.31 t ha^{-1} respectively and interquartile range of 0.85 - 1.46 t ha^{-1} (Figure 4.33C).

Plants fertilized with 20 and 40 kg P ha^{-1} had statistically similar grain yield in 2015 and 2016 which were significantly higher than the grain yield of the unfertilized plants. In 2017, the order of performance in respect of grain yield was 40 kg P ha^{-1} > 20 kg P ha^{-1} > 0 kg P ha^{-1} (Table 4.44). Plants that received 40 kg P ha^{-1} had the mean grain yield of 1.67 t ha^{-1} and the median value of 1.74 t ha^{-1} in 2015. The interquartile range of the grain yield obtained in this plants was 1.26 - 2.07 t ha^{-1}. Next in performance were plants treated with 20 kg P ha^{-1} with the mean grain yield of 1.58 t ha^{-1} and the median value of 1.52 t ha^{-1}. The interquartile range of the plants that received 20 kg P ha^{-1} was 1.24 - 1.85 t ha^{-1}. The least grain yield was recorded in the unfertilized plants with the mean grain yield of 0.81 t ha^{-1} and the median value of 0.82 t ha^{-1}. The interquartile range of the unfertilized plants was 0.58 - 1.00 t ha^{-1} (Figure 4.34A).

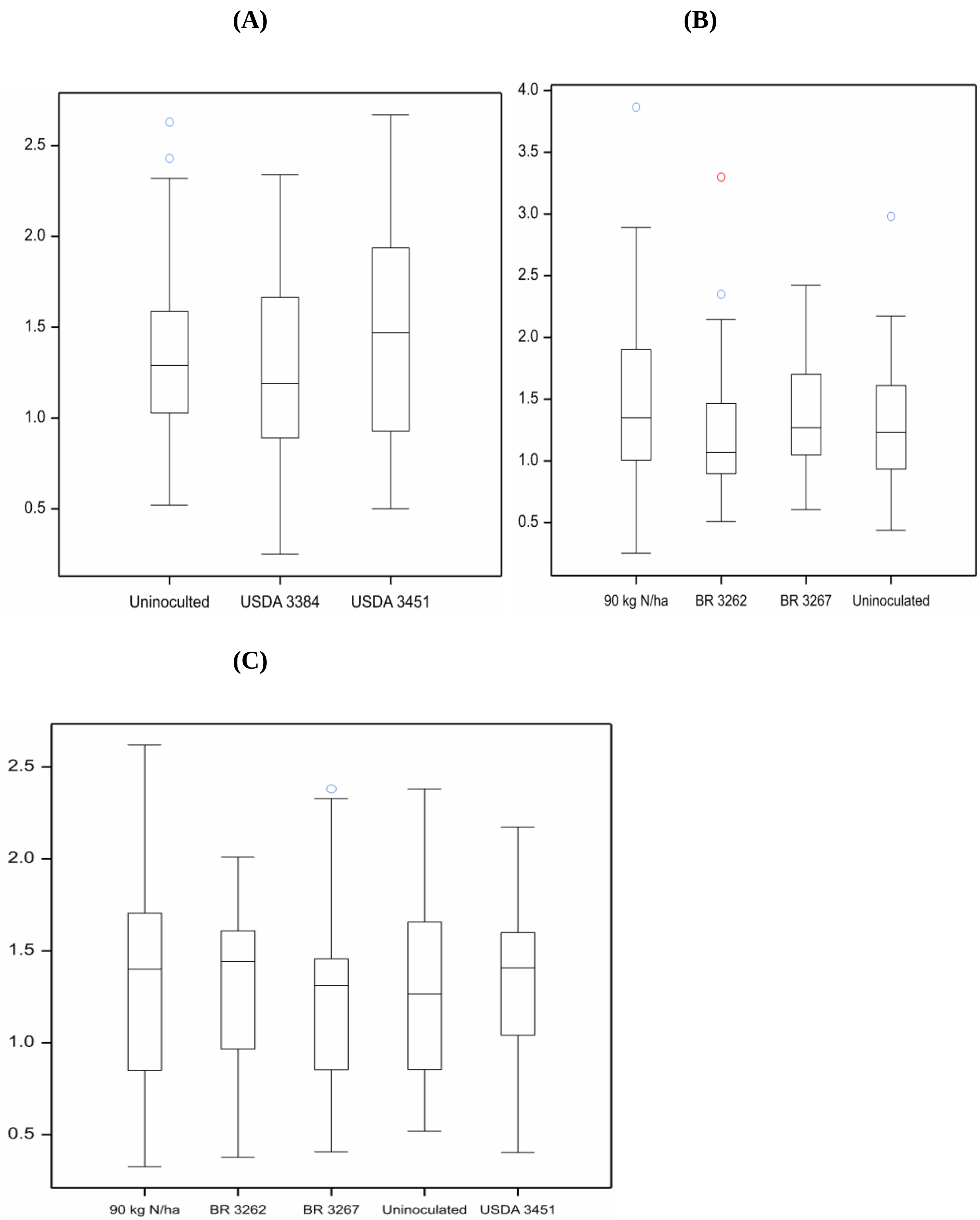


Figure 4.33: Box plots showing the grain yield of the different rhizobia inoculation treatments in (A) 2015 (B) 2016 (C) 2017.

Similar trend was observed in 2016. Plants that received 40 kg P ha^{-1} had the mean, median and interquartile range of 1.69, 1.55 and 1.30 - 2.05 t ha^{-1} respectively. Plants that received 20 kg P ha^{-1} had the mean, median and interquartile range of 1.53, 1.39 and 1.09 - 1.84 t ha^{-1} respectively. Plants that were not fertilized with P had the mean, median and interquartile range of 0.88, 0.90 and 0.71 - 1.07 t ha^{-1} respectively (Figure 4.34B).

In 2017 plants that received 40 kg P ha^{-1} equally had the highest mean, median and interquartile range of 1.58, 1.59 and 1.33 - 1.92 t ha^{-1} respectively. Next in performance were plants that received 20 kg P ha^{-1} which had the mean, median and interquartile range of 1.40, 1.41 and 0.86 - 1.06 t ha^{-1} respectively. Plants that did not receive P fertilizer had the least mean, median and interquartile range of 0.94, 0.86 and 0.68 -1.05 t ha^{-1} respectively (Figure 4.34C).

In 2015 and 2017, IT99K-573-1-1 produced significantly higher grain yield than IT93K-452-1 and TVX-3236 plants which produced statistically similar grain yield. In 2016 however, there was no significant difference between the grain yields of the three varieties (Table 4.44).

IT99K-573-1-1 plants had the mean grain yield value of 1.61 t ha^{-1} and the median value of 1.7 t ha^{-1} in 2015. The lower and upper quartile grain yield values were 1.10 and 2.13 t ha^{-1} respectively. This was followed by IT93K-452-1 with the mean and median values of 1.26 and 1.29 t ha^{-1} respectively. The lower and upper quartile grain yield values of IT93K-452-1 plants were 0.96 and 1.54 t ha^{-1} respectively. TVX-3236 plants had the least mean grain yield (1.19 t ha^{-1}) with the median value of 1.19 t ha^{-1}. The lower and upper quartile grain yield values were 0.82 and 1.44 t ha^{-1} respectively (Figure 4.35A).

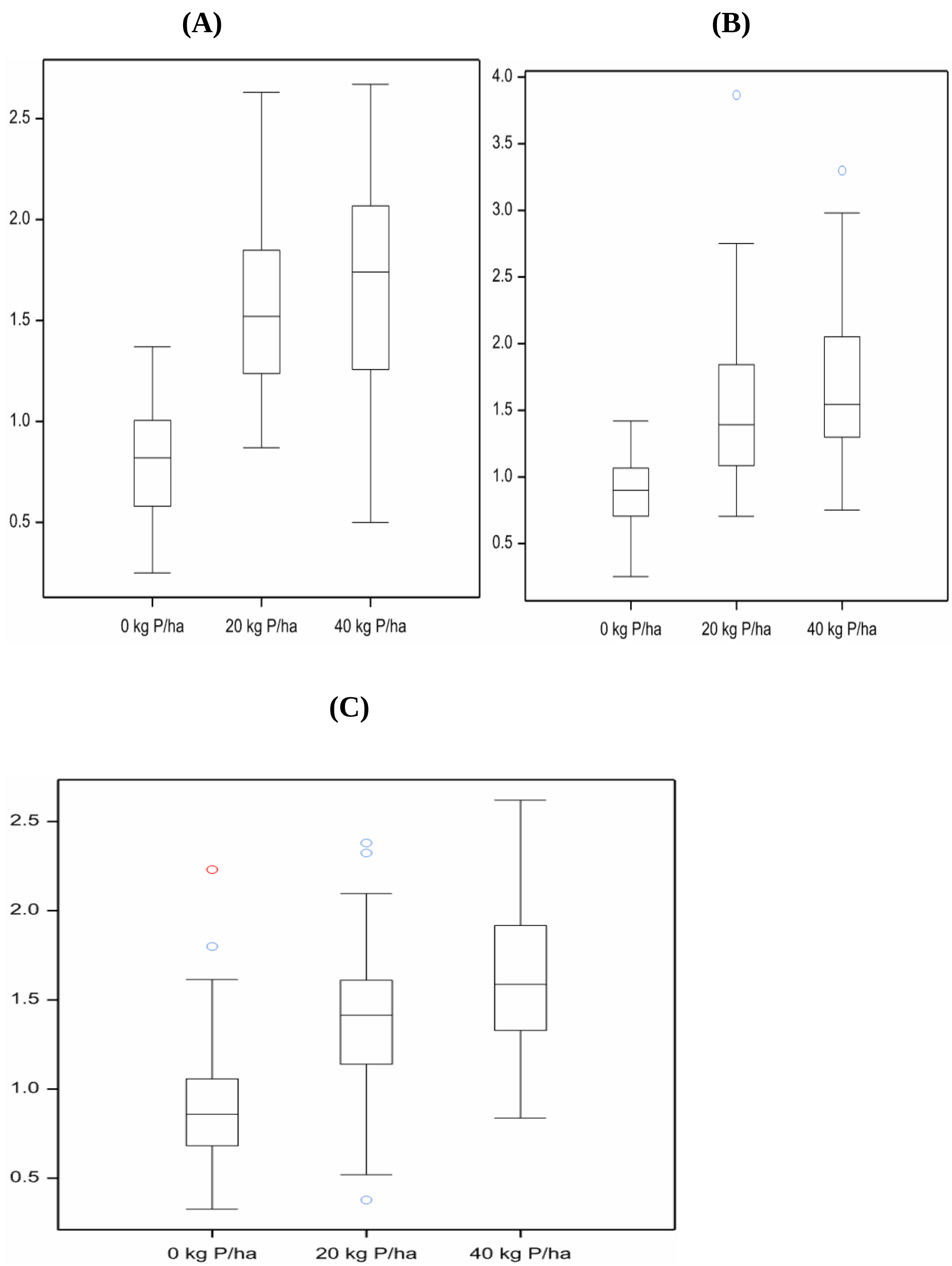
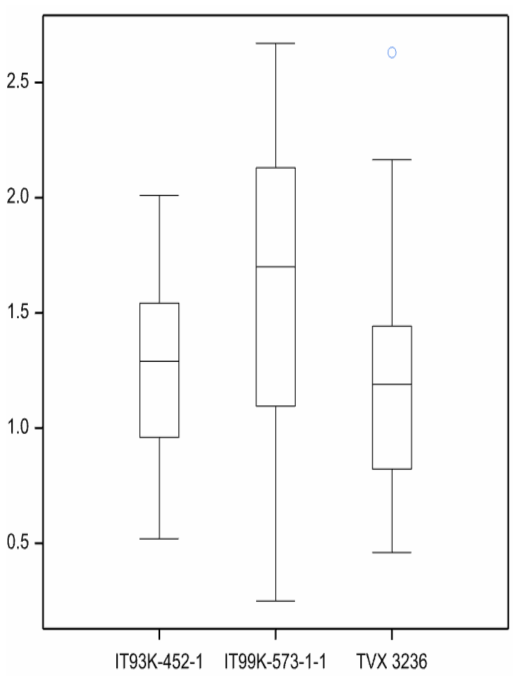


Figure 4.34: Box plots showing the grain yield of the different phosphorus treatments in (A) 2015, (B) 2016 (C) 2017.

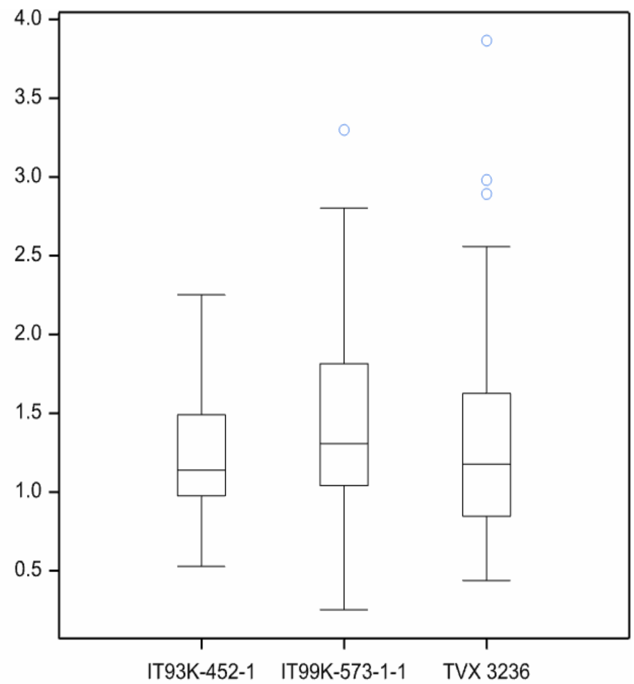
In 2016, IT99K-573-1-1 plants similarly had the highest grain yield with the mean and median values of 1.44 and 1.31 t ha^{-1} respectively. The lower and upper quartile grain yield values of IT99K-573-1-1 plants were 1.04 and 1.82 t ha^{-1} respectively. However, TVX-3236 was second in performance with the mean and median values of 1.42 and 1.18 t ha^{-1} respectively. The lower and upper quartile grain yield values of TVX-3236 plants were 0.85 and 1.63 t ha^{-1} respectively. The least grain yield was obtained in IT93K-452-1 variety with the mean and median values of 1.25 and 1.14 respectively. The lower and upper quartile grain yield values obtained in this variety were 0.98 and 1.49 t ha^{-1} respectively (Figure 4.35B).

Similar trend was observed in 2017 (Figure 4.35C) with IT99K-573-1-1 having the highest mean grain yield value of 1.46 t ha^{-1}. The median, lower and upper quartile grain yield values were 1.53, 0.98 and 1.96 t ha^{-1} respectively. TVX-3236 was next in performance with the mean grain yield value of 1.23 t ha^{-1}. The median grain yield value was 1.27 t ha^{-1} and the lower and upper quartile values were 0.85 and 1.60 t ha^{-1} respectively. Similarly, IT93K-452-1 plants had the mean grain yield value of 1.23 t ha^{-1}. The median, lower and upper quartile values of 1.24, 0.89 and 1.48 t ha^{-1} respectively were obtained.

