Original article (Orijinal araştırma)

Determination of insecticide residues in "Bayramiç Beyazı" nectarines and their risk analysis for consumers[1]

"Bayramiç Beyazı" nektarinlerde insektisit kalıntılarının belirlenmesi ve tüketiciler için risk analizi

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Abstract

In this study, insecticide residues on "Bayramiç Beyazı" nectarines were investigated with the use of QuEChERS method and LC-MS/MS analysis. Analytical method was verified through SANTE 11312/2021 Guidelines. The limit of quantification were below the MRLs for 12 insecticides. Method recovery was identified as 89.6%. Such a value was within the SANTE recovery (60-140%) limits. Nectarine samples were collected from Çanakkale open markets between 15 June-30 September, 2022 and analyzed at ÇOMÜ Agriculture Faculty-Pesticide Laboratory (Çanakkale-Türkiye). Abamectin, acetamiprid, deltamethrin, etoxazole, novaluron, pyriproxyfen, spirodiclofen, tetramethrin and thiacloprid residue levels were below the MRLs. On the other hand, dimethoate, imidacloprid and omethoate residues exceeded their MRLs only in one sample each. The maximum residues of acetamiprid, deltamethrin, etoxazole and novaluron were about 1/2, 1/5, 1/10, and 1/70 of the MRLs in one sample, respectively. Risk assessments revealed that exposure levels for adults were low (hazard quotient, HQ ≤ 1), with the exception of omehoate residues. Omethoate posed a chronic risk to human health through consumption of nectarines. For the remaining 11 insecticides, there was no risk for human health. However, the highest acute HQ were found for dimethoate even though its HQ was less than or equal to 1 The use of dimethoate is in the process of being banned in Türkiye, while omethoate (metabolite of dimethoate) is already banned. Presence of omethoate residue may be due to the degradation product of dimethoate.

Keywords: Acute and chronic risk assessment, Bayramiç Beyazı, insecticide residues, nectarin, QuEChERS

Öz

Bu çalışmada QuEChERS yöntemi ile "Bayramiç Beyazı" nektarinlerde insektisit kalıntıları araştırılmıştır. Analiz metodu SANTE 11312/2021'e göre doğrulanmıştır. 12 adet insektisitin LOQ limiti, MRL değerlerinin altında bulunmuştur. Metodun geri kazanımı %89.6 olmuştur. Bu rakamlar SANTE geri alımları (%60-140) ile uyumludur. Nektarin örnekleri 15 Haziran-30 Eylül 2022 arasında Çanakkale pazarından toplanmış ve ÇOMÜ Ziraat Fakültesi-Pestisit Laboratuvarı(Çanakkale-Türkiye)'nda analizleri yapılmıştır. Abamectin, acetamiprid, deltamethrin, etoxazole, novaluron, pyriproxyfen, spirodiclofen, tetramethrin ve thiacloprid kalıntı seviyeleri MRL değerlerinin altındadır. Öte yandan, sadece birer örnekte dimethoate, imidacloprid ve omethoate kalıntıları MRL değerlerini aşmıştır. Acetamiprid, deltamethrin, etoxazole ve novaluronun kalıntıları, birer örnekte MRL'lerin sırasıyla 1/2, 1/5, 1/10 ve 1/70'i olarak bulunmuştur. Risk değerlendirmeleri, yetişkinler için, omehoate hariç, maruz kalma düzeylerinin düşük olduğunu ortaya çıkarmıştır (tehlike katsayısı, HQ ≤ 1). Nektarin tüketiminde insan sağlığı açısından omethoate riskli bulunmuştur. Geri kalan 11 insektisit için insan sağlığı açısından herhangi bir risk bulunmamıştır. Bununla birlikte, HQ≤1 olmasına rağmen en yüksek akut HQ dimethoate için bulunmuştur. Zaten Türkiye'de dimethoate yasaklanma sürecindedir, dimethoate'ın metaboliti olan omethoate kullanımı ise yasaklanmıştır, Omethoate kalıntısı bulunması dimethoate'ın parçalanma ürünü olmasından kaynaklanabilir.

Anahtar sözcükler: Akut ve kronik risk değerlendirmesi, Bayramiç Beyazı, insektisit kalıntıları, nektarin, QuEChERS

Introduction

The "Bayramiç Beyazı" nectarine has been grown as an endemic species for years in Bayramiç District (located in Kazdağları region) of Çanakkale province. With a unique color, taste, smell, aroma and long shelf life, it differs from the other nectarine varieties. The geographical registration approval of the "Bayramiç Beyazı" nectarine has been published in the Official Journal of the European Union (EU) on April 16, 2021. It is the 1st product of Çanakkale with an EU-geographical indication. This fruit is produced in an area of 5500 da with 250 thousand trees. An average of 13 to 15 kt of nectarine is produced annually. In recent years, its cultivation has reached an important level because it can be marketed at high prices in big cities such as İstanbul and İzmir (Anonymous, 2022). Pests including *Grapholita molesta* (Busck, 1916) (Lepidoptera: Torticidaae), *Anarsia lineatella* (Zeller, 1839) (Lepidoptera: Gelechiidae)*, Myzus persicae* (Sulzer, 1776) (Hemiptera: Aphidiidae), *Sphaerolecanium prunastri* (Boyer de Fonscolombe, 1834) (Hemiptera: Coccidae*), Nilotaspis halli* (Green, 1923) (Homoptera: Diaspididae) are the most common harmful insects in nectarine cultivation. While farmers want to protect nectarines against these harmful pests, they prefer chemical control as it gives fast and effective results. So acetamiprid, cyantraniliprole, emamectin benzoate, malathion pyriproxyfen and spinosad pesticides are used against these pests (BKÜ, 2023a). However, ignorant spraying is very harmful to the environment and human health (Ambrus et al., 2023). It is possible to receive alerts about these pesticides from the Rapid Alert System for Food and Feed portal (RASFF, 2023).

