



## Levels of organochlorine pesticides in the chewable and leftover parts of the khat (*Catha edulis*) sold in local markets of Jimma Town, Southwest Ethiopia

Fitsum Tamirat, Tsegaye Girma Asere, Bereket Tesfaye & Abera Gure

**To cite this article:** Fitsum Tamirat, Tsegaye Girma Asere, Bereket Tesfaye & Abera Gure (03 Oct 2024): Levels of organochlorine pesticides in the chewable and leftover parts of the khat (*Catha edulis*) sold in local markets of Jimma Town, Southwest Ethiopia, International Journal of Environmental Analytical Chemistry, DOI: [10.1080/03067319.2024.2407053](https://doi.org/10.1080/03067319.2024.2407053)

**To link to this article:** <https://doi.org/10.1080/03067319.2024.2407053>



Published online: 03 Oct 2024.



Submit your article to this journal [↗](#)



View related articles [↗](#)



View Crossmark data [↗](#)



# Levels of organochlorine pesticides in the chewable and leftover parts of the khat (*Catha edulis*) sold in local markets of Jimma Town, Southwest Ethiopia

Fitsum Tamirat , Tsegaye Girma Asere , Bereket Tesfaye and Abera Gure

Department of Chemistry, College of Natural Sciences, Jimma University, Jimma, Ethiopia

## ABSTRACT

This study aimed to determine the residual levels of organochlorine pesticides (OCPs) in chewable (tender leaves) and leftovers (mature leaves) of khat sold in local markets in Jimma town, southwest Ethiopia. Samples were collected from various local khat market places in Jimma town, based on their growing origin. A modified QuEChERS method was used to extract pesticides from the samples before their analysis by GC–MS. The findings indicated that p,p′-dichlorodiphenyldichloroethylene (p,p′-DDE) and p,p′-dichlorodiphenyltrichloroethane (p,p′-DDT) were detected in 88.3% and 76.6% of chewable samples, and in 86% and 69% of khat leftovers samples. The p,p′-DDE and p,p′-DDT levels found in chewable khat ranged from 10.12–45.39 and 8.53–34.15 µg/kg, respectively. While in leftover khat they ranged from 30.12–45.04 and 6.73–32.16 µg/kg, respectively. It was found that the concentration of p,p′-DDE is greater than its precursor compound p,p′-DDT for each sample, indicating past use of DDT in the study area. None of the detected analytes exceeded the maximum residue level (MRL) values established by the FAO/WHO or European Commission (EC) of 50 µg/kg. However, no information is available regarding these pesticides' collective and synergetic negative impacts on consumers.

## ARTICLE HISTORY

Received 15 July 2024

Accepted 16 September 2024

## KEYWORDS

QuEChERS; organochlorine pesticides; DDT; DDE; khat; GC–MS

## 1. Introduction

Pesticides are often used in agriculture to control harmful pests including insects, plant diseases, weeds, and other organisms that threaten to our food supply and health during various stages of food production, such as processing, storage, transport, and marketing [1,2]. Despite their benefits, pesticides can have negative effects on the environment and human health due to their toxic and persistent nature [3]. The global public health sectors are worried about pesticide residues in food and other natural resources because of their toxicity even at low concentrations [4].

Among the different classes of pesticides, organochlorine pesticides (OCPs) are known for their long half-life, persistence and high toxicity to humans and other animals [5]. The food chain allows OCPs to transfer and accumulate at higher trophic levels [6]. Due to their toxicity, persistence, and potential for biomagnification, OCPs like dichlorodiphenyltrichloroethane

(DDT) and hexachlorocyclohexane (HCH) have been banned from use in agriculture since the 1980s and the Stockholm Convention, has classified them as persistent organic pollutants (POPs) [7]. However, in several developing nations, such as Ethiopia, these pesticides have been used until recently in agriculture to control agricultural pests and combat malaria vectors [8]. The extensive use of OCPs can result to environmental contamination, bioaccumulation in food chains, and adverse health effects in humans and wildlife. Their persistence poses challenges for biodiversity and agricultural sustainability [9]. Literature indicates that Ethiopian khat cultivators use DDT to kill pests and keep the leaves shiny and attractive to customers [8]. Khat is an evergreen, blooming tree with mild narcotic properties, primarily cultivated in East Africa. Every day, five to ten million people chew khat leaves as a stimulant, mainly in the Middle East and Eastern Africa [10,11]. Its leftovers are used as a source of animal feed in Ethiopia [12–14].

Several studies have been conducted to determine the levels of OCP residues in khat cultivated in different regions of Ethiopia [15–17]. For instance, Mekonen et al. [8] reported high levels of OCP residues such as *p,p'*-DDE, *p,p'*-DDD, *o,p'*-DDT, and *p,p'*-DDT in chewable parts of khat samples collected from various districts of Jimma Zone, southwest Ethiopia. The samples from Dedo, Saka, Kersa, Mana, and Sokoru districts had the highest levels of DDT residue, with amounts of 149.00, 137.00, 103.00, 73.00 and 71.70 µg/kg, respectively. Samples collected from Gomma district had 41.20 µg/kg. The results indicated that the concentration of the detected OCPs in this district exceeded the maximum residue limits set by FAO/WHO and EC.