(A)



(B)



(C)

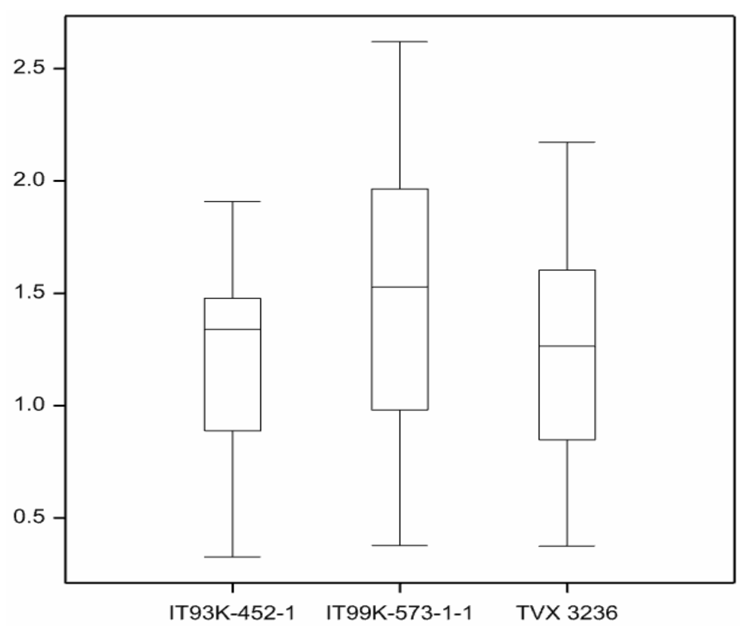


Figure 4.35: Box plots showing the grain yield of the different varieties in (A) 2015, (B) 2016 (C) 2017.

The interaction between rhizobia inoculation and phosphorus application on grain yield in 2015 is presented in Figure 4.36A. P application increased the grain yield of uninoculated plants and plants inoculated with USDA 3451 up to 20 kg P ha^{-1} application after which there was a decline at 40 kg P ha^{-1}. In plants inoculated with USDA 3384 however, grain yield increased significantly with increase in P application up to 40 kg P ha^{-1}.

The interaction between rhizobia inoculation and variety on grain yield is presented in Figure 4. 36 B. When IT93K-452-1 plants were inoculated with USDA 3451, significantly higher grain yield was recorded than when uninoculated. However, uninoculated IT99K-573-1-1 plants produced significantly higher grain yield than when the same variety was inoculated with USDA 3384 strain. In TVX-3236 variety, there was no significant difference between the grain yield of the inoculated and uninoculated plants.

The interaction between phosphorus and variety on grain yield in 2015 is presented in Figure 4.36 C. IT93K-452-1 plants and TVX-3236 plants produced the highest grain yield when fertilized with 20 kg P ha^{-1}. IT99K-573-1-1 plants however had its highest grain yield when fertilized with 40 kg P ha^{-1}.

The interaction between rhizobia inoculation, phosphorus application and variety on grain yield in 2015 was significant. All the uninoculated plants of the three varieties produced their highest grain yield when fertilized with 20 kg P ha^{-1}. IT93K-452-1 and TVX-3236 plants inoculated with USDA 3451 had their highest grain yield when fertilized with 20 kg P ha^{-1}. IT99K-573-1-1 plants inoculated with the same strain produced the highest grain yield at 40 kg P ha^{-1}. When the three

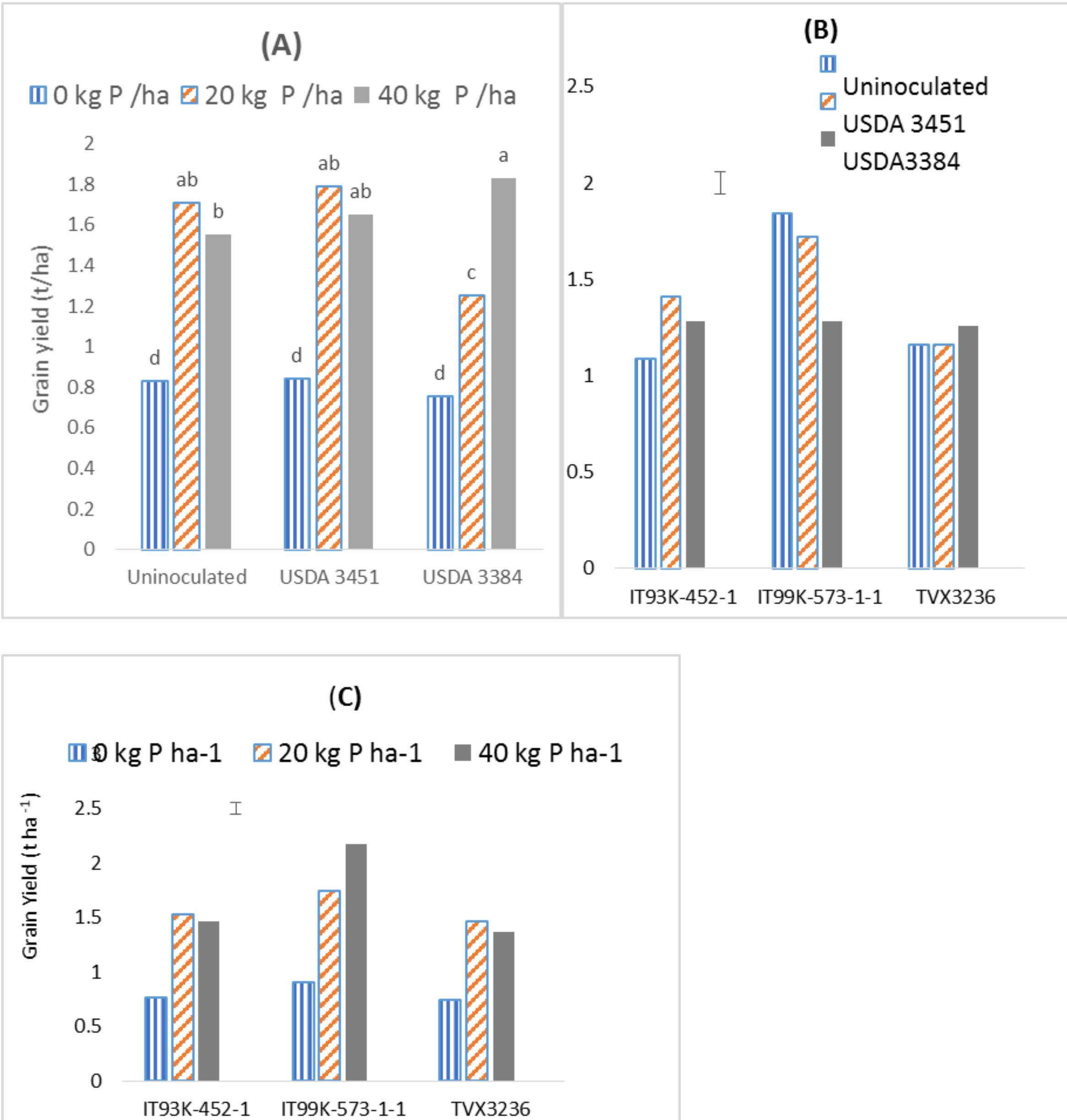


Figure 4.36: Interaction effects of (A) rhizobia inoculation and phosphorus (B) rhizobia inoculation and variety (C) phosphorus and variety on grain yield of cowpea in 2015

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

I- represent LSD at P=0.05.

varieties were inoculated with USDA 3384, the significantly highest grain yield was obtained at 40 kg P ha^{-1}. However, in IT9K-452-1 plants, the grain yield obtained at 20 and 40 kg P ha^{-1} was similar. The unfertilized plants generally had the least grain yield among all the treatment combination.

Among the unfertilized plants, uninoculated and USDA 3451 inoculated IT99K-573-1-1 plants had grain yield above 1 t ha^{-1} (1.09 and 1.17 t ha^{-1} respectively). The least (0.46 t ha^{-1}) was recorded in unfertilized IT99K-573-1-1 plants inoculated with USDA 3384. Among all the treatment combinations, 20 kg P ha^{-1} fertilized uninoculated IT99K-573-1-1 plants and 40 kg P ha^{-1} fertilized IT99K-573-1-1 plants inoculated with USDA 3451 strain had the highest grain yield (2.32 t ha^{-1}) (Figure 4.37).

ix. Grain Harvest Index

The grain harvest index of cowpea varieties as influenced by rhizobia inoculation and phosphorus application in the three years is presented in Table 4.45. In the three years, there was no significant difference between the grain harvest index of the plants that received the various inoculation treatments.

In 2015, the unfertilized plants had significantly higher grain harvest index than the fertilized plants. In 2016 however, the grain harvest index of plants that received 20 and 40 kg P ha^{-1} were similar and significantly higher than the value recorded in the unfertilized plants. In 2017, plants fertilized with 40 kg P ha^{-1} had the highest grain harvest index which was significantly higher than the value obtained in the unfertilized plants but similar to the value recorded in 20 kg P ha^{-1} fertilized plants.

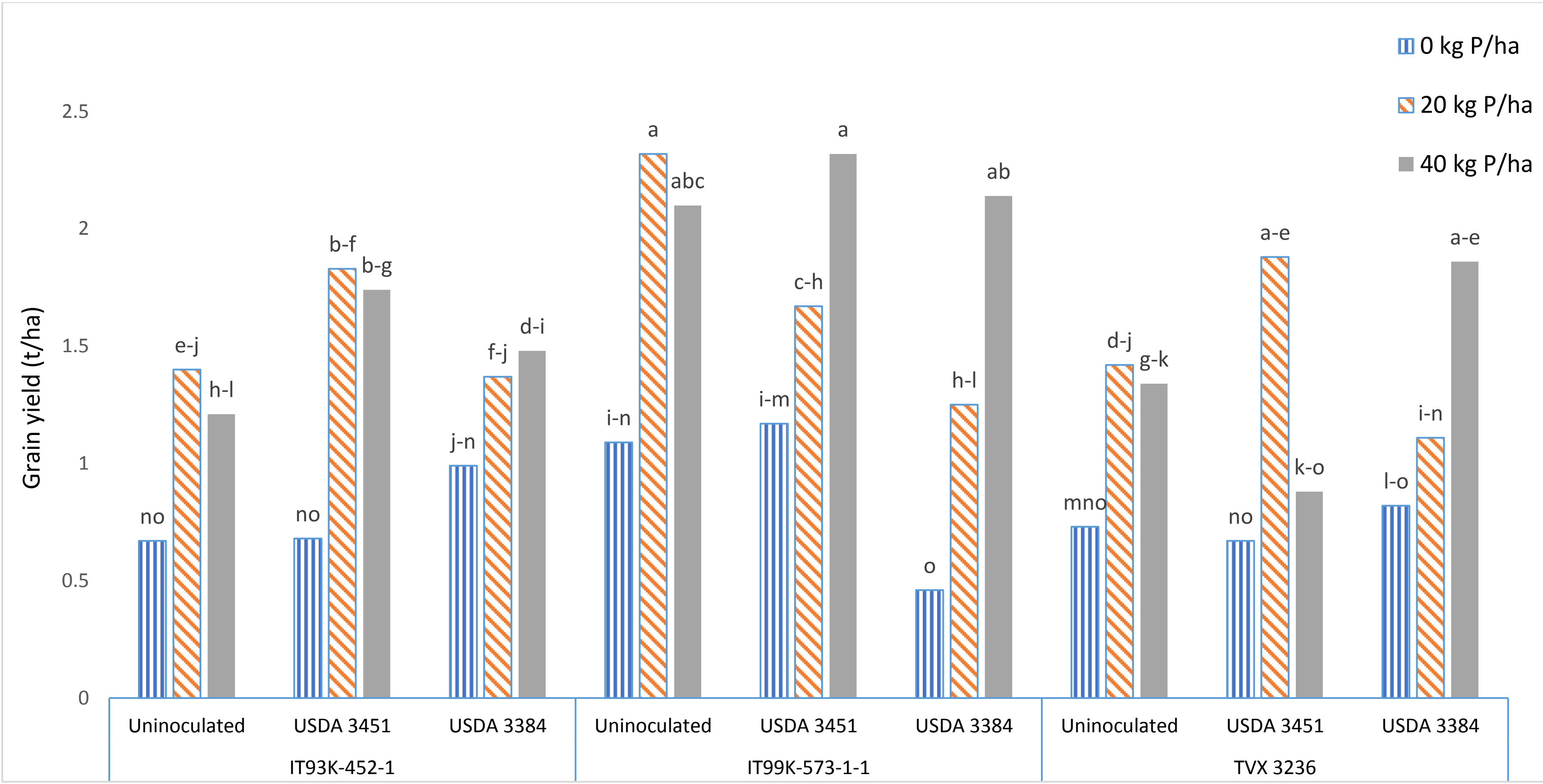


Figure 4.37: Interaction effects of rhizobia inoculation, phosphorus application and varieties on grain yield of cowpea in 2015

Bars carrying similar alphabets are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test

Table 4.45: Grain harvest index of three cowpea varieties as affected by rhizobia inoculation and phosphorus application

	Cropping seasons		
	2015	2016	2017
Treatments			
Inoculation (I)			
Uninoculated	0.32a	0.29a	0.36a
USDA 3451	0.34a		0.39a
USDA 3384	0.31a		
BR 3262		0.28a	0.38a
BR 3267		0.30a	0.39a
90 kg N ha^{-1}		0.30a	0.37a
M S D	0.06		
Phosphorus (P)			
0 kg ha^{-1}	0.36a	0.26b	0.36b
20 kg ha^{-1}	0.30b	0.31a	0.38ab
40 kg ha^{-1}	0.31ab	0.31a	0.40a
M S D	0.06	0.04	0.03
Variety (V)			
IT93K-452-1	0.29b	0.26b	0.36a
IT99K-573-1-1	0.36a	0.30ab	0.38a
TVX-3236	0.31ab	0.31a	0.39a
M S D	0.06	0.04	0.03
Interactions			
I × P	NS	NS	NS
I × V	NS	NS	NS
P × V	NS	NS	NS
I × P × V	NS	NS	NS

Means followed by different alphabets within a column and factor are significantly different at P=0.05 Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, NS- not significant at P=0.05

IT99K-573-1-1 plants had the highest grain harvest index in 2015 but the value was similar to what was recorded in TVX-3236 plants and significantly higher than the value recorded in IT93K-452-1 variety. In 2016, TVX-3236 plants had the highest grain harvest index which was similar to the value recorded in IT99K-573-1-1 plants but significantly higher than the value recorded in IT93K-452-1 plants. In 2017, there was no significant difference between the grain harvest index of the three varieties.

x. Agronomic Efficiency

The agronomic efficiency of cowpea varieties as influenced by rhizobia inoculation in the three years is presented in Table 4.46. In 2015, there was no significant difference between the agronomic efficiency of the inoculated and uninoculated plants both at 20 and 40 kg P ha^{-1} application. In 2016 however, plants fertilized with 90 kg N ha^{-1} had significantly higher agronomic efficiency than the inoculated and uninoculated plants whose agronomic efficiency were similar at 20 kg p ha^{-1} application. When 40 kg P ha^{-1} was applied however, there was no significant difference between the agronomic efficiency of the inoculated, uninoculated and 90 kg N ha^{-1} fertilized plants. In 2017 also, plants fertilized with 90 kg N ha^{-1} equally had the highest agronomic efficiency which was significantly higher than the value recorded in plants inoculated with BR 3262 but similar to the efficiency of the uninoculated and other inoculated plants at 20 kg P ha^{-1} application. At 40 kg P ha^{-1} application, the highest agronomic efficiency was equally recorded in plants fertilized with 90 kg N ha^{-1} and the least in the uninoculated plants but there was no significant difference between the values recorded in the inoculated and the uninoculated plants and likewise between the inoculated and 90 kg N ha^{-1} fertilized plants. Generally, the agronomic efficiency recorded at 40 kg P ha^{-1} was lower than the values recorded at 20 kg P ha^{-1}.

