African Entomology 2023, 31: e14027 (8 pages)
https://doi.org/10.17159/2254-8854/2023/a14027
RESEARCH ARTICLE

AFRICAN ENTOMOLOGY
ISSN (online) 2224-8854

T. V. PheroLure®: Volatile emission by semiochemical lures and the impact thereof on the volatile profile of a commercial tomato field

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Pheromone-based or semiochemical lures for insect detection and monitoring in agriculture is common practice. Many countries exempt these devices from regulatory requirements, but not South Africa. The question arises whether the pheromone/semiochemical lures influence the naturally occurring compounds significantly, to justify concern for human toxicity and ecotoxicity. T.V. PheroLure® is a novel five-component lure developed by Insect Science (Pty) Ltd. used for monitoring African bollworm, *Helicoverpa armigera* (an important insect pest on tomatoes). T.V. PheroLure® is a volatile organic compound (VOC) blend impregnated in a polyethylene bulb. The influence of T.V. PheroLure® on the volatile profile of a tomato field was evaluated in a commercial growing area of South Africa. Tomato VOCs were collected before, during and after the application of six T.V. PheroLures® in yellow bucket funnel traps randomly distributed over 1 ha. VOCs were collected from planting until harvest (22 weeks) at five randomly selected sites. Collection also took place in adjacent tomato fields where no T.V. PheroLure® was applied. The constituents of T.V. PheroLure® had no significant influence on the naturally occurring VOCs observed in the tomato field. The results suggest that the concern for toxicity and ecotoxicity is unjustified when using semiochemical devices for monitoring purposes. The natural physiology of the plant, rather than T.V. PheroLure®, influenced the VOCs observed in a tomato field.

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DATES
Received: 31 May 2022
Accepted: 08 November 2022

KEYWORDS
insect monitoring
toxicity
South Africa
volatile organic compound
VOC

INTRODUCTION

The world is moving ever increasingly towards environmentally friendly approaches in agriculture when it comes to crop protection products (Vurro et al. 2019). Using semiochemicals for monitoring, suppression and control of insect pests will become even more prevalent in the near future. For South Africa to remain relevant as an agricultural export country, it is essential that research concerning semiochemical-based products be conducted where necessary. South Africa currently has more stringent regulations concerning the registration of semiochemical-based agricultural remedies. Even more stringent than some other countries, including the European Union and United States of America in some regards (LII 2004; DAFF 2015a, b; OECD 2018).

The use of pheromone-based or semiochemical lures and devices for detection of insect pest populations and monitoring in agriculture is a common practice (Witzgall et al. 2010). Pheromone- or semiochemical-based lures used for detection and population monitoring of insects in agriculture, are classified as agricultural remedies and are therefore regulated by South African Law under Act No. 36 of 1947 (DAFF 2015a, b). The registration of these agricultural remedies are required in order to sell and use these lures and devices. One of the concerns from the South African registration authority, the Department of Agriculture, Forestry and Fisheries (DAFF) is that the emittance of these biological products will influence the natural background volatile organic compound (VOC) presence. The current timeline set for the registration of agricultural remedies, is 418–627 working days, which is between one and a half to three years (DAFF 2015a). If semiochemical-based agricultural remedies are exempt from toxicological and ecotoxicological requirements, the rate of the registration process could increase significantly.

The requirements for the registration of an agricultural remedy in South Africa include that toxicity and ecotoxicity studies be conducted (DAFF 2015b), which is a challenge for semiochemical-based agricultural remedy registrations. Most VOCs used in lures are complex and, in most cases, considered to be of no concern to human and environmental health due to the method of application and because they are not found to exceed naturally occurring concentrations (OECD 2003, 2018). These devices are usually seen as products with non-toxic, target-specific modes of action, and active ingredients that occur naturally (OECD 2003, 2018).

Plant volatiles or VOCs play an important role in plant-plant interactions, plant-pest interactions and has a major impact on atmospheric chemistry (Tholl et al. 2006; Baldwin 2010; Holopainen and Blande 2012). VOCs can impact plant-pest interactions in various ways, inter alia, attracting or repelling a pest or influencing the behaviour of specific pests (Baldwin 2010; Witzgall et al. 2010; Holopainen and Blande 2012; Frérot et al. 2017). Utilised correctly within a semiochemical-based product, VOCs can play a vital role in Integrated Pest Management (IPM) programmes (Baldwin 2010; Witzgall et al. 2010; Holopainen and Blande 2012).