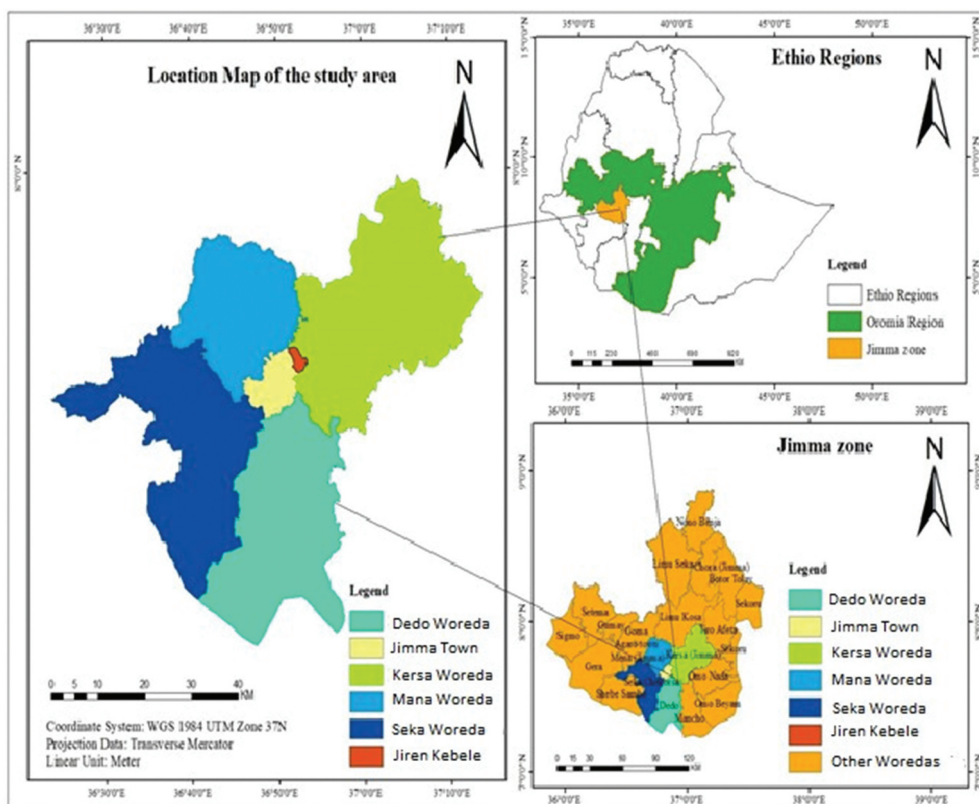
A study conducted on khat samples collected from Ezha and Enamor districts of Gurage zone, South Nations, Nationalities and Peoples Regional (SNNPR) State of Ethiopia also showed the presence of *p, p'*-DDT and its metabolites (*p, p'*-DDE and *p, p'*-DDD) in the samples [18]. The average concentrations of *p, p'*-DDT in khat samples from Ezha and Enamor districts were 5.77 and 4.77 µg/kg, respectively. The average concentrations of DDT metabolites, *p, p'*-DDD and *p, p'*-DDE, varied between the two areas. However, the levels of pesticides in the leftover khat have not yet been studied. In Ethiopia, khat leftovers are usually given to animals, like goats and sheep as feed. Additionally, the levels of OCP residues in khat samples grown in the present study area have not been recently investigated. Therefore, assessing the levels of OCP residue in both the chewed and discarded portions of khat samples from this area was crucial.

This study aimed to assess the residual levels of OCPs in both chewable and leftover parts of khat samples collected from local markets in Jimma town, southwest Ethiopia. A modified QuEChERS sample preparation technique was used and refined for extraction. The analytical sample preparation procedure was validated, and employed to monitor OCPs in khat samples. The presence of OCP residues in khat samples was identified and quantified using a GC-MS instrument.

## 2. Materials and method

### 2.1. Study area and sampling site

The study was conducted in Jimma town, located in southwestern Ethiopia. Samples were collected from local marketplaces in the town, based on their cultivars, or the districts from where they were supplied (Figure 1). Specifically, khat supplied from four districts



**Figure 1.** The map showing the districts that provided khat to Jimma town markets.

including Kersa, Seka, Dedo, and Mana districts of Jimma Zone, and Jiren Kdebele from Jimma town were considered in the study.

## 2.2. Sampling techniques

Before sampling, information was collected about the origins of the khat leaves that are typically sold and consumed at local khat marketplaces in Jimma town. The samples were purposefully selected from various local khat marketplaces in the town, including Mercator, Ajip, Koche, Central, Sekaber, and Areboch Tera. The chewable khat from these marketplaces came from four districts: Kersa, Seka, Dedo, and Mana of the Jimma zone as well as Jiren Kebele from Jimma town (Figure 1).

A total of 120 khat leaf samples were collected, consisting of 60 chewable (tender) leaves and 60 mature (leftover) leaves. Each khat sample, weighing about 300 to 350 g, was packed in polyethene plastic bags, labelled, and transported to the Analytical Chemistry Laboratory of Jimma University. Composite samples were then prepared based on their cultivar sources and stored in a refrigerator at 4°C until further analysis.

