Heliyon 10 (2024) e30068

Contents lists available at ScienceDirect

Heliyon

journal homepage: www.cell.com/heliyon

Research article

Mango headspace volatiles trigger differential responses of the mango fruit fly *Ceratitis cosyra* and its parasitoids

Raphael Njurai Miano [a,c,*], Teun Dekker [b], Egmont Rohwer [c], Tibebe Dejene Biasazin [b], Shepard Ndlela [a], Abdullahi Ahmed Yusuf [d], Xavier Cheseto [a], Samira A. Mohamed [a]

[a] *International Centre of Insect Physiology and Ecology (icipe), P.O. Box 30772-00100, Nairobi, Kenya*
[b] *Unit of Chemical Ecology, Department of Plant Protection Biology, Swedish University of Agricultural Sciences, P.O. Box 102, 230 53, Alnarp, Sweden*
[c] *Department of Chemistry, Faculty of Natural and Agricultural Science, University of Pretoria, Private Bag X20, Hatfield, 0028, South Africa*
[d] *Department of Zoology and Entomology, Faculty of Natural and Agricultural Science, University of Pretoria, Private Bag X20, Hatfield, 0028, South Africa*

ARTICLE INFO

Keywords:
Tree-attached mango
Tritrophic interaction
Parasitoids
Psyttalia cosyrae
GC-MS
Semiochemical

ABSTRACT

Before the introduction of *Bactrocera dorsalis* (Hendel) to sub-Saharan Africa, *Ceratitis cosyra* (Walker) was economically the most important pest in mango farming. Its native natural enemy, the solitary parasitoid *Psyttalia cosyrae* (Wilkinson), played a crucial role in *C. cosyra* bio-control, later complemented by the exotic parasitoids *Diachasmimorpha longicaudata* (Ashmead) and *Fopius arisanus* (Sonan) among Integrated Pest Management (IPM) systems. To understand the *in situ* mango-*C. cosyra*-parasitoid tritrophic interaction, we assessed the responses of the fruit fly and the three parasitoids to headspace volatiles from various mango conditions. These conditions included non-infested mature unripe mangoes, *C. cosyra*-infested mangoes, 7th- and 9th-day post-infestation mangoes, non-infested ripe mangoes of three varieties (Kent, Apple, and Haden), and clean air (blank). We also compared the fruit fly's performance in the mango varieties and identified the chemical profiles of mango headspace volatiles. *Ceratitis cosyra* was attracted to both infested and non-infested mangoes (66–84 % of responsive *C. cosyra*) and showed superior performance in Kent mango (72.1 % of the 287 puparia recovered) compared to Apple and Haden varieties. *Fopius arisanus* displayed a stronger attraction to the volatiles of *C. cosyra*-infested mangoes (68–70 %), while *P. cosyrae* and *D. longicaudata* were significantly attracted to the 9th-day post-infestation mangoes (68–78 %) compared to non-infested mango volatiles. Gas chromatography-mass spectroscopy showed substantial quantitative and qualitative differences in volatile profiles among mango treatments. Esters predominated in non-infested ripe, 7th- and 9th-day post-infestation mangoes, while monoterpenes and sesquiterpenes were most dominant in the other treatments. The *in situ* experiments underscored varying preferences of the species for mango headspace volatiles and their subsequent treatments. These results provide valuable insights for further exploration, specifically in identifying the key volatiles responsible for species

* Corresponding author. P.O. Box 30772, Nairobi, Kenya.
E-mail addresses: mianorn@gmail.com (R.N. Miano), teun.dekker@slu.se (T. Dekker), egmont.rohwer@up.ac.za (E. Rohwer), tibebe.dejene@slu.se (T.D. Biasazin), sndlela@icipe.org (S. Ndlela), abdullahi.yusuf@up.ac.za (A.A. Yusuf), xcheseto@icipe.org (X. Cheseto), sfaris@icipe.org (S.A. Mohamed).

https://doi.org/10.1016/j.heliyon.2024.e30068
Received 23 August 2023; Received in revised form 14 April 2024; Accepted 18 April 2024
Available online 23 April 2024

responses, to facilitate the development of applicable selective semiochemicals for managing species of African fruit fly.

1. Introduction

Frugivorous Tephritid fruit flies pose a significant challenge to the horticultural industry globally [1,2]. Among the Afrotropical native *Ceratitis* species, *Ceratitis cosyra* (Walker) (Diptera: Tephritidae, also referred to as the marula fruit fly), stands out as the most destructive species affecting mango cultivation in sub-Saharan Africa [3,4]. Although the pest is considered to be polyphagous [5], it exhibits a marked preference for mango, *Mangifera indica* L (Anacardiaceae). Infestation by this pest can lead to mango yield losses of up to 30 % if left unmanaged [6]. Other high-value fruits vulnerable to attacks by *C. cosyra* include common guava (Myrtaceae); custard apples, soursop and *Annona muricata* L. (both Annonaceae) and Avocado, *Persea americana* Miller (Lauraceae) [7]. Beyond direct fruit losses, the implication of *C. cosyra* infestations extends to export restrictions in quarantine-sensitive markets [8], given its status as a quarantine pest.

Following the invasion and widespread prevalence of *Bactrocera dorsalis* (=*B. invadens*) (Diptera: Tephritidae) [9,10] reports indicate that *C. cosyra* has been displaced by the former [11]. Nevertheless, *C. cosyra* remains a formidable challenge to mango cultivation due to its adaptability across a wide geographical range. Unlike *B. dorsalis*, which primarily inhabits low-lying areas, *C. cosyra* exhibits a wider altitudinal distribution. While it may have been displaced at lower elevations, it continues to dominate as a pest in higher elevations. For example, in Kenya, it was reported that *C. cosyra* is distributed across the country at altitudes from 20 to 2,100 masl [7]. The pest continues to be a major menace to important export mango varieties [12,13]. Furthermore, *C. cosyra* attacks an important and highly cherished Marula (Sclerocarya birrea) fruit found in many African countries [4].

In Africa, the management of fruit flies relies heavily on non-sustainable synthetic chemical insecticides, a practice with significant consequences for One Health [14,15]. Efforts have been made to explore biocontrol management agents of *C. cosyra*. For example, certain isolates of *Metarhizium anisopliae* (Metsch.) have demonstrated high potency against *C. cosyra* [16]. In laboratory studies, *Psyttalia cosyrae* (Wilkinson) (Hymenoptera: Braconidae) exhibited over 40 % parasitism on this pest [17]. However, field parasitism of *C. cosyra* by this parasitoid is very low [7]. Following the introduction of *Fopius arisanus* (Sonan), and *Diachasmimorpha longicaudata* (Ashmead) (both Hymenoptera: Braconidae) for biological control of *B. dorsalis* in Africa [18], these parasitoids formed new associations with *C. cosyra* showing outstanding performance and complementing the role of indigenous parasitoids [19]. For example, in a choice test involving six fruit fly species, *C. cosyra* was identified as the most preferred and accepted host for *D. longicaudata* [20]. Another bio-based strategy explored for the control of *C. cosyra* is the use of host marking pheromone, tripeptide glutathione (GSH) [21] as an oviposition deterrent. Its application under field conditions resulted in the reduction of *C. cosyra* infestation by up to 75 % [22].

However, unlike the other fruit flies of the genus *Bactrocera*, *Anastrepha* and *Rhagoletis,* research on semiochemicals (plants-produced volatiles) for potential use in suppressing *Ceratitis* fruit flies (except for *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae)) is scarce. Semiochemicals play various vital roles in the bi and tritrophic, (host plant-herbivores-parasitoid) communication [23,24]. In tephritid communities, it is well-documented that flies use plant semiochemicals to locate suitable host plants as oviposition sites [25,26]. Similarly, fruit fly parasitoids exploit host plant volatiles and herbivores-related volatiles for host habitat and host location [27–29].

Understanding the bi- and tritrophic interaction of fruit flies-parasitoid systems mediated by semiochemicals emitted from infested and non-infested first trophic levels (fruits) is fundamental for developing sound and sustainable management strategies. While studies in laboratory settings provide valuable information, investigations conducted in field settings are crucial for a more accurate reflection of the plant-herbivore-parasitoid interaction in nature. In this context, we have investigated the attraction and subsequent performance (in terms of the number of puparia recovered) of *C. cosyra* on tree-attached mango fruits of different ripening and infestation stages for three mango varieties (Kent, Apple and Haden). Furthermore, we assessed the response of *P. cosyrae*, *F. arisanus* and *D. longicaudata* to infested and non-infested fruits of the three mango varieties. Furthermore, we identified changes in volatile chemical composition following *in situ* infestation.

2. Materials and methods

2.1. Field experimental mango fruits

This study was conducted in an open field in Kirinyaga County, one of the mango-producing regions in Kenya. A mango orchard located in Mwea-East Sub-County, (00°41′39.8″ S 037°24′26.7″ E, 1158 m ASL) was selected for *in situ* experiments. The orchard contained various mango varieties, including local varieties (10 trees); Apple mango variety (36 trees); Kent mango variety (13 trees), Ngowe mango variety (8 trees), Mukarati mango variety (4 trees), Tommy Atkin mango variety (4 trees), Haden mango variety (6 trees), and Van Dyke mango variety (4 trees). Two mango trees of each of the Apple, Haden and Kent varieties, with immature mango fruits, were identified and exempted from routine mango sprays for use in the trials. To protect the mango fruits from insect pests, they were enclosed in fine white nets on 20 × 20 × 20 cm frames as elaborated in Miano et al. [13]. This enclosure not only created a conducive environment with adequate air circulation but also facilitated ease of handling during subsequent mango assessments. Each cage held a minimum of four mangoes, with a total of at least 32 mangoes secured for each mango variety. Duduthrin 1.75 EC powder,

containing Lambda-cyhalothrin as an active ingredient (Twiga Chemical Industries Ltd, Nairobi, Kenya), was strewed at the base of each tree every month to control crawling insects. The tree-attached mangoes that reached non-infested physiological maturity were used for *in situ* trials [13].

2.2. Ceratitis cosyra and parasitoid colonies

Ceratitis cosyra, *P. cosyrae*, *D. longicaudata*, and *F. arisanus* were reared from an established colony at the insectary of the International Centre of Insect Physiology and Ecology (*icipe*) (01° 13′ 25.3″ S, 36° 53′ 49.2″ E; 1600 m ASL, Nairobi, Kenya). The colony was maintained at 26 ± 2 °C temperature, 50–60 % RH, and a 12:12 h (L: D) photoperiod cycle. The experimental *C. cosyra* and parasitoids were reared by first exposing nine ripe mangoes (Apple variety) to 600 *C. cosyra* adults (♂: ♀ = 1:1) in a Perspex cage (30 ×30 ×30 cm) for 4 h. Subsequently, three of the freshly-infested mangoes were exposed for 19 h to egg parasitoid, *F arisanus* (200 adults; ♂: ♀ ratio = 1:1, aged between 8 and 15 days), for parasitism [13]. The parasitized and non-parasitized infested mangoes were separately incubated for larvae development. After six days of incubation, a set of three non-parasitized mangoes were separately exposed to adults of *P. cosyrae* and *D. longicaudata* (200 adults; ♂: ♀ = ratio 1:1, aged between 8 and 15 days), contained in Perspex cages (30 ×30 ×30 cm) for three days to achieve maximum parasitism. Each Perspex cage featured a fine net-sleeved window (18 cm diameter) for food and water provision. On the opposite side, a fine white net was mounted to allow air circulation [13]. Upon pupation and eclosion, the adult flies were separated into cages according to species and the rearing procedure was repeated during the study period. Adult *C. cosyra* were fed on a mixture of fine sugar and enzymatic yeast hydrolysate (ratio 3:1), and water using the protocol explained in Miano et al. [13]. Parasitoids were fed on 70 % *Eco* honey (*icipe*, Nairobi, Kenya) and water. The experimental fruit flies and parasitoids were maintained under room conditions to facilitate easy adoption in the field [13].