Tomatoes are one of the most produced crops in the world and South Africa's annual tomato production is estimated around 600 000 tonnes (PHI 2020). African bollworm (ABW) [*Helicoverpa*

armigera Hubner (Lepidoptera: Noctuidae)], seen as a major pest on this economically important crop, could be considered one of the world's most destructive crop pests (Ravi et al. 2005; Prinsloo and Uys 2015; Pinto et al. 2017).

Researchers have reported more than 400 VOCs in the ripening of the tomato fruit including limonene, phenethyl alcohol, phenyl-acetaldehyde and methyl salicylate, four of the components used in the Insect Science (Pty) Ltd. T.V. PheroLure® (Pyne and Wick 1965; Dalal et al. 1967; Viani et al. 1969; Buttery et al. 1971; Petro-Turza 1986; Buttery et al. 1987; Baldwin et al. 2000; Tikunov et al. 2005; Beltran et al. 2006; Mayer et al. 2008; Baldwin 2010; Wang et al. 2016; Wang et al. 2018). Literature pertaining to the presence of methyl 2-methoxybenzoate, the fifth component, as a VOC in tomatoes is lacking. Of these VOCs identified, it has been found that less than 10% play a significant role in the aromas of the tomatoes (Du et al. 2015; Wang et al. 2016; Wang et al. 2018). However, in South Africa studies specifically on these VOCs in tomatoes have not been done. Identification of background concentrations for specific VOCs are very difficult to determine and therefore natural background data is usually not available (OECD 2017). For VOCs, this may be more complex as several factors contribute to the emission of VOCs. Factors that influence VOC emissions may include, but are not limited to weather conditions, maintenance to the plant and spray programmes (OECD 2017).

The aim of this study was to determine the influence of current VOCs in tomato fields when VOCs were introduced with the T.V. PheroLure®, a novel five-component lure, under commercial tomato production conditions. Specifically, to demonstrate whether the influence of plant volatiles released into the field is significant or not.

METHODS and MATERIALS

Study design and area

A qualitative research design was used to achieve the research aim by determining the background Volatile Organic Compound (VOC) presence in a commercial tomato field and the influence of the VOCs emitted by the T.V. PheroLure® thereon.

Sampling of VOCs in this study has been done in parallel with an agricultural remedy registration trial over and above the chemical spray programme used in the field. An agricultural remedy registration trial needs to be done according to the relevant DAFF guidelines (DAFF 2015a, b). It is required that these trials should have at least four replicates in order to have a meaningful statistical difference, and that the error degrees of freedom (df) should be 12 or more (DAFF 2015b), that is, df = $(t-1)(r-1) \geq 12$ where t, is the number of treatments, and r is the number of replicates. If similar products are already available on the market, it is required that the new agricultural remedy must be compared to the commercially available remedy or remedies (DAFF 2015a, b). Therefore, the trial in this study consisted of five different treatments (Table 1) with six replicates on a one hectare site (one treatment every 20 m and every third row) following a completely randomised design (CRD) (Insect Science 2018). The treatments in each replicate were then rotated on a weekly basis.

Table 1. Five different treatments used in the registration trial

	Treatments
1	T.V. PheroLure® (2000 mg) – VOC lure
2	ABW PheroLure® (1 mg) – Pheromone lure
3	ABW PheroLure® with T.V. PheroLure® (1 mg + 2000 mg) – VOC and pheromone lure combined
4	ABW lure (Chempac (Pty) Ltd) (3.5 mg) – Pheromone lure
5	Control – No lure

This study only focused on compounds of one product, namely, T.V. PheroLure® from Insect Science, in a single crop namely tomatoes (*Solanum lycopersicum*).

Within this CRD for the agricultural remedy registration trial, five random sites were selected for sampling the volatile content in the air as shown in Figure 1.

Figure 1 indicates the trial layout as well as the random sampling site selection. The study was conducted on a ZZ2® commercial tomato farm block Rivierland 4B (GPS: 23°32′29.2″S 30°14′13.8″E), Jachtpad, Mooketsi in the Limpopo province, Republic of South Africa.

