

Chapter 3: Microplastics in Small Ruminants I

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Abstract:

The research work in the Province of Guayas (Ecuador) aimed to evidence the presence of microplastics in sheep based on the analysis of 200 samples divided between feces, sources of water, and diet types given. Different methods were applied for the different analyses. The findings were shocking, with microplastics found in every single sample. The most significant factors, such as water origin and feed category, were assessed, which indicated that they are both sources of pollution in sheep. Statistically, dependent and independent factors related with the contamination in these animals were detected. However, one of the biggest concerns is due to their omnipresence, microplastics have now become an enormous problem for both human health and the environment, in part because of their slow disintegration. The build-up of the microplastics is a significant issue as the plastic items typically have a very short useful life and are generally single use, yet they remain in the environment for years without breaking down.

Keywords: Feces, Food, Microplastics, Sheep, Water

1 Introduction

Environmental pollution has gradually intensified – be it on land or water – and is now a big problem that impacts in many ways ecosystems. In fact, the prime culprit is plastic, which is used on a large scale across the world because it is strong, it is lightweight, it is versatile. It is made of organic and synthetic substances and can be formed into wide range of shapes and solids, which is one of the advantages that sets it apart from other materials that could potentially be used for the same purpose, such as metal, paper, or glass (Naspug & Zhiñin, 2022).

Put simply, plastic is everywhere in our lives, from the shack of our food and the clothes we wear (more often than not): synthetic, to car tires and on to medical devices. In a few words, utilization and exploitation of this material have grown exponentially, resulting in the direct accumulation of plastic in landfills and oceans (Naspug & Zhiñin, 2022).

Plastics are physically stable, but they have suffered physical and chemical degradation, which has caused it to fragment into pieces with a size lower than 5 mm, becoming microplastics. Although a standardized definition does not yet exist, microplastics (MPs) are produced both from terrestrial and marine sources and are delivered to marine areas via run-off and activities carried out by man, including tourism, waste water treatment plants, and industrial processing, among others (Torres et al., 2023).

The presence of MPs contributes to the problem of the bioaccumulation throughout the trophic levels of the food chain (Torres et al., 2023).

In research by Narváez and Tipán (2022) it was reported that for the first time, in a United Nations Environment Program as a theme of concern, MPs were introduced in 2014. Today MPs remain an issue that is common, from the bottom of the earth to the tops of mountains.

Indeed, recent studies have emphasized public health implications of MPs and their possible toxic effects which can cause diseases like cancer, cardiopathies and in particular nervous or reproductive disorders (Naspug & Zhiñin, 2022).

Additionally, MPs are also substrates for microorganisms and parasites, in turn, potentially, elevate their impact on environmental health. Castañeta et al. (2020) have speculated that MPs function as a "Trojan horse," in which plastic molecules (microplastics and nano plastics) are accidentally consumed or inhaled, introducing hazardous compounds into living organisms. Hence MPs are known to be the carriers of pollutants and pathogens.

The aim of this study was to encourage responsible actions and environmental awareness in the Guayas Province, protecting animals, the environment and preventing health hazards in humans, for a cleaner and unpolluted future.

The specific aim of this study is to assess the microplastics found in water sources and diet in sheep in the Province of Guayas, Ecuador.

The specific objectives are:

- To determine the microplastics concentrations in the various sources of water provided to the sheep in the study.
- To measure the amounts of microplastics in the various diets the sheep were fed to

- To estimate the quantity of non-relational microplastics present in sheep feces apart from water sources and types of diet.

II. Theoretical Framework

Origin of Plastics

Plastic was created about 150 years ago to fulfill human needs in the daily life (Calderón et al., 2020). “Plastic” is a general term for all kinds of synthetic polymers which are insoluble in water and can be formed into any desired shape when heated to a certain point and cooled ensuring they will hold their shape until disposed (Iannacone et al., 2022).

Plastics, by contrast, are now produced on a mass scale to promote consumption. The majority of the plastic that is produced goes into bottles, packaging, and disposable goods (Jaén & Bano, 2019).

