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Identification of chemical cues of *Camellia sinensis* (Ericales: Theaceae) and alternate host plants for preference by tea mosquito bug *Helopeltis theivora* (Hemiptera: Miridae)

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Abstract

The tea mosquito bug (TMB), *Helopeltis theivora* (Hemiptera: Miridae) is a polyphagous pest causing serious economic loss in tea plantations of north east India. The push-pull companion cropping system can be used as a component of integrated pest management system to control the pest. We examined the host plant choice of *H. theivora* for *Duranta erecta* (Lamiales: Verbenaceae), *Senna tora* (Fabale: Fabaceae), *Lawsonia inermis* (Myrtales: Lythraceae), *Psidium guajava* (Myrtales: Myrtaceae) and *Melastoma malabathricum* (Myrtales: Melastomataceae) including the primary host, *Camellia sinensis* (Ericales: Theaceae) using two-choice olfactometric bioassay. The growth index and relative growth index of *H. theivora* were calculated for different host plants. *H. theivora* preferred the odor released from the mechanically damaged leaves of *C. sinensis* and *P. guajava*. The further study on biochemical constituents of all the host plants reflected the role of soluble sugar, total flavonoids and terpenoids as cues for host preference by *H. theivora* which indicate a scope for electroantennographic assessment in future. The information generated for *P. guajava* preference can be utilized in trap cropping or trap formulating approaches for management of *H. theivora*.

Keywords: *C. sinensis*; Olfactometric bioassay; Growth index; Flavonoids; *H. theivora*

1. Introduction

Phytophagous insects use their olfactory sensilla in their search for host plants for either oviposition or feeding (Pophof et al.2005; Bora et al.2013). Once they land on the host plant, they use their gustatory sensilla for perceiving taste (Bora et al. 2015, Bora et al. 2016) and prefer the host plant where the progeny performs the best (Molleman et al.2020). The olfactory sensilla are shown to respond to the volatile organic chemicals mostly in the form of C6 derivatives or mono-, sesquiterpenoids emanated by the host plants into the environment in proximity (Deka and Bora 2014; Picket and Khan 2016), the gustatory sensilla respond to the various constitutive or induced chemicals present in the leaves. Plants evolve the mechanism for synthesis of defensive chemicals and even mechanical damage led to greater emission of volatile chemicals. The polyphagous insects in nature on the other hand are under strong selection pressure as the sustainability of their progeny depend on the chemical constituents of the host plants which vary from species to species. Host acceptance and host related performance are often tightly connected. When the insect pests use the volatile chemicals primarily for host acceptance, they use the plant primary and secondary metabolites as nutrients essential for the growth, development, performance and sustainability (Hopkins *et al*., 2017; Stec et al.2021). The carbon-based nutrient, carbohydrates in general act as a major source of energy for various insect activities including flight and soluble sugars act as a powerful feeding stimulant (Genc 2006). The secondary metabolites are mainly produced in plants as defensive compounds against pathogens and herbivores (Fraenkel, 1959) of which plant terpenoid and phenolic compounds constitute two major groups (Lattanzio et al. 2000; Lattanzio 2008, Boncan et al. 2020). They are known to regulate the feeding and oviposition by acting as a deterrent or stimulant in many herbivorous

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insects (Simmonds 2001; Van Loon et al. 2002; Awmack and Leather 2002; Mierziak et al. 2014). Estimation of selected bio-constituents therefore provides valuable cues regarding host plant metabolite preference of the herbivore.

The tea mosquito bug (TMB), *Helopeltis theivora* Waterhouse (Hemiptera: Miridae) is a polyphagous pest causing serious economic loss in tea plantations of North East India. The insect sucks the cell sap of tender shoots of *Camellia sinensis* (Ericales: Theaceae) causing a heavy production loss of up to 10-50%. Tea is an important cash crop providing the most popular globally acknowledged beverage having antioxidant potency owing to the presence of a considerable amount of phenolic compounds. Because the tea plantations are maintained in a monoculture system, they provide favourable habitat for *H. theivora* and other major pests and this necessitates the application of a good amount of pesticides. However, the use of pesticides in tea gardens reduces the quality of tea and has been a matter of serious concern for decades in the case of tea exported from North East India. Being polyphagous, the insect feeds on large number of plants (Saroj et al.2016). The polyphagous nature of an insect pest favours the strategy of trap cropping, which is advocated as effective option of integrated pest management programmes (Sarkar et al.2018, Dara 2019). The trap cropping is widely used in push-pull system of insect pest management in which the trap crop pulls the insect pests (Khan et al.2016). The IPM programmes in tea plantations of NE India has not incorporated any trap cropping option till now (Roy et al.2018). A systematic study for identification of potential trap crop and their use in tea plantations may reduce the economic and environmental challenges caused by *H. theivora*. Such an objective also necessitates the determination of chemical factors associated with host plant selection by *H. theivora.*

Therefore, in the present study, we examined the preference of *H. theivora* for primary and secondary host plants through olfactometric assay, studied its' performance by rearing on the plants and analysed selected phytochemical constituents for having a chemical basis of understanding. The study is expected to reveal information about the potential secondary host plant for *H. theivora* and its phytochemical constituents generally associated with insect-plant interaction.

