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# POTENTIAL FOR INTEGRATED BOTANICALS AND HOST RESISTANCE IN MAIZE FOR MANAGEMENT OF FALL ARMYWORM IN KENYA

POTENTIEL DE L'INTÉGRATION DES PRODUITS BOTANIQUES ET DE LA RÉSISTANCE DES HÔTES DANS LE MAÏS POUR LA LUTTE CONTRE LA LIGUEUSE D'AUTOMNE AU KENYA

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### **ABSTRACT**

The Fall Armyworm (FAW), Spodoptera frugiperda, has been a devastating invasive pest across Africa since 2016; severely affecting maize (Zea mays L.) by attacking the crop from emergence to cob formation. Farmers increasingly rely on the use of excessive synthetic insecticides, which can lead to environmental harm, human health risks, and the development of pest resistance. There is a need to minimise reliance on synthetic pesticides in agroecosystems and opt for safer, bio alternatives. The objective of this study was to evaluate an integrated botanical system and host plant resistance for managing FAW for maize production in Kenya. A field experiment was conducted to evaluate combined Brachiaria cultivars (Xaraes and Mulato II) and Tithonia diversifolia botanical extracts; and host maize accessions resistance on FAW proliferation in maize production in Kenya. The findings revealed no significant difference (P>0.05) between T. diversifolia botanical and synthetic insecticide in controlling FAW populations in maize. Therefore, T. diversifolia was a promising biopesticide; while Bracharia cultivars (Xaraes and Mulato II) were effective as alternate trap plants in the push-pull systems. The integration of T. diversifolia, Brachiaria trap crops (Xaraes), and maize host resistance (MA4) proved to be the most effective strategy of FAW. Thus, this eco-friendly IPM approach has the potential to reduce insecticide dependence, preserve biodiversity and offers an alternative sustainable FAW management in maize production.

Key Words: IPM, Spodoptera frugiperda, Tithonia diversifolia, Zea mays

### RÉSUMÉ

La chenille légionnaire d'automne (CLA), Spodoptera frugiperda, est un ravageur invasif dévastateur en Afrique depuis 2016. Elle affecte gravement le mais (Zea mays L.) en attaquant la culture de la levée à la formation des épis. Les agriculteurs ont de plus en plus recours à un usage excessif d'insecticides de synthèse, ce qui peut entraîner des dommages environnementaux, des risques pour la santé humaine et le développement de résistances aux ravageurs. Il est nécessaire de minimiser le recours aux pesticides de synthèse dans les agroécosystèmes et d'opter pour des alternatives biologiques plus sûres. L'objectif de cette étude était d'évaluer un système botanique intégré et la résistance des plantes hôtes pour la gestion de la CLA dans la production de maïs au Kenya. Une expérience au champ a été menée pour évaluer la combinaison de cultivars de Brachiaria (Xaraes et Mulato II) et d'extraits botaniques de Tithonia diversifolia, ainsi que la résistance des accessions de maïs hôtes sur la prolifération de la CLA dans la production de maïs au Kenya. Les résultats n'ont révélé aucune différence significative entre l'insecticide botanique et synthétique de T. diversifolia dans la lutte contre les populations de CLA dans le maïs. Par conséquent, T. diversifolia s'est avéré un biopesticide prometteur, tandis que les cultivars de Bracharia (Xaraes et Mulato II) se sont révélés efficaces comme plantes pièges alternatives dans les systèmes push-pull. L'intégration de *T. diversifolia*, de cultures pièges de Brachiaria (Xaraes) et de la résistance de l'hôte maïs (MA4) s'est avérée la stratégie la plus efficace contre la CLA. Ainsi, cette approche de lutte intégrée respectueuse de l'environnement a le potentiel de réduire la dépendance aux insecticides, de préserver la biodiversité et offre une alternative durable de gestion de la CLA dans la production de maïs.

Mots Clés: Lutte intégrée, Spodoptera frugiperda, Tithonia diversifolia, Zea mays

### INTRODUCTION

The Fall armyworm (FAW), Spodoptera frugiperda (J.E. Smith) (Insecta: Lepidoptera: Noctuidae), is an invasive insect pest that affects more than 80 plant species, causing significant yield damage (Bakry and Abdel-Baky, 2023). It has a higher preference for maize (Zea mays L.), but also affects other major cultivated crops, including sorghum, rice, sugarcane, cabbage, beetroot, finger millet, and pasture grass (Girsang et al., 2020). When not properly controlled, it is reported to cause yield losses of up to 100%, particularly in sub-Saharan Africa, SSA (De Groote et al., 2020; Lowry et al., 2022).

Farmers in SSA have limited knowledge about options for combating this new pest (Yigezu and Wakgari, 2020). Additionally, predators have taken time to build up to counter FAW spread, thus exacerbating the severity of their outbreaks (Akeme *et al.*, 2021). In response to the infestation, farmers in SSA have resorted to foliar sprays with synthetic

pesticides, all of which have hitherto posed long-term risks, including the development of pest resistance and adverse environmental and human health effects (Brühl *et al.*, 2022). This situation necessitates the development of alternative strategies for effective and sustainable pest management (Curl *et al.*, 2020; Ngegba *et al.*, 2022).