2.3. Responses of test insects (C. cosyra, D. longicaudata, F. arisanus, P. cosyrae) to volatiles of C. cosyra-infested and non-infested mangoes

A two-choice wind tunnel described in Ref. [13] was used to assess the response of *C. cosyra* and the parasitoids, *in situ*. Experimental mangoes were placed in mango holders crafted from Perspex glass which had an open oven bag (Lifetime Brands Europe Limited, KitchenCraft, Birmingham, UK) on top for secure placement of the mangoes. The mango holders were equipped with air inlets and outlets connected to the two-choice wind tunnel. A vacuum field pump (Analytical Research System Inc. Gainesville, Florida, USA) was used to pump clean air to each arm of the tunnel at 350 mL min $^{-1}$ and was drawn from the centre at 700 mL min $^{-1}$. Ten females (either of 8–14 day-old *C. cosyra* or 8–12 day-old parasitoids, *F. arisanus*, *D. longicaudata*, or *P. cosyrae*) of each test insect species were randomly selected from cages containing adults (♂: ♀ = 1:1). These individuals were put in releasing vials and allowed to acclimatize for 10 min. Subsequently, the test insects were released through the insect release point at the centre of the base of the wind tunnel (one species at a time) and given 20 min to choose. Insects that moved to or beyond the 30 cm mark from the insect release point were deemed to have chosen while those that remained within the 30 cm mark were considered non-responsive. Seven replicates were conducted for each insect species' choice test. To avoid positional bias, the treatment and control arms were changed between runs, and then clean air was pumped through the apparatus to remove odors from the previous experiment and to stabilize the air flow rate for 20 min. The two-choice experiment tests for each insect species are displayed in Table 1.

On the oviposition day, considered day one for freshly-infested mangoes (CC1), 15 *C. cosyra* females (8–14-day old) were randomly selected from a mixed adult population (♂: ♀ = 1:1). These individuals were released into a holder containing four mangoes and given

Table 1
The two-choice experiment tests which were performed among different treatments of volatiles for each insect species. K = Kent; A = Apple; H = Haden; NU = physiologically mature non-infested unripe mango; CC1 = *Ceratitis cosyra* freshly-infested mango; CC7 and CC9 = 7th and 9th-day post-infestation mango; NR1 = non-infested ripening mango; NR2 = non-infested ripe mango ("X" indicates the two-choice experiment tests performed for each insect species under the specified conditions).

Test Condition	*C. cosyra*	*F. arisanus*	*D. longicaudata*	*P. cosyrae*
Blank vs Blank	X	X	X	X
KNU vs Blank	X	X		
KNU vs KCC1	X	X		
KCC7 vs KNR1	X		X	X
KCC9 vs KNR1	X		X	X
KNR2 vs Blank	X	X	X	X
ANU vs Blank	X	X		
ANU vs ACC1	X	X		
ACC7 vs ANR1	X		X	X
ACC9 vs ANR1	X		X	X
ANR2 vs Blank	X	X	X	X
HNU vs Blank	X	X		
HNU vs HCC1	X	X		
HCC7 vs HNR1	X		X	X
HCC9 vs HNR1	X		X	X
HNR2 vs Blank	X	X	X	X

20-min to acclimatize before conducting the day's assays. The insects remained with the mangoes for 11 h. The flies were removed and placed in a separate cage in the evening. To ascertain oviposition, the mangoes were assessed using a ×10 hand lens to detect fruit fly punctures. The experimental mangoes were returned to the netted cage every day to prevent them from any further attack. On the 10th day post-oviposition, *C. cosyra*-infested mangoes were harvested and incubated to assess the reproduction of the fruit fly. Non-infested mangoes ripened several days after harvesting the infested ones i.e. Kent-15 days, Apple-9, and Haden-11, which made it impossible to compare the attractiveness of the insects between infested mangoes and non-infested ripe ones.

2.4. *In situ* collections of tree-attached mango headspaces

In situ, the volatile collection was done simultaneously with behavioral experiments using dynamic headspace trapping (DHT) systems. The fine netting and cages were removed, and four tree-attached mangoes were placed in an oven bag (Lifetime Brands Europe Limited, KitchenCraft, Birmingham, UK). Clean humidified air was pumped in and drawn out at 250 mL min^{-1} using the field pumps described in section **2.3**. Volatiles were trapped for 11 h between 07:00 and 18:00 local time using HayeSep-Q mixed-phase sorbents (30 mg, copolymers of polydimethylsiloxane-divinylbenzene, PDMS-DVB) that were pre-cleaned with GC-grade dichloromethane (DCM). Headspace volatile collections included (i) clean air (an empty oven bag sampled as a method blank); (ii) non-infested mature unripe mangoes (NU); (iii) *C. cosyra* freshly-infested mangoes (CC1); (iv) the 2nd-day post-infestation (CC2); (v) the 7th-day post-infestation (CC7); (vi) the 9th-day post-infestation (CC9); (vii) non-infested ripening mangoes (NR1) and non-infested ripe mangoes (NR2). After collection, the terminals of HayeSep-Q sorbents cartridges (with their respective headspace volatile organic compounds) were sealed in Teflon tape (MAAT, UK), wrapped in aluminium foil, and put in a cool box containing dry ice [13] before transporting to *icipe* laboratories, Nairobi. The headspace volatiles contained in the sorbent cartridges were eluted in 200 μL of 99.9 % dichloromethane, via high-purity nitrogen gas, into 2 mL glass vials. The eluents were stored at −80 °C until further analysis. The sorbent cartridges were then purged with nitrogen gas.

2.5. *Chemical analysis of tree-attached mango fruit headspaces*

The chemical analysis of tree-attached mango fruit headspaces was done using gas chromatography-mass spectrometry (GC-MS), on a 7890A gas chromatograph linked to a 5975C mass selective detector (Agilent Technologies Inc., Santa Clara, CA, USA) which was equipped with an HP-5 MS (5 % phenyl-methylpolysiloxane) 30 m × 0.25 mm ID × 0.25 μm film thickness column. For each headspace collection, 1 μL of eluent was injected into the splitless mode (270 °C) GC for analysis where helium was used as the carrier gas at a flow rate of 1.2 mL min^{-1}. The temperature profile was: 35 °C for the first 5 min followed by an increase of 10 °C/min to 280 °C and then held for 10.5 min. The mass selective detector and the quadrupled temperature were respectively retained at 230 °C and 180 °C, while the electron impact (EI) mass spectra were obtained at 70 eV. Furthermore, the mass range of 40–550 m/z was used to analyze the fragment ions in the full scan mode, and the filament delay time was set at 3.3 min. The elution solvent (DCM), blank runs and empty system collections were similarly analyzed to remove the solvent, column and adsorbent contaminant peaks or peak areas.

For the qualitative identification of compounds, the MS data were compared to those of reference spectra published in the library–MS databases Chemecol, the National Institute of Standards and Technology (NIST 05, 08, 11), and Adams. The experimental retention indices (RI) for each of the compounds were also computed using the Van den Dool and Kratz equation of C_5–C_{31} straight-chain alkanes and compared with literature values [30,31]. Some compounds were further authenticated using retention times of standard run under the same GC-MS conditions (Supplementary Fig. S1).

Quantification of headspace volatiles was achieved using calibration curves of α-pinene and α-humulene (purity >98 %, Sigma-Aldrich® Solutions, St. Louis, MO) prepared in concentrations ranges between 2.25 and 1000 ηg/μl. The equation $y = 2036653.8x - 5127153.0$; $R^2 = 0.9963$ from α-pinene was used to semi-quantify compounds with retention times below 16.0 min, while the equation $y = 1127808.7x - 5512234.2$; $R^2 = 0.9991$ for α-humulene semi-quantified compounds with retention times equal and above 16.0 min [21,32,33]. The concentrations obtained were converted to ηg/mango/hr [13].

3. Statistical analyses

Responses of *C. cosyra, F. arisanus*, *D. longicaudata*, and *P. cosyrae* to headspace volatiles of non-infested (ripe and unripe) and *C.* cosyra-infested mangoes: Chi-square goodness of fit was used to analyze behavioral assay data to determine whether the number of insects that landed on either arm of the olfactometer were statistically different.

Performances of *C. cosyra* in the three varieties of mangoes: The number of puparia from each variety of the three mangoes was averaged. Data was then subjected to one-way ANOVA followed by pairwise comparisons using Tukey's HSD posthoc test in R software [34].

Chemical profiles of mango headspace volatiles: The identified compounds from each mango treatment were counted and compared using Pearson's Chi-square tests followed by Chi-square multi-comparison tests in version 0.9–80 RVAideMemoire () in R [35].

To determine the distribution normality of volatile release rates, data from mango variety treatments were first compared using the Shapiro-Wilk test followed by Barlett's test for homogeneity of variances. Lacking normal distribution, the data were further subjected to the Kruskal-Wallis non-parametric rank-sum test and the pairwise comparison post hoc Dunn test in R [13,36]. Furthermore, non-metric multidimensional scaling (NMDS), similarity percentages (SIMPER) analysis, and the one-way analysis of similarities (ANOSIM) of the Bray–Curtis dissimilarity matrix [37] in *Past 3* software [38] were used for comparing the headspace chemical profile

release rates of the mango varieties.

In addition, to find the variation in the volatile release rate per mango variety, each dataset was separately analyzed using NMDS and SIMPER, and the top 30 discriminant compounds were visualized using graphs and NMDS biplots. Then the average volatile release rates of the 30 discriminant compounds were calculated and auto-scaled using the equation $y = log_{10}\ (x+1,$ where x represents the average headspace volatile release rate), and used to draw a differentiation heatmap cluster. To further understand the trend in the changes of headspace volatiles per mango variety, the average volatile release rates of each common compound across the treatments were summed up, and the percentage of each to the total was computed as follows (example):

$$\%\ release\ rate = \frac{volatile\ release\ rate\ of\ X_{NU}}{Volatile\ release\ rates\ (X_{NU} + X_{CC1} + X_{CC2} + X_{CC7} + X_{CC9} + X_{NR1} + X_{NR2})}\ x\ 100;$$

where X represents the relative release rate of a given compound in a treatment, NU represents non-infested unripe, CC represents *C. cosyra* infested, 1 = freshly-infested, 2–9 = nth day post-infestation, NR1 represents non-infested ripening, NR2 represents non-infested ripe.

Results were then visualized in graphs. This was done to figure out how these compounds (especially terpenes which are generally associated with plant defense mechanisms) change with time as a result of the treatments.

4. Results

4.1. Responses of C. cosyra, F. arisanus, D. longicaudata, and P. cosyrae to headspace volatiles of non-infested and C. cosyra-infested mangoes

In all three mango varieties, *C. cosyra* exhibited a significant attraction to volatiles from non-infested unripe mangoes (NU; Kent and Apple, $P < 0.01$; Haden, $P < 0.05$) and ripe mangoes (NR2; $P < 0.001$) compared to blanks (air) (Fig. 1A, **1 B**, and **1 C**). Additionally, *C. cosyra* showed increased attraction to headspace volatiles from conspecific freshly-infested mango fruits (CC1; $P < 0.001$ for Kent and Apple and $P < 0.01$ for Haden) compared to unripe non-infested mangoes (NU). On the 7th-day post-infestation, volatiles from infested mangoes were more attractive to *C. cosyra* (Apple, $P < 0.001$; Kent and Haden, $P < 0.01$) Similarly, the attractiveness of *C. cosyra* was higher for the 9th-day post-infestation mango headspace volatiles (CC9; Haden-$P < 0.01$, Apple- $P < 0.01$ and Kent-$P < 0.05$) compared to the volatiles of ripening mangoes (NR1; Fig. 1 A, **1 B**, and **1 C**).