The QuEChERS (Quick-Easy-Cheap-Efficient-Rugged-Safe) method, developed by Anastassiades et al. (2003), is generally used for pesticide residue analyses of vegetable and fruits (Lehotay et al., 2005; AOAC, 2007; Polat & Tiryaki, 2019; Polat, 2021; Çatak & Tiryaki, 2020; Balkan & Yılmaz, 2022a). However, a further verification of the method should be conducted if it is to be used by local laboratories (Omeroglu et al., 2012).

Dülger & Tiryaki (2021) used QuEChERS method to investigate boscalid, chlorpyrifos and tebuconazole residues on nectarine and peach samples collected from Çanakkale markets. The overall recovery was determined as 113.5% with a relative standard deviation (RSD) of 17.3% for peach samples and 113.61% with an RSD of 11.4% for nectarine samples. These values were a fit for the recovery (60-140%) and repeatability (RSD ≤ 20%) limits of SANTE. Researchers reported that residue levels did not exceed maximum residue limits (MRLs) in any samples. Chronic exposure levels were low and pesticide residues did not pose any health risks.

Camara et al. (2020) investigated pesticide residues in various fruit juices and assessed dietary risk exposure. Long term chronic risk assessment indicated that potential consumer risk for the imidacloprid pesticide was practically negligible for human health, with the risk Quotients (RQ) of 0.044 and 0.000 for unprocessed peach and peach juice, respectively.

Chatzicharisis et al. (2012) investigated insecticide residue levels on peach and nectarine samples. Decreasing residue levels were seen over time following application. Bupirimate, chlorpyrifos, fenoxycarb, iprodione and pirimicarb residue levels on peach samples were lower than relevant EU-MRLs while chlorothalonil residues were below the quantification limit (LOQ).

Pesticide residues were investigated in peach and nectarine samples imported into the United Arab Emirates. Residues above MRL were found in half of the analyzed samples. The pesticides reported were dimethoate and omethoate from Organophosphorus Class (Osaili et al., 2022). In another study conducted in China, 18 pesticide residues were found in peaches, and acetamprid residues exceeding the EU-MRL were found in 2 samples. Imidacloprid, pyriproxyfen and spirodiclofen residues were less than EU-MRL. Acute and chronic dietary exposure assessment indicated that potential dietary risk induced by the pesticides was not significant for Chinese consumers (Zhang et al., 2021).

Galietta et al. (2011) conducted a study in Uruguay about dissipation curves of pesticide in peach samples. Recovery rates of azoxystrobin, acetamiprid and thiacloprid were observed as 95.3, 98.6 and 80.6%, respectively. Dissipation curves revealed that the time required for insecticide residues to go below MRL was 10-12 days for thiacloprid and 25 days for acetamiprid. Kaya & Tuna (2019) reported that chlorantraniliprole, deltamethrin, phosmet and spirodiclofen residue levels in peach samples were less than the corresponding MRL. However, Ersoy et al. (2011) reported that chlorpyrifos residue levels on peach samples were greater than the MRL. Choi et al. (2011) conducted a study on chlorfluazuron residues in peach samples, which indicated that residues were lower than the MRL in all samples.

To control the safe and efficient use of pesticides, their residues should regularly be monitored in food and environmental samples. The samples should be taken randomly and dietary risk assessment should be performed (Ambrus et al., 2023). On the other hand, improper use of insecticides may result in serious risks on human health. Extended periods of exposure to insecticides may cause cancers, headaches, nausea and endocrine disorders (Yousefi et al., 2022). Therefore, dietary risk assessment of insecticides has recently gained a great attention (Gebara et al., 2011; Marete et al., 2020; Chen et al., 2021). For dietary risk assessments, both acute and chronic risks to the consumer health are evaluated. Dietary risk assessments are performed based on daily food consumption and detected pesticide residue data on foodstuffs. For short-term acute diatery risk assessments, acute reference dose (ARfD, mg/kg bw/day) values are used. Then, estimated short-term intake (ESTI, mg/ kg bw/day) and acute hazard quotient (HQ) are calculated. For long-term chronic dietary risk assessments, acceptable daily intake (ADI, mg/kg bw/day) values are used. Then, estimated daily intake (EDI, mg/ kg bw/day) and chronic HQ values are calculated. HQ values of >1 indicate a potential risk for human health (EFSA, 2007; Balkan & Yılmaz, 2022b).

In this study, insecticide residues on/in "Bayramiç Beyazı" nectarines sampled from Çanakkale open markets were investigated with the QuEChERS method. The method was verified through SANTE (Directorate-General for Health and Food Safety) Guidelines. Total 377 pesticides were analysed in the LC-MS/MS system (located in Çanakkale Food Control Directorate) and insecticides residues above LOQ, were evaluated. Consumer acute and chronic risk assessment for insecticides were also performed.

