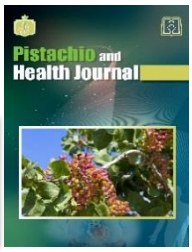
PHJ, Vol. 1, No. 2, Spring 2018, 1-6

Pistachio and Health Journal

Journal homepage: http://phj.rums.ac.ir



ORIGINAL ARTICLE

Assessment of Acetamiprid and Chlorpyrifos Residues on Fresh and Dried Pistachio Nuts

Ali Dini[a], Ali Alizadeh[b*], Ezat Alizadeh[b]

[a] *Pistachio Safety Research Center, Rafsanjan University of Medical Sciences, Rafsanjan, Iran.*
[b] *Department of Plant Protection, Faculty of Agriculture, Vali-e-Asr University of Rafsanjan, Rafsanjan, Iran.*

Introduction: In the recent years, the security and safety of foods have taken on high importance, due to the presence of pesticide residues in agricultural products. In this research, Chlorpyrifos and Acetamiprid residues have been investigated in the fresh, dried, split and non-split shelled pistachio from the variety of Ouhadi.

Material and method: The QuEChERS method was used for the extracting and cleaning up of the pesticides. The concentrations of Chlorpyrifos and Acetamiprid were determined using HPLC and Ion Mobility Spectrometer (IMS), respectively.

Results and discussion: The results showed that the amounts of Chlorpyrifos and Acetamiprid residues in the split samples were higher compared with those of the non-split shells, and the amounts of both pesticide residues in the dried samples were lower compared with those of the fresh ones. Chlorpyrifos and Acetamiprid residue levels in the fresh split samples were 2 and 2.5 times higher than the Maximum Residue Level (MRL), respectively. In the dried split and non-split samples, the amounts of both pesticides were less than those of MRL. As a matter of fact, the sun drying process reduces the amount of pesticide residues in pistachio nuts.

Conclusion: the pesticides were normally applied to control pistachio pests and even after the pre-harvest interval, as suggested in previous research, the amounts of pesticide residues in fresh split samples were higher than MRL; therefore, there should also be some more serious hygienic controls such as pesticide residues in fresh and split pistachio nuts. In addition, it seems one should be more careful when using fresh and split pistachio nuts.

Keywords: Acetamiprid; Chlorpyrifos; HPLC; Pesticide residues; QuEchERs method; IMS

1. Introduction

Iran is the largest producer of pistachio nuts in the world. This type of economically important nuts are produced in various regions of Iran [1]. Pesticide residues in agricultural products can be one of the major challenges to producers [2]. The management and control of pesticide residues in foods such as fruits and vegetables, as they are consumed raw and fresh, is a major international issue, since the public health, the environment and the foreign trade aspects of crops are affected by it [3, 4].

The Codex Food is an international organization that has set out a series of rules for the maximum pesticide residue levels permitted in foods. The objective of that organization is to ensure the health of consumers and uphold healthy farming practices through the introducing of rules on the maximum permissible levels of pesticide residues in foods, and also advance healthy food trades by governmental and non-governmental organizations [5]. The maximum concentration determined by this commission has been established as an MRL of the general law, in accordance with the general conditions existing in the world, and it can be used as a general guide for countries with no MRL laws, so the differences can be identified by comparing codex standards with other MRLs as determined by other countries [6]. The determination of a large number of pesticides in different agricultural products is necessary to formulate rules to be applied to pesticides in a country [7]. The increasing public concerns over human health threats caused by the presence of pesticide residues in the diet pattern of people has recently led to the introduction of more regulations on the quality control and food security [8]. Many countries have determined the maximum residual levels of pesticides in various foodstuffs due to their increasing awareness of the harmful carcinogenic, mutagenic and teratogenic effects on human [9], and in some developing countries, such regulations are not yet in place.

