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Full Length Research Paper

Passion Fruit Seedlings in the Humid Tropics of Nigeria: Relationship between Moisture Regimes and Fungal Diversity/Composition

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Yellow Passion Fruit (*Passiflora edulis* F. *flavicapa*, Deg) is a highly valued fruit crop across the globe but highly susceptible to F*ungal* attack in the soil. Pot experiment was conducted in a screen house to determine the effects of moisture regime on the level of Fungi diversity and composition in the soils and its subsequent impact on the growth and development of passion fruit. The experiment was conducted in a complete randomized design with four moisture regimes as treatments (100%, 65%, 25% and 0%) each replicated three times. Pre and post planting soil microbial analysis, plant moisture content and dry matter analyses were carried out. Fungal count (CFU/G) population was highest in 25% moisture regime (1.7×10^5 CFU/G), and decreased in the order 100% > 65% > 0% moisture regimes, although *Fusarium* species as well as others were observed at the end of 21 days in 65% and 0% moisture regimes. Significantly at P<0.05, higher fresh and dry weights of the plant were observed in 65% moisture regime (375% and 78% respectively) compared with other moisture regimes. In addition, high shoot and root ratio were obtained at the same moisture regime. Therefore, for optimum growth performance, Passion fruit in the humid tropics of Nigeria requires less saturated soil. However, to enhance the growth performance of *P. edulis* F. *flavicapa* and reduce fungal incidence in the humid tropics of Nigeria, soil water requirement by irrigation should be maintained at field capacity for at least once in every three days.

Keywords: Fungi, Moisture content, Passion fruit, Seedling

INTRODUCTION

Yellow passion fruit is a tropical crop that is grown for its high earnings in some African countries such as Kenya, South Africa, Uganda, Togo, as well as in America, Europe (Holland, England) Australia (Maluf et al., 1988), amongst others in commercial quantities enough for export to other parts of the world (Crfg., 1989). It is high in Vitamin A (3002IU) and Vitamin C (70.8mg). It is also medicinal as its extract can be used to cure Asthma, Hypertension and fight chronic inflammations (Anonymous., 2000). Of all the various organisms which have the potential to affect passion fruit leading to severe loss, *Fungi* species, (mostly soil-borne fungi) ,are major

Table 1. Soil Nutrient analysis

Chemical properties					Physical properties		
N(%)	**P (mg/20g)**	**K (mg/20g)**	**T.O.C (%)**	**SMC (%)**	**Silt (%)**	**Clay (%)**	**Sand (%)**
0.007	0.26	2.73	5.3	35	5	9.6	85.4

Soil pH = 6.2

Table 2. Soil Microbial Analysis of Soil from Ibadan, Oyo state

Ibadan soil Sample / Serial Dilution	Observation / Colour	Inference
2-1	Green, white, brown and black.	*Penicillum*, *Fusarium* and *ASpergillus* species suspected
2-2	Green, Black and White	*Penicillum*, *Aspergillus* and *Fusarium* species suspected
2-3	White and Green	*Fusarium* and *Penicillum* species suspected
3-1	White and Green	*Fusarium* and *Penicillum* species suspected
3-2	Green and Brown	*Penicillum* and *Aspergillus* species suspected
4-1	Black	*Aspergillus* species suspected
4-2	Black	*Aspergillus* species suspected
4-3	Black, Green, White and Pink	*Aspergillus*, *Penicillum*, and *Fusarium* species suspected
4-4	Black, Green and White	*Aspergillus*, *Penicillum*, and *Fusarium* species suspected
5-1	Green	*Penicillum* specie suspected
5-2	Black and Green	*Aspergillus* and *Penicillum* species suspected
5-3	Black	*Aspergillus* specie suspected
5-4	Black	*Aspergillus* specie suspected
5-5	Black	*Aspergillus* specie suspected

contributors. Soil moisture, which affects growth of plants, however, plays a major role in the growth of soil-borne fungi. Yellow passion fruit is susceptible to *Fusarium* species attack, which result in fusarium wilt. This fungi amongst others tend to thrive well in soils with low water potential (Maria.et. al., 2009), thus passion fruit crop could wilt due to low moisture content or due to the population of diverse fungi species in the soil. Fargana and Ghaffar (1991) reported that the population of *Fusarium* species is higher in the rhizosphere of plants growing at 25% soil moisture content than those grown at 50%, 75% and 100%. This study focuses on the impact of different moisture regimes on fungal diversity, and subsequent impact on the growth and development of Yellow Passion Fruit seedlings in the humid tropics of Nigeria.

