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RESEARCH ARTICLE

Effect of storage conditions on removal of 32 pesticide residues in detox waters

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Abstract
Detox water, a beverage widely consumed for weight management and health benefits, is typically prepared using fruits and vegetables such as green apple, parsley, cucumber, mint, and lemon. This study evaluated the impact of storage conditions (ambient temperature and 4 °C) over 72 h on the concentrations of 32 pesticide residues commonly found in these ingredients. Changes in pesticide residue levels and their removal rates were analyzed during storage. Results demonstrated a consistent decrease in pesticide residue concentrations with longer storage durations under both conditions. Notably, at ambient temperature, the pesticide clofentezine exhibited the highest removal rate of 89% after 72 h. These findings provide valuable insights into the behavior of pesticide residues in aqueous media under varying storage conditions, contributing to improved safety practices for detox water preparation and storage.

Keywords Detox water · Health · Pesticide · Removal rate · Storage temperature

Introduction

Fruits and vegetables are vital sources of nutrients in human diets, playing a key role in maintaining health and preventing diseases. The World Health Organization (WHO) recommends a daily intake of at least 400 g (or five portions) of fruits and vegetables to lower the risk of certain noncommunicable diseases (NCDs), such as cardiovascular disorders and specific cancers (WHO, 2019). However, the widespread use of pesticides in agriculture raises concerns about contamination in these essential foods, posing potential risks to human health (Philippe et al. 2021). Pesticides have been associated with adverse health outcomes, including cancer, neurodegenerative diseases like Parkinson's and Alzheimer's, reproductive and respiratory disorders, and endocrine disruption (Sabarwal et al. 2018; Valcke et al. 2017).

In recent years, the growing emphasis on weight management and the prevention of obesity has popularized various diet programs. Among these, detox diets have emerged as a prominent trend, aiming not only to support weight control but also to promote toxin elimination, boost immunity, and improve overall health (Klein and Kiat 2015). A key component of detox diets is detox water, a beverage made by infusing water with fruits, vegetables, and herbs. These ingredients release phytochemicals into the water, offering potential health benefits (Ariyawansa and Ramanathan 2021). Additionally, detox waters are low in fat, high in fiber, and cholesterol-free, making them a valuable tool in reducing the risk of obesity (Snyder and Clum 2014).

The ease of preparation has made detox water highly appealing to consumers. It is typically made by blending or infusing washed fruits and vegetables and is stored at either room temperature or in the refrigerator for consumption at regular intervals. Commonly used ingredients include cucumber, mint, green apple, lemon, and parsley. However, these fruits and vegetables are often contaminated with pesticide residues. Researches has identified several pesticides commonly detected in produce, including acetamiprid, azoxystrobin, boscalid, clofentezine, malathion, and tebuconazole (Toptanci et al. 2021; Zhang et al. 2023; Bakırcı et al. 2014). Notably, malathion is the most commonly found pesticide in lemons (Aslantas et al. 2023), while tebuconazole

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and pyrimethanil are often present in apples (Zhao et al. 2023; Kowalska et al. 2022).

Despite the growing popularity of detox water, there is limited research on the behavior and removal rates of pesticide residues in aqueous media during storage. This study aims to address this gap by examining the removal behavior of 32 commonly found pesticide residues in detox water under different storage conditions (ambient and cold temperature) over various time intervals. Pesticide residue concentrations were quantified using advanced analytical techniques, including LC–MS/MS and GC–MS/MS, combined with the QuEChERS method. The methodology was validated following SANTE guidelines (SANTE/12682/2019). This study is the first to provide a comprehensive analysis of the effects of time and temperature on the removal rates of pesticide residues in detox water, offering critical insights for improving food safety and consumer health.

