# Determination of resistance improving potentials of cotton whitefly *Bemisia tabaci* (Genn.) (Hemiptera: Aleyrodidae) biotypes against cyantraniliprole

Isse Hassan Ali¹, Utku Yükselbaba²\*

**Citation:** Ali I.H., Yükselbaba U. (2025): Determination of resistance improving potentials of cotton whitefly *Bemisia tabaci* (Genn.) (Hemiptera: Aleyrodidae) biotypes against cyantraniliprole. Plant Protect. Sci., 61: 191–200.

Abstract: Bemisia tabaci is a significant insect pest that causes extensive agricultural damage. The MEAM1 (Middle East-Asia Minor) and MED (Mediterranean) genetic groups of  $B.\ tabaci$  are the most prevalent cryptic species. This study investigated the potential for resistance development in the MED and MEAM1 genetic groups of  $B.\ tabaci$  against cyantraniliprole. Additionally, multiple-resistance development within each genetic group for pyriproxyfen, spirote-tramat, and acetamiprid insecticides was determined. To assess the susceptibility of the first-larval stage of  $B.\ tabaci$ , we employed a systemic uptake method with cyantraniliprole. Additionally, we utilized a leaf-dipping method to apply acetamiprid, spirotetramat, and pyriproxyfen to second-instar larvae, adults, and eggs of  $B.\ tabaci$ , respectively, to determine the  $LC_{50}$  values for each insecticide. Each genetic group was subjected to six selections using cyantraniliprole. After six rounds of selection, a 1.8-fold resistance was observed in the  $B.\ tabaci$  MEAM1 population, whereas the MED population exhibited a 1.4-fold resistance. While  $B.\ tabaci$  MED and MEAM1 genetic groups exhibited very low resistance to cyantraniliprole, it's important to note that no multiple-resistance was observed with pyriproxyfen, spirotetramat, or acetamiprid in either group. These findings provide valuable insights for future monitoring and developing insecticide resistance management strategies for  $B.\ tabaci$ .

Keywords: cross-resistance diamide; insecticide; resistance management

The cotton whitefly, *Bemisia tabaci* (Genn.), is globally recognized as an important insect in the Aleyrodidae family, known for its harmful effects on crops (Mound & Halsey 1978; Gerling 1990). *Bemisia tabaci* has been reported to cause damage to more than 600 plant species and to be the vector of more than 200 plant viruses, most notably

TYLCV (De Barro et al. 2011; Navas-Castillo et al. 2011). The cotton whitefly impacts plants by feeding on plant sap and excreting honeydew, thus preventing the plant from photosynthesizing and causing yield and quality loss. They have been reported to directly cause substantial economic damage or transmit viral diseases (Martin & Mound 2007;

<sup>&</sup>lt;sup>1</sup>Department of Plant Protection, Faculty of Agriculture, Akdeniz University, Institute of Natural and Applied Sciences, Antalya, Türkiye

<sup>&</sup>lt;sup>2</sup>Department of Plant Protection, Faculty of Agriculture, Akdeniz University, Antalya, Türkiye

<sup>\*</sup>Corresponding author: uyukselbaba@akdeniz.edu.tr

Supported by the Akdeniz University' scientific research projects coordination unit project number FYL-2021-5682. The abstract of this study was presented as an oral presentation at the IV. Balkan Agricultural Congress (August 31 – September 2, 2022, Edirne, Turkey).

<sup>©</sup> The authors. This work is licensed under a Creative Commons Attribution-NonCommercial 4.0 International (CC BY-NC 4.0).

De Barro et al. 2011). *B. tabaci* is a species complex with several genetic groups (Perring 2001; De Barro et al. 2011). However, MED and MEAM1 genetic groups are the two most common cryptic species (De Barro et al. 2011). Reproductive and feeding behaviours, host plant preferences, relationships with plant viruses, and especially insecticide sensitivity are all associated with these genetic groups in *B. tabaci*. (Xie et al. 2014).

Insecticides play a pivotal role in controlling B. tabaci and are extensively used as a primary strategy to manage this pest. Consequently, insecticide resistance has been reported in many regions worldwide, affecting most classes of insecticides, including organophosphates, pyrethroids, and neonicotinoids (Wang et al. 2010; Basit et al. 2013; Dângelo et al. 2018; Roy et al. 2019). Furthermore, these insecticides have been shown to pose a relatively high level of toxicity to non-target organisms, including arthropods and even humans (Zeng et al. 2013; Cimino et al. 2016). To mitigate the development of insecticide resistance, it is essential to develop novel modes of action that selectively impact pests while sparing non-target species and natural enemies (Sattelle et al. 2008). The ryanodine receptor (RyR) is a target for creating new insecticides focusing on selectivity and minimal mammalian toxicity, owing to variations among insect orders (Qi & Casida 2013; Qi et al. 2014; Casida 2015). Diamide insecticides influence the regulation of calcium release from intracellular stores to the sarcoplasmic reticulum, leading to muscle contractions and functioning as ryanodine receptor modulators, ultimately paralyzing the targeted pests (Sattelle et al. 2008).

