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COMPARISON OF DETOXIFICATION ENZYMES OF *BEMISIA TABACI* (HEMIPTERA: ALEYRODIDAE) BIOTYPES B AND Q AFTER VARIOUS HOST SHIFTS

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ABSTRACT

To overcome host plant defense mechanisms, herbivorous insects have developed a series of strategies, which include changes in reliance on various classes of detoxification enzymes. There are few relevant experimental studies on detoxification enzymes of whitefly biotypes B and Q when shifting to different host plants. Here we report changes in the activities of carboxylesterase (CarE), cytochrome-P450-dependent monooxygenase (P450), and glutathione S-transferase (GST) of *B. tabaci* biotypes B and Q, 24 h after these biotypes had been transferred from cucumber (*Cucumis sativus* L.; Cucurbitaceae) to numerous other host species belong to 29 families. The aim of this study was to compare the differential utilization of the above detoxifying enzymes by these 2 *B. tabaci* biotypes when the latter were subjected to shifts between host species. The GST activities of these 2 biotypes did not change significantly 24 h after being shifted from cucumber to various other hosts. However after such recent shifts from cucumber to various other host species, most values of CarE and P450 of biotype Q were significantly higher than those of biotype B at $P < 0.05$. The experiments revealed that *B. tabaci* biotypes utilize different defense strategies of differentially inducing detoxification enzymes when facing a variety of host shifts.

Key Words: carboxylesterase, CarE, cytochrome-P450-dependent monooxygenase, P450, glutathione S-transferase, GST, short-term induction

RESUMEN

Para superar los mecanismos de defensa de las plantas hospederas, insectos herbívoros han desarrollado una serie de estrategias, que incluyen cambios en la dependencia de las diversas clases de enzimas de desintoxicación. Existen pocos estudios experimentales pertinentes a las enzimas de desintoxicación de biotipos B y Q de la mosca blanca *B. tabaci*, cuando se cambian a diferentes plantas hospederas. Aqui informamos de los cambios en las actividades de la carboxilesterasa (CarE), monooxigenasa dependiente de citocromo-P450 (P450) y transferasa-S de glutatión - (GST) de los biotipo B y Q de *B. tabaci*, 24 horas después de que estos biotipos habían sido trasladados del pepino (*Cucumis sativus* L.; Cucurbitaceae) a un extenso numero de otras especies hospedadoras que pertenecen a otras 29 familias. El objetivo de este estudio fue comparar la utilización diferencial de las enzimas desintoxicantes mencionadas anteriormente de estos 2 biotipos de *B. tabaci* cuando fueron sometidos a un cambio de la especie hospedera. La actividad de GST en estos 2 biotipos no cambió significativamente 24 horas después de que estos fueron trasladados de pepino a otros hospederos. Sin embargo, después de estos trasladados recientes de pepino a varias otras especies hospederas, la mayoría de los valores de CarE y P450 del biotipo Q fueron significativamente más altos que los de biotipo B en $P < 0.05$. Los experimentos revelaron que los biotipos de *B. tabaci* utilizan diferentes estrategias de defensa al inducir diferencialmente las enzimas de desintoxicación cuando se enfrentan a una variedad de traslados de hospederos.

Palabras Clave: carboxilesterasa, CarE, monooxigenasa-dependiente de citocromo-P450, P450, transferasa-S de glutatión, GST, inducción de corto plazo

The silverleaf whitefly, *Bemisia tabaci* (Gennadius) (Hemiptera: Aleyrodidae), is a destructive pest of agricultural and horticultural crops, infesting more than 600 plant species in temperate and tropical regions(Xu et al. 2011; Xu et al. 2013; Yang et al. 2013). This pest consists of a species complex of at least 28 species, which include morphologically indistinguishable biotypes A, B, Q and Ms. Among these biotypes of *B. tabaci* B and Q are the 2 most invasive pests, causing great losses in crop yield and quality by sucking plant sap and transmitting viral diseases(Perring 2001;

Jones 2003; Liu et al. 2007; Chu et al. 2010; Camara et al. 2013; Sun et al. 2013). In the past 20 years, the status of *B. tabaci* has risen considerably due to the widespread invasions of biotypes B and Q (De Barro et al. 2011; Liu et al. 2012).

