Original scientific paper
Journal of Central European Agriculture, 2024, 25(4), p.1033-1042
DOI: /10.5513/JCEA01/25.4.4355

Assessing the laboratory efficacy of cypermethrin, deltamethrin, and gel baits against the German cockroach (*Blattella germanica* L.)

Osjetljivost smeđeg žohara (*Blattella germanica* L.) na cipermetrin, deltametrin i gel mamce u laboratorijskim uvjetima

Krešimir ŠIMUNAC[1], Luka MUSTAPIĆ[1], Renata BAŽOK[2], Petra MUSTAPIĆ[3], Martina KADOIĆ BALAŠKO[4] (✉)

[1] Croatian agency for agriculture and food, Centre for plant protection, Gorice 68b, 10 000 Zagreb, Croatia

[2] Department of Agricultural Zoology, University of Zagreb Faculty of Agriculture, Svetošimunska cesta 25, 10000 Zagreb, Croatia

[3] ID EKO d.o.o., Porečka 11, 10000 Zagreb, Croatia

[4] Zeleni prsten Public Institution of Zagreb County, 151. samoborske brigade HV 1, 10430 Samobor, Croatia

✉ Corresponding author: martina.kadoic.balasko@zeleni-prsten.hr

Received: April 8, 2024; Accepted: September 3, 2024

ABSTRACT

The German cockroach (*Blattella germanica* L.) is a significant pest in urban areas and is known for its ability to resist insecticides. Today, resistance to insecticides makes the control of agriculturally, economically and medically important insect pests considerably more difficult. This study represents the first investigation on the resistance of *B. germanica* in Croatia. After control failures with pyrethroid insecticides, a population of *B. germanica* was collected from a public place in Zagreb, Croatia. A susceptible population, which was kept under laboratory conditions for over 20 years without contact with insecticides, was obtained from Austria. The experiment was conducted on adults according to the IRAC protocol (No. 037). Active ingredients (AIs) commonly used to control this pest were tested: cypermethrin and deltamethrin as solutions and imidacloprid and chlorfenapyr in the form of gel baits. The results for cypermethrin and deltamethrin showed little to no efficacy in the site population after 72 hours, even at a dose 40 times higher than the dose that achieved full efficacy in the laboratory population. The efficacy of two gel baits with chlorfenapyr and imidacloprid after 72 hours was 40% and 68% higher in the laboratory population than in the site population, respectively. Neither the insecticides nor the gel baits showed sufficient efficacy in the site population of *B. germanica*. Considering these findings and observed resistance, it is essential to implement an anti-resistance strategy including the use of insecticides from various chemical groups, mechanical, biological and biotechnological measures as well as accompanying measures to reduce pest populations.

Keywords: anti-resistance strategies, biocides, *Blatella germanica*, insecticides, IRAC, resistance

SAŽETAK

Smeđi žohar (*Blattella germanica* L.) važan je štetnik u gradskim sredinama i poznat po svojoj otpornosti na insekticide. Otpornost na insekticide značajno otežava suzbijanje štetnika koji su važni u poljoprivredi i javnom zdravstvu. Ovaj rad predstavlja prvo istraživanje o otpornosti smeđeg žohara na insekticide u Hrvatskoj. Nakon neuspješnog suzbijanja pomoću insekticida iz skupine piretroida, populacija žohara prikupljena je na javnoj lokaciji u Zagrebu i donesena u laboratorij na daljnja istraživanja. Osjetljiva populacija koja je više od 20 godina uzgajana u laboratorijskim uvjetima bez doticaja s insekticidima nabavljena je iz Austrije. Laboratorijski pokusi provedeni su na odraslim jedinkama obje populacije prema IRAC protokolu (No. 037). U pokusu su testirane aktivne tvari koje se najčešće koriste za suzbijanje ovog štetnika cipermetrin i deltametrin u obliku sredstva te imidakloprid i klorfenapir u obliku gel mamaca. U istraživanju je dokazana rezistentnost smeđeg žohara na aktivne tvari cipermetrin i deltametrin kod kojih učinkovitost nije postignuta čak ni na 40 puta većoj dozi od doze s kojom je postignuta potpuna učinkovitost na laboratorijskoj populaciji. U slučaju gel mamaca učinkovitost na laboratorijskoj populaciji u odnosu na populaciju s javnog mjesta je 40% veća za aktivnu tvar klorfenapir te 68% veća za imidakloprid. S obzirom na rezultate istraživanja i dokazanu pojavu rezistentnosti kod populacije prikupljene u Zagrebu, potrebno je poduzeti sve mjere koje mogu doprinijeti smanjenju populacije štetnika. To uključuje primjenu sredstava različitog mehanizma djelovanja, mehaničke, biološke i biotehnološke mjere te sve dodatne mjere koje mogu pridonijeti smanjenju populacije štetnika.

Ključne riječi: antirezistentna strategija, biocidi, *Blatella germanica*, insekticidi, IRAC, rezistentnost

INTRODUCTION

German cockroach (*Blattella germanica* L.) is a serious invasive pest in urban areas such as households, apartments, hospitals, schools, food processing facilities and other urban facilities worldwide (Menasria et al., 2014; Nalyanya et al., 2014; Fardisi et al., 2017.). It poses a risk to human health and well-being by scattering fecal matter and carcasses in closed residences (Shahraki et al., 2013). Due to the high fertility rates, it reaches high infestations rapidly, it can be a mechanical vector and reservoir for various pathogenic agents and can cause incidents of asthma by allergens. Among the food-borne pathogens, it can transmit species of *Salmonella*, *Shigella flexneri* (Castellani and Chalmers 1919), *Escherichia coli* (Migula, 1895), *Staphylococcus aureus* (Rosenbach, 1884) and *Bacillus cereus* (Frankland & Frankland 1887) (Tachbele et al., 2006). Species *Blattella germanica* is a highly adaptive species due to its extremely generalist feeding behavior and its ability to withstand nutritional imbalances and abiotic stressors (Raubenheimer and Jones, 2006). Furthermore, *B. germanica* forage randomly since they cannot detect food or water distantly, which forces their colonies to spread out and colonize new areas quickly. Additionally, *B. germanica* populations can persist in severely toxic areas thanks in part to its highly mutagenous genome (Muhka et al., 2011).