Plastic arose in the mid-19th century, largely out of a desire for something other than ivory for billiard balls. Polystyrene, polyvinyl chloride, polyethylene, polyethylene terephthalate, polymethyl methacrylate, polyamide and similar materials have come to be familiar materials in modern society. For some specialists, the present situation is even akin to the “plastic age” (Prieto, 2022).

And so most of the plastic is very damaging to the environment, and to humans -- as well as to plants and animals. Nevertheless, some nations have awoken to this issue and they have passed bills to work on planning to restrict the misuse of plastics (Prieto, 2022).

The effects of plastic pollution on the environment could be at biochemical/molecular through ecosystem and population (Landrigan et al., 2023).

Classification of Plastics

Plastics may be also classified according to Morocho and Pozo [2] that are:

Synthetic plastics: Created using materials based on petroleum.

Natural plastics: are those that originate from nature using cellulose and casein material.

Plastics can be categorized by their response to heat as:

The plastics are: Thermosetting: Made using an aldehyde, these are heat-treated, molded, solidified and remelted; they become hard materials and can't melt.

- Thermoplastic: When heated will soften and cool to become rigid, can be reheated without permanent change in its properties. They are thermostable at 150 °C and are hence recyclable.

- Elastomers: They can expand to eight times their original length and return to their original shape after being stretched. They degrade at moderate temperatures.

Most Persistent Plastics

Plastics have been in clothes, toys, tools and now are also in our food and drinks — often in plastic packaging too — since their invention. These cassettes also keep medical instruments and equipment sterile. Nevertheless, the universal use and production of plastic have resulted in serious environmental disparities in terms of pollution (National Institutes of Health [NIH], 2024).

While some plastics can be recycled, the vast majority are taken to landfills, where they break down into smaller and smaller bits. Plastic is seeping into oceans, rivers, lakes and the environmental surrounds (Martínez et al., 2023).

Microplastics (MPs) have been discovered in human lungs, intestines, blood, feces, placenta and even breast milk. But, with all of them out there, we still do not know if these plastic particles have any negative effects on human health at all (NIH, 2024).

Urli et al. (2023) reported that the exposure to MPs leads to physical, chemical and microbiological alterations in the organs of the animals. These hazardous properties of nanoparticles can transcend biological membranes, like skin, blood brain barrier (BBB), bowel, placenta and even testicular tissue, to exert direct toxic effects by the dimension of the particles.

Microplastics

MPs are generated by breakdown of plastic. According to Pérez et al. (2024), they are released into ecosystems, where they may be ingested by species along the food chain. Operationally MPs are defined as particles < 5 mm with respect to classification.

Ding et al. (2024) concluded that MPs have already been identified in the environment, water, and soil surfaces.

The above per Celi et al.: Celi et al., the slow degradation and accumulation of MPs in ecosystems are a major environmental threat. There are a few plastic products that are used for short periods before being thrown away, and plastic does not often last long in the environment. It can take decades to manifest itself both in ecosystems and in human communities. They include: Microplastics; Others are intentionally produced synthetics (Chen et al., 2023). Primary Micro plastics: These are synthetically produced synthetic polymer particles such as small beads or granules. They are added to the product for example, cosmetics, and washing agents, and several other products as microbeads. The plastic particles, either beads or pellets as emphasized by Castañeta et al., are then washed into water bodies such as rivers and oceans. However, as Villamar: 2022 indicated, primary MPs can be also found in sizes between 2 mm and 5 mm. Secondary Microplastics (MPs) Synthesized from plastic particles which are often accidentally lost into water bodies from larger particles referred to as microplastics. On the other hand, microplastic breaks down into MPs. Textiles and clothing also generate MPs because physical and chemical wear factors during laundering emit microfibers that are discharged into untreated washing water as revealed by Chen et al. Increase the number of Plastics and Microplastics Approximately 400 million tons of plastic are produced worldwide annually. Most of it is not properly discarded due to inefficiencies in the solid waste management system. Thus, the major issue is poor planning, as pointed out by Vidal et al.