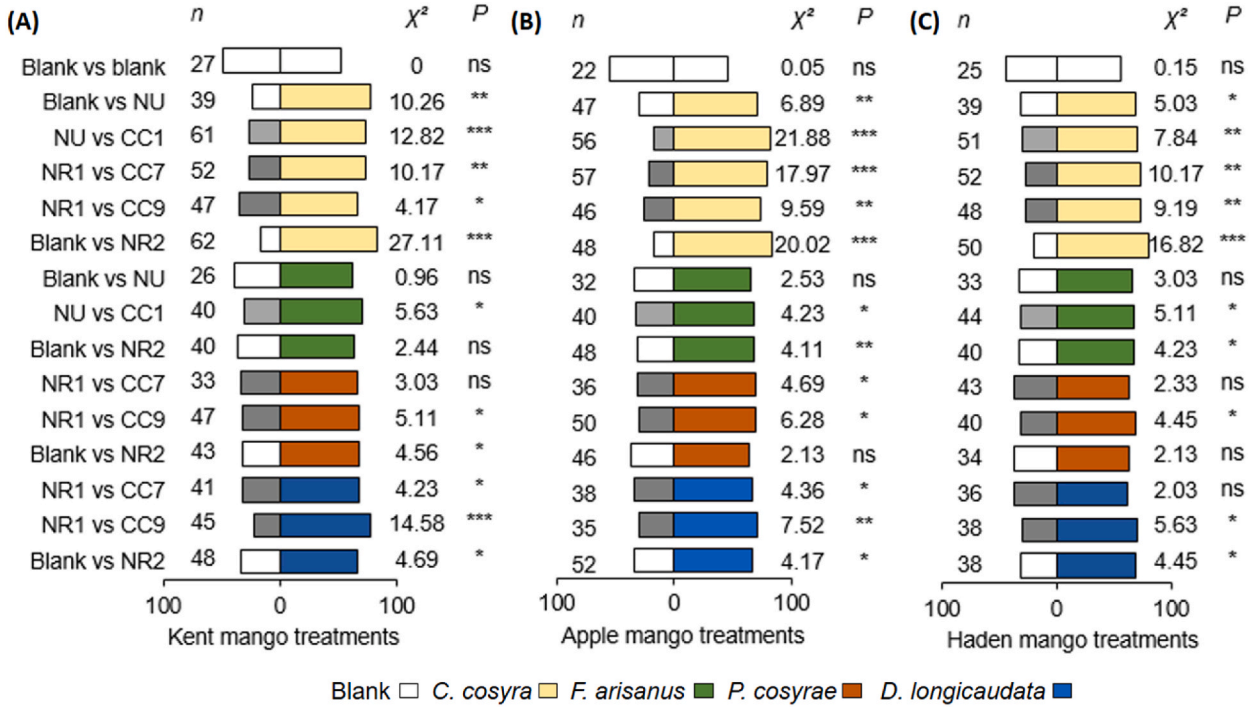

Fig. 1. Response (%) of *Ceratitis cosyra, Fopius arisanus*, *Psyttalia cosyrae***,** and *Diachasmimorpha longicaudata* to headspace volatiles of **(A)** Kent, **(B)** Apple, and **(C)** Haden mango varieties (NU = non-infested unripe; NR1 = non-infested ripening; NR2 = non-infested ripe mango; CC1 = day 1 of *C. cosyra* infestation; CC7 and CC9 = 7th-day and 9th-day post-infestation mangoes; CC═*C. cosyra*; n represents the number of responsive insects; *P*, the level of significant difference with *ns* = significantly equal, and *, **, *** = significance differences of $P < 0.05$, $P < 0.01$, and $P < 0.001$, respectively using the Chi-square goodness of test at $\alpha = 0.05$).

For the three varieties of mango, a significant number of female *F. arisanus* were attracted to headspace volatiles of freshly-infested mango fruits (CC1; $P < 0.05$) compared to non-infested unripe ones. Additionally, *F. arisanus* showed attraction to headspaces of non-infested ripe Apple ($P < 0.01$) and Haden ($P < 0.05$) mangoes compared to blanks-. However, the volatiles from non-infested unripe fruit (NU) of the three varieties, and non-infested ripe Kent fruit (NR2 Kent), were not attractive to *F. arisanus* ($P > 0.05$) compared to blank (Fig. 1 A, **1 B**, and **1 C**).

For the indigenous parasitoid, *P. cosyrae*, a significantly greater number of female wasps were attracted to the 7th-day post-infestation Apple mango volatiles (CC7; $P < 0.05$), but not to the 7th post-infestation Kent or Haden mango volatiles ($P > 0.05$), when compared to non-infested ripening ones. The volatiles of the 9th-day post-infestation (CC9) of the three mango varieties attracted *P. cosyrae* ($P < 0.05$) compared to the volatiles of their counterpart non-infested ripening mangoes (NR1). The wasps of this parasitoid were also attracted to non-infested Kent ripe mangoes (NR2; $P < 0.05$) compared to the blank whereas there was no significant attraction to non-infested ripe Apple and Haden mangoes (NR2; $P > 0.05$) when compared to blank (Fig. 1 A, **1 B**, and **1 C**).

The parasitoids *D. longicaudata* and *P. cosyrae* responded equally to headspace volatiles of the 7th-day post-infestation mango of Apple and Haden varieties compared to their respective non-infested ripening ones (NR1). On the other hand, unlike that for *P. cosyrae,* a significantly greater number of *D. longicaudata* was attracted to headspaces of the 7th-day post-infestation Kent mangoes (CC7; $P < 0.05$) compared to those attracted to non-infested ripening ones (NR1). Additionally, *D. longicaudata* was significantly attracted to the 9th-day post-infestation mangoes (CC9; Haden, $P < 0.05$; Apple, $P < 0.01$; Kent-$P < 0.001$) relative to the non-infested ripening (NR1). Non-infested ripe mango headspaces for the three varieties attracted more *D. longicaudata* (NR2; $P < 0.05$) compared to blanks (Fig. 1 A, **1 B**, and **1 C**).

As mentioned earlier, infested mangoes detached from the tree quite earlier than the non-infested mangoes, making comparisons of the responses of test insects to infested versus non-infested mangoes impossible. For example, from the day the infested mangoes were harvested, non-infested Kent mangoes took 15 more days to ripen, while Apple took 9, and Haden took 11 days, which made it impossible to compare the attractiveness of the insects to the infested and non-infested ripe mangoes.

4.2. Performances of *C. cosyra* in the three varieties of mangoes

The performance of *C. cosyra*, as indicated by the average number of recovered puparia exhibited significant variation ($F = 260.1$, $df = 2$, $P < 0.0001$) among the mango varieties (Fig. 2). Among the 287 recovered puparia, Kent mango showed the highest yield (72.1 %), whereas Haden had the lowest (10.8 %). There was no significant difference between puparia recovered from Apple and Haden varieties of mango.

4.3. Chemical profiles of headspace volatiles of the three mango varieties

A total of 184 compounds were tentatively identified from different treatments of the three varieties of mango. Of these, 69 were esters, 34 sesquiterpenes, 25 monoterpenes, 13 alcohols, 11 monoterpenoids, 11 aldehydes, 9 ketones, 4 organic acids, 2 Benzenoids, 2 sesquiterpenoids, 2 diterpenoids, 1 lactone and 1 furanone (Supplementary Table 1). Myrcene, α-Pinene, δ-3-carene, β-pinene, α-gurjunene, β-copaene, (*E*)-caryophyllene, δ-cadinene, and α-humulene were present in each treatment of the mango varieties (Supplementary Table 1). Furthermore, ethyl propionate, methyl butanoate, 2-methyl-1-butanol, 2-methyl propyl ethanoate, ethyl 2-methyl prop-2-enoate, and ethyl 3-hydroxy butanoate were common compounds detected in the headspaces at 7th- and/or 9th-days post-infestation on all mango varieties.

We also observed qualitative and quantitative differences in the headspace volatile constituents, which varied among treatments and time of volatiles collection for each mango variety (Fig. 3).

Among the treatments of the three varieties of mangoes, the number of identified compounds differed significantly ($\chi^2 = 41.328$, $df = 6$, $P < 0.001$ for Kent variety; $\chi^2 = 28.722$, $df = 6$, $P < 0.001$ for Apple variety; and $\chi^2 = 54.287$, $df = 6$, $P < 0.001$ for Haden variety), being highest for 7th- and day 9th-day post-infestation for both Kent and Apple varieties, and for day 9th-day post-infestation for

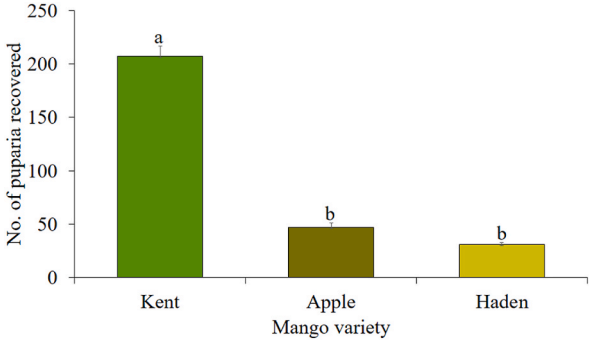

Fig. 2. Performance of *Ceratitis cosyra* on three mango varieties. Different letters on the bars indicate a significant difference (One-way ANOVA test followed by Tukey's HSD posthoc test).

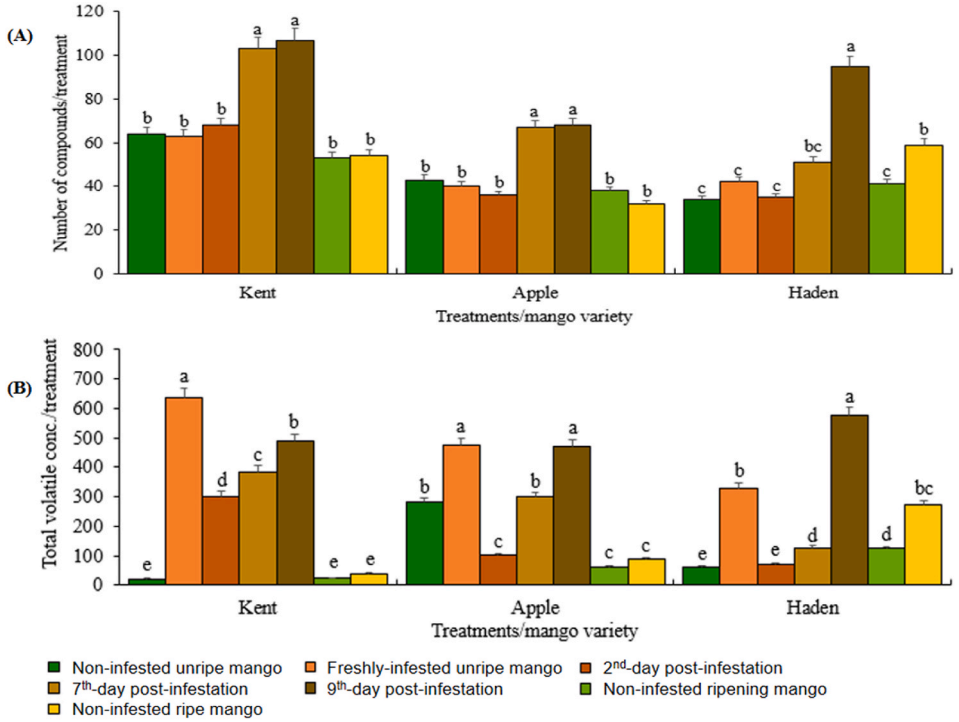

Fig. 3. The number of identified compounds of each mango treatment (Pearson's Chi-square and Chi-square multi-comparison tests) **(A)**; Total volatile release rates/mango treatment of each variety of mango (Kruskal-Wallis rank-sum test followed by pairwise posthoc Dunn test at $\alpha = 0.05$) **(B)**. Different letters on bars from the same variety indicate a significant difference.

Haden (Fig. 3 A). Also, the total volatile release rates varied among treatments of each mango variety ($\chi^2 = 25.012$, $df = 6$, $P < 0.00034$; $\chi^2 = 22.374$, $df = 6$, $P < 0.001036$; and $\chi^2 = 24.502$, $df = 6$, $P < 0.000422$, for Kent, Apple and Haden, respectively) (Fig. 3 B), being highest for freshly-infested fruits for Kent while it was highest for 9th-day post-infestation for Haden variety. For the Apple mango variety, both of these treatments (freshly-infested fruits and day 9 post-infestation) had the highest release rates. Generally, the total volatile release rates of both non-infested unripe and ripe mangoes were lower than those of infested ones (Fig. 3 B), especially in the case of the Kent variety.