Materials and Methods

Chemicals and Reagents and Insecticide solutions

Standards for insecticides were supplied from Dr. Ehrenstorfer GmbH (Wesel, Germany) and Chem Service (West Chester, PA, USA). QuEChERS extraction [6 g anhydrous magnesium sulfa ($MgSO_4$); 1.5 g anhydrous sodium acetate (NaOAC)] and clean-up kits [1.2 g $MgSO_4$, 400 mg primary and secondary amines (PSA, 40 μm particle size) and 400 mg C_{18}] were used. The other solvents and reagents including acetonitrile (MeCN) and acetic acid (HAc) were at analytical grade. Stock solution of insecticides (400 μg/mL) were used to prepare working solutions (1.0 μg/mL) through series of dilutions. Calibration (matrix match standards) was performed on blank nectarines. Calibration solutions of matrix-matched (MC) were prepared with MeCN (1-1000 pg/μL) (Poole, 2007). Spiking solutions corresponding to 1 x LOQ and 10 x LOQ were prepared. For MC and quantifications, representative apple matrix was used (CAC, 2003; SANTE, 2021).

Instruments

An LC-MS device was used for chromatographic analyses (Waters I Class Plus UPLC + Xevo TQ-S micro MS Detector; ESI + mode). The device is connected with Acquity UPLC BEH C_{18} column (1.7 mm, 100 x 2.1 mm). Flow rate, injection volume and total run time were 0.35 mL/min, 1 μL and 15 minutes, respectively. A gradient program including 10 mM ammonium acetate ($NH_4CH_3CO_2$) in methanol (B) and 10 mM $NH_4CH_3CO_2$ in water of pH 5 (A) was used. Insecticide retention times (tR), precursor ion and fragment ions are given in Table 1.

The other materials used in the present study included precise balance (± 0.0001 g) (Shimadzu ATX224), centrifuge (Hettich EBA 280, 4500 rpm), vortex (VELP scientifica), centrifuge tubes, Agilent GC vials (1.5 mL), blender and N_2 stream.

Verification of QuEChERS-AOAC Official Method 2007.01

Method verification was performed in accordance with verification parameters of SANTE, such as linearity, recovery, precision and LOQ parameters (SANTE, 2021). Blank nectarine samples of 1 kg were homogenized with a blender. For recovery tests, 15 g blank nectarine samples were spiked with 100 µL of insecticide spike solutions (in MeCN) corresponding 1 x LOQ and 10 x LOQ level of insecticides. Tests were conducted in five replicates (five replicate analytical portions). Resultant mixtures were vortexed for 30 seconds and left standing for 15 minutes for interaction of insecticides with the sample. Figure 1 presents the further analytical steps taken in analyses. MC calibration curve was used to quantify insecticides. The rates of recovery were calculated as the ratio of measured concentration to spiked concentration. Recovery and precision of the method were assessed based on SANTE European Guidelines (SANTE, 2021). Linearity of the method was checked for the range 1-1000 pg/µL.
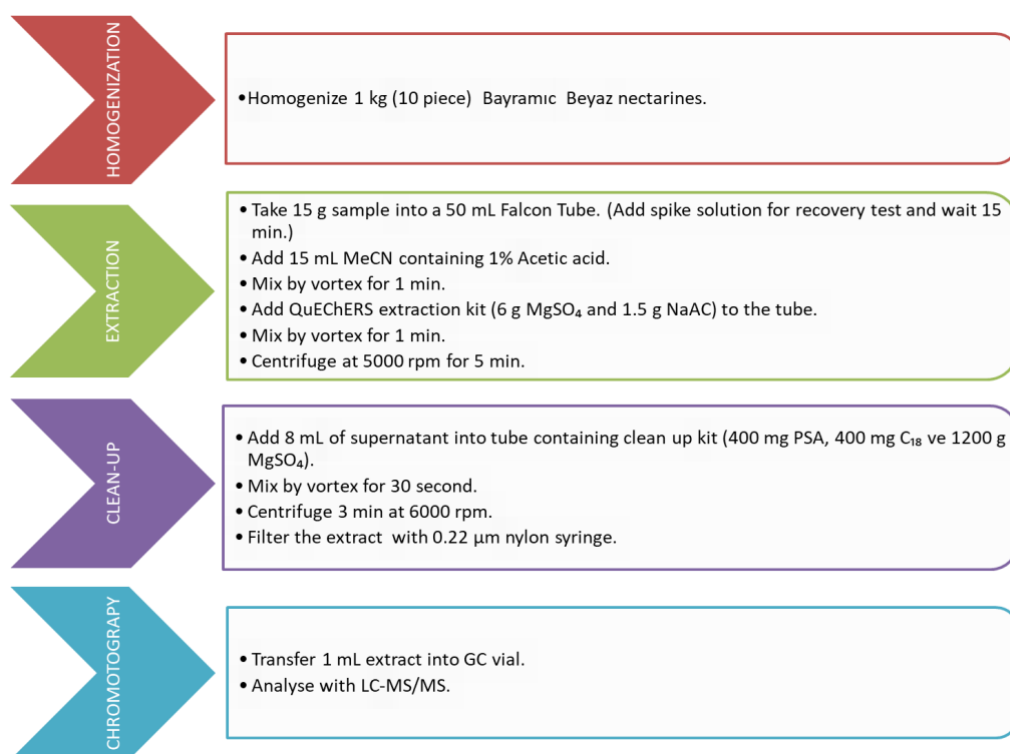
HOMOGENIZATION
- Homogenize 1 kg (10 piece) Bayramıc Beyaz nectarines.

EXTRACTION
- Take 15 g sample into a 50 mL Falcon Tube. (Add spike solution for recovery test and wait 15 min.)
- Add 15 mL MeCN containing 1% Acetic acid.
- Mix by vortex for 1 min.
- Add QuEChERS extraction kit (6 g $MgSO_4$ and 1.5 g NaAC) to the tube.
- Mix by vortex for 1 min.
- Centrifuge at 5000 rpm for 5 min.