The first record of *B. tabaci* in China was in 1949, but no significant damage was reported until the 1990s (Zhou 1949; Luo et al. 2002).In the mid-1990s, *B. tabaci* outbreaks throughout most of China were those of biotype B designated a new species, *B. argentifolii* Bellows & Perring, in the USA (Bellows et al. 1994), it is still referred to as *B. tabaci* biotype B in China (Wu et al. 2002). In 2003, biotype Q was found in Kunming and subsequently in Beijing and Henan (Chu et al. 2006). Since then, biotype Q has gradually displaced the established biotype B as the predominant *B. tabaci* strain in most of China (Pan et al. 2011).

To overcome host plant defense mechanisms, herbivorous insects synchronously develop a series of strategies, including a change in the activities and structures of different detoxification enzymes. The constitutive, induced direct and induced indirect defenses of the host plant can affect the various physiological and behavioral traits of herbivorous insects. In insects, detoxification of xenobiotics is accomplished by 3 multigene families: carboxylesterases (CarE), cytochrome P450 monooxygenases (P450) and glutathione-S-transferases (GST) (Claudianos et al. 2006). Due to the use of xenobiotic compounds by the host and increased insecticide resistance, detoxification enzymes should be studied because their role in xenobiotic compound metabolism was well established (Schuler 1996; Byrne et al. 2000, 2003; Riley & Tan 2003; Liang et al. 2007; Howe & Jander 2008; Xie et al. 2011).

Although researchers have proposed that biotype Q has a higher capacity to utilize host plants than biotype B, there are few relevant experimental studies of detoxification enzymes of biotypes B and Q upon various host plants. We determined and compared the activities of the detoxifying enzymes including CarE, P450 and GST from *B. tabaci* biotypes B and Q 24 h after they were transferred from cucumber, *Cucumis sativus* L. (Cucurbitales: Cucurbitaceae), to 55 other host plant species of 29 families. The objectives of this study were to compare how *B. tabaci* biotypes B and Q utilize detoxification enzymes when coping with short-term host shifts.

Materials and Methods

Whiteflies and Plants

Bemisia tabaci biotypes B and Q were collected in 2010 from cucumber, *C. sativus*, in the fields of the Dongsheng Garden, Liwan District, Guangzhou. Biotypes B and Q whiteflies were maintained on cucumber and continuously reared for 10 generations without exposure to any insecticide in the greenhouse. The *B. tabaci* populations were identified using an mtDNA CO I marker (Luo et al. 2002)#. All plant species were collected in 2011 from the South China Botanical Garden, Chinese Academy of Science, Tianhe District, Guangzhou.

Chemicals

The reagents used for the experiments included α-naphthylacetate (α- NA), Eserine, AlbuMin bovine V (BSA), Coomassie Brilliant Blue G250, 1-chloro-2,4-dinitrobenzene (CDNB),reduced glutathione (GSH), Fast Blue B salt, Sodium dodecyl sulfate (SDS), EDTA, 1,4-dithioerythritol (DTT), phenylmethanesulfonyl fluoride (PMSF) and 4-nitroanisole (PNA),which were all bought from Sigma. Reduced β-nicotinamide adenine dinucleotide phosphate (NADPH), Triton X-100 was bought from Shanghai Yanhui Biotechnology LLC. Other reagents were bought from Guangzhou Qianhui Bose Instrument Co., Ltd.

