Revista Veterinaria. 2025; 36(2): 1-8
https://doi.org/10.30972/vet.3628605
Artículo Original

Revista Veterinaria
Facultad de Ciencias Veterinarias-UNNE
ISSN: 1668-4834 / ISSN-e: 1669-6840

Bovine milk and microplastics: Revealing the invisible with advanced microscopy

González-Puetate, I.[1]; Pinela Castro, D.[1]; Carpio Zorrilla, G.M.[1]; Moreno Verdesoto, M.K.[1]; Zambrano Guerra, M.d.C.[1]; Cedeño Reyes, P.P.[1]; Marín-Tello, C.[2]; Vásquez-Kool, J.[3]; Yánez-Avalos, D.[4]*

[1]Facultad de Medicina Veterinaria y Zootecnia, Universidad de Guayaquil. Ecuador. [2]Grupo de investigación en Fisiología y Fisiopatología del Metabolismo de Alimentos en la Ruta de la Investigación Nutricional (MARINUTRI). Facultad de Farmacia y Bioquímica. Universidad Nacional de Trujillo. Perú. [3]Department of Health, Human and Life Sciences Shaw University. 118 East South Street. Raleigh, North Carolina. 27601. USA. [4]Fauna, Conservation and Global Health Research Group, Universidad Regional Amazónica Ikiam, Km 7 Muyuna-Tena 1501-Ecuador. darwin.yanez@ikiam.edu.ec

Abstract
This study assessed the presence of microplastics (MP) in bovine milk marketed in Ecuador, considering milk type, packaging material, and the analytical methodology employed. A total of 35 samples of raw, whole, and semi-skimmed milk packaged in Tetra Pak, plastic pouches, or sold unpackaged were analyzed. Samples were subjected to direct observation (DO), density separation with supersaturated saline solution (SSS), filtration (FSSS), and chemical digestion with 10% NaOH (CD). Raw, unpackaged milk exhibited the highest MP concentrations (241.05 ± 66.18 MP L^{-1}), significantly exceeding those found in semi-skimmed (115.14 ± 24.21 MP L^{-1}) and whole milk (65.83 ± 14.19 MP L^{-1}), suggesting that industrial processing may partially reduce contamination. Unpackaged milk also showed higher MP levels compared to milk in Tetra Pak (92.38 ± 20.56 MP L^{-1}) and plastic pouches (88.60 ± 19.66 MP L^{-1}), indicating a protective effect of packaging. Among analytical methods, DO yielded the highest apparent MP concentrations (390.86 ± 48.48 MP L^{-1}), while SSS, SSSF, and CD reported lower values. CD, corroborated by scanning electron microscopy (SEM), was found to be the most sensitive and reliable technique due to reduced interference. In conclusion, raw and unpackaged milk contained significantly higher levels of MPs, potentially due to factors such as contaminated feed and deteriorated milking equipment. These findings highlight the need to reinforce sanitary control during production, improve processing and packaging practices, and standardize analytical methodologies to ensure dairy safety and protect public health.

Key words: Livestock, contamination, polymer, animal milking products.

Leche bovina y microplásticos: revelando lo invisible con microscopía avanzada

Resumen. Este estudio evaluó la presencia de microplásticos (MP) en leche bovina comercializada en Ecuador, considerando el tipo de leche, el tipo de envase y la técnica analítica empleada. Se analizaron 35 muestras de leche cruda, entera y semidescremada, en envases Tetra Pak, funda plástica y sin envasar. Las muestras fueron sometidas a observación directa (OD), separación por densidad con solución salina sobresaturada (SSS), filtración (SSSF) y digestión química con NaOH al 10% (DQ). Los resultados revelaron que la leche cruda y sin envasar presentó las mayores concentraciones de MP (241,05 ± 66,18 MP L^{-1}), superando ampliamente a la leche semidescremada (115,14 ± 24,21 MP L^{-1}) y entera (65,83 ± 14,19 MP L^{-1}), lo que sugiere que el procesamiento industrial reduce parcialmente la contaminación. En cuanto al envasado, las muestras sin empaque registraron niveles más elevados frente a Tetra Pak (92,38 ± 20,56 MP L^{-1}) y funda (88,60 ± 19,66 MP L^{-1}), destacando el papel protector del envase. La OD arrojó las concentraciones más altas (390,86 ± 48,48 MP L^{-1}), mientras que SSS, SSSF y DQ reportaron niveles más bajos; esta última

Recibido: 23 abril 2025. **Aceptado:** 20 julio 2025

demostró ser la técnica más sensible y precisa, al minimizar interferencias, hecho confirmado mediante microscopía electrónica de barrido (SEM). En conclusión, la leche cruda y sin envasar contiene niveles más altos de microplásticos, probablemente debido a fuentes como piensos contaminados y equipos de ordeño deteriorados. Estos hallazgos subrayan la necesidad de fortalecer el control sanitario en la producción, optimizar procesos industriales y envasado, y estandarizar metodologías analíticas para garantizar la inocuidad del producto lácteo y proteger la salud pública.

Palabras clave: Ganadería, contaminación, polímero, producto lácteo de origen animal.