To date, five diamide insecticides have been commercialized, namely flubendiamide (a phthalic diamide) and chlorantraniliprole, cyclaniliprole, tetraniliprole, and cyantraniliprole (all belonging to the Anthranilic diamides class). Among the earliest diamides, flubendiamide and chlorantraniliprole displayed high selectivity for Lepidopteran pests (Jeanguenat 2013). Cyantraniliprole, part of the new anthranilic diamide class, represents the second xylem systemic active substance. It offers a broad control spectrum, encompassing Lepidoptera, Hemiptera, Coleoptera, and other orders (Caballero et al. 2013; Barry et al. 2015). Due to its unique mode of action, the use of cyantraniliprole has been reported not to induce cross-resistance with conventional insecticides, making it a valuable tool for delaying resistance development to other insecticides in *B. tabaci* (Grávalos et al. 2015). While numerous studies have explored whitefly susceptibility to various insecticides, research on resistance to cyantraniliprole remains limited (Wang et al. 2019). In China, shortly after the introduction of cyantraniliprole, reports indicated low to moderate resistance in the MED genetic group of B. tabaci (Yao et al. 2017; Wang et al. 2018). However, no change in sensitivity to cyantraniliprole has been observed in European countries such as Spain, Italy, and Greece (Grávalos et al. 2015). In contrast, high levels of resistance to cyantraniliprole have been documented in P. xylostella populations in China and Brazil (Liu et al. 2015; Ribeiro et al. 2017). Resistance to more than sixty active substances has been detected in Bemisia tabaci (Horowitz et al. 2020). Cyantraniliprole represents a novel insecticide with a unique mode of action against B. tabaci, offering the potential for use in rotation with several other insecticides to proactively delay the development of resistance (Wang et al. 2018). This study aimed to determine the potential for resistance development in B. tabaci exposed to a small number of ongoing selection pressures with cyantraniliprole and the development of multiple resistances as a result of selection. In addition, the study aimed to compare the potential for resistance development to cyantraniliprole between the MED and MEAM1 genetic groups. The goal is to contribute to developing resistance management strategies against cyantraniliprole in B. tabaci MEAM1 and MED. To achieve this, the MEAM1 and MED genetic groups of B. tabaci were subjected to six rounds of selection with cyantraniliprole using a systemic uptake method. Additionally, the study investigates the development of multiple resistances to different insecticides (pyriproxyfen, spirotetramat, and acetamiprid) within each genetic group due to selection with cyantraniliprole.

# MATERIALS AND METHODS

# Insecticides

In the study, insecticides were used for all bioassays, including diamide cyantraniliprole (CIRCA-DEN DuPont<sup>™</sup>, USA, 200 g/L), tetramic acid spirotetramat (Bayer MOVENTO, Germany, 100 g/L), neonicotinoid acetamiprid (MOSPILAN Sumi Agro, 20%), and insect growth regulator pyriproxyfen (AD-MIRAL Sumitomo, Japan, 50 g/L). Commercially

available formulations of these insecticides were employed in the study.

## **Cotton plants**

Carisma (Progen Hatay/Turkiye) cotton seeds were planted in a peat-perlite mixture within 180 mL plastic cups. These seeds were planted in climate chambers at a temperature of  $26 \pm 1$  °C, relative humidity of  $60 \pm 10$ %, light intensity at 2 400 lux, and a 16:8 (L:D) lighting period. No insecticides were applied during this period to use the cotton plants in the study. Cotton plants aged 25-30 days were employed for whitefly reproduction and insecticide bioassays.

#### **Insects**

The study collected *B. tabaci* MED and MEAM1 populations from the Gaziler and Serik districts of Antalya province in 2019. Genetic groups were identified based on mitochondrial cytochrome oxidase I (mtCOI) sequence information. These populations were reared on cotton plants in controlled climate chambers with conditions set at  $26 \pm 1^{\circ}$ C temperature,  $60 \pm 10\%$  relative humidity, 2 400 lux light intensity, and a 16:8 hour L:D photoperiod. The reared Meam1 and MED main stock populations were named B-mn and Q-mn, respectively, and lethal concentration values for all insecticides were determined before initiating selection studies. The Meam1 and MED populations from the selection studies were coded as B-sel and Q-sel, respectively.

# Insecticide bioassays

Determination of LC<sub>50</sub> values of Bemisia tabaci genetic groups against cyantraniliprole. A systemic uptake method was used to determine the lethal concentration values of the first larval stage of genetic groups against cyantraniliprole. Following the procedure outlined by Li et al. (2012), approximately 15 B. tabaci adults were placed in small cages called 'Clip Cages', using a mouth aspirator to collect individuals from the population. They were left for 24 h for oviposition. After this period, the adults were removed, and the white eggs on the leaves were counted and recorded. Shoots containing whitefly eggs were cut, leaving about 12 cm from the leaf, using clean scissors. These shoots were then placed in tubes containing 40 mL of various insecticide solutions, including Triton-X, with a pure Triton-X solution as the negative control. Subsequently, the tubes were placed in climate chambers with conditions set at  $26 \pm 1$  °C temperature,  $60 \pm 10\%$  relative humidity, and a 16:8 h (L:D) photoperiod for 12 days (Li et al. 2012). After 12 days of insecticide application, the leaves were examined under a stereo microscope. Larvae that had developed at least to the  $2^{\rm nd}$  larval stage were considered alive, while dried larvae and those that had not progressed beyond the 1st stage were categorized as affected or dead. Mortality was determined by subtracting the number of surviving larvae from the total number of eggs.

