Phytoparasitica (2021) 49:983–1002
https://doi.org/10.1007/s12600-021-00934-8

ORIGINAL ARTICLE

Insecticide resistance status of *Hyposidra talaca* (Lepidoptera: Geometridae) in major tea growing zone of India

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Received: 18 July 2020 / Accepted: 24 May 2021 / Published online: 14 June 2021

Abstract Management of the black looper *Hyposidra talaca* (Walker), the most demolishing foliage feeder of tea in Himalayan foothills, is based on the use of chemical insecticides, though poor field efficacy of various commercially formulated products has recently been reported. In the present study, insecticide resistance of *H. talaca* to some traditional and newer insecticides was evaluated from north-eastern tea growing belt of India. Six populations of *H. talaca* were collected in three consecutive years, 2017 to 2019, from five locations (Dooars, Kalimpong, Sikkim, Assam, and Darjeeling). These areas represent the major and commercially exportable best-quality Indian tea production belts. The Darjeeling and Assam populations showed low to very high lethal concentration ratios (LCRs) (16.67 to 140.32, respectively) to bifenthrin, deltamethrin and diflubenzuron while, highest LCR to quinalphos was observed in the Dooars population (119.81). Similarly, very low to extremely high LCRs to emamectin benzoate and flubendiamide (4.00 to 65.25 and 16.43 to 148.94, respectively) were observed in all six populations. However, pyridalyl (LCR ≤ 77.09) and spinetoram (LCR ≤ 82.03) showed higher toxicity than that of cyantraniliprole (LCR ≤ 120.98) to field populations of *H. talaca*, irrespective of locations. The pairwise correlation coefficients of log LC_{50} values revealed that emamectin benzoate was significant but negatively correlated with bifenthrin. LCRs to the tested insecticides were heterogeneous and highly variable among locations and years. Specific resistance management strategies should be established, especially in locations where *H. talaca* has developed

Key Message

- Scanty of information is available on *Hyposidra talaca* (Walker) resistance to insecticides.
- Resistance to outdated and novel insecticides was examined in different field populations collected in tea
- Extremely high levels of resistance against organophosphate and pyrethroid insecticides.
- Moderate to high resistance levels against emamectin benzoate and flubendiamide.
- Spinetoram and emamectin benzoate were less toxic compared to older molecules for populations collected from organic plantations.
- Varied levels of resistance with respect to locations and years.

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very high levels of resistance to newer chemistry insecticides.

Keywords Black looper · Susceptibility · Organophosphate · Pyrethroids · New molecules · Cross resistance

Introduction

The black inch worm or black looper *Hyposidra talaca* (Walker) (Lepidoptera: Geometridae) is considered one of the most destructive polyphagous pests to various forest flora species in Australasian tropics, covering China, India, New Guinea, Indonesia, Taiwan, Hong Kong, Malaysia, Thailand and Australia (Intachat et al., 2001; Mathew et al., 2005; Singh & Singh, 2004; Winotai et al., 2005). In 2006, *H. talaca* was first detected in tea [*Camellia sinensis* (L.) Kuntze] over the Sub-Himalayan zone of West Bengal (Das & Mukhopadhyay, 2008), but this pest has emerged as the major defoliator of tea plantations in entire North-East India by 2008 (Chutia et al., 2012; Das & Mukhopadhyay, 2009). The larval instars feed on both young and mature leaves of bushes, and under severe infestations, they cause up to 90 percent damage to crop (Basu Majumder, 2010). Beside various biological, environmental and agro-climatic issues, repeated use of synthetic pesticides is recognized as one of the major reasons of the invasion of tea plantations by this new looper pest, *H. talaca* (Das & Mukhopadhyay, 2014; Mukhopadhyay & Roy, 2009).

Based on occupation and production value, tea is the most important crop in the eastern Himalayan Terai-Dooars and Assam and Darjeeling foothills (Biggs et al., 2018). Tea plantations have occupied an area around 350,000 ha in the eastern Himalayan zone of India which is accountable for 70 per cent of the total national tea production (Gohain et al., 2012; Laskar & Thappa, 2015). Owing to the severity of *H. talaca* in North-Eastern tea plantations of India, the application of organophosphate (OP) and synthetic pyrethroid insecticides became the most effective way for the control of this pest (Basu Majumder et al., 2012; Sannigrahi & Talukdar, 2003). However, as a result of non-rational spray strategies and overuse of active ingredients with a similar mode of action, the efficacy of insecticide for black looper management is being threatened by the possibility of resistance development (Saha, 2016).

In the north-eastern tea growing regions, estate owners solely rely on chemical insecticides and can carry out up to 12 rounds of foliar applications from March to October to mitigate lepidopteran pests like *H. talaca* (Walker), *Buzura suppressaria* (Guenee), *Eterusia aedea* (Linnaeus), *Cydia leucostoma* Meyrick etc. (Gurusubramanian et al., 2008). As bifenthrin, deltamethrin, diflubenzuron and quinalphos are broad-spectrum insecticides, they have been extensively used for tea pests' management (Gurusubramanian et al., 2008). Indeed, as a result of their low market price, these molecules have been applied since 2008 for managing *H. talaca* on tea in North-East India. During the first years of utilization, reports of control failure have come since 2013 (Nain, 2015). With the approval by the Central Insecticide Board, Government of India, of new active ingredients such as emamectin benzoate and flubendiamide to control lepidopteran pests (NPATG, 2016; PPC, 2019), growers have new tools for a more rational insecticide control of *H. talaca*. Despite of this, there is a rising concern that insecticide resistance to the old and new generation insecticides become a common phenomenon (Roy et al., 2017; Saha, 2016). In this sense, a baseline of the resistance status of *H. talaca* field populations is essential for development insecticide resistance management strategies in the study but also other tea growing areas threaten by this pest. In the present study, resistance status to some conventional and newer insecticides, frequently used against lepidopteran caterpillars in the tea gardens, were determined for *H. talaca* of six field populations collected in Namchi, Tumsong, Kalchini, Kamalpur, Harishpur and Kumai.

Materials and methods

Insecticides

The commercial insecticides selected and tested were bifenthrin 7.9 SC (Bifen I/T, Oldham Chemicals Company Inc.), cyantraniliprole 10.26 w/w OD (Benevia, DuPont India Pvt. Ltd.), deltamethrin 2.8 EC (Decis, Bayer Crop Science), diflubenzuron 25 WP (Bi-Larv, Bayer Crop Science), emamectin benzoate 5 SG (Proclaim, Syngenta India), flubendiamide

480 SC (Fame, Bayer Crop Science), pyridalyl 10 EC (Sumipleo, Sumitomo Chemical Co. Ltd.), quinalphos 25 EC (Ekalux, Syngenta India), and spinetoram 11.7 SC (Delegate, Dow Agrosciences Pvt. Ltd.). Cyantraniliprole, pyridalyl and spinetoram are new generation biorational insecticides; emamectin benzoate and flubendiamide are recently approved new active ingredients against the tea-looper complex in India (MUP, 2020); and bifenthrin, deltamethrin, diflubenzuron and quinalphos are conventional molecules belonging to synthetic pyrethroid, insect growth regulator (IGR) and organophosphates (OP) insecticides, respectively.

