**Research Article** 

# Studies on the Suitability of Soil Solirazation as a Physical Control Mechanism to Manage Phytonematode Population in the Soil and its Effect on Plant Growth

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# ABSTRACT

Plant Growth-Promoting *Rhizobacteria* (PGPR) are able to promote plant growth and/or induce local and systemic resistance against biotic and abiotic stresses, but the stability and durability of their efficiency still need more investigation. The present work aims to identify a compatible-PGPR-mixture effective to stimulate wheat growth, resistance against *Mycosphaerella graminicola*, the causal agent of *Septoria tritici* leaf Blotch (STB), and tolerance to drought stress.

The interactions between twenty-six PGPR and four wheat cultivars with different resistance levels to STB, in individual and co-inoculations, were tested. The results demonstrated higher external and internal root colonisation potential of *Paenibacillus sp.* strain B2 (PB2) in a mixture, referred hereafter as Mix-3, with strains *Arthrobacter sp.* SSM-004 and *Microbacterium sp.* SSM-001, and without an impact of wheat genotype and growth stage, as observed in its individual inoculations. Only with Mix-3 was wheat growth promotion observed. Interestingly, PB2 and Mix-3 eliminated the negative impact of drought stress on the Foliar Dry Biomass (FDB) and Root Dry Biomass (RDB).

Only in a mixture of three-compatible-PGPR (Mix-3) was plant growth promotion observed and the tolerance induced to drought stress was more effective. However, it seems that resistance induced against STB is PB2-dependent.

Keywords: Mycosphaerella graminicola; Drought stress; Induced systemic resistance; Plant growth-promoting rhizobacteria

# INTRODUCTION

Soil solarization is described by many scholars and researchers as a non-chemical, soil treatment that utilizes solar radiation and a thin film of transparent mulch, usually of polyethylene, to heat the soil so that it reaches temperatures (ranging from 38°C to 50°C to a depth of about 10 cm to 20 cm) detrimental to soil-borne pests [1,2]. Soil solarization has been studied as an alternative to chemical soil fumigants like methyl bromide or 1, 3-dichloropropane, which disinfect the top soil layer but are also harmful to non-target organisms. Some of the pioneering work done on soil solarization has been conducted in Hawaii. Hagan, 1933 tried to increase the soil temperature with cellophane films to control nematodes in pineapple fields [3]. However, soil solarization did not become popular until six years later, when Groshevoy 1939 demonstrated that exposing soil to solar heat controlled soil-borne pathogens. In 1971, Adams became the first to use polyethylene mulch to control a soil-borne pathogen, black root rot (*Thielaviopsis basicola*), on sesame. Pullman and Devay 1977 published work that indicated there were wide applications for soil solarization. In 1981, Pullman et al. coined the term "soil solarization". Since then, many scholars and researchers have demonstrated the use of soil solarization for the management of nematodes and other soil-borne pathogens [4].

Tomato (*Lycopersicon esculentum*) is one of the most widely consumed fresh vegetable in the world. They are fruits, but they are commonly referred to as vegetable. Fresh-market tomatoes are a popular and versatile fruit vegetable, making significant contributions to human nutrition throughout the world for their content of sugars, acids, vitamins, minerals, lycopene and other carotenoids, among

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other constituents. Being a climacteric and perishable vegetable, tomatoes have a very short life span, usually 2-3 weeks. Tomatoes are consumed widely throughout the world and their consumption has recently been demonstrated to possess health benefits because of their rich content of phytonutrients [5].

Phytonematodes are plant-parasitic nematodes belonging to the group Nematoda. They have global habitat especially problematic in areas with tropical or sub-tropical type of climate. About 2000 plants worldwide are susceptible to infection by phytonematodes and they cause approximately 5% of global crop loss [6]. Second stage larvae of phytonematodes infect plant roots, causing the development of a variety of histological and chemical changes in the plant body and drain the plant's photosynthate and nutrients. Infection of young plants may be lethal, while infection of mature plants causes decreased yield.

Phytonematodes are important limiting factor in plant growth. Majority of the plant species, which account for the world's major food supply, is susceptible to attack from phytonematodes. They are capable of causing economic losses in terms of quantity and quality of the crops. The crop losses caused by phytonematodes in economic terms are estimated to be about \$ 157 billion annually to the world agriculture [1]. Yield losses due to root-knot nematodes (*Meloidogyne* spp.) range from 35.0% to 39.7% [7].