CLEAN-UP
- Add 8 mL of supernatant into tube containing clean up kit (400 mg PSA, 400 mg C_{18} ve 1200 g $MgSO_4$).
- Mix by vortex for 30 second.
- Centrifuge 3 min at 6000 rpm.
- Filter the extract with 0.22 µm nylon syringe.

CHROMOTOGRAPY
- Transfer 1 mL extract into GC vial.
- Analyse with LC-MS/MS.

Figure 1. Analytical ssteps QuEChERS-AOAC Official Method 2007.01.

Collecting samples and analyses

Nectarines were taken from different stands in the open markets of Çanakkale province for 14 weeks between 15 June-30 September, then the analyses were performed. The nectarine samples of about 1 kg were homogenized and 15 g of analytical portions (in triplicates) were taken. Analytical steps taken are presented in Figure 1. Total 210 analyses (5 stands/week x 14 weeks x 3 analytical portions) were performed. The analyses of the spiked samples and collected samples from the market were performed with the use QuEChERS method (AOAC, 2007). Chromatographic analyses of 377 pesticides were performed in the LC-MS/MS system and insecticide residues above LOQ were evaluated in the study.

Methodology for assessing dietary intake of insecticides

Acute and chronic risks to consumer health were estimated based on the previous studies (Chen et al., 2011; Soydan et al., 2021). Annual nectarine consumption per person was taken as 7.3 kg (i.e., 0.02 kg of nectarine per day) in Türkiye (TSI, 2022). The average body weight of an adult is taken as 60 kg in toxicological research (EFSA, 2019; WHO, 2021; Calderon et al., 2022). ADI (mg/kg bw/day) and ARfD (mg/kg bw/day) values of insecticides were taken from IUPAC Pesticide Properties DataBase (PPDB, 2022). ARfD values of insecticides were used for short-term acute dietary risk assessments (Liu et al., 2016; Malhat et al., 2021). ESTI (mg/ kg bw/day) and acute HQ values were calculated with the use of the following equations.

$$ESTI, mg/kg\ bw\ /\ day = \frac{Daily\ nectarine\ consumption, kg/day*high\ residue, mg/kg}{Body\ weight} x100 \qquad (1)$$

$$Acute\ HQ = \frac{ESTI, mg/kg\ bw\ /\ day}{ARfD, mg/kg\ bw\ /\ day} x100 \qquad (2)$$

Similar to acute risk assessments, ADI values were used for long-term chronic dietary risk assessments. EDI (mg/ kg bw/day) and chronic HQ values were calculated with the use of the following equations.

$$EDI, mg/kg\ bw\ /\ day = \frac{Daily\ nectarine\ consumption, kg/day*mean\ residue, mg/kg}{Body\ weight} x100 \qquad (3)$$

$$Chronic\ HQ = \frac{EDI, mg/kg\ bw\ /\ day}{ADI, mg/kg\ bw\ /\ day} x100 \qquad (4)$$

The level of concern for HQ value was set as 1. Therefore, HQ values of ≥ 1 represents a risk for human health and HQ values of < 1.0 presents non-potential risk for human health.

Results and Discussion

Method verification

Matrix-matched calibration curves of 12 insecticide standards were linear over the 1-1000 pg/µL concentration ranges with various determination coefficient (R^2 ≤0.999). R^2, retention times (tR, min) (ranged between 4.49-11.55 min) and MC line equations (4-point level) of all insecticides are also provided in Table 1. For quantification of the insecticides, regression equations of matrix-matched calibration curves (analytical function) were used. LOQs and MRLs of all insecticides are provided in Table 2. These LOQ values were quite lower than the MRLs.

Trueness and precision of the method are assessed as recovery (Q %) and repeatability (RSD %), respectively (Tiryaki, 2016; TURKAK, 2022). Percent recovery values together with standard deviation (SD) and RSD for Bayramiç Beyazı nectarine samples are given in Table 2. Individual recovery (mean recovery of 1 x LOQ and 10 x LOQ spiking levels with 5 replicate analyses) of each pesticide and their RSDs were provided in the table. Insecticide recoveries from nectarine samples varied between 65.2-115.3% with relative standard deviations (RSDs) of between 2.35-7.3%. The number of recovery data (n) was 10 for each insecticide. The overall method recovery was identified as 89.6% with a RSD of 11.8% (n=120). Present LOQ values (Table 2) also revealed that the method could detect insecticide residues lower than the MRL (Table 3) set by the EU (2022).

Present recovery values comply with method verification parameteres (SANTE, 2021; EURACHEM, 2014). Dülger & Tiryaki (2021) identified mean recovery of boscalid, chlorpyrifos and tebuconazole as 113.6% with an RSD of 11.4 % for nectarine and 113.5% with an RSD of 17.3% for peach. Galietta et al. (2011) measured the mean recovery of azoxystrobin, thiacloprid and acetamiprid in peaches as 95.3, 80.6, 98.6%, respectively.

These findings revealed that QuEChERS method may offer an accurate and rapid tool in detection of insecticide residues in Bayramiç Beyazı nectarine samples.