The 30 topmost discriminant compounds of the volatiles of the three mango varieties contributed to 88.81 % of the total dissimilarities (Bray-Curtis similarity percentage, SIMPER, Fig. 4 A). The compounds that significantly contributed to the separation and clustering were δ-3-carene (22.1 %), myrcene (14.1 %), α-pinene (9.3 %), ethyl octanoate (8.3 %), and β-phellandrene (4.2 %). Headspace volatiles from all treatments were successfully grouped into defined clusters, with overlaps between Kent and Haden headspaces (NMDS: k = 2, stress = 0.1159, Fig. 4 B, Supplementary Fig. S2A). One-way analysis of similarity, ANOSIM, indicated a significant difference among headspace volatile release rates of the treatments for the three mango varieties ($R = 0.9654$, $P = 0.0001$). Most of the 30 top discriminant compounds were associated with the headspaces of *C. cosyra*-infested mangoes (Fig. 4C).

Among the treatments of Kent mango, a total of 135 compounds were detected out of which 23 compounds were shared in all treatments. Methyl benzoate, cyclooctanone, pinocarvone, 6-camphenol, *p*-methyl acetophenone, 3-caren-10-al, (*Z*)-3-hexenyl salicylate, benzyl benzoate, and benzyl salicylate were present in the headspace of the freshly-infested and/or day 2 post oviposition, while ethyl 2-methyl propionate, 2,3-butanediol, ethyl 3-methyl butanoate, methyl tiglate (methyl 2-methyl-2-butenoate), *n*-hexanol, 2-heptanone, methyl hexanoate, 2-methyl propyl butanoate, *m*-cymenene, and (2-*endo*,3-*exo*)-3-methyl bicyclo[2.2.1] heptane-2-carboxaldehyde were among the 36 compounds that were detected only from the 7th- and/or 9th-day post-infestation headspace volatiles. Moreover, 19 compounds were detected in infested and non-infested ripe mangoes which included isopentyl formate, (*Z*)-but-2-enoic acid, (*Z*)-ethyl but-2-enoate, ethyl 2-methyl butanoate, ethyl tiglate (ethyl 2-methyl-2-butenoate), ethyl hexanoate, ethyl hex-(2*E*)-enoate, ethyl heptanoate, phenyl ethyl alcohol, and methyl octanoate.

The top 30 most discriminant compounds as per SIMPER of NMDS (Fig. 5 A), accounted for 90.9 % of the dissimilarity contribution. Of these compounds, α-phellandrene, ethyl octanoate, myrcene, ethyl-(4*E*)-decenoate, and limonene contributed 57.4 %.

A significant difference was registered among the headspaces' volatile release rates for Kent mango treatments (ANOSIM, $R = 0.9715$, $P = 0.0001$; Fig. 5 B; Supplementary Fig. S2B). The most discriminant compounds were associated with headspaces of infested Kent mangoes (Fig. 5 B). Among the compounds that were most discriminant was δ-3-carene (C62) which was mostly more abundant in almost all treatments of Kent (Fig. 5C). Furthermore, the heatmap shows that the selection of compounds was spread in almost all possible categories, for example, compounds that appeared in all treatments and those that did not, compounds with a difference in abundance, and compounds from different classes among others.

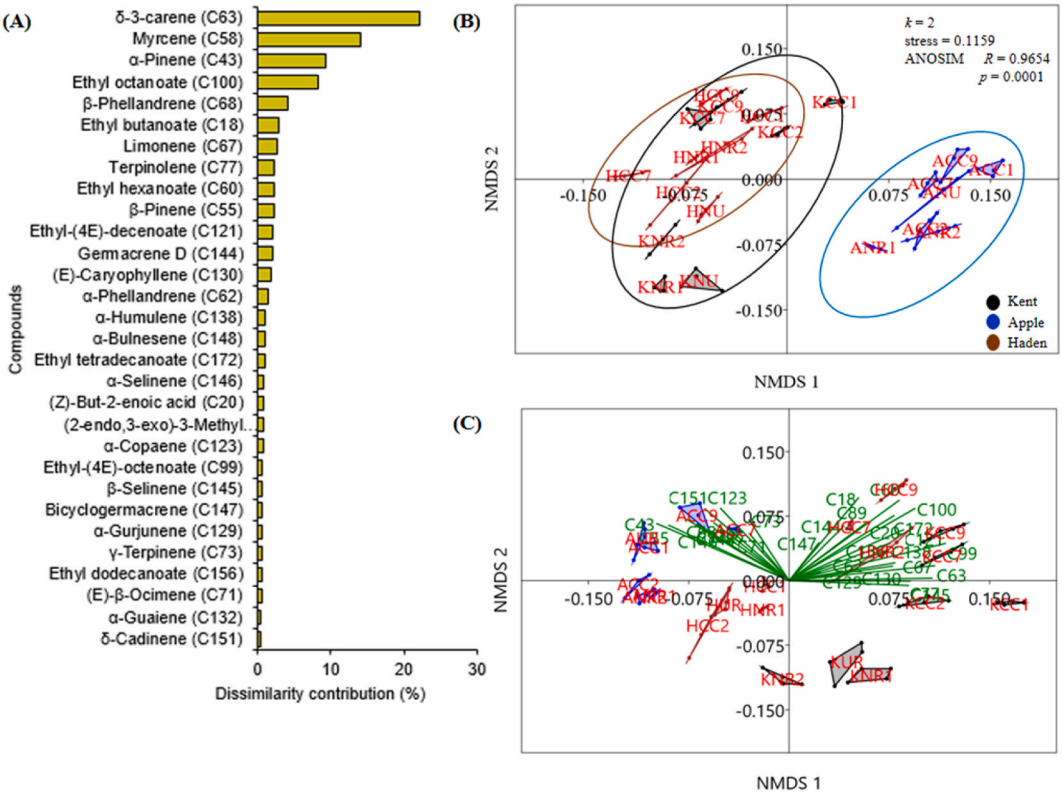

Fig. 4. **(A)** The most discriminant compounds for the 3 mango varieties based on similarity percentage (SIMPER) of NMDS. **(B)** NMDS plot shows the scattering of compounds of the treatments from the three mango varieties in the Bray–Curtis dissimilarity matrix (k = 2; stress = 0.1159). **(C)** NMDS biplots showing the spread of the selected 30 most discriminant compounds within the headspaces (H = Haden; A = Apple; K = Kent; CC = *Ceratitis cosyra*; NU = non-infested unripe mango; NR1 = non-infested ripening mango; NR2 = non-infested ripe mango; 1 = *C. cosyra* freshly-infested; 2, 7, and 9 = the nth days of *C. cosyra* post-infestation).

A total of 82 compounds were identified from Apple mango headspaces out of which 28 were common in all treatments. Verbenone, 6,7-epoxymyrcene, and caryophyllene oxide were the only compounds that were added on the day of infestation relative to those of non-infested mango headspace. Furthermore, 31 compounds were only identified from the 7th- and/or 9th-day post-infestation mango headspaces which included ethyl propionate, *n*-propyl acetate, isopentyl formate, ethyl butanoate, ethyl 2-methyl propionate, 2-methyl propyl ethanoate, 2-methyl-1-butanol, 2,3-butanediol, ethyl 2-methyl butanoate, and (*Z*)-ethyl but-2-enoate but not in the other treatments. Ethyl octanoate, aromadendrene, and bicyclogermacrene were the only common compounds among the infested and the non-infested ripe Apple mango headspace volatiles.

In addition, the 30 topmost discriminant compounds as per SIMPER of NMDS contributed 97.0 % of the total dissimilarity contribution (Fig. 6 A). Myrcene, α-pinene, α-bulnesene, β-pinene, and β-phellandrene were the top five discriminant compounds contributing 69.9 %.

Unlike in Kent mango treatments, in Apple mango, there was no clear separation of the top most discriminating compounds of NMDS biplots as either from infested or non-infested headspaces ($k = 2$, stress = 0.05093; Fig. 6 B; Supplementary Fig. S2C). However, ANOSIM registered a significant difference in volatiles of all treatments of the Apple variety of mango ($R = 0.6882$, $P = 0.0001$; Fig. 6 B) The heatmap (Fig. 6C) shows that most discriminant compounds were selected from most classes of compounds where α-pinene and myrcene were the most dominant compounds.

Haden mango had 109 identified compounds out of which 22 were common in the headspace volatiles of all treatments. α-Fenchene, (*Z*)-β-ocimene, *p*-methyl acetophenone, and caryophyllene oxide were additional compounds identified from the headspaces of freshly *C. Cosyra* infested conspecifics relative to non-infested unripe mango compounds. Moreover, of the 7th- and/or 9th-day post-infestation mango headspaces, 38 additional compounds were identified relative to those of non-infested mangoes. Among the additions were isopentyl formate, 3-pentanone, acetoin, ethyl propionate, methyl butanoate, butanoic acid, 2-methyl-1-butanol, (*Z*)-ethyl but-2-enoate, (3*Z*)-hexenol, 4-hydroxy-2-pentanone, and methyl tiglate (methyl 2-methyl-2-butenoate). Additionally, 25 compounds were common in infested and non-infested ripe mango headspaces. These compounds included ethyl 2-methyl propionate, ethyl 2-methyl butanoate, ethyl pentanoate, ethyl tiglate (ethyl 2-methyl-2-butenoate), 2-methyl propyl butanoate, ethyl hexanoate, ethyl hex-(2*E*)-enoate, ethyl heptanoate, and methyl octanoate among others.

The 30 most discriminant compounds, selected by SIMPER of NMDS (Fig. 7 A), accounted for 91.1 % of the total dissimilarity

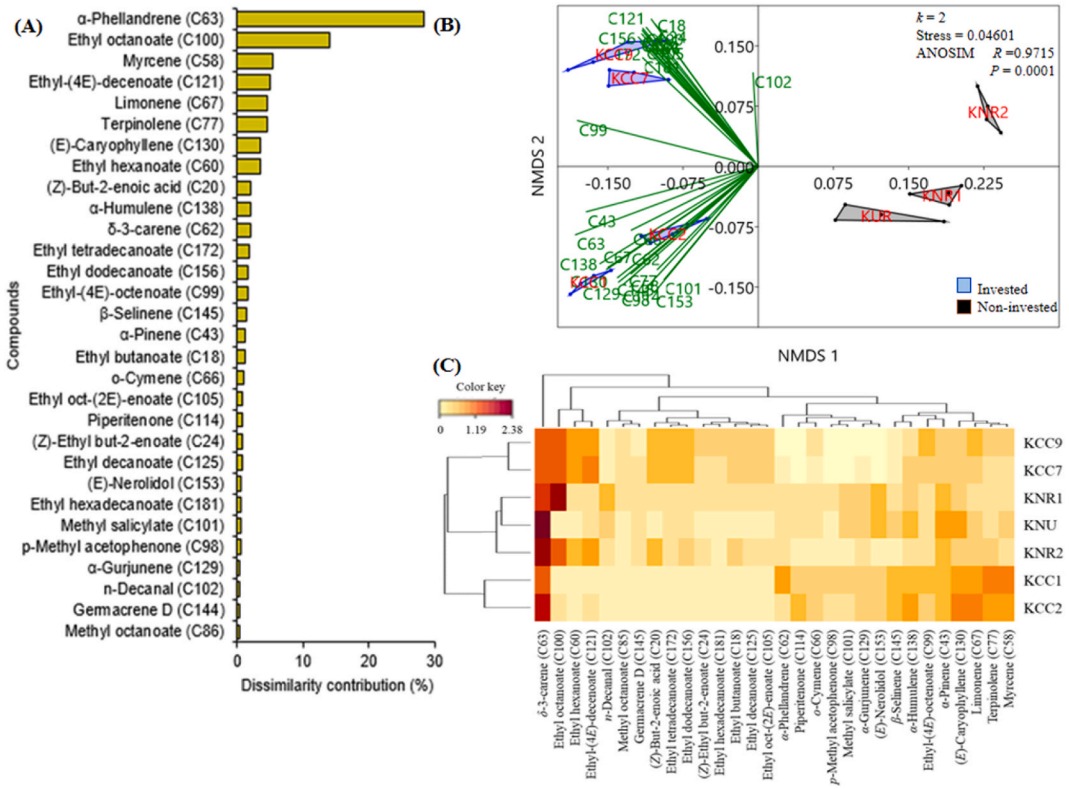

Fig. 5. **(A)** The most discriminant volatiles of all treatments of Kent variety of mango based on the similarity percentage in decreasing order of importance. **(B)** NMDS biplots show differentiations in the treatments of the 30 most discriminant compounds. **(C)** Heatmap of volatile release rates of the 30 selected compounds. The dark brown color represents the highest mean volatile release rate. (K = Kent variety; KCC2 = 2nd-day post-infestation Kent mango; KCC1 = *Ceratitis cosyra* freshly-infested mango; KNR2 = non-infested ripe mango; KNU = non-infested unripe mango; KNR1 = non-infested ripening mango; KCC7 = 7th-day, and KCC9 = 9th-day post-infestation mangoes).

contribution. Of these compounds, germacrene D, ethyl hexanoate, ethyl butanoate, ethyl octanoate, and δ-3-carene, contributed 63.3 %. There was a significant variation (ANOSIM, $R = 0.9269$, $P = 0.0001$) in the volatile release rates among the treatments of Haden mangoes (Fig. 7 B; Supplementary Fig. S2D) but like in apple mango, there was no clear separation of clusters of non-infested and infested mango volatiles. The selected compounds are spread in almost all categories e.g. classes of compounds, and release rates amongst others (Fig. 7C). δ-3-Carene had the highest release rate among the selected compounds, except on the 7th-day post-infestation mangoes when ethyl butanoate was the major compound (Fig. 7C). However, about 90 % of the 30 compounds were associated with headspaces of non-infested ripe Haden mangoes (HNR2), freshly-infested mangoes (HCC1), the 7th-day (HCC7), and the 9th-day (HCC9) post-infestation mangoes (Fig. 7 B).

