Behavioral Responses of Bemisia Tabaci Cryptic Species Med to Three Host Plants and Their Volatiles

zhe liu
Yangzhou University  https://orcid.org/0000-0003-3076-1057

WENBIN CHEN
Yangzhou University

SHUAI ZHANG
Yangzhou University

HAN CHEN
Yangzhou University

HONGHUA SU
Yangzhou University

TIANXING JING
Yangzhou University

YIZHONG YANG (✉️ yzyang@yzu.edu.cn)
Yangzhou University

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Abstract

*Bemisia tabaci* Gennadius (Hemiptera: Aleyrodidae) is a worldwide pest that damages more than 900 host plant species. The infestation behavior of this pest is affected by the volatile organic compounds (volatiles) of different plants and their growth stage. We investigated the chemical constituents of the volatiles extracted from three plants (*Gossypium hirsutum*, *Abutilon theophrasti* and *Ricinus communis*) at different growth stages (pre-flowering, *florescence* and fruiting) and their effects on the behavior of adult *B. tabaci*. The selectivity studies on three plants showed that the *B. tabaci* preferred *piemarker*. The olfactometer studies showed that growth periods of the three plants also affected the preference of *B. tabaci*. Volatiles of *piemarker* and cotton plant had different levels of attraction to adults during all stages. Volatile substances released by castor at stage of *florescence* have a repellent effect on *B. tabaci*. In the plant VS plant combination, the adults showed the strongest preference to volatiles from before and during anthesis of *piemarker*, followed by cotton, and then castor. A total of 23, 22 and 18 compounds were detected from volatiles of *piemarker*, cotton and castor, respectively, and proportions among the compounds changed during different stages of plant development. The olfactory responses of *B. tabaci* to volatile compounds showed that linalool and high concentration of leaf acetate had strong trapping effect on this pest, while 1-nonanal had significant repellent effect at high concentration. This study indicates that different plants and their growth stage affects their attractiveness or repellency to *B. tabaci* adults which is mediated by plant constitutive and dynamic changes. The compounds obtained by analysis screening can be used as potential attractants or repellents to control Mediterranean (MED) *B. tabaci*.

Introduction

*Bemisia tabaci* Gennadius (Hemiptera: Aleyrodidae) is a worldwide pest which is known to damage more than 900 host species (Lee 2020). It was first observed on tobacco in Greece in 1889 (Brown et al. 1995; Gennadius 1889). With the introduction of *Euphorbia pulcherrima* in China beginning at the end of last century, *B. tabaci* has gradually become an important agricultural pest in local area. It is a complex population with many cryptic species (at least MEAM1 (Middle East-Asia Minor I) and MED (Mediterranean), formerly biotypes B and Q, respectively) (Dickey et al. 2013). Although chemical control is still the main method for against *B. tabaci* currently, its widespread application has brought negative impacts on the environment and potential threat to human health (Liu et al. 2016). Therefore, many scientific researchers now focus on how to use natural enemies, microorganisms and semiochemicals from plants to control this pest.

In the interaction between plants and insects, the latter mainly rely on olfaction to sense extrinsic environment, and the plant volatiles provide important clues for insects to locate hosts for food or oviposition (Bernays and Chapman 1994; Liu et al. 2019). The olfactory system of insects has high sensitivity and specificity for plant volatiles (de Bruyne and Baker 2008). Previous study has reported that the preference of herbivorous insects to host plants is due to the presence of certain one or several attractive components in the host plant volatiles or the absence of repellent components (Li et al. 2002).
Piemarker, *Abutilon theophrasti*, as a Malvaceae family plant, is generally more attractive to pests than cotton (Chen et al. 2020; Liu et al. 2016). Castor *Ricinus communis*, containing ricin, its extract is mainly used to insecticide material around the world (Hua et al. 2013). However, recent studies and our field research have shown that plants attract or repel insects only at a certain growth stage (Knolhoff and Heckel 2014; Luo et al. 2018; Seiter et al. 2013). This may be related to the change of volatiles at different growth stages, but few studies have been reported. Therefore, it is necessary to study the host localization behavior of MED *B. tabaci* and its chemical mechanism in order to provide theoretical basis for the application of behavioral control methods to control this pest, to reduce the economic losses caused by it. The purpose of this study was to select the companion plants and their optimal growth period with regulatory effects on *B. tabaci*, in order to establish an accurate push-pull strategy for *B. tabaci* on cotton.

