

## Quail Meat Under Threat: Hidden Microplastics Pose Risks to Public Health and Environment

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### Abstract

**Aim of study:** This study aimed to determine the presence of microplastics in tissues and organs of quails (*Coturnix coturnix*) and to evaluate the potential risks of microplastic contamination in terms of human consumption and environmental impacts.

**Materials and Methods:** Organ and tissue samples were analyzed from five laying quails that had died naturally. Samples were treated with 10% KOH and filtered in a laminar flow cabinet. Microplastics were identified using light microscopy and FTIR spectroscopy.

**Results:** Microplastics in filament, fragment, and film forms were detected in quail tissues and digestive system contents. Polyethylene and polyvinyl stearate polymers were the most common types of microplastics. The highest microplastic density was found in intestinal contents. The presence of microplastics in edible tissues (breast and leg meat) was identified, posing potential risks for human consumption.

**Conclusion:** The presence of microplastics in quail meat and tissues poses potential risks for human consumption and highlights the prevalence of environmental pollution.

**Keywords:** Environmental pollution, microplastic, microplastics contamination, public health, quail.

### Bıldırcın Eti Tehdit Altında: Gizli Mikroplastikler Halk Sağlığı ve Çevre İçin Risk Oluşturuyor

#### Öz

**Çalışmanın Amacı:** Bu çalışma, bıldırcınlarda (*Coturnix coturnix*) doku ve organlarda mikroplastik varlığını belirleyerek, insan tüketimi ve çevresel etkiler açısından mikroplastik kirliliğinin olası risklerini değerlendirmeyi amaçlamaktadır.

**Materyal ve yöntemler:** Ölü olarak temin edilmiş beş yumurtacı bıldırcından alınan organ ve doku örnekleri analiz edildi. Örnekler 10% KOH ile muamele edilip, laminar akış kabininde filtrelendi. Mikroplastikler ışık mikroskobu ve FTIR spektroskopisiyle tanımlandı.

**Bulgular:** Çalışmada bıldırcın dokularında ve sindirim organları içeriğinde filament, fragment ve film formunda mikroplastikler tespit edildi ve polietilen ile polivinil stearat polimerleri en yaygın mikroplastik çeşitleri olarak tespit edildi. En yüksek mikroplastik yoğunluğu bağırsak içeriğinde görüldü. Yenilebilir dokularda (göğüs ve but eti) mikroplastik varlığı saptandı, insan tüketimi için risk oluşturabileceği belirlendi.

**Sonuç:** Bıldırcın eti ve dokularında mikroplastik varlığı insan tüketimi için potansiyel risk oluşturmakta, çevresel kirliliğin yaygınlığını göstermektedir.

**Anahtar Kelimeler:** Çevresel Kirlilik, Mikroplastik, Mikroplastik Kontaminasyonu, Halk Sağlığı, Bıldırcın.



## Introduction

Microplastics, defined as plastic particles smaller than 5 mm, are recognized as a pervasive pollutant that affects not only aquatic ecosystems but also terrestrial environments and their inhabitants (Cole et al., 2015; Olubusoye et al., 2023; Sun et al., 2019). Studies have shown that microplastic can disrupt the ecosystem health through breaking the food chain (Bhusare et al., 2024; Olubusoye et al., 2023). The bioaccumulation of microplastics throughout ecosystems, along with their cascading effects across the food web, impacts not only individual species but also extends beyond aquatic environments to terrestrial ecosystems, thereby influencing broader ecosystem dynamics (Cole et al., 2015; Davis & Raja, 2020). The accumulation of microplastics in terrestrial environments is concerning, as they can be transported into aquatic systems via runoff, perpetuating the pollution cycle (Lehner et al., 2019; Paudel et al., 2024). Moreover, ingesting microplastics by terrestrial organisms, including mammals, raises significant concerns regarding bioaccumulation and toxicity (Bhusare et al., 2024; Jeong et al., 2024; Dong et al., 2023). The ingestion of microplastics by terrestrial organisms occurs through contaminated food, water, and air. Studies have demonstrated that microplastics can accumulate in the tissues of mammals and poultry, leading to potential risks such as reproductive toxicity and metabolic disorders (Prata et al., 2021; Bhusare et al., 2024; Paudel et al., 2024).