Table 4.46: Agronomic efficiency of three cowpea varieties as affected by rhizobia inoculation

	Cropping seasons					
	2015		2016		2017`	
Treatments						
Inoculation (I)	**20 kg P ha^{-1}**	**40 kg P ha^{-1}**	**20 kg P ha^{-1}**	**40 kg P ha^{-1}**	**20 kg P ha^{-1}**	**40 kg P ha^{-1}**
Uninoculated	3.78a	2.14a	5.25b	4.09a	3.99ab	1.72b
USDA 3451	5.53a	2.11a			3.33ab	2.2ab
USDA 3384	2.75a	3.49a				
BR 3262			5.35b	4.02a	2.32b	2.20ab
BR 3267			3.75b	3.84a	4.56ab	3.74ab
90 kg N ha^{-1}			11.47a	4.02a	6.37a	4.82a
MSD	3.50	2.10	4.83	3.11	4.03	2.78
Variety (V)						
IT93K-452-1	4.37a	2.58a	5.20a	3.31a	5.37a	2.30a
IT99K-573-1-1	3.50a	3.82a	6.71a	4.26a	2.93a	3.01a
TVX-3236	3.49a	1.34a	7.46a	4.41a	4.04a	3.62a
MSD	3.50	2.10	4.19	2.68	3.43	2.15
Interactions						
I × V	NS		NS		NS	

Means followed by different alphabets within a column and factor are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference , NS- not significant at P=0.05

There was no significant difference between the agronomic efficiency recorded among the three varieties in the three years

4.1.2.6 Relationships between growth, physiological and yield attributes of cowpea varieties as influenced by rhizobia inoculation and phosphorus application

i. Correlation between growth and yield attributes as influenced by rhizobia inoculation and phosphorus application

The correlation matrix between growth and yield attributes as influenced by rhizobia inoculation and phosphorus application in the three years is presented on Table 4.47.

Among the growth attributes, the correlation between stem diameter, leaf area, number of leaves, vine length, shoot biomass yield and grain yield were positive and highly significant with the shoot biomass yield having the strongest relationship (correlation coefficient (r) =0.61**) with grain yield. Among the yield attributes, 100 seed weight, pod length and number of pods per plant had highly significant and positive correlation with grain yield. Shell percentage had highly significant but negative correlation with grain yield. Number of pods per plant had the strongest relationship (r=0.79**) with grain yield. The correlation between number of days to 1st flower at sight, days to 50% flowering and grain yield were significant but they were negatively correlated.

The correlation between stem diameter, leaf area, number of leaves, vine length and shoot biomass yield were positive and highly significant having r values of 0.64, 0.56, 0.57 and 0.71 respectively. Days to 1st flower at sight and 50% flowering had highly significant but negative correlation with shoot biomass yield. Shoot biomass yield correlated significantly and positively with all the grain yield attributes except number of seeds per pod with number of pods per plant having the strongest relationship (r=0.51) (Table 4.47).

Table 4.47: Correlation matrix between growth and yield attributes of cowpea as influenced by rhizobia inoculation and phosphorus application

	1	2	3	4	5	6	7	8	9	10	11	12	13
1													
2	0.73**												
3	0.55**	0.42**											
4	0.76**	0.81**	0.51**										
5	-0.42**	-0.50**	0.04	-0.47**									
6	-0.46**	-0.49**	-0.00	-0.45**	0.88**								
7	-0.19*	0.23**	-0.24**	-0.14	0.16	0.09							
8	0.51**	0.66**	-0.08	0.53**	-0.63**	-0.65**	-0.11						
9	0.38**	0.52**	0.05	0.38**	-0.38**	-0.03**	-0.03	0.59**					
10	0.34**	0.22**	0.58**	0.29**	-0.02	-0.04	-0.29**	-0.03	-0.01				
11	-0.04	-0.14	0.15	-0.05	0.15	0.15	0.03	-0.34**	0.18*	0.01			
12	0.64**	0.56**	0.57**	0.71**	-0.27**	-0.30**	-0.16*	0.36**	0.19*	0.51**	-0.09		
13	0.50**	0.44**	0.53**	0.45**	-0.21*	-0.20*	-0.41**	0.27**	0.22**	0.79**	0.04	0.61**	

*= Significant at 5%, **=Highly significant at 1%, 1=Stem diameter, 2= Leaf area, 3= Number of leaves, 4= vine length, 5=Days to 1st flower at sight, 6=Days to 50% flowering 7=Shell percentage 8= 100-seed weight, 9= Pod length, 10= Pod per plant, 11=Seed per pod, 12= Shoot biomass yield, 13 =Grain yield

ii. Correlation between N-fixation and Yield Attributes of Cowpea as Influenced by Rhizobia Inoculation and Phosphorus Application

Table 4.48 shows the correlation matrix between N-fixation and yield attributes of cowpea as influenced by rhizobia inoculation and phosphorus application .The correlation between N uptake, nodule dry weight, number of nodules, NDFA, N-fixed and grain yield were positive and highly significant having the r values of 0.49, 0.45, 0.40, 0.41 and 0.42 respectively. N uptake, nodule dry weight, number of nodules, N derived from atmosphere and N- fixed correlated positively and significantly ($P<0.01$) with shoot biomass yield with N-uptake having the highest r value ($r=0.79$). Number of nodules and dry weight correlated positively and significantly with the quantity of N-fixed.

iii. Correlation between physiological and yield attributes of cowpea as influenced by rhizobia inoculation and phosphorus application

Table 4.49 shows the correlation between physiological attributes and yields of cowpea as influenced by rhizobia inoculation and phosphorus application. Leaf area index and crop growth rate correlated significantly ($P<0.01$) and positively with grain yield having r value of 0.58 and 0.78 respectively. All the physiological attributes correlated positively and significantly with shoot biomass yield with leaf area index having the highest r value (0.70) followed by crop growth rate (0.56).

iv. Correlation between environmental factors and photosynthetic activities of cowpea as influenced by rhizobia inoculation and phosphorus application

Among the environmental factors, photosynthetic active radiation (PAR) had highly significant ($P<0.01$) but negative relationship with Phi 2 (Table 4.50). The relative humidity, chlorophyll and

Table 4.48: Correlation matrix between N-fixation attributes and biomass and grain yield of cowpea as influenced by rhizobia inoculation and phosphorus application

	1	2	3	4	5	6	7	8	9
1									
2	0.40**								
3	0.33**	0.79**							
4	0.01	-0.01	0.04						
5	0.05	0.11	0.05	-0.15					
6	0.83**	0.18	0.13	0.04	0.03				
7	0.97**	0.28**	0.21*	-0.02	0.07	0.87**			
8	0.83**	0.49**	0.37**	-0.01	0.14	0.70**	0.79**		
9	0.49**	0.45**	0.40**	0.12	0.03	0.41**	0.42**	0.61**	

*= Significant at 5%, **=Highly significant at 1%, 1=N uptake, 2= Nodule dry weight, 3= Number of nodules, 4= % effectiveness, 5=Seed protein, 6= N derived from atmosphere, 7= N fixed, 8= Shoot biomass yield, 9 =Grain yield

Table 4.49: Correlation matrix between physiological attributes and biomass and grain yield of cowpea as influenced by rhizobia inoculation and phosphorus application

	1	2	3	4	5	6	7	8	9
1									
2	0.81*								
3	0.91**	0.89**							
4	0.15	0.16	0.10						
5	0.11	0.12	0.13	0.56**					
6	-0.31**	-0.21*	-0.33**	-0.28**	-0.19*				
7	0.26**	0.21*	0.27**	0.45**	0.25**	-0.76**			
8	0.25**	0.23*	0.28**	0.70**	0.56**	0.46**	0.17*		
9	0.11	0.11	0.11	0.58**	0.78**	0.11	0.07	0.61**	

*= Significant at 5%, **=Highly significant at 1%, 1=chlorophyll a, 2= chlorophyll b, 3= total chlorophyll, 4= leaf area index, 5=crop growth rate, 6= Net assimilation rate, 7= leaf area ratio, 8= Shoot biomass yield, 9 =Grain yield

Table 4.50: Correlation matrix between environmental factors and photosynthetic attributes of cowpea as influenced by rhizobia inoculation and phosphorus application.

	1	2	3	4	5	6	7	8	9
1									
2	-0.86**								
3	-0.37**	-0.05							
4	0.36**	0.07	0.04						
5	0.64**	-0.02	0.17*	0.17*					
6	-0.07	0.04	0.07	0.12	-0.12				
7	0.20*	-0.21*	-0.57**	-0.1	-0.18*	0.39**			
8	-0.28**	-0.17*	0.79**	0.11	0.13	0.10	-0.77**		
9	0.31**	0.13	-0.79**	-0.18*	-0.04	0.20*	0.51**	-0.93**	

*= Significant at 5%, **=Highly significant at 1%, ns= Not significant, 1=Relative humidity, 2= Ambient temperature, 3= Photosynthetic active radiation, 4= Leaf angle, 5=Leaf temperature difference, 6= Chlorophyll, 7= Phi no, 8= Phi npq, 9 =Phi 2

Phi no correlated positively and significantly with Phi 2. The strongest relationship was however observed between photosynthetic active radiation, phi npq and Phi 2 with the coefficient value of -0.79 and -0.93 respectively.

The relative humidity, ambient temperature and Phi no had significant but negative relationship with Phi npq among which the relationship with Phi no was the strongest (r=0.77). The PAR on the other had highly significant and positive relationship with Phi npq (r=0.79). The relative humidity and chlorophyll correlated positively and significantly with Phi no. Negative but significant relationship existed between Phi no and the ambient temperature, PAR, leaf temperature among which the relationship with PAR was the strongest (-0.57**).

v. Regression between physiological traits and grain yield of cowpea varieties as influenced by rhizobia inoculation and phosphorus application

Table 4.51 shows the linear regression between some physiological traits and grain yield. The result showed that Phi 2, CGR and LAI positively influenced grain yield and explained 67.29% of the variation in grain yield (R^2= 67.29%). Phi 2, CGR and LAI could significantly predict grain yield with P values of 0.036, 0.000 and 0.009 respectively. From the results, the predicting regression model was -281+ 941Phi 2 + 132.4CGR + 240.1 LAI indicating that each unit increase in Phi 2 is associated with 941.2 unit increase in grain yield holding CGR and LAI constant, and each unit increase in CGR is associated with 132.4 units increase in grain yield holding Phi 2 and LAI constant and each unit increase in LAI is associated with 240.1 unit increase in grain yield holding Phi 2 and CGR constant. The model was highly significant (P=0.000) and among the three variables CGR is the most important predictor having a t-value of 10.14 compared to the value 2.12 and 2.67 obtained in Phi 2 and LAI respectively.

Table 4.51: The regression coefficients of physiological variables in predicting grain yield of cowpea under rhizobia inoculation, phosphorus application and varietal differences

Variable	Coefficient	SE	T-value	P-Value
Constant	-281	211	-1.33	0.18
Phi 2	941	443	2.12	0.036
CGR	132.4	13.1	10.14	0.000
LAI	240.1	89.8	2.67	0.009

Regression P-value - 0.000
R^2 - 67.29%
Adjusted R^2 - 66.49%

4.1.3 Experiment 3: Physiological responses of cowpea varieties to cowpea sequential cropping system

4.1.3.1 Growth parameters of cowpea varieties in cowpea sequential cropping system

Among the varieties sown in the first sequence in 2016, Kanannado variety had significantly higher number of leaves than IT93K-452-1 but similar to the values recorded in the other varieties. In 2017, Kanannado equally had the significantly highest number of leaves followed by TVX-3236 which had similar number of leaves with IT99K-573-1-1 and Oloyin varieties. The least was recorded in IT93K-452-1 similar to IT90K-76 varieties (Table 4.52). In the second sequence, there was no significant difference between the number of leaves of the five varieties.

The longest and shortest vines in the first sequence were recorded in Kanannado and TVX-3236 varieties in both years but the value obtained in TXV-3236 was statistically similar to the vine length of the remaining varieties in 2016 but significantly lower than the value obtained in IT93K-452-1 in 2017. In the second sequence, Oloyin had significantly longer vines than the remaining varieties which had similar vines in 2016. In 2017, there was no significant difference between the vine lengths of the five varieties sown in the second sequence (Table 4.52)

IT93K-452-1, Kanannnado and Oloyin plants had similar leaf area that were significantly larger than the leaves of IT99K-573-1-1, TVX-3236 and IT90K-76 leaves which were similar among the varieties sown in the first sequence in 2016. In 2017, Kanannnado and Oloyin varieties had similar leaf sizes that were significantly larger than the leaves of the remaining varieties. In the second sequence, there was no significant difference in the leaf area of the five varieties in 2016. In 2017, Oloyin had the largest leaves which were similar to the leaf area of IT90K-76 and IT93K-452-1 varieties (Table 4.52).