Determination of susceptibility of Bemisia tabaci populations to pyriproxyfen. We determined the LC<sub>50</sub> values of populations against pyriproxyfen using the leaf-dipping method, following the procedure described by Li et al. (2003). To assess the impact of Pyriproxyfen on egg hatching, we collected at least 15 B. tabaci adults from the population in a mixed form using a mouth aspirator and placed them in small cages. These cages were then affixed to the undersides of cotton leaves using clips. They were kept in climate chambers maintained at a temperature of 26  $\pm$  1 °C, 60  $\pm$  10% relative humidity, and a 16:8 h (L:D) photoperiod for 24 h to allow whiteflies to lay eggs on the leaves. After 24 h, we removed the *B. tabaci* adults and counted and noted the eggs on the leaves using a stereo microscope. Each leaf had at least 35 eggs. Leaves with whitefly eggs were then immersed for 10 sec in 100 mL of various insecticide solutions containing Triton-X, with a pure Triton-X solution as the negative control. These plants were kept in the climate chamber for 12 days (Li et al. 2003). After 12 days, we examined the leaves under a microscope and counted the surviving larvae on the leaves. Egg hatching rates were calculated by subtracting the number of live larvae from the initial total number of eggs.

**Determination of the susceptibility of** *Bemisia tabaci* **populations to spirotetramat.** For spirotetramat,  $LC_{50}$  values of populations were determined using the leaf-dipping method on the second larval stage of *B. tabaci*, following the procedure described by Bielza et al. (2019). At least 20 *B. tabaci* adults were collected from the population, gathered in mixed form using a mouth aspirator and placed in small cages. These cages were affixed to the lower surfaces of cotton leaves with clips and maintained in climate chambers at a temperature of  $26 \pm 1$  °C,  $60 \pm 10\%$  relative humidity, and a 16:8 h (L:D) photoperiod for 24 h, allowing whiteflies to lay eggs on

the leaves. After this period, the *B. tabaci* adults were removed, and the cotton plants were left undisturbed for 10 days in the same climate chamber to allow the eggs to hatch and the whitefly larvae to develop into the 2<sup>nd</sup> stage (Bielza et al. 2019). Following these 10 days, using an insect needle under a microscope, larval stages other than the 2<sup>nd</sup> stage were removed, and the 2<sup>nd</sup> stage larvae of B. tabaci on the leaves were counted and recorded. Leaves bearing the 2<sup>nd</sup> stage larvae were then briefly immersed for 10 sec in 100 mL of various insecticide solutions containing Triton-X, with a pure Triton-X solution serving as the negative control. These plants were kept in the climate chamber for an additional 6 days. After 6 days, the leaves were examined, and larvae that had advanced beyond the 2<sup>nd</sup> stage were considered alive, while dried larvae and those that had not progressed beyond the 2<sup>nd</sup> stage were categorized as affected and deceased. Mortality rates of the larvae were calculated by subtracting the number of surviving 2<sup>nd</sup> instar larvae from the initial total count.

Determination of the susceptibility of Bemisia tabaci populations to acetamiprid. We employed the leaf dip method, as described by Elbert and Nauen (2000), to determine the LC<sub>50</sub> values of B. tabaci adults against acetamiprid. Cotton leaves, each with a diameter of 3.5 cm, were cut and briefly immersed for 5 sec in 100 mL of various insecticide solutions containing Triton-X. A pure Triton-X solution was used as a negative control. Subsequently, the leaves were allowed to dry for approximately two hours on a damp napkin. These dried leaves were then positioned in Petri dishes containing 1.5% agar with the lower surface facing upwards. B. tabaci adults were transferred from cotton plants in breeding cups to the prepared leaf discs by gently shaking them. The Petri dishes were covered with gauze to facilitate whiteflies' respiration and prevent their escape. The Petri dishes were maintained in climate chambers at  $26 \pm 1$  °C,  $60 \pm 10\%$  relative humidity, and a 16:8 h (L:D) photoperiod for 72 h. After this period, the leaves were examined under a stereo microscope, and mortality rates of B. tabaci adults were calculated. Adults who exhibited no movement when touched with a brush and did not display any signs of movement were considered dead. Mortality rates were determined by subtracting the number of surviving adults from the total number of adults.

Selection of Bemisia tabaci genetic groups with cyantraniliprole. The selection of populations with cyantraniliprole was conducted as described in the section 'Determination of LC<sub>50</sub> Values of Bemisia tabaci Genetic Groups Against Cyantraniliprole'. Each selection involved the use of at least 1 000 eggs. After determining the lethal concentration values for the MED and MEAM1 groups, each genetic group was separately exposed to a dose of 0.4 mg/L. Surviving individuals were transferred to new cages for reproduction and subsequent selection studies after each selection. Whitefly populations were bred in these cages to ensure sufficient reproduction, and reselection was initiated in the earliest part of the 2<sup>nd</sup> generation. Each selection was replicated at least 10 times, and this process was repeated six times for each group.