INTRODUCTION

Microplastic (MP) pollution has become a common phenomenon widespread in the marine ecosystem, as noted by the United Nations Environment Programme (2021). Their presence has been widely documented in molluscs and fish, organisms that act as bioindicators of environmental impact (Iannacone 2021, Jaén-Rodríguez 2024), as well as in freshwater environments, such as rivers (Cháves Velasco et al. 2023). However, PM contamination has transcended aquatic ecosystems, evidencing its presence in farm animals, including ruminants, poultry and swine, which represents a critical link in the food chain (Beriot et al. 2021, Aardema et al. 2024, Caicedo et al. 2024, Fernández et al. 2024, González-Puetate et al. 2024, Loor et al. 2024). This contamination is due, in part, to the presence of PMs in feed for animal consumption, as well as supply systems, in production (Prata and Dias-Pereira 2023, Rivas et al. 2023). Given this scenario, it is of vital importance to study the production and marketing stages of food intended for human consumption, since the potential transfer of PPMs occurs throughout the food chain (Da Costa Filho et al. 2021, Jin et al. 2021, Valarezo et al. 2024). In addition, the presence of microplastics has been detected in human tissues and organs, which reinforces the urgency to address this problem from a global public health perspective (Yang et al. 2023, Lemos 2024).

In the livestock sector, research on the presence of microplastics (MPs) in bovine milk has become relevant due to its implications for food safety. Pacheco and Palacios Moreno (2023) demonstrated that milking equipment can release MPs that are transferred to milk, and that lack of maintenance significantly increases contamination in raw milk. Kutralam-Muniasamy et al. (2020), after analyzing 26 studies worldwide, found that dairy products contain between 1 and 10 PM particles per litre. The most common polymers identified were polyethylene (PE), polypropylene (PP) and polyethylene terephthalate (PET). Migration of microplastics (MP) from plastic packaging is one of the main sources of pollution. It also underlines the urgent need to standardize analytical methodologies in order to improve the comparability of the data obtained (Shruti et al. 2023). The presence of PMs directly affects food safety and quality, as these particles can act as a substrate for bacterial growth, which would compromise not only the quality of milk, but also that of by-products (Fernández et al. 2020, Sangkham et al. 2022, Nihan Tavşanoğlu et al. 2025). In this context, the present study aims to assess possible differences in PM contamination between raw and processed milk, using advanced microscopy techniques to detect these invisible particles and better understand their impact on dairy production.

This research is especially relevant to veterinary medicine and food safety, providing crucial information to mitigate risks associated with microplastic contamination in dairy products for human consumption.

MATERIALS AND METHODS

This study is experimental, quantitative, and descriptive explanatory in scope. It analyzes microplastics in milk using various detection and quantification techniques.

Types of samples. Milk samples were taken in supermarkets and at a dairy farm, obtaining a total of 35 samples distributed in five groups: whole milk in tetrapak (7), whole milk in bags (7), semi-skimmed milk in tetrapak (7), semi-skimmed milk in bags (7) and raw milk (7), in the province of Guayas, Ecuador, in 2024. Sterilised glass containers covered with aluminium foil were used for storage to avoid cross-contamination. Each group of milk samples was subjected to the four analytical techniques with seven (7) replicates each.

Sample preparation and techniques. The samples collected were processed at the Laboratory of the Faculty of Veterinary Medicine of the University of Guayaquil. For the direct observation technique (DO), 50 mL of milk was used from each of the group of samples was subjected to temperature (60 °C) (Criollo 2019). Fifty µL were filtered and observed under an optical microscope at a field of 10X (Martínez and Ramos 2020). Density separation with NaCl supersaturated saline solution (SSS) (Giri et al. 2024), for which 2 mL of filtered milk (filter paper) was placed with 28 mL of SSS and incubated at 37 °C for 4 hours, which allowed the separation of microplastics due to density differences (Cusba et al. 2025). For the third technique, a second filtration of the supersaturated NaCl solution (FSSS) was performed. Subsequently, a chemical digestion (CD) with 10% NaOH was performed, 20 mL of this solution was added to 0.5 mL of filtered milk to remove organic matter without degrading microplastics, incubating at 60 °C for 72 hours (Rodríguez Guamán 2023). A 6:4 mixture of 98% ethanol and distilled water, previously filtered through laboratory paper, is then prepared in a ratio of 6:4 and distributed into 50 mL test tubes. To each one, 1 mL of the supernatant of each sample is carefully added, using a dedicated glass pipette per sample to avoid cross-contamination. The solution is shaken vigorously to ensure homogeneous distribution and incubated for 12 hours under buoyant conditions. Due to their lower density, microplastics tend to accumulate on the surface of the mixture. Subsequently, a drop of the surface fraction is

taken with individual pipettes and deposited on previously identified slides, adding the coverslip for observation under light microscopy (Villao Rodríguez 2022). Sample processing was carried out in a contamination-free controlled environment chamber at a temperature of 24 to 25 °C (BINDER, ED115, Germany) (Culcay et al. 2025).

