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RESEARCH PAPER

Assess the microplastic prevalence and characterization in bivalve (*Lamellidens marginalis*): implications for freshwater ecosystems and human health

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Abstract

Pioneer research work is designed to assess the characterization of microplastic pollution in *Lamellidens marginalis,* freshwater bivalve, from Jhargram district of West Bengal, India. Bivalves, known as their ecological and economical importance, also act as bioindicator for various environmental contaminates like MPs and heavy metals. A total of 373 bivalve samples, with body weights ranging from 24 to 58 g and lengths between 4.5 and 8 cm, were analyzed. The average MP concentration in the gastrointestinal (GI) tract and muscle tissue was found to be 3.76 and 1.48 MPs/gm respectively. MPs were categorized by color primarily red, black, blue, green and white in decreasing order of frequency and by size ranging from 50 to 1000 µm, categorised into 50–100, 101–200, 201–300, 301–500, and 501–1000 µm groups. Morphotypes were identified including fibers, fragments and sheets in decreasing manner. Stereo microscopy and Raman micro-spectroscopy analysis revealed the highest MP abundance in study site III (SIII) and the lowest in SV, with notable spatial variation in the physical and chemical properties of MPs. This study is in unique in nature due to its comprehensiveness, particularly on polymer types, physical and chemical characteristics of microplastic. Findings of the present investigation will help for effective management strategies of microplastic pollution in near future.

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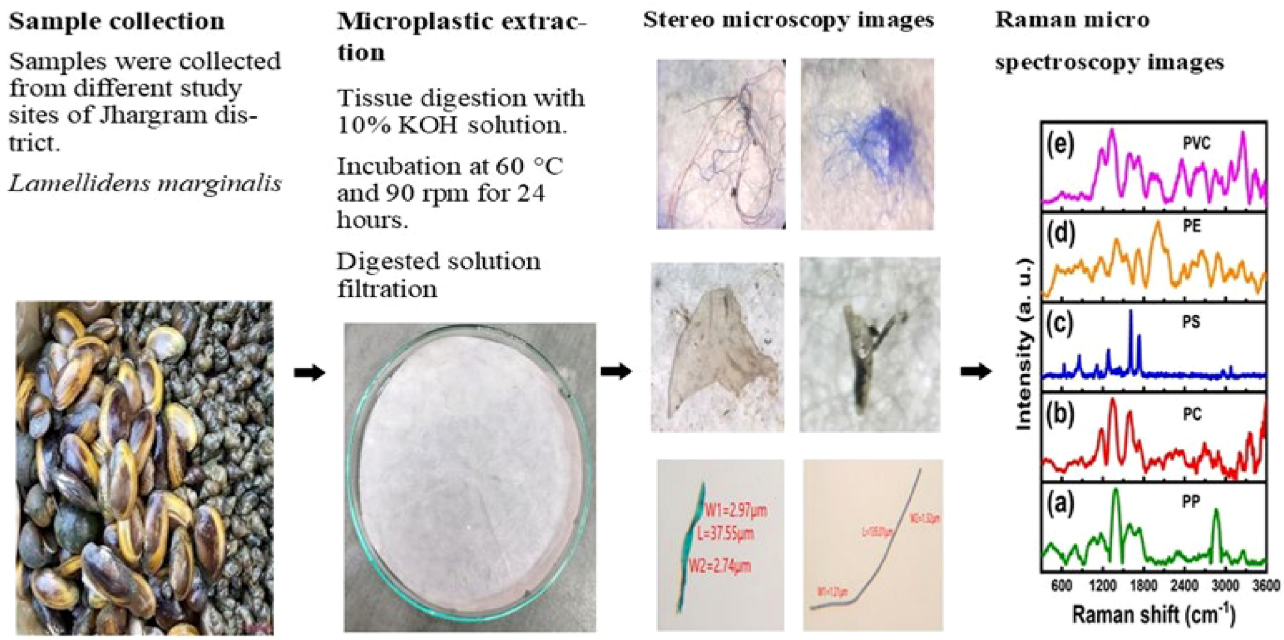
Graphical abstract



Keywords Plastic pollution · Freshwater bivalve · *Lamellidens marginalis* · Bio-accumulations · Human health

Introduction

The extensive usage of plastic in daily life, driven by its large-scale applications has led to a corresponding increase in plastic pollution. In present time, microplastics (MPs) have become ubiquitous pollutants in aquatic environments, causing significant threats to ecological systems. MPs, defined as plastic particles smaller than 5 mm, has convoked global attention due to its potential environmental impacts due to longer weathering period and widespread distribution, in aquatic as well as other ecosystems (Li et al. 2018; Skalska et al. 2020). Moreover, MPs can act as vectors that can facilitate the transport of pathogens, invasive species, heavy metals and other organic contaminants that adhere to their surfaces (Hermabessiere et al. 2017). Regardless of the fact that riverine ecosystems are approximately contributing up to 80% of the total plastic pollution entering into the world's oceans they remain largely unexplored (Meijer et al. 2021). The concentration of plastic waste has been rapidly increasing, primarily because of the design for single use plastic. Previous studies have found that 19–23 million metric tons (Mt) of plastic waste entered into aquatic ecosystems globally in the year of 2016, with prognosis indicating that this figure could rise to 53 Mt annually by 2030 (Smaal et al. 2019).

Children in developing countries like India and Africa, mainly under 5 years of age have been suffered from Protein Energy Malnutrition (PEM). Most frequent diseases are kwashiorkor and marasmus mainly found in children those who are suffer from PEM (Kumari 2017). To overcome this nutritive gap marine as well as freshwater bivalves would play an excellent role as they contain large number of proteins, vitamin B12, omega-3 fatty acids, zinc etc. (Wright and Kelly 2017). Bivalves including oysters, scallops or mussels can be collected from natural waterbodies (river, oceans) and also can be farmed in suitable environments. In India more than 50 species of mussels are distributed in the freshwater ecosystem, the genus *lamellidens* comprise nine species with two sub species. Among the genus, the most abundant freshwater mussel is *Lamellidens marginalis*. According to the IUCN, *L. marginalis* is classified as a "least concern" organism (Madhyastha et al. 2010). The meat yield percentage can vary from 16.92 to 21% in *L. marginalis* with crude protein, fat, moisture and ash content comprising 7.19, 1.64, 83.92 and 3.70% respectively. Additionally, it also acquires inorganic minerals like Mn, Cu and Zn with large amount of essential and non-essential amino acids like Glutamate in highest amount with 67.62%, Aspartate being second with 13.77%, then lysine with 5.11%, Serin with 4.26%, threonine with 2.52%, Arginine with 1.37%, methionine with 1.27% phenylalanine with 1.24%, valine, isoleucine, histidine, glycine, tyrosine, cysteine, proline, and nearly 32 types of fatty acids are also found. (Jamaluddin et al. 2016; Moniruzzaman et al. 2021; Sarma et al. 2022).

Because of the above-mentioned qualities and easy availability of *L. marginalis* it is widely used as edible muscle in Indian subcontinent. Bivalves not only provide significant nutritional benefits to humans but also have substantial economic impacts on local communities.