It is currently estimated that up to 245 million tons of plastic particles are being deposited into the marine environment per year by tourism, wastewater, fishing vessels, or industrial discharges. As a result, MPs are found in large quantities in marine ecosystems, many of them having a variety of properties and in large amounts. The presence of these particles, with a large range of sizes and subtypes in the ocean, contributes to the critical negative consequences on marine organisms and eco-systemic processes. Without adequate processing, a vast amount of plastic waste unintentionally gets dumped into the environment. Plastic debris can degrade in the environment due to biotic and abiotic causes. However, because biodegradable plastics such as polypropylene, polyvinyl chloride, polylactic acid, and bio-based cellulose acetate have occupied a large section of the market. Numerous corrective efforts have been executed to reduce the consequences of both degradable and non-degradable plastics despite the several advantages of plastic. The main sizes are as follows: The main sizes are as follows: Nano plastics: 1 nm – 1 μ Microplastics: 1 μ – 5 mm Meso plastics: 1 mm – 1 cm. MPs can occur in a variety of forms, including particles, pellets, foams, beads, flakes, filaments, and shreds and may be relevant to the initial form of the primary plastic and the degradative processes, including harsh environmental conditions. Scientists classify plastic molecules by size using the following hierarchical designation.

Macroplastics: 1 cm to 100 cm

MPs also differ in color, size and shape. Available Colors are: clear, red, black, green, blue and white. This information is particularly relevant for studies on aquatic organisms since it has been hypothesized that species could preferentially ingest MPs of certain color. Pink, purple, yellow, brown and other (Cverenkárová et al., 2021) are also less common colors.

Formation of Microplastics

Microplastics, especially less than 5 mm in size, have been in the forefront of public and scientific concern for impact, particularly on aquatic biota. While the chronic impacts on MP exposure are less clear, studies have found MPs in the gastrointestinal tracts of humans and animals (Zhang & Chenxi, 2023).

Distribution of Microplastics

In general, MP load is greater in the coastal zone than in the open sea. There are several sinks to retain plastic items in different environments: during the dry season, and water bodies (i.e., rivers) and seawater during the rainy season. Upon being transported, MPs usually settle in water sediments, and their concentration in sediments is projected to be five times higher from the top to the bottom of the water sediments (López & Fermín, 2019).

The vertical profiles of MPs are difficult to anticipate. Seawater has a constant density of ~ 1.025 , so substances with a density greater than 1 would normally sink, and those with a density less than 1 would float. That is to say, MPs go from surface to the bottom, including water and sediments (López & Fermín, 2019).

The retention and ingestion of MPs may clog the digestive system, reduce energy reserves, alter hormone levels, suppress growth, and retard sexual maturity Lino and Janín, 2020. MPs contain chemical compounds that are ineffective in terms of nutrition and often accumulate in tissues, where they concentrate contaminants. Several other kinds of damage can be identified: for example, “in the liver, inflammation and the accumulation of deposits and increased antioxidant systems such as catalase and signs of stress to the liver,” in some species Lino and Janín, 2020, can be found. Furthermore, MPs, due to their size, shape, or color, have proved to be able to be confused with natural food Caguana, 2021. Individuals who consume MPs may also form complexes with residual organic molecules or extractive toxicants that sometimes leak into the creatures. Such a condition causes these particles to become more harmful in theory based on their

size, with the smaller particles being more toxic. The more poisonous monomers are the ones that contain less MP Caguana, 2021. NPs are more toxic than MPs, while more substantial plastics are ejected quicker. Plastic continues to dominate manufacturing worldwide today, accounting for around 90% of all smoked. The most commonly used and abundant polymers are as follows: Polyvinyl chloride is a low-cost monomer derived from vinyl chloride Quinteros, 2022. High-density polyethylene is a chemically simple polymer derived from chemically straightforward ethylene Polyethylene terephthalate is a thermoplastic polymer Polyethylene has lightweight and excellent adhesion qualities. Polystyrene, bought under the generic term crystal plastic or plastic glass Microplastics are part of polystyrene and are commonly used. While such plants continue to receive one of the highest benefits for each user, their usage zones rise. The consumption of manufactured goods and their anticoagulant effects can be seen in the study of...ero.

Studies show that biological soil members such as earthworms transport plastic molecules through soil. Plowing and sowing processes in agriculture could also embed plastics into the soil biota. Another direct source of contamination due to contaminated crop is through the use of plastic materials and this is of concern, particularly in the aerial part of plants (Garrido & Constanzo, 2022).

