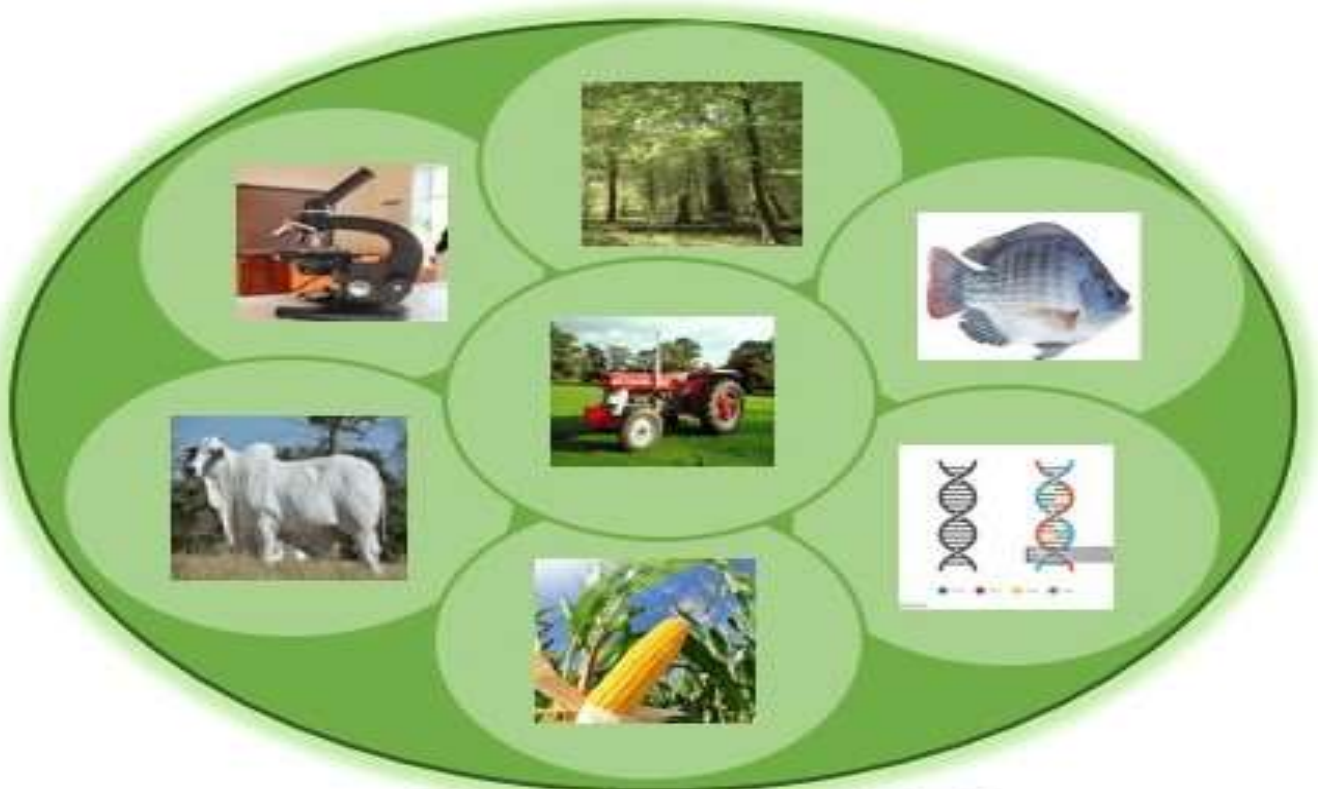




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CONTACT:

The Editor,
Kebbi Journal of Agriculture and Natural Sciences,
Faculty of Agriculture,
Kebbi State University of Science and Technology Aliero,
PMB 1144, Birnin kebbi, Nigeria.
Email: kejaanseditor@ksusta.edu.ng, kejaans.foa@gmail.com.
Phone: +234 8039370546

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INFLUENCE OF *TRICHODERMA VIRIDE* ON SOIL AVAILABLE PHOSPHORUS USING ROCK PHOSPHATE AND SINGLE SUPER PHOSPHATE IN SEMI-ARID ENVIRONMENT

G.A. Abubakar, and Attah Abubakar

Department of Soil Science and Land Resources Management, Faculty of Agriculture,
Usmanu Danfodiyo University, Sokoto, Nigeria