Table 1. Retention times (tR), calibration ranges, calibration curve equations, determination coefficients (R^2) and selected ion groups of the analyzed insecticides

İnsecticide	tR*, min	Calibration range, pg/µL	Calibration equation y=a+bx	Determination co-efficient, R^2	Precursor ion, m/z (CE**)	Fragment ion, m/z (CE)
Abamectin	11.5	5-1000	y=1030.1+813.014x	0.99955	890.4 > 305.2 (13)	890.4 > 567.3 (25)
Acetamiprid	4.9	1-100	y=7256.9+127041x	0.99996	223.1 > 125.9 (21)	223.1 > 55.9 (15)
Deltamethrin	11.3	3-600	y=813.5 +3904.8x	0.99987	523.0 > 280.9 (15)	523.0 > 506.0 (9)
Dimethoate	4.8	1-100	y=7458.5+113468x	0.99948	230.0 > 198.9 (9)	230.0 > 124.9 (20)
Etoxazole	11.0	1-100	y=9280.6+201567x	0.99991	360.2 > 140.9 (48)	360.2 > 112.9 (60)
Imidacloprid	4.4	1-100	y=1659.5+20360.1x	0.99874	256.0 > 209.0 (15)	256.0 > 175.0 (20)
Novaluron	10.3	1-200	y=-22.5 +7292.69x	0.99992	493.0 > 158.0 (18)	4 93.0 > 141.0 (48)
Omethoate	2.7	1-200	y=7881 + 107889x	0.99980	214.0 > 124.9 (20)	214.0 > 182.9 (10)
Pyriproxyfen	10.7	1-100	y= 56099+270369x	0.99831	322.3 > 95.4 (16)	322.1 > 227.1 (15)
Spirodiclofen	11.2	1-200	y=-05.91+ 11752.9x	0.99992	411.4 > 313.2 (11)	411.4 > 71.3 (18)
Tetramethrin	10.5	1-100	y=253436+58830.9x	0.99975	332.2 > 164.1 (25)	332.2 > 135.1 (16)
Thiacloprid	5.4	1-100	y=3346.4+176582x	0.99999	253.0 > 125.6 (20)	253.0 > 89.9 (39)

*tR, retention time (min); *** CE, Collision Energy (V)

Table 2. Spiking levels and recovery (including SD and RSD) values of insecticides obtained in method verification studies

İnsecticide	Spike level, µg/kg	Found , µg/kg	Recovery, %	Mean Recovery, % (As a tool for truness)	SD	RSD,% (As a tool for precision)
Abamectin	5 50	4.5 41.5	89.4 83.0	86.2	5.6	6.5
Acetamiprid	1 10	0.8 7.7	86.0 76.9	81.5	6.7	8.2
Deltamethrin	3 30	3.1 28.5	104.9 94.9	99.9	7.3	7.3
Dimethoate	1 10	0.9 7.4	93.4 73.6	83.5	11.4	13.6
Etoxazole	1 10	1.0 8.8	99.6 87.7	93.7	7.8	8.3
Imidacloprid	1 10	0.99 9.8	98.2 96.8	97.9	5.5	5.6
Novaluron	1 10	0.8 6.8	77.6 68.2	72.9	6.0	8.2
Omethoate	1 10	0.9 9.1	97.4 90.6	94.0	5.7	6.0
Pyriproxyfen	1 10	0.9 8.3	91.4 82.8	87.1	6.3	7.2
Spirodiclofen	1 10	1.1 9.2	109.8 92.5	101.1	9.6	9.5
Tetramethrin	1 10	0.8 8.2	85.8 82.1	83.9	4.4	5.2
Thiacloprid	1 10	0.9 8.9	99.0 89.7	94.4	6.1	6.4

Recovery range: 65.2-115.3; RSD range: 2.3 - 7.3%; Overall recovery (Accuracy): 89.6% with an RSD of 11.8% (n=120).

Residues in the "Bayramiç Beyazı" nectarines

Totally, 210 analytical portions, [70 samples (14-week x 5-stand, coded as A, B, C, D and E) and 3 replicates] were analyzed. A total of 12 insecticides, namely abamectin, acetamiprid, deltamethrin, dimethoate, etoxazole, imidacloprid, novaluron, omethoate, pyriproxyfen, spirodiclofen, tetramethrin, thiacloprid were detected in "Bayramiç Beyazı" nectarine samples.

The detected insecticide residues were below their MRLs, except for dimethoate, imidacloprid and omethoate. The insecticide (totally 8) residues detected only in a few samples and their details are given

in Table 3. Abamectin, imidacloprid and spirodiclofen residues (in one sample each) were found to be above the LOQs (below the MRLs) with the residue levels of 6.1, 5.0 and 32.0 µg/kg, respectively. Pyriproxyfen and thiacloprid residues (in two sample each) were found to be above the LOQs. Tetramethrin residues (above the LOQ) were found (106.6 µg/kg) only in one sample (12th week, Stand D). There is no specified MRL value for tetramethrin in nectarine. In one sample (2nd week, Stand E), dimehoate residue was found approximately 10 times (97.8 µg/kg) of the MRL value. Imidacloprid residues were found approximately 2 times (17.7 µg/kg) of the MRL in one sample (6th week, Stand B). Omethoate residue was found slightly over (10.2 µg/kg) the MRL values in one sample (2nd week, Stand A).

Omethoate and thiacloprid were banned in Türkiye on 31 August 2012 and 30 June 2022, respectively. Dimethoate is also in the process of being banned(BKÜ, 2023b).

Table 3. Insecticide residues observed in "Bayramiç Beyazı" nectarines and comparison with LOQ and MRL values

Insecticide	Below the MRL Residue, µg/kg (Number of sample detected)	MRL-exceeding Residue, µg/kg (Number of sample detected)	LOQ, µg/kg	MRL, µg/kg
Abamectin	6.1 (1)	-	5	20
Dimehoate	-	97.8 (1)	1	10
Imidacloprid	5.0 (1)	17.7 (1)	1	10
Omethoate	-	10.2 (1)	1	10
Pyriproxyfen	8.1 (1) 7.1 (1)	-	1	500
Spirodiclofen	32 (1)	-	1	2000
Tetramethrin		106.6 (1)*	1	-
Thiacloprid	85.5 (1) 26.5 (1)	-	1	500

*There is no specified MRL value for nectarine.

