CHARACTERIZATION OF SELECTED KENYAN COWPEA CULTIVARS FOR TOLERANCE TO ACIDITY

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DECLARATION

Declaration by the candidate

I hereby declare that this thesis is my original work and has not been presented for a degree award in any other university. No part of this work should be reproduced without prior permission from the author and/or University of Eldoret.

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This thesis is dedicated in memory of my beloved father, Mr. Erastus Sang and my mother, Mrs. Marcella Sang and also with love and gratitude to all family members.

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ABSTRACT

Acid soils are normally deficient in available phosphorus and have high levels of soluble aluminium. Crops vary in sensitivity to acidity and associated stresses. Kenyan varieties of cowpea have not been tested for tolerance to acidity and phosphorus deficiency. This study was conducted to assess the response of selected cowpea cultivars to acidity and aluminium stress under laboratory and field conditions. A total of nine cowpea cultivars namely UOE-COWPEA-1, UOE-COWPEA-2, UOE-COWPEA-3, UOE-COWPEA-4, UOE-COWPEA-5, KENKUNDE-1, K-80, M-66 and KVU-27-1, were screened in culture solution containing 0 µM and 185 µM $AlCl_3$ at a pH of 4.3 and four which showed a greater ability to modify the culture solution pH and a higher relative net root length were further evaluated in the field. pH values of the culture solution were taken daily for a period of six days without adjustment. Root and shoot lengths were measured and recorded after six days. The field experiment were laid out in RCBD, where the two main plots were either limed (4 t/ha) or not limed. The treatments were phosphorus (TSP) (0.06 t/ha), lime, phosphorus plus lime and control (-P, -L) and four cowpea cultivars; UOE-COWPEA-2, KVU 27-1, K-80 and KENKUNDE-1 were grown for a duration of four months. Soil samples were analyzed prior to and after planting. Plant height, total number of leaves per plant, total number of branches per plant, leaf area per plant, plant biomass, number of pods per plant, pod length, number of seeds per pod, 100 seed weight and seed yield per plant were assessed using standard procedures. The nine cowpea cultivars screened increase the pH of the culture solution as the days progressed. The increase in the culture solution pH could be due to cowpea cultivars secreting chelating agents that reduce H^+ thereby increasing the culture solution pH and reducing Al toxicity. UOE-COWPEA-4 cultivar grown at 185 µM Al was the most tolerant with higher relative net root length (75.6%) while UOE-COWPEA-1 cultivar was the most Al sensitve (63.7%). Lime increased the soil pH from 5.23 to 6.37 while both P+L increased soil P to the maximum of 28.93 mg/kg with K-80 cultivar. UOE-COWPEA-2 produced the greatest number of leaves (52) and number of pods per plant (59) while K-80 yielded the highest total seed weight per plant (42g) with KENKUNDE-1 recording the greatest number of seeds per pod (12) all under phosphorus plus lime treatment. UOE-COWPEA-2, UOE-COWPEA-4, UOE-COWPEA-5, K-80 and KEN-KUNDE-1 cultivars that showed greater promise in modifying the pH, superior growth and yield attributes, could be grown in acidic soils. The cultivars can be tested further to establish their stability in alleviating acidity.

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LIST OF ACRONYMS AND ABBREVIATIONS

N- Nitrogen

NRL- Net root length

RNRL- Relative net root length

BNF- Biological nitrogen fixation

H- Hydrogen

Al- Aluminium

Fe- Iron

P- Phosphorous

pH- Potential hydrogen

Kg- Kilogram

Ha- Hectares

K- Potassium

Mg- Magnesium

Ca- Calcium

CEC- Cation exchange capacity

Mn- Manganese

Al^{3+}- Aluminium ion

Mn^{2+}- Manganese ion

Mo- Molybednum

Na- Sodium

NH_4^+- Ammonium ion

NO_3^- Nitrate

OH^-- Hydroxide

RCBD- Randomized complete block design

TSP- Triple super phosphate

DAP- Days after Planting

LA- Leaf Area

ANOVA- Analysis of variance

CHAPTER ONE

INTRODUCTION

1.1 Background Information

Cowpea (*Vigna unguiculata* L. Walp) is an indigenous African grain legume globally cultivated in tropical and subtropical Africa (Adeyemi *et al.,* 2020). Cowpea is an important source of dietary protein, vitamins, minerals and roughage to millions of people in rural and urban settings in Africa (Asiwe *et al.*, 2020), used as vegetables, grains and to lesser extent as a fodder crop (Nkhoma *et al*., 2020). Several products such as tender green pods and leaves of cowpea are consumed as fresh green vegetables (Gerrarro *et al.,* 2019), which can also be either fermented or sun-dried (Owade *et al*., 2020). Cowpea green leaves and mature dry grains contain 27 to 43% and 21 to 33% of protein respectively (Ddamulira *et al*., 2015; Carvalho *et al.,* 2017). The leaves are also richer in vitamins, macro and micronutrients, flavonoids, antioxidants, ß-carotene, fatty acids (1.1-3.0%), essential amino acids, lysine and tryptophan, carbohydrates (56.0-65.7%), and dietary fibre (1.7-4.5%) when compared to cereals (Gondwe *et al*., 2019; Owade *et al*., 2020). Cowpea is therefore, among the most suitable African indigenous crop in the provision of a healthy balanced diet and addressing nutritional deficiencies among the resource-constrained people (Goncalves *et al*., 2016).

Further, cowpea earns income to peasant farmers in rural areas through the sale of leaves and seeds (Kebedo and Bekeko, 2020; Tolba *et al*., 2023). It also improves and sustains the fertility of acidic soils (Ajayi *et al*., 2018), by its ability to fix atmospheric nitrogen (N) to the soil through symbiotic nitrogen fixation (BNF) with *Bradyrhizobium spp* (Namatsheve *et al.,* 2020; Asiwe and maimela, 2021). Hence, it reduces the reliance by farmers on expensive commercially produced fertilizers (Sun *et al*., 2019). Cowpea is mostly intercropped with cereals such as maize or sorghum due to its shade tolerance characteristics (Diana *et al*., 2023) or planted in rotation with cereals and other crops to boost soil fertility and add nutrients to degraded soil through nitrogen fixing property (Asiwe and maimela, 2021), thus, increasing cereal crop yields. Cowpea also is used as green manure (Binacchi *et al*., 2022) and improves soil properties which enhances weed control, soil cover, and protection from soil erosion.

Cowpea's deep taproot system and short maturity period make it drought tolerant and adaptable to acidic soils (Mekonnen *et al*., 2022) and low rainfall conditions where many other crops fail (Carvalho *et al*., 2017). However, best yields are obtained in fertile soils that drain well and have an ideal pH of 5.5 - 6.5 (Mogale *et al*., 2023; Singh *et al*., 2023). Despite the cowpea benefits as source of food, its production is limited by abiotic factors such as acidic soils and aluminium toxicity (Bolarinwo *et al.*, 2021). Most Kenyan soils are infertile and usually acidic characterized with high levels of hydrogen (H), aluminium (Al) and iron (Fe) ions (Keino *et al*., 2015). Soils with a pH below 5.0 are acidic and limit leguminous plants' symbiosis with the bacterium and the formation and functioning of nodules (Sankar *et al*., 2021).

Acidity reduces the availability of N and P nutrients which are responsible for cowpea production (Gurmessa, 2021). Deficiency of N and P in the soil reduces photosynthesis and early root development, low microbial populations and poor nitrogen fixation in crops, leading to low yields (Amba *et al*., 2011). Phosphorus is important for establishment of nodulation and N_2 fixation in legumes and its deficiency in the soil causes poor nodulation and poor nitrogen fixation reducing the capability of the strains of rhizobia to extract and incorporate P from the external environment (Desta *et al*., 2015; Wolde-meskel *et al*., 2018). Therefore, the availability of P in the soil is important in nodulation, nitrogen fixation and nodule activity.

Addressing soil acidity and aluminium toxicity is important to alleviate N and P deficiency in the soil, but inorganic fertilizers are generally not affordable to small scale farmers to apply them at the proper rates. Under favorable conditions, biological nitrogen fixation (BNF) by legume crops like cowpea in cropping systems is one of the affordable options to increase not only soil N levels, but also improvement of soil productivity (Rego *et al*., 2015). Biological nitrogen fixation (BNF) is a cheaper means of improving soil fertility and productivity (Horn and Shimelis, 2020). The use of legumes in cropping systems and associated BNF contributes to continuous supply of soil nitrogent content (Kebede, 2020). Cowpea can fix about 337 Kg N/ha (Yahaya, 2019) through BNF in a symbiotic association with *Bradyrhizobium spp* contributing to soil fertility (Horn and Shimelis, 2020), thus BNF is an important process for improving and maintaining of soil fertility however, it is limited in soils with low P availability because of the high P requirement for BNF (Nijar *et al.*, 2016). Nitrogen fixation associated with bacteria depends on soil phosphorus availability (Vardien *et al.*, 2014). Legumes require P during BNF in the form of ATP which is the source of energy (Schulze *et al*., 2006). It is also essential for nodule activity, growth and functioning on legumes in signal transduction and membrane biosynthesis (Divito and sadras, 2014). Phosphorus deficiency reduces nitrogen accumulation and alters photosynthate allocation during BNF in cowpea legumes (Isidra- Arellano *et al.*, 2018). The amount of N fixed, depends on the type of legume, legume genotype and the microsymbiont (Stagnari *et al.*, 2017). Yusuf *et al*., (2006) also showed variability in BNF ability of legume plants as a result of the difference in genotype, environment like soil fertility and crop management including spacing and weeding (Woomer *et al*., 2014; Gyogluu *et al.*, 2016). Although cowpea is tolerant to acidic soils, its mechanism of tolerance is not well documented and its yield has remained low in most tropical regions including Kenya.

1.2 Statement of the problem

Soil acidity associated with toxic levels of aluminium (Al) and deficiency of phosphorus (P) are major constraints to cowpea productivity in tropical Africa, including Kenya. Although cowpea is tolerant to stress, Kenyan cultivars have not been systematically characterized for their ability to tolerate aluminium toxicity and low P. Identifying acid-tolerant cowpea cultivars would provide farmers with cost-effective alternatives to fertilizers for maintaining yield in acidic soils.

1.3 Justification of the study

Although cowpea is widely grown in Kenya, there has been no systematic evaluation of local cultivars for tolerance to acidic soils conditions that dominate most of the arable lands in Kenya. Identifying and promoting cultivars with inherent tolerance will provide a low-cost adaptation strategy for smallholder farmers who cannot afford lime and fertilizer inputs. This work aligns with Kenya’s vision 2030 goals and climate-smart agriculture strategies by contributing to improved food security and resilience under soil- related production constraints.

1.4 Objectives of the Study

1.4.1 General objective

The overall objective of this study was to characterize Kenyan cowpea cultivars adapted to acidic soils that could be used to improve productivity of acid soils.

1.4.2 Specific objectives

i. To determine the effect of growth of selected cowpea cultivars on the culture solution pH supplented with aluminium.
ii. To evaluate the effect of selected cowpea cultivars on pH and soil nutrient profiles when grown on acidic soils.
iii. To determine the growth and yield responses of selected cowpea cultivars to lime and phosphorus application.

1.5 Research Hypotheses

1. Cowpea cultivars do not raise the pH of the culture solution supplemented with aluminium
2. Selected cowpea cultivars do not affect soil chemical properties
3. There exist genotypic differences in growth and production of cowpea cultivars in acidic soils

CHAPTER TWO

LITERATURE REVIEW

2.1 Origin and Distribution of Cowpea

Cowpea is an African native legume widely cultivated and consumed in tropical and sub-tropical areas of Africa, Latin America, South East Asia and in the Southern United States (Adeyemi *et al*., 2020). Although, the precise origin of cultivated cowpea is not known, it is presumed to have originated from Africa before 2500 BCE (Herniter *et al*., 2020) and was domesticated in Southern Africa; later it was spread to East and West Africa and Asia (Agbogidi, 2010). The Southern African region and the semi-arid west and central Africa are reportedly considered the center of diversity of *Vigna unguiculata* (Rivas *et al.,* 2016) where people in the areas of center of diversity consume cowpea and other pulses as supplementary to their daily diet. Currently, cowpea is among the most commonly cultivated lowland pulse crops in Kenya for food (Habte *et al*., 2018).

2.2 Botany and Ecology of Cowpea

Cowpea (*Vigna unguiculata* (L.) Walp) is a dicotyledonae with 2n=22 chromosomes (Lonardi *et al*., 2019). Cowpea belongs to the family fabaceae and sub-family faboidea (Singh, 2020). The crop is characterised by a variety of plant traits; a plant height of 48–61 cm when grown under favourable conditions (Singh *et al*., 2003); leaves and leaf types are oppositely arranged on the stem which are trifoliate and usually dark green, shiny and show considerable variation in size and are linear-lanceolate to ovate in shape. Cowpea cultivars have thicker stems which are indeterminate with branch apices. The stems may be smooth, striate or hairy with purple shades. Cowpea have also well developed long taproot root system with many lateral roots in the soil surface (Madamba *et al.,* 2006).

Cowpea have varying growth habits which can be erect, semi-erect, trailing or climbing to bushy type depending on the cowpea genotype. Their inflorescence is arranged in racemose or intermediate at the distal ends of long peduncles that arise from the leaf axils. Flowers are large, conspicuous and brightly coloured, which may be purple, pink, white, blue, or yellow. Two to three pods per peduncle are common and often 4 or more pods can be carried on a single peduncle when growing conditions are favourable. The presence of a long peduncle is a distinguishing feature of cowpea and a characteristic which facilitates hand harvesting. Pods vary in size, shape, color and texture which may be long, smooth, cylindrical and curved or straight. The number of seeds per pod varies between 8 to 20 and the seed weight weighing between 8-32 grams. Cowpea seeds have a variety of seed-coat colours and textures and mostly pendant but some erect hilums (Choudhary *et al*., 2023).

Cowpea is grown on a wide range of soils but shows preference for sandy soils, which tend to be less restrictive on root growth (Vanlauwe *et al*., 2019). It is well adapted to most environmental stresses and drought as compared to other crop species (Tankari *et al.*, 2021). Cowpea is tolerant to low fertility (Alemu *et al*., 2016) and acidic soils of a wide range of pH (4.5- 5.5) as compared to other grain legumes (Ddamulira *et al*., 2015).

2.3. Economic Importance of Cowpeas

Cowpea plays a significant role in nutrition, income generation and food security to millions of people in urban and rural areas. The crop is economically significant as a source of income for many small scale farmers in rural areas (Tolba *et al*., 2023; Kebede and Bekeko, 2020). Cowpea products such as fresh leaves, green pods and protein rich green seeds are consumed as vegetables (Binacchi *et al*., 2022). Mature seeds are usually consumed as a protein source in many households (Santos *et al*., 2020) while the remaining parts are used as animal fodder (Kebede and Bekeko, 2020), green manure (Binacchi *et al*., 2022) and as cover crops (Mwenda *et al*., 2023). The dual-purpose cowpea cultivars are utilised for both grain and fodder to improve the health status of both humans and domestic animals in semi-arid sub-Saharan Africa (Akplo *et al*., 2023; Faye *et al*., 2024). As a green manure, cowpea is capable of fixing free atmospheric nitrogen into ammonia through biological nitrogen fixation hence improving soil fertility by providing soil nitrogen (Owade *et al*., 2020). In intercropping production systems, the spreading intermediate type of cowpea suppresses the germination of weeds and prevents soil erosion (Das *et al*., 2018). Additionally, cowpea builds up soil organic matter and fixes organic carbon and nitrogen into the soil when included in crop rotations thus improving soil fertility and water retention capacities (Beker *et al*., 2023; Faye *et al*., 2024). Cowpea also improves biological soil fertility and crop productivity through interaction with rhizobia bacteria (Wekesa *et al.*, 2022). Cowpea production therefore, has played a significant role in improving food security, living standards and malnutrition in sub-saharan Africa and contributes significantly to economic productivity and environmental sustainability in Africa (Cardona- Ayala *et al.,* 2020).

