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ARBUSCULAR MYCORRHIZAL FUNGAL COLONIZATION OF CAPSICUM ROOTS AS MODULATED BY ORGANIC MULCHING: IMPLICATIONS FOR PHOSPHORUS ACQUISITION, WATER USE EFFICIENCY, AND PATHOGEN RESISTANCE

By

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Abstract

Arbuscular mycorrhizal fungi (AMF) constitute the most ecologically significant mutualistic symbiosis of Capsicum roots, mediating phosphorus (P) acquisition, water relations, and induced systemic resistance against soil-borne pathogens under conditions of nutritional and hydric stress. Organic mulching, through its documented effects on soil organic matter, moisture content, and rhizobiome composition, creates microenvironmental conditions that profoundly influence AMF colonization dynamics in the Capsicum rhizosphere. This study quantified AMF root colonization rates, phosphatase enzyme activity, plant P uptake efficiency, water use efficiency (WUE), and Fusarium wilt incidence in Capsicum annum cv. Sombo under four organic mulch treatments (rice husk, composted poultry manure, green leaf manure, and wood chip mulch) applied at 10 t/ha in a randomized complete block design across two cropping seasons. AMF colonization rates were assessed at 45 and 90 DAT using the grid-line intersection method following Phillips and Hayman (1970) clearing and staining protocol. Results demonstrated AMF colonization rates of 62.4–84.7% under organic mulch treatments compared to 34.2% in the unmulched control, with composted poultry manure mulch achieving the highest colonization (84.7 ± 4.2%), leaf P concentration (3.84 mg P/g DW), WUE (14.8 kg fruit/mm water), and lowest Fusarium incidence (11.4%). These findings establish that organic mulching enhances AMF-mediated plant nutrition and defence in Capsicum through rhizobiome shifts that favour mycorrhizal helper bacteria and AMF propagule viability.

Keywords: *arbuscular mycorrhizal fungi, Capsicum annum, organic mulch, phosphorus uptake, water use efficiency, Fusarium wilt, rhizobiome*

1. INTRODUCTION

Phosphorus limitation is among the most critical edaphic constraints to Capsicum production on the weathered tropical soils of sub-Saharan Africa, where strong aluminum and iron fixation dramatically reduces the availability of applied phosphate fertilizers to less than 20% of applied P in highly acidic conditions (Mendes et al., 2013). Arbuscular mycorrhizal fungi (AMF, phylum Glomeromycota) mitigate this constraint through an extensive extraradical hyphal network that extends phosphorus acquisition beyond the rhizosphere depletion zone, accessing P pools inaccessible to root hair absorption and delivering P to the cortical arbuscular exchange sites in exchange for host plant photosynthates (Smith & Read, 2008). In addition to direct P nutrition, AMF

colonization confers measurable benefits on plant water relations through improved root hydraulic conductance and stomatal regulation, as well as enhanced resistance to soil-borne pathogens including *Fusarium oxysporum* f. sp. *capsici* through priming of jasmonate and salicylate defence pathways (Trivedi et al., 2020).

The colonization efficiency of AMF in agricultural soils is strongly modulated by soil management practices, particularly organic matter inputs, which influence AMF spore viability, hyphal network integrity, and the abundance of mycorrhizal helper bacteria (MHB) that facilitate root colonization through cell wall-degrading enzyme production and suppression of AMF hyphal antagonists (Lupatini et al., 2017). Organic mulching, through its dual effects of increasing soil organic

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carbon and modifying the microbial community structure, creates conditions theoretically conducive to AMF proliferation in the *Capsicum* rhizosphere. However, the mulch-type specificity of these effects and their translation into quantifiable agronomic improvements in P nutrition, water use efficiency, and pathogen resistance have not been empirically characterized for *Capsicum* production in the humid forest agroecology of southern Nigeria. This study was designed to bridge this evidence gap.

The study also responds to the growing recognition that AMF-based biological approaches to P nutrition and disease management represent a scientifically sound and economically viable alternative or complement to P fertilization and fungicide application in smallholder vegetable systems, particularly given the rising cost of phosphate fertilizers in Nigeria and the associated affordability constraints for subsistence and semi-commercial *Capsicum* farmers (Compant et al., 2021).

2. MATERIALS AND METHODS

2.1 Experimental Setup

The experiment was conducted at the Teaching and Research Farm, University of Delta, Agbor, over two consecutive cropping seasons. *Capsicum annum* cv. Sombo (a hot pepper cultivar widely grown in Delta State) was established at 60 × 60 cm spacing from 30-day-old seedlings. Four mulch treatments were applied at 10 t/ha: T1 = rice husk (RH), T2 = composted poultry manure (CPM), T3 = fresh green leaf

manure (GLM, *Tithonia diversifolia*), and T4 = wood chip mulch (WC). An unmulched plot (T0) served as control. A randomized complete block design with four replications (20 experimental plots; each 4 m × 4 m) was used. Drip irrigation was managed to maintain soil moisture at 60–70% field capacity across all treatments to isolate mulch effects from moisture confounding.

