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Effect of Different Soil Fertilities on Cowpea Mosaic Virus Disease Incidence

Chrispo Makheti Mutebi[1]* and Vincent Moranga[2]

1 Kenya Agricultural and Livestock Research Organization, HRI Kibos Center, P.O. Box 1490-40100, Kisumu Kenya
2 Kenya Accreditation Service, P.O. Box 47400-00100, GPO, Nairobi, Kenya

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ABSTRACT

The present study was done to determine the effect of different soil fertilities on cowpea mosaic virus disease incidence. The fertilities were; 50 kg/ha Diammonium phosphate (18.46.0), 50 kg/ha Mavuno (10.26.10) that is a mineral fertilizer that has been extensively utilized in western Kenya, 3 tons/ha farmyard manure (FYM), FYM/Mavuno combination with half rate of the latter at 25 kg/ha, FYM/DAP combination with half rate of the latter at 25 kg/ha and unfertilized control, all applied at planting time. Certified cowpea seed of K80 variety was planted using the different fertilizers in a randomized complete block design (RCBD). Percent cowpea mosaic disease incidence (DI) data was collected and analyzed through one-way ANOVA. Fisher's LSD post-hoc test at 5% level of confidence was used to evaluate the nature of the differences in mean DI. The results of cowpea mosaic DI shown as descriptive statistics collected from the six fertilizer treatments indicated that FYM/DAP combination had the smallest mean for DI ($M = 12.55$), while DAP alone had the highest DI mean ($M = 33.97$). Associations among groups in the ANOVA table showed that the fertilizer effects were significant ($p < .005$). The measure of effect size showed that 92% of the variance in DI was accounted by the type of fertilizer treatment applied. In conclusion, application of DAP combined with FYM at 25 kg/ha and 1.5 tons/ha was found to be helpful in lowering the cowpea mosaic virus infection in the region where this study was carried out, though these could be used with knowledge of soil reaction.

Abbreviations: ANOVA, Analysis of variance; **DAP**, Diammonium phosphate; **DI**, Disease incidence; **FYM**, Farmyard manure; **HRI,** Horticulture Research Institute; **LSD**, Least significant difference; **KALRO**, Kenya Agricultural and Livestock Research Organization; **NRF**, National Research Fund; **RCBD**, Randomized complete block design; **SPSS**, Statistical package for social sciences

Introduction

Cowpea (*Vigna unguiculata* L. Walp), also known as Black eye pea, Southern pea and Crowder pea, is a legume of African origin and a major food crop in many developing countries in the tropical and sub-tropical regions of the world (Ayodele, 2019). This is due to its high-quality dietary protein content, acceptable palatability and low cost of production (Kareem and Taiwo, 2007). Cowpea belongs to the family Fabaceae and subfamily Faboideae (Hughes et al., 2003). It follows an epigeal emergence pattern which makes it prone to seedling injury (Shiringani,

*Corresponding author's email: cmutebi@yahoo.com
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2007). It has trifoliate leaves with a smooth surface that is dull to shiny (Davis et al., 1991).

The cowpea production is constrained by a range of abiotic and biotic factors, including viral diseases, with over 140 viruses identified as naturally infecting the crop (Hughes and Soyinka, 2003). However, only 15 viruses have been reported as seed-borne (Hampton et al. 1997). These viruses are widely distributed in most cowpea producing areas of the world due to cowpea seed trade and movement of germplasm for trials (Salem et al., 2010). Insects also transmit a majority of seed-borne viruses. Accordingly, infected mature plants can serve as the initial foci for secondary virus spread among field-grown plants (Johansen et al., 1994). Efficient and accurate diagnosis and discarding of them is the key to mitigate the consequences of seed transmission of the viruses. Currently, scouting the field to look for virus-like symptoms is a standard diagnostic method adopted by seed banks (Salem et al., 2010).

