

Effects of Time of Planting, Plant Population Density and Weeding Regime on the Incidence and Severity of Cowpea Mottle Virus Disease (CPMoV) on Cowpea [*Vigna unguiculata* (L) Walp.]

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Abstract

Field experiments were carried out in 2011 and 2012 planting seasons to evaluate the effects of time of planting, weeding regime and plant population density on the incidence and severity of cowpea mottle virus on cowpea. The experiment followed factorial treatments fitted into a randomized complete block design. The application of the factors considered in the experiment, were at two levels: time of planting cowpea (1st August and 15th August, 2011 and 2012), plant population (106,666 plants/ha and 53,333 plants /ha), and weeding regime (chemical and mechanical). The results showed that planting cowpea at intervals of 1st August and 15th August, was of no significant effect on virus incidence (33.1 % and 34.3% for 2011; 34.6% and 35.7% for 2012), also the severity index value (2) were similar. Weeding regime and plant population density, on the other hand, significantly influenced the virus incidence but not severity. Plant population of 106,666 plants/ha, had the significantly lower virus incidence of 28.0% and 31.7% for 2011 and 2012. Virus incidence was also lower in plants that had chemical and mechanical weeding (31.6% and 33.8% for 2011 and 2012 respectively). The treatment combination effect indicated that significantly higher crop yields were obtained at early planting, at the population of 106,666 plants/ha, with chemical and mechanical weeding. The results indicate that sowing early, at moderately high plant population density, combined with an efficient weeding regime, as a cultural control practice, is effective in ameliorating cowpea mottle virus disease for an improved cowpea yield.

Keywords: Cowpea Mottle Virus , *Vigna unguiculata*, Incidence, Severity, Time of planting, Plant Population, Weeding regime, Cultural control.

Introduction

Cowpea (*Vigna unguiculata* [L.] Walp) is an annual tropical grain legume that plays an important role in the nutrition of people in developing countries of the tropics and subtropics, especially in sub-Saharan Africa, Central Asia, and South America (Kareem and Taiwo, 2007). The popularity of cowpea is due to its importance in the diet of man and is a cheap source of plant protein (Alabi *et al.*, 2003).

Cowpea grows in a wide range of soil pH and temperature (18-28°C) compared to other legumes and also has a considerable adaptation to high temperatures and drought compared to other crop species (Ehlers and Hall, 1996). The majority (64%) of the cowpea production, 12.5 million tons worldwide takes place in the sub-Saharan Africa (Langyintuo *et al.*, 2003).

Cowpea yields are generally poor in Africa at the farm level, hardly above 300kg ha⁻¹ (Omongo *et al.*, 1997). Nigeria accounts for over 70% of the total world cowpea production, amounting to over 5 million tons in 2002 on a total surface area of 14 million hectares (Grubben and Denton, 2004). Diseases, induced by various pathogenic groups - fungi, bacteria, viruses, nematodes and parasitic flowering plants parasitic are the major constraints to cowpea production in Nigeria (Asiwe, 2005, Mbong *et al.*, 2012).

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Viral diseases of cowpea have been reported to cause appreciable losses in yield if the plants are infected in early growth stages (Booker *et al.*, 2005). In Nigeria, some of the viruses that have been reported on legumes include: *Bean common mosaic virus* (BCMV), genus *Potyvirus* (Hughes and Tarawali, 1999); *Cowpea aphid-borne mosaic virus* (CABMV), genus *Potyvirus* (Shoyinka *et al.*, 1997), *Peanut mottle virus* (PMoV), genus *Potyvirus* and *Bean yellow mosaic virus* (BYMV), genus *Potyvirus* (Yusuf, 1999); *Cucumber mosaic virus* (CMV), genus *Cucumovirus*. *Cowpea mosaic virus* (CPMV), genus *Comovirus* and *Cowpea mottle virus* (CPMoV), genus *Carmovirus* have also been reported from *Vigna unguiculata* (Odedara, 2011).

Cowpea mottle virus (CPMoV) genus *Carmovirus* was first reported from Nigeria in 1963 and it is sap-and seed-transmissible and vectored by beetles including *Ootheca mutabilis* and *Paraluperodes quaternius* (Shoyinka *et al.*, 1979). The virus induces distortion and reduction in leaf size and witches broom syndrome in cowpea. It also causes mottling and bright yellow mosaic. CPMoV is most commonly found in the riverine area of the middle belt of Nigeria, a region with southern Guinea savanna type of climate. Surveys conducted in Nigeria between 1991 and 1993 showed incidences of 9.8%, 0%, and 7.9% and prevalences of 29.2%, 0%, and 40.9% for the three years, respectively (Taiwo, 2003).

