

RELATIVE

A THESIS

THE AWAR

SUITABILITY OF PHOSPHORUS ANALYTICAL METHODS FOR A FERRALSOL

Emmanuel Odama

BSc. Agriculture (Hons) Makerere University

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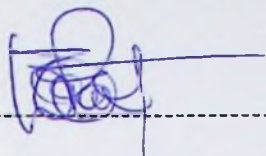


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DECLARATION

I, Emmanuel Odama, hereby declare that this thesis is my original work, and has never been presented to this University or any other university for any award of a degree.

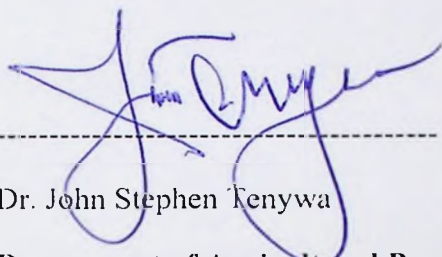


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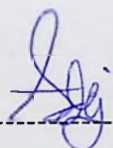
Dr. John Stephen Tenywa

Department of Agricultural Production

Makerere University

29/11/12

Date



Dr. Alice Amoding Katusabe

Department of Agricultural Production

Makerere University

29/11/2012

Date

DEDICATION

To the Father of my Lord Jesus Christ for His unmerited Love and Grace

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ABSTRACT

Declining soil fertility is a fundamental impediment to agricultural growth and a major reason for the slow growth in food production in sub-Saharan Africa. Among the primary plant macronutrients, phosphorus is the most important nutrient element, limiting agricultural production. In order to manage soil phosphorus objectively, the existing stocks in the soil should be known. It is on this basis that informed effective recommendations for intervention can be made. However, among the challenges in achieving reliable information lies the use of the most efficient analytical methods that simulate closely with plant nutrient uptake and growth. The most widely used soil P extractants are Bray I, Bray II, Olsen, Mehlich I, II and III. These methods were largely developed targeting temperate soils and eventually extended for use under tropical soils without the necessary evaluation for relative effectiveness and efficiency. This study was, thus, geared to evaluating the common methods of testing for soil available P in laboratories in Uganda so as to identify the most appropriate method. Treatments administered included six P rates namely, 0 (Control), 20, 40, 60, 80 and 100 kg P ha⁻¹. Three soil phosphorus extraction methods used included Bray I, Bray II and Mehlich III. The experiment was set in a greenhouse at Abi Zonal Agricultural Research and Development Institute, Arua, Uganda. Maize variety Longe V was used as the test crop. Data collected were analysed with the GenStat software, and correlations were run between available P fractions extracted using the different methods against dry matter and plant growth parameters. Mehlich III presented the highest correlation coefficient with stem girth ($r = 0.63$), number of leaves ($r = 0.733$) and plant P ($r = 0.254$). There was a strong positive correlation between Bray I and Mehlich III methods ($r = 0.975$), thus either could be used to quantify extractable P in Ferralsols with mica, kaolinite and quartz mineralogy of 1.57% organic carbon and pH 5.6.

CHAPTER ONE

INTRODUCTION

1.1 Background

Declining soil fertility is a fundamental impediment to agricultural growth and a major reason for the slow growth in food production in sub-Saharan Africa (Henao and Baanante, 2006). Among the primary plant macronutrients limiting agricultural production in most regions of the world is phosphorus (Hofman and Cleemput, 2004) and, thus its efficient use is a pending task in sustainable agriculture. Unlike nitrogen that can be naturally replenished by legumes in symbiosis with rhizobia, phosphorus can only be sourced externally, thus its deficiency is a major constraint to agricultural production in many regions of Africa (Henao and Baanante, 2006).

Phosphorus deficiency occurs in soils with high fixation capacities (Warren, 1992), and in tropical soils, it occurs also largely due to low inherent presence of P-containing minerals (Bunemann, 2003). Continuous and intensive cropping without commensurate nutrient replenishment is also reported to exacerbate the low P content of many soils (Sanchez, 2002). This situation is particularly severe in East Africa where human population continues to surge against diminishing resources. For example, Smaling *et al.* (1997) and Henao and Baanante (2006) reported that in East Africa, nutrient depletion ranges from high to very high with alarming figures in countries such as Kenya, Ethiopia, Uganda and Rwanda where annual depletion rates exceeded 66 kg P ha^{-1} . Beside the high nutrient depletion, soils in East Africa are also dominated by acid phosphorus-fixing soils in the order of Ferralsols, Acrisols and Nitisols

(Deckers 1993), and phosphorus becomes a major limiting nutrient for crop production in many soils across the region.

Yost and Eswaran (1990) reported that P-adsorbing Ferralsol is a dominant soil type occupying about 70% of Uganda; on which most of the farming is practiced. According to Sanchez *et al.* (1989), Ferralsols are rich in sesquioxides, which are major actors in fixing P in forms that are plant-unavailable. Also, they consist of low-activity clays with low cation exchange capacity and base saturation (FAO, 2006).

1.2 Statement of the problem

In order to manage soil P objectively, it is imperative that the existing stocks in the soil are known. It is on this basis that informed effective recommendations for intervention can be made. However, the challenge in achieving reliable information lies in the use of the most efficient analytical methods that simulate closely with plant nutrient uptake and utilisation. The most widely used soil P tests are Bray I (Bray and Kurtz, 1945), Mehlich I (Nelson *et al.*, 1953), and Olsen (Olsen *et al.*, 1954). Other common extraction procedures include Bray II, Mehlich II and III, and Resin (Fixen and Grove, 1990). These methods were developed targeting largely temperate soils and eventually extended for use under tropical soils without pre-evaluation for relative effectiveness and efficiency. Most of the tropical soils are unique in physical and chemical properties in that (i) there is excessive weathering and leaching, which contrast with the limited weathering and leaching in the temperate soils; and (ii) tropical soils are largely rich in sesquioxides, which are principal in fixing P, while temperate soils are rich in the silicate clays and are high in cation exchange capacity (CEC). (iii) Furthermore, although temperate soil pH may range from high acidity to alkaline, most soils there have pH values > 5.5. The reverse is

true of most soils in the humid tropics where the pH values are < 5.5 , due to high values of exchangeable Al^{3+} , thus contributing to P fixation.

Owing to the several P extraction methods being employed in Uganda, soil phosphorus analysts provide widely dissimilar results across different laboratories in the country and most parts of sub-Saharan Africa. As a result, comparison of results and sharing of equipment across laboratories is not feasible. This has ultimately made it impossible to interpret results uniformly among laboratories, yet all these efforts target the same farming communities. Consequently, synergies for sharing soil P analysis results and other P related information are basically lacking across soil laboratories in most parts south of the Sahara. Therefore, the need to evaluate the different methods for available P analysis widely used cannot be over-emphasised. This study was, thus geared to evaluating the common method(s) of testing for soil extractable P in laboratories in Uganda so as to identify the most appropriate method(s).

1.3 Objective

The objective of this study was to identify the most suitable method for determination of soil extractable phosphorus for a Ferralsol in Uganda

CHAPTER TWO

LITERATURE REVIEW

2.1 Soil phosphorus pools

Soil extractable phosphorus analysis is paramount because phosphorus provides plants with a means of using the energy harnessed by photosynthesis to drive its metabolism (Hibberd *et al.*, 2008). Phosphorus occurs in soil in several inorganic and organic forms (Troeh and Thompson, 2005). Inorganic phosphorus includes apatitic minerals, secondary precipitates formed with Ca, Fe and Al and free phosphate ions (H_2PO_4^- , HPO_4^{2-} , PO_4^{3-} all denoted here as $\text{PO}_4\text{-P}$) attached to sorption surfaces or dissolved in the soil water (Turner *et al.*, 2002). Organic phosphorus (Po) includes a group of organic molecules having P as a part of their structure. Orthophosphate esters are Po compounds having an ester linkage joining $\text{PO}_4\text{-P}$ to organic moiety and they are further divided into mono- and di-esters according to the number of ester groups attached to each $\text{PO}_4\text{-P}$. The largest group of Po in most soils is the mono-esters (Turner *et al.*, 2006).

Although analysis for N and K are fairly straightforward, analysis for available P is rather complicated by the competing pools in the soil as well as fixation agents such as sesquioxides, surfaces, and exchangeable Ca^{2+} and Mg^{2+} in the upper pH range. The amount of P extracted from soil is, therefore, dependent upon many factors. Among those are soil pH, clay content, and concentrations of calcium, iron, and aluminum (Cox, 2001). Therefore, the effectiveness of a phosphorus analytical method is determined by, among other factors, the pH of an extractant (Haney *et al.*, 2006). Several methods for determining available soil P have been developed and most of these methods involve the use of an extracting agent.

2.2 Extraction of phosphorus plant availability

Availability of phosphorus for plant utilisation is not a function of its total amount in the soil, but the rate of its release from the soil surface into the soil solution. Nelson *et al.* (1953) reviewed routine soil test procedures used in estimating P availability to plant and divided them into two broad categories; namely, (i) extraction methods and (ii) biological methods. Extraction methods aim at categorising soil P into different pools that respond similarly to an extractant-induced change in chemical environment. Single extractions are used to give an estimate of a certain pool of P, whereas sequential extractions aim at characterising P in more detail and P into pools that are separated by some chemical property (Nelson *et al.*, 1953). When an extracting solution is added to soil, there are four basic reactions by which P is removed from the solid phase. These include, (a) desorbing soil P from the sorption sites by creating circumstances where desorption is enhanced, (b) replacement of soil P with a compound having a stronger sorption affinity, (c) complexing of cations binding P, and (d) solubilisation of sorption components (Nelson *et al.*, 1953). Given the fact that biological methods, which involved field or greenhouse experiments with use of higher plants as available P indicators were time-consuming, the development of extraction procedures expanded.

In 1953, the chemical extraction methods used included water, carbon dioxide-saturated water, acids, bases, salts, buffered solutions, electrodialysis and ion exchangers (Nelson *et al.*, 1953). A large variety of extraction tests are now used all over the world for extracting plant-available P from soil. Behind each procedure in use, there have been extensive studies in the temperate region on the ability of the method to produce P concentrations that correlate well with the P uptake by plants and are, thus, calibrated with P-fertiliser rate recommendations. However, the

extraction tests used for bio-availability estimations of P often serve at the same time as tests to estimate the availability of other important nutrients and might even be developed to extract other nutrients from soil (Nelson *et al.*, 1953).

Water is probably the simplest extractant used for plant available P at field pH using an ion exchange membrane in resin extraction method. The small amounts of soil P extracted by water (mainly P in dissolved forms) and difficulties related to chemical analysis limit the use of water as an extractant (Nelson *et al.*, 1953). Other extractants suggested for soil P testing include organic acids that were assumed to simulate organic acids excreted by plant roots, and mineral acids that, according to Nelson *et al.* (1953), were originally selected largely based on the analytical suitability. Olsen *et al.* (1954) suggested bicarbonate extraction for predicting plant availability of P in calcareous soils. In calcareous soils, the main function of NaHCO_3 in extracting P is to decrease the Ca^{2+} activity by forming CaCO_3 (Olsen *et al.*, 1954). However, in acid soils it is more likely that the solution pH, buffered to 8.5, promotes desorption of P. Some other additional extractants being used for available soil P testing across laboratories in sub-Saharan Africa include Bray I (Bray and Kurtz, 1945), Mehlich I (Mehlich, 1953) and Truog among others.