Gómez et al. (2019) detail how microfibers adhere to atmospheric dust which subsequently can deposit on food and may result in direct MP contamination.

Dawson et al. (2019) had already pointed out that the occurrence of MPs as beads can imply that they can be transferred through the organisms undigested, being expelled from the body or entering the tissues in smaller size.

Microplastics in Water

MPs may enter aquatic systems by natural means (e.g. rain, river flow and wind). Once they are in water, MPs may aggregate and be transported by freshwater systems or be sequestered in sediments according to their density, size, shape, structure, composition, and buoyancy (Elizarraraz et al., 2022).

As Bollaín and Vicente (2019) emphasize, the products with the description 'biodegradable' and 'oxodegradable' do not substantially decrease plastic emissions into the water and neither they reduce physical and chemical environmental damage.

On average MPs in water are 1.9 mm, while in sediments they are smaller, with a mean size of 1.6 mm. This could be attributed to the fact that sediment-derived plastics are

prone to experience photodegradation more rapidly, in response to higher temperatures and oxygen concentrations (Acosta et al., 2022).

The shape of MPs present in water is dependent on local practice. In household waste and shredded fishing nets, for example, filaments may originate, and granules can be found in hygiene products or in the fragmentation of larger biodegradable plastics (Acosta et al., 2022).

Similarly, Kumar et al., 2024, isolated MP fragments obtained from commercial plastic products manufacturing and meanwhile produced from macro plastics by breaking down them. Gravity Filtration Technique. Filtration is a physical process used in water treatment. Water is passed through a filter medium, which retains the solid part and allows the liquid to pass through a driving force into water filtration. Because of the driving force into the filtration, it becomes gravity filtration.

The involvement of a filtering medium, which is mostly filter paper, is crucial in this type of filtration. Products such as water go through the filtration medium through gravity. Therefore, gravity plays a significant role because it is the power that drives water through the filter due to this fact, it is a straightforward filtration, making gravity cheap since no energy is needed to run.

Microplastics in Feces Microplastics can be incorporated into agricultural soil through irrigation dirtied with plastic. Most sheep farming practices involve grazing, which comprises straw and stubble fields with little grasses. Only a few sheep farms have fodder grounds that grew specifically for their animals that are made to meet the demand to provide pasture.

The period of food remains in the ruminant's digestive system is almost 35 hours; therefore, plastic ingested while grazing can be transported and ended up being excreted in another field. This problem becomes a concern when sheep from a plastic-contaminated field are shifted to a stubble field or natural area. Modified Willis Molloy Technique The Willis Molloy technique is an ancient technology widely used in Veterinary Medicine for parasitology. The technique has been remodeled for MPs detection in animal feces. MP is detected by dispersing the used breeding material of sheep uniformly, based on the principal flotation mechanism used in suspension using gravity.

The flotation sediment is now transferred into a beaker, and more water is added, the solution is stirred well using a glass stick. A pipette is then put into the water in the beaker to separate the flotation. SEM pychoiting used at INSPI validated this technique.

III. Methods

Methodology

Unit of Analysis

- Fecal samples
- Water source samples
- Examples of diet types in sheep (pasture)

Type of Research

- Observational, cross-sectional, descriptive, analytic, Non-experimental field based.

Variables

Dependent Variables

- Microplastics in water sources, diet, and feces.

Independent Variables

- Origin of water sources/diet type

Statistical Analysis

In the statistical section of the study, percentage method with frequency distribution tables was employed to analyze data with SPSS statistical software (version 26).

SPSS A data analysis method can solve algebra, arithmetic, and trigonometric operations, resulting, depending on the context, in the data processing easily, dynamically, and efficiently. This allows the researcher to conduct full statistical studies (Mayorga et al., 2021).

Research Procedure

The study took four months. Sampling and collecting samples in the field were conducted from April to May, and laboratory analyses were conducted. The actual thesis write-up happened from June-July. The following steps were taken:

The following study was authorized to be conducted at the laboratory of the Faculty of Veterinary Medicine and Zootechnic, University of Guayaquil.