Maize (Zea mays L.) is among the top five crops grown and consumed globally; ranking second after sugarcane, and is considered the number one crop in East Africa (Santpoort, 2020). The production of the crop in the region has been heavily impaired by the rapidly surging populations of the FAW, which affect the crops at all stages of growth. The use of botanicals in managing FAW in maize is a suitable alternative as their mode of action is more specific, compared to the mostly broadspectrum synthetic pesticides (Njavwa Mwila, 2021). Moreover, the risk of botanicals to human health and the environment is much lower than their synthetic pesticide counterparts (Rioba and Stevenson, 2020).

Crude extracts from neem, pepper, China berry, castor plant seeds, and Tithonia diversifolia have been successfully used as botanicals for control of different crop pests globally (Egbuna et al., 2020). Azadrachta indica insecticides are widely used in agriculture, due to their effectiveness in controlling pests, coupled with their antibacterial and germicidal properties that protect plants from a variety of pests. Neem extracts have also been proven to be effective bio-pesticidal in crop production, which also do not leave hazardous residual effects on crop plants (Ukoroije and Otayor, 2020). Similarly, T. diversifolia species, which are known to contain sesquiterpene lactones and diterpenoids, possess biological properties that repel and control insect pests.

Despite these bio-pesticide innovations in crop production, little effort has been made to evaluate their effects on the FAW in maize under African conditions. The objective of this study was to evaluate an integrated botanicals and host plant resistance for managing FAW for maize production in Kenya.

### MATERIALS AND METHODS

**Study area.** The study consisted of a laboratory, greenhouse and field experiment, all conducted at the Kenya Agricultural Livestock Research Organisation, Sugar Research Institute (KALRO - SRI), Kibos, in Western Kenya. The Institute is located in Kisumu County, Western Kenya, at 0.0699° S and 34.8169° E; and at approximately 1,224 m above sea level (Mutonyi and Khaemba, 2022).

The soil type of the experimental field is Eutric Vertisols, dark, predominantly smectiterich, clays with shrinking and swelling properties. The mean annual temperatures in Kibos ranges between 21 and 23°C; while rainfall ranges between 1200 and 1500 mm per annum.

The laboratory component. The laboratory component was carried out at the SRI Entomology Laboratory at KALRO -SRI Kibos, in Western Kenya. Two botanical extracts (A. indica and T. diversifolia) were evaluated for suppressing the FAW, using the leaf dip assay method (Phambala et al., 2020). The factors investigated were botanical extracts at four levels, namely, A. indica, T. diversifolia, pesticide, and the control (distilled water without botanicals). The other factor was the accessions of maize, MA2 (an early-maturing accession) and MA4 (a late-maturing accession).

The experiment was laid out in a completely randomised design (CRD), with three replications. The experiment was repeated two times.

# Experimental plants for laboratory assay.

Two maize accessions (MA2 and MA4) from Bayer East Africa, were planted in plastic bags each of 1 m<sup>3</sup> capacity, at a spacing of 75 cm between rows and 25 cm between plants. The set up was laid out in an insect-proof greenhouse, to avoid invasion from pests.

Soil used in the greenhouse was obtained from a nearby fallow field, with known cropping history. The bulk soil sample was pulverised using a fork hoe and sieved to remove large objects and plant debris, using a 4 mm mesh sieve. Twenty-one days after emergence, leaves were harvested, washed with tap water, and cut into 3 cm x 3 cm pieces, sufficient for replicating three times per treatment (Osae *et al.*, 2022).

# Sourcing and rearing of fall armyworm. A

FAW starter colony was obtained from the International Centre of Insect Physiology and Ecology (ICIPE-Mbita), with approximately 400 eggs supplied in plastic rearing jars. The jars were kept in the entomology laboratory rearing room, at a temperature of  $25 \pm 2^{\circ}$ C,  $50 \pm 10\%$  RH, and under a 12-hour natural

photoperiod. After 2-3 days, the eggs began to hatch into first-instar larvae, which were then transferred individually to plastic rearing jars containing artificial diet supplied with the eggs (Ashok *et al.*, 2021).

The larvae were moved early in the morning to minimise diet contamination, using a sterilised camel hairbrush that had been rinsed in distilled water after being sterilised in 70 % ethanol. The date of inoculation and the initial number of larvae inoculated were recorded.

The larvae stayed in the artificial feed and moved through all its instars, until they pupated. The pre-pupal stage was transferred to a plastic jar layered with 3 cm of autoclaved soil for pupation. The pupae were collected and placed in moistened petri dishes in oviposition cages at similar conditions to those for hatching above. Sterile cotton soaked in 10% honey solution was placed in the Petri dishes as a food source for emerging adults.

The wall of the oviposition cage was lined with wax paper as an oviposition media. After 3 days, egg batches were collected from the oviposition cages and the above rearing cycles repeated for the period of the trials (Ge *et al.*, 2021).

