

Authentication of Microplastic Accumulation in Customary Fruits and Vegetables

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Abstract

The environment has become a major source of plastic pollution. Microplastics have been well documented in aquatic ecosystems as an increasing pollutant of worldwide relevance, but little is known about their effects on the terrestrial environment, particularly in agroecosystems. Researches have recently proven that microplastics can affect human health and are found in human organs and tissues. In this present study, two different types of fruits like grapes (*Vitis vinifera*) and banana (*Musa paradisiaca*), vegetables like brinjal (*Solanum melongena*) and potato (*Solanum tuberosum*) were collected from local market in Trichy, Tamil Nadu, India and analysed for microplastics accumulation. Further, we identified the microplastic size through stereomicroscope, in grapes and banana the microplastic size was 0.002 mm and 0.01 mm respectively as well as vegetables like potato and brinjal the microplastic size was 0.002 mm and 0.01mm. The identified microplastics had been chemically characterized by FT-IR and viewed through SEM.

1. Introduction

Plastic pollution has become a major scientific and societal problem. Because of the rapid increase in plastic manufacture and consumption. Despite a gradual growth in plastic recycling and energy recovery, the majority of plastic wastes are released into the environment on a global scale (Blasing and Amelung, 2018). Microplastics (MPs) have recently been the subject of environmental research due to their small size and potential impact on terrestrial ecosystems. Microplastic is described as a plastic with a diameter of less than 5 mm and comes in a range of morphologies, including beads, fragments, fibres, and films. MPs are divided into two categories: primary MPs, which are formed in small quantities and discharged into the environment, and secondary MPs, which are degraded from bigger plastic wastes due to water, wind, sunshine, and other environmental factors. Primary MPs are released directly into the environment, whereas secondary MPs are formed by the breakdown of large-size chunks. Microplastics can enter agroecosystems through a variety of routes, including fertiliser coatings (Heuchan *et al.*, 2019), wastewater irrigation (Zhang and Liu, 2018), compost addition and biosolids application (Nizzetto *et al.*, 2016; Weithmann *et al.*, 2018), and, most importantly, the use of mulching film (Nizzetto *et al.*, 2016). (Liu *et al.*, 2014; Qi *et al.*, 2018). In general, MPs decomposition in soil is extremely slow, requiring hundreds, if not thousands, of years to complete (Zubris and Richards, 2005; Andrady, 2011).

Microplastics can enter the body of an animal, particularly a human, through ingestion and inhalation, where they can accumulate in numerous organs and cause health problems, such as cell damage or inflammatory and immunological problems (Dick Vethaak *et al.*, 2021). Many toxic and dangerous substances are found in microplastics, including bisphenol A, phthalates, antiminitroxide, brominated flame retardants, polyfluorinated chemicals, and others, all of which pose a major risk to human health and the environment. Different human health issues such as eye irritation, vision loss, breathing difficulties, respiratory problems, liver dysfunction, cancers, skin diseases, lungs problems, headache, dizziness, birth effect, reproductive, cardiovascular, genotoxic and gastrointestinal problems are all linked to the use of toxic plastics (Ram proshad *et al.*, 2018).

Agricultural systems are the final beneficiaries of a variety of contaminants, including microplastics, whose impacts are relatively unclear (Razzaghi *et al.*, 2018). In general, there is a lack of information of material fate and detrimental consequences in the agricultural system, which leads to food chain failure and an unknown channel of human exposure. Microplastics, depending on their size and type, can penetrate the seed, root, culm, leaves, and fruit cells (Dietz and Hertz, 2011). There is very few microplastic accumulation studies on regular consumed fruits and vegetables so we focused in this present study.

Currently, lacking of information on this subject due to the absence of data about the microplastics presence in edible fruits and vegetables. So we decided and applied our method to evaluate the microplastics <5mm presence in common edible fruits and vegetables. The aim of this study is to investigate the presence of microplastics present in regularly consumed vegetables like brinjal (*Solanum tuberosum*) and potato (*Solanum melongena*) and fruits of grapes (*Vitis vinifera*) and banana (*Musa paradisiaca*) and also identified and characterized the microplastics through various analytical methods.

2. Materials And Methods

2.1 Sample Collection and Preparation

Two types of samples like fruits (Grapes and Banana) and vegetables (Brinjal and Potato) were collected from local market at Tiruchirapalli, Tamil Nadu, India. Samples were put into cleaned aluminium tray and kept in closed conditions. Collected samples were sliced in the same day by using sterilized steel knife. The sliced fruits and vegetables samples were kept on clean sterilized petri plates. Then the samples were dried in hot air oven, at 60°C temperature for one day. The dried samples were powdered using clean sterilized mortar and pestle. Then grained samples were air dried for four days after that samples were extracted. Only glass equipment and containers were utilised, any plastic material and any product with an inorganic carbon chemical structure (containers, caps, pipettes, filters, holders) were avoided. Still no standardized protocol for extracting microplastics in fruits and vegetables. Therefore, we implemented a methodology (Fabio Corradini *et al.*, 2019).