Corresponding Author: garba.aliyu@udusok.edu.ng +23408062662255

ABSTRACT

This study aimed to evaluate the influence of *Trichoderma viride* on soil available Phosphorus when used with rock phosphate (RP) and single super phosphate (SSP) in a controlled screen house at Department of Crop Science, Usmanu Danfodiyo University Sokoto. The experiment consisted of six treatments: *Trichoderma viride* alone (TV), rock phosphate alone (RP), single super phosphate alone (SSP), rock phosphate combined with *Trichoderma viride*, single super phosphate combined with *Trichoderma viride*, and a control. The experiment was laid out in a Randomized Complete Block Design (RCBD), replicated three times for 40 days. Soil properties such as pH and available Phosphorus were determined before and after the experiment. Results indicated that, soil pH across treatments remained slightly acidic to neutral, which is suitable for nutrient management. Specifically, *Trichoderma viride* alone achieved the highest mean pH of 6.98, followed by the combination of RP + TV, which recorded 6.96. Additionally, both TV and combination of RP + TV have the highest available Phosphorus levels of 0.16 mg/kg, outperforming the SSP and control treatments. These findings suggest that *Trichoderma viride* effectively enhances soil health and Phosphorus availability, supporting its application in sustainable agricultural practices.

Keywords: *Trichoderma viride*, Available Phosphorous, Rock Phosphate, Single Super Phosphate, Inoculation

Introduction

Phosphorus (P) is the second major element, essential for plant growth and development after nitrogen (N). The concentration of Phosphorus in plant dry matter varies from 0.05 to 0.5% (Malhotra *et al.* 2018). Phosphorus plays a major role in plants such as energy transfer, cell division, photosynthesis, and activation of different plant enzymes that resulted in better seed germination, membrane integrity, optimum plant metabolism, formation of plant reproductive parts, and

nutrient plants (Razaq *et al.* 2017; Yamaji *et al.* 2017). Soil is a vast source of Phosphorus for terrestrial plants, but the availability is primarily governed by the release of soluble inorganic phosphate ions (H_2PO_4^- , HPO_4^{2-} and PO_3^{3-}) in soil-solution, reaction with dominant soil-cations (Fe^{2+} , Fe^{3+} , Al^{3+} , Ca^{2+} , and Mg^{2+}), type of soils, and soil-pH (Penn and Camberato 2019). In agricultural fields, the majority of the Phosphorus (natural source, externally applied, or both) in top soils gets converted to plant unavailable forms and

interferes with crop uptake. Low use efficiency of Phosphorus (15–20%) in field crops has resulted in higher consumption of fertilizer-P (Malhotra *et al.* 2018).

Available soil phosphate (P) deficiency has been widely recognized as a factor that severely impairs soil quality and plant performance in agriculture and forestry ecosystems in the tropics (Turner *et al.*, 2006; León and Osorio, 2014). To overcome this problem, it is necessary to apply high amounts of P fertilizers; however, this is inefficient and costly. Although rock phosphates (RP) can be used effectively in high P fixing soils (Osorio and Habte, 2013), the low reactivity of most RP restricts their widespread use (Shrivastava *et al.*, 2007; Ojo *et al.*, 2007). The use of phosphate solubilizing microorganisms (PSM) to enhance RP dissolution has been considered a viable approach to improve the efficacy of RP as a source of P for agricultural purposes (Singh and Reddy, 2011; Osorno and Osorio, 2014).

Rock phosphate is an important natural source of P and is used as raw material for the production of chemical phosphatic fertilizers (Reddy *et al.* 2002). Apatitic rocks, it contains a high concentration of P and may contain a wide range of minor chemical elements, some of which are useful for plant nutrition (IAEA, 2002; Szilas *et al.*, 2007). Rock phosphate is extremely relevant for replenishing P, Ca, and Mg. Given that African nations cannot afford to buy pricey water-soluble phosphatic fertilizers, their value is even more crucial (Appleton, 2002; Straaten, 2007). The direct application of rock Phosphate is considered inefficient due to limited solubility of the most of the phosphate compounds in rock phosphate, despite the above mentioned the direct use of rock Phosphate is still a possible option if it is applied after certain treatments for activation such as partial acidulation (Ahmad *et al.* 2019).

Biological solubilization of rock phosphate is environmentally friendly, inoculation of phosphate-solubilizing fungi and mycorrhizal fungi helps in releasing Phosphorus from rock phosphate and transforming to available forms during composting. Moreover, it improves the physicochemical, biochemical, and biological soil properties (Santi *et al.*, 2000). The higher amount of soluble Phosphorus was attained from composts inoculated with *Aspergillus Niger* plus *Trichoderma* with or without farmyard manure and was much better than the superphosphate in maintaining the grown maize with available Phosphorus (Hellal *et al.*, 2013).