**Phytochemical screening of botanical extracts.** Fully-grown leaves of *Azadirachta indica* and *Tithonia diversifolia*, were collected from Kibos fields and thoroughly washed with clean water, to remove soil and debris. The

leaves were then air-dried in the shade for 7 days. They were ground separately into a fine powder using a mortar and pestle. For phytochemical screening, 50 g of each powdered sample was soaked in 250 ml of absolute ethanol for 24 hours (Phambala *et al.*, 2020). The mixtures were filtered with Whatman No. 1 filter paper to obtain crude extracts, which were then concentrated by open-air evaporation at room temperature to let the ethanol evaporate gradually.

Various reagents were used for qualitative detection of phytochemicals. These included (i) Dragendorff's reagent for alkaloids, sodium hydroxide and sulfuric acid for flavonoids, ferric chloride for tannins and phenols, distilled water for saponins, chloroform and sulfuric acid for terpenoids, and (ii) Fehling's solutions for glycosides (Ngegba *et al.*, 2022). Positive results were indicated by specific colour changes or precipitate formation, allowing for the identification of secondary metabolites in the samples (Table 1).

For bioassays, fresh leaves were again collected from the same location, washed and chopped into smaller pieces (50 mm diameter). Each botanical sample was crushed into a fine paste, using a mortar and pestle. Subsequently, 75 g of the paste was mixed with one litre of distilled water in a conical flask and left to stand for 24 hours.

The resulting mixtures were first filtered through muslin cloth, and then through

TABLE 1. I	Phytochemical	properties of b	ootanicals (A.	. indica and T. di	iversifolia) extracts

Phytochemical	A. indica	T. diversifolia	
Alkaloids	+	+	
Flavonoids	+	+	
Glycosides	+	+	
Tannins	+	+	
Saponins	+	+	

<sup>(+)</sup> indicates presence of phytochemical compounds

Whatman No. 1 filter paper to obtain fine aqueous extracts. Distilled water and Lambdacyhalothrin (50 g L<sup>-1</sup> served as negative and positive controls, respectively. All prepared solutions were stored at 4 °C until used in subsequent laboratory assays.

Larval orientation. To evaluate the repellency or attraction of botanical extracts, a larval orientation assay was performed to observe the behavioral response of FAW larvae, when exposed to treated and untreated leaves. Maize leaf discs, measuring 3 cm by 3 cm, were prepared from fresh leaf samples of the two maize accessions grown in the greenhouse, using a blade and a ruler. The discs were soaked separately for 30 minutes in the various bioassays, including a control with distilled water. After soaking, the leaf discs were placed side by side on the choice test petri dishes, with four larvae positioned in the centre to assess their preference (orientation), leaf area consumed and mortality, according to Morales et al. (2021).

The maize accessions and treatments were arranged in a completely randomised design (CRD), with three replications. The process was repeated for all larval instars, using different maize accessions separately. Numerical data for preference, dislike and mortality were collected after 24 hours and recorded in Excel data sheets.

A larva was considered dead if it could not turn itself after being placed on its dorsal surface (Kumar *et al.*, 2021). Leaf area consumed data was also recorded from the 9 cm<sup>2</sup> leaves and subjected for further analysis to determine the exact area consumed by the FAW.

Field integration of botanicals and host resistance. A field experiment was carried out at the KALRO - SRI trial farms as an integrated pest management (IPM) system against FAW, initially during long rains season (trial 1) and repeated during short rains season (trial 2) in the year 2023. The experiment was

a 3x3x2 factorial, laid in a randomised complete block design (RCBD), with each treatment replicated three times. The first factor was botanical extracts application at three levels namely, *T. diversifolia* extracts: known synthetic pesticide (Lambda cyhalothrin 50 g L<sup>-1</sup>) as positive control and no botanical (NB) as negative control. The second factor was the fodder crop, *Brachiaria* cultivars, at three levels, grown on the border of the plots, namely: Mulato II, Xaraes and without border plant (WBP).

This field study was done at the KALRO-Non-Ruminant Research Institute (KALRO-NRRI), Kakamega. The third factor consisted of two maize accessions, MA2 (an early-maturing type) and MA4 (a late-maturing type), obtained from Bayer East Africa.

Treatment application. Brachiaria cultivars were planted 3 weeks prior to planting of maize, at a spacing of 30 cm between plants and 100 cm from the designated maize plots, to provide a buffer zone that minimised the interaction effect (Njarui et al., 2021). The Xareas and Mulatto II splits were randomised completely within the replicates, according to layout provided for the experiment.

Three weeks after the trap crop (Brachiaria cultivars) had been established, maize accessions were planted within the designated randomised Brachiaria plots. The maize plot size measured 200 cm x 150 cm, with an inter-row spacing of 75 cm and intrarow spacing of 25 cm, making up a total of 3 rows of 8 plants each. During the establishment of Brachiaria and maize, Di ammonium phosphate fertiliser (DAP-18:46:0) was applied at a rate of 100 kg ha<sup>-1</sup> at six weeks after planting

After planting, the crops were raised using standard agronomical practices (Agri *et al.*, 2022). Immediately after planting maize, 100 FAW adult moths that had been reared in the entomology laboratory at SRI Kibos, as described above, were released in the field, to allow for egg laying and hatching into larvae.