The determination of MRL in agricultural products is an important issue that is expressed as the concentration of the toxin per a kilogram of the fruit weight, and it is also the maximum allowable concentration of pesticides in foods not hazardous to human health. MRL has been determined for different pesticides at a range of 0.01-10 µg/g [10] being set at 0.05 µg/g for pistachio nuts by codex [11], and one of the most important factors that affect pesticide residues in agricultural products is pre-harvest intervals (PHI).

Food processing treatments such as washing, peeling, drying or cooking lead to a significant reduction of pesticide residues. Foods can be dried in several ways, for example by the sun or in an oven; a food dryer can also be used. Sun

* Corresponding author: Ali Alizadeh,
Email: alializadeh2004@gmail.com
Tel.: +98 343 1322940

DOI: 10.22123/PHJ.2018.108287.1002
Received: 24 November 2017
Accepted: 28 April 2018

drying is a simple and traditional method for preserving foods as compared with the oven or hot air drying methods, and it has more decisive effects on pesticide residue dissipation [12]. Athanasopoulos et al. (2005) reported that the drying of grapes lead to 64.2–71.9% losses of Methamidophos due to the evaporation of pesticides during the process [13].

One of the most important methods of reducing the potential threats of pesticide residues to human health is to monitor and control pesticide residues in various foodstuffs [14]. Today, the pesticide residue treatment in agricultural products is considered as an important priority to ensure the food safety of consumers. The purpose of this research is to monitor and control Chlorpyrifose and Acetamiprid residues and also to identify these pesticide residues in the fresh and dried split and non-split shelled pistachio nuts in14 days after being sprayed. In addition, the researchers of this study intend to investigate the effects of the drying process on the amounts of Chlorpyrifos and Acetamiprid residues in pistachio nuts.

2. Materials and Methods

2.1. Sample preparation

The spraying of pistachio trees was carried out with the commercial formulation of Chlorpyrifos (EC 40. 8% of the Policom Company of Iran) and Acetamiprid (SP of 20% of the Moshkfam Company of Iran) at the rate of 1.5-2 grams in 1000 liters and 200 to 250 grams in 1000 liters of water, respectively.

The processes of sampling were carried out after 14 days of spraying in several pistachio-cultivated farms in Rafsanjan town. All of the round pistachio cultivars (Ouhadi) have been selected from the specimens. Samples were selected based on a random selection method, and the chances of selecting the split and non-split samples were similar, and sampling with the X pattern of one of the four randomly selected trees was performed by removing 4 clusters from each row [15]. Following the gently mixing of the samples, one kilogram of pistachios was randomly taken as the final sample of the mixed harvest. Pistachio samples went through peeling, and then 200 grams of both fresh split and non-split shell samples were randomly taken again and finally, 50 grams of them were selected as the final sample after the seed-washing process and separating the endocarp. To prepare dried samples, one kilogram of the split and non-split shell samples was dried under sun-drying conditions for 3 days after peeling and washing them. Other preparation stages were as similar as the steps followed for preparing fresh samples. Specimens were immediately transferred to the laboratory for homogeneous mixing, and they were then kept in the dark at -20 °C to be further used in the experimental analysis.

2.2. Chemicals and reagents

Chlorpyrifos and Acetamiprid standards were procured from Sigma (MO, U.S.A). Acetone, acetonitrile (Caledon laboratories Ltd, Canada) and water were of the high-performance liquid chromatography (HPLC) grade. Sodium chloride, magnesium sulfate and silica gel (80-60 mesh) (Merck, Darmstadt, Germany) were used in the present research.

2.3. Extraction and cleanup

The QuEChERS method was used to extract the samples [16]. In this method, for the extraction of pesticides, 50 grams of the samples were mixed and homogenized. Afterwards, 10 grams of specimen were taken and poured into a 50-mL volumetric flask after 10 mL of acetonitrile, 1 gram of sodium chloride and 4 grams of anhydrous magnesium sulfate were added, and mixed together. In order to remove impurities, the solution was centrifuged at 3000 rpm for 10 minutes, and the supernatant was used for the cleanup process. Firstly, the supernatant was passed through a glass column filled with 4 grams of silica gel [80-60 mesh] and then, the column was washed with acetone to remove oil and other impurities. Next, 30 mL of acetonitrile were used to extract the analyte from the column, and it was then collected in a special container; the extract was concentrated to 1 mL by blowing a stream of nitrogen gas. The sample was then prepared for measurement using IMS and HPLC [17].