2.3 Phosphorus extracting reagents

In selecting an extractant for a particular soil and nutrient, the important consideration is the degree of correlation between the extracted nutrient and plant growth (Watson and Mullen, 2007). Thomas and Peaslee (1973) discussed the characteristics of suitable extractors of P. According to them, the extractant must:

- (a) rapidly dissolve and/or desorb soil P and be time independent;

- (b) maintain organic matter and soil clay in a flocculated state;
- (c) avoid precipitation of dissolved P and/or hydrolysis of organic P;
- (d) contain no excess salts, buffers or ions that interfere with analytical determinations;
- (e) extract meaningful quantities of other nutrient ions as well as P; and
- (f) be easy to prepare, store and dispose of.

Among the methods, Bray I (Bray and Kurtz, 1945), Mehlich I (Nelson *et al.*, 1953), and Olsen (Olsen *et al.*, 1954) are mainly employed in most parts of the world, sub-Saharan Africa inclusive.

2.3.1 Bray I extractant

The Bray and Kurtz P-1 (0.03 M NH_4F + 0.025 M HCl) soil phosphorus analysis solution was developed by Roger H. Bray and Touby Kurtz of the Illinois Agricultural Experiment Station in 1945 (Frank *et al.*, 1998). and is now widely used in most parts of the world. Phosphorus extracted by the Bray and Kurtz P-1 method has been shown to be well correlated with crop yield response on most acid and neutral soils in the Midwestern and North Central United States. For acid soils, the fluoride in the Bray and Kurtz extractant enhances P release from aluminium phosphates by decreasing exchangeable Al^{3+} activity in solution through the formation of various Al-F complexes. Fluoride is also effective in suppressing the re-adsorption of solubilised P by soil colloids. The acidic nature of the extractant (pH 2.6) also contributes to dissolution of available P from Al, Ca, and Fe-bound forms in most soils. The Bray soil test is not suitable for:

- (i) clay soils with a moderately high degree of base saturation;
- (ii) silty clay loam or finer-textured soils that are calcareous or have a high pH value (pH > 6.8) or have a high degree of base saturation;

- (iii) soils with a calcium carbonate equivalent > 7% of the base saturation; or
- (iv) soils with large amounts of lime (> 2% CaCO₃) (Frank *et al.*, 1998).

In soils such as these, the acidity of the extracting solution can be neutralised unless the ratio of extractant to soil is increased considerably. Additionally, CaF₂, formed from the reaction of soluble Ca²⁺ in the soil with F added in the extractant, can react with and immobilise soil P (Thomas, 2009). Both types of reactions reduce the efficiency of P extraction and result in low soil test P values. The Bray and Kurtz extractant can dissolve P from rock phosphates; therefore, it should not be used in soils recently amended with these materials, as it will overestimate available P. A Bray and Kurtz P-1 value of 25 to 30 mg P kg⁻¹ soil is often considered optimum for plant growth, although Holford (1980) reported lower critical values for highly buffered soils. Bray-P1 test was, however, approved by the United States Department of Agriculture's (USDA) North Central Regional Soil Testing Research Committee only for the acid/neutral soils of the North Central Region of the United States (Frank *et al.*, 1998). The correlation between the amounts of P extracted from the soil with the P taken up by the plant ranged from $r = 0.73$ to $r = 0.96$ for alkaline soil conditions of North Central U.S. (Kamprath and Watson, 1980).

2.3.2 Bray II extractant

Bray II (0.03 M NH₄F + 0.1 M HCl) is more acidic than Bray I because of relatively higher concentrations of HCl. Due to the relatively strong acidic reagents of Bray II, it can dissolve a substantial amount of undissolved rock phosphate during soil extraction (Bationo *et al.*, 1991). Hence, it can overestimate rock phosphate availability with respect to water-soluble P. Therefore, the Bray II method is not recommended for rock phosphate and water-soluble P as it

can overestimate available P. It is not a good method in neutral to alkaline soils and calcareous soils since it will over-estimate the soil available phosphorus levels. Given that the Bray methods were recommended as such for acid soils without consideration of the unique difference in chemical composition and extent of weathering, there is the need to re-evaluate the methods targeting tropical, heavily weathered soils, rich in sesquioxides and poor in layer silicate clays.

2.3.3 Mehlich I, II and III extractants

Mehlich and his co-workers introduced a combination of HCl and H₂SO₄ acids (Mehlich I) to extract P from soils (Mehlich, 1953). Mehlich I extracts P from aluminium, iron, and calcium phosphates and is best suited to acid soils (pH < 6.5) with low CEC (<10 cmol kg⁻¹) and organic matter contents (< 5%). It is, thus, a good extractant of calcium phosphates, and to some lesser extent extracts some Al-P. Kuo (1996) reported that the Mehlich I soil test was unreliable for calcareous or alkaline soils because it extracts large amounts of non-labile P in soils with pH > 6.5. The test was unreliable also for soils that had recently been amended with rock phosphate; and soils with high CEC or high base saturation. In soils such as these, the acidity of the Mehlich I solution is neutralised, thus reducing the capability of the dilute acid to extract P. Cope and Evans (1985) similarly cautioned that in soils where rock phosphate has been applied, care should be taken in using this extraction reagent because it will dissolve some unreacted rock phosphate, resulting in overestimation of the P supply and underestimation of the fertiliser P requirements. Similar reductions in P extraction efficiency were attributed to clay and hydrous aluminium and iron oxides (Lins and Cox, 1989).

In light of the fact that Mehlich I did not correlate well with crops on neutral to alkaline soils where apatite was the predominant source of available P, and Mehlich II did not extract copper

adequately and caused corrosive damage to the laboratory and equipment, Mehlich III extraction was developed by Mehlich in 1984 as an improved multi-element extractant for P, K, Ca, Mn, Cu, Fe, Mn and Zn (Mehlich, 1984). The extractant is composed of 0.2 M CH₃COOH, 0.25 M NH₄NO₃, 0.015 M NH₄F, 0.013M HNO₃, and 0.001 M ethylene diamine tetraacetic acid (EDTA). In the Mehlich III procedure, phosphorus is extracted by reaction with acetic acid and fluoride compounds. Exchangeable K⁺, Ca²⁺, Mg²⁺ and Na⁺ are extracted by the action of ammonium nitrate and nitric acid. Finally, the micronutrients (Cu²⁺, Zn²⁺, Mn²⁺ and Fe²⁺) are extracted by NH₄⁺ and the chelating agent EDTA. The Mehlich III soil test is similar in principle to the Bray and Kurtz P-I test because it is an acidic solution that contains ammonium fluoride. It is more effective than the Mehlich I soil test for predicting crop response to P on neutral and alkaline soils because the acidity of the extractant is neutralised less by soil carbonates (Tran and Simard, 1993).

Several studies showed that the Mehlich III soil test was highly correlated with P extracted from soils by the Bray and Kurtz P-I, Mehlich I, and Olsen P methods (Tran *et al.*, 1990). Since Mehlich III extractant is neutralised less by carbonate compounds in soil than the double acid (Mehlich I) and the Bray I, and is less aggressive towards apatite or other calcium phosphates than the double acid and Bray II extractants, the extractant was approved by the USDA's North Central Regional Soil Testing Research Committee number 13 (NCR-13) for the acid/neutral soils of the North Central Region of the United States (Frank *et al.*, 1998). The correlation between the amount of P extracted and plant uptake of P ranges from $r = 0.83$ to $r = 0.99$ for North Central U.S.A soils. Much as the Mehlich test gave relatively precise results in the United States and Canada, its effectiveness and the suitability in the humid tropics, which has unique

edaphic components, has not been effectively evaluated. This, therefore, leaves the adoption of this method in its entirety into the tropical region a question worth addressing due to the differing soil chemical and physical properties in these two regions.

2.3.4 Olsen extractant

The sodium bicarbonate soil test P method also known as Olsen P method was developed by Sterling R. Olsen and co-workers in 1954 (Olsen *et al.*, 1954). Their work was the result of a search for an extractant that would correlate crop response to fertiliser on calcareous soils. This procedure was recommended for calcareous soils, particularly those containing more than 2% calcium carbonate, but has been shown in some research to be reasonably effective for acidic soils (Fixen and Grove, 1990). The method is based on the use of the HCO_3^- , CO_3^{2-} and OH^- in the pH 8.5; 0.5M NaHCO_3 solution to decrease the solution concentrations of soluble Ca^{2+} by precipitation as CaCO_3 and soluble Al^{3+} and Fe^{3+} by formation of Al and Fe oxyhydroxides, thus increasing P solubility. In calcareous soils, the main function of NaHCO_3 in extracting P is to decrease the Ca^{2+} activity by forming CaCO_3 (Olsen *et al.*, 1954). However, in acid soils it is more likely that the solution pH, buffered to 8.5, promotes desorption of P.

The conditions under which the extraction is conducted can influence the amount of P extracted from the soil. Increasing the speed and time of the shaking will usually result in greater amounts of P being extracted (Olsen and Sommers, 1982). Increasing temperature of extraction will also increase the amount of P extracted. Olsen *et al.* (1954) reported that extracted inorganic P increased by $0.43 \text{ mg P kg}^{-1}$ soil for each 1°C increase in temperature between 20 and 30°C in soil testing between 5 and 40 mg P kg^{-1} soil. It is, therefore, important that if the results are to be interpreted in terms of making management recommendations, the condition of extraction must

be similar to those used for the calibration of the soil test. If the results are for a comparative purpose between samples, then uniformity of extraction conditions between sample extractions is of greater importance than selecting a specific shaking speed, duration, and temperature of extraction. Given that the Olsen method was designed for calcareous alkaline soils, it is basically unsuitable for soils of the humid tropics; whose pH tends to be way less than 7.

2.4 Definition and characteristics of Ferralsols

Ferralsols are soils that are developed in strongly weathered parent materials. They represent the classical, deeply weathered, red or yellow soils of the humid tropics (FAO, 2006). These soils have diffuse horizon boundaries, a clay assemblage dominated by low-activity clays (mainly kaolinite) and a high content of sesquioxides (FAO, 2006). Many Ferralsols are known as: *Oxisols* (USDA Classification); *Latosolos* (Brazil Classification); *Alítico*, *Ferrítico* and *Ferralítico* (Cuba Classification); *Sols ferralitiques* (France Classification); and *Ferralitic soils* as known by the Russian Federation (FAO, 2006). According to Vidal-Torrado and Cooper (2008), taxonomically, Ferralsols are soils having a *ferralitic* horizon at some depth between 25 and 200 cm from the soil surface. They lack a *nitic* horizon within 100 cm from the soil surface, and they lack a layer which fulfils the requirements of an *argic* horizon and which has the upper 30 cm, 10 percent or more water-dispersible clay unless the soil material has *geric* properties or more than 1.4 percent organic carbon (Yerima and Van Ranst, 2005).

Ferralsols are usually deep soils because both the intensity and the duration of weathering have been considerable. The most favourable conditions for the formation of kaolinite are found under free drainage, when silica and bases produced by the weathering of the parent materials can be

freely leached out of the profile (Van Wambeke, 1974). These circumstances are conducive to good aeration, under which iron is immobilised in the oxidised stage. It stains the smallest soil particles with yellowish or reddish colours. Most Ferralsols are strongly coloured iron oxides, which also contribute to the aggregation of clay and silt which creates porosity and most oxic horizons are friable and have a well aerated structure (Van Wambeke, 1974). Air and water can usually circulate freely through Ferralsols. Rainfall acceptance by these profiles is usually faster than by most other soils of comparable texture; it also leaches quite rapidly to deeper layers that are beyond the reach of the common rooting of most cultivated crops (Van Wambeke, 1974).