Residue levels of the remaining 4 insecticides, which were below the MRLs, are presented in Figure 2-5. Maximum acetamiprid residue (102.4 µg/kg, half of the MRL) was detected in the 1st week of Stand-C (Figure 2). Maximum deltamethrin (34.6 µg/kg, about 1/5 of the MRL, Figure 3) residue was detected in the 10th week of Stand-D, etoxazole (9.7 µg/kg, about 1/10 of the MRL, Figure 4) in the 12th week of Stand-D and novaluron (28.3 µg/kg, about 1/70 of the MRL, Figure 5) in the 9th week of Stand-C samples. Novaluron was banned in Türkiye on 30 June 2022.

Dülger & Tiryaki (2021) reported that boscalid, chlorpyrifos and tebuconazole residues in peach and nectarine samples were below the corresponded MRLs. Maximum boscalid residue in peach samples was measured as 566.8 µg/kg and maximum boscalid residue in nectarine samples was measured as 322.1 µg/kg. Maximum values for tebuconazole were 47.5 and 56.9 µg/kg, respectively. Chlorpyrifos residue levels were all below LOQ. In another study, chlorpyrifos residue levels greater than the corresponding MRL were reported for peach samples (Ersoy et al., 2011). In a previous study, Galietta et al. (2011) indicated that 25 and 12 days were required to pass after application for acetamiprid and thiacloprid residues below MRL in peach samples. Soydan et al. (2021) investigated pesticide residues in fruits and analyzed total 92 samples. Of the analyzed samples, 23.9% had residues below the LOQ, 57.6% had residues exceeding MRL and 18.4% had residues below the MRL. Osaili et al. (2022) found omehoate and dimethoate residues above the MRL in half of the nectarine samples they analyzed. In another study, 18 pesticide residues were found in peaches. Acatamprid residues exceeding the EU-MRL were found in 2 samples. Imidacloprid, pyriproxyfen and spirodiclofen residues were less than EU-MRL (Zhang et al., 2021).
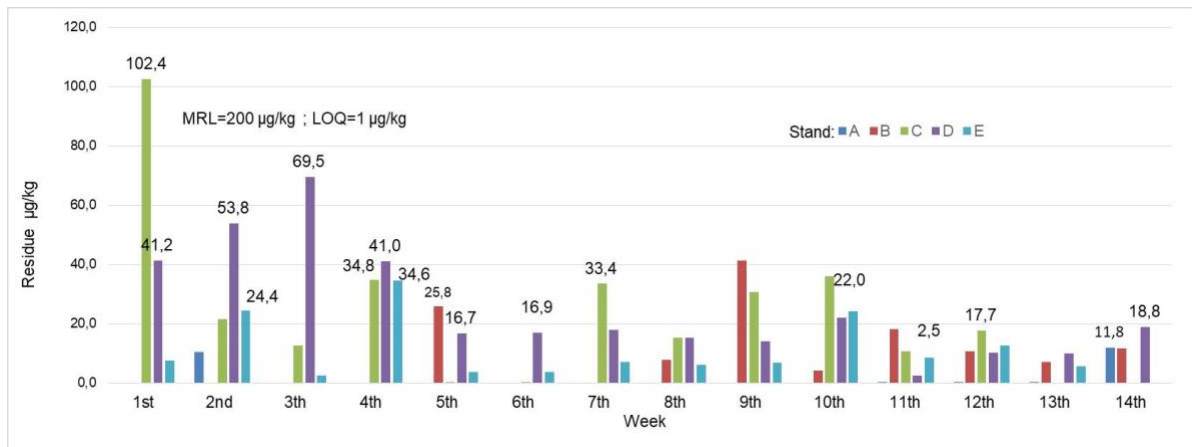
Figure 2. Stand and week-based acetamiprid residues in "Bayramiç Beyazı" nectarines.
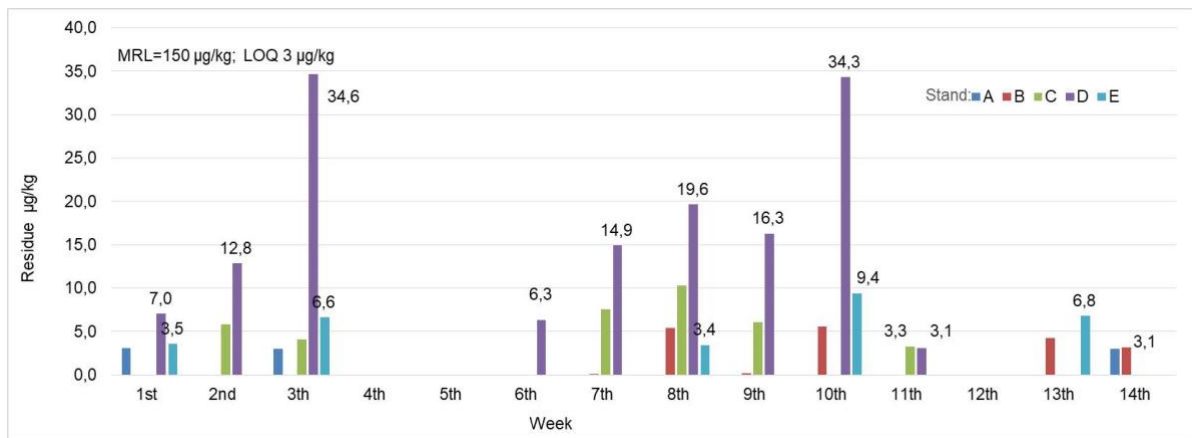