Table 4.52: Growth parameters of cowpea varieties in cowpea sequential cropping system at 50% flowering

	2016				2017			
	NOL	VL	LA	SG	NOL	VL	LA	SG
First planting								
IT93K-452-1	42.33b	194.67b	253.12a	1.09b	36.44d	155.57b	190.79b	1.01b
IT99K-573-1-1	73.00ab	192.33b	225.75b	1.14b	51.33bc	151.88bc	186.31b	0.96b
TVX-3236	71.00ab	147.67b	207.66b	1.41b	56.22b	92.88c	171.36b	0.91b
Kanannado	106.67a	276.23a	259.19a	2.10a	79.78a	216.62a	243.15a	1.44a
Oloyin	80.67ab	155.33b	259.06a	1.35b	46.67bc	152.32bc	244.44a	1.01b
IT90K-76	79.33ab	158.00b	218.18b	1.33b	42.11cd	111.68bc	177.89b	0.97b
MSD (0.05)	4.48	56.41	19.09	0.37	8.35	60.20	26.65	0.40
Second planting								
IT93K-452-1	40.60a	95.08b	165.13a	1.29a	32.56a	93.19a	196.17ab	0.95a
IT99K-573-1-1	52.81a	123.70b	201.83a	1.46a	34.67a	94.90a	177.03b	0.94a
TVX-3236	44.56a	104.38b	170.30a	1.23a	32.11a	69.27a	176.71b	0.86a
Oloyin	47.33a	195.43a	161.40a	1.34a	29.33a	77.71a	233.91a	1.03a
IT90K-76	52.67a	93.84b	170.96a	1.54a	31.78a	83.18a	194.59ab	0.99a
MSD (0.05)	22.20	45.11	93.06	0.76	17.17	35.48	42.83	0.18

Means followed by different alphabets within a column and sequence of planting are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, NOL- number of leaves, VL-vine length, LA—leaf area, SD- stem diameter.

Kanannado had significantly thicker stem than the remaining varieties which had similar stem dimeter at 50% flowering in both years. In the second sequence, there was no significant difference in the stem diameter of the five varieties (Table 4.52)

4.1.3.2 Physiological attributes of cowpea varieties in cowpea sequential cropping system

i. Crop growth rate

Kanannado had the significantly highest growth rate similar to IT99K-573-1-1 at the early growth stage in the first sequence of 2016. At mid growth stage, IT99K-573-1-1, IT90K-76 and Oloyin had similar growth rate that were significantly higher than the remaining varieties which had similar growth rate. At late growth stage, there was no significant difference between the growth rate of the six varieties. In 2017, IT93K-452-1, IT99K-573-1-1 and IT90K-76 had similar growth rate that were significantly higher than the value recorded in the remaining varieties which had similar growth rate at the early growth stage. At mid growth stage, IT90K-76, IT93K-452-1 and TVX-3236 had similar growth rate that were significantly higher than the values recorded in Kanannado and Oloyin varieties. At late growth stage, TVX-3236 had the highest growth rate similar to IT90K-76. Oloyin had the least similar to the value recorded in IT93K-452-1, IT99K-573-1-1 varieties (Table 4.53).

In the second sequence, IT93K-452-1 had the highest growth rate similar to IT99K-573-1-1 and IT90K-76 varieties but significantly higher than the values recorded in Oloyin and TVX-3236 which had the least growth rate in 2016. At mid growth stage, the growth rate of the varieties were similar except that IT90K-76 and IT99K-573-1-1 had significantly higher growth rate than Oloyin variety which had the least growth rate. At late growth stage, TVX-3236 had the highest growth rate followed by IT99K-573-1-1 which were significantly higher than the remaining varieties which had similar lowest growth rate. In the early growth stage in 2017, all the varieties except TVX-3236 had significantly higher growth rate than Oloyin variety Table 4.53).

Table 4.53: Crop growth rate of cowpea varieties in cowpea sequential cropping system

		2016			2017	
	Stage 1	Stage 2	Stage 3	Stage 1	Stage 2	Stage 3
First planting						
IT93K-452-1	11.40c	14.83b	14.16a	10.34a	13.93ab	11.89cd
IT99K-573-1-1	18.82a	30.00a	14.84a	8.23b	12.19bc	12.14bcd
TVX-3236	14.25bc	20.14b	13.19a	7.34b	13.84a	14.13a
Kanannado	19.08a	16.90b	17.81a	11.10a	10.85c	12.39bc
Oloyin	13.52bc	35.05a	14.78a	8.17b	10.62c	11.10d
IT90K-76	14.48b	35.23a	14.15a	10.89a	14.23a	13.27ab
MSD (0.05)	3.00	6.91	9.53	1.10	1.91	1.22
Second planting						
IT93K-452-1	4.92a	6.22ab	5.38c	4.37a	5.93a	5.42b
IT99K-573-1-1	4.61ab	7.13a	6.63b	4.42a	6.37a	6.65a
TVX-3236	3.22c	6.76ab	7.89a	3.68ab	5.85a	6.36a
Oloyin	3.79bc	5.94b	6.12bc	3.44b	4.33b	5.22b
IT90K-76	4.85ab	6.97a	5.15c	4.55a	5.70a	5.31b
MSD (0.05)	1.06	0.92	1.13	0.90	1.06	0.75

Means followed by different alphabets within a column and sequence of planting are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, stages 1, 2 and 3- early, mid and late growth stages respectively.

At mid growth stage, all the varieties had similar and significantly higher growth stage than Oloyin variety. At late growth stage, TVX-3236 and IT99K-573-1-1 had similar growth rate that were significantly higher than the remaining varieties which had similar lower growth rate.

ii. Leaf area index

Table 4.54 shows the LAI of the cowpea varieties sown in two sequence of planting. The LAI of the varieties were statistically similar in the first sequence in 2016 except that the LAI of Kanannado were significantly higher than the LAI of Oloyin and IT90K-76 at 3 WAS, TVX-3236 at 5 WAS and IT93K-452-1 at 7 WAS. In 2017, there was no significant difference between the LAI of the six varieties at 3 WAS. At 5 WAS, Kanannado variety had the significantly highest LAI (7.91) followed by IT99K-573-1-1 (5.93) which had similar LAI with Oloyin variety (5.88). IT93K-452-1 had the least LAI (3.27) which was statistically similar to the value recorded in IT90K-76 variety (4.73). At 7 WAS, Kanannado variety maintained the highest the LAI (9.11) but it was similar to the value recorded in Oloyin variety (7.61). IT93K-452-1 maintained the least LAI (4.65) which was statistically similar to the value recorded in IT90K-76 variety (4.54).

In the second sequence, IT90K-76 had significantly higher LAI than the remaining varieties which had similar LAI at 3 and 5 WAS in 2016. There was no significant difference between the LAI of the varieties at 7 WAS in 2016 as well as at 3 and 7 WAS in 2017. At 5 WAS in 2017, the LAI of the varieties were similar except that Oloyin variety had significantly higher LAI than TVX-3236 which had the highest and lowest LAI respectively.

Table 4.54: Leaf area index of cowpea varieties in cowpea sequential cropping system

	2016			2017		
	Sampling period					
	3 WAS	5 WAS	7 WAS	3 WAS	5 WAS	7 WAS
First planting						
IT93K-452-1	1.92ab	4.42b	7.10b	0.80a	3.27e	4.65d
IT99K-573-1-1	1.88ab	5.94ab	11.10ab	1.32a	5.93b	6.38bc
TVX-3236	1.61ab	4.80b	9.8ab	0.94a	4.88cd	6.43bc
Kanannado	2.48a	7.92a	18.03a	1.20a	7.91a	9.11a
Oloyin	1.27b	6.15ab	13.85ab	1.34a	5.88bc	7.61ab
IT90K-76	1.23b	5.41ab	11.52ab	1.00a	4.73d	4.99cd
MSD (0.05)	0.88	3.07	7.27	0.99	1.02	1.57
Second planting						
IT93K-452-1	0.23b	1.92b	4.50a	0.47a	1.43ab	4.33a
IT99K-573-1-1	0.29b	2.54b	7.54a	0.44a	1.35ab	4.08a
TVX-3236	0.21b	1.57b	3.37a	0.29a	1.08b	3.78a
Oloyin	0.32b	2.69b	5.13a	0.69a	1.90a	4.51a
IT90K-76	0.91a	4.87a	5.88a	0.58a	1.68ab	4.04a
MSD (0.05)	0.41	1.81	5.73	0.52	0.71	3.21

Means followed by different alphabets within a column and sequence of planting are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, WAS- weeks after sowing

iii. Net assimilation rate

IT90K-76 plants had significantly higher net assimilation rate than IT93K-452-1, TVX-3236 and Kanannado which had the least NAR at reproductive stage among the plants sown in the first sequence of 2016. At vegetative stage, the NAR observed among the varieties were similar. In 2017, IT93K-452-1 had the significantly highest NAR followed by IT90K-76 which had similar NAR with the remaining varieties except Kanannado which maintained the least NAR (Table 4.55).

In the second sequence, there was no significant difference among the NAR of the varieties except at reproductive stage in both years in which TVX-3236 had significantly higher NAR than IT90K-76 in 2016 and TVX-3236 and IT99K-573-1-1 had similar NAR that were significantly higher than the NAR of Oloyin in 2017 (Table 4.55).

iv Photosynthetic attributes

The quantum yield of photosystem II, ratio of light that goes towards non-photochemical quenching, ratio of light that is lost through non-regulated processes and leaf temperature of the varieties were not significantly different in both the first and second planting (Table 4.56).

4.1.3.3 Yield attributes of cowpea varieties in cowpea sequential cropping

i. Shoot biomass yield

Kanannado variety had the significantly highest biomass yield (9.16 t ha^{-1}) among the varieties sown in the first planting in 2016. This was followed by IT90K-76, Oloyin and IT99K-573-1-1 which had statistically similar biomass yield (7.23, 6.92 and 6.76 t ha^{-1} respectively). The least (4.17 t ha^{-1}) was recorded in TVX-3236 variety. In 2017, Kanannado maintained the highest

Table 4.55: Net assimilation rate of cowpea varieties in cowpea sequential cropping system

	2016		2017	
Treatments	Vegetative stage	Reproductive stage	Vegetative stage	Reproductive stage
First planting				
IT93K-452-1	3.1×10^{-4}a	1.7×10^{-4}bc	6.6×10^{-4}a	2.3×10^{-4}a
IT99K-573-1-1	5.4×10^{-4}a	2.6×10^{-4}ab	3.2×10^{-4}a	1.0×10^{-4}bc
TVX-3236	4.4×10^{-4}a	2.1×10^{-4}bc	5.5×10^{-4}a	1.4×10^{-4}b
Kanannado	3.9×10^{-4}a	1.2×10^{-4}c	5.7×10^{-4}a	6.9×10^{-5}c
Oloyin	5.8×10^{-4}a	2.6×10^{-4}ab	3.4×10^{-4}a	9.1×10^{-5}bc
IT90K-76	6.6×10^{-4}a	3.4×10^{-4}a	6.2×10^{-4}a	1.5×10^{-4}b
MSD (0.05)	4.0×10^{-4}	1.0×10^{-4}	6.0×10^{-4}	6.0×10^{-7}
Second planting				
IT93K-452-1	1.5×10^{-3}a	1.8×10^{-4}ab	5.1×10^{-4}a	2.2×10^{-4}ab
IT99K-573-1-1	8.8×10^{-4}a	1.5×10^{-4} ab	5.5×10^{-4}a	2.5×10^{-4}a
TVX-3236	2.4×10^{-3}a	3.0×10^{-4} a	7.2×10^{-4}a	2.8×10^{-4}a
Oloyin	9.7×10^{-4} a	1.2×10^{-4} ab	3.7×10^{-4}a	1.2×10^{-4}b
IT90K-76	2.8×10^{-4}a	7.5×10^{-5} b	4.3×10^{-4}a	1.8×10^{-4}ab
MSD (0.05)	2.6×10^{-3}	2.0×10^{-4}	6.0×10^{-4}	1.0×10^{-4}

Means followed by different alphabets within a column and sequence of planting are significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference.

Table 4.56: Photosynthetic attributes of cowpea varieties in cowpea sequential cropping system

Treatments	Phi 2 (mol e^- mol^{-1} quanta)	Phi NPQ	Phi no	Leaf temp. diff (°C)
First planting				
IT93K-452-1	0.52a	0.28	0.31a	-3.60a
IT99K-573-1-1	0.53a	0.15a	0.33a	-4.95a
TVX-3236	0.53a	0.15a	0.31a	-4.69a
Kanannado	0.53a	0.18a	0.30a	-3.45a
Oloyin	0.57a	0.13a	0.30a	-3.68a
IT90K-76	0.54a	0.15a	0.31a	-3.27a
MSD (0.05)	0.13	0.23	0.05	2.58
Second planting				
IT93K-452-1	0.41a	0.32a	0.29a	-3.59a
IT99K-573-1-1	0.42a	0.33a	0.28a	-2.77a
TVX-3236	0.42a	0.35a	0.24a	-2.72a
Oloyin	0.40a	0.29a	0.27a	-2.41a
IT90K-76	0.41a	0.31a	0.28a	-2.81a
MSD (0.05)	0.10	0.11	0.13	2.36

Means followed by the same alphabets within a column and sequence of planting are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference, temp. diff.- temperature difference biomass yield (8.03 t ha^{-1}). The least was recorded in Oloyin variety (4.17 t ha^{-1}) which had statistically similar values with IT99K-573-1-1, TVX-3236 and IT90K-76 varieties.In the second planting, there was no significant difference between the biomass yields of the five varieties in 2016. In 2017, IT99K-573-1-1 had the significantly highest biomass yield (2.97 t ha^{-1}) similar to IT93K-452-1. The least was recorded in TVX-3236 (1.52 t ha^{-1}) statistically similar to the values recorded in Oloyin and IT90K-76 varieties (Table 4.57).

ii. Grain yield attributes

Among the plants sown in the first planting, IT93K-573-1-1 had the significantly longest pods in the first planting in both two years followed by Oloyin and IT90K-76 pods which had similar pod length. Kanannado plants produced the significantly shortest pods in the two years (Table 4.58). In the second planting, all the varieties had similar pod lengths except TVX-3236 in both years. In 2016 however, IT93K-452-1 had similar pod length with TVX-3236 plants.

Kanannado had the significantly heaviest seeds among the varieties sown in the first planting in the two years. This was followed by the values recorded in the remaining varieties which were similar except TVX-3236 which had the significantly lightest seeds (Table 4.58). In the second planting, Oloyin plants had the heaviest seeds which was similar to the value recorded in IT99K-573-1-1 in 2016. In 2017, the seed weight of all the varieties except TVX-3236 were similar. TVX-3236 had the significantly lightest seeds in the two years.