Bivalves also provide a food source for birds, serve as habitats for various marine species and play a critical role in carbon and nitrogen sequestration, also help in maintaining water quality (Smaal et al. 2019). However, due to their filter-feeding behaviour, bivalves are exposed to various contaminants like MPs. According to various field investigation, mussels from more than twenty different countries had MPs in them (Pazos et al. 2020; Li et al. 2020). The first line of exposure towards MPs through direct ingestion causing various physiological damage like injury or clogging of gills and gastrointestinal (GI) tract. MPs accumulation has been spotted in various organ like GI tract, soft tissue, valves, gills, haemolymph and hepatopancreas, in which fibers shaped MPs found to be prevalent one (Naji et al. 2018).

Approximately 16% of all animal protein consumed comes from seafood products (FAO 2013), which may be a significant source of MPs exposure for humans. Among seafood sources, bivalves may be the biggest source of MPs consumption for humans because they are always eaten as of whole. According to estimates, shellfish consumption in Europe might expose consumers to intake up to 11,000 MPs particles annually. According to (Van Cauwenberghe and Janssen 2014), consumers in countries with low shellfish consumption consume 1800 MPs particles annually on average, which is still a significant amount of exposure. According to (Smith et al. 2018), MPs can harm human health through toxicological pathways that include both chemical and physical impacts. According to Wright and Kelly 2017, the bio-persistence of MPs in human tissue can cause oxidative stress, inflammation, genotoxicity, apoptosis, and necrosis. Human lung tissue extracted from lung cancer patients has been shown to have MPs fibers (Pauly et al. 1998). Oxidative stress was identified as one of the mechanisms of cytotoxicity at the cellular level in a study of the effects of MPs on human brain and epithelial cells (Schirinzi et al. 2017).

Hence, Improved MPs management is necessary for bivalve, fisheries and food security. Aim of the current investigation is to determine the characterization of the MPs that bivalves consume are connected to variations in the sampling sites. Can these variations serve as a basis for utilizing bivalves as bioindicators of MPs pollution and whether fresh water bivalve species *L. marginalis* is fit to human consumption. This work comprises a combined study of field-based data collection and literature review of previous works to examine the proposed hypothesis, that are: 1. Quantitative variations in the concentration and abundance of different types (shape, size colour and polymer types) of MPs in freshwater bivalve *L. marginalis* across the selected study sites in respect to the spatial variation of MPs pollution level. 2. *L. marginalis* can be used as bioindicator for MPs pollution.

Material and methods

Study sites

Jhargram district has been selected as the study site for this work. On April 4, 2017, it is separated from Paschim Medinipur District to become the 22nd district of West Bengal. Jhargram district has its latitude and longitudinal extent at 21°52' N to 22° 48' N and 86°34' E to 87°20' E. It is located in the southwestern part of West Bengal. Sharing its borders with Jharkhand state to the west, Odisha to the south-west, Bankura district to the north, Purulia district to the north-west and Paschim Medinipur shares it's east and south-east bordar. The district has a total area size of 3,037.90 square kilometres (1,172.94 square miles) with 370/sq. kilometres of population density. There are eight Community Development blocks, with Jhargram as a subdivision, which also serves as the district headquarters. Kangsabati, Silaboti, Dulung and Keleghai river basins are situated in this district with receiving 160 cm annual rainfall and it holds numerous freshwater (ponds, lakes, streams, wetlands) ecosystem. (Mishra and Chandrakar 2023).

Over 40% of people in the Jhargram district are directly or indirectly dependent on agriculture, while 30% are engaged in collective activities related to forests. As a result, this region exhibits a lower level of social and economic development (Mishra and Chandrakar 2022). Hence a huge number of local people rely on aquatic resources not only for food but economic assistance also. Selected study sites for this work were: (1) Gopiballabpur (22.2059°N, 86.8947°E), (2) Jhargram Town (22.4562°N, 86.9944°E), (3) Jamda (22.3748°N, 87.0131°E), (4) Nayagram (22.0323°N, 87.1746°E), (5) Lalgarh (22.5833°N, 87.0499°E) and (6) Belpahari (22.6321°N, 86.7643°E) comprising both rural and urban areas of the district. Every study site receives a number of pollutants from various sources including industrial waste runoff, farmland runoff, domestic sewage, river discharge and one of the biggest sources of pollutants in Jhargram district is the tourist activities.

SI, Gopiballavpur, is a town and falls under gram panchayat area of Gopiballavpur I community development block situated on the banks of Subarnarekha River, although this place is developed, it is a busy town with prominent plastic pollution can be seen from various sources. SII, Jhargram, town is the headquarter of the Jhargram district it is under a municipality area and famous as a tourist destination, with multiple factories and mills like, sponge iron factory around 13 km away from town, Tara soap factory, Jhagram rice mill, Jhargram paper mill, Kanchan oil industry etc. causing a significant amount of air, water, soil and noise pollution from which MP and heavy metal pollution can definitely be seen in surrounding places. SIII, Jamda, approximately two kilometres from Jhargram town is under Jhargram sub-division, it's also a town area where locals of neighbouring villages sell bivalves in marketplace. SIV, Nayagram, is a village and a gram panchayat area falls under Nayagram Community development block. Even though the Subarnarekha River is close to Nayagram, the region is prone to drought. SV, Lalgarh, is also a gram panchayat area falls under Binpur I community development block and distance from district headquarter is 20 km. SVI, Belpahari is a village in Binpur community development block, it is a hill range area surrounded in the north by Kankrajhor hill and Subarnarekha River situated in the south. Because of the diversified natural beauty, this place attracts a huge number of tourist and plastic pollution due to tourism is prominent in many places, it is mostly village area where locals collect and sell small fishes and edible molluscan including bivalves.

During the monsoon season in June and July of 2023, commercially important bivalve species *L. marginalis* were collected from of the above mentioning study sites. These six study sites with their ten subsites of each were chosen to provide an inclusive representation of the conditions within the study region (Fig. 1).

Sample collection

In the month of June and July of 2023, total of 373 *L. marginalis* commercially important bivalve more specifically mussel specimens were collected from the selected study sites. After being properly wrapped in aluminium foil and placed in Ziplock bags, the live samples were brought to the laboratory in cooler boxes and kept there at − 20 °C for the subsequent MPs examination. All lab surfaces and equipment were thoroughly cleaned with ethanol and distilled water prior to processing. After being carefully cleansed with water, the bivalves were patted dry using paper towels (Sathish et al. 2020). Shell length of each sample was measured on a centimetre scale, with lengths ranging from 4.5 to 8 cm and the weights of each sample was recorded