Preparation of Host Leaves

Leaf discs of host plants were placed with their adaxial surfaces downwards onto a bed of agar (2 mL of 30 g L^{-1}) in a flat-bottomed glass tube (90 mm in length, 35 mm in diam). More than 60 adults, which were similar in size and age, were selected at random from a population for experiments on various host shifts and were transferred into the glass tube. Each treatment included 3 replicates. The apparatus was inverted to allow the leaf to be placed in a natural orientation and incubated at 25 °C and 60% RH and 12:12 h L:D. A total of 10 adults were collected for further measurement of detoxification enzymes after a 24 h host shift. Adult whiteflies collected from different transferred host plants were placed in liquid nitrogen for 10 min and then transferred to a -80 °C freezer for later use.

Measurement of Carboxylesterase(CarE) Activities

Carboxylesterase(CarE) activities were measured by the method of Byrne et al. (2000). Ten adult whiteflies from various host shifts were individually homogenized in 50mL of buffer (pH 6.0) containing 0.2 M phosphate and 1 g L^{-1} Triton X-100. Each sample was adjusted to 150 mL with the same buffer without Triton X-100 and maintained at 4 °C for 30 min to enhance CarE dissolution. Then, 50 mL were added to a fresh 96-well microplate. After adding 200 mL of solution containing 1 mM α-NA and 6 g L^{-1} Fast Blue RR salt to each well, the action was conducted by 12-channel multipipettes. CarE activities were determined continuously at 450 nm for 15 min. The results were expressed in nmol·min^{-1}·$mgPro^{-1}$, based on the OD450 and protein content.

Measurement of Glutathione S-Transferase (GST) Activities

Glutathione S-transferase (GST) activities were detected using 1-chloro-2,4-dinitrobenzene (CDNB) and reduced glutathione (GSH) as substrates and adopting the method of Stumpf &Nauen (2002). Ten adult whiteflies were manually homogenized in 300 mL Tris/HCl buffer (0.05 M, pH 7.5) and centrifuged at 10,000 × g at 4 °C for 5 min. The reaction solution, which contained 100 mL each of the supernatant, CDNB in ethanol (0.1% v/v) and GSH in buffer, for a final concentration of 0.4 mM CDNB and 4 mM GSH per well in 300 mL. The non-enzymatic sample of CDNB with GSH without homogenate was used as the control. GST activities were assayed using a kinetic microplate reader with 15 replicates, and every strain was measured at 340 nm for 5 min. The results were expressed innmol·min^{-1}·mgPro^{-1} based on the OD_{450} and protein content.

Measurement of Cytochrome-P450-Dependent Monooxygenase Activities

Cytochrome-P450-dependent monooxygenase (P450)activities were determined using 4-nitroanisole (PNA) as a substrate and adopting the method of Feng et al. (2010).Ten adult whiteflies were homogenized in buffer (0.1M, pH7.5) containing1mM EDTA, 0.1 mM DTT, 1 mM PTU and 1 mM PMSF. The homogenate solutions were centrifuged at 13,000 × g for 10 min at 4 °C. The resulting supernatants were centrifuged for 30 min as before. Using ultra-pure water, 700 mL of supernatant was adjusted to 1 mL. The 1 mL solution was used as a crude enzyme source. A 750 mL mixture including 375 mL of 2 mM PNA, 37.5 mL of 9.6 mM NADPH and 337.5 mL of the crude enzyme source was shaken constantly for 30 min at 34 °C. Two hundred mL of this mixture was then added to a well of a 96-well microplate and the absorbance was read at 405 nm. The non-enzymatic sample of PNA and NADPH without homogenate was used for the control. The results show the p-nitrophenol product (nmol·min^{-1}·mgPro^{-1}).

Measurement of the Protein Content

Protein content of the sample was determined using the method of Bradford (1976). The OD_{595} value was assayed in a 96-well microplate at 595nm. The protein content of the sample was calculated using a standard curve.

Data Analysis

Our purpose was to determine and compare the activities of the detoxifying enzymes, CarE, P450 and GST, of *B. tabaci* biotypes B and Q at 24 h after they had been transferred from cucumber to 55 other host plant species belonging to 29 families. Thus the data pertaining to CarE, P450 and GST activities, were expressed as the mean ± standard error (SE). To examine significant differences between biotypes B and Q, statistical analyses of the data obtained from each treatment was performed on 3 replicates using a Pair-Sample T Test with $P<0.05$, (SPSS Inc., Chicago, Illinois, USA).

