THE DYNAMIC OF PLASTIC AND MICROPLASTIC WASTE IN THE ISHMI RIVER WAS ANALYZED USING VIBRATIONAL METHODOLOGY, THE FTIR NICOLET 6700, AND RAMAN SPECTROSCOPY

Alda Osmeni[1*], Ardian Maci[2], Arjana Ylli[3], Ramadan Firaj

[1]European University of Tirana, Department of Informatics and Information Technology, Tirana, Albania;*
[2]Agriculture University of Tirana, Department of Environment and Natural Resources, Kamëz, Albania;
[3]University of Tirana, Faculty of Natural Science, Department of Biotechnology, Bulevardi Zogu I Pare, Tirana, Albania;
[4]University of Tirana, Institute of Applied Nuclear Physics, Department of Analytical Methods, Tirana, Albania;

[*]Corresponding Author Alda Osmeni, e-mail: alda.osmeni@uet.edu.al; alda.osmeni@uet.edu.al; amaci@ubt.edu.al; arjana.ylli@fshn.edu.al; ramadan.firaj@unitir.edu.al;

Received June 2024; Accepted July 2024; Published August 2024;

DOI: https://doi.org/10.31407/ijees14.303

ABSTRACT

Albania is considered one of the countries with the highest percentage of untreated plastic in the Mediterranean, about 73%. Approximately 14% of total waste in our country is plastic witch consist in a daily production of plastic waste amounts to 198 tons of low-density and 111 tons of high-density plastic. Plastic - a "silent invader" can stay in the environment for hundreds of years, slowly breaking down into pieces smaller than 5mm called microplastics (MP). MP can seep into ecosystems, contaminate water sources, soil and biota consequently all food chains. Furthermore, the widespread use of plastic in everyday products means that plastic pollution affects almost every corner of the globe, making it an insidious and widespread threat. There is a pressing need for enhanced monitoring and information regarding microplastic pollution in both freshwater and marine environments in Albania, along with an evaluation of the primary sources of plastic pollution in rivers and lakes. The Ishmi River, stretching 74 km and flowing through Rinas, Fushë Kruja, ultimately reaching the Adriatic Sea via Cape Rodoni, is identified by the National Environmental Agency as the most polluted basin. The primary contributors to pollution in the Ishmi River are plastic and primary microplastic waste originating from domestic products, local activities along the river basin, and natural processes. Through this paper, the reader is presented with the dynamics of plastic polymers and microplastics present in five different stations of the river, involving specific steps such as collection, chemical treatment, density separation, and sample filtration. This study aimed to evaluate the importance of using vibrational methodology in determining the dynamics of plastic polymers in freshwater ecosystems by using comparative techniques, such as the FTIR Nicolet 6700 and Raman Spectroscopy.

Keywords: Plastics /Microplastics, sampling, vibrational methodology, Ishmi river

INTRODUCTION

Many plastics are discarded into the aquatic environment due to massive production and imperfect waste management (Filgueiras & Pedrotti, 2019). Plastic waste in the aquatic environment degrades into numerous tiny plastic fragments/fibers/spheroids/ granules/pellets/flakes/beads between 0.1 and 5000 mm in size (G. Peng et al., 2018). These particles are known as microplastics (MPs).

Plastics are used in different Sectors of the industry for packaging (40%), building and construction (~20%), automotive industries (~10%), electrical and electronic industries (~6%), agriculture (3 to 4%), household, leisure, sports (~4%), and others (~17%; this includes plastics for furniture, medical applications, machinery and mechanical engineering, technical parts.) (Maddela et al., 2023). Approximately 70% of all plastics produced are waste, and only 9% are recycled(Bergmann et al., n.d.). There is also an urgent need to gain knowledge on the environmentally relevant concentrations of microplastics and, therefore, to understand microplastic degradation, interaction, and impact on the environment (Maddela et al., 2023), food chain, and health (Vethaaka & Legler, 2021).

In our country, current microplastic research needs more reliable data on concentrations of microplastics in the marine and freshwater environment (Strategjia_ndersektorial_e mjedisit2013_2020, n.d.). Environmental plastics encompass a diverse array of litter, and their characteristics are often delineated based on various factors such as size, origin, shape, polymer type, and color in existing literature (Wagner et al., 2014).However, there is currently no universally accepted classification system. Through this study, we reached to know the dynamics of different plastic polymers present in different stations of Ishmi River (G. Peng et al., 2018). Another essential step of the study was to underscore the critical importance of accurately employing the digestive preparation technique when analyzing freshwater samples for microplastics (Löder & Gerdts, 2015).

This knowledge ensures that microplastics are effectively isolated, contaminants are minimized, and safety measures are followed, enhancing the quality and reliability of the (Hurley et al., 2018) research. It also critically reviewed the methodology presently (Marine et al., 2015) used to evaluate plastic/microplastics pollution in the Ishmi river environment, focusing on the most convenient techniques and approaches recently applied for identifying plastic polymers. After an overview of the laboratory's non-selective sampling approaches and sample processing, the reader was introduced to detection techniques currently applied to plastic and microplastics (Robertson, 2018). The polymer type of environmental (micro)plastics can be identified using techniques like Fourier-transform infrared spectroscopy (FT-IR) or Raman spectroscopy.