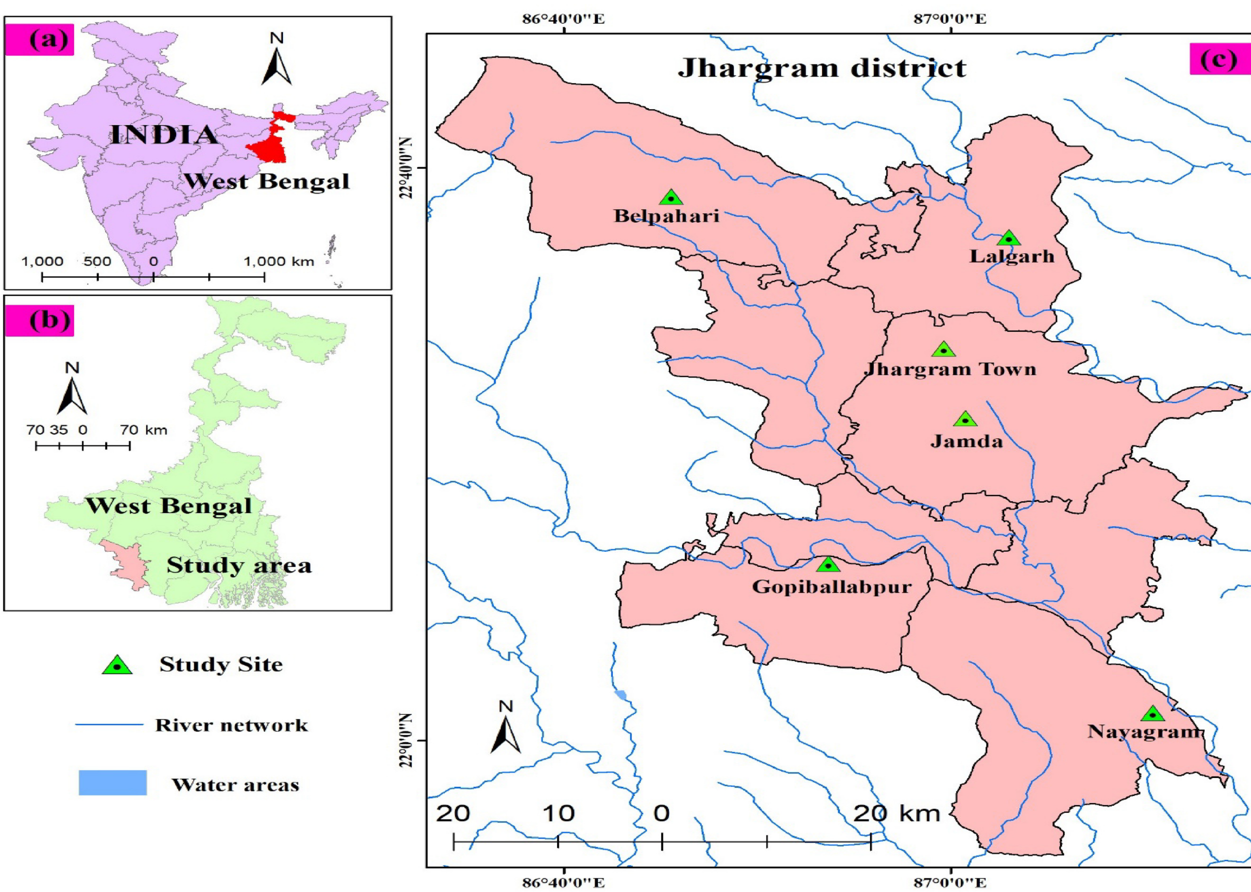


Fig. 1 Map of Study area. **a** India, **b** West Bengal. **c** Jhargram district with six sampling sites

separately using a digital weighing balance (weighing balance), with weights ranges between 24 and 58 g. After the shell water was completely drained from each sample a clean scalpel and a pair of sessors were used to dissect the GI tract and other muscle tissues separately. The wet weight of the GI tract and muscle tissues was recorded individually.

Microplastic extraction

MP extraction from GI tract and other muscle tissue of bivalves was performed using an established procedure by (Ding et al. 2018; Song et al. 2022). The GI tract and other muscle tissues of bivalves were carefully extracted and weighed. Two separate 250 mL conical flask containing the individual GI tract and muscle was filled with 100 mL of 10% KOH for each prepared sample. After that, the conical flasks were quickly wrapped in aluminium foil, sonicated for 5 min, and then put in a shaking incubator at 60 °C at 90 rpm for 24 h. After digestion, the solution was filtered through a Whatman GF/F glass fiber membrane (47 mm in diameter, 0.7 µm pore size) without cooling. The filter membrane was then put in Petri dishes and allowed to heat at 60 °C for 24 h (Joyce and Falkenberg 2023) before further analysis.

Quality control

According to Ding et al. (2021), standard precautions were taken to avoid external contaminations, including double cleaning of glass containers and counter tops with distilled water and 70% ethyl alcohol, wearing nitrile gloves and a 100% cotton lab coat throughout all the procedures. Carefully filtering solutions, and covering every instrument with aluminium foil were done when not used. Every sample has been treated in a laminar flow and properly washed with Milli-Q water both before dissection. The identification of MPs was carried out in a closed laboratory with a glass cover over the stereomicroscope was placed. Blanks with the same volume of 10% KOH but no tissues were processed concurrently with the samples to adjust for procedural contaminations. Furthermore, the blanks were placed straight into the air to identify the airborne contaminants while the suspected MPs were found and identified using a stereomicroscope and Raman spectroscopy. Less than half of the blank conical had one or two MP fibers. Because the average number of MPs in the blanks was less than 1, the results were qualitatively taken into account when interpreting the data but were not deducted from the experimental outcome.

Physical characterization by stereomicroscopy study:

A Dewinter stereomicroscope was used to visually inspect the dried filter papers at 40 × magnification. To conduct a comprehensive analysis, the filters were separated into four quadrants and each quadrant was inspected twice (Ramaremisa et al. 2024). The shape (fibers, pieces, or sheets) and color (red, black, blue, green, and white) of the suspected particles were used to categorize them. When visual inspection proved inadequate, hot needle test method was used to confirm the presence of MPs (Hurt et al. 2020). Using the BSI Lab software, images of MPs were taken under a stereo microscope along with their length and width based on observations, MPs were categorized into different length and width groups. The quantity of MPs was quantified separately for gut contents and muscle tissues. Visual observations showed that fibers were the most prevalent type of MP found in the sample, followed by fragments and sheets. Among the MPs that were seen, black and red MPs predominated, followed by blue, white, and green MPs. Depending on the study site, characteristics and abundance of MPs vary substantially.

Chemical characterization by Raman microspectrometer

Following the visual identification under a stereo microscope, extracted MPs were analysed using a Raman microspectrometer for chemical characterization or polymer identification. A subset comprising approximately 20–25% of the extracted particles was selected for spectroscopic examination. The selection process was random but it prioritizes the morphological prevalence. More specifically, the analysed MPs included approximately 87 fibers (ranging from 50 to 1000 µm), 60 fragments (ranging in between 100 and 1000 µm) and 52 sheets (ranging from 100 to 1000 µm). For conducting Raman spectroscopy analysis, MPs were carefully transferred from filter paper onto a scotch tape already attached to a glass slide. A glass coverslip was then placed over the sample to avoid contamination. Each MP was first examined under the stereomicroscope and then observed using the microspectrometer for Raman characterization. (Malloggi et al. 2024a, b).

Raman spectroscopy has been performed in WiTec made, model Alpha300R with 532 nm and 10mw of LASER power. Regular calibration of the wavelength was performed by focusing on a silicon wafer and analyzing its first photon band. To enhance signal sensitivity and prevent polymer degradation, acquisition parameters were adjusted for each particle type. The Raman microspectrometer was controlled using LabSpec 6 software (Ramaremisa et al. 2022). The chemical composition of the MPs and their additives was determined by comparing reference and sample spectra using the SLOPP Library of MPs and the KnowItAll® Informatics System software (John Wiley & Sons, Inc., New Jersey, U.S.A.) (Ramaremisa et al. 2022).