MATERIALS AND METHODS

Plant Materials

Two weeks old, Yellow Passion Fruit seedlings used to conduct the experiment were gotten from the Nigerian Institute of Horticulture (NIHORT) Ibadan, Oyo state, Nigeria. The seedlings were derived from fruits that were harvested at the NIHORT Institute.

Experiment Location and dimension

The experiment was conducted in a Screen-house (12m x 10m) at the University of Port Harcourt in Choba community, Rivers state, Nigeria. Poly-pots of 40cm x

Table 3. Soil Microbial Analysis on Soils from University of Port Harcourt, Choba

Choba Soil Sample/Serial Dilution	Observation	Inference
2-1	Green	*Penicillum spp* suspected
2-2	Green	*Penicillum spp* suspected
2-3	Green and Surface white	*Penicillum* and *Rhizopus* species suspected
3-1	Green and Pink	*Penicillumsp* suspected
3-2	Green and yellow-black	*Penicillum* and *Aspergillus* species suspected
3-3	Green and Brown	*Penicillum* and *Aspergillus* species suspected
3-4	Green, Black and white	*Penicillum*, *Aspergillus* and *Fusarium* species suspected
4-1	White and brown	*Fusarium* and *Aspergillus* species suspected
4-2	White	*Fusarium* specie suspected
5-1	Black and white	*Aspergillus* and *Fusarium* species suspected

Table 4. Fungi Composition analysis (Microscopic (MC) observations carried out on one representative culture)

S/No.	Culture ID	MC. Observation	Inference
1	Ibadan 5-5	Presence of conidophores, simple terminating in a globose bearing phailides at the apex	*Aspergillus* confirmed
2	Ibadan 3-3	Non-septate Hyphae, simple pore and leathery	*Rhyopus spp* confirmed
3	Ibadan 3-2	Simple pores, conidophores broken off due to age of the culture. Septate hyphae	*Aspergillus flavus* confirmed
4	Ibadan 5-1	Branched conidophores, conidia is ovoid and one celled	*Penicillum spp* confirmed
5	Ibadan 3-1	Extensive and cotton like mycelium simple conidiophores ovoid and oblong conidia	*Fusarium spp* confirmed
6	Choba 4-2	Extensive and cotton like mycelium simple conidiophores ovoid and oblong conidia	*Fusarium spp* confirmed
7	Choba 5-2	Presence of conidophores, simple terminating in a globosebearing phialides at the apex	*Aspergillus niger* confirmed
8	Choba 2-2	Branched conidophores, conidia is ovoid and one celled	*Penicillum spp* confirmed

Table 5. Fresh and Dry weight of Leaf after 4 weeks of planting

Parameters/%SMC	100%	65%	25%	0%	S.E.D	L.S.D (5%)
Leaf Dry Weight (g)	6.70	.15	4.448	4.55	1.083	2.286
Leaf Fresh Weight (g)	24.7	38.7	12.6	12.1	5.56	11.74

25cm in size were used for the experiment, filled to a weight of 10kg of top soil gotten from the University of Port Harcourt premises. The internal environmental condition of the Screen-house was temperature- 30^{o}C, light intensity-817.3^{x10} Lux, air speed- 1.5KM/H, Relative humidity- 77.6% and a soil pH of 6.2.

Pre-Experimental Laboratory Analysis

a. Soil nutrient analysis

Soil Physico-Chemical analyses conducted on the soil samples to determine the nutrient status shows the following;

The soil status indicated low nutrient according to Bray and Weil, (1999). N.P.K. (15:15:15) Fertilizers were applied by ring method to the soil in the poly-pots at the rate of 68g per 10kg of soil weight.

b. Soil Microbial analysis

Before and after the experiment, fungi diversity was determined by culturing soil samples using Potato Dextrose Agar (PDA) as media and allowed to colonies

Table 6. Leaf Moisture content (dry and fresh weight basis), at the end of the experiment.

Parameters/%SMC	100%	65%	25%	0%	S.E.D	L.S.D (5%)
Leaf Dry Weight (%)	269	375	183	165	32.2	68.2
Leaf Fresh Weight (%)	73	78	65	62	4.64	9.83

Table 7. Stem biomass at the end of the experiment (Three weeks after)

Parameters/%SMC	100%	65%	25%	0%	S.E.D	L.S.D (5%)
stem Dry Weight (g)	7.43	7.23	5.43	6.10	1.23	2.58
Stem Fresh Weight (g)	23.6	35.9	13.4	13.8	5.50	11.52

Table 8. Shoot Biomass 2 weeks into experiment

Parameters/%SMC	100%	65%	25%	0%	S.E.D	L.S.D (5%)
Fresh weight (g)	24.1	37.0	13.0	12.9	3.73	7.54
Dry Weight (g)	7.1	7.6	4.6	5.3	0.82	1.65

Table 9. Shoot: Root Ratio 3 weeks after experiment

Parameters/%SMC	100%	65%	25%	0%
Fresh weight (g)	1.99	2.09	1.60	1.52
Dry Weight (g)	1.83	1.90	1.35	1.51

for seven days. The process was replicated three times in different serial dilutions ranging from 10-^{2}ml to 10^{-5}ml with the aid of a pipette. (Barnet and Hunter, 1968). Data generated was by visual observation and a Light Microscope (XSZ-107BN) was used to ascertain fungi diversity and their composition in the soil used for the experiment.