This was intended to allow random infestation to take place when the maize was three weeks old (Oliveira *et al.*, 2022).

The crop was monitored for foliar damage starting from the third week after planting. It was then sprayed with a *T. diversifolia* botanical extract at a concentration of 75 g L<sup>-1</sup>. The extract was evaluated alongside Lambda cyhalothrin 50 g L<sup>-1</sup> insecticide (positive control) and a negative control, which consisted of distilled water. Subsequent spraying and data collection occurred weekly until the crop reached six weeks after planting.

**Data collection.** Data were collected from eight plants in the middle row of each plot, to avoid interference by the border effects. Data on leaf damage by FAW larvae (damage severity), leaf area surface, and yield were also recorded. For damage severity, the rating was based on a visual assessment scale of Davis' 0 to 9 Whorl and Furl; where 0 = No visible damage; and 9 = Whorl and Furl leaves almost totally destroyed (Sisay *et al.*, 2019). Leaf area (LA) was calculated individually from the 8 plants, by measuring the length of the tagged leaves (from the leaf collar to the leaf tip) and the width (maximum leaf width), as per the following equation:

LA = 0.75LW

Where:

LA = the area of the single leaf in cm<sup>2</sup>; L = Length of the leaf from the leaf collar to the tip (cm); W = Maximum leaf width (cm); and 0.75 = Leaf regression co-efficient that is related to the shape of the leaf (Song and Jin 2020).

Data for yield were taken as the weight of the total grain harvested from the 8 cobs from the 8 plants per plot and weighed separately using a digital tare scale (Kern Hus 300K 1000). The moisture content was taken per sample, using a moisture meter (Twist Grain Pro, Draminski, Poland), and adjusted to the required 13.5% moisture content.

**Data analysis.** Data exploration was carried out to single out outliers, if any, and checked for conformity to normality, using graphical methods (QQ plot, box plot). Thereafter, the normality test was done using the Shapiro-Wilk test for location and the Levene test for dispersion.

Analysis of variance was carried out on the area consumed by fall army larvae, preference, and mortality. Field data, including damage severity, leaf area and yields, were analysed using analysis of variance. The posthoc tests were carried out using Tukey, with alpha of 0.05. Pearson correlation coefficients (PCCs) were computed by taking the laboratory data, except for Neem, and correlated with field data by taking the seasonal average, with the assumption of a border crop, to quantify the strength and orientation of relationships among variables. A heatmap of the correlation matrix was generated, using Seaborn in Python.

### RESULTS

# Botanical extracts under laboratory conditions

Leaf area consumed. The laboratory experiment for testing the efficacy of Azadirachta indica (Neem) and *Tithonia diversifolia* (Tithonia), demonstrated significant differences across all the treatments, in suppressing the Fall Armyworm. In maize accession 2, the control treatment recorded a significantly higher leaf area consumed (4.5 cm<sup>2</sup>), compared to all other treatments. The Neem treatment, followed by a significant increase in leaf area consumed (2.3 cm<sup>2</sup>); while Tithonia treatment resulted in a significant decrease in area consumed (1.5 cm<sup>2</sup>). The treatment with the synthetic pesticide had the least significant leaf area consumed (0.5 cm<sup>2</sup>), indicating the highest suppression of FAW.

Maize accession 4 resulted in a similar trend, where the negative control treatment had a significantly high leaf area consumed (4.5 cm²), compared to all other treatments. This was followed by Neem treatment, which had a significantly lower leaf area consumed (3.5 cm²). Tithonia treatment followed with a significant leaf area eaten (2.5 cm²); while the synthetic pesticide treatment showed the least significant leaf area consumed (0.2 cm²).

Based on the comparison between maize accessions, MA2 and MA4 treatments with negative and positive showed no significant differences, while treatments with botanical extracts displayed significant differences across the tested maize accessions. Specifically, maize accession 4 showed a significantly higher leaf area eaten; than accession 2 which showed a significantly lower leaf area eaten (Fig. 1).

**Preference.** The leaf dip and choice test experiment demonstrated that both botanical extracts [Azadirachta indica (Neem) and Tithonia diversifolia (Tithonia)] resulted in significant preference of the FAW larvae, compared to the negative control, indicating

their potential efficacy in suppressing FAW under laboratory conditions. In maize accession MA2, the control treatment recorded significantly high larval preference (3.3 larvae), compared to the Neem extract treatment (2.5 larvae), Tithonia treatment (2.1 larvae), and the synthetic pesticide treatment (1.6 larvae). There was no significant difference between treatments with Neem, Tithonia and the synthetic pesticide; although, there were variations in terms of the number of larvae. In MA4, the control and Neem treatments showed significantly higher larval preference (3.0 larvae), compared with Tithonia (2.2 larvae) and the synthetic pesticide (1.4 larvae), the latter of which showed no significant (p>0.05) differences. There were no significant differences observed across the two maize accessions, either (Fig. 2).