Although with low yields due to limited inputs, cowpea forms a significant part of the cropping system's dietary protein and an essential component in semi-arid regions of sub-Saharan Africa (Kyei-Boahen et al., 2017). Ajeigbe et al. (2012) reported cowpea as a major protein source for urban and rural dwellers. The protein content of the leaves is between 27-43%, while that of dry grain is 21-33%, according to Abudulai et al. (2016). In addition, cowpea has been reported to be a good source of fodder for livestock (Kamara et al., 2012).

At the farm level, nitrogen fixation is a major nutrient replenishment source for the soil (Ddamulira et al., 2015). However, declining cowpea yields have been due to continuous land cropping without external inputs (FAOSTAT, 2016). Besides, viral attacks in cowpea render the crop unsightly and thus reduce the marketable value of the produce (FRM, 2015. The resultant loss in quality, vigour, colour and consistency can be termed as spoilage (Garcha, 2018). It has been suggested that growing tolerant cultivars using improved crop management practices such as time of planting and plant population, residue management, tillage and inputs, such as crop protection chemicals, mineral fertilizers, and *Rhizobium* inoculants, would go a long way towards addressing the low yields (Kyei-Boahen et al., 2017). Soil fertility has been reported to have a bearing on disease incidence. Mutebi and Ondede (2021) reported that nitrogen fertilization at optimum rates could potentially be used as additional means of suppressing *Cercospora moricola* in mulberry. Parthasarathy (2015), in a study on more than 400 diseases and pests, found that in general, phosphorus fertilization tended to improve plant health, with reductions in disease incidents recorded in 65% of cases. However, it was found that the regime of phosphorus used depended on a range of factors, including the type of crop and pathogen (Parthasarathy, 2015). On the other hand, Kyei-Boahen et al. (2017) reported that the application of phosphorus fertilizer to nitrogen-fixing legumes on phosphorus-deficient soils increased nitrogen fixation.

Reports have indicated that more than one virus infected cultivated cowpea at the same crop (Quinn and Myers, 2002; Hughes et al., 2003). Seven viruses are known to infect cowpea worldwide. According to András et al. (2014), viral infections can be identified on the basis of the symptoms appearing on test plants. These are; *Cowpea aphid borne mosaic virus* (CABMV) genus *Potyvirus*, *Cowpea golden mosaic virus* (CPGMV) genus *Begomovirus*, *Southern bean mosaic virus* (SBMV) genus *Sobemovirus*, *Blackeye mosaic virus* (BlCMV) genus *Potyvirus*, *Cucumber mosaic virus* (CMV) genus *Cucumovirus*, *Cowpea mottle virus* CMeV) genus *Carmovirus* and *Cowpea yellow mosaic virus* genus *Comovirus* (Hughes et al., 2003). With the exception of CPGMV and CMeV, all other viruses are known to be seed-borne (Bashir and Hampton, 1996; Salem et al., 2010). The resultant effect of the viral infection is expressed in poor pod formation, chlorosis, vein clearing, necrosis, leaf deformation and mild to severe mosaic and mottle (Figure 1).

Cowpea plants may be infected with one or with mixed infections of viruses of different strains. A virus facilitates an increase in the concentration of a co-infecting virus and assists it in its replication (Wang et al., 2002; Mukasa et al., 2006). More severe symptoms are expressed in the host plant, and in some cases, the symptoms induced by one virus species may mark the expression of the symptom caused by the other virus (Mathews, 1991; Mukasa et al., 2006). Whitney and Gilmer (1974) attributed the transmission of the mosaic virus in cowpeas to; thrips (two species: *Sericothrips occipitalis* and *Taeniothrips sjostedti*), chrysomelid beetles (two species: *Ootheca mutabilis* and *Paraluperodes quanternus*), beetles (*Nematocerus acerbus*), grasshoppers (*Cantantops spissus* and *Zonocerus variegatus*). Masarapu et al. (2014) reported that aphids and whiteflies transmit cowpea mosaic viruses. Cowpea mosaic virus-like other viral pathogens in plants cause severe losses in yield and quality of cultivated plants worldwide. These losses and the resulting financial damage can be limited by controlling epidemics using measures that minimize virus infection sources or suppress virus spread (Jones, 2003).