Hyposidra talaca reference strain

An insecticide-susceptible population of *H. talaca* from Bidhan Chandra Krishi Viswavidyalaya (BCKV), Nadia, West Bengal, India, designated as Lab/Htal-IN, was employed as the reference strain in the present bioassays. This population was obtained by single-pair crosses using individuals collected from Sikkim University, Sikkim, India in 2014 and reared on green tea leaves in the insect and mite culture laboratory at 28 ± 2 ˚C temperature and 75–80% relative humidity with 14:10 (L:D) photoperiod for five years. This Lab/Htal-IN strain had never been exposed to the chemical insecticides. Thus, the mortality data obtained from laboratory bioassay were used as an allusion for baseline susceptibility of various insecticides.

Collection of field populations

Field populations of *H. talaca* at third to fifth instar larvae were collected from six different tea estates [Namchi tea garden, Ravangla (27.1012 ˚N, 88.2060 ˚E); Tumsong tea garden, Darjeeling (27.0353 ˚N, 88.1755 ˚E); Kalchini tea estate, Dooars (26.4920 ˚N, 89.5265 ˚E); Kamalpur tea estate, Darjeeling (26.7073˚N, 88.3085˚E); Harishpur tea estate, Namrup (27.2044 ˚N, 95.2656 ˚E); and Kumai tea estate, Kalimpong (26.9951 ˚N, 88.8287 ˚E)] in three major tea growing provinces (West Bengal, Assam, and Sikkim) of north-eastern India during 2017, 2018, and 2019 (Fig. 1). In each site, field individuals were collected by walking in a zig-zag pattern covering a 3 ha area (Tong et al., 2013). Collected individuals were taken to the BCKV laboratory. The larvae were reared separately under the laboratory condition on fresh and pesticide-free tender tea foliage in growth chambers at 28 ± 2 ˚C temperature, 75–80% relative humidity and 14:10 (L:D) photoperiod (Basu Majumder, 2010). The collected healthy larvae were individually transferred to tea shoots bearing 2–3 leaves and a bud with the help of a camel-hair brush. Each shoot placed in 50 ml Erlenmeyer flask (BRL_4980021, Borosil Glassworks Ltd.) containing pure water, was tightly plugged with absorbent cotton to retain it properly. The flasks were then preciously arranged in glass jars (diameter, 15 cm; height, 20 cm) tightly roofed with insect rearing nylon mesh, fitted with the help of a rubber band (9 flasks in each jar) and placed in the growth chambers. Replacement of shoots along with the cleaning of faecal matter was done after every 24 h and pupae were collected on alternate days. After the adult emergence, a pair of male and female moths were taken and taxonomic identification was carried out using the key of Hampson (1985) and Holloway (1993). The rest of adults were taken into oviposition chambers with nylon mesh sides for adequate ventilation and fed on a 10 per cent honey-protinex solution absorbed onto a clinical cotton ball. All the field-collected populations were fostered for at least one generation to obtain a homogenous population with sufficient numbers of larvae before the bioassays were conducted.

Bioassays

A standard "leaf disk bioassay" method was followed (Paramasivam & Selvi, 2017) by taking newly moulted 2nd instar larvae to all tested insecticides except diflubenzuron. Serial dilutions as mg L^{-1} of commercial formulations of the test insecticides were prepared using 0.1% Triton X-100 (a non-ionic wetting agent) in double-distilled water. Fresh tea leaves were clipped into circular pieces (5 cm diameter) and dipped in the serially diluted insecticide solutions for 10 s. Then the treated leaves were air-dried at room temperature for 10 min and placed with their abaxial surface upwards in each Petri plate lined with moistened Whatman no. 1 filter paper. Leaf disks that were dipped in sterile diluents only were used as controls. Ten 2nd instar larvae of *H. talaca* were introduced in each Petri plate and sealed with a

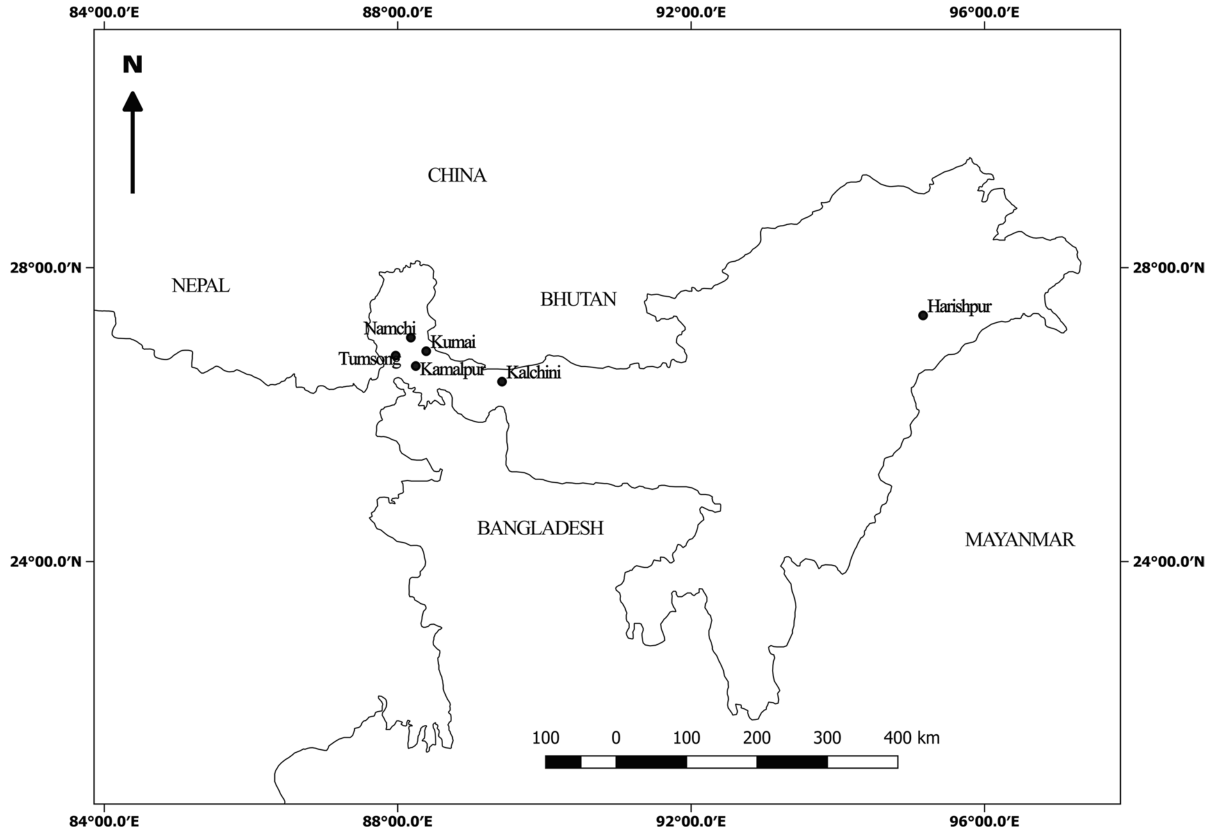

Fig. 1 Collection sites of *H. talaca* field populations from different tea plantations in north-east India

nylon mesh net. Nine different concentrations of each insecticide plus the control treatment were set. All the treatments were replicated four times to calculate the concentration mortality line (Das et al., 2010). The Petri plates were then kept at the aforesaid controlled conditions. Mortality data were recorded after 48 h exposure to bifenthrin, cyantraniliprole, deltamethrin, emamectin benzoate, flubendiamide, and quinalphos and after 72 h with pyridalyl and spinetoram. The "topical application" method of bioassay was employed for diflubenzuron by taking third instar larvae, just a day after moulting (Berry et al., 1993; Durmusoglu et al., 2015). Larvae were weighed and treated topically on the thoracic junctions with one of the nine dosages of diflubenzuron (10 larvae per concentration), ranging from 0.5–1200 mg L^{-1} and replicated four times. In the present study, 0.1 µl insecticide solution per mg larval body weight was applied, using a hand micro-applicator (Burkard Manufacturing Co. Ltd., UK) equipped with one 50-II micro-syringe (MS-N50; Shizuoka, Japan). Larvae treated similarly with the sterile solution only, served as control. Treated larvae were immediately placed on respective fresh tea shoots in 50 ml Erlenmeyer flask with all the aforementioned arrangements and monitored daily. Mortality of treated larvae was assessed when they failed to moult or did not react to soft piercing after emerging as adults by the survivors. Larvae were assumed to be dead if no coordinated movement was observed after probing with a soft hair brush (Gurusubramanian & Bora, 2007).