Observation of the samples. For the quantification and characterization of microplastics with the optical detection technique, a Nikon Eclipse Ei optical microscope at 10X was used (Pérez 2023). For the analysis of the milk samples, scanning electron microscopy (SEM) was used to obtain three-dimensional images of the plastic particles under high vacuum conditions. For this purpose, the samples were coated with a thin layer of gold using the JEOL JPC1200 Fine Coater and subsequently analyzed with the JSM IT500 SEM system (Amano and Diaz 2015). These samples shall be placed for 3 days in a contamination-free controlled environment chamber at a temperature of 24 to 25 °C (BINDER, ED115, Germany). The slides shall then be placed in an oven for 24 hours at a temperature of 40 °C (day 1), 24 hours at a temperature of 60 °C (day 2) and 24 hours at a temperature of 80 °C. From day 4 to day 6 they shall be maintained at a temperature of 100 °C. Subsequently, the plates shall be subjected to 150 °C for 3 hours to complete critical point drying (CPD) without bubble formation and deformation, which is essential to preserve the structural integrity of the particles (Culcay et al. 2025).

Implementation of positive and negative controls. Positive control: This will consist of the inclusion of samples containing known plastic particles, such as polyethylene or polypropylene microplastics, in ultrapurified water to verify the ability of analytical techniques, such as density separation with supersaturated NaCl saline (SSS), filtration of supersaturated NaCl solution (FSSS), chemical digestion (CD) with 10% NaOH as well as scanning electron microscopy (SEM), to detect these particles. This control will allow validation of the efficiency and sensitivity of the detection process, confirming that the methods to be used will be suitable for contamination by plastic contaminants (Lusher et al. 2017, Van-Cauwenberghe et al. 2015, Culcay et al. 2025).

Negative control. This will involve the use of samples without plastic particles such as contaminant-free ultrapurified water to detect any cross-contamination during the analysis process. The negative control samples shall be observed in density separation with supersaturated NaCl saline (SSS), filtration of supersaturated NaCl solution (FSSS), chemical digestion (CD) with 10% NaOH as well as scanning electron microscopy (SEM), to confirm that the plastic particles present in the experimental samples are not due to handling or collection equipment (Sunil et al. 2024, Culcay et al. 2025).

To calculate the microplastic concentration (MP/mL) in the milk samples analyzed, an equation adjusted based on the volume observed in each technique was used. In DO, where 50 µL (0.05 mL) were analyzed, a correction factor of 20 was applied. In density separation with NaCl-supersaturated saline (SSS) y(FSSS), a final volume of 2 mL was considered, with a factor of 0.5. For CD 10% NaOH, 0.5 mL of final sample was used, applying a correction factor of 2. The concentration of microplastics (CMP) was determined using the equation CMP=NMP×F, where NMP is the number of particles counted and F the factor corresponding to each technique.

Statistical analysis. SPSS software was used in the study, applying the Shapiro-Wilk, Levene, and Kruskal-Wallis methods. Descriptive statistics and box-and-whisker plots were also used to compare groups, assess data distribution, and detect potential outliers.

RESULTS AND DISCUSSION

This study evaluates the concentration of microplastics in different types of milk, packaging and detection techniques, analyzing their impact on quantification. Statistical methods were used to evaluate significant differences in the results obtained with optical microscopy (OM) and scanning electron microscopy (SEM) to confirm the presence of plastic particles in the samples.

Table 1. Descriptive statistics of microplastic concentrations by milk type, packaging type, and detection technique.

Variable		Minimum	Maximum	Mean	SD
Milk Type	Whole Milk	1.00	460.00	65.83 ± 14.19	106.20
	Semi Milk	2.50	840.00	115.14 ± 24,21	181.18
	Raw Milk	7.50	1060.00	241.05 ± 66.18	350.21
Package Type	Tetrapack	1.50	700.00	92.38 ± 20.56	153.89
	Bag	1.00	840.00	88.60 ± 19.66	147.15
	Without	7.50	1060.00	241.05 ± 66.18	350.21
Type of technique	DO	60.00	1060.00	390.86 ± 48.48	286.82
	SSS	1.00	27.00	8.23 ± 1.17	6.93
	FSSS	6.00	116.00	33.26 ± 5.13	30.36
	CD	4.00	134.00	50.06 ± 6.05	35.77

As shown in Table 1, raw milk had the highest average concentration of microplastics (241.05 MP mL^{-1}), followed by semi-skimmed milk (115.14 MP mL^{-1}) and whole milk (65.83 MP mL^{-1}), suggesting that industrial processes could contribute to reducing the presence of these particles. However, milking equipment represents a key source of contamination in dairy production, especially when it is not properly maintained. Pacheco and Palacios Moreno (2023)

demonstrated that these hygienic practices are essential to avoid the release of plastic particles during milking. Furthermore, Van-der Veen et al. (2022) identified not only milking equipment but also feed as the main routes of entry of microplastics into cattle, after detecting their presence in blood, milk and feed samples. Likewise, Da Costa Filho et al. (2021) documented the presence of PMs in pasteurised, UHT and powdered milk, with concentrations between 1 and 10 particles per litre, identifying polyethylene (PE), polypropylene (PP) and polyethylene terephthalate (PET) as the predominant polymers, mainly associated with packaging and industrial processing.