The necessity of their participation in this study was explained to the residents at local farms which were visited at the cantons of Pedro Carbo and Naranjal, in Guayas Province.

In total, 100 feces samples, 50 feeding type samples, and 50 water sources samples given to animals were obtained.

Laboratory Procedure:

Fecal Sample Protocol

The method described by Guerrero et al. was used and modified according to the Willis Molloy procedure. (2020). This method was corroborated by scanning electron microscopic analysis at INSPI.

- A NaCl solution at the relationship with the solubility was prepared.
- The feces from Pedro Carbo and Naranjal were collected into glass containers, weighed with the aid of a digital balance scale and watch glass. Samples were weighed (2g) and introduced inlet and dumped into graduated cylinders.
- 28 ml of the supersaturated NaCl solution was poured into each cylinder using a flask and mixed with a glass rod to form a homogeneous mixture.
- Aluminum foils were used to cover the opening of each cylinder to avoid the cross-contamination of airborne MPs.
- MPs to rise to the top of the cylinders for 4 hours at 37.5°C.
- Finally, a drop of the supernatant was immobilized on a microscope slide with a pipette, covered with a large cover glass, and was observed from the microscope with 10 and 40× magnification.

Food Sample Protocol

- The food items where the animals grazed (e.g. barley grass, "retama noequina") were picked up and stored in glass containers.
- All samples were well marked to minimize confusion.
- In the laboratory the food was cut and weighed at 1 g per sample. It was transferred to a graduated cylinder 10 ml of 10% sodium hydroxide (NaOH) solution and homogenized and covered with aluminum foil to avoid contamination of the environment. The sample was then allowed to cure for 72 hours in an oven at 60° C.
- The organic matter decomposed in presence of the sodium hydroxide (NaOH) allowed the MPs to float.
- After the incubation, 1 ml of the supernatant was extracted and combined with 9 ml of ethanol, homogenized, and covered again with aluminum foil to prevent evaporation and contamination. The culture was incubated for 24 hr at 37.5°C.

- A droplet of the supernatant was ultimately applied to a microscope slide using a pipette and overlaid with a cover slip and viewed with a microscope at a 10x and 40x magnification.

Protocol for Water Samples

It was carried out based on Sánchez's (2022) protocol, gravity filtration.

- The sample was taken in the source where the animal's drunk water (Bucay river / water reservoir) in a glass container.
- The sample was labeled with relevant information to distinguish and prevent mixing up of the samples.
- 10ml of each of the water samples was taken with a pipette and filtered with filter paper.
- The filter paper was afterwards dried in an oven.
- After drying the filter paper, the latter was then placed under an optical microscope, at 40x times magnification, to detect microplastics (MPs).

IV. Result and Discussion

For this study, it investigated 200 samples, which consisted of feces samples, various diet types, and drinking water sources fed for the sheep. On the basis of this detailed investigation, we were able to quantitatively interpret the microplastics (MPs) detected in every sampled item.

Presence of MPs in Different Types of Samples	Frequencies		
	FA	FAA	FR
Feces	100	100	50%
Food	50	150	25%
Water	50	200	25%
Total	200		100%

The dependent variables (water sources, diet type and fecal samples) show 100% presence of MPs (Table 1).

This finding is in line with that of the study by González-Puetate et al. (2024) that examined 300 fecal samples from ruminants (100 of each species) and detected 80% of MPs in sheep. They also verified the identification of these plastic particles in other

species, recording 54% of cattle and a higher plenitude of goats, in which the percentage was 93%.

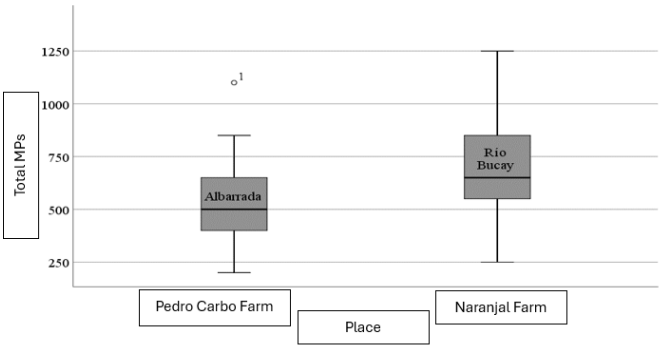
Nevertheless, the results differ from Celi et al. to some extent. (2021) who in 2019 investigated MPs in drinking water and reported positive findings in 80% of cases.