Microplastics in animals living in terrestrial ecosystems can disrupt various physiological parameters, thereby posing a significant threat to animal health. The presence of microplastics in the food chain constitutes a direct risk to the health of terrestrial organisms, as these particles can carry toxic substances capable of impairing growth, reproduction, and overall well-being

(Jeong, et al., 2024; Paudel et al., 2024; Bhusare et al., 2024; Prata et al., 2021). Moreover, the long-term effects of microplastic exposure in mammals and poultry are still under investigation; however, existing evidence suggests that it may lead to chronic health issues, including inflammation and oxidative stress (Mahmud et al., 2024; Li et al., 2024). Terrestrial mammals and poultry are increasingly exposed to microplastics, which have become ubiquitous across various ecosystems. The primary exposure routes include ingestion via the digestive tract and, to a lesser extent, inhalation of particles through the respiratory system. Studies have demonstrated that these particles can accumulate in various tissues, including the liver, kidneys, and gastrointestinal system (Deng et al., 2017; Salikova et al., 2024; Yong et al., 2020).

The ingestion of microplastics in animals typically occurs through the consumption of contaminated feeds and water. Once ingested, microplastics can translocate from the intestine to the lymphatic and circulatory systems, reaching various organs and tissues (Palaniappan et al., 2021; Smith et al., 2018). Studies have reported the presence of microplastics, such as polyethylene (PE), polyvinyl chloride (PVC), polypropylene (PP), polystyrene (PS), etc., in animal tissues, raising concerns about their potential toxicity and long-term health impacts. For instance, microplastics have been associated with oxidative stress, inflammation, and disruptions in metabolic processes (He & Yin, 2023; Roman et al., 2024; Salikova et al., 2024).

Additionally, evidence is growing that microplastics may affect reproductive health, with potential transgenerational effects observed in mammalian models (He & Yin, 2023; Mills et al., 2023). Beyond mammals, the presence of microplastics in poultry feed and manure has been documented, heightening concerns about their accumulation in edible poultry tissues,

particularly from a public health perspective. Research has indicated that microplastics can be found in poultry meat, posing a risk of human exposure through the consumption of contaminated products (Kadac-Czapka et al., 2023; Ma & Li, 2023).

Despite the expanding literature on microplastics in poultry, significant research gaps remain, and the full extent of their health effects on poultry, as well as their implications for food safety and human health, are not yet fully understood (Lackner & Branka, 2024; Lu et al., 2022; Ma & Li, 2023). Recent studies on microplastics in poultry have predominantly focused on chickens, with limited research on other avian species that are consumed by humans and integral to natural food chains, such as wild birds (Cusworth et al., 2023; Jasińska et al., 2023; Lackner & Branka, 2024; Lu et al., 2022).

This study highlights the potential for microplastics to contribute to broader health issues, mainly through wildlife contamination and the potential bioaccumulation in the food chain, ultimately impacting humans (Blackburn & Green, 2021). Consequently, this study addresses the overlooked issue of microplastic contamination in the organs and tissues of quails (*Coturnix coturnix*), which both humans and wildlife consume. We have investigated the presence of microplastics in various tissues and organs, including the liver, spleen, ovaries, pancreas, heart, gizzard, intestines, intestinal contents, gizzard contents, breast and leg meat, and visceral fat, providing valuable insight into microplastic contamination in quails.

## Material and Methods

### Animals

This study was conducted on five laying quails (*Coturnix coturnix*) in the egg-laying period, which had died naturally or due to disease and

were obtained from the same farm in Kastamonu Province, Türkiye. To prevent environmental microplastic contamination, necropsy procedures were performed under a fume hood, and the necessary tissues and contents were collected.

The following organs and tissues were sampled for the study: gizzard and its contents, intestine (including the duodenum, jejunum, and ileum), intestinal contents, liver, spleen, visceral fat, breast muscle, leg muscle, heart muscle, ovary, and pancreas.

### Tissue Extraction

To extract potential microplastics from the tissues, an alkaline digestion process was applied using filtered 10% KOH (w/v) (Rani et al., 2023). Tissues were finely chopped into small pieces with a knife and placed in 500 mL glass beakers.

All glassware used during the extraction process was washed sequentially with filtered distilled water, filtered ethanol (Absolute for Analysis; CAS No: 64-17-5, Merck, Germany), filtered acetone (CAS No: 67-64-1, Isolab, Germany), and again filtered distilled water. To prevent contamination, the glassware was covered with aluminum foil.

Filtered 10% KOH (200 mL) was added to the tissue samples, then incubated at 60°C in an oven for 24 hours with periodic gentle shaking. After the incubation, the samples were vacuum-filtered under a Laminar Flow cabinet with only one individual present to prevent overcrowding and air movement. In this study, glass fiber with a pore size of 1.2 µm, preheated at 300°C, was used (Filter-Lab MFV3-047).

### Gastrointestinal Content Extraction

Gizzard and intestinal contents collected from the animals were treated with 10% KOH at 60°C for 24 hours to break down organic materials. The extract was then transferred to glass tubes and centrifuged at 1000 rpm for 5 minutes to remove

coarse particles, such as feed materials. The liquid fractions were subsequently vacuum-filtered under a Laminar Flow cabinet using filters preheated at 300°C.