2. Material and methods

2.1. Insect and plant materials

The nymphs of *H. theivora* were collected from the selected tea estates of Assam, India. The tender shoots of a primary host plant, *C. sinensis* (TV23, a variety), and alternate host plants *Duranta erecta* (Lamiales: Verbenaceae), *Senna tora* (Fabales: Fabaceae), *Lawsonia inermis* (Myrtales: Lythraceae), *Psidium guajava* (Myrtales: Myrtaceae) and *Melastoma malabathricum* (Myrtales: Melastomataceae) were collected from the gardens maintained in outdoor at ambient temperature. The nymphs were then reared on freshly collected tender shoots of the host plants maintained in water containing plastic bottles. The plants were covered with glass chimneys with the upper ends closed with a muslin cloth to prevent escaping of the insects. The chimneys were kept in the laboratory at room temperature, photoperiod (10D:14L) and relative humidity (75± 2%). The shoots of primary and alternate host plants were replaced every 24 hrs.

2.2. The analysis of growth index and relative growth index of *H. theivora* on different host plants

The collected nymphs of *H. theivora* were reared in the laboratory on *C. sinensis* and the selected alternate host plants. To ensure mating and egg deposition by the female, the newly moulted adult males and females were kept together in separate chambers. Soon after hatching, with the help of a soft brush, the first instar nymphs were transferred to tender shoots of the plants. When they completed one generation in laboratory condition, five pairs of adult insects from the F1 generation were used as parent stock for the study of growth indexes. The total number of eggs, nymphs and adults was counted daily and the study was executed for subsequent three generations each of which was maintained with five replications (May to November).

The growth index (GI) and relative growth index (RGI) was calculated to quantify the development of *H. theivora* on different host plants following the method of Zhang et al. (1993) with modification. The data obtained for the three life stages viz. eggs, nymphs and adults of *H. theivora* on the respective host plants were averaged to calculate GI and RGI for all the three generations. The GI and RGI for the third generation was calculated by averaging the data of five replications maintained. Only the third generation was chosen to calculate GI and RGI due to the steady performance of the insect in all the host plants which were much comparable to each other than the first and second generations. The formulae used for calculating GI and RGI:

$$\mathrm{GI}=\frac{\sum_{i=1}^{i_{max}}[n_{(i)}\times i]+\sum_{i=1}^{i_{max}}[n'_{(i)}\times(i-1)]}{N\times i_{max}}$$

Where i is the stage number, $n_{(i)}$ is the number of live nymphs at stage i, $n'_{(i)}$ is the number of dead nymphs at stage i, i_{max} is the highest attained stage of the insect which is 3 in the present study i.e., adult stage and N is total number of insects in the group.

After obtaining the GI for each host plant the RGI was determined by using the following formula:

$$\text{RGI}=\frac{\text{GI of each alternate host plant}}{\text{GI of the primary host plant (}C.sinensis\text{)}}$$

2.3. Olfactory response of *H. theivora* to the leaf odor of the host plants

The olfactory orientation of *H. theivora* was assessed by following the method of Blackmer et al. (2004) with modifications. A Y-tube olfactometer consisting of two glass arms of 12 cm length and 3.0 cm diameter laid at 100^0 angle connected by a base tube of 22 cm length was used. A constant humidified and filtered airflow was maintained by a pump connected to an air-flow meter. The glasswares were washed properly in detergent solution, rinsed with distilled water and dried in oven at 100 °C before performing the tests. Every test was done independently using insects not used in previous choice tests or after an interval of 24 hours. Moreover, the Y-tube was cleaned with ethanol after each test. The leaves were collected from uninfested host plants. The two choice tests were performed by keeping a ball of five freshly crushed leaves of the plant options in different combinations in the odor chambers separately. The laboratory reared fifth instar nymphs of *H. theivora* were released on the middle arm inside the base tube of the olfactometer. Twenty insects were used for each initial test. The initial response of nymphs identified as "walking into one of the arm within one minute" was recorded. If the nymph did not make any choice within one minute of its release the test was discarded. After the initial odor preference tests done using twenty insects, another set of tests was performed between *C. sinensis* and all the other potential plants using fifty insects independently for each test. To conduct the behavioural trials for odor preference, the insects were collected from tea gardens and were kept on tender leaves of *C. sinensis* for 24 hours in laboratory condition for acclimatization.