Viruses must have one or more effective means of spread from an infected to uninfected plants if they are to persist in host populations and not be at the risk of total extinction, although there are certainly many tropical viruses that spread naturally into or within crops, but for which no vector has yet been determined (Brunt *et al.*, 1990). Rates of transmission however are dependent on host species and cultivars, time of infection, environmental conditions and the strain of the virus (Thresh, 1974; Shukla *et al.*, 1994).

The importance of cultural control practices such as, crop residue disposal, planting dates, removal of alternate hosts and plant population density have been

adapted to deal with plant-virus problems. Cock (1986) contended that when these cultural measures are properly and extensively employed, it could reduce pesticide use by as much as 50%.

The objective of this study therefore, was to evaluate the effect of the use of cultural practices such as varying the time of planting, chemical / manual weeding and plant population density manipulation, on the incidence and severity of cowpea mottle virus disease in cowpea.

Materials and Methods

Experimental design

A factorial treatment fitted into a randomized complete block experimental design was established at the Teaching and Research Farm of the University of Ilorin – Nigeria in 2011 and 2012 cropping seasons. The factors considered were time of planting, plant population density and weeding regime effect on virus incidence on cowpea variety Ife - Bimpe, which is a widely cultivated variety in the guinea savannah agroecology.

Field layout

Land preparation followed the conventional method of ploughing, harrowing and ridging. Each experimental plot consisted of nine ridges, and each of this ridge was 25m long. The seeds of the cowpea cultivar used in the experiment (Ife- Bimpe) was obtained from the National seed service center Ilorin, Kwara State - Nigeria.

Agronomic practices

The seeds were planted at the rate of four seeds per hole and thinning to two plants per stand was done at fourteen days after planting. Cypermethrin dimethrate was sprayed at the rate of 50g a.i ha⁻¹ at 4weeks after seedling emergence and twice after flowering to control insects.

Treatment allocation

Planting was done twice at two weeks interval (1st August and 15th August 2011 and 2012 respectively), at plant population densities of 106,666 plants/ha (0.75 x 0.25m) and 53,333 plants /ha (0.75 x 0.50m). Chemical weed control was achieved by the application of a mixture of metolachlor and prometryn at 1.25 + 0.08kg a.i/ha using the knapsack sprayer. The mechanical control was done by hand weeding with the hoe. The weeding operations were carried out at the 4th and 6th week after planting.

Collection of Data

Data were collected on growth and yield attributes, while the virus disease incidence was estimated based on 20 plant observations per plot and calculated based on Allen et al (1983) formula, that is:

Percentage incidence =

$$\frac{\text{Number of infected plants (symptomatic plants)}}{\text{number of Plants sampled (20)}} \times 100\%$$

Virus severity was scored on a severity index scale of 1-5, based on extent of leaf damage and percentage number of leaves showing symptoms, whereby 1 = 1-20% (very mild), 2 = 21-40% (mild), 3 = 41-60% (severe), 4 = 61-80% (very severe) and 5 = 81-100% (almost dead).

The cowpea pods harvested at maturity from each plot at 75 – 80 days after planting were manually threshed and weight of grains appropriately measured using electronic weighing balance (Model Kerro No. Ka – 3002c).

Statistical analysis

Data were analyzed for variance at 5% level of significance following factorial treatment fitted into randomized complete block experimental design. Treatments were separated using the New Duncan's Multiple Range Test. The statistical package for the

social sciences (SPSS) version 11.0 was used for the analysis.

Results

Effects of treatments on viral disease incidence

The results showed that time of planting of August 1st 2011 and August 15th 2012 did not significantly affect cowpea mottle virus disease incidence for both years. The virus incidence in plots planted on August 1st was 33.1% for 2011 and 34.6% for 2012 planting seasons respectively. Cowpea planted 2 weeks later, had a virus incidence of 34.3% (2011) and 35.7% (2012). The severity index of the virus for both years was rated mild with a severity score of 2 (Tables 1a and 1b).

Evaluation of the effect of plant population and weeding regime for year 2011 and 2012 (table 1a and 1b), showed that both treatments, significantly influenced the incidence of mottle virus diseases on cowpea. The population of 106,666 plants/ha consistently produced the significantly lowest virus incidence for 2011 (28.0%) and 2012 (31.7%), while a significantly higher incidence was recorded at the population of 53,333 plants/ha (39.3% for 2011 and 42.9% for 2012 respectively). A combination of chemical with manual weeding was the most effective in suppressing the virus incidence. The results showed that the treatment produced the significantly lowest virus incidence for 2011 (31.6%) and 2012 (33.8), compared with chemical weeding alone (35.7% for 2011 and 38.2% for 2012).