Sample collection

Sampling at the random five sampling points (Figure 1) was done by the adsorption of the compounds on sorbent tubes (Tenax® TA) which were later thermally desorbed, with a Thermal desorber (TD) and analysed using Gas chromatography-Mass spectrometry (GC-MS). Tenax® TA stainless steel tubes (dimensions 89 mm × 4.5 mm inner diameter × 6.5 mm outer diameter and packed with 197 mg of a sorbent) were used for the VOC collection (Joubert 2011; Marks 2014). Tenax® TA has a compound range of n-$C_{6/7}$ to n-C_{30} with a high volatility range of 100 °C up to 450 °C, making it thermally stable (Joubert 2011; Marks 2014). Other reasons why the Tenax® TA tubes were selected for the analyses were because the tubes are hydrophobic, making it more suitable for very humid conditions and the material can be conditioned to give very low background signals, smaller than 1 ng/compound (Joubert 2011; Marks 2014). The above-mentioned properties allowed for the sampling of volatile and semi-volatile compounds (Joubert 2011). Tenax® TA tubes were received conditioned and capped from Chemetrix (Pty) Ltd which is the distributor for Markes International in South Africa (Marks 2014).

The VOCs were adsorbed on the Tenax® TA tubes by using a BioVOC Breath Sampler (Markes International in South Africa), which is a kind of syringe (displacement volume of 0.130 l), hereafter referred to as the syringe. VOCs were adsorbed on the tube when the screw-in plunger was pulled out to fill the syringe. Once the syringe had been filled, the tube was removed, and the screw-in plunger was used to discharge the remaining air in the syringe.

Sampling was done by adsorbing the air content sampled with the syringe onto stainless steel Tenax® TA tubes. Thermal desorption was then utilised, and the desorbed VOCs were

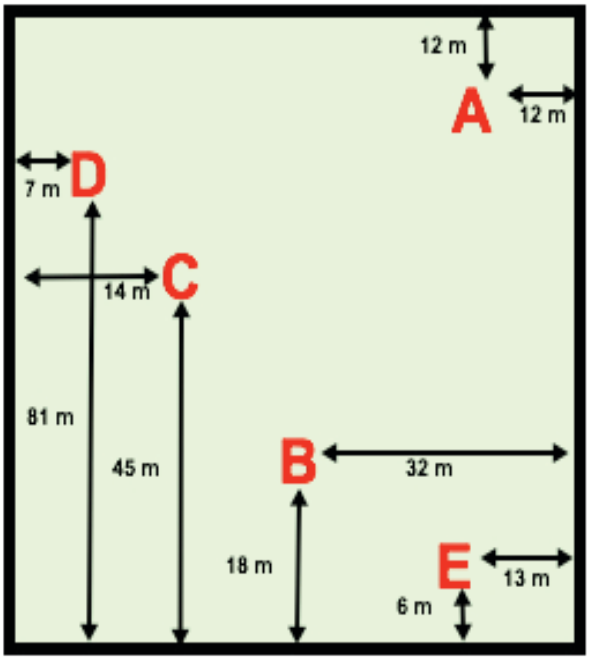

Figure 1. Random site selection for sampling of volatiles

transferred to the GC-MS column for analysis using MSD ChemStation D.03.00.611 software (Agilent Technology, California, USA) and the NIST05 mass spectral library was used for identification of the compounds.

Objective (i): Samples of the natural air were taken in the tomato field before any loaded traps were placed in the tomato fields, as well as throughout the trial. Samples were taken at the randomly selected sites within the 1 ha block (Figures 1 and 2). The aforementioned samples were used as the initial control samples in this experiment. The pre-determined 90 syringe samples were taken per Tenax® TA tube at a sampling site for the initial assessment.

Objective (ii): VOC sampling of the selected tomato field with the loaded traps were done following Objective (i). The VOCs were sampled by adsorbing 90 syringes of air from the designated sampling sites on the tomato field onto stainless steel Tenax® TA tubes (Joubert 2011). Samples were collected in the field by sampling 90 syringes of air at the opening in the T.V. PheroLure®-loaded Yellow Bucket Funnel Trap® followed by the thermal desorption of the Tenax® TA tubes, after which time the desorbed VOCs was transferred to the GC-MS column for analysis.

This procedure was repeated once a week on the same day for the first seven weeks of the trial. Thereafter monitoring was done each fortnight up to week 15. The final three samples were taken at week 20, week 21 (one week after loaded traps were removed) and week 22 at harvest time, before the tomato fields were reworked. Table 2 shows the sampling schedule of the VOCs in the tomato field for the duration of the trial.