2.4. Determination of Chlorpyrifos residues

The Liquid Chromatography (LC) Analysis was performed to measure the Chlorpyrifos residues using the reverse-phase system (Knauer, Germany) equipped with a UV-Vis detector at 254 nm, and the Dionex LC column (C18, 250 mm × 4.6 mm, 5 µm) was set at 36°C. The binary mixture of water /acetonitrile (60:40) was used as a mobile phase with the flow rate of 1ml/min and the retention time for Chlorpyrifos was 3.1 minutes. A calibration curve was prepared using the working standard solutions through pipetting appropriate volumes into a set of 20-ml calibrated volumetric flasks, diluted with acetonitrile at the 0.01, 0.02, 0.04, 0.08, 0.16 µg/g concentration. The calibration curve was plotted in Fig. 1. It was constructed before the analysis to check the pilot for linearity (r^2= 0.998), and it was used for the quantification of Chlorpyrifos(Fig. 1). The concentration of pesticides in samples was also calculated using the sub-peak area, and it was placed in the standard calibration curve of pesticides; the chromatogram of Chlorpyrifos pesticides extracted by the QuEChERS method as also been shown in Fig. 2.

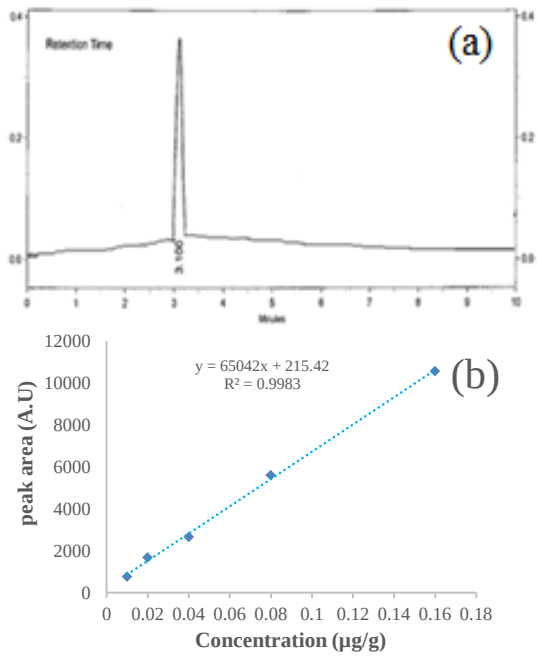


Fig. 1. The chromatogram of the Chlorpyrifos standard (a) and the calibration curve of the Chlorpyrifos pesticide (b)

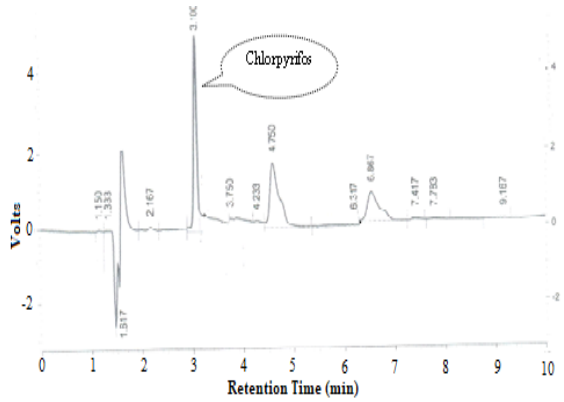


Fig. 2. The chromatogram of Chlorpyrifos pesticide extracted using the QuEChERS method