2.5 Regional distribution of Ferralsols

The worldwide extent of Ferralsols is estimated at some 750 million ha, almost exclusively in the humid tropics on the continental shields of South America (especially Brazil) and Africa (especially Congo, Democratic Republic of the Congo, southern Central African Republic, Angola, Guinea, eastern Madagascar and Uganda). Outside the continental shields, Ferralsols are restricted to regions with easily weathering basic rock and a hot and humid climate, for example, in Southeast Asia (FAO, 2006).

Weathering and soil genesis in the tropics lead to the formation of Ferralsols characterised by a colloidal fraction dominated by low activity clays and sesquioxides. These sesquioxides may have variable charge depending on their crystallinity (Paramanathan and Eswaran, 1980). Associated properties of these soils are the very low amounts of weatherable minerals; a profile morphology characterised by its uniformity in terms of lack of distinct horizonation. If there is sufficient Fe in the original material, the soils are reddish. The soils also usually have a weak

expression of pedal structure and few marks of other soil forming processes such as clay accumulation through translocation (Paramanathan and Eswaran, 1980). Due to advanced weathering, weatherable minerals are almost absent in Ferralsols, with the accumulation of gibbsite and iron oxyhydrates present in concretionary forms or as part of the fine earth fraction of the soils (Paramanathan and Eswaran, 1980).

2.6 Chemical properties of Ferralsols

2.6.1 Cations, anions and organic matter

In Ferralsols, the exchange of cations and anions between the solid and liquid phases is conditioned by the type and quantity of the clay, the oxides and the organic matter (Yerima and Van Ranst, 2005). From an analytical point of view, organic matter content is best compared in the upper 0 to 15 cm of the soil profile. Generally, soils with more than 2.5% organic carbon are considered high in organic matter. Medium values are 1 to 2.5% and below 1% carbon the soil is considered as low in organic matter (Yerima and Van Ranst, 2005).

The cation exchange capacity of ferrallitic soils is related to three sources: (i) the silicate clay minerals, (ii) the organic matter, and (iii) the sesquioxides (Yerima and Van Ranst, 2005). Kaolinite, the main clay mineral in Ferralsols, has a very small permanent charge. During weathering, this charge becomes saturated with aluminium. The poor chemical attributes of Ferralsols is attributed to the dominance of kaolinite and the sesquioxides clays with low cation exchange capacities (Yerima and Van Ranst, 2005). Organic matter is concentrated at the surface and its charge is pH-dependent. It has been estimated that the CEC is 260 cmol kg⁻¹ carbon at pH 7 and 85 cmol kg⁻¹ carbon at pH 4.5 (Yerima and Van Ranst, 2005).

2.6.2 Particle charges

The third kind of bonding energies related to the sesquioxides develops a positive charge. This charge is also pH dependent and increases from the zero point of charge of goethite at pH 4.5, to greater values at lower pH. As a result of this, at the extreme ultimate stage (acid group), the charge of the mineral colloidal soil fraction (silicate clays plus sesquioxides) may be zero or positive at pH levels below the iso-electric point (Yerima and Van Ranst, 2005). Driessen and Dudal (1991) revealed that the variable charge is due to: (i) dissociation of H^+ ions from molecules located at the periphery of the exchange complex (creating negative sites), or (ii) protonation which results in a positive charge. Protons may, for example, be released by acid groups at the edges of clay particles, or by carboxylic or phenolic groups in the organic matter, or by aluminium and iron hydroxides. Dissociation of H^+ ions is strongest at high concentration of OH in the soil solution and causes the pH dependent part of the CEC. The CEC of the ferralic B-horizon may, by definition, not exceed 16 cmol kg^{-1} of the clay; the CEC is determined in a $1M \text{ HN}_4\text{OAc}$ solution buffered to pH 7 (Yerima and Van Ranst, 2005).

A Ferralsol with 50% clay in its B-horizon contains less than 8 cmol kg^{-1} at pH 7.0 (Yerima and Van Ranst, 2005). However, the field pH of Ferralsols is normally around 5, and the base saturation is low (15-30% with depth). The effective CEC of Ferralsols is much lower than CEC; the actual cation adsorption is often a mere 3 or 4 cmol kg^{-1} soil. Protonation of hydroxylic groups at low pH values may boost the anion exchange capacity (AEC) of the Ferralsols to the extent that the AEC equals or exceeds the CEC (Driessen and Dudal, 1991).

2.6.3 Phosphorus and cations

The total phosphorus content of Ferralsols mostly varies between 20 and 300 mg kg⁻¹ with values of 20 – 40 for the percentage of organic P. Low amounts of total phosphorus in parent materials of Ferralsols are often responsible for P-deficiency related soil fertility constraints (Yerima and Van Ranst, 2005). Since there are only few external sources of P, the amount of total P in soils is proportional to those of the parent rocks (Yerima and Van Ranst, 2005).

Although these highly weathered soils have a disadvantage in that the quantities of nutrient cations such as potassium, ammonium, calcium and magnesium that a soil can hold are comparatively low, the holding capacity can be easily regulated by artificial means. It has been estimated that by raising the pH from its original of 5 to 7 simply through the application of alkaline materials, such as lime, the holding capacity of some Ferralsols for cations can be increased by 70% with an accompanying increase in holding intensity (Yu and Zhang, 1990).

CHAPTER THREE

MATERIALS AND METHODS

3.1 Nature of study

A greenhouse experiment was conducted at Abi Zonal Agricultural Research and Development Institute (ZARDI) in north-western Uganda during July 2008 to October 2009, in plastic buckets of 10-litre capacity previously perforated with holes of approximately half a centimetre to permit aeration and free drainage.

3.2 Study soil

Bulk soil was dug from a field at Abi ZARDI ($3^{\circ} 4.58' \text{ N}$ and $30^{\circ} 56.74' \text{ E}$; altitude of 1206 m asl) using a hand hoe at a depth of 0 – 30 cm in an area not fertilised for more than 10 years, and whose productivity was known to be low. The soil was freed of visible materials such as roots, litter and stones manually, and was thereafter air-dried on distilled water-cleaned polythene sheet under a roofed, well-aerated shelter for 7 days. Soil clods were, thereafter, pulverised using mortar and pestle and screened through 2 mm sieve before being weighed using a spring balance of 100 kg-capacity. From the bulk soil, a soil sample of about 500 g was taken for preliminary characterisation for pH, total N, available P, organic matter content and exchangeable cations at the National Agricultural Research Laboratories, Kawanda using routine methods as described by Okalebo *et al.* (2002). Eight kilograms of the bulk soil, freed of roots and other unwanted inert materials, was taken into plastic buckets of 10-litre capacity previously washed using distilled water.

3.3 Clay mineralogy

In order to ascertain that the study soil was a Ferralsol, a sample was taken for mineralogical analysis of the clay fraction with X-ray diffraction (XRD) method (Moore *et al.*, 1989) at the University of California, Davis in U.S.A. At the laboratory, the soil was gently crushed to coarse sand-sized fractions using a mortar and pestle. A clay fraction ($< 2 \mu\text{m}$) was separated out from the shale by disaggregating and dispersing the soil in distilled water and immediately washed by centrifugation. The fraction of $< 2 \mu\text{m}$ was isolated by centrifugation and the suspension were air-dried on glass slides. After air-drying, the slides were mounted on the goniometer and scanned from 2 to $32^\circ 2\theta$. The specimen was prepared from (i) potassium saturation heated to 25°C (K 25), (ii) potassium saturation heated to 350°C (K 350), (iii) potassium saturation heated to 550°C (K 550), (iv) magnesium saturation (Mg), and (v) glycerol solvation (GL). The Mg and K saturation treatments were done to create a monoionic exchange complex so as to control cation saturation, which causes variable spacing of the crystal planes. Glycerol was used because of its tendency to solvate the interlayer regions of the clay minerals. Digital data generated were interpreted using Diffrac plus software of the Bruker Analytical X-ray system, which comprises a search-match routine based on a Powder Diffraction File. The peaks observed from the X-ray diffractometer were interpreted and the dominant mineral compound determined.

3.4 Treatments and study design

Treatments administered included 6 P rates: 0 (Control), 20, 40, 60, 80 and 100 kg P ha^{-1} in the form of Triple Superphosphate (TSP). The equivalents of the fertiliser corresponding to the rates applied per pot included: 0, 0.08, 0.16, 0.24, 0.32 and 0.4 g P per pot . The TSP fertiliser was thoroughly mixed with the potted soils using a hand trowel. At planting, each pot received a

blanket application of 60 kg N ha⁻¹ and 40 kg K ha⁻¹ in the forms of urea and muriate of potash, respectively. These translated into 0.24 g per pot and 0.16 g per pot of N and K, respectively. Four seeds of maize (Longe V) were then planted per pot at 2 – 3 cm depth. Three soil phosphorus extraction methods used included Bray I, Bray II and Mehlich III. The experiment was laid in completely randomized design with 4 replications and the study was repeated 3 times.

3.5 Test crop

Maize (*Zea mays* L.), variety Longe V, was used as the test crop. Longe V is rich in quality protein, and was developed in 1998 by the National Agricultural Crops Resources Research Institute (NaCRRRI). The variety is drought tolerant, disease resistant, responsive to fertiliser treatments, and matures in about 105 – 110 days (Krivanek *et al.*, 2007). Also, it yields up to 5,000 kg grain ha⁻¹, which is quite high by the national yield data statistics (Krivanek *et al.*, 2007).

3.6 Crop management

The pots were watered (distilled water) to field capacity (25%) 3 times a week using a watering can and well kept in the greenhouse. After germination, when the plants were 10 cm high, they were thinned to 3 plants per pot. Every pot was kept free of weeds by hand-weeding. No inorganic chemicals (insecticides and pesticides) were used on the maize crop as there was no pest or disease infestation on the crop.

3.7 Parameters measured

At 8 weeks after planting (WAP), data were taken on maize stem girth, number of leaves per plant and leaf area. The stem girth was measured at 30 cm up on the maize plant from the soil level on the 3 maize plants per pot and an average obtained; the number of leaves was counted from one of the 3 plants per pot randomly selected; and two fully expanded leaves per each of the 3 plants were selected per pot to take measurements for the leaf area determination. Leaf area was taken at 8 WAP when most of the leaves had fully expanded and no new leaves were expected to emerge since the maize was starting to flower at this stage. It was also assumed that by the 8th WAP, the maize crop had consumed 75% of its P requirement since it had already attained 25% of its expected biomass.

The leaf area was determined by the non-destructive length x width measurement method (Saxena and Singh, 1965) using the relation: Leaf area = 0.75 (length x width); where 0.75 is a correction factor (McKee, 1964). Six leaves were measured with a carpenter's tape for each pot before the leaf area was determined. Simultaneously, 1 plant per pot was taken from above-ground level, chopped into 10 – 15 cm pieces using a hand knife, packed in labelled paper bags and delivered for laboratory analysis at the National Agricultural Research Laboratories, Kawanda. While at the lab, the samples were oven-dried at 65 °C for 48 hrs and the dry weight taken. Thereafter, the samples were ground and sub-samples in triplicates were digested using sulphuric acid and hydrogen peroxide for the determination of total phosphorus analysis using the three study analytical methods as described by Okalebo *et al.*, 2002.