The most prevalent polymers in the environment, reflecting global production rates, include high- and low-density polyethylene (HD/LD-PE), polyethylene terephthalate (PET), polypropylene (PP), polystyrene (PS), and polyvinyl chloride (PVC), polyamide fibers (nylon) (Wagner et al., 2014). An overview of plastic size and type was also made in Ishmi River water and sediments. Finally, this work presented a case study emphasizing verifying the synthetic polymer origin of potential plastics and microplastics by FTIR Nicolet 6700 and Raman spectroscopy.

MATERIALS AND METHODS

Study area.

Collecting river water samples for microplastic pollution analysis required careful planning and execution to ensure accurate and representative results (Li et al., 2021a). Before deciding on a sampling location, we needed to familiarize ourselves with the situation of the Ishmi River, its characteristics, and potential sources of microplastics in the area.

Figure 1 Map of Ishmi River

Figure 1 Map of Ishmi River

Table 1. Coordinates of sample stations.

Station	Name	Coordinates
Station I	Rinas Bridge	N 41°26'11.8'' E019° 41' 47.6"
Station II	Gjola Bridge	N41°28'06.3" E 019°41'33.1"
Station III	Paskuqan Bridge	N 41° 21'03.5" E019°48'28.4"
Station IV	Ish Dogana Kashar	N 41° 21'28,5" E019°44'39,2"
Station V	Salmer Bridge	N 41° 32'22,4" E019°36'38,9"

After that, five appropriate sampling sites(not the only one) were identified that represented different types of areas (Ministria e Mjedisit 2020, n.d.)(e.g., urban, industrial, rural) along the river to capture potential variations in plastic/microplastic contamination.

Sample collection

Plastic Sample collection of river water and sediments

Plastic waste accumulates in aquatic ecosystems through various direct and indirect sources. Primary land-based plastic pollution sources are freshwater input, residential and domestic activities, tourism, and other economic actions, including harbor operations.

Many plastic pieces were in river water, and sediments were grabbed with skimmer net water.

Microplastic Sample collection of river water and sediments

Water samples were collected in each station's coastal area and the third station's coast and center area of the river. For laboratory treatment, 2-4 L surface seawater was collected using sterilized glass bottles at about 10 cm depth (Zheng et al., 2021).

A careful estimation of microplastic concentration in sediment samples may require a definition of sampling depth of 1 – 5 cm. Each station took 2-4 kg of sediments in sterilized glass bottles (Prata et al., 2019). All samples were taken back to the laboratory and stored at 4 °C until processing analysis.

Plastic/Microplastic Separation from samples

Plastic purification

All the big plastic particles were washed with H_2O_2 30% and were dissolved in water, relying on organic matter. Samples were dried at room temperature on a Petri plate before analysis.

Microplastic Separation from samples

The research on marine MP is advanced, but there are immense gaps in knowledge.

Regarding freshwater microplastics (Wagner et al., 2014), separation is a critical step that prepares microplastics for accurate and meaningful analysis, contributing to a deeper understanding of microplastic pollution in aquatic environments.

Samples may be subjected to a separation step (Prata et al., 2019) for three main reasons:

- Separation helps concentrate microplastics from a larger volume of water, making detecting and analyzing them easier.
- Separation removes other organic and inorganic materials present in the sample.
- Concentrating microplastics through Separation allows for a clearer view under a microscope, aiding in the proper identification and classification of microplastics.

Microplastic particles can be separated from matrices with higher densities by flotation with high-density saturated salt solutions.

Water samples

To avoid the influence of organic matter in the water sample, was added H_2O_2 30%. Peroxide digest the organic matter (Li et al., 2021b) and ensure for better quality of microplastic waste in water. The baker was then placed in a constant-temperature oscillation box and vibrated at 100 r/min at 70 °C for 24h.

After vibration, the supernatant was taken and mixed with NaCl at a 1l x300gr ratio. It was placed in a constant-temperature oscillation box and vibrated at 100 r/min at 70 °C for 24h.

The solution is homogenized, and NaCl is perfectly dissolved in water to minimize the sample volume for more manageable and efficient results; all the sample content was thrown in a separatory funnel and left for 24 hours.

After decanting big particles and separating densities, only 1/3 of the water column (surface part) was filtered by a vacuum pump through 2,2 μm filter paper. Before analysis, the filter papers were put in a culture dish in the oven and dried at 30 °C for 72 h.

Sediment samples

An optimized quantity of sediment samples was thrown in bakers and put in the oven at 90 degrees for the time needed for total drying. The dried sample sieve in 500-75 μm for 30 minutes.(Adomat & Grischek, 2021) The obtained samples were thrown in a saturated water solution with NaCl and left for 72h on the magnetic plate for a perfectly homogeneous; all the sample content was thrown in a separatory funnel and left for 24 hours. After decanting big particles and separating densities, only 1/3 of the water column (surface part) was treated with H_2O_2 30% at 60 °C in the magnetic plate (Klein et al., 2015). After 48h, the solution was thrown in a separatory funnel and let for 72 h. After the second decantation, the water column was filtered by a vacuum pump through 2,2 μm filter paper. The filter papers were put in a culture dish in the oven and dried at 30 °C for 72 h.