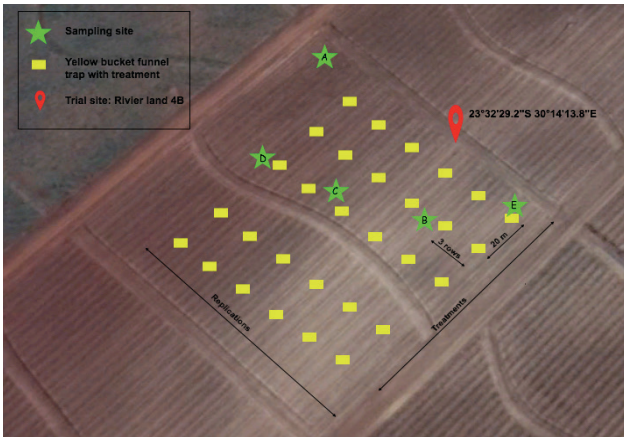

Figure 2. Registration trial layout with random sampling points (Google Maps 2018)

Table 2. Sampling schedule of the VOCs in the tomato field throughout the duration of the trial

Week of sampling	Sample
0	1 (Control sample – Before traps were put in the field)
1	2
2	3
3	4
4	5
5	6
6	7
7	8
9	9
11	10
13	11
15	12
20	13
21	14 (One week after loaded traps have been removed).
22	15 (Harvest of crop/before land is reworked)

Objective (iii): Following the conclusion of the trial, all data was summarised and statistically analysed in which the VOCs emitted by the tomato plants was compared against the VOCs emitted by the loaded traps over the 20-week lifespan of the T.V. PheroLure®. These results were also compared to the samples taken at week 21 at which time all the loaded traps had been removed from the field.

Objective (iv): The final analysis was done to compare the VOCs present before (samples 1–10) and during harvest (samples 11–15). At sample 15 the tomato fruit had been harvested (See Table 2).

This analysis was initially expected to be used to determine the natural background VOCs in the tomato field. However, the five specific compounds of T.V. PheroLure® were not detected in the 15 syringe samples. Therefore, the number of syringes used per sampling was increased to 30 and later 90 syringe samples. Ninety syringes ensured that the specific VOCs could be detected in low levels and that the sorbent material was not saturated, which ensured no chromatographic errors. This was done to determine the natural background VOCs in the tomato field using the TD-GC-MS and the data was used to determine the release of the specific five compounds of the T.V. PheroLure®. This was done at each sampling point of the tomato plants, as well as random samples throughout the trial in the surrounding fields (blank samples).

Samples analysis

Thermal desorption

A Unity desorption unit (TDU) (Markes International Ltd., Pontyclun, UK), feeding into the injection port of the GC, was utilised for the TD of the sampled VOCs retained on the packing material in the sample tube. The TDU was controlled by Unity Thermal Desorption System Control Software, version 2.0. The TD was pre-purged with helium for 2 minutes after which time the VOCs were desorbed from the tubes at 280 °C for 10 min at a flow rate of 30 ml min^{-1} splitlessly. Thereafter it was cryo-focused on a general-purpose hydrophobic cold trap (C4/5 to C30/32) at –10 °C, with a split ratio of 4.6:1. The trap was heated to 300 °C at a rate of 100 °C.s^{-1} and kept at the maximum temperature for 3 minutes. The desorbed volatiles were transferred to the GC column through a heated fused silica line at 190 °C.

GC-MS Analysis

GC-MS analysis was performed on an Agilent Technology (Little Falls, California, USA) 6890 series GC system, equipped with an Agilent 5973 MS detector coupled to the TDU. The respective peaks were recorded and integrated using MSD ChemStation D.03.00.611 software (Agilent Technology, California, USA). Volatilised compounds were separated with a Zebron ZB-5 fused silica capillary column (5% phenyl-dimethylpolysiloxane, 30 m × 0.25 mm inner diameter × 0.25 µm film thickness) (Phenomenex, Torrance, California, USA) and detected with the MS detector set at 300 °C. Helium was used as the carrier gas and the column flow was measured as 4.2 ml min^{-1}, at 40 °C at a constant pressure of 15 psi. The oven temperature was programmed to start at 40 °C and increased to 70 °C at a rate of 2.5 °C min^{-1} and held at this temperature for 5 minutes. A second ramp consisted of an increase to 154 °C at a rate of 6 °C min^{-1}, where it was held for 1 minute, followed by an increase to 250 °C at 10 °C min^{-1}, held for 2 minutes. Finally, the oven temperature was increased to 300 °C at 10 °C min^{-1} and held for 5 minutes.