Overall, a strong increase in the amounts of common compounds in Kent mango following *C. cosyra* infestation (Fig. 8 A; Supplementary Fig. S3A). However, the release rates of these compounds in non-infested ripening and ripe mangoes were generally lower than those of non-infested unripe mangoes. The trend for release rates of volatiles differed in Apple and Haden mangoes, where non-infested ripe and unripe mangoes released substantial amounts of volatiles, although in most cases lower than infested mangoes. (Fig. 8 B and **8 C**; Supplementary Fig. S3B and S3C).

5. Discussion

Tephritid fruit flies and their parasitoids rely on semiochemicals for locating suitable hosts for oviposition, and many are known for their broad host range [18,39]. *Ceratitis cosyra*, a member of Tephritid, is a notorious mango pest [3]. The parasitoid *P. cosyra* has a natural association with this pest [17], while *F. arisanus* and *D. longicaudata* have positively adapted as an alternative parasitoid in the control of *C. cosyra* following its introduction and release in Africa [20,40]. In this study, we explored the *in situ* tritrophic interactions of *C. cosyra* and its parasitoids with the mango.

Our results indicate that *C. cosyra* dissimilarly responded to headspace volatiles from ripe and unripe mangoes compared to their respective blanks (clean air). Similar results have been reported for *B. dorsalis* [13]. Surprisingly, *C. cosyra* exhibited a preference for volatiles from infested over non-infested unripe mangoes, indicating the ability to discriminate between them. Perhaps, volatiles emitted as a result of *C. cosyra* infestation masked oviposition volatiles, which otherwise could serve as host marking pheromone (HMP) to deter conspecifics of this species, as recently documented by Cheseto et al. [21,22]. Another explanation is that the volatiles

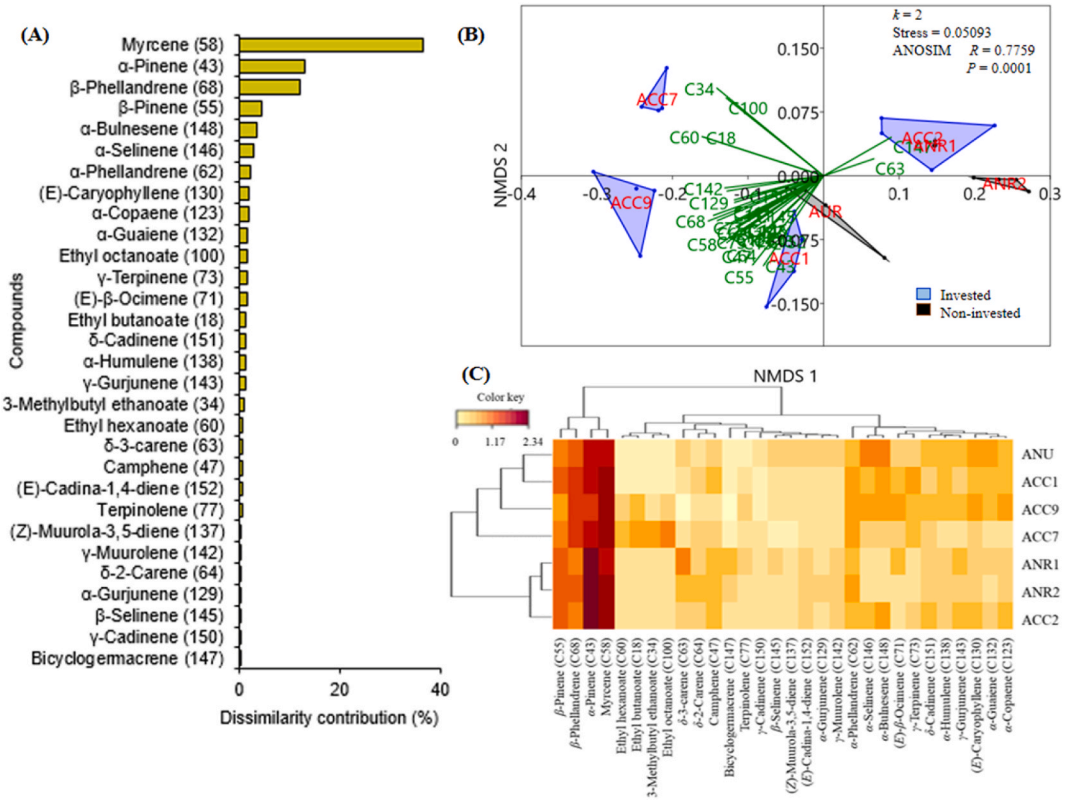

Fig. 6. **(A)** The most discriminant volatiles of all treatments of Apple mango variety based on the similarity percentage in decreasing order of importance. **(B)** NMDS biplots show the differentiation in the mango treatments of the 30 most discriminant compounds. **(C)** Heatmap of volatile release rates of the 30 selected compounds. The dark brown color represents the highest mean volatile release rate. (A = Apple variety; ACC2 = 2nd-day post-infestation mango; ANR2 = non-infested ripe Apple mango; ANR1 = non-infested ripening Apple mango; ACC7 = 7th-day, and ACC9 = 9th-day post-infestation mangoes; ACC1 = *Ceratitis cosyra* freshly-infested Apple mango and ANU = non-infested unripe Apple mango).

released by infested fruit may indicate the presence of an oviposition substrate or the presence of punctures that would facilitate subsequent oviposition by *C. cosyra*. To support this argument, during data correction at the field, photos (Fig. 9) were taken in which a female *C. cosyra* successfully made an oviposition puncture and proceeded to oviposit (Fig. 9 A). Another conspecific is spotted smelling the newly made oviposition puncture (Fig. 9 B) with an ulterior motive of chasing the ovipositing fruit fly (Fig. 9C). It was later joined by a second conspecific (D). The invested mango attracted more fruit flies leading to multiple oviposition punctures (Fig. 9 E). Later, on incubating the tree-detached infested mango, many larvae of different developmental stages emerged from the rotting mango indicating that more fruit flies managed to oviposit on it at different time intervals (Fig. 9 F). Sometimes, a *C. cosyra*-infested Kent mango attracts the fruit fly larvae' natural enemy which feeds on the part containing larvae yet adjacent to it are clean non-infested fruit (Fig. 9 G). Interestingly, most *in situ*-infested mangoes give forth to hundreds of adults. The emergence of adults mostly takes a span of fourteen to seventeen days but with many casualties of larvae and puparia which could be attributed to lack of enough feeds.

These examples indicate that *C. cosyra* may indeed prefer ovipositing in preexisting oviposition punctures instead of making new ones or ovipositing on an already infested mango despite having host-marking pheromones just like what has been reported of a congeneric species, *C. capitata* [41]. Similarly, *B. dorsalis* was more attracted to mangoes with ovipositing conspecific females in a field set-up [13]. Conversely, *B. zonata* (Saunders) (Diptera: Tephritidae) was reported to be more attracted to non-infested guava than infested ones [42]. On the other hand, the oviposition choice of *B. tryoni* (Froggatt) (Diptera: Tephritidae) was not dependent on the infestation status when offered high-quality guava [43].

The results have also demonstrated that the parasitoid species used in this study were attracted to headspace volatiles emitted from the host fruit of their host. *Fopius arisanus* was attracted to the headspace of freshly *C. cosyra*-infested mangoes which agrees with earlier findings in which the egg parasitoid was attracted to other fruit fly-freshly-infested hosts [13,27,29,44]. The parasitoid also prefers younger host's eggs for parasitism compared to aged ones [45,46]. The significant number of responsive *F. arisanus* to headspaces of non-infested ripe mango fruits (except for Kent) was reported by Miano et al. [13] on the same varieties of mangoes and by Altuzar et al. [47] who used non-infested guava. The lower response of *F. arisanus* to headspace volatile of ripe Kent mango compared to the other two varieties could be explained by the fact that this variety is a poor host for the parasitoid's coevolved host insect, *B dorsalis* [13].

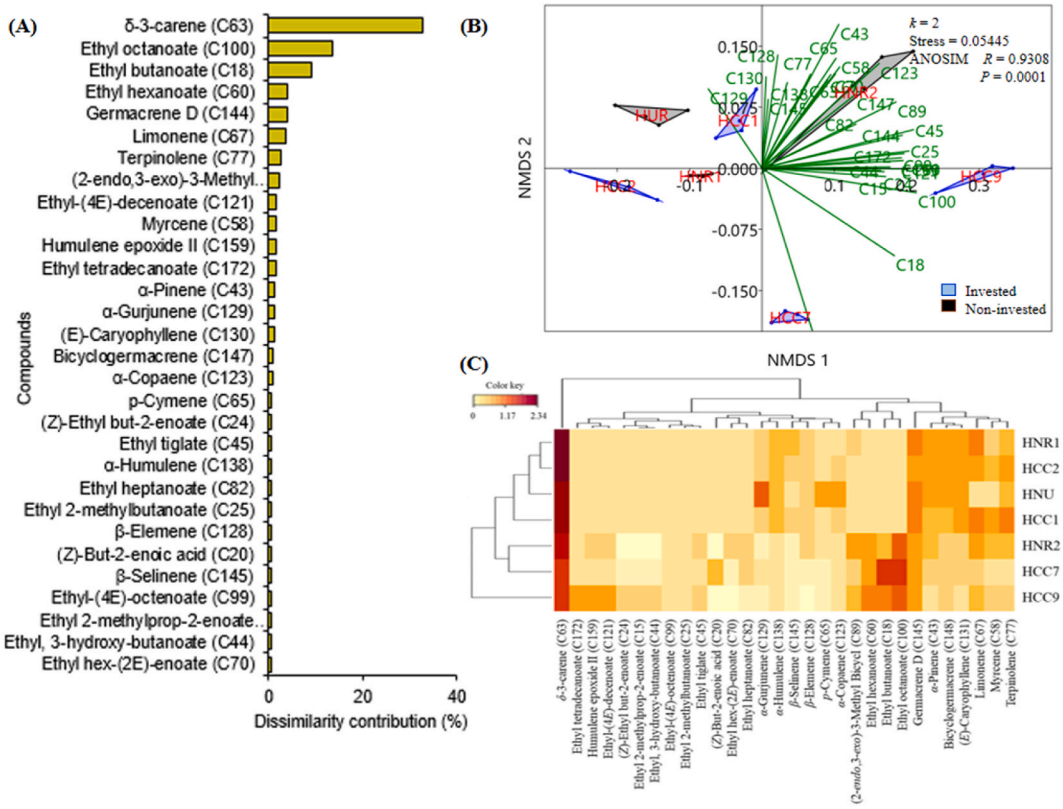

Fig. 7. **(A)** The most discriminant volatiles of all treatments of Haden mango variety based on the similarity percentage in decreasing order of importance. **(B)** NMDS biplots show the differentiation in the Haden mango treatments of the 30 most discriminant compounds. **(C)** Heatmap of volatile release rates of the 30 selected compounds. The dark brown color represents the highest mean volatile release rate. (H = Haden; HCC9 = 9th-day post-infestation; HCC7 = 7th-day post-infestation; HNR2 = non-infested ripe; HCC1 = *Ceratitis cosyra* freshly-infested Haden mango; HNU = non-infested unripe mango before infestation; HCC2 = 2nd-day post-infestation mango; and HNR1 = non-infested ripening Haden mango).