The control of plant diseases using classical pesticides raises serious concerns about food safety, environmental quality, and pesticide resistance (Dordas, 2008). According to Schumann et al. (2018), mineral nutrients are essential for the growth and development of plants and microorganisms and are crucial factors in plant-disease interactions, how each nutrient affects a plant's response to disease, whether positively or negatively, is unique to each plant-disease complex. Fertilizer application has been reported to affect the development of plant disease under field conditions directly through the nutritional status of the plant and indirectly by affecting the conditions which can influence the development of the disease, such as dense foliage, changes in light interception and humidity within the crop stand (Dordas, 2008). Nutritional factors that favour the growth of host plants also favour virus multiplication (Schumann et al., 2018). This holds particularly for nitrogen and phosphorus. However, despite the rapid multiplication of the virus, visible symptoms of the infection do not necessarily correspond to an increase in nutrient supply to the host plant. In fact, the symptoms of viral infections sometimes disappear when N supplies are large, even though the entire plant is infected. Schumann et al. (2018) have further reported that nutritional deficiencies discolour leaf surfaces and increase susceptibility to pests. For instance, the Asian citrus psyllid, *Diaphorina citri*, tends to settle on yellow reflecting surfaces (i.e., surfaces that appear yellow in colour to the human eye).

Smallholder farmers' enhancement of soil nutrients mainly depends on applying farmyard manure (FYM) since it is cheap and readily available in their fields. However, insufficient supply is made by combining the FYM with phosphatic fertilizers such as DAP and Mavuno (Sanginga and Woomer, 2009). Mavuno (10.26.10) is the common name of a mineral fertilizer extensively advanced in western Kenya. Its use of the local minerals in its production makes it less expensive than other mineral fertilizers (Sibusisiwe et al., 2013). Fertilizer manufacturing and blending are shifting to ensure that fertilizers have the major macronutrients and the secondary macronutrients (Sanginga and Woomer, 2009). Ademba et al. (2015), in a study on phosphate fertilizers and manure on maize field, reported that the phosphate fertilizers and manure combination increased growth vigour, grain, dry matter, and harvest index. Lacroix et al. (2017) reported that environmental supplies particularly of nitrogen and phosphorus may alter virus epidemiology indirectly by changing host phenotype or the dynamics of co-infecing pathogens.

Farmyard manure has been reported to be efficient and effective in improving soil fertility, especially when integrated with organic fertilizers (Belay et al., 1997). Studies have shown that the use of various rates of FYM and low rates of inorganic fertilizers are better than the application of either the inorganic fertilizers or FYM alone (Ademba et al., 2015). Nitrogen is known to be a chlorophyll component that promotes vegetative growth and green colouration of foliage (Jones, 1983).

The approach tried in the control of the mosaic virus in cowpeas is the one that is generally applied to all viral diseases in plants. It is based on practices such as sources of infection, avoidance or control of vectors and modification of cultural practices (Varma, 1993). For effective management, an integrated approach is essential in sustainable agriculture (Varma, 1993). When various control measures that act in different ways are used together, their effects complementarily result in far more effective overall control (Jones, 2003). However, cultural methods have not yet been established, particularly those manipulating plant nutrition that can form part of the integrated approach. The objective of this study was to investigate how mavuno and DAP fertilizer combination with farmyard manure affects mosaic virus disease incidences in cowpea.

Materials and Methods

Location of the study

The experiment was laid out on-station at KALRO, HRI Kibos research center located at 0° 3' 0" S and 34° 51' 0" E with an altitude of approximately 1173 meters above sea level. The annual rainfall average at the location is 1290 mm.

Seedbed preparation and management of the experimental crop

The certified K80 cowpea seeds were sown directly into a seedbed prepared to a fine tilth. Planting was done at the beginning of the long rain season at a spacing of 20 cm between plants and 45 cm between rows in plots of 4.5 m including a path of 50 cm in between the 3 replicates. The plots were randomly assigned a fertilizer treatments. Each hole was planted with 2 to 3 seeds. The plots were weeded, and thinned to one plant per hole at 14 days after emergence.