The treatment combination effect on viral incidence for 2011 and 2012 (tables 1a and 1b), showed that the significantly lowest incidences were recorded in the plots planted on August 1st, at the population of 106,666 plants/ha, which crops had the regime of chemical and manual weeding. The viral incidences were 25.8% (2011), and 28.2% (2012). The severity of the virus however, ranged from mild (2), to severe (3).

Effects of treatments on selected growth attributes

The results showed that time of planting had significant effect on plant height. The plants sown on August 1st had significantly taller plants with the mean value of 52.8cm (2011) and 50.6 (2012) in comparison to those planted two weeks later. Weeding regime and plant population density however, did not have any significant effect on cowpea plant height (Table 2).

The mean number of leaves per plant was however significantly influenced by both the weeding regime and the time of planting. The combination of chemical weeding with manual weeding produced the significantly highest mean number of leaves per plant for 2011 (42.3) and 2012 (39.6), compared with the use of chemical weeding alone. In the same vein, planting early (August 1st) had the highest mean number of leaves per plant for both years (43.9 and 39.6), compared with late planting on August 15th, which had an average of 34.4 (2011) and 31.2 (2012) leaves per plant. Meanwhile, the plant population density per se did not significantly affect the mean number of leaves per plant. In year 2011, the values were 40.2 and 39.9 for the population of 106,666 and 53,333 plants/ha, while for 2012, the corresponding mean number of leaves per plant was 35.1 and 34.8 respectively.

The treatment combination effects (table 2), showed that plants sown early (August 1st), at the plant population density of 106,666plants/ha, with a regime of chemical and manual weeding produced the significantly tallest plants. The table showed that for 2011 and 2012 seasons, the mean heights of plants for this combination was 55.9cm and 49.9cm respectively. This same treatment combination produced the significantly highest number of leaves per plant for 2011(48.3) and 2012 (39.5).

Effects of treatments on selected yield attributes

The results showed that combining chemical

The results showed that combining chemical with manual weeding consistently produced the significantly highest values for all yield attributes. The grain weight/ha for the treatment for 2011 and 2012, was 576.4kg and 547.8kg respectively, while the corresponding grain weight/ha for chemical weeding alone was 555.9kg and 510.9kg. Time of planting considerations indicated that yield attributes was significantly higher in the early plantings (August 1st). The grain weight/ha was 629.9kg (2011) and 597.4kg (2012). Plant population density evaluation for the 2years also indicated that density of 106,666plants/ha, produced significantly higher grain weight/ha of 703.5kg and 701.3kg (Table 3).

The treatment combination effect on yield showed that, early planting (August 1st), at the population density of 106,666plants/ha, coupled with chemical and manual weeding, produced the significantly highest yields for grain weight per plot(2.3kg and 2.2kg), grain weight per plant (12.9g and 11.2g), and grain weight per hectare (849.9kg and 820.7kg).

Discussion

Kucharek and Purcifull (2001), reported that several weeds are hosts and sources of viruses and the removal of weeds from cropping areas, reduce potential sources of viral inoculum. The present study found that the combination of chemical with manual weeding (resulting in less weeds), caused a significant reduction in virus incidence and higher crop yields. Manual weed control, being an important aspect of the smallholder crop production system, requires a lot of human labour. Environmental and economic concerns have led to increased interest in chemical / manual weed control and reduced herbicide use (Edwards 1987). The low virus incidence observed in the chemical/manually weeded plots is attributable to effective removal of weeds which serve as sources of inoculum or as alternate host of viruses. Cohen *et al.*(1988), identified two weed species as sources of Tomato yellow leaf curl

virus(TYLCV) inoculum, and suggested eradication of weeds bordering plants in the management of virus diseases.

This present study also showed that virus incidence was higher at a lower plant population. Fargette *et al.* (1990), in a similar experiment, found the incidence of African cassava mosaic virus (ACMV), to be the greatest at the lowest plant population density. The manipulation of plant populations especially at relatively high plant densities have been shown by Ogenga-Latigo *et al.* (1992), to affect the dynamics of virus vectors that transmit viruses to plant crops. The concept of increasing the density of crop plants per unit area to decrease disease incidence is based on the principle that given a fixed number of vectors, the more crop plants there are per unit area, the smaller the proportion of plants those insects can infect (Broadbent, 1969).