In Croatia, *B. germanica* is most often controlled in small to medium food shops, restaurants, fast foods and similar small catering establishments with the usage of gel bait insecticides (Balta et al., 2010). Bait insecticides are highly effective, pest specific, pose little risk for non-target organisms and are practical in insecticide-sensitive and small areas such as restaurant kitchens, food stocks or pantries. Nevertheless, continuous use of gel bait insecticides for the control of *B. germanica* can cause behavioral resistance (i.e. bait aversion) (Wang et al., 2004). Due to the high effectiveness and low toxicity to mammals, the use of pyrethroid insecticides in the control of *B. germanica* has also become frequent. However, repeated use of these insecticides has resulted in the development of resistance, causing low effectiveness of control in some field populations of *B. germanica* (Cochran 1989; Scott et al., 1990; Atkinson et al., 1991; Valles and Yu, 1996; Dong et al., 1998). Resistance of key pests to insecticide AIs is an immense problem in pest control practices worldwide, with serious impacts in agriculture as well as in urban areas. In the United States alone, pesticide resistance accounts for $1.5 billion in economic losses per year (Pimentel, 2005). Insecticide-resistant field populations of *B. germanica* have been reported for decades (Bennett and Spink 1968; Koehler and Patterson 1986; Cochran 1987; Umeda et al. 1988; Cochran 1989; Hemingway et al., 1993a, 1993b; Valles and Yu 1996; Lee at al. 1996; Scharf et al. 1997; Scharf et al. 1998; Wei et al. 2001; Holbrook et al. 2003; Gondhalekar et al. 2011, 2013; Ko et al. 2016; Naqqash et al. 2016). So far, *B. germanica* populations have been proven resistant to 43 AIs in total, including reported cross-resistance (Whalon et al., 2016). Therefore, resistance management has become an integral part of any cockroach-integrated pest management (IPM) protocol.

The work presented here tackled the issue of urban pest management posed by potentially resistant on-site collected cockroach populations in the public area of farmer's markets and small food-related shops in its vicinity in Zagreb, Croatia. The issue reported there was the evident non-effectiveness of control of cockroach populations. Hence, the goals of this study were: i) to test the laboratory and site population of the cockroaches to four insecticides, two liquid formulations (EC and SC) and two gel baits (GD) ii) to determine the dose-mortality data for lethal concentration (LC) and topical lethal dose (LD) estimation of resistance on-site populations collected and iii) based on the results of the laboratory testing, to recommend insecticide AIs for subsequent cockroach management having in mind anti-resistance strategies. The resulting information from the bioassay presented here is of practical importance as it determined insecticides, concentrations and doses for use in future cockroach management. To our knowledge, this is the first bioassay of this kind and the first report of insecticide-resistant *B. germanica* populations in Croatia.

MATERIALS AND METHODS

Insect collection and rearing

This study involved two *B. germanica* populations. The laboratory population, bred in a controlled laboratory environment for over 20 years, was acquired from Kwizda Agro Gmbh (Leobendorf, Austria). The site population was collected from a public place in Zagreb, Croatia, following control failures with cypermethrin and deltamethrin. The samples were collected at a farmers' market using an automatic aspirator. Approximately 50 individuals at various developmental stages were gathered and transported to the laboratory in a plastic container at room temperature. In the laboratory, the insect species was identified using the dichotomous key by Choate et al. (2008). After identification, the necessary number of adult individuals for trials were reared. Both populations were maintained in the Laboratory for Zoology, Centre for Plant Protection. The cockroaches were housed in plastic boxes within a walk-in environment, maintained at 25-27 °C and 60% relative humidity, with a 12:12h light: dark photoperiod. Each population was kept in separate plastic boxes of the same size. To prevent the cockroaches from escaping, the upper interior surface of the plastic boxes was coated with a dilution of baby oil and petroleum jelly. The cockroaches were provided with a continuous supply of dog food, water, and cardboard for harbourage.

Insecticides

Two insecticide preparations, Cymina 10 (cypermethrin; active ingredient – 1%, Colkim Srl, Italy) and Deltasect (deltamethrin; active ingredient – 2.5%, Sharda Europe B.V.B.A., Belgium), were utilized in the experiment. These insecticides were selected due to concerns raised by pest management specialists regarding their efficacy. The insecticides were dissolved in distilled water to create stock solutions. Testing involved four different concentrations, and an untreated control group, in addition, for the site population we added two more concentrations (Table 1). Each concentration was applied in five replicates with five adult cockroaches per replicate.

Two gel baits, Imidasect gel (imidacloprid; active ingredient – 2.15%, Sharda Europe B.V.B.A., Belgium) and Mythic gel (chlorfenapyr; active ingredient – 0.4%, BASF Croatia d.o.o., Croatia), were also tested. Gel baits were examined at a single concentration, resulting in two variants, along with an untreated control group (Table 1). For the gel baits, each variant was set up in five replicates with five adult cockroaches per replicate.

