



E-ISSN: 2320-7078
P-ISSN: 2349-6800
JEZS 2017; 5(6): 97-103
© 2017 JEZS
Received: 17-09-2017
Accepted: 18-10-2017

Purnima Das
Assistant Professor, Department
of Entomology, Assam
Agricultural University,
Jorhat, Assam, India

Surajit Kalita
Assistant Professor, Department
of Entomology, Assam
Agricultural University,
Jorhat, Assam, India

Lakshmi Kanta Hazarika
Professor and Head, Department
of Entomology, Assam
Agricultural University,
Jorhat, Assam, India

Tea clonal preference by *Helopeltis theivora* (Hemiptera: Miridae)

Purnima Das, Surajit Kalita and Lakshmi Kanta Hazarika

Abstract

Helopeltis theivora Waterhouse (Miridae : Hemiptera) is a polyphagous insect having a characteristic diet preference, as revealed through *ex situ* and *in situ* screening of Tocklai Vegetative (TV) clones. Second instars caused the most feeding lesions (193.00 ± 8.91), but not the damage, amongst the developmental stages, and were capable of discriminating tested TV clones. Stadia of five instars were almost similar but were significantly different from adult longevity (15.58 to 18.25 days); adults exhibited not only female-biased sexual dimorphism in longevity but also of feeding potential. *In situ* screening resulted in identification of TV1 and TV7 as the susceptible clones, while TV6, TV12, TV14 and TV19 as resistant ones; these data were at par with that of *ex situ* screening in the laboratory, therefore, this method may be useful for large scale screening.

Keywords: tocklai vegetative clones, *Helopeltis theivora*, sexual dimorphism, feeding preference, feeding potential, *ex situ* resistance screening

1. Introduction

Tea, *Camellia sinensis* (L.) O. Kuntze (Family: Theaceae), is an intensively managed perennial monoculture crop cultivated on over 2.71 million ha in large- and small-scale plantations situated between latitudes 41°N and 16°S across Asia, Africa, Latin America, and Oceania^[1] and plays an important role in national economy of many of these countries. Globally, 1031 pest species attack tea, however, in recent years, mirids are causing havoc to the tea industry. Altogether 41 species of mirids in the genus *Helopeltis* (Hemiptera: Miridae) had been described in Asia, Australia, and Africa, out of which *H. theivora* Waterhouse, also called the tea mosquito bug (TMB)^[1], caused 11% to 100%^[2] crop loss in Asia. Being polyphagous, the nymphs and adults suck cell sap from tender stems, young leaves, and buds, forming reddish brown circular feeding lesions (FL). In severe infestations, damaged leaves with 76 to 210 FLs curl upward and desiccate and cause die back. TMB must have a preference for a particular diet, which needs to be evaluated and is an important aspect of host plant resistance (HPR). Research on is in progress in various tea research organizations around the world, which have been thoroughly reviewed^[1], where it was clearly shown that "Little progress was made on selection and breeding of pest resistant cultivars", though transgenic technology paved the way for developing a cultivar engineered with *rolb*, *Bt* and chitinase genes. Rigorous screening of the existing cultivars needed to understand reaction of clones to key pests so as to identify desirable moderately resistant clones as well as to know the mechanisms involved there. Tea cultivars are morphometrically and genetically variable^[2,3], and pests react differently^[1]. Our study was undertaken to see if TMB has a preference to categorise the Tocklai Vegetative clones into groups based on damage caused by *H. theivora* and also to describe the resistance mechanisms.

2. Materials and methods

2.1. Tea clones used

Thirty Tocklai Vegetative (TV 1 to TV 30) clones are maintained in the Experimental Garden for Plantation Crops (EGPC), Assam Agricultural University, Jorhat for the last 30 years. The top three leaves and the bud of each of the clones were used for experiments.

2.2. Mass culture of TMB

TMB adults were collected from the EGPC, Jorhat and were reared in the detached TV1 shoots each consisted of three leaves and a bud (Fig. 1) at $24 \pm 2^{\circ}\text{C}$, 85-90% RH and 12:12 L:D cycle

Correspondence
Lakshmi Kanta Hazarika
Professor and Head, Department
of Entomology, Assam
Agricultural University,
Jorhat, Assam, India

in the Physiology Laboratory, Department of Entomology, AAU, Jorhat. The shoots, on which eggs were deposited were isolated every morning and incubated for hatching. Immediately after hatching, 1st instars were maintained by providing fresh TV1 shoots. Likewise, different instars and adults were also maintained, which were further utilized for screening of TV clones.

2.3. Feeding potential assessment

Three freshly detached shoots of TV1 clone were wrapped with cotton and placed in a glass vial (Make : General; Size: 4 x 7 cm), partially filled with sterilized double distilled water to keep the shoots afresh; each vial was placed on a 15 cm diameter Petri-plate, one 1st instar, immediately after emergence, was released to the shoots, which were caged with hurricane lantern glass chimney (Make: RM; Size: 9 cm x 20 cm), covered with muslin cloth to prevent its escape; this was replicated for six times. Mirid bug feeds on plant tissues by evacuating cell contents, thus produce feeding lesion (FL). After 24 hours, shoots were replaced with fresh ones until the 1st instar molts. Numbers of FL on the bud, 1st to 3rd leaf were recorded as well as diameter of the lesions daily. Mean diameter was calculated by taking ten samples from each replication and the area of each FL was determined by using the formula: Area (A) = πr^2 (where, r= radius [$\frac{1}{2}$ x diameter] of lesion). Likewise, data were taken for each developmental stage (2nd to 5th instar and adult male and female), which were of course not recorded during molting. Second to 5th instars and adult males were also assessed. Furthermore, total number of FLs on the bud and also on three leaves was recorded separately for each developmental stage.

