

Effect of mulches on virus severity, growth and yield of tomato (*Solanum lycopersicum*) inoculated with cucumber mosaic virus

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Abstract: Tomato plays a major role in human nutrition but viruses cause severe constraints in terms of yield and quality. There is however meager information on the effectiveness of mulches to ameliorate virus severity in tomato. Greenhouse experiment was conducted to assess the effect of polythene middies and grass mulches on virus severity, growth and yield of tomato mechanically inoculated with cucumber mosaic virus (CMV). Tomato seeds were sown in plastic pots filled with sandy-loam soil previously steam-sterilised. Seedlings were inoculated with CMV 21 days after germination and no-mulch as experimental control. The results indicated mulching significantly ($P \geq 0.05$) ameliorated virus severity. At 7th WAI polythene middies and grass mulches had virus severity of 12.4% and 16.5% respectively compared with 20% non-mulch plants. Vine length was tallest in the polythene middies (13.3 – 58.1 cm) and grass mulch (14.8 – 49.9cm). The average number of leaves (38.4) and stem girth (3.1cm) were significantly low for non-mulch compared to other treatments. The effect on yield indicated fruit weights being significantly ($P \geq 0.05$) higher for polythene middies (322.3g) and grass (225.1g) mulches compared to non-mulch (164.5g). The study recommends polythene middies and grass as mulch in managing viral infection in tomato.

Keywords: Mulch; rural farmer; sustainable control; tomato; virus management

1. Introduction

Tomato (*Lycopersicon esculentum* Mill.) is an annual warm-season crop that belongs to *Solanaceae* family with origins in South America. It is one of the important vegetable crops cultivated in the world with high consumption rate and nutritionally important because of its high contents of antioxidants including carotenoids, lycopene and phenolic compounds, which offer a lot of health benefits for the consumers (Nahar & Gretmacher, 2002). The fruit is a good source of energy and plays major roles in human nutrition. It is also an excellent source of phosphorus, iron and vitamins A, B and C (Varela et al., 2013; Naika et al., 2005). Tomato is widely cultivated in Nigeria by subsistence farmers and the average daily consumption in most Nigerian homes is about 18% of the whole consumed vegetables (Aja, 2012). Fresh market tomato fruit quality can be

described by physical characteristics such as color, shape, size, defects, firmness and flavor (Etissa et al., 2014). However, tomato yield and quality are influenced by various production constraints such as varietal characteristics, diseases, insects, soil factors, climate variability and cultural practices (Tshiala & Olwoch, 2010).

Diseases are considered to be among the most serious factors responsible for low yield of tomato. The crop is susceptible to more than 200 diseases out of which 40 are caused by viruses (Lukyanenko, 1991), as virus diseases are regarded as one of the most important pathogenic tribulations affecting production in many countries (Ribeiro et al., 2009; Davinoa et al., 2009). Globally, viruses cause severe constraints on the productivity of tomato in terms of yield and quality (Dasgupta et al., 2003). There are about 75 viruses

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which infect tomato and cucumber mosaic virus (CMV) is reported to be among the most devastating in Nigeria (Li et al., 2009; Ayo-John & Odedara, 2017).

Cucumber mosaic virus (CMV) with its 59 strains causes damage to tomato production by infecting and reducing yield all over the world (Kaper & Waterworth, 1981; Jorda et al., 1992). CMV has an extensive host range and is transmitted by aphids in a non-persistent manner. The virus is not seed borne in tomato and does not persist in plant debris in the soil, but is spread by aphids from infected plants (Jiang et al., 2004). The CMV Genome consists of RNA; single-stranded; linear; of three parts; largest (or only) genome parts the largest 3.389 kb; the 2nd largest 3.035 kb; the 3rd largest 2.197 kb. Virions isometric; not enveloped; 29 nm in diameter; rounded in profile; without a conspicuous capsomere arrangement. Virions contain 18 % nucleic acid; 82 % protein; 0 % lipid (Brunt, 1996).

Insect vectors that spread diseases in tomato are known to identify host plants by color, shape, size, and floral volatiles. The use of pesticides to control aphids that act as vectors of viruses is usually ineffective (Riley & Pappu, 2000). Insecticides may actually enhance the spread of aphid-borne viruses by stimulating vector activity (Charles et al., 2005). Many insecticides stimulate the aphid's nervous system, causing it to move from plant to plant very rapidly. This results in the infection of more plants than would occur in those visited by a non-intoxicated aphid, which settles down and feeds on one plant before moving to another plant. This occurs before the insecticide has a lethal impact on the aphid. In contrast, whitefly infestations can be somewhat relieved by a systemic insecticide. However, the development of resistance to systemic and other insecticides among insect vectors is a major concern (Elbert & Nauen, 2000).