Bioassays

Before setting up the experiment, 80 glass Petri dishes (140 mm) were prepared on the bottom of which a filter paper previously soaked in water was placed. To induce a state of calmness and reduced activity in the cockroaches, plastic boxes containing the insects were subjected to a 5-minute immersion in ice. Once the cockroaches became more manageable, they were carefully transferred to glass petri dishes. For the application of insecticide solutions, a topical approach was employed. Specifically, 1 µl of the insecticide solution was injected into the intersegmental membrane between the second and third abdominal sternites of adult male cockroaches using a precise 10-µl micropipette (Eppendorf-Netheler-Hinz GmbH, Hamburg, Germany), following the guidelines outlined in IRAC Method No. 037 (IRAC, 2023). Control groups of cockroaches were treated with distilled water. To investigate the potential resistance or tolerance of the site population to gel bait insecticides, gel bait feeding bioassays were conducted, adhering to the protocol established by Wang et al. (2004) with some modifications. The assays were designed in a 'no-choice' format, wherein no alternative food sources were provided. Cockroaches were placed in Petri dishes a day before the introduction of the gel bait. Each dish received approximately 0.5 g of gel bait, while control dishes were supplied with dog food. Throughout the experiments, the experimental units were housed in a walk-in environmental chamber set at a constant temperature of 26 °C, relative humidity of 60%, and a photoperiod of 12:12 (L: D) hours. This controlled environment aimed to standardize the conditions and ensure the reliability of the experimental outcomes. Mortality assessments were conducted at 24, 48, and 72 hours after application, whereby cockroaches that could no longer move or were unable to right themselves were considered dead. The efficacy of cypermethrin, deltamethrin, chlorfenapyr and imidacloprid on all variants including control was calculated according to Schneider-Orelli (1947):

$$Efficacy\ (\%) = \frac{mortality\ on\ treatment\ (\%) - mortality\ on\ control\ (\%)}{100 - mortality\ on\ control\ (\%)} x\ 100\%$$

Statistical analysis

Data on efficacy were statistically evaluated by ANOVA and ranked using Duncan's new multiple-range test. The analysis was done by using the statistical software ARM 9® (Gylling Data Management, 2023). The calculation of efficacy was only applied to those variants and dates for which a statistically justified difference was found between untreated control and variants in the trials, i.e. $P<0.05$. In case of an uneven data distribution, data were transformed with log (x + 1) transformation.

Table 1. Insecticidal treatments used against *B. germanica* adults in the laboratory trial (Zagreb, 2023)

Treatment No.	Insecticide Product	Active Ingredient (a.i.)	Insecticide Dose	Dose of a.i. (g)	Dose (% of recommended)
1	Cymina 10	cypermethrin	20 µl/10 ml	0,2	20
2	Cymina 10	cypermethrin	50 µl/10 ml	0,5	50
3	Cymina 10	cypermethrin	100 µl/10 ml	1	100
4	Cymina 10	cypermethrin	200 µl/10 ml	2	200
5*	Cymina 10	cypermethrin	400 µl/10 ml	4	400*
6*	Cymina 10	cypermethrin	800 µl/10 ml	8	800*
7	Deltasect	deltamethrin	20 µl/10 ml	0,0625	20
8	Deltasect	deltamethrin	50 µl/10 ml	0,125	50
9	Deltasect	deltamethrin	100 µl/10 ml	0,25	100
10	Deltasect	deltamethrin	200 µl/10 ml	0,5	200
11*	Deltasect	deltamethrin	400 µl/10 ml	1	400*
12*	Deltasect	deltamethrin	800 µl/10 ml	2	800*
13	Untreated control	N/A	N/A	N/A	N/A
14	Imidasect gel	imidacloprid	0,5 g	0,01	100
15	Mythic gel	chlorfenapyr	0,5 g	0,002	100
16	Untreated control (dog food)	N/A	N/A	N/A	N/A

* a dose used only on-site population

RESULTS AND DISCUSSION

This study is the first research on the efficacy of insecticides against *B. germanica* in Croatia. At the request of a company specializing in public pest control and due to repeated failures in cockroach control, we decided to investigate the effectiveness of the most commonly used insecticides and gel baits for cockroach control in Croatia.

The topical bioassay results revealed a complete absence of efficacy in pyrethroid insecticides administered to the site population of *B. germanica* in this experiment (Table 2). Even at concentrations of 400% and 800% of both insecticides, there was minimal effectiveness observed after 72 hours (Figures 1 and 2).

Table 2. Mean number of dead individuals of the *B. germanica* for site and laboratory population

Population	Active ingredient	Dose of a.i (g)	24 h	48 h	72 h
Site	Cypermethrin	0,2	0[f]	0[d]	0[e]
		0,5	0[f]	0[d]	0[e]
		1,0	0[f]	0[d]	0,16[de]
		2,0	0[f]	0,16[d]	0,16[de]
		4,0	0[f]	0,28[d]	0,59[d]
		8,0	0[f]	0,34[d]	0,54[d]
	Deltamethrin	0,0625	0[f]	0[d]	0[e]
		0,125	0[f]	0[d]	0[e]
		0,25	0[f]	0[d]	0,16[de]
		0,5	0[f]	0[d]	0,16[de]
		1,0	0[f]	0[d]	0,16[de]
		2,0	0[f]	0[d]	0[e]
	Imidacloprid	0,01	0,47[e]	1,18[c]	1,37[c]
	Chlorfenapyr	0,002	0,16[f]	1,44[c]	2,51[b]
Laboratory	Cypermethrin	0,2	5[a]	5[a]	5[a]
		0,5	5[a]	5[a]	5[a]
		1,0	5[a]	5[a]	5[a]
		2,0	5[a]	5[a]	5[a]
	Deltamethrin	0,0625	3,98[bc]	4,79[a]	5[a]
		0,125	4,56[ab]	5[a]	5[a]
		0,25	5[a]	5[a]	5[a]
		0,5	5[a]	5[a]	5[a]
	Imidacloprid	0,01	3,77[c]	4,59[a]	4,79[a]
	Chlorfenapyr	0,002	1,53[d]	2,24[b]	4,59[a]
	Duncan's LSD P = 0.05		0.280 - 0.713	0.395 - 0.963	0.403 - 0.979
	Treatment F		1.847*	2.527*	1.399
	Treatment Prob (F)		0.017*	0.001*	0.123

Prior to analysis, the data were transformed using a log(x + 1) transformation
Means followed by same letter in the same column do not significantly differ
* Adjusted means