Period taken to complete each stage (developmental period, DP), cumulative number of lesions and damaged area were also assessed for determining damage potential of each stage.

2.4. Ex-situ screening of TV clones

We further observed that 2nd instar could produce the highest FLs, therefore, it was used for screening 30 different TV clones in the laboratory. Thus, a freshly detached shoot of TV clones 1 to 30 was subjected to feeding by a single 2nd instar, under the similar experimental set up as described under section 2.3. Each clone was replicated three times and the number of FLs on three leaves and the bud were recorded after 24 hrs.

2.5. In-situ screening of TV clones

TV1 to TV19 clones are being maintained at EGPC, Jorhat. Each clone was maintained row wise in a block and thus there are 19 blocks. From each block, ten randomly selected tea bushes were plucked, out of which healthy and infested shoots were counted during the month of March, April, May and June at seven day interval consecutively for two years (2013 and 2014). TMB population build up with the onset of pre-monsoon showers coupled with growing of new shoots in the tea bushes, as such *in situ* screening was done during March to June. These data were converted to % shoot damaged and subjected to statistical analysis for *in situ* screening of TV clones.

The vegetative clones hereby tested for their reaction against TMB were rated based on infested % shoots [4] as given below.

Table 1

% infested shoots	Category
0	Immune (I)
1-10	Highly Resistant (HR)
11-20	Resistant (R)
21-35	Moderately Resistant (MR)
36-50	Susceptible (S)
51-100	Highly Susceptible (HS)

2.6. Statistical analysis

Data on feeding potentiality and *ex-situ* screening of TV clones to TMB were subjected to analysis of variance (ANOVA) using completely randomized block design. *In situ* reaction of TV clones to TMB were subjected to ANOVA using randomized block design. The data recorded on means of each experiments were compared and separated through DMRT using the SPSS computer statistical software (Ver. 20.0). Correlation between FL size and number was calculated as well as regression analysis was done between FL numbers and stage of the insect.

3. Results

3.1. Feeding potential

During the process of sap sucking from the three leaves and the bud, Table 2 shows how many lesions (FLs) were produced by the nymphs and adults of TMB along with the damaged area. Diameter of each of the FL differed significantly which ranged between 0.74 mm (1st instar) to 2.50 mm (Adult female) (Table 2). The 1st, 2nd and 3rd instars sucked the sap through 0.43 mm, 1.08 mm and 1.06 mm diameter FLs, respectively; whereas the 4th and 5th instars produced 2.34 mm and 3.17 mm diameter FLs, respectively; FLs were significantly bigger than those caused by 3rd instars

but smaller than those caused by adults. Cumulative FLs varied significantly between developmental stages, the order, however was 2nd instar (193.00) > 1st instar (185.67) > adult female (178.33) > adult male (171.67) > 4th instar (149.67) > 5th instar (147.33) and 3rd instar (134.83) and followed a regressive pattern (Table 2, Fig. 3) showing that a 2nd instar could produce the highest numbers of FL. Similarly, nacroded are varied significantly between developmental stages, which were arranged in a descending order : 1st instar (81.96 mm²) < 3rd instar (137.08 mm²) < 2nd instar (211.22 mm²) < 4th instar (363.09 mm²) < 5th instar (465.96 mm²) < adult male (797.52 mm²) < adult female (886.44 mm²) (Table 2).

3.2. Ex-situ screening of TV clones

Data on *ex-situ* feeding preference of 2nd instar TMB over 24 hrs on different clones (TV1 to TV30) are presented in Table 3. Based on site of feeding it was observed that the 1st leaf was the most preferred, followed by 2nd leaf, the bud and the 3rd leaf. Amongst the 30 clones, TV1 was the most preferred on which 75.33 FL/day, whereas TV6 was the least preferred recording 14.17 FLs. Remaining clones reacted differently, number of lesions caused to each of the clones varied significantly (Table 3). Among the screened TV clones, none of them was found to be completely resistant against TMB;

most were susceptible to *H. theivora* under no choice condition.

3.3. *In-situ* screening of TV clones

Field infestation data of TMB on TV1 to19 were presented in Table 4 during March, April, May and June, 2013 and 2014. Analysis of pooled data revealed that there was a month wise variation of infestation, June being the period on which highest infestation was observed in all the tested clones. Clone wise infestation was also significantly different during various months. Based on the per cent shoot infestation (Fig. 3), TV6, TV12, TV14 and TV19 were the least preferred, whereas TV1 and TV7 were the most preferred. Based on the Kavitha and Reddy (2012) [4], these clones can be rated under three categories - resistant (TV6, TV12, TV14 and TV19), moderately resistant (TV2, TV3, TV4, TV5, TV8, TV9, TV11, TV13, TV15, TV16, TV17, TV 18) and susceptible (TV1 and TV7) (Table 4).