The high responses of *P. cosyrae* and *D. longicaudata* to advanced-stage infested mango headspace volatiles (7th- and 9th-day post-infestation) is not unexpected, as these parasitoids prefer late larval instars of their host [28]. The higher attraction of *D. longicaudata* to *C. cosyra* 9th-day post-infestation mangoes of the Kent variety compared to Apple and Haden varieties could be attributed to the higher preference of this host for Kent mangoes, indicated by the greater number of recovered puparia from this variety, potentially triggering the production of a higher number of headspace volatiles compounds, as shown in figure (4 **A**). Nunez-Campero et al. [48] reported that host density greatly influences the number of parasitoids that would visit the host fruit of the fruit fly. Although we do not have a conclusive explanation for the differential varietal response of these parasitoids to 7th-day post-infestation mangoes, with a higher response to the headspace of Kent and Apple varieties for *D. longicaudata* and only the headspace volatile of Apple variety for *P. cosyrae*, we can hypothesize that the development rate of *C. cosyra* may vary with mango varieties, thus affecting the volatiles (in term of quantities/qualities) that emitted from the different varieties. Here, we have reported a stronger attraction of *D. longicaudata* to a wider range of mango treatments. This finding can explain previous laboratory findings in which *D. longicaudata* reproduction outperformed its native parasitoid *P. cosyrae* [19].

The differential performance of *C. cosyra* on the different mango varieties in terms of the number of recovered puparia suggests that *C. cosyra* prefer the Kent mango variety as an oviposition substrate compared to varieties of Apple and Haden mangoes. Despite reports of *B. dorsalis* displacing *C. cosyra* from its habitats [11], our findings suggest their reproduction is variety-oriented and hence holds significant importance in mango farming. For instance, in the mango season in Kenya, Kent mango matures last compared to other commercial varieties. Therefore, we hypothesize that the lower performance of *C. cosyra* in the early maturing varieties of Apple and Haden mangoes, assures a slow increase in its population as farmers take more precautionary measures managing *B. dorsalis* in their orchards. This increase is followed by an invasion of its favorite late-maturing Kent mango resulting in considerable crop damage, sometimes leading to 100 % crop loss. A close inspection of the mango orchard where this study was conducted indicated a decline in *B. dorsalis* population during the maturing and ripening of the Kent mango variety. This finding will advise policymakers on the best framework for IPM measures to implement where different species of fruit flies pose a problem for a given crop.

The findings also align with the preference/performance hypothesis which states that "female insects will evolve to oviposit on hosts on which their offspring fare best" [49,50]. Diatta et al. [51] reported similar observations on *B. dorsalis*' performance (=*invadens*) among mango varieties while more recently, Miano et al. [13] demonstrated that the puparia recovered from mangoes

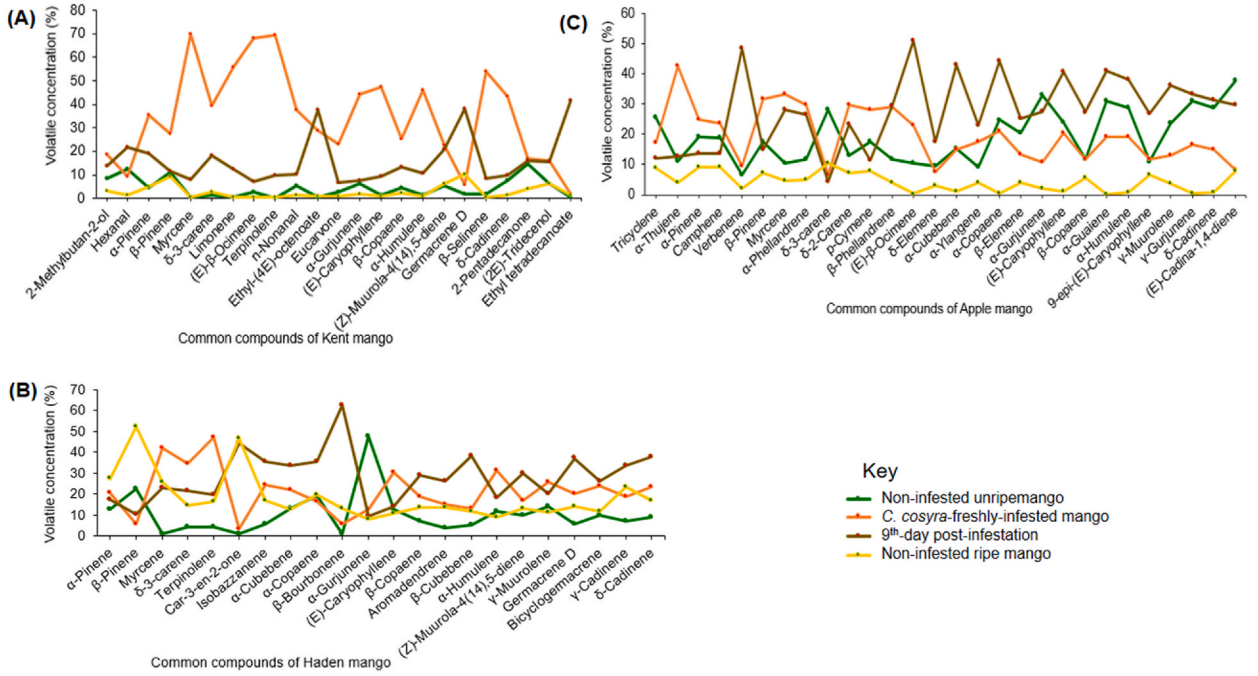

Fig. 8. Percentages of the average volatile release rates of each common compound (relative to the total) of non-infested ripe mangoes; 9th-day *C. cosyra* post-infestation mangoes; *C. cosyra* freshly-infested mangoes; and non-infested unripe mangoes for the three varieties, **(A)** Kent; **(B)** Apple; and **(C)** Haden.

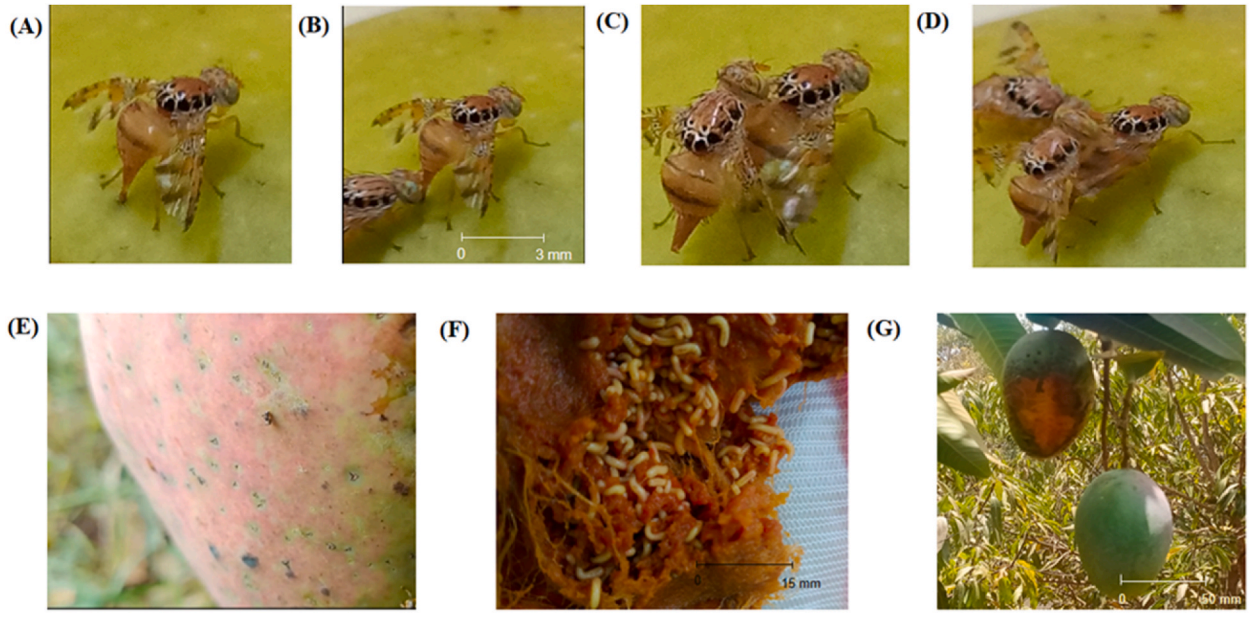

Fig. 9. Female *Ceratitis cosyra* ovipositing on a punctured mango (**A**). Another conspecific smelling the newly made oviposition puncture (**B**) with an ulterior motive of chasing the ovipositing fruit fly (**C**) and (D). Infested mango with multiple oviposition punctures (**E**). Rotten mango with many larvae of different developmental stages (**F**). *Ceratitis cosyra* infested Kent mango having been fed on by fruit fly larvae' natural enemy (**G**).

varied with varieties, whereas *B. dorsalis* failed to perform in Kent variety but did better in the Apple mango variety.

This study demonstrated that the behaviors of *C. cosyra*, *P. cosyrae*, *D. longicaudata* and *F. arisanus* are influenced by headspace odors. The quantitative and qualitative differences in headspace compounds, with notable overlaps among varieties, as well as across treatments, were also reported for the same varieties of mangoes before and after *B. dorsalis* infestation [13]. The changes in the

chemical profile have been linked to the genetic characteristics of a given mango variety [52,53] and the treatments for the same fruit variety [13,25,54]. In this study, a lower number of esters were detected from headspaces of non-infested unripe mangoes compared to those of their respective non-infested ripe mangoes, where defense-related terpenes were generally reduced both in numbers and release rates in ripe mangoes. Fruit ripening signifies readiness for seed dispersal and is connected to the attraction of predators, insects and different microorganisms, mostly characterized by the release of attractive chemical signals [55,56]. *Ceratitis cosyra* may, therefore, be similarly attracted to ripe mangoes due to the increase in esters. Interestingly, among the experimental varieties of mangoes, the majority of most discriminant compounds by SIMPER of NMDS were esters, suggesting their possible significance in the attraction of *C. cosyra* and its parasitoids.

The number of compounds produced by *C. cos*yra freshly-infested mango was not significantly different from compounds of non-infested unripe mangoes although we have reported a general increase in their release rates. War et al. [57] associated changes in volatile content after the herbivorous attack of a plant with defense mechanisms of the plant against the herbivorous. Here, we have reported an increased attractiveness of freshly-infested mango to *C. cosyra* conspecifics. Similar results were reported for *B. dorsalis* on mangoes with ovipositing *B. dorsalis* [13], *C. capitata* on kumquat, *Fortunella japonica* Swingle (Rutaceae) [41], and *Scirtothrips dorsalis* (Hood) (Thysanoptera: Thripidae) on Bell pepper, *Capsicum annuum* L (Solanaceae) [58]. Females of *C. cosyra* were also attracted to the other treatments regardless of infestation status. Probably, compounds like δ-3-carene, myrcene, *p*-cymene, (*E*)-ocimene, (*Z*)-ocimene, α-terpinolene, *allo*-ocimene, ethyl butanoate, ethyl hexanoate, γ-octalactone, ethyl 2-methylprop-2-enoate, ethyl tiglate, phenyl ethyl alcohol, 1-octen-3-ol, and ethyl octanoate which have been associated with other fruit fly attraction [59,60] were responsible for the attraction of *C. cosyra* demonstrated in this study. In addition to the production of the new compounds, the attractiveness of *C. cosyra* to infested mangoes reported here could be attributed to the increase in release rates of the most common compounds.

