



## Deltamethrin and Imidacloprid Resistance Levels of *Leptinotarsa decemlineata* (Coleoptera: Chrysomelidae) Populations Collected from Afyonkarahisar, Turkey

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### ARTICLE INFO

Received: 04.12.2019

Accepted: 13.06.2020

**Keywords:** *Leptinotarsa decemlineata*, LD<sub>50</sub>, imidacloprid, deltamethrin, insecticideresistance

### ABSTRACT

Colorado potato beetle *Leptinotarsa decemlineata* (Say) (Coleoptera: Chrysomelidae) is an important pest that can develop resistance to insecticides. In this study, imidacloprid and deltamethrin resistance levels were determined in ten Colorado potato beetle (CPB) populations collected from Afyonkarahisar province, which is one of the important potato production areas in Turkey, during the season of 2018. The third instar larvae and topical application method were used in the bioassay experiments. The dead live counts were made after seventy-two hours and LD<sub>50</sub> values of the populations were determined. Resistance ratios of imidacloprid and deltamethrin were calculated by the ratio of LD<sub>50</sub> values of field populations to LD<sub>50</sub> of susceptible population. Resistance rates determined for imidacloprid in Sandıklı-1, Sandıklı-2, Sandıklı-3, Şuhut-1, Şuhut-2, Şuhut-3, Dinar, Central, Bolvadin, Emirdağ populations are 3.96, 7.43, 11.33, 14.33, 27.31, 25.08, 9.12, 2.34, 7.46, 8.72-fold, respectively; and 25.86, 36.50, 35.09, 60.11, 77.17, 72.82, 40.07, 9.41, 45.86, 56.59-fold, respectively for deltamethrin. As a result, high insecticide resistance was determined in CPB populations collected from potato production areas in Afyonkarahisar province. It is thought that insecticide resistance management protocols should be implemented in CPB control in these areas.

## Afyonkarahisar İlinden Toplanılan *Leptinotarsa decemlineata* (Coleoptera: Chrysomelidae) Popülasyonlarında Deltamethrin ve İmidacloprid Direnç Seviyeleri

### MAKALE BİLGİSİ

Alınış tarihi: 04.12.2019

Kabul tarihi: 13.06.2020

**Anahtar Kelimeler:** *Leptinotarsa decemlineata*, LD<sub>50</sub>, imidacloprid, deltamethrin, insektisit direnci

### ÖZET

Patates böceği *Leptinotarsa decemlineata* (Say) (Coleoptera: Chrysomelidae) pestisitlere karşı direnç geliştirebilen önemli bir tarımsal zarardır. Bu çalışmada ülkemizin önemli patates üretim merkezlerinden biri olan Afyonkarahisar ilinden 2018 yılı üretim sezonunda toplanan 10 adet patates böceği popülasyonunda imidacloprid ve deltamethrin direnç düzeyleri belirlenmiştir. Bioassay deneylerinde 3. dönem larvalar üzerinde topical aplikasyon uygulaması yapılmıştır. Ölü canlı sayımları uygulamadan 72 saat sonra yapılmış ve LD<sub>50</sub> değerleri belirlenmiştir. Popülasyonların direnç oranları arazi popülasyonlarının LD<sub>50</sub> değerlerinin, hassas popülasyonun LD<sub>50</sub> değerine oranlanmasıyla bulunmuştur. Sandıklı-1, Sandıklı-2, Sandıklı-3, Şuhut-1, Şuhut-2, Şuhut-3, Dinar, Merkez, Bolvadin, Emirdağ popülasyonlarının imidacloprid için belirlenen direnç oranları sırasıyla 3.96, 7.43, 11.33, 14.33, 27.31, 25.08, 9.12, 2.34, 7.46, 8.72-kat; deltamethrin için belirlenen direnç oranları ise sırasıyla 25.86, 36.50, 35.09, 60.11, 77.17, 72.82, 40.07, 9.41, 45.86, 56.59-kat olarak tespit edilmiştir. Çalışma sonunda Afyonkarahisar ilinde patates böceği popülasyonlarına göre farklı oranlarda insektisit direnci geliştiği belirlenmiştir. Sonuç olarak bölgede insektisit direnç yönetim programlarına uygun savaşım yöntemleri uygulanmalıdır.