### Contamination Control

To monitor potential airborne contamination of the liquids used in the study, filters obtained under a Laminar Flow cabinet were used as negative controls.

### Quantification and Classification of Microplastics

After drying, the filters were examined under a light microscope (Leica DM500). Suspected microplastic particles were classified as filaments, fragments, and films. All particles count was recorded. Each observed particle was photographed and scaled (Leica ICC50W). For further microplastic characterization, Fourier-transform infrared (FTIR) spectroscopic analysis was performed. FTIR analyses (ATR-FTIR, Perkin Elmer, Spectrum-two, USA) were carried out in the 600-4000  $\text{cm}^{-1}$  range with a resolution of 4  $\text{cm}^{-1}$  and 32 scans in absorption mode. The spectra were compared with the library (Fiveash Data Management, Inc. 2006-2008) database for validation. A match rate of 70% or higher was used as the criterion for polymer identification.

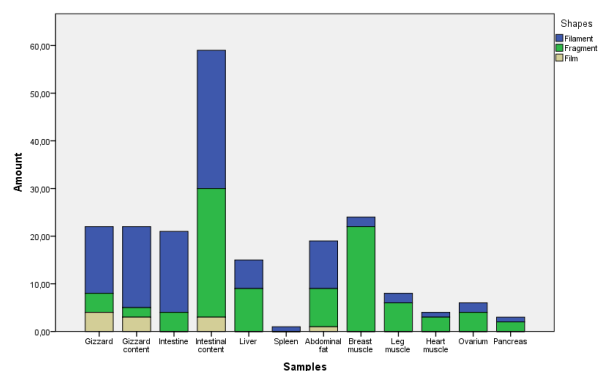
### Statistical Analysis

Statistical analyses were performed to evaluate the differences in microplastic density across tissues and animals. To test for statistically significant differences, One-Way ANOVA was applied for both tissue- and animal-level comparisons. For post-hoc comparisons, Tukey's HSD test was applied to identify specific contributing to significant differences when ANOVA results indicated significance. All statistical analyses were conducted using IBM SPSS Statistics 23, with a significance level of  $p \leq 0.05$ .

## Results

No contamination was detected in the control filters from the materials used in the study. The Petri dishes used were sequentially washed with distilled water, ethanol, acetone, and distilled water, covered with aluminum foil, and confirmed to be free from airborne contamination under microscopic examination.

Analyses of the filters obtained after the extraction of tissue and content samples from the animals revealed the presence of microplastics (Tables 1 and 3). The results variability in microplastic levels among animals from the same facility. Microscopic examination of tissue and content extract identified microplastic particles in the forms of filament, fragment, and film (Figure 1). Total microplastic amounts for the animals (Q1, Q2, Q3, Q4, and Q4) were 61, 44, 37, 28, and 34, respectively (Figure 2). However, there was no statistically significant difference in the microplastic amounts among the quails ( $p = 0.351$ ).



**Figure 1.** The distribution of microplastic shapes by tissues (particle count/sample).

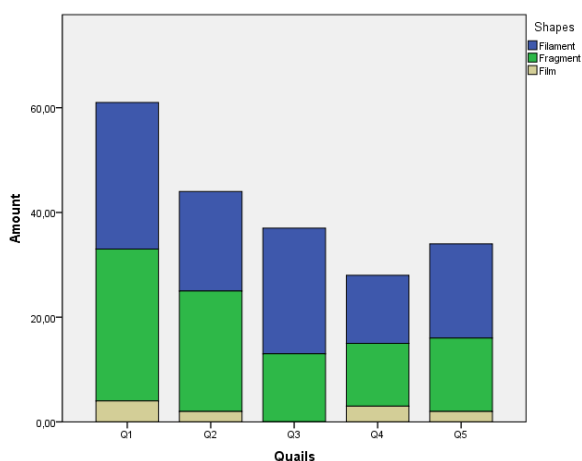
Significant differences in microplastic presence were observed among tissue and content samples ( $p < 0.05$ ). Among potential contamination routes, gastrointestinal contents (gizzard and intestinal contents) exhibited the highest microplastic presence. Notably, the small intestine contents had the highest density,  $11.8 \pm 3.27$  (Table 2).

**Table 1.** Microplastic counts in animals. All particles classified as filament, fragment, and film. Groups sharing the same superscript letter indicate no statistically significant differences ( $p > 0.05$ ). Different superscripts denote statistically significant differences ( $p < 0.05$ ).