Source of farm inputs

The mineral fertilizer i.e. DAP and Mavuno (NPK 10.26.10) plus certified cowpea seeds were purchased from local marchants. The organic manure was composited from chicken droppings, and crop residues (maize, rice and sorghum). The compost was prepared under the researcher's supervision to ensure standardization (their main ingredients – N – P – K should be tested for repeatability of experiment).

Treatments and experimental design

A randomized complete block design (RCBD) with three replicate per treatment was used in this experiment. Six soil fertility treatments were used are shown in Table 1.

For the full inorganic fertiliser rate, 5.0 g of DAP (50 kg/ha) or mavuno was applied per hole, while 2.5 g of DAP or mavuno were applied per hole at half rate when they were combined with FYM.

Table 1. Soil fertility treatment for the study on cowpea mosaic virus disease incidence

Treatment	Soil fertility option	
	Inorganic fertilizer	FYM
1	None (Control)	None (Control)
2	None	3 tons/ha FYM
3	50 kg/ha Mavuno (10.26.10)	0
4	25 kg/ha Mavuno (10.26.10)	1.5 tons/ha FYM
5	50 kg/ha DAP	0
6	25 kg/ha DAP 18.46.0)	1.5 tons/ha FYM

Data collection

Cowpea mosaic virus disease incidence (DI) was scored at the 10th week after planting. It was done within the plot after the guard plants were marked out by excluding a row of plants at each end of the plot and two plants at each of the other ends of the plot. The total number of within the plot was counted for assessment of cowpea mosaic virus symptoms. The number of plants showing cowpea mosaic virus infection (Fig. 1, 2) was also counted. Disease incidence was determined in all the plots using the formular described by Manandhar et al. (2016) as the following:

$$Disease incidence = \frac{Number of\ diseased plants}{Total\ number of\ plants observed} X100$$

Host response for each treatment was recorded as shown in Table 2 based on the scale developed by Manaandhar et al. (2016).

Fig. 1. Cowpea leaves with Mosaic symptoms (Source: Author's gallery)

Fig. 2. Close-up photo of cowpea mosaic vein clearing symptom (Source: Author's gallery)

Table 2. Host reaction for the different soil fertility treatments according to the scale developed by Manandhar et al. (2016)

Disease incidence	Host response
0-10	Resistant
11-30	Moderately resistant
31-60	Moderately susceptible
61-100	Susceptible

Data analysis

To test the hypothesis whether fertilizer treatments had an effect on cowpea mosaic virus DI, an independent between groups ANOVA was performed using SPSS version 2019. This was followed up with Fisher's LSD post-hoc tests using Least Significant Difference (LSD) at 5% probability level.

Results

The results of the descriptive statistics associated with DI across the six treatments are ploted in Figure 3. It was observed that farmyard manure/DAP fertilizer treatment was associated with the numerically smallest mean DI (*M = 12.55*) while DAP fertilizer treatment was associated with the numerically highest mean DI (*M = 33.97*). Combining Mavuno fertilizer with FYM elicited the second highest mean DI (*M = 32.33*).

The results of the host response for the different soil fertility options are shown in Table 3.