In order to avoid contact between samples and other plastic products, resulting in experimental errors, distilled water was used to clean all experimental equipment (Adomat & Grischek, 2021) to avoid the influence of dust and fine fibers in the air on the experimental results; the samples have placed the samples in Petri dishes covered with glass lids.

Quality control

To ensure better results and to avoid contamination of samples during the sampling process, cotton clothes, laboratory gloves, and sterilized glass bottles were used. In the laboratory, nitrile gloves

and cotton clothes were worn during the experiment, analysis, and determination. All the utensils in the experiment were washed with ultra-pure water (W. Zhang et al., 2017).

Before any chemical treatment, water samples were saved in the fridge, and sediment samples were dried in the oven at constant room temperature.

Methodology in use

The technology used to analyze the prepared samples is:

1. Microscope (Kozo XJPG304, Sony TCC-8.1version 7.3.1.7 40x100 zoom) which was used to locate the remains in the sample and photograph them.

2. The spectrometer is a Nicolet 6700 type from the Thermo Electron company. This spectrometer allows us to obtain spectra in the NIR, MIR and FIR region of the spectral scale.

This system works in two geometries, in transmission and reflection geometry. We worked on geometry with reflection. The basis of this method is the Michelson interferometer.

From the radiation detected by the detector, the interferogram is taken, which in the action of Fourier transformations, which the system uses, will give the absorption spectrum of IR radiation, which we see in a computer.

The i-Raman EX provides a spectral coverage range from 100 cm-1 to 2,500 cm-1, enabling you to measure across the entire fingerprint region. The system's small footprint, lightweight design, and low power consumption ensure research-grade Raman analysis capabilities at any location. The i-Raman EX comes equipped with a fiber probe and an XYZ-positioning-stage with probe holder. It can be used with a range of sampling accessories to facilitate measurements on a wide range of different samples. With the i-Raman EX, always can have a high precision Raman solution for qualitative and quantitative analysis without fluorescence.

RESULTS

Observation and identification of microplastics

After the filter paper containing the filter material dried, a microscope (Kozo XJPG304, Sony TCC-8.1version 7.3.1.7 40x100 zoom) observed all suspicious microplastics on the filter paper, the size and shape of the microplastics were identified and photographed, and ImageJ software was used for size calculation.

The microplastics observed on the filter paper under a microscope showed the following characteristics: 1) no fixed shape, 2) no apparent spatial structure, and 3) uniform diameter (fiber). Some microplastics larger than 2 mm were selected from each sample, and 70 suspected microplastics were selected for FTIR analysis. All microplastics smaller than 5 mm are recorded in the microscope. According to the different sizes, the microplastics were divided into three categories: Fibers <100µm, 100µm-200µm and >200µm and Fragments <100µm², 100µm²-500µm² and >500µm².

Polymer identification of plastics

Microplastics larger than 2 mm were carefully selected for scrutiny of their chemical composition using a Fourier-transform infrared (FTIR) spectrometer (Nicolet 6700) in attenuated total reflection (ATR) mode and Raman (BWS485III). The spectra, obtained within the scan range of 400–4000 cm−1, underwent thorough investigation and were subtracted from the baseline. These spectra were subsequently compared with the library polymer spectra to ascertain the specific polymer type.

Table 2. Microsoft Office Excel 2010 is used to analyze and compute data at each point and create charts. (Leusch et al., 2023).

Nr	Type of plastic waste	Type of polymer	FTIR Mach %	Raman Match%
1	Plastic bag type 1	Polyethylene	73%	96.10%
2	Plastic bag type 2	Polyvinyl acetat/ Polyester	46,6%/ 43,7%	33,2%
3	Plastic bottle	Polystyrene	58%	68,2%
4	Plastic bottle cap	Polyethylene	73%	96.10%
5	Tie	Polyethylene/ Calcium Carbonate	37%/29,7%	none
6	Plastic Toy type 1	Polypropylene Atactic	79,39%	81,17%
7	Plastic Toy type 2	Alcyd resine / Polyester	64,8%/ 59,94%	none
8	Clothes scraps	Poly(Ethylen;propylen)	66,86%	none
9	Detergent bottle	Polymethacrylate / Acrylic	66,57%/ 66,12%	57,11%
10	Garbage bags	Acrylic/ Polybutadien	42,6%/ 48%	none
11	Artificial grass carpet	Polypropylene Atactit/ Syndiotactic	81,7%/ 73,39%	78%

Morphological characteristics of microplastics

Five sampling points (S1-S5) were set up in the Ishmi basin (Fig. 1). Laboratory analysis showed that microplastics were detected in each sampling point, indicating that microplastics had polluted the Ishmi basin of the Ishmi River. Filter paper of microplastic samples obtained from the laboratory process was analyzed using the same techniques. Image-J software analyzed microplastics' size and shape in the Ishmi River.

During the microscopic observation phase, various colors of microplastics were identified, including white, transparent, black, blue, green, and red. The diversity of microplastic colors serves as an indicator of pollution sources (Fig 2) (D. Zhang et al., 2021).