## Data analysis

Following the insecticide bioassays,  $LC_{50}$  and  $LC_{90}$  values of the populations and their 95% confidence intervals were calculated through probit analysis using POLO-PC [Leora Software 2008 (version 2.0)]. Populations were considered significantly different when the 95% confidence intervals of their  $LC_{50}$  values did not overlap. When necessary, mortality rates were corrected based on the control results using the Abbott formula (Abbott 1925). After the selections, resistance ratios were calculated by dividing the final  $LC_{50}$  value of the populations by the initial  $LC_{50}$  value.

## **RESULTS**

Lethal concentration values of MED and MEAM1 populations, both before and after selection, are provided in Table 1. Before and after selection, the probit analysis values of genetic groups to pyriproxyfen, spirotetramat, and Acetamiprid are presented in Tables 2, 3, and 4, respectively. Additionally, the lethal concentration values of the genetic groups are presented graphically in Figure 1.

When Table 1 examined the end of six selections with cyantraniliprole, the B-sel and Q-sel populations slightly increased their  $LC_{50}$  levels compared to their respective B-mn and Q-mn populations. In the case of the B-sel MEAM1 population, the  $LC_{50}$  value rose from 0.298 mg/L to 0.553 mg/L, resulting in a 1.8-fold resistance ratio; similarly, for the Q-sel MED population, the  $LC_{50}$  value in-

 $Table~1.~LC_{50}~values~of~\textit{Bemisia~tabaci}~MEAM1~and~MED~populations~before~and~after~selection~with~cyantraniliprole~and~after~selection~with~after~sel$ 

Population code	п	Slope ± SE	LC <sub>50</sub> (mg/L) (%95 CL)	LC <sub>90</sub> (mg/L) (%95 CL)	RD (mg/L)	$X^2$	df	Н	RR
B-mn	2 364	$2.65 \pm 0.12$	0.298 (0.260–0.330)	0.906 (0.780–1.090)	100	78.32	38	2.06	1.0
B-sel	2 093	$2.42 \pm 0.13$	0.553 (0.480–0.640)	1.869 (1.470–2.610)	100	63.12	23	2.74	1.8
Q-mn	2 749	$2.04 \pm 0.09$	0.353 (0.310-0.390)	1.500 (1.210–1.990)	100	73.01	35	2.09	1.0
Q-sel	2 857	$3.15 \pm 0.22$	0.478 (0.420–0.540)	1.220 (0.990–1.670)	100	88.52	31	2.86	1.4

Table 2. Multiple-resistance status to pyriproxyfen in MEAM1 and MED populations after cyantraniliprole selection

Population code	п	Slope ± SE	LC <sub>50</sub> (mg/L) (%95 CL)	LC <sub>90</sub> (mg/L) (%95 CL)	RD (mg/L)	$X^2$	df	Н	RR
B-mn	3 805	$0.38 \pm 0.03$	1 698.994 (711.200–5 056.600)	3 941 556.068 (498 962–122 094 518)	50	148.12	41	3.61	1.0
B-sel	2 707	$0.31 \pm 0.02$	1 034.052 (575.600–2 004.400)	13 460 317.720 (3 270 945–80 769 818)	50	37.62	31	1.21	0.6
Q-mn	3 415	$0.48 \pm 0.03$	1 005.880 (308.100-4 193.700)	1 192 967.830 (104 016–300 225 805)	50	268.40	38	7.09	1.0
Q-sel	1 854	$0.42 \pm 0.03$	387.239 (244.100–633.300)	438 259.790 (165 602–1 476 474)	50	26.51	30	0.88	0.4

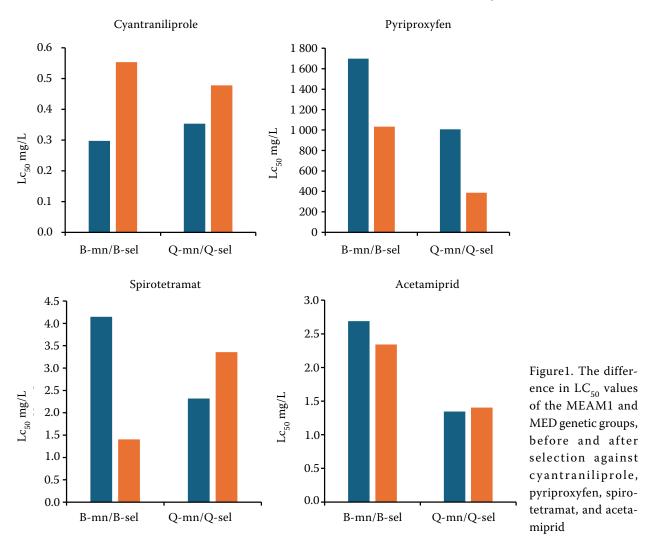
Table 3. Multiple-resistance status to spirotetramat in MEAM1 and MED populations after cyantraniliprole selection