Statistical analysis

The collected VOC data were analysed using soft independent modelling of class analogy (SIMCA), a multivariate statistical analysis (MVA). SIMCA was used to perform a principal component analysis (PCA) to determine significant differences

between the different data profiles and an orthogonal projection to latent structures-discriminant analysis (OPLS-DA) to determine differences between two specific groups.

RESULTS and DISCUSSION

Resulting chromatographic data was filtered by eliminating irrelevant compounds based on the following criteria: (i) Identification using the NIST05 library; (ii) with a certainty of less than 60%; (iii) a retention time (RT) less than 5 min to eliminate volatiles associated with laboratory gasses (< 5 minutes); (iv) chemicals associated with the chemical spray programme the farmer used and (v) compounds associated with the tar poles used in the tomato fields. The final result of the elimination process concluded that 72 VOCs remained for interpretation and analysis.

The filtered chromatographic data was then exported to a Microsoft Excel spreadsheet and analysed using soft independent modelling of class analogy (SIMCA) which is a multivariate statistical analysis (MVA). A principal component analysis (PCA) model was initially constructed from the chromatographic data, to enable the identification of clusters, groups and outliers (Sandasi et al. 2011). The PCA model displayed a very small variation between collected data (R^2_{cum} = 0.151 and Q^2_{cum} = 0.022 for a two principal component model). The scores scatter plot for the first two principal components (Figure 3) demonstrates that the data points are loosely clustered together with a few outliers identified. This grouping and separation of data points indicate that there are chemical compositional differences between these groups.

Orthogonal projections to latent structures-discriminant analysis (OPLS-DA) is a regression technique that improves the separation obtained by rotating PCA-components (Wishart 2008). OPLS-DA can also be used to identify variables that are responsible for class discrimination (Bylesjö et al. 2006; Mehl et al. 2014).

The PCA scores plot of all filtered chromatographic data obtained, indicated that the data clustered together with only a separation occurring at two instances in the tomato field at, Week 0 (blue circled) and Week 5 and 6 (orange circled) (Figure 3). Possible explanations for these observations include the physiology of the plant and environmental change. These explanations are discussed in more detail in the conclusion.

Determination of the natural background VOCs present in the tomato field utilising GC-MS.

The natural VOCs present in a tomato field were determined by taking samples at Week 0, before traps were put in the field, as well as throughout the trial in the surrounding field (blank samples). Table 3 indicates the VOCs identified by the NIST05 library as the natural VOCs present in a commercial tomato field (Rivierland 4B) at a NIST05 library certainty of more than 60%. The specific T.V. PheroLure® VOCs are marked red.

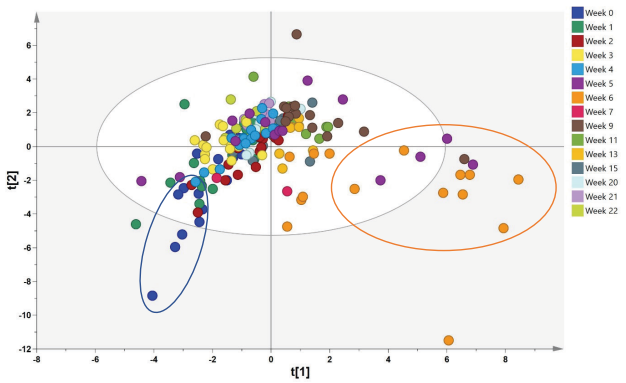

Figure 3. PCA scores scatter plot with VOC data collected weekly for the duration of the trial, indicating VOC profile differences observed at Week 0 (blue circled) and Week 5 and 6 (orange circled)

Loadings plots (Figure 4 Part A) derived from the OPLS-DA model display the differences in the *y*-variables (orthogonal), in this case the separated volatile compounds, in relation to each other. This type of plot identifies compounds with similar information in relation to the *x*-variables (predictive), sampling dates and sites. The compounds represented in the loadings plot are arranged in ascending order. The compounds displayed on the left side of the plot are associated with week 0 samples and the compounds on the right are associated with blank samples.

As shown in Figure 4 Part B and C, the plots indicate that the background VOCs that are associated with week 0 samples include: nonanoic acid, 2-octanone, n-decanoic acid and phenol. The compounds associated with blank samples include: isopropyl myristate, 1-nonadecene, 1-hexadecanol and farnesyl acetaldehyde. Similar results were also reported by two independent research groups (Wang et al. 2016; Wang et al. 2018). The red highlighted columns indicate the specific T.V. PheroLure® compounds. From Figure 4 Part A it can be seen that the contribution of these compounds, to the separation based on total volatiles, are very small and negligible.