As a compliment to chemical control of aphids that are vectors of viruses, cultural control measures such as the use of mulches may be considered as part of virus management strategy (Stapleton & Summers, 2002). Farmers and horticulturists naturally use mulching as a method of improving the condition of agricultural soils by covering the soil surface with different kinds of materials (Chakraborty et al., 2008). Improvement of the soil physical environment contributes to better plant production and covering of the ground with mulch adds organic matter to the soil, reduces weed growth, reduces or eliminates erosion and enhances plant health (Bot & Benites, 2005).

Researchers have reported that silver colored plastic mulch effectively reduces insect vectors (Kelley, 2009). Polyethylene and cover cropping consequently now

show promise as methods for excluding insects and reducing viral diseases (Webb & Linda, 1992). There is however modest information on the effectiveness of using polyethylene middies and grasses as mulches for virus control in tomato. The objectives of this study therefore were to: (1) establish the effectiveness of polyethylene middies and grass mulches in ameliorating the severity of CMV in tomato under greenhouse conditions; (2) assess the effect of polyethylene middies and grass as mulches on the growth and yield of tomato mechanically inoculated with CMV.

2. Materials and methods

2.1. Experimental design and inoculation procedure

The tomato variety used was Tomato Rio Grande obtained from Techni seed limited Kano, Kano State, Nigeria. Four seeds were sown per 10-litre (50 cm diameter) in plastic pots filled with sandy-loam soil, previously steam-sterilised at 121°C for 60 minutes. CMV infected leaf samples were obtained from the International Institute for Tropical Agriculture (IITA) Ibadan-Nigeria and the virus extracted by homogenisation. This was achieved by macerating the infected leaf samples using mortar and pestle in 0.01 M phosphate buffer at pH 7.2 at the rate of 1g leaf sample to 5ml of buffer (Balogun & Aliyu, 2005). One part virus infected tissue was ground up in a small mortar with 2 to 5 parts buffer, generally 0.01M phosphate buffer, pH 7.2. The inoculum was kept cool and used immediately. The buffer was prepared in the following way: Solution A: 1.36gKH₂PO₄ in 1000ml H₂O. Solution B: 1.78gNa₂V-PO₄ x 2 I-0 in 1000ml H₂O. 51.0 ml of solution B mixed with 49.0 ml of solution A gives 100 ml of 0.01 M phosphate buffer solution pH 7.2 (Noordam, 1973).

Plants were inoculated at 21 days after germination (DAG) by slight dusting of the primary leaves with carborundum (silicon carbide, 400-600mesh) and celite (diatomaceous earth) to increase infection by providing wounds for the entry of virus particles. The abrasive was finely dusted over the leaf surface and also suspended in the inoculum (0.5-1% w/v) before inoculation after which the leaves were then rubbed with the extracted juice of the inoculum, using cotton wool. The inoculation was carried out by moistening 1 or 2 fingers covered with hand gloves in inoculum and rubbing gently onto the leaves, while supporting them with the other hand. The plants were rinsed with distilled water thereafter to reduce inoculation stress (Aliyu, 2018). The treatments (polyethylene middies, grass mulch and no-mulch) were then applied, replicated 5 times and arranged in the green house in a Completely Randomised Design.

2.2. Data collection

Data were collected on weekly basis after inoculation for vine length, number of leaves, number of leaves showing virus disease symptoms, number of flowers, number of fruits and fresh fruit weight at harvest. Percentage disease severity was determined by using a modified formula-grading scheme of Merritt et al. (1999).

2.3. Statistical analysis

All collected data were subjected to analysis of variance (ANOVA) using the Statistical Package for the Social Sciences SPSS version 15.0 and the significant means separated using The New Duncan Multiple Range Test at 5 % level of probability.

3. Results

3.1. Effect of treatments on virus severity

Mulches had significant effect on ameliorating virus severity and the effect became apparent from the 3rd WAI with 3.1% and 3.5% virus severity for polythene middies and grass mulches, respectively (Table 1). These values were significantly ($P \geq 0.05$) lower than 7.5% obtained in no-mulch. At the 4th WAI, polythene middies mulch had the significantly lowest virus severity (4.1%) compared to the grass mulch (5.9%) and no-mulch (9.8%) treatments. This trend continued to the

7th WAI with 12.4%, 16.5% and 20.0% virus severity for polythene middies, grass mulches and no-mulch, respectively (Table 1).