Data analyses

Concentration-mortality regressions for the Lab/Htal-IN population and each insecticide tested were estimated assuming a normal distribution of the binomial variable 'probability of response' based on probit analysis using PoloPlus statistical software version

2.0 (LeOra Software Company, USA). Same procedure was also followed in the concentration–response experiments for different field-collected populations and the tested insecticides. Lethal concentration ratios (LCR) (relative toxicity ratios) and 95% confidence limits were calculated for LC_{50} to compare changes in susceptibility between laboratory-susceptible (Lab/Htal-IN) and field strains (Vanaclocha et al., 2019). Lethal concentrations (LC_{50}) were considered significantly different when LCR confidence limits did not include 1 (Robertson et al., 2007).

Among the tested insecticides, a cross-resistance mechanism was estimated by pairwise correlation coefficients of log LC_{50} values of the field strains by the Pearson correlation through the XL-Stats computer program (Tong et al., 2013).

Results

Toxicity of insecticides to the *H. talaca* reference strain

The LC_{50} values of the nine insecticides for Lab/Htal-IN are depicted in Table 1. Toxicity was highest for spinetoram but was also high for cyantraniliprole, emamectin benzoate and flubendiamide. Results of bioassays for conventional insecticides showed that deltamethrin was less toxic than bifenthrin (toxicity ranking) in pyrethroids tested, while diflubenzuron followed by quinalphos were proved to have lowest toxicities with LC_{50} values of 3.52 mg L^{-1} and 3.24 mg L^{-1}, respectively. Among the newer generation novel molecules tested, pyridalyl was the least toxic against the laboratory strain of *H. talaca*.

Resistance of *H. talaca* field strains to four traditional insecticides

The resistance to bifenthrin in *H. talaca* was the lowest in a population collected from Namchi, while the highest resistance was obtained in Harishpur population collected during May 2019 (Table 2). For deltamethrin, the LCRs for populations collected in 2019 from Tumsong and Harishpur were 107.52 and 111.41 respectively, similar to that acquired for the Kamalpur population collected in 2017 (98.81). The LC_{50} values of the Namchi populations, collected in 2017, 2018 and 2019, bioassayed with diflubenzuron were significantly higher than that of the reference population with LCRs ranging from 25.34 to 60.63. However, the field strains of *H. talaca* in all six locations revealed higher LCRs in 2018 (ranged from 38.80 to 117.10) compared to 2019 (LCRs ranged from 10.92 to 42.79). Regarding quinalphos, the LC_{50} values for Kalchini population collected in 2018 and 2019 and Harishpur and Kumai populations collected in 2019 were between 95.90 mg L^{-1} and 139.58 mg L^{-1}.

Resistance of *H. talaca* field strains to two new recommended insecticides

Toxicities of new recommended insecticides against different field populations of *H. talaca* are listed in Table 3. Populations collected from Namchi, Tumsong, Kalchini, Harishpur and Kumai in 2017–2019 showed varying levels of resistance to emamectin benzoate, while Kamalpur strain exhibited an increasing trend. For flubendiamide, the LC_{50} values for Harishpur populations collected in 2017 and 2019 and Kalchini population collected in 2018 were

Table 1 Baseline susceptibilities of Lab/Htal-IN population to selected insecticides

Insecticides	*n* total	Slope (±SE)	χ^2	df	LC_{50} (mg L^{-1})	Rank[a]	95% limits
Bifenthrin 7.9 SC	440	1.82 ± 0.29	0.23	4	1.27	6	0.82–1.44
Cyantraniliprole 10.26 OD	400	2.56 ± 0.35	1.12	4	0.72	2	0.45–1.24
Deltamethrin 2.8 EC	440	1.79 ± 0.48	2.59	4	2.86	7	2.03–3.90
Diflubenzuron 25 WP	520	1.18 ± 0.30	1.26	5	3.52	9	1.28–4.19
Emamectin benzoate 5 SG	400	1.91 ± 0.52	1.42	4	0.84	3	0.61–1.23
Flubendiamide 480 SC	400	2.42 ± 0.31	0.85	4	0.93	4	0.58–1.32
Pyridalyl 10 EC	360	0.86 ± 0.22	0.54	5	1.12	5	0.88–2.07
Quinalphos 25 EC	440	0.94 ± 0.15	2.16	4	3.24	8	2.72–4.08
Spinetoram 11.7 SC	360	1.69 ± 0.27	1.67	4	0.58	1	0.33–1.02

[a]Toxicity ranking of each insecticide for Lab/Htal-IN according to their estimated LC_{50}

Table 2 Toxicity of four traditional insecticides against different field strains of *Hyposidra talaca* from north-eastern tea growing zone of India

Insecticides	Tea estate, location and province	Collection time	*n* total	Slope (±SE)	χ^2	df	LC_{50} (mg L^{-1})	95% CL	LCR_{50}	95% CL
Bifenthrin 7.9 SC	Namchi, Ravangla, Sikkim	Oct, 2017	320	1.36 ± 0.21	1.13	4	2.91	0.71–4.56	2.29	1.19–3.65
		Nov, 2018	320	1.28 ± 0.15	1.54	4	9.02	4.18–13.43	7.10	5.32–10.96
		May, 2019	320	1.72 ± 0.32	0.68	4	5.38	2.62–7.18	4.23	2.82–6.39
	Tumsong, Darjeeling, West Bengal	Oct, 2017	320	2.03 ± 0.41	1.39	4	21.18	8.39–37.91	16.67	10.15–28.57
		Oct, 2018	320	1.44 ± 0.22	1.22	4	32.12	12.26–52.54	25.29	18.51–39.82
		Apr, 2019	320	1.62 ± 0.35	0.53	4	58.20	27.67–73.16	45.82	33.85–58.29
	Kalchini, Dooars, West Bengal	Nov, 2017	320	1.49 ± 0.28	0.76	4	4.26	2.11–7.25	3.35	2.23–6.05
		Nov, 2018	320	1.13 ± 0.19	1.29	4	12.05	7.14–23.88	9.48	6.19–14.36
		Apr, 2019	320	1.91 ± 0.24	2.07	4	17.86	9.27–32.16	14.06	9.56–19.25
	Kamalpur, Darjeeling, West Bengal	Oct, 2017	320	1.39 ± 0.21	0.53	4	56.74	37.18–72.43	44.67	38.76–52.89
		Oct, 2018	320	1.82 ± 0.35	0.33	4	119.59	66.38–131.67	94.16	74.24–114.38
		Apr, 2019	320	1.50 ± 0.31	1.45	4	93.24	80.50–152.22	73.41	59.35–96.84
	Harishpur, Namrup, Assam	Nov, 2017	320	2.32 ± 0.33	1.52	4	105.30	69.84–140.12	82.91	65.57–110.40
		Oct, 2018	320	1.36 ± 0.18	1.98	4	82.49	52.25–118.34	64.95	42.86–91.32
		May, 2019	320	1.59 ± 0.26	1.21	4	178.21	106.51–213.73	140.32	118.42–176.13
	Kumai, Kalimpong, West Bengal	Oct, 2017	320	1.30 ± 0.29	0.26	4	44.38	28.51–72.84	34.94	21.29–49.11
		Nov, 2018	320	1.71 ± 0.32	1.68	4	79.74	43.91–102.47	62.78	49.13–88.57
		May, 2019	320	1.52 ± 0.22	1.52	4	110.19	89.15–133.20	86.76	61.19–104.28