The number of seeds per pod of IT93K-452-1, IT99K-573-1-1 and IT90K-76 in the first planting in 2016 were similar and significantly higher than the values recorded in Kanannado and Oloyin variety. Kanannado had the significantly least number of seeds per pod. In the second planting all the varieties produced statistically similar number of seeds per pod in 2016. In 2017, IT93K-452-

Table 4.57: Shoot biomass yield (t ha^{-1}) of cowpea varieties in cowpea sequential cropping system

Treatments	2016	2017
First planting		
IT93K-452-1	4.39d	5.61b
IT99K-573-1-1	6.76b	4.80bc
TVX-3236	5.35c	5.07bc
Kanannado	9.16a	8.03a
Oloyin	6.92b	4.17c
IT90K-76	7.23b	4.82bc
MSD (0.05)	0.88	0.92
Second planting		
IT93K-452-1	1.91a	2.40ab
IT99K-573-1-1	2.37a	2.97a
TVX-3236	2.00a	1.52c
Oloyin	1.64a	1.93bc
IT90K-76	1.72a	1.80bc
MSD (0.05)	1.22	0.73

Means followed by the same alphabets within a column and sequence of planting are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference

1, TVX-3236 and IT90K-76 had significantly higher number of seeds per pod than IT99K-573-1-1 plants (Table 4.58).TVX-3236 and IT93K-452-1 had the similar grain harvest index (0.18 and 0.17 respectively) which were significantly higher than the remaining varieties sown in the first planting of 2016. This was followed by IT99K-573-1-1 and IT90K-76 which equally had similar grain harvest index. The least was recorded in Oloyin and Kanannado which had the significantly least grain harvest index (0.05 and 0.02 respectively). In 2017, IT90K-76 plants and TVX-3236 had the highest grain harvest index (0.26 and 0.25 respectively) followed by IT93K-452-1 and IT99K-573-1-1 (0.16 and 0.14 respectively). The significantly lowest grain harvest index was recorded in Kanannado variety (0.05) which was similar to Oloyin variety (0.09) (Table 4.58).

In 2016 second planting, IT90K-76 had the significantly highest grain harvest index (0.34) similar to IT99K-573-1-1 (0.29). The least was recorded in TVX-3236 and Oloyin variety which had similar grain harvest index (0.18 and 0.20 respectively). In 2017, all the varieties had similar grain harvest index except that TVX-3236 and IT90K-76 had significantly higher grain harvest index than Oloyin variety which had the least grain harvest index (Table 4.58).

The shelling percentage of the varieties were only significant in 2017 (Table 4.58). IT93K-452-1 had significantly higher shelling percentage than Kanannado, Oloyin and IT90K-76 in the first planting. In the second planting however, the shelling percentage of IT93K-452-1 was only significantly greater than TVX-3236.

Table 4.58: Grain yield attributes of cowpea varieties sown in cowpea sequential cropping system

Treatments	Pod length (cm)		100 seed weight (g)		Seed per pod		Grain harvest index		Shell percentage (%)	
	2016	2017	2016	2017	2016	2017	2016	2017	2016	2017
First planting										
IT93K-452-1	15.88b	15.59b	13.09bc	15.14b	14.43a	12.17a	0.17a	0.16bc	20.30a	25.72a
IT99K-573-1-	19.89a	17.98a	15.19b	14.68b	14.10a	13.13a	0.13b	0.14c	19.73a	24.70ab
TVX-3236	13.50c	12.90c	8.58d	10.65c	15.03a	12.43a	0.18a	0.25ab	19.14a	21.34abc
Kanannado	8.69d	10.21d	17.91a	21.51a	8.10c	9.07b	0.05c	0.05d	17.32a	20.02bc
Oloyin	14.23bc	14.81b	12.73bc	15.08b	10.87b	12.07a	0.02c	0.09cd	17.16a	19.61bc
IT90K-76	16.28b	14.56b	12.41c	15.61b	15.87a	12.63a	0.11b	0.26a	21.33a	18.23c
MSD (0.05)	2.12	1.64	2.52	3.84	2.04	2.35	0.03	0.09	5.71	5.56
Second planting										
IT93K-452-1	15.06ab	15.58a	15.20b	17.62a	12.2a	12.17a	0.27bc	0.34ab	21.59a	27.42a
IT99K-573-1-	16.83a	17.40a	16.46ab	16.79a	10.77a	9.83b	0.29ab	0.36ab	24.76a	23.30ab
TVX-3236	12.22b	11.29b	8.88c	9.38b	11.40a	12.13a	0.18d	0.40a	19.93a	21.25b
Oloyin	15.53a	15.90a	19.36a	19.25a	11.93a	11.37ab	0.20cd	0.25b	19.63a	23.14ab
IT90K-76	15.41a	15.50a	13.76b	16.11a	11.97a	11.83a	0.34a	0.41a	19.10a	22.37ab
MSD (0.05)	2.94	2.67	4.04	3.78	2.08	1.67	0.07	0.16	6.18	5.44

Means followed by the same alphabets within a column and sequence of planting are not significantly different at P=0.05 using Waller-Duncan K-ratio T-test, MSD- Minimum significant difference

iii. Grain yield

Among the plants sown in the first planting in 2016, TVX-3236 had the highest grain yield (1.84 t ha^{-1}). This was followed by the value recorded in IT99K-573-1-1 variety (1.17 t ha^{-1}). The values recorded in IT93K-452-1 and IT90K-76 were similar (1.08 and 1.07 t ha^{-1} respectively) and significantly higher than the yield obtained in Kanannado (0.46 t ha^{-1}) and Oloyin (0.13 t ha^{-1}) which had the least grain yield (Figure 4.38A). In 2017, TVX-3236 equally had the highest grain yield (1.83 t ha^{-1}) which was similar to the value recorded in IT90K-76 (1.88 t ha^{-1}). The value obtained in IT93K-452-1 and IT99K-573-1-1 were similar (1.14 and 0.96 t ha^{-1} respectively) and significantly higher than the value recorded in Kanannado and Oloyin varieties which had similar and the least grain yield (0.44 and 0.42 t ha^{-1} respectively) (Figure 4.38B).

IT99K-573-1-1 had the highest grain yield (1.84 tha^{-1}) among the plants sown in the second planting in 2016. This was similar to the value recorded in IT90K-76 (1.64 t ha^{-1}). The least was obtained in Oloyin variety (0.52 t ha^{-1}) which had similar value with TVX-3236 in the second planting (0.55 t ha^{-1}) in 2016. In 2017, IT93K-573-1-1 had the significantly highest grain yield (1.25 t ha^{-1}) and the value obtained in the remaining varieties except Oloyin were similar. Oloyin had the least grain yield (0.52 t ha^{-1})

iv. Cumulative grain yield

In 2016, IT99K-573-1-1 plants had the highest cumulative grain yield (3.00 t ha^{-1}) which was statistically similar to the value obtained in IT90K-76 (2.70 t ha^{-1}) but significantly higher than the values recorded in IT93K-452-1 and TVX-3236 (2.18 and 2.02 t ha^{-1} respectively). Kanannado had the least cumulative grain yield similar to Oloyin variety (0.65 t ha^{-1}) (Figure 4.38C).

In 2017, IT90K-76 had the highest cumulative yield (3.58 t ha^{-1}) similar to TVX-3236 (3.50 t ha^{-1}). The values obtained in IT99K-573-1-1 and IT93K-452-1 were intermediate (2.95 and 2.77 t ha^{-}

^{1} respectively) but statistically similar to the value recorded in the TVX-3236 and IT90K-76. Kanannado variety had the significantly lowest cumulative yield (0.55 t ha^{-1}) which was statistically similar to the value recorded in Oloyin variety (1.15 t ha^{-1}) (Figure 4.38D).

4.1.3.4 Profitability of cowpea varieties in cowpea sequential cropping system

Among the varieties sown in the first planting in 2016, TVX-3236 was the most profitable with the cost-benefit ratio of 1.01 and gross margin of N312,660 (Table 4.59) . The remaining varieties except Kanannado and oloyin had the GM of between 209,500 and 259,000 and profit / cost ratio of between 0.67 and 0.70. Kanannado and Oloyin varieties had negative GM (-N60,000 and -N197,850 respectively). In the second planting, all the varieties had positive gross margin with IT99K-573-1-1 being the most profitable having the gross margin of N658, 130 and profit / cost ratio of 3.32 followed by IT90K-76 which had the gross margin of 557,445 and profit / cost ratio of 2.99 . The least profitable was Oloyin with the GM of N74,785 and profit / cost ratio of 0.43. TVX-3236 was not as profitable in the second planting as in the first with a profit / cost ratio of 0.57 and GM of 97,250 which is 49% reduction in gross margin compared to the first planting (Table 4.59).

In 2017, IT90K-76 and TVX-3236 were the most profitable in the first planting with GM of N748,610 and 726,645 respectively and profit / cost ratio of 2.35 and 2.30 respectively. Kanannado and Oloyin were not profitable with a GM of –N35, 575 and –N43,095 respectively and profit margin of -0.11 and -0.15 respectively. IT99K-573-1-1 maintained the most profitable variety in the second planting in 2017 with the GM of N524,831 and profit / cost ratio of 2.73 (Table 4.60) . Though Oloyin maintained the least profitability with GM of 132,026 and profit margin of 0.78, there was a great increase (820%) in the gross margin of Oloyin in the second planting over the first planting.

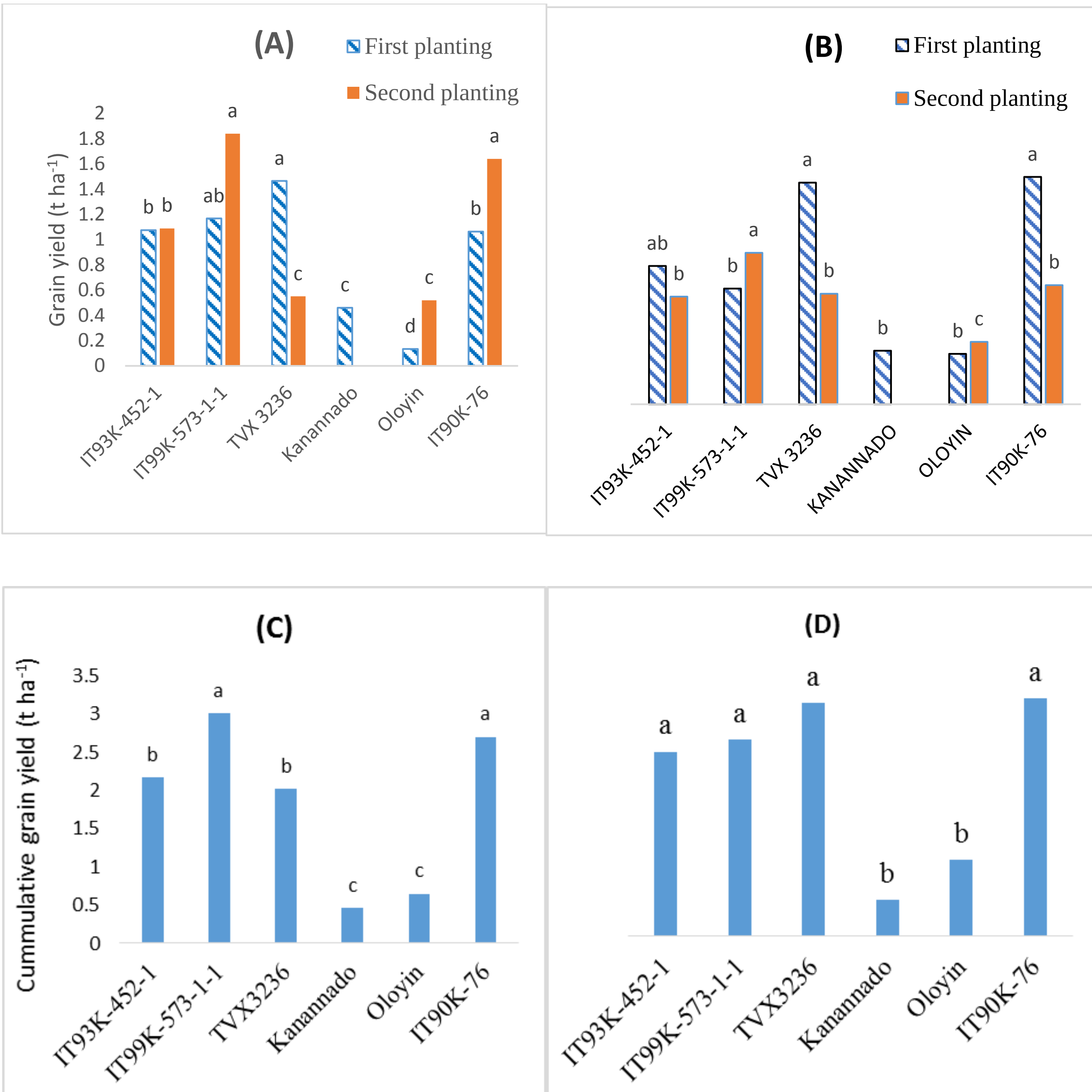


Figure 4.38 A & B: Grain yield of cowpea varieties in cowpea sequential cropping system in 2016 and 2017 respectively; C & D: cumulative grain yield of cowpea varieties in cowpea sequential cropping system in 2016 and 2017 respectively

Bars carrying similar alphabets in the same series are not significantly different using Waller Duncan K-ratio T-test at P=0.05

Cumulatively, planting the varieties in cowpea sequential cropping system increased the profitability of the varieties than the traditional practice of planting once by 63.34 and 106.31% for IT93K-452-1 in 2016 and 2017 respectively, 39.35 and 47.62% for IT99K-573-1-1 in 2016 and 2017 respectively, 321.50 and 213.34% for TVX-3236 in 2016 and 2017 respectively, 37.89 and 195.80% for IT90K-76 in 2016 and 2017 respectively. IT99K-573-1-1 was the most profitable with the cumulative GM of N917,130 in 2016 (Table 4.59) however in 2017, IT90K-76 was the most profitable with the cumulative GM of N1,133,967 and Kanannado maintained the least profitable with a loss of -60000 and –N33,575 in 2016 and 2017 respectively.