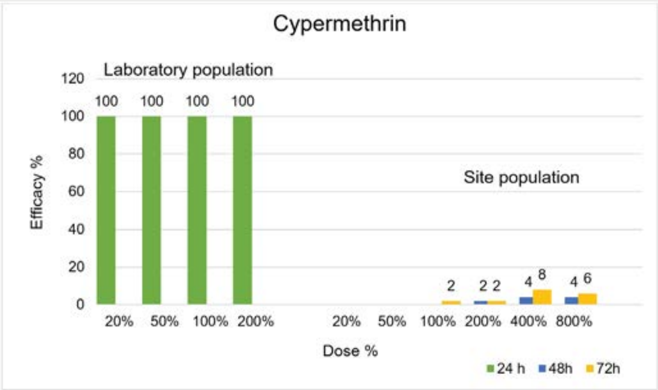

Figure 1. Efficacy of cypermethrin on site and laboratory population of *B. germanica*

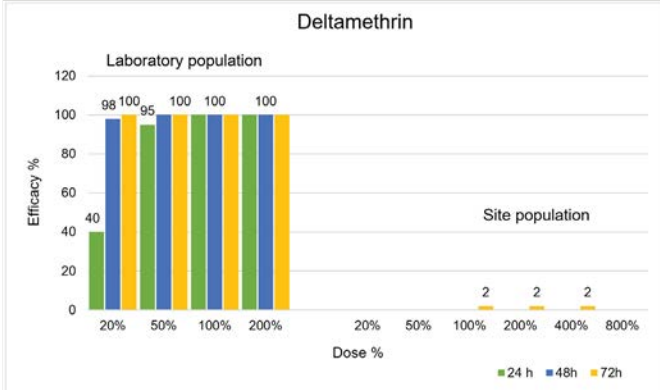

Figure 2. Efficacy of deltamethrin on site and laboratory population of *B. germanica*

In contrast, in the laboratory setting where cypermethrin was applied, across all doses, complete mortality of the cockroach population was achieved within 24 hours (Figure 1). Similarly, deltamethrin exhibited significant lethality, with doses of 50%, 100%, and 200% resulting in complete mortality within 24 hours, and at the 20% dose after 48 hours (Figure 2).

Our study revealed low to no efficacy of pyrethroid insecticides in *B. germanica* site population collected in Zagreb County. These results were not expected especially on 400% and 800% doses. We assumed there would be a lack of efficacy considering several studies have reported that the widespread use of pyrethroid insecticides has led to the emergence of resistance to this class of insecticides in *B. germanica* populations (Anspaugh et al. 1994; Cochran 1999; Valles et al. 2000). However, these results showed that there is a serious problem with insecticide resistance in *B. germanica* site population which requires our attention.

Insecticides have played a crucial role in cockroach management for decades; however, resistance has challenged effective control efforts since the 1950s (Cochran et al., 1952; Bennett et al., 1968; Scharf et al., 1997; Wei et al., 2001; Wang et al., 2004; Gondhalekar et al., 2012; Zhu et al., 2016). Cochran (1995) pointed to the emergence of pyrethroid resistance in cockroach populations, suggesting that these important insecticides may become ineffective in the foreseeable future. In addition, cross-resistance of cockroaches to pyrethroids has been documented. Wei et al. (2001) found that a cockroach strain from Alabama, USA, showed a high level of resistance and cross-resistance to pyrethroid insecticides such as deltamethrin and permethrin after failed control attempts with pyrethroid insecticides. In addition, several mechanisms of insecticide resistance have been documented in *B. germanica*, including enzymatic detoxication, insensitivity to the target site, reduced cuticular penetration, and behavioral avoidance (Anspaugh et al., 1994; Dong and Scott, 1994; Scharf et al., 1997; Scharf et al., 1998; Valles et al., 2000; Wei et al., 2001; Zhu et al., 2016; Fardisi et al., 2017). Although the specific mechanisms of insecticide resistance in the *B. germanica* populations examined in this study have not been defined, the findings presented herein offer preliminary insights for future characterization.

Proposed strategies for managing resistance in *B. germanica* encompass the rotation of various products or the utilization of combination products featuring multiple modes of action, rather than relying solely on single AI products with singular modes of action. Scharf et al (1998) indicate that organophosphates might be most effective when used before pyrethroids in rotational resistance prevention or management programs.

Implementing rotation on a single-generation basis and incorporating a third insecticide or mixture could also be effective strategies to maintain control and prevent the development of significant resistance to any single active ingredient. However, only rotation of various modes of action may not be sufficient to avoid resistance development. In this research, we also tested two gel baits commonly used for suppressing *B. germanica*. Cockroach baits, often used in pest control efforts, are effective in reducing the health risks associated with cockroaches for people and in cutting down on pesticide use in cities (Curl, 2011; Rabito et al., 2017). However, it's important to be aware that cockroaches can quickly become resistant to these bait treatments (Harbison et al., 2003; Wang et al., 2004; Gondhalekar et al., 2013). In this research, chlorfenapyr gel bait showed low efficacy on the site population of *B. germanica* with an efficacy of 52% after 72 hours (Figure 3) and even lower in the case of imidacloprid gel bait with an efficacy of 28% (Figure 4). The laboratory population showed a sensitivity of 92% for chlorfenapyr and 96% for imidacloprid after 72 hours.

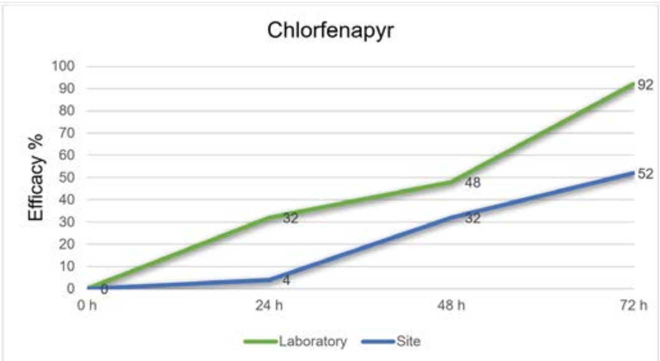

Figure 3. Efficacy of chlorfenapyr gel bait on site and laboratory population of *B. germanica*

Resistance is genetically determined by features that allow insects to withstand insecticide dosages that are lethal to other individuals of the same species (Tabashnik et al., 2014). According to Saipollizan and Ab Majid (2021), this occurs frequently in German cockroach communities and it's not unexpected that certain species show resistance to 8–12 different pesticides.