2.4 Cowpea production

Global estimate shows that the total production of cowpea in Africa was 9,475,644 tonnes in 2022, mostly in sub-Saharan Africa (Nigeria 4,133 t, 708 t, Niger 2,865,884 t, Burkina Faso 829,204 t, Ghana 319,960 t, and Mali 250,317 t) according to FAOSTAT (2024), with Nigeria being the world's largest producer of cowpea production. The major cowpea producing East African countries are Kenya, Tanzania, South Sudan and Uganda (Owade *et al*., 2020). Kenya produces about 246,870 tonnes on 298,120 hectares, with 85% in the arid and semi-arid lands (Muniu, 2023) with the key cowpea production regions being Kisii, Kitui, Taita Taveta, Migori, Bungoma, Kakamega, Kwale, Makueni, Machakos, Tharaka Nithi and Kilifi (Binacchi *et al*., 2022; Mwenda *et al*., 2023). The average yield of cowpea in Kenya remains very low at 828.1 kg/ha compared to potential yield of 1500 to 3000 kg/ha under optimal conditions (FAOSTAT, 2021). Low production of cowpea is contributed by infertile soils which are acidic characterized with high levels of aluminium and nutrient deficiencies (Krah *et al*., 2019).

2.5 Factors affecting soil fertility

Soil fertility is the capacity of soil to supply essential nutrients for plant growth and optimize crop yield over extended period of time (Vanlauwe *et al.*, 2017). Low soil fertility especially soil acidity associated with aluminium toxicities and nutrient deficiencies such as N and P due to continous cultivation, with little or no soil nutrient replenishment (Vanlauwe *et al*., 2017) are the major factors limiting crop productivity in most farming systems. Though, legumes play a significant role in sustainable agriculture through their ability to improve soil fertility and health as they fix atmospheric nitrogen and add organic matter (Korir *et al.*, 2017), its production in tropical and sub-tropical regions is low due to low soil fertility which affect BNF. Cultivation of leguminous crops such as cowpea can be an alternative source of N as it is renewable and ecofriendly (Dwivedi *et al.*, 2015).

2.5.1 Soil acidity and aluminium (Al) toxicity

Soil acidity is a major limiting factor to cowpea growth and production. Approximately, 40% of the world's arable land are acidic in many sub- tropical and tropical areas (Phukunkamkaew *et al*., 2021) and more than 50% of the world’s potentially arable lands (Asfawu *et al.*, 2024). Soil acidity affects many physical, chemical reactions, and biological processes within soil, which controls the nutrient availabilty, the toxicity of metal elements, and the metabolic functions of plants, microorganisms, and soil fauna (Hartemink and Barrow, 2023). Nutrients such as calcium, magnesium and potassium are easily leached from acidic soils, leading to decreased soil fertility (Li *et al*., 2020). Acidic soils tend to fix phosphorus resulting to low bioavailabilty which is important for cowpea growth and yield leading to long term reliance on phosphorus application to ensure crop yield (Wang X *et al*., 2023). Soil acidity induces aluminum (Al) toxicity and increases bioavailability of toxic metals which affects crop yields and quality (Rahman and Upadhyaya, 2021; Ur Rahman *et al*., 2024). Low pH inhibits enzyme activity and disturbs soil microbial community structure and function which finally changes carbon, nitrogen, phosphorus and sulphur cycles in soil (Lee *et al*., 2022; Hu e*t al*., 2024). It also affects the symbiotic relationship between rhizobia and legume crops including cowpea resulting to reduced nodule formation, development and nitrogen fixation thus limiting plant growth (Namatsheve *et al*., 2020).

Aluminium is ranked first among metals and the third most abundant element in the earth's crust (Shetty *et al.*,2021). Aluminium is non-phytotoxic when the soil pH is neutral or slightly acidic since it exists in the insoluble oxides or aluminosilicate. However, the phytotoxic form occurs mainly in soils with pH values below 5.0, resulting to the release of Al^{3+} (Casierra-Posada *et al*., 2021). The phytotoxic species becomes soluble in the soil as acidity increases and can negatively affect plant growth and development (Casierra-Posada *et al*., 2021 and Wei *et al*., 2024). Aluminium toxicity is a major factor limiting crop productivity on acid soils (Kushwala *et al*., 2017). The phytotoxic- aluminium (Al^{3+}) inhibits root elongation in Al- sensitive plants due to its quick inhibition of cell division and cell expansion in root meristems (Phukunkamkaew *et al*., 2021). Consequently, this limits root extension growth thus affects the ability of plants to absorb water and nutrients (Alemu *et al.*, 2022) leading to poor growth and substantial decrease in yield (Du *et al*., 2020). Al- toxicity coupled with low pH is a major factor limiting crop productivity (Gurmessa *et al*., 2021).

Manganese toxicity is the second most important growth limiting factor after Al for plants grown in acidic soil (Zhao *et al.*, 2017). Manganese toxicity in the soil disrupt many physiological processes in plant roots such as enzyme activity and metabolic functions (Pradeep *et al*., 2020). Excess of Mn in the soil result in oxidative stress, which harm root cell membranes and contribute to the development of lesions (Zhou *et al*., 2022). The lesions weaken the root’s structural integrity and hinder their capacity to efficiently absorb water and nutrients. Mn poisoning which arises in acidic soils, results to necrosis and chlorosis in plant tissues, hence reducing plant vigor and yield (Ofoe *et al*., 2023). In addition to Al and Mn toxicities, nutrient deficiency especially phosphorus deficiency is another major constraint in acidic soils, often acting synergistically with Al toxicity.

2.5.2 Nutrient deficiency

Soil acidity greatly affects plant development and yield, since the essential nutrients becomes limited under acidic environments. Many agricultural soils in the tropics and sub tropics are low in both total nitrogen (N) and available phosphorus (P) which are essential plant nutrients due to soil acidity (Olego *et al*., 2022) including Uasin Gishu soils in Kenya. Depletion of soil nutrients is caused by continuous cultivation, with little or no soil nutrient replenishment (Muui *et al.*, 2013). Legumes such as cowpea require adequate supply of major nutrients including nitrogen, phosphorus, potassium, sulphur, calcium, manganese

(Magani and Kuchinda, 2009). The lower nutrient's availability occurs due to the tendency of toxic Al and Fe to fix P in the soil into insoluble compounds (Warke and Wakgari, 2024) rendering the availabilty of P for plant absorption to decrease. Soil acidity limits the availability of P in the soil restricting plant growth and crop yield (Gurmessa *et al*., 2021). Under acidic conditions also, other nutrient elements such as Ca, Mg and K are easily leached in the soil leading to their low concentaction in the soil (Gilbert *et al*., 2007).

Nitrogen is one of the essential nutrients and plays a key role in promoting plant growth and a key component of proteins, nucleic acids and phospholipids in the metabolism of essential substances within the plant (Kwena *et al*., 2019). Nitrogen is also an important component of chlorophyll, linked to photosynthesis, is a constituent of proteins, enzymes, chlorophyll, and growth regulators to plants and its deficiency causes reduced growth, leaf yellowing, reduced branching and smaller trifoliate leaves in legumes (Onmwonga *et al.*, 2010).

Phosphorus (P) is a major essential nutrient in plant growth (Magadlela *et al.*, 2016; Perez-Fernandez *et al*., 2017). Phosphorus is important in cowpea production because it plays a significant role in physiological processes such as stimulating growth, initiation of nodules formation and the efficiency of rhizobium-legume symbiosis (Yadav *et al*., 2017). It also helps in establishing seedlings faster and hastens maturity and quality of pulse crops (Mohammed *et al*., 2021). Phosphorus is also important in metabolic processes including energy generation, respiration, membrane synthesis, photosynthesis, activation and inactivation of enzymes, signaling and carbohydrate metabolism (Zhang *et al*., 2014). Its deficiency limits the formation of nodules in the legume roots affecting nitrogen fixation and consequently the seed yield potential of legume crops, thus reducing crop production

(Asuming-Brempong *et al.*, 2013). Cultivation of legumes like cowpea in cropping systems reduces the use reliance by farmers on inorganic fertilizers and would act as an alternative source of nutrients to the soil due to its ability to convert and fix appreciable amount of atmospheric nitrogen into the soil (Mndzebele *et al*., 2020; Mohammed *et al*., 2022).

2.5.3 Native rhizobia populations

Rhizobia is a gram - negative rhizobacteria among the micro-organisms present in the cowpea rhizosphere and establishes a symbiotic association with cowpea (Hamza and Alebejo, 2017). Native rhizobia are important in legume BNF and play a significant role in growth and yield of most leguminous crops by supplying N and relieve the use of nitrogenous fertilizers in these crops (Deb *et al.*, 2015). Soil acidity has an impact on microbial activity and soil biodiversity in which both play a critical role in preserving soil fertility and structure. Beneficial microorganisms, such as N-fixing bacteria and mycorrhizal fungi do flourish in environments that are neither too acidic nor too alkaline (Sanchez- Galindo *et al*., 2022). These organisms have a high level of sensitivity to changes in soil pH and are hindered in their activity and variety by acidic environments (Xiao *et al.*, 2020). When the pH of the soil decreases, the activity and number of these microorganisms decrease which in turn, reduces their capacity to promote plant development (Choma *et al*., 2020). The decrease in bacterial populations decreases bacterial activity hindering the breakdown process, resulting in the buildup of organic materials in the soil (Yang *et al.*, 2021). Soil acidity therefore, affects rhizobia growth, survival and their abundance in the soil compromising symbiotic efficiency, nodulation and nitrogen fixation (Yang *et al.*, 2021). Yakubu *et al.* (2010) reported that low phosphorus content of the soil restricts rhizobia population and legume root development, which in turn, can lower the N_2 fixing potential. The rhizobacteria in the soil are able to solubilize sparingly soluble phosphates by releasing chelating organic acids (Vessey *et al*., 2004). The performance of cowpea in the soil depends on the rhizospheric characteristics where it forms a beneficial association with micro-organisms present in the rhizosphere (Abdel-Fattah *et al*., 2016).

Higher rhizobia populations are supported in soils with higher organic matter content (Ngokota *et al.*, 2008). Kimiti and Odee (2010) also reported that native rhizobia population counts also varies with the time of soil sampling, nutrient input and cowpea variety used. Cowpea cultivation can enhance native soil rhizobia populations such that other legumes nodulated by *Bradyrhizobium* spp like soybean, can also benefit since cowpea cultivation is known to stimulate proliferation of rhizobia in a field (Mulongoy and Ayanawa, 1985). The ability of cowpea to fix nitrogen in the soil therefore, is influenced by the population of rhizobia in the soil, effectiveness of the rhizobia and availability of phosphorus; where these conditions are not meet, nitrogen fixation may not be optimal thereby reducing yield (Fening and Danso, 2002). Therefore, the higher the population of rhizobia, the more chances for nodule infection hence higher biological nitrogen fixation capacity.

2.6 Cultivar variation in response to Acidity and Aluminium stress

Tolerance to soil acidity, high Al levels and P deficiency varies across plant species and between cultivars of the same species (Fukuda *et a*l., 2007). The exact mechanisms by which certain plants tolerate high Al levels, soil acidity and P deficiency is still unknown (Liu *et al*., 2022). Several hypotheses have been suggested that acid tolerant plants have adaptive mechanisms that enable them to survive in high Al levels and acidic P deficient soils. Tolerant plant species prevent excess Al ions absorption from entering the root apical cells or detoxify aluminium ions once it has been absorbed (Garzon *et al*., 2011). Tolerant plant species also exude organic acids anions from their roots such as malate, citrate and oxalate that chelate Al^{3+} ions (Zhang *et al*., 2021). Other plants have high root elongation hence uptake of water and nutrients is enhanced.

Research done shows that, cowpea employs certain mechanisms of tolerating Al toxicity and phosphorus deficiency like genotypic variability for root traits where cowpea plants form an extensive root system by producing a higher total root length, which is also an important adaptive trait in cowpea cultivars that enhances phosphorus uptake from low P soils (Zhu and Lynch, 2004). Rothe (2014) reported that a large surface area in cowpea genotypes roots is responsible for tolerance to low soil P. The association formed between soil mycorrhizal fungi and cowpea in some genotypes is also known to enhance uptake of nutrients (Saidou *et al.*, 2012) in soils with low fertility. Martin *et al.*, (2007), reported that aluminium induced the secretion of organic acids anions such as malate, citrate and oxalate from cowpea roots. These root exudates maintain a beneficial relationship between plants and microorganisms by regulating the soil microbial community in the rhizosphere (Zhang *et al.*, 2021). The organic acids are also involved in Al detoxification and P acquisition of plants and alteration in rhizosphere pH (Lopez-Bucio *et al*., 2000).

Aluminium toxicity and low P in soils can be corrected by applying lime (CaO) and P fertilizer to the soils. Application of lime raises the soil pH thus mitigating the negative effects of aluminum ions that are highly soluble in acidic soils and increases available P leading to better root development and nutrient uptake thus allowing cowpea to thrive in environments that would otherwise hinder growth. However, liming and P fertilizer are generally expensive to small scale farmers to apply them at the proper rates and only limited to the sub surface soil due to low solubilty of lime (Anderson and Bell, 2019). Despite extensive research on soil amendments to improve acidic soils, cultivar tolerance remains underexplored in Kenya, where resource-poor farmers cannot always afford soil amendments. Therefore, this study aims to characterize selected Kenyan cowpea cultivars that can tolerate acidic soils associated with Al toxicities and P deficiency and which would assist in improving the growth and production of cowpea hence contributing to food nutrition and security in the country.

CHAPTER THREE

MATERIALS AND METHODS

3.1 Description of Site of Study

Field work was carried out at the University of Eldoret farm, situated 9 km North-East of Eldoret town in Uasin Gishu County, Kenya. The University is located at a Latitude of 0^{0}35’0.59’’N and a Longitude of 35^{0}18’34.6’’E at an altitude of 2139 m above sea level. The area receives an average rainfall of between 900 mm – 1300 mm per annum. The mean temperature is 17.5^{o}C with maximum of 25^{o}C in February and a minimum of 10^{o}C in July (Jaetzold *et al.,* 2006). The laboratory experiments were carried out at the University of Eldoret Biological Science and Soil Science laboratories.

3.2 Cowpea Seed Materials

A total of nine cowpea cultivars commonly grown in Kenya as described in Table 3.1 below were screened for low pH and aluminium toxicity in the laboratory and four cowpea cultivars namely UOE-COWPEA-2, KVU 27-1, K-80 and KENKUNDE-1 which showed a greater ability to modify the culture solution pH and a higher relative net root length were further evaluated in the field.

Table 3.1. The description of the cowpea cultivars used in the study

Cultivar	Source	Seed colour	Growth habit	100 Seed weight (g)
UOE-COWPEA-1	Eldoret market	Dark brown	Determinate	12.61
UOE-COWPEA-2	Bumala market-Busia county	Brown with white spots	Indeterminate	12.42
UOE-COWPEA-3	Eldoret market	White with black eyes	Determinate	11.48
KENKUNDE-1	Kenya Seed Company	Red brown	Indeterminate	12.18
UOE-COWPEA-4	Sega market- siaya	Black with white eyes	Determinate	7.94
Katumani-80	Kenya Agricultural and Livestock Research Organisation	Creamy brown	Determinate	12.32
Machakos-66	Kenya Agricultural and Livestock Research Organisation	Creamy brown	Determinate	12.80
UOE-COWPEA-5	Bumala market- Busia	Creamy brown	Determinate	14.49
KVU 27-1	Kenya Agricultural and Livestock Research Organisation	Maroon	Determinate	12.42

3.3 Screening Cowpea Cultivars for Tolerance to Aluminium Toxicity and Low pH Levels in Nutrient Solution Culture

3.3.1 Screening of cowpea cultivars to acidity and Al toxicity

Cowpea seeds to be screened were sorted, soaked in soapy sterile distilled water for five minutes and surface sterilized with 0.1% sodium hypochlorite for 8 minutes. Thereafter, the seeds were rinsed eight times with sterilized distilled water to ensure all the traces of chloride were removed. The sterilized seeds of each cultivar were separately placed in between sterilized paper towels moistened with sterile distilled water in labeled petri dishes. The seeds were then pre-germinated in the dark in an incubator set at 26^{o}C for three days.

For each cowpea cultivar, fifteen pre-germinated healthy seedlings with root length between 1.5 cm to 2.5 cm were transferred into a constantly aerated three (3) liters growth trays using an aquarium air pump containing freshly prepared 1/5 Hoagland Nutrient solution adjusted to a pH of 4.3 and supplemented with 0 µM or 185 µM $AlCl_3$. Plants were grown in a growth chamber maintained at 28/25^{0}C day/night temperature, 16-h photoperiod using fluorescent tubes, 60% relative humidity and 300 µmol/m^{2}/s light intensity for the entire experimental period. The experiment were laid out in a completely randomized design and each treatment was replicated three times. The seedlings were acclimatized for twenty-four hours in the growth medium then the initial root and shoot length of each seedling per cultivar and treatment were measured and recorded. pH measurements were taken daily for 6 days without adjustment. Root and shoot lengths were measured and number of lateral roots were counted after six days and recorded. The data collected were used to calculate growth indices: Net root length (NRL) and Relative Net root length (RNRL). The NRL was calculated as:

$$\mathrm{NRL} = \mathrm{FRL} - \mathrm{IRL} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots.\text{Equation 1}$$

Where FRL is the final root length in both Al treated and control plants and IRL is the initial root length.