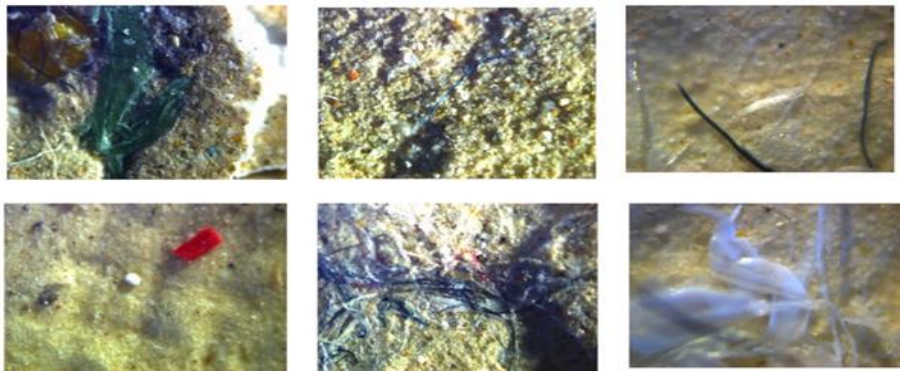


Figure 2. Different microscope images of MP shapes and colors present in Ishmi Basin.

MP in all different colors was found in all stations. That means that the pollution sources of these microplastics were overwhelmingly comprehensive. At the same time, different types of MP shapes were identified. In this study, according to previous literature research (D. Zhang et al., 2021), the shape of microplastic could be divided into four types: fragments, fibers, particles, and others. The distribution of microplastics with different colors and shapes was detailed in figure 3 and 4, concluded from the supplementary information.

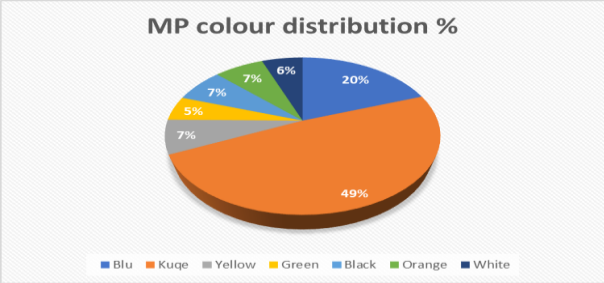


Figure 3. MP color distribution of five different stations

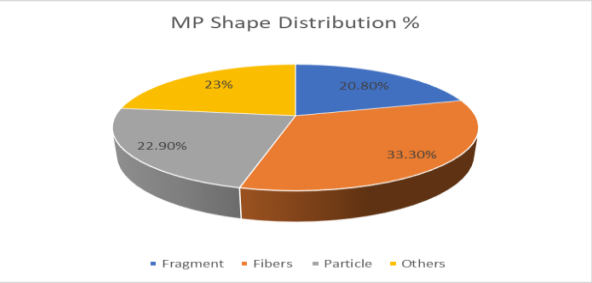


Figure 4. MP shape distribution of five different stations

The ratio of microplastic to fiber, fragment, particle, and others is 33.3% %, 20.8%, 22.9%, and 23%, respectively (Fig 4). The proportion of types shows that, for typical microplastics, the proportion of fibrous microplastics is the largest.

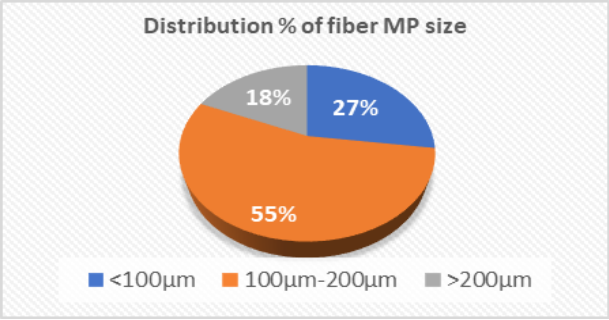


Figure 5. MP fiber size distribution of five different stations

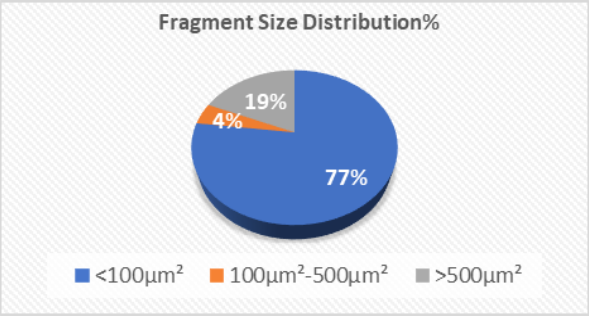


Figure 6. MP fragment size distribution of five different stations

It can be seen from the Fig. 5 data that the proportion of microplastics in shape fiber with the size of 100–200 μm is the most, and that of microplastics more significant than 200 μm is the least; at the same time, the fragments with the most widespread surface are <100μm² (Fig.6) (B. Peng et al., 2022). Because larger microplastics can produce several smaller fiber fragments during degradation, the enormous abundance is often small- and medium-sized microplastics.

Studies have shown that a liter of laundry wastewater may contain more than 100 kinds of plastic fibers, which municipal sewage pipes transport to rivers (Browne et al., 2011; Claessens et al., 2011). Human activities are along the Ishmi River and a national road along the river. These fibrous microplastics, which are more present in colors red, blue, and black, may come from wastewater from washing human clothes (Leslie et al., 2017), products of automobile tires, and suspended atmospheres (Claessens et al., 2011).

Compared with other rivers in different studies (Leslie et al., 2017; D. Zhang et al., 2021), the fibrous microplastics in Ishmi River did not originate from fishing activities because the whole river was closed to fishing because it is the most polluted river in Albania (Agjensia Kombetare e Mjedisit, n.d.).