Every part of higher plants (roots, stems, trunk, and leaves) is susceptible to attack by one or more plant parasitic nematode (PPN) species. PPN are a serious challenge to farmers and home gardeners. The use of soil fumigants for pest control is often undesirable due to their residual toxicity in plants and soils and unfavourable effects on humans and animals. The complexity of treatment, high cost of chemicals and increasing interest for organic agriculture demands unconventional ways of phytonematode management [8].

Soil solarisation is a tested method in many tropical and subtropical countries; however, location specific studies are lacking in Nigeria in general and Northern Nigeria in particular. It is hypothesized that the use of this method allows the plants to developed healthy root and shoot system and improves the growth of plant and soil structure which needs to be investigated and documented.

The aim of this research was to study the suitability of soil solarisation as a physical control mechanism to manage phytonematode population in soil and its effect on plant growth in Northern Nigeria.

# MATERIALS AND METHODS

#### Study area

The study was conducted from June to September 2019 in the experimental fields of Yusuf Maitama Sule University, Kano. Kano is a state located in the north-western Nigeria and borders Katsina state to the north-west, Jigawa state to the north-east and Bauchi and Kaduna states to the south [9]. It's coordinated positioning lies between latitude 130° in the North and 110° in the South and longitude 80° in the West 100° in the East. Kano State is made up of forty-four local government areas (administrative units). The total land area of Kano state is 20,760 km2 with the population of approximately 10 million people with majority of them belonging to the agrarian society. Kano has a mean height of about 472.45 m above sea level with an average rainfall of 696.4 mm/year. In Kano, the month with the most daily hours of sunshine is May with an average of 11.32 hours of sunshine. In total there are 350.88 hours of sunshine throughout May. The month with the fewest daily hours of sunshine in Kano is January with an average of 10.42 hours of sunshine a day. In total there are 312.57 hours of sunshine in January [10]. On an average around 3748 hours of sunshine are counted in Kano throughout the year and there are 123.29 hours of sunshine per month (Figure 1).

# Site layout

The experimental site was divided into 12 beds of  $1 \times 3$  m beds each and labelled T1, T2, T3 and Control. There were three replicates for each treatment i.e., 3 plots for each treatment were used [11]. Each bed was covered with a different thickness of polythene sheets. Untreated and uncovered beds served as control. All the beds were dug approximately up to 20 cm of depth (Figure 2).





# Polythene sheet

Polythene sheets of different thickness *viz.* 50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m, to cover the soil beds so as to find the best option for soil solarisation was used. The size of the sheets was 1.5 m × 3 m for all the nine beds [12].

# Soil sampling

The soil samples were collected using an auger from all the 4 plots weekly to monitor the effect of soil solarisation. The soil samples are collected at a depth of 10 cm from each of the plots where polythene sheets were covered and from the control. The samples were collected weekly to quantify the presence, number and types of phytonematodes in the soil for 5 weeks starting from May 2018. The same field was planted with tomato plants having 9 plants in each bed after soil solarisation. Observations were recorded on structured format and were duly digitalized weekly. The plants were left to a height of 20 cm when the growth parameters were noted [13,14].

The temperature of the soil was measured using digital soil thermometer which was inserted in the soil and removed after few minutes. The readings on the thermometer were then recorded.

The plots were then compared among themselves after the duration of time [15].

# Phytonematodes extraction from the soil

Each soil sample was thoroughly mixed, and a subsample of 100 ml soil was assayed for nematodes [16]. For the root samples, a subsample of 10 g roots were washed, chopped into 1 to 2 cm pieces and ground for 1 to 2 minutes using a blender and nematodes were extracted using the above mentioned technique. Phytonematodes were recovered after an incubation period of 48 hrs and identified to genus level in a count plate under a binocular microscope. To confirm the observation with a magnifying binocular microscope and refine the identification a post-lethal observation of individuals killed by heat was carried out. Nematodes were fixed in 4% formalin, processed in glycerol, and mounted on a glass slide based on a standard procedure. Observations were done under the microscope at X4, X10, X40 and X100 following the identification keys. Three subsamples with equal volume (10 ml) were counted three times, and the average was used to estimate the nematode population prevalence and densities. Prevalence, computed as frequency, of the recorded genera in each zone was calculated according to Luquini 2019.

Prevalence: [(number of field positive for genus)/(total number of fields in zone)] x100

#### Identification of phytonematodes

A sample of nematode is carefully placed on a glass slide and a drop of water was added. The glass slide was placed under a microscope and viewed at different magnification until a clear image was obtained. *Meloidogyne* species were identified using perennial pattern observations under compound microscope. An available key was consulted for identification of phytonematodes 002E [17].

# Recording of growth parameters (Duraisamy, 2017)

The growth of the parameters was defined and plants were observed on the following parameters after they attain a height of 20 cm [18-25].