4. Discussion

Every instar, 1st to 5th of TMB, maintains a relatively steady DP ranging between 2.38 to 2.88 days (Table 2), which is an interesting deviation from the normal rule of growth and development, and is perhaps a common phenomenon in mirids reared in the laboratory [5-7]. No studies have detailed information on time taken by each instar, this is the first report of this kind. DP of each instar differed significantly with the adult longevity (AL) of both the sexes, however, sexual dimorphism with respect to AL in the TMB is also evident, adult female being lived significantly longer (18.25 days) than its counterpart (15.58 days). Sexual dimorphism biased towards the fair sex is common in insects [8] including plant sucking bugs. After eggs being fertilized, they are to be laid by females in batches, therefore, adult female TMBs have to live longer than the adult males to breed successfully.

TMB nymphs and adults can evacuate cell sap through pectinase-lyased lesions on the cell walls without breaking them. Hence, mirid lesions do not alter cell shape [9]. In addition, they inject lipase and proteinase into the plant tissues and suck the sap through the stylet, as a result a FL is formed on the feeding site, diameter of FL corresponds with the stage, suggesting bigger or older the individual, larger is the FL diameter, adults having the largest area (4.70 mm² to 4.97 mm²) followed by the 5th instar (3.17 mm²) (Table 2)

Length of the stylet also plays an important role in the FL formation and FL-area, because an adult has a longer stylet and can extend the same to penetrate many cells at a time resulting larger necrotic area of almost 5 mm² and produces 171.67 to 178.33 FLs but correspondingly fewer numbers.

FL numbers varies significantly between stages. On a regression analysis performed between stage and numbers of FL, we found a significant negative regression ($r^2= 0.5675$, $df=5$, $p=0.05$) (Fig. 2), which proves our hypothesis that older the insect, lesser the numbers of lesions caused. Number-wise FLs caused by the 2nd instar was significantly the highest (193.0) amongst the stages. Cumulative area necrosed (CAN) represents the damaging capacity of a stage based on which stage specific feeding potential (FP) is determined. But the CAN of the 4th and 5th instar was the largest (363.09 to 465.96 mm², respectively). It is expected that as the nymph grows, the stylet also lengthens; as such it spends longer time per probe that is true for adults also. Further it is interesting to note significant difference of CAN between two sexes, compared to adult males, females are more destructive, which might be related to higher longevity of the adult females and

to their need of high protein requirements for egg production [13, 14]. We have studied stage-wise DP and FP of *H. theivora*, which were not considered in earlier studies [10-12]. Based on FL-numbers, we found that 2nd instar is the most FL producing stage; therefore, it was utilized for *ex situ* screening of clones. Data presented in Table 3 clearly suggests that 2nd instar prefers the 1st leaf > 2nd leaf > leaf bud > 3rd leaf under no choice situation. This suggests existence of stage specific variability in feeding site preference in order to reduce the intraspecific competition and thus may result in resource partitioning [15, 16].

Reaction of TV clones to TMB 2nd instar was significantly different from one clone to another, which might be because of physical characteristics of respective clone as well as presence of polyphenols, other resistant conferring compounds and genes. Our present observation revealed that TV1 was the susceptible clone [17, 18, 1]. Further there is a discrepancy between our present and earlier studies that we found TV9, TV22, TV25 and TV26 to be preferred next to TV1 (Table 3), whereas Sundaraju and Babu (1999) designated them as more susceptible [17]; likewise, TV12 and TV23 were recognized as the most susceptible [12], but our data suggested these to be less preferred over TV1. This kind of variable results demands designing a full proof common screening technique. It is of course true that variation in plant resistance to insects exists which depends on species of the insect, plant materials and the environment. Plant tissues behave differently towards insect attack, phenology being the main player; in some cases, vegetative stages being succulent are susceptible to one group of insect; but resistant to others because younger leaves are containing higher mono- and poly-phenols than older leaves. It might due to the fact that TMB infestation led to decrease in phenylalanine ammonia lyase activity and polyphenol content [1]. Moreover, leaf structure and texture of the clones play roles in conferring resistance against TMB. TV1 was found to be mostly preferred on the basis of FL-inflicted on the 1st, 2nd and leaf bud as well as cumulative FLs. TV6 stood out be least preferred, which had already been recognized as one of the moderately resistant clone against tea pests including red spider mite [19, 1]. This shows that TMB can choose its own brand.

Laboratory screening data were further confirmed with *in situ* screening as such this study has a lot of significance in TMB host plant resistance studies in order to design management strategy. Field or *in situ* screening of 19 TV clones showed that clones had reacted differently towards TMB attack. Clearly TV1 and TV7 are the susceptible clones, while TV6, TV12, TV14 and TV19 are resistant. We found that there are no clones under immune and highly resistant categories, there are some clones which can be placed under moderately resistant. *Ex situ* and *in situ* screening data are almost at par especially with respect to the most preferred and least preferred clones, therefore, the *ex situ* screening technique we followed can be considered as a standard technique for screening. It can further perhaps be reinforced by associating it with molecular techniques to identify RAPD and SCAR markers [20, 21] to ease the problems associated with *in situ* screening. This kind of comparative *in situ* and *ex situ* assessment of resistance is important to confirm that TMB has a choice. Furthermore, since *ex situ* screening corresponds with that of the *in situ*. At present host plant resistance (HPR) programme in tea is very weak, and fails to understand the mechanism of resistance as well as to identify resistance conferring genes. Another aspect which has not been

attempted is “describing likely changes in proteomes from insects in response to cultivar switching and insect resistance

management”^[1], which needs further studies in order “to add new dimensions to the HPR programmes in tea”^[1].