The fragment microplastics may have come from frosted particles in bags and plastic containers and in people's cleaning products (Claessens et al., 2011). According to the different shapes and colors of MP detected in samples, we may accept that the primary sources of MP waste are packing cushions and insulation materials of various products, plastic products used in agricultural planting, such as agricultural film and impermeable plastic film, or they may be broken from improper treatment or discarded plastic bags(Zhou et al., 2020).

Many particles (other 23%) needed to be better identified. They could be covered with MP particles or quartz particles. (Rafael Paiva Nunes Junior, 2021) This is very important to know in the future and needs further investigation to improve methodology.

Polymer identification of microplastics

Fourier identified the type of polymer transform infrared spectroscopy (FT-IR), widely used to identify unknown particles due to its excellent reliability in determining the chemical constitution.

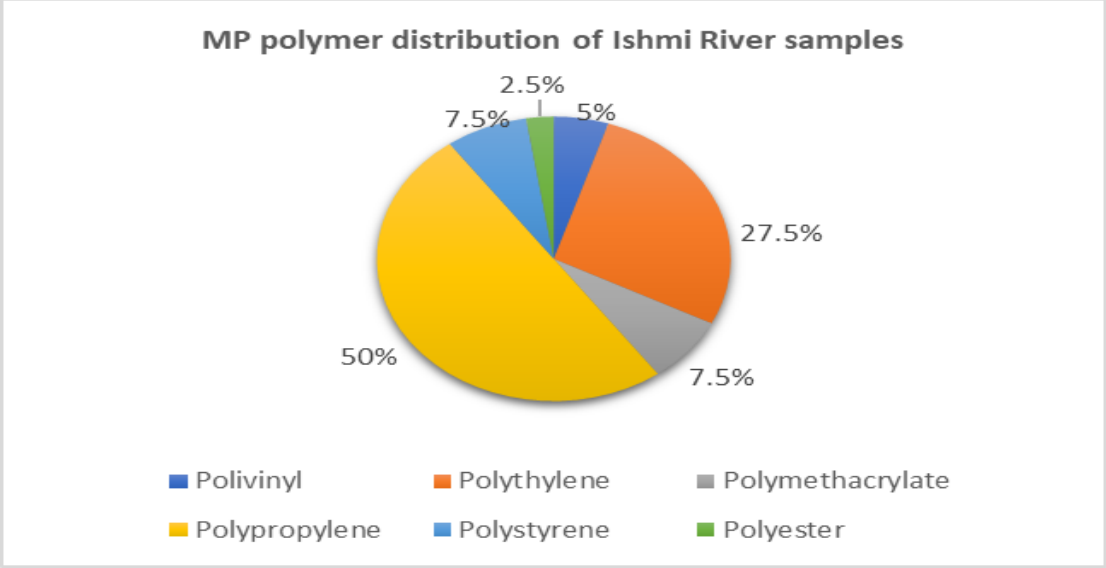
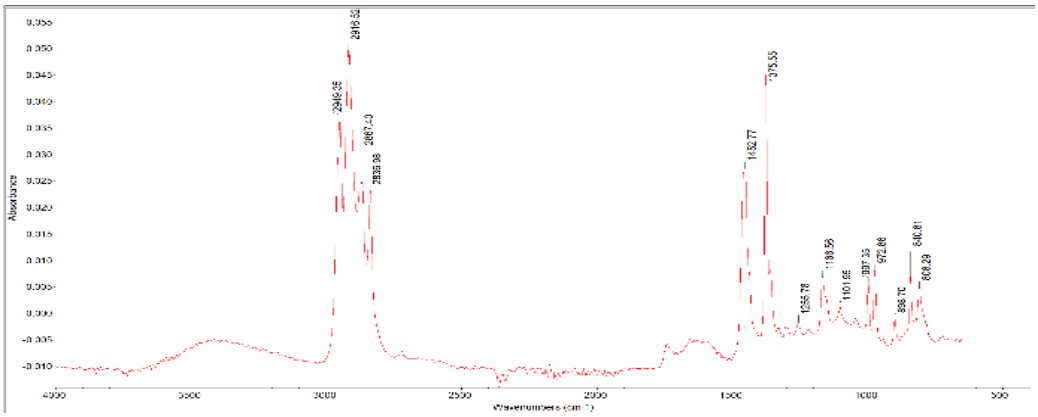
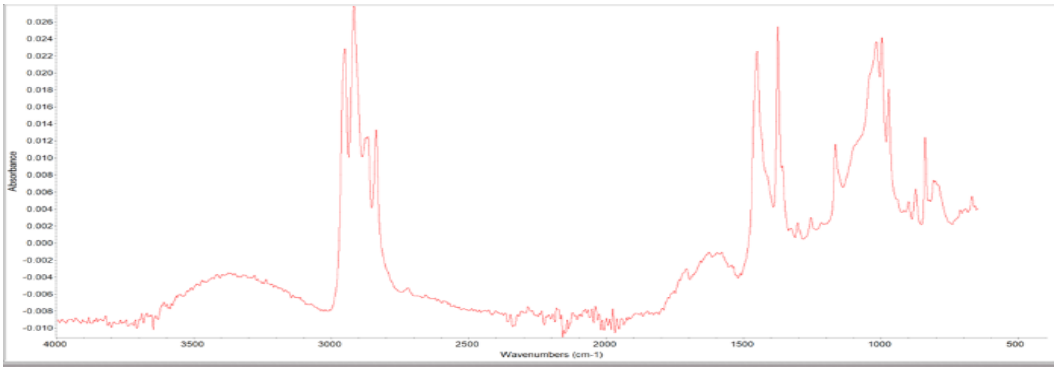