At the same time (8 WAP), soil in each pot was emptied on polythene sheets and mixed thoroughly using a hand trowel. Soil samples were obtained from the mixed soil through quarter sampling. Sub-samples obtained were air-dried for 7 days and eventually analysed for P using the three study analytical methods, namely Bray I, Bray II and Mehlich III.

3.8 Statistical analysis

Data collected were subjected to analysis of variance using GenStat software (Version 7). Fisher's Protected Least Significant Differences at 5% probability level was used to separate means where significant differences were evident. Correlations were run between P for the different extraction methods against plant growth parameters.

CHAPTER FOUR

RESULTS

4.1 Characteristics of study soil

The chemical and physical characteristics of the study soil are presented in Table 1. The P analysis was done using Bray I, Bray II and Mehlich III extraction procedures. The soil pH was slightly acidic. The soil textural class was sandy loam. Bray I P, total nitrogen and organic matter were below the critical limit for crop growth.

Table 1. Pre-study chemical and physical properties of the soil

Soil properties	Test soil	Critical limit for crop growth*
pH(H ₂ O)	5.6	5.5
Organic matter (%)	2.7	3.0
Total N (%)	0.17	0.2
Bray I P (mg kg ⁻¹)	10.18	15.0
Bray II P (mg kg ⁻¹)	10.61	NA
Mehlich III P (mg kg ⁻¹)	7.05	NA
Exchangeable cations (cmol kg ⁻¹)		
Ca ²⁺	8.77	0.8
Mg ²⁺	0.89	0.22
K ⁺	0.29	0.4
Sand (%)	77.1	
Clay (%)	15.6	
Silt (%)	7.3	
Textural name	Sandy loam	

*(Okalebo *et al.*, 2002) NA = Data not available

4.2 Clay mineralogy

There were three prominent peaks generated by the X-ray diffractometer for the dominant clay minerals in the study soil; namely, mica at 10 Å, and kaolinite (7.0 Å and 3.5 Å) (Figure 1). Mica produced peaks that remained stable regardless of any heat or saturation treatment. On the other hand, kaolinite produced peaks that disappeared with 550° C heat treatment. Kaolinite was,

however, unaffected by glycerol solvation. The fourth and shorter peak at 3.3 Å indicated detection of quartz (Figure 1).

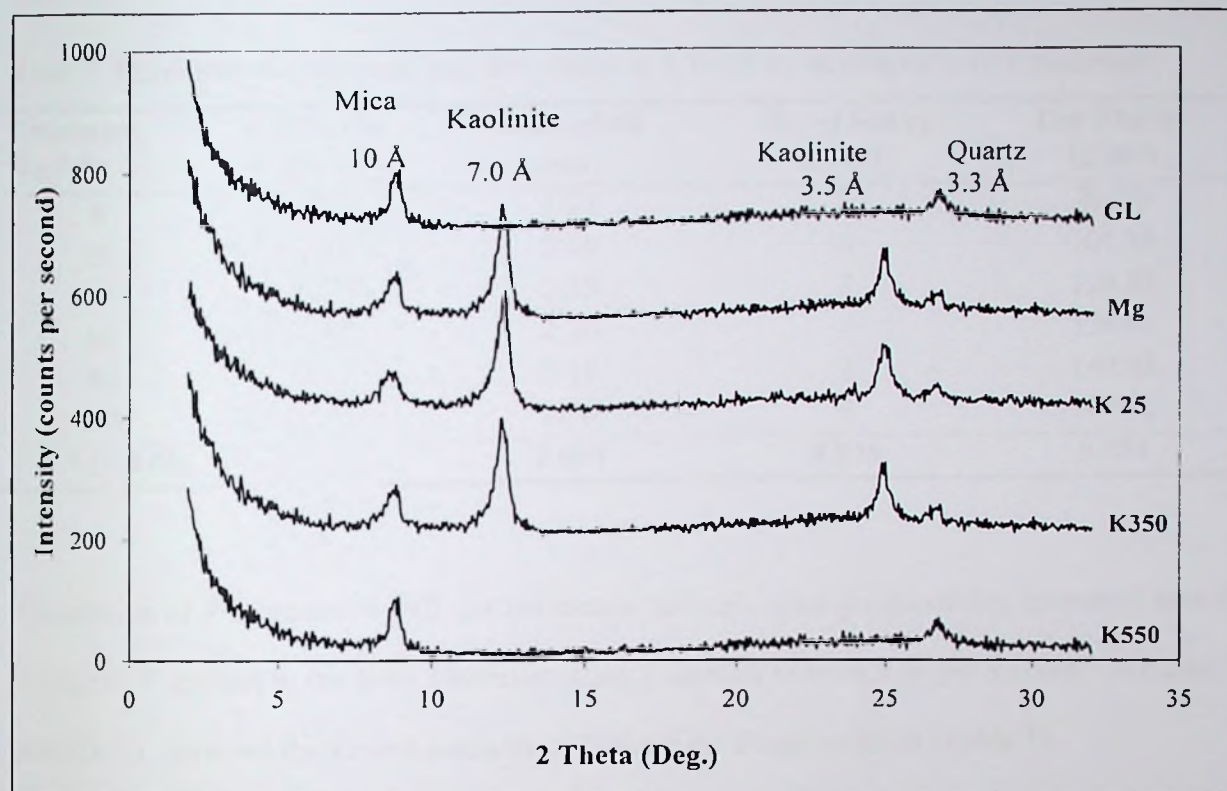


Figure 1. Sequence of X-ray diffraction patterns for the clay fraction of the study soil obtained from Abi ZARDI, Arua district, Uganda.

4.3 Performance of plant parameters at different rates of phosphorus

After statistical analysis, data in Tables 2 and 3 were used to plot correlation graphs so as to explain the responses in relation to the three extraction procedures used. The varying amount of P applied to the pots did not cause a significant change in the leaf area and the number of leaves. However, the highest average leaf area was obtained at 60 kg P ha⁻¹ treatment, and a constant average number of leaves were observed from 40 kg to 80 kg P ha⁻¹ treatments (Table 2). A significant increase in the stem girth and the dry matter were observed with increase in the

amount of P administered along the treatments. Both the stem girth and the dry matter showed their highest average values at the 100 kg P ha⁻¹ treatment (Table 2)

Table 2. Plant growth parameter and dry matter at 8 WAP at varying rates of P treatment

Treatment (kg P ha ⁻¹)	Leaf area (m ²)	Stem girth (cm)	No. of leaves (count)	Dry Matter (g/pot)
0	1.41	1.93	5	70.29
20	1.60	2.10	6	101.14
40	1.83	2.35	7	124.89
60	2.30	2.30	7	139.47
80	1.82	2.10	7	149.63
100	1.91	2.60	8	165.99
L.S.D (0.05)	0.871	0.004	0.975	9.774

The amount of P extracted by all the extraction methods used progressively increased with the amount of P applied in the pots. However, Bray I seemed to extract larger amounts of P and the Mehlich III extracted the lowest amounts of P along the P rates applied (Table 3).

Table 3. P fractions extracted by Bray I, Bray II and Mehlich III reagents from soil treated with varying rates of applied inorganic P.

Treatment (kg P ha ⁻¹)	Bray I P	Bray II P	Mehlich III P
	 (mg kg ⁻¹)		
0	10.18	10.61	7.05
20	20.00	18.54	12.08
40	18.10	14.10	10.35
60	15.43	15.82	9.10
80	24.08	22.02	14.30
100	23.23	16.43	17.58
L.S.D (0.05)	10.98	9.29	7.04

4.4 Extractible soil P and plant parameters

Correlations of data for maize plant on stem girth, number of leaves and leaf area with the three methods of P extraction showed various relationships and strengths, as presented in the following subsections:

4.4.1 Stem girth

There were positive correlations between stem girth and Bray I, Bray II and Mehlich III extractible P (Figure 2). Mehlich III method presented the greatest correlation coefficient ($r = 0.630$), while Bray II showed the lowest correlation coefficient ($r = 0.369$).

4.4.2 Number of leaves and leaf area

Among the three methods used to extract available P, Mehlich III again correlated highest with the number of leaves ($r = 0.733$) followed by Bray I (Figure 3). Bray I showed the highest correlation with leaf area ($r = 0.505$) while Bray II correlated the lowest with leaf area (Figure 4).

4.5 Extractible soil P and maize dry matter

A positive correlation was observed between dry matter of the test crop and extractable P obtained by the three extraction methods used in this study, that is, Bray I, Bray II and Mehlich III. However, the highest correlation coefficient between P extracted and the test method used was observed with Bray I ($r = 0.81$), followed by Mehlich III ($r = 0.79$) and Bray II showed the lowest correlation efficient (Figure 5).

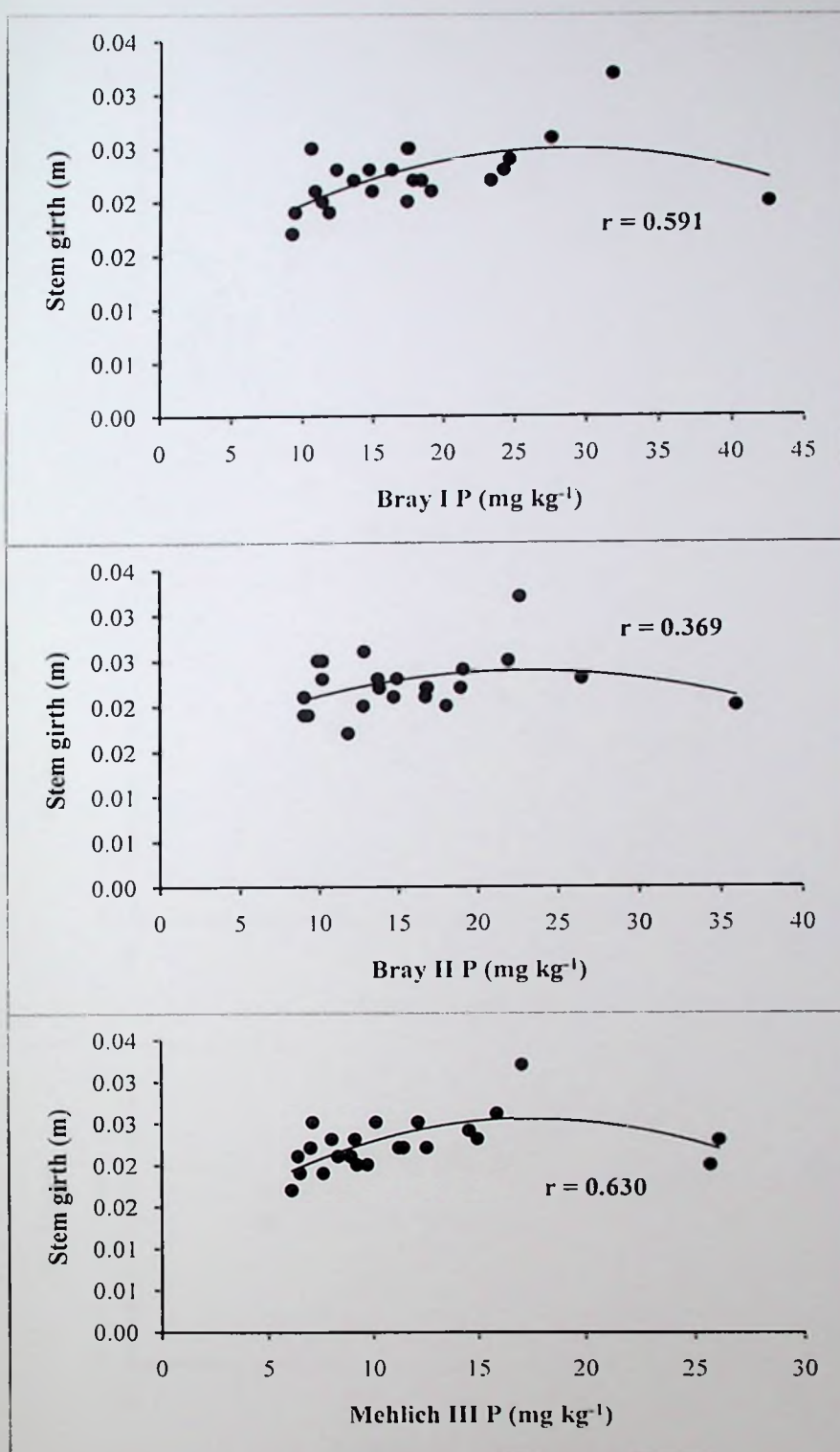


Figure 2. Correlations between stem girth and Bray I, Bray II and Mehlich III extractants for soil obtained from Abi ZARDI, Arua district, Uganda.

