Australian Journal of Crop Science 3(3):167-172 (2009) ISSN: 1835-2707

# Combined effect of host plant resistance and insecticide application on the development of cowpea viral diseases

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#### Abstract

Cowpea [*Vigna unguiculata* (L) Wap] is an important food crop which is widely grown in the Soudano-sahelian region of Cameroon. An integrated disease control approach involving insecticide treatment and plant host resistance was used to control virus-induced diseases, which are the most cowpea yield-limiting factor in this part of the country. A field experiment was conducted at Guirin-Maroua location in which three cowpea varieties (CRSP, LORI and VYA) that differ in their resistance to cowpea viral diseases were treated with Cyperdim 220 EC insecticide at different doses (1.75, 1.25 and 0.95 l/ha). In this experiment, severity of cowpea viral diseases including SMVD, YMVD, ABMVD and GMVD, were assessed. Population size of thrips (*Megalurothrips sjostedti*) and larvae of *Maruca testuladis*, two main vectors of cowpea viral diseases were evaluated. An evaluation of cowpea grain yield was also made. Visual diagnosis which was confirmed by ELISA test showed that only SMVD, YMVD, ABMVD were present during these investigations. Both viral diseases and the population of vectors reduced with combined treatment consisting of the less susceptible cowpea variety VYA and the highest insecticide dose (1.75 l/ha). This treatment combination also produced the highest cowpea grain yield (29.5 t/ha), a yield that was almost 3 times higher than the control (10.2 t/ha). Our results suggest that cowpea viral diseases that prevail in the Soudano-sahelian region of Cameroon are likely to be under control if less susceptible cowpea varieties such as VYA are treated with some insecticides including Cyperdim 220 EC.

Keywords: Cyperdim 220 EC; integrated management; natural defence; Vigna unguiculata; viruses

*Abbreviations:* ABMVD\_aphid-borne mosaic virus disease; DAP\_days after planting; ELISA\_enzyme linked immuno-sorbent assay; GMVD\_golden mosaic virus disease; JIRCAS\_ Japan international research centre for agricultural sciences; IITA\_international institute of tropical agriculture; IRAD\_institute of agricultural research for development; SMVD\_severe mosaic virus disease

#### Introduction

Cowpea, *Vigna unguiculata* (L) Walp, is a dominant staple crop in some Sahelian countries. World production of dry grains amounted to 5 millions tons in 2002 on a total surface area of 14 million hectares. Of that production, Central and West Africa contributed 64 %. The main producers of cowpea are Senegal, Nigeria, Niger, Burkina-Faso and Cameroon, though some regions of South

Africa, South Asia, Central and South America, have high production capacities (Grubben and Denton, 2004). Cowpea is mainly grown to produce dry grains, but about 25 % is consumed on-farm or marketed as green pods. The grains, fresh peas and hay have high protein content (25 %) and serve as a valuable source of proteins for human and animal nutrition (Tarawali et al., 1997). On the basis of dry

| Table 1.  | Virus  | content   | in  | cowpea  | cultivars |
|-----------|--------|-----------|-----|---------|-----------|
| according | to ELI | SA test ( | λ=4 | 05 nm). |           |

|         | C    | Cowpea variet | ies |
|---------|------|---------------|-----|
| Viruses | CRPS | LORI          | VYA |
| SMV     | +++  | ++++          | ++  |
| YMV     | +++  | ++++          | +   |
| ABMV    | +++  | ++++          | +   |
| GMV     | -    | -             | -   |

++++: Very high content; +++: High content; ++: mean content; +: less content; -: no virus

weight, a combination of 75 % cereal and 25 % cowpea diet provides sufficient proteins for adults (Mbaye, 2007). Cowpea roots have Rhizobium that is able to fix nitrogen even in very poor soils (organic matter less than 0.2 %, sand content > 85 %). Cowpea is shade-tolerant and therefore compatible as an intercrop (Singh and Sharma, 1996). All these qualities have contributed to the widespread distribution of cowpea in the semi-arid regions of Central and West Africa in general, and in the Soudano-sahelian zone of Cameroon in particular, where other food legumes do not perform well. Conditions of drought resulting in water deficiency and stress are common in these areas and affects cowpea production. Although parasitic agents of different nature adversely affect cowpea plants in these regions, viral diseases rank first and result in grain yield losses ranging from 20 to 60 % (Ouzounov, 1988; Taïwo, 2003). Four main types of viral diseases have been reported on cowpea crop; SMVD, YMVD, ABMVD and GMVD (Singh and Allen, 1997; Büchen-Osmond, 2006; Taïwo et al., 2007). Symptoms of these diseases which are observable on cowpea leaves include: mosaics, blotchiness, deformation, blisters and chlorotic variegations. In some cases, cowpea plants are stunted (William, 1975). Field dissemination of cowpea viral diseases is mediated by stinking, sucking, and crushing insect vectors (Abdullahi et al., 1998; Adipala et al., 2000).