Population code	п	Slope ± SE	LC <sub>50</sub> (mg/L) (%95 CL)	LC <sub>90</sub> (mg/L) (%95 CL)	RD (mg/L)	$X^2$	df	Н	RR
B-mn	1 782	0.99 ± 0.05	4.150 (2.550–6.270)	81.122 (52.380–138.370)	100	89.83	34	2.64	1.0
B-sel	749	$0.80 \pm 0.06$	1.403 (0.890–2.140)	56.042 (31.920–113.260)	100	22.50	22	1.02	0.3
Q-mn	2 197	$0.68 \pm 0.03$	2.320 (1.060–4.500)	181.630 (83.850–504.780)	100	202.95	35	5.80	1.0
Q-sel	1 627	$0.60 \pm 0.03$	3.360 (1.930–5.910)	452.393 (176.950–1 660.410)	100	83.86	22	3.80	1.4

Table 4. Multiple-resistance status to acetamiprid in MEAM1 and MED populations after cyantraniliprole selection

Population code	п	Slope ± SE	LC <sub>50</sub> (mg/L) (%95 CL)	LC <sub>90</sub> (mg/L) (%95 CL)	RD (mg/L)	$X^2$	df	Н	RR
B-mn	972	1.90 ± 0.18	2.689 (2.350–3.300)	12.69 (10.12–17.17)	60	45.09	44	1.02	1.00
B-sel	1 276	$1.95 \pm 0.14$	2.342 (2.000–2.700)	10.59 (8.45–14.31)	60	85.18	45	1.90	0.90
Q-mn	581	1.49 ± 0.11	1.346 (0.860–2.060)	9.69 (5.56–22.95)	60	96.70	22	4.40	1.00
Q-sel	641	1.51 ± 0.11	1.404 (0.970–1.97)	9.86 (6.26–18.90)	60	70.02	22	3.18	1.04

n – number of individuals used in bioassay; RD – recommended doze; H – heterogeneity; RR – resistance ratio;  $X^2$  – Chisquare; Df – degrees of freedom



creased from 0.353 mg/L to 0.478 mg/L, resulting in a 1.4-fold resistance ratio (Table 1 and Figure 1). The 95% confidence intervals of B-sel and Q-sel populations did not overlap with their respective main stock (B-mn and Q-mn), confirming the significance of the change in  $LC_{50}$  values in the two *B. tabaci* genetic groups.

After the selection studies, the multiple-resistance of cyantraniliprole with pyriproxyfen, spirotetramat and acetamiprid insecticides was determined as the following.

After six selections, the  $LC_{50}$  value of pyriproxyfen in the B-sel population changed from 1 698.994 mg/L to 1034.052 mg/L, resulting in a 0.6-fold resistance ratio. This indicates that the B-sel population lost 0.4-fold of its resistance ratio. Similarly, an increase in susceptibility was observed in the Q-sel population as the  $LC_{50}$  value changed from 1 005.88 mg/L to 387.239 mg/L, resulting in a 0.4-fold resistance ratio (Table 2 and

Figure 1). Upon examining Table 2, it becomes apparent that the 95% confidence interval of the B-sel and Q-sel population's  $LC_{50}$  values overlaps with their respective main stock. Consequently, it was determined that there is no cross-resistance of pyriproxyfen with cyantraniliprole in the *B. tabaci* MEAM1 and MED populations.

Before the selection, the  $LC_{50}$  value of spirotetramat in the B-mn population was 4.150 mg/L. After the selection, this value decreased to 1.403 mg/L, resulting in a 0.3-fold resistance ratio. Unlike the  $LC_{50}$ , the value of spirotetramat in the MED population increased from 2.32 mg/L to 3.36 mg/L, resulting in a 1.4-fold resistance ratio (Table 3 and Figure 1). Notably, there is an overlap in the 95% confidence interval between the initial and final  $LC_{50}$  values in the two populations. This suggests that no cross-resistance between spirotetramat and cyantraniliprole was observed in the *B. tabaci* MEAM1 and MED populations.

The results obtained from the bioassay show that the  $LC_{50}$  value of the B-sel population against acetamiprid changed from 2.689 mg/L to 2.342 mg/L at the end of the selections, resulting in a 0.9-fold resistance ratio (Table 4 and Figure 1). Conversely, the  $LC_{50}$  value of the Q-sel population against acetamiprid increased from 1.346 mg/L to 1.404 mg/L, leading to a 1.04-fold resistance ratio. Despite these changes, overlaps between the initial and final LC50 values showed no cross-resistance between acetamiprid and cyantraniliprole in both the *B. tabaci* MEAM1 and MED populations.