The effect of the time of planting in this study, showed that virus incidence was however higher in the cowpea plants sown 2weeks later. The dissemination of virus infection throughout a host is the consequence of multiple processes that include replication of viral nucleic acids, protein synthesis and targeting, and cell–cell spread of infectious entities (Citovsky and Zambryski, 1991). Mushegian and Koonin (1993), had shown that plant virus infection spreads from epidermal to mesophyll cells within 4–10days, depending on temperature and other experimental conditions. This therefore could be the reason for the slightly lower virus incidence observed in early planted cowpea plants as compared with those planted 2weeks later.

This study has shown that cultural practices which have received disproportionately little attention from researchers, possibly due to the difficulty of testing by conventional methods, could play a significant role in cowpea mottle virus disease suppression in cowpea. The outcome of the 2year study clearly indicated that planting early in August with moderately high plant population density of 106,666plants/ha, coupled with an effective control of weeds using manual and chemical means, not only suppressed virus

thesources of Tomato yellow leaf curl virus disease in cowpea, but also lead to higher growth and yield gains.

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Table 1a Main and Treatment combination effects of weeding, time of planting and plant population density on Cowpea Mottle Virus Disease on cowpea in 2011 season

Weeding Regime	% virus Incidence	Severity index
Chemical weeding only	35.7b	2
Chemical + manual weeding	31.6a	2
Non - weeded	50.2c	3
Time of planting		
1 (August 1 st)	33.1	2
2 (August 15 th)	34.3	2
	NS	
Plant population density		
106,666plants/ha	28.0a	2
53,333plants/ha	39.3b	2
Plant population density X Time of planting X Weeding Regime		
106,666plants/ha x August 1 st x Chemical only	27.6ab	2
106,666plants/ha x August 1 st x Chemical + manual weeding	25.8a	2
106,666plants/ha x August 1 st x No-weeding	39.2d	2
106,666plants/ha x August 15 th x Chemical only	29.5ab	2
106,666plants/ha x August 15 th x Chemical + manual weeding	29.2ab	2
106,666plants/ha x August 15 th x No- weeding	42.1d	3
53,333plants/ha x August 1 st x Chemical only	36.7cd	2
53,333plants/ha x August 1 st x Chemical + manual weeding	32.5bc	2
53,333plants/ha x August 1 st x No-weeding	51.4g	3
53,333plants/ha x August 15 th x Chemical only	46.5f	3
53,333plants/ha x August 15 th x Chemical + manual weeding	41.3de	3
53,333plants/ha x August 15 th x No- weeding	63.2h	4

NS = Not significant

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

Table 1b: Main and Treatment combination effects of weeding, time of planting and plant population density on Cowpea Mottle Virus Disease on cowpea in 2012 season

Weeding Regime	% virus Incidence	Severity index
Chemical weeding only	38.2b	2
Chemical + manual weeding	33.8a	2
Non - weeded	54.4c	3
Time of planting		
1 (August 1 st)	34.6	2
2 (August 15 th)	35.7	2
	NS	
Plant population density		
106,666plants/ha	31.7a	2
53,333plants/ha	42.9b	3
Plant population density X Time of planting X Weeding Regime		
106,666plants/ha x August 1 st x Chemical only	30.1ab	2
106,666plants/ha x August 1 st x Chemical + manual weeding	28.2a	2
106,666plants/ha x August 1 st x No-weeding	41.8d	3
106,666plants/ha x August 15 th x Chemical only	32.5ab	2
106,666plants/ha x August 15 th x Chemical + manual weeding	31.9ab	2
106,666plants/ha x August 15 th x No- weeding	45.7d	3
53,333plants/ha x August 1 st x Chemical only	38.9cd	2
53,333plants/ha x August 1 st x Chemical + manual weeding	36.2bc	2
53,333plants/ha x August 1 st x No-weeding	54.7g	3
53,333plants/ha x August 15 th x Chemical only	50.0f	3
53,333plants/ha x August 15 th x Chemical + manual weeding	44.1de	3
53,333plants/ha x August 15 th x No- weeding	62.6h	4

NS = Not significant

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

Table 2 Main and Treatment combination effects of weeding, time of planting and plant population density on selected growth attributes of cowpea for 2011 and 2012 seasons