### 2.3. Chemicals and reagents

OCP standards, such as hexachlorocyclohexane ( $\alpha$ -HCH (99.5%),  $\beta$ -HCH (99.5%),  $\delta$ -HCH (99.5%)), Aldrin ( $\geq 98.8\%$ ),  $\gamma$ -Chlordane (98.8%),  $p,p'$ -DDE (99.99%),  $p,p'$ -DDT (98.9%), and methoxychlor (MC) (97.7%) were purchased from Sigma Aldrich (St. Louis, MO, USA). Analytical grade solvents: acetonitrile and hexane, and reagent grade salts: magnesium sulphate ( $\text{MgSO}_4$ ) and sodium chloride ( $\text{NaCl}$ ) were supplied by Loba Chemie Pvt. Ltd. (Mumbai, India). Florisil, 60–100 mesh, Merck, the residual grade was obtained from BDH Chemical Ltd. (Poole, England).

Stock standard solutions were prepared with concentrations of 1000 mg/L of  $\beta$ -HCH,  $\delta$ -HCH, and  $\alpha$ -HCH, 800 mg/L of aldrin as well as 400 mg/L of  $p,p'$ -DDE,  $p,p'$ -DDT,  $\gamma$ -chlordane, and MC in hexane. An intermediate standard solution with a concentration of 20 mg/L of each OCP was then prepared in hexane. Working solutions were daily prepared by diluting to the necessary concentrations. The solutions were stored below 4°C when not in use for analysis.

### 2.4. Instrument and apparatus

An Agilent 8890 gas chromatography (GC) coupled with an Agilent 5977B single quadrupole mass detector (Q-MS) and an Agilent G4513A autosampler (Agilent Technologies, USA) was used for analysis of OCPs. An HP-5 MS capillary column coated with 5% diphenyl 95% dimethylsiloxane (30 m, 0.25 mm i. d. and 0.25  $\mu\text{m}$  film thickness) supplied by (Agilent Technologies, USA) was used for separation of the analytes. The centrifuges, model D-6072 from KARL KOLB (Germany) and PLC02 in (Taiwan), and a vortex mixer, model an FB15024 from Fisher Scientific, Belgium were used during the experimental activities.

### 2.5. Operating condition of GC – MS

The targeted OCPs were analysed by GC-MS, following the operating conditions reported in the literature [19,20]. Briefly, helium gas (99.9999%) was used as the carrier gas with a flow rate of 1 mL/min. The sample, 1  $\mu\text{L}$ , was injected in splitless mode, with the injector port temperature set at 280°C. The GC oven temperature was programmed to start at 100°C, ramped at 15 °C/min to 200°C (kept constant for 5 min), then ramped at 4 °C/min to 250°C (held for 2 min), and finally ramped at 10 °C/min to 270°C (held for 10 min).

The MS operated in electron ionisation mode with an ionisation energy of 70 eV. The GC-MS transfer line, ion source, and quadrupole temperatures were set at 250°C, 230°C and 150°C, respectively. The MS was scanned from  $m/z$  45 to 500 at 150 s/scan; with a 3 min solvent delay. Selected ion monitoring (SIM) mode was used with two qualifier ions and one quantitative ion. Quantitative ion, qualifier ions, and retention time of the studied pesticides are presented in Table 1.

### 2.6. QuEChERS method

A modified QuEChERS method, as reported by Collimore and Bent [21] was utilised with slight modifications for the extraction and preconcentration of the targeted OCPs from the samples. A homogenised fresh sample, 10 g, was placed into a 50-mL Falcon tube. After adding 15 mL of

**Table 1.** Lists of OCPs molar mass, quantitative ion, qualifier ions and retention time the ions.

Analytes	Molar mass (g/mol)	Quantitative ion (m/z)	Qualifier ion (m/z)	Retention time (min)
$\alpha$ -HCH	290.83	219	189, 109	7.8
$\beta$ -HCH	290.83	219	189, 109	8.5
$\delta$ -HCH	290.83	219	189, 109	10.08
Aldrin	364.9	263	66, 293	12.44
Methoxchlor	345.6	240	227, 274	24.88
$\gamma$ -Chlordane	409.8	272	65, 373	15.032
<i>p, p'</i> -DDE	318.2	246	176, 318	17.38
<i>p, p'</i> -DDT	354.49	212	165, 235	19.25

acetonitrile, the mixture was vortexed for approximately a minute. Subsequently, 4 g of  $\text{MgSO}_4$  and 1 g of NaCl were added, followed by centrifugation at 5000 rpm for 5 minutes. Then, 7 mL of the supernatant was transferred into a 15-mL Falcon tube containing 250 mg of Florisil and 750 mg of anhydrous  $\text{MgSO}_4$ . The contents were vortexed for a minute and then centrifuged for 5 minutes at 5000 rpm. Next, 4 mL of the n-hexane layer were withdrawn and dried using a rotary evaporator. Finally, the dried extract was redissolved in 4 mL of hexane, from which 1 mL was transferred to a GC-autosampler vial for injection of 1  $\mu\text{L}$  of the sample into the GC-MS instrument.

## 2.7. Method validation

External calibration curves were constructed ranging from 5 to 50  $\mu\text{g}/\text{kg}$  to quantify pesticide residues in khat samples. All calibration curves had coefficient determinations ( $R^2$ ) of 0.995 or higher. The limits of detection and quantitation (LOD and LOQ) of each analyte were determined as 3 and 10 times the signal-to-noise ratio, respectively.

The precision of the analytical method was evaluated in terms of intra-day and inter-day precisions. For the intra-day precision study, six samples spiked with a mixture of OCPs at a concentration of 10  $\mu\text{g}/\text{kg}$  of each analyte were extracted on the same day. Similarly, for the inter-day precision study, spiked samples were extracted on different days and analysed by GC-MS. A recovery study was conducted to evaluate the extent of analytes loss, particularly during extraction, by spiking the samples with a 10  $\mu\text{g}/\text{kg}$  mixed standard solution.