Results

Table 1 shows the enzyme responses at 24 h following each host shift for carboxylesterase (CarE), cytochrome-P450-dependent monooxygenase (P450) and glutathione S-transferase (GST)from biotypes B and Q on 55, 35 and 34 plant species, respectively. The values shown are the outcome of the insects being transferred from cucumber to a different host species.

These results revealed that the post-transfer values of P450 were the highest, whereas those of GST were the lowest, except when whiteflies were shifted to white mulberry, *Morus alba* L. (Moraceae). CarE, P450 and GST values for both biotypes B and Q were generally observed in the following order, P450 > CarE > GST (Table 2). In addition, CarE and P450 values were generally higher in biotype Q than in biotype B. The value of P450 activity in biotype Q, which was 77.22 ± 5.46 nmol·min^{-1}·mgPro^{-1}, was the highest on the South American climber, *Mikaniamicrantha* Kunth (Compositae).

Pair-Samples T Test indicated that there were significant differences in P450 and CarE activities between biotypes B and Q at $P < 0.05$. However, no significant differences in GST activities were observed between biotypes B and Q, as the P value was 0.208 and thus larger than our cutoff for significance of $P < 0.05$ (Table 3).

Discussion

In these experiments, the activities of carboxylesterase (CarE), cytochrome P450-dependent monooxygenase (P450) and glutathione S-transferase (GST) of biotypes B and Q on 55, 35 and 34 plant species, respectively were shown to change when these insects were moved from cucumber onto a new host species. The various values obtained reflect effects of short-term host shifts. However, the data show a regularity with the post-shift values of P450 being the greatest and those of GST being the smallest, except for when the whiteflies were shifted to the white mulberry, *M. alba* L. In addition, the CarE and P450 values of biotype Q were greater than those of biotype Bat 24 h after each host shift. These results demonstrate that the effect of host shifts on whiteflies

TABLE 1. CARBOXYLESTERASE (CARE), CYTOCHROME-P450-DEPENDENT MONOOXYGENASE (P450) AND GLUTATHIONE S-TRANSFERASE (GST) ACTIVITIES OF *BEMISIA TABACI* BIOTYPES B AND Q AT 24 HOURS AFTER BEING SHIFTED FROM *CUCUMIS SATIVUS* TO EACH OF THE PLANT SPECIES LISTED.