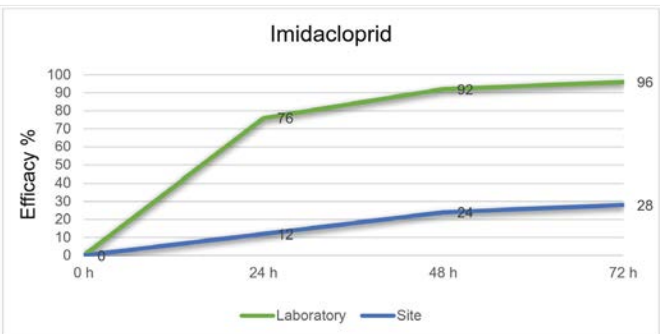

Figure 4. Efficacy of imidacloprid gel bait on site and laboratory population of *B. germanica*

This occurrence may be attributed to the high frequency of application and a wide range of chemical insecticides. This also occurred with *Leptinotarsa decemlineata* Say, which is resistant to 56 different compounds belonging to all major insecticide classes (Kadoić Balaško et al., 2020a), and *Cydia pomonella* L. being resistant to 22 active compounds (Kadoić Balaško et al., 2022). However, the resistance to gel baits appears to be behavioral driven rather than physiological. We observed the lack of feeding of site population on gel baits compared to the control group provided with dog food. This behavioral shift towards avoidance of gel baits is likely a response to the high selection pressure imposed by exposure to these baits. Cases of behavioral resistance on gel baits in *B. germanica* have been reported also by Wang et al. (2004). Similar cases of behavioral resistance have been documented in other insect pests. The western corn rootworm developed resistance to crop rotation by laying its eggs in soybean fields (Levine et al., 2002), and Sarfraz et al. (2005) suggest that the diamondback moth may choose where to lay its eggs to avoid lethal doses of foliar insecticides in the field. Behavioral resistance has also been found in *Aedes aegypti* (Kongmee et al., 2004), the horn fly, *Haematobia irritans* (Lockwood et al., 1985) and house flies, *Musca* spp. (Smith and Yearian, 1964). Behavioral resistance poses new challenges for cockroach control experts. Simply switching between gel baits with different ingredients won't solve the resistance problem. Cockroaches may also avoid other parts of gel baits, not just the AIs if they're repeatedly exposed to them. In addition, adjusting the formulation of gel baits could help make them more effective against *B. germanica* in practice. The ability of cockroaches to develop behavioral resistance repeatedly demonstrates the importance of (IPM) for the long-term and sustainable control of cockroaches.

Noureldin (2016) in his research reports a significant effect of the IPM technique against *B. germanica* with a reduction of 100% after 12 weeks. IPM is an alternative to conventional chemical pest control methods (Olkowski et al., 1991). It uses non-chemical approaches as well as educational measures and utilizes extensive knowledge of pest life cycles and their interactions with the environment to guide pest control. The basic principle of IPM is that pest populations can be controlled by disrupting their basic survival needs, such as air, moisture, food and shelter. This can be accomplished by sealing cracks and crevices to prevent pests from entering buildings and by strategically placing baits and gels that are least toxic to humans and pets.

Maintenance, hygiene, education and training are the main pillars of IPM. In addition, it's becoming increasingly important to include biological and biotechnical measures in pest control. Zhang et al. (2022) discovered that German cockroaches' resistant to beta-cypermethrin were more susceptible to infection by the fungus *Metarhizium anisopliae* (Metschn.). Parasitoid wasps such as *Tetrastichus hagenowii* (Ratzeburg), known as oothecal parasitoid wasps, have shown promise as natural enemies for indoor cockroach control (Pan and Zhang, 2020). El-Kady et al. (2014) found that *Steinernema carpocapsae* (Weiser) and its symbiont *Xenorhabdus bacteria* exhibit strong pathogenicity against adult *B. germanica* under laboratory conditions. A study by Tian (2015) showed the potential of ozone technology for the control of *B. germanica*, which proved to be very promising. Virić Gašparić and Lemić (2020) also see ozone as an alternative method to insecticides against a number of pests, especially storage pests. However, further research especially with *B. germanica* is needed. Moreover, the area-wide mass trapping technique has been shown to be highly effective in reducing insecticide use and establishing IPM plans. In combination with other control measures, this approach also holds significant potential for minimizing codling moth damage levels (Kadoić Balaško et al., 2020b) but it demonstrated good efficacy in the control of the sugar beet weevil also (Drmić et al., 2017). These results show promising avenues for the implementation of iIPM strategies for *B. germanica*. Further research, in particular, laboratory and field trials of the effectiveness of the aforementioned alternative methods on *B. germanica* is needed.

CONCLUSION

Although *B. germanica* is an important pest in households and public spaces, there is a lack of similar research and knowledge about its effective control in Croatia. Our current findings prove practical implications for the development of insecticide-resistant *B. germanica* site populations. Compared to laboratory populations of *B. germanica*, site-collected populations showed high levels of resistance to cypermethrin and deltamethrin insecticides and to gel baits containing chlorfenapyr and imidacloprid. These results indicate the need for a more careful future selection and rotational use of these insecticides in their application for controlling *B. germanica* site populations. In order to find suitable alternatives to the use of insecticides, further research is needed to determine their potential efficacy and resistance mechanisms, which in turn may provide useful information for sustainable and effective resistance management.