Materials and methods

Reagents and chemicals

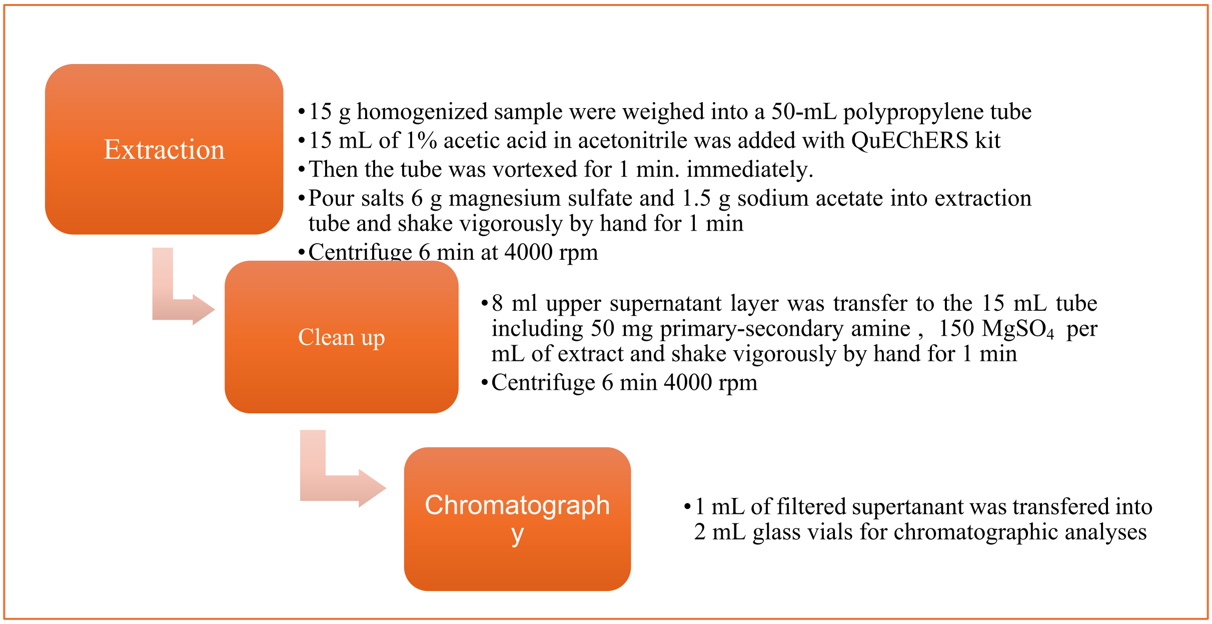
All reagents were of analytical grade. All the standards were of high purity (> 95.0%) and standards were supplied by Chem. Service (West Chester, USA). Pesticide reference standards were used for analyzing pesticide residues. Individual stock solutions were prepared in acetonitrile and stored at – 18 °C. The QuEChERS kits (part no. 5982–5755) with 6 g magnesium sulfate, 1.5 g sodium acetate, and 15-mL centrifuge tubes with 1200 mg magnesium sulfate and 400 mg primary-secondary amine (PSA) for dispersive solid-phase extraction (dSPE, part no. 5982–5058) were purchased from Agilent Technologies (USA).

Preparation of detox water

The green apple, parsley, cucumber, mint, and lemon were collected in year 2023 from local market of Balikesir, Turkey. Before analyses, vegetables and fruits were hand washed under running water then they were dried on filter paper until excessing all water. Before making detox water, vegetables and fruits were analysed before experiments to make sure that they were from any of the pesticides to be evaluated in this study. Detox water was from fresh vegetables and fruits (one green apple, a few sprigs of parsley, one cucumber, a few sprigs of mint, and one lemon) cut and put in a Vestel Mix and Go Smoothie Blender (Manisa, Turkey) and blending them. Fresh detox water was sprayed at a concentration of 50 µg kg^{-1} pesticide active substances.

Sample extraction and clean-up

Pesticide residues was determined using extraction and clean up procedures in QuEChERS AOAC Method 2007.01 were performed according to Lehotay (2007). Analytical steps of QuEChERS-AOAC Method 2007.01 are shown in Fig. 1.



- 15 g homogenized sample were weighed into a 50-mL polypropylene tube
- 15 mL of 1% acetic acid in acetonitrile was added with QuEChERS kit
- Then the tube was vortexed for 1 min. immediately.
- Pour salts 6 g magnesium sulfate and 1.5 g sodium acetate into extraction tube and shake vigorously by hand for 1 min
- Centrifuge 6 min at 4000 rpm

- 8 ml upper supernatant layer was transfer to the 15 mL tube including 50 mg primary-secondary amine , 150 $MgSO_4$ per mL of extract and shake vigorously by hand for 1 min
- Centrifuge 6 min 4000 rpm

- 1 mL of filtered supertanant was transfered into 2 mL glass vials for chromatographic analyses

Fig. 1 Analytical procedure of the QuEChERS-AOAC official method 2007.01

LC–MS/MS and GC–MS/MS instruments and conditions

Pesticide residue determination was performed using Shimadzu 8045 LC–MS/MS (Shimadzu, Kyoto, Japan) and Agılent 8890A-7000D GC–MS/MS (Agilent Technologies USA):

- LC–MS/MS analyses was performed with Restek Biphenyl (2.1 mm × 100 mm, 2.7 µm) column. Samples were analyzed with the mobile phase (A) water with ammonium formate 5 mM, and 0.1% formic acid, and (B) methanol. Mobil phase was same for all analytes. The final LC–MS/MS method conditions are listed in Table S1. Chromatographic parameters of pesticides analyzed by LC–MS/MS are listed in Table S2.
- The Agilent 7890A-7000D GC–MS/MS model was used for pesticide residue analyses. For pesticide separation, HP-5MS column (30 m i.d. × 250 µm × 0.25 µm) was used. The column was set at a constant flow rate of 3 mL/min using helium as carrier gas. The ion source and transfer line temperature was set at 280 °C and injection volume was 2 µL. General GC–MS/MS information for analyzed pesticide are listed in Table S3.

Statistical analysis

Removal rates of residuals pesticides were evaluated by the following equations:

$$Removal\ rate\ (\%) = \left(m_{before} - m_{before}\right)/m_{before} \times 100 \quad (1)$$

m_{before} and m_{after} refers to the mass of the residual pesticides in detox water before and after the storage, respectively.

Cluster analyses was conducted using Origin 2025 software (OriginLab Corporation, USA). Heatmap is a graphical representation of data where the individual values contained in a matrix are represented as colors. Cluster analysis was carried out in the form of heat map to explore the difference of removal rate under different storage conditions.