All the treated plots were planted with local variety of tomato seedlings and were observed over a period of five (5) weeks for different growth parameters *viz*. Root and shoot weight, root and shoot length and number of branches, buds, flowers and fruits were recorded. Untreated plots served as control. Volumetric data was collected every week and compared with the control.

#### Statistical analysis

The collected data were processed using Microsoft Excel and R statistical programing language (R Development Core Team 2021) software. Two-way Analysis of Variance (ANOVA) was conducted to see if differences exist across the treatments and controls. This was followed Turkey Post Hoc test to observe where the differences are [26-30].

# RESULTS

# Nematological studies

Ten (10) major plant parasitic nematodes were recorded from the experimental site in the pre-treatment samples. It was found that IInd stage juvenile of *Meloidogyne incognita* was highest in number (17.67/100 ml soil) followed by *M. javanica* (11.00/100 ml soil). These two spp. proves to be the highest in terms of frequency (no. of times each spp. appears in the total sample done) as well with 100% and 91.67% recording from the total samples taken (Table 1).

*Paratrichodorus spp.* was recorded as the least number of phytonematodes present in the soil (1.33/100 ml soil) while Longidorus spp. was the least occurring spp. in terms of occurrence [31].

Table 1: The types,	number and frequenc	of different phytonematod	es recorded from experimental	site in 100 ml soil.
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S/No.	Types of phytonematodes	Number/100 ml of soil	Frequency (%)
1	Meloidogyne incognita	17.67	100.00
2	Meloidogyne javanica	11.00	91.67
3	Rotylenchus reniform	9.67	75.00
4	Ditylenchus dipsaci	8.33	83.33
5	Ditilenchuis destructer	7.00	75.00
6	Pratylenchus spp.	5.00	58.33
7	Trichodorus spp.	4.67	83.33
8	Xiphinema spp.	8.67	83.33
9	Paratrichorus spp.	1.33	66.66
10	Longidorous spp.	9.00	33.33

Table 2 depicts percentage reduction in nematode population in soil, solarised over a period of 5 weeks using different thickness of polythene sheets *viz.* 50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m. It was found that polythene sheet having a 50  $\mu$ m was the best in controlling phytonematodes below economic threshold level. It was found that there is gradual increase in the percentage reduction in nematode population from different thickness of polythene sheets from week 1 to week 5 (Table 2).

Two-way ANOVA showed significant differences across the treatments and controls (df=5, p<0.01). Turkey Post Hoc test reveals the differences that exist between the treatments and controls (Table 3 and Figure 3).

#### Growth parameters

**Length of root:** The mean root lengths after the observation period were 6.82 cm, 6.40 cm and 5.96 cm for the plots treated with 50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m thick polythene sheets, respectively (Figure 4). While the average root lengths for untreated control were 5.76, 5.80 and 5.46 cm (average 5.67 cm). The mean difference in root lengths when compared with the average root length of control plants were 20.21%, 12.81% and 5.05% for plants under 50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m thick polythene treated plots, respectively. The percentage differences were on the positive side of the enhancement over control.

**Length of shoot:** The mean shoot lengths over a period of five weeks were 21.04 cm, 19.42 cm and 18.78 cm under plots treated with 50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m polythene sheets, respectively. During the same period the average control shoot height was registered as 18.26 cm. In terms of percentage difference, it again registered 15.24%, 6.37% and 2.86% increase for plants grown in plots treated with 50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m polythene sheets, over the average shoot length of control (Figure 4).

**Root weight:** The mean root weights were 4.64 g, 4.04 g and 3.48 g under plots treated with 50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m polythene sheets, respectively, over period of five weeks. The average root weight of tomato plants was 2.90 g for control during the same period of time. The root weight registered an increasing trend under treated plots with 50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m thick polythene sheets treated plots showing a growth of 60.00%, 39.31% and 20.00% respectively over control [32].

**Shoot weight:** An overall, pronounced positive enhancement in the shoot weights of plant grown on plots treated with different (30  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m) thicknesses of polythene sheets were observed. Mean shoot weight of plants grown on plots treated with 50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m thick polythene sheets were 37.20 g, 33.42 g and 31.00 g over a period of five weeks. The average shoot weight of controlled plants was 22.01 g during the same period. The total difference was 69.04%, 51.86% and 40.87% for the three different polythene thicknesses respectively (Figure 5).

Number of branches: The mulching treatment with different thickness of polythene (50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m) used to treat the beds yielded positive results in terms of enhancement in number of branches being produced by the plants. Specifically, it was found that over a period of five weeks' time, plants sown in beds covered with a 50  $\mu$ m sheets showed 26.82% higher number of branches than controlled. Likewise, plants sown o beds covered earlier with 75  $\mu$ m and 100  $\mu$ m sheets, showed 18.47% and 10%, 84%, higher number of branches.