#### **DISCUSSION**

Understanding resistance levels, cross-resistance patterns, and resistance mechanisms to a new insecticide is essential for designing sustainable pest management strategies. Cyantraniliprole, a novel anthranilic diamide insecticide, has yet to witness resistance in many significant pests (Wang et al. 2019). In this study, we observed very low resistance development in B. tabaci populations following selections with cyantraniliprole. A two-year susceptibility follow-up of B. tabaci MED and MEAM1 populations in the USA revealed their high susceptibility, with LC<sub>50</sub> levels ranging from 0.015 to 0.191 mg/L and resistance ratios ranging from 0.94 to 11.94-fold to cyantraniliprole (Li et al. 2012). Likewise, Grávalos et al. (2015) conducted a seven-year study monitoring changes in sensitivity to cyantraniliprole in B. tabaci MED populations in Spain. They determined that MED populations remained highly sensitive to cyantraniliprole, with LC<sub>50</sub> values between 0.011 and 0.116 mg/L and a resistance rate of 10.5-fold. In their study, Xie et al. (2014) examined the sensitivity of two field B. tabaci egg and larval stages to cyantraniliprole. They found that both stages were highly sensitive, with observed LC<sub>50</sub> levels ranging between 5.74 and 8.65 mg/L for eggs and less than 0.1 mg/L for larvae.

In Turkey, this study marks the first assessment of the potential for developing resistance to cyantraniliprole, and it revealed very low resistance development. The findings align with only a limited number of similar research efforts. Unlike the current study, Wang et al. (2019) reported a significant 138.4-fold increase in resistance in the *B. tabaci* MED population after 16 generations of selection

with cyantraniliprole. In contrast, our study reveals a low resistance rate, which may be attributed to the number of selections conducted. If our B. tabaci populations were subjected to more extensive selection pressure, similar to Wang et al. (2019), we might observe a higher resistance rate. While there aren't enough studies on the selection of B. tabaci's MED and MEAM1 genetic groups with cyantraniliprole, research on the selection of B. tabaci against various insecticides does exist (Grávolos et al. 2015). For instance, Horowitz et al. (2005) observed an approximately 30-fold increase in resistance development in the Q biotype after 26 generations of selection of *B. tabaci* Q biotype against acetamiprid in Israel in 1999. They further reported moderate resistance rates of around 50fold and 40-fold against acetamiprid and thiamethoxam, respectively, after 21 generations of selection in the Q biotype population collected from the same location in 2002. Their study differs from ours in several ways. Acetamiprid and thiamethoxam have different modes of action compared to cyantraniliprole, and they subjected the B. tabaci population to more extensive selection pressure. Additionally, it's worth noting that cyantraniliprole is a novel insecticide, which may account for the limited resistance development observed in our study. These findings highlight the potential of cyantraniliprole as an effective insecticide that can be used in rotation with other insecticides. In a study by Yükselbaba (2015), resistance rates of 213-fold and 1.35-fold were determined in B. tabaci Q and B biotypes, respectively, after five selections with cypermethrin. Additionally, they observed resistance rates of 15.3-fold and 4-fold in B. tabaci Q and B biotypes, respectively, after seven selections with pyriproxyfen. Their study indicated that the Q biotype might develop higher resistance to cypermethrin and pyriproxyfen in a shorter timeframe than the B biotype.

Our current study observed that the potential for resistance development in MEAM1 and MED populations was nearly the same after selection with cyantraniliprole. Resistance development to cyantraniliprole was expected to be low because *B. tabaci* populations used in this study had not been previously exposed to this novel insecticide. Cyantraniliprole has been approved for use against *B. tabaci* in Turkey since 2015. Because its use is relatively recent, it is expected to be more effective against *B. tabaci* than other insecticides currently

in use in Turkey. Furthermore, it operates through a unique mode of action distinct from other insecticides. This makes it an efficient alternative for controlling insects with piercing-sucking mouthparts. However, it should not be used consistently, as prolonged and exclusive use can lead to the development of resistance.

The development of resistance to cyantraniliprole has been reported in other pest species in China. Sang et al. (2016) discovered significant regional variations in sensitivity to cyantraniliprole in Spodoptera litura (Fabricius) populations collected from different areas in southern China. They observed that prolonged exposure to the insecticide could lead to resistance development in S. litura. Likewise, Zhang et al. (2014) identified  $LC_{50}$  values exceeding 10 μg/g in three Bactrocera dorsalis (Hendel) field populations when exposed to cyantraniliprole. This resulted in a 19.44-fold increase in resistance after 14 selection cycles against the insecticide. The study also investigated multiple resistance to pyriproxyfen, spirotetramat, and acetamiprid in cyantraniliprole-selected B. tabaci MED and MEAM1 populations. No cross-resistance was observed in the cyantraniliprole-selected MEAM1 and MED populations against pyriproxyfen, spirotetramat, and acetamiprid, as their confidence intervals overlapped. Before selection, all B. tabaci populations displayed high resistance to pyriproxyfen and moderate resistance to spirotetramat and acetamiprid. However, no multiple-resistance to cyantraniliprole was observed in the resistant populations. Grávolos et al. (2015) conducted seven generations of selection with laboratory populations of *B. tabaci* using cypermethrin, azadirachtin, buprofezin, pymetrozine, pyridaben, pyriproxyfen, spiromesifen, and thiamethoxam and they found no cross-resistance to cyantraniliprole. Li et al. (2012) determined that there was no cross-resistance between chlorantraniliprole, cyantraniliprole, imidacloprid, and pyriproxyfen in B. tabaci B-biotype field populations and Q-biotype laboratory populations in Arizona. Due to its high efficacy and lack of multiple-resistance to other insecticides, cyantraniliprole may be crucial in mitigating pesticide resistance in B. tabaci (Grávolos et al. 2015).