An overwhelming number of MPs were detected in all three studies. This is worrying because sources of environmental contamination are certainly having a greater impact on the numbers of sheep in Guayas province.

Table 2
Statistical Table of the Quantification of MPs in the Different Water Sources Supplied to Sheep

Location	Variable	Parameter	Estimate	S.E.	n	95% CI (Lower)	95% CS (Upper)
1	Total MPs	Mean	550.00	39.69	25	468.09	631.91
2	Total MPs	Mean	698.00	49.28	25	596.28	799.72

Figure 2
Graph of the Quantification of Microplastics (MPs) in the Different Water Sources Supplied to Sheep



The average MPs estimate from the 50 tested samples of different water sources was determined to be 624 MPs/l. Among these, 25 samples corresponded to the Pedro Carbo Canton with a mean of 550 MPs/L and 25 samples to the Naranjal Canton with a mean of 698 MPs/L.

One corresponds to the findings for the mean difference and the statistical analysis with t-test was $p=0.023$, indicating to the existence of statistically significant differences between the studied variables.

These findings are similar to those found by Paredes and Fuentes (2019) in the city of Quito, with 24 drinking water samples analyzed had 75% of the samples with MPs. Conversely, in a study by Roca and Suárez (2024) on 100 sheep, it was revealed that prevalence of MPs was greater in rivers (82.60%), followed by irrigation ditches (81.80%), and wells (72.70%) indicating sources of water negatively or positively affect the quantities of MPs, consistent with findings of this study.

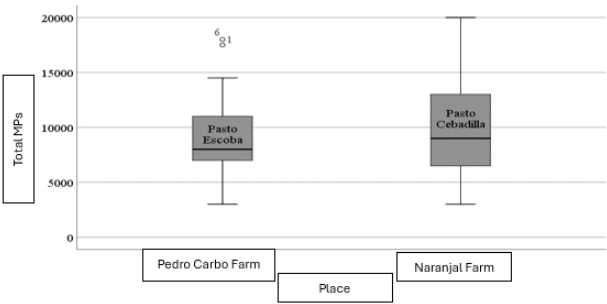
Table 3

Quantitative table of microplastics in the different types of diets supplied to the sheep.

Location	Variable	Parameter	Estimate	S.E.	n	CI Lower (95%)	CI Upper (95%)
1	Total MPs	Mean	9,320.00	738.74	25	7,795.32	10,844.68
2	Total MPs	Mean	9,460.00	904.58	25	7,593.04	11,326.92

Figure 3

Quantitative graph of microplastics in the different types of diets supplied to the sheep.



Based on 50 samples of various diets analyzed, the mean estimated load of MPs (weighted by the number of fed days' use of each diet) in food ingested by the animals daily was 9,390 MPs kg food⁻¹. Twenty-five of the specimens were collected in the

Canton of Pedro Carbo, with the average value of 9320 MPs/kg, and 25 other samples from the Canton of Naranjal, average 9460 MPs/kg.

The statistical comparison (Student t test) of the variables from the Table 3 was that of $p=0.90$, which shows there is no statistically significant difference between them.

Contrastingly, a study by Chusan and Cruz (2023) on two grazing systems with 100 ruminant’s animals reports that grazing system 1 that is similar to the free grazing was highly contaminated (72% of the samples contained MPs) in contrast to grazing system 2 where animals grazed in controlled paddocks, only 36% were colonized with MPs. Likewise, Beriot et al. (2021) studied MPs in low-density conditions by visual identification in 1,000 of sheep and showed that 92% of the analyzed feces samples were MPs positive, indicating that the MPs particles in the soil were consumed by the animals when eating grass.