Table 2 (continued)

Insecticides	Tea estate, location and province	Collection time	*n* total	Slope (± SE)	χ^2	df	LC_{50} (mg L^{-1})	95% CL	LCR_{50}	95% CL
Deltamethrin 2.8 EC	Namchi, Ravangla, Sikkim	Oct, 2017	320	1.17 ± 0.23	0.34	4	4.32	2.92–10.36	1.51	0.25–3.19
		Nov, 2018	320	1.88 ± 0.22	2.53	4	54.12	28.52–82.96	18.92	12.28–31.41
		May, 2019	320	1.57 ± 0.39	1.36	4	31.39	13.71–56.32	10.97	6.39–17.11
	Tumsong, Darjeeling, West Bengal	Oct, 2017	320	1.32 ± 0.28	1.72	4	142.67	88.16–179.12	49.88	32.53–69.71
		Oct, 2018	320	1.89 ± 0.49	0.46	4	178.91	112.63–208.51	62.55	48.56–90.19
		Apr, 2019	320	1.54 ± 0.31	1.37	4	307.52	266.25–346.80	107.52	89.30–141.08
	Kalchini, Dooars, West Bengal	Nov, 2017	320	1.79 ± 0.32	0.29	4	15.19	7.20–38.16	5.31	2.52–8.94
		Nov, 2018	320	1.34 ± 0.20	1.56	4	112.84	76.47–141.30	39.45	22.54–56.82
		Apr, 2019	320	1.62 ± 0.54	1.56	4	28.76	12.06–52.90	10.05	4.32–17.58
	Kamalpur, Darjeeling, West Bengal	Oct, 2017	320	2.11 ± 0.31	1.82	4	282.61	220.51–341.91	98.81	70.18–135.27
		Oct, 2018	320	1.52 ± 0.30	0.69	4	156.83	98.23–178.55	54.83	42.25–68.72
		Apr, 2019	320	1.70 ± 0.23	1.17	4	207.59	170.18–289.30	72.58	51.27–96.52
	Harishpur, Namrup, Assam	Nov, 2017	320	1.33 ± 0.35	0.32	4	123.68	77.36–156.84	43.24	28.42–66.58
		Oct, 2018	320	1.65 ± 0.10	1.91	4	108.95	82.16–175.28	38.09	28.39–52.35
		May, 2019	320	1.70 ± 0.23	2.65	4	318.65	196.51–464.25	111.41	86.11–139.44
	Kumai, Kalimpong, West Bengal	Oct, 2017	320	1.82 ± 0.42	0.54	4	86.17	29.12–112.58	30.12	22.26–43.78
		Nov, 2018	320	1.45 ± 0.31	0.33	4	168.32	116.33–208.34	58.85	38.11–76.29
		May, 2019	320	1.56 ± 0.28	0.90	4	132.05	79.77–161.10	46.17	31.32–57.86

Table 2 (continued)

Insecticides	Tea estate, location and province	Collection time	*n* total	Slope (± SE)	χ^2	df	LC_{50} (mg L^{-1})	95% CL	LCR_{50}	95% CL
Diflubenzuron 25 WP	Namchi, Ravangla, Sikkim	Oct, 2017	360	1.29 ± 0.30	1.89	4	180.16	95.54–226.29	51.18	34.52–78.03
		Nov, 2018	360	1.81 ± 0.34	1.31	4	213.43	139.37–268.55	60.63	46.11–89.15
		May, 2019	360	1.62 ± 0.42	0.82	4	89.20	21.13–132.18	25.34	18.19–37.51
	Tumsong, Darjeeling, West Bengal	Oct, 2017	360	2.02 ± 0.23	2.72	4	292.42	150.07–395.11	83.07	68.25–113.58
		Oct, 2018	360	1.57 ± 0.22	2.61	4	377.15	191.51–558.86	107.14	90.11–129.08
		Apr, 2019	360	1.84 ± 0.15	0.58	4	128.63	71.25–180.30	36.54	21.47–48.62
	Kalchini, Dooars, West Bengal	Nov, 2017	360	2.38 ± 0.35	0.88	4	46.18	22.17–98.70	13.11	9.28–18.50
		Nov, 2018	360	1.71 ± 0.36	1.69	4	162.26	108.11–223.42	46.09	32.15–65.40
		Apr, 2019	360	1.54 ± 0.25	1.53	4	87.47	39.31–159.26	24.84	17.24–38.38
	Kamalpur, Darjeeling, West Bengal	Oct, 2017	360	1.34 ± 0.17	1.49	4	263.71	179.23–308.18	74.91	61.05–93.77
		Oct, 2018	360	1.50 ± 0.36	0.61	4	196.33	110.97–276.46	55.77	41.25–74.38
		Apr, 2019	360	1.31 ± 0.24	1.35	4	127.58	31.29–289.67	36.24	24.56–50.91
	Harishpur, Namrup, Assam	Nov, 2017	360	1.67 ± 0.30	1.61	4	301.49	137.51–390.77	85.65	59.57–126.08
		Oct, 2018	360	1.29 ± 0.12	0.76	4	412.20	269.81–630.72	117.10	92.56–145.29
		May, 2019	360	2.61 ± 0.34	0.44	4	150.63	75.22–238.45	42.79	27.11–61.34
	Kumai, Kalimpong, West Bengal	Oct, 2017	360	2.09 ± 0.52	0.28	4	92.16	23.12–148.29	26.18	16.20–35.72
		Nov, 2018	360	1.58 ± 0.39	1.59	4	136.58	80.67–179.90	38.80	28.42–53.70
		May, 2019	360	1.36 ± 0.34	2.65	4	38.44	10.58–56.58	10.92	6.95–15.67

Table 2 (continued)