ACKNOWLEDGMENTS

The authors thank the student Lovro Brtan for his help in setting up the experiment, Maja Pintar and Mladen Šimala for their help in maintaining the *B. germanica* populations and Jennifer Bernard from Kwizda Agro, Austria for providing the laboratory population of *B. germanica*.

REFERENCES

Anspaugh, D. D., Rose, R. L., Koehler, P. G., Hodgson, E., Roe, R. M. (1994) Multiple mechanisms of pyrethroid resistance in the German cockroach, *Blattella germanica*. Pesticide Biochemistry and Physiology, 50, 138–148.
DOI: https://doi.org/10.1006/pest.1994.1066

Atkinson, T. H., Wadleigh, R. W., Koehler, P. G., Patterson, R. S. (1991) Pyrethroid resistance and synergism in a field strain of the German cockroach (Dictyoptera: Blattelidae). Journal of Economic Entomology, 84, 1247-1250.
DOI: https://doi.org/10.1093/jee/84.4.1247

Balta, S., Salihović, H., Pašić, E., Mraković, E. (2010) Experiences in German cockroaches elimination in health facilities. In: Zbornik radova 22. Znanstveno Stručno Edukativni Seminar DDD i ZUPP 2010: Djelatnost dezinfekcije, dezinsekcije, deratizacije i zaštite uskladištenih, Pula, Croatia, pp. 249-254.

Bennett, G. W., Spink, W. T. (1968) Insecticide resistance of *B. germanica* from various areas of Louisiana. Journal of Economic Entomology, 61, 426–431.

Choate, P., Burns, S., Olsen, L., Richman, D., Perez, O., Patnaude, M., Pluke, R. (2008) A Dichotomous key for the identification of the cockroach fauna (Insecta: Blattaria) of Florida. Florida entomologist, 72 (4), 612-617.

Cochran, D. G. (1999). Cockroaches: their biology, distribution and control. World Health Organization (No. WHO/CDS/CPC/WHOPES/99.3).

Cochran, D. G., Grayson, J. M., Levitan, M. (1952) Chromosomal and cytoplasmic factors in transmission of DDT resistance in the German cockroach. Journal of Economic Entomology, 45, 997. DOI: https://doi.org/10.1093/jee/45.6.997

Cochran, D. G. (1987) Selection for pyrethroid resistance in the German cockroach. Journal of Economic Entomology, 80, 1117–1121. DOI: https://doi.org/10.1093/jee/80.6.1117

Cochran, D. G. (1989) Monitoring for insecticide resistance in field-collected strains of the German cockroach (Dictyoptera: Blattellidae). Journal of Economic Entomology, 82, 336-241. DOI: https://doi.org/10.1093/jee/82.2.336

Cochran, D. G. (1995) Insecticide resistance. Understanding and controlling the German cockroach. Oxford University Press, New York, pp. 171-192.

Curl, G. (2011) Strategic analysis of the U.S. structural pest control industry. Specialty Product Consultants, LLC, Mendham, N. J.

Dong, K., Scott, J. G. (1994) Linkage of kdr-type resistance to the para-homologous sodium channel gene in *B. germanica* (*Blattella germanica*). Insect Biochemistry and Molecular Biology, 24, 647–654. DOI: https://doi.org/10.1016/0965-1748(94)90051-5

Dong, K., Valles, S. M., Scharf, M. E., Zeichner, B., Bennet, G. W. (1998) The knockdown resistance (kdr) mutation in pyrethroid-resistant *B. germanica*. Pesticide Biochemistry and Physiology, 60, 195-204. DOI: https://doi.org/10.1006/pest.1998.2339

Drmić, Z., Tóth, M., Lemić, D., Grubišić, D., Pospišil, M., Bažok, R. (2017) Area-wide mass trapping by pheromone-based attractants for the control of sugar beet weevil (*Bothynoderes punctiventris* Germar, Coleoptera: Curculionidae). Pest management science, 73 (10), 2174-2183. DOI: https://doi.org/10.1002/ps.4601

El-Kady, G. A., El-Bahrawy, A. F., El-Sharabasy, H. M., El-Badry, Y. S., El-Ashry, R. M. A., Mahmoud, M. F. (2014) Pathogenicity and reproduction of the entomopathogenic nematode, *Steinernema carpocapsae* (Wieser) in the German cockroach, *Blattella germanica* L. (Dictyoptera: Blattellidae). Egyptian Journal of Biological Pest Control, 24 (1), 133.

Fardisi, M., Gondhalekar, A. D., Scharf, M. E. (2017) Development of diagnostic insecticide concentrations and assessment of insecticide susceptibility in German Cockroach (Dictyoptera: Blattellidae) field strains collected from public housing. Journal of Economic Entomology, 110 (3), 1210–1217. DOI: https://doi.org/10.1093/jee/tox076

Gondhalekar, A. D., Scharf, M. E. (2012) Fipronil resistance mechanisms in a multi-resistant field strain of the German cockroach. Journal of Medical Entomology, 49, 122–131. DOI: https://doi.org/10.1603/ME11106

Gondhalekar, A. D., Scherer, W., Saran, R. K., Scharf, M. E. (2013) Implementation of an indoxacarb susceptibility monitoring program using field-collected German cockroach isolates from the United States. Journal of Economic Entomology, 106, 945–953. DOI: https://doi.org/10.1603/EC12384

Gondhalekar, A. D., Song, C., Scharf, M. E. (2011) Development of strategies for monitoring indoxacarb and gel bait susceptibility in the German cockroach. Pest Management Science, 67, 262–270. DOI: https://doi.org/10.1002/ps.2057

Gylling Data Management Inc. (2023) (ARM 2023 ® GDM Software, Revision 2023.6, November 29, 2023 (B = 23424); Gylling Data Management Inc.: Brookings, SD, USA, 2023.

Harbison, B., Kramer, R., Dorsch, J. (2003) Stayin' alive. Pest Control Technology, 83, 24–29.