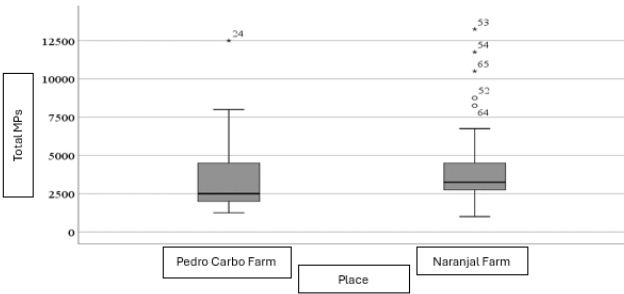
Table 4

Determination of the amount of microplastics in sheep fecal samples not associated with water sources or diet types

Location	Variable	Parameter	Estimate	S.E.	n	CI Lower (95%)	CI Upper (95%)
1	Total MPs	Mean	3,430.00	314.61	50	2,797.76	4,062.24
2	Total MPs	Mean	4,130.00	361.00	50	3,404.55	4,855.45

Figure 4

Graphical representation of the quantity of microplastics in sheep fecal samples not related to water sources or diet types.



In Table, an average of 3,780 MPs per 750g feces were found out of 100 samples investigated. Of the total, 50 samples belonged to the Canton of Pedro Carbo, with an average of 3,430 MPs/750g, and the other 50 samples from the Canton of Naranjal had an average of 4,130 MPs/750g.

Although these results show a slight numerical difference, the statistical test using the t-test resulted in a p-value of 0.14, showing no difference from a statistical point of view for the compared variables.

Similarly, Beriot et al. (2021) showed that all of the soil samples investigated were contaminated with microplastics, with concentrations of $2,116 \pm 1,024$ particles kg^{-1} dry weight. The same study was one of the firsts to assess the presence of MPs in animal feces, and all the flocks analyzed presented MPs with 997 ± 971 particles of MPs/kg of feces. In some instances, it was noticed that animals consumed plastic products by grazing in vegetable fields, where they adhered to plants.

Jie et al. (2020) have shown that microplastics serve as vectors that promote bioaccumulation of hydrophobic organic contaminants through soil organisms. In evaluating this bioaccumulation, considerable reliance was placed on field studies that reported estimates of contaminant exposure and quantifiable contamination within ovine species and laboratory studies that provided estimates of contaminant exposure, absorption, and retention within this species.

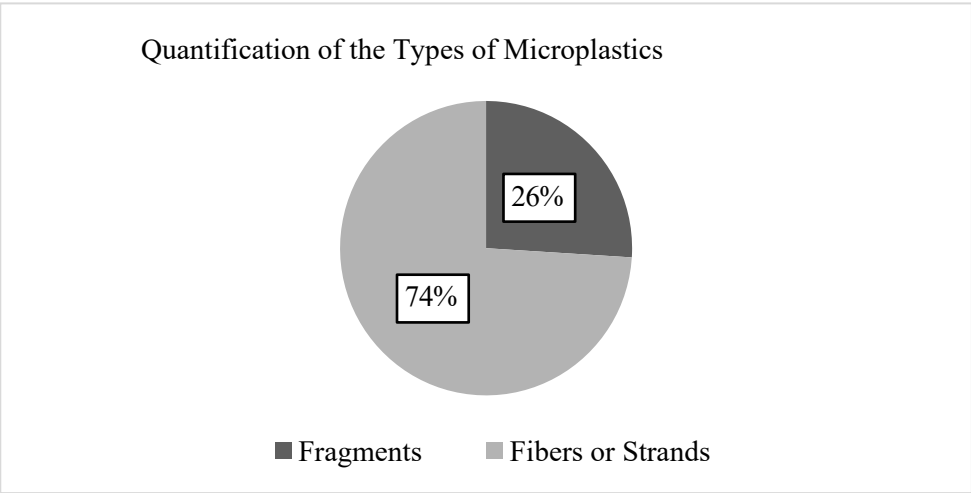
About the quantitative relation between the MPs in sheep feces types and the origin and type of diet (Objective 3), the quantity of MPs excreted per day in animals coming from the Pedro Carbo and the Naranjal cantons does not show differences of statistically significance with a p-value of 0.83.

Table 5
Statistical Table of the Quantification of Microplastic Types

Microplastic Types Quantified	Frequency	Percentage
Fragments	403	26%
Fibers or Lines	1136	74%
Total	1539	100%

Figure 5

Representative Graph of the Quantification of Microplastic Types



The statistical outcomes from the quantification of MP types (microplastics) – fibers and fragments – are presented in Fig. 74% were found to be fibers, which were more dominant than the fragments (26%) in 200 tested samples from feces, diets, and water sources.