Finally, we analysed the data to assess the preference of *H. theivora* to different host plants using the grand average of preference in comparison to *C. sinensis* which was taken as the reference(r) standard. The reference grand mean ($\bar{X}$r) ± CLr (Confidence limit for r) was calculated by averaging the preference percentage to *C. sinensis* as fixed option in all the olfactrometric tests carried out with the alternate host plants following the method of Sadeh et al.2019 with modification. The mean preference of *H. theivora* towards the alternate host plants were compared with the reference grand mean ($\bar{X}$r) to partition the host plants into different preference categories. While comparing the mean preference percentage of *H. theivora* towards the alternate host plants with the reference grand mean ($\bar{X}$r), 16% being the lowest preference percentage when *C. sinensis* was used as the fixed option in the binary bioassay, the values 0-15% were excluded in computing the confidence limits of preference.

2.4. Phytochemical analysis

Estimation of total soluble sugar: The dried leaf samples were depigmented using 100% acetone, ultra-turax. Sugar was extracted with 1.5 ml of 80% ethanol by centrifuging and the supernatant was used for estimation of soluble sugar by anthrone method (Nag 2015). For preparation of anthrone reagent 1 g of anthrone was dissolved in 500 ml of 72% sulphuric acid. Standard glucose solution was prepared by dissolving 100 mg glucose in 100ml distilled water. From the stock solution of glucose 10ml was taken and diluted with 100 ml of distilled water to get the working standard (100 µg/ml). Soluble sugar content was determined with the help of standard graph prepared by using the working standard.

Estimation of total terpenoid: The total terpenoid content in the leaf sample was determined by following the method of Ferguson (1956). The leaf powder of each sample was soaked in ethanol for 24 hours. The filtrate was extracted with petroleum ether and the ether extract was treated as total terpenoids for estimation by weighing method.

Estimation of total polyphenol content: The polyphenol content of each shade dried leaf sample was extracted with aqueous ethanol with vigorous shaking following the method of Baba and Malik (2015) and determined by the Folin-Ciocalteau method. The standard solution was prepared by dissolving 100 mg catechol in 100 ml distilled water. From this solution working standard was prepared (100 µg/ml). Along with the working standard different aliquots of the samples were also prepared. There after 0.5 ml Folin-ciocalteau and 2 ml of 20% sodium carbonate was added and the test tubes were kept in boiling water bath for one minute and the polyphenol content was determined using a standard calibration curve. The results were expressed as a gram of catechin equivalent per 100 g dry weight.

Estimation of total flavonoid: The total flavonoid was estimated according to the method of Woisky and Salatino (1998) with modification. Each plant sample was extracted with aqueous ethanol followed by shaking at 200 rpm for 24 hours.

Then the working standard was prepared (100 µg/ml) and different aliquots of the plant samples as well as working standard were taken in different test tubes. After making the volume of each test tube up to 3 ml by adding distilled water 0.1 ml 10% aluminium chloride, 0.1 ml of 1M potassium acetate and 1.5 ml 95% ethanol were added. Finally, the total flavonoid was estimated after incubation of 30 minutes by colorimetric assay, the results were expressed as a gram of quercetin equivalent per 100 g dry weight.

2.5. Statistical analysis

The percentage of preference of the leaf odor of the host plants was analysed by Pearson Chi square and where the assumption of normality was questionable nonparametric Mann-Whitney U or Wilcoxon test was used. The variation of biochemical constituents of host plants were examined by one way ANOVA. Post hoc mean separation was executed with LSD and Tukey's HSD test. All the statistical analysis were performed using SPSS software version 17.0 (SPSS Inc., Chicago, IL).