So far, management of cowpea viral diseases has been undertaken by means of resistant varieties and chemicals (Perring et al., 1999). The use of the host plants resistance is the most effective way to control plant diseases caused by viruses (Nelson, 1988). With some cropped species such as cowpea, existing cultivars can only offer partial disease resistance (Mahabeer et al., 1995). Despite their adverse effects on the environment, vector-killing chemicals are widely used to fight against cowpea viral diseases. However, both methods are often less effective when used separately (Fontem et *al.*, 1996; Murphy and Bowen, 2006). In this work, cowpea varieties with variable disease resistance were combined with insecticide applications in attempts to effectively control some viral diseases. The objectives were to confirm disease resistance levels of the three cowpea cultivars currently cultivated in the Soudano-sahelian zone of Cameroon and to examine viral disease suppression associated with combination of cowpea resistance and use of insecticide.

#### Materials and methods

#### Pedo-climatic conditions

The study was carried out during the 2004 growing season at the Maroua (Cameroon) research centre. The experiment was conducted on a clay-sandy soil, with pH 5.6. This region has a Soudano-sahelian climate with a monomodal rainfall pattern, 7 to 8 months dry season (from December to June) and 4 to 5 months rainy season (from July to November). Average monthly rainfall within the growing season was 225 mm and average daily temperatures ranged from 26 to 32 °C.

### Cowpea varieties, land preparation, crop management and experimental set up

Three cowpea varieties; two from IRAD Maroua (CRSP and LORI) and one local variety (VYA) were chosen based on their differences in sensitivity to viral diseases.

A factorial experiment was conducted using a split plot design. Cyperdim 220 EC insecticide (Dimethoate 200 g/l and Cypermethrine 20 g/l) (factor 1) consisting of four doses  $(T_0 = 0, T_1 =$ 1.75,  $T_2 = 1.25$  and  $T_3 = 0.95$  l/ha) and cowpea (factor 2) consisting of three varieties (CRSP, LORI and VYA) were experimented. Insecticide treatments were assigned to main plots while cowpea varieties were assigned to subplots within each main plot. The subplot size was 16.2 m<sup>2</sup> and had 42 cowpea stands planted at 0.8 m x 0.5 m apart. The various treatments were repeated in three blocks of 226 m<sup>2</sup> (10 m x 22.6 m) each. Seeds were planted manually using a hoe. Handweedings were done twice before harvest. Insecticide treatments were applied using a hand sprayer and began at 35 days after planting as soon as the first symptoms appeared. Treatments were made over a 40 days period at 10 days intervals.

## Detection of viruses and estimation of viral infections

Detection of SMV, YMV, ABMV and GMV was done using the ELISA procedure described by Bar-Joseph and Garnsey (1981) and the results were read using a suitable photometer at 405 nm. Viral infections were evaluated at 10 days intervals as from the  $34^{th}$  DAP using visual observation. Symptoms were scored on a set of 20 cowpea plants selected randomly from middle rows within each subplot. Viral infection was estimated using a 1 to 5 standard evaluation scale (EES) in which 1 = no symptoms of infection on all the leaves and 5 = all leaves show symptoms of infection. Severity of infection was calculated according to the

formula:  $S = \frac{\sum (ab)}{N}$  (Tchoumakov and

Zaharova, 1990). Where: S (expressed in EES) = Severity of infection on the plot;  $\Sigma$  (ab) = Sum of the multiplications of number of diseased plants (a) with the corresponding degree of infection (b) expressed in EES; N = Total number of diseased plants studied on the plot.

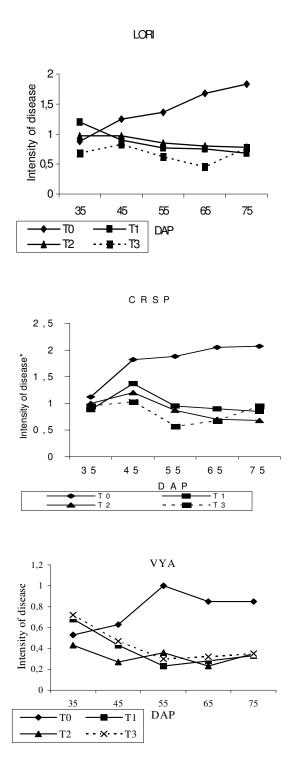
#### Evaluation of insect population, plant damages and yield of dry grains

The study of insects was based on two groups: thrips (*Megalurothrips sjostedti*) and larvae of *Maruca testulalis*, because these are the most dominant groups of vectors involved in disease dissemination in the zone under study (Singh and Allen, 1997). Evaluation was done at the 50 % flowering phase, when half of the plants bore an open flower at 65 DAP. Fifty flowers were randomly selected from 20 plants in the within middle rows then, the number of damaged flowers was determined and the percentage of infested flowers established.