Fungi of the genus *Trichoderma* belong to the phylum *Ascomycota*, subdivision *Pezizomycotina*, class *Sordariomycetes*, order *Hypocreales* and family *Hypocreaceae* (Waghunde *et al.*, 2016) *Trichoderma* spp. are common rhizosphere fungus which have been regarded as beneficial fungi due to the function of promoting the growth of plants and pathogen suppression ability (Harman *et al.*, 2004). Therefore, *Trichoderma* spp. are often used as biocontrol agents and biofertilizers to improve crop yields (Herrera *et al.*, 2017). *Trichoderma* spp. have the function of promoting plant growth mainly because of the ability to dissolve nutrients from the soil, changing the root structure, secreting indoleacetic acid, cytokinin, gibberellin, gibberellins, and zeatin (He *et al.*, 2020). Recently, (Zhang *et al.*, 2016) observed that *Trichoderma* species could increase the activity of antioxidant enzyme in plants such as peroxidase, superoxide dismutase, and catalase, however, decrease the content of the hydrogen peroxide and the superoxide radical, which enhanced disease resistance of plants. (Rudresh *et al.* 2005) reported that different species of *Trichoderma* were able to dissolve tricalcium phosphate (TCP) *in vitro* and *in vivo*, including *Trichoderma viride*, *Trichoderma virens* and *Trichoderma*

harzianum. However, there have been few studies to conduct on the solubilization of insoluble Phosphorus in soil by *T. viride* and the effect of interaction with plants on growth. In particular, studies on the promotion of insoluble organic Phosphorus uptake to plants by *T. viride* are rarely reported. Therefore, it is necessary to clarify that *T. viride* has the property of solubilizing/ mineralizing insoluble Phosphorus.

Single superphosphate (SSP) is a popular Phosphorus fertilizer that has been widely used in agriculture for many years. It is a simple, low-cost source of Phosphorus that provides an immediate and readily available nutrient supply for plants. Its ease of application and quick nutrient release make it a practical choice for enhancing plant nutrition and improving crop production. However, it is important to consider soil pH and nutrient interactions when using single superphosphate to ensure optimal nutrient uptake by plants. The cost of producing fertilizers, especially phosphate fertilizers, has increased, markedly worldwide (Straaten, 2002, 2006; Chien *et al.*, 2011). Finding alternative sources of fertilizer that can offer P or other elements for the sustainable production of crops is doable, especially if it's a long-term, low-cost plan that takes into account the needs of impoverished farmers. (Appleton, 2002; Zapata and Roy, 2004; Straaten, 2006). This current work aims to study the influence of *T. viride* on available phosphorous in soil from rock phosphate and to investigate the feasibility of using rock phosphate bio-enriched by microbial strains as an alternative to phosphate chemical fertilizers and its effected-on soil properties and plant growth.

Materials and Methods

The experiment was conducted at crop science garden, Usmanu Danfodiyo University, Sokoto State Nigeria. Sokoto State is Located

at Latitude 13⁰¹' North, Longitude 5⁰¹⁵' East, and at elevation of 350m above sea level of the Sudan savanna agro-ecological zone characterized by three months of rainfall. The vegetation of the of the area is semi-arid with average annual rainfall of 655.85mm, with relative humidity of 48.54 between mid-May to early June and reach its peaks in August or September. Average temperature of 36°C. Dry season starts in mid-October and ends in Aprils, while the coldest months are November to January which is characterized by dry harmattan wind (NIMET, 2022). The treatment consist of three levels of *Trichoderma viride* (0.25g, 0.5g and 0.75g/pot), Rock phosphate (0.02g, 0.04g and 0.06g/pot), Single Super phosphate (0.2g, 0.4g and 0.6g/pot), combination of Rock phosphate + *Trichoderma viride*, Single super phosphate + *Trichoderma viride* and control (where no TV, RP, or SSP) was applied and laid out in a Randomized Complete Block Design (RCBD) replicated three times. The incubation period lasted for forty days (40) which soil samples were collected and taken to the laboratory for analysis. Soil pH was determined in distilled water, with soil-water ratio of (1:1) using pH meter as described by (Thomas, 1996). Available Phosphorus was determined using Bray No 1 method as described by (Kuo, 1996). The data collected were subjected to descriptive statistics.