RNRL was calculated as:

$$\mathrm{RNRL} = \frac{\mathrm{NRL}_{\mathrm{Al}}}{\mathrm{NRL}_{\mathrm{C}}} \text{ x } 100 \ \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots.\text{Equation 2}$$

Where NRL_{Al} is net root length in Al, and NRL_{C} is net root length in control

Fresh root and shoot harvested at the end of the sixth day were dried in an oven at 68°C for 48 hours to determine dry weight.

3.4 Field Experimental Design

The experimental field was ploughed, harrowed and divided into four blocks. Each block was subdivided into twelve plots measuring 4 m by 3 m as shown in Figure 3.1 below. The experiment was arranged in a split plot, where the first two main blocks were not limed while the last two blocks were limed using agricultural lime (CaO) at a rate of 4.8 kg/m^2. Two cowpea cultivars, UOE-COWPEA-2 and KENKUNDE-1 that showed tolerance to acidity and Al toxicity in culture solution and two other cultivars, KVU 27-1 and K-80 that were sensitive to acidity and aluminium stress were used. The treatments tested on each of the four cowpea cultivars were phosphorus (Triple super phosphate ($Ca(H_2PO_4)_2$. H_2O) application alone (+P) at a rate equivalent to 60Kg/ha, lime (+L) application alone, combination of phosphorus and lime applications (+P & +L) and control (-P, -L). The treatment combinations and cowpea cultivars were arranged in a randomized complete block design (RCBD) with three replications (Figure 3.1).

Block 1	T1					T2					T1			
	2	4	1	3		1	4	3	2		4	3	2	1
Block 2	T2					T1					T2			
	2	1	3	4		2	1	4	3		1	2	3	4
Block 3	T3					T4					T3			
	3	4	2	1		4	3	2	1		3	1	4	2
Block 4	T4					T3					T4			
	1	2	3	4		1	3	4	2		4	1	2	3

Figure 3.1. Experimental layout showing treatments and cowpea cultivar combinations

Treatments: T1= Control T2=TSP T3=Lime T4=TSP+ Lime

Cowpea cultivars: 1. UOE-COWPEA-2 2. KVU 27-1 3. K-80 4. KENKUNDE-1

3.5 Soil Sampling and Analysis

3.5.1 Field soil sampling

Soil sampling at the University farm was done following a procedure described by Barker and Pilbeam (2007). Surface litter was removed and the soil collected at a depth of 0-20cm from five (5) random spots in a zigzag sampling approach per plot. The samples were air-dried in the greenhouse before laboratory analysis. The samples were mixed thoroughly and a composite from each individual plot prepared, sub-sampled and kept in labeled paper bags. The dried samples were ground then sieved through a 60 mm and 20 mm mesh. They were then subjected to chemical analysis at the Soil Science and Biological Science laboratories. Soil samples were collected before planting, at flowering and at crop physiological maturity stages from the rhizosphere soil for pH assessment and nutrient composition.

3.5.2 Determination of soil pH

Soil pH was determined with a calibrated glass electrode and pH meter (Hanna instrument HI99121 model) in a 1:2.5 (w/v) soil: water suspension as described by Okalebo *et al.*, (2002). Fifty (50) ml distilled water was added to 20 g of air -dry soil. The mixture of soil – water suspension was stirred thoroughly for ten minutes and allowed to settle for 30 minutes. After calibrating the pH meter with buffer solution at pH 4.0 and 7.0, the pH of the supernatant was measured and recorded.

3.5.3 Digestion of soil samples for chemical analysis

The determination of total nitrogen and phosphorus in soils was done after wet digestion (Okalebo *et al*., 2002). Wet digestion reagents were prepared from analytical grade chemicals.

Air dried soil samples were passed through 60 mm sieve and weighed to obtain 0.3 g of sub-samples. The weighed 0.3 g sub-samples were placed in dry clean digestion tubes and mixed with 4.4 ml of digestion reagent. Blanks containing 4.4 ml of the digestion reagent were run as a check. The mixture was heated to 70^0C for one hour, followed by 360^0C in a Kjelderm digester (CSB 204-Gerhardt). The contents were allowed to cool and topped to 50 ml with distilled water.

3.5.4 Determination of total nitrogen

The total nitrogen in soil samples was determined using ammonium distillation and titration method (Okalebo *et al.,* 2002). Steam distillation apparatus was set up and steam was passed through for 30 minutes. Steam blank was checked by collecting 50 ml distillate and titrated with N/140 HCl.

Ten (10) ml aliquot of the final sample digest was transferred into the reaction chamber and mixed with 10 ml of 40% NaOH solution. Distillation was carried out and the distillate collected in 5 ml of 1M boric acid containing four drops of mixed indicator. The distillation continued for about two minutes until the indicator turned green.

The distillate was titrated with N/140 HCl to a pink end point and the titre was then recorded, T. A blank distillation and titration were also carried out. The percentage nitrogen (N) in the soil sample was calculated using the equation described by Leeghood (1993);

$$\%N = \frac{T*0.1}{W} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \text{Equation 3}$$

Where;

T = corrected ml of N/140 HCl (titrant).

W=weight (g) of soil sample used

3.5.5 Determination of available phosphorus

Three (3) grams of fine (2mm) air dry soil was weighed into 250 ml clean polythene bottle and mixed with 50 ml of the Olsen extracting solution (1.0 M Sodium hydrogen carbonate $NaHCO_3$ at pH 8.5). It was stoppered well and shaken on a reciprocal shaker for 30 minutes. The suspension was filtered through the Whatman No.42 filter paper, and the filtrate was used for the colorimetric P measurements.

Ten (10) ml of P standard solutions or the trial filtrates were pipetted into 50 ml volumetric flasks and mixed with 5 ml of 0.8 M boric acid. Then 10 ml of ascorbic acid reagent was added and the solution diluted to 50 ml with distilled water to form a blank. The contents were stoppered, mixed well and allowed to stand for 1 hour. The absorbance of the solution was measured with a spectrophotometer maker (21D spectrophotometer, Milton Roy) at

880 nm. Standard absorbance readings (Appendix 3) were used to prepare a standard curve, which was used to convert sample absorbance into equivalent concentration (ppm). The amount of phosphorous in the samples was calculated by subtracting the blank from the obtained result and multiplying by the dilution factor.

$$\mathrm{P(mg/\,kg)} = \frac{(\mathrm{a\text{-}b})^{*}\mathrm{v}^{*}\mathrm{f}^{*}1000}{1000^{*}\mathrm{W}} \text{……………………………………….Equation 4}$$

where a = the concentration of P in the sample; b = the concentration of P in the blank; v = volume of the extract solution; f = dilution factor; w = weight of the sample in g.

3.5.6 Determination of soil organic carbon

The percentage organic carbon (% C) in the soil was determined using the Nelson and Somers (1975) oxidation method. A sample of 0.3 g of dried ground soil was weighed into an Erlenmeyer flask. Ten (10) ml of 0.167M potassium dichromate ($K_2Cr_2O_7$) solution and 7.5 ml of 1.0M concentrated sulphuric acid was added to the samples in the Erlenmeyer flask. The flask was then swirled to ensure full contact of the soil with the solution after which it was allowed to stand for 30 minutes to cool. The unreduced $K_2Cr_2O_7$ remaining in solution after the oxidation of the oxidizable organic material in the soil sample was titrated against 0.2M ferrous ammonium sulphate solution after adding 200ml of distilled water, 10 ml of orthophosphoric acid and 2 ml of diphenylamine indicator solution was added till colour changed from brown to green end point.

The percent organic carbon was calculated as;

$$\%\ \mathrm{C} = \frac{0.3[10-(\mathrm{XN})]*1.33}{\mathrm{W}} \text{……………………………………….Equation 5}$$

Where;

X=ml of $Fe(NH4)_2(SO4)_2$ required for the titration

N= normality of $Fe(NH4)_2(SO4)_2$

W= weight (g) of soil sample

3.5.7 Extraction of exchangeable cations in soil samples

Potassium (K), Sodium (Na) and Calcium (Ca) were determined by extraction of the soil samples with 1M ammonium acetate (NH_4OAc) solution. Five (5) grams of air dried soil was weighed into a clean plastic bottle with a stopper and 100 ml of 1 M ammonium acetate (NH_4OAc) solution pH (7) was added. The contents were shaken for 30 minutes and filtered through No.42 Whatman filter paper. The soil extract was then diluted ten (10) times to fall within measureable range of Flame photometry (FP) (Model No. EQ-855A) and Atomic absorption spectrophotometer (AAS) that was used to determine K, Na and Ca (Okalebo *et al*., 2002).

3.5.8 Determination of Magnesium

The soil extract solution in section 3.5.7.1 above, was diluted to 25-fold and two (2) ml was pipetted into a 50 ml volumetric flask. Five ml of 5000 ppm of strontium (Sr) was added and filled up to the mark with 1 M NH_4OAc extracting solution. The solution was sprayed into the flame of atomic absorption spectrophotometer. The standard working solutions were used in the calibration of the flame photometer (FP) and Atomic Absorption Spectrophotometer (ASS) (Okalebo., 2002). Standard curve for Mg^{2+} was constructed from the respective readings. The concentrations of K, Na, Ca and Mg in the soil sample expressed in mg/kg were determined as;

Mg/kg K, Na, Ca and Mg in soil= $\frac{(a-b)*v*f*1000}{1000*w}$ ……………………………….Equation 6

Where;

a=concentrations of K, Na, Ca and Mg in the sample extract

b = concentration of element in the blank

v = volume of the extract

w = weight of the soil sample in g

f = dilution factor.

3.6 Evaluation of Selected Cowpea Cultivars in Acidic Soils

3.6.1. Sowing and seedling management

Four seeds were sown per hole at a depth of 3-4 cm and with a spacing of 20 cm apart between plants and 60 cm between rows. Urea was applied as a starter dose to all the plots at the rate equivalent to 15 kg /ha of urea. The seedlings were thinned to one per hole after two weeks of planting. The experimental area was weeded regularly with a hoe before maturity to enable the plants to develop under non-competing conditions. Insect pests and fungal pathogens were chemically controlled using Duduwar by spraying at 2 weeks after seedling emergence and thereafter, at 10-day intervals following the procedure of Awe (2008).

Ten seedlings per plot were selected randomly and tagged from the four center rows and were used to assess plant height, number of branches, number of leaves and leaf area per plant at 60, 75 and 90 days after planting (DAP). Plant height was measured using a meter rule from the ground surface to the terminal bud, the number of branches and number of leaves per plant was determined by visual counting. For each trifoliate leaf, the length and broadest width of the leaflets for three randomly selected plants were measured using a ruler. Leaf length was measured from lamina tip to the point of intersection of the lamina and the petiole, along the midrib. Leaf width was measured from end to end between the widest lobes of the lamina perpendicular to the mid rib. The linear measurements were then used to estimate the average leaf area per cultivar following the model below as described by Bhatt and Chanda (2003).

LA (cm^2) = 0.88 (L +W) +0.11 ……………………………. Equation 8

Where LA = Leaf area; L= Length of the leaf midrib; W = Maximum leaf width.

Three other seedlings were randomly selected and tagged to determine plant biomass at flowering. At physiological maturity, ten plants per plot were also randomly harvested to determine the number of pods per plant, pod length, number of seeds per pod, pod dry weight, 100 seed weight and dry seed yield per plant (kg/ha).

3.6.2 Data Analysis

The data collected from the laboratory and field trials were subjected to analysis of variance (ANOVA) using Microsoft Excel 2021 and Statgraphics centurion where a P-value of less or equal to 0.05 was considered statistically significant. Separation of means was done using Tukey’s test. Data was presented in Tables and Figures.

CHAPTER FOUR

RESULTS

4.1 The Effect of Cowpea Cultivars and Al Stress on pH of Solution Culture

The cowpea cultivars growth in nutrient solution with or without aluminium induced variations in pH of nutrient culture solution. Generally, all the nine (9) cowpea cultivars increased the pH of the solution as the number of days progressed. The growth of cowpea cultivars at 0 µM Al induced a higher change in pH compared to when grown in 185 µM Al concentration (Figure 4.1).

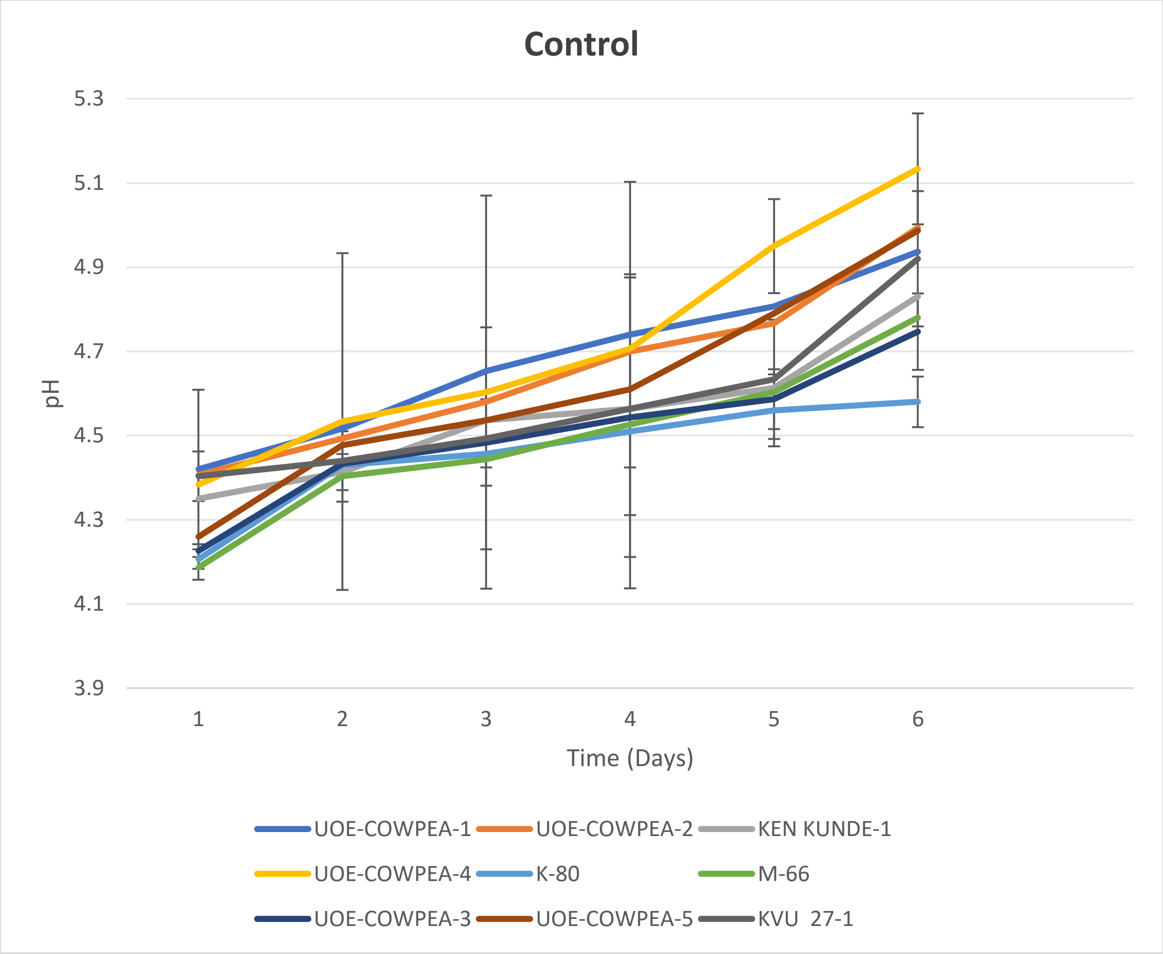

Figure 4.1. Changes in solution culture pH as influenced by different cowpea cultivars grown in acidic solution culture without aluminium

The growth of UOE-COWPEA-4 in the culture solution without Al caused the highest change in pH from 4.3 to 5.13 (raised pH by 0.83) compared to other cultivars while the cultivar K-80 induced the least change in pH from 4.21 to 4.58 (raised pH by 0.37) as shown in Figure 4.1 above.