In relation to the type of packaging, unpackaged milk showed the highest concentration (241.05 MP mL^{-1}), outperforming milk packaged in bags (88.59 MP mL^{-1}) and in tetrapak (92.37 MP mL^{-1}), suggesting that the packaging material may play a role in both contamination and protection from external sources, supporting Da Costa Filho et al. (2021).

In terms of analytical techniques, direct observation (DO) yielded the highest detection values (390.85 MP mL^{-1}), while density separation (SSS: 8.22 MP mL^{-1}), filtration (FSSS: 33.25 MP mL^{-1}) and chemical digestion with NaOH (CD: 50.05 MP-mL^{-1}) showed considerably lower concentrations. This technical variability is in line with the analysis by Kutralam-Muniasamy et al. (2020), who, in a review of 26 global studies, highlighted the need for standardisation of analytical methods to improve comparability and reliability of results.

Furthermore, statistical analyses revealed that the techniques used showed a non-normal distribution ($p = 0.000$ for OD, SSS and FSSS; and $p = 0.002$ for CD), and that variances were not homogeneous (Levene's test, $p = 0.000$), which justified the use of non-parametric tests. The Kruskal-Wallis test showed statistically significant differences in PM concentrations according to the type of milk ($p = 0.001$), the type of packaging ($p = 0.013$) and the analytical technique used ($p = 0.000$).

Overall, the results obtained confirm that the degree of processing of the milk, the packaging material and the analytical technique used have a significant influence on the detection and quantification of microplastics.

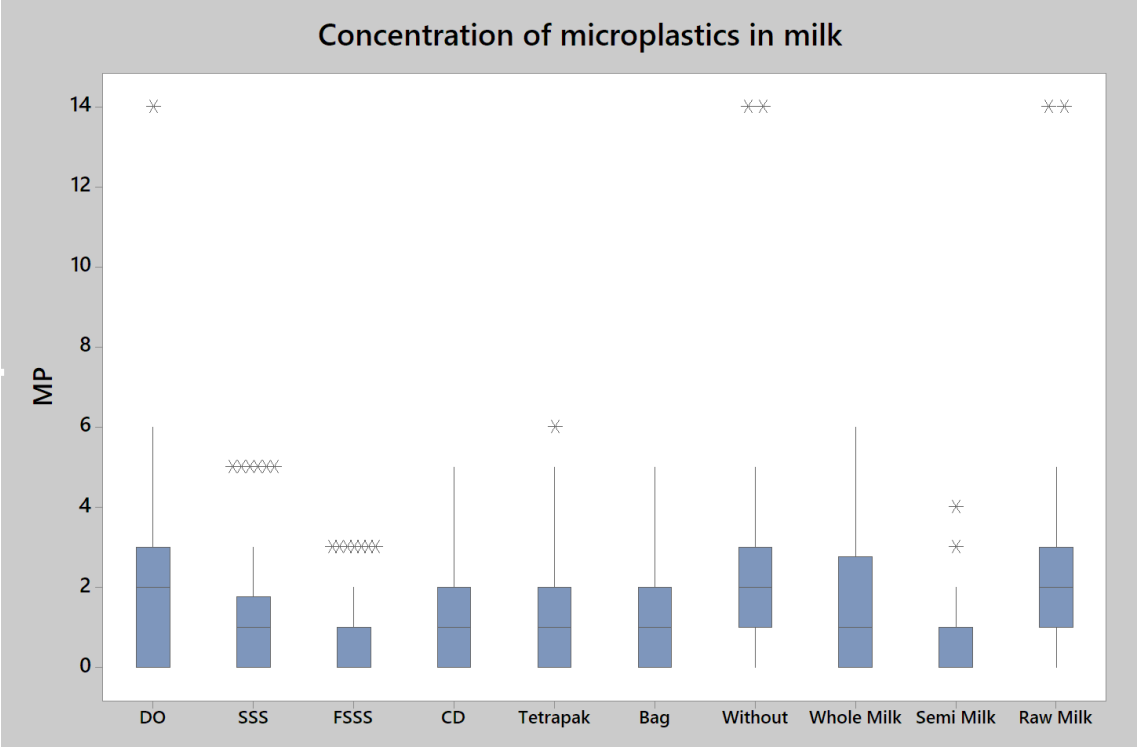

Figure 1. Concentration of microplastics in milk according to type of packaging, type of milk and analysis technique.

Figure 1 shows that unpackaged milk has the highest concentrations of microplastics, reaching up to 133 µg L^{-1}, while samples packaged in tetrapak or bags exhibit significantly lower levels, generally below 200 µg L^{-1}. These results suggest that the analysis process may influence the detection of microplastics, as factors such as handling, filtration and chemical digestion affect the final quantification. Regarding the type of milk, raw milk showed the highest concentrations (up to 133 µg L^{-1}), while whole and semi-skimmed milk showed significantly lower values, usually below 50 µg L^{-1}, indicating that industrial treatments could contribute to reduce the microplastic load. In terms of analytical technique, OD yielded the highest concentrations (up to 1,200 µg L^{-1}), while methods based on SSS, FSSS and CD showed much lower values, confirming that the methodological approach has a substantial impact on the detection and quantification of these particles.