Hemingway, J., Dunbar, S. J., Monro, A. G., Small, G. J. (1993a) Pyrethroid resistance in *B. germanica*: resistance levels and underlying mechanisms. Journal of Economic Entomology, 86, 1931–1938. DOI: https://doi.org/10.1093/jee/86.6.1931

Hemingway, J., Small, G. J., Monro, A. G. (1993b) Possible mechanisms of organophosphorus and carbamate insecticide resistance in *B. germanica* from different geographical areas. Journal of Economic Entomology, 86, 1623–1630. DOI: https://doi.org/10.1093/jee/86.6.1623

Holbrook, G. L., Roebuck, J., Moore, C. B., Waldvogel, M. G., Schal, C. (2003) Origin and extent of resistance to fipronil in the German cockroach, *Blattella germanica* (L.). Journal of Economic Entomology, 96, 1548–1558. DOI: https://doi.org/10.1093/jee/96.5.1548

IRAC Susceptibility Test Methods Series. Insecticide Resistance Action Committee, Test Methods. (2023) Available at: https://irac-online.org/teams/methods/documents [Accessed 20 March 2023].

Kadoić Balaško, M., Mikac, K. M., Bažok, R., Lemic, D. (2020a) Modern techniques in Colorado potato beetle (*Leptinotarsa decemlineata* Say) control and resistance management: history review and future perspectives. Insects, 11 (9), 581. DOI: https://doi.org/10.3390/insects11090581

Kadoić Balaško, M., Bažok, R., Mikac, K. M., Lemic, D., Pajač Živković, I. (2020b) Pest management challenges and control practices in codling moth: A review. Insects, 11 (1), 38. DOI: https://doi.org/10.3390/insects11010038

Kadoić Balaško, M., Bažok, R., Mikac, K. M., Benítez, H. A., Suazo, M. J., Viana, J. P. G., Lemić, D., Živković, I. P. (2022) Population genetic structure and geometric morphology of codling moth populations from different management systems. Agronomy, 12 (6), 1278. DOI: https://doi.org/10.3390/agronomy12061278

Ko, A. E., Bieman, D. N., Schal, C., Silverman, J. (2016) Insecticide resistance and diminished secondary kill performance of bait formulations against *B. germanica*. Pest Management Science, 72, 1778–1784. DOI: https://doi.org/10.1002/ps.4211

Koehler, P. G., Patterson, R. S. (1986) A comparison of insecticide susceptibility in seven nonresistant strains of the German cockroach, *Blattella germanica*. Journal of Medical Entomology, 23, 298–299. DOI: https://doi.org/10.1093/jmedent/23.3.298

Kongmee, M., Prabaripai, A., Akratanakul, P., Bangs, M. J., Chareonviriyaphap, T. (2004) Behavioural responses of *Aedes aegypti* (Diptera: Culicidae) exposed to deltamethrin and possible implications for disease control. Journal of medical entomology, 41 (6), 1055-1063. DOI: https://doi.org/10.1603/0022-2585-41.6.1055

Lee, C. Y., Yap, H., Chong, N. L., Lee, R. S. T. (1996) Insecticide resistance and synergism in field collected *B. germanica* in Peninsular Malaysia. Bulletin of Entomological Research, 86, 675–682. DOI: https://doi.org/10.1017/S0007485300039195

Levine, E., Spencer, J. L., Isard, S. A., Onstad, D. W., Gray, M. E. (2002) Adaptation of the western corn rootworm to crop rotation: Evolution of a new strain in response to a management practice. American Entomologist, 48, 94–107.

Lockwood, J. A., Byford, R. L., Story, R. N., Sparks, T. C., Quisenberry, S. S. (1985) Behavioural resistance to the pyrethroids in the horn fly, *Haematobia irritans* (Diptera: Muscidae). Environmental Entomology, 14 (6), 873-880. DOI: https://doi.org/10.1093/ee/14.6.873

Menasria, T., Moussa, F., El-Hamza, S., Tine, S., Megri, R., Chenchouni, H. (2014) Bacterial load of German cockroach (*Blattella germanica*) found in hospital environment. Pathogens Global Health, 108 (3), 141–147. DOI: https://doi.org/10.1179/2047773214Y.0000000136

Muhka, D. V., Mysina, V., Mavropulo, V., Schal, C. (2011) Structure and molecular evolution of the ribosomal DNA external transcribed spacer in the cockroach genus Blattella. Genome, 54 (3), 222–234. DOI: https://doi.org/10.1139/G10-112

Nalyanya, G., Gore, J. C., Linker, H. M., Schal, C. (2014) German cockroach allergen levels in North Carolina schools: Comparison of integrated pest management and conventional cockroach control. Journal of Medical Entomology, 46 (3), 420–427. DOI: https://doi.org/10.1603/033.046.0302

Naqqash, M. N., Gökçe, A., Bakhsh, A., Salim, M. (2016) Insecticide resistance and its molecular basis in urban insect pests. Parasitology Research, 1–11. DOI: https://doi.org/10.1007/s00436-015-4898-9

Noureldin, E. M. (2016) Integrated pest management (IPM) for the German cockroach (*Blattella germanica* L.) in Jeddah province, Saudi Arabia. Biosciences Biotechnology Research Asia, 7 (2), 657-665.

Olkowski, W., Darr, S., Olkowski, H. (1991) Common-Sense Pest Control. Newtown, CT: Taunton Press.