3. Results

3.1. Behavioural assay for Odour preference

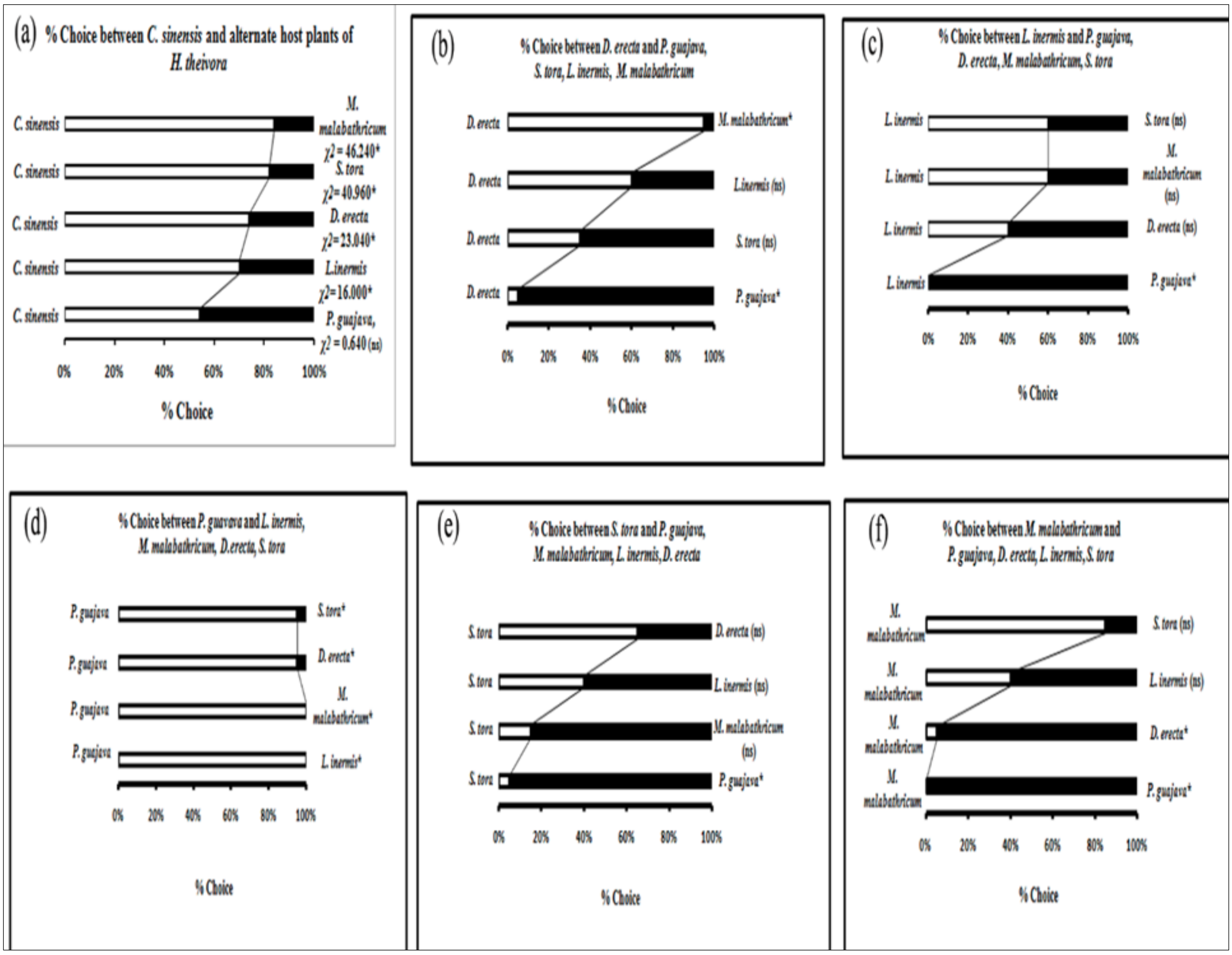

Figure 1 Behavioral response of H. theivora nymphs in olfactometric two choice bioassay in (a) *C. sinensis* (b) *D. erecta* (c) *L. inermis* (d) *P. guajava* (e) *S. tora* (f) *M. malabathricum* '*' indicates $p<0.0001$, ns indicates 'Not significant' in Chi square and Mann Whitney U test

When odor preference of *H. theivora* for other host plants with respect to *D. erecta* was studied, the attraction was significant towards *C. sinensis* ($\chi^2_{C. sinensis}=23.04$, $p<0.0001$) (Fig-1 a) and *P. guajava* ($U_{P.guajava} = 20.00$, $p< 0.0001$), but the nymphs showed significant attraction towards *D. erecta* ($U_{D. erecta}= 20.00$, $p<0.0001$) when it was given a choice with *M. malabathricum* (Fig-1b).

When odor preference of *H. theivora* was studied between *L. inermis* and the other host plants, the nymphs became significantly attracted to *C. sinensis* ($\chi^2_{C. sinensis}$=16.00, p<0.0001) (Fig-1a) and *P. guajava* ($U_{P.guajava}$ = 0.00, p<0.0001)(Fig-1c) .

