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Bioactivity of Aqueous Extract Mixtures of Nigerian Plants for Controlling the Pod Borer, *Maruca vitrata* Fab. and Pod Sucking Bug, *Clavigralla tomentosicollis* Stal. on Cowpea, *Vigna unguiculata* (L.) Walp.*

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Abstract: Mixtures of aqueous extracts of various plants were tested for their efficacy in the control of *Maruca vitrata* Fab. and *Clavigralla tomentosicollis* Stal under field conditions using cowpea variety SAMPEA 7. The materials involved included leaf extracts of *Tagetes erecta* L., in mixtures with *Eucalyptus citriodora* Denn, *Azadirachta indica* A. Juss and *Gmelina arborea* L; stem bark extract of *G. arborea* in mixtures with *E. citriodora*, *A. indica* stem bark extracts; leaf extract of *Hyptis suaveolens* Poit. mixed with *Vernonia amygdelina* L. and *Cymbopogon citratus* Staph leaves. These plant materials were mixed in 10:10% w/w. The stem bark mixtures of *G. arborea* + *A. indica* extracts and the leaf extract mixtures of *T. erecta* + *A. indica* caused significantly higher reduction of *M. vitrata* larvae and *C. tomentosicollis* (adults/nymphs) compared with the other extracts mixture treatments and the untreated control. Cowpea pods were better protected from pests damage on plots treated with these two extract mixtures leading to increased grain yields. This pest management strategy is suitable for low-income agriculture practiced in the developing countries.

Key words: Bioactivity, plant extract mixtures, *M. vitrata*, *C. tomentosicollis*, control, cowpea

Introduction

Cowpea, *Vigna unguiculata* (L.) Walp is a popular and nutritionally important grain legume in tropical countries where it supplies the bulk of the protein intakes in the menu of most people. Cowpea yields are generally low in Africa due to heavy infestation by insect pests and diseases. The most damaging of all pests are those occurring during the flowering and podding periods of which the legume pod borer, *Maruca vitrata* Fab. (Lepidoptera: Pyralidae) and a complex of pod and seed suckers predominated mainly by *Clavigralla tomentosicollis* Stal. (Hemiptera: Coreidae) and the flower bud thrips, *Megalurothrips sjostedti* Trybom (Thysanoptera: Thripidae) are the most important ones (Jackai and Daoust, 1986; Singh *et al*., 1990). Synthetic insecticides are the most effective, economical and acceptable control measures on cowpea in spite of their serious environmental consequences (Booker, 1965; Jackai *et al*., 1985). In addition to its environmental concerns, the current economic crisis in Africa accentuated by high exchange rate of the local currencies and low purchasing power of the farmers have influenced research into alternative pests control strategies of which botanicals are a component part.

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Many plants in the African landscape are being investigated for insecticidal attributes for control of pests of food crops (Schmutterer, 1990; Saxena, 1989) and some successes have been reported. Although, most of these trials were based on storage protection of cowpea (Ofuya, 1986; Ogunwolu and Idowu, 1994; Oparaeke, 1997; Dike and Mshelia, 1997) and maize (Kossou, 1989), more interests have been generated in the application of Plant-Based Insecticides (PBIs) against field pests. For instance, neem based preparations have been extensively studied to control field pests of crops such as cassava (Olaifa and Adenuga, 1988), cowpea (Jackai *et al*., 1992; Tanzubil, 1991; Emosairue and Ubana, 1998) and okra (Emosairue and Uguru, 1998). Since PBIs are slow acting mortality agents and the application of extracts from a single plant source may not likely produce satisfactory results, the "best mix" approach using insecticidal plants of various species is advocated. This involves the most logical combination of different plant species that might be compatible in mixture formulations for management of these pests. A limited database is available on the insecticidal activities of mixtures of naturally growing plants in Africa for control of crop pests. This study intends to fill this vacuum as it reports the results of part of an ongoing research work on cowpea protection in the Institute for Agricultural Research, using botanical mixtures.