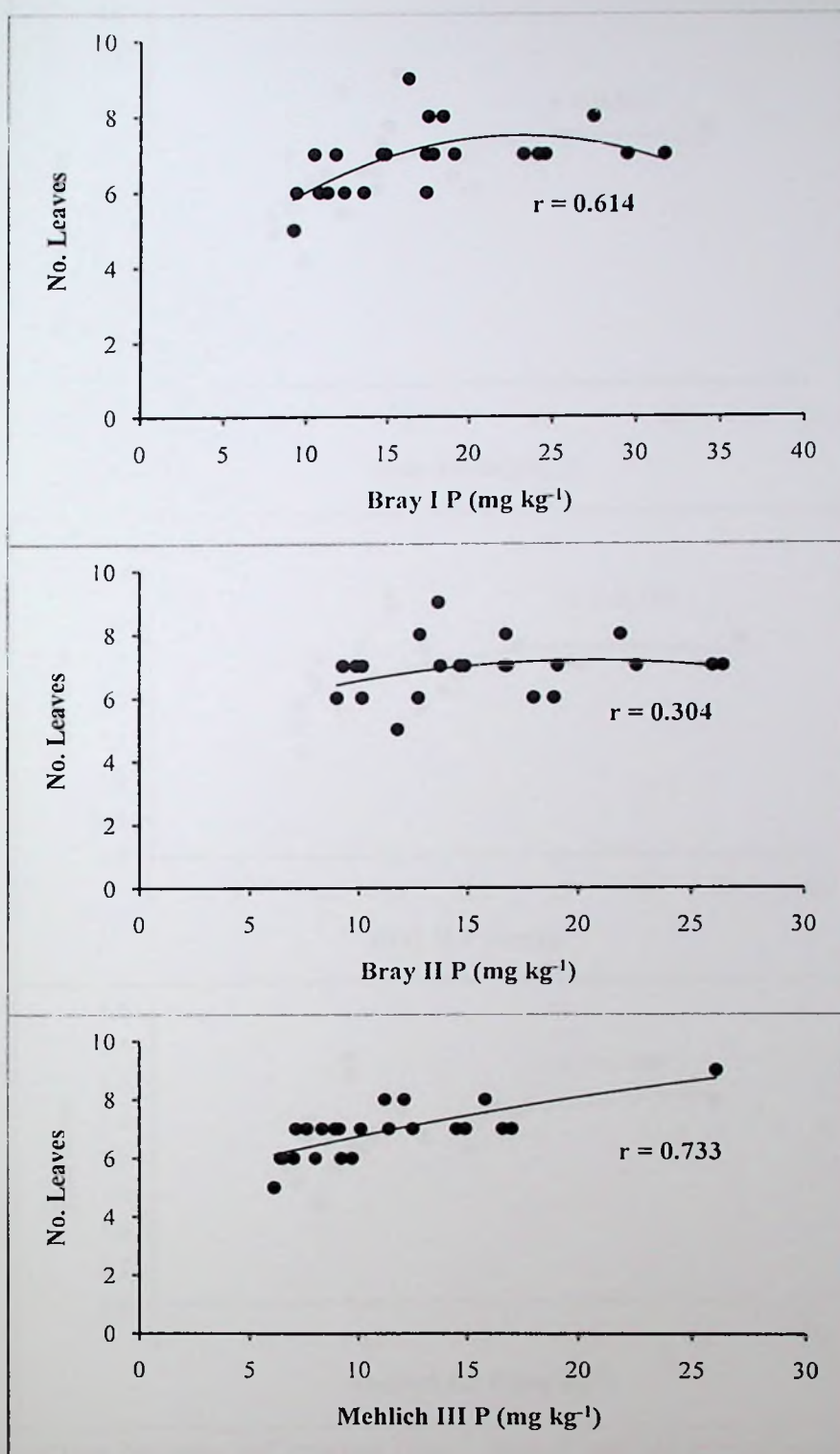


Figure 3. Correlations between number of leaves and Bray I, Bray II and Mehlich III extractants for soil obtained from Abi ZARDI, Arua district, Uganda.

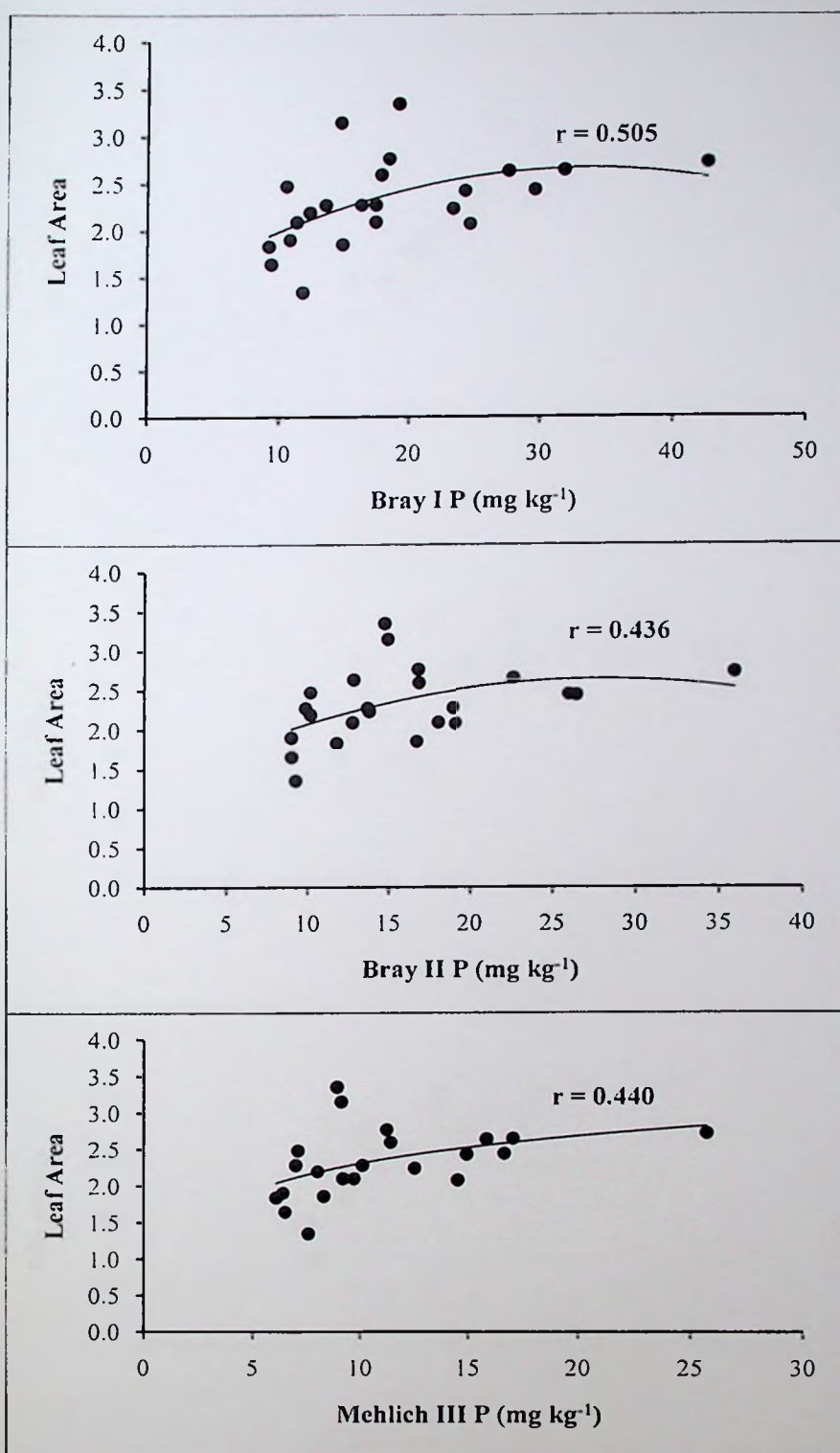


Figure 4. Correlations between leaf area and Bray I, Bray II and Mehlich III extractants for soil obtained from Abi ZARDI, Arua district, Uganda. The correlation coefficients were not significantly different among extracting agents.