## 2.8. Statistical analysis

The results were presented as the average of replicate measurements along with their standard deviations. One-way ANOVA was used to compare the levels of OCPs in khat samples collected from different locations, with a significance level of  $p < 0.05$ . The Tukey HSD comparison test was used to compare the variations in the total DDT concentration among different khat leftover khat samples. IBM SPSS Statistics, version 26 was used for the statistical analysis of the data.

# 3. Results and discussion

## 3.1. Method validation

Table 2 shows the analytical performance characteristics of the method used to analyse OCP residues in both the chewable and leftover parts of the khat samples. As can be seen,

**Table 2.** Method validation results for khat residue analysis: regression coefficient, linear dynamic range (LDR), LOD and LOQ, intra-day and inter-day precision RSD (%), and recovery (%).

Precision (RSD), (%) <i>n</i> = 6									
Pesticides	R <sup>2</sup>	LDR (µg/kg)	LOD (µg/kg)	LOQ (µg/kg)	Intra-day		Inter-day		Recovery (%)
					Chewable	Leftover	Chewable	Leftover	
α-HCH	0.996	16–50	4.8	16	8.987	7.722	8.568	7.132	83.2
β-HCH	0.996	12–50	3.6	12	7.837	9.142	4.036	8.717	86.3
δ-HCH	0.997	6–50	1.8	6	6.748	4.058	2.809	4.104	81.2
Aldrin	0.997	16–50	4.8	16	9.015	8.785	7.264	9.904	90.4
Methoxychlor	0.997	8–50	2.4	8	8.695	1.589	8.291	5.648	93.3
γ-Chlordane	0.997	7–50	2.2	7	5.376	2.140	5.126	9.326	103.3
<i>p</i> , <i>p'</i> -DDE	0.997	14–50	4.2	14	1.027	2.716	3.455	7.450	101.8
<i>p</i> , <i>p'</i> -DDT	0.995	10.3–50	3.1	10.3	6.182	1.018	6.691	4.506	82.0

wide linear ranges, precision, and accuracy (recoveries) studies were performed after the QuEChERS step. For this purpose, spiked khat samples were analysed at the level of 10 µg/kg. Table 2 displays the results of the validation parameters.

As per the guidelines set by the European Commission, the percentage of recovery must fall within the range of 70% to 120% with a relative standard deviation (RSD) of no more than 20% [22]. The study found that the recoveries of the OCPs ranged from 81.2% for δ-HCH to 103.3% for γ-Chlordane (Table 2), indicating that the method is suitable for determining the eight OCPs in both chewable and leftover khat samples. Figure 2 displays representative GC-MS chromatograms for the standard, unspiked and spiked khat samples. The limits of detection (LODs) ranged from 1.8 to 4.8 µg/kg while the limits of quantification (LOQs) ranged from 6 to 16 µg/kg. Hence, the utilised technique proved to be trustworthy in identifying and measuring OCP residues in the khat samples.

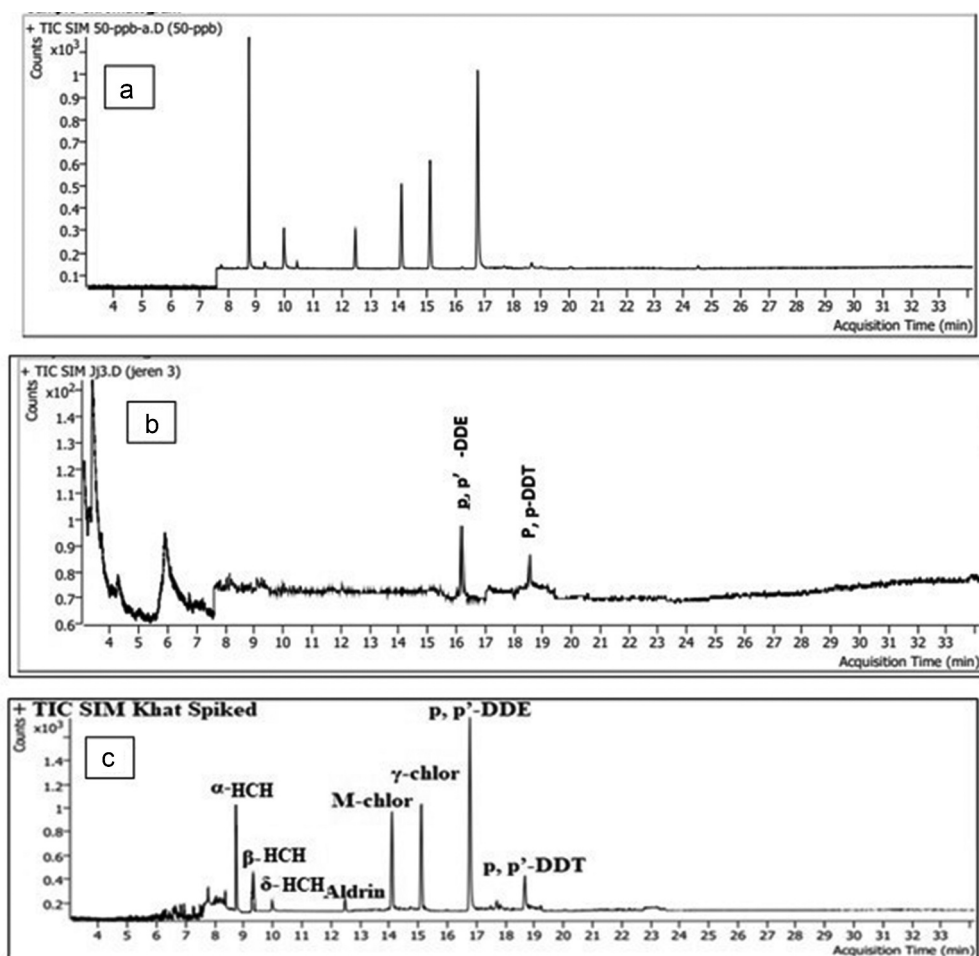
Standard solutions of the OCP mixture produced in pure solvent were used to plot the calibration curves [23]. Examining the y-intercept of the linear regression line for the response versus concentration and the correlation coefficient (R<sup>2</sup>) are common ways to assess the acceptability of linear data. In general, a correlation coefficient that falls between 0.995 and 0.999 is regarded as accepted. The y-intercept should be lower than a few percent of the response achieved at the target level [24]. As Table 1 shows, all of the pesticide calibration curves exhibited good linearity and correlation coefficients (R<sup>2</sup> > 0.99).