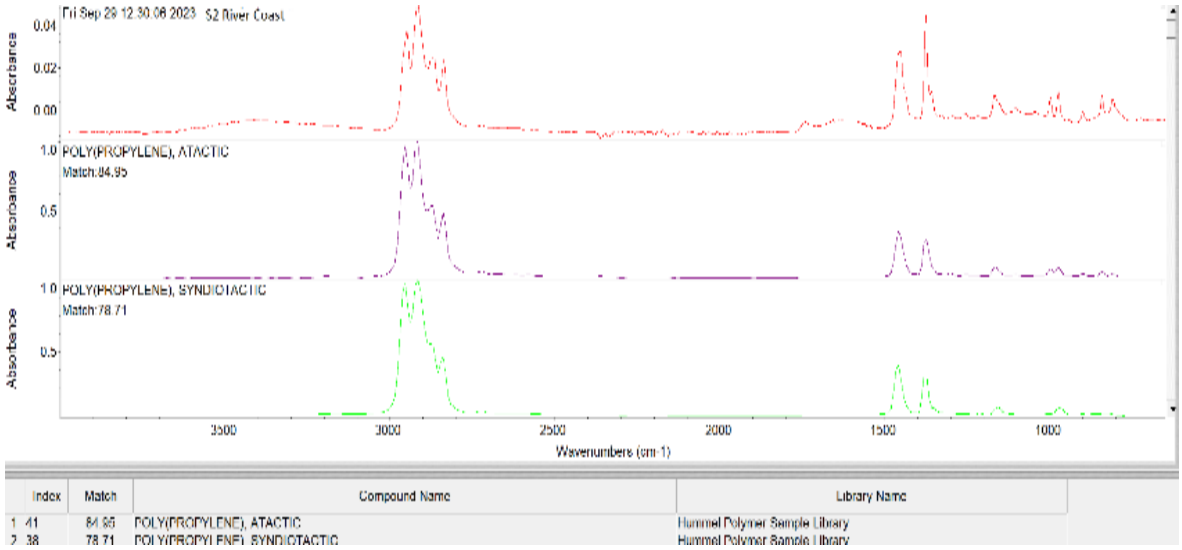
Figure 7. MP polymer distribution of Ishmi River samples



a)



b)



c)

Figure 2 a) Polypropylene FTIR absorbance spectra, b) Polyethylene FTIR absorbance spectra, c) Polypropylene compatibility FTIR library absorbance spectra.

Because of their low cost, these polymers (Yakushev et al., 2021) are the most common polymers used in packaging materials. The lower production costs make plastics an attractive option for many manufacturers.
Polyvinyl (Polyvinyl Chloride—PVC) (Ibor et al., 2023): PVC is versatile and often used in construction and packaging. It is typically considered cost-effective.

Polyethylene: PE is the most widely used plastics known for its low cost. It is commonly used in packaging, containers, and various other applications.

Polymethacrylate (Polymethyl Methacrylate - PMMA): PMMA, commonly known as acrylic, is used in products like plexiglass and is known for its optical clarity. While it may not be as inexpensive as other plastics, its cost can vary depending on the specific application.

Polypropylene: PP is a versatile and cost-effective plastic used in packaging, textiles, and other applications. Its relatively low production cost makes it popular in various industries.
Polystyrene: PS is affordable and widely used in packaging, disposable products, and insulation.
Polyester: PET is a diverse polymer used in textiles, packaging, and other applications.

After FTIR analysis, filter paper was analyzed using Raman spectroscopy. After using both techniques, the following table shows the polymer compatibility with the library.

Table 1. FTIR and Raman MP polymer compatibility.

Polymer type	FTIR match %	Raman Match%
Polyethylene	75,72% - 81,7%	96,1%
Polypropylene	79,29% - 86,3%	81,17%
Polystyrene	54,46% - 70,24%	68,2%
Polyvinyl chloride	46,93%	none
Polymethyl Methacrylate	62,87%	58,4%
Polyester	46,8%	none

The results show that the information provided by both techniques is highly credible. The exemption applies to polyvinyl chloride and polyester, which did not give any Raman match results.

Discussion

Our manual screening of bright-field microscopic images for areas with heterogeneous particle appearance revealed an overall abundance of microplastics in the samples at moderate levels. Intriguingly, a significant portion of particles turned out to be quartz particles, a finding unexpected as they were intended to be excluded during density separation. Substantial sand grains in the samples raise questions and need further investigation (Rafael Paiva Nunes Junior, 2021). Careful attention should be given to high-density salt, favoring higher-density options like NaI over NaCl. This strategic choice facilitates qualitative differentiation between microplastic waste and the remaining solution.

Acknowledgement. This work was supported by Institute of Applied Nuclear Physics, Tirana, Albania.