The dried fruit and vegetable samples were weighed at 5g and deposited in glass centrifuge tubes with a capacity of 50 ml. Each tube was added 20 ml of deionized water, and the samples were swirled for 30 seconds. The supernatant was filtered using Whatmann No.42 filter paper after centrifugation at 2000 rpm for 15 minutes. The precipitate was agitated and centrifuged after 20 ml of sodium chloride was added. The supernatant was filtered through the same Whatmann No.42 filter paper for the second time. For the final extraction, 20 ml of a concentrated zinc chloride solution was added to the centrifuge tubes with the precipitate. The samples were swirled for 30 seconds since the ZnCl₂ solution had a higher viscosity than the other solutions. The supernatant was filtered using Whatmann No.42 filter paper after centrifugation at 2000 rpm for 15 minutes. After that, filter paper were placed on Petri dish for optical inspection in stereo microscope (Fabio Corradini *et al.*, 2019).

2.2 Identification of Microplastics

2.2.1 Stereomicroscopic Analysis

The extracted microplastic particle was taken into stereomicroscopical analysis. Under a magnifying lamp, samples were optically sorted and inspected under a stereomicroscope. (Leica M50) by the following method of Sarah piehl *et al.*, 2018.

2.3. Characterization of Microplastics

2.3.1 FT-IR Analysis

Fourier transform infrared spectroscopy can provide a distinct infrared spectrum for a specific chemical bond. Various samples have different bond compositions, so its is used to identify an unknown substance by comparing its spectrum with the spectra of known materials. FT-IR has become one of the most widely utilised techniques in chemical characterization of microplastics due to its great dependability. The visible microplastic particles in fruits and vegetables extracted samples were taken for FT-IR analysis (Perkin Elmer, Spectrum RX, USA). Spectra in the frequency range of $4000-400\text{ cm}^{-1}$ was used to record the spectrum of the extracted microplastics (Jung *et al.*, 2018).

2.3.2 Scanning Electron Microscope Analysis

The extracted microplastic particles were taken by the previously defined method (Fabio corradini *et al.*, 2019). The samples were gold-coated on the surfaces of microplastic particles while micrographs were taken. After fixation, the samples were placed in a deep vacuum and examined using a scanning electron microscope (SEM) (ZEISS EVO, Carl Zeiss microscopy). The SEM photograph had revealed that the microplastic present in the samples. The SEM micrograph also showed at high power of the electron beam.

3. Results

3.1 Sample Collection and Preparation

This study reports were revealed the presence of microplastics in regular intake of edible fruits and vegetables. Fruits and vegetable samples were collected and dried to sieved for extraction of microplastics particles.

3.1 Identification of Microplastics

3.1.1. Stereomicroscopic Analysis

The microplastic accumulation in fruits and vegetables sample was identified through stereomicroscope. In stereomicroscope microplastic size was measured, grape fruit microplastic size is 0.002 mm and

banana 0.01 mm and vegetables samples like potato microplastic size is 0.002 mm and brinjal size is 0.01 mm (Figure.1).

3.2 Characterization of Microplastics

Extracted microplastics were transferred to the FT-IR spectra to determine the different chemical groups were present in fruits and vegetables. In banana sample has shown spectral range at 3444.68 cm^{-1} , 1633.55 cm^{-1} refers to C=O stretching, 693 cm^{-1} peak range represent aromatic CH and 1271.00 cm^{-1} peak range refers to C-N stretching. In grapes sample has shown spectral range at 3436.06 cm^{-1} and 1633.82 cm^{-1} refers to C=O stretching, 1270.76 cm^{-1} refers to NH bending. In potato sample has shown spectral range at 3435.84 cm^{-1} and 1634.11 cm^{-1} represents C=O stretching, 684 cm^{-1} represents C=O bending. In brinjal sample has shown spectral range at 3435.82 cm^{-1} C=O stretching, 1120.93 cm^{-1} and 696 cm^{-1} peak range represent acrylonitrile butadiene styrene (ABS) group (Table.1 and Figure 2).

3.3. Scanning Electron Microscope Analysis

Electron images of microplastic particles (a, b, c, d) were shown in figure.3. Resulting spectra of the SEM analysis demonstrated the presence of microplastics presence in fruits and vegetables. In SEM results were show that microplastic size was found in fruits like grapes $2\text{ }\mu\text{m}$ and banana is $10\text{ }\mu\text{m}$. Vegetables like potato sample microplastic size is $2\text{ }\mu\text{m}$ and brinjal microplastic size is $10\text{ }\mu\text{m}$.