Although resistance development was observed after six selections of *B. tabaci* MED and MEAM1 genetic groups with cyantraniliprole, resistance levels were found to be very low. Low resistance patterns to insecticides have practical implications

for managing insects, as they increase the efficacy and sustainability of control strategies.

In areas where whitefly populations are resistant to certain insecticides, cyantraniliprole can be adopted as an effective solution for several years, reducing the need for frequent and high-dose applications of these products and may contribute significantly to insecticide resistance management. Since the development rate of resistance to cyantraniliprole is lower compared to other insecticides, it can remain effective for an extended period. Less use of insecticides in insect control slows the development of resistance. Long-term efficacy and a reduced number of applications have lower environmental impacts, as fewer applications mean less exposure of non-target organisms and surrounding ecosystems to chemicals.

#### **CONCLUSION**

To the best of our study, no research has been conducted in Turkey on the resistance-improving potentials of *B. tabaci* biotypes against cyantraniliprole. In this study, all populations were highly susceptible to cyantraniliprole, and no multiple resistance was observed between cyantraniliprole and other insecticides, confirming the effectiveness of this insecticide. In general, the results presented in this study suggest that cyantraniliprole can be an effective alternative in controlling *B. tabaci* MED and MEAM1 genetic groups. It is recommended that excessive and continuous cyantraniliprole use be avoided to prevent potential resistance development.

#### Data availability

The datasets conducted and analyzed in this study are available from the authors upon reasonable request.

# **Declaration of Interests**

The authors declare no conflicts of interest in conducting and presenting this research.

## REFERENCES

Abbott W.S. (1925): A method of computing the effectiveness of an insecticide. Journal of Economic Entomology, 18: 265–267.

- Barry J.D., Portillo H.E., Annan I.B., Cameron R.A., Clagg D.G., Dietrich R.F., Watson L.J., Leighty R.M., et al. (2015): Movement of cyantraniliprole in plants after foliar applications and its impact on the control of sucking and chewing insects. Pest Management Science, 71: 395–403.
- Basit M., Saeed S., Saleem M.A., Denholm I., Shah M. (2013): Detection of resistance, cross-resistance, and stability of resistance to new chemistry insecticides in *Bemisia tabaci* (Homoptera: Aleyrodidae). Journal of Economic Entomology, 106: 1414–1422.
- Bielza P., Moreno I., Belando A., Grávalos C., Izquierdo J., Nauen R. (2019): Spiromesifen and spirotetramat resistance in field populations of *Bemisia tabaci* Gennadius in Spain. Pest Management Science, 75: 45–49.
- Caballero R.R., Cyman S.S., Schuster D.J., Portillo H.E., Slater R.R. (2013): Baseline susceptibility of *Bemisia tabaci* (Genn.) biotype B in southern Florida to cyantraniliprole. Crop Protection, 44: 104–108.
- Casida J.E. (2015): Golden age of RyR and GABA-R diamide and isoxazoline insecticides: common genesis, serendipity, surprises, selectivity, and safety. Chemical Research in Toxicology, 2: 560–566.
- Cimino A.M., Boyles A.L., Thayer K.A., Perry M.J. (2016): Effects of neonicotinoid pesticide exposure on human health: a systematic review. Environmental Health Perspectives, 125: 155–162.
- Dångelo R.A.C, Michereff-Filho M., Campos M.R., da Silva P.S., Guedes R.N.C. (2018): Insecticide resistance and control failure likelihood of the whitefly *Bemisia tabaci* (MEAM1; B biotype): a Neotropical scenario. Annals of Applied Biology, 172: 88–99.
- De Barro P.J., Liu S.S., Boykin L.M., Dinsdale A.B. (2011): *Bemisia tabaci*: a statement of species status. Annual Review of Entomology, 56: 1–19.
- Elbert A., Nauen R. (2000): Resistance of *Bemisia tabaci* (Homoptera: Aleyrodidae) to insecticides in southern Spain with special reference to neonicotinoids. Pest Manage. Science, 56: 60–64.
- Gerling D. (1990): Whiteflies. Their Bionomics. Pest Status and Management. Intercept Andover, 1: 2.
- Grávalos C., Fernández E., Belando A., Moreno I., Ros C., Bielza P. (2015): Cross-resistance and baseline susceptibility of Mediterranean strains of *Bemisia tabaci* to cyantraniliprole. Pest Management Science, 71: 1030–1036.
- Horowitz A.R., Kontsedalov S., Khasdan V., Ishaaya I. (2005): Biotypes B and Q of *Bemisia tabaci* and their relevance to neonicotinoid and pyriproxyfen resistance. Archives of Insect Biochemistry and Physiology, 58: 216–225.
- Horowitz A.R., Ghanim M., Roditakis E., Nauen R., Ishaaya I. (2020): Insecticide resistance and its management in *Bemisia tabaci* species. Journal of Pest Science, 93: 893–910.