FAW larval mortality. The laboratory bioassay results showed clear differences in FAW larval mortality, across the treatments, including maize accessions. In accession MA2, the synthetic pesticide caused the highest larval mortality overall (1.6 larvae); particularly the negative control and Neem extract treatments

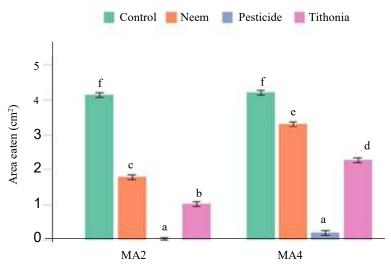


Figure 1. Mean leaf area eaten ( $\pm$ SE) by fall armyworm larvae in two maize accessions (MA2 and MA4) under different treatments. Control was treated with distilled water. Treatments with the same letter are not significantly different at ANOVA and Tukey (P < 0.05).

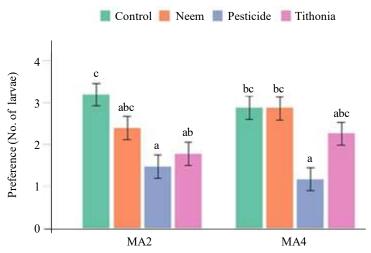


Figure 2. Mean preference ( $\pm$ SE) by fall armyworm larvae in two maize accessions (MA2 and MA4) under different treatments. Treatments with same letters are not significantly different at ANOVA and Tukey (P<0.05).

(0.3 larvae). This was, however, not significantly different from the Tithonia extract (1.0 larvae).

In accession MA4, the synthetic pesticide treatment recorded a significantly high larval mortality (2.8 larvae), which differed from the Tithonia treatment (1.7 larvae). Tithonia extract was significantly different from both Neem treatments, positive and negative control treatments, respectively (0.3 larvae).

When both maize accessions were compared, the synthetic pesticide treatment in accession MA2 and the Tithonia treatment in accession MA4 showed no significant differences in larval mortality. However, the results demonstrated that the synthetic pesticide consistently produced the most significant larval mortality; while Tithonia had a more significant effect than Neem, particularly in MA4 (Fig. 3).

# Integrating botanical and maize host resistance

Foliar damage severity. The results showed clear differences in foliar severity across various treatment combinations. Synthetic pesticide-treated plots consistently recorded

the lowest damage scores in both maize accessions, confirming their strong protective effect. For example, maize accession 4 under pesticide + Xaraes showed a damage severity of  $2.78/\pm/0.3$  in Trial 1 and  $3.71/\pm/0.31$  in Trial 2. Conversely, NB (no botanical) plots consistently exhibited the highest damage levels, especially in maize accession 2, where the values reached  $6.71/\pm/0.6$  and  $6.85/\pm/0.54$  under WBP in Trials 1 and 2, respectively. Botanical treatments with T. diversifolia provided moderate protection, with foliar damage scores ranging from 4.35 to 5.56, depending on the trap crop and maize accession (Table 2).

The performance of the trap crop also varied, with Xaraes generally associated with lower damage scores than Mulato II. Maize accession 4 experienced less damage overall compared to accession 2, indicating better inherent resistance to FAW (Table 2).

**Leaf area.** Differences in leaf area across treatments were indicative of the varying levels of plant recovery and vigor following FAW attack. Pesticide treatments, especially those combined with Xaraes, supported the highest leaf area values, reaching 469.52/ ±/ 61.13/

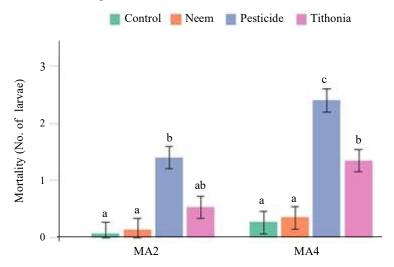


Figure 3. Mean morality ( $\pm$ SE) by fall armyworm larvae in two maize accessions (MA2 and MA4) under different treatments. Treatments with same letters are not significantly different at ANOVA and Tukey (P<0.05).

cm² in accession 4 (Trial 1). Although botanical treatments were less effective, they still resulted in moderate to high leaf area retention, with *T. diversifolia* grown borders achieving values above 400/ cm² in several combinations. Negative control treatments had the lowest recorded leaf area (291.36/±/39.98/cm²) in maize accession 2 under WBP. Among the trap crops, Xaraes combinations generally resulted in larger leaf areas, compared to Mulato II, suggesting better pest suppression (Table 2). Maize accession 4 generally exhibited greater leaf area than maize accession 2 in all the treatments (Table 2).