Determination of the VOCs emitted into the atmosphere by T.V. PheroLure® utilising GC-MS.

T.V. PheroLure® consists of five specific VOCs (limonene, methyl 2-methoxybenzoate, phenethyl alcohol, phenylacetaldehyde and methyl salicylate). When the T.V. PheroLure® loaded Yellow Bucket Funnel Traps® (YBFT) were sampled, only methyl salicylate was detected at levels greater than 1% of the total sample volume. Therefore, the VOCs released from the T.V. PheroLure® is in such low concentrations that the method used in this study, could not detect these compounds. T.V. PheroLure® loaded YBFT data was collected in the field by sampling ninety syringes of air at the opening of the loaded YBFT (Figure 5). The VOCs detected from the selected sampling sites (A–E, Figure 1), corresponded with the natural background VOCs reported in Table 3, for which similar results were also reported in other studies (Wang et al. 2016, Wang et al. 2018).

The VOCs detected and identified with the NIST05 library (certainty of >60 %) at levels greater than 1% of total sample volume when sampling T.V. PheroLure® loaded YBFT are shown in Table 4.

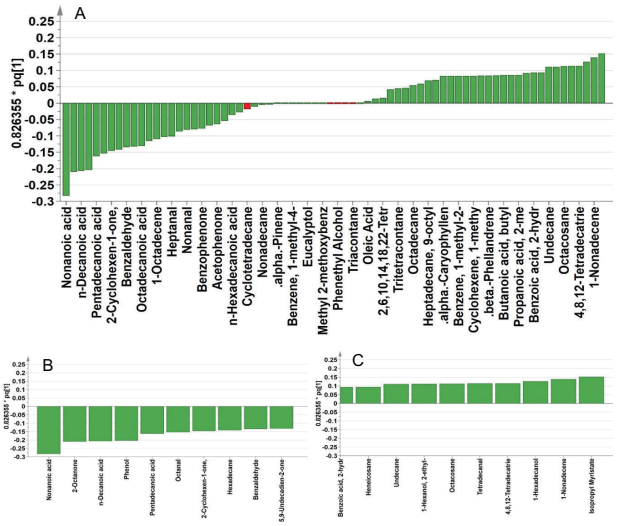

Figure 4. Part A: OPLS-DA loadings plot for chromatographic data of Week 0 compared to blank samples. Part B: enlargement of ten compounds on left (week 0 variables) and Part C: ten compounds on right (blank variables). (*Red indicates five specific VOC compounds of T.V. PheroLure®)

Table 3. Natural background VOCs identified in the commercial tomato field (Rivierland 4B).

1	alpha-Caryophyllene	37	Heptadecane
2	alpha-Phellandrene	38	Heptadecane, 9-octyl-
3	alpha-Pinene	39	Heptanal
4	beta-Phellandrene	40	Hexacosane
5	(+)-4-Carene	41	Hexadecanal
6	1-Docosene	42	Hexadecane
7	1-Hexadecanol	43	Hexanoic acid
8	1-Hexadecene	44	Hexatriacontane
9	1-Hexanol, 2-ethyl-	45	Isopropyl myristate
10	1-Nonadecanol	46	Limonene
11	1-Nonadecene	47	Methyl 2-methoxybenzoate
12	1-Octadecanol	48	Methyl salicylate
13	1-Octadecene	49	n-Decanoic acid
14	2-Cyclohexen-1-one, 2-methyl-5-(1-methylethenyl)-, l-	50	n-Hexadecanoic acid
15	2-Octanone	51	Nonadecane
16	2,6,10,14,18,22-Tetracosahexaene, 2,6,10,15,19,23-hexamethyl-, (all-***E***)-	52	Nonanal
17	4,8,12-Tetradecatrienal, 5,9,13-trimethyl- (Farnesyl acetaldehyde)	53	Nonanoic acid
18	5,9-Undecadien-2-one, 6,10-dimethyl-	54	Octacosane
19	Acetophenone	55	Octadecane
20	Benzaldehyde	56	Octadecanoic acid
21	Benzene, 1-methyl-2-(1-methylethyl)-	57	Octanal
22	Benzene, 1-methyl-4-(1-methylethyl)-	58	Oleic acid
23	Benzoic acid, 2-hydroxy-, 3-methylbutyl ester	59	Pentadecanoic acid
24	Benzophenone	60	Phenethyl alcohol
25	Butanoic acid, butyl ester	61	Phenol
26	Caryophyllene	62	Phenylacetaldehyde
27	Cyclohexene, 1-methyl-4-(1-methylethylidene)-	63	Propanoic acid, 2-methyl-, 2-ethyl-3-hydroxyhexyl ester
28	Cyclotetradecane	64	Squalene
29	Decanal	65	Tetracosane
30	Docosane	66	Tetradecanal
31	Dotriacontane	67	Tetradecanoic acid
32	***E***-14-Hexadecenal	68	Triacontane
33	Eicosane	69	Tricosane
34	Eucalyptol	70	Tridecane
35	Heneicosane	71	Tritetracontane
36	Heptacosane	72	Undecane