Insecticides	Tea estate, location and province	Collection time	*n* total	Slope (±SE)	χ^2	df	LC_{50} (mg L^{-1})	95% CL	LCR_{50}	95% CL
Quinalphos 25 EC	Namchi, Ravangla, Sikkim	Oct, 2017	360	1.54 ± 0.22	1.81	4	8.21	3.13–17.50	2.53	1.07–4.57
		Nov, 2018	360	1.32 ± 0.16	0.67	4	67.52	38.71–83.56	20.83	11.82–29.34
		May, 2019	360	1.26 ± 0.35	0.32	4	15.86	9.26–31.09	4.89	1.94–8.29
	Tumsong, Darjeeling, West Bengal	Oct, 2017	360	1.89 ± 0.29	1.46	4	42.82	12.22–56.17	13.21	6.38–21.05
		Oct, 2018	360	2.01 ± 0.21	3.12	4	39.37	18.57–49.46	12.15	7.88–18.60
		Apr, 2019	360	1.64 ± 0.24	1.62	4	137.63	83.21–153.81	42.47	31.18–58.21
	Kalchini, Dooars, West Bengal	Nov, 2017	360	1.59 ± 0.42	1.33	4	192.25	79.38–223.90	59.33	46.27–82.54
		Nov, 2018	360	1.44 ± 0.34	0.43	4	388.20	110.17–516.72	119.81	85.48–153.11
		Apr, 2019	360	1.07 ± 0.31	0.59	4	342.57	168.26–423.59	105.73	92.03–122.52
	Kamalpur, Darjeeling, West Bengal	Oct, 2017	360	1.18 ± 0.12	2.27	4	24.80	8.92–38.03	7.65	3.81–11.49
		Oct, 2018	360	2.32 ± 0.28	0.69	4	120.53	93.54–186.47	37.20	30.71–47.92
		Apr, 2019	360	1.51 ± 0.42	1.82	4	149.31	70.62–208.29	46.08	22.40–67.82
	Harishpur, Namrup, Assam	Nov, 2017	360	1.62 ± 0.11	1.09	4	276.96	88.71–326.20	85.48	69.62–101.30
		Oct, 2018	360	1.56 ± 0.25	0.63	4	210.36	101.87–332.39	64.92	51.25–73.97
		May, 2019	360	1.49 ± 0.34	1.72	4	452.25	176.91–683.64	139.58	108.83–161.21
	Kumai, Kalimpong, West Bengal	Oct, 2017	360	1.37 ± 0.32	1.54	4	105.12	49.07–136.16	32.44	23.29–42.18
		Nov, 2018	360	2.09 ± 0.27	2.05	4	68.83	32.54–102.27	21.24	13.72–35.09
		May, 2019	360	1.55 ± 0.33	1.36	4	310.72	171.39–426.15	95.90	71.91–126.58

LCR_{50} confidence limits not including 1 are indicative of significant differences on LC_{50} between the Lab/Htal−IN population and field collected populations

Table 3 Toxicity of two new recommended insecticides against different field strains of *Hyposidra talaca* from north-eastern tea growing zone of India

Insecticides	Tea estate, location and province	Collection time	*n* total	Slope (±SE)	χ^2	df	LC_{50} (mg L^{-1})	95% CL	LCR_{50}	95% CL
Emamectin benzoate 5 SG	Namchi, Ravangla, Sikkim	Oct, 2017	320	1.42 ± 0.25	0.75	4	31.60	24.03–39.44	37.61	21.18–52.75
		Nov, 2018	320	1.51 ± 0.31	1.56	4	26.82	20.95–35.06	31.92	23.26–42.83
		May, 2019	320	1.32 ± 0.26	1.89	4	39.16	18.71–56.32	46.61	27.54–61.56
	Tumsong, Darjeeling, West Bengal	Oct, 2017	320	1.49 ± 0.09	2.61	4	7.11	2.77–9.56	8.46	3.86–12.25
		Oct, 2018	320	1.52 ± 0.18	0.19	4	4.59	2.32–7.80	5.46	2.69–9.16
		Apr, 2019	320	1.12 ± 0.36	0.32	4	3.36	1.03–6.21	4.00	2.21–7.72
	Kalchini, Dooars, West Bengal	Nov, 2017	320	1.26 ± 0.29	1.54	4	26.51	17.46–39.16	31.55	18.55–44.81
		Nov, 2018	320	1.35 ± 0.24	1.39	4	21.93	9.20–30.77	26.10	19.46–33.68
		Apr, 2019	320	1.76 ± 0.16	1.15	4	49.20	32.63–61.98	58.57	35.39–73.54
	Kamalpur, Darjeeling, West Bengal	Oct, 2017	320	1.82 ± 0.31	0.93	4	15.28	7.51–24.82	18.19	10.22–29.86
		Oct, 2018	320	2.15 ± 0.54	1.76	4	25.69	20.25–42.18	30.58	21.65–39.76
		Apr, 2019	320	1.40 ± 0.13	2.51	4	28.36	16.33–38.73	33.76	17.26–48.26
	Harishpur, Namrup, Assam	Nov, 2017	320	1.67 ± 0.40	1.42	4	30.75	19.52–36.30	36.60	20.55–54.19
		Oct, 2018	320	1.92 ± 0.21	0.34	4	26.24	16.68–47.18	31.23	19.18–38.26
		May, 2019	320	1.83 ± 0.33	1.66	4	40.81	21.94–76.65	48.58	34.51–70.23
	Kumai, Kalimpong, West Bengal	Oct, 2017	320	1.54 ± 0.16	0.72	4	16.78	7.55–26.64	19.97	11.27–32.64
		Nov, 2018	320	2.17 ± 0.43	0.81	4	41.58	24.81–59.93	49.50	38.35–64.12
		May, 2019	320	1.69 ± 0.30	0.36	4	8.06	3.13–14.25	9.59	5.36–14.78

Table 3 (continued)

Insecticides	Tea estate, location and province	Collection time	*n* total	Slope (±SE)	χ^2	df	LC_{50} (mg L^{-1})	95% CL	LCR_{50}	95% CL
Flubendiamide 480 SC	Namchi, Ravangla, Sikkim	Oct, 2017	320	1.34 ± 0.23	1.42	4	15.28	7.13–21.32	16.43	9.94–26.51
		Nov, 2018	320	1.89 ± 0.21	1.70	4	26.52	18.57–29.11	28.51	19.26–37.14
		May, 2019	320	1.74 ± 0.26	0.32	4	31.18	21.51–42.03	33.52	23.65–45.18
	Tumsong, Darjeeling, West Bengal	Oct, 2017	320	1.55 ± 0.34	0.29	4	45.82	38.31–57.83	49.26	34.52–66.21
		Oct, 2018	320	1.86 ± 0.33	1.59	4	36.50	17.32–61.58	39.24	27.13–55.29
		Apr, 2019	320	1.32 ± 0.21	1.80	4	72.39	41.62–90.19	77.83	62.27–91.08
	Kalchini, Dooars, West Bengal	Nov, 2017	320	2.22 ± 0.29	1.13	4	138.52	93.74–155.66	148.94	116.51–177.82
		Nov, 2018	320	1.31 ± 0.22	0.96	4	88.24	46.51–107.82	94.88	82.05–114.75
		Apr, 2019	320	1.54 ± 0.31	0.56	4	104.57	75.03–132.10	112.44	89.82–130.56
	Kamalpur, Darjeeling, West Bengal	Oct, 2017	320	2.50 ± 0.29	2.73	4	37.51	20.11–56.28	40.33	29.11–52.31
		Oct, 2018	320	1.32 ± 0.31	1.58	4	30.97	25.15–39.68	33.30	20.29–44.95
		Apr, 2019	320	2.07 ± 0.20	3.35	4	51.08	27.55–71.31	54.92	41.17–75.46
	Harishpur, Namrup, Assam	Nov, 2017	320	1.09 ± 0.32	0.83	4	68.71	42.17–85.78	73.88	62.78–90.06
		Oct, 2018	320	1.81 ± 0.35	1.74	4	48.65	34.52–77.93	52.31	40.57–68.58
		May, 2019	320	1.94 ± 0.29	0.34	4	89.54	47.08–120.81	96.27	71.25–122.24
	Kumai, Kalimpong, West Bengal	Oct, 2017	320	1.38 ± 0.45	1.62	4	21.37	13.29–28.08	22.97	14.75–28.96
		Nov, 2018	320	1.54 ± 0.32	0.81	4	33.59	18.37–44.54	36.11	21.39–48.60
		May, 2019	320	1.67 ± 0.23	1.29	4	70.92	41.15–92.34	76.25	60.34–89.47

LCR_{50} confidence limits not including 1 are indicative of significant differences on LC_{50} between the Lab/Htal−IN population and field collected populations

between 68.71 and 88.24 mg L^{-1}. However, LCRs for Kalchini populations collected in 2017 and 2019 were relatively higher (> 110) than for Namchi populations (ranged from 16.43 to 33.52).