**Grain yield.** Grain yield data were consistent with the performance patterns observed in damage severity and leaf area, highlighted above (Table 2). Pesticide treatments consistently led to the highest yields, with the pesticide + Xaraes combinations in accession 4 reaching  $1724.33/ \pm / 98.16/ \text{ kg ha}^{-1}$  in long rains (trial 1) and  $1522/ \pm / 125.29/ \text{ kg ha}^{-1}$  in short rains (trial 2). Botanical treatments, especially those using *T. diversifolia*, also

supported relatively high yields, (e.g. 1608.67/ ±/ 46.69/ kg ha<sup>-1</sup> in accession 4). Negative control (NB) plots had the lowest and most variable yields, particularly in accession 2; while some negative control treatments in accession 4 surprisingly yielded above 1400/ kg ha<sup>-1</sup>. Among the trap crops, Xaraes consistently increased yield more than Mulato II, when combined with either synthetic pesticides or botanical extracts. Maize accession 4 outperformed accession 2 in most treatments, highlighting the benefit of using resistant varieties in IPM strategies (Table 2).

Correlations between variables. A strong positive correlation occured between pest preference and area consumed by the FAW larvae (r = 0.87). Both variables were negatively correlated with insect mortality (r = -0.64 to -0.71) and leaf area (r = -0.60 to -0.61). Notably, insect mortality showed moderate positive correlations with yield (r = 0.58) and leaf area (r = 0.57). Additionally, damage severity exhibited a moderate positive relationship with yield (r = 0.61) (Fig .4).

TABLE 2. The Mean of Maize Parameters under different IPM combinations

Treatments	Border plant	Damag	Damage severity	Leaf area (cm²)	$a (cm^2)$	Yield (kg ha <sup>-1</sup> )	g ha <sup>-1</sup> )
		Trial 1	Trial 2	Trial 1	Trial 2	Trial 1	Trial 2
				Maize accession 2			
T. diversifolia	WBP	5.38±0.5ab	4.54±0.62abcd	410.09±72.46	400.50±41.39	1147±150.68bc	1208.7±54.91
	Xaraes	5.56±0.5ab	5.46±0.68abcd	388.1±30.19	331.12±55.59	1139.33±217.75bc	1196.6±152.33
	Mulato II	5.45±0.6ab	5.66±0.65abcd	339.95±39.10	452.03±50.00	834.33±28.66bc	1126.7±162.85
Pesticide	WBP	3.90±0.3bc	3.35±0.29d	$340.22\pm40.20$	450.89±52.11	1560.67±225.92bc	1117.3±257.11
	Xaraes	4.08±0.4bc	3.34±0.6d	$340.62\pm38.56$	439.32±53.19	1359±180.08bc	1475.3±166.03
	Mulato II	4.44±0.4abc	3.75±0.41cd	$388.61\pm40.14$	428.79±45.95	1307.33±118.53bc	1085±398.08
NB	WBP	6.71±0.6a	6.85±0.54ab	291.36±39.98	403.23±62.46	728.66±54.77b	1295±184.52
	Xaraes	6.32±0.6ab	6.95±0.51ab	300.34±32.71	468.63±59.11	1189±370.24bc	1011±287.68
	Mulato II	5.71±0.5ab	7.21±0.55a	356.12±39.15	402.47±39.83	1175±69.17bc	899.6±305.73
				Maize accession 4			
T. diversifolia	WBP	4.35±0.3abc	4.73±0.6abcd	356.90±44.36	346.02±47.64	1608.67±46.69bc	1559.3±263.78
	Xaraes	4.91±0.4abc	4.28±0.72bcd	336.24±40.77	431.69±53.92	1499.66±26.36bc	1638.3±185.93
	Mulato II	5.53±0.4ab	4.57±0.43abcd	334.05±45.66	470.67±56.61	1450.33±120.40bc	1504.3±144.71
Pesticide	WBP	2.75±0.3b	3.80±0.30cd	353.32±53.57	417.11±54.85	1604.67±137.07bc	1661.3±92.89
	Xaraes	2.78±0.3b	3.71±0.31cd	469.52±61.13	383.34±47.35	1724.33±98.16a	1522±125.29
	Mulato II	4.08±0.3bc	3.51±0.42cd	327.64±46.36	467.40±71.86	1389.33±376.64bc	1689±197.53
NB	WBP	6.21±0.4ab	6.90±0.61ab	343.6±40.10	371.05±46.57	1476.33±36.35bc	1223±215.80
	Xaraes	5.80±0.5ab	5.83±0.48abcd	384.55±51.36	393.33±46.69	1665.33±89.37bc	1499.3±224.52
	Mulato II	6.09±0.5ab	6.19±0.66abc	315.46±41.41	369.63±50.76	1525.33±50.34bc	1469.3±143.72

Means with same letters across the column are not statistically different. column means without statistical letters showed no significant effect across the treatments. anova (p<0.05) and tukey hsd post hoc. Trial 1(long rains season) and Trial 2 (short rains season)



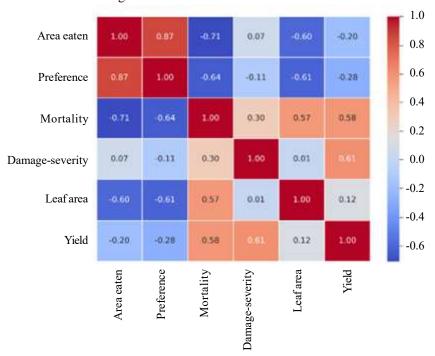


Figure 4. Correlation heatmap for variables measured in laboratory and field experiments.