Results and discussion

Method validation

The analytical method was validated by evaluating the recovery, linearity, precision, limit of detection (LOD), and limit of quantification (LOQ). Matrix-matched calibration curves for each analyte was used for linearity of this method validation. The calibration curves were prepared in red pepper blank acetonitrile extracts using the multi-residue working solutions and filling up the volume with blank samples' extracts (SANTE/12682/2019 2020). The calibration curves in red pepper were prepared in blank matrix using concentration of 5, 10, 20, 50, and 100 µg kg^{-1}. Good linearity was achieved in the range studied. The LOD and LOQ were defined respectively as the signal corresponding to three and ten times the noise ratio, determined experimentally from fortified samples (SANTE/12682/2019). Good linearity was achieved in all cases with correlation coefficients better than 0.990. Good recovery values was achieved in the range studied. The accuracy of this method for all tested pesticide in the range of 70–120% and the values fulfilled the requirements of Document SANTE/12682/2019 (European Commission, 2019). Table S4 shows the results for RMSbias, RSD$_{\text{WR}}$, combined uncertainty (U_c), and expanded uncertainty (U_{exp}).

The removal rate of pesticides during storage at ambient temperature

The removal rates of 32 pesticide residues in detox water during ambient storage are presented in Fig. 2. Initial pesticide concentrations ranged from 0.04 to 0.06 mg kg^{-1}, which decreased to 0–0.03 mg/kg after 72 h of storage. After 24 h at ambient temperature, 13 pesticides (clothianidine, emamectin benzoate, pyridaben, pyridaphenthion, spinosad [D], thiacloprid, azoxystrobin, carbendazim, difenoconazole, fenamidone, imazalil, metalaxyl, triadimefon, linuron, clofentezine) exhibited removal rates exceeding 50%. Among these, azoxystrobin showed the highest reduction rate at 72%, while fenamidone had the lowest at 18%.

Removal rates increased substantially after 48 h, with all pesticides except tebuconazole exceeding a 50% reduction. At this point, kresoxim-methyl exhibited the highest removal rate (79%), whereas ametocradin displayed the lowest (55%). By the end of 72 h, reduction rates ranged between 42% (Tebuconazole) and 89% (Clofentezine). Notably, emamectin benzoate, pyridaben, triadimefon, and clofentezine achieved removal rates above 80%. These findings align with Karaca (2019), who reported significant reductions in azoxystrobin, carbendazim, and trifloxystrobin residues during cold storage of table grapes. Similarly, studies by Bian et al. (2018, 2021, 2024) observed a decline in malathion and diazinon residues during ambient storage of cucumber, cowpea, and celery. As storage time increased, the pesticide removal rates generally improved under ambient temperature conditions. As shown in Fig. 2, pesticide concentrations consistently decreased with rising temperatures. However, some pesticides did not exhibit a steady reduction. This irregularity may be attributed to the inherent lack of homogeneity in detox water, which is not a uniform liquid. Homogeneity is a critical factor for accurate pesticide analysis. Consequently, during storage, certain pesticides, such as ametocradin, may