### 1. Introduction

Potato (*Solanum tuberosum* L.), which belongs to Solanaceae family, is an industrial plant that is produced in almost every part of the world and has an important place in human and animal nutrition. *Leptinotarsa decemlineata* is the major pest in potato production areas. *L. decemlineata*, which is a polyphagous pest that damages every period of the potato plant, damages the plant by eating the leaf epidermis during the first and second instar larvae and by eating the leaves completely in the third and fourth instar larvae periods and even via opening galleries

in the tubers (Radcliffe, 1982). Neonicotinoid and pyrethroid group insecticides are used extensively in the control of *L. decemlineata* in Turkey. However, long-term use of broad-spectrum synthetic insecticides is known to have negative effects on the environment, human health and beneficial organisms. It is known that the CPB can develop resistance easily to insecticides (Pimentel et al., 1992; Cox et al., 1995; Mansour et al., 2004; Stankovic et al., 2004; Nauen and Denholm, 2005; Alyokhin et al., 2007; Kumargal et al., 2012; Bozdoğan and Bahadıroğlu, 2014).

Imidacloprid active ingredient was introduced to the market in Turkey in 1991 and it is placed at 4a group in the IRAC mode of action list (IRAC, 2019). The neonicotinoid insecticides, including imidacloprid, act as nicotinic acetylcholine receptors (nAChRs) in the central nervous system in pests (Bai et al., 1991).

The active ingredient deltamethrin is placed at 3A group containing pyrethroids /pyrethrins in IRAC grouping. In this group the active ingredients act on the nervous and muscular system of insects and are known as sodium channel modulators (IRAC, 2019). The active substance deltamethrin is the first synthesized pyrethroid active ingredient and has been used for a long time in order to control of agricultural pests (Zamojska et al., 2011). Pyrethroid insecticides are among the most common insecticide groups used in chemical control against agricultural pests in the world (Khambay and Jewess, 2004).

## 2. Material and Method

### 2.1. *Leptinotarsa decemlineata* populations

Colorado potato beetle populations were collected from potato production areas in Afyonkarahisar province during potato production season in 2018. 3, 3, 1, 1, 1, 1 populations were collected from Sandikli, Suhut, Dinar, Central, Bolvadin, Emirdag districts, respectively. In the field, CPB adults were collected and put into air-permeable boxes and the date and locations of the populations were written on the boxes. A susceptible CPB population obtained from the London Research and Development Center (Canada) used as a reference population. Potato plants were grown in pots in a climate room to feed the populations. Adult individuals collected from the field were taken to cages with potato plants and waited for eggs to lay. The climate room conditions were to  $25 \pm 1$  °C, 16 : 8 (L / D) hour photoperiod and  $60 \pm 10\%$  humidity.

### 2.2. Insecticides

Commercial insecticides with imidacloprid active ingredient (Confidor SC 350; Bayer CropScience) and deltamethrin active ingredient (Decis EC; Bayer CropScience) were used for resistance tests.

### 2.3. Topical bioassays

In bioassay tests, Zhao et al. (2000) was used by adapting the method. In all bioassay tests, LD50 values were determined via using the topical application method to the 3rd instar larvae. Insecticide applications were carried out with 6 doses + 1 control and 3 replications for each dose. Each replication consists of a total of five 3rd instar larvae. The doses were prepared by serial dilutions of 50% each time and only distilled water was used in the control. The applications were made on the top of the abdomen as 1 µl by micropipette. After the application, the individuals were placed to the Petri dishes which have air-permeable covers and moved to a climate room. Dead live counts were made after seventy-two hours. In this period fresh potato leaflets were provided for nutrition.

## 2.4. Statistical analysis

The data obtained from the result of the dead live counts was analyzed by using POLO computer package program (LeOra software, 1994). The resistance ratio was calculated with comparing the LD<sub>50</sub> values of the resistant populations to the LD<sub>50</sub> value of the susceptible population.