When odor preference of *H. theivora* was studied between *P. guajava* and the other host plants, the variations of preference of nymphs to *C. sinensis* and *P. guajava* was not significant (Fig-1a). But the nymphs were significantly attracted to *P. guajava* when the choice was given with *L. inermis* ($U_{P.guajava}$=0.00, p<0.0001), *M. malabathricum* ($U_{P.guajava}$=0.00, p<0.0001), *D. erecta* ($U_{P.guajava}$=20.00, p<0.0001) and *S. tora* ($U_{P.guajava}$=20.00, p<0.0001) (Fig-1d).

When odor preference of *H. theivora* was studied for *S. tora*, the nymphs became significantly attracted towards *C. sinensis* ($\chi^2_{C. sinensis}$ =40.96, p<0.0001) (Fig-1a) and *P. guajava* ($U_{P.guajava}$=20.00, p<0.0001) (Fig-1e) with respect to *S. tora.*

When the odor preference of *H. theivora* was studied between *M. malabathricum* and the other host plants, the attraction of *H. theivora* nymphs to *C. sinensis* ($\chi^2_{C. sinensis}$=46.24, p<0.0001) (Fig-1a), *P. guajava* ($U_{P.guajava}$= 0.00, p<0.0001) and *D. erecta* ($U_{D. erecta}$= 20.00, p<0.0001) were significant with respect to *M. malabathricum* (Fig-1f).

The reference standard was found to be 72.80 ± 11.97 with 95% upper and lower confidence limits (CL) of 83.27 to 62.33 with a total variation of 16.44% based on the coefficient of variation (CV). By comparing the mean preference for each alternate host plant ± CL ($\bar{X}$a ± 95% CLa) to the reference grand-mean ± CL ($\bar{X}$r ± 95% CLr) the host plants were divided into three categories, 'High', 'Moderate' and 'Low' preference groups. The 'High' preference group had a mean preference greater than the reference grand-mean ± 95% upper CL ($\bar{X}$H >$\bar{X}$r + 95% CLr) and included only *P. guajava*. The 'Moderate' preference group had a mean preference between the reference grand-mean ($\bar{X}$r) and the lower CL ($\bar{X}$r >$\bar{X}$M>$\bar{X}$r – 95% CLr) and included *D. erecta* and *M. malabathricum*. The 'Low' preference group had a mean preference lower than the reference grand-mean *(*$\bar{X}$L <$\bar{X}$r – 95% CLr) and included *L. inermis* and *S. tora* (Fig-2).

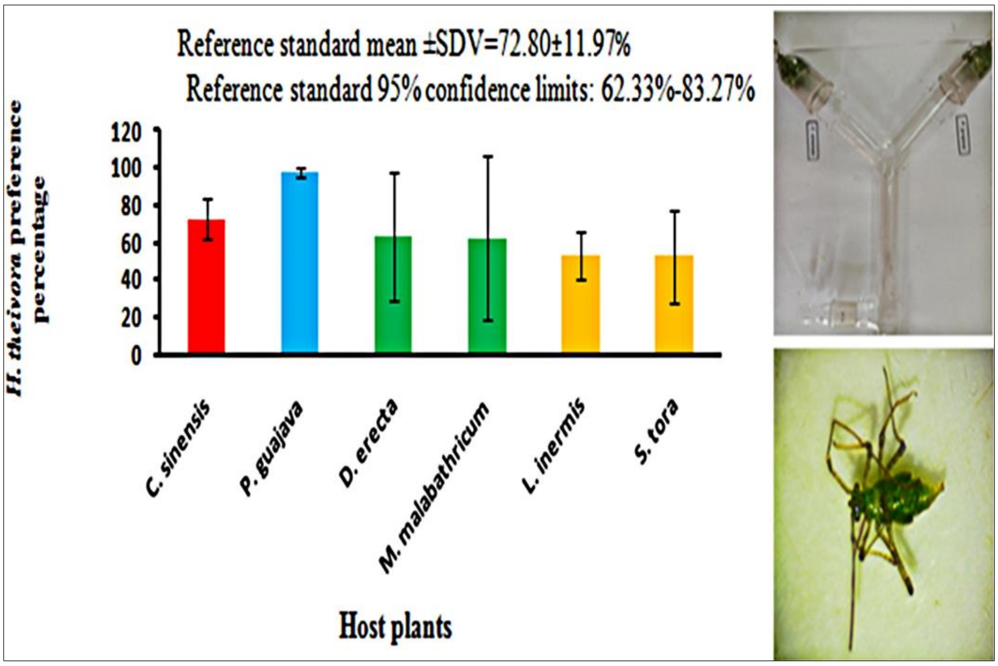

Figure 2 Preference of *H. theivora* to *C. sinensis* and alternate host plants following olfactory choice bioassay. Error bars represent 95% CL. The categorization was done in relation to $\bar{X}$r±95% CL, grand mean of preference percentage to *C. sinensis*. High (blue), moderate (green) and low (yellow)