Resistance of *H. talaca* field strains to three novel bio-rational insecticides

The LCRs for cyantraniliprole ranged from 5.98 to 120.98 for all the field populations collected and bioassayed in 2017–2019 (Table 4). The LC_{50} value for Harishpur strain collected in 2018 was 73.83 mg L^{-1}, resembling that obtained for the Kalchini strain collected in 2019 (87.11 mg L^{-1}). For pyridalyl, all LC_{50} values were significantly different from that of the Lab/Htal-IN. LCRs ranged from 9.09 to 77.09, 13.47 to 39.26 and from 18.91 to 48.63 for all the field populations collected in 2017, 2018 and 2019, respectively. Out of six field strains bioassayed for spinetoram, Kalchini strain showed a very low level of resistance consistently (LCRs 5.96 to 9.03), while other populations displayed moderate to high level with LCRs in the range of 19.31 to 82.03 was observed.

Pairwise correlations between log LC_{50} values of tested insecticides

Correlation between conventional insecticides and new-generation biorational molecules was not significant ($P < 0.05$) except emamectin benzoate, which was significant but negatively correlated with bifenthrin (Table 5). A significant correlation was audited between bifenthrin, deltamethrin and quinalphos ($P < 0.01$), whereas resistance to deltamethrin exhibited no correlation with resistance to other molecules except bifenthrin ($P < 0.05$). Lack of cross-resistance was observed for cyantraniliprole, diflubenzuron, flubendiamide, pyridalyl and spinetoram in field populations of *H. talaca*.

Discussion

Insecticide resistance of *H. talaca* has been studied very briefly and has rarely been documented (Roy et al., 2021; Saha, 2016). The occurrence of traditional insecticides resistance along with new-generation biorational molecules in *H. talaca* from major tea growing zones of North-East India is reported here for the first time. In the present study, it has been observed that LCRs were heterogeneous among the insecticides and also variable among seasons, years and locations. This indicates the potentiality of *H. talaca* field populations to develop resistance to a wide range of insecticides.

In the case of pyrethroids, most of the field populations especially Kamalpur and Harishpur showed very high LCRs (> 90) to both bifenthrin and deltamethrin except the populations collected from Namchi (≤ 18.92). A similar level of poor toxicity of cypermethrin (LC_{50} > 250 mg L^{-1}), another old-generation pyrethroid compound, was also found in the population of *H. talaca* collected in tea ecosystem of Darjeeling (Das et al., 2010). This could be linked to the common reliance on the use of synthetic pyrethroids against looper pest complex in these regions (Roy et al., 2017). The high LCRs to both bifenthrin and deltamethrin observed in the present study may be attributable to the impolitic use of different synthetic pyrethroid insecticides (eight to ten applications per year) in most of the Darjeeling and Assam tea gardens (Gurusubramanian et al., 2008). Moreover, the report of poor field efficacy of deltamethrin at the recommended dose against *H. talaca* from North-Eastern tea estates of India (Basu Majumder et al., 2012), *Spodoptera litura* from China (Tong et al., 2013) and *Helicoverpa armigera* from Pakistan (Hussain et al., 2014), corroborates our findings.

High to very high LCRs (> 100) to diflubenzuron encountered in the present investigation in Tumsong and Harishpur populations of *H. talaca*, could be imputed to the widespread usage of this IGR compound by the tea growers over a long period (Gurusubramanian et al., 2008). In these areas, diflubenzuron is among the most used IGR insecticides for the control of various insect pests of tea like tea mosquito bug, thrips, jassids etc. (Saha, 2016). In contrast, populations of *H. talaca* collected from Kumai, showed LCRs for diflubenzuron between 10.92 and 38.80. The incidence of high efficacy of diflubenzuron in Kalimpong region is transparent from the results of previous literatures (Ghatak & Reza, 2007; Gurusubramanian & Borthakur, 2005), and this could be related to the decreasing resistance to this molecule. Besides, we hypothesized that the application of some ready-mix insecticides (diflubenzuron + deltamethrin, novaluron + indoxacarb and

Table 4 Toxicity of three novel insecticide molecules against different field strains of *Hyposidra talaca* from north-eastern tea growing zone of India

Insecticides	Tea estate, location and province	Collection time	*n* total	Slope (± SE)	χ^2	df	LC_{50} (mg L^{-1})	95% CL	LCR_{50}	95% CL
Cyantraniliprole 10.26 OD	Namchi, Ravangla, Sikkim	Oct, 2017	320	1.16 ± 0.12	1.32	4	4.31	2.67–9.11	5.98	2.36–9.10
		Nov, 2018	320	1.23 ± 0.23	1.58	4	14.56	5.28–27.12	20.22	9.18–32.55
		May, 2019	320	1.72 ± 0.29	1.16	4	13.27	7.13–19.62	18.43	12.26–25.29
	Tumsong, Darjeeling, West Bengal	Oct, 2017	320	2.15 ± 0.32	0.25	4	49.66	23.78–67.37	68.97	56.34–81.58
		Oct, 2018	320	0.92 ± 0.07	0.89	4	18.23	7.98–26.16	25.31	14.98–36.47
		Apr, 2019	320	1.54 ± 0.11	2.54	4	24.13	11.21–32.84	33.51	20.09–45.25
	Kalchini, Dooars, West Bengal	Nov, 2017	320	1.33 ± 0.30	1.23	4	41.29	24.11–62.54	57.34	41.13–70.24
		Nov, 2018	320	1.88 ± 0.31	1.61	4	59.37	21.38–68.29	82.45	69.51–95.47
		Apr, 2019	320	1.71 ± 0.22	0.83	4	87.11	61.22–102.37	120.98	93.66–152.80
	Kamalpur, Darjeeling, West Bengal	Oct, 2017	320	0.89 ± 0.10	1.65	4	10.72	4.34–16.20	14.88	6.43–21.38
		Oct, 2018	320	1.34 ± 0.22	0.55	4	22.68	13.76–42.37	31.50	23.26–41.18
		Apr, 2019	320	1.74 ± 0.36	0.72	4	30.27	15.19–39.77	42.04	27.45–59.76
	Harishpur, Namrup, Assam	Nov, 2017	320	1.57 ± 0.24	1.56	4	38.79	16.32–59.61	53.87	40.21–68.85
		Oct, 2018	320	1.30 ± 0.30	2.30	4	73.83	45.88–137.09	102.54	88.13–117.39
		May, 2019	320	1.52 ± 0.28	1.14	4	60.18	31.57–93.20	83.58	71.50–93.08
	Kumai, Kalimpong, West Bengal	Oct, 2017	320	0.76 ± 0.25	0.51	4	19.58	10.81–31.26	27.19	21.25–35.42
		Nov, 2018	320	1.61 ± 0.33	0.63	4	24.27	12.71–46.50	33.70	19.56–41.31
		May, 2019	320	1.58 ± 0.30	1.52	4	45.62	24.48–85.44	63.36	48.12–75.06