Quail Samples	Filament					Fragment					Film					Total									
	Q1	Q2	Q3	Q4	Q5	Q1	Q2	Q3	Q4	Q5	Q1	Q2	Q3	Q4	Q5										
Gizzard	3	4	3	3	1	1	2	0	0	1	1	0	0	3	0	22									
Gizzard content	3	3	5	3	3	1	0	0	1	0	0	2	0	0	1	22									
Intestine	6	2	3	2	4	0	2	0	0	2	0	0	0	0	0	21									
Intestinal content	6	5	8	4	6	7	5	5	3	7	2	0	0	0	1	59									
Liver	2	1	0	0	3	2	2	1	2	2	0	0	0	0	0	15									
Spleen	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1									
Abdominal fat	2	4	2	1	1	2	2	2	1	1	1	0	0	0	0	19									
Breast muscle	1	0	1	0	0	8	5	4	4	1	0	0	0	0	0	24									
Leg muscle	1	0	1	0	0	2	4	0	0	0	0	0	0	0	0	8									
Heart muscle	1	0	0	0	0	2	1	0	0	0	0	0	0	0	0	4									
Ovary	1	0	1	0	0	3	0	1	0	0	0	0	0	0	0	6									
Pancreas	1	0	0	0	0	1	0	0	1	0	0	0	0	0	0	3									
<b>Total</b>	28	19	24	13	18	29	23	13	12	14	4	2	0	3	2	204									
<b>Microplastic Amounts (Mean <math>\pm</math> S.D)</b>																									
<b>Quail</b>	<b>Q1</b>					<b>Q2</b>					<b>Q3</b>					<b>Q4</b>					<b>Q5</b>				
<b>Amounts</b>	5.08 $\pm$ 3.27 <sup>a</sup>					3.66 $\pm$ 3.05 <sup>a</sup>					3.08 $\pm$ 3.62 <sup>a</sup>					2.33 $\pm$ 2.42 <sup>a</sup>					2.83 $\pm$ 4.10 <sup>a</sup>				

**Table 2.** Microplastic densities by tissues (particle count/sample), classified by density group. Groups sharing the same superscript letter indicate no statistically significant differences ( $p > 0.05$ ). Different superscripts denote statistically significant differences ( $p < 0.05$ ).

Sample	Groups based on microplastic density	Microplastic Amounts (Mean $\pm$ S.D)
Intestinal content	<b>Group 1 (Highest Density)</b>	11.8 $\pm$ 3.27 <sup>a</sup>
Gizzard	<b>Group 2 (Moderate Density)</b>	4.40 $\pm$ 1.81 <sup>b</sup>
Gizzard content		4.40 $\pm$ 0.54 <sup>b</sup>
Breast muscle		4.80 $\pm$ 2.86 <sup>b</sup>
Liver		3 $\pm$ 1.58 <sup>b</sup>
Abdominal fat		3.8 $\pm$ 1.78 <sup>b</sup>
Intestine		4.20 $\pm$ 1.79 <sup>b</sup>
Pancreas		<b>Group 3 (Low Density)</b>
Heart muscle	<b>Group 4 (Lowest Density)</b>	0.80 $\pm$ 1.30 <sup>b</sup>
Ovarium		1.20 $\pm$ 1.78 <sup>b</sup>
Leg muscle		1.60 $\pm$ 1.81 <sup>b</sup>
Spleen		0.20 $\pm$ 0.45 <sup>c</sup>



**Figure 2.** The distribution of microplastic shapes by animals (particle count/animal).

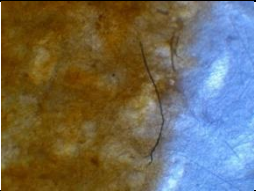
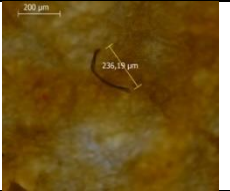
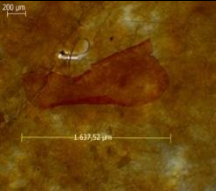

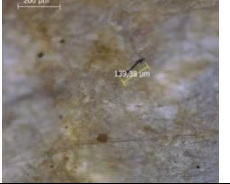


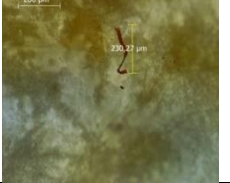
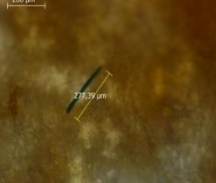




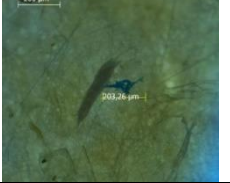
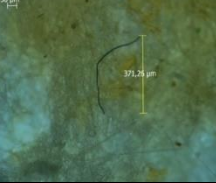