Statistical analysis

Here, MPs abundance in freshwater bivalves *L. marginalis* was measured in terms of MPs per gram for muscle tissue as well as for digestive tract or gut tissue (MPs/gm). Furthermore, in-depth investigations were carried out to describe the MPs characteristics according to their shape, size, colour, and polymer type across the selected study sites. These characteristics may help in identifying different types of possible MPs contamination sources in the environment.

Result and discussion

Quantitative analysis of MPs in commercially important bivalve

Bivalves and other filter-feeding organisms are good bioindicators for a variety of environmental pollutants because they exhibit bioaccumulation of different pollutants, including MPs, high concentration of MPs can be seen in bivalves because of their filter-feeding behaviour and limited excretion ability (Su et al. 2022). In this study, we compare six study sites residing in Jhargram district, West Bengal, India. The concentration and abundance of MPs, as well as their physical and chemical properties in *L. marginalis*, vary significantly between the study sites. A total of 373 *L. marginalis* was collected from the six study sites, and 757 MPs were extracted for further investigation. By examining the physical parameters of bivalves *L. marginalis*, the average length, width, total body weight and tissue weight were presented as mean ± SD (Table 1).

If we consider each study site for these values than the SI sites have the average body length of 7.37 ± 0.60 cm, width of 3.23 ± 0.59 cm, average total body weight was 47.10 ± 11.09 gm, and mean tissue weight was around 3.85 ± 0.32 gm. In SII, average body length of 6.17 ± 1.48 cm, width of 3.17 ± 0.47 cm, average total body weight was 38.53 ± 6.41 gm, and mean tissue weighted around 3.65 ± 0.40 gm. In SIII, average body length of 6.53 ± 0.91 cm, width of 3.50 ± 0.62 cm, average total body weight was 40.83 ± 10.36 gm, and mean tissue weight was 3.74 ± 0.48 gm. In SIV, average body length of 6.50 ± 1.32 cm, width of 3.40 ± 0.53 cm, average total body weight was 42.33 ± 7.52 gm, and mean tissue weighted around 3.99 ± 0.55 gm, In SV, average body length of 6.41 ± 1.25 cm, width of 3.23 ± 0.68 cm, average total body weight was 46.67 ± 5.65 gm, mean tissue weight was around 4.30 ± 0.27 gm. In SVI, average body length of 5.67 ± 1.26 cm, width of 3.03 ± 0.58 cm, average total body weight was 30.87 ± 3.57 gm, and mean tissue weight was around 3.67 ± 0.37 gm. The bivalve samples were dissected for the separation of gut content and muscle sample for better understanding of MPs accumulation trend and concentration rate at different body parts of *L. marginalis*. During filter feeding bivalve use their gills, by the combine effect of water current and ciliary movement of gills filter food particles with contaminants enters the GI tract (Moreschi et al. 2020). Therefore, gills and GI tract come directly in contact with the water and pollutants hence pollutant accumulation in those part should become more prominent while absorption of contaminants like MPs can also be seen in different tissue layers (Moreschi et al. 2020).

We also found quite a difference in our findings where concentration of MPs in gut content is quite higher than the other muscle tissues, such as in SI, average concentration of MPs in gut content is 3.81 MPs/gm, in SII 4.45 MPs/gm, in SIII 6.28 MPs/gm, in SIV 3.45 MPs/gm, in SV 1.38 MPs/gm and in SVI 2.05 MPs/gm, While the concentration of MPs in other muscle tissues were 1.50 MPs/gm in SI, in SII 1.94 MPs/gm, in SIII 1.82 MPs/gm, in SIV 1.20 MPs/gm, in SV 0.66 MPs/gm, and in SVI 1.11 MPs/gm (Fig. 2). Thus, it signifies that primary accumulation occurs in the GI tract and then absorption to different organs occurs in *L. marginalis*. In support of this finding, reported that MPs and NPs (Nano plastics) have been found in various parts

Table 1 Physical characteristics (length, width, total body weight, tissue weight) with separately dissected gut and muscle weight and microplastic concentration in *L. marginalis* in different study sites

Study sites	Mean shell length (cm)	Mean shell width (cm)	Mean bivalve weight (gm) (shell + soft tissue)	Mean soft tissue weight (gm)	Total dissected muscle and gut weight (gm)		MP/gm	
					Gut	Muscle	Gut	Muscle
SI (Gopiballabpur)	7.37 ± 0.60	3.23 ± 0.59	47.10 ± 11.09	3.85 ± 0.32	21.8	32.6	3.81	1.5
SII (Jhargram Town)	6.17 ± 1.48	3.17 ± 0.47	38.53 ± 6.41	3.65 ± 0.40	22.9	34.5	4.45	1.94
SIII (Jamda)	6.53 ± 0.91	3.50 ± 0.62	40.83 ± 10.36	3.74 ± 0.48	22.6	31.2	6.28	1.82
SIV (Nayagram)	6.50 ± 1.32	3.40 ± 0.53	42.33 ± 7.52	3.99 ± 0.55	24.3	29.8	3.45	1.2
SV (Lalgarh)	6.41 ± 1.25	3.23 ± 0.68	46.67 ± 5.65	4.30 ± 0.27	24.5	28.6	1.38	0.66
SVI (Belpahari)	5.67 ± 1.26	3.03 ± 0.58	30.87 ± 3.57	3.67 ± 0.37	25.8	27.9	2.05	1.11

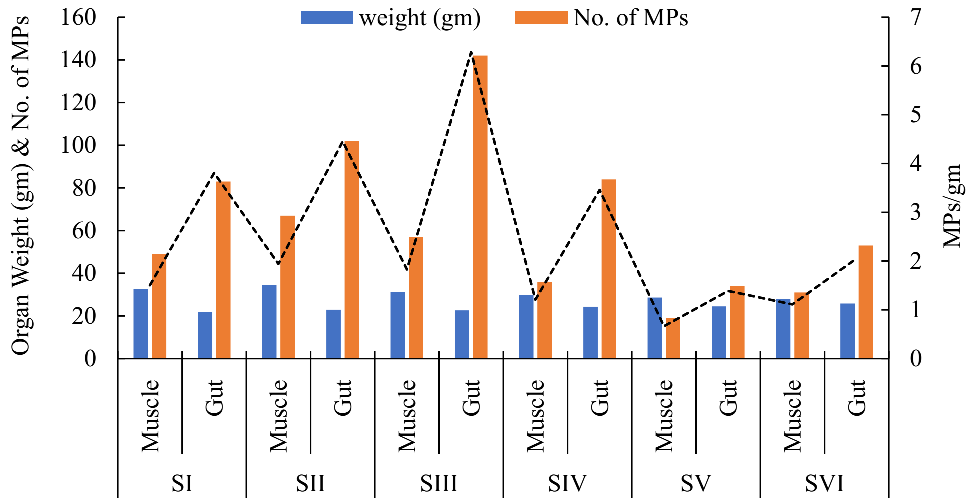


Fig. 2 Comparation of dissected GI tract (gut) and muscle tissue weight with extracted MPs/gm in different study sites

of bivalves while the main focus resides on digestive tract (Merzel et al. 2020). Some other studies also proposed the presence of MPs in foot, mantle, visceral mass, muscle and hepatopancreases in bivalves (Gaspar et al. 2018; Li et al. 2020). With the difference of MPs concentration in various body parts of *L. marginalis,* variance in MPs concentration in different study sites also recorded. Findings of this study showed that MPs concentrations differed considerably among the several study sites and body parts of *L. marginalis*, indicating that anthropogenic activities and surrounding environmental conditions affecting MPs presence in different study sites and accumulated by *L. marginalis*. As all bivalve samples were of the same species and collected in similar seasonal duration, hence the differences in MPs abundance emphasize on the role of various site-specific factors, such as pollutant concentration in the environment more specifically in the freshwater ecosystem, sources of MPs as well as anthropogenic activities in determining MPs distribution. This study shows the efficacy and importance of bivalves as bioindicators for site-specific pollution monitoring.