2.5. The determination of the Acetamiprid pesticide residue

In order to measure the residue of Acetamiprid, IMS was used. In this analysis, a syringe pump (LSP01-A) was used to make the drowning gas (N_2) enter the machine at the flow rate of 0.5 ml/min. Standard solutions at different concentrations of pesticides (0.005, 0.01, 0.02, 0.06, 0.12µg/g) were prepared by pipetting appropriate volumes of the working solution into a set of 20-ml calibrated volumetric flasks, and acetonitrile was applied to the volumes to dilute them, and 10µL was injected into the device, and then calibration curves were plotted (Fig. 3b). The concentration of the pesticide was calculated using the sub-peak area of samples, and placing it in the standard calibration curve equation. Analyte spectrum has a peak of about 9.4 ms that has been well separated from the reactant ion peaks. The ion mobility spectrum of the spiked sample was extracted based on the QuEChERS procedure as shown in Fig. 3a.

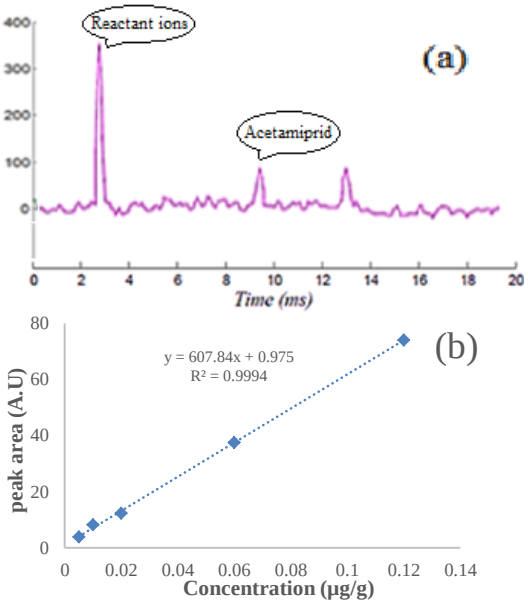


Fig. 3. The spectrum of the Acetamiprid pesticide extracted using the QuEChERS method (a) and the calibration curve of the Acetamiprid pesticide (b)

2.6. Quality assurance

To evaluate the reliability of results, in addition to using validated methods, internal quality control experiments were also performed. The accuracy and precision of the methods were verified. Therefore, the method demonstrated a good performance at low statutory limits. Moreover, the limit of quantification (LOQ) was acceptable, implying the reliability of the generated data (Table 1). The average recoveries and the relative standard deviation for the reproducibility (RSDr) of the analytical method applied for the measurement of various pesticides in pistachio nuts are given in table 1. The limit of detection for each pesticide was calculated using equation 1.

Eq. 1:
$$LOD = \frac{(3S_{y/x})}{m}$$

$S_{y/x}$ is the return error of x to y, and m is the slope of the calibration curve. $S_{y/x}$ was calculated using Equation 2.

Eq. 2:
$$S_{y/x} = \left[\frac{\sum_{i=1}^{n}(yi-yj)^2}{x-2}\right]^{1/2}$$

In this equation, yi is the individual points of the calibration line, yj is the quantity coincided on the calibration line, and x is the number of calibration curve points.

Table 1. Performance characteristics of the analytical method

Pesticide/Method	Sample	LOD[a] (µg/g)	LOQ[b] (µg/g)	No.[c]	Spike level (µg/g)	Average recovery (%)	RSDr[d] (%)
Chlorpyrifos/HPLC	Fresh split	0.008	0.024	5	0.035	79.12	2.7
	Dried split			5	0.035	86.7	2.6
	Fresh non-split			5	0.035	80.2	2.4
	Dried non-split			5	0.035	87.3	2.1
Acetamiprid/IMS	Fresh split	0.004	0.01	5	0.035	94.9	2.2
	Dried split			5	0.035	98.5	0.6
	Fresh non-split			5	0.035	94.8	2.7
	Dried non-split			5	0.035	97.5	0.6

[a] Limit of detection.
[b] Limit of quantification.
[c] Number of the spiked pistachio nut samples analyzed using HPLC or IMS.
[d] Relative standard deviation for reproducibility.