Table 2: Damage potentiality of *TMB* on TV1 clone under no-choice situation in the laboratory

Developmental stage	Mean ± SEM				
	Developmental period (days) (a)	FL			Cumulative necrosed area / stage (mm ²) (a x c x d)
		\$ Diameter (mm) (b)	\$ Area / FL (mm ²) (c)	Nos./stage (d)	
1st Instar	2.88±0.11 ^c	0.74±0.03 ^e	0.43±0.04 ^e	185.67±6.82 ^{ab}	81.96±9.92 ^e
2nd Instar	2.38±0.11 ^c	1.16±0.06 ^d	1.08±0.12 ^d	193.00±8.91 ^a	211.22±30.00 ^d
3rd Instar	2.58±0.15 ^c	1.11±0.15 ^d	1.06±0.30 ^d	134.83±8.60 ^c	137.32±31.45 ^{de}
4th Instar	2.46±0.19 ^c	1.71±0.11 ^c	2.34±0.28 ^c	149.67±22.67 ^{bc}	363.09±99.49 ^{cd}
5th Instar	2.50±0.17 ^c	2.01±0.03 ^b	3.17±0.10 ^b	147.33±8.95 ^{bc}	465.96±27.14 ^c
Adult (Male)	15.58±0.26 ^b	2.45±0.04 ^a	4.70±0.16 ^a	171.67±17.72 ^{abc}	797.52±65.97 ^b
Adult (Female)	18.25±1.15 ^a	2.50±0.11 ^a	4.97±0.18 ^a	178.33±11.19 ^{ab}	886.44±109.18 ^a
SEd	0.38	0.07	0.20	10.84	52.50
CD (P=0.01)	0.74	0.14	0.39	21.24	102.89
CD (p=0.05)	1.91	0.37	1.02	54.59	264.44

- ^s Data presented are the mean of 60 samples ; FL, Feeding lesion
- Mean data were compared by Turkey Test (P<0.05)
- Means followed by same letter are not significantly different

Table 3: Reaction of TV clones to 2nd instar *TMB* (pooled data of 2013 and 2014)