The growth of UOE-COWPEA-5 at 185 µM Al induced the highest change in pH when compared to other cultivars with an increment from 4.03 to 5.06 (raised pH by 1.03). The cultivar K-80 still induced the least pH change from 4.32 to 4.53 (raised pH by 0.21) in solution culture supplemented with 185 µM Al (Figure 4.2).

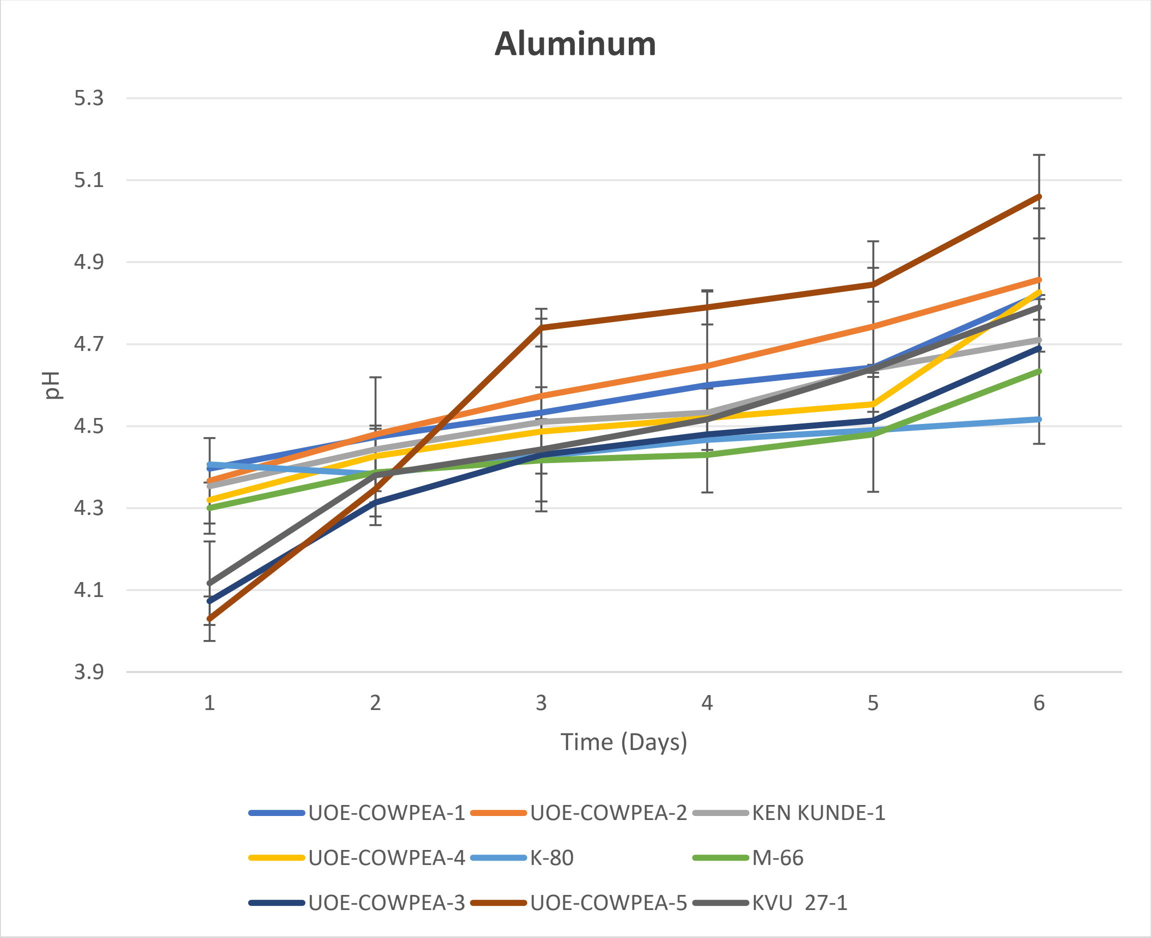

Figure 4.2. Changes in solution culture pH as influenced by the different cowpea cultivars grown in acidic solution culture with 185 µM aluminium

ANOVA was used to assess the effect of cowpea cultivars and Al toxicity on pH of culture solution at P≤0.05. The cultivar, treatment and the number of days had a significant statistical effect on pH of the culture solution. The interactions were however, not statistically significant at 95% confidence level (Table 4.1).

Table 4.1: ANOVA table on effects of cowpea cultivars and Al toxicity on pH of culture solution

Source of Variation	Sum of Squares	Df	Mean Square	F-Ratio	P-Value
MAIN EFFECTS					
A:CULTIVAR	1.76022	8	0.220028	5.75	0.0000
B:TREATMENT	0.336078	1	0.336078	8.78	0.0034
C:Days	9.3339	5	1.86678	48.76	0.0000
INTERACTIONS					
AB	0.301126	8	0.037641	0.98	0.4500
AC	1.00077	40	0.025019	0.65	0.9455
BC	0.058558	5	0.011712	0.31	0.9090
ABC	0.545143	40	0.013629	0.36	0.9999
RESIDUAL	8.26892	216	0.038282		
TOTAL (CORRECTED)	21.6047	323			

4.2 Response of Cowpea Cultivars to Al Concentration in Culture Solution

There was a significant difference (*P≤0.05*) observed in the net root lengths of cowpea cultivars at 0 µM Al and 185 µM Al (Table 4.2). The cowpea cultivars UOE-COWPEA-2 and UOE-COWPEA-1 had significantly higher net root lengths while M-66 had the least at 0 µM Al. At 185 µM Al, a significant difference was also observed among the cultivars. UOE-COWPEA-2 had the highest net root length whereas M-66 had the least (Table 4.2).

Table 4.2: Response of Cowpea Cultivars to Al Concentraction in Culture Solution

	Control (0 uM Al)			Aluminum (185 uM Al)		
CULTIVAR	**NRL**	**SL**	**NO LR**	**NRL**	**SL**	**NO LR**
UOE-COWPEA-1	13.91±3.2^{ab}	10.60±2.5^{a}	59.40±13.8^{ab}	8.67±2.5^{b}	11.05±1.5^{a}	41.27±10.3^{bc}
UOE-COWPEA-2	14.25±1.9^{b}	10.40±2.4^{a}	60.67±10^{ab}	10.32±2.3^{ab}	9.33±2.4^{a}	45.10±13.1^{bc}
UOE-COWPEA-3	10.88±2.3^{ab}	10.82±2.1^{a}	81.30±21.9^{c}	7.40±2.8^{bc}	11.12±2.2^{a}	57.16±14.4^{ab}
KEN KUNDE-1	12.48±2.3^{ab}	8.76±2.1^{a}	59.89±13.7^{ab}	9.29±2.5^{b}	8.83±1.8^{a}	45.30±10.5^{bc}
UOE-COWPEA-4	10.75±2.2^{ab}	10.64±1.5^{a}	45.33±8.9^{bc}	8.13±2.3^{bc}	10.38±1.9^{a}	35.40±7.9^{c}
K-80	9.45±2.5^{bc}	9.49±1.9^{a}	49.80±10.4^{abd}	6.04±2.1^{bc}	9.29±2.2^{a}	36.19±10.9^{bcd}
M-66	8.22±2.0^{bc}	9.70±1.5^{a}	51.92±11.3^{abd}	5.42±1.3^{c}	9.30±2.5^{a}	35.39±10.5^{bcd}
UOE-COWPEA-5	10.93±3.4^{ab}	9.50±2.4^{a}	53.33±12.9^{abd}	6.71±2.1^{bc}	9.27±1.7^{a}	39.64±8.7^{bcd}
KVU 27-1	11.23±2.9^{ab}	12.03±2.2^{a}	63.96±17.9^{ab}	8.40±2.8^{bc}	11.88±1.6^{a}	47.10±12.4^{abd}

Means with similar letters are not significantly different at $p< 0.05$. Means were separated using the Turkey test. The data presented are means±SD

There was no significant difference on shoot lengths among the cultivars at 0 µM Al and 185 µM Al. KVU 27-1 cultivar had longer shoot length while KEN-KUNDE-1 cultivar had the least at 0 µM Al (Table 4.2). At 185 µM Al, KVU 27-1 cultivar recorded a higher shoot length whileKEN-KUNDE-1 cultivar recorded the least (Table 4.2).

The number of lateral roots was not significantly different at 0 µM Al and 185 µM Al among the cultivars. UOE-COWPEA-3 cultivar recorded the highest number of lateral roots while UOE-COWPEA-4 cultivar recorded the least (Table 4.2). At 185 µM Al, the number of lateral roots were significantly reduced by aluminium stress and no significant difference was observed among the cultivars. UOE-COWPEA-3 cultivar still recorded a higher number of lateral roots while UOE-COWPEA-4 and M-66 cultivars recorded the least (Table 4.2).

There was no significant difference among the cowpea cultivars on relative net root length (RNRL) response to the aluminium treatment. UOE-COWPEA-4, KVU 27-1, KEN KUNDE-1 and UOE-COWPEA-2 (75.6 %, 75.1%, 74.2%, and 72.7%) cultivars had higher RNRL whereas UOE-COWPEA-1 and UOE-COWPEA-5 cultivar (63.9%) and 63.7%) had the least (Figure 4.3).

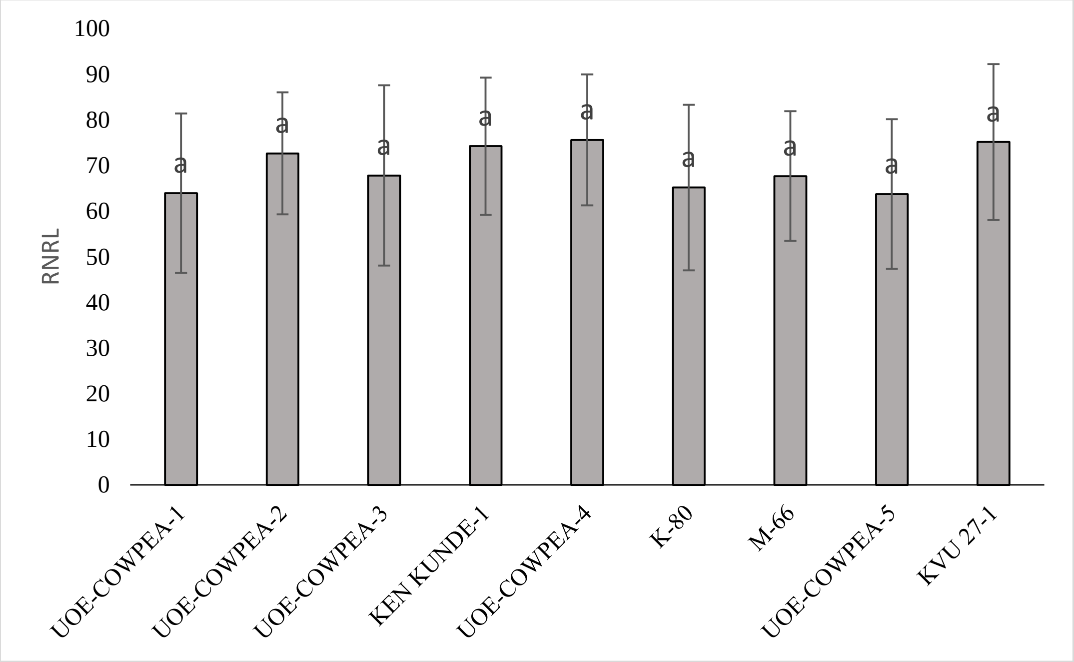

Figure 4.3. Effect of low pH and Al toxicity on cowpea cultivars Relative net root length

Bars with similar letters are not significantly different at P≤0.05

4.3 Effects of Low pH and Al Toxicity on cowpea Plant Biomass

The root dry biomass was significantly different among the cultivars and treatments at 0 µM Al and 185 µM Al (Table 4.3). The root dry biomass was significantly lower at 185 µM Al relative to control. UOE-COWPEA-1 and UOE-COWPEA-5 recorded a higher root dry biomass at 0 µM Al while UOE-COWPEA-4 recorded the least. At 185 µM Al, UOE-

COWPEA-1 had higher root dry biomass and UOE-COWPEA-4 still had the least (Table 4.3).

Table 4.3: Effects of Low pH and Al Toxicity on cowpea Plant Biomass

	Control (0 uM Al)			**Aluminum (185 uM Al)**		
CULTIVAR	**DRB**	**DSB**	**RB:SB**	**DRB**	**DSB**	**RB:SB**
UOE-COWPEA-1	0.05±0.03^{a}	0.26±0.16^{a}	0.2±0.04^{a}	0.04±0.03^{b}	0.2±0.13^{b}	0.21±0.06^{a}
UOE-COWPEA-2	0.04±0.03^{ab}	0.24±0.15^{a}	0.19±0.07^{a}	0.03±0.02^{c}	0.18±0.11^{b}	0.19±0.08^{b}
UOE-COWPEA-3	0.05±0.03^{a}	0.23±0.15^{a}	0.21±0.05^{a}	0.04±0.03^{b}	0.16±0.1^{bc}	0.25±0.1^{b}
KEN KUNDE-1	0.05±0.03^{ab}	0.32±0.22^{d}	0.21±0.05^{a}	0.04±0.03^{b}	0.22±0.16^{d}	0.24±0.04^{c}
UOE-COWPEA-4	0.04±0.02^{ab}	0.14±0.08^{b}	0.31±0.39^{b}	0.03±0.02^{d}	0.11±0.06^{e}	0.22±0.07^{ab}
K-80	0.04±0.03^{ab}	0.26±0.17^{a}	0.18±0.08^{ab}	0.03±0.02^{b}	0.18±0.12^{b}	0.18±0.06^{ab}
M-66	0.04±0.03^{ab}	0.26±0.17^{a}	0.16±0.03^{ab}	0.04±0.02^{b}	0.21±0.14^{b}	0.18±0.06^{ab}
UOE-COWPEA-5	0.05±0.04^{a}	0.28±0.17^{a}	0.2±0.12^{a}	0.03±0.02^{b}	0.19±0.12^{b}	0.2±0.08^{b}
KVU 27-1	0.05±0.03^{a}	0.24±0.15^{a}	0.21±0.05^{a}	0.04±0.02^{ab}	0.19±0.12^{b}	0.21±0.07^{ab}

Means with similar letters are not significantly different at p< 0.05. Means were separated using the Turkey test. The data presented are means±SD

Key: DRB= Dry Root Biomass, DSB= Dry Shoot Biomass, RB:SB= Root to Shoot Ratio

Shoot dry biomass was also significantly different among the cultivars and treatments. KEN-KUNDE-1 cultivar recorded a higher shoot dry biomass and UOE-COWPEA-4 recorded the least at 0 μM Al. At 185 μM Al also, M-66 cultivar recorded higher shoot dry biomass and the least UOE-COWPEA-4 as shown in Table 4.3 above.

Higher root to shoot ratio was observed at 0 µM Al than at 185 µM Al except for KEN KUNDE-1 cultivar. A significant difference was observed on root to shoot ratio among the cultivars and treatments (Table 4.3). At 0 µM Al UOE-COWPEA-4 had higher root to shoot ratio while M-66 cultivar had the least, and at 185 µM Al KEN-KUNDE-1 cultivar had higher root to shoot ratio while UOE-COWPEA-1 and M-66 cultivars had the least root to shoot ratio (Table 4.3).

4.4 The Effect of Cowpea Cultivars on Soil pH and Selected Soil Nutrient Profiles When Grown on Acidic Phosphorus Deficient Soils

Soil chemical properties

The soil from the experimental field before planting had the following properties; soil pH 5.0, soil available P; 5.51 mg/kg, % total nitrogen; 0.13, % organic carbon; 1.58, exchangeable cations (cmol/kg soil): (C, 2.46; Mg, 1.19; K, 0.54 and Na, 0.52).

4.4.1 Soil pH

The soil pH in the study site at the University farm ranged from 4.6 to 5.3. The results for soil rhizosphere pH at flowering and physiological maturity stage are presented in Table 4.4 below. At the physiological maturity stage, the rhizosphere soil pH was higher for all the cultivars and treatments compared with the flowering stage.

All the four cowpea cultivars grown in the field increased the rhizosphere pH by different values. At control treatment, UOE-COWPEA-2 cultivar increased the soil pH and the least K-80 cultivar by 0.4 units though they were not significantly different from each other at physiological maturity stage (Table 4.4).