The results obtained show that the concentration of microplastics in milk varies considerably according to the type of milk, the type of packaging and the analytical technique used, in agreement with previous findings. Regarding the type of milk, raw milk showed the highest mean concentration (241.05 ± 66.18 MP L^{-1}), with a maximum value of 1,060 MP L^{-1}, far exceeding semi-skimmed milk (115.14 ± 24.21 MP L^{-1}) and whole milk (65.83 ± 14.19 MP L^{-1}). These data coincide with those reported by Ortega and Solís (2023), who reported an average concentration of 83 MP L^{-1} in raw milk. On the

other hand, Díaz et al. (2020) found significantly lower values in skimmed milk (40 MP L^{-1}), suggesting that industrial processing could contribute to reduce the microplastic load, possibly as a result of filtration and solids separation processes during production. However, Kiruba et al. (2022) reported elevated concentrations in processed milk (164 - 512 MP L^{-1}), indicating that other factors, such as milk origin, collection conditions and particularities of the industrial process, may also significantly influence microplastic contamination levels.

In relation to the type of packaging, unpackaged milk showed the highest concentration of microplastics (241.05 ± 66.18 MP L^{-1}), reaching a maximum value of 1060 MP L^{-1}, while samples packaged in tetrapack (92.38 ± 20.56 MP L^{-1}) and pouch (88.60 ± 19.66 MP L^{-1}) showed significantly lower concentrations. These results are in agreement with those reported by Díaz et al. (2020), who pointed out that the packaging material can influence the presence of microplastics. However, the high concentration detected in unpackaged milk suggests that other factors, such as handling and exposure to the environment prior to packaging, could also be responsible for contamination.

Regarding the analytical technique used, OD yielded the highest concentrations (390.86 ± 48.48 MP L^{-1}), in contrast to the considerably lower values obtained by density separation -SSS: 8.23 ± 1.17 MP L^{-1}; FSSS: 33.26 ± 5.13 MP L^{-1} and CD with 10% NaOH (50.06 ± 6.05 MP L^{-1}). These results are congruent with the findings of Visentin et al. (2024), who applied filtration and alkaline digestion techniques for the extraction of microplastics from milk powder, obtaining more accurate and consistent data. In this context, chemical digestion is positioned as one of the most effective methodologies for the quantification of microplastics by reducing interferences and improving the accuracy of particle identification.

The analysis showed that whole milk contained between 1 and 9 fibers, semi-skimmed milk between 1 and 5, and raw milk between 1 and 3. Moreover, regarding packaging, tetrapack and pouches reached up to 56 fibers, while samples without packaging did not exceed 28. This indicates that processing and packaging type may influence the greater presence of microfibers in milk.