Materials and Methods

The investigation was conducted at the Research Farm of the Institute for Agricultural Research, Ahmadu Bello University, Zaria located in Northern Guinea savanna of Nigeria, for the periods 2000 and 2001 under rain fed conditions. The plant materials used for the study included leaves of *Tagetes erecta*, L. (Compositae), *Vernonia amygdelina* L. (Compositae), *Hyptis suaveolens* Poit (Lamiaceae), *Cymbopogon citratus* Staph (Graminae), leaves and stem barks of *Gmelina arborea* L (Verbanaceae), *Eucalyptus citriodora* Denn (Myrtaceae) and *Azadirachta indica* A. Juss (Meliaceae). These materials were collected around the institute's Head Office and were dried under the shade for 72 h. Each plant material weighing 500 g was pounded in a mortar with pestle and the appropriate combinations of plant powders (Table 1) were poured into plastic buckets containing 3.5 l hot water (70%) and soaked overnight. The extract mixtures were then filtered the next day with 1.0 l tap water using a double folded muslin cloth. Each of the extract mixtures received 250 mL of 20% w/v of soap and starch solutions to improve their rain fastness and spread on plant surfaces. The solutions were labeled and taken to the field for spraying.

The experimental site had an area of about 0.45 ha. The field was prepared by spraying glyphosate at the rate of 5.0 l ha^{-1} and allowed to stand for 21 days before harrowing and ridging at 0.75 m apart. Nine plots measuring 6.0×5.0 m were marked out in three replicates using randomized block design. Each plot consisting of five ridges (three inner ridges and two discards) was separated by a 1.5 m wide border along the ridge and two unplanted ridges. Cowpea variety SAMPEA 7, an improved, semi determinate, medium duration maturing crop (75-85 days) with a semi erect growth habit was used as the test material. SAMPEA 7 is reported to be highly susceptible to all major insect pests of cowpea in this ecological zone. The growth period synchronizes with the peak populations of these noxious pests (Amatobi, 1994). Three seeds of this cowpea variety were sown at an intra-row spacing of 0.25 m in the first week of August 2000 and 2001. The plots were immediately sprayed with Galex (Metalachlor 250 g a.i + Metabromuron 250 g a.i) at 2.5 kg a.i ha^{-1} to control volunteer weeds. The seedlings were thinned to two plants per planting hole 2 to 3 Weeks after Sowing (WAS). Compound fertilizer NPK (15:15:15) was top dressed at 37.5 kg a.i ha^{-1} two WAS. At four WAS, a mixture of benomyl and mancozeb was applied weekly for four weeks at 0.33 kg a.i ha^{-1} to control fungal diseases such as scabs, brown blotch and septoria leaf spots The field was manually weeded once with a hoe at 5 WAS. Spraying of both extracts mixtures and synthetic insecticide commenced at seven WAS (at flower bud initiation or at the onset of flowering) when the peduncles were 2-10 cm long. CP 3 Knapsack sprayers were used during insecticide application and four weekly schedules were conducted. A single cowpea row was sprayed per pass or trip for good coverage. There were some unsprayed plots used as the control check. If rain fell within 2 h of application of extract mixtures a repeat spraying was conducted the next day.

Ten plants were randomly inspected for signs of phytotoxicity two days after each spraying. *M. vitrata* larvae were sampled before each spraying from 7.00-9.00 am by random picking of 20 flowers from plants located within the three inner ridges per plot. The flowers were placed in vials containing 30% alcohol and taken to the laboratory where they were dissected the next day and the number of *Maruca* pod borers (MPBs) found was counted. Adults and nymphs of *C. tomentosicollis* were assessed by visual counting of the insect found on plants located in three 1.0×1.0 m^2 quadrants, which were randomly placed within the inner ridges per plot. Cowpea pod damage was also assessed at 10 WAS when the pods have attained between 75-85% physiological maturity using the formula below:

$$\%\text{ Pod damage} = \frac{\text{Total pods produced/plant} - \text{Number of undamaged pods/plant}}{\text{Total number of pods produced/plant}} \times 100$$

Cowpea pods were harvested dry within the three inner ridges. The pods were threshed, winnowed and the grains weighed. Data collected were subjected to appropriate transformation (square root or arcsine for percentage data) before Analysis of Variance was done and means were separated by SAS-SNK ($p<0.05$) test (SAS, 1989).