Soil Moisture Regimes (Treatments)

Soil moisture was maintained at field capacity for the following treatments; 100%, 65%, 25% and 0% SMC. Soil Moisture Content or water holding capacity was determined using Agvise (2010) method.
Water holding capacity = mass of soil at saturation/mass of dry soil x100

Moisture content determination

Percentage moisture content of the plant was calculated in fresh weight (FW) and dry weight (DW) basis in grams.

Experimental design

The experiment was conducted in a complete randomized design comprised of four treatments replicated three times to make a total of 15 potted plants of yellow passion fruit seedling and each replicate comprise of the different Soil Moisture levels. Treatments were randomly assigned to each plots with 60 pots per replicate, thus making it 180 pots for the entire experiment. Every pot was watered to field capacity (i.e. 100%) at the start of the experiment. Treatments were randomly assigned to the various plots in each replicate.

Plant sampling method

Two weeks and three weeks after the experiment, two seedlings each from the different treatments in each replicate respectively were randomly selected and used to determine dry matter content and level of partitioning through destructive sampling method.

DATA COLLECTION

Data was obtained for leaf fresh and dry weight, stem fresh and dry weight using the following method as described by Noogle et al., (2000). Shoot and root ratio data was also recorded.
Fresh weight (FW) = FW - DW / FW x 100
Dry Weight (DW) = FW - DW / DW x 100
Data Analysis
Data was subjected to one way Analysis of Variance (ANOVA) without blocking on GenStat Discovery 3 Software.

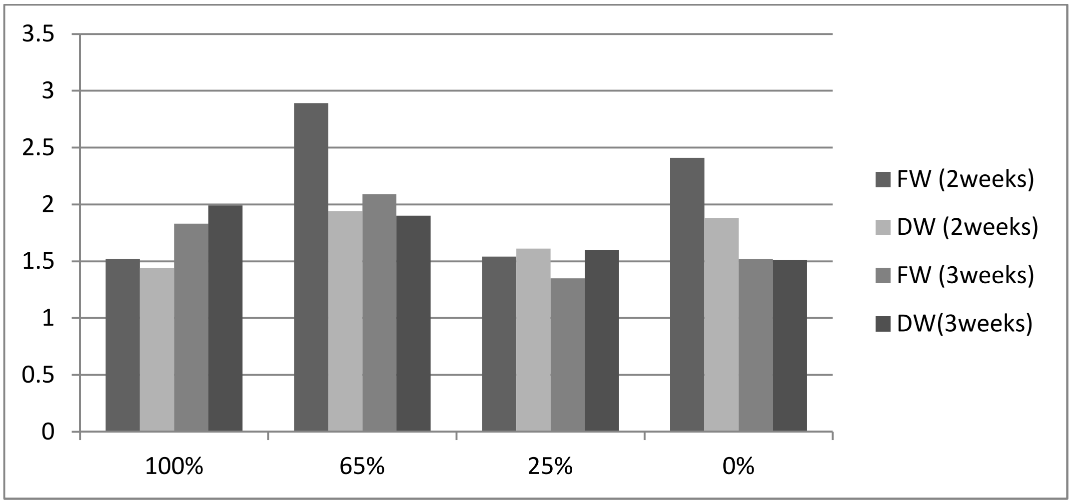

Figure 1. Shoot/Root Ratio determined at two different periods.