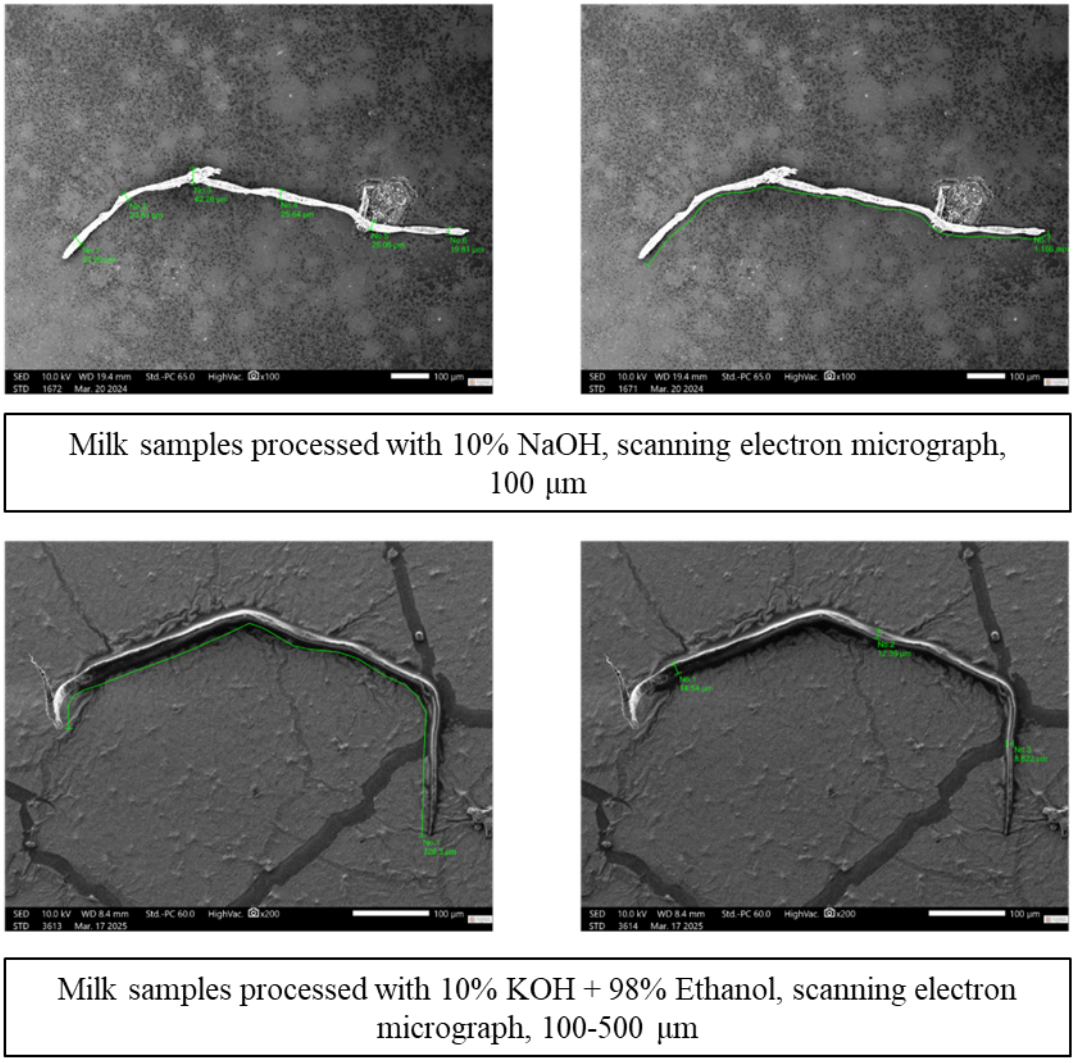

Figure 2. Plastic particles observed by SEM after digestion.

In Figure 2, scanning electron microscopy (SEM) allowed visualization of plastic particles in the samples treated by chemical digestion, with an average size of 20.64 ± 3.81 µm in width and a length of up to 1,166 µm. The addition of 98% ethanol facilitated the removal of residual organic matter, significantly improving the sharpness of

the images, especially at a magnification of 100 µm. This optimization favored a more accurate identification of the particles, which showed an average size of 13.54 ± 0.62 µm in width and lengths ranging from 693.85 to 1,439 µm.

In this research, the presence of microplastics (MPs) in bovine milk was identified by optical microscopy at 10x magnification. Most of the particles observed were fibres (93%), while fragments accounted for only 7%. Similar results were reported by Ortega and Solis (2023), who identified two types of PMs: fibres in 64% of the samples and fragments in 36%. In addition, an average of 32 fibres and 2 fragments per 50 µL were detected in raw milk, and in industrialised milk, 12 fibres and 1 fragment in the same volume. Consistently, in raw milk Ortega and Solis (2023) identified that fibres accounted for 64% of the PMs found. Kiruba et al. (2022) found that fragments were predominant (55%) in industrialized milk. Colours were also analyzed, and most of the fibres observed in this study were transparent (85%), while fragments presented red tones in 39% of the cases. In a complementary way, Valarezo et al. (2024) indicated that in samples of animal origin, transparent fibres represented 70% of the total, while in industrial samples, red tones predominated in 40% of the cases.

These findings reinforce the need to further study microplastic pollution and its bioaccumulation due to its potential impact on the food chain. In this context, farm animals can be considered both indicators of contamination and environmental sentinels (Prata and Dias-Pereira 2023, Aardema et al. 2024, Ferré et al. 2024, Culcay et al. 2025). Also, the detection of microplastics in breast milk (Carpi-Souza et al. 2025) raises important implications within the 'One Health' approach (Corte Pause et al. 2024), suggesting a possible interconnected exposure between animals, humans and the environment.

This study shows that raw, unpackaged milk contains more microplastics than processed milk, indicating that industrial processing and good packaging help to reduce such contamination. The main sources appear to be poorly maintained feed and milking equipment. In addition, among the different forms of analysis, chemical digestion was found to be the most sensitive for detecting these particles. All this reinforces the importance of taking care at every stage of the production process to provide safer food and protect the health of those who consume it.

ORCID

González-Puetate, I. ivan.gonzalezp@ug.edu.ec, https://orcid.org/0000-0001-9930-0617
Pinela Castro, D. douglas.pinelaca@ug.edu.ec, https://orcid.org/0009-0008-0237-7916
Carpio Zorrilla, G.M. genesicaz2001@gmail.com, https://orcid.org/0000-0002-9039-0182
Moreno Verdesoto, M.K. melissak.morenov@gmail.com, https://orcid.org/0000-0002-3442-8959
Zambrano Guerra, M.d.C. maria.zambranog@ug.edu.ec, https://orcid.org/0009-0006-9104-6929
Cedeño Reyes, P.P. pedro.cedenor@ug.edu.ec, https://orcid.org/0000-0001-5976-5225
Marín-Tello, C. cmarin@unitru.edu.pe, http://orcid.org/0000-0001-6862-1805
Vásquez-Kool, J. jorge.vasquezkool@shawu.edu, https://orcid.org/0000-0002-0858-4907
Yánez-Avalos, D. darwin.yanez@ikiam.edu.ec, https://orcid.org/0000-0001-9306-3489