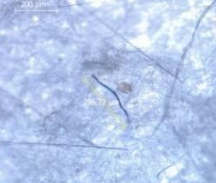

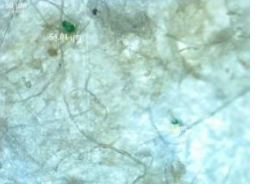

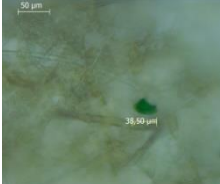
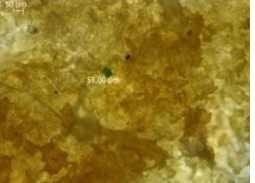

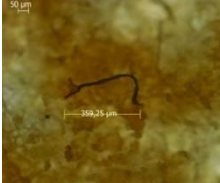


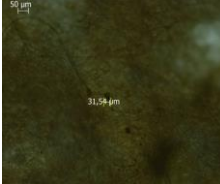

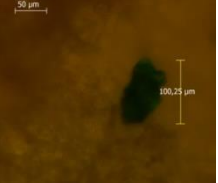



Microplastic analysis revealed that the majority of the microplastics were in filament and fragment forms. Microplastic levels in gizzard tissue were found to be similar to those in gizzard contents, averaging 4.40 $\pm$ 1.81. The average microplastic presence in intestinal tissue (including duodenum, jejunum, and ileum) was notably lower than that in intestinal contents, at 4.20 $\pm$ 1.79.

In edible tissues, such as breast and leg muscles, average microplastic amounts were 4.80 $\pm$ 2.86 and 1.60 $\pm$ 1.81, respectively ( $p = 0.324$ ). Among the other tissues, the spleen exhibited the lowest microplastic presence, with an average value of

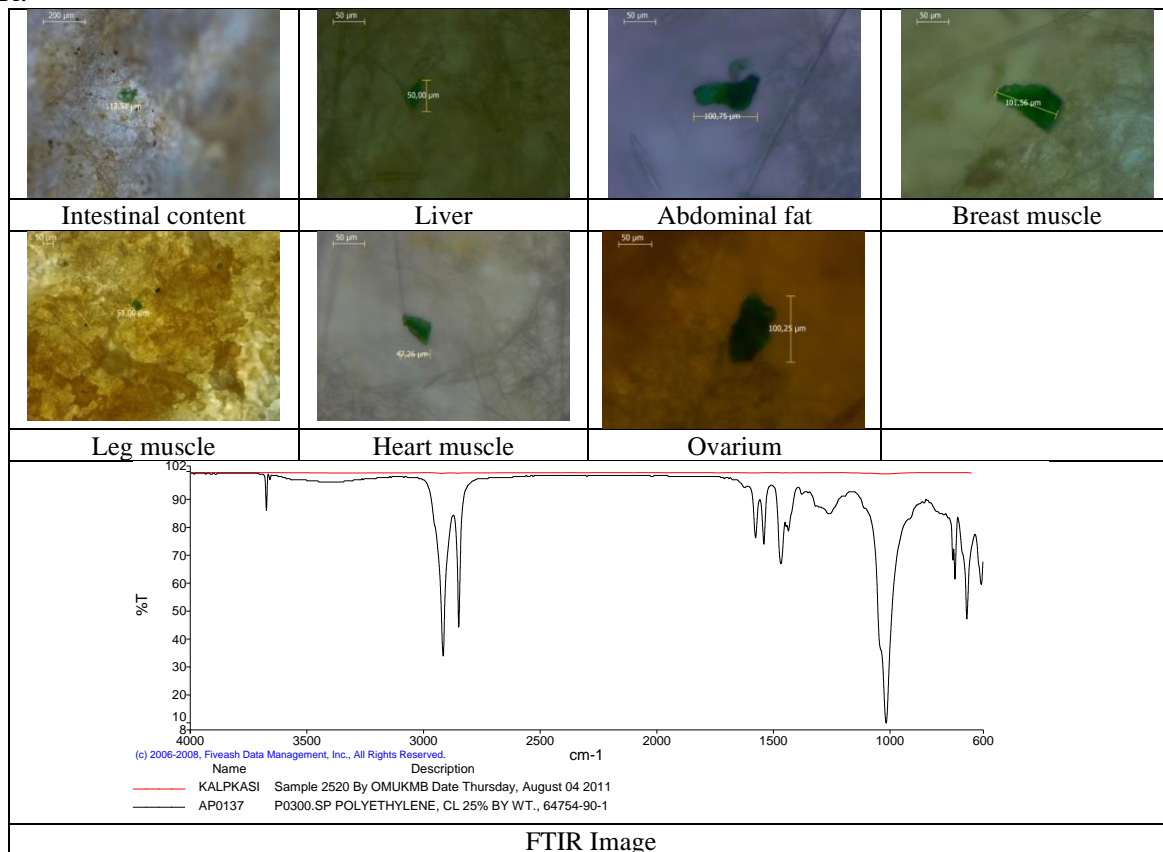
0.20 $\pm$ 0.45 (Table 2). The size of microplastic particles detected across tissues and contents ranged from 30 to 1600  $\mu$ m.