3.2. Growth Index and RGI

The GI of all three generations of *H. theivora* were 0.54, 0.55, 0.45, 0.36, 0.44 and 0.40 respectively for *C. sinensis*, *D. erecta*, *L. inermis*, *P. guajava*, *M. malabathricum* and *S. tora*. The RGI of *D. erecta* (1.01) was followed by *L. inermis* (0.83), *P.* guajava (0.87), *M. malabathricum* (0.73) and *S. tora* (0.66). The GI of the third generation of *H. theivora* on different host plants were 0.55, 0.56, 0.48, 0.47, 0.45, and 0.42 respectively for *C. sinensis*, *D. erecta*, *P. guajava, L. inermis*, *S. tora*, and *M. malabathricum*. In this case also, the RGI was maximum in *D. erecta* (1.01), followed by *P. guajava* (0.87) and *L. inermis* (0.85) (Table-1).

Table 1 GI and RGI of *H. theivora* with respect to different host plants

Plants	**For all three generations of *H. theivora***		**For only third generation of *H. theivora***	
	GI	**RGI**	**GI**	**RGI**
C. sinensis	0.54	-	0.55	-
D. erecta	0.55	1.01	0.56	1.01
L. inermis	0.45	0.83	0.47	0.85
S. tora	0.36	0.66	0.45	0.82
P. guajava	0.44	0.81	0.48	0.87
M. malabathricum	0.40	0.73	0.42	0.76

3.3. Phytochemical analysis

The highest amount of soluble sugar was higher in *P. guajava* (5.57±0.65) as compared to the other host plants. The terpenoid content was the highest in *L. inermis* (0.625±0.021, $p<0.0001$) and the lowest amount was recorded in *M. malabathricum* (0.397±0.019, $p<0.0001$). The variations in polyphenol level were significant and the highest amount was recorded in *C. sinensis* (9.02±1.73, $p<0.0001$). The level of flavonoid was significantly high in *P. guajava* (1.89±0.49, $p=0.044$) and the lowest amount was deteded in *S. tora* (0.74±0.10, $p=0.044$) (Table-2).

Table 2 Average bioconstituents of different host plants of *H. theivora*

Plants	**Percentages of different bioconstituents (Mean ± SEM)**			
	Soluble sugar	**Terpenoid**	**Polyphenol**	**Flavonoid**
C. sinensis	$4.53^{ab}\pm0.81$	$0.426^{a}\pm0.027$	$9.02^{b}\pm1.73$	$1.84^{ab}\pm0.18$
D. erecta	$5.21^{ab}\pm0.74$	$0.574^{bc}\pm0.024$	$2.54^{a}\pm0.46$	$1.46^{ab}\pm0.22$
L. inermis	$4.04^{ab}\pm0.95$	$0.625^{c}\pm0.021$	$2.97^{a}\pm0.32$	$1.28^{ab}\pm0.15$
S. tora	$2.26^{a}\pm0.44$	$0.554^{bc}\pm0.037$	$2.35^{a}\pm0.28$	$0.74^{a}\pm0.10$
P. guajava	$5.57^{b}\pm0.65$	$0.495^{ab}\pm0.024$	$4.97^{a}\pm0.97$	$1.89^{b}\pm0.49$
M. malabathricum	$4.99^{ab}\pm0.23$	$0.397^{a}\pm0.019$	$4.61^{a}\pm1.14$	$1.40^{ab}\pm0.30$
SED	0.96	0.06	2.37	0.66
CD (0.05)	1.71	0.11	3.94	1.10

Means followed by different letters within a column are significantly different ($p<0.05$) (in case of soluble sugar it was near significant, $p = 0.05$) (Post hoc tests, LSD and Tukey), SED: Standard error of difference and CD: Critical Difference represent column wise differences, NS: Not significant, SEM: Standard error of mean.