All cowpea pods from 20 plants within the middle rows in each subplot were harvested at 80 DAP. They were next dried under sunlight for 3 days and then manually cracked to separate the grains from the shells. The weight of these two yield components were thereafter determined using a balance.

#### Statistical analysis of data

Analysis of variance of effects of various treatments was performed using the software analysis system. Average effects of treatments assigned to subplots were compared within each main plot. Treatment mean difference was determined according to Duncan multiple rang test at P = 0.05. The aptitude of the method used to describe the data of infections was appreciated by Fisher's test with  $F_{cal}$  and  $F_{tab}$  values. Once  $F_{cal} > F_{tab}$ , the result was significant.



*Fig 1.* Evolution of viral infection on the three cowpea varieties under the influence of different doses of Cyperdim 220 EC.  $T_0 = \text{control}$ ,  $T_1 = 1.75$  l/ha,  $T_2 = 1.25$  l/ha,  $T_3 = 0.95$  l/ha.

| Factors               | RG (t/ha)                    | RGr (t/ha)                   | RC (t/ha)                  | RS (t/ha)                    |
|-----------------------|------------------------------|------------------------------|----------------------------|------------------------------|
| Insecticide doses     |                              |                              |                            |                              |
| То                    | $16.7 \pm 13.8$ <sup>b</sup> | $10.2 \pm 7.9$ <sup>b</sup>  | $4.5 \pm 3.4$ <sup>b</sup> | $5.6 \pm 6.3$ <sup>c</sup>   |
| T1                    | $39.8 \pm 5.5^{a}$           | $31.0 \pm 4.2^{a}$           | $8.1 \pm 1.8$ <sup>a</sup> | $23.1 \pm 3.5^{a}$           |
| T2                    | $36.8 \pm 15.3^{a}$          | $28.2 \pm 11.8^{a}$          | $7.9 \pm 3.4^{a}$          | $19.8 \pm 8.4$ <sup>b</sup>  |
| Т3                    | $37.1 \pm 21.1^{a}$          | $28.5 \pm 16.7$ <sup>a</sup> | $7.8 \pm 4.5^{a}$          | $20.5 \pm 13.7$ <sup>a</sup> |
| LSD (5 %)             | 5.4                          | 4.8                          | 0.9                        | 3.9                          |
| F <sub>cal</sub>      | 9.4                          | 10.6                         | 7.5                        | 11.8                         |
| F <sub>tab</sub> (5%) | 3.0                          | 3.0                          | 3.0                        | 3.0                          |
| Varieties             |                              |                              |                            |                              |
| CRSP                  | 33.2 ± 14.4 b                | 25.7 ± 12.2 b                | $6.8 \pm 2.1 \text{ b}$    | 17.0 ± 9.6 b                 |
| LORI                  | 18.8± 11.7 c                 | $14.0 \pm 9.4 \text{ c}$     | $4.0 \pm 2.2$ b            | 9.7 ± 7.9 c                  |
| VYA                   | 45.8 ± 10.3 a                | 33.7 ± 14.5 a                | 10.4 ±1,8 a                | 25.2 ± 8.8 a                 |
| LSD (5 %)             | 12.6                         | 9.2                          | 2.9                        | 7.2                          |
| Fcal                  | 20.3                         | 15.1                         | 34.5                       | 15.2                         |
| Ftab (5 %)            | 3.4                          | 3.4                          | 3.4                        | 3.4                          |

| Table 2. Effect of insecticide applications and cowpea varieties on yields RG: Yield in dry pods; |
|---|
| RGr: Yield in dry grains; RC: Yield in shell; RS: Yield in grains after sorting for seeds         |

Means on the same column followed by the same letter are not significantly different (P = 0.05) according to Duncan Multiple Range Test.

#### Results

### *Effect of insecticide applications and cowpea host resistance on viral infections*

In this study three cowpea viral diseases (SMVD, YMVD and ABMVD) were identified based on visual observation and ELISA test (Table 1). Viral disease severity was generally higher in control plots than in treated plots irrespective of cowpea variety (Fig. 1). Disease spread was faster in control than in treated plots, and infection severity was more pronounced on CRSP and LORI than on VYA. No significant disease reduction was observed for all cowpea varieties 10 days after the first insecticide application. However, the next insecticide application made at 75 DAP significantly reduced disease severity.