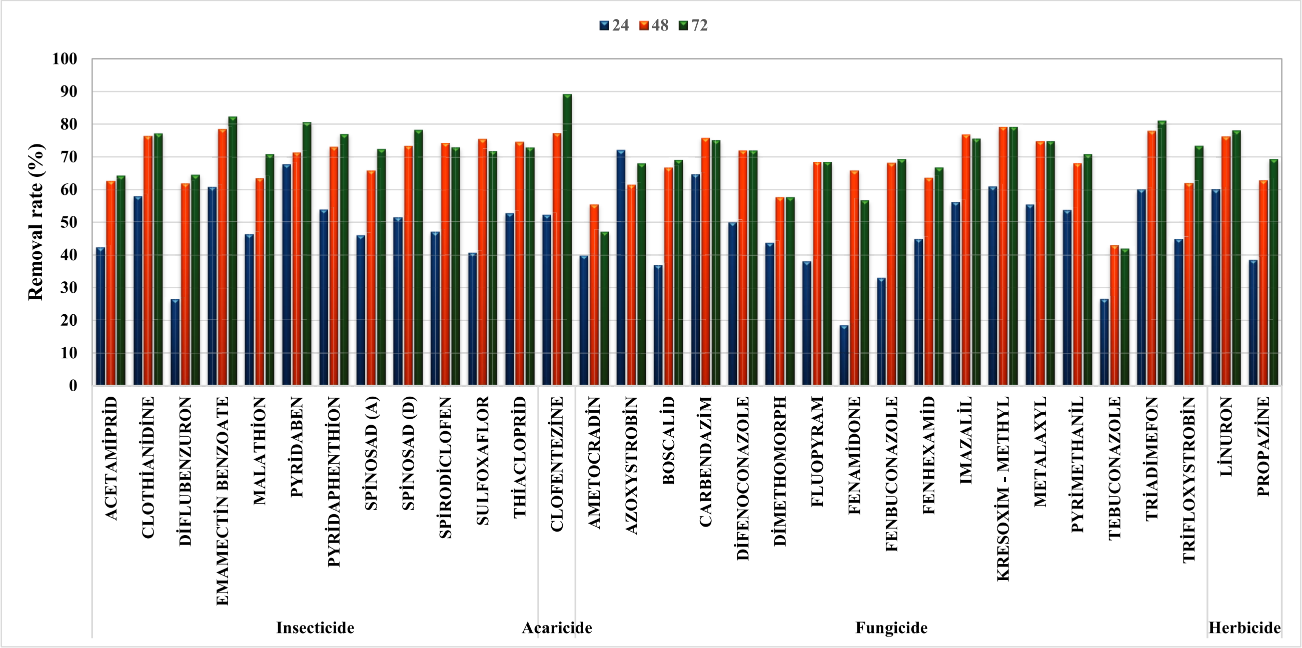


Fig. 2 Removal rates of 32 pesticides after storage at ambient temperature

not volatilize consistently, leading to fluctuations in their concentration over time.

To further analyze the behavior of pesticide residues, heatmaps were generated to illustrate trends over 24, 48, and 72 h under both ambient and cold storage conditions (Fig. 3).

The heatmaps provided a clear visualization of how storage time influenced pesticide removal. Vertical clustering analysis grouped storage durations into two main clusters: 24 h in one cluster, and 48 and 72 h in another. This indicates that storage time became a significant factor in pesticide removal after 24 h. For example, tebuconazole demonstrated limited removal throughout storage, forming a distinct group in the clustering analysis, while other pesticides showed greater reductions with extended storage. Horizontal clustering of pesticides further revealed two primary groups: tebuconazole as one group and the remaining pesticides as another. This analysis highlights the critical role of extended storage in enhancing the removal of pesticide residues from detox water, particularly for most pesticides except tebuconazole.

The removal rates of pesticides during storage at 4 °C

The reduction rates of 32 pesticide residues in detox water during cold storage are presented in Fig. 4. Initial pesticide concentrations ranged from 0.04 to 0.06 mg kg^{-1}. After 72 h of storage at 4 °C, pesticide residue levels ranged from 0.02 to 0.04 mg kg^{-1}. Overall, the reduction rates increased with storage duration. After 24 h, 15 pesticides (diflubenzuron, spinosad [A], spinosad [d], sulfoxaflor, ametoctradin, boscalid, difenoconazole, dimethomorph, fluopyram, fenamidone, fenbuconazole, fenhexamid, imazalil, propazine, and clofentezine) showed no reduction, indicating slower removal at 4 °C.

By 48 h, triadimefon exhibited the highest reduction rate at 61%, whereas ametoctradin had the lowest at 4%. At the end of 72 h, the removal rates exceeded 50% for only seven pesticides (emamectin benzoate, pyridaphenthion, carbendazim, kresoxim-methyl, metalaxyl, triadimefon, and linuron). Among these, kresoxim-methyl displayed the highest removal rate (70%), while ametoctradin again showed the lowest (6%). The specific reduction rates of key pesticides after 72 h at 4 °C were as follows: acetamiprid (38%), azoxystrobin (36%), boscalid (14%), malathion (40%), pyrimethanil (49%), and tebuconazole (44%). These results are consistent with previous studies. For instance, Bian et al. (2021) reported fluctuations in pesticide levels, including dichlorvos, diazinon, and malathion, in cowpeas stored at varying temperatures (– 20 °C, 4 °C, and ambient). Nevertheless, reductions were observed across all storage conditions, with 72-h cold storage resulting in similar pesticide reduction rates to those reported in our study. Our findings also align with Bilkova et al. (2022), who observed that Boscalid exhibited higher stability during low-temperature storage of sweet cherries (1.2–1.6 °C), while acetamiprid and tebuconazole demonstrated greater reductions compared to boscalid by the end of storage.