4. Discussion

While growth of the insect depends on the plant chemical constituents, the approach of the insect to the hostplant followed by subsequent landing for feeding and oviposition depends on the odour emanated by the plant. The odour comprises blends of volatile organic chemicals (VOC) which may be insect pheromones or plant VOCs. The plant VOCs are released in enhanced quantity when the plant is damaged by herbivory or other infestations. Therefore, we hypothesized that the freshly crushed leaves will provide an odour of stronger intensity than a set of whole leaves plucked to be able to stimulate the olfactory sensilla of *H. theivora*. Literature reveals preferential olfactometric response of *H. theivora* to different volatile organic chemicals including C6 derived hydrocarbons (Sachin et al. 2009). Our odor choice olfactometric assay using the freshly crushed host plant leaves revealed *P. guajava* as the most strongly preferred while *D. erecta* and *M. malabathricum* as the moderately preferred alternate host plants. For a species of phytophagous insect, all host plants are not equally seductive to it and hence, on the basis of observed preference host plants may be arranged in rank (Singer et al. 1992). The host plant preference of stink bug, *Sinopla perpunctatus* (Hemiptera: Acanthosomatidae) was studied by Pietrantuono et al. (2014) and they gave ranking to two most preferred host plant of this insect species. Therefore, in the context of the odor preference test of the present study and analysis we concluded that, *P. guajava* can be considered as the most preferred alternate host plant for *H. theivora*.

Comparison of growth inhibiting and promoting effects of plant chemicals, such as primary metabolites and allelochemicals particularly in the case of insects with plant-sap sucking mouthpart is difficult and for such studies were introduced two cost effective tools, GI and RGI (Zhang et al.1993). In our study, when we examined the growth of *H. theivora* in third generation on the different host plants, the GI on *D. erecta* was closer to *C. sinensis* and the RGI was the maximum in *D. erecta* followed by *P. guajava*. In our earlier study concerning preference of *H. theivora* for alternate host plants among *D. erecta, L. inermis* and *S. tora*, we recorded *D. erecta* as the most preferred after *C. sinensis* (Borthakur et al.2016). Further, we recorded that, the survivorship of *H. theivora* on *P. guajava* was lower as compared to *C. sinensis* and *D. erecta*, but in case of *P. guajava* it enhanced in the third generation as compared to the first generation (Fig 3) (Borthakur et al.2016).

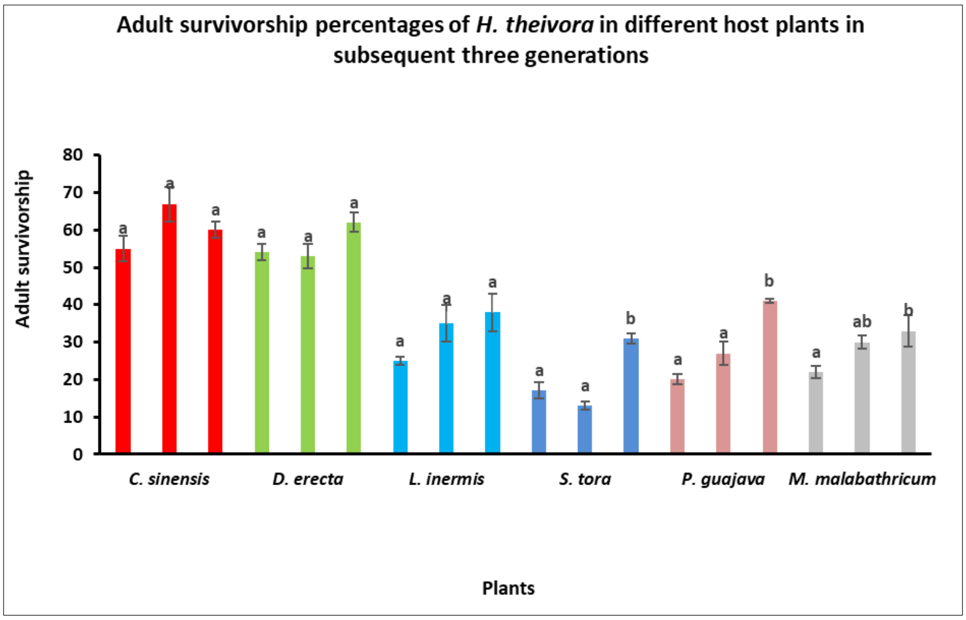

Figure 3 Graphical representation of adult survivorship percentages of *H. theivora* in *C. sinensis* (red bars), *D. erecta* (light green bars), *L. inermis* (light blue bars), *S. tora* (light blue-gray bars), *P. guajava* (light orange bars) and *M. malabathricum* (light gray bars) in three subsequent generations. Different letters given above the bars represent significant differences of adult survivorship percentage within three generations of *H. theivora* in each of the host plant ($p<0.05$) (Post hoc tests, LSD and Tukey)