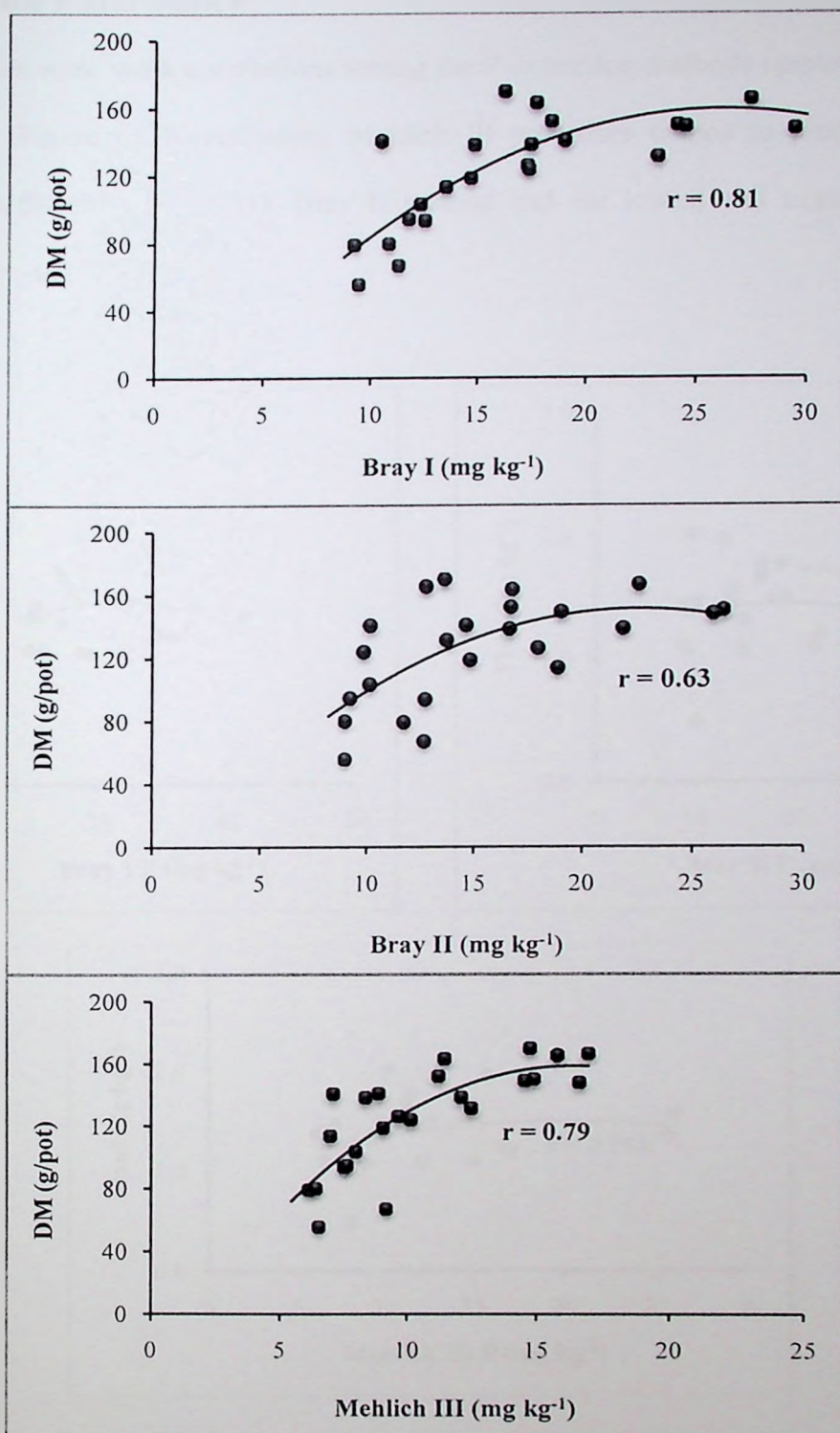


Figure 5. Correlations between dry matter and Bray I, Bray II and Mehlich III extractants for soil obtained from Abi ZARDI, Arua district, Uganda.

4.6 Extractable P and plant P

In general, there were weak correlations among the P extraction methods employed and plant P concentration (Figure 6). Nevertheless, Mehlich III procedure tended to present the highest correlation coefficient ($r = 0.254$). Bray II method had the lowest and negative correlation coefficient ($r = -0.191$) (Figure 6).

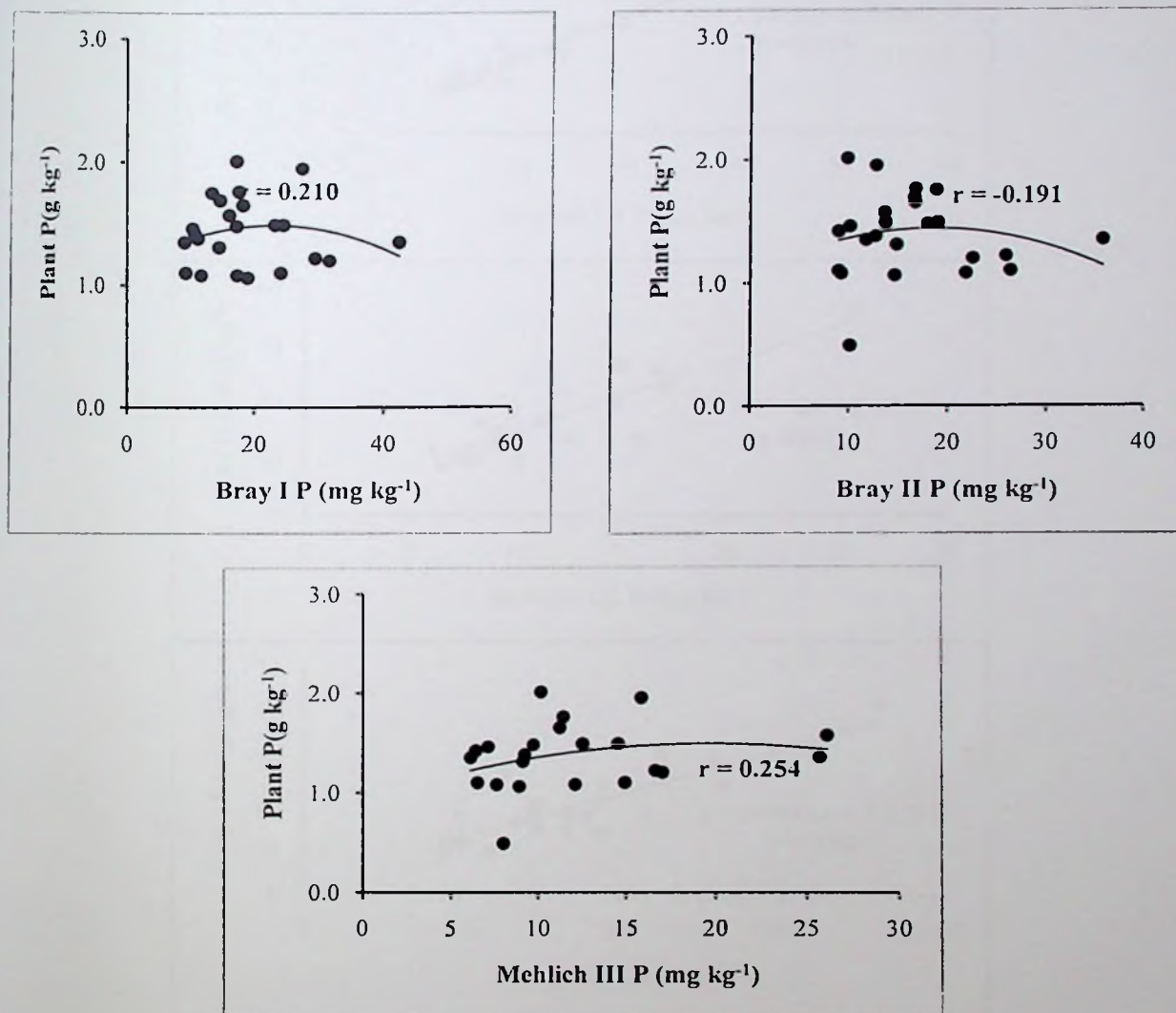


Figure 6. Correlations between plant P and Bray I, Bray II and Mehlich III extractants for soil obtained from Abi ZARDI, Arua district, Uganda.

The interrelationships between the three extractants showed that Mehlich III correlated strongly with Bray I than Bray II (Figure 7). The correlation between Bray I and II was also not so strong ($r = 0.814$), compared to the correlation coefficient between Bray I and Mehlich III ($r = 0.975$), and between Bray II and Mehlich III ($r = 0.834$).

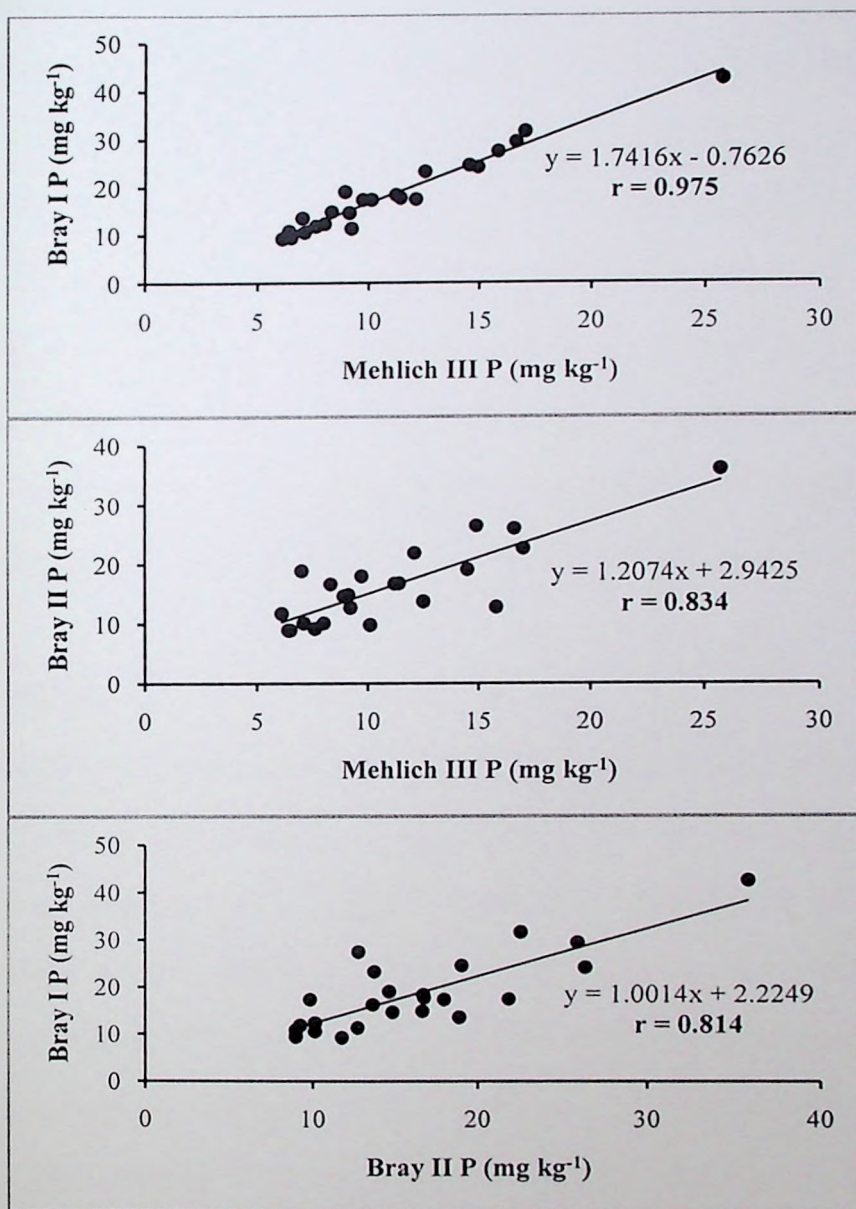


Figure 7. Interrelationships between Bray I, Bray II and Mehlich III extractants for soil obtained from Abi ZARDI, Arua district, Uganda.

CHAPTER FIVE

DISCUSSION

5.1 Characteristics of the study soil

The soil pH of the study soil was slightly acidic but was within the range for optimum maize production. The observed low organic matter content can be attributed to land use and intensity of cropping of the sites where the soils were collected. The extractable phosphorus (Bray I) with a mean value of 10.18 mg kg^{-1} was lower than the critical limit as described by Okalebo *et al.*, 2002. This could have been as a result of the strong retention of P, a characteristic problem of Ferralsols (FAO, 2006), thus making the extractable fraction so limited. Exchangeable calcium and magnesium values in the study soil appeared adequate but the exchangeable potassium was below the critical level, based on the 0.4 cmol kg^{-1} as described by Okalebo *et al.*, 2002.

5.2 Clay mineralogy

The study soils exhibited a high reflection intensity from spacing near $7.0 \text{ \AA}$ and $3.5 \text{ \AA}$ which corresponds to kaolinite (Brindley and Brown, 1980). Therefore, the findings of the X-ray diffraction analysis (Figure 1) purely support the fact that the study soil was ferralitic in its mineralogy. Yerima and Van Ranst (2005) noted that kaolinite is the main clay mineral in Ferralsols. This, according to them, is sandwiched with high levels of sesquioxides, thus, making the soils have a low fertility; especially due to the low cation exchange capacities (5 cmol/kg). Deckers *et al.* (1998) pointed out earlier that inactivation of phosphorus is a major problem ($>85\%$ phosphate fixing capacity) in Ferralsols.