The method's precision, both intra- and inter-day, was examined by adding a mixture of 10 µg/kg OCPs into the blank sample. Six [6] spiked samples were conducted during a single day of extraction to assess the method's reproducibility (intra-day). Table 1 indicates that the RSD value of all pesticides was less than 10%. The method's repeatability (inter-day) was examined by analysing six [6] spiked samples on different extraction days. Additionally, the RSD value was less than 10%. This suggests that the employed method being used is fairly reproducible and repeatable [25,26].

### 3.2. Concentration of OCP residues in chewable khat leaves

In all the studied chewable khat leaves *p*, *p'*-DDT and *p*, *p'*-DDE were detected at varying concentration levels (Table 3). However, residues of other target analytes were not detected. This may indicate that they are either not present at all in the samples, or present at concentration levels below the detection limits of the method.





**Figure 2.** Representative GC–MS chromatograms for pesticide standards (a); unspiked (b); and spiked khat (c) samples.

**Table 3.** Pesticide residues (mean  $\pm$  SD) detected in the chewable khat leaf samples.

Sample	OCPs concentration ( $\mu\text{g/kg}$ )	
	<i>p, p'</i> -DDT	<i>p, p'</i> -DDE
Dedo	24.14 $\pm$ 0.05 <sup>a</sup>	35.03 $\pm$ 0.05 <sup>c,d</sup>
Seka	12.49 $\pm$ 0.54 <sup>b</sup>	40.15 $\pm$ 0.00 <sup>d</sup>
Beda Buna (Kersa)	11.17 $\pm$ 0.02 <sup>b</sup>	34.07 $\pm$ 0.06 <sup>c</sup>
Jiren	8.53 $\pm$ 0.36 <sup>b</sup>	10.12 $\pm$ 0.0 <sup>b</sup>
Buture (Mana)	34.15 $\pm$ 0.03 <sup>c</sup>	45.39 $\pm$ 0.09 <sup>e</sup>

Different letters represent significant differences at  $p < 0.05$ .

The results of the one-way ANOVA analysis show a significant variation ( $p < 0.01$ ) in the levels of *p, p'*-DDT, and *p, p'*-DDE among the sources of khat leaf samples (the five districts in the Jimma zone). The Tukey HSD comparison tests indicated a statistically significant



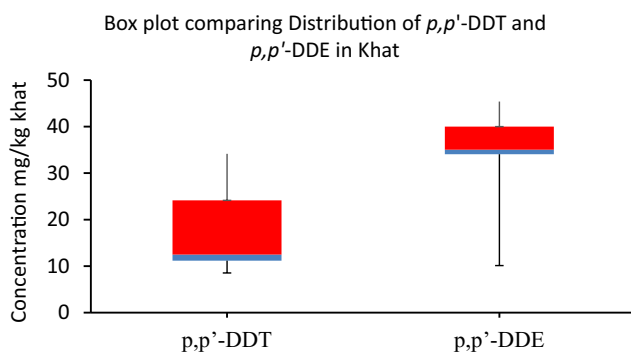
difference in  $p,p'$ -DDT levels between Dedo and Buture (Mana) compared to the other samples, with a  $p$ -value  $<0.01$ . However, there was no significant difference in the mean concentrations of  $p,p'$ -DDT from Seka, Kersa, and Jiren ( $p \geq 0.181$ ). The highest concentration of  $p,p'$ -DDT was detected in Buture (Mana) at  $34.15 \mu\text{g/kg}$ , followed by Dedo at  $24.14 \mu\text{g/kg}$ , Seka at  $12.49 \mu\text{g/kg}$ , Beda Buna (Kersa) at  $11.17 \mu\text{g/kg}$ , and Jiren with  $8.53 \mu\text{g/kg}$ .

The contamination of khat with  $p,p'$ -DDT in the Jimma Zone probably resulted from the past use of  $p,p'$ -DDT for public health purposes, especially in controlling disease vectors. It may have also occurred due to the disposal of obsolete pesticides containing organo-chlorine chemicals like  $p,p'$ -DDT [8,27]. This can result in the pollution of environmental compartment including agricultural products such as khat.