3.2. Effect of treatments on vine length of tomato inoculated with CMV

The effect of the applied treatment on vine length of tomato inoculated with CMV revealed that from the 1st to the 7th WAI the tallest vines were in the polythene middies (13.3 – 58.1 cm) and grass mulches (14.8 – 49.9cm), while the shortest plants (11.5 – 42.1 cm) were found in no-mulch treatment (Table 2).

3.3. Effect of treatments on stem girth of tomato inoculated with CMV

There were no significant differences in the stem girth of the treatment plants at t 1st, 2nd, 4th, 5th and 6th WAI (Table 3). However, significant differences were observed at the 3rd and 7th WAI. The highest values were in the polythene middies and grass mulches for the 3rd (2.2 and 2.4 cm) and 7thWAI (3.3 and 3.7 cm), respectively.

3.4. Effect of treatments on average number of leaves per plant on tomato inoculated with CMV

Effect of the treatments on average number of leaves per plant (Table 4) became manifested at the 3rd WAI, with the highest number of leaves recorded on grass mulch

Table 1: Effect of treatments on virus severity

TREATMENT	WEEKS AFTER INOCULATION						
	wk 1	wk 2	wk 3	wk 4	wk 5	wk 6	wk 7
Polythene middies	0.7	1.5	3.1 ^b	4.1 ^c	7.4 ^c	9.6 ^c	12.4 ^c
Grass mulch	0.6	1.6	3.5 ^b	5.9 ^b	11.8 ^b	13.6 ^b	16.5 ^b
No-mulch (control)	0.8	3.0	7.5 ^a	9.8 ^a	17.8 ^a	18.6 ^a	20.0 ^a
S.E.M	0.031	0.383	0.519	0.548	0.945	0.855	0.928

Means within a column followed by the same superscript letter are not significantly different using The New Duncan Multiple Range Test at $P \geq 0.05$

Table 2: Effect of treatments on vine length (cm)

TREATMENT	WEEKS AFTER INOCULATION						
	wk 1	wk 2	wk 3	wk 4	wk 5	wk 6	wk 7
Polythene middies	13.3 ^{ab}	20.6 ^a	30.3 ^a	41.1 ^a	50.4 ^a	57.4 ^a	58.1 ^a
Grass mulch	14.8 ^a	22.1 ^a	32.8 ^a	39.4 ^a	43.4 ^b	49.3 ^b	49.9 ^b
No-mulch (control)	11.5 ^b	17.6 ^b	20.3 ^b	22.0 ^b	34.4 ^c	40.8 ^c	42.1 ^c
S.E.M	0.516	0.665	1.390	1.874	1.497	1.566	1.515

Means within a column followed by the same superscript letter(s) are not significantly different using The New Duncan Multiple Range Test at $P > 0.05$

Table 3: Effect of treatments on stem girth (cm)

TREATMENT	WEEKS AFTER INOCULATION						
	wk 1	wk 2	wk 3	wk 4	wk 5	wk 6	wk 7
Polythene middies	1.5	1.6	2.2 ^a	2.8	2.9	3.2	3.3 ^{ab}
Grass mulch	1.7	1.9	2.4 ^a	2.8	3.3	3.6	3.7 ^a
No-mulch (control)	1.6	1.7	1.7 ^b	2.7	2.8	3.0	3.1 ^b
S.E.M	0.0423	0.5161	0.0923	0.0660	0.113	0.121	0.118

Means within a column followed by the same superscript letter(s) are not significantly different using The New Duncan Multiple Range Test at $P > 0.05$

Table 4: Effect of treatments on the average number of leaves per plant

TREATMENT	WEEKS AFTER INOCULATION						
	wk 1	wk 2	wk 3	wk 4	wk 5	wk 6	wk 7
Polythene middies	13.1	23.1	33.9 ^a	53.9 ^a	58.5 ^a	59.1 ^a	60.1 ^a
Grass Mulching	16.6	24.3	36.5 ^a	41.3 ^b	44.3 ^b	44.1 ^b	43.3 ^b
No-mulch (control)	14.8	20.9	25.9 ^b	30.4 ^c	37.0 ^c	37.6 ^c	38.4 ^c
S.E.M	0.8113	1.095	1.371	2.536	2.274	2.291	2.377

Means within a column followed by the same superscript letter are not significantly different using The New Duncan Multiple Range Test at $P \geq 0.05$

(36.5) but this value was not significantly ($P \geq 0.05$) different from the number of leaves on the polythene mulch (33.9). This observable trend continued to the 7th WAI with the polythene middies treatment having the highest average number of leaves per plant (60.1). This was followed by grass mulch (43.3) and no-mulch (38.4).