In this study, we aimed to determine (i) whether cotton *Gossypium hirsutum*, piemarker and castor had a differential attractiveness to adult *B. tabaci* using leaf disc test and free-choice assays, and (ii) whether different growth stage of those three plants had a differential attractiveness to adult *B. tabaci* using olfactometer, and (iii) volatiles of the three host plants at different stage are collected and analyzed, as well as (iv) identify chemical volatiles which attract or repel the insect.

## Materials And Methods

### Plant growth

Cotton plant, *Gossypium hirsutum* SGK321 seeds were obtained from the Institute of Plant Protection, Chinese Academy of Agricultural Sciences. Piemarker plant *Abutilon theophrasti* seeds were from Wang Zheng seedling sales center of China and castor plant *Ricinus communis* seeds were from Shouguang wentian seed industry co., ltd. They were grown in pots (size:13cm in diameter) in a 2:1 mixture of seedling substrate and soil. All plants were nurtured in a greenhouse and tested when they reached five true leaves. All plants were watered weekly. They were maintained at 28 ± 1°C with 65-75% RH and a 16:8 (L:D) at a light intensity of 1400–1725 lux, no pesticides were applied to the pants before or during the experiments.

### Insect rearing

The *B. tabaci* (cryptic species MED), were mass reared on cotton plant in mesh cages (60cm×60cm ×100cm). They were maintained at 28 ± 1°C with 65-75% RH and a 16:8 (L:D). The *B. tabaci* was regularly identified by mt DNA Co I gene sequence.

### Free-choice test in cages

The preference test of *B. tabaci* adults were determined on different plants using the free-choice test experiment. Three pots with cotton, piemarker and castor plants were arranged randomly in each iron framed cage (60cm×60cm×100cm) covered with the fine mesh nylon net to keep whiteflies from escape.
Twenty adults were introduced in the cage. Five replicates per treatment were set. To observe the preference study, 20 adults were introduced in the cage uniformly. In order to ensure the free movement of whiteflies, five paper plates with a diameter of 10cm and a height of 0.5cm were hung on the top of the cage, which were divided into five equidistant sites: east, south, west, north and middle. The numbers of adults on each plant were recorded at 1 h, 2 h, and 24 h after release (Liu et al. 2020; Zhou et al. 2008).

**Leaf disc test**

Referring to the method of Zhou et al. (2008), a filter paper sprayed with water was placed at the bottom of the glass petri dish (16cm diameter). The leaves of the plants under test were cut into round leaves with a hole puncher (1cm diameter). Two circular plates were taken from each plant and placed at different intervals in a petri dish. Fifty *B. tabaci* which were starved for 4 hours already released into a petri dish, and host selection of *B. tobaci* was observed 2 hours later. The experiment was repeated for 5 times. In order to reduce the influence of the light source on the test, the device is placed in separate environmental chambers with the 10000 lux intensity top light source at 28±3°C, 70%±5% RH.

**Olfactory choice test with Y-Tube olfactometer**

The behavioral response of *B. tabaci* to volatile compounds of three plants was evaluate by a Y-tube olfactometer bioassays, following the equipments and procedure as previously described by Akol et al. (2010) and Saad et al. (2015) with slightly modifications. Y-tube via odor sources using a QC-3 atmospheric sampler (0.2-3L/min) (Beijing Municipal Institute of Labour Protection, China) for circulating the volatile organic compounds. The air flow was adjusted and measured by a glass rotameter.