When phosphorus was applied, the soil pH increased, KVU 27-1 raised the pH by 0.9 units and the least Ken-kunde-1 cultivar by 0.5 units. The soil rhizosphere pH recorded was higher compared to that of control treatment (Table 4.4).

Table 4.4: pH of Soils sampled from the Rhizosphere of different Cowpea Cultivars

CULTIVAR	TREATMENTS	PH R1	PH R2	PH R3
UOE-COWPEA-2	Control	4.87±0.06^{a}	5.00±0.09^{a}	5.50±0.1^{a}
	Phosphorus	5.07±0.05^{b}	5.63±0.28^{b}	5.72±0.31^{a}
	Lime	4.96±0.04^{a}	5.69±0.36^{b}	5.88±0.38^{a}
	P+L	5.28±0.0^{c}	6.05±0.32^{b}	6.12±0.48^{b}
KVU27-1	Control	5.02±0.01^{a}	5.21±0.14^{a}	5.54±0.15^{a}
	Phosphorus	4.57±0.65^{ab}	5.38±0.07^{b}	5.46±0.09^{a}
	Lime	4.95±0.02^{a}	5.45±0.22^{ab}	5.93±0.51^{ab}
	P+L	5.09±0.27^{ab}	5.72±0.06^{b}	6.16±0.4^{b}
K-80	Control	5.06±0.03^{b}	5.45±0.13^{b}	5.50±0.16^{b}
	Phosphorus	5.06±0.03^{b}	5.45±0.13^{b}	5.50±0.16^{b}
	Lime	5.23±0.02^{b}	5.90±0.52^{b}	6.37±0.47^{b}
	P+L	4.97±0.04^{a}	6.03±0.62^{b}	6.03±0.56^{b}
Ken-Kunde-1	Control	4.95±0.01^{a}	5.34±0.26^{b}	5.51±0.09^{b}
	Phosphorus	5.07±0.04^{b}	5.39±0.45^{b}	5.55±0.27^{b}
	Lime	5.19±0.02^{ab}	5.90±0.58^{ab}	5.93±0.33^{ab}
	P+L	5.00±0.03^{c}	5.89±0.25^{b}	5.89±0.22^{b}

The data presented are means±SD. Means with similar letters are not significantly different at p< 0.05.

Key: pHR1=pH before planting, pHR2= pH at flowering and pHR3= pH at physiological maturity.

Application of lime also resulted to an increase in rhizosphere soil pH with K-80 cultivar raising the soil pH by 1.1 units higher than the values obtain when P alone was applied and the least with KENKUNDE-1 by 0.7 units and a significant difference was observed among the cultivars (Table 4.4).

Application of combined phosphorus and lime also increased the soil pH higher compared to sole application of phosphorus and at control but similar to lime application alone. KVU 27-1 and K-80 cultivar raised the soil pH by 1.1 units each and the least with UOE-COWPEA-2 cultivar by 0.8 units. A significant difference was also observed among the cultivars and treatments (Table 4.4).

4.4.2 Soil nutrients

a) Total soil Nitrogen

Total soil nitrogen content was higher at flowering stage when compared to when the crop was at physiological maturity stage as shown in Table 4.5. Cowpea cultivars slightly increased the total nitrogen content in the soil with K-80 increasing by 0.04 units though they were not significantly different from each other. The application of P to the soil also resulted in a slight increase of total N content and no significant difference was observed among the cultivars with K-80 cultivar increasing by 0.09 units above the control values. Lime application also increased the total N content in the soil but there was no significant difference observed among the cultivars. The values recorded upon lime application was higher compared to control and P application alone with UOE-COWPEA-2 increasing by 0.08 units.

The interaction of lime and phosphorus also resulted to an increase in total N content with K-80 cultivar increasing by 0.1 units, though there was no significant difference observed among the cultivars. The values recorded by the interaction of P+L were similar to those recorded when lime alone was applied. The application of treatments increased soil total N values in the order; combined L and P with K-80 cultivar (0.22%) followed by L with

UOE-COWPEA-2 and then P application alone with K-80 (0.19%) and the lowest was observed under control with K-80 cultivar (0.17%) as shown in Table 4.5.

Table 4.5: Effect of Cultivars and Treatments on Percent Total Nitrogen in the soil.

CULTIVAR	TREATMENTS	PH R1	PH R2	PH R3
UOE-COWPEA-2	Control	0.14 ± 0.01^{a}	0.16 ± 0.01^{a}	0.17 ± 0.01^{a}
	Phosphorus	0.14 ± 0.01^{b}	0.18 ± 0.01^{b}	0.17 ± 0.01^{a}
	Lime	0.11 ± 0.02^{a}	0.19 ± 0.02^{b}	0.16 ± 0.01^{a}
	P+L	0.12 ± 0.02^{c}	0.19 ± 0.01^{b}	0.17 ± 0.01^{b}
KVU27-1	Control	0.14 ± 0.01^{a}	0.16 ± 0.01^{a}	0.16 ± 0.01^{a}
	Phosphorus	0.14 ± 0.01^{a}	0.19 ± 0.01^{b}	0.16 ± 0.01^{a}
	Lime	0.13 ± 0.01^{a}	0.20 ± 0.01^{b}	0.17 ± 0.01^{b}
	P+L	0.13 ± 0.02^{ab}	0.21 ± 0.01^{ab}	0.16 ± 0.01^{ab}
K-80	Control	0.13 ± 0.01^{b}	0.17 ± 0.01^{b}	0.17 ± 0.01^{b}
	Phosphorus	0.12 ± 0.02^{b}	0.19 ± 0.01^{b}	0.15 ± 0.01^{b}
	Lime	0.15 ± 0.01^{b}	0.21 ± 0.02^{b}	0.17 ± 0.01^{b}
	P+L	0.12 ± 0.03^{a}	0.22 ± 0.01^{b}	0.16 ± 0.01^{b}
Ken-Kunde-1	Control	0.14 ± 0.01^{a}	0.17 ± 0.01^{b}	0.16 ± 0.01^{b}
	Phosphorus	0.12 ± 0.01^{b}	0.16 ± 0.01^{b}	0.17 ± 0.01^{b}
	Lime	0.14 ± 0.01^{c}	0.19 ± 0.01^{b}	0.17 ± 0.00^{ab}
	P+L	0.14 ± 0.01^{ab}	0.19 ± 0.01^{b}	0.17 ± 0.01^{b}

The data presented are means±SD. Means with similar letters are not significantly different at $p < 0.05$.

Key: R1=Total Nitrogen before planting, R2=Total Nitrogen at flowering and R3=Total Nitrogen at physiological maturity

b) Available soil phosphorus

Table 4.6, shows the effect of treatments and cultivars growth on soil phosphorus. The growth of cowpea cultivars and treatment application significantly influenced the available P in the soil, at flowering and at physiological maturity stages. The available P recorded at physiological maturity stage was higher as compared to at flowering stage for all the four cultivars. At control treatment, KEN KUNDE-1 cultivar increased the available P with an increment of 12.7 mg/kg at physiological maturity stage compared to other cultivars.

Table 4.6: Effect of cultivar and treatment on available phosphorus.

CULTIVAR	TREATMENTS	PH R1	PH R2	PH R3
UOE-COWPEA-2	Control	5.68 ± 0.05^{a}	14.33 ± 0.25^{b}	16.27 ± 0.45^{c}
	Phosphorus	5.63 ± 0.29^{a}	17.67 ± 0.4^{d}	18.77 ± 0.4^{e}
	Lime	5.34 ± 0.02^{a}	20.20 ± 1.21^{e}	22.17 ± 1.34^{ef}
	P+L	5.47 ± 0.18^{a}	22.9 ± 1.93^{e}	26.17 ± 0.32^{g}
KVU27-1	Control	5.54 ± 0.18^{a}	13.83 ± 0.4^{b}	15.63 ± 0.67^{c}
	Phosphorus	5.5 ± 0.16^{a}	21.4 ± 6.09^{cd}	20.9 ± 0.46^{fg}
	Lime	5.48 ± 0.15^{a}	24.0 ± 1.9^{b}	21.0 ± 0.75^{b}
	P+L	5.37 ± 0.03^{ab}	23.3 ± 1.44^{ab}	25.8 ± 0.5^{ab}
K-80	Control	5.38 ± 0.25^{b}	14.4 ± 0.26^{b}	13.9 ± 0.4^{b}
	Phosphorus	5.78 ± 0.21^{b}	17.33 ± 1.42^{b}	17.03 ± 1.14^{b}
	Lime	5.44 ± 0.11^{b}	19.67 ± 1.4^{b}	22.1 ± 0.89^{b}
	P+L	5.63 ± 0.28^{a}	20.6 ± 1.21^{b}	28.93 ± 0.5^{b}
Ken-Kunde-1	Control	5.47 ± 0.11^{a}	14.6 ± 0.1^{b}	18.17 ± 0.15^{b}
	Phosphorus	5.22 ± 0.11^{b}	19.33 ± 0.83^{b}	19.4 ± 0.95^{b}
	Lime	5.75 ± 0.13^{c}	24.23 ± 2.77^{b}	21.2 ± 1.15^{ab}
	P+L	5.50 ± 0.18^{ab}	22.03 ± 1.07^{b}	26.07 ± 0.59^{b}

The data presented are means±SD. Means with similar letters are not significantly different at p< 0.05.

Key: R1=Available Phosphorus before planting, R2=Available Phosphorus at flowering and R3=Available Phosphorus at physiological maturity

KVU 27-1 cultivar application with P alone increased the soil available P by 15.4 mg/kg higher than that of control. UOE-COWPEA-2 and K-80 cultivars applied with lime application also significantly increased the amount of available P recording the highest values with an increment of 16.8 and 16.7 mg/kg each. The values recorded are higher than that of control and P alone but lower than that of combined P and L. Combined P and L also increased the amount of soil available P. K-80 cultivar applied with combined P and L increased the soil available P higher with increment of 23.3mg/kg.

c) Soil Organic Carbon

The results for the effect of cultivar and treatments on soil organic carbon is shown in Table 4.7. The organic carbon content was higher at physiological maturity stage compared to flowering stage. The growth of cowpea cultivars increased the soil organic carbon and a significant difference was also observed among the cultivars. At control treatment, UOE-COWPEA-2 cultivar increased the soil organic carbon by 1.05 units and KENKUNDE-1 recorded the least by 0.14 units (Table 4.7).

Table 4.7: Effect of cultivar and treatment percentage carbon.

CULTIVAR	TREATMENTS	PH R1	PH R2	PH R3
UOE-COWPEA-2	Control	1.52±0.14^{ab}	1.67±0.04^{bc}	2.57±0.13^{f}
	Phosphorus	1.59±0.03^{a}	1.83±0.01^{b}	1.97±0.1^{f}
	Lime	1.62±0.02^{a}	2.0±0.02^{d}	2.15±0.02^{e}
	P+L	1.59±0.05^{a}	1.93±0.03^{d}	2.13±0.1^{de}
KVU27-1	Control	1.59±0.05^{a}	1.75±0.06^{b}	1.85±0.03^{d}
	Phosphorus	1.63±0.02^{a}	1.87±0.02^{d}	1.95±0.07^{d}
	Lime	1.67±0.02^{a}	2.0±0.05^{d}	1.95±0.02^{e}
	P+L	1.38±0.13^{c}	2.1±0.04^{e}	2.08±0.15^{e}
K-80	Control	1.51±0.12^{abc}	1.82±0.02^{bb}	2.25±0.02^{b}
	Phosphorus	1.51±0.16^{abc}	1.80±0.07^{b}	1.85±0.1^{b}
	Lime	1.49±0.16^{abc}	1.92±0.08^{b}	1.86±0.05^{b}
	P+L	1.7±0.05^{a}	1.83±0.1^{ab}	1.95±0.04^{e}
Ken-Kunde-1	Control	1.66±0.11^{ab}	1.73±0.04^{ab}	1.8±0.06^{ab}
	Phosphorus	1.67±0.03^{a}	1.94±0.09^{b}	2.07±0.07^{be}
	Lime	1.59±0.05^{a}	1.93±0.03^{b}	2.13±0.1^{be}
	P+L	1.55±0.06^{a}	2.08±0.04^{e}	1.94±0.02^{b}

The data presented are means±SD. Means with similar letters are not significantly different at $p < 0.05$.

Key: R1=Soil Organic Carbon before planting, R2=Soil Organic Carbon at flowering and R3=Soil Organic Carbon at physiological maturity

Application of P fertilizer alone also increased the soil organic carbon with KENKUNDE-1 recording the highest (2.07%) while KVU 27-1 cultivar recorded the least (1.95%). Lime application also increased soil organic carbon content in the soil and a significant difference was observed among the cultivars (Table 4.7). The value recorded upon lime application was higher compared to control and P application. KENKUNDE-1 cultivar recorded a higher organic carbon content (2.23%) and KVU 27-1 resulted in the least (2.08%). The interaction of lime and phosphorus also resulted in an increase in soil organic carbon. A significant difference was observed among the cultivars. Values recorded by the interaction of P+L were higher than those recorded when lime alone was applied. KVU 27-1 cultivar recorded a higher value (2.25%) while K-80 cultivar recorded the least (2.08%) in the P+L treatment combination (Table 4.7).

4.4.3 Exchangeable cations

The growth of cowpea cultivars and treatments application at flowering stage and at physiological maturity stage had a significant influence on Na^+, Mg^{2+}, Ca^{2+} and K^+ in the soil (Table 4.8).

a) Sodium

The initial level of Na^+ in the soil before planting ranged between 0.46 to 0.60 mg/kg. The growth of cowpea cultivars and treatment application increased the amount of Na^+at flowering and physiological maturity stages. Cowpea cultivars growth increased the amount of Na^+ in the soil, though there was no significant difference ($P>0.05$) observed among the cultivars. Treatments application had a significant effect ($P\leq0.05$) on the amount of Na^+. Application of P+L increased the amount of sodium higher with UOE-COWPEA-

2 and Ken-Kunde-1 cultivars from 0.60 to 0.73 mg/kg and 0.59 to 0.73 mg/kg and the least was recorded in control plots with K-80 cultivar from 0.50 to 0.53 mg/kg at mid planting. At physiological maturity stage, the amount of sodium in the soil was higher compared to flowering. Plots applied with P+L recorded higher amount of sodium with KVU 27-1 cultivar from 0.54 to 1.14 mg/kg and the least was recorded in control plots with K-80 cultivar from 0.50 to 0.56 mg/kg (Table 4.8).

b) Magnesium

The level of Mg^{2+}in the soil ranged between 0.87 to 1.42 mg/kg. There was no significant difference ($P>0.05$) observed among the cultivars at flowering and at physiological maturity stages. Application of treatments increased the amount of Mg^{2+}content in the soil and a significant difference ($P\leq0.05$) was observed at the two stages of growth. Higher Mg^{2+}content was recorded in plots with P+L with KENKUNDE-1 cultivar from 0.87 to 1.64 mg/kg at flowering and physilogical maturity stages. The least amount of Mg^{2+}content was recorded in control plots with K-80 (1.11 to 1.21 mg/kg) and KVU 27-1 cultivar (1.02 to 1.22 mg/kg) at flowering and physiological maturity stages (Table 4.8).

c) Calcium

The initial soil status of Ca^{2+} was 2.23 mg/kg to 2.91 mg/kg. The growth of cowpeas increased the Ca^{2+} content though no significant difference ($P\leq0.05$) was observed among the cultivars. Ca^{2+} content was higher at flowering compared to physiological maturity stage with Ken-Kunde-1 cultivar (2.73 to 11.45 mg/kg) recording the higher amount and the least K-80 cultivar from 2.91 to 5.95 mg/kg. A significant difference ($P\leq0.05$) was observed in the Ca^{2+} content in the soil upon treatments application. The amount of Ca^{2+}

content was higher at physiological stage compared to flowering stage. Plots applied with combined lime and phosphorus recorded higher Ca^{2+} content with KVU 27-1 cultivar (2.30 mg/kg to 18.63 cmol/kg) and the least was recorded in control plots with K-80 cultivar (2.91 mg/kg to 5.33 mg/kg) at physiological maturity stage (Table 4.8).

d) Potassium

The initial amount of K^+ in the soil ranged from 0.51 to 0.66 mg/kg. Cultivars growth and treatments application increased the amount of K^+ content in the soil at flowering and physiological stage though a higher level was recorded at physiological stage. There was no significant difference ($P>0.05$) observed among the cultivars on K^+content. K-80 cultivar (0.55 to 0.74 mg/kg) recorded a higher amount and the least KVU 27-1 cultivar (0.52 to 0.64 mg/kg). A significant difference ($P\leq0.05$) was observed on treatments. Plots applied with P+L recorded higher amount of K^+content with K-80 cultivar from 0.55 to 1.04 mg/kg and the least was recorded in control plots with KVU 27-1 cultivar from 0.52 mg/kg to 0.54 mg/kg (Table 4.8).