The concentration of MPs in the gut content of bivalves was substantially higher than that in the muscle tissue in each study site, suggesting a greater tendency for MPs to accumulate in the digestive system. SIII (6.28 MPs/gm) showed the highest concentration of MPs within bivalve gut content followed by SII (4.45 MPs/gm), SI (3.81 MPs/gm), SIV (3.45 MPs/gm), SVI (2.05 MPs/gm) and SV (1.38 MPs/gm), respectively. While SII (1.94 MPs/gm) showed the highest concentration in case of dissected muscle tissue, followed by SIII (1.82 MPs/gm), SI (1.50 MPs/gm), SIV (1.20 MPs/gm), SVI (1.11 MPs/gm) and SV (0.66 MPs/gm). These results imply that MPs are not only site-dependent but also concentrate more in particular tissues, such the gut. This could be due to variations in the exposure pathways or retaining durations of MPs within bivalve bodies in different habitats.

Physical characteristics of MPs

MPs are known to be plastic particles smaller than 5 mm in diameter, still it exhibits various physical and chemical characteristics like different shape (fiber (filament), fragment, film (sheet, flake), and sphere (beads, pellet) (Kolandhasamy et al. 2018), size (1 nm to 5 mm in diameter) (Berglund et al. 2019), color (transparent, black, blue, red, green, yellow, grey, purple etc.) (Berglund et al. 2019) and polymer types (some common polymer types are: PVC, PET, PE, PP, PS, EVA, PA, PC, PTT, CA, PBA, PUR, PO, PEG, PMMA, PTFE etc.) (Chakraborty et al. 2023).

MPs obtained from different study sites were analysed for physical characteristics using a stereo microscope. Additionally, the results show a significant difference between them. In this study, MPs were categorized by shape primarily into three types, fibers, fragments, and sheets (Fig. 3). The most prevalent shape was fibers, occupying 44.70% in SI, 48.52% in SII, 46.73% in SIII, 52.50% in SIV, 49.06 in SV and 44.05% in SVI followed by fragments exhibiting the second most abundant shape exhibiting 31.82% in SI, 34.91% in SII 23.62% in SIII, 31.67% in SIV, 32.08% in SV and 34.52% in SVI. Although sheets were less frequent, but their presence can also be seen for instance in SI 23.48%, in SII 16.57%, in SIII 29.65%, in SIV 15.83%, in SV 18.87% and finally in SVI 21.43% (Fig. 4) (Table 2). We can divide MPs into two categories based on their formation. Firstly, the primary MPs that are deliberately created for use in various kinds of industries, cosmetic and personal care products, secondly the secondary MPs that are produced from large plastic particles by natural or anthropogenic degradation process (Lozano et al. 2021). Hence it explains the various kinds of shapes exhibiting by MPs. And the abundance of fibrous MPs in bivalves can be explained by their retention and accumulation traits. According to (Sathish et al. 2020) fiber-shaped MPs have slower excretion rates than other

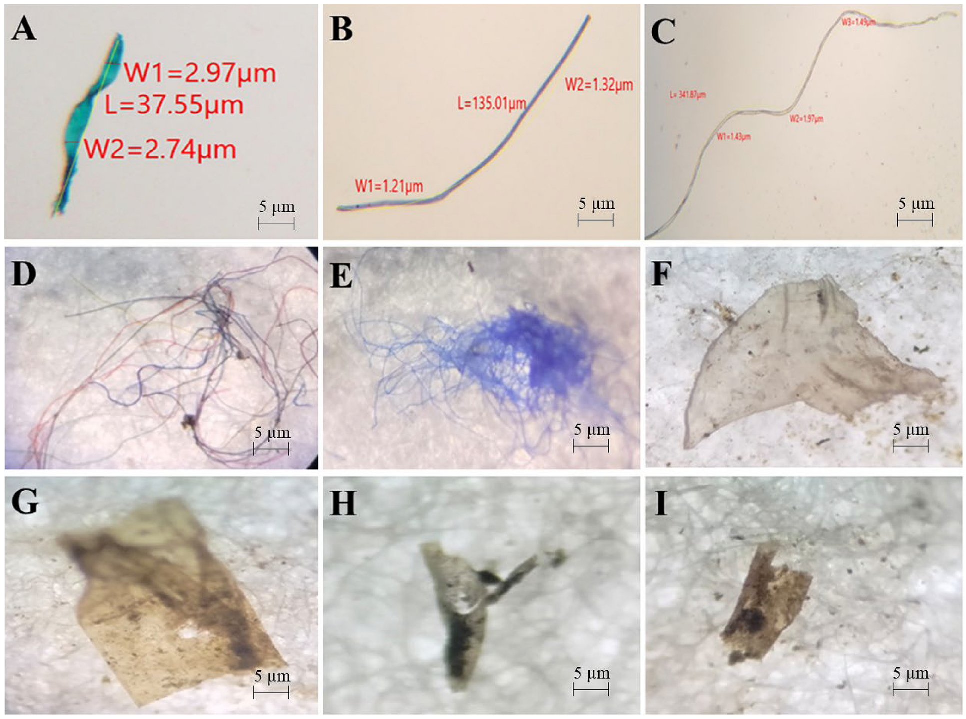


Fig. 3 Stereomicroscopic view of MPs. Where, **a, b, c** represent the length and width of MPs, **d, e** fibrous types of MP, **f, g** fragment shaped of MPs, H,I: sheet type of MPs

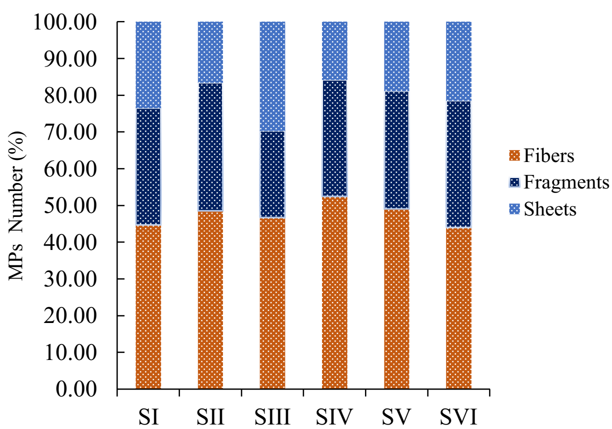


Fig. 4 Shape (Fibers, Fragments and sheets) distribution of MPs in different study sites

shape of MP, which let them stay longer in bivalve bodies. In gills and hepatopancreatic system, fibers tend to accumulate and are more difficult to remove through normal excretory mechanisms (Renzi et al. 2018). Because fibrous MPs are frequently found in common sources such discarded fishing nets, broken rope, and sewage wastewater particularly in coastal areas, environmental variables also have a role in their occurrence in bivalves (Wang et al. 2019; Sathish et al. 2020).