Results

Extracts from different plants mixed at 10:10% w/w exerted different levels of efficacy on *Maruca* Pod Borers (MPBs) and Pod Sucking Bugs (PSBs) pre dominated by *C. tomentosicollis*. The stem bark mixtures of *G. arborea* + *A. indica* and the leaf extract mixtures of *T. erecta* + *A. indica* were most effective in reducing the numbers of MPBs (<1.0 larva/flower and/or pod) and PSBs (<1.5 bugs/plant) in the two years of investigation. These extract mixtures significantly ($p<0.05$) lowered the numbers of the tested insect pests compared with the untreated control and some other plant extract mixtures. Although, all the plant extracts mixture treatments were significantly superior to the untreated control (Table 1), the leaf mixtures of *T. erecta* + *G. arborea*, *T. erecta*+*E. citriodora*, *H. suaveolens*+*V. amygdelina* and stem bark mixtures of *G. arborea* + *E. citriodora* were not effective against the two pests. The synthetic insecticide treated plots caused the greatest reduction ($p<0.05$) in the population of MPBs and PSBs and was superior to all the plant extracts mixture treatments.

Similar trends were observed for pod damage and grain yields assessments where plots treated with the stem bark mixtures of *G. arborea* + *A. indica* and the leaf extract mixtures of *T. erecta* + *A. indica* had lower pod damage ($p<0.05$) and higher grain yields ($p<0.05$) compared with plots treated with other plant extract mixtures and the untreated control. The greatest pod damage and lowest grain yield were found in plots sprayed with leaf extract mixtures of *T. erecta* + *G. arborea*, *T. erecta* + *E. citriodora* and *H. suaveolens* + *V. amygdelina*. Although, all the plant extracts mixture treatments were superior to the untreated check they were inferior to the synthetic insecticide

Table 1: Mean number of *M. vitrata* and *C. tomentosicollis* on cowpea treated with aqueous plant extracts mixtures in 2000 and 2001 seasons in Northern Guinea savanna of Nigeria

	M. vitrata/per flower and/or pod		*C. tomentosicollis*/plant	
Treatment	2000	2001	2000	2001
T. erecta + *E. citriodora* leaves	1.5 b	1.6 b	2.9 bc	3.3 bc
T. erecta + *A. indica* leaves	0.8 cd	0.9 cd	1.3 d	1.5 d
T. erecta + *G. arborea* leaves	1.7 b	1.8 b	3.1 b	3.4 b
G. arborea + *E. citriodora* barks	1.4 b	1.5 b	2.8 c	2.9 c
G. arborea + *A. indica* barks	0.8 d	0.8 d	1.2 d	1.3 d
H. suaveolens + *V. amygdelina*	1.4 b	1.6 b	2.8 c	3.0 c
H. suaveolens + *C. citratus* leaves	1.1 c	1.2 c	2.0 c	2.2 c
Uppercott	0.2 e	0.3 e	0.3 e	0.4 e
Control (0.0)	3.9 a	4.2 a	4.7 a	5.8 a
SE±	0.09	0.08	0.09	0.08

Means in a column followed by similar superscript (s) are not significantly different by SAS - SNK, (p<0.0) test

Table 2: Mean number of pods damaged and grain yields of cowpea treated with aqueous plant extracts mixtures in 2000 and 2001 seasons in Northern Guinea savanna of Nigeria

	% Pod Damage/plant		Grain yield (Kg ha^{-1})	
Treatment	2000	2001	2000	2001
T. erecta + *E. citriodora* leaves	32.3 bc	32.0 bc	414.4 c	410.3 c
T. erecta + *A. indica* leaves	19.6 f	19.4 f	679. 5 f	672.7 f
T. erecta + *G. arborea* leaves	33.9 b	33.6 b	333.2 b	329.9 b
G. arborea + *E. citriodora* barks	27.8 d	27.5 d	445.2 de	440.8 de
G. arborea + *A. indica* barks	17.8 f	17.6 f	701.9 f	694.8 f
H. suaveolens + *V. amygdelina*	30.5 cd	30.2 cd	439.6 d	435.2 d
H. suaveolens + *C. citratus* leaves	23.8 e	23.5 e	548.8 e	543.3 e
Uppercott	11.4 g	11.2 g	1120.3 g	1107.3 g
Control (0.0)	90.4 a	89.5 a	197.8 a	195.3 a
SE±	0.94	0.93	6.32	6.3