The compounds ethyl butanoate, ethyl propionate, ethyl 2-methyl butanoate, ethyl-(4*E*)-decenoate, ethyl 3-methyl butanoate, and α-copaene were tentatively identified in this study. These compounds are also produced by marula, *S. birrea* fruit, found in most parts of sub-Sahara Africa and the preferred wild host of *C. cosyra* [4] and their elevated release rates following *C. cosyra* mango infestation may explain to some extent the attractiveness of the infested mango to more conspecifics. Similarly, the increase in headspace volatiles release rate by freshly-infested mangoes and the increased number of esters produced by ripe mangoes could be responsible for the increased attraction of *Fopius arisanus*. Miano et al. [13] reported similar results where *F. arisanus* was increasingly attracted to *B. dorsalis* freshly-infested mango. On the same note, *D. longicaudata* could have been attracted by the headspace volatiles of 7th-day, 9th-day and ripe mangoes because of the presence of volatiles which included ethyl 2-methyl prop-2-enoate, 2-methyl-1-butanol, ethyl propionate, methyl butanoate, 2-methyl propyl ethanoate, and ethyl, 3-hydroxy-butanoate as well as some terpenes that could be attractive. Eben et al. [61] and the references therein demonstrated how infestation levels and the volatiles produced do influence the host-seeking behavior of parasitoids. More *D. longicaudata* were attracted to *C. cosyra* 9th-day post-infestation Kent mangoes compared to those attracted to Apple and Haden mangoes. It is interesting to note that this variety is also more preferred by *C. cosyra* as indicated by the higher number of puparia recovered and this might have triggered the production of the higher number of headspace volatiles compounds shown in Figure (4 **A**). Previous studies have reported that the host density highly influences the number of parasitoids that visit the fruit fly's host [48].

Our study provides new insights into the behaviors of *C. cosyra* and its parasitoids and how mango headspace semiochemicals correlate using *in situ* experimental approaches. The systematic follow-up of the responses of this fruit fly and parasitoids towards non-infested and conspecific post-infestation mangoes, accompanied by elucidation of changes in headspace chemical profiles, lays a good foundation for future studies. *C. cosyra* and its parasitoids are generally attracted by the headspaces of non-infested and *C. cosyra*-infested mangoes. We highly recommend further studies on the olfactory responses of the insects to mango headspaces and their individually identified compounds to improve fruit fly management techniques.

6. Conclusion and further research

For the first time, we have investigated and reported on the *in situ* responses of *C. cosyra* and its parasitoids to tree-attached mangoes, supported by the performance of this fruit fly together with subsequent changes in headspace volatile composition. The attraction of *C. cosyra* to infested mangoes indicates its readiness to take advantage of existing oviposition punctures or inability to use the host-marking pheromones. While there is the suggestion that *C. cosyra* could be in the process of being displaced by *B. dorsalis,* our study demonstrates that Kent is the most preferred candidate for the former fly's performance (unlike what has been reported for the latter), which is important in advising the currently used IPM strategies. Our results also indicate notable differences in the chemical profiles of the headspaces among the mango varieties and treatments which have direct consequences on the responses of *C. cosyra* and its parasitoid. Most compounds were detected in increasing quantities as post-infestation days progressed where esters were the most prevalent compounds. This was contrary to the decrease in the quantities of monoterpenes as non-infested mangoes ripened, while those of esters increased. This calls for further studies on how individual volatiles may contribute to fruit fly and parasitoid attraction to provide an evolutionary ecological backdrop to olfactory studies and informed leads for developing selective attractants for combatting fruit fly pests and/or enhancing ecosystem services of their parasitoids.

Ethics approval and consent to participate

This was sourced from the National Commission for Science, Technology, and Innovation (NACOSTI; License No: NACOSTI/P/20/6447) of Kenya. In addition, consent for the study was granted by the mango orchard owner.

Data availability

Data will be availed by the corresponding authors on request.

CRediT authorship contribution statement

Raphael Njurai Miano: Writing – original draft, Visualization, Validation, Software, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Teun Dekker:** Writing – review & editing, Visualization, Validation, Supervision, Resources, Project administration, Funding acquisition, Conceptualization. **Egmont Rohwer:** Writing – review & editing, Visualization, Validation, Supervision. **Tibebe Dejene Biasazin:** Writing – review & editing, Visualization, Validation, Supervision, Formal analysis. **Shepard Ndlela:** Writing – review & editing, Validation, Resources. **Abdullahi Ahmed Yusuf:** Writing – review & editing, Validation, Supervision, Funding acquisition. **Xavier Cheseto:** Writing – review & editing, Visualization, Validation, Supervision. **Samira A. Mohamed:** Writing – review & editing, Visualization, Validation, Supervision, Resources, Project administration, Funding acquisition, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

This study received financial support from the Australian Centre for International Agricultural Research (ACIAR) for the project "Alien Invasive Fruit Flies in Southern Africa: Implementation of a Sustainable IPM Programme to Combat Their Menaces (grant number 109040)"; the International Development Research Centre (IDRC); the Norwegian Agency for Development Cooperation (NORAD), the Section for Research, Innovation, and Higher Education for the project "Combatting Arthropod Pests for Better Health, Food and Climate Resilience (grant number RAF-3058 KEN-18/0005)". The authors also greatly acknowledge the financial support provided by the Swiss Agency for Development and Cooperation (SDC); the Swedish International Development Cooperation Agency (Sida); the Federal Democratic Republic of Ethiopia; the Australian Centre for International Agricultural Research (ACIAR); and the Government of the Republic of Kenya. The first author (RNM) was sponsored by the collaboration support through Swedish Research Links grants 2020-05344; the NORAD (grant number RAF-3058 KEN-18/0005); and the Post Graduate Bursary Fund of the University of Pretoria.

The views expressed herein do not necessarily reflect the official opinion of the donors.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.heliyon.2024.e30068.

References

[1] S. Ekesi, S.A. Mohamed, M. De Meyer, Fruit Fly Research and Development in Africa-*Towards a Sustainable Management Strategy to Improve Horticulture*, Springer International Publishing, Cham, Switzerland, 2016, pp. 1–778, https://doi.org/10.1007/978-3-319-43226-7.

[2] W. Jaleel, L. Lu, Y. He, Biology, taxonomy, and IPM strategies of *Bactrocera* tau Walker and complex species (Diptera; Tephritidae) in Asia: a comprehensive review, Environ. Sci. Pollut. Res. 25 (2018) 19346–19361, https://doi.org/10.1007/s11356-018-2306-6.

[3] G.J. Steck, *Ceratitis cosyra* (Walker) (Diptera: Tephritidae), Fla. Dept. Agric. Consum. Serv. Div. Plant Ind. Entomol. Circ. No. 403 November/December 2000 (2000) 1–2. http://syndication.freshfromflorida.com/content/download/10788/141019/ent403.pdf.

[4] N. Gikonyo, S. Lux, P. Nemeye, Variation in volatiles from fruits of mango and marula attractive to the mango fruit fly, *Ceratitis cosyra* (Walker), East Cent. African J. Pharm. Sci. 6 (2005) 3–8, https://doi.org/10.4314/ecajps.v6i1.9691.

[5] C.W. Weldon, L. Boardman, D. Marlin, J.S. Terblanche, Physiological mechanisms of dehydration tolerance contribute to the invasion potential of *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae) relative to its less widely distributed congeners, Front. Zool. 13 (2016) 1–15, https://doi.org/10.1186/s12983-016-0147-z.

[6] S.A. Lux, S. Ekesi, S. Dimbi, M. Samira, M. Billah, Mango-infesting fruit flies in Africa: perspectives and limitations of biological approaches to their management, in: Biological Control in IPM Systems in Africa, CABI Publishing, Wallingford UK, 2003, pp. 277–293.

[7] R.S. Copeland, et al., Geographic distribution, host fruit, and parasitoids of African fruit fly pests *Ceratitis anonae, Ceratitis cosyra, Ceratitis fasciventris*, and *Ceratitis rosa* (Diptera: Tephritidae) in Kenya, Ann. Entomol. Soc. Am. 99 (2006) 261–278, https://doi.org/10.1603/0013-8746(2006)099[0261:GDHFAP]2.0.CO;2.

[8] B.N. Barnes, Monitoring and control of fruit flies in South African fruit orchards, in: Proceedings of the Indian Ocean Commission, Regional Fruit Fly Symposium, Flic en Flac, 2000, pp. 147–152. *Mauritius, 5th-9th June 2000*.

[9] S.A. Lux, R.S. Copeland, I.M. White, A. Manrakhan, M.K. Billah, A new invasive fruit fly species from the *Bactrocera dorsalis* (Hendel) group detected in East Africa, Int. J. Trop. Insect Sci. 23 (2003) 355–361, https://doi.org/10.1017/s174275840001242x.

[10] R.A.I. Drew, K. Tsuruta, I.M. White, A new species of pest fruit fly (Diptera: Tephritidae: Dacinae) from Sri Lanka and Africa, Afr. Entomol. 13 (2005) 149–154.

[11] S. Ekesi, M.K. Billah, P.W. Nderitu, S.A. Lux, I. Rwomushana, Evidence for competitive displacement of *Ceratitis cosyra* by the invasive fruit fly *Bactrocera invadens* (Diptera: Tephritidae) on mango and mechanisms contributing to the displacement, J. Econ. Entomol. 102 (2009) 981–991, https://doi.org/10.1603/029.102.0317.

[12] A.M. Akol, C. Masembe, B.E. Isabirye, C.K. Kukiriza, I. Rwomushana, Oviposition preference and offspring performance in Phytophagous fruit flies (Diptera: Tephritidae): the African Invader, *Bactrocera invadens*, Int. Res. J. Hortic. 1 (2013) 1–14, https://doi.org/10.12966/irjh.05.01.2013.

[13] R.N. Miano, S.A. Mohamed, X. Cheseto, S. Ndlela, T.D. Biasazin, A.A. Yusuf, E. Rohwer, T. Dekker, Differential responses of *Bactrocera dorsalis* and its parasitoids to headspaces of different varieties of tree-attached mango fruits and the associated chemical profiles, Front. Ecol. (2022) 1–21, https://doi.org/10.3389/fevo.2022.1021795.

[14] S. Adebayo, R.R. Uddinii, A. Mubarak, Mango farmers' perception on the effect of fruitflies infestation, Int. J. Agric. Sci. 11 (2021) 193–200.

[15] C. Akotsen-Mensah, et al., Pest management knowledge and practices of mango farmers in Southeastern Ghana, J. Integr. Pest Manag. 8 (2017) 13, https://doi.org/10.1093/jipm/pmx008.

[16] S. Dimbi, N.K. Maniania, S. Ekesi, Horizontal transmission of *Metarhizium anisopliae* in fruit flies and effect of fungal infection on egg laying and fertility, Insects 4 (2013) 206–216, https://doi.org/10.3390/insects4020206.

[17] S.A. Mohamed, W.A. Overholt, R.A. Wharton, S.A. Lux, E.M. Eltoum, Host specificity of *Psyttalia cosyrae* (Hymenoptera: Braconidae) and the effect of different host species on parasitoid fitness, Biol. Control 28 (2003) 155–163, https://doi.org/10.1016/S1049-9644(03)00099-9.

[18] S. Ekesi, S.A. Mohamed, M. De Meyer, In and out of Africa: parasitoids used for biological control of fruit flies-Towards a sustainable management strategy to improve horticulture. https://doi.org/10.1007/978-3-319-43226-7, 2017.

[19] S. Ndlela, S.A. Mohamed, A.G.A. Azrag, P.N. Ndegwa, G.O. Ong'amo, S. Ekesi, Interactions between two parasitoids of Tephritidae: *Diachasmimorpha longicaudata* (Ashmead) and *Psyttalia cosyrae* (Wilkinson) (Hymenoptera: Braconidae), under laboratory conditions, Insects 11 (2020) 1–16, https://doi.org/10.3390/insects11100671.

[20] S.A. Mohamed, S. Ekesi, R. Hanna, Evaluation of the impact of *Diachasmimorpha longicaudata* on *Bactrocera invadens* and five African fruit fly species, J. Appl. Entomol. 132 (2008) 789–797, https://doi.org/10.1111/j.1439-0418.2008.01350.x.