The test was carried out in a dark room so that adults cannot receive visual cues from the plants. A 300 lux LED light was placed 0.5 m vertically above the Y-tube olfactory meter. The adult whiteflies were released within 1.0 cm base of the Y-tube for after 4 h starvation and their responses assessed for 3 min. If it cross 1/3 of any of the two arms of the Y-tube branch and stayed a least of 30 seconds and did not return was considered as a positive-responsive individual. Otherwise, they were considered as non-responsive insect. For each 10 adults detected, the two lateral branches of the Y-tube were inverted 180°, which was a repeat. Each olfactory test was repeated 5 times, and a total of 50 adults were tested. The test time is arranged from 8:00 to 18:00. During the test, the temperature of the darkroom used for the test is kept at 26°C and the humidity is about 65%.

**Volatile compounds collection and analysis**

Different plants released dynamic headspace volatiles were collected at pre-flowering, florescence and fruiting. Similarly, the pot of plant was removed and the roots with soil were packed in tin foil. The plant was transferred individually to 10 L cylindrical glass chambers. Before trapping volatile, the activated charcoal-filtered air entered the chamber at 1.0 L/min with a vacuum pump for more than 30 min. Filtered out the air of the system and then the kept plant in the glass container for 1 h before trapping volatiles. During the volatile collection, activated charcoal purified air was pumped through Teflon tubing into the
system at a flow rate of 1 L/min through a glass tube filled 200 mg, 60-80 mesh Tenax TA adsorption column (Nazrul et al. 2017). The samples were cyclic collection for 4h and collected 4 plants (replicates) per treatment.

The trapped volatiles were extracted from the adsorbent tube using n-hexane (Sinopharm Chemical Reagent Co. Ltd., Shanghai, China) in 4 times, 100μL each time, and 7.03ug/mL n-octane (Aladdin Co. Ltd., Shanghai, China) was added to piemarker and cotton sample as an internal standard, and 5.87ug/mL methyl salicylate (Aladdin Co. Ltd., Shanghai, China) was added to castor sample as an internal standard. Samples were stored at –20 °C until they were analyzed through GC-coupled mass spectrometry (GC-MS).

For GC-MS, Trace ISQ (Thermo Fisher, USA) for detection, identification, and quantification of plant volatile components. The chromatographic column was DB-5 MS column (30 m×0.25 mm×0.25μm). A helium (99.999%) was used as a carrier gas with a flow rate of 1.0 ml min-1 with constant mode. Heating procedure: The column temperature was maintained to 50°C for 1 min, the oven temperature was increased from 50 to 150°C at a rate of 5°C min-1 and held for 2 min, and then from150 to 250°C at a rate of 10°C min-1 and held for 2 min.

**The olfactory responses of B. tabaci to volatile compound**

The volatiles compounds with significant changes in the of growth period of three plants and their standard samples was selected (Linalool, CAS: 78-70-6, Purity 95%; leaf acetate, CAS: 3681-71-8, Purity 98%; and 1-nonanal, CAS: 124-19-6, Purity 96%. All purchased from Aladdin Co. Ltd., Shanghai, China.) were diluted with n-hexane solution of 100, 10 and 1 (μL/mL), respectively. 20 μL of single volatile solution was accurately absorbed and dropped on 1.5cm × 2cm qualitative filter paper, and put into the bottle connected by one arm of the "Y" shaped tube. A qualitative filter paper dripping with 20 μL n-hexane was put into the bottle on the other arm as a control. The test method is the same as olfactory selection test.

**Statistical analyses**

IBM SPSS 25.0 as used to conduct all statistical analyses. Among copy numbers (4 replicates), ANOVA with Tukey HSD post hoc multiple pairwise statistical comparison and Kruskale Wallis test (non-parametric test) was used for analyzing the proportion of adults per plant in free-choise test and leaf disc test. For the Y-tube olfactory test, the hypothesis was that the pest showed no preference (i.e., a 25:25 response) for each arm. Data produced from Y-tube olfactometer choice bioassays were analyzed by $\chi^2$ test. Those who did not make a choice on host plants were not included in the selection rate.