In this method, for the analysis and measurement of residual pesticide residues in the fresh and dry pistachios, due to the presence of intrusive matrices and compounds and also the high fat content in the pistachios, 100% of the residues in the pistachio seeds is not extracted; as a result, it is highly recommended to use the column chromatography method to extract pesticides from the oil in new methods [18]. Before carrying out the operation on the main samples, a recovery test must be first performed [19]. Recoveries of Chlorpyrifos and Acetamiprid were recorded by analyzing a blank pistachio nut sample spiked at 0.035 µg/g for each pesticide. Thus, the amount of the residues of each pesticide was measured and the recovery percentage was calculated according to Equation 3; pesticide levels were also corrected based on the recovery values.

Eq. 3:

$$\%\text{Recovery} = \frac{\text{The amount of pesticide measured in the spiked sample}}{\text{The amount of pesticide added}} \times 100$$

2.7. Data analysis

Data were analyzed using software SAS 9.1.3. The normality of data was verified using the Kolmogrov-Smirnov test. The ANOVA test was used to evaluate the means. T-test (LSD) with the 95% significance level was utilized for the comparison of means. SigmaPlot 12.0 software was used to plot charts.

3. Results

To determine the degree of diagnosis and the accuracy of the device, the test was carried out where the limit of detection (LOD) averaged 0.008 µg/g for Chlorpyrifos and 0.001 µg/g for Acetamiprid. Therefore, the method demonstrated a good performance at low statutory limits. Furthermore, the limit of quantification (LOQ) was acceptable (Table 1) implying the reliability of the data generated. The results of average recoveries, and the relative standard deviation for the reproducibility (RSDr) of the analytical method applied for the measurement of various pesticides in pistachio nuts are given in Table 1. The average recoveries were 83.3% for Chlorpyrifos and 96.4% for Acetamiprid, and the RSDr values were within the acceptable ranges, indicating the high accuracy and precision of the analytical method used. The results of the determination of the recovery rate showed that using the column chromatography method to extract pesticides from the supernatant, the acceptable amount of pesticides could be extracted from the intrusive matrices of pistachio nuts. According to Table (2), Chlorpyrifos residues in fresh split and non-split shell treatments were determined to be 0.1 µg/g and 0.029 µg/g, respectively. Moreover, the residue of this pesticide was 0.0275 µg/g in split dried samples, and the amount of Chlorpyrifos in non-split shell dried samples was less than LOD.

The Acetamiprid pesticide residue in split and non-split fresh samples was equal to 0.128 µg/g and 0.048 µg/g, respectively. In split and non-split dried samples, the pesticide residue was equal to 0.045 µg/g and 0.0035 µg/g, respectively, being lower than MRL. The amounts of Chlorpyrifos and Acetamiprid residues were obtained in different treatments and were compared with the reported MRLs from the Codex Food Commission (0.05 µg/g). As it is shown in the results, in fresh split samples, the Chlorpyrifos and Acetamiprid pesticide residues were 2 and 2.5 times more than the MRL reported by the Codex Food Commission, respectively.

As it is shown in Table 2, the dried pistachio nuts had lower pesticide residues compared with the fresh pistachio nuts. Sun drying leads to around a 72.5% decline and a 64.8% decline in Chlorpyrifos and Acetamiprid residues from the present 0.1 µg/g and 0.128 µg/g rates at split samples, respectively, while the sun drying process of the non-split shell sample leads to a 100% decline and a 92.7% decline in Chlorpyrifos and Acetamiprid residues from the present 0.029 µg/g and 0.048 µg/g in the fresh samples, respectively.