## 3. Results

Imidacloprid resistance ratios of the populations are given in Table 1. The resistance ratios determined for imidacloprid in Sandikli 1, Sandikli 2, Sandikli 3, Suhut 1, Suhut 2, Suhut 3, Dinar, Central, Bolvadin, Emirdag populations were 3.96, 7.43, 11.33, 14.33, 27.31, 25.08, 9.12, 2.34, 7.46, 8.72-fold, respectively. It is showed that populations with slope > 2 are homogeneous and populations with < 2 are heterogeneous (Yu, 2008). Accordingly, it is seen that the Sandikli 3, Suhut 1, Suhut 2, Suhut 3, and Bolvadin populations are more homogeneous. On the other hand, Sandikli 1, Sandikli 2, Dinar, Central, and Emirdag populations were determined to be more heterogeneous. As long as insecticide selection pressure take in heterogeneous pest populations, resistance development may increase more.

LD50 values and resistance ratios determined against deltamethrin in the populations are given in Table 2. Resistance ratios determined for deltamethrin in Sandikli 1, Sandikli 2, Sandikli 3, Suhut 1, Suhut 2, Suhut 3, Dinar, Central, Bolvadin, Emirdag populations were 25.86, 36.50, 35.09, 60.11, 77.17, 72.82, 40.07, 9.41, 45.86, 56.59-fold, respectively. According to the slope values, the populations Suhut 1, Suhut 2, Suhut 3 and Bolvadin are homogeneous; and other populations are heterogeneous. As long as deltamethrin application continue take in populations collected from Sandikli, Dinar, Central, and Emirdag districts, resistance to deltamethrin in CPB populations may reach to higher levels of resistance.

## 4. Discussion

Colorado potato beetle is a major pest in potato production fields and is one of the most important factors limiting potato production (Bolter and Jongsma, 1995; Gruden et al., 1998). One of the factors, which makes it difficult to control of the CPB, is the insecticide resistance because it increases the production cost. Insecticide resistance is an important factor that limits the effectiveness of chemical control.

In this study, the resistance levels of deltamethrin were determined between 9.41-77.17-fold and imidacloprid resistance levels were 3.96-27.31-fold in CPB populations collected from Afyonkarahisar province by topical application. Also, this is the first study in terms of insecticide resistance of *Leptinotarsa decemlineata* in Afyonkarahisar province which is one of the important potato production areas of Turkey.

Daniella et al. (2017) classified insecticide resistance into three categories: low resistance ( $R < 5$ ), medium resistance ( $5 \leq R \leq 10$ ) and high resistance ( $R > 10$ ). According to the scale, it is seen that high levels of deltamethrin resistance

in pest populations. The number of studies of insecticide resistance in CPB development in Turkey is very few, and the literature is out of date. Atak and Atak (1977), investigated the susceptibility of wintered and late summer

adults to the active ingredient azinphosmethyl, trichorphon, and propoxur; and they stated that the difference between 1974 and 1975 and it is not important.

Table 1. LD<sub>50</sub> values and resistance ratios of imidacloprid in *Leptinotarsa decemlineata*

Population	n*	Slope±SE	LD <sub>50</sub> (mg a.i l <sup>-1</sup> ) (95% CL)	R**
Sandikli 1	90	1.620 ± 0.389	21.17 15.21-30.14	3.96
Sandikli 2	90	1.820 ± 0.418	39.66 28.11-49.12	7.43
Sandikli 3	105	4.446 ± 1.005	60.49 50.45-72.15	11.33
Suhut 1	105	9.407 ± 2.922	76.51 58.78-89.12	14.33
Suhut 2	105	7.903 ± 2.288	145.81 124.75-160.11	27.31
Suhut 3	105	8.413 ± 2.288	133.93 111.98-152.56	25.08
Dinar	90	1.471 ± 0.399	51.89 39.99-60.46	9.12
Central	105	1.705 ± 0.359	12.5 7.45-15.65	2.34
Bolvadin	110	3.240 ± 0.819	39.86 25.14-48.23	7.46
Emirdag	90	1.610 ± 0.406	46.59 34.56-55.56	8.72
Susceptiblepopulation	105	1.489 ± 0.299	5.34 3.15-6.89	-

\*: Total number of individuals

\*\* : Resistanceratio

Table 2. LD<sub>50</sub> values and resistance ratios for deltamethrin of *Leptinotarsa decemlineata* populations.