Vásquez et al. (2021) highlighted in Fig. 4), the major contributions to differences in MP distributions could be attributed to fibers from dry season (54.19%) and rainy season (86.29%) while fragments from transposition period (55.66%). Investigations in wastewater systems showed that comparable results were obtained in other countries, where the 87% of MPs were fibers; whilst fragments represented only the 6% (Calderón et al., 2020).

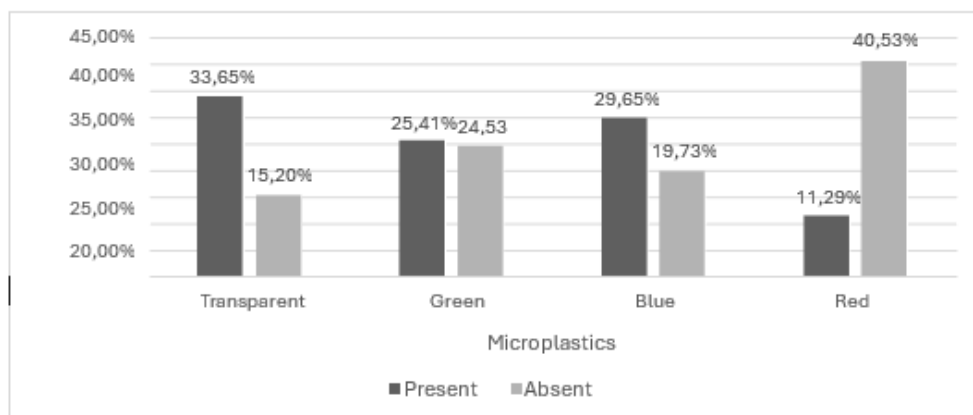
Table 6

Statistical table of microplastic quantification by colors.

Quantification of MPs by Colors	Frequencies		Percentages	
	Present	Absent	Present	Absent
Transparent	143	57	33,65%	15,20%
Green	108	92	25,41%	24,53%
Blue	126	74	29,65%	19,73%
Red	48	152	11,29%	40,53%

Total	425	375	100,00%	100,00%
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Figure 6
Representative graph of the quantification of microplastics by color



When MPs were quantified according to colour, transparent MPs (33.65%) were found to be the most dominant in 200 samples studied, followed by blue (29.65%), green (25.41%) and red (11.29%).

Rosas et al. (2022) transparent MPs are defined from plastics (i.e., from polyethylene (PE), polypropylene (PP), polyethylene terephthalate (PET), polyvinyl chloride (PVC)). In contrast, blue MPs were mainly made of polyethylene, polyethylene terephthalate and polyvinyl chloride.

Ríos (2023), in his investigation collected high density polyethylene plastic packings (detergent bottles, tubes), reported 86% MP present in a freshwater fish, zebrafish (*Danio rerio*). The percentage of contributions received, by color, was: 55% in blue, 15% in black, 8% in red, 6% in yellow, 1% in green and 1% in white.

V. Conclusions

According to the findings, the present study discovered the existence of MP in 100% of the variables tested in this study, which would be indicative that the sheep are exposed to particle of MP daily.

The investigation of the sources of water revealed that MP contamination in rivers is more severe due to more ineffective waste pollutant control. Water reservoirs

(albarradas) are not excluded from this pollution, but the cycle of impurities is different. Diets samples are also contaminants, but as indicated by the findings, there are no significant differences in the number of MPs detected among diets than between waters and feces, respectively.

In the same way, the dog fecal samples do not show differences in the quantities of MPs excreted daily in individuals from both Pedro Carbo and Naranjal, given that the numerical value does not translate into a statistical disparity.

This work is a contribution to the knowledge on MP contamination and also a call for environmental responsibility to protect the animal and human health and ensure best management practices to rugouware products that will be pointed to human needs. But while MPs are a so-called emerging contaminant, there is still plenty of work to do on research and MPs.

Acknowledgments: Jamilet C., Erika P.

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