### Influence of insecticide treatment and cowpea variety on population size of vectors

Rate of flower infestation by thrips varied according to insecticide dose and cowpea variety (Fig. 2). All three treatments  $T_1$ ,  $T_2$  and  $T_3$  significantly reduced flower infestation as compared to the control  $T_0$  (95.7 %). However,  $T_1$  with a flower infestation of 69.0 % seemed to be more effective when compared with  $T_2$  and  $T_3$  which had 84.0 and 77.7 % infestation respectively. Cowpea varieties did not vary in their infestation level by thrips. In contrast, susceptibility to *M. testulalis* varied among cowpea cultivars (Fig. 2). Cowpea flower infestation levels by *M. testulalis* from insecticide treated plants ranged between 5.0 to 19.0 %, 6.7 to 23.0 % and 5.0 to 24.7 % for  $T_1$ ,

 $T_2$  and  $T_{3,}$  respectively. These variations were shown to be significantly different at P = 0.05.

### Effect of insecticide treatment on cowpea grains yield

Cowpea grains yield significantly increased as a result of insecticide application. However, increasing insecticide dose did not significantly increase yields of cowpea grains (Table 2). Grain yield also varied according to cowpea variety. VYA variety gave the highest yield (45.8 t/ha) followed by CRSP variety (33.0 t/ha) while variety LORI gave the lowest yield in grains (18.8 t/ha) (Table 2).

#### Discussion

Virus diseases seriously reduce cowpea production in the Soudano-sahelian zone of Cameroon. In this study, attempts were made to control these cowpea diseases using a combination of insecticide treatment and host plant resistance. Three cowpea viral diseases (SMVD, YMVD and ABMVD) that were previously described in this zone of Cameroon (Ntoumkam and Kitch, 1994) and in some Sahelian regions in Nigeria (Taiwo and Akinjogunla, 2006) were studied. Three currently grown cowpea varieties (CRSP, LORI and VYA) that have different levels of resistance to viral diseases were treated with Cyperdim 220 EC to see whether such an integrated disease control approach could satisfactorily lower cowpea yield losses induced by viral diseases. Cowpea varieties treated with insecticides showed reduced disease incidence. Disease severity was shown to decrease

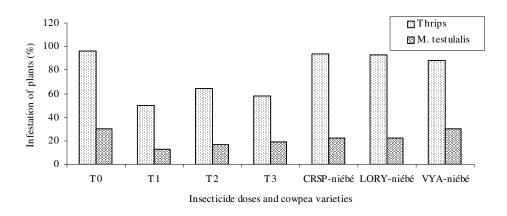


Fig 2. Effect of different insecticide doses and varieties on the pest population 60 DAP

progressively with increase in insecticide application frequency, and this agrees with a report by IITA (1994) and Chatzivassiliou (2008) in similar works. The results show that 4 to 7 sprays of insecticide would be enough to achieve an effective control of cowpea viral diseases and this corroborates the data obtained by Pedersen et al. (2007) in management of soybean virus diseases. Insecticide application, despite some of its possible environmental inconveniences, is widely used against devastating insects that transmit viral infections.

Insecticide-treated cowpea plants were generally less infested by thrips than untreated plants (controls). The level of flower infestation by thrips was shown to decrease with increase in insecticide application. It is known that cowpea flowers do not remain open for long. This physiological feature makes insecticide flow inside flowers very poor (Singh and Allen, 1997; Ofuya, 1997). High insecticide application rates likely reduces the effect of this floral physical barrier, and might explain, at least partly, why the level of cowpea flower infestation by thrips was inversely linked to insecticide application rates (Fereres, 2000). Considering the effect of cowpea varieties on the population of thrips, it was noticed that cowpea varieties from IRAD collection (CRSP-cowpea and LORI-cowpea) were more sensitive to thrips and showed about 93.50 % level of infestation. This confirms an early report by IITA (1994). The local variety, VYA-cowpea on the contrary was less sensitive to thrips. Application of Cyperdim 220 EC slightly reduced the population of *M. testulalis* larvae. However, the results showed that application of Cyperdim 220 EC at 10 days

intervals better reduces insect population than the use of variety resistance alone (Irwin et al., 2000; Tanzubil et al., 2008). Plants that received Cyperdim 220 EC showed a great difference in grain yield. This could be explained by the protection provided by insecticide doses against insects that transmit viruses, and this corroborates data obtained by Pasquet and Baudoin (1997) in improvement of cowpea production. Test cowpea varieties differed in their yield potential. The observed differences between the test varieties were probably due to their variable ability to resist pests and diseases (Mbouemboué, 1994). Both methods of plant protection; chemical and host resistance had very significant effects on viral diseases reduction and increase in cowpea yield. Therefore, a combination of these two methods could result in better control of cowpea viral diseases and production of relatively virus-free seeds for planting by poor-resource farmers.

#### Acknowledgements

Authors are thankful to Pr. Pomazkov Y.I., Laboratory of Plant Virology, Department of Crop Protection of the Russian People's Friendship University for help to achieve the ELISA test.

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