When microplastic presence was grouped by tissue type, intestinal contents had the highest density, followed by the gizzard, gizzard contents, and breast muscle. The liver, abdominal fat, and intestines formed the third group, while the spleen, pancreas, and heart muscle exhibited the lowest levels. Microscopic examination of green-colored fragments with average length of 67.07 $\pm$ 29.89  $\mu$ m observed in the intestinal content, liver, abdominal fat, breast muscle, leg muscle, heart muscle, and ovaries, was followed by FTIR analysis. FTIR scans confirmed that these fragments were composed of PE (Table 3-4). Filament and film-like particles identified in gizzard and gizzard contents, as well as filament and fragment particles in the liver, heart muscle, intestinal content, and abdominal fat tissues, were also classified as PE (Table 5). Additionally, other microplastic particles were predominantly identified as polyvinyl stearate (PVS) based on FTIR spectra.

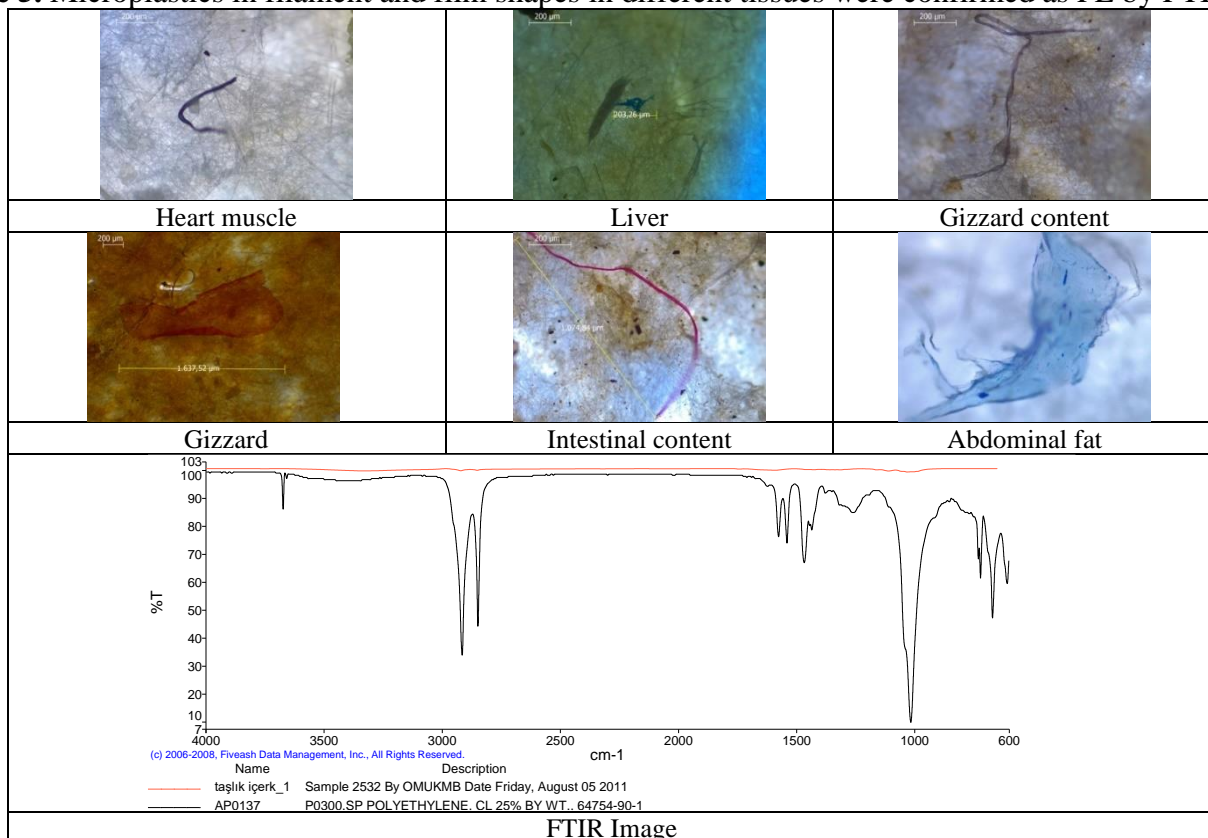
**Table 3:** Microplastic particle images in filament, fragment, and film shapes by tissue.