Kaolinite is almost omnipresent in soil clay assemblages, especially in Ferralsols. Its relative importance is usually related to rainfall or age alteration; the higher the rainfall, the more kaolinite (Velde and Muenier, 2008). Further, kaolinite is devoid of potassium, and hence not a reservoir of this precious element. Therefore, it is often more of a hindrance to agricultural soil use than anything else (Velde and Muenier, 2008). This observation largely agrees with the low crop yields noted in the field from which the soil was sampled. Over the years, the productivity of this soil is observed to be low, which probably could be due to the poor nutrient reserves typical to Ferralsols.

5.3 Extractible P and plant parameters

The strong relationships exhibited by Mehlich III with both stem girth ($r = 0.630$, Fig. 2) and number of leaves ($r = 0.733$, Fig. 3) suggest that Mehlich III was a good extractant among the three. It was the best in mimicking P uptake by plants. However, the direct relationship observed between the increase in stem girth and the amount of P extracted by the reagents (Figure 2) could be linked to the effect of P on the cell elongation and expansion of vascular bundles. Work done by Sarker *et al.* (2010) on effects of phosphorus deficiency on anatomical structures in maize (*Zea mays* L.) showed that P deficient plants decreased in stem diameter and that the vascular bundles shrank. The gradual increment in number of leaves observed, which was limited to about 8 leaves per plant, could be explained by the fact that this is largely determined genetically. In a similar work, Ali (1998) reported that phosphorus level did not significantly affect maize foliage per plant.

The best correlation coefficient of Mehlich III observed over the other two extractants tested could be so because Mehlich III simulated closely plant P uptake under the natural soil conditions. The fact that Bray I correlated relatively better with the plant parameters tested than Bray II (Figures 2, 3 and 4), could be as a result of the less acidic nature of the solution (0.025 M HCl), which better simulated plant P uptake. Bray II, with a higher acid content (0.1 M HCl), could have extracted much more of the extractable P thus, overestimating the available P content, resulting to the low correlation with plant growth parameters. The fluoride in the Bray I extractant enhances P release from aluminium phosphates by decreasing Al^{3+} activity in solution through the formation of various Al-F complexes (Ketterings *et al.*, 2002). Fluoride is also effective at suppressing the re-adsorption of solubilised P by soil colloids (Ketterings *et al.*, 2002).

The case where the amount of soil P extracted by Mehlich III procedure correlated highly with P extracted by Bray I ($r = 0.975$, Fig. 7) implies that the two reagents are not very different in simulating plant P uptake. This is in tandem with work done by Tran *et al.* (1990), who noted that the amount of Mehlich III P is similar to that extracted by the Bray I method on most non-calcareous soils. Mallarino (2003) also noted that Mehlich III and Bray I soil test results were highly correlated in neutral to acid soils. The only contrast was that Mehlich III extracted slightly more P than Bray I in their case, while Bray I extracted more P in the present study.

5.4 Extractible soil P and maize dry matter

The maize dry matter content increased significantly over the control with all the P application rates (Table 2). The increase in dry matter was progressive with successive increase in the

application rates of P treatments up to 100 kg ha^{-1} . Similar result has been reported by Alias *et al.*, 2003 who noted an increase in stover yield of maize with increasing level of P up to 125 kg P ha^{-1} . The increased dry matter due to P application might be attributed to stimulation of root growth and development and increased root surface area which might have resulted in proper uptake and utilisation of all other nutrients by the plant, thus resulting in higher dry matter production. Pellerin *et al.* (2000) reported that the increase in dry matter production following P application is a result of improved root system, increased leaf area index and its subsequent effect on photosynthetically active radiation absorption and carbohydrate nutrition of plants. All the three P extraction methods used in this study showed strong positive correlations with the dry matter content (Figure 5). Bray I gave the highest correlation ($r = 0.81$) followed by Mehlich III ($r = 0.79$) and the lowest correlation was showed by Bray II. However, the correlation coefficients given by Bray I and Mehlich III were not significantly different meaning that both extractants simulated P uptake resulting in dry matter accumulation in a similar way. This result agrees with work done by Mallarino (2003) who observed a similar relationship between Bray I and Mehlich III in neutral of acid soils.

5.5 Soil P extracted by different reagents

The difference in the extractible P fractions using the three methods of P extraction demonstrates the varied P extracting powers of the extractants on a Ferralsol. This seems to be linked to the varying acid concentrations in the extracting agents. For example, Bray II ($0.03 \text{ M NH}_4\text{F} + 0.1 \text{ M HCl}$) is more acidic than Bray I ($0.03 \text{ M NH}_4\text{F} + 0.025 \text{ M HCl}$) because of relatively higher concentrations of HCl (Frank *et al.*, 1998), thus it extracted the greatest amount of available P (Table 3). In Mehlich III procedure, phosphorus is extracted by reaction with acetic acid and

fluoride compounds (Mehlich, 1984). It is also possible that the differences in the amount of P extracted by the three extractants are due to their selectivity in solubilising different P fractions to varied extents (Dhillon *et al.*, 1993). For example, NH_4F selects to solubilise only Al-bound P (Soinne, 2009). Therefore, a multi-element extractant such as Mehlich III is more suitable for use. The extractant is composed of 0.2 M CH_3COOH , 0.25 M NH_4NO_3 , 0.015 M NH_4F , 0.013 M HNO_3 , and 0.001 M ethylene diamine tetraacetic acid (Mehlich, 1984).

5.6 Extractible soil P and plant P

The correlations between the three extractants used and the P extracted from the maize plant were rather weak (Figure 6). However, Mehlich III tended to give the best correlation coefficient amongst the three extractants tested ($r = 0.25$), followed by Bray I ($r = 0.21$) (Figure 6). Similar work done by Hailin *et al.* (2009) on inter-laboratory validation of the Mehlich III method for extraction of plant-available phosphorus indicated that, for the determination of plant-available P in soil the Mehlich III method is both accurate and precise when standardised procedures are used. Conclusions drawn after the work done by Ketterings and Flock (2005) also indicated that Mehlich III solution is a multi-element extractant that is suitable for removing P and other elements in both acidic and neutral soils, a factor that has boosted its popularity in university and commercial soil laboratories as a standard soil test. Furthermore, Watson and Mullen (2007) provided that phosphorus extracted by using the Mehlich III method is assumed to mimic P taken up by plants. In some soils, this extractant has been found to have a detection limit of 1.0 mg kg^{-1} for P.

Therefore, the weak coefficients could be a result of the biomass dilution effect that might have confounded the plant P concentration response. Nutrient uptake and internal mobility affect the nutrient concentrations in plant tissues (Scheffe, 2008). Thus, concentration and dilution occur due to the difference between plant growth and nutrient absorption and movement of the nutrients within and between plant parts (Scheffe, 2008). Under normal growing conditions, nutrient absorption and plant growth closely parallel each other during most of the vegetative growth period (Scheffe, 2008).

CHAPTER SIX

CONCLUSION AND RECOMMENDATIONS

Conclusion

This study has demonstrated that Mehlich III and Bray I are most suitable for extracting available P in Ferralsols with mica, kaolinite and quartz mineralogy; 1.57% organic carbon and pH 5.6. Despite their equal competence for P extraction from the study Ferralsols, Bray I is a single-nutrient extractant, while Mehlich III is a multi-element extractant. It is suitable for extracting P, exchangeable K^+ , Ca^{2+} , Mg^{2+} and Na^+ , and micronutrients (Cu^{2+} , Zn^{2+} , Mn^{2+} and Fe^{2+}) in one go.

Recommendations

Mehlich III is recommended for the extraction of phosphorus in Ferralsols and could be adopted as the extractant of choice for the soils laboratories in Uganda where Ferralsols is the dominant soil type.

Since the costs involved in using the given extractants was not evaluated, there is need to determine the relative costs of the two procedures so as to arrive at a more rationalised procedure to be recommended to the laboratories. It might also be prudent to conduct further investigation to determine the best relationship between maize grain yield and methods of P extraction under field conditions.

REFERENCES

- Ali, A. 1998. Effect of nitrogen and magnesium on growth, yield and quality of hybrid maize (*Zea mays* L.). M.Sc. Thesis, Department of Agronomy, University of Agriculture Faisalabad, Pakistan. 81p.
- Alias, A., M. Usman, E. Ullah and E. A. Waraich. 2003. Effects of different Phosphorus levels on the growth and yield of two cultivars of maize (*Zea mays* L.). International Journal of Agriculture and Biology 5(4):632-634.
- Bationo, A., W. E. Baethgen, C. B. Christianson and A. U. Mokuwunye. 1991. Comparison of five soil testing methods to establish phosphorus sufficiency levels in soil fertilised with water-soluble and sparingly soluble P sources. Fertiliser Research 28(3): 271-279.
- Bray, R. H. and L. T. Kurtz. 1945. Determination of total, organic and available forms of phosphorus in soils. Soil Science 59:39-45.
- Brindley, G. W., and G. Brown. 1980. Crystal structures of clay minerals and their X-ray identification. Mineralogical society, 41 Queen's Gate, London SW7 5HR, 495 p.
- Bunemann, E. K. 2003. Phosphorus dynamics in a Ferralsol under maize-fallow rotations: The role of the soil microbial biomass. Ph.D. Thesis, Swiss Federal Institute of Technology Zurich, Switzerland. 162p.
- Cope, J. T. and C. E. Evans. 1985. Soil testing. Advances in Soil Science 1:201-228.
- Cox, M. S. 2001. The Lancaster soil test method as an alternative to the Mehlich 3 soil test method. Plant and Soil 166(7): 484-489.
- Deckers, J. A., F. O. Nachtergaele and O. C. Spaargaren. 1998. World Reference Base for Soil resources: Introduction. International Soil Reference and Information Centre (ISRIC),

Wageningen. Food and Agriculture Organisation of the United Nations (FAO), Rome, Italy. 165p.

Deckers, J. 1993. Soil fertility and environmental problems in different ecological zones of the developing countries in Sub-Saharan Africa. p. 37-52. *In*: Van Reuler H. and W. H. Prins (eds.). The role of plant nutrients for sustainable food crop production in Sub-Saharan Africa. Dutch Association of Fertiliser producers. Leidschendam, The Netherlands.

Dhillon, N. S., H. S. Handal and G. Dev. 1993. Studies on P nutrition of rice-wheat cropping sequence in a samana sandy loam. *Indian Society of Soil Science Journal* 41: 784-786.

Driessen, P. M. and R. Dudal. 1991. The Major Soils of the World. Lecture notes on their geography, formation, properties and use. Agricultural University Wageningen and Katholieke Universiteit Leuven, Belgium. 310p.

FAO (Food and Agriculture Organisation). 2006. World Reference Base for Soil Resources, by IUSS-ISRIC-FAO. World Soil Resources Report No. 103. Rome, Italy.

Fixen, P. E. and J. H. Grove. 1990. Testing soils for phosphorus. p. 141-180. *In*: Westerman R.L. (ed.). Soil testing and plant analysis, 3rd ed. SSSA Book Series No. 3. Soil Science Society of America. Madison, WI, USA.

Frank, K., D. Beegle and J. Denning. 1998. Recommended phosphorus tests. p. 21-23. *In*: Brown J. R. (ed.) Recommended chemical soil test procedures for the north central region. North Central Regional Publication No. 221 (revised). Missouri Agricultural Experiment Station SB 1001. Columbia, Missouri, USA.