The levels of  $p,p'$ -DDE in the khat samples showed significant variation ( $p < 0.01$ ) except in samples collected from Dedo. The highest concentration was detected in Buture (Mana) at  $45.39 \mu\text{g/kg}$ , followed by Seka at  $40.15 \mu\text{g/kg}$ , Dedo at  $35.03 \mu\text{g/kg}$ , Beda Buna at  $34.07 \mu\text{g/kg}$ , and Jiren at  $10.12 \mu\text{g/kg}$ . The reason why the Jiren sample had the lowest concentration of  $p,p'$ -DDE ( $10.12 \mu\text{g/kg}$ ) compared to the others could be due to lower contamination of  $p,p'$ -DDT in samples from that area.

The detected concentrations of  $p,p'$ -DDT and  $p,p'$ -DDE in the present findings as well as the Mekonen et al. [8] earlier report show notable trends in pesticide pollution levels in the samples. The findings demonstrate a decrease in  $p,p'$ -DDT levels for Seka ( $12.49 \mu\text{g/kg}$ ) and Beda Buna (Kersa) ( $11.17 \mu\text{g/kg}$ ) compared to the earlier study's results ( $18.83 \mu\text{g/kg}$  and  $16.71 \mu\text{g/kg}$ , respectively). Results from Dedo indicated a minor drop from  $25.97 \mu\text{g/kg}$  to  $24.14 \mu\text{g/kg}$ , whereas Buture (Mana) samples displayed a notable rise from  $10.31 \mu\text{g/kg}$  to  $34.15 \mu\text{g/kg}$ . On the contrary, there has been a significant decrease in the levels of  $p,p'$ -DDE in the samples from various locations, with Dedo dropping from  $113.65 \mu\text{g/kg}$  to  $35.03 \mu\text{g/kg}$ , Seka from  $64.13 \mu\text{g/kg}$  to  $40.15 \mu\text{g/kg}$ , Beda Buna (Kersa) from  $68.94 \mu\text{g/kg}$  to  $34.07 \mu\text{g/kg}$ , and Buture (Mana) from  $60.12 \mu\text{g/kg}$  to  $45.39 \mu\text{g/kg}$ .

Similarly, the box-whisker plot in post hoc tests (Figure 3) showed a significant variation in the distribution of  $p,p'$ -DDT and  $p,p'$ -DDE in the chewable khat samples ( $p < 0.05$ ). The khat samples exhibited a higher prevalence of the metabolite  $p,p'$ -DDE. The enhanced spread of  $p,p'$ -DDE in plant matrices arises from a combination of uptake mechanisms, chemical durability, and environmental factors.



**Figure 3.** Distribution of the concentration of  $p,p'$ -DDT and  $p,p'$ -DDE in khat samples.

Huang et al. [28] stated that the DDE/DDT or DDT/DDE ratio is used to differentiate between pesticide residues from past or current applications. The current study shows that the amount of *p, p'*-DDT was lower than the amount of its main metabolite *p, p'*-DDE, as demonstrated in Figure 3. This aligns with the results from Mekonen et al. [8] and indicates a decrease in *p, p'*-DDT usage on khat crops in the area under investigation. Moreover, multiple studies documented the presence of *p, p'*-DDE and *p, p'*-DDD in environmental samples, suggesting that *p, p'*-DDT could have been utilised previously in that area [29–31]. Over time, the original *p, p'*-DDT transforms into *p, p'*-DDE and *p, p'*-DDD, but there has been no recent use of *p, p'*-DDT in that area. Hence, the past utilisation of DDT for public health purposes, like controlling disease vectors, could have played a role in the contamination of khat in the Jimma zone.

### 3.3. Concentrations of OCP residues in the leftover khat samples

Residue concentrations of eight OCPs were measured in leftover samples of khat collected from the Mana, Kersa, and Seka districts in the Jimma zone of southwest Ethiopia. Samples from Dedo and Jiren were excluded due to insufficient presence of mature leaves. The findings are presented in Table 4. Similar to the result observed in the analysis of OCPs in chewable khat, most of the pesticides analysed, such as  $\alpha$ -HCH,  $\beta$ -HCH,  $\delta$ -HCH, aldrin, methoxychlor, and  $\gamma$ -chlordane, were not detected in the samples. However, the predominant contaminant *p, p'*-DDT and its metabolite, *p, p'*-DDE were found in all the samples. The widespread detection of *p, p'*-DDT and *p, p'*-DDE is likely due to their chemical properties, historical usage patterns, and environmental persistence compared to other organochlorine pesticides.

One-way ANOVA analysis indicated significant variations in *p, p'*-DDT among the khat leftover samples ( $p < 0.01$ ). The sample from Mana (Buture) khat had the highest concentration of *p, p'*-DDT, while Seka had a notably higher amount than Kersa (Beda Buna). On the other hand, Seka's khat leftover samples exhibited the highest concentration of *p, p'*-DDE at 45.04  $\mu\text{g/kg}$ , followed by Mana at 35.06  $\mu\text{g/kg}$ , and Kersa at 30.12  $\mu\text{g/kg}$ . The findings revealed significant differences in the concentrations of both *p, p'*-DDT and *p, p'*-DDE across the sampled locations. These results highlight the need for targeted monitoring in these areas to address OCP contamination and protect environmental and public health.