Red indicates five specific VOC compounds of T.V. PheroLure

Figure 5. Yellow Bucket Funnel Trap® loaded with T.V. PheroLure® (Insect Science 2018)

Comparing of the differences in VOCs present in the presence of T.V. PheroLure® compared to naturally occurring background VOCs released by the tomato plants.

Sampling was done at the five selected sampling sites (A–E, Figure 1) for the duration of the trial (22 weeks). The resulting chromatographic data obtained was grouped into two groups: Group 1 (samples without T.V. PheroLure®) and Group 2 (samples with T.V. PheroLure®). An OPLS-DA model was created, but unfortunately a poor model was obtained (R^2Y = 0.238 and Q^2 = –0.231) with no separation observed between the groups, indicating that the two groups had similar compounds at same concentrations levels present as shown in Figure 6. As previously indicated, the samples taken in Week 0 (B) and Week 5 and 6 (A) (Figure 6) differed from the rest of the samples. In both instances, in Week 0 (B) and Week 5 and 6 (A), one would therefore expect the VOCs present to differ from each other.

Samples encircled and marked A, on Figure 6 refers to all the samples taken on the very first day of sampling. The tomato

Table 4. VOCs detected in T.V. PheroLure loaded YBFT

1	2,6,10,14,18,22-Tetracosahexaene, 2,6,10,15,19,23-hexamethyl-, (all-***E***)-
2	Decanal
3	Hexadecanoic acid
4	Isopropyl myristate
5	Methyl salicylate
6	Nonanal
7	Nonanoic acid
8	Octadecanoic acid
9	Octana
10	Tetradecanoic acid

Red indicates specific VOC compound of T.V. PheroLure®

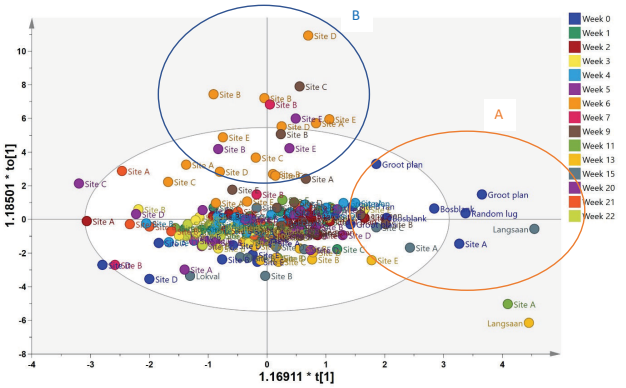

Figure 6. OPLS-DA score plot for VOCs present with and without T.V. PheroLure® present. A: Week 0; B: Week 5 and 6

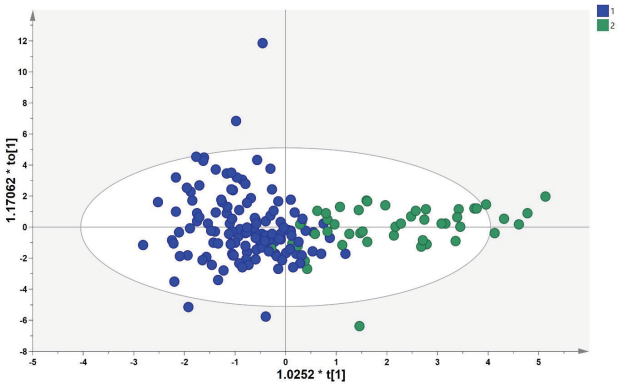

Figure 7. OPLS-DA scores plot comparing VOC emitted before (Group 1: Blue, Week 0–Week 11) and during harvest (Group 2: Green, Week 13–Week 22)