### **DISCUSSION**

# **Botanical extracts and FAW populations under laboratory conditions**

Leaf area consumed. The hypothesis that the botanical extracts significantly affect the area consumed by FAW larvae, under laboratory conditions, has well been confirmed by the findings of the present study (Fig. 1). The synthetic pesticide treatment protected the leaf area fully from conservation by FAW larvae. This suggests that, although the synthetic pesticide treatment was nearly 100% effective in preventing leaf feeding damage, the two botanical extracts (A. indica and T. diversifolia) also showed efficacy in reducing the leaf area damage by FAW larvae, compared to the no botanical treatment. In fact, T. diversifolia exhibited greater protective efficacy than A. indica in this case.

Similar results were reported by George-Onaho (2023), who, while studying the effect of *T. diversifolia* extracts on pink Stem Borer (Sesamia calamistis) in maize, reported that

the botanical extract reduced the leaf damage the borer, in maize plants (George-Onaho, 2023). This is attributed to phenolic acids compounds in *T. diversifolia*, notably for their anti-feedant properties, which prevent insect feeding. Phenolic acids disrupt the feeding behavior of insects, making the plant material treated less attractive and less palatable to pests (Nino *et al.*, 2021; Othman and Latip, 2021; Ngegba *et al.*, 2022; Guo *et al.*, 2024).

Preference. The hypothesis that selected selective botanical extracts significantly deter the FAW preference for maize accessions under laboratory bioassays, was confirmed by the results from this study (Fig 2). The findings showed that treatment without botanical was highly preferred by the pest; followed by *A. indica* treatment, while *T. diversifolia* was the least preferred. This indicates that, although *A. indica* may have had some repellant property, *T. diversifolia* tended to be less attractive to FAW larvae than to the other treatments.

Gitahi et al. (2021) evaluated preventive properties of botanical extracts of T. diversifolia and Vernonia lasiopus against maize weevil (Sitophilus zeamais) and concluded that the extracts displayed repellant mechanisms by having most insects prefer untreated leaf areas of the petri dish; yet disliking the extract-treated areas. This is attributed to sesquiterpene lactones, which is distinguished for their repellent properties (Gitahi et al., 2021). They exhibit strong insecticidal and deterrent activities, making them effective in reducing pest damage on treated plants.

FAW larval mortality. The hypothesis that FAW larval mortality affects FAW larval mortality under different botanical treatments was confirmed by the leaf dip bioassay; whereby the mortality rate was highest under the synthetic pesticide treatment; followed by T. diversifolia and lastly by the treatment without the botanical extracts. These results agree with a study done by Ajao et al. (2021) whereby T. diversifolia extracts were evaluated for mortality and repellence on maize weevils (S. zeamais) and the beetle (Callosobruchus maculatus) (Ajao et al., 2021). According to the authors, the extracts significantly repelled and killed the insects (Ajao et al., 2021). This was attributed to the presence of saponins, primarily responsible for insect mortality. Saponins are glycosides that disrupt cell membranes in insects, leading to their death (Qasim et al., 2020). They act as natural insecticides by interfering with the physiological processes of insects, ultimately leading to their mortality.

# Integrating botanicals with host resistance for FAW control (field study)

Foliar damage severity. The findings of this study strongly support the hypothesis that integrating selected botanical extracts with trap crops significantly enhances maize tolerance to FAW under field conditions (Table 4).

Among the variables assessed, foliar damage severity served as a direct indicator of FAW infestation pressure and the crop's capacity to withstand or recover from pest attacks (Table 2).

The lowest damage levels were consistently observed in synthetic pesticide-treated plots; although notable reductions were also evident in plots treated with *T. diversifolia* in combination with the trap crop Xaraes (Table 2). In maize accession MA4, these integrated treatments produced foliar damage levels statistically comparable to some synthetic pesticide treatments, suggesting a beneficial interaction between the botanical and the trap crop. Xaraes outperformed Mulato II, possibly due to stronger FAW attraction or more favourable growth characteristics for pest diversion.

These results agree with those of Kumar et al. (2022), who reported significant reductions in larval feeding damage using Brachiaria cultivars as trap crops in an integrated pest management (IPM) strategy (Kumar et al., 2022). Similarly, Midega et al. (2018) demonstrated that Brachiaria spp. effectively diverted FAW in a push-pull system, thus enhancing maize protection (Midega et al., 2018).

In contrast, treatments without botanicals (NB) showed the highest damage levels, particularly in the more susceptible accession MA2. This highlights the importance of proactive FAW management, especially in varieties with low inherent resistance. Supporting evidence from Rioba and Stevenson (2020) confirms the pesticidal properties of *T. diversifolia*, attributed to its bioactive compounds that interfere with insect physiology and behaviour, thereby reducing feeding damage.