		CarE ($nmol \cdot min^{-1} \cdot mgPro^{-1}$)		P450 ($nmol \cdot min^{-1} \cdot mgPro^{-1}$)		GST ($nmol \cdot min^{-1} \cdot mgPro^{-1}$)	
Family	Plant species	biotype B	biotype Q	biotype B	biotype Q·	biotype B	biotype Q
Cucurbitaceae	*Cucumis sativus* L.	52.49 ± 1.27	56.36 ± 3.04	65.35 ± 2.58	71.20 ± 4.31	9.09 ± 3.88	11.57 ± 1.63
	Benincasahispida	55.81 ± 1.30	56.98 ± 2.30	—	—	—	—
	Cucurbita moschata	42.01 ± 5.71	41.56 ± 1.12	51.47 ± 4.38	50.94 ± 12.30	24.38 ± 3.00	25.77 ± 2.26
	Lagenariasiceraria	41.21 ± 4.50	44.66 ± 5.36	—	—	—	—
	Luffa cylindrical	44.11 ± 6.72	45.12 ± 1.22	50.78 ± 2.94	54.55 ± 7.98	9.21 ± 1.71	13.16 ± 2.30
	Luffaacutangula	40.80 ± 2.11	38.20 ± 3.10	—	—	—	—
	Momordica charantia	54.01 ± 6.49	55.56 ± 2.52	58.71 ± 16.91	57.39 ± 4.42	7.55 ± 1.12	7.16 ± 1.34
Cruciferae	*Raphanussativus*var. *longipinnatus*	46.04 ± 7.07	43.01 ± 5.23	68.91 ± 18.93	77.20 ± 9.66	8.38 ± 2.66	11.24 ± 0.66
	Brassica parachinensis	25.64 ± 0.22	29.32 ± 2.50	56.27 ± 3.19	58.77 ± 6.71	18.02 ± 1.47	22.46 ± 3.38
	Brassica campestris subsp. chinensis var. *communis*	49.76 ± 4.54	56.11 ± 6.33	56.24 ± 19.44	61.51 ± 5.44	15.66 ± 1.24	16.12 ± 1.33
Labiatae	*Menthahaplocalyx*	44.17 ± 0.62	47.39 ± 5.33	—	—	—	—
	Perillafrutescens	28.37 ± 5.85	35.68 ± 4.22	49.67 ± 5.78	55.26 ± 3.55	7.96 ± 3.55	8.54 ± 2.12
	Solenostemonscutellarioides	52.91 ± 5.50	55.11 ± 0.78	—	—	—	—
	Salvia splendens	67.56 ± 6.52	66.41 ± 3.66	—	—	—	—
Malvaceae	*Hibiscus rosa-sinensis*	55.64 ± 0.78	63.40 ± 5.46	55.68 ± 12.60	58.45 ± 10.34	9.97 ± 2.15	15.13 ± 7.23
Moraceae	*Morus alba*	77.66 ± 6.38	79.56 ± 1.55	51.25 ± 2.96	56.47 ± 5.59	17.71 ± 3.66	19.59 ± 4.11
	Ficus virens	—	—	61.19 ± 15.62	65.71 ± 9.32	—	—
Oleaceae	*Jasminumsambac*	32.49 ± 5.07	35.12 ± 5.87	—	—	—	—
Solanaceae	*Capsicum annuum*	33.88 ± 5.97	31.62 ± 2.30	54.06 ± 10.74	55.53 ± 12.49	30.66 ± 4.86	26.40 ± 3.55
	Daturastramonium	66.18 ± 3.87	66.88 ± 6.63	—	—	—	—
	Lyciumchinense	25.59 ± 0.07	25.03 ± 1.22	—	—	—	—
	Lycospersicumesculentum	44.49 ± 3.56	47.36 ± 5.55	68.83 ± 9.46	71.11 ± 13.20	5.24 ± 0.44	4.91 ± 1.62
	Solanum melongena	46.13 ± 2.94	45.89 ± 1.10	64.14 ± 13.83	65.83 ± 10.34	10.02 ± 1.62	7.16 ± 1.54
Umbelliferae	*Apiumgraveolens var. dulce*	25.56 ± 0.20	34.56 ± 1.26	64.48 ± 13.93	63.14 ± 16.48	16.74 ± 1.79	15.68 ± 3.83
Verbenaceae	*Durantarepens*	52.47 ± 5.75	55.63 ± 1.47	—	—	—	—

Notes: The values were the average of 3 replicates. Values were mean ± SD. "-" was not determined.

TABLE 1. (CONTINUED) CARBOXYLESTERASE (CarE), CYTOCHROME-P450-DEPENDENT MONOOXYGENASE (P450) AND GLUTATHIONE S-TRANSFERASE (GST) ACTIVITIES OF *BEMISIA TABACI* BIOTYPES B AND Q AT 24 HOURS AFTER BEING SHIFTED FROM *CUCUMIS SATIVUS* TO EACH OF THE PLANT SPECIES LISTED.