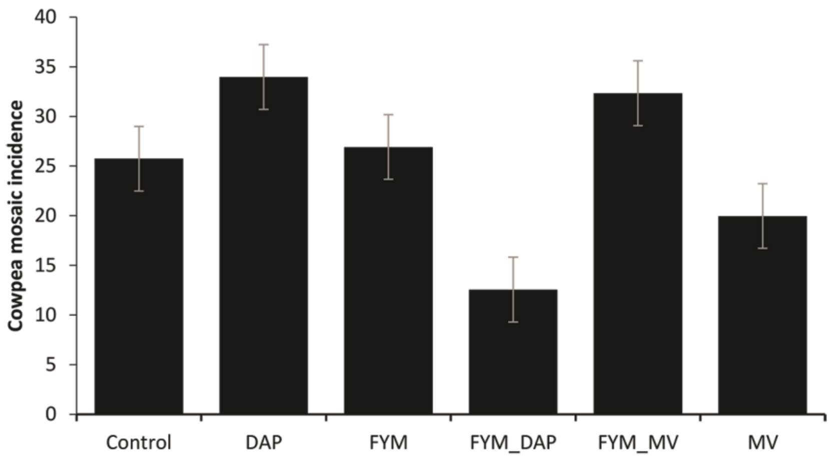

Fig. 3. Cowpea mosaic virus incidence

It was observed that the control, FYM, Mavuno and DAP/FYM treatments were moderately resistant while Mavuno/FYM combination and DAP treatmnts were moderately susceptible according to the scale developed by Manandhar et al. (2016) as indicated in Table 2. The results of the ANOVA yielded a statistically significant effect, $F(5, 12) = 27.23$, $p < .001$, $\eta^2 = 0.92$. Following the statistically significant result, the null hypothesis of no difference among the six treatment means was rejected. Further, it was observed that 92% of the variance in DI was accounted for the type of fertilizer treatment applied as evidenced from the partial eta squared (η^2) result of 0.92.

Table 3. Host reaction to the different soil fertilities according to the scale developed by Manandhar *et al.* (2016)

Treatment	Soil fertility option		Host response
	Inorganic fertilizer	FYM	
1	None (Control)	None (Control)	Moderately resistant
2	None	3 tons/ha FYM	Moderately resistant
3	50kg/ha Mavuno (10.26.10)	0	Moderately susceptible

Treatment	Soil fertility option		Host response
	Inorganic fertilizer	FYM	
4	25kg/ha Mavuno (10.26.10)	1.5 tons/ha FYM	Moderately susceptible
5	50kg/ha DAP	0	Moderately resistant
6	25kg/ha DAP 18.46.0)	1.5 tons/ha FYM	Moderately resistant

Discussion

The control treatment was moderately resistant. The obtained results from current study suggested that though certified seeds of cowpea was used, the natural virus inocula in the environment was able to infect the plants but the plants resisted quite well. Farmyard manure, Mavuno and DAP/FYM combination were also moderately resistant. The findings obtained from the current nvestigation suggest that these soil fertility options did not bring about change in the host plant resistance. However, since the DI values were statistically significant, DAP/FYM fertilizer combination could be taken as inherently better than the rest of the fertility options, suggesting that this combination of fertilizers was best treatments able to significantly lower the cowpea mosaic incidence. In accordance with our findings Dordas (2008) reported that application of fertilizer affects development of plant disease under field conditions.

Farmyard manure/DAP combination contains high amount of N that makes the symptoms of the viral infections to disappear even though the entire plant may still be latently infected and therefore asymptomatic in conformity with the findings of Schumann et al. (2018).

The high cowpea mosaic DI associated with Mavuno/FYM combination was in conformity with the results of studies repoted by Lacroix et al. (2017) who reported that environmental supplies particularly of N and P may alter virus epidemiology indirectly by changing host phenotype or the dynamics of co-infecing pathogens. As compared to DAP/FYM combination, Mavuno/FYM combination results in lower supplies of both nitrogen and phosphorus that could potentially change the dynamics of the co-infecting viruses, which is in accordance with the findings of Lacrix et al. (2017). This is further reinforced by the fact that a huge percentage (92) of the variance in cowpea mosaic virus DI was accounted for the type of fertilizer treatment applied – this huge DI account for treatment did not result in a clear cut separations treatment effects on DI.

Conclusion

The present investigation suggested that application of DAP combined with FYM at 25 kg/ha and 1.5 tons/ha respectively can be helpful in lowering the cowpea mosaic virus infection in the region where this study was carried out. This fertilizer application option could however be used with soil reaction tests.

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Conflict of interest

The authors indicate no conflict of interest in this work.

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