Results and Discussion

Effect of Treatments on Soil pH

The effect of treatments on soil pH is shown in Table 1. Slight differences were recorded in the pH from the treatments under *Trichoderma viride*, having close range mean soil pH of 6.96 – 6.98. Both TV₁ and TV₂ had similar mean soil pH of 6.98, while TV₃ maintained the same pH as the control (6.96). The results according to the ratings by Esu (1991) is slightly acidic- neutral in pH, which is

recognized as optimal for most crops and beneficial microbial activity (Bolan *et al.*, 2003). This study conforms with the findings of Gururani *et al.* (2012) who found that *Trichoderma spp* enhances soil pH by suppressing mycorrhizal fungi that contribute to soil acidity. Similar findings by Ghisalberti *et al.* (2020) also suggests that certain strains of trichoderma can produce compounds that alleviate soil acidity. This results therefore affirms that the presence of trichoderma may stabilize pH levels, thereby promoting conditions conducive to nutrient availability. The rock phosphate (RP) treatments led to a gradual decrease in soil pH, with RP₃

recording the lowest pH value of 6.89. while RP₁ and RP₂ had pH of 6.91 and 6.92 respectively. All the RP treatments compared with the control falls within the low-medium category for soil pH base on the ratings by (Esu 1991). The decrease in pH with the addition of rock phosphate, especially RP₃, aligns with the findings of Baker *et al.* (1990) who observed that rock phosphate application could lead to acidification of the soil profile over time. This behaviour can also be linked to the release of organic acids via microbial interaction, which could help solubilize Phosphorus but at the same time lower soil pH (Fardeau and Mallet, 1989).

Table 1: Effects of *Trichoderma viride* on Soil pH Using Rock Phosphate and Single Super Phosphate.

Treatments	pH means
Control	6.96
TV ₁ (0.02g)	6.98
TV ₂ (0.04g)	6.98
TV (0.06g)	6.96
RP ₁ (0.02g)	6.92
RP ₂ (0.04g)	6.91
RP ₃ (0.06g)	6.89
SSP ₁ (0.2g)	6.86
SSP ₂ (0.4g)	6.84
SSP ₃ (0.6g)	6.82
RP ₁ + TV ₁	6.90
RP ₂ + TV ₂	6.86
RP ₃ + TV ₃	6.96
SSP ₁ + TV ₁	6.87
SSP ₂ + TV ₂	6.89
SSP ₃ + TV ₃	6.82

TV indicates *Trichoderma viride*, RP = Rock phosphate, SSP = Single Super phosphate.

The single super phosphate (SSP) treatments demonstrated a more pronounced decrease in pH, with values ranging from 6.82 to 6.86 compared with the control treatment. This trend suggests that SSP contributes more to soil acidification than rock phosphate, which might be detrimental to soil health over time.

The observed lower pH values correlate with the findings of Hegarty *et al.* (2005), who demonstrated that phosphoric acid fertilizers can significantly acidify soils due to their inherent acidity. Other findings by Hinsinger (2001) observed that continued use of SSP may

result in a more significant decline in soil health, thereby affecting nutrient uptake.

The combinations of rock phosphate (RP) and *Trichoderma viride* (TV) maintained more favourable pH levels (6.89 – 6.96), RP₃ + TV₃ (6.96) had similar pH as the control treatment, thereby showed better pH maintenance. This result concurs with the findings of Pokharel and Shrestha (2022) who suggested that trichoderma may ameliorate the acidity imparted by rock phosphate.

Conversely, the combinations with single super phosphate (SSP) and *Trichoderma viride* (TV) resulted in lower mean pH values (6.82 – 6.89), further emphasizing the acidifying nature of SSP when used alone or in combination with Trichoderma.

Effect of treatments on available Phosphorus (AP)

Available Phosphorus as influenced by treatments is shown in Table 1. The results of

available Phosphorus on the trichoderma treatments showed that, TV₂ and TV₃ maintained Phosphorus levels similar to the control treatment (0.15mg/kg and 0.14mg/kg, respectively), suggesting varied effectiveness of Trichoderma strains or conditions under which they were applied. While TV₁ yielded the highest mean available Phosphorus level of 0.16mg/kg, indicating that the application of *T. viride* can enhance Phosphorus availability. This is in concomitant with the findings of Zhao *et al.* (2015) who stated that *Trichoderma spp* have the ability to dissolve phosphates through enzyme production, which improves Phosphorus availability for plants. The interaction between Trichoderma and soil microbes can lead to increased mineralization and mobilization of Phosphorus.