		CarE ($nmol \cdot min^{-1} \cdot mgPro^{-1}$)		P450 ($nmol \cdot min^{-1} \cdot mgPro^{-1}$)		GST ($nmol \cdot min^{-1} \cdot mgPro^{-1}$)	
Family	Plant species	biotype B	biotype Q	biotype B	biotype Q·	biotype B	biotype Q
Vitaceae	*Vitisvinifera*	36.08 ± 2.17	37.28 ± 1.66	52.10 ± 9.68	51.62 ± 8.46	24.28 ± 1.55	31.28 ± 10.47
Rosaceae	*Rosa chinensis*	54.27 ± 6.16	56.06 ± 3.39	65.39 ± 30.22	70.29 ± 17.44	13.49 ± 2.75	16.39 ± 5.17
Passifloraceae	*Passifloraedulis*	39.75 ± 4.34	44.22 ± 1.35	56.74 ± 13.71	55.30 ± 9.73	12.94 ± 2.74	8.75 ± 2.11
Rubiaceae	*Gardenia jasminoides*	61.32 ± 3.51	66.22 ± 2.25	57.80 ± 15.97	61.12 ± 13.87	19.84 ± 0.93	21.81 ± 7.96
Apocynaceae	*Allamandacathartica*	55.95 ± 3.53	54.10 ± 4.46	—	—	—	—
Euphorbiaceae	*Codiaeumvariegatum*	51.02 ± 1.22	51.99 ± 2.56	59.10 ± 14.97	57.31 ± 5.27	11.49 ± 1.27	10.51 ± 5.70
	Manihotesculenta	54.03 ± 3.34	49.31 ± 3.88	—	—	—	—
Rutaceae	*Citrus madurensis*	41.96 ± 3.32	40.49 ± 1.01	—	—	—	—
	Clausenalansium	37.94 ± 4.29	38.64 ± 2.22	57.70 ± 11.06	63.74 ± 16.30	14.73 ± 2.28	14.71 ± 5.47
	Fortunella margarita	39.75 ± 0.95	38.11 ± 4.44	57.17 ± 17.09	68.98 ± 21.43	16.73 ± 2.74	20.32 ± 3.86
	Murrayapaniculata	54.63 ± 5.96	53.11 ± 1.23	—	—	—	—
Fabaceae	*Lablab purpureus*	60.58 ± 4.86	67.89 ± 5.39	—	—	—	—
	Vigna unguiculata	55.56 ± 5.23	54.44 ± 3.32	—	—	—	—
	Phaseolus vulgaris var. *humilis*	40.43 ± 5.32	38.12 ± 0.67	62.37 ± 10.71	63.76 ± 11.25	19.42 ± 1.30	12.43 ± 3.28
	Erythrinaarborescens	47.22 ± 5.29	45.33 ± 5.54	50.99 ± 5.35	51.12 ± 8.93	9.99 ± 1.29	17.83 ± 0.58
	Codariocalyxmotorius	55.45 ± 4.74	58.02 ± 1.33	—	—	—	—
Compositae	*Gerbera jamesonii*	56.45 ± 5.62	56.03 ± 5.77	—	—	—	—
	Mikaniamicrantha	30.78 ± 4.41	38.98 ± 5.13	71.57 ± 10.96	77.22 ± 5.46	24.40 ± 2.95	30.78 ± 11.69
	Lactuca sativa var. *angustana*	25.48 ± 0.12	28.46 ± 1.33	—	—	—	—
	Wedeliachinensis	57.94 ± 4.43	65.64 ± 6.11	58.04 ± 19.21	55.53 ± 15.32	12.34 ± 2.58	10.96 ± 2.69
Oxalidaceae	*Averrhoacarambola*	71.05 ± 4.51	76.89 ± 5.29	63.73 ± 18.62	69.41 ± 13.55	9.59 ± 1.64	9.01 ± 1.43
	Oxalis corniculata	76.11 ± 19.96	70.55 ± 6.32	47.07 ± 2.99	48.44 ± 10.20	20.64 ± 2.29	21.05 ± 5.32
Convolvulaceae	*Ipomoea batatas*	36.83 ± 5.27	41.47 ± 2.55	59.71 ± 18.44	62.36 ± 14.31	11.19 ± 1.57	8.43 ± 0.76
	Ipomoea nil	43.30 ± 3.91	45.67 ± 1.39	—	—	—	—
Boraginaceae	*Carmona microphylla*	55.39 ± 7.95	61.84 ± 4.25	57.70 ± 22.76	61.23 ± 15.35	20.11 ± 3.21	16.89 ± 7.31
Bombacaceae	*Pachiraaquatica*	44.91 ± 2.16	44.44 ± 3.68	52.17 ± 2.98	52.45 ± 6.25	24.09 ± 1.66	25.86 ± 6.99

Notes: The values were the average of 3 replicates. Values were mean ± SD. "-" was not determined.