- Jeanguenat A. (2013): The story of a new insecticidal chemistry class: the diamides. Pest Management Science, 69: 7–14.
- Li X.C., Degain B.A., Harpold V.S., Marcon P.G., Nichols R.L., Fournier A.J, Naranjo S.E., Palumbo J.C., et al. (2012): Baseline susceptibilities of B and Q biotype *Bemisia tabaci* to anthranilic diamides in Arizona. Pest Management Science, 68: 83–81.
- Li Y., Dennehy T.J, Li X., Wigert M.E. (2003): Susceptibility of Arizona whiteflies to chloronicotinyl insecticides and IGR's: new developments in the 1999 season. In: Proc from Beltwide Cotton Conferences, January 6–10, 2003, National Cotton Council, Memphis, TN, 2: 1325–1330.
- Liu X., Ning Y., Wang H., Wang K. (2015): Cross-resistance, mode of inher-itance, synergism, and fitness effects of cyantraniliprole resistance in *Plutella xylostella*. Entomologia Experimentalis et Applicata, 157: 271–278.
- Martin J.H., Mound L.A. (2007): An annotated check list of the world's whiteflies (Insecta: Hemiptera: Aleyrodidae). Zootaxa, 1492: 1–81.
- Mound L.A., Halsey S.H. (1978): Whitefly of the World., Wiley, New York.
- Navas-Castillo J., Fiallo-Olive E., Sanchez-Campos, S. (2011): Emerging virus diseases transmitted by whiteflies. Annual Review of Phytopathology, 49: 219–248.
- Qi S., Casida J.E. (2013): Species differences in chlorant-raniliprole and flubendiamide insecticide binding sites in the ryanodine receptor. Pesticide Biochemistry and Physiology, 107: 321–326.
- Qi S., Lümmen P., Nauen R., Casida J.E. (2014): Diamide insecticide target site specificity in the Heliothis and Musca ryanodine receptors relative to toxicity. Journal of Agricultural and Food Chemistry, 62: 4077–4082.
- Perring T.M. (2001): The *Bemisia tabaci* species complex. Crop Protection, 20: 725–737.
- Ribeiro L.M.S., Siqueira H.A.A, Wanderley-Teixeira V., Ferreira H.N., Silva W.M., Silva J.E., Teixeira A.A.C. (2017): Field resistance of Brazilian *Plutella xylostella* to diamides is not metabolism mediated. Crop Protection, 93: 82–88.
- Roy D., Bhattacharjee T., Biswas A., Ghosh A., Sarkar S., Mondal D., Sarkar P.K. (2019): Resistance monitoring for conventional and new chemistry insecticides on *Bemisia tabaci* genetic group Asia-I in major vegetable crops from India. Phytoparasitica, 47: 55–66.
- Sang S., Shu B.S., Yi X., Liu J., Hu M.Y., Zhong G.H. (2016): Cross resistance and baseline susceptibility of *Spodoptera litura* (Fabricius) (Lepidoptera: Noctuidae) to cyantraniliprole in the south of China. Pest Management Science, 72: 922–928.
- Sattelle D.B., Cordova D., Cheek T.R. (2008): Insect ryanodine receptors: molecular targets for novel pest control chemicals. Invertebrate Neuroscience, 8: 107–119.

- Wang R., Wang J., Che W., Luo C. (2018): First report of field resistance to cyantraniliprole, a new anthranilic diamide insecticide, on *Bemisia tabaci* MED in China. Journal of Integrative Agriculture, 17: 158–163.
- Wang R., Wang J., Che W., Sun Y., Li W., Luo C. (2019): Characterization of field-evolved resistance to cyantraniliprole in *Bemisia tabaci* MED from China. Journal of Integrative Agriculture, 18: 2571–2578.
- Wang Z.Y., Yan H.F., Yang Y.H., Wu Y.D. (2010): Biotype and insecticide resistance status of the whitefly *Bemisia tabaci* from China. Pest Management Science, 66: 1360–1366.
- Xie W., Liu Y., Wang S., Wu Q., Pan H., Yang X., Guo L., Zhang Y. (2014): Sensitivity of *Bemisia Tabaci* (Hemiptera: Aleyrodidae) to several new insecticides in China: Effects of insecticide type and whitefly species, strain, and stage. Journal of Insect Science, 14: 1–7.

- Yao F.L, Zheng Y., Huang X.Y., Ding X.L., Zhao J., Desneux N., He Y., Weng Q. (2017): Dynamics of *Bemisia tabaci* biotypes and insecticide resistance in Fujian province in China during 2005–2014. Science Reports, 7: 40803.
- Yükselbaba U. (2015): Determination of the potential of B and Q biotypes of the cotton whitefly *Bemisia tabaci* (Genn.) (Hemiptera: Aleyrodidae) to develop resistance to different insecticides and their enzyme activities. [PhD thesis], Akdeniz university, Antalya.
- Zeng G., Chen M., Zeng Z. (2013): Risks of neonicotinoid pesticides. Science, 340: 1403.
- Zhang R.M., He S.Y., Chen J.H. (2014): Monitoring of *Bactrocera dorsalis* (Diptera: Tephritidae) resistance to cyantraniliprole in the south of China. Journal of Economic Entomology, 107: 1233–1238.

Received: November 7, 20232 Accepted: November 20, 2024 Published online: December 5, 2024