Table 4 (continued)

Insecticides	Tea estate, location and province	Collection time	*n* total	Slope (± SE)	χ^2	df	LC_{50} (mg L^{-1})	95% CL	LCR_{50}	95% CL
Pyridalyl 10 EC	Namchi, Ravangla, Sikkim	Oct, 2017	320	1.36 ± 0.31	0.91	4	10.19	7.32–21.57	9.09	5.61–14.75
		Nov, 2018	320	2.19 ± 0.22	0.49	4	15.09	10.11–23.32	13.47	8.52–16.97
		May, 2019	320	1.54 ± 0.25	1.56	4	28.12	12.27–42.80	25.10	16.45–34.68
	Tumsong, Darjeeling, West Bengal	Oct, 2017	320	1.34 ± 0.26	1.73	4	37.39	21.76–65.36	33.38	17.98–44.58
		Oct, 2018	320	1.65 ± 0.34	3.32	4	28.72	15.81–37.51	25.64	12.88–38.41
		Apr, 2019	320	2.34 ± 0.29	0.56	4	24.80	13.67–40.25	22.14	15.25–31.74
	Kalchini, Dooars, West Bengal	Nov, 2017	320	1.82 ± 0.39	0.71	4	86.35	45.76–172.19	77.09	63.44–92.50
		Nov, 2018	320	1.28 ± 0.32	1.54	4	40.08	20.11–58.24	35.78	23.64–48.37
		Apr, 2019	320	1.97 ± 0.26	1.59	4	54.47	22.44–72.89	48.63	29.58–61.96
	Kamalpur, Darjeeling, West Bengal	Oct, 2017	320	2.25 ± 0.29	0.72	4	29.37	15.50–42.23	26.22	14.11–38.42
		Oct, 2018	320	0.95 ± 0.20	2.69	4	26.92	20.10–35.16	24.03	15.61–33.28
		Apr, 2019	320	0.89 ± 0.18	1.32	4	21.18	9.51–31.60	18.91	10.62–27.65
	Harishpur, Namrup, Assam	Nov, 2017	320	1.54 ± 0.26	0.31	4	49.27	31.25–71.16	43.99	31.80–59.31
		Oct, 2018	320	1.62 ± 0.23	0.19	4	43.98	23.31–87.24	39.26	26.62–54.90
		May, 2019	320	1.70 ± 0.17	1.72	4	32.20	18.22–40.60	28.75	20.08–37.89
	Kumai, Kalimpong, West Bengal	Oct, 2017	320	1.31 ± 0.39	2.15	4	30.13	17.21–44.18	26.90	18.29–35.43
		Nov, 2018	320	1.50 ± 0.25	0.56	4	31.79	19.45–52.26	28.38	18.92–38.47
		May, 2019	320	1.32 ± 0.33	1.73	4	22.85	10.21–37.56	20.40	12.25–30.41

Table 4 (continued)

Insecticides	Tea estate, location and province	Collection time	*n* total	Slope (±SE)	χ^2	df	LC_{50} (mg L^{-1})	95% CL	LCR_{50}	95% CL
Spinetoram 11.7 SC	Namchi, Ravangla, Sikkim	Oct, 2017	320	1.81 ± 0.31	1.31	4	18.92	11.15–24.61	32.62	18.21–46.77
		Nov, 2018	320	1.74 ± 0.34	1.64	4	47.58	15.78–72.53	82.03	68.45–102.62
		May, 2019	320	1.32 ± 0.30	0.79	4	31.77	20.36–43.19	54.77	41.53–70.58
	Tumsong, Darjeeling, West Bengal	Oct, 2017	320	0.90 ± 0.22	1.92	4	39.62	23.18–53.37	68.31	55.26–79.66
		Oct, 2018	320	2.10 ± 0.26	0.54	4	44.91	34.50–69.07	77.43	58.42–94.35
		Apr, 2019	320	1.25 ± 0.27	0.36	4	27.39	19.20–38.26	47.22	32.76–61.58
	Kalchini, Dooars, West Bengal	Nov, 2017	320	1.69 ± 0.20	1.56	4	3.46	1.92–5.06	5.96	2.84–9.73
		Nov, 2018	320	1.34 ± 0.19	1.18	4	4.18	2.88–5.67	7.20	3.42–10.55
		Apr, 2019	320	1.58 ± 0.31	1.68	4	5.24	2.63–7.49	9.03	4.67–16.33
	Kamalpur, Darjeeling, West Bengal	Oct, 2017	320	1.79 ± 0.22	0.23	4	45.82	29.77–59.93	79.00	63.25–95.40
		Oct, 2018	320	1.26 ± 0.54	0.50	4	19.79	9.56–28.89	34.12	19.65–45.81
		Apr, 2019	320	0.82 ± 0.27	1.37	4	25.51	14.33–38.20	43.98	31.58–58.72
	Harishpur, Namrup, Assam	Nov, 2017	320	2.01 ± 0.31	2.40	4	22.09	14.52–30.45	38.08	25.61–52.93
		Oct, 2018	320	1.56 ± 0.22	1.23	4	29.13	18.29–38.55	50.22	38.97–61.06
		May, 2019	320	1.84 ± 0.20	0.58	4	20.11	7.21–27.94	34.67	20.74–47.64
	Kumai, Kalimpong, West Bengal	Oct, 2017	320	1.59 ± 0.18	3.07	4	11.20	4.26–19.82	19.31	11.23–28.59
		Nov, 2018	320	1.70 ± 0.13	0.22	4	32.51	21.18–42.05	56.05	42.08–73.42
		May, 2019	320	1.21 ± 0.34	1.68	4	19.87	12.38–26.94	34.25	23.11–44.97

LCR_{50} confidence limits not including 1 are indicative of significant differences on LC_{50} between the Lab/Htal−IN population and field collected populations

Table 5 Pairwise correlation coefficient comparison among log LC_{50} values of the selected insecticides on different field strains of *Hyposidra talaca*

	Bifenthrin (SCM)	Cyan-traniliprole (RRM)	Deltamethrin (SCM)	Diflubenzu-ron (ICB)	Emamectin benzoate (GLUCL)	Flubendiamide (RRM)	Pyridalyl (UMOA)	Quinalphos (ACHI)
Bifenthrin (SCM)								
Cyan-traniliprole (RRM)	0.294^{NS}							
Deltamethrin (SCM)	$0.538^{0.011}$	0.456^{NS}						
Diflubenzuron (ICB)	0.153^{NS}	0.629^{NS}	0.251^{NS}					
Emamectin benzoate (GLUCL)	$-0.461^{0.029}$	0.158^{NS}	-0.462^{NS}	0.859^{NS}				
Flubendiamide (RRM)	0.259^{NS}	0.546^{NS}	0.080^{NS}	0.013^{NS}	0.259^{NS}			
Pyridalyl (UMOA)	-0.426^{NS}	0.406^{NS}	0.135^{NS}	0.094^{NS}	0.381^{NS}	0.198^{NS}		
Quinalphos (ACHI)	$0.294^{0.008}$	0.324^{NS}	$0.309^{0.003}$	0.330^{NS}	0.485^{NS}	-0.451^{NS}	0.296^{NS}	
Spinetoram (NACHR)	0.189^{NS}	0.580^{NS}	-0.065^{NS}	0.028^{NS}	0.311^{NS}	0.120^{NS}	0.328^{NS}	0.047^{NS}

Superscripts impart the significance of regression

SCM Sodium Channel Modulators, *RRM* Ryanodine Receptor Modulators, *ICB* Inhibitors of Chitin Biosynthesis affecting CHS1, *GLUCL* Glutamate−Gated Chloride Channel Allosteric Modulators, *UMOA* Unknown Mode of Action, *ACHI* Acetylcholinesterase Inhibitors, *NACHR* Nicotinic Acetylcholine Receptor Allosteric Modulators−Site 1

lufenuron + emamectin benzoate) has allowed the Kalimpong population of *H. talaca* to remain susceptible to diflubenzuron. For example, susceptibility of *Plutella xylostella* (L.) to bifenthrin increased when this insecticide was mixed with indoxacarb, spinosad and emamectin benzoate (Attique et al., 2006). Similarly, spraying cotton with pyrethroid and organophosphate combinations has apparently prevented the development of pyrethroid resistance in the *Helicoverpa armigera* (Hübner) (Martin et al., 2000).