4. Discussion

In the split shells, the amount of the pesticide residues is higher than that of the non-split shell ones, whereas all samples are taken from a region. Because the pistachio hard skin (endocarp) is a barrier against the penetration of pesticides and prevents them from penetrating into the edible part of the nuts [kernel], the non-split endocarp can reduce the penetration rate of the pesticides into the kernel; as a result, the amount of the pesticide residues was expected to decrease significantly in the non-split shell

treatments of pistachio nuts [17].

Table 2. Results of Chlorpyrifos and Acetamiprid pesticide residues on the pistachio kernel

Pesticide	Treatment	Remaining amount (µg/g)	MRL (µg/g)
Chlorpyrifos	Fresh split	0.1 ± 0.0170	0.05
	Dried split	0.0275 ± 0.0082	0.05
	Fresh non-split	0.0170 ± 0.029	0.05
	Dried non-split	< LOD	0.05
Acetamiprid	Fresh split	0.128 ± 0.0236	0.05
	Dried split	0.045 ± 0.0129	0.05
	Fresh non-split	0.048 ± 0.0185	0.05
	Dried non-split	0.0049 ± 0.0004	0.05

Phopin et al. [2017] reported that the rind of mangosteen (*Garcinia mangostana L.*) had barrier effects on the penetration by pesticides [dimethoate, metalaxyl and carbofuran]. The pesticide residues cannot penetrate into it, and the Mangosteen pulp would be less harmful to the consumers [20]. The pistachio cultivar selected in this research was a kind of round pistachio (Ouhadi). Ouhadi is one of the most important varieties of pistachio cultivars that is very famous and is cultivated in approximately 60-70% of Rafsanjan pistachio nut gardens. It is a species of pistachios with a premature fruit that can be harvested at the beginning of September [21]. The properties mentioned make the fruit split faster and make the crust slice off in a form named "early-split". It is one of the most important and harmful complications in pistachio trees and can make the product get exposed to pesticides more easily. Therefore, premature varieties can also affect the amount of residual pesticides and make them remain in the product. The Acetamiprid and Chlorpyrifos pesticides that have intrusive effects remain inside the bony crust and stay there for longer than the duration of the PHI that is reported so that can even exceed the limit of 21 days (data are not shown) [22]. According to the results achieved by Yakhdani et al. on Amitraz and Phosalone in the pistachio seeds, a slight decrease in the amount of residual pesticides was shown in the short term [17]. While this interval has been observed in the current study, the remaining residue is due to the organic phosphorous pesticides being less soluble and more stable in water [23]. It also seems that the length of the pre-harvest intervals differs for different pesticides, and that it depends on their physical and chemical structures. This factor also varies depending on the formulation of pesticides and spraying methods [24-26]. Environmental factors such as temperature, the humidity of the environment and sunlight can also affect the length of time (the period of PHI); the type and the variety of the plant and its morphological and physiological characteristics can also be effective during PHI [27-29].

The amount of the pesticide residue in the dried samples was less than MRL, and dried pistachio nuts had lower pesticide residues as against the fresh fruit. In previous studies, the effect of sun drying on the reduction of pesticide residues has been reported [30]. Sun drying has been found to reduce bitertanol by 50% and dimethoate by 80% of residues in the apricot fruit and raisin, respectively [12, 31]. The decrease in pesticide residues was attributed to the heat that could cause evaporation and degradation due to photo-degradation; sun drying has also a greater effect on the pesticide degradation as against other drying methods such as oven drying [31, 32], therefore in this research, the sun drying of pistachio nuts reduced a significant amount of Chlorpyrifos and Acetamiprid.

5. Conclusions

The present results indicate that the QuEChERS pretreatment method accompanied by the column extraction in combination with HPLC and the IMS analysis technique proved to be an accurate, convenient, simple and reliable tool for the determination of Chlorpyrifos and Acetamiprid residues in pistachio nut matrices. Early split is one of the most important and harmful complications in pistachio trees that makes pistachio seeds split prematurely when the fruit is ripped in August, and due to favorable reproductive conditions of insects, farmers are simultaneously forced to use insecticides. This process leads to direct exposure of pistachio kernel to the pesticide and it remains for a longer period of time in the product. It also increases PHI more than the periods reported in previous researches. The results show that pesticide residues in the fresh split samples remain higher than MRL. Based on the results obtained, sun drying leads to pesticide residue dissipation, due to the evaporation and photo-degradation of pesticides. Therefore, these findings may suggest that more attention should be paid to the consumption of fresh and split shelled pistachio nuts.