TV Clones	Mean ± SEM numbers of FL/day				
	1st Leaf	2nd Leaf	3rd Leaf	Leaf Bud	Total
TV1	43.67±1.67 ^a	20.67±1.17 ^a	0.67±0.33 ^a	10.33±2.60 ^a	75.33±3.37 ^a
TV2	29.00±0.76 ^{bc}	9.33±1.69 ^{defg}	0.67±0.67 ^a	1.67±0.88 ^{cd}	40.67±1.30 ^{bcd}
TV3	31.83±2.33 ^{bcde}	7.00±0.50 ^{fg}	1.17±0.60 ^a	3.33±1.67 ^{bcd}	43.33±2.46 ^{bcd}
TV4	22.00±0.58 ^{efghijk}	20.33±0.73 ^a	0.00±0.00 ^a	1.67±0.88 ^{cd}	44.00±1.32 ^{bcd}
TV5	33.50±0.76 ^{bc}	8.33±0.83 ^{efg}	1.00±1.00 ^a	0.33±0.17 ^d	43.17±1.17 ^{bcd}
TV6	8.00±1.00 ^m	5.00±1.61 ^g	1.17±0.73 ^a	0.00±0.00 ^d	14.17±3.18 ^h
TV7	11.00±1.76 ^{klm}	11.00±1.89 ^{cdef}	0.00±0.00 ^a	0.00±0.00 ^d	22.00±3.55 ^{fgh}
TV8	12.67±1.76 ^{ijkl}	20.83±0.17 ^a	1.00±1.00 ^a	0.67±0.67 ^d	35.17±2.13 ^{cdef}
TV9	12.33±0.17 ^{ijklm}	7.00±1.04 ^{fg}	0.00±0.00 ^a	3.17±0.60 ^{bcd}	22.50±1.26 ^{efgh}
TV10	32.00±1.15 ^{bcde}	11.17±1.17 ^{cdef}	0.00±0.00 ^a	1.00±0.58	44.17±1.09 ^{bcd}
TV11	35.67±1.45 ^{bcd}	9.83±0.60 ^{defg}	0.00±0.00 ^a	3.33±2.03 ^{bcd}	48.83±1.59 ^{bc}
TV12	42.00±2.50 ^{bc}	10.33±1.20 ^{defg}	0.00±0.00 ^a	0.00±0.00 ^d	52.33±1.36 ^b
TV13	11.67±1.01 ^{klm}	9.17±2.46 ^{defg}	0.00±0.00 ^a	9.00±1.26 ^{ab}	29.83±3.90 ^{defgh}
TV14	26.33±0.88 ^{defgh}	9.17±1.86 ^{defg}	0.00±0.00 ^a	3.33±1.67 ^{bcd}	38.83±1.20 ^{bcde}
TV15	27.33±1.20 ^{cdef}	12.00±1.76 ^{cdef}	1.00±1.00 ^a	0.50±0.50 ^d	40.83±1.48 ^{bcd}
TV16	23.33±1.33 ^{efghi}	12.67±1.45 ^{bcdef}	1.33±0.88 ^a	2.00±1.00 ^{cd}	39.33±0.88 ^{bcd}
TV17	28.67±1.17 ^{bcde}	9.50±2.29 ^{defg}	0.00±0.00 ^a	3.33±0.33 ^{bcd}	41.50±3.69 ^{bcd}
TV18	24.17±3.68 ^{bcdef}	16.33±2.62 ^{abc}	0.00±0.00 ^a	3.33±0.83 ^{bcd}	43.83±3.09 ^{bcd}
TV19	13.67±1.59 ^{hijklm}	13.33±1.33 ^{bcde}	0.00±0.00 ^a	2.00±1.15 ^{cd}	29.00±3.33 ^{defgh}
TV20	17.67±1.20 ^{klm}	8.33±1.64 ^{efg}	0.83±0.44 ^a	3.00±0.58 ^{cd}	29.83±1.42 ^{fgh}
TV21	14.67±1.33 ^{lm}	9.67±2.62 ^{defg}	1.67±0.60 ^a	1.00±1.00 ^{cd}	27.00±2.84 ^{fgh}
TV22	23.00±3.55 ^{hijkl}	17.67±2.46 ^{ab}	1.67±0.73 ^a	6.50±1.04 ^{ab}	48.83±6.22 ^{bc}
TV23	14.67±3.09 ^{lm}	11.50±2.65 ^{cdef}	0.83±0.44 ^a	1.67±1.20 ^{cd}	28.67±4.28 ^{defgh}
TV24	10.00±2.31 ^{lm}	9.50±2.89 ^{defg}	0.00±0.00 ^a	1.33±0.67 ^{cd}	20.83±5.78 ^{fgh}
TV25	17.00±1.15 ^{ghijkl}	10.33±0.88 ^{defg}	0.17±0.17 ^a	1.67±0.88 ^{cd}	29.17±1.01 ^{defgh}
TV26	26.00±1.61 ^{cdef}	11.17±1.09 ^{cdef}	0.17±0.17 ^a	3.67±0.67 ^{bcd}	41.00±2.02 ^{bcd}
TV27	12.33±0.44 ^{ijklm}	14.50±2.08 ^{bcde}	1.33±0.67 ^a	2.67±0.73 ^{cd}	30.83±3.38 ^{defg}
TV28	19.00±2.52 ^{ghijk}	11.67±1.92 ^{cdef}	1.33±0.67 ^a	3.00±1.53 ^{cd}	35.00±5.07 ^{cdef}
TV29	13.50±2.18 ^{ijkl}	7.83±0.60 ^{efg}	0.00±0.00 ^a	0.33±0.33 ^d	21.67±1.76 ^{fgh}
TV30	19.67±4.67 ^{ghijkl}	7.33±1.45 ^{fg}	0.00±0.00 ^a	2.67±0.33 ^{cd}	29.67±2.96 ^{defgh}
SEd	2.28	1.98	0.58	1.21	3.43
CD (P=0.01)	4.46	3.88	NS	2.37	6.73
CD (p=0.05)	11.417	9.97	NS	6.08	17.29

- Data presented are the mean of 3 replications
- Mean data were compared by Turkey Test (P<0.05)
- Means followed by same letter are not significantly different

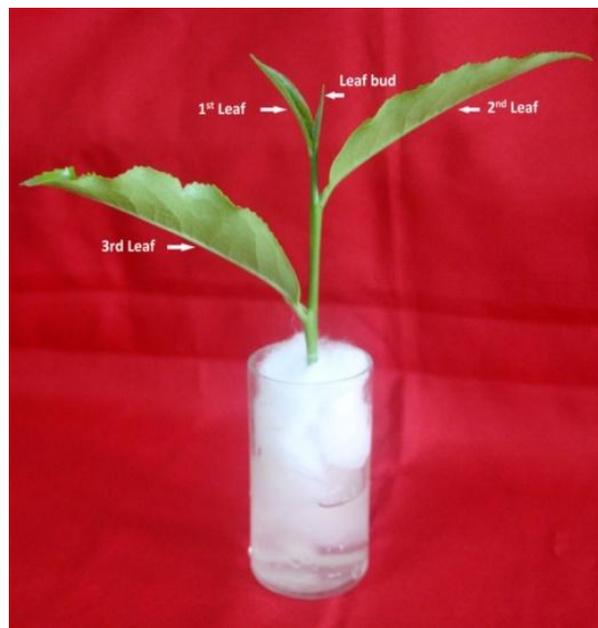
Table 4: *In situ* screening of TV clones against TMB (Pooled data of 2013 and 2014)