Figure 3. Stand and week-based deltamethrin residues in "Bayramiç Beyazı" nectarines.
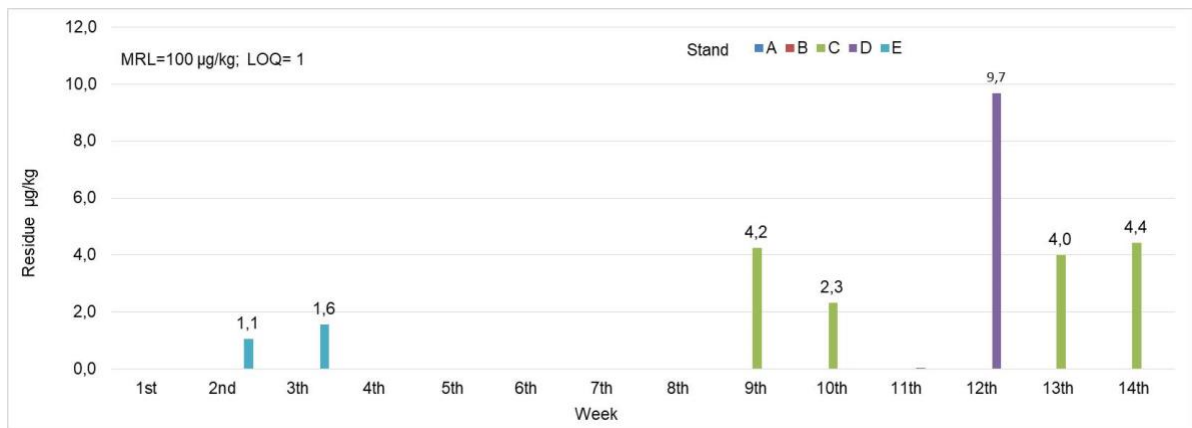

Figure 4. Stand and week-based etoxazole residues in "Bayramiç Beyazı" nectarines.
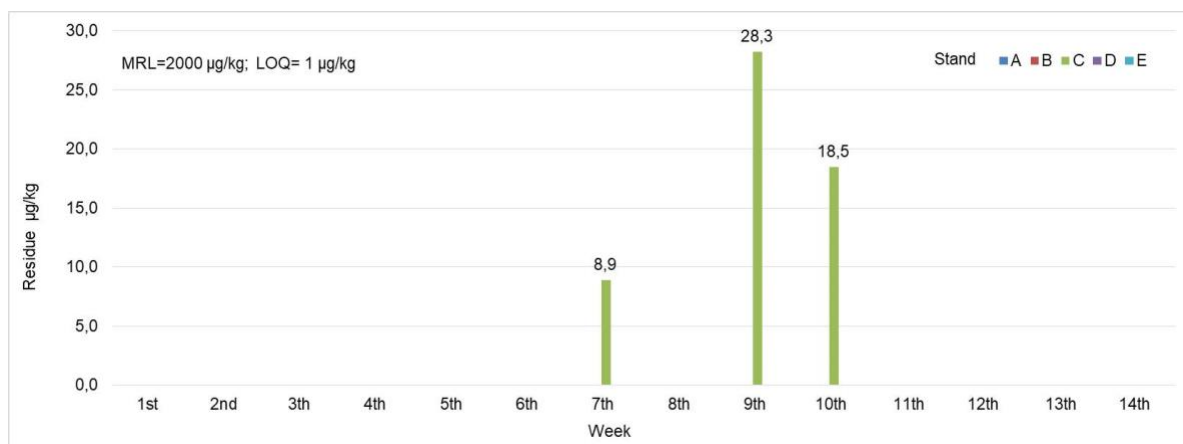
Figure 5. Stand and week-based novaluron residues in "Bayramiç Beyazı" nectarines.

Risk assessment for dietary intake of insecticides

For acute and chronic risk assessments, ESTI (mg/kg bw/day) and acute HQ were used for short-term risk and EDI (mg/kg bw/day) and chronic HQ were used for long-term risks. Resultant values are provided in Table 4. For all the insecticides, the number of residue figures and residue ranges (including mean residue) were also given in the table.

The ESTI values (calculated with Equation 1) for short-term risk ranged from 0.21E-05 to 3.45E-05. The acute HQ (calculated with Equation 2) of insecticides ranged between 0.0077 to 0.3449. The highest values of acute exposure (HQ_{acute}) were found for dimethoate (0.3449). This value was followed by Omethoate, acetamiprid, thiacloprid and deltamethrin insecticides with the HQ_{acute} values of 0.1742, 0.1386, 0.0989 and 0.0513, respectively. The EDI values (calculated with Equation 3) for long-term risk ranged from 0.09E-05 to 0.65E-05. The chronic HQ (calculated with Equation 4) of insecticides ranged between 0.0025 to 1.1285. The highest values of chronic exposure ($HQ_{chronic}$) were found for omethoate (1.1285). Therefore, with a $HQ_{chronic}$ value of ≥ 1, omethoate represents a risk for human health. This value was followed by abamectin, novaluron, thiacloprid, dimethoate, and deltamethrin insecticides with the HQ values of 0.0813, 0.0618, 0.0511, 0.0474, and 0.0327, respectively (Table 4). Deltamethrin, dimethoate and thiacloprid are moderately hazardous (Classs II), while omethoate and abamectin are highly hazardous (Class Ib) insecticides (WHO, 2019).