Hailin, Z., S. Kariuki, J. L. Schroder, M. E. Payton and C. Focht. 2009. Interlaboratory validation of the Mehlich 3 method for extraction of plant-available phosphorus. *Journal of AOAC International*. 92(1):91-10.

- Haney, R. L., E. B. Haney, L. R. Hossner and J. G. Arnold. 2006. Development of a new soil extractant for simultaneous phosphorus, ammonium, and nitrate analysis. *Communications in Soil Science and Plant Analysis* 37(11/12):1511-1523.
- Henao, J. and C. Baanante. 2006. Agricultural Production and Soil Nutrient Mining in Africa. Summary of IFDC Technical Bulletin, IFDC, Muscle Shoals, Alabama. USA.
- Hibberd, J. M., J. E. Sheehy and J. A. Langdale. 2008. Using C-4 photosynthesis to increase the yield of rice – rationale and feasibility. *Current Opinion in Plant Biology* 11: 228–231.
- Hofma, G. and O. V. Cleemput. 2004. Soil and Plant Nitrogen. International Fertiliser Industry Association (IFA). Paris, France, 48p.
- Holford, I.C.R. 1980. Greenhouse evaluation of four phosphorus soil tests in relation to phosphate buffering and labile phosphate in soils. *Soil Science Society of America Journal* 44:555-559.
- Kamprath, E. J. and M. E. Watson. 1980. Conventional soil and tissue tests for assessing the phosphorus status of soils. p. 433-469. *In*: Khasawneh F. E. (ed.). The role of phosphorus in agriculture. American Society of Agronomy; Madison, WI, USA.
- Ketterings, Q. M., K. J. Czymmek, W. S. Reid and R. F. Wildman. 2002. Conversion of modified Morgan and Mehlich-3 soil tests to Morgan soil test values. *Soil Science* 167:830-837.
- Ketterings, Q.M. and M. Flock. 2005. Comparison of Bray-1 and Mehlich-3 tests in high phosphorus soils. *Soil Science* 170: 212-219.
- Krivanek A. F., H. De Groote, N. S. Gunaratna, A. O. Diallo and D. Friesen. 2007. Breeding and disseminating Quality Protein Maize (QPM) for Africa. *African Journal of Biotechnology*. 6(4): 312-324.

- Kuo, S. 1996. Phosphorus. p. 869-919. *In*: Sparks, D. L. (ed.) Methods of Soil Analysis: Part 3- Chemical Methods. SSSA, Madison, WI, USA.
- Lins, I. D. G. and F. R. Cox. 1989. Effects of extractant and selected soil properties on predicting the optimum phosphorus fertiliser rate for growing soybeans under field conditions. *Communications in Soil Science and Plant Analysis* 20:310-333.
- Mallarino, A. P. 2003. Field calibration for corn of the Mehlich-3 soil phosphorus test with colorimetric and inductively coupled plasma emission spectroscopy determination methods. *Soil Science Society of America Journal* 67:1928-1934.
- McKee G. W. 1964. A coefficient for computing leaf area in hybrid corn. *Agronomy Journal*. 56: 240-241.
- Mehlich, A. 1953. Determination of P, Ca, Mg, K, Na and NH_4 by North Carolina Soil Testing Laboratories. Raleigh, University of North Carolina, USA.
- Mehlich, A. 1984. Mehlich 3 soil test extractant: A modification of Mehlich II extractant. *Communications in Soil Science and Plant Analysis* 15: 1409-1416.
- Moore, D. M., R. C. Reynolds and J. Reynolds. 1989. Sample preparation techniques for clay minerals. p. 179-201. *In*: X-Ray diffraction and identification and analysis of clay minerals. D.M. Moore and R.C. Reynolds Jr., (ed). Oxford University Press, New York, NY.
- Nelson, W. L., A. Mehlich and E. Winters. 1953. The development, evaluation, and use of soil tests for phosphorus availability. *Agronomy Journal* 4:153-158.
- Okalebo, J. R., K. W. Gathua and P. L. Woomer. 2002. Laboratory Methods of Soil and Plant Analysis: A Working Manual, 2nd Ed. Maxwell EPZ (Kenya) Ltd., Nairobi, Kenya. 128p.

- Olsen, S. R. and L. E. Sommers. 1982. Phosphorus. p. 403-430. *In*: Page A.L. *et al.* (ed.) Methods of soil analysis. Part 2. 2nd ed. Agronomy Monograph 9. ASA and Soil Science Society of America, Madison, WI., USA.
- Olsen, S.R., C.V. Cole, F.S. Watanabe, and L.A. Dean. 1954. Estimation of available phosphorus in soils by extraction with sodium bicarbonate. USDA Circular 939. U.S. Government Printing Office, Washington D.C., USA.
- Paramanathan, S. and H. Eswaran. 1980. Morphological properties of Oxisols. p. 35-44. *In*: Theng, B.K.G. (ed.). Soils with Variable Charge. New Zealand Soil Science Society, Lower Hutt, New Zealand.
- Pellerin, S., A. Mollier and D. Plénet. 2000. Phosphorus deficiency affects the rate of emergence and number of maize adventitious nodal roots. *Agronomy Journal* 92: 690-697.
- Sanchez, P.A. 2002. Soil fertility and hunger in Africa. *Science* 295: 2019-2020.
- Sanchez, P. A., C. A. Palm, L. T. Szott, E. Cuevas and R. Lal. 1989. Organic input management in tropical agroecosystems. p. 125 – 152. *In*: Coleman D. C. *et al.*, (eds.) Dynamics of soil organic matter in tropical ecosystems. Niftal Project, Paia Hawaii, USA.
- Sarker, B. C., J. L. Karmoker and P. Rashid. 2010. Effects of phosphorus deficiency on anatomical structures in maize (*Zea mays* L.). *Bangladesh Journal of Botany* 39(1): 57-60.
- Saxena, M. C. and Y. Singh. 1965. A note on leaf area estimation of intact maize leaves. *Indian Journal of Agronomy* 10: 437-439.
- Scheffe, L. 2008. How to Collect and Interpret Plant Tissue Samples. (Website: http://www.nm.nrcs.usda.gov/technical/handbooks/iwm/NM_IWM_Field_Manual/Section

n05/5d-How_to_Collect_and_Interpret_Plant_Tissue_Samples.pdf. Visited on 18th December 2010).

- Smaling, E. M. A., S. M. Nandwa and B. H. Janssen. 1997. Soil fertility in Africa is at stake. p. 47- 61. *In*: Replenishing soil fertility in Africa, SSSA Special Publication Number 51. Buresh, R.J., P.A Sanchez and F. Calhoun (Eds.). SSSA, ASA. Madison, Wisconsin, USA.
- Soinne, H. 2009. Extraction methods in soil phosphorus characterisation: limitations and applications. Pro terra, no. 47, University of Helsinki. Department of Applied Chemistry and Microbiology. Helsinki, Finland. (Website: [helda.helsinki.fi/bitstream/handle/10138/20847/ extracti.pdf?sequence=1](http://helda.helsinki.fi/bitstream/handle/10138/20847/extracti.pdf?sequence=1). Visited on 16th Dec 2010).
- Thomas, J. S., 2009. Soil Test Phosphorus: Principles and Methods. p. 9-19. *In*: Kovar, J. L. and G.M. Pierzynski (eds.) Methods of Phosphorus Analysis for Soils, Sediment, Residuals, and Waters. 2nd Edition. Southern Cooperative Series Bulletin No. 408, Virginia Tech University, USA.
- Thomas, G. W. and D. E. Peaslee. 1973. Testing soils for phosphorus. p 115-132. *In*: Walsh L. M. and J. D. Beaton (eds). Soil Testing and Plant Analysis. SSSA. Inc. Madison, Wisconsin, USA.
- Tran, T. S., M. Giroux, J. Guilbeault and P. Audesse. 1990. Evaluation of Mehlich-III extractant to estimate the available P in Quebec soils. Communications in Soil Science and Plant Analysis. 21(1/2): 1-28.
- Tran, T. Sen and R. R. Simard. 1993. Mehlich III extractable elements. p. 43-49. *In*: Carter, M.R. (ed.) Soil Sampling and Methods of Analysis. Canada Society of Soil Science, Ottawa, Ontario, Canada.

- Troeh, F. R. and L. M. Thompson. 2005. *Soils and Soil Fertility* (6th ed.). Blackwell Publishing, United Kingdom. 489p.
- Turner, B. L., S. Newman and K. R. Reddy. 2006. Overestimation of organic phosphorus in wetland soils by alkaline extraction and molybdate colourimetry. *Environmental Science and Technology* 40: 3349–3354.
- Turner, B. L., M. Papházy, P.M. Haygarth and I. D. McKelvie. 2002. Inositol phosphates in the environment. *Philosophical Transactions of the Royal Society of London B* 357: 449–469.
- Van Wambeke, A. 1974. Management Properties of Ferralsols. *Soils Bulletin*, 23. FAO, Rome, Italy. p. 129.
- Velde, B. and A. Meunier. 2008. *The origin of clay minerals in soils and weathered rocks*. Springer, Heidelberg, New York. ISBN 9783-540-75633-0. 406p.
- Vidal-Torrado, P and M. Cooper. 2008. Ferralsols. p. 238-240. *In*: Chesworth, W. (Ed.). *Encyclopedia of Soil Science*. Springer. Dordrecht, The Netherlands.
- Warren, G. 1992. Fertiliser Phosphorus Sorption and Residual Value in Tropical African Soils. *NRI Bulletin* 37. Natural Resources Institute, Chatham, England.
- Watson, M. and R. Mullen. 2007. *Understanding Plant-Available Phosphorus*, Fact Sheet 3373, The Ohio State University Cooperative Extension, Columbus, Ohio, USA.
- Yerima, B. P. K. and E. Van Ranst. 2005. *Major Soil Classification Systems Used in the Tropics: Soils of Cameroon*. Trafford Publishing, Canada. 176p.
- Yu T. R. and X. N. Zhang. 1990. Chemistry of variable charge soils. p. 494-513. *In*: *Soils of China* (Institute of Soil Sciences, (ed.) Science Press, Beijing, China.

Yost, D. and H. Eswaran. 1990. Major land resources areas of Uganda. Washington, D.C: Soil Management Support Services, USAID, USA.

APPENDICES

Appendix 1. Analysis of variance table for Bray I extractant

Source	Degrees of freedom	Sum of squares	Mean square	F-value	P-value
Replications	3	57.17	19.06		
Treatments	5	633.28	126.66	7.35	0.001
Residual	15	258.31	17.22		
Total	23	948.76			

Coefficient of variation = 24.3%

Appendix 2. Analysis of variance table for Bray II extractant

Source	Degrees of freedom	Sum of squares	Mean square	F-value	P-value
Replications	3	57.53	19.18		
Treatments	5	306.39	61.28	3.75	0.021
Residual	15	245.33	16.36		
Total	23	609.25			

Coefficient of variation = 26.8%

Appendix 3. Analysis of variance table for Mehlich III extractant

Source	Degrees of freedom	Sum of squares	Mean square	F-value	P-value
Replications	3	17.57	5.86		
Treatments	5	220.45	44.09	15.77	<0.001
Residual	15	41.94	2.79		
Total	23	279.97			

Coefficient of variation = 17.3%