TV Clones	Shoot infestation (Mean % \pm SEM)			
	March	April	May	June
TV1	21.14 \pm 1.33 ^a	34.94 \pm 0.83 ^a	37.46 \pm 1.56 ^c	47.19 \pm 1.43 ^a
TV2	14.04 \pm 1.11 ^{defg}	21.88 \pm 0.37 ^{cdefg}	30.38 \pm 1.16 ^{cd}	45.94 \pm 1.61 ^{bcd}
TV3	13.38 \pm 0.30 ^{defg}	17.70 \pm 0.78 ^h	26.52 \pm 1.06 ^{ef}	44.84 \pm 1.61 ^{bcd}
TV4	18.91 \pm 1.02 ^{abc}	24.58 \pm 0.56 ^{bc}	35.75 \pm 1.10 ^{bc}	49.51 \pm 1.20 ^{ab}
TV5	14.44 \pm 1.12 ^{cdefg}	21.97 \pm 0.75 ^{cdef}	39.08 \pm 19.45 ^b	41.56 \pm 0.74 ^{de}
TV6	13.89 \pm 0.50 ^{defg}	18.00 \pm 0.92 ^{gh}	17.44 \pm 1.11 ^{hi}	23.65 \pm 0.97 ⁱ
TV7	16.44 \pm 0.91 ^{abcde}	25.03 \pm 0.58 ^{bc}	41.71 \pm 1.31 ^a	42.19 \pm 0.96 ^{abc}
TV8	13.29 \pm 0.70 ^{defg}	22.00 \pm 0.71 ^{cdef}	26.34 \pm 0.60 ^{ef}	41.22 \pm 0.81 ^{de}
TV9	17.28 \pm 1.22 ^{abcde}	20.38 \pm 0.45 ^{defgh}	25.99 \pm 0.62 ^{efg}	37.86 \pm 1.32 ^{ef}
TV10	15.16 \pm 0.83 ^{bcdef}	20.85 \pm 0.97 ^{cdefgh}	29.71 \pm 0.78 ^e	41.41 \pm 1.08 ^{de}
TV11	19.76 \pm 0.85 ^{ab}	19.32 \pm 0.77 ^{efgh}	20.29 \pm 0.78 ^{ghi}	26.69 \pm 1.11 ^{hi}
TV12	10.19 \pm 0.50 ^f	21.19 \pm 0.38 ^{cdefh}	15.91 \pm 0.94 ⁱ	25.75 \pm 0.58 ⁱ
TV13	16.20 \pm 0.22 ^{bcde}	22.92 \pm 0.72 ^{cde}	23.83 \pm 1.19 ^{fgh}	41.97 \pm 1.10 ^{cde}
TV14	10.39 \pm 0.52 ^{fg}	21.69 \pm 1.01 ^{cdefg}	20.79 \pm 1.57 ^{ghi}	24.81 \pm 0.43 ⁱ
TV15	17.83 \pm 1.07 ^{abcd}	19.15 \pm 0.92 ^{efgh}	26.01 \pm 1.03 ^{efg}	31.63 \pm 0.96 ^{gh}
TV16	12.84 \pm 0.85 ^{efg}	23.74 \pm 0.82 ^{bcd}	30.75 \pm 1.15 ^{cd}	42.50 \pm 0.44 ^{cde}
TV17	18.06 \pm 0.83 ^{abcd}	27.47 \pm 0.60 ^b	20.20 \pm 0.84 ^{ghi}	33.53 \pm 0.65 ^{fg}
TV18	12.53 \pm 0.93 ^{efg}	21.91 \pm 0.85 ^{cdef}	25.66 \pm 0.38 ^{efg}	22.69 \pm 1.28 ⁱ
TV19	15.13 \pm 1.61 ^{bcdef}	18.94 \pm 0.73 ^{fgh}	21.51 \pm 0.60 ^{fghi}	25.58 \pm 0.79 ⁱ
S.Ed.	1.00	0.90	0.95	1.42
CD (P=0.01)	1.97	1.76	1.86	2.78
CD (p=0.05)	5.05	4.53	4.78	7.16

- Data presented are the mean of 40 samples per month
- Mean pooled data were compared by Turkey Test (P<0.05)
- Means followed by same letter are not significantly different

Table 5: Rating of TV clones based on *in situ* screening against TMB

Group Name	TV clones
Immune (I)	0
Highly Resistant (HR)	0
Resistant (R)	TV6, TV12, TV14, TV19
Moderately Resistant (MR)	TV2, TV3, TV4, TV5, TV8, TV9, TV11, TV13, TV15, TV16, TV17, TV 18
Susceptible (S)	TV1, TV7
Highly Susceptible (HS)	0