Sample	Microplastic Images		
<b>Gizzard</b>			
<b>Gizzard content</b>			
<b>Intestine</b>			
<b>Intestinal content</b>			
<b>Liver</b>			
<b>Spleen</b>			
<b>Abdominal fat</b>			
<b>Breast muscle</b>			
<b>Leg muscle</b>			
<b>Heart muscle</b>			
<b>Ovarium</b>			
<b>Pancreas</b>			

**Table 4:** Microplastics, green-colored and in fragment shapes in different tissues were confirmed as PE by FTIR.

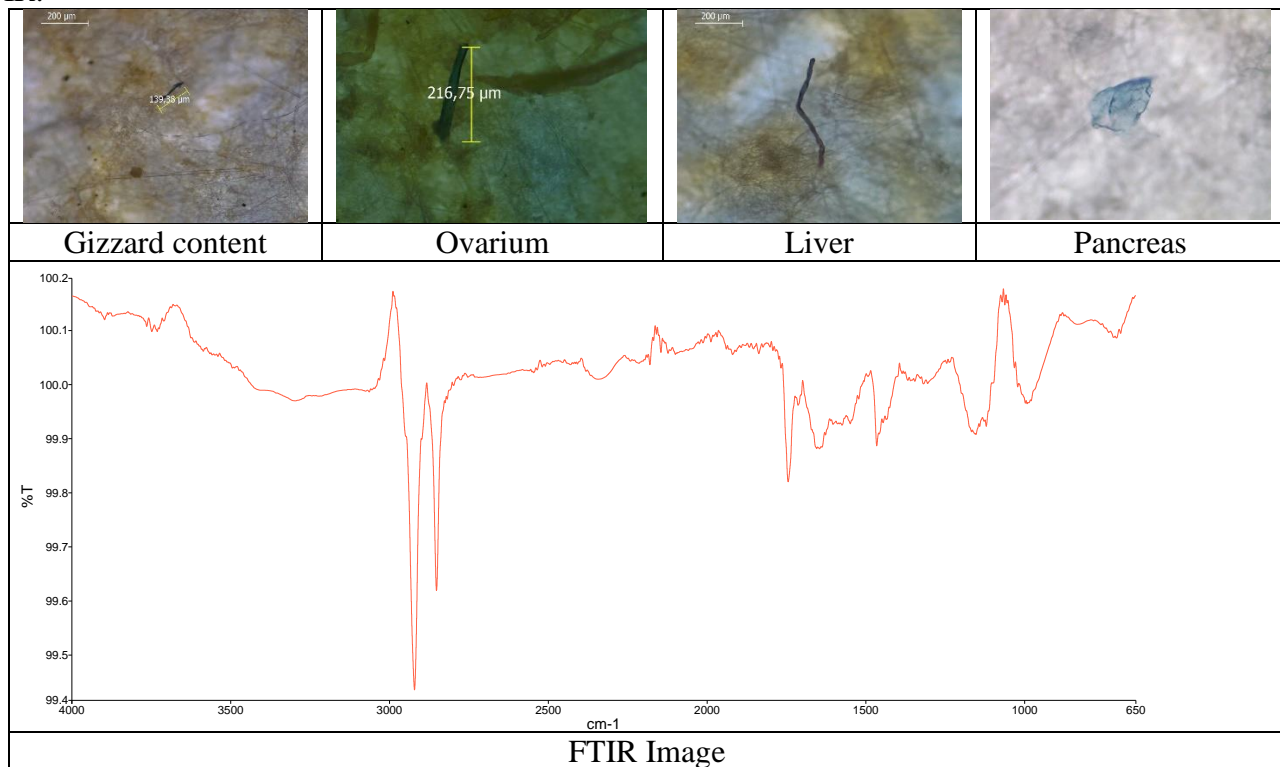


**Table 5.** Microplastics in filament and film shapes in different tissues were confirmed as PE by FTIR.





**Table 6.** Microplastics in filament and fragment-like shapes in different tissues were confirmed as PVS by FTIR.



The presence of microplastics in edible tissues such as breast and leg muscles raises concerns about human exposure through consumption. Based on the findings, estimated microplastic exposure through breast muscle was calculated as 0.48 particles/gram, while leg muscle exposure was 0.16 particles/gram.