The study's chewable and leftover khat samples had average concentrations of *p, p'*-DDT and *p, p'*-DDE that were below the FAO/WHO MRLs of 100–200  $\mu\text{g/kg}$  for different agricultural food items (FAO/WHO Food Standards Codex Alimentarius) [32] or an EC MRL value of 50  $\mu\text{g/kg}$  [33]. However, MRLs do not represent safety level;

**Table 4.** Residual concentrations (mean  $\pm$  SD) of OCPs detected in leftover khat samples.

Sample	OCPs concentration ( $\mu\text{g/kg}$ )	
	<i>p, p'</i> - DDT	<i>p, p'</i> - DDE
Mana (Buture)	32.16 $\pm$ 0.06 <sup>c,d</sup>	35.06 $\pm$ 0.07 <sup>c</sup>
Kersa (Beda Buna)	6.73 $\pm$ 0.01 <sup>a</sup>	30.12 $\pm$ 0.06 <sup>d</sup>
Seka	22.21 $\pm$ 0.04 <sup>b</sup>	45.04 $\pm$ 0.09 <sup>e</sup>

Different letters represent significant differences at  $p < 0.05$ .

rather, they highlight legal concerns around the use of pesticides, such as the unlawful use of out-of-date or prohibited pesticides, the use of substandard formulations, or contamination from a variety of sources, including those that are meant to safeguard the public's health [34].

In general, finding trace amounts of  $p,p'$ -DDT and its metabolite,  $p,p'$ -DDE in khat leftovers that are used to feed goats and sheep raises significant concerns. Table 4 shows that the concentration of  $p,p'$ -DDT was notably less than its primary metabolite,  $p,p'$ -DDE. This indicates that although  $p,p'$ -DDT is not being used presently, there is proof of past use in the area, potentially impacting the environment and agriculture. Because OPCs are not easily metabolised or eliminated,  $p,p'$ -DDT and  $p,p'$ -DDE can accumulate in the bodies of animals such as goats and sheep, possibly increasing in concentration as they are passed along the food chain.

## 4. Conclusion

The present study investigated the concentrations of eight OCPs in chewable and leftover khat samples obtained from Jimma town's market in southwest Ethiopia. The OCPs were extracted from the samples using a modified QuEChERS technique, and then analysed by using GC-MS. The method's high linearity ( $R^2 \geq 0.995$ ), acceptable percentage recovery (70–120%), and low limits of detection and quantification revealed its suitability for analysing OCPs in khat samples. Examination of the eight targeted OCPs in khat samples demonstrated that  $p,p'$ -DDT and  $p,p'$ -DDE were present in every sample, suggesting a higher prevalence of usage in the region where the khat was cultivated. Most chewable khat samples contained higher levels of  $p,p'$ -DDT and its metabolite ( $p,p'$ -DDE) pesticide residues when compared to leftover khat samples. Moreover, a significant level of  $p,p'$ -DDE was found in the khat samples compared to the precursor compound ( $p,p'$ -DDT), suggesting the previous usage of DDT in the specific area. Despite the levels of OCPs being below the CA and EU guidelines, there may still be potential health hazards for both consumers and animals. Hence, it is crucial to perform a health risk assessment on the identified OCP residues in khat samples from this study.

## Acknowledgments

The author wishes to thank the Chemistry Department at Jimma University for giving access to laboratory resources for the research.

## Disclosure statement

No potential conflict of interest was reported by the author(s).