Pan, X. Y., Zhang, F. (2020) Advances in biological control of the German cockroach, *Blattella germanica* (L.). Biological Control, 142, 104104. DOI: https://doi.org/10.1016/j.biocontrol.2019.104104

Pimentel, D. (2005) Environmental and economic costs of the application of pesticides primarily in the United States. Environment, Development and Sustainability, 7 (2), 229–252. DOI: https://doi.org/10.1007/s10668-005-7314-2

Rabito, F. A., Carlson, J. C., He, H., Werthmann, D., Schal, C. A. (2017) Single intervention for cockroach control reduces cockroach exposure and asthma morbidity in children. Journal of Allergy and Clinical Immunology, 140, 565–570. DOI: https://doi.org/10.1016/j.jaci.2016.10.019

Raubenheimer, D., Jones, S. A. (2006) Nutritional imbalance in an extreme generalist omnivore: tolerance and recovery through complementary food selection. Animal Behaviour, 71 (6), 1253–1262. DOI: https://doi.org/10.1016/j.anbehav.2005.07.024

Saipollizan, Q. A., Ab Majid, A. H. (2021) Laboratory efficacy and toxicology of two commercial insecticides (deltamethrin and fenitrothion) against two German cockroach field strains. Toxicology Reports, 8, 1849-1855. DOI: https://doi.org/10.1016/j.toxrep.2021.11.005

Sarfraz, M., Dosdall, L. M., Keddie, B. A. (2005) Evidence for behavioural resistance by the diamondback moth, *Plutella xylostella* (L.). Journal of applied entomology, 129 (6), 340-341. DOI: https://doi.org/10.1111/j.1439-0418.2005.00969.x

Scharf, M. E., Kaakeh, W., Bennett, G. W. (1997) Changes in an insecticide-resistant field population of German cockroach after exposure to an insecticide mixture. Journal of Economic Entomology, 90, 38–48. DOI: https://doi.org/10.1093/jee/90.1.38

Scharf, M. E., Neal, J. J., Bennett, G. W. (1998) Changes of insecticide resistance levels and detoxication enzymes following insecticide selection in the German cockroach, *Blattella germanica* (L.). Pesticide Biochemistry and Physiology, 59, 67–79. DOI: https://doi.org/10.1006/pest.1997.2311

Schneider-Orelli, O. (1947) Entomologisches Pratikum. HR Sauerlander and Co, Aarau.

Scott, J.G., Cochran, D.G., Siegfried, B.D. (1990) Insecticide toxicity, synergism, and resistance in the German cockroach (Dictyoptera: Blattellidae). Journal of Economic Entomology, 83, 1698-1701. DOI: https://doi.org/10.1093/jee/83.5.1698

Shahraki, G. H., Parhizkar, S., Nejad, A. R. S. M. (2013) Cockroach infestation and factors affecting the estimation of cockroach population in urban communities. International Journal of Zoology, 1-6. DOI: https://doi.org/10.1155/2013/649089

Smith, W. W., Yearian, W. C. (1964) Studies of behaviouristic insecticide resistance in houseflies. Journal of the Kansas Entomological Society, 37 (1), 63-77.

Tabashnik, B. E., Mota-Sanchez, D., Whalon, M. E., Hollingworth, R. M., Carrière, Y. (2014) Defining terms for proactive management of resistance to Bt crops and pesticides. Journal of economic entomology, 107 (2), 496-507. DOI: https://doi.org/10.1603/EC13458

Tachbele, E., Erku, W., Gebre-Michael, T., Ashenafi, M. (2006) Cockroach-associated food-borne bacterial pathogens from some hospitals and restaurants in Addis Ababa, Ethiopia: distribution and antibiograms. Journal of Rural and Tropical Public Health, 5, 34-41.

Tian, Y. (2015). Potential of ozone technology for German cockroach (*Blattella germanica* (L.)) management. Open Access Theses. 621.

Umeda, K., Yano, T., Hirano, M. (1988) Pyrethroid-resistance mechanism in German cockroach, *Blattella germanica* (Orthoptera: Blattellidae). Applied Entomology and. Zoology, 23, 373–380. DOI: https://doi.org/10.1303/aez.23.373

Valles, S. M., Dong, K., Brenner, R. J. (2000) Mechanisms responsible for Cypermethrin resistance in a strain of German cockroach, *Blattella germanica*. Pesticide Biochemistry and Physiology, 66, 195–205. DOI: https://doi.org/10.1006/pest.1999.2462

Valles, S. M., Yu, S. J. (1996) Detection and biochemical characterization of insecticide resistance in the German cockroach (Dictyoptera: Blattellidae). Journal of Economic Entomology, 89, 21-26. DOI: https://doi.org/10.1093/jee/89.1.21

Virić Gašparić, H., Lemić, D. (2020) Budućnost uporabe ozona u poljoprivredi. Glasilo biljne zaštite, 20 (6), 571-583.

Wang, C., Scharf, M. E., Bennett, G. W. (2004) Behavioral and physiological resistance of the German cockroach to gel baits (Blattodea: Blattellidae). Journal of Economic Entomology, 97,2067–2072. DOI: https://doi.org/10.1093/jee/97.6.2067

Wei, Y., Appel, A. G., Moar, W. J., Liu, N. (2001) Pyrethroid resistance and cross-resistance in the German cockroach, *Blattella germanica* (L). Pest Management Science, 57, 1055–1059. DOI: https://doi.org/10.1002/ps.383

Whalon, M. E., Mota-Sanchez, M., Hollingworth, R. M. (2016) Arthropod resistant to pesticides database (ARPD). Available at: https://www.pesticideresistance.org/display.php?page=species&arId=215 [Accessed 08 September 2023].

Zhang, X. C., Jiang, M., Zang, Y. N., Zhao, H. Z., Liu, C. X., Liu, B. R., .Xue, H., Schal, C., Lu, M. X., Zhao, D. Q., Zhang, X. X., Zhang, F. (2022) *Metarhizium anisopliae* is a valuable grist for biocontrol in beta-cypermethrin-resistant *Blattella germanica* (L.). Pest Management Science, 78(4), 1508-1518. DOI: https://doi.org/10.1002/ps.6769

Zhu, F., Lavine, L., O'Neal, S., Lavine, M., Foss, C., Walsh, D. (2016) Insecticide Resistance and Management Strategies in Urban Ecosystems. Insects, 7 (1), 2. DOI: https://doi.org/10.3390/insects7010002