Table 4.59: Profitability of cowpea varieties sown in sequence in 2016

	IT93K-452-1	IT99K-573-1-1	TVX-3236	Kanannado	Oloyin	IT90K-76
			1st planting			
Total cost	308,500	309,500	306,500	308,000	286,700	303,500
Total revenue	516,000	566,500	617,160	246,000	88,850	516,500
Gross margin	209,500	259,000	312,660	-60,000	-195,850	215,000
NFI	207,500	257,000	310,660	-62,000	-197,850	213,000
Profit / cost ratio	0.67	0.83	1.01	-0.20	-0.69	0.70
			2nd planting			
Total cost	186,500	198,500	171,500	-	174,500	189,500
Total revenue	517,280	856,630	268,750	-	249,285	756,945
Gross margin	330,780	658,130	97,250	-	74,785	567,445
NFI	330,780	658,130	97,250	-	74,785	567,445
Profit / cost ratio	1.77	3.32	0.57	-	0.43	2.99
			Cummulative			
Total cost	495,000	508,000	478,000	308,000	461,200	493,000
Total revenue	1,033,280	1,423,130	885,910	246,000	338,135	1,273,445
Gross margin	540,280	917,130	409,910	-60,000	-121,065	782,445
NFI	538,280	915,130	407,910	-62,000	-123,065	780,445
Profit / cost ratio	1.09	1.80	0.85	-0.20	-0.27	1.58

Table 4.60: Profitability of cowpea varieties sown in sequence in 2017

	Varieties					
	IT93K-452-1	IT99K-573-1-1	TVX-3236	Kanannado	Oloyin	IT90K-76
			1st planting			
Total cost	309,500	308,500	315,500	314,500	296,500	318,500
Total revenue	663,060	556,455	1,042,145	278,925	253,405	1,067,110
Gross margin	355,560	249,955	728,645	-33,575	-41,095	750,610
NFI	353,560	247,955	726,645	-35,575	-43,095	748,610
Profit / cost ratio	1.14	0.80	2.30	-0.11	-0.15	2.35
			2nd planting			
Total cost	181,500	192,500	178,500	-	169,500	181,500
Total revenue	515,946	717,331	520,031	-	301,526	564,857
Gross margin	334,446	524,831	341,531	-	132,026	383,357
NFI	334,446	524,831	341,531	-	132,026	383,357
Profit / cost ratio	1.84	2.73	1.91	-	0.78	2.11
			Cummulative			
Total cost	491,000	501,000	494,000	314,500	466,000	500,000
Total revenue	1,179,006	1,273,786	1,562,176	278,925	554,931	1,631,967
Gross margin	690,006	774,786	1,070,176	-33,575	90,931	1,133,967
NFI	688,006	772,786	1,068,176	-35,575	88,931	1,131,967
Profit / cost ratio	1.40	1.54	2.16	-0.11	0.19	2.26

4.2 Discussion

The significant increase in shoot biomass yield obtained in plants that received 90 kg N ha^{-1} is an indication that cowpea performance is limited by N availability in the Nigeria savannas. Several researchers have also confirmed that although cowpea fixes N, it can also benefit from external N supply especially in the tropical region where N is inherently low in the soil (Abayomi *et al.,* 2008, Namvar *et al.*, 2011). This is truly reflected in the result of the soil analysis of the soils obtained from the twenty locations in Nigeria savannas with N ranging from 0.1-1.3 g kg^{-1} which is very low according to the rating of Chude *et al.* (2012). This result agrees with the report of Fening *et al*. (2001) who assessed the potential of improving N fixation in cowpea in Ghananian soils**.** The authors reported that all the 45 cowpea cultivars used showed significant response to increasing N fertilizer application to a particular level**,** indicating that N fixation was not providing the plants with sufficient N for optimum growth and yield**,** hence indicating the need for inoculation in cowpea to improve the N fixed. Abayomi *et al*. (2008) reported that though cowpea fixes N, it may suffer from temporary N deficiency once the cotyledonary N reserve is exhausted. It takes about two to three weeks before the onset of nodule formation in this crop and nodules do not start fixing nitrogen immediately they are formed. Also at the pod filling stage, nodules generally lose their ability to fix nitrogen because assimilate is preferably transferred to the developing seeds rather than the nodules. Hence legumes could benefit from external N supply at this period if the soil N is low (Lindemann and Glover, 2015).

The insignificant difference between the number of nodules and weight of the inoculated and uninoculated plants indicated that the elite strains used are equally competitive and infective. They successfully competed for the nodule sites; the failure of the nodulation to translate to higher nitrogen fixation and biological yield indicates that they may not be as effective as the indigenous rhizobia. Similar result was reported by Batista *et al.* (2017) where BR 3267 inoculated plants produced similar number and weight of nodules as the control plants.

A rhizobium could be infective but not effective. A rhizobia strain is infective if it has the ability to form nodules with a particular legume while effectiveness is the ability of those nodules to fix nitrogen. To achieve a positive response to rhizobium inoculation, the introduced rhizobium must be infective, competitive and effective. The rhizobium must be able to compete with native rhizobia for the infection sites of the host legumes. Successful competition for nodule sites by native rhizobia is one of the reasons for the failure to achieve a response to inoculation with elite rhizobia strains (Theis *et al*., 1991). Date (2000) reported that the quality of the native rhizobia can affect the plants response to inoculation. A higher population of symbiotically competitive indigenous rhizobia will have an advantage over the introduced strains because it is already adapted to the environmental conditions of the area. Theis *et al*. (1991) reported that the response to inoculation and competitive success of inoculant rhizobia are inversely related to number of indigenous rhizobia. Slattery and Pearce (2002) in their report stated that where there are low (<50 rhizobium bacteria /g soil) naturalized populations of rhizobia specific to a target legume, the introduction of new strains by seed inoculation is normally successful. On the other hand, inoculation into soils where naturalized rhizobia population is high ($>10^3$ Rhizobium bacteria g/m soil) may result in non-response to inoculation. In addition, improper handling and poor quality could contribute to lack of response to rhizobia inoculation in cowpea. The success of commercial inoculants is dependent on the number of viable bacteria available to participate in the infection process at the point of use (Catroux *et al*., 2001). In this research, the plants inoculated with the elite strains equally nodulated well close to the crown of the roots of the plant and the percentage of the infected and active nodules were not different from that of the control plants indicating that the inoculants used were of good quality and the inoculation was successful but the strains were just not as efficient in fixing nitrogen as the native strain in the study area. Pule-Meulenberg *et al.* (2010) reported that cowpea bradyrhizobia vary significantly in their nitrogen fixing ability. The result obtained in this study agrees with the findings of Otieno *et al*. (2007) who observed that inoculation with rhizobia increased number of nodulation and nodule weight but the increase in nodulation did not translate to increase in plant growth or grain yield in common bean, green gram, lima bean and lablab. The authors asserted that the rhizobium strain used for the inoculation may be infective but not effective enough in bringing about the desired change.

Giller (2001) reported that the ability of the plant to form nodules as a result of symbiosis with a rhizobium is not enough to obtain an effective N fixation. Similarly, Omiroua *et al*. (2016) reported that the native rhizobia inoculant used in their study was more competitive than the elite inoculum in occupying the cowpea nodules but contrary to the result in this study, the applied inoculum was more efficient in fixing nitrogen. This may be attributed to the variation in native rhizobia composition and effectiveness. Omiroua *et al*. (2016) reported that the population of soil native rhizobia may vary in composition and effectiveness. This may be an indication that the indigenous rhizobia strains in the study area are more effective than all the elite strains used in this study. Lots of variation exists in the response of legumes to rhizobia inoculation (Giller, 2001) and not many positive response have been recorded particularly in cowpea probably because cowpea is a promiscuous legume and the bradyrhizobium that can infect the crop are numerous in African soils Batista *et al*. (2017). Mpepereki *et al*. (2000) reported that cowpea is the most promiscuous legume which has been intensively studied**,** nodulating with a wide range of fast and slow growing rhizobia. Furthermore, not many commercial inoculants have been developed for cowpea compared to some other legumes like soya beans. However, contrary to the reports obtained in this study, some researchers have reported increase in growth and yield of cowpea in response to rhizobia inoculation (Fening and Danso, 2001; Arumugam *et al.*, 2010; Nyoki and Ndakidemi, 2014). Giller (2001) attributed the inconsistency in reponse to rhizobia inoculation to different method of inoculation, use of different inoculum strains and emphasized that inconsistent results were obtained even when the same researchers carried out several experiments. Soil and other environmental factors could aslo contribute to the inconsistent results obtained in response to inoculation. Aliyu *et al*. (2013) reported that soil pH and nutrient status could affect response to rhizobia inoculation.

The higher values of nodulation and N-fixation parameters obtained at 20 and 40 kg P ha^{-1} application compared to the unfertilized confirms the assertion that adequate P nutrition is needed for legumes (Weisany *et al*., 2013). High phosphorus supply has been reported to be required for nodulation (Elkoca *et al*.**,** 2007) as nodules themselves are strong sink for Weisany *et al.* (2013) reported that the phosphorus content per unit dry weight is considerably higher in the nodules than in the roots and biomass**,** particularly at low external phosphorus supply. Phosphorus is known to promote early root formation and the formation of lateral**,** fibrous and healthy roots**,** which play an important role in N_2 fixation, water and nutrient uptake (Niu *et al*.**,** 2012). This explains the higher N-uptake obtained in this study as P application rate increased.

Increase in grain yield by 49-52% as a result of phosphorus application further confirms the importance of phosphorus in legume productivity. Several workers have reported that P increased the grain yield of legumes. Amjad *et al*. (2004) reported that pod yield of peas increased with increasing phosphorus rates and the authors obtained the highest yield at 69 kg P_2O_5 ha^{-1}. Ndakidemi *et al*. (2006) reported that application of P to inoculated plants increased the grain yield of soybean and that application of 26 kg P ha^{-1} increased the profit margins by 84% - 102% in two districts of Tanzania. Magani and Kuchinda (2009) reorded an increase in the productivity of cowpea as a result of phosphorus application and therefore recommended the application of 37.5 kg P ha^{-1} in the northern Guinea savanna of Nigeria. This is more than the optimum rate obtained in this study. The optimum phosphorus rate obtained in this study (20 kg P ha^{-1}) agrees with the recommendation of Chude *et al*. (2012) for cowpea in soils with medium P content.

The suppression of nodulation by application of N fertilizer in this study is not surprising as N fertilizer has been reported to reduce number of nodules and mass in legumes by many researchers. Leguminous plants can utilize both symbiotically fixed N and mineral N absorbed from the soil to meet its need. If there is enough mineral N in the soil to meet the need of the plant, nodulation and N-fixation is reduced. Plants tend to stop nitrogen fixation when the soil nitrogen is high. Havlin *et al*. (2005) and Zhou *et al*. (2006) reported that excess nitrate in the soil can reduce nitrogenase activity and hence, reduce nitrogen fixation. Laws and Graves (2005) reported that nitrogen inhibits nodulation and reversibly suppresses nitrogen fixation in nodules of *Almus mauritima*. Otieno *et al.* (2007) similarly reported that nitrogen fertilizer application significantly reduced nodulation in lablab, common bean and green gram. They stated further that the effect of nitrogen fertilizer application on legume is dependent on species, parameters being measured and other environmental factors. Zhou *et al*. (2006) similarly reported that excess N supply may cause some negative effects on soybean plants.

Though the uninoculated plants fixed marginally higher nitrogen than the inoculated plants in this study (37-57 kg N ha^{-1}), the amount fixed is still far below the potential N-fixed reported in cowpea. Bationo *et al.* (2002) reported that with efficient soil fertility management, cowpea can fix up to 88 kg N ha^{-1} which will result in an increase in nitrogen use efficiency on the succeeding cereal crop. Giller (2001) reported a potential N-fixed of 200 kg N ha^{-1}. This confirms that the indigenous rhizobia strains in the study area is not fixing enough nitrogen and hence, the need for rhizobia inoculation of cowpea with highly effective strains to bridge the N-fixation gap that exists. Enhanced nitrogen fixation through inoculation provides an alternative to the application of chemical nitrogenous fertilizers which is more expensive and not environmentally friendly. Studies on cereal–cowpea rotation revealed that yields of cereals succeeding cowpea could double compared to continuous cereal cultivation (Bationo *et al*., 2002) as a result of the left over N from the preceding cowpea.

The fact that producers inoculate their legume crops to improve soil fertility without spending so much on N fertilizer have been documented by several researchers (Kennedy *et al.,* 2004; Matiru and Dakora, 2004). Microbial inoculants can be used as an economic input to increase crop productivity; fertilizer doses can be lowered and more nutrients can be harvested from the soil (Chen, 2006; Rosen and Allan, 2007). Nigeria farmers can equally exploit this to cut down on the use of chemical N fertilizers. Otieno *et al*. (2007) reported that some African countries such as Rwanda, Malawi, Egypt and Zimbabwe have turned to efficient exploitation of biological nitrogen fixation (BNF) by legumes in their farming system in an attempt to cut down on fertilizer expenses. Odame (1997) reported that rhizobium strains fixed more nitrogen as compared with applying a recommended 90 kg of mineral nitrogen fertilizer per ha in common beans. A great advantage of rhizobia inoculation is that it is much cheaper than mineral nitrogen fertilizer (Chianu *et al.,* 2010). Nyoki and Ndakidemi (2014) similarly reported that inoculation may be up to 15 times cheaper than commercially produced nitrogen fertilizers. The result of this study emphasizes the need to develop and test more rhizobia inoculants to bridge the N-fixation and productivity gap that exists in cowpea. Not many inoculants have been developed for cowpea compared to some other legumes like soybean and common bean which is not as common as cowpea in Nigeria. It has been reported that cowpea is the most economically important indigenous African legume that supplies 40% of the daily dietary protein of most of Nigerians (Muleba *et al*., 1997).

Grain yield is the integration of many characters that affect plant growth throughout the growing season. Yield characters such as number of flowers per plant, seed per pod, 100-seed weight, number of spikelet per spike have been found to be associated with the final yield of crop (Maunde *et al*., 2015). In this study, number of seed per pod, pod length, 100- seed weight and number of pods per plant exhibited significant and positive relationship with grain yield among the yield characters. However, number of pod per plant had the highest correlation coefficient (r=0.79**) with grain yield. Maunde *et al*. (2015) similarly reported that though 100-seed weight, number of seeds per pod, shell percentage correlated significantly with seed yield in groundnut, the contribution of harvest index and number of pods per plant to seed yield were the highest (r=0.74**). This result indicates that selecting cowpea cultivars with high pod loading capacity for cowpea improvement will improve cowpea productivity. Manggoel *et al*. (2012) identified number of penduncles per plant, number of flower per plant, 100-seed weight and number of pods per plant as selection criteria for obtaining good parental lines in cowpea breeding program. The authors recorded a correlation coefficient of r=0.74 between grain yield and number of pod per plant. Correlation coefficient provides a measure of association between assessed characters thus enabling researchers to identify the more important and lesser important characters that must be considered in breeding as well as elicit the essentiality of these characters during plant selection for increased yield (Mbah and Okoro, 2015). Maunde *et al*. (2015) similarly reported that examination of relationships between yield characters tend to give an insight into the importance of and possibility of exploiting these characters to improve the yield of crops through the use of certain agronomic practices and breeding programs. The significant and positive relationship that existed between grain yield and stem diameter, leaf area, number of leaves and shoot biomass yield indicated these parameters could equally be used as indices for selecting for high grain yield in cowpea. The significant negative correlation that existed between number of days to flowering and grain yield could be an indication that early flowering could be used as an index for selecting cowpea for high grain yield.