Herbivores select host plants based on biophysical and chemical profiles for feeding, oviposition, or sustenance. While selecting the plant, the herbivorous insect may use the principle of preference –performance hypothesis, or alternately the female may take her choice decision to maximize her fitness rather than the fitness of her offspring (Prager et al. 2014). The chemical factors satisfy the insect's demand for nutrients and may act as phagostimulant, inhibitor, oviposition attractant, or deterrent through the participation of the insect's chemosensory and gustatory receptors. The effectiveness of the compound(s) depends on both its quantity and quality. In our study, we examined the metabolites considered as critical for plant-insect interaction in many earlier studies. We detected the highest sugar concentration in *P. guajava* with no significant difference among the plants except *S. tora* in which it was the lowest. Based on RGI calculated for three generations of *H. theivora*, we detected the poorest performance of the insect on *S. tora*. Similarly, *D. erecta* had high sugar content and supported *H. theivora* with the highest RGI. Therefore, the soluble sugar content could be a crucial factor for the growth of *H. theivora*. Earlier studies have also shown soluble sugars to be one of the main factors for hemipteran in host plant acceptance as food. The host plants use low sugar content as a mechanism of resistance against pests (Golan and Najda 2011). Therefore, the soluble sugar was a crucial factor for host acceptance and eventually for the growth of *H. theivora*.

There is a consensus agreement that terpenoid production and emission is tightly associated with trophic interactions (Boncan et al. 2020). Terpenoids act as oviposition stimulants in herbivores and some can also regulate reproductive maturation in insects (McClure and Hare 1984; Awmack and Leather 2002; Van 2007). In our study, the Post hoc (LSD) analysis revealed no significant difference between the terpenoid amount of *C. sinensis* and *P. guajava.* In the present study, *P. guajava* supported *H. theivora* with the second and third highest RGI respectively in the third and all the three generations of *H. theivora*. *M. malabatrichum* possessed the lowest amount of total terpenoid and *H. theivora* showed the lowest performance on the plant in terms of RGI of the third generation. *L. inermis* with the highest level of terpenoids, supported *H. theivora* with the third highest and second highest RGI in third and in all the three generations respectively. The results suggested that the terpenoids may be involved in growth performance of *H. theivora*. Thus, terpenoids could serve as determining factors for the host suitability of *H. theivora* for either oviposition or sustenance or both.

That phenolics adversely affect growth and oviposition of hemipteran insect is well studied (Lattanzio et al. 2000). But there is no rule of thumb for effects of contents of natural flavonoids and chlorogenic acids against insect herbivores. They may stimulate, inhibit feeding, oviposition and growth of insect herbivores. Phenolic acids extracted from black currant, sour cherry, walnuts and green husks prolonged the prereproductive period of *S. avenae* and reduced their daily fecundity (Chrzanowski et al.2012). But, the chlorogenic acid is reported to act as phagostimulant (Simmonds 2001). Rutin at a moderate dose acted as phagostimulant to *Schistocerca americana* without influencing the growth pattern (Bernays et al. 1991). In an electropenetrographic study rutin and quercetin are reported to enhance phloem probing activity of *Myzus persicae* (Stec et al.2021). In our study, the polyphenol content was significantly high in *C. sinensis* (9.02%) and the amount is comparable with the *C. sinesis* var *assamica* (12%) grown in Mississippi (Zhang et al.2020). The flavonoid contents of alternate host plants except for *S. tora* statistically resembled that of *C. sinensis*.

The present study has revealed two information,

- *P. guajava* is a strongly preferred alternate host plant for *H. theivora* followed by *D. erecta* in terms of odor preference and sustainability. The information generated from the study can be utilized in trap cropping approaches for management of *H. theivora* and
- The metabolites flavonoids, phenolic acid, terpenoids and soluble sugar content of the host plants may influence the host preference by *H. theivora* for feeding and reproduction. The findings if are used for electroantennographic study of *H. theivora*, can provide highly valuable information for integrated management of *H. theivora*.

5. Conclusion

The present study revealed that *H. theivora* preferred *C. sinensis* and *P. guajava* the most among the other host plants in olfactometric bioassay. The soluble sugar, total flavonoids and terpenoids are reflected as the cues for host preference by *H. theivora*. This indicates that in future, there is a scope for electroantennographic assessment. The alternate host preferred by *H. theivora* could be a basis of formulating new management strategy to control the pest.

Compliance with ethical standards

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Disclosure of conflict of interest

The authors have no conflict of interest.

Contribution of author

The authors equally contributed to the manuscript.

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