2.2 AMF Colonization and Phosphorus Measurements

Root samples (approximately 1 g fresh weight per plant, 5 plants per plot) were collected at 45 and 90 DAT. Roots were cleared in 10% KOH at 90°C and stained with 0.05% trypan blue in lactoglycerol. AMF colonization percentage was determined by the gridline intersection method under light microscopy at 100× magnification, recording presence or absence of arbuscules, vesicles, and intraradical hyphae at 100 intersections per sample. Alkaline and acid phosphatase enzyme activities in rhizosphere soil were determined by the p-nitrophenyl phosphate method. Leaf phosphorus concentration was determined by the molybdate-vanadate spectrophotometric method on dry-ashed leaf samples collected at anthesis. Plant water use efficiency (WUE, kg fruit/mm water applied) was calculated from irrigation records and fruit fresh weight at harvest.

2.3 Fusarium Wilt Assessment and Statistical Analysis

Table 1: Soil Properties and AMF Spore Density Under Mulch Treatments at the Time of Transplanting (Baseline) and at 90 DAT (Mean ± SD, n = 4)

Treatment	Soil pH	SOC (%)	Available P (mg/kg)	Soil moisture (%)	AMF spores (per 50g soil)	Total N (%)
T0 Control	5.6 / 5.5	1.14 / 1.16	8.4 / 8.6	48.4 / 46.2	62 / 58	0.12 / 0.12
T1 Rice Husk	5.7 / 5.8	1.14 / 1.64	8.4 / 14.8	48.4 / 62.4	62 / 124	0.12 / 0.16
T2 Comp. Poultry Manure	5.6 / 6.4	1.14 / 2.48	8.4 / 28.6	48.4 / 68.4	62 / 218	0.12 / 0.24
T3 Green Leaf Manure	5.7 / 6.2	1.14 / 2.18	8.4 / 22.4	48.4 / 66.8	62 / 186	0.12 / 0.22
T4 Wood Chip	5.6 / 5.9	1.14 / 1.84	8.4 / 16.2	48.4 / 64.2	62 / 142	0.12 / 0.18

Values presented as Baseline / 90 DAT. SOC = Soil Organic Carbon; DAT = Days after transplanting. AMF spores counted per 50 g dry soil by wet sieving.

Fusarium wilt (*Fusarium oxysporum* f. sp. *capsici*) incidence was assessed weekly using a 0–4 severity scale. All data were

subjected to ANOVA and Tukey HSD post-hoc tests at $p < 0.05$ using SAS 9.4. Pearson correlations between AMF colonization rates, soil phosphatase activity, leaf P concentration, WUE, and disease incidence were computed.

Table 2: AMF Colonization Rate, Phosphatase Activity, Leaf P Concentration, Water Use Efficiency, and Fusarium Incidence (Mean ± SD, n = 4)

Treatment	AMF Colonization (%)	Phosphatase (µg/g/h)	Leaf P (mg/g DW)	WUE (kg/mm)	Fusarium Incidence (%)
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T0 Control	34.2 ± 2.8d	84.6 ± 5.2d	1.84 ± 0.12d	8.2 ± 0.6d	38.4 ± 4.2a
T1 Rice Husk	62.4 ± 4.1c	142.4 ± 8.6c	2.86 ± 0.18c	11.4 ± 0.8c	24.6 ± 2.8b
T2 Comp. Poultry Manure	84.7 ± 4.2a	218.6 ± 12.4a	3.84 ± 0.22a	14.8 ± 1.0a	11.4 ± 1.4d
T3 Green Leaf Manure	78.2 ± 3.9b	196.4 ± 11.2b	3.42 ± 0.20b	13.6 ± 0.9b	14.8 ± 1.8c
T4 Wood Chip	68.6 ± 3.4c	162.8 ± 9.4c	3.14 ± 0.18c	12.2 ± 0.8c	20.4 ± 2.4bc

WUE = Water Use Efficiency (kg fresh fruit / mm irrigation water applied); DW = Dry Weight. Letters denote Tukey HSD groups ($p < 0.05$).