Table 2: Effects of *Trichoderma viride* on Available Phosphorus Using Rock Phosphate and Single Super Phosphate.

Treatments	Available Phosphorus(mg/kg) mean
Control	0.15
TV ₁ (0.25g)	0.16
TV ₂ (0.05g)	0.15
TV ₃ (0.075g)	0.14
RP ₁ (0.02g)	0.14
RP ₂ (0.04g)	0.15
RP ₃ (0.06g)	0.15
SSP ₁ (0.2g)	0.13
SSP ₂ (0.4g)	0.13
SSP ₃ (0.6g)	0.13
RP ₁ + TV ₁	0.15
RP ₂ + TV ₂	0.15
RP ₃ + TV ₃	0.16
SSP ₁ + TV ₁	0.12
SSP ₂ + TV ₂	0.12
SSP ₃ + TV ₃	0.12

TV indicates *Trichoderma viride*, RP = Rock phosphate, SSP = Single Super phosphate.

The available Phosphorus from rock phosphate (RP) treatments ranged from 0.14 - 0.15mg/kg. Notably, RP₂ and RP₃ sustained levels similar to the control, indicating that while rock phosphate may provide some Phosphorus, its effectiveness is limited without biological enhancement. The relatively stable Phosphorus levels observed with rock phosphate aligns with research by Tandon (1995), which indicates that rock phosphate can steadily supply Phosphorus, but its solubility is heavily influenced by soil pH and microbial activity. Other findings by Bolan (2003) also observed that rock phosphates effectiveness is often limited in acidic soils, making the presence of beneficial microbes such as *Trichoderma* crucial for maximizing Phosphorus availability.

Single super phosphate (SSP) treatments showed the lowest mean Phosphorus levels in the entire experimental setup, ranging from 0.12 – 0.13mg/kg. This suggests that even though SSP typically provides readily available Phosphorus, its effectiveness may be limited under the experimental conditions used, possibly due to increased soil acidity. Ghorbani *et al.* (2010) reveals that lower available Phosphorus levels from SSP treatments correlate with a decline in pH and suggest less Phosphorus solubility in the acidic conditions created.

The Phosphorus levels in combinations involving rock phosphate (RP) and *Trichoderma viride* (TV) indicated potential synergistic effect where Phosphorus availability was maintained or increased (0.15 – 0.16mg/kg). RP₃ + TV₃ had the highest mean available Phosphorus of 0.16mg/kg. This synergistic effect observed with *Trichoderma viride* combined with rock phosphate exemplifies how biological amendments can enhance nutrient solubilization (Zhao *et al.*, 2015).

In contrast, *Trichoderma viride* (TV) combinations with single super phosphate

(SSP) resulted in lower Phosphorus levels (0.12mg/kg) in all treatments compared to the control, indicating that the acidifying nature of SSP combined with biological amendments may inhibit Phosphorus solubilization. While it can also provide readily available Phosphorus, the associated soil acidification limits its long-term viability in nutrient management and therefore underscores the necessity for integrated soil fertility management strategies that consider both soil chemistry and microbial ecology (Hinsinger, 2001; Ghorbani *et al.*, 2010).

Conclusion

The treatments as influence by pH had a significant effect on soil pH through maintaining a slightly acidic – neutral pH across treatments which is recognize as optimal for most crops and beneficial microbial activity. *Trichoderma viride* treatments alone and treatment from combine application of rock phosphate + *Trichoderma* recorded the highest mean pH levels. Similarly on the available Phosphorus, trichoderma alone and its combine application with rock phosphate (RP + TV) also performed better compared with other treatments. This study further clarified the pivotal role of *Trichoderma viride* in managing soil pH and enhancing Phosphorus availability when combined with different Phosphorus source. Therefore, it can be concluded that the successful integration of biological agents like *Trichoderma* with rock phosphate applications may lead to optimized nutrient availability and enhance soil health. Nevertheless, *Trichoderma viride* have shown potential as decomposers to enhance nutrient cycling. These findings highlight the potential of integrating rock phosphate and microbial inoculants for farmers, researchers and other relevant stakeholders such as fertilizer industries to improve soil fertility in semi-arid agricultural systems. Further studies are

recommended to explore long-term effects and optimize application strategies for broader adoption in Sub-Saharan Africa.

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