Both acute and chronic risk of insecticides with no ARfD and ADI values in PPDB Database (tetramethrin) were not assessed. Similarly, acute risk assessments of insecticides with no ARfD values in PPDB Database (etoxazole, novaluron, pyriproxyfen, spirodiclofen) were not performed. Although MRL- exceeded residues were found for dimethoate and imidacloprid, risk assessment revealed that there was no consumer acute and chronic health risk of all the insecticides because HQ values of all the insecticides (except omethoate) were less than 1.0. Even if their HQs were below 1, the highest acute risk was found for dimethoate with acute HQ of 0.3449. There is a risk for human health for omethoate since HQ values of ≥ 1. The lowest acute HQ and chronic HQ values belonged to imidacloprid and pyriproxyfen residues, respectively (Table 4).

Balkan & Yılmaz (2022b) performed dietary risk assessments for 260 compound residues of leafy vegetables. Pesticide residues were detected in 57.6% of samples. Of the samples, five had residue levels of above MRLs, however they posed no short and long-term risks on consumer health. The greatest risk was detected for acetamiprid with HQ_{acute} of 0.97% and for cypermethrin with $HQ_{chronic}$ of 0.29%. Our findings were in agreement with Kanbolat et al. (2023)'s findings, indicating omethoate and dimethoate cause acute and chronic toxicity for consumers.

Soydan et al. (2021) performed chronic health risk assessments for pesticide residues of vegetable and fruits. The lowest EDI values ranged from 357E-5 to 898000E-5. Lower HQ values were observed in strawberry, grape and dried apricot with a value of 0.01, although HQ value of 32 out of 62 insecticides tested was almost 0.

Dülger & Tiryaki (2021) performed consumer dietary chronic risk assessments for tebuconazole boscalid and chlorpyrifos residues on nectarine and peach matrices. Pestice residues did not exceed the MRLs in any samples. Risk assessments based on WHO method revealed that chronic exposure levels of insecticides were low and there was no risk to human health.

Chronic risk assessment for imidacloprid residues in peach and peach juice was carried out by Camara et al. (2020). The potential consumer risk for the imidacloprid pesticide was negligible for human health. In another study, acute and chronic dietary exposure assessment for peach indicated that potential dietary risk induced by the acetamprid, imidacloprid, pyriproxyfen and spirodiclofen pesticides were not signifcant for Chinese consumers (Zhang et al., 2021)

Table 4. Chronic and acute risk assessments of insecticides for "Bayramiç Beyazı" nectarine

Compound	Number of residue data	Residue range (mean resdue). ug/kg	Short term Acute dietary risk			Long-term Chronic Dietary risk		
			ARfD**	ESTI**	HQ $_{Acute}$	ADI**	EDI**	HQ$_{Chronic}$
Abamectin*	3	5.79-6.4 (6.1)	0.005	0.21E-05	0.0425	0.0025	0.20E-05	0.0813
Acetamiprid	147	1.6-103.9 (19.62)	0.025	3.47E-05	0.1386	0.0250	0.65E-05	0.0262
Deltamethrin	72	3.0-38.5 (9.8)	0.025	1.28E-05	0.0513	0.0100	0.33E-05	0.0327
Dimethoate	24	1.0-103.5 (14.2)	0.010	3.45E-05	0.3449	0.0010	0.47E-05	0.0474
Etoxazole	20	1.0-10.7 (4.05)	NL***	-		0.0400	0.13E-05	0.0034
Imidacloprid	6	4.9-18.6 (11.3)	0.080	0.62E-5	0.0077	0.0600	0.38E-05	0.0063
Novaluron	9	8.4-29.2(18.5)	NL	-	-	0.0100	0.62E-05	0.0618
Omethoate	3	9.7-10.4(10.1)	0.002	0.35E-5	0.1742	0.0003	0.34E-05	1.1285
Pyriproxyfen	6	6.5-8.5(7.6)	NL	-		0.1000	0.25E-05	0.0025
Spirodiclofen	81	1.1-36.7(2.6)	NL	-		0.0150	0.09E-05	0.0058
Tetramethrin	3	96.0-115.5(106.6)	NL	-		NL	-	
Thiacloprid	27	1.0-89.0(15.3)	0.030	2.97E-05	0.0989	0.0100	0.51E-05	0.0511

* Abamectin ARfD and ADI values were taken from EFSA (2020).
** The unit of ARfD, ESTI, ADI and EDI is "mg/kg‑bw/day".
*** NL: not listed; there was no specified ARfD and/or ADI in PPDB (2022).

Conclusion

Usage of insecticides is an important component of agricultural activities and significantly reduce labor costs for pest control. However, these chemical substances pose important risks on environment and human health. This study was conducted to investigate abamectin, acetamiprid, deltamethrin, dimethoate, etoxazole, imidacloprid, novaluron, omethoate, pyriproxyfen, spirodiclofen, tetramethrin, thiacloprid residues in "Bayramiç Beyazı" nectarines sampled from Çanakkale-Türkiye open market. The QuEChERS AOAC 2007.01 was efficiently used for the analyses of 12 insecticide residues on nectarine sample matrix. Method validation criteria were met. Residue levels of 9 insecticides were below the MRLs, whereas, in one sample each, dimethoate (approximately 10 times of MRL), imidacloprid (approximately 2 times of MRL) and omethoate (slightly over MRL) residues exceeded their MRLs. Diatery risk assessments revealed that present insecticide (except omethoate) concentrations did not pose any risks on human health. Omethoate was found to pose a chronic risk for human health. The highest acute HQ values were found for dimethoate, even if their HQ was ≤ 1. These two insecticides belong to the Organophosphate Class and should be taken into consideration. The use of dimethoate is in the process of being banned in Türkiye, while omethoate (metabolite of dimethoate) is already banned. Omethoate can cause residue as a degradation product of dimethoate.

Acknowledgements

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