Table 4.8. Effect of cowpea growth and treatments on Na+, Mg2+, Ca2+ and K+ in the soil.

			Before planting			**Mid planting**				**Physiological maturity stage**			
	Treatment	UOE-COWPEA-2	**KVU27-1**	**K-80**	**KEN-KUNDE-1**	UOE-COWPEA-2	**KVU27-1**	**K-80**	**KEN KUNDE-1**	UOE-COWPEA-2	**KVU27-1**	**K-80**	**KEN KUNDE-1**
	Control	0.54±0.05a	0.54±0.02b	0.50±0.06a	0.50±0.07a	0.55±0.02a	0.58±0.01a	0.53±0.03a	0.55±0.03a	0.61±0.03a	0.72±0.02a	0.56±0.02a	0.63±0.03a
	Phosphorus (P)	0.54±0.02a	0.46±0.04a	0.49±0.05a	0.46±0.03a	0.61±0.02b	0.63±0.03b	0.63±0.12b	0.63±0.01b	0.63±0.01a	0.73±0.02a	0.62±0.02b	0.64±0.02a
Sodium	Lime (L)	0.55±0.03a	0.49±0.07a	0.47±0.06a	0.47±0.04a	0.69±0.02b	0.63±0.06b	0.68±0.02b	0.71±0.02c	0.71±0.02b	0.81±0.03b	0.72±0.02c	0.74±0.03b
	P+L	0.60±0.02b	0.54±0.02b	0.50±0.01a	0.59±0.01b	0.73±0.02c	0.64±0.01b	0.67±0.01b	0.73±0.04c	0.73±0.01b	1.14±0.02c	0.73±0.01c	0.76±0.02b
	Control	1.29±0.04b	1.02±0.16a	1.11±0.10a	1.22±0.02c	1.36±0.13a	1.22±0.03a	1.21±0.04a	1.26±0.04a	1.27±0.01a	1.22±0.09a	1.23±0.06a	1.25±0.10a
Magnesium	Phosphorus (P)	1.24±0.02b	1.29±0.01c	1.24±0.01b	1.26±0.05c	1.36±0.04a	1.30±0.03b	1.25±0.04a	1.40±0.03b	1.28±0.05a	1.36±0.07b	1.31±0.18b	1.42±0.02b
	Lime (L)	1.12±0.02a	1.21±0.03b	1.23±0.03ab	1.11±0.03b	1.36±0.06a	1.31±0.03b	1.28±0.01a	1.62±0.07c	1.37±0.06b	1.38±0.02b	1.37±0.01b	1.63±0.06c
	P+L	1.18±0.01a	1.42±0.01d	1.25±0.01b	0.87±0.03a	1.37±0.21a	1.44±0.03c	1.29±0.48a	1.64±0.02c	1.41±0.03b	1.41±0.03c	1.42±0.02c	1.64±0.03c

	Control	2.66±0.07b	2.85±0.11b	2.91±0.03c	2.73±0.04c	9.38±0.02b	9.36±0.02a	5.95±0.04a	11.45±0.03a	7.91±0.04a	7.28±0.03a	5.33±0.03a	7.22±0.02a
Calcium	Phosphorus (P)	2.26±0.02a	2.38±0.05a	2.23±0.03a	2.37±0.03a	7.75±0.02a	9.64±1.15b	7.90±0.04b	11.74±0.03b	8.15±0.04a	14.43±0.01b	6.69±0.11b	10.82±0.02b
	Lime (L)	2.23±0.03a	2.43±0.03a	2.37±0.09b	2.43±0.07b	9.87±0.02c	9.81±0.01b	9.95±0.01c	12.45±0.03c	10.08±0.01b	16.96±0.01c	12.37±0.03c	11.42±0.02b
	P+L	2.28±0.13a	2.30±0.13a	2.49±0.04b	2.42±0.19ab	10.10±0.18d	16.38±0.01c	11.38±0.02d	13.19±0.02d	11.25±0.01c	18.63±0.03d	16.59±0.03d	14.09±0.02c
	Control	0.53±0.03a	0.52±0.02a	0.55±0.02a	0.54±0.02a	0.54±0.02a	0.54±0.02a	0.66±0.03a	0.62±0.04a	0.64±0.04a	0.64±0.02a	0.74±0.04a	0.66±0.05a
Potassium	Phosphorus (P)	0.53±0.01a	0.54±0.02a	0.52±0.02a	0.57±0.02a	0.57±0.06a	0.54±0.02a	0.69±0.03a	0.66±0.03b	0.75±0.05b	0.66±0.02b	0.83±0.02b	0.72±0.02a
	Lime (L)	0.52±0.02a	0.54±0.04a	0.55±0.02a	0.51±0.05a	0.61±0.04b	0.63±0.03b	0.88±0.04b	0.73±0.03b	0.77±0.02b	0.77±0.04c	0.92±0.02c	0.81±0.01b
	P+L	0.55±0.03a	0.55±0.03a	0.55±0.01a	0.56±0.02a	0.66±0.02b	0.65±0.02b	1.04±0.04c	0.78±0.02c	0.78±0.03b	0.75±0.02c	0.93±0.06c	0.83±0.02b

Means of soil exchangeable cations with three replicates at a significant level of ($P \leq 0.05$) ± Standard Deviation using Tukeys Test.*Means in the same column with the same superscript are not significantly different.

4.5 Effect of Lime and Phosphorus Application on Vegetative Growth

4.5.1 Plant height

Both cultivars and treatments significantly ($P \leq 0.05$) affected plant height (Table 4.9). Plant height is a cultivar dependent trait as certain cultivars were significantly ($P \leq 0.05$) taller than others. At 60 DAP, there was no significant difference observed among the cultivars. At 75 and 90 DAP, a significant difference among the cowpea cultivars and the treatments were observed. There was a significant increase on plant height among cowpea cultivars in all the sampling days. At control treatment, UOE-COWPEA-2 cultivar recorded the highest plant height in all the sampling days whereas K-80 recorded the least plant height in all the days.

Plots applied with phosphorus fertiliser had taller plants compared to control treatment. UOE-COWPEA-2 cultivar recorded the highest plant height while K-80 cultivar recorded the least. Plots applied with lime treatment also produced tall plants but with reduced height compared to that of phosphorus treatment. UOE-COWPEA-2 cultivar still recorded a greater plant height while K-80 recorded the least. Plots that were applied with a combination of phosphorus and lime had taller plants compared to control and lime treatments but lower than that of phosphorus treatment. A significant difference was observed among the cultivars with UOE-COWPEA-2 cultivar still recording the highest plant height and K-80 cultivar recording the least.

Table 4.9. Effect of treatments on plant height

Cultivar	Treatment	Plant height (cm)		
		60 DAP	75DAP	90DAP
UOE-COWPEA-2	Control	9.66±1.13^{a}	16.04±2.41^{b}	21.02±3.02^{b}
	Phosphorus	10.90±2.34^{a}	16.35±2.55^{b}	22.33±4.35^{b}
	Lime	9.87±1.56^{a}	15.75±2.34^{a}	21.66±3.68^{b}
	P+L	10.77±2.09^{a}	17.42±5.98^{b}	23.21±4.11^{b}
KVU 27-1	Control	9.33±1.52^{a}	11.42±1.87^{a}	13.85±2.06^{a}
	Phosphorus	10.24±1.58^{a}	13.19±2.40^{a}	17.76±3.24^{a}
	Lime	10.61±1.29^{a}	13.11±1.67^{a}	16.22±1.81^{a}
	P+L	11.02±1.46^{a}	14.27±1.79^{a}	17.65±2.23^{a}
K-80	Control	9.00±1.23^{a}	11.42±1.37^{a}	13.71±1.64^{a}
	Phosphorus	9.46±1.49^{a}	12.26±1.75^{a}	15.22±3.18^{a}
	Lime	9.37±1.61^{a}	12.05±1.69^{a}	14.45±1.92^{a}
	P+L	9.84±1.61^{a}	13.11±2.39^{a}	16.18±3.47^{a}
Ken kunde-1	Control	8.84±1.33^{a}	12.89±2.25^{a}	16.86±3.00^{a}
	Phosphorus	9.82±1.15^{a}	13.77±2.28^{a}	19.14±4.68^{ab}
	Lime	9.15±1.77^{a}	12.96±2.00^{a}	16.64±2.74^{a}
	P+L	10.28±1.55^{a}	14.53±3.07^{a}	19.48±4.79^{ab}

*Means in the same column with the same superscript are not significantly different ($P \leq 0.05$) ± Standard Deviation.

DAP= Days after Planting

4.5.2 Shoot branching

The results on number of branches per plant at 60, 75 and 90 DAP are presented in Table 4.10. The number of branches formed at 60 DAP, was not significantly differently among the cultivars. At 75 DAP, there was a progressive increase in the number of branches compared to at 60 DAP which differed significantly ($P \leq 0.05$) among the cultivars. UOE-COWPEA-2 cultivar produced the highest number of branches. At 90 DAP, there was a significant difference ($P \leq 0.05$) observed on the number of branches among the cultivars. UOE-COWPEA-2 cultivar formed the highest number of branches while KVU 27-1 cultivar produced the least number of branches. The number of branches was not different among the treatments. At 60 DAP, there was no significant effect of treatment on number of branches, however, plots that were applied with combined phosphorus and lime recorded more shoot branching than in the rest of the treatments.

Table 4.10. Effect of treatments on the number of branches per plant

Cultivar	Treatment	60 DAP	75 DAP	90 DAP
UOE-COWPEA-2	Control	3.57±0.94^{a}	5.77±1.59^{ab}	7.87±1.72^{b}
	Phosphorus	4.37±1.43^{ab}	6.03±1.53^{ab}	8.83±2.77^{bc}
	Lime	4.30±1.18^{ab}	6.00±1.02^{ab}	8.17±1.66^{bc}
	P+L	4.56±0.86^{ab}	6.07±1.25^{ab}	8.97±2.49^{c}
KVU27-1	Control	2.83±1.53^{a}	4.17±0.87^{a}	5.93±1.31^{a}
	Phosphorus	4.00±1.23^{ab}	5.33±1.32^{a}	7.10±1.67^{b}
	Lime	2.97±1.03^{a}	4.43±1.01^{a}	6.13±1.36^{a}
	P+L	4.03±1.09^{ab}	5.43±1.31^{a}	7.53±2.67^{ab}
K-80	Control	4.13±1.01^{ab}	5.80±1.21^{a}	7.50±1.19^{b}
	Phosphorus	4.33±1.03^{ab}	5.63±0.88^{a}	7.53±1.69^{b}
	Lime	3.80±1.18^{a}	5.63±1.07^{a}	7.43±1.89^{b}
	P+L	4.70±0.87^{b}	6.13±1.22^{b}	7.73±1.74^{b}
Ken-Kunde-1	Control	3.23±1.25^{a}	5.23±1.43^{a}	6.97±1.71^{ab}
	Phosphorus	3.47±1.57^{a}	5.63±1.38^{a}	6.83±1.34^{ab}
	Lime	4.27±1.28^{ab}	5.70±1.15^{a}	7.40±1.52^{ab}
	P+L	4.63±0.99^{b}	6.10±1.06^{b}	8.33±1.62^{bc}

*Means in the same column with the same superscript are not significantly different ($P\leq0.05$). Data are means ± Standard Deviation from three replicates.
DAP= Days after Planting

4.5.3 Number of leaves

The number of leaves per plant differed significantly ($P\leq0.05$) among cultivars. The cultivar, UOE-COWPEA-2 consistently produced the highest number of leaves whereas KVU 27-1 had the least (Table 4.11). The number of leaves per plant also differed significantly with treatment application. Plots with P+L and P treatments produced the highest number of leaves when compared to control treatment for all the cultivars. At 90 DAP, the effect of P+L treatment was significantly higher compared to other treatments.

Table 4.11. Effect of treatments on the number of leaves per plant

Cultivar	**Treatment**	**60 DAP**	**75DAP**	**90DAP**
UOE-COWPEA-2	Control	5.67 ± 1.81^{a}	23.23 ± 5.72^{bc}	41.53 ± 11.42^{c}
	Phosphorus	7.33 ± 1.76^{b}	26.53 ± 6.21^{bc}	50.47 ± 10.91^{c}
	Lime	7.17 ± 2.56^{ab}	28.87 ± 7.52^{bc}	47.67 ± 14.30^{c}
	P+L	7.97 ± 7.97^{b}	29.43 ± 6.13^{c}	52.40 ± 18.67^{c}
KVU27-1	Control	4.23 ± 0.57^{a}	12.73 ± 3.05^{a}	19.87 ± 5.53^{a}
	Phosphorus	5.50 ± 1.66^{a}	17.10 ± 4.64^{ab}	29.87 ± 10.18^{ab}
	Lime	4.77 ± 1.79^{a}	15.53 ± 4.35^{a}	24.17 ± 7.82^{a}
	P+L	6.23 ± 2.38^{ab}	18.03 ± 4.83^{ab}	30.57 ± 13.73^{b}
K-80	Control	5.70 ± 1.89^{a}	19.67 ± 7.12^{b}	32.47 ± 11.85^{ab}
	Phosphorus	7.33 ± 2.03^{ab}	22.17 ± 6.58^{b}	30.36 ± 11.19^{ab}
	Lime	6.37 ± 2.14^{ab}	19.87 ± 5.81^{b}	36.97 ± 12.83^{b}
	P+L	8.63 ± 2.93^{c}	23.23 ± 6.57^{b}	38.87 ± 15.91^{b}
KENKUNDE-1	Control	5.23 ± 1.36^{a}	20.73 ± 5.07^{b}	36.97 ± 10.76^{b}
	Phosphorus	6.07 ± 1.76^{ab}	22.40 ± 5.74^{b}	37.10 ± 8.45^{b}
	Lime	6.10 ± 1.56^{ab}	26.63 ± 8.49^{bc}	46.43 ± 14.13^{c}
	P+L	7.63 ± 2.32^{bc}	29.00 ± 6.31^{c}	51.57 ± 12.79^{c}

***Means in the same column with the same superscript are not significantly different ($P\leq0.05$) ± Standard Deviation.**
DAP= Days after Planting

The control treatment had the least effect on number of leaves in all the sampling days for all the cultivars. Similarly, plots applied with phosphorus also produced plants with higher number of leaves. UOE-COWPEA-2 cultivar still produced the highest number of leaves and KVU 27-1 cultivar producing the least. Lime treatment also increased the number of leaves per plant but lower than P and P+L treatment and higher than control treatment. UOE-COWPEA-2 had higher number of leaves and the least KVU 27-1.

4.5.4 Leaf area

At 60, 75 and 90 DAP, leaf area for cultivars was significant throughout the sampling days (Table 4.12). At 60 DAP, a significant difference was observed among the cowpea cultivars on leaf area. At 90 DAP, the leaf area was higher compared to that measured at 75 and 60 DAP, with UOE-COWPEA-2 cultivar recording the highest (13.46 cm^2) and K-80 cultivar recording the least 9.45 cm^2.The leaf area did not differ significantly among the treatments.

Table 4.12. Effect of cultivars and treatments on leaf area in cm^2

Cultivar	Treatment	60DAP	75DAP	90DAP
UOE-COWPEA-2	Control	8.28 ± 0.86^{ab}	10.91 ± 1.65^{b}	12.87 ± 1.45^{b}
	Phosphorus	8.54 ± 1.79^{ab}	11.38 ± 1.72^{b}	11.61 ± 1.72^{b}
	Lime	8.68 ± 1.91^{ab}	10.38 ± 1.69^{b}	12.48 ± 2.41^{b}
	P+L	9.23 ± 1.21^{b}	11.61 ± 1.31^{b}	13.46 ± 1.38^{b}
KVU27-1	Control	6.45 ± 0.86^{a}	7.77 ± 1.19^{a}	9.55 ± 1.28^{a}
	Phosphorus	6.94 ± 1.18^{a}	8.51 ± 1.14^{a}	10.24 ± 1.64^{ab}
	Lime	6.54 ± 1.43^{a}	7.85 ± 1.37^{a}	9.79 ± 1.34^{a}
	P+L	7.31 ± 1.01^{a}	9.21 ± 0.96^{ab}	10.34 ± 1.19^{ab}
K-80	Control	6.91 ± 0.72^{a}	7.69 ± 0.69^{a}	9.45 ± 1.14^{a}
	Phosphorus	7.94 ± 1.16^{ab}	9.12 ± 0.65^{ab}	9.98 ± 1.05^{a}
	Lime	7.34 ± 0.63^{a}	8.78 ± 1.02^{ab}	9.77 ± 0.90^{a}
	P+L	8.11 ± 0.99^{ab}	9.46 ± 0.87^{ab}	10.17 ± 1.08^{ab}
KENKUNDE-1	Control	7.05 ± 1.17^{a}	8.49 ± 1.29^{ab}	9.99 ± 1.74^{ab}
	Phosphorus	7.65 ± 1.35^{a}	8.92 ± 1.92^{ab}	10.62 ± 1.18^{ab}
	Lime	7.99 ± 1.22^{ab}	9.65 ± 1.08^{ab}	9.86 ± 1.25^{a}
	P+L	8.75 ± 1.12^{ab}	10.24 ± 0.89^{b}	11.97 ± 0.68^{ab}

*Means in the same column with the same superscript are not significantly different ($P \leq 0.05$) ± Standard Deviation.