CONCLUSION

This case study underscores the critical need for spectroscopic techniques like FTIR in microplastic analysis. Specifically, FTIR spectroscopy and Raman spectroscopy emerged as up-and-coming methods for confirming the polymer origin of microplastic particles.

The microscopic method used to evaluate the residues was successful, as the microplastic residues (referred to in the literature) were visible. The type of polymer residues was identified from (Primpke et al., 2018; Schymanski et al., 2018), and the results were obtained from the two spectroscopic techniques, FTIR and Raman. A detailed evaluation of the samples found that the most widespread MP is in the form of fibers with a size of 100-200µm, while the fragments of the most evidenced waste are < 100µm². The polymers present in the samples are polypropylene, polyethylene, and polystyrene.

FTIR spectroscopy furnishes data across a broader spectrum of wavenumbers, encompassing fingerprint regions and single bonds, whereas Raman spectroscopy yields more detailed information within the fingerprint area. These two analytical techniques, FTIR and Raman, are complementary. When employed together, they offer a holistic understanding of a sample's chemical composition. The complementary nature of these techniques is advantageous in terms of sample compatibility. Some samples may be more suitable for one technique depending on factors such as sample transparency, fluorescence, or water content.

REFERENCES

1. Adomat, Y., & Grischek, T. (2021). Sampling and processing methods of microplastics in river sediments - A review. In *Science of the Total Environment* (Vol. 758). Elsevier B.V. https://doi.org/doi.org/10.1016/j.scitotenv.2020.143691;
2. Agjensia Kombetare e Mjedisit. (n.d.). *Environmental report, 2020*;
3. Bergmann, M., Gutow, L., & Klages, M. (n.d.). *Marine Anthropogenic Litter*.;
4. Browne, M. A., Crump, P., Niven, S. J., Teuten, E., Tonkin, A., Galloway, T., & Thompson, R. (2011). Accumulation of Microplastic on Shorelines Woldwide: Sources and Sinks. *Environmental Science & Technology*, *45*(21), 9175–9179. https://doi.org/10.1021/es201811s;
5. Claessens, M., Meester, S. De, Landuyt, L. Van, Clerck, K. De, & Janssen, C. R. (2011). Occurrence and distribution of microplastics in marine sediments along the Belgian coast. *Marine Pollution Bulletin*, *62*(10), 2199–2204. https://doi.org/10.1016/j.marpolbul.2011.06.030;
6. Filgueiras, A., & Pedrotti, M. L. (2019). *Standardised protocol for monitoring microplastics in seawater*. https://doi.org/10.13140/RG.2.2.14181.45282;
7. Hurley, R., Woodward, J., & Rothwell, J. J. (2018). Microplastic contamination of river beds significantly reduced by catchment-wide flooding. *Nature Geoscience*, *11*(4), 251–257. https://doi.org/10.1038/s41561-018-0080-1;
8. Ibor, O. R., Mpama, N.-O. L., Okoli, C. P., Ogarekpe, D. M., Edet, U. O., Ajang, R. O., Onyezobi, C. E., Anyanti, J., Idogho, O., Aizobu, D., & Arukwe, A. (2023). Occurrence, identification and characterization of plastic pollution from an open solid waste dumpsite in Calabar, Southern Nigeria. *Environmental Advances*, *11*, 100338. https://doi.org/10.1016/j.envadv.2022.100338;
9. Klein, S., Worch, E., & Knepper, T. P. (2015). Occurrence and spatial distribution of microplastics in river shore sediments of the rhine-main area in Germany. *Environmental Science and Technology*, *49*(10), 6070–6076. https://doi.org/10.1021/acs.est.5b00492;
10. Leslie, H. A., Brandsma, S. H., van Velzen, M. J. M., & Vethaak, A. D. (2017). Microplastics en route: Field measurements in the Dutch river delta and Amsterdam canals, wastewater treatment plants, North Sea sediments and biota. *Environment International*, *101*, 133–142, https://doi.org/10.1016/j.envint.2017.01.018;