**Fig 1:** Detached shoot with three leaf and a bud

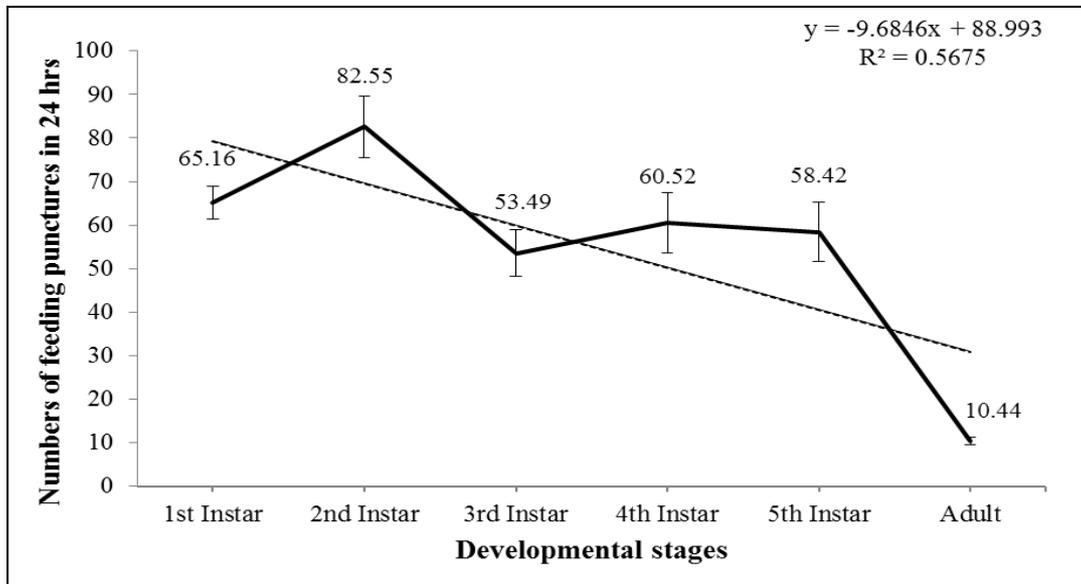


Fig 2: Developmental stage-wise damage potential of TMB

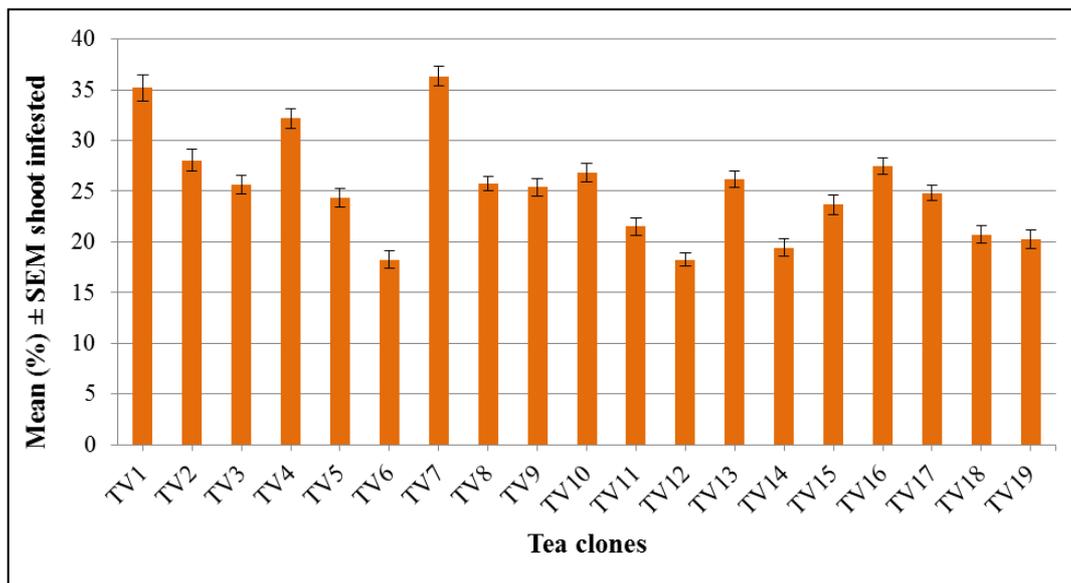


Fig 3: In situ screening of TV clones against *Helopeltis theivora* (pooled data of 2013 and 2014)

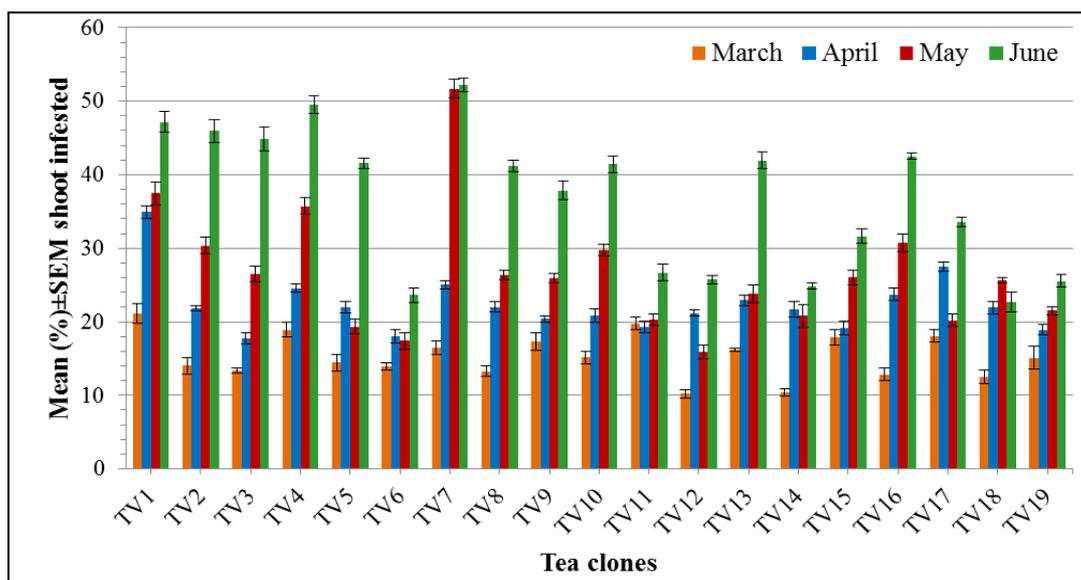


Fig 4: In situ screening of TV clones against *Helopeltis theivora* based on average of March, April, May and June (2013 and 2014)

5. Conclusion

TMB, *Helopeltis theivora*, 2nd instar caused the highest number of feeding lesions, thus it was utilized for *ex situ* screening of tea clones. It revealed that TV1 and TV7 were susceptible to TMB, while TV6, TV12, TV14 and TV19 were moderately resistant, similar results were obtained from the field (*in situ*) screening as well. Thus the latter group can be utilized for HPR studies as well as may be included as a strategy in designing a tea IPM programme.