Weeding Regime	Plant ht (cm) at flowering 2011	Plant ht (cm) at flowering 2012	Number of leaves per plant at flowering 2011	Number of leaves per plant at flowering 2012
Chemical weeding only	47.9a	44.7a	35.9b	33.7b
Chemical + manual weeding	48.3a	45.5a	42.3a	39.6a
Non - weeded	30.2b	26.8b	27.7c	24.1c
Time of planting				
1 (August 1 st)	52.8a	50.6a	43.9a	39.6a
2 (August 15 th)	39.8b	36.4b	34.4b	31.2b
Plant population density				
106,666plants/ha	47.6	46.8	40.2	35.1
53,333plants/ha	46.7	45.9	39.9	34.8
	NS	NS	NS	NS
Plant population density X Time of planting X Weeding Regime				
106,666plants/ha x August 1 st x Chemical only	53.2ab	48.2ab	44.2b	36.6b
106,666plants/ha x August 1 st x Chemical + manual weeding	55.9a	49.9a	48.3a	39.5a
106,666plants/ha x August 1 st x No- weeding	39.6d	34.4e	32.5e	24.7e
106,666plants/ha x August 15 th x Chemical only	36.2de	32.1ef	29.4f	21.4f
106,666plants/ha x August 15 th x Chemical + manual weeding	45.1bc	41.7c	38.7d	29.9d
106,666plants/ha x August 15 th x No- weeding	22.8f	19.1g	17.7gh	10.1h
53,333plants/ha x August 1 st x Chemical only	50.9ab	46.6b	41.6bc	33.3c
53,333plants/ha x August 1 st x Chemical + manual weeding	51.2ab	47.9ab	41.7bc	32.9cd
53,333plants/ha x August 1 st x No- weeding	35.6e	32.4ef	29.8f	21.6f
53,333plants/ha x August 15 th x Chemical only	34.2e	31.1f	28.6f	20.0g
53,333plants/ha x August 15 th x Chemical + manual weeding	43.9bc	39.9cd	40.7bcd	33.2c
53,333plants/ha x August 15 th x No- weeding	22.4f	17.6h	16.5h	9.1h

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

Table 3: Main and Treatment combination effects of weeding, time of planting and plant population density on selected yield attributes of cowpea for 2011 and 2012 season.

Weeding Regime	Grain wt/plot (Kg) 2011		Grain wt/plot (Kg) 2012	Grain wt/plant (g) 2011		Grain wt/plant (g) 2012
Chemical weeding only	0.9b	0.7b	12.0b	11.6a	555.9b	510.9b
Chemical + manual weeding	1.4a	1.3a	13.2a	12.3a	576.4a	547.8a
Non - weeded	0.2c	0.1c	9.6c	8.4b	254.3c	220.7c
Time of planting						
1 (August 1 st)	1.8a	1.9a	13.0a	12.9a	629.9a	597.4a
2 (August 15 th)	0.5b	0.7b	10.7b	11.2ab	502.5b	498.7b
Plant population density						
106,666plants/ha	1.2	1.3a	14.3a	13.1a	703.5a	701.3a
53,333plants/ha	1.1	0.9b	11.2b	12.9ab	428.8b	450.1b
NS						
Plant population density X Time of planting X Weeding Regime						
106,666plants/ha x August 1 st x Chemical only	2.2a	1.9a	11.6b	10.8ab	767.2b	699.9b
106,666plants/ha x August 1 st x Chemical + manual weeding	2.3a	2.2a	12.9a	11.2a	849.9a	820.7a
106,666plants/ha x August 1 st x No-weeding	0.1d	0.7d	9.9cd	8.9d	245.5ij	248.7h
106,666plants/ha x August 15 th x Chemical only	1.3b	1.5b	10.8c	10.1ab	505.7d	487.4de
106,666plants/ha x August 15 th x Chemical + manual weeding	1.4b	1.7b	11.9b	9.9bc	691.3c	525.6c
106,666plants/ha x August 15 th x No- weeding	0.2d	0.3d	7.7e	6.5e	200.8k	220.4ij
53,333plants/ha x August 1 st x Chemical only	0.5c	0.6c	10.2c	8.9d	443.9fg	420.9f
53,333plants/ha x August 1 st x Chemical + manual weeding	0.6c	0.8c	11.5b	10.6ab	458.4ef	444.5ef
53,333plants/ha x August 1 st x No-weeding	0.2d	0.1d	7.4e	6.8e	213.6jk	199.5jk
53,333plants/ha x August 15 th x Chemical only	0.5c	0.6c	9.9cd	8.2d	388.6h	380.8g
53,333plants/ha x August 15 th x Chemical + manual weeding	0.5c	0.7c	11.0b	9.9bc	424.4g	420.2f
53,333plants/ha x August 15 th x No- weeding	0.1d	0.2d	7.0e	6.7e	199.7l	201.1j

NS = Not significant

Means within a column in the same group that are followed by the same letter(s) are not significantly different using the New Duncan multiple Range Test at $P \geq 0.05$.

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