11. Leusch, F. DL., Lu, H.-C., Perera, K., Neale, P. A., & Ziajahromi, S. (2023). Analysis of the literature shows a remarkably consistent relationship between size and abundance of microplastics across different environmental matrices. *Environmental Pollution*, *319*, 120984, https://doi.org/10.1016/j.envpol.2022.120984;
12. Li, J., Ouyang, Z., Liu, P., Zhao, X., Wu, R., Zhang, C., Lin, C., Li, Y., & Guo, X. (2021a). Distribution and characteristics of microplastics in the basin of Chishui River in Renhuai, China. *Science of The Total Environment*, *773*, 145591. https://doi.org/10.1016/j.scitotenv.2021.145591;
13. Li, J., Ouyang, Z., Liu, P., Zhao, X., Wu, R., Zhang, C., Lin, C., Li, Y., & Guo, X. (2021b). Distribution and characteristics of microplastics in the basin of Chishui River in Renhuai, China. *Science of the Total Environment*, *773*. https://doi.org/10.1016/j.scitotenv.2021.145591;
14. Löder, M. G. J., & Gerdts, G. (2015). Methodology used for the detection and identification of microplastics—a critical appraisal. In *Marine Anthropogenic Litter* (pp. 201–227). Springer International Publishing. https://doi.org/10.1007/978-3-319-16510-3_8;
15. Maddela, N. R., Ramakrishnan, B., Kadiyala, T., Venkateswarlu, K., & Megharaj, M. (2023). Do Microplastics and Nanoplastics Pose Risks to Biota in Agricultural Ecosystems? In *Soil Systems* (Vol. 7, Issue 1). MDPI. https://doi.org/10.3390/soilsystems7010019;
16. Marine Debris Program, N. (2015). *Laboratory Methods for the Analysis of Microplastics in the Marine Environment: Recommendations for quantifying synthetic particles in waters and sediments*;
17. Peng, B., Hossain, K. B., Lin, Y., Zhang, M., Zheng, H., Yu, J., Meng, X., Wang, J., Cui, Y., Wu, B., Lou, L., & Cai, M. (2022). Assessment and sources identification of microplastics, PAHs and OCPs in the Luoyuan Bay, China: Based on multi-statistical analysis. *Marine Pollution Bulletin*, *175*, 113351. https://doi.org/10.1016/j.marpolbul.2022.113351;
18. Peng, G., Xu, P., Zhu, B., Bai, M., & Li, D. (2018). Microplastics in freshwater river sediments in Shanghai, China: A case study of risk assessment in mega-cities. *Environmental Pollution*, *234*, 448–456. https://doi.org/10.1016/j.envpol.2017.11.034;
19. Prata, J. C., da Costa, J. P., Duarte, A. C., & Rocha-Santos, T. (2019). Methods for sampling and detection of microplastics in water and sediment: A critical review. In *TrAC - Trends in Analytical Chemistry* (Vol. 110, pp. 150–159). Elsevier B.V. https://doi.org/10.1016/j.trac.2018.10.029;
20. Primpke, S., Wirth, M., Lorenz, C., & Gerdts, G. (2018). Reference database design for the automated analysis of microplastic samples based on Fourier transform infrared (FTIR) spectroscopy. *Analytical and Bioanalytical Chemistry*, *410*(21), 5131–5141. https://doi.org/10.1007/s00216-018-1156-x;
21. Rafael Paiva Nunes Junior. (2021). *Multivariate statistics applied to microplastic analysis techniques*. Dissertação de Mestrado apresentada à Faculdade de Ciências da Universidade do Porto em Química;
22. Robertson, I. (2018). *Optimising the Workflow for Microplastic Analysis by FTIR Microscopy Microplastics Method Workshop*;
23. Schymanski, D., Goldbeck, C., Humpf, H.-U., & Fürst, P. (2018). Analysis of microplastics in water by micro-Raman spectroscopy: Release of plastic particles from different packaging into mineral water. *Water Research*, *129*, 154–162. https://doi.org/10.1016/j.watres.2017.11.011;
24. Vethaaka, A. D., & Legler, J. (2021). Microplastics and human health Knowledge gaps should be addressed to ascertain the health risks of microplastics. *Science Magazine*, *371*(6530);
25. Wagner, M., Scherer, C., Alvarez-Muñoz, D., Brennholt, N., Bourrain, X., Buchinger, S., Fries, E., Grosbois, C., Klasmeier, J., Marti, T., Rodriguez-Mozaz, S., Urbatzka, R., Vethaak, A. D., Winther-Nielsen, M., & Reifferscheid, G. (2014). Microplastics in freshwater ecosystems: what we know and what we need to know. *Environmental Sciences Europe*, *26*(1), 12. https://doi.org/10.1186/s12302-014-0012-7;
26. Yakushev, E., Gebruk, A., Osadchiev, A., Pakhomova, S., Lusher, A., Berezina, A., van Bavel, B., Vorozheikina, E., Chernykh, D., Kolbasova, G., Razgon, I., & Semiletov, I. (2021). Microplastics distribution in the Eurasian Arctic is affected by Atlantic waters and Siberian rivers. *Communications Earth & Environment*, *2*(1), 23. https://doi.org/10.1038/s43247-021-00091-0;
27. Zhang, D., Fraser, M. A., Huang, W., Ge, C., Wang, Y., Zhang, C., & Guo, P. (2021). Microplastic pollution in water, sediment, and specific tissues of crayfish (Procambarus clarkii) within two different breeding modes in Jianli, Hubei province, China. *Environmental Pollution*, *272*, 115939. https://doi.org/10.1016/j.envpol.2020.115939;

28. Zhang, W., Zhang, S., Wang, J., Wang, Y., Mu, J., Wang, P., Lin, X., & Ma, D. (2017). Microplastic pollution in the surface waters of the Bohai Sea, China. *Environmental Pollution*, *231*, 541–548. https://doi.org/10.1016/j.envpol.2017.08.058;
29. Zheng, Y., Li, J., Sun, C., Cao, W., Wang, M., Jiang, F., & Ju, P. (2021). Comparative study of three sampling methods for microplastics analysis in seawater. *Science of the Total Environment*, *765*. https://doi.org/10.1016/j.scitotenv.2020.144495;
30. Zhou, G., Wang, Q., Zhang, J., Li, Q., Wang, Y., Wang, M., & Huang, X. (2020). Distribution and characteristics of microplastics in urban waters of seven cities in the Tuojiang River basin, China. *Environmental Research*, *189*, 109893. https://doi.org/10.1016/j.envres.2020.109893;