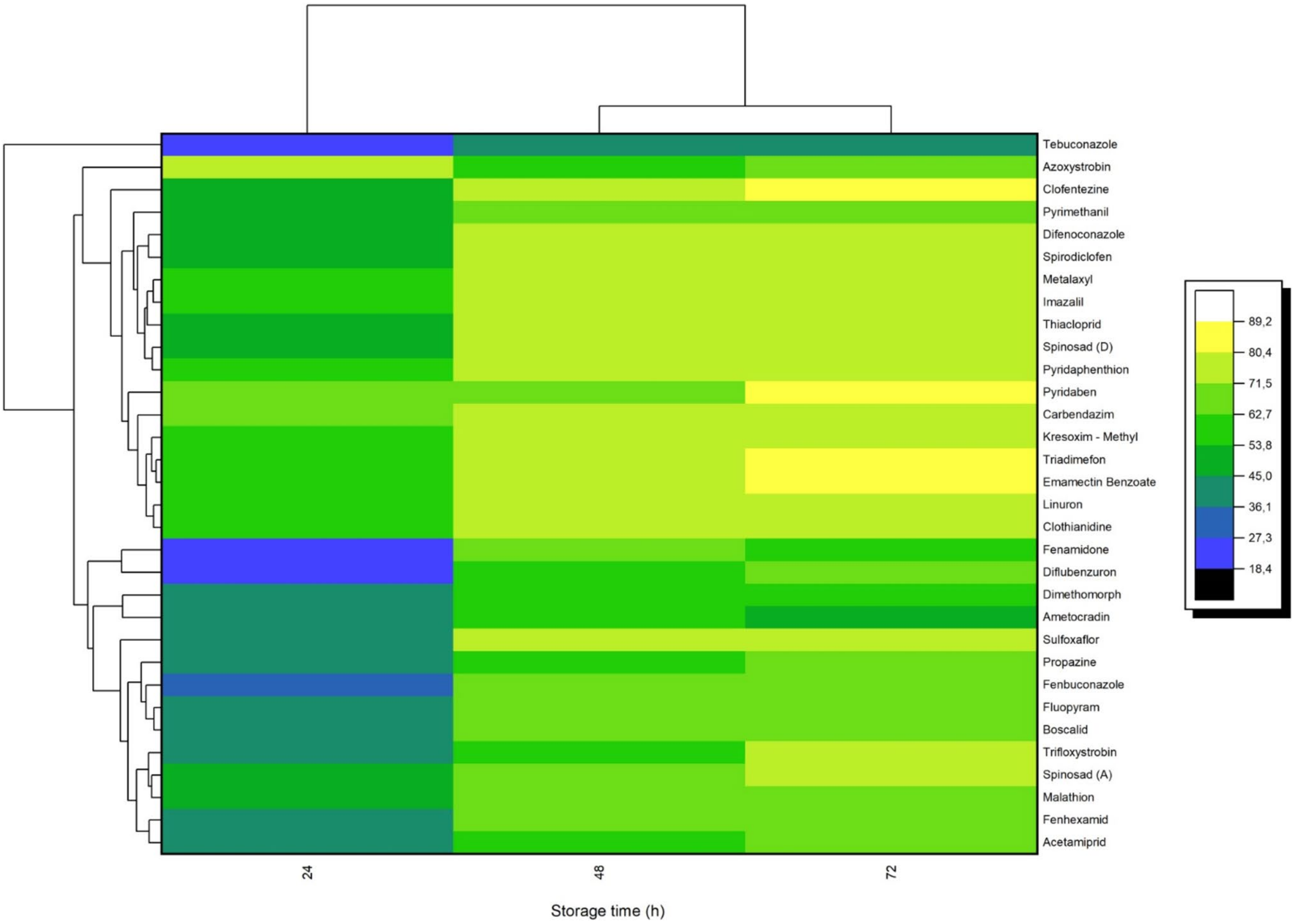


Fig. 3 Heatmap of the removal rates of pesticides at ambient temperature

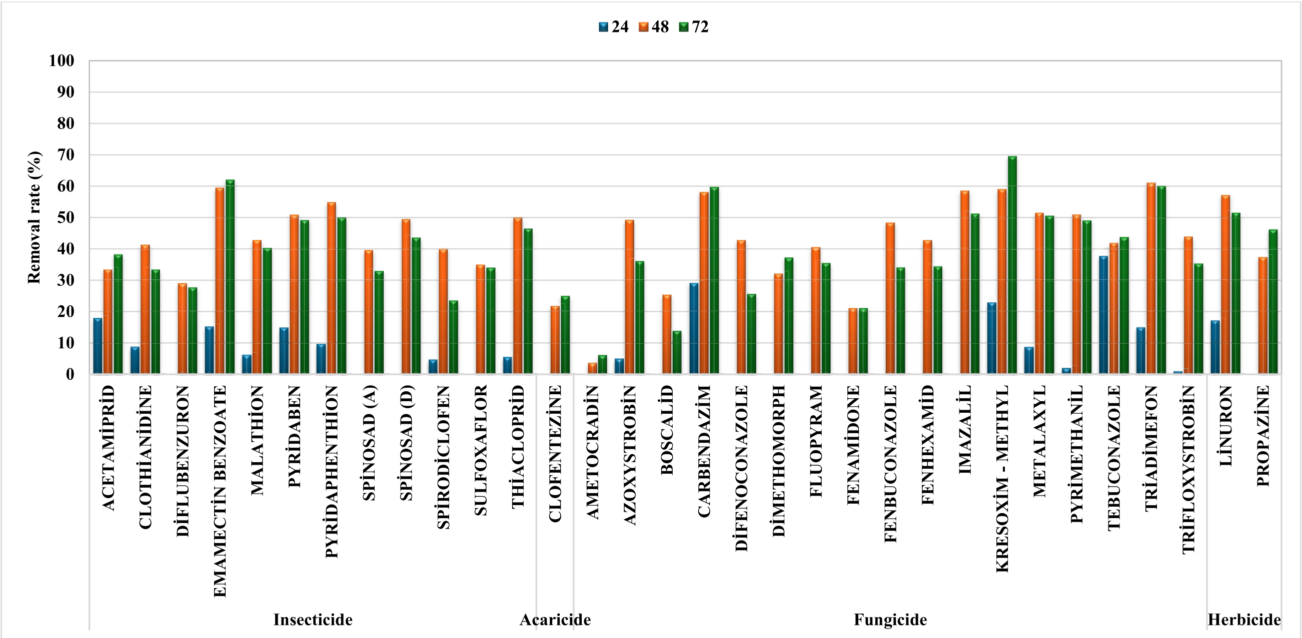


Fig. 4 Removal rates of 32 pesticides after storage at cold temperature

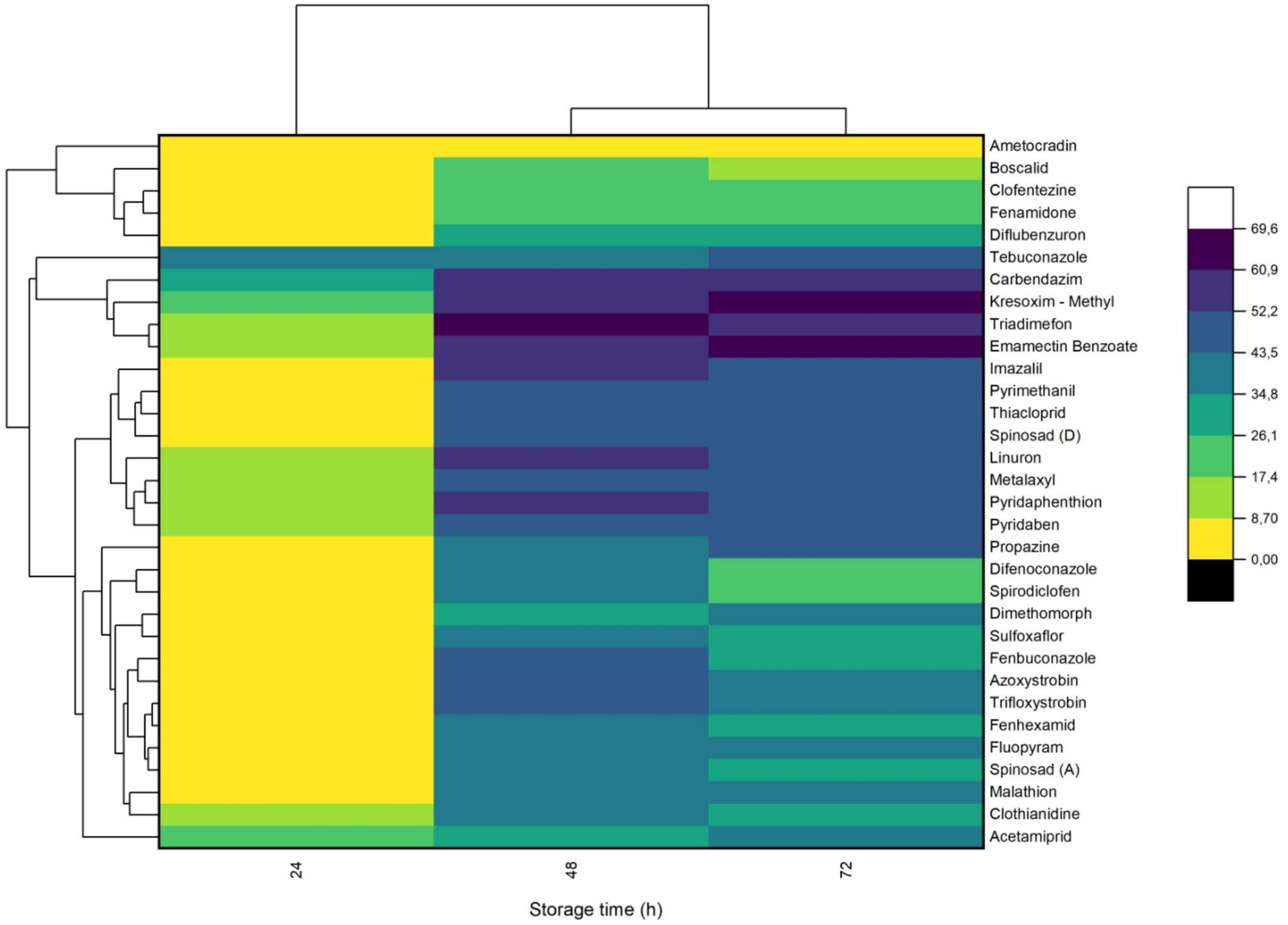


Fig. 5 Heatmap of the removal rates of pesticides at cold temperature

Clustering analysis (Fig. 5) highlighted distinct groupings for storage durations. Similar to ambient storage, two main clusters were identified: one for 24 h and another encompassing 48 and 72 h. Pesticide distribution patterns were also analyzed horizontally, revealing two clusters. The first included ametoctradin, boscalid, clofentezine, fenamidone, and diflubenzuron, while the second contained the remaining pesticides.

As shown in Fig. 6, the removal rates of pesticides increased progressively with storage time, confirming the influence of storage conditions on pesticide behavior. Notably, reductions in pesticide residues were greater under ambient storage compared to cold storage. These observations align with findings by Liang et al. (2012), who reported significant reductions in pesticide residues during ambient storage (25 °C) of cucumber compared to cold storage (4 °C). Similar trends were observed by Bian et al. (2021), who noted higher reductions in diazinon and malathion in cowpeas stored at ambient temperature compared to cold conditions. The differences in pesticide reduction rates between ambient and cold storage may be attributed to variations in enzymatic activity in homogenized detox water samples. Enzymes likely exhibit higher activity under ambient conditions, accelerating the degradation of pesticide residues, as suggested by Bian et al. (2021). These findings underscore the importance of storage conditions in determining pesticide residue levels in detox waters and provide a basis for optimizing preparation and storage practices to ensure consumer safety.