DAP= Days after Planting

However, plots that were applied with phosphorus had higher leaf area compared to control and lime treatment. At the three time points, plots that had combined phosphorus and lime had higher leaf area compared to control treatment for all the cultivars. The effect of phosphorus treatment on leaf area was similar to that of combined phosphorus and lime. The effect of lime treatment was similar to that of control treatment.

4.5.5 Dry matter production

Table 4.13, shows the results on effects of treatments on cultivar dry matter production, root dry weight, shoot dry weight, leaf dry weight and number of lateral roots. Treatments application had significant effect on dry matter production. Plots applied with P+L recorded higher plant dry biomass with the highest content in UOE-COWPEA-2 cultivar and the least in control treatment with KVU 27-1 cultivar. Similarly, the highest root dry weight was recorded by UOE-COWPEA-2 cultivar in plots applied with combined P and L and the least was recorded in control plots with KVU 27 -1 cultivar. Shoot dry weight was also higher in plots with P+L with UOE-COWPEA-2 cultivar followed with lime plots and plots with P alone with UOE-COWPEA-2 cultivar and the least in control treatment with KVU 27-1 cultivar. Combining P and L or L alone had also a significant effect on stem and leaf biomass. UOE-COWPEA-2 cultivar recorded the highest stem and leaf dry biomass while the control treatment recording the least with KVU 27-1 cultivar. Plots with P fertilizer recorded higher number of lateral roots followed by control treatment with K-80 cultiva and was the least in plots with lime alone for KVU 27-1 cultivar.

Table 4.13. Effect of cultivars and treatments on dry matter accumulation

Cultivar	**Treatment**	**Plant Biomass (g)**	**Root Biomass (g)**	**Shoot Biomass (g))**	**Stem Biomass (g)**	**Leaf Biomass (g)**	**Number of Lateral Roots**
UOE-COWPEA-2	Control	29.18±14.38^{b}	3.38±2.38^{b}	25.36±12.70^{b}	14.64±8.93^{b}	9.23±3.72^{a}	16.33±5.10^{a}
	Phosphorus	41.63±14.31^{c}	3.54±1.59^{b}	37.92±13.44^{c}	21.89±9.87^{c}	16.84±5.44^{b}	17.11±4.76^{a}
	Lime	49.87±22.08^{c}	5.10±2.38^{b}	45.05±18.48^{c}	26.73±14.18^{c}	18.34±4.86^{b}	15.67±2.69^{a}
	P+L	77.29±35.36^{d}	7.03±3.04^{b}	70.15±33.38^{d}	44.92±15.69^{d}	24.64±9.86^{b}	15.00±6.32^{a}
KVU27-1	Control	5.94±2.66^{a}	0.86±0.42^{a}	5.77±3.32^{a}	1.74±0.75^{a}	3.29±1.47^{a}	18.00±6.63^{a}
	Phosphorus	14.12±2.55 ^{b}	1.88±0.73^{b}	12.18±6.28^{b}	6.42±4.86^{a}	5.59±2.66^{a}	19.67±2.60^{a}
	Lime	18.00±9.25^{b}	2.64±0.97^{b}	14.21±9.36^{b}	8.05±6.91^{a}	7.35±3.03^{a}	15.22±5.04^{a}
	P+L	22.83±2.36^{b}	2.70±2.23^{b}	20.06±19.19^{b}	10.64±1.63^{a}	8.72±6.21^{a}	17.78±7.01^{a}
K-80	Control	10.78±6.18^{a}	2.31±1.53^{b}	8.86±4.10^{a}	3.49±2.12^{a}	5.91±2.99^{a}	20.33±6.12^{a}
	Phosphorus	19.00±11.69^{ab}	2.85±1.80^{b}	16.22±10.33^{ab}	8.38±6.70^{a}	7.75±3.95^{a}	21.22±7.51^{a}
	Lime	25.39±18.32^{b}	3.54±2.00^{b}	21.68±16.23^{b}	11.63±11.78^{b}	9.81±4.74^{a}	16.22±4.84^{a}
	P+L	24.19±11.38^{b}	3.85±1.68^{b}	20.28±9.81^{b}	10.66±5.26^{ab}	9.38±4.76^{a}	17.78±3.63^{a}
Ken-Kunde-1	Control	27.75±17.21^{b}	2.46±1.75^{b}	24.99±16.18^{b}	13.85±13.02^{ab}	9.84±5.45^{a}	14.67±7.78^{a}
	Phosphorus	29.67±15.74^{b}	2.71±1.33^{b}	26.98±15.29^{b}	15.27±11.99^{ab}	10.75±4.98^{a}	16.89±4.46^{a}
	Lime	42.85±24.19^{cb}	3.70±2.06^{b}	39.16±22.19^{c}	21.42±11.99^{c}	12.41±5.84^{ab}	16.33±6.60^{a}
	P+L	42.02±12.31^{c}	3.66±2.39^{b}	38.08±11.44^{c}	21.09±9.91^{c}	13.71±5.11^{b}	17.89±4 .59^{a}

*Means in the same column with the same superscript are not significantly different ($P \geq 0.05$) ± Standard Deviation.

4.6 Effect of Lime and Phosphorus Application on Yield and yield components

4.6.1 Number of pods per plant

There was a significant difference observed (P≤0.05) among the cultivars on number of pods per plant (Table 4.14). At control, K-80 cultivar produced the highest number of pods per plant whereas KVU 27-1 cultivar produced the least. The number of pods per plant also differed significantly (P≤0.05) among the treatments. The control treatment produced significantly lower number of pods compared to plots with P and a combination of P+ L treatments. The effect of phosphorus treatment on number of pods per plant was most pronounced in UOE-COWPEA-2 and K-80. The effect of lime treatment on number of pods per plant was similar to that of control treatment. The effect of combined phosphorus and lime on the number of pods per plant was higher compared to control and lime treatment, and the cultivars UOE-COWPEA-2 and K-80 still were outstanding (Table 4.14).

4.6.2 Pod length

Cowpea cultivars showed significant variations in mean pod length (Table 4.14). Pod length was significantly different among the cowpea cultivars. KVU 27-1 had the longest and KEN-KUNDE-1 had the shortest. There was no significant effect (P≤0.05) of treatment on pod length.

Table 4.14. Effect of treatments on number of pods per plant, pod length and pod dry weight

Cultivar	**Treatment**	**PN**	**PL(cm)**	**PDW(g)**
UOE-COWPEA-2	Control	27.10 ± 10.03^{b}	13.48 ± 2.24^{ab}	0.650 ± 0.39^{b}
	Phosphorus	42.77 ± 8.72^{bc}	14.49 ± 4.84^{ab}	0.557 ± 0.16^{ab}
	Lime	28.47 ± 8.16^{b}	14.82 ± 2.17^{ab}	0.626 ± 0.17^{b}
	P+L	59.00 ± 13.25^{c}	14.04 ± 2.16^{ab}	0.582 ± 0.19^{ab}
KVU27-1	Control	18.93 ± 10.02^{a}	14.48 ± 2.43^{ab}	0.631 ± 0.21^{b}
	Phosphorus	29.30 ± 8.26^{ab}	15.25 ± 7.25^{b}	0.642 ± 0.58^{b}
	Lime	19.17 ± 6.92^{a}	15.05 ± 2.35^{b}	0.686 ± 0.20^{b}
	P+L	36.97 ± 11.05^{b}	14.65 ± 2.61^{ab}	0.693 ± 0.22^{b}
K-80	Control	33.60 ± 7.87^{a}	13.23 ± 1.72^{ab}	0.497 ± 0.13^{a}
	Phosphorus	42.23 ± 10.98^{b}	13.01 ± 1.72^{ab}	0.497 ± 0.12^{a}
	Lime	35.57 ± 8.82^{a}	13.67 ± 1.78^{ab}	0.779 ± 0.89^{c}
	P+L	56.2 ± 13.99^{c}	13.73 ± 1.51^{ab}	0.535 ± 0.12^{ab}
Ken-Kunde-1	Control	21.07 ± 6.97^{ab}	11.79 ± 1.79^{a}	0.738 ± 0.32^{abc}
	Phosphorus	31.33 ± 6.65^{b}	12.13 ± 1.62^{a}	0.544 ± 0.13^{ab}
	Lime	21.00 ± 5.22^{ab}	12.26 ± 1.48^{a}	0.605 ± 0.13^{b}
	P+L	42.80 ± 10.99^{bc}	12.59 ± 1.57^{a}	0.615 ± 0.14^{b}

*Means in the same column with the same superscript are not significantly different ($P\leq0.05$) ± Standard Deviation. **PN**= pod number, **PL**= pod length and **PDW**= pod dry weight.

4.6.3 Number of seeds per pod, hundred seed weight and total seed weight per plant.

The results for the number of seeds per pod and hundred seed weight are shown in Table 4.15. There was no significant difference observed among the cultivars and treatments on the number of seeds per pod. However, KEN-KUNDE-1 cultivar recorded a higher number of seeds per pod compared to other cultivars. There was also no significant effect ($P\leq0.05$) of treatments on the number of seeds per pod. A significant difference was observed among the cowpea cultivars on seed weight per plant with K-80 cultivar recording higher whereas

KVU 27-1 recorded the least. There was a significant effect ($P \leq 0.05$) of treatments on seed weight per plant. Combined phosphorus and lime recorded higher total seed weight per plant compared to lime, phosphorus and control treatments in all the cultivars with K-80 cultivar recording higher and the least with KVU 27-1 cultivar.

Table 4.15. Effects of cultivar and treatments on seed yield

Cultivar	**Treatment**	**NSP**	**100SWPC(g)**	**Total SWPP(g)**
UOE-COWPEA-2	Control	9.87 ± 2.97^{a}	12.03 ± 2.36^{a}	29.21 ± 6.04^{a}
	Phosphorus	10.44 ± 2.51^{a}	12.98 ± 1.32^{a}	36.72 ± 4.14^{b}
	Lime	10.82 ± 2.62^{a}	12.44 ± 1.37^{a}	40.70 ± 1.46^{c}
	P+L	10.99 ± 2.54^{a}	11.41 ± 1.33^{a}	41.23 ± 7.88^{b}
KVU27-1	Control	10.59 ± 2.93^{a}	11.23 ± 2.79^{a}	21.93 ± 3.53^{a}
	Phosphorus	10.95 ± 3.05^{a}	13.44 ± 1.34^{a}	28.82 ± 6.85^{a}
	Lime	11.73 ± 2.95^{a}	12.42 ± 1.29^{a}	22.65 ± 1.27^{a}
	P+L	11.19 ± 3.33^{a}	11.47 ± 1.48^{a}	30.73 ± 6.85^{a}
K-80	Control	11.13 ± 2.76^{a}	11.66 ± 0.94^{a}	34.02 ± 3.62^{b}
	Phosphorus	11.03 ± 2.85^{a}	11.57 ± 0.92^{a}	38.15 ± 6.16^{b}
	Lime	12.00 ± 8.32^{a}	11.13 ± 0.90^{a}	40.76 ± 4.04^{c}
	P+L	11.80 ± 2.25^{a}	10.97 ± 0.76^{a}	42.76 ± 2.98^{b}
Ken-Kunde-1	Control	12.04 ± 2.68^{a}	13.35 ± 1.63^{a}	31.66 ± 2.81^{b}
	Phosphorus	12.05 ± 2.71^{a}	13.55 ± 1.67^{a}	33.52 ± 5.75^{b}
	Lime	12.50 ± 2.55^{a}	13.61 ± 1.11^{a}	33.29 ± 4.18^{b}
	P+L	12.27 ± 2.64^{a}	13.08 ± 1.59^{a}	41.48 ± 4.92^{b}

*Means in the same column with the same superscript are not significantly different ($P \geq 0.05$) ± Standard Deviation.**NSP=** Number of seed per pod. **100SW=** Weight of 100 seeds per cultivar and **Total SWPP**= Total Seed Weight per plant

CHAPTER FIVE

DISCUSSION

5.1 Culture solution pH changes

All the nine cowpea cultivars screened increase the pH of the culture solution as the days progressed, but the magnitude of pH change differed by cultivar. This suggests genetic variation in rhizosphere chemistry due to secretion of chelating agents that reduce H^+ thereby increasing the culture solution pH and reducing Al toxicity with UOE-COWPEA-5 showing the strongest alkalinizing effect and therefore greater potential for Al-detoxification. Yang *et al*., (2019) attributed such an increase in pH to the relative higher Al-detoxification capability of some genotypes when compared to those that cause smaller changes in pH of the growth medium. The independence of the responses to acidity and aluminium toxicity among cowpea cultivars is also in agreement with the findings of Kidd and Proctor (2001), who showed that plant species adapt differently to H^+ and Al^{3+} toxicity as a result of the difference in the nature of soil parent materials and where the species originated. Pinheiro de Carvalho *et al*. (2003) also noted that there were significant differences among cowpea genotypes on rhizosphere pH modification upon exposure to 100 µM and 200 µM Al.

Cowpea root growth was significantly inhibited after exposure to Aluminium concentration of 185µM Al. This is attributed to the fact that Al toxicity limits cell expansion and cell division inhibiting root elongation in plants. Kochian *et al*., (2024) reported on the detrimental effects of Al toxicity on crop growth, including rapid inhibition of root elongation. The inhibition may be attributed to excess Al binding tightly to the cell walls of plant root cells impacting root development, (Singh *et al.*, 2017).

The substantial cultivar difference in RNRL highlights their tolerance to Al stress. Cowpea cultivars with a higher relative net root length namely, UOE-COWPEA-4, KVU 27-1, Ken-Kunde-1 and UOE-COWPEA-2 were classified as tolerant to low pH and Al toxicity while those with lower RNRL namely, UOE-COWPEA-1 and UOE-COWPEA-5 were classisied as moderately tolerant. The results of the study are in line with the report of Aguilera *et al*., (2016) who reported that RRL was strongly and negatively correlated with soil exchangeable Al and used it to differentiate between sensitive and tolerant wheat cultivars, with sensitive cultivars exhibiting the lowest RRL. Negusse *et al*., (2022) also demonstrated that chickpea RRL was significantly affected by varying Al rates a and was a reliable criterion for distinguishing tolerant from sensitive varieties.

There was no significant effect of aluminium concentration on cowpea shoot elongation. Giannakoula *et al*. (2008) and Giongo and Bohnen (2011) reported that in the presence of Al^{3+}, P is precipitated in the root apoplast, reducing Al translocation to the aerial parts of the plant, resulting in little effect of Al concentrations on shoot elongation. Through the mechanism, P nutrient inhibit Al effects in the root system, favoring seedling growth and greater accumulation in the root. Mattiello *et al.* (2008) also observed similar results while studying root growth and P and Al absorption in coffee plants, which they concluded that the accumulation of Al in the root system and restriction of its transport to the shoots are important factors in relation to plant tolerance to aluminum, providing evidence that the Al element can be accumulated in the roots, preventing its toxicity from reaching other plant parts (Grifferty and Barrington, 2000).