REFERENCES

1. Aardema H, Vethaak A, Kamstra JH, Legler J. Farm animals as a critical link between environmental and human health impacts of micro- and nanoplastics. *Microplast. Nanoplast*. 2024; 4(5).
2. Amano Y, Díaz L. Introducción a la microscopía electrónica: Principios y aplicaciones. 3rd ed. INSPI; 2015.
3. Beriot N, Peek J, Zornoza R, Geissen V, Huerta Lwanga E. Low density-microplastics detected in sheep faeces and soil: A case study from the intensive vegetable farming in Southeast Spain. *Sci Total Environ*. 2021; 755(Pt 1): 142653.
4. Caicedo Olvera J, Paguay Paguay E, Valle Mieles E. Evaluación de la presencia de microplásticos en fuentes de agua y tipos de dieta en ovinos en Guayas, Ecuador [Tesis de grado]. Universidad de Guayaquil; 2024.
5. Carpi-Souza OC-S, Rocha LVMD, Tavares MIB. Microplásticos presentes no leite ofertado na primeira infância. *Braz J Health Rev*. 2025; 8(1): e76878.
6. Cháves Velasco JJ, Arias Hoyos A, Muñoz Solarte DM, Romero Puentes RB. Estudio de microplásticos en muestras de agua y sedimentos de un río urbano del suroccidente de Colombia. *Investig Innov Ingenierías*. 2023; 11(2): 120-130.
7. Criollo K. Determinación de la presencia de microplásticos en leche descremada [Tesis de grado]. Universidad Central del Ecuador; 2019.
8. Corte Pause F, Urli S, Crociati M, Stradaioli G, Baufeld A. Connecting the dots: Livestock animals as missing links in the chain of microplastic contamination and human health. *Animals.* 2024; *14*(2): 350.
9. Culcay I, Yánez D, Delgado J, Montalvo M, Díaz R, Marini P. Detección de partículas plásticas en sangre de equinos mediante Microscopía Electrónica de Barrido. *Revista Científica*, FCV-LUZ. 2025; 35.
10. Cusba J, Pacheco C, Ibarra-Gutiérrez K, Saldarriaga-Vélez JF, Obando-Madera P, Espinosa-Díaz LF. Coastal populations and river discharges as sources of microplastic pollution of the Ciénaga Grande de Santa Marta, Colombian Caribbean. *Environ Monit Assess.* 2025; 197(4): 347.
11. Da Costa Filho PA, Andrey D, Eriksen B, Peixoto RP, Carreres BM, Ambühl ME, Descarrega JB, Dubascoux S, Zbinden P, Panchaud A, Poitevin E. Detection and characterization of small-sized microplastics (≥5 µm) in milk products. *Sci Rep*. 2021; 11(1): 24046.
12. Díaz M, Conesa J, Fullana A. Microplastics in honey, beer, milk and refreshments in Ecuador as emerging contaminants. *Sustainability*. 2020; 12(14): 5514.