## Discussion

Previous studies have reported microplastic contamination in edible seafood, primarily resulting from environmental sources (Bergami et al., 2016). This raised interest in the presence of microplastics in terrestrial ecosystems, especially in animals farmed for human consumption. Recent research has confirmed microplastic contamination in feces and tissue samples of domestic animals (Susanti et al., 2021; Beriot et al., 2021; Wu et al., 2021). In this study, microplastic presence was confirmed in five laying quails (*Coturnix coturnix*) obtained from the same farm and raised for human consumption.

Microplastics in filament, fragment, and film shapes were identified in the gastrointestinal contents and various tissues of the animals.

These findings, consistent with previous studies, indicate that microplastic contamination is not confined to aquatic ecosystems but is rapidly and extensively spreading in terrestrial ecosystems as well. Considering these findings, terrestrial products, like aquatic products, are significantly contributor to human exposure to microplastics and potential health risks.

Quails are widely used for meat and egg production (Lukanov & Pavlova, 2020). They are classified into three types: laying (light), dual-purpose, and meat (heavy). This study did not analyze egg samples from the laying quails due to the potential presence of other animals in the same cage. However, the detection of PE polymers in the ovaries, with particle sizes ranging from 51-100.25 µm, suggest that such particles could potentially be transferred via eggs developing in

the female reproductive tract. Meat-type quails are predominantly farmed in countries such as Spain, France, Italy, and Portugal, as well as the USA and China (Dalle Zotte & Cullere, 2024). In Türkiye, quail meat production was reported as 103 tons in 2019 (TUIK, 2019), although its consumption is less common compared to other countries.

The average microplastic levels in edible tissues such as breast and leg muscles were found to be  $4.80 \pm 2.86$  and  $1.60 \pm 1.81$ , respectively ( $p = 0.324$ ). These values correspond to potential human exposure rates of 0.48 particles/gram for breast muscle and 0.16 particles/gram for leg muscle (Domenech & Marcos, 2021). Consequently, this study is the first to report potential microplastic exposure through quail meat, highlighting the risks associated with their consumption in regions where quail products are popular, such as European countries.

Microplastic exposure in intensively farmed animals is likely caused by air, water, and feed contamination (Walkinshaw et al., 2022; Beriot et al., 2021; Chen et al., 2020). Wu et al. (2021) identified filament and fragment-shaped microplastics, such as PE and PP, larger than 1000  $\mu\text{m}$  in chicken feces. They suggested that possible sources of PE in feces might include the inner linings of feed bags and drinker equipment. In this study, the similarity of microplastic types (PE, PSA) in the gastrointestinal system contents further support the notion that these are likely contamination sources. The transfer of microplastics through the intestinal wall is largely size-dependent (Li et al., 2024). Particles smaller than 150  $\mu\text{m}$  are reported to be more likely absorbed via endocytosis by intestinal epithelial cells (Yong & Du, 2023). Based on measurements, PE particles averaging  $67.07 \pm 29.89$   $\mu\text{m}$  in length, found in intestinal contents, liver, abdominal fat, breast and leg muscle, heart muscle, and ovaries, are likely transported from feed to other tissues through

gastrointestinal system. Although the lengths of other particles suggest a limited passage through the gastrointestinal wall, their diameters might allow transit under intestinal movement. PVS particles are also presumed to follow similar pathways to spread throughout the body.

Based on the study findings, filament-shaped particles were the most prevalent, followed by fragments and films ( $p < 0.000$ ). The presence of filament-shaped particles strengthens the assumption that they might originate from plastic packaging products (Ivleva et al., 2017). The detection of PE and PVS polymers predominantly suggests that the feed and watering materials used in the poultry industry are primary contributors to microplastic contamination. However, air and water contamination cannot be disregarded.

Detecting microplastics in farmed quails, which are consumed as human food, indicated an additional potential route for human exposure to microplastics. These findings emphasize the need for strategies to reduce plastic use and contamination, which could have broader implications for environmental and public health policies. Furthermore, the presence of microplastics in the gastrointestinal system and edible tissues suggests that greater caution is needed regarding feed contamination with microplastics. Stakeholders in animal farming should adopt stricter measures to minimize plastic use. Additional efforts to prevent microplastic contamination, such as implementing methods to degrade microplastics during feed preparation or within the gastrointestinal system, are becoming essential.

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## Ethical Statement

This study was approved by the Kastamonu University Animal Experiments Local Ethics Committee (E-16498365-605-2400143033).

## Author Contributions

Investigation: V.D., Ç.S. and S.G.; Material and Methodology: V.D., Ç.S. and S.G.; Supervision: V.D.; Visualization: M.S.A.; Writing-Original Draft: V.D. and Ç.S.; Writing- review & Editing: V.D., Ç.S., S.G. and M.S.A.

## Conflict of Interest

The authors declared that there is no conflict of interest.

## Data Availability Statement

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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