TABLE 1. (CONTINUED) CARBOXYLESTERASE (CARE), CYTOCHROME-P450-DEPENDENT MONOOXYGENASE (P450) AND GLUTATHIONE S-TRANSFERASE (GST) ACTIVITIES OF *BEMISIA TABACI* BIOTYPES B AND Q AT 24 HOURS AFTER BEING SHIFTED FROM *CUCUMIS SATIVUS* TO EACH OF THE PLANT SPECIES LISTED.

		CarE ($nmol \cdot min^{-1} \cdot mgPro^{-1}$)		P450 ($nmol \cdot min^{-1} \cdot mgPro^{-1}$)		GST ($nmol \cdot min^{-1} \cdot mgPro^{-1}$)	
Family	Plant species	biotype B	biotype Q	biotype B	biotype Q·	biotype B	biotype Q
Araceae	*Colocasiaesculenta*	44.70 ± 1.22	44.06 ± 6.34	—	—	—	—
Ericaceae	*Rhododendron simsii*	57.83 ± 0.63	55.91 ± 1.14	55.91 ± 13.23	58.39 ± 11.17	21.51 ± 3.88	20.53 ± 7.60
Magnoliaceae	*Michelia alba*	51.11 ± 6.33	55.64 ± 5.31	59.20 ± 21.72	56.33 ± 7.15	12.89 ± 3.33	8.45 ± 2.30
Caprifoliaceae	*Lonicera Japonica*	39.40 ± 1.83	41.21 ± 2.85	53.89 ± 3.28	57.28 ± 7.19	4.83 ± 1.24	7.64 ± 3.29
Caricaceae	*Carica papaya*	48.03 ± 6.28	51.41 ± 7.21	64.78 ± 17.41	65.22 ± 21.44	13.25 ± 2.59	14.11 ± 5.58
Basellaceae	*Basella alba*	44.51 ± 2.23	43.15 ± 5.17	—	—	—	—
Asclepiadaceae	*Telosmacordata*	—	—	59.37 ± 19.41	61.94 ± 17.85	17.38 ± 2.39	21.60 ± 3.79

Notes: The values were the average of 3 replicates. Values were mean ± SD. "-" was not determined.

TABLE 2. MULTIPLE MEANS COMPARISON OF PAIRED SAMPLES BETWEEN CARBOXYLESTERASE (CARE), GLUTATHIONE S-TRANSFERASE (GST), AND CYTOCHROME-P450-DEPENDENT MONOOXYGENASE (P450) ACTIVITIES OF *BEMISIA TABACI* BIOTYPES B AND Q AT 24 HOURS AFTER BEING SHIFTED FROM *CUCUMIS SATIVUS* TO EACH OF THE PLANT SPECIES LISTED IN TABLE 1.

Descriptives				ANOVA				
Paired	N	M ± S D		Sum of Squares	df	Mean Square	F	Sig.
P450-CarE	33	11.00 ± 15.09 a*	Between Groups	17276.17	2	8638.08	47.84491	<0.001
CarE-GST	33	31.85 ± 14.85 b	Within Groups	17332.17	96	180.54		
P450-GST	33	42.86 ± 9.65 c	Total	34608.33	98			

*Harmonic means in the column followed by different letters are significantly different based on Duncan's test at $\alpha = 0.05$.

TABLE 3. PAIRED SAMPLES T TESTS FOR CARBOXYLESTERASE (CARE), GLUTATHIONE S-TRANSFERASE (GST), AND CYTOCHROME-P450-DEPENDENT MONOOXYGENASE (P450) ACTIVITIES OF *BEMISIA TABACI* BIOTYPES B AND Q AT 24 HOURS AFTER BEING SHIFTED FROM *CUCUMIS SATIVUS* TO EACH OF THE PLANT SPECIES LISTED IN TABLE 1.