Conflict of interest

The authors declare no conflicts of interest.

References

1.Dini A, Khazaeli P, Roohbakhsh A, Madadlou A, Pourenamdari M, Setoodeh L, Askarian A, Doraki N, Farrokhi H, Moradi H. Aflatoxin contamination level in Iran's pistachio nut during years 2009–2011. Food Control. **2013**;30[2]:*540-4*.

2.Larsen AE. Agricultural production in the 21st century: Land-use, diversity, pests and pesticides: University of California, Santa Barbara; **2015**.

3.Cortés Aguado S, Sánchez-Morito N, Garrido Frenich A, Martínez Vidal J, Arrebola F. Screening Method for the Determination at Parts Per Trillion Levelsof Pesticide Residues in Vegetables Combining Solid-Phase Microextraction and Gas

Chromatography-Tandem Mass Spectrometry. Analytical Letters. **2007**;40[15]:*2886-914*.
4.Melo LF, Collins CH, Jardim IC. High-performance liquid chromatographic determination of pesticides in tomatoes using laboratory-made NH 2 and C 18 solid-phase extraction materials. Journal of Chromatography A. **2005**;1073[1]:*75-81*.
5.Cooper J, Leifert C, Niggli U. Handbook of organic food safety and quality: Elsevier; **2007**.
6.Núñez L, Tadeo J, García-Valcárcel A, Turiel E. Determination of parabens in environmental solid samples by ultrasonic-assisted extraction and liquid chromatography with triple quadrupole mass spectrometry. Journal of Chromatography A. **2008**;1214[1]:*178-82*.
7.Yenisoy-Karakaş S. Validation and uncertainty assessment of rapid extraction and clean-up methods for the determination of 16 organochlorine pesticide residues in vegetables. Analytica Chimica Acta. **2006**;571[2]:*298-307*.
8.Schmidt RH, Rodrick GE. Food safety handbook: John Wiley & Sons; **2003**.
9.Sharif Z, Man YBC, Hamid NSA, Keat CC. Determination of organochlorine and pyrethroid pesticides in fruit and vegetables using solid phase extraction clean-up cartridges. Journal of Chromatography A. **2006**;1127[1]:*254-61*.
10.Arrebola F, Vidal JM, Mateu-Sánchez M, Álvarez-Castellón F. Determination of 81 multiclass pesticides in fresh foodstuffs by a single injection analysis using gas chromatography–chemical ionization and electron ionization tandem mass spectrometry. Analytica Chimica Acta. **2003**;484[2]:*167-80*.
11.Tadeo JL. Analysis of pesticides in food and environmental samples: CRC Press; **2008**.
12.Cabras P, Angioni A, Garau VL, Melis M, Pirisi FM, Cabitza F, Pala M. Pesticide residues in raisin processing. Journal of Agricultural and Food Chemistry. **1998**;46(6):*2309-11*.
13.Anastassiades M, Lehotay SJ, Stajnbaher D, Schenck FJ. Fast and easy multiresidue method employing acetonitrile extraction/partitioning and dispersive solid-phase extraction for the determination of pesticide residues in produce. Journal of AOAC International. **2003**;86(2):*412-31*.
14.Wang S, Yu C, Zhang Y, Wang J, Duan Z, Zhang J. Tube-immunoassay for rapid detection of carbaryl residues in agricultural products. Journal of Environmental Science and Health Part B. **2006**;41(5):*693-704*.
15.Lee PW, Barefoot A, Murphy J, Aizawa H. Handbook of residue analytical methods for agrochemicals: Wiley Chichester, West Sussex, England; Hoboken, NJ; **2003**.
16.Anastassiades M, Lehotay SJ, Štajnbaher D, Schenck FJ. Fast and easy multiresidue method employing acetonitrile extraction/partitioning and "dispersive solid-phase extraction" for the determination of pesticide residues in produce. Journal of AOAC international. **2003**;86(2):*412-31*.