The significant and positive correlation that existed between shoot biomass yield and leaf chlorophyll, leaf area index, crop growth rate, net assimilation rate and leaf area ratio confirms the importance of these traits in assessing the rate dry matter accumulation in plants. Among these traits, CGR and LAI had highly significant and positive relationship with grain yield which was also confirmed by the regression analysis in which CGR, LAI and Phi 2 significantly predicted 67% of the variation in grain yield among the physiological traits indicating that these three traits could be used as a strong indicator of grain yield and can be a veritable tool in predicting grain yield even before harvest. This equally indicates that these physiological traits can be used as selection criteria in breeding programs thereby improving the efficiency of breeding programs by identifying appropriate indices to select cowpea cultivars (Evans and Fisher, 1999). CGR been the strongest predictor in this study corroborates the findings of Mbah and Okoro (2015) who opined that selection aimed at improving CGR would invariably lead to increase in yield.

The negative correlation between PAR and Phi 2 obtained in this study is similar to the result obtained by Harb *et al*. (2018) who reported reduction in Phi 2 and electron transport rate in *Pterocladiella capillacea* as irradiance increased. This could be attributed to energy associated with high PAR exceeding the photochemical demand or energy dissipation capacity of the plant. Photosensitivity, photo tolerance, photo-inhibition, and photo damage processes could occur when the energy received exceeds the photochemical demand or energy dissipation capacity of plants

(Hanelt and Figueroa, 2012). Photosynthetic photo inhibition occurs under excessive light availability where the irradiance is greater than the acclimation capacity, causing a reduction of photosynthetic activity (Hou and Hou, 2013). Under long periods of high light intensity, an accumulation of excess excitation energy usually occurs, consequently inducing photo stress conditions. Under this circumstance plants pigments such as carotenoids and chlorophyll a are destroyed (Beach *et al*., 2000).

In this study however, there was negative correlation between Phi 2 and phi npq which is a measure of the light that goes towards regulatory processes to reduce damage to the plant. Indicating that if the light energy is more than what can be harvested by photosystem II for manufacture of food, there was efficient regulated non photochemical quenching of excess of energy to prevent photo damage. This confirms the photo tolerance efficiency of cowpea plants as a typical tropical crop. Bukhov *et al.* (1998) reported that increase in phi npq may be a mechanism to downregulate photosynthetic electron transport so that production of ATP and NADPH would be in equilibrium with the decreased demand in the Calvin cycle in heat-treated leaves and to also avoid over-reduction of QA.

The reduced chlorophyll content and quantum yield of photosysytem II in P unfertilized plants obtained in this study confirms the assertion of Singh and Reddy (2016) who reported that decrease in chlorophyll concentration is a mechanism to avoid excessive light harvesting to protect photosystem II from photo damage under P deficiency. The authors reported further that under stress conditions, such as nutrient stress, decline in CO_2 assimilation also reduces the consumption of the chemical energy created in the light reaction of photosynthesis, which leads to the over excitation of photosystem II reaction center due to continuing photon absorption by chlorophyll molecules. This excess energy must be dissipated for optimum performance of photosystems and to avoid photo inhibition. Plants possess mechanisms to dissipate the excess excitation energy as heat, re-emite as chlorophyll fluorescence or by non-radiative mechanisms (Ivanov *et al.*, 2008). This explains why phi npq was in turn significantly higher in the P unfertilized plants.

Photosynthetic efficiency that was significantly higher in the P fertilized than unfertilized plants confirms the importance of P in photosynthesis. Fleisher *et al.* (2012) related the increase in photosynthesis activities as a result of P to the regulatory role of P in CO_2 assimilation pathway, increase in the amount and activity of rubisco, and ribulose-1,5-bisphosphate (RuBP) regeneration capacity. Photosynthetic processes are dependent on the phosphate precursors, inorganic phosphate or phosphorylated intermediates, such as ADP, ATP, NADP(H) and sugar phosphates, essential for energy transfer. Therefore, P deficiency affects plant photosynthetic capacity due to its direct effect on the tissue P status and especially phosphorus homeostasis in the cytosol and chloroplasts (Warren, 2011).

Kanannado variety could not be planted in two plantings because it was photoperiod sensitive and a late maturing variety. Kanannado is not an improved variety but it is highly relished by people in the study area because of its size of seed and taste. IT99K-573-1-1, IT90K-76, IT93K-452-1 and TVX-3236 are all improved variety, they are not photoperiod sensitive and they are relatively early maturing compared to Kanannado. This explains why the four varieties were able to complete two life cycle within the growing season. The wide gap between the yield of the improved and unimproved varieties used in this study emphasizes the need to use improved seed varieties by farmers and the need to strengthen the formal seed system to meet the farmers need in this wise. Alene and Manyong (2006) reported that productivity gains achievable from improved agricultural technologies such as use of improved varieties have not been fully exploited by farmers in developing countries. Timu *et al*. (2014) reported that the major attributes driving rapid adoption of crop varieties are taste, drought tolerance, yield, ease of cooking and the variety's ability to fetch a price premium. It is therefore important that breeders should focus not only on yield but other non-yield attributes like taste and ease of cooking to fully exploit the benefits of improved varieties.

The negative gross margin obtained for Kanannado and Oloyin especially in the first planting indicated that if farmers were to pay for all the inputs and labour on the farm, the farmer will produce Oloyin and Kanannado varieties at a loss. Oloyin planted in the first planting were highly susceptible to pythium wilt and pod rot and incidence of mosaic virus was high in Kannanado variety. This could have contributed to the significantly lower grain yield obtained in this two.

The grain yield obtained in this study is much higher than the average of 450 kg ha^{-1} obtained on farmers' fields (Omotosho**,** 2014). This could be as a result of better management practices, improved varieties and the cropping system used in this experiment. Most farmers in Nigeria don't plant cowpea as a major and sole crop. It's majorly planted as a minor crop in relay cropping system or as an intercrop with cereals (Bationo *et al*., 2002). The cropping system adopted by the farmer and the management practices are contributory factors to the low yield obtained on farmer's field. Alene and Manyong (2006) similarly reported that there is a big yield gap between the potential yield of cowpea and what is actually achieved on farmers' fields. The authors attributed the wide gap to poor extension services, institutional and cultural constraints and farmers' long adaptability to traditional practices and hence limited ability and willingness to achieve full adjustment of input levels. Pingali and Heisey (1999) reported that even when farmers have access to modern inputs such as improved seeds, they often lack the agronomic and crop management technologies and knowledge that are crucial for bridging the yield gap. Alene and Manyong (2006) emphasized that to bridge the yield gap that exists in cowpea, use of improved varieties by farmers must be accompanied with adoption of the right cropping system and management practices.

CHAPTER FIVE

5.0 CONCLUSION AND RECOMMENDATION

5.1 Conclusion

From the result of these experiments, it can be concluded that nitrogen limits the productivity of cowpea in Nigeria savannas. There was variation in the response of the cowpea varieties to rhizobia inoculation and N application in the soils of the different locations. Soils of Kontagora, Garatu, Abaji, Kaduna-south, Kuje, Kumbotso, Gezawa, Gwagwalada, Dawakin Kudu, Kaduna-north and Sabongida responded positively to N application. The cowpea varieties successfully formed symbiosis with the elite rhizobia strains indicating that rhizobia inoculation is capable of overcoming the N-limitation in cowpea if highly effective strains than what was used in this study is used. The strains tested in this study were not more effective than the indigenous rhizobia strain resident in the soil of the study area in increasing the photosynthetic efficiency, N-fixation, growth and productivity of cowpea. Application of phosphorus significantly increased the photosynthetic efficiency, growth, N-fixation, shoot biomass and grain yield of cowpea with application of 20 kg P ha^{-1} being the optimum increasing grain yield by 49-95% over the unfertilized plants. IT99K-573-1-1 maintained the highest productivity amongst the varieties however, TVX-3236 appeared to be more P efficient having the optimum growth rate, nodule weight and grain yield at lower P rates. The photosynthetic active radiation correlated significantly ($p<0.01$) and positively with ratio of light that goes towards non-photochemical quenching (phi npq) but negatively ($P<0.01$) with quantum yield of photosystem 2 (phi 2). The relative humidity and chlorophyll correlated significantly and positively with quantum yield of photosystem 2. Among the growth parameters, stem diameter, leaf area and number of leaves had positive and significant correlation with grain yield. Pod length, 100-seed weight and number of pods per plant exhibited significant and positive relationship with grain yield among the yield characters though number of pods per plant maintained the highest correlation coefficient with grain yield. Crop growth rate, leaf area index and quantum yield of photosystem II explained 67.29% of the variation in grain yield hence, these three physiological characters could significantly predict grain yield. Furthermore, these aforementioned growth, yield and physiological characters could be exploited to improve the productivity of cowpea through agronomic practices and plant breeding program.

All the varieties were successfully planted in two plantings in a growing season except Kanannado and there was significant variation in the growth and yield attributes of the cowpea varieties in sequential cropping system. IT93K-452-1, IT99K-573-1-1, TVX-3236 and IT90K-76 all had significantly higher ($P \leq 0.05$) grain yield and higher profitability than Oloyin and Kanannado variety which were produced at a loss in the first planting.

5.2 Recommendation

Crop growth rate, leaf area index and quantum yield of photosystem II, number of pods per plant should be exploited to improve the productivity of cowpea through agronomic practices and plant breeding programs. Application of 20 kg P ha^{-1} is recommended for the optimum performance of cowpea in the study area. More effective rhizobia inoculants for cowpea should be developed and tested in the study area. Farmers should be encouraged to adopt double cropping of improved, early and medium maturing varieties of cowpea which is capable of increasing the productivity and profitability of cowpea in the study area.

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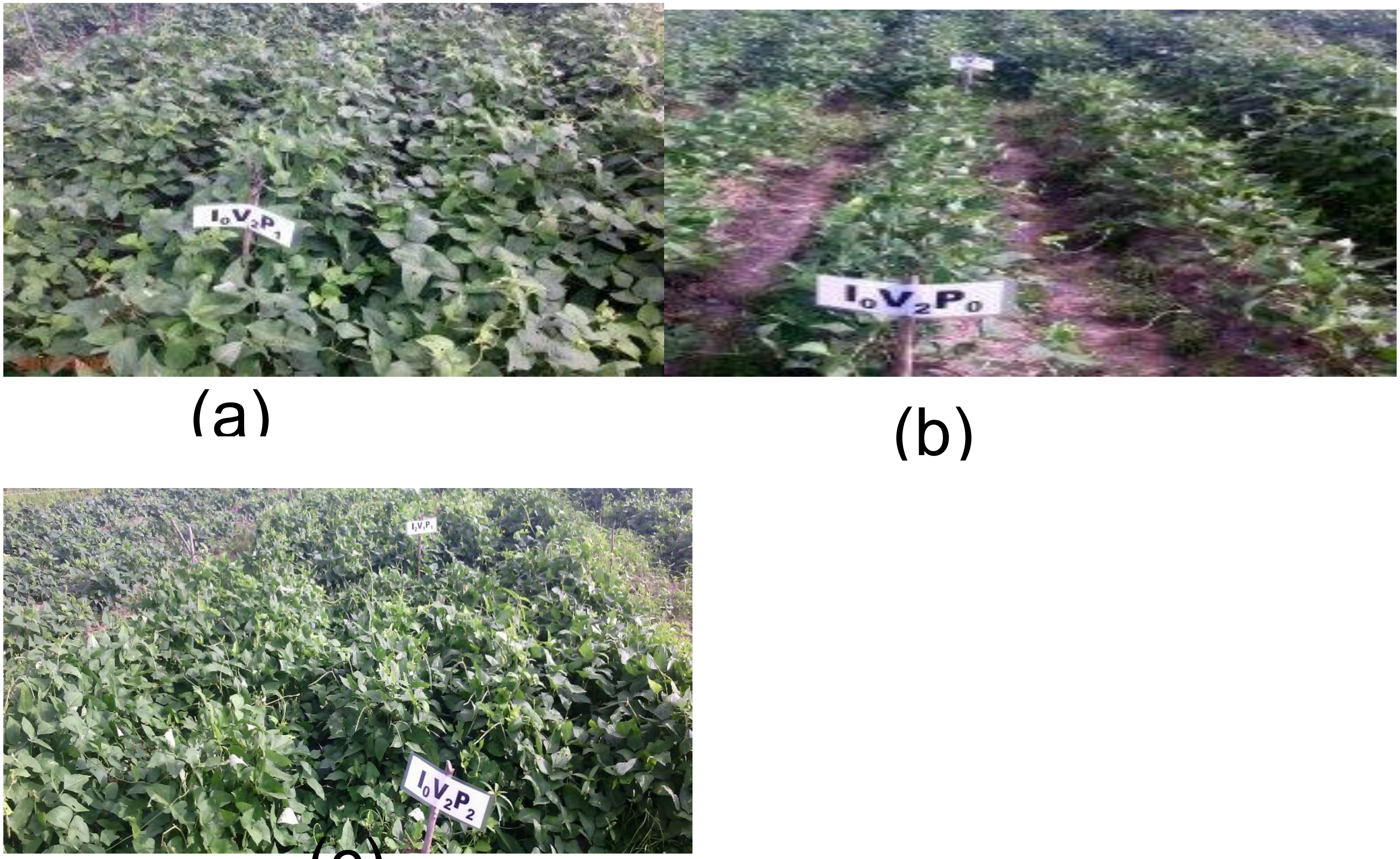
APPENDICES

Appendix A

Monthly meteorological data for 2015 -2017 cropping seasons

	2015				2016				2017			
	Total Rainfall	Relative humidity	Min. Temp.	Max. Temp.	Total Rainfall	Relative humidity	Min. Temp.	Max. Temp.	Total Rainfall	Relative Humidity	Min. Temp.	Max. Temp.
Months												
May	109.30	64.98	24.47	37.00	61.40	69.96	21.58	33.43	172.80	67.17	24.52	36.60
June	138.70	76.97	21.62	32.85	176.10	72.75	24.90	31.75	171.00	72.74	23.60	30.05
July	173.50	78.02	23.20	30.62	136.70	77.57	24.23	31.06	243.00	76.74	23.17	31.00
August	227.50	89.14	22.23	29.59	224.30	82.47	22.47	31.00	210.40	81.78	22.21	30.69
September	209.90	75.56	22.55	30.78	307.30	81.10	31.07	30.84	130.20	73.62	21.24	30.66
October	308.00	79.53	23.38	32.52	203.60	74.45	22.45	32.80	24.40	75.60	21.25	33.26
November	10.20	48.99	22.8	36.03	24.90	56.27	22.32	36.07	0.00	49.49	19.50	36.45