3.5. Effect of treatments on average number of flowers on tomato inoculated with CMV

The average number of flowers per plant was significantly higher in polythene middies from the 4th to 7th WAI (2.4 to 18.3) than grass mulch and no-mulch with 0.5 – 12.9 and 0.0 – 7.5, respectively (Table 5).

Table 5: Effect of treatments on average number of flowers per tomato plant

TREATMENT	WEEKS AFTER INOCULATION			
	wk 4	wk 5	wk 6	wk 7
Polythene middies	2.4 ^a	10.6 ^a	13.9 ^a	18.3 ^a
Grass mulching	0.5 ^b	4.3 ^b	9.0 ^b	12.9 ^b
Non-mulching	0.0 ^b	1.1 ^c	4.5 ^c	7.3 ^a
S.E.M	0.279	0.890	0.893	1.000

Means within a column followed by the same superscript letter are not significantly different using the New Duncan multiple Range Test at $P \geq 0.05$

3.6. Effect of treatments on yield parameters in tomato inoculated with CMV

Polythene middies mulch had the significantly higher number of fruits recorded for the 8th WAI (12.6) than the grass mulch and no-mulch treatments with 7.9 and 4.4, respectively (Table 6). Similarly, the trend was also observed in the respective fruit weights of the treatments.

Table 6: Effect of treatments on yield components

TREATMENT	Average number of fruits at 7 WAI	Average number of fruits at 8 WAI	Average fruit wt per plant (g)
Polythene middies	8.4 ^a	12.6 ^a	322.3 ^a
Grass mulching	4.8 ^b	7.9 ^b	225.1 ^b
No-mulch (control)	0.6 ^c	4.4 ^c	164.5 ^c
S.E.M	0.697	0.778	14.211

Means within a column followed by the same letter are not significantly different using The New Duncan Multiple Range Test at $P \geq 0.05$

4. Discussion

This research was conducted to compare the effectiveness of polyethylene middies and grass mulches in ameliorating the severity of virus diseases. It was also designed to determine the effect of the mulches on growth and yield

components in tomato inoculated with CMV. The results obtained from the present study indicated that polythene middies used as mulch ameliorated the severity of CMV in tomato in comparison to no-mulching. The finding is in tandem with Kapoor (2012) who also found polythene mulches to be effective in reducing virus severity in bell pepper. Murphy et al. (2009) also reported achieving decreasing virus severity using reflective plastic as mulch for watermelon infected with *Water melon mosaic virus*. The study also found using grass mulch to be effective in reducing virus severity in viral inoculated tomato. This finding is in agreement with Rwezaula et al. (2005) who confirmed that grass mulch excelled in the control of pathogenesis in tomato.

The mechanism by which the mulches decreased viral severity is attributable to the effective weed suppression. Weeds serve as alternate hosts for viruses and provide optimum environment for virus proliferation. Mulches on the other hand are widely used to suppress weeds and cause reduction in weed abundance on crops. It is therefore possible to assume that the condition of fewer weeds within the plant canopy created by mulching resulted in lower virus severity compared to the control. This view is corroborated by Momol et al. (2002) and Shtienberg et al. (2010).

The study indicated that the mulches were effective in improving plant growth and number of leaves in virus inoculated tomato compared to control. Similar findings were also made by Makus et al. (1994). It can be suggested that growth parameters positively affected by mulching is due to higher soil moisture conservation and reduced water stress. This view is in agreement with Gordon et al. (2008) and Wadas et al. (2009). They reported plants sown on mulched soil were much taller than those grown on non-mulched soil. It was elucidated that mulched soil has higher air temperature and provided better conditions and nutrients for growth.

The yield components were also enhanced in the mulched tomato compared to the non-mulched plants. Ravinder et al. (1997) and Singh et al. (2009) reported that mulching significantly improved the number of fruits per plant and reduced the percentage of fruit abortion compared to non-mulched control. It can be suggested that yield increases observed in the mulched plots is associated with improved microclimate both beneath and above the soil surface.

5. Conclusion and recommendation

The use of polythene middies and grass as mulches is suitable and recommended for ameliorating CMV infection in tomato. This would provide a wholesome

approach to managing tomato virus diseases in the greenhouse especially for resource poor farmers and consequent increased production.

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