13. Fernández P, Prieto M, Fernández P, López M, Álvarez A. Biopelículas y persistencia microbiana en la industria alimentaria. *Arbor*. 2020; 196(795): 538.
14. Fernández M, García V, Valle-Mieles E. Evaluación de la presencia de microplásticos en fuentes de agua y tipos de dieta en porcinos [Tesis de grado]. Universidad de Guayaquil; 2024.
15. Ferré DM, Carracedo RT, Pedrosa A, Caliri MN, Ludueña HR, Gorla NB. Los bovinos domésticos como potenciales bioindicadores ambientales: Análisis del epitelio oral y aplicación en el ensayo de micronúcleos. *Rev vet*. 2024; 35(2): 22-29.
16. Giri S, Lamichhane G, Khadka D, Devkota H. Microplastics contamination in food products: Occurrence, analytical techniques and potential impacts on human health. *Curr Res Biotechnol*. 2024; 7: 100190.
17. González-Puetate I, Martínez-Cepeda G, Torres-Lasso P, Chávez-Toledo K, Guevara G. Microplásticos en materia fecal de rumiantes en Ecuador. *Cien Vet*. 2024; 26(2): 114-129.
18. Iannacone J, Principe F, Minaya D, Panduro G, Carhuapoma M, Alvariño L. Microplastics in marine fishes of economic importance in Lima, Peru. *Rev Investig Vet Perú*. 2021; 32(2): 20038.
19. Jaén-Rodríguez C, Luna-Pacompea N, Juárez-Laguna F, Alvariño L, Iannacone J. Caracterización de microplásticos en *Thaisella chocolata* de un puerto del sur peruano. *Rev vet*. 2024; 35(2): 86-92.
20. Jin M, Wang X, Ren T, Wang J, Shan J. Microplastics contamination in food and beverages: Direct exposure to humans. *J Food Sci*. 2021; 86(7): 2816-2837.
21. Kiruba R, Preethi M, Aganasteen R, Rithick Raj M, Hannah Thabitha C, Monica P, Sakthivel J, Levince C, Naseera Banu I. Identification of microplastics as emerging contaminant in branded milk of Tamil Nadu State, India. *Asian J Biol Life Sci. 2022; 11*(1): 181-187.
22. Kutralam-Muniasamy G, Pérez F, Elizalde I, Shruti V. Branded milks – Are they immune from microplastics contamination? *Sci Total Environ*. 2020; 714: 136823.
23. Lemos L. Exposição isolada e combinada de partículas microplásticas de poliestireno e de metilmercúrio leva a alterações na arquitetura e cobertura de muco no cólon e ao aumento do dano oxidativo nos intestinos de ratos Wistar [Tesis de grado]. Universidade Federal do Rio Grande do Sul; 2024.
24. Loor R, Ochoa J, Valle-Mieles E. Evaluación de la presencia de microplásticos en fuentes de agua, tipos de dieta en equinos [Tesis de grado]. Universidad de Guayaquil; 2024.
25. Lusher A, Hollman P, Mendoza-Hill J. *Microplastics in fisheries and aquaculture: Status of knowledge on their occurrence and implications for aquatic organisms and food safety* (FAO Fisheries and Aquaculture Technical Paper No. 615). Food and Agriculture Organization of the United Nations. 2017. https://www.fao.org/3/i7677e/i7677e.pdf
26. Martínez R, Ramos E. Factores relevantes de la contaminación ambiental por nanoplásticos [Tesis de grado]. Universidad César Vallejo; 2020.
27. Tavşanoğlu ÜN, Akca G, Pekmez T, Kankılıç GB, Çırak T, Çağan AS, Kotiloğlu SÖ, Grossart HP. Increasing microplastics pollution: An emerging vector for potentially pathogenic bacteria in the environment. *Water Res*. 2025; 274: 123142.
28. Ortega F, Solís M. Determinación de la presencia de microplásticos y plomo en muestras de leche cruda [Tesis de grado]. Universidad Politécnica Salesiana; 2023.
29. Pacheco FX, Palacios Moreno M. Impacto de plásticos (macro y microplásticos) en el ecosistema de playas en el cantón Playas (General Villamil), Provincia del Guayas, Ecuador [Tesis de grado]. Universidad del Pacífico; 2023.
30. Pérez E. Detección de micro-plásticos en el medio marino mediante backscattering ultrasónico de alta frecuencia [Tesis de maestría, Universidad Politécnica de Madrid]. 2023. http://hdl.handle.net/10261/353570
31. Prata JC, Dias-Pereira P. Microplastics in terrestrial domestic animals and human health: Implications for food security and food safety and their role as sentinels. *Animals*. 2023; 13(4): 661.
32. Rivas J, Gracia M, Gómez J. Los plásticos y el daño a la salud de los seres vivos y a los ecosistemas. *Biocenosis*. 2023; 34(1): 93-103.
33. Rodríguez Guamán JT. Detección de microplásticos por metodología digestiva alcalina en *Anadara tuberosa* y *Anadara similis* (concha negra) proveniente de la zona de manglar de San Lorenzo, Esmeraldas-Ecuador [Tesis de grado]. Universidad Central del Ecuador; 2023.
34. Sangkham S, Faikhaw O, Munkong N, Sakunkoo P, Arunlertaree C, Chavali M, Mousazadeh M, Tiwari A. A review on microplastics and nanoplastics in the environment: Their occurrence, exposure routes, toxic studies, and potential effects on human health. *Mar Pollut Bull*. 2022; 181: 113832.
35. Shruti VC, Pérez-Guevara F, Kutralam-Muniasamy G, Elizalde-Martínez I. A meta-analysis of microplastic pollution in commercial dairy products: Occurrence, sources, and potential health risks. *Environ Pollut*. 2023; 318: 120854.
36. Sunil M, Mithun N, Charles M, Chidangil S, Kumar S, Lukose J. Visualization and characterisation of microplastics in aquatic environment using a home-built micro-Raman spectroscopic set up. *J. Environ. Manage.* 2024; *354*: 120351.
37. United Nations Environment Programme. From Pollution to Solution: A Global Assessment of Marine Litter and Plastic Pollution. Synthesis. 2021.
38. Valarezo C, Cárdenas Z, Vásquez Ponce V, González-Puetate I, Calle Atariguana D. Miel de abeja: Estudio para determinar la presencia de polietileno en muestras artesanales e industrializadas. *LATAM Rev Latinoam Cienc Sociales Humanidades*. 2024; 5(2): 1645-1653.
39. Van-Cauwenberghe L, Devriese L, Galgani F, Robbens J, Janssen CR. Microplastics in sediments: A review of techniques, occurrence and effects. *Mar. Environ. Res.* 2015; *111*: 5-17.
40. Van-der Veen I, Van Mourik LM, Van-Velzen MJ, Groenewoud QR, Leslie HA. *Plastic particles in*

livestock feed, milk, meat and blood: A pilot study (Report EH22-01). Department of Environment & Health, Vrije Universiteit Amsterdam. 2022. https://www.plasticsoupfoundation.org/rapporten/Final-Report-pilot-study-plastic-particles-in-livestock-feed-milk-meat-and-blood.pdf

41. Visentin E, Manuelian CL, Niero G, Benetti F, Perini A, Zanella M, Pozza M, De Marchi M. Characterization of microplastics in skim-milk powders. *J Dairy Sci*. 2024; 107(8): 5393-401.
42. Villao Rodríguez JR. *Microplásticos en mejillones (Mytella guyanensis), capturados en Puerto El Morro, provincia del Guayas-Ecuador, durante noviembre y diciembre de 2021* (Trabajo de titulación, Universidad Estatal Península de Santa Elena). Repositorio UPSE. 2022. https://repositorio.upse.edu.ec/handle/46000/8106
43. Yang Y, Xie E, Du Z, Peng Z, Han Z, Li L, Zhao R, Qin Y, Xue M, Li F, Hua K. Detection of various microplastics in patients undergoing cardiac surgery. *Environ Sci Technol*. 2023; 57(30): 10911-10918.