6. Acknowledgment

The authors are thankful to the AAU authority for providing necessary facilities and fund against conduct of research on tea pest management. We also thank Dr. Randy Gaugler, Centre for Vector Biology, Rutgers University, New Brunswick for reading the manuscript and for his suggestions.

7. Reference

- Hazarika LK, Bhuyan M, Hazarika BN. Insect pests of tea and their management. Annual Review of Entomology. 2009; 54:267-84.
- Muraleedharan N. Pest control in Asia. In: KC Wilson, MN Clifford (eds.). Tea: Cultivation to Consumption, Chapman & Hall, London. 1992, 375-411.
- Banerjee B. Botanical classification of tea. In: KC Wilson, MN Clifford (eds.). Tea: Cultivation to Consumption. Chapman & Hall, London. 2006, 25-51.
- Kavita K, Reddy KD. Screening techniques for different insect pests in crop plants. International Journal of Bioresource and Stress Management. 2012; 3:188-195.
- Reyes TM, Gabriel BP. The life history and consumption habits of *Cyrtorhinus lividipennis* Reuter (Hemiptera: Miridae). Philippine Entomologist. 1975; 3:79-88.
- Wheeler AG. Biology of the plant bugs (Hemiptera : Miridae): Pests, predators, opportunist. Cornell University Press. 2001, 507.
- Udikeri SS, Kranthi KR, Patil SB, Modagi SA, Vandal NB. Bionomics of mired bug, *Creontiades biseratense* (Distant) and oviposition pattern in *Bt* cotton. Karnataka Journal of Agricultural Sciences. 2010; 23:153-156.
- Nunn CL, Lindenfors P, Pursall ER, Rolff J. On sexual dimorphism in immune function. Philosophical Transactions of the Royal Society of London Series B: Biological Sciences. 2009; 364:61-69.
- Miles PW. Plant-sucking bugs can remove the contents of the cells without mechanical damage. Experientia. 1987; 43:937-939.
- Baurah M, Hazarika LK, Ahmed B, Kalita S. Effect of *Beauveria bassiana* (Bals.) Vuill. on feeding and growth of *Helopeltis theivora* Waterhouse (Hemipetra : Miridae). Journal of Agriculture Science Society of NE India. 2006; 46:23-26.
- Bhuyan M, Bhattacharyya PR. Feeding and oviposition preference of *Helopeltis theivora* (Hemiptera: Miridae) on tea in Northeast India. Insect Science. 2006; 13:485-488.
- Roy S, Muraleedharan N, Mukhapadhyay A, Handique G. The tea mosquito bug, *Helopeltis theivora* Waterhouse (Heteroptera: Miridae): its status, biology, ecology and management in tea plantations. International Journal of Pest Management. 2015; 61:179-197.
- Smith CM, Khan ZR, Pathak MD. Techniques for evaluating insect resistance in crop plants. Lewis Publishers, Boca Raton, New York. 1994, 321.
- Awmack CS, Leather SR. Host plant quality and fecundity in herbivorous insects. Annual Review of Entomology. 2002; 47:817-844.
- Hazarika LK, Deka M, Bhuyan M. Oviposition behaviour of the rice hispa *Dicladispa armigera* (Coleoptera: Chrysomelidae). International Journal of Tropical Insect Science. 2005; 25:50-54.
- Radhika V, Kost C, Bartram S, Heil M, Boland W. Testing the optimal defence hypothesis for two indirect defences: extrafloral nectar and volatile organic compounds. Planta. 2008; 228:449-457.
- Sundararaju D, Sundara Babu PC. *Helopeltis* spp. (Heteroptera: Miridae) and their management in plantation and horticultural crops of India. Journal of Plantation Crops. 1999; 27:155-74.
- Roy S, Mukhopadhyay A, Gurusubramanian G. Varietal Preference and Feeding Behaviour of Tea Mosquito Bug (*Helopeltis theivora* Waterhouse) on Tea Plants (*Camellia sinensis*). Academic Journal of Entomology. 2009; 2:01-09.
- Hazarika LK, Sharma M, Saikia MK, Borthakur M. Biochemical basis mite resistance in tea. In: Proceedings of National Conference on Insect Biochemistry and Molecular Biology, Trivandrum, 1995.
- Saha D, Mukhopadhyay A, Bahadur M. Effect of host plants on fitness traits and detoxifying enzymes activity of *Helopeltis theivora*, a major sucking insect pest of tea. Phytoparasitica. 2012; 40:433-444.
- Suganthi M, Senthilkumar P, Arvinth S, Rajkumar R, Chandrashekara KN. RAPD and SCAR markers linked to tea mosquito resistance in tea. Journal of Crop Improvement. 2014; 28:795-803.