[21] X. Cheseto, D.L. Kachigamba, S. Ekesi, M. Ndung'u, P.E.A. Teal, J.J. Beck, B. Torto, Identification of the ubiquitous antioxidant tripeptide glutathione as a fruit fly semiochemical, J. Agric. Food Chem. 65 (2017) 8560–8568, https://doi.org/10.1021/acs.jafc.7b03164.

[22] X. Cheseto, H. Kirwa, S.A. Mohamed, S. Ekesi, J.J. Beck, B. Torto, Field evaluation of glutathione and glutamic acid as host marking pheromones for control of Tephritid fruit flies in a mango orchard in Kenya, Pest Manag. Sci. (2023), https://doi.org/10.1002/ps.7331.

[23] P.D.K. Jayanthi, V. Kempraj, R.M. Aurade, T. Kumar Roy, S. KS, A. Verghese, Computational reverse chemical ecology: virtual screening and predicting behaviorally active semiochemicals for *Bactrocera dorsalis*, BMC Genom. 15 (2014) 1–7, https://doi.org/10.1186/1471-2164-15-209.

[24] N.M.A. El-ghany, Semiochemicals for controlling insect pests, J. Plant Protect. Res. 59 (2019) 1–11, https://doi.org/10.24425/jppr.2019.126036.

[25] J.P. Cunningham, M.A. Carlsson, T.F. Villa, T. Dekker, A.R. Clarke, Do fruit ripening volatiles enable resource specialism in polyphagous fruit flies? J. Chem. Ecol. 42 (2016) 931–940, https://doi.org/10.1007/s10886-016-0752-5.

[26] T.D. Biasazin, S.L. Herrera, F. Kimbokota, T. Dekker, Translating olfactomes into attractants: shared volatiles provide attractive bridges for polyphagy in fruit flies, Ecol. Lett. 22 (2019) 108–118, https://doi.org/10.1111/ele.13172.

[27] X. Wang, R.H. Messing, Foraging behavior and patch time allocation by *Fopius arisanus* (Hymenoptera : Braconidae), an egg-larval parasitoid of Tephritid fruit flies, J. Insect Behav. 16 (2003) 593–612.

[28] A. Harbi, L. De Pedro, F.A.A. Ferrara, J. Tormos, B. Chermiti, F. Beitia, B. Sabater-Munoz, *Diachasmimorpha longicaudata* parasitism response to medfly host fruit and fruit infestation age, Insects 10 (2019) 1–12, https://doi.org/10.3390/insects10070211.

[29] P. Cai, Y. Song, D. Huo, J. Lin, H. Zhang, Z. Zhang, Chemical cues induced from fly-oviposition mediate the host-seeking behaviour of an effective egg parasitoid of *Bactrocera dorsalis* (Diptera: Tephritidae), within a tritrophic context, Insects 11 (2020) 231.

[30] H. van Den Dool, P.D. Kratz, A generalization of the retention index system including linear temperature programmed gas-liquid partition chromatography, J. Chromatogr. (1963) 463–471.

[31] R.P. Adams, Identification of Essential Oil Components by Gas Chromatography/mass Spectroscopy, Allured Publishing Corporation 362 South Schmale Road Carol Stream, Illinois 60188-2787 USA, 1996.

[32] P.K. Njuguna, L.K. Murungi, A. Fombong, P.E.A. Teal, J.J. Beck, B. Torto, Cucumber and tomato volatiles: influence on attraction in the melon fly *Zeugodacus cucurbitae* (Diptera: Tephritidae), J. Agric. Food Chem. 66 (2018) 8504–8513, https://doi.org/10.1021/acs.jafc.8b03452.

[33] R.N. Miano, P.M. Ayelo, R. Musau, A. Hassanali, S.A. Mohamed, Electroantennogram and machine learning reveal a volatile blend mediating avoidance behavior by *Tuta absoluta* females to a wild tomato plant, Sci. Rep. 12 (2022) 1–16, https://doi.org/10.1038/s41598-022-13125-0.

[34] V.O. Nyasembe, P.E.A. Teal, W.R. Mukabana, J.H. Tumlinson, B. Torto, Behavioural response of the malaria vector *Anopheles gambiae* to host plant volatiles and synthetic blends, Parasites Vectors 5 (2012) 1–11.

[35] RStudio Team, RStudio: Integrated Development for R; RStudio, PBC, Boston, MA, USA, 2020, p. 2021.

[36] A. Dinno, Nonparametric pairwise multiple comparisons in independent groups using Dunn's test, STATA J. 15 (2015) 292–300, https://doi.org/10.1177/1536867X1501500117.

[37] F. Rohart, B. Gautier, A. Singh, K.A. Lê Cao, mixOmics: an R package for 'omics feature selection and multiple data integration, PLoS Comput. Biol. 13 (2017) 1–19, https://doi.org/10.1371/journal.pcbi.1005752.

[38] D. Hammer, D.A.T. Harper, P.D. Ryan, PAST: paleontological statistics software package for education and data analysis, Palaeontol. Electron. (2001) 1–9.

[39] S. Ekesi, M.K. Billah, A Field Guide to the Management of Economically Important Tephritid Fruit Flies in Africa, ICIPE Science Press, Kenya, 2006, p. 145.

[40] M. Kibira, H. Affognon, B. Njehia, B. Muriithi, S.A. Mohamed, S. Ekesi, Economic evaluation of integrated management of fruit fly in mango production in Embu County, Kenya, African J. Agric. Resour. Econ. 10 (2015) 343–353.

[41] D.R. Papaj, A.L. Averill, R.J. Prokopy, T.T.Y. Wong, Host-marking pheromone and use of previously established oviposition sites by the Mediterranean fruit fly (Diptera: Tephritidae), J. Insect Behav. 5 (1992) 583–598, https://doi.org/10.1007/BF01048006.

[42] M. Binyameen, A. Hamid, I. Afzal, M. Sajjad, M. Azeem, S. Muhammad, Z. Zahid, M. Sarwar, S. Ali, S. Thomas, C.B. Fredrik, Role of fruit volatiles of different guava varieties in attraction and oviposition behaviors of peach fruit fly, Bactrocera zonata Saunders, Arthropod. Plant. Interact. 15 (2021) 95–106, https://doi.org/10.1007/s11829-020-09796-z.

[43] R. Silva, A.R. Clarke, Aversive responses of Queensland fruit flies towards larval-infested fruits are modified by fruit quality and prior experience, J. Insect Physiol. 131 (2021) 104231, https://doi.org/10.1016/j.jinsphys.2021.104231.

[44] J. Pérez, J.C. Rojas, P. Montoya, P. Liedo, A. Castillo, Anastrepha egg deposition induces volatiles in fruits that attract the parasitoid *Fopius arisanus*, Bull. Entomol. Res. 103 (2013) 318–325, https://doi.org/10.1017/S0007485312000739.

[45] R. Moretti, M. Calvitti, Mortality by parasitization in the association between the egg-pupal parasitoid *Fopius arisanus* and *Ceratitis capitata*, BioControl 48 (2003) 275–291, https://doi.org/10.1023/A:1023610421270.

[46] M.F. Karlsson, E.O. de Souza, P.M. Ayelo, J.A. Zannou, G.S.B. Mègnigbèto, A.H. Bokonon-Ganta, Interspecific competition between egg parasitoids: native *Fopius caudatus* and exotic *Fopius arisanus*, in *Ceratitis cosyra*, Biol. Control 117 (2018) 172–181, https://doi.org/10.1016/j.biocontrol.2017.11.010.

[47] A. Altuzar, P. Montoya, J.C. Rojas, Response of *Fopius arisanus* (Hymenoptera: Braconidae) to fruit volatiles in a wind tunnel, Fla. Entomol. 87 (2004) 616–618, https://doi.org/10.1653/0015-4040(2004)087[0616:ROFAHB]2.0.CO;2.

[48] S.R. Nunez-Campero, S. Benitez-Vieyra, D.E. Gorla, S.M. Ovruski, Ecology and population biology changes in *Diachasmimorpha longicaudata* (Hymenoptera: Braconidae) functional response as a consequence of host density choice, Ann. Entomol. Soc. Am. (2016) 1–7, https://doi.org/10.1093/aesa/saw045.

[49] S. Gripenberg, P.J. Mayhew, M. Parnell, T. Roslin, A meta-analysis of preference-performance relationships in phytophagous insects, Ecol. Lett. 13 (2010) 383–393, https://doi.org/10.1111/j.1461-0248.2009.01433.x.

[50] D. Carrillo, A. Birke, L. Guillen, J.E. Peña, Pests of Mango, Handbook of Mango Fruit: Production, Postharvest Science, Processing Technology and Nutrition, John Wiley & Sons Ltd, 2017, pp. 61–90.

[51] P. Diatta, J.Y. Rey, J.F. Vayssieres, K. Diarra, E.V. Coly, M. Lechaudel, I. Grechi, S. Ndiaye, O. Ndiaye, Fruit phenology of citruses, mangoes and papayas influences egg-laying preferences of *Bactrocera invadens* (Diptera: Tephritidae), Fruits 68 (2013) 507–516, https://doi.org/10.1051/fruits/2013093.

[52] B. Gonçalves, I. Oliveira, E. Bacelar, M.C. Morais, A. Aires, F. Cosme, J. Ventura-Cardoso, R. Anjos, T. Pinto, Aromas and flavours of fruits, Intech (2016) 9–31, https://doi.org/10.5772/57353.
[53] K. Shimizu, T. Matsukawa, R. Kanematsu, K. Itoh, S. Kanzaki, S. Shigeoka, S. Kajiyama, V. Pride, Volatile profiling of fruits of 17 mango cultivars by HS-SPME-GC/MS combined with principal component analysis, Biosci. Biotechnol. Biochem. 85 (2021) 1789–1797, https://doi.org/10.1093/bbb/zbab097.
[54] S. Nair, Z. Singh, S.C. Tan, Aroma volatiles emission in relation to chilling injury in 'Kensington Pride' mango fruit, J. Hortic. Sci. Biotechnol. (2015) 866–873, https://doi.org/10.1080/14620316.2003.11511711. ISSN. 0316.
[55] A. Rodríguez, B. Alquézar, L. Peña, Fruit aromas in mature fleshy fruits as signals of readiness for predation and seed dispersal, New Phytol. 197 (2013) 36–48, https://doi.org/10.1111/j.1469-8137.2012.04382.x.
[56] W. Jaleel, R. Saeed, M.Z. Shabbir, R. Azad, S. Ali, M.U. Sial, D.M. Aljedani, H.A. Ghramh, K.A. Khan, D. Wang, Y. He, Olfactory response of two different Bactrocera fruit flies (Diptera: Tephritidae) on banana, guava, and mango fruits, J. King Saud Univ. Sci. 33 (2021) 1–7, https://doi.org/10.1016/j.jksus.2021.101455.
[57] A.R. War, M.G. Paulraj, T. Ahmad, A.A. Buhroo, B. Hussain, S. Ignacimuthu, H.C. Sharma, Mechanisms of plant defense against insect herbivores, Plant Signal. Behav. 7 (2012) 1306–1320.
[58] S. Shivaramu, P.D.K. Jayanthi, V. Kempraj, R. Anjinappa, B. Nandagopal, A.K. Chakravarty, What signals do herbivore-induced plant volatiles provide conspecific herbivores? Arthropod. Plant. Interact. 11 (2017) 815–823, https://doi.org/10.1007/s11829-017-9536-2.
[59] P.D.K. Jayanthi, C.M. Woodcock, J. Caulfield, M.A. Birkett, T.J. Bruce, Isolation and identification of host cues from mango, *Mangifera indica*, that attract gravid female oriental fruit fly, *Bactrocera dorsalis*, J. Chem. Ecol. 38 (2012) 361–369, https://doi.org/10.1007/s10886-012-0093-y.
[60] T.D. Biasazin, S. Larsson Herrera, F. Kimbokota, T. Dekker, Translating olfactomes into attractants: shared volatiles provide attractive bridges for polyphagy in fruit flies, Ecol. Lett. 22 (2019) 108–118, https://doi.org/10.1111/ele.13172.
[61] A. Eben, B. Benrey, J. Sivinski, M. Aluja, Host species and host plant effects on preference and performance of *Diachasmimorpha longicaudata* (Hymenoptera: Braconidae),", Environ. Entomol. 29 (2000) 87–94, https://doi.org/10.1603/0046-225x-29.1.87.