Table 10. Post-Experiment Studies on Soil Fungi

% SMC TREATMENTS	ORGANISMS	FUNGI COUNT (CFU/G)
100%	*Aspergillus niger* *Penicillum spp.* *Rhizopus spp.* *Clodosporium sp.*	$4.3x10^4$
65%	*Penicillum spp.* *Aspergillus spp.* *Fusarium spp.* *Aspergillus niger*	$3.6x10^4$
25%	*Aspergillus spp.* *Aspergillus flavus* *Mucor sp.* *Penicillum*	$1.7x10^5$
0%	*Aspergillus spp.* *Aspergilus flavus* *Fusarium spp.*	$2.3x10^4$

RESULTS

Plant Biomass

Leaf fresh and dry weights reduced in treatments 25% and 0% moisture regimes as compared to 100% and 65% moisture regimes. Plants in 65% moisture regime were significantly (p=0.005) higher in leaf fresh and dry weights than other moisture regimes as indicated in table 5

Leaf moisture was generally 165% and 62% and above on dry and fresh weight basis respectively (Table 6). At the end of the experiment, leaf moisture content for 100% and 65% moisture regimes were higher compared to 25% and 0% moisture regimes, by at least 219% and 17%. Therefore, under reduced soil moisture treatments, leaf turgidity declined.

Stem

At the end of the experiment, stem fresh weight was significantly (p=0.05) higher in treatments where moisture levels were adequate (i.e. 100% and 65%) than the others (i.e. 25% and 0%). There was no significant difference in dry weight. However, the trend shows that plants in 100% and 65% moisture regimes had higher dry weight or biomass.

For all the treatment, stem moisture content was generally greater by 126% and 56% dry and fresh weight bases respectively. Plants in 65% moisture level treatment had higher moisture content than 25% and 0%

moisture level treatments by at least 92% and 13%.

Shoot Biomass

Shoot moisture content was higher in 100% and 65% moisture regimes at two weeks of the experiment as shown in table 8. However, 65% moisture regime showed high intake of water as well as high in assimilates when compared to other moisture regimes.

Figure 1 shows a drastic decrease in moisture intake with 65% moisture regimes being higher in both fresh weight and dry weight at the close of the third week (3 weeks). The chart also shows that shoot root ratio was higher in the 0% moisture regime in comparison to the 100% and 25% at two weeks of the experiment. Whereas, 3 weeks after the experiment 65% moisture showed higher gain in shoot/root development.

Post Experimental Soil Microbial analysis

At the start of the experiment: *Penicillum* spp, *Aspergillus niger*, *Aspergillus flavus*, *Rhizopus* spp and *Fusarium* spp where identified from samples of the soil used for research and the soil in which they were transported with from Ibadan, through visual and microscopic observation. However, at the end of the experiment fungi count was higher in 25% moisture regime as compared to the other moisture regimes (see table 10).

DISCUSSION

The increase of Fungi colony growth at low moisture levels agrees with the observation of Hare and Parry., (2008). However, the major wilt-causing organism in passion fruit – *Fusarium* species, was not identified in soil samples of 100% Soil Moisture regime and in 25% Soil moisture regime. Maximum fungal growth was observed in 25% soil moisture regime, which corresponds with the report of Maria et al., (1989) who observed that fungi species grow better in severe water deficit conditions that inhibit plant growth. The fact that Passion fruit growth was better in 65% SMC treatment, which had a greater fungi count as well as diverse fungi than 100% soil moisture regime, suggest that the fungi concentration and diversity may not account primarily for the poor growth and performance of the plant in the lower soil moisture treatments. Also in regards to the shoot-root ratio, it is assumed that the Passion fruit seedling may have high tolerance level for soil moisture deficit. It is believed that the plant may have developed longer roots, which it uses to absorbed unavailable water from soil capillaries, although further research is needed to confirm this theory on Passion fruit. Literatures on other plants indicate that plants in severe water stress can absorb moisture by developing longer roots that can reach unavailable water from the soil during periods of drought. However, at the end of the experiment the 0% moisture regime plants, drastically reduced in both anatomical growth and assimilate of the plant, which is an indication of wilting. Thus, Passion fruit not infested by *Fusarium* species or any fungi specie can withstand drought conditions in the tropics and still maintain good growth and productivity over a period of two weeks. Adequate irrigation on Passion fruit can also improve the growth and development of the crop and limit the growth of soil fungi such as *Fusarium* species.

CONCLUSION

From the study, growth of passion fruit seedlings responds negatively to low soil moisture content in a humid tropical environment in Nigeria. However, Passion fruit can tolerate one week of moisture stress and even two weeks and resume normal growth and development after water is re-applied to the soil. By three weeks, the effect on growth due to moisture stress will be significant. Leaf area, shoot weight, number of nodes and leaf weight as well as Root – Shoot ratio will decline as soil moisture content reduces but root growth and development will not decline.

Water deficit also increases the growth of fungi diversity more importantly *Fusarium* specie which was however, observed to be at a decline at three weeks of water deficit. It is unclear as to whether the necrosis observed is due to the presence of other fungi in the soil, therefore it suggest that research be carried out to ascertain the effects of these fungi on the growth of Passion fruit seedlings in a humid tropical environment of Nigeria.

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