The compounds were identified by comparing the fragmentation patterns from the mass spectra with the databases of the library NIST 2014 (National Institute of Standards and Technology, Washington, DC, USA) database. Exploration of the GC data collected from different stage of piemarker, cotton and castor samples were preliminarily conducted by principal component analysis (PCA). Classification models
using partial least square-discriminant analysis (PLS-DA) were generated and validated. Class modelling of three plants were finally performed by Soft Independent Model Class Analogy (SIMCA) respectively, to test the differences in the plant volatiles of different growth stage (Deconinck et al. 2018).

Results

Preference of *B. tabaci* to three plants

In free-choice assay, *B. tabaci* adults showed a significant difference in number among different plant treatments (Fig.1). In general, piemarker plant attracted a significantly higher number of *B. tabaci* adults than cotton and castor after 1 h (F=11.165, P=0.01), 2h (F=23.681, P=0.001) and 24h (F=44.432, P<0.0001) release, and no statistical difference between castor and cotton. Post-release time influenced the number of adults on different host plants. The number of *B. tabaci* on piemarker increased with the time, and increased from 50.5 % to 60.6% after 24 h release, while the number of whiteflies on castor decreased from 21.1% to 12.4% after 24 h release. However, stable *B. tabaci* population dynamics were detected in cotton, and the percentage of this pests on the host after 24, 48 and 72 h were 28.4%, 28.7% and 27.0%, respectively. The results of petri dish test are consistent with that of free-choice test. The selectivity of *B. tabaci* to piemarker (66.61%) was the highest, significantly higher than that of cotton (22.39%) and castor (11.00%) (Fig.2).

Response of *B. tabaci* to pre-flowering volatiles of three plant

The statistical analysis indicated respond preferentially of *B. tabaci* to volatiles from piemarker and cotton at pre-flowering stage compared to air. The attraction of piemarker and cotton to adults were the most significant (67.6%, $c^2=11.7$, $P<0.001$) and extremely significant (64.6%, $c^2=7.96$, $P<0.01$) respectively, than that of air flow. However, there was no obvious reaction between caster at pre-flowering stage(46.4% $c^2=0.38$, $P>0.05$) and air flow choses by adults(Fig.3 a).

The *B. tabaci* responded preferentially to volatiles emitted by piemarker (61.7%, $c^2=5.02$, $P<0.05$), when compared to castor (38.3%). They also responded favourably to volatiles from piemarker(60.8%, $c^2=4.26$, $P<0.05$), compared to cotton (39.2%). The difference between cotton and castor was extremely significant, in which 66.5% of *B. tabaci* chose cotton, and only 33.5% of them on castor ($c^2=10.24$, $P<0.01$) (Fig.3 a).

Response of *B. tabaci* to florescence volatiles of three plant

During flowering, the attraction of piemarker and cotton to *B. tabaci* were significantly greater than that of air flow, but castor was more resistant to them. Compared with air, The selection rate of *B. tabaci* on piemarker was 63.3% ($c^2=6.56$, $P<0.05$), cotton 61.7% ($c^2=5$, $P<0.05$) and castor 34.7% ($c^2=8.76$, $P<0.01$) (Fig.3 b).
The *B. tabaci* responded preferentially to the volatiles emitted by piemarker during florescence stage in combination of plant vs plant. Between the combinations of piemarker and castor, the selection rate of *B. tabaci* for the former and the latter was 64.1% and 35.9% ($c^2=7.36, P<0.01$), respectively. When the combination of piemarker and cotton was selected, 64.5% of the adults chose piemarker and 35.5% chose cotton ($c^2=7.84, P<0.01$). There were also significant differences between cotton and castor. Among them, 63.3% of *B. tabaci* responded favourably to volatiles from cotton, while only 36.7% to castor ($c^2=6.56, P<0.05$) (Fig.3 b).