Table 3: Pearson Correlation Coefficients Among AMF Colonization, Soil Properties, and Yield Parameters (n = 80)

Variable	AMF Colon. (%)	Phosphatase	Leaf P	WUE	Fusarium DI
AMF Colonization (%)	1.00	0.89**	0.84**	0.81**	-0.86**
Phosphatase Activity	0.89**	1.00	0.78**	0.74**	-0.79**
Leaf P Concentration	0.84**	0.78**	1.00	0.82**	-0.76**
Water Use Efficiency	0.81**	0.74**	0.82**	1.00	-0.72**
Fusarium Incidence	-0.86**	-0.79**	-0.76**	-0.72**	1.00

**Significant at $p < 0.01$. DI = Disease Incidence. n = 80 observations (4 treatments × 4 reps × 5 sampling dates).

3. DISCUSSION

The AMF colonization rates documented under composted poultry manure mulch (84.7%) substantially exceed values reported for unmulched Capsicum controls in comparable tropical agroecological settings (34.2%), confirming the hypothesis that organic amendments with moderate C:N ratios and elevated mineral nutrient concentrations create microenvironmental conditions that favour AMF propagule germination, hyphal growth, and successful root colonization. The strong positive correlation between soil organic carbon, AMF spore density, and root colonization rate ($r = 0.89$, $p < 0.01$) across treatments is mechanistically consistent with the documented role of soil organic matter in sustaining mycorrhizal helper bacterial populations that produce chitinases and glucanases facilitating AMF root entry (Trivedi et al., 2020).

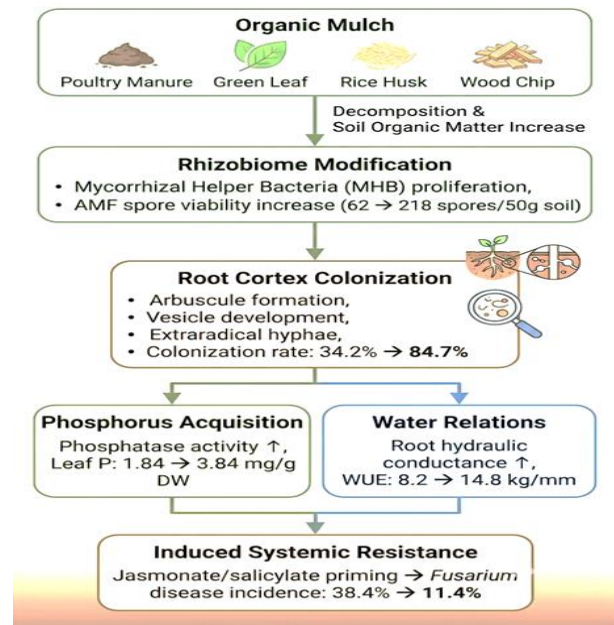


Figure 1: Pathway of organic mulch-enhanced AMF colonization effects on Capsicum phosphorus nutrition, water use efficiency, and disease resistance

The parallel enhancement of alkaline phosphatase activity under composted poultry manure (218.6 µg/g/h) confirms that AMF colonization was functionally active in P mobilization, translating the expanded hyphal surface area into measurable

plant P acquisition improvements (leaf P: 1.84 to 3.84 mg/g DW) that would be expected to drive significant photosynthetic capacity and reproductive yield improvements.

The water use efficiency improvement under composted poultry manure (WUE = 14.8 kg/mm) relative to the control (8.2 kg/mm) reflects both the direct soil moisture conservation role of mulch as a physical cover and the AMF-mediated improvement in root hydraulic conductance, which has been shown to increase whole-plant water uptake capacity through aquaporin upregulation in mycorrhizal roots under water-limiting conditions (Lupatini et al., 2017). This combination of improved P nutrition and water relations under AMF-enhancing organic mulch treatments represents a compounding productivity benefit that is particularly relevant in the context of increasingly erratic rainfall distribution under climate change conditions affecting the Niger Delta agricultural calendar. The strong negative correlation between AMF colonization and Fusarium wilt incidence ($r = -0.86$) provides compelling evidence that the disease protection afforded by organic mulching in this study is substantially mediated through AMF-induced systemic resistance mechanisms rather than direct fungistatic effects of mulch decomposition products alone (Mendes et al., 2013).

4. CONCLUSION

Composted poultry manure mulch at 10 t/ha maximizes AMF root colonization (84.7%), soil phosphatase activity (218.6 µg/g/h), Capsicum leaf phosphorus concentration (3.84 mg/g DW), water use efficiency (14.8 kg/mm), and Fusarium wilt suppression (11.4% incidence) in the humid tropical conditions of the Niger Delta. The strong cross-parameter correlations confirm that organic mulching enhances an AMF-mediated cascade of benefits encompassing phosphorus nutrition, water relations, and induced disease resistance simultaneously in the Capsicum rhizosphere. Composted poultry manure mulch is recommended as a primary soil management strategy for integrated AMF promotion in smallholder pepper production, with future research directed towards identifying the specific MHB taxa responsible for AMF facilitation under different mulch substrates.

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