			N	Paired Correlations	Samples	Paired Differences*			
Samples		Mean ± SD		Correlation	Sig.	Mean ± SD	t	df	Sig. (2-tailed)
CarE: B-Q	biotype B	47.68 ± 12.27	55	0.961	0.000	-1.683 ± 3.444	-3.625	54	0.001
	biotype Q	49.36 ± 12.25	55						
P450:B-Q	biotype B	58.12 ± 5.86	35	0.896	0.000	-2.478 ± 3.189	-4.597	34	<0.001
	biotype Q	60.60 ± 7.10	35						
GST: B-Q	biotype B	15.20 ± 6.26	34	0.872	0.000	0.087 ± 3.084	-1.285	33	0.208
	biotype Q	15.96 ± 7.11	34						

Notes: * Paired Samples T Test At $\alpha = 0.05$

is very complicated. Feeding on different hosts affects the levels of expression of the different detoxifying enzymes, CarE, P450 and GST.

Our findings are in accord with previous results, which showed the effect of host induction in biotype B of CarE and GST activities after inter-species transfer(Liang et al. 2007; Deng et al. 2013). Feng et al. (2010) demonstrated that biotype B can develop resistance to thiamethoxam and that enhanced detoxifying capacities of CarE and P450 were the major causes for this resistance. These results are also in agreement with the important role P450 plays in detoxification of host phytochemicals in herbivorous insects(Despres et al. 2007; Li et al. 2007; Alon et al. 2010; Castaneda et al. 2010; Zhou et al. 2010; Schuler 2011; Deng et al. 2013). P450s and their associated P450 reductases can mediate resistance to all classes of insecticides (Feyeseisen, 2005). Li et al. (2002) showed that jasmonate and salicylate could induce the expression of cytochrome P450 genes in herbivores and increase the production of this detoxifying enzyme. The known cross-resistance between some neonicotinoid insecticides (e.g. imidacloprid) and pymetrozine in *B. tabaci* is associated with its hydroxylation by constitutively over-expressed *CYP6CM1*, a cytochrome P450 enzyme (Karunker et al. 2008; Longhurst et al. 2013; Nauen et al. 2013).

In various insect species, genes encoding members of the CarE, P450 and GST families have been most frequently associated with resistance to a range of different insecticides (Li et al. 2007). The increased population of biotype Q in many countries may be due to the application of insecticides because biotype Q possesses greater resistance to insecticides than biotype B (Horowitz et al. 2005; Dennehy et al. 2010). Relatively higher resistance to insecticides in biotype Q has been suggested to be a key factor in its displacement of biotype B in many regions in southern Spain and Israel (Pascual & Callejas 2004; Khasdan et al. 2005). Similarly, recent reports on the status of insecticide resistance of biotypes B and Q in China, show that biotype Q is significantly more resistant to nearly all commonly applied insecticides than biotype B (Luo et al. 2010; Wang et al. 2010; Kontsedalov et al. 2012; Rao et al. 2012; Yuan et al. 2012). Sun et al. (2013) demonstrated that the displacement of biotype B by Q takes a few generations following the application of imidacloprid. Further more, the higher the dosage of insecticide applied, the more rapid the displacement. Our results suggest that the activities of CarE, P450 and GST from biotypes B and Q only contribute partially to the dominance of biotype Q over biotype B.

The best assay time was based on preliminary experiments which determined 24 h post host shift from *C. sativus* to other host plants to be the suitable time. Although the time assayed allows

only a short-term induction, all whiteflies detoxifying enzymes were compared with the whitefly cucumber population, with the results showing a certain regularity. Our results, together with previous studies, support that *B. tabaci* biotypes utilizes multiple detoxification enzymes when encountering various host shifts.

Endnotes

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