Appendix B

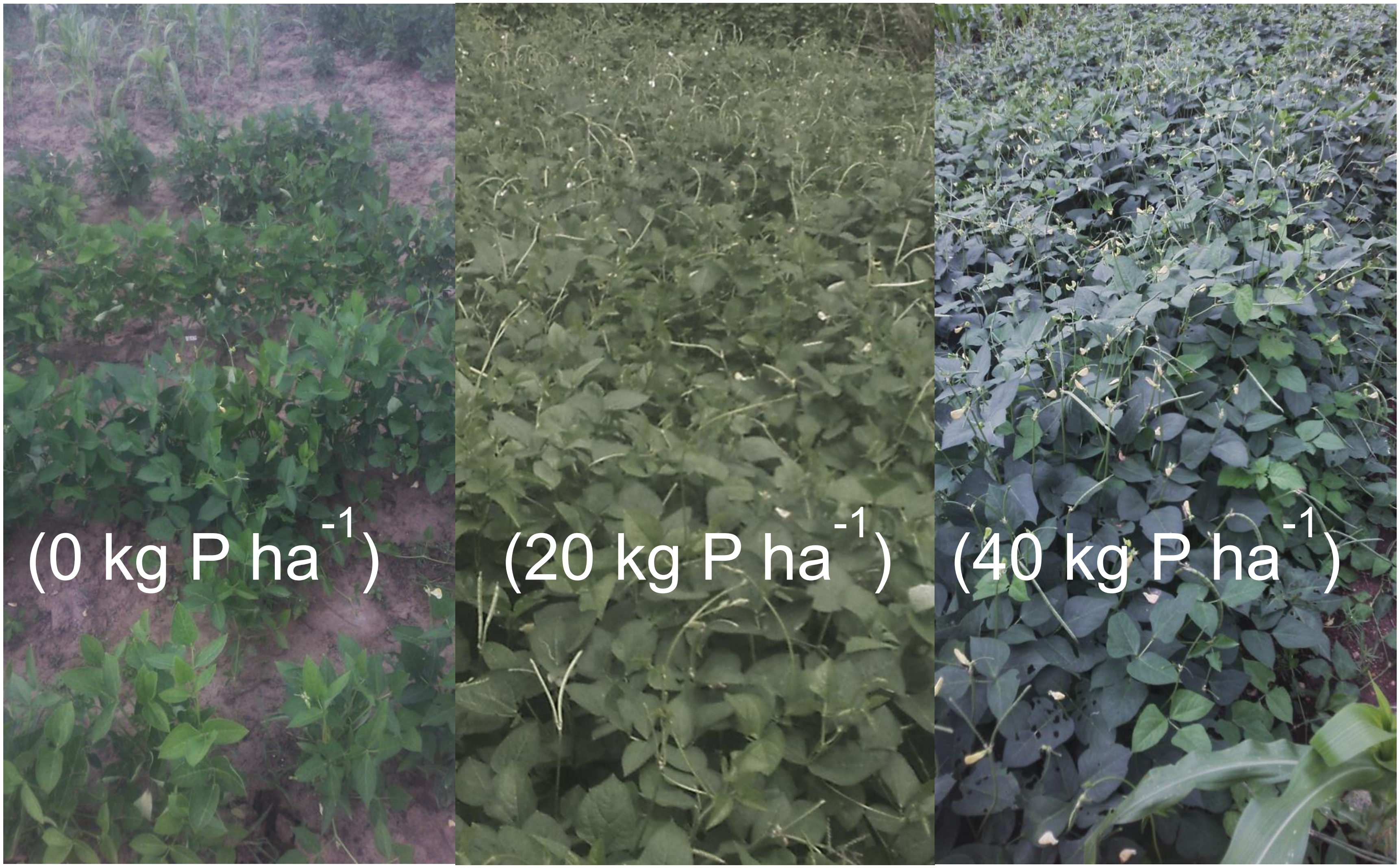


Experimental plot showing the P effect (a)- P-unfertilized plants, (b) and (c)– plants fertilized with 20 and 40 kg P ha^{-1} respectively, covering up the inter and intra-row spacing .

Appendix C

Phosphorus effect on nodulation

Appendix D



Experimental plots showing the P effect on the pod set

Appendix E

Farmers visit to the experimental plot

Appendix F

Soil physical and chemical properties of the experimental sites

Soil parameters	Farm 1	Farm 2	Farm 3
Particle size distribution			
Sand (%)	74.30	70.40	75.80
Silt (%)	10.50	11.50	88.00
Clay (%)	15.20	18.10	15.40
Texture	Sandy loam	Sandy loam	Sandy loam
Chemical properties			
pH (H_2O)	5.99	6.52	6.50
Organic carbon (g kg^{-1})	1.35	0.60	1.27
Total Nitrogen (g kg^{-1})	0.15	0.18	0.10
Available phosphorus (mg kg^{-1})	20.07	18.90	18.50
Exchangeable bases (cmol kg^{-1})			
Ca	0.65	0.85	0.70
Mg	0.95	0.90	1.80
Na	0.25	0.26	0.24
K	0.17	0.19	0.16
ECEC	2.02	2.20	2.90

Appendix G

Profitability of cowpea varieties sown in the first sequence in 2016

		Varieties				
Items	1	2	3	4	5	6
Variable cost			(N/ha)			
Seed	15,000	15000	12000	18000	15000	15,000
Fertilizer	82,000	82000	82000	82000	82000	82,000
Herbicide	4,500	4500	4500	7500	4500	4,500
Pesticide	17,500	17,500	17,500	12500	17,500	12,500
Labour						
Land clearing	20,000	20000	20000	20000	20000	20,000
Ridging	25,000	25000	25000	25000	25000	25,000
Herbicide spraying	6,000	6000	6000	6000	6000	6,000
Pesticide spraying	31,500	31,500	31,500	27000	31,500	31,500
Weeding	30,000	30,000	30,000	45,000	30,000	30,000
Fertilizer application	20,000	20000	20000	20000	20000	20,000
Harvesting	20,000	20000	20000	15000	8000	20,000
Threshing	10,000	10000	10000	5000	4000	10,000
Storage	5,000	6000	6000	3000	1200	5,000
Transportation	20,000	20,000	20,000	20,000	20,000	20,000
Total variable cost	**306,500**	**307,500**	**304,500**	**306,000**	**284,700**	**301,500**
Fixed cost						
Land rentage	2,000	2000	2000	2000	2000	2,000
Total fixed cost	**2000**	2000	2000	2000	2000	**2000**
Total cost	**308,500**	**309,500**	**306,500**	**308,000**	**286,700**	**303,500**
Revenue						
Grain yield	486,000	526500	586160	207000	59850	481,500
Stover yield	20,000	30000	20000	34000	26000	25,000
Chaff	10,000	10000	11000	5000	3000	10,000
Total revenue	**516,000**	**566,500**	**617,160**	**246,000**	**88,850**	**516,500**
Gross margin	209,500	259,000	312,660	-60,000	-195,850	215,000
NFI	207,500	257,000	310,660	-62,000	-197,850	213,000
Profit / cost ratio	0.672609	0.83037	1.01357	-0.2013	-0.6901	0.70181

1=IT93K-452-1, 2=IT99K-5773-1-1, 3=TVX-3236, 4= Kanannado, 5=Oloyin, 6=IT90K-76, NFI=Net financial income

Appendix H

Profitability of cowpea varieties sown in the second sequence in 2016

	Varieties				
Items	1	2	3	4	5
Variable cost			(N/ha)		
Seed	15,000	15,000	12,000	15,000	15,000
Fertilizer	6,000	6,000	6,000	6,000	6,000
Herbicide	4,500	4,500	4,500	4,500	4,500
Pesticide	12,500	12,500	12,500	12,500	12,500
Labour					
Ridging	25,000	25,000	25,000	25,000	25,000
Herbicide spraying	6,000	6,000	6,000	6,000	6,000
Pesticide spraying	22,500	22,500	22,500	22,500	22,500
Weeding	30,000	30,000	30,000	30,000	30,000
Fertilizer application	10,000	10,000	10,000	10,000	10,000
Harvesting	20,000	25,000	15,000	15,000	20,000
Threshing	10,000	15,000	5,000	5,000	13,000
Storage	5,000	7,000	3,000	3,000	5,000
Transportation	20,000	20,000	20,000	20,000	20,000
Total variable cost	**186,500**	**198,500**	**171,500**	**174,500**	**189,500**
Fixed cost					
Land rentage					
Total fixed cost					
Total cost	**186,500**	**198,500**	**171,500**	**174,500**	**189,500**
Revenue					
Grain yield	494,280	828630	247,550	232785	736,245
Stover yield	13,000	16,500	15,700	11,000	10,000
Chaff	10,000	11,500	5,500	5,500	10,700
Total revenue	**517,280**	**856,630**	**268,750**	**249,285**	**756,945**
Gross margin	330,780	658,130	97,250	74,785	567,445
NFI	330,780	658,130	97,250	74,785	567,445
Profit / cost ratio	1.773619	3.31552	0.56706	0.42857	2.99443

1=IT93K-452-1, 2=IT99K-5773-1-1, 3=TVX-3236, 4=Oloyin, 5=IT90K-76, NFI=Net financial income

Appendix I

Profitability of cowpea varieties sown in the second sequence in 2017

Items	1	2	3	4	5
		Varieties			
Variable cost			(N/ha)		
Seed	15,000	15,000	12,000	15,000	15,000
Fertilizer	6,000	6,000	6,000	6,000	6,000
Herbicide	4,500	4,500	4,500	4,500	4,500
Pesticide	12,500	12,500	12,500	12,500	12,500
Labour					
Ridging	25,000	25,000	25,000	25,000	25,000
Herbicide spraying	6,000	6,000	6,000	6,000	6,000
Pesticide spraying	22,500	22,500	22,500	22,500	22,500
Weeding	30,000	30,000	30,000	30,000	30,000
Fertilizer application	10,000	10,000	10,000	10,000	10,000
Harvesting	15,000	20,000	15,000	10,000	15,000
Threshing	10,000	15,000	10,000	5,000	10,000
Storage	5,000	6,000	5,000	3,000	5,000
Transportation	20,000	20,000	20,000	20,000	20,000
Total variable cost	**181,500**	**192,500**	**178,500**	**169,500**	**181,500**
Fixed cost					
Land rentage					
Total fixed cost					
Total cost	**181,500**	**192,500**	**178,500**	**169,500**	**181,500**
Revenue					
Grain yield	489,946	688831	503,531	284025.5	542,157
Stover yield	16,000	17,000	11,000	12,000	12,000
Chaff	10,000	11,500	5,500	5,500	10,700
Total revenue	**515,946**	**717,331**	**520,031**	**301,526**	**564,857**
Gross margin	334,446	524,831	341,531	132,026	383,357
NFI	334,446	524,831	341,531	132,026	383,357
Profit / cost ratio	1.842678	2.72639	1.91334	0.77891	2.11216

1=IT93K-452-1, 2=IT99K-5773-1-1, 3=TVX-3236, 4=Oloyin, 5=IT90K-76, NFI=Net financial income

Appendix J

Cumulative profitability of cowpea varieties sown in sequence in 2016

		Varieties				
Items	1	2	3	4	5	6
Variable cost			(N/ha)			
Seed	30,000	30,000	24,000	18000	30,000	30,000
Fertilizer	88,000	88,000	88,000	82000	88,000	88,000
Herbicide	9,000	9,000	9,000	7500	9,000	9,000
Pesticide	30,000	30,000	30,000	12500	30,000	25,000
Labour						
Land clearing	45000	20000	20000	20000	20000	20000
Ridging	25000	50000	50000	25000	50000	50000
Herbicide spraying	12000	12000	12000	6000	12000	12000
Pesticide spraying	54000	54000	54000	27000	54000	54000
Weeding	60000	60000	60000	45,000	60000	60000
Fertilizer application	30000	30000	30000	20000	30000	30000
Harvesting	40000	45000	35000	15000	23000	40000
Threshing	20000	25000	15000	5000	9000	23000
Storage	10000	13000	9000	3000	4200	10000
Transportation	40,000	40,000	40,000	20,000	40,000	40,000
Total variable cost	**493,000**	**506,000**	**476,000**	**306,000**	**459,200**	**491,000**
Fixed cost						
Land rentage	2000	2000	2000	2000	2000	2000
Total fixed cost	2000	2000	2000	2000	2000	2000
Total cost	**495,000**	**508,000**	**478,000**	**308,000**	**461,200**	**493,000**
Revenue						
Grain yield	980280	1355130	833710	207000	292635	1217745
Stover yield	33000	46500	35700	34000	37000	35000
Chaff	20000	21500	16500	5000	8500	20700
Total revenue	**1,033,280**	**1,423,130**	**885,910**	**246,000**	**338,135**	**1,273,445**
Gross margin	540,280	917,130	409,910	-60,000	-121,065	782,445
NFI	538,280	915,130	407,910	-62,000	-123,065	780,445
Profit / cost ratio	1.087434	1.80144	0.85337	-0.2013	-0.2668	1.58305

1=IT93K-452-1, 2=IT99K-5773-1-1, 3=TVX-3236, 4= Kanannado, 5=Oloyin, 6=IT90K-76, NFI=Net financial income

Appendix K

Cumulative profitability of cowpea varieties sown in sequence in 2017

		Varieties				
Items	1	2	3	4	5	6
Variable cost			(N/ha)			
Seed	30000	30000	24000	18,000	30000	30000
Fertilizer	88000	88000	88000	82000	88000	88000
Herbicide	9000	9000	9000	4500	9000	9000
Pesticide	30000	30000	30000	17,500	30000	30000
Labour						
Land clearing	20000	20000	20000	20000	20000	20000
Ridging	50000	50000	50000	25000	50000	50000
Herbicide spraying	12000	12000	12000	6000	12000	12000
Pesticide spraying	54000	54000	54000	31,500	54000	54000
Weeding	60000	60000	60000	45000	60000	60000
Fertilizer application	30000	30000	30000	20000	30000	30000
Harvesting	35000	40000	40000	15,000	25000	40000
Threshing	20000	25000	23000	5000	10000	23000
Storage	11000	11000	12000	3,000	6000	12000
Transportation	40,000	40,000	40,000	20,000	40,000	40,000
Total variable cost	**489,000**	**499,000**	**492,000**	**312,500**	**464,000**	**498,000**
Fixed cost						
Land rentage	2000	2000	2000	2000	2000	2000
Total fixed cost	2000	2000	2000	2000	2000	2000
Total cost	**491,000**	**501,000**	**494,000**	**314,500**	**466,000**	**500,000**
Revenue						
Grain yield	1119006	1215786	1512176	243925	513430.5	1576267
Stover yield	38000	36500	31000	30,000	31000	31000
Chaff	22000	21500	19000	5000	10500	24700
Total revenue	**1,179,006**	**1,273,786**	**1,562,176**	**278,925**	**554,931**	**1,631,967**
Gross margin	690,006	774,786	1,070,176	-33,575	90,931	1,133,967
NFI	688,006	772,786	1,068,176	-35,575	88,931	1,131,967
Profit / cost ratio	1.401234	1.54248703	2.1622996	-0.1131	0.19084	2.263934

1=IT93K-452-1, 2=IT99K-5773-1-1, 3=TVX-3236, 4= Kanannado, 5=Oloyin, 6=IT90K-76, NFI=Net financial income