Number of leaves: The enumeration data showed an inconsistent counting over a period of five weeks, where by the plants sown in beds covered with a 50  $\mu$ m sheets showed 17.40% higher number of leaves than in plants sown in controlled beds. However, plants sown in beds which were earlier covered with 75  $\mu$ m and 100  $\mu$ m polythene sheets registered a negative growth of 3.58% and 10.85% respectively (Figure 6).

Number of buds: The numbers of buds were high in all the plants sown in beds covered with different thickness of polythene sheets (50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m) in comparison to plants sown in controlled plants. The plants sown on beds covered earlier by 50  $\mu$ m sheets showed an increase of 41.29% than controlled bed's plants. A total of 13.23 and 28.73% increase in the total number of buds were also recorded in plants sown on beds covered with 75  $\mu$ m and 100  $\mu$ m polythene sheets [33-35].

Number of flowers: The flower settings over a period of five weeks' time showed some marked variation. It was found that average no. of flowers over the said period were 3.81% higher than control, in plants planted over the beds earlier covered with 50 µm polythene sheets. However, plants sown in beds earlier covered with 75 µm and 100 µm sheets showed a negative growth of 22.86% and 29.52% over control (Figure 7).

#### Sultan Z, et al.

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 Table 2: Shows the average percentage reduction in phytonematodes population under different polythene sheet recorded over 5 weeks under field conditions.

C /NI	TT (1, 1, 1)	Average no. of PPN in 100 ml soil								
5/ No.	Types of phytonematodes	T1	C1	% dec.	T2	C2	% dec.	T3	C3	% dec.
1	Meloidogyne incognita	3.67	10.33	64.52	5.67	10.00	43.33	10.00	10.67	6.25
2	Meloidogyne javanica	2.67	8.67	69.23	3.67	8.00	54.17	8.00	7.67	-4.35
3	Rotylenchus reniform	3.00	10.33	70.97	4.00	10.00	60.00	10.00	10.67	6.25
4	Ditylenchus dipsaci	2.00	6.33	68.42	2.67	6.67	60.00	6.67	5.67	-17.65
5	Ditilenchuis destructer	2.33	6.33	63.16	3.67	6.33	42.11	6.33	6.67	5.00
6	Pratylenchus spp.	3.00	8.33	64.00	3.67	9.33	60.71	9.33	8.00	-16.67
7	Trichodorus spp.	2.67	8.67	69.23	5.33	9.00	40.74	9.00	8.33	-8.00
8	Xiphinema spp.	4.33	10.33	58.06	6.67	11.00	39.39	11.00	11.67	5.71
9	Paratrichorus spp	4.00	8.67	53.85	5.67	8.00	29.17	8.00	9.67	17.24
10	Longidorous spp.	3.33	9.00	62.96	5.33	9.33	42.86	9.33	9.00	-3.70
	Total	31.00	86.99	64.37	46.35	87.66	47.12	87.66	88.02	0.01

Table 3: Tukey Post Hoc test across the treatments and controls.

	Difference	Lower	Upper	Parallel adjustment
C2-C1	0.067	-1.7781322	1.91213224	0.9999979
C3-C1	0.168	-1.6771322	2.01313224	0.9997980
T1-C1	-5.599	-7.4441322	-3.75386776	0.0000000
T2-C1	-4.064	-5.9091322	-2.21886776	0.0000004
T3-C1	-1.598	-3.4431322	0.24713224	0.1255917
C3-C2	0.101	-1.7441322	1.94613224	0.9999837
T1-C2	-5.666	-7.5111322	-3.82086776	0.0000000
T2-C2	-4.131	-5.9761322	-2.28586776	0.0000003
T3-C2	-1.665	-3.5101322	0.18013224	0.0991851
T1-C3	-5.767	-7.6121322	-3.92186776	0.0000000
T2-C3	-4.232	-6.0771322	-2.38686776	0.0000001
T3-C3	-1.766	-3.6111322	0.07913224	0.0681427
T2-T1	1.535	-0.3101322	3.38013224	0.1552498
T3-T1	4.001	2.1558678	5.84613224	0.0000006
T3-T2	2.466	0.6208678	4.31113224	0.0029928



Figure 3: Differences across treatments and controls.