Considerable resistance to quinalphos observed in the present study could be attributed to the excessive application of various OP insecticides in the tea ecosystem of Kalchini and Harishpur. *H. talaca* can be subjected to up to six applications of profenophos, dimethoate and phosalone per year (Gurusubramanian et al., 2008), used to mitigate the tea red spider mite and some phloem sap-sucking insect pests, which can also flourish cross-resistance with quinalphos. Considering the verity that the aforesaid molecules are OP compounds, the similar chemistry as quinalphos, this is not a sudden consequence. Similar circumstance has been reported for *Diaphorina citri* (Pardo et al., 2018), *Spodoptera exigua* (Ishtiaq et al., 2012) and *Quadraspidiotus perniciosus* (Buzzetti et al., 2015), whereby the OP compounds showed the highest resistance for the populations collected at places with the severe application of insecticides belonging to this class.

Members of new recommended insecticides against *H. talaca* (NPATG, 2016; PPC, 2019) exhibited varying levels of resistance, which will be conducive in creating management strategies. However, it is difficult to explain why emamectin benzoate and flubendiamide resulted in higher resistance in the Dooars and Assam populations, and comparison of the log LC_{50} values of tested molecules showed incident of correlation within emamectin benzoate and bifenthrin, which suggest a possibility of cross-resistance mechanism. This situation could be associated with two factors. First, the detoxification augmentation causes metabolism resistance and involves some enzymes like general esterases (GEs) and glutathione S-transferase (GSTs) have various isoenzymes

(Das & Mukhopadhyay, 2014; Prasad & Mukhopadhyay, 2015; Roy et al., 2021). Cross-resistance might be possible when a single molecule selects specific isoenzymes, which can act on other molecules. We hypothesized that the significant correlation between emamectin benzoate and bifenthrin is due to the association of bifenthrin selected specific isoenzymes with the emamectin. A significant higher correlation between emamectin benzoate and abamectin has been reported from *Spodoptera litura* in Pakistan (Ahmad et al., 2008), our study does not derive this result, although both the molecules bind to the GABA-gated chloride channel. Second, lack of recommended dose-dependent application of both emamectin benzoate and flubendiamide in the tea estates of Dooars and Assam compared to Darjeeling since 2016 (Roy et al., 2021). A piece of relevant information should be kept in mind that Darjeeling tea is plucked in the form of "two leaves and a bud" to produce high-quality green, black, white and oolong tea through partial oxidation (Gohain et al., 2012), while Dooars and Assam tea are used to produce CTC (crush, tear, curl) tea through full oxidation process and fine plucking is not done (Laskar & Thappa, 2015). For this reason, degradation of applied insecticides is found to be more spontaneous in Dooars and Assam tea than that of Darjeeling tea while processing (Bajwa & Sandhu, 2014; Pan et al., 2015), which might be attributable to the improper usage of flubendiamide and emamectin benzoate by the tea growers of former and subjected to higher LCR than the later.

Like flubendiamide, high LCRs to cyantraniliprole found in both Kalchini and Harishpur populations of *H. talaca*, could be linked with cross-resistance between these compounds belong to the same chemical group or act of similar detoxification enzymes. Pyridalyl and spinetoram exhibited low to high LCRs in different field-collected strains of *H. talaca*. The minimal usage of newer molecules is also associated with their high market price, which many small tea growers could not afford. However, it is important to note that blending newer molecules with traditional insecticides is a very common practice among the tea growers (Saha, 2016), may annul the benefits of insecticide combinations. Such irrational tank-mix formulations can also result in cross or multiple resistance that may exaggerate across another class of chemistry, further ensnaring pest management (Ahmad et al., 2009). On the other hand, an interesting thing observed in the present study that the Namchi populations of *H. talaca* showed low levels of toxicity against emamectin benzoate and spinetoram compared with OP and pyrethroid insecticides. The high LCRs to these novel molecules may be attributable to the significant use of some microbial derivative insecticides in Sikkim. In January 2016, the Sikkim government declared the state as the first organic state of India (Gopi et al., 2016; Meyer, 2019). On this circumstance, many farmers possibly started to use some green level insecticides like emamectin benzoate and spinetoram as biologically originated "semi-organic" inputs to combat various insect and mite pests of crops including tea (Buragohain, 2020; Rao, 2017), which could be linked to the present observation.

H. talaca has recently emerged as a serious defoliator of tea in North-Eastern India and the control of this pest has relied solely on chemical insecticides. Due to the lack of suitable resistance management strategies, calendar-based application of pesticides is a general practice among the tea producers, which could be the most probable cause for the development of insecticide resistance. However, the findings gathered in the present study have potentially significant conjugations for insecticide resistance management. The implementation of a resistance management plan is suggested, especially in locations where *H. talaca* has developed very high levels of resistance to newer insecticides. Some alternative management practices like crop sanitation (Roy et al., 2013), augmentation of natural predators and parasitoids (Sinu et al., 2011), use of microbial insecticides like nuclear polyhedrosis virus (Ghosh et al., 2015; Sinu et al., 2015) and some botanicals (Roy et al., 2015) and rotation of insecticide classes (Roy et al., 2017) could be included in the plan. Also, the use of highly resistant molecules could be suspended at the concerned areas for a few years to increase the susceptibility of *H. talaca* populations against those molecules in the coming future. Therefore, an extensive IPM programme along with appropriate insecticide resistance management techniques will be the ideal option for successful control of this black looper on tea in India.

Acknowledgements Authors would like to thank University Grants Commission, New Delhi, India for providing the required fund and Dr. N. S. Bag, Associate Professor, Sikkim University for providing the laboratory reared population of *Hyposidra talaca*, respectively. Additionally, authors are also grateful to American English expert for final editing of the manuscript, which has profited to illuminating the text. We are also thankful to the anonymous reviewers and Cesar Monzo Ferrer, Associate Editor for suggesting substantial improvements.

Authors' contributions The work was carried out in collaboration with all authors. Author DR and AS conceived and designed the research work. DR, AS and GC conducted the laboratory experiments and collected data. AB and PKS analyzed data. DR, AB and GC wrote the manuscript. All authors read and approved the final manuscript.

Funding The study was funded by University Grants Commission, New Delhi, India. The grant number of the concerned funding is RGNF-2017–18-SC-WES-43747.

Declarations

Ethics approval This article does not contain any studies with human or other animal subjects.

Conflicts of interest The authors have declared that no conflict of interest exists.

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