Conclusion

This study investigated the removal rates of 32 pesticide residues in detox water under ambient and cold storage conditions over 24, 48, and 72 h. Storage time and temperature were found to significantly influence pesticide degradation. Ambient storage conditions, due to higher temperatures, facilitated greater pesticide volatilization and degradation compared to cold storage. However, certain pesticides with low removal rates demonstrated strong

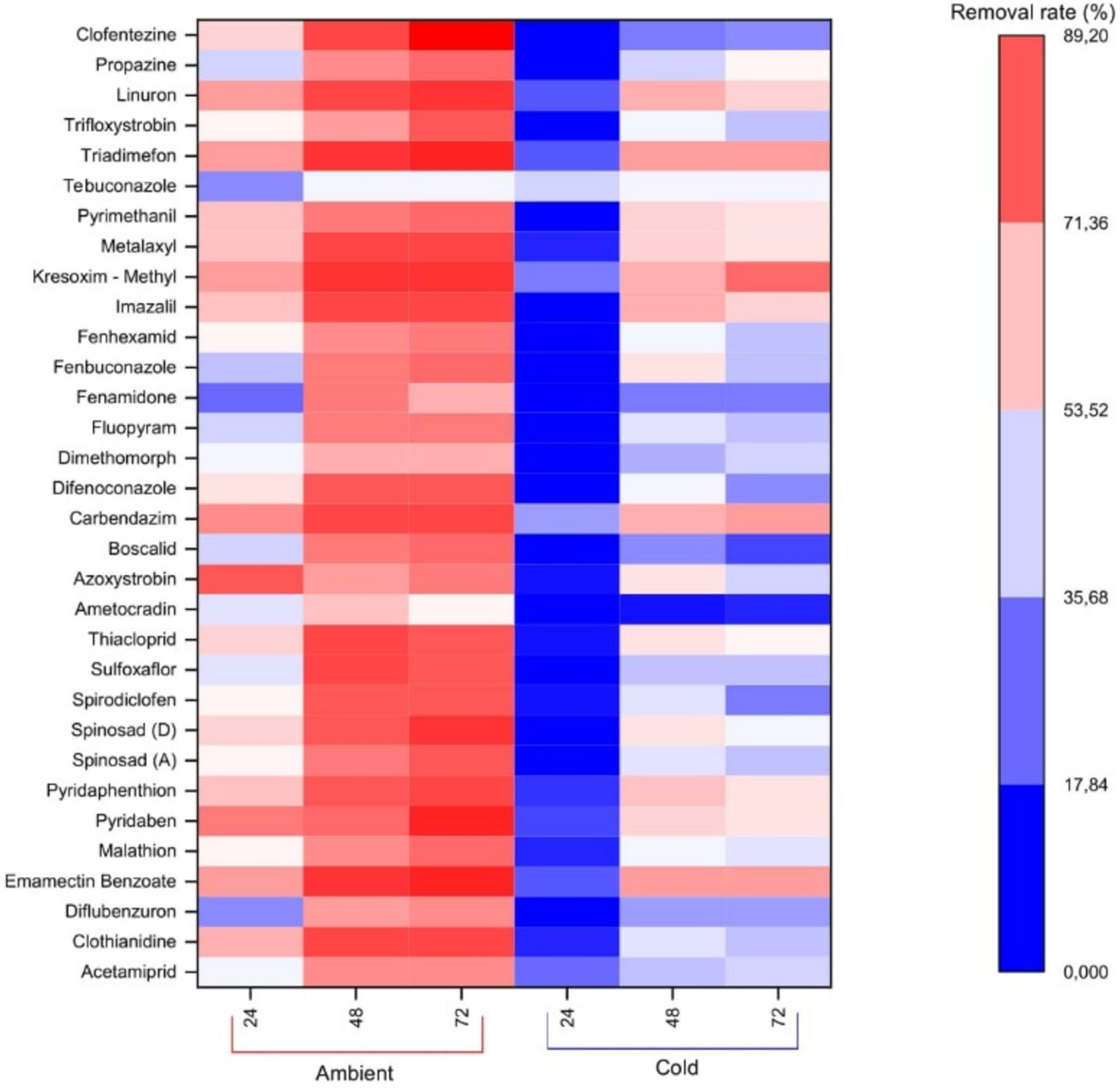


Fig. 6 Heatmap of the removal rates of pesticides in detox water after storage under ambient and cold storage conditions

resistance to degradation, underscoring their persistence even under favorable conditions for volatilization.

These findings provide valuable insights into the degradation behavior of pesticide residues in detox water and highlight the importance of storage conditions in mitigating pesticide contamination. By identifying persistent pesticides, this study contributes to the understanding of food safety risks associated with detox water preparation and storage. Future research should focus on exploring enzymatic and chemical factors influencing pesticide degradation in aqueous media and evaluating alternative methods to enhance pesticide removal for safer consumption.

Supplementary Information The online version contains supplementary material available at https://doi.org/10.1007/s11356-025-36842-1.

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Author contribution All authors contributed to the study conception and design. Material preparation, data collection, and analysis were performed by Saliha Gökduman. The first draft of the manuscript was written by Sezer Kıralan and all authors commented on previous versions of the manuscript. All authors read and approved of the final manuscript.