Population	n*	Slope±SE	LD <sub>50</sub> (mg a.i l <sup>-1</sup> ) (95% CL)	R**
Sandikli 1	105	2.820±0.452	55.36 35.45-70.21	25.86
Sandikli 2	90	2.120±0.514	78.12 52.10-85.21	36.50
Sandikli 3	105	2.166±1.112	75.10 45.48-95.78	35.09
Suhut 1	105	7.103±2.881	128.65 95.14-158.78	60.11
Suhut 2	105	5.702±2.144	165.15 101.12-198.17	77.17
Suhut 3	90	6.741±2.122	155.85 98.21-185.85	72.82
Dinar	105	1.890±0.225	85.75 62.14-100.12	40.07
Central	105	1.912±0.233	20.15 10.98-28.12	9.41
Bolvadin	90	3.225±0.615	98.15 75.12-115.45	45.86
Emirdag	90	1.952±0.365	121.12 98.78-141.14	56.59
Susceptiblepopulation	105	1.489±0.299	2.14 1.12-4.45	-

\*: Total number of individuals

\*\* : Resistanceratio

In other study, Erdogan and Gurkan (1997), were investigated to azinphos-methyl and chlorpyrifos-ethyl

(organic phosphorus), cypermethrin and deltamethrin (pyrethroids), endosulfan (chlorinated hydrocarbons), and

carbofuran (carbamates) resistances in *Leptinotarsa decemlineata* populations collected from the provinces Bolu, Nevşehir, and Tekirdağ. The highest resistance level in this study was found to be 225.92, 58.83, 90.42 in deltamethrin for Bolu, Nevşehir, and Tekirdağ, respectively.

In the world, current studies are possible on insecticide resistance in CPB. This data can be useful for insecticide resistance management (IRM) strategies. Baker et al. (2007), between the years, 1999 - 2005 found imidacloprid resistance in CPB populations ranging between 6.8 - 22.9-fold. Zamosjka et al. (2011) determined deltamethrin resistance between 1.18 and 4.89-fold in *L. decemlineata* populations collected from Poland. Sladan et al. (2012) detected 1.0 - 645.3-fold bensultap, 1.0-60.0-fold cypermethrin, and 1.0 - 82.9-fold imidacloprid resistance in CPB populations in Serbia. Huseth and Groves (2013) determined imidacloprid resistance in CPB populations ranging from 0.3 - 79.02-fold. Baker et al. (2014) found imidacloprid resistance ranging from 7.6 - 71.0-fold in CPB populations in the USA. Clements et al. (2016) reported that the CPB populations had imidacloprid resistance between 1.84 and 27.03-fold. Crossley et al. (2018) determined the LD50 values of CPB populations collected from the USA. In our study, imidacloprid and deltamethrin resistance were determined in CPB populations similar to world literature.

Despite the widespread and effective use of imidacloprid until 2018 production season, the active ingredient was banned in open areas due to the damage to bees in Turkey since 24 December 2018 (Anonymous, 2019). Because the active ingredient was not banned during the production season, it is considered that it does not affect the validity of the study. At least in this study, imidacloprid resistance was generally determined in potato production areas of Afyonkarahisar province. If the imidacloprid wasn't banned, Adequate use of the correct insecticides is important for reducing production costs and environmental benefits. In addition, it is considered that the insecticide resistance studies should be extended in the reduction of pesticide costs which are a heavy burden on the national economy; the use of insecticides that are not yet lost susceptibility to pests and the rotation of insecticides which have different active ingredients will provide economic benefit. In this respect, it is thought that the reduction to use of deltamethrin, which was found to resistant in CPB populations, and the rotation with other active substance insecticides, will both provide financial contributions to producers and reduce negative effects on human and environmental health.

## 5. Acknowledgements

We would like to thank Dr. Ian Scott and Sophie Krolikowski from Agriculture and Agri-Food Canada for provide susceptible *Leptinotarsa decemlineata* population and Suleyman Demirel University, Scientific Research Projects Coordination Unit for financing this project with FYL-2018-6054.

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