## ORCID

Fitsum Tamirat  <http://orcid.org/0009-0002-6301-2300>

Tsegaye Girma Asere  <http://orcid.org/0000-0003-0126-9098>

Bereket Tesfaye  <http://orcid.org/0000-0002-7119-9497>

Abera Gure  <http://orcid.org/0000-0002-2777-3500>

## References

- [1] J. Beddington, *Philos. Trans. R. Soc. B. Biol. Sci.* **365** (1537), 61–71 (2010). doi:10.1098/rstb.2009.0201
- [2] C.A. Damalas and I.G. Eleftherohorinos, *Int. J. Environ. Res. Public Health* **8** (5), 1402–1419 (2011). doi:10.3390/ijerph8051402
- [3] C.O. Ogah and H.B. Coker, *J. Appl. Pharm. Sci.* **2** (9), 93–97 (2012). doi:10.7324/JAPS.2012.2919
- [4] T.S. Kathpal and K. Beena, *Environ. Monit. Assess.* **151** (1), 19–26 (2009). doi:10.1007/s10661-008-0210-0
- [5] B.O. Botwe, *J. Environ. Issues Agric. Dev. Ctries.* **3** (2), 1–6 (2007).
- [6] Y.B. Man, J.K.Y. Chan, S.C. Wu, C.K.C. Wong and M.H. Wong, *Sci. Total Environ.* **463**, 264–273 (2013). doi:10.1016/j.scitotenv.2013.06.011
- [7] K. Feng, B.Y. Yu, D.M. Ge, M.H. Wong, X.C. Wang and Z.H. Cao, *Chemosphere* **50** (6), 683–687 (2003). doi:10.1016/S0045-6535(02)00204-7
- [8] S. Mekonen, A. Ambelu, B. Negassa and P. Spanoghe, *Regul. Toxicol. Pharmacol.* **87**, 64–70 (2017). doi:10.1016/j.yrtph.2017.05.008
- [9] H. Nakata, M. Kawazoe, K. Arizono, S. Abe, T. Kitano, H. Shimada, W. Li and X. Ding, *Arch. Environ. Contam. Toxicol.* **43** (4), 473–480 (2002). doi:10.1007/s00244-002-1254-8
- [10] S. Geografiska, A. Series, H. Geography, G. Dessie and P. Kinlund, *Anthropol. Geogr.* **90** (2), 187–203 (2008). <https://www.jstor.org/stable/4>
- [11] M. Fj and D. Cg, *Mayo Clin. Proc.* **85** (11), 971–973 (2010). doi:10.4065/mcp.2010.0658
- [12] Y. Mekasha, A. Tegegne and H. Rodriguez-Martinez, *Reprod. Domest. Anim.* **43** (4), 437–444 (2008). doi:10.1111/j.1439-0531.2007.00931.x
- [13] Y. Getinet and M. Yoseph, *J. Anim. Plant. Sci.* **24** (1), 35–42 (2014).
- [14] Z. Woldu, D. Belew and T. Benti, *J. Agric. Sci. Technol. B* **5** (3), 149–169 (2015). doi:10.17265/2161-6264/2015.03B.001
- [15] S.A. Atnafie, N.Y. Muluneh, K.A. Getahun, A.T. Woredikal and W. Kahaliw, *J. Environ. Public Health.* **2021**, 1–8 (2021). doi:10.1155/2021/4680573
- [16] T. Hailemariam, T. Belete, T. Hailu, D. Ayele and S. Ligani, *J. Anal. Bioanal. Tech.* **09** (1), (2018). doi:10.4172/2155-9872.1000395
- [17] G. Regassa and C. Regassa, *Afr. J. Environ. Sci. Technol.* **15** (January), 16–26 (2021). doi:10.5897/AJEST2020.2916
- [18] Hailemariam T., Belete, T., Hailu, T., Ayele, D., *Analytical & Bioanalytical Techniques Analytical & Bioanalytical Techniques Quantification of Organochlorine Pesticide Residues in Chewable Part of Chata Edulus in Gurage Zone*, (SNNPR, Ethiopia, October 2018).
- [19] B. Tesfaye, A. Gure, T.G. Asere and G.J. Molole, *Microchem. J.* [Internet]. **195** (July), 109428. Available from: (2023). doi:10.1016/j.microc.2023.109428
- [20] K.A. Ago, S.A. Kitte, G. Chirfa and A. Gure, *Heliyon* **9** (1), e12954 (2023). doi:10.1016/j.heliyon.2023.e12954
- [21] W.A. Collimore and G.A. Bent, *Environ. Monit. Assess.* **192** (2), (2020). doi:10.1007/s10661-020-8072-1
- [22] P.R.J. Fao, W.H.O. Food, S. Programme, C. Alimentarius, S. Cicg R.O.F. The, (24–29 April 2017).
- [23] D.M. Bliesner, *Validating Chromatographic Methods: A Practical Guide*, (John Wiley & Sons, Hoboken, NJ, USA, 2006).
- [24] A.B. Gobo, M.H.S. Kurz, I.R. Pizzutti, M.B. Adaime and R. Zanella, *J. Braz. Chem. Soc.* **15** (6), 945–950 (2004). doi:10.1590/S0103-50532004000600024
- [25] Sanco. No. SANCO/12495/2011. Doc No SANCO/12495/2011. 41 (2011).
- [26] European Commission. SANCO/3103/2000. *Quality Control Procedures for Pesticide Residues Analysis. Guidelines for Residues Monitoring in the European Union*, (2000), pp. 15–17.
- [27] D. Daba, A. Hymete, A.A. Bekhit, A.M.I. Mohamed and A.E.-D.A. Bekhit, *Bull. Environ. Contam. Toxicol.* **86** (3), 336–341 (2011). doi:10.1007/s00128-011-0207-1
- [28] Z. Huang, Y. Li, B. Chen and S. Yao, *J. Chromatogr. B* **853** (1–2), 154–162 (2007). doi:10.1016/j.jchromb.2007.03.013
- [29] EFSA. *Efsa. J.* **8** (7), 1646 (2010).

- [30] O.I. Kalantzi, R.E. Alcock, P.A. Johnston, D. Santillo, R.L. Stringer G.O. Thomas, Jones, K. C. Environ. Sci. Technol. **35** (6), 1013–1018 (2001). doi:[10.1021/es0002464](https://doi.org/10.1021/es0002464)
- [31] I.A. Merlin Junior, J.S. Santos, L.G. Costa, R.G. Costa, A. Ludovico, Rego F.C., and E.H. Santana. Arch. Latinoam Nutr. **65** (3), 193–198 (2015).
- [32] Codex Alimentarius; Pesticide database [Internet], (2014). [https://www.fao.org/fao-who-codexalimentarius/codex-texts/dbs/pestres/pesticide-detail/en/?p\\_id=21](https://www.fao.org/fao-who-codexalimentarius/codex-texts/dbs/pestres/pesticide-detail/en/?p_id=21). (accessed Feb 28, 2024).
- [33] EU pesticides database. European Commission [Internet]. [<https://ec.europa.eu/food/plant/pesticides/eu-pesticides-database/start/screen/mrls>]. (accessed Feb 28, 2024).
- [34] R.J. Fussell, in Thermo. Fish Sci. Hemel. Hempstead, UK (2016).