Leaf area. Leaf area served as a valuable indicator of plant recovery and photosynthetic potential, following pest attack. Generally, treatments that effectively reduced foliar damage, also supported greater leaf area

development (Table 2). The largest leaf areas were observed in plots treated with synthetic pesticides or integrated combinations, particularly of *T. diversifolia* + Xaraes and pesticide + Xaraes.

Although synthetic pesticide treatments yielded the highest leaf area mean overall, integrated non-chemical treatments also maintained substantial no-damaged leaf surface area, demonstrating their potential as sustainable alternatives strategies. Mulato II was less effective in maintaining leaf area, compared to Xaraes, mirroring its performance in suppressing foliar damage.

Maize accession MA4 consistently maintained higher leaf area under pest pressure, compared to MA2, indicating superior host resistance. This is consistent with earlier work by Prasanna *et al.* (2022), who emphasised the importance of combining host plant resistance with ecological control strategies for managing FAW in African maize systems (Prasanna *et al.*, 2022).

**Grain yield.** Although the synthetic pesticide treatments resulted in the highest yields overall, integrated treatments particularly those combining *T. diversifolia* with Xaraes also performed impressively (Table 2). In long rains, the accession MA4 treated with *T. diversifolia* + Xaraes achieved yields above 1600 kg ha<sup>-1</sup>, closely rivaling pesticide-treated plots.

This suggests that integrated approaches not only mitigate pest damage but also maintain productive capacity under pest pressure. Plots without botanical treatments, especially in MA2, experienced severe yield reductions, underscoring the economic risks of unmanaged FAW infestations. Again, Xaraes outperformed Mulato II, reinforcing its perceived suitability as a trap crop in maize-based IPM systems.

Genetic resistance played a significant role in performance across treatments. Accession MA4 consistently outperformed MA2 in both damage tolerance and yield, confirming that host plant resistance is a foundational component of sustainable pest management (Mookiah *et al.*, 2021). These results are consistent with findings by Job *et al.* (2022), who reported that maize hybrids with enhanced resistance traits exhibited improved yield and lower pest damage under FAW and stem borer pressure (Job *et al.*, 2022).

Seasonal differences also tended to influence the outcomes of the study (Table 2). The long-rain and short-rain seasons presented varying levels of pest pressure and crop performance, likely driven by differences in temperature, humidity, and other factors behind climatic variables. Other studies also support this observation, emphasising the need for adaptive IPM strategies responsive to seasonal and environmental variability (Scheiner and Martin, 2020; Sharma *et al.*, 2020; Skendžiæ *et al.*, 2021).

Correlations between variables. A strong positive correlation (r = 0.87) was observed between pest preference and leaf area consumed by the FAW larvae, suggesting that maize accessions preferred by pests tended to experience higher levels of herbivory. Both variables were negatively correlated with insect mortality (r = -0.64) and leaf area (r = -0.64) 0.71), suggesting that increased herbivore preference and feeding are associated with reduced pest mortality and reduced foliage (Fig. 4). Notably, insect mortality showed moderate positive correlations with yield (r = 0.58), implying that higher pest mortality may contribute to improved maize accession health and productivity. Additionally, damage severity exhibited a moderate positive relationship with yield (r = 0.61), which may indicate plant host resistance based on physiological or genetic properties. These findings underscore the role of pest dynamics in shaping crop performance and highlight potential targets for integrated pest management strategies.

#### **CONCLUSION**

#### ACKNOWLEDGEMENT

The integration of crude botanical extracts, Brachiaria spp., and host plant resistance shows promise in managing FAW in maize, non-chemically. Pesticide treatments, particularly when combined with resistant maize varieties, are highly effective in reducing damage severity and maintaining high yields. However, non-pesticide treatments, especially those involving *T. diversifolia* and *Brachiaria* spp., also demonstrate moderate to high effectiveness, making them viable options for integrated pest management (IPM). Bracharia cultivar Xaraes outperforms Mulato II as a trap crop, in the IPM strategy against FAW in maize. Xaraes show lower damage severity, higher leaf area, and higher maize yield, both with and without pesticide treatments, making it a more effective option for managing FAW.

The efficacy of host plant resistance in managing FAW is evident, as maize accession 4; thus, demonstrating superiority to Maize accession 2. These findings support the development of an IPM strategy that reduces overreliance on chemical pesticides and promotes sustainable agriculture, thereby increasing maize productivity. Based on the results, it is recommended to integrate T. diversifolia, Brachiaria trap crops (Xaraes), and host plant resistance using Maize accession 4 (a late-maturing variety) for effective IPM against FAW. The use of T. diversifolia and Brachiaria spp. has shown moderate to high effectiveness in reducing damage severity and maintaining yields, making them viable nonpesticide options. Maize accession 4 demonstrates strong inherent resistance, further enhancing the overall resilience of the crop. There is a need for further investigations under multiple environments and using different maize varieties to arrive at the most optimal conclusions.

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