MPs also exhibit wide range of size difference in this study. Majority of the MPs resides in the 101–200 µm class, while 501–1000 µm class was the least abundant in case of each study site. In study sites SI, 17.42% of MPs resides in 50–100 µm class, 43.94% resides in 101–200 µm class, 14.39% residing in 201–300 µm class, 18.18% in 301–500 µm class and 6.06% in 501–1000 µm class. In SII, 20.12% MPs present in 50–100 µm class, 34.91% present in 101–200 µm class, 16.57% in 201–300 µm class, 23.08% on 301–500 µm class and finally 5.33% present in 501–1000 µm class. In SIII, 24.62% present in 50–100 µm class, 40.20% resides in 101–200 µm class, 18.59% resides in 201–300 µm class, 11.06% in 301–500 µm class and 5.53% present in 501–1000 µm class. In SIV, 20.83% MPs present in 50–100 µm class, 42.50% resides in 101–200 µm class, 19.17% can be seen in 201–300 µm class, 13.33% in 301–500 µm class and 4.17% resides in 501–1000 µm class. In SV, 22.64% MPs present in 50–100 µm class, 39.62% present in 101–200 µm class, 16.98% are in 201–300 µm class, 15.09% in 301–500 µm class and 5.66% present in 501–1000 µm class and finally in case of SVI 26.19% MPs present in 50–100 µm class, 32.14% present in 101–200 µm class, 20.24% in 201–300 µm class, 13.10% resides in 301–500 µm class and finally 8.33% resides in 501–1000 µm class (Fig. 5), based on MPs length then the findings of Sathish et al. (2020) supported this result. Some previous study on various invertebrate revealed that, this size preference is likely due to selective ingestion, as smaller MPs particle exhibit similarity to their natural food sources (Leslie et al. 2013; Van Cauwenberghe et al. 2015). These documentations demonstrate that because of higher availability and easy incorporation smaller particle (10–1000 µm) ingestion become more frequent and longer fibers exhibit potential

Table 2 Microplastic concentration with their shape, size, colour and polymer variation in different bivalve species across the world

Study sites	Bivalve species	Microplastic concentration (MP/g or MP/Individual)	Microplastic characteristics (shape, length, colour)	Reference
Jhargram, West Bengal, India	*Lamellidens marginalis*	3.76 MPs/gm in gut and 1.47 MPs/gm in muscle tissue	Shape (fibers, fragments and sheets); Length (50–1000 µm with predominancy of (101–200 µm); Colour (red, black, blue, green and white)	Present study
Santa Catarina Is land Bay, Brazil	Pacific oyster (*Magallana gigas*), brown mussel (*Perna perna*), and scallops (*Nodipecten nodosus*)	Oysters (4.11 ± 2.67 items/ind.); mussels (1.63 ± 0.74 items/ind.); scallops (1.44 ± 1.42 items/ind	Shape (fibers and fragments); Length (72.5 to 5138.1 µm with predominance of 1000–5000 µm); Colour (blue black, gray, red, green and orange)	Brocardo et al. (2025)
Southwest coast of India (Kochi, Munroe Island, Ashtamudi lake, coastal region in Kollam distric, Enayam beach, Kanyaku mari)	*Villorita cyprinoides, Crassostrea madrasensis, Paphia mala barica, Perna perna*	15.33 ± 15.56 MP/gm	Shape (fibers, fragments, films, beads/pellet) Length (10 µm to > 1000 µm with predominancy of > 1000 µm followed by 500–1000 µm); Colour (transparent, blue, black	Abisha et al. (2024)
Beyhan Dam and Hydroelectric Power Plant, Turkey	Zebra mussel (*Dreissena polymorpha*)	3.47 ± 2.62 MP/gm (highest) and 1.06 ± 0.97 MP/gm (lowest)	Shape (fibers and fragments); Length (1000 µm -2000 µm); Colour (black, blue, grey and red)	Atamanalp et al. (2023)
Mumbai, Ratnagiri, and Sindhudurg beaches of Maharashtra	Yellow clams (*Meretrix casta*)	55.04 ± 32.15 MPs items/g in soft tissue and 138.61 ± 94.74 MPs items/individual	Shape (fibers, fragments, films, pellet); Length (11 µm to 3.8 mm, 9 µm to 2.88 mm, and 20 µm to 1.94 mm yet mostly belong to 1–100 µm range); Colour (translucent, black, red, brown, blue, green)	Naidu et al. (2022)
Qingdao, China	Scallop (*Chlamys farreri*), mussel (*Mytilus galloprovincialis*), oyster (*Crassostrea gigas*) and clam (*Ruditapes philippinarum*)	0.5 and 3.3 items/individual or 0.3 and 20.1 items/gm of soft tissue	Shape (fiber, fragment, film, and granule); Length (< 100 µm to 5000 µm); Colour (transparent, blue, black, white, gray, red, pink)	Ding et al. (2021)
Ariyankuppam, Panithittu, and Chunnambar in Pondicherry, India	*Perna viridis* and *Meretrix meretrix*	(0.18 ± 0.04, 1.84 ± 0.61, and 1.76 ± 0.48) MPs/gm and (0.50 ± 0.11, 1.75 ± 0.35, and 4.80 ± 1.39) MPs/individual	Shape (micro-fragments); Length (majority smaller than 100 µm); Colour (not mentioned)	Dowarah et al. (2020)
Seoul, Gwangju, and Busan in South Korea	Oyster (*Crassostrea gigas*), mussel (*Mytilus edulis*), Manila clam (*Tapes philippinarum*) and scallop (*Patinopecten yessoensis*)	Manila clam (0.34 ± 0.31 particles/gm, 0.03–1.08 particles/gm); oyster (0.07 ± 0.06 particles/gm, 0–0.19 particles/gm); scallop (0.08 ± 0.08 particles/gm, 0.01–0.17 particles/gm); mussel (0.12 ± 0.11 particles/gm, 0–0.35 particles/gm	Shape (fragments, fibers, films); Length (< 100 µm to 300 µm)	Cho et al. (2019)

Table 2 (continued)

Study sites	Bivalve species	Microplastic concentration (MP/g or MP/Individual)	Microplastic characteristics (shape, length, colour)	Reference
Germany	*Mytilus edulis; Crassostrea gigas*	*Mytilus edulis* (0.36 ± 0.07 particles / gm); Crassostrea gigas (0.47 ± 0.16 particles/gm)	Shape (fibre- and particle-shaped); length (5–10 µm, 11–15 µm, 16–20 µm, 21–25 µm and > 25 µm); Colour (red, blue, green)	Van Cauwenberghe and Janssen (2014)

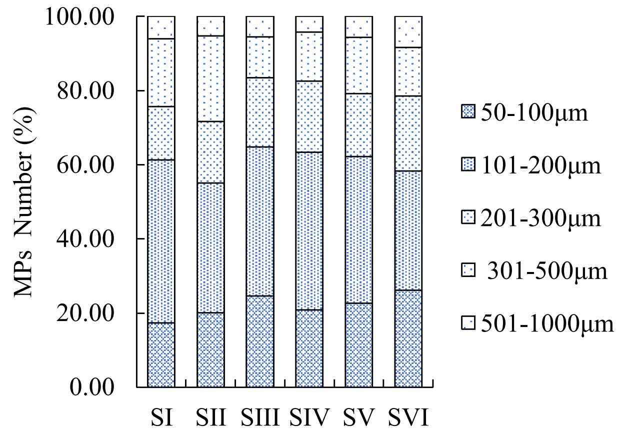


Fig. 5 Size wise distribution of MPs in different study sites

to become entangled in various parts of the GI tract, causing difficulty in excretion (Atamanalp et al. 2023). These results provide insight into why fiber-shaped and smaller MPs are more common in bivalve bodies by highlighting physiological and environmental factors as drivers of fiber retention (Table 2).