**Response of *B. tabaci* to fruiting volatiles of three plant**

The *B. tabaci* responded preferentially to volatiles from piemarker and cotton when compared with to air, respectively, but no significant difference between castor and air (Fig.4 a). Compared with air, the selection rate of *B. tabaci* was 61.1% for piemarker ($c^2=4.5, P<0.05$), 62.0% for cotton ($c^2=5.3, P<0.05$) and 52.6% for castor ($c^2=4.5, P<0.05$).

The *B. tabaci* adults have no significant preference to the volatiles between piemarker (48.9%) and castor plants (51.1%, $c^2=0.02, P>0.05$). They also no responded preferentially to between piemarker (46.5%) and cotton plants (53.5%, $c^2=0.36, P>0.05$), as well as between cotton (52.2%) and castor (47.8%, $c^2=0.12, P>0.05$)(Fig.4 a).

**Qualitative analysis of three plant volatiles in different periods**

The results showed that more than 30 different compounds were detected, including alcohols, aldehydes, esters, terpenes and other compounds (Fig.5). In total, 23 volatiles were identified from the piemarker plants, with 21 detected before flowering, 22 at flowering, and 19 at fruiting. Twenty-two compounds were tentatively identified from three growth stage volatiles released by cotton, at the pre-owering, florescence and fruiting volatiles compounds was 19, 21 and 19 kinds, respectively. A total of 18 compounds were detected in castor during the three stages, including 14 at pre-owering, 17 at flowering and 14 at fruiting stage (Fig.5).

**PLS-DA analysis of different periods volatiles of three plant**

The partial least squares-discriminant analysis (PLS-DA) showed that the plant volatiles contents were clearly separated among pre-owering, florescence and fruiting of piemarker. The first two significant PLS components explained 43.3% and 47.1% of the total variance, respectively (Fig. 6 a). In this model, the following seven volatile compounds linalool, unknow, ethyl caprylate, ethyl nonylate, butyl acrylate, 1,3,7-ocimene, 3-hexadecanol with VIP values $\geq 1.0$ contributed most to the separation among different periods volatiles of piemarker (Fig. 6 b).

The PLS-DA also showed a clear separation among pre-owering, florescence and fruiting of cotton. The first two significant PLS components explained 46.3% and 49% of the total variance, respectively (Fig.7 a). The first component and the second component showed a clear separation among three volatiles of
different periods of cotton. In this model, the following nine volatile compounds leaf acetate, unknow, ethyl caprylate, naphthalene, ethyl nonanoate, nonane, 1,3-xylene, dodecyl aldehyde, octanal with VIP values $\geq 1.0$ contributed most to the separation among different periods volatiles of cotton (Fig. 7b).

The PLS-DA showed that the first component and the second component showed a clear separation among pre-flowering, fluorescence and fruiting volatiles of castor. The first two significant PLS components explained 43.5% and 44.5% of the total variance, respectively (Fig. 8a). In this model, the following six volatile compounds 1-nonanal, ethyl caprate, 2-butyl-1-octanol, butyl acrylate, 3-hexadecanol, naphthalene with VIP values $\geq 1.0$ contributed most to the separation among different periods volatiles of castor (Fig. 8b).

The olfactory responses of *B. tabaci* to standard samples of volatile compound

The Figure (4b) showed that among the three compounds, the *B. tabaci* responded clear preference to linalool with the concentration of 1, 10 and 100μL/mL, and the selection rates were 64.4% ($c^2=7.72$, $P<0.01$), 61.2% ($c^2=4.56$, $P<0.05$) and 60.4% ($c^2=3.92$, $P<0.05$), respectively. In addition, leaf acetate was also significantly attractive to this pest at 100μL/mL, and the selection rate was 60.9% ($c^2=4.32$, $P<0.05$). However, there was no significant difference between 1 and 10μL/mL ($c^2=0.02$, $P>0.05$; $c^2=0.28$, $P>0.05$). At concentration of 100μL/mL, 1-nonanal showed the obvious repellant against *B. tabaci*, the selection rate only 32.5% ($c^2=11.56$, $P<0.001$). And there was no significantly preference of adults to medium and low concentrations ($c^2=1.87$, $P>0.05$; $c^2=0.02$, $P>0.05$).