17.Dehghani Yakhdani D. Investigating and measuring the residue of two insecticides in the pistachio kernel (MS Thesis)**1998**.
18.Goodspeed DP, Chestnut L. Determining organohalides in animal fats using gel permeation chromatographic cleanup: repeatability study. Journal-Association of Official Analytical Chemists. **1991**;74(2):*388-94*.
19.Hassanzadeh N, Bahramifar N, Esmaili-Sari A. Residue content of carbaryl applied on greenhouse cucumbers and its reduction by duration of a pre-harvest interval and post-harvest household processing. Journal of the Science of Food and Agriculture. **2010**;90(13):*2249-53*.
20.Phopin K, Wanwimolruk S, Prachayasittikul V. Food safety in Thailand. 3: Pesticide residues detected in mangosteen (Garcinia mangostana L.), queen of fruits. Journal of the Science of Food and Agriculture. **2017**;97(3):*832-40*.
21.Panahi B, Talaie A, Mirdamadiha F. Determination of the best time of harvest in different commercial Iranian pistachio nuts. Options Méditerranéennes, Série A. **2005**;*63:215-9*.
22.Rogers ME. General pest management considerations. Citrus Industry. **2008**;*89:12-7*.
23.Arjmandi R, Tavakol M, Shayeghi M. Determination of organophosphorus insecticide residues in the rice paddies. International Journal of Environmental Science & Technology. **2010**;7(1):*175-82*.
24.Alister C, Araya M, Becerra K, Saavedra J, Kogan M. Preharvest Interval Periods and their relation to fruit growth stages and pesticide formulations. Food chemistry. **2017**;221:*548-54*.
25.Liu C, Wan K, Huang J, Wang Y, Wang F. Behavior of mixed formulation of metalaxyl and dimethomorph in grape and soil under field conditions. Ecotoxicology and Environmental Safety. **2012**;84:*112-6*.
26.Alister C, Araya M, Morandé J, Volosky C, Saavedra J, Cordova A, Kogan M. Effects of wine grape cultivar, application conditions and the winemaking process on the dissipation of six pesticides. Ciencia e Investigación Agraria. **2014**;41(3):*375-86*.
27.Mandal S, Das S, Bhattacharyya A. Dissipation study of thiophanate methyl residue in/on grapes (Vitis vinifera L.) in India. Bulletin of Environmental Contamination and Toxicology. **2010**;84(5):*592-5*.
28.Huo R, Salazar J, Hyder 3 K, Xu X-M. Modelling non-systemic pesticide residues in fruits with initial deposit variability and weather effects. Food Additives and Contaminants. **2007**;24(11):*1257-67*.
29.Marín A, Oliva J, Garcia C, Navarro S, Barba A. Dissipation rates of cyprodinil and fludioxonil in lettuce and table grape in the field and under cold storage conditions. Journal of Agricultural and Food Chemistry. **2003**;51(16):*4708-11*.
30.Kaushik G, Satya S, Naik S. Food processing a tool to pesticide residue dissipation–A review. Food Research International. **2009**;42(1):*26-40*.
31.Cabras P, Angioni A, Garau VL, Melis M, Pirisi FM, Cabitza F, Cubeddu M. Pesticide residues on field-sprayed apricots and in apricot drying processes. Journal of Agricultural and Food Chemistry. **1998**;46(6):*2306-8*.
32.Athanasopoulos PE, Pappas C, Kyriakidis NV, Thanos A. Degradation of methamidophos on soultanina grapes on the vines and during refrigerated storage. Food Chemistry. **2005**;91(2)*:235-40*.