Means in a column followed by similar superscript (s) are not significantly different by SAS - SNK, (p<0.0) test

treatment (Table 2). Field observation also indicated that none of the plant extracts mixtures exhibited signs of phytotoxicity or discolouration of the cowpea plants throughout the periods.

Discussion

The effects of different plant extract mixtures on *M. vitrata* and *C. tomentosicollis* have been shown in this study. The greatest reduction in MPBs and PSBs numbers was found in plots treated with stem bark mixtures of *G. arborea* + *A. indica* and leaf extracts of *T. erecta* + *A. indica*. The same extract mixtures caused the greatest reduction in pod damage leading to higher grain yields on treated plots. Although, their values were significantly ($p<0.05$) inferior to the synthetic insecticide treatment, they were however significantly superior ($p<0.05$) to the untreated check in pod damage reduction and higher grain yields.

These findings were similar to earlier work conducted by some researchers (Jung, 1938; Allen *et al*., 1944; Yepsen, 1976; Snoek, 1984), which showed that plant extract mixtures possess higher insecticidal efficacy (where compatible) than a single plant material. In this study, the differences between the untreated check, which harboured more pests than the extract mixtures treatments, were significant. This could be ascribed to the persistence of the extract mixtures sprays which minimized latter reinfestation on the treated plots.

Phylogenically, plants belonging to the families Myrtaceae (for example, *Eucalyptus* sp.), Meliaceae (e.g., neem), Compositae (bitter leaf and African marigold), Lamiaceae (e.g., African bush tea), Graminae (lemon grass) and Verbanaceae (*Gmelina* sp.) possess the highest bioactivity against insect pests (Grainge *et al*., 1986). The poor performance of some of the plant species belonging to these families was unexpected. The reason might be due to the pattern of distribution of bioactive compounds among some genera and species in the plant kingdom as well as the geographical location of such plants (Grainge *et al*., 1986). Secondly, some parts of plants contain more concentrations of the active principles than others, for example, the seed and stem bark of neem possess more Azadirachtin, Nimbin and Salannin than the leaf portion (Saxena, 1989; Kossou, 1989; Schmutterer, 1990). This explains why the stem bark mixtures of *G. arborea* + *A. indica* was more effective in controlling MPBs and PSBs than their leaf equivalents in mixtures with leaf materials from other plant species. The issue of active principles compatibility can not be ignored since the stem bark mixtures of *G. arborea* + *E. citriodora* did not follow this trend possibly due to incompatibility factor in bioactive materials present in the two plants species. However, the poor performance of some extract mixtures against the two insect pests may not be the true reflection of the insecticidal potentials of the plants. The screening criteria adopted for the two pests might not permit recording the effect of semiochemical of chronic effects of the plant extract mixtures on developmental and reproductive physiology of these pests (Williams and Mansingh, 1993). For instance, *A. indica* and some other plants possess growth regulatory, antifeedant, repellent and semiochemical activities on different insects pests (Schmutterer *et al*., 1980; Schmutterer and Ascher, 1984; Grainge *et al*., 1986). The present investigation indicates that stem bark mixtures of *G. arborea* + *A. indica* and leaf extract mixtures of *T. erecta* + *A. indica* can significantly reduce MPBs and PSBs on cowpea plants.

In Nigeria where this trial was conducted, all the plants are readily available, cheap, safe and technologically friendly for farmers' use to protect their crops. The possession of highly lethal compounds by *G. arborea* + *A. indica* offers good opportunity for developing the mixture as an alternative to the synthetic insecticides which are environmentally unfriendly and expensive for limited resource farmers in the country. Further reserch is needed to assess the efficacy of different proportional mixtures of these plant materials and their spraying schedules to ensure increased grain yields and better quality grains for farmers.

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