Aluminium stress significantly reduced plant biomass. Aluminium concentration had a negative significant effect on dry root and shoot biomass in all the cultivars. The reduced root biomass could be associated with damage to root cell wall and plasma membrane impairing nutrient uptake (Kochian *et al*., 2015; Rahman *et al* 2018) in the cowpea cultivars. Hayes *et al*., (2020) reported that $AlCl_3$ reduced root biomass by 22.3% and 9.96%. Qu *et al*. (2020) also reported that Al toxicity reduced root length, diameter, volume and overall plant biomass by hindering protein biosynthesis and reducing carbohydrate content in Al stressed *Camellia oleifera Abel*. Other researchers also observed that Al ions interact with absorption, translocation, allocation, and metabolic activity of nutrients such as Ca, N, K, Mg, P, Mn, Fe, Cu and B (Ren *et al*., 2022; Tscuchiya *et al.*, 2021; Xia *et al*.,2020).

5.2 Soil Chemical properties

The soil pH in the field ranged between 4.6 to 5.3 against 5.5 to 6.5 suitable for cowpea production (Osipitan *et al*., 2021). These results are indications that soils are acidic and need to be corrected through sapplication of agricultural lime application to neutralize the acid effects, for enhanced nutrient availability and optimum crop yield. The high acidity reported here agree with the findings of Okalebo *et al*. (2002) and Osundwa *et al*., (2013) who reported low soil pH of 5.0 at the University of Eldoret farms and its environs.

5.2.1 Effect of cultivars and treatments on soil pH and mineral nutrients

Results showed that cowpea cultivars, application of phosphorous, lime and phosphorous + lime treatments increased the soil rhizosphere pH which in turn led to an increase in the amount of nitrogen, available P, soil organic carbon and exchangeable cations in the soil. Cowpea cultivars increased the soil rhizosphere pH. These could be attributed to the activity of nitrogen-fixing bacteria in cowpea root nodules which releases hydroxide ions (OH^-) into the soil through the process of nitrogen fixation and helps neutralize acidity and increase the soil pH (Vasconceles *et al*., 2020). Cowpea roots also are known to release organic acids such as malic and oxalic acids into the rhizosphere especially in acidic soils. These exudates increase the availability of nutrients and stimulates the growth of microorganisms which influenced the soil pH through metabolic processes hence increase in soil pH. The results of the study are in agreement with the findings of (Menezes-Blackburn *et al*., 2016) who observed that the exudation of organic acids stimulates microbial activity in the rhizosphere, which contributes to increased P availability. Increased rhizosphere pH led to an increase exchangeable mineral nutrient elements. The increase in soil total N observed under control treatment was also attributed to the addition of organic matter to the soil with the falling of leaves from cowpea cultivars. Amba *et al.* (2011) recorded a significant increase in total nitrogen content as a result of addition of organic matter to the soil with the falling of leaves. Cowpea cultivars also were able to increase soil organic C which might be due to the falling of leaves and their decomposition at physiological stage. Amba *et al*., (2011) reported an increase in soil C and attributed the increase to the falling of leaves which added organic C to the soil.

Application of P alone from TSP fertilizer increased soil pH, total nitrogen, available phosphorus, soil organic carbon and exchangeable cations in the present study. Increase in soil pH after P application without lime might be attributed to contribution of Ca from TSP ($Ca(H_2PO_4)_2$). Application of P also led to increased total soil N. This is because application of P to the soil promotes the colonization and activity of rhizobacteria which in turn increases the amount of immobilized N in the soil. High total soil N due to P fertilization was also reported by Zhang *et al.*, (2024) and Hussain *et al*., (2021) who observed an increase in total N when P fertilizer was applied compared to the control.

Lime application significantly increased the soil pH which in turn increased the contents of total nitrogen, soil available P, soil organic carbon and exchangeable Ca, Mg and K. This is because lime applied chemically reacts with soil acids, causing the H^+ to be neutralized and resulting in the production of water and carbon dioxide, increasing the soil pH. The results of the study are similar to the studies of Ameyu (2019) who reported an increased soil pH under lime treatment. Lime treatment enhances the availability of nutrients, total nitrogen, available phosphorus and soil organic carbon by increasing the soil pH and improving the solubility of nutrients. Gurmessa *et al*. (2021) reported that lime application increased soil pH by neutralizing soil acidity enhancing P availabilty. Cowpea is also characterized by having the ability to host rhizobacteria, which contribute to the fixation of nitrogen for legume crops hence increasing the amount of N in the soil (Sun *et al*., 2019 ; Lengwati *et al*., 2020). Soils that are acidic make it difficult for beneficial bacteria to thrive and fix atmospheric nitrogen in the soil. A tolerant cowpea cultivar can still establish symbiosis and effective N-fixation under acidic soil conditions. In the absence of tolerance liming would be crucial for enhanced crop production in the acidic soils. Liming of acid soils raises the soil pH which in turn facilitates the release of phosphate ions into the soil solution (Kisinyo, 2016) making it available for plant absorption. Liming increases the availability of P by mineralizing organic P, and makes P more readily available to plants (Ameyu, 2019). The significant increase of soil organic C after lime application also might be associated to general improvement of soil condition which might have enhanced proliferation of soil microbial biomass and activity in the soil. Application of lime led to a small increase in the exchangeable cations. This is attributed to the increase in soil pH and the release of the initially occluded amorphous and inter layer substitutive charge via Ca ions which induced deprotonation of the variable charge minerals and functional groups of humic substances (Ameyu, 2019). The negative charges of the soil exchange complex are displaced by basic cations due to neutralization of a portion of the soil's acidity as a result of lime application (Achalu *et al.,* 2012). Jafer and Hailu (2017) reported a small increase in exchangeable Mg and K when lime was applied to acidic soils.

Combined application of lime and phosphorus resulted in a significant pH increase which also increased the contents of total nitrogen, soil available P and soil organic carbon. Increase in exchangeable Ca was due to its release from the liming material and TSP fertilizer. Kiboi *et al*. (2010) and Kisinyo *et al*. (2012) also reported that application of combined Lime and P increases the soil rhizosphere pH which increased the soil available phosphorus. This might be due to the residual effect of phosphorus fertilizer because of its low mobility in the soil and also lime applied contributed to raising soil pH thus increased P availability. Gupta. (2011) reported on low mobility of P in acid soils.

5.3 Effect of acidic P deficient soil on cowpea growth

Cowpea plant height was a cultivar dependent trait as certain cultivars were taller than others even upon application of treatments. The plant height values determined in the study were within the ranges reported by Aboyomi *et al*. (2008) who reported that cowpea plant height was between 20.21 and 59.12 cm but lower than the plant height values reported by Ugur *et al*. (2011) who found out that plant height values was between 101.0 cm and 122.4 cm in cowpea genotypes.

Phosphorus application increased the plant height of cowpea cultivars which could be attributed to the fact that phosphorus is an essential element in plants that is required in large quantities in shoot and root tips of growing plants where cell division and metabolism is higher (Ndakidemi and Dakora, 2007). This result is in conformity to those observed by Nkaa *et al*. (2014) and Nyoki *et al*. (2013).

The number of branches per plant in the study varied significantly ($p<0.05$) with the cowpea cultivar. Variation in the number of branches per cultivar could be due to genetic variation of the cultivars (Magani and Kuchinda, 2009). Karikari and Arkoful (2015) also observed that there was a varietal difference in the number of branches per plant throughout the experimental period. Treatment effect was also significant on the number of branches produced per plant, where greater number of branches was observed on cultivars treated with combined phosphorus and lime and phosphorus alone. This could be attributed to increased soil pH as a result of liming which ensured the availabilty of phosphorus in the soil solution. Nkaa *et al*. (2014) observed that phosphorus application significantly increased the number of branches per plant, however, this observation is contrary to that of Karikari and Arkoful, (2015).

The number of trifoliate leaves per plant differed significantly ($p<0.05$) with cultivar and treatment application. This is because lime helps to improve soil pH, making phosphorus more available for uptake thus promoting leaf growth and production. The results in the study is in conformity to that of Ayodele and Oso, (2014) who observed and reported that phosphorous application increased the number of leaves per plant however, it is in contrary to that of Karikari and Arkoful, (2015).

The leaf area was significantly influenced by cultivars and treatments. Application of combined phosphorous and lime led to larger leaf areas per plant. This can be explained by improved nutrient availabilty especially phosphorus which allows for greater leaf expansion and development. The results in the study are in conformity to the results observed by Ayodele and Oso (2014).

Total dry matter production varied significantly among the cowpea cultivars. This shows that the cultivars had unequal growth and dry matter production potential. Dry matter production is influenced by genotype. The results of the study are similar to the report of Addo-Quaye *et al*. (2011); Kana *et al*. (2021) and Karikari and Arkorful (2015).

Treatment had a significant effect on the dry matter production. The results of the study indicate that the interactions of cultivar with combined phosphorus and lime had greater dry matter production compared with other treatments. The increase in total dry biomass in response to applied lime and phosphorus is attributed to the fact that, liming of acidic soils increased the soil pH. This enhances the availability of essential elements such as P, N, Ca and Mg for plant uptake thus increased vegetative growth and increased dry matter production (Matthew and Thampatti, 2007). Sarker *et al*. (2014) reported that application of combined lime and TSP increased shoot and dry weights of cowpea cultivars.

Dry matter partitioning in the above ground parts such as leaves and stem varied significantly among the cowpea cultivars. This could be attributed to phenological difference among the varieties. Dry matter production was higher in plots applied with combined phosphorus and lime. Increased dry matter production due to liming and applied P on above ground parts is attributed to the fact that lime ameliorated soil acidity and increased the available P along with applied P which had a positive effect on vegetative growth reflected in number of leaves, number of branches and leaf area of cowpea cultivars. Cowpea cultivars had greater accumulation of assimilates in the stem than in the leaves. Variation in dry matter distribution among the ground parts confirms that dry matter partitioning among the sinks of a plant is primarily regulated by the sinks themselves (Marcelis, 1996).

5.4 Effect of cowpea cultivar and treatments application on yield attributes

Number of pods per plant significantly varied among cultivars. K-80 cultivar produced the highest number of pods per plant while KVU 27-1 cultivar produced the least number of pods per plant. Plots with combined phosphorus and lime and phosphorus alone produced the highest number of pods per plant with UOE-COWPEA-2 and K-80 cultivar recording a higher number of pods per plant compared to the other cultivars. This is because phosphorus is important in photosynthesis, pod development and grain filling in leguminous crops hence responsible for nodulation in cowpea. The results in this study are in conformity to the results observed by Magani and Kuchinda (2009) where the number of pods per plant increased as a result of P application.

Cultivars and treatment application had no significant effect on pod length, number of seeds per pod and 100-seed weight per plant. Though, KENKUNDE-1 cultivar produced a higher number of seeds per pod but was not statistically different from the other cultivars while UOE-COWPEA-2 cultivar produced the least. The number of seeds per pod could be attributed to genetic variation of the varieties (Acquah, 2007). Application of treatments also had no significant effect on number of seeds per pod although the values recorded under combined P and L, P alone and L alone were higher compared to the control. KENKUNDE-1 cultivar had higher 100-seed weight while KVU 27-1 cultivar had the least but they were not statistically different from ecah other. These results were found to be in conformity with those of Agboola and Obigbesan (2001) who reported that phosphorus application did not significantly influence cowpea yield and yield components. Cultivars and treatment application had a significant effect on seed weight per plant. K-80 cultivar recorded higher seed weight per plant at control treatment. This could be attributed to the genetic variation of the cultivars. Application of combined phosphorus and lime with K-80 cultivar recorded higher seed weight per plant with the control treatment with KVU 27-1 cultivar producing the least values. This is attributed to the fact that liming the soil increased soil pH which led to availabilty of P in the soil and the applied P which is important in seed formation and grain filling (Haruna, 2011). This compares favorably with the report of other researchers (Ndor *et al*., 2012; Haruna and Usman, 2013) who reported a significant increase in seed weight per plant of cowpea in response to P application.

CHAPTER SIX

CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions

These findings confirm that certain Kenyan cowpea cultivars, UOE-COWPEA-2, UOE-COWPEA-5, KEN KUNDE-1 and K-80 possess traits that confer resilience to soil acidity associated with Al toxicity and phosphorus deficiency, making them promising candidates for cultivation in acid soils. This study provides the first systematic characterization of Kenyan cowpea cultivars for tolerance to aluminium stress and phosphorus deficiency in acidic soils.

6.2 Recommendations

1. Farmers in acidic regions should be encouraged to cultivate UOE-COWPEA-2, UOE-COWPEA-5, KEN KUNDE-1 and K-80, combined with lime and phosphorus application where resources allow.

2. County governments and seed agencies should fast-track multiplication and dissemination of these tolerant cultivars.

3. Breeding programs should incorporate these tolerant cultivars into crossing programs for developing low P-acid soil adapted cultivars.

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APPENDICES

Appendix I: Hoagland Nutrient Solution

i) Composition of the modified 1/4 strength Hoagland* nutrient solution for P screening

Compound	Molecular Weight	Concentration of stock solution	Conc. stock soln.	Vol. of stock soln L^{-1} of final solution
	g mol^{-1}	mM	g L^{-1}	mL
Macronutrients				
KNO_3	101.10	1000	101.10	1.0
$Ca(NO_3).4H_2O$	236.16	1000	236.16	1.0
NH_4NO_3	80.04	1000	80.04	0.5
$MgSO_4.7H_2O$	246.48	1000	246.48	0.25
KH_2PO_4	136.09	1000	136.09	0,0.1or 0.32**

Micronutrients

KCl	74.55	25	1.864
H_3BO_3	61.83	12.5	0.773
$MnSO_4.H_2O$	169.01	1.0	0.169
	0.5		
$ZnSO_4.7H_2O$	287.54	1.0	0.288
$CuSO_4.5H_2O$	249.68	0.25	0.062
$NaMoO_4. 2H_2O$	241.95	0.25	0.060
NaFeEDTA	367.00	53.7	19.71
	1		

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* **Source**: Modified from Taiz & Zeiger 1998.

**: To reflect the P treatments of 0 (-P), 160 µM (+P) or 320 µM (++P)

Notes: 1. The macronutrients are added separately from stock solutions to avoid precipitation during preparation of the nutrient solution. A combined stock solution is made up containing all micronutrients except iron. Iron is prepared and added to the nutrient solution separately.

2. Adjust the pH of the final solutions accordingly

3. Adjust the P levels accordingly for the germination experiment

ii) Composition of a modified Hoagland*nutrient solution for growing plants

Compound	molecular weight $g\ mol^{-1}$	Concentration of stock solution mM	Conc. stock soln. $g\ L^{-1}$	Vol. of stock soln L^{-1} of final solution mL
Macronutrients				
KNO_3	101.10	1000	101.10	1.2
$Ca(NO_3).4H_2O$	236.16	1000	236.16	0.8
$NH_4H_2PO_4$	115.08	1000	115.08	0.4
$MgSO_4.7H_2O$	246.48	1000	246.48	0.2
Micronutrients				
KCL	74.55	25	1.864	
H_3BO_3	61.83	12.5	0.773	
$MnSO_4.H_2O$	169.01	1.0	0.169	0.4
$ZnSO_4.7H_2O$	287.54	1.0	0.288	
$CuSO_4.5H_2O$	249.68	0.25	0.062	
H_2MoO_4	161.97	0.25	0.040	
NaFeEDPTA	558.50	53.7	30	0.3

* **Source**: Taiz & Zeiger 1998.

Appendix II. Preparation of Chemical Reagents for Soil Chemical Analysis from Analytical Reagents

[3]0.42 g of selenium powder was mixed with 14 g of lithium sulphate and placed in 350 ml of 30% H_2O_2. The suspension was thoroughly mixed and 420 ml of concentrated H_2SO_4 was carefully added while cooling in ice bath. The resultant reagent was stored in bottles at $2\text{-}4^0$ C.

[4]8.1 ml conc. HCl was put in 1 litre of distilled water to make 0.1N HCl and diluted to give N/140 HCl. 73 ml of 0.1N HCl diluted to 1 litre results in **N/140 HCl.**

[5]400g NaOH was dissolved in distilled water and diluted to water to make 40% NaOH.

[6]10 g of boric acid was dissolved in distilled water and diluted to 1 litre to make1% Boric acid.

[7]0.99g bromocresol green, 0.066g methyl red and 0.011g thymolblue were dissolved in 100 ml ethanol to form the **mixed indicator**.

[8]42 g of $NaHCO_3$ was dissolved in 1 litre of distilled water to make o.5 M sodium bicarbonate at a pH of 8.5. The pH was adjusted to 8.5 using 1 M sodium hydroxide solution prepared by dissolving 40 g of NaOH in 1 litre of distilled water. 49.4g of boric acid powder was dissolved and diluted to 1 litre with distilled water to make 0.8M boric acid.

Appendix III. Similarity Report

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Certificate of Plagiarism Check for Thesis

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