Data availability This is not applicable.

Declarations

Ethical approval This is not applicable.

Consent to participate This is not applicable.

Consent for publication The authors declare their satisfaction with the publication of the submitted work.

Competing interests The authors declare no competing interests.

References

Ariyawansa GP, Ramanathan R (2021) Antioxidative potential and phytochemical content of detox water. Advanced Journal of Graduate Research 10:41–50. https://doi.org/10.21467/ajgr.10.1.41-50

Aslantas S, Golge O, González-Curbelo MÁ, Kabak B (2023) Determination of 355 pesticides in lemon and lemon juice by LC-MS/MS and GC-MS/MS. Foods 12:1812. https://doi.org/10.3390/foods12091812

Bakırcı GT, Acay DBY, Bakırcı F, Ötleş S (2014) Pesticide residues in fruits and vegetables from the Aegean region, Turkey. Food Chem 160:379–392. https://doi.org/10.1016/j.foodchem.2014.02.051

Bian Y, Liu F, Chen F, Sun P (2018) Storage stability of three organophosphorus pesticides on cucumber samples for analysis. Food Chem 250:230–235. https://doi.org/10.1016/j.foodchem.2018.01.008

Bian Y, Wang B, Liu F, Wang Y, Huang H (2021) Effect of storage states on stability of three organophosphorus insecticide residues on cowpea samples. J Sci Food Agric. https://doi.org/10.1002/jsfa.11257

Bian Y, Yu Z, Zhang A, Qi X, Li C, Wang S, Liang L (2024) New insights to improve the storage stability of pesticide residues in analytical samples. Microchem J 199:110009. https://doi.org/10.1016/j.microc.2024.110009

Bilkova A, Knapova P, Suran P, Kwiecien J, Svec F, Sklenarova H (2022) Effect of storage conditions on content of pesticide residues in sweet cherries. Food Chem 13:100185. https://doi.org/10.1016/j.fochx.2021.100185

European Commission (2019) Guidance document on analytical quality control and method validation procedures for pesticide residues and analysis in food and feed. SANTE/12682/2019

Karaca H (2019) The effects of ozone-enriched storage atmosphere on pesticide residues and physicochemical properties of table grapes. Ozone Sci Eng 41:404–414. https://doi.org/10.1080/01919512.2018.1555449

Klein AV, Kiat H (2015) Detox diets for toxin elimination and weight management: a critical review of the evidence. J Hum Nutr Diet 28:675–686. https://doi.org/10.1111/jhn.1228

Kowalska G, Pankiewicz U, Kowalski R (2022) Assessment of pesticide content in apples and selected citrus fruits subjected to simple culinary processing. Appl Sci 12:1417. https://doi.org/10.3390/app12031417

Lehotay SJ (2007) Determination of pesticide residues in foods by acetonitrile extraction and partitioning with magnesium sulfate: collaborative study. J AOAC Int 90(2):485–520. https://doi.org/10.1093/jaoac/90.2.485

Liang Y, Wang W, Shen Y, Liu Y, Liu XJ (2012) Effects of home preparation on organophosphorus pesticide residues in raw cucumber. Food Chem 133:636–640. https://doi.org/10.1016/j.foodchem.2012.01.016

Philippe V, Neveen A, Marwa A, Basel AYA (2021) Occurrence of pesticide residues in fruits and vegetables for the Eastern Mediterranean Region and potential impact on public health. Food Control 119:107457. https://doi.org/10.1016/j.foodcont.2020.107457

Sabarwal A, Kumar K, Singh RP (2018) Hazardous effects of chemical pesticides on human health–cancer and other associated disorders. Environ Toxicol Pharmacol 63:103–114. https://doi.org/10.1016/j.etap.2018.08.018

Snyder M, Clum L (2014) The truth about beverages. In: Water infusions: refreshing, detoxifying and healthy recipes for your home infuser, 1st edn. Ulysses press, California, pp 17–23

Toptanci İ, Kiralan M, Ramadan MF (2021) Levels of pesticide residues in fruits and vegetables in the Turkish domestic markets. Environ Sci Pollut Res 28:39451–39457. https://doi.org/10.1007/s11356-021-13538-w

Valcke M, Bourgault MH, Rochette L, Normandin L, Samuel O, Belleville D, Phaneuf D (2017) Human health risk assessment on the consumption of fruits and vegetables containing residual pesticides: a cancer and non-cancer risk/benefit perspective. Environ Int 108:63–74. https://doi.org/10.1016/j.envint.2017.07.023

World Health Organization (2019) Guidelines for nutrition interventions. World Health Organization, Geneva

Zhang J, Sheng X, Cao J, Fang S, Liu X, Liu X, Weng R (2023) Occurrence and risk exposure assessment of multiple pesticide residues in edible mint in China. J Food Compos Anal 116:105071. https://doi.org/10.1016/j.jfca.2022.105071

Zhao H, Li R, Hu J (2023) Frequently used pesticides and their metabolites residues in apple and apple juice from markets across China: occurrence and health risk assessment. LWT 178:114610. https://doi.org/10.1016/j.lwt.2023.114610