Discussion

In this study we investigated the response of *B. tabaci* to chemical cues released by different plants at their different growth stages. Results showed that the piemarker plants attracted *B. tabaci* adults, while the castor repelled against them. However, attraction/avoidance only occurred during the certain growth period of plants, which was closely related to the release of volatiles by plants.

It was found that *B. tabaci* showed a strong preference to piemarker in the free-choice test. Moreover, the selection rate of adults increased with time increase, and adults showed obvious host selectivity in the process of diffusion. In the leaf disc test, the selection rate of *B. tabaci* to piemarker reached 66.61%, which was consistent with the results of free-choice test. The *B. tabaci* showed obvious preference for piemarker in previous behavioral studies. For instance, Lin et al. (2006) found that *B. tabaci* significantly preferred piemarker in the piemarker vs air treatment with a selection rate of 68.1%, but showed no obvious preference when compared with cucumber or tobacco. Wang et al. (2016) also found that piemarker and cotton had strong attractiveness to *B. tabaci* among 13 host plants.

Our study indicates the physiological stage of the host plant influences the host-foraging behavior of the whitefly. The “Y” olfactometer test demonstrated that in the plant VS air flow treatment, compared with cotton, piemarker is more attractive to *B. tabaci* in the pre-flowering stage. In the plant VS plant treatment,
at pre-flowering and flowering stage, most adults chose piemarker, followed by cotton, and then castor. but the preference of *B. tabaci* to the three plants was not significantly different during fruiting stage which suggests that the piemarker, as an attractor of pests, was more attractive at the pre-flowering and flowering stages. Mohammed et al. (2021) studied the influences of the volatiles from different parts of brinjal plants on the behavior of adult *Leucinodes orbonalis*, and found that adults responded differently to the volatiles extracted from fruits, leaves, shoots and flowers. In addition, we detected the castor had a repellent effect on *B. tabaci* at the flowering stage. In the castor VS air flow treatment, only 35.9% of adults chose castor at the flowering stage while there was no significant difference in avoidance (preference) of *B. tabaci* at pre-flowering stage and fruiting stage. Luo et al. (2018) investigated the population of *B. tabaci* in the castor trap belt, its neighboring cotton fields and castor-free cotton fields, and found that there was no significant difference in the number of *B. tabaci* on the three areas between June and July. However, in August and September, the number of whiteflies on castor and cotton adjacent to castor was 1205 and 1580 per 100 plants, respectively, significantly lower than that of control cotton (4697 per 100 plants). It also proved that castor had repellent effects on *B. tabaci* at a certain period.

The change of composition and content of volatiles at different growth stages of plants may be responsible for the host-seeking behavior of whiteflies. The volatiles play a key role in the process of host recognition, which is an important clue for insects to identify and locate food and natural enemies (Renou et al. 2019). In this study, more than 30 different compounds were detected, including alcohols, aldehydes, esters and terpenes (23 in piemarker, 22 in cotton, and 18 in castor). This is almost consistent with previous reports (Chen 2013; Rizwangul 2018; Zhang et al. 2016). In addition, the proportion of some volatile compounds in the three plants changed significantly at different growth stages, which may be the reason for the difference in preference for *B. tabaci* at different growth stages.

The olfactory responses of *B. tabaci* to volatile compounds such as linalool, leaf acetate and 1-nonanal were determined. It found that linalool and high concentration of linalool acetate had strong trapping effects on cryptic species MED *B. tabaci*, while 1-nonanal had significant repellent effects at high concentration. The result indicates that concentration of the compounds is also an important factor affecting host selection. Therefore, it indicates that linalool, leaf acetate and 1-nonanal can be used as potential attractants or repellents to control *B. tabaci*.

In general, our research proves that different plants and their growth stages affect the attraction (repellent) to *B. tabaci* adults and that the attraction (repellent) cue is the volatiles emitted from pre-flowering, fluorescence or fruiting plants. The three volatile compounds (linalool, leaf acetate and 1-nonanal) could be used as potential attractants or repellents in future.