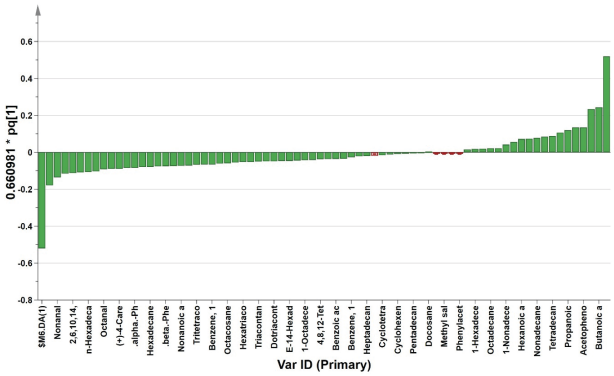

Figure 8. OPLS-DA loadings plot indicating the contribution of the variables to separation of VOCs before and during harvest. (*Red indicates five specific VOC compounds of T.V. PheroLure®)

plants were very young and planted the day before. Thus, one would expect that VOCs associated with the plant will be negligible in this sampling. Samples circled and labelled B refer to the samples taken during the flowering stage of the plants. This indicates that there is a higher incidence of VOCs present in the natural atmosphere during the flowering stage of the tomato plants possibly due to volatile aromas released by the flowers. It would be expected that the VOCs present in the tomato field during the flowering stage would differ from the natural VOC profile of the plant in the absence thereof.

Comparison of the VOCs present in a tomato field before and during harvest.

Harvesting of the tomato fruit commenced in Week 13 (sample 11). The resulting chromatographic data was therefore divided into two groups to create an OPLS-DA model for further analysis. Group 1 (blue: Week 0–Week 11) and Group 2 (green: Week 13–Week 22). Figure 7 is the OPLS-DA scores plot, displaying clear separation between the two groups.

The OPLS-DA loadings plot (Figure 8) indicates that the compounds decanal, nonanal, 5,9-undecadien-2-one, 6,10-dimethyl-2,6,10,14,18,22-tetracosahexaene, contributed to the separation according to the samples taken before harvest, whereas compounds that contributed to the separation during harvest include, butanoic acid butyl ester, undecane, acetophenone and isopropyl myristate.

The separation observed can be contributed to many factors of which one possibility may be the absence of fruit and flowers on the plants. Other factors which can also influence the VOC emissions are changes in the environment, such as humidity, excessive rainfall or drought, which may lead to increased volatile emissions (Holopainen and Gershenzon 2010).

The results demonstrate that differences in volatile compounds are most prominent in Week 0 and Week 5 and Week 6 (Figure 3) as well as before and during harvest of the tomato fruit (Figure 7). These changes may be contributed to changes in the physiological stages of the plant or environmental changes rather than the use of semiochemical-based products like T.V. PheroLure®. VOCs reported in the loading plots (Figure 4) indicated that apart from a slight increased contribution of limonene, there was no significant influence observed from the specific T.V. PheroLure® compounds on the natural background VOCs found in the tomato field. This is confirmed by Figure 7 indicating differences only for Week 0 and Week 5 and Week 6.

CONCLUSION

Volatile organic compounds (VOCs) in a commercial tomato field were sampled to determine the influence of semiochemical-based lures and devices such as T.V. PheroLure® could have on the natural background VOCs found in this tomato field.

Apart from a slight increased contribution of limonene, there was no significant influence observed from the T.V. PheroLure® compounds on the natural background VOCs found in the tomato field. There were differences observed between Week 0 and Week 5 and 6 samples. The differences in VOC presence observed in Week 0 and Week 5 and 6 of sampling could possibly be contributed to the physiology of the plant (plant age and flowering) rather than the use of the T.V. PheroLure®. Furthermore, the changes in VOCs observed before and during harvest may be contributed to the change in environment of the tomato plant.

There were no significant differences observed when comparing the VOCs present with and without T.V. PheroLure® in the tomato field. However, differences were observed during Week 0 and Week 5 and 6 of sampling.

Thus, the results indicate that the VOCs introduced into the field by agricultural devices do not significantly influence the naturally occurring background VOCs. Therefore, the rate of the registration process may be significantly improved if the toxicological and ecotoxicological requirements set by South African authorities are waived.

ACKNOWLEDGEMENTS

This work was supported by the University of South Africa and Insect Science (Pty) Ltd. Opinions expressed in this publication and the conclusions arrived at, are those of the authors and are not necessarily attributed to the funders. The authors would like to gratefully acknowledge Dr Wilma Augustyn (Tshwane University of Technology) for data interpretation and assistance in data analysis and Dr Divan van Greunen for consultation and discussion.

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