While considering MPs color, throughout the study sites, black and red were the most dominant colors found within bivalve *L. marginalis*. In SI, 27.27% red, 24.24% blue, 24.24% black, 9.85% green and 14.39% white colored MPs were found. In case of SII, 31.95% red, 20.12% blue, 26.63% black, 11.24% green and 10.06% white colored MPs were found. In SIII, 29.65% red, 18.59% blue, 29.15% black, 12.06% green and 10.55% white colored MPs are present. In SIV, 24.17% red, 19.17% blue, 32.50% black, 14.17% green and 10.00% white MPs are prevalent. In SV, 20.75% red, 16.98% blue, 32.08% black, 20.75% green and 9.43% white colored MPs were observed and finally in SVI, 32.14% red, 17.86% blue, 26.19% black, 10.71% green and 13.10% white MPs were seen (Fig. 6). Some other studies also support the prevalence of dark colour MPs mainly black, blue and red in their report, but in contrast many studies also reported that the white, transparent or light colour MPs were predominant in various bivalves (Ding et al. 2021; Hoellein et al. 2021). Hence some study claim that bivalves have color preference while ingesting their food and some claim that they cannot uptake fibers based on the colour. Different sources of plastics commonly found in coastal areas may help to explain the frequent occurrence of these colours. It is more likely that these polymers will enter aquatic ecosystems as contaminants as they are often coloured for specific uses (Table 2).

Chemical characteristics of MPs

Raman spectroscopy, despite its complex physics, is now a user-friendly method for quick quantitative analysis, aided

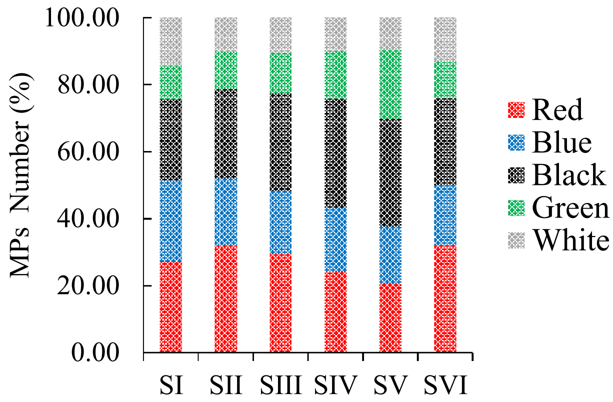


Fig. 6 Colour (Red, Black, Blue, Green and white) distribution of MPs in different study site

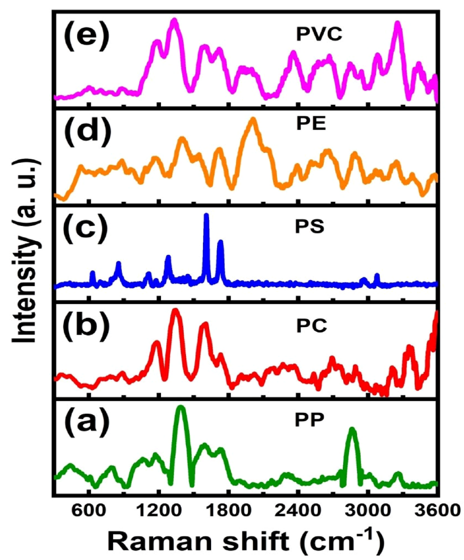


Fig. 7 Raman shifts of polymer types in *Lamellidens marginalis* of the current investigation

by automated tools and algorithms. Sample signals can be identified by comparing them with databases, and no extra processing is needed to distinguish polymer types if only qualitative analysis is required. MPs fall into different categories: polyethene (PE), polypropylene (PP), polystyrene (PS), Polycarbonate (PC) and Polyvinylchloride (PVC) with distinct signals typically appearing between 300 and 3600 cm^{-1} (Fig. 7). In the Raman spectrum (Fig. 7a), six distinct peaks appeared at key fingerprint regions: 458, 809, 1040, 1167, 1371, and 2871 cm^{-1}. These peaks closely match the characteristic peaks of PP. Specifically, the 458 cm^{-1} peak corresponds to the stretching of the C–C vibration, while the peaks at 809, 1040, 1167, 1371, and 2871 cm^{-1} are associated with C–C skeletal, C–H rocking vibration in plane, C=C stretching vibration, C–H bending vibration, and C–H stretching, respectively (Table 3). Five distinct peaks were observed in the Raman spectrum (Fig. 7b) at specific fingerprint regions: 888, 1178, 1603, 1774, and 2885 cm^{-1}. These peaks align closely with the characteristic peaks of PC. The peak at 888 cm^{-1} corresponds to the stretching of the [O–(C=O)–O] band. Peaks at 1178, 1603, 1774, and 2885 cm^{-1} are attributed to C–H in-plane bending, C=C stretching, C–N stretching, and C–H stretching, respectively (Table 3). In the Raman spectrum (Fig. 7c), six distinct peaks appeared at key fingerprint regions: 631, 858, 1170, 1280, 1614 and 1726 cm^{-1}. These peaks closely match the characteristic peaks of PS. Specifically, the 631 cm^{-1} peak corresponds to the stretching of the C–C vibration, while the peaks at 858, 1170, 1280, 1614 and 1726 cm^{-1} are associated with C–H bending vibration out-of-plane in the benzene ring, C–H twisting vibration in-plane, C–H bending vibration, C=C stretching vibration, respectively (Table 3).

In the Raman spectrum (Fig. 7d), four distinct peaks appeared at key fingerprint regions: 1132, 1429, 1729 and 2881 cm^{-1}. These peaks closely match the characteristic peaks of PE. Specifically, the 1132 cm^{-1} peak corresponds to the stretching of the C–C vibration within the polyethylene chain, characteristic of the polymer's skeletal structure while the peaks at 1429, 1729 and 2881 cm^{-1} are associated with CH_2 bending or scissoring vibrations due to polyethylene's methylene groups, C=O stretching vibration due to oxidation and due to C–H stretching in the CH_2 groups, respectively (Table 3). In the Raman spectrum (Fig. 7e), five distinct peaks appeared at key fingerprint regions: 610, 694, 1325, 1724 and 2911 cm^{-1}. These peaks closely match the characteristic peaks of PVC. 610 and 694 cm^{-1} peaks corresponds to the stretching of the C–Cl vibration, while the peaks at 1325, 1724 and 2911 cm^{-1} are associated with CH_2 twisting or wagging vibrations within the polymer chain, C=O stretching, C–H stretching in the CH_2 groups, respectively (Table 3). Table 4 summaries the previously reported data to supported our current investigation.