**Figure 4:** Effect of different thickness (50 µm, 75 µm and 100 µm) polythene sheet on growth parameters (root and shoot length), on tomato plants over a period of five weeks, under field conditions. **Note:** (**1**) Length root (cm); (**1**) Length root (cm); (**1**) Height root (cm); (**1**) Height root (cm); (**1**) Height root (cm); (**1**) Height root (cm).



**Figure 5:** Effect of different thickness (50 µm, 75 µm and 100 µm) polythene sheet on growth parameters (root and shoot weight), on tomato plants over a period of five weeks, under field conditions. **Note:** (**1**) Root weight (cm); (**1**) Root weight (cm); (**1**) Shoot weight (cm); (**1**) Shoot weight.





**Figure** 7: Effect of different thickness of polythene sheets used for soil solarization and its effect on growth parameters (no. of buds, flowers and fruits) observed on host plant (tomato) over a period of five weeks, under field conditions.. **Note:** (**■**) No. of buds; (**■**) No. of buds (cm); (**■**) No. of buds (cm); (**■**) No. of flowers (cm); (**■**) No. of flowers (cm); (**■**) No. of flowers; (**■**) No. of fruits; (**■**) No. of fruits; (**■**) No. of fruits.

# DISCUSSION

In Table 1, the types, number and frequency of phytonematodes where presented. 10 phytonematodes were observed in which Meloidogyne incognita has the highest number of occurrence and Paratrichorus spp. has the lowest number of occurrence. Many scientists like Abad 2008, Bello 2020, Berry 1972 have reported that M. incognita population was highest in irrigated fields of Nigeria. In Table 2, there is a significant reduction in the number of phytonematodes which is in line with many observations done in many countries of the world. Shut 2021 also reported from Nigeria that the overall population of phytonematode in solarized soil was significantly higher than in unsloarized soil, which is in line with the present finding. Table 2 also shows that the plots that were covered with polythene sheet with the thickness of 50 µm have the highest reduction in phytonematodes population and that of 75 µm. The polythene sheet with 100 µm how ever showed the least reduction in phytonematode population which is line with the work of Porter and Merriman 1983 on solarisation that shows that there is significant reduction in phytonematodes population in solarized soil. The findings were also supported by work of Al-Kayssi and Karaghouli, 2002.

It also shows the various soil temperatures on different thickness of polythene sheets over 5 weeks, there is gradual increase in temperature among the plots covered with polythene sheets and it was found that the polythene sheet with the thickness 50  $\mu$ m has the highest temperature rise while the polythene sheets with thickness of 100  $\mu$ m has the lowest temperature. Carson and Otoo 1996 in their work of non-chemical management concluded that the plastic layers with thinner thickness (50  $\mu$ m) have the lower light reflect compared to the thicker 100  $\mu$ m and the medium 75  $\mu$ m, so it could increase the soil temperature more than the other two. The present findings are in line with the presented reference.

Table 3 shows the records of growth parameters that were taken during the survey in three different plots covered with 50  $\mu$ m, 75  $\mu$ m and 100  $\mu$ m polythene sheets thick. The plot that was covered with 50  $\mu$ m polythene sheet shows the highest number of leaves, fruits, and flowers content followed by 75  $\mu$ m with the medium content then 100  $\mu$ m with the least the correlated with the work

of Katan, 2000.

The percentage reduction in the number of phytonematode was significantly affected by the different thickness level of polythene sheets. The fact that the observed variance ratio (F=9.48), was greater than the table values (F=3.35, P<0.05) suggests that there was a highly significant treatment effect. The post-experimental analysis LSD, indicated that the difference between all the three treatment means (50 µm and 75 µm, 50 µm and 100 µm, 75 µm and 100 µm) were greater than the LSD value (3.99). This indicated that the mean of 100 µm (36.23) was significantly lower than the other treatment means. The differences between the means were significant as they exceeded 3.99. Since the mean of treatment 50 µm (44.69) was greater than other treatment means, it can be concluded that it induced greatest percentage reduction in number of phytonematode followed by 75 µm, while 100 µm resulted in a significantly lower percentage reduction in phytonematode.

# CONCLUSION

From the results obtained, it can be concluded that planting tomato on solarized mulched plot was better in the control of phytonematode infestation. Under the warm weather conditions of Northern Nigeria, solarization was proved to be an effective technique for an environmentally sustainable and cost-effective disinfestation of soil. It is therefore, recommended as a potential alternative to chemical measure in controlling phytonematode infestation and in maximizing tomato yield under field conditions in Nigeria. This can be attributed to the overall enhancement in various growth parameters such as; root length, shoot length, number of branches, number of leaves, number of flowers, number of fruits and yield respectively. It is however, recommended that further investigation be evaluated across different locations with varied soil types in Nigeria.

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