**Declarations**

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**Figures**

**Figure 1**

Preference of *Bemisia tabaci* to different host plants in cage. Different lowercase letters a and b indicate significant differences in the number of *Bemisia tabaci* on different hosts at the same time ($P \leq 0.05$).

**Figure 2**

Preference of *Bemisia tabaci* to different host plants in petri dishes. Different lowercase letters a and b indicate significant differences in the number of *Bemisia tabaci* on different hosts ($P \leq 0.05$).
Figure 3

Attracting effects of different host plants on *Bemisia tabaci* at pre-flowering(a) and flowering(b) stage. The data in the histogram represents the percentage of individuals who acted in each treatment. *Significant difference (P<0.05), **extremely significant difference (P<0.01), *** most significant difference (P<0.001) and ns shows no significant difference (P>0.05), c² test.*
Figure 4

Behavioral response of *Bemisia tabaci* to different host plants at fruiting stage(a), and to a single volatile componen(b). The data in the histogram represents the percentage of individuals who acted in each treatment. *Significant difference ($P \leq 0.05$), and ns shows no Significant difference($P > 0.05$), $c^2$ test.
Figure 5

The heat map is used to compare volatile components of three host plants in different periods, 1, 2 and 3 represent the pre-flowering, flowering and fruiting stages of piemarker respectively; 4, 5 and 6 represent the pre-flowering, flowering and fruiting stages of cotton respectively; 7, 8 and 9 represent the pre-flowering, flowering and fruiting stages of castor respectively. The values represent the mean percentages of the peak area relative to the peak area of the internal standard.

Figure 6

(a). Partial least squares discriminant analysis (PLS-DA) of pre-flowering (PF), florescence (FL) and fruiting (FR) of piemarker plant volatile compounds. The score plot display the grouping pattern according to the first two components and the ellipse defines the Hotelling's $T^2$ confidence interval (95%)
Figure 7

(a). Partial least squares discriminant analysis (PLS-DA) of pre-flowering (PF), fluorescence (FL) and fruiting (FR) of cotton plant volatile compounds. The score plot display the grouping pattern according to the first two components and the ellipse defines the Hotelling's $T^2$ confidence interval (95%) for the observations. (b). VIP value of cotton plant volatile compounds. Compounds ID: 1. leaf acetate, 2. unknow, 3. ethyl caprylate, 4. naphthalene, 5. ethyl nonanoate, 6. nonane, 7. 1,3-xylene, 8. dodecyl aldehyde 9. octanal, 10. α-caryophyllene 11. linalool, 12. ethylbenzene, 13. DMNT, 14. 6-methyl-5-hepten-2-one, 15. unknow, 16. 3,7-dimethyl-1,3,6-octatriene, 17. caryophyllene, 18. decanal, 19. 1-1-nonanal, 20. 2-ethylhexyl acrylate, 21. methyl benzoate, 22. benzaldehyde, 23. decane, 24. ethyl caprate.

Figure 8

a. Partial least squares discriminant analysis (PLS-DA) of pre-flowering (PF), fluorescence (FL) and fruiting (FR) of castor plant volatile compounds. The score plot display the grouping pattern according to the first two components and the ellipse defines the Hotelling's $T^2$ confidence interval (95%) for the observations. (b). VIP value of castor plant volatile compounds. Compounds ID: 1. 1-1-nonanal, 2. ethyl caprate, 3. 2-butyl-1-octanol, 4. butyl acrylate, 5. 3-hexadecanol, 6. naphthalene, 7. ethylbenzene, 8. 3,7-dimethyl-1,3,6-octatriene, 9. decanal, 10. butyl acetate, 11. dodecyl aldehyde, 12. 1,3-xylene, 13. d-Longifolene, 14. 2-ethyl hexanol, 15. methyl benzoate, 16. (-)-thujopsene, 17. 6-methyl-5-hepten-2-one, 18. unknow, 19. decane 20. unknow.