Despite the fact that MPs contamination in bivalves has been extensively documented, there is a lack of quantitative data about the possible harm that MPs can cause to human health. Based on the concentration of MPs found in bivalves, a number of previous studies estimated how much MPs humans consume each year through their usual diet (Wang et al. 2021; Zhang et al. 2022). According to Cho et al. (2019), Italy is estimated to have the highest yearly dietary exposure to MPs from mollusc intake (11,970 items/person/year) followed by Canada, China and Franch with (8407 items/person/year), (6636 items/person/year) and (1139 items/person/year) respectively with various other countries with less concentration. Hence the potential chemical and particle toxicity of MPs affecting human health should not be overlooked. Accordingly, a

Table 3 Summaries of all functional groups found in Raman spectra in different microplastic

Microplastic type	Raman peaks (cm^{-1})	Band assignment	Reference
PP	458	Stretching of the C–C vibration	Nava et al. (2021)
	809	C–C skeletal	
	1040	C–H rocking vibration in plane	
	1167	C=C stretching vibration	
	1371	C–H bending vibration	
	2871	C–H stretching	
PC	888	Stretching of the [O–(C=O)–O] band	Zimmerer et al. (2019)
	1178	C–H in plane bending	
	1603	C=C stretching	
	1774	C=O stretching	
PS	631	Stretching of the C–C vibration	Alam et al. (2019); Fang et al. (2021)
	858	C–H bending vibration out-of-plane in the benzene ring	
	1170	C–H twisting vibration in-plane	
	1280	C–H bending vibration	
	1614	C=C stretching vibration	
	1726	C=C stretching vibration	
PE	1132	Stretching of the C–C vibration within the polyethylene chain	Lenz et al. (2015), Absher et al. (2019), Dong et al. (2020), Ripken et al. (2021)
	1429	CH_2 bending or scissoring vibrations due to polyethylene's methylene groups	
	1729	C=O stretching vibration due to oxidation	
	2881	C–H stretching in the CH_2 groups	
PVC	610	Stretching of the C–Cl vibration	Dong et al. (2020), Ripken et al. (2021)
	694	Stretching of the C–Cl vibration	
	1325	CH_2 twisting or wagging vibrations within the polymer chain	
	1724	C=O stretching	
	2911	C–H stretching in the CH_2 groups	

hazard ranking model was created for polymer hazard risk evaluation based on the chemical composition of polymers (Lithner et al. 2011). This model has been applied to evaluate the possible health hazards associated with MPs in human body (Fang et al. 2019; Ding et al. 2020). In this study PVC, PE, PS, PC and PP were found where PVC exhibits the highest hazard score of 10,551 posing the highest-level V of hazard score that can cause carcinogenicity, reproductive toxicity in human, followed by PC in level IV causing skin sensation, skin corrosion, acute toxicity, reproductive toxicity, serious eye damage and specific target organ toxicity-single exposure. PS, PE and PP falls under level II, II and I respectively causing various harmful effect on human health including acute toxicity, eye irritation, skin irritation for PS, specific target organ toxicity-single exposure for PE more specifically it can be seen in both low-density polyethylene (LDPE) and high-density polyethylene (HDPE) and finally PP exhibit flammable gaseous nature for the propylene monomer present in it (Lithner et al. 2011).

Conclusion

In the present study, we investigated the occurrence and prevalence of Microplastics (MPs) in the commercially significant freshwater bivalves *L. marginalis* within the Jhargram district of West Bengal, India. Finding of the present work are significantly dependent on pollution level of selected study sites, and eventually accumulation of MPs within tissues of *L. marginalis* due to their feeding behavior.

The observations in MP concentration across spatial locations highlights the intricate interactions between environmental inputs and biological responses. These findings further emphasize the utility of *L. marginalis* as a potential bioindicator for assessing microplastic pollution in freshwater ecosystems. The species' ability to bioaccumulate MPs, coupled with its widespread commercial and ecological significance, positions it as a valuable tool for monitoring aquatic ecosystem health and human exposure to plastic-derived contaminants through the food chain.

Table 4 Different types of MPs in various bivalve species with their identification method around the world

Study Site	Bivalve species	Microplastic Identification method	Polymer types	Reference
Jhargram district, West Bengal, India	*Lamellidens marginalis*	Raman micro spectroscopy	PVC, PP, PE, PC, PS	Present study
Tuscan coast, Italy	*Donax trunculus*	Raman spectroscopy	PP, PE, PET	Malloggi et al. (2024a, b)
North Ionian Sea	*Mytilus galloprovincialis*	ATR-FTIR spectroscopy, Micro-Raman spectroscopy, and SEM–EDS Analysis	LDPE, PVB, PBMA	Miserli et al. (2023)
Panguil Bay, Southern Philippine	*Meretrix meretrix, Donax sp. Katelysia hiantina*	ATR-FTIR Spectroscopy	ABS, PET, CA, PE, PS, PP, PVC	Bonifacio et al. (2022)
Sal estuary, Goa	*Paphia malbarica, Perna viridis and Crassostrea sp.*	μ-FTIR Spectroscopy	Polyacetylene, Polyacrylamide, PVC, EVOH	Saha et al. (2021)
Shazikou and Huangdao, China	*Chlamys farreri, Mytilus galloprovincialis, Crassostrea gigas, Ruditapes philippinarum*	μ-FTIR Spectroscopy	PVC, PET, CPE, PVDF, PVDC-PE, PVE, PE, PEI, PVDC-PAN	Ding et al. (2021)
Hong Kong	*Perna viridis*	Raman microspectroscopy	PP, PET, PS, PE	Leung et al. (2021)
Coast of Gulf of Mannar, India	*Donax cuneatus*	FTIR-ATR Spectroscopy	PE, PP, PP-PE, PET, PA, PEST, PVC, PS, RA, PVA	Sathish et al. (2020)
Ariyankuppam, Chunnambar and Panithittu estuaries in Puducherry coastline	*Perna viridis and Meretrix meretrix*	Nile red staining and fluorescent microscope quantification and Raman Spectroscopy	PU, PVC, PVCA, PES, PET, ABS, SBR, PVK and PEVA	Dowarah et al. 2020
Fishery markets of Seoul, Gwangju, Busan. South Korea	*Crassostrea gigas, Mytilus edulis, Tapes philippinarum, Patinopecten yessoensis,*	μ-FTIR	PE, PS, PP, PET, PVC, PTFE, PEVA, PPS, SAN, SEBS	Cho et al. (2019)
Northern Bavaria, Germany	*Unio pictorum*	Raman microspectroscopy	PP, PVC, PET	Domogalla-Urbansky et al. (2019)

Given the substantial implications of MP contamination on human health and the ecological balance of aquatic habitats, this study calls for more detailed investigations into the mechanisms of MP bioaccumulation. Future research should prioritize the investigation of potential sources of MPs pollution, their degree and mode of contamination, and also their long-term impacts on bivalve physiology and ecosystem dynamics. Such focused studies are essential to comprehensively understand the relationship between surrounding environmental conditions and the extent of MPs contamination in freshwater bivalves.

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Data availability The datasets used and analysed during the current study are available from the corresponding author on reasonable request.

Declaration

Conflict of interest The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Ethics approval Not applicable.

Consent for publication Not applicable.

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