4. Discussion

Microplastics have long been recognised as a serious environmental problem due to their widespread use and slow decomposition. In our experiment we detected the microplastics accumulation in fruits and vegetables. Two types of fruits and vegetables were taken and analyzed for microplastics accumulation. It is the preliminary study for accumulation of microplastics.

In this study, we have detected the MPs at $2\text{ }\mu\text{m}$ and $10\text{ }\mu\text{m}$ size in fruits and $2\text{ }\mu\text{m}$ and $10\text{ }\mu\text{m}$ size in vegetables respectively. The size of the MPs is less than 5mm in diameter a similar study was reported by Dyachenko *et al.* (2016) they stated the presence of MPs in secondary wastewater treatment plant effluent through stereomicroscope. Sarah Piehl *et al.* (2018) have reported the presence of MPs in soil through stereomicroscopic analysis. Gea Oliveri Conti *et al.* (2020) have been detected microplastics at the size $1.51\text{ }\mu\text{m}$ in carrot and $2.52\text{ }\mu\text{m}$ in lettuce. Qingrunliu *et al.* (2020) had stated the presence of MPs in fruits and vegetables as following in apple at the size $2.17\text{ }\mu\text{m}$, common pear at size $1.99\text{ }\mu\text{m}$ broccoli at size $2.10\text{ }\mu\text{m}$, potato at size $1.51\text{ }\mu\text{m}$ through SEM. These are the similar results reported in recent research related to the size of the MPs.

Mecozzi *et al.* (2016) have been reported that nylon group peak was identified and shows C=O stretching in 3350 cm^{-1} to 3600 cm^{-1} at FT-IR spectra analysis. Similar results were found in our banana, grapes, potato and brinjal samples at spectral range 3444.68 cm^{-1} , 3436.06 cm^{-1} , 3435.84 cm^{-1} , 3435.82 cm^{-1} refers to C=O stretching resulted nylon polymer, spectral peak at $1250\text{--}1350\text{ cm}^{-1}$ shows C-N stretching

resulted high density polyethylene terephthalate polymer group present on it. Similar peaks were found in our banana sample shows peak range 1271.00 cm^{-1} has C-N stretching it refers to high density polyethylene terephthalate polymer group. At 1050 cm^{-1} - 1150 cm^{-1} peak range C-N stretch was found and refers to polyethylene oxide group. Same results were found in banana sample at range 1121.33 cm^{-1} shows C-N stretch it refers to polyethylene oxide group. At peak range 1050 cm^{-1} - 1150 cm^{-1} C-N stretch was found and refers to polyethylene oxide group. In brinjal same results found in the peak range 1120.93 cm^{-1} and refer to polyethylene oxide group.

Jung *et al.* (2018) have reported that 1634 cm^{-1} peaks was present and it represents nylon group. Moreover, our results were similar in banana sample the peak range was 1633.55 cm^{-1} and shows C=O stretching and it represents nylon group and 694 cm^{-1} peak range represent aromatic CH out of plane bending belongs to polystyrene group eventhough Similar result was found in banana sample at peak range 693 cm^{-1} . In grapes, potato and brinjal 1633.55 cm^{-1} , 1270.76 cm^{-1} , 1270.83 cm^{-1} , 1633.82 cm^{-1} , 1634.11 cm^{-1} peak ranges are found it shows C=O stretching that represents nylon group. The peak range 698 cm^{-1} was representing aromatic CH out-of-plane bending resulted acrylonitrile butadiene styrene (ABS) group. Similar results were found in brinjal sample at peak range 696.45 cm^{-1} and represents acrylonitrile butadiene styrene (ABS) group.

Qingrun Liu *et al.* (2021) have reported microplastics enter the soil ecosystem mostly through mulching and littering. After mulching, film and other wastes decompose into microplastics/nanoplastics (M/NPs), which are subsequently transported to plants (fruits and vegetables) and animals (meat and milk) via sewage sludge, composting, and irrigation. Microplastics is easily penetrate through seeds, roots, stems, leaves, fruits and plant cells because of their size and type under controlled conditions the uptake of microplastics has been reported. MPs can pass through plant cell walls and membrane barriers, obstruct cell pores and cause cell death that can be reported by Raza Ullah *et al.* (2021). Li *et al.* (2019) had reported polystyrene MPs can be transfer and accumulated in the roots of raw vegetables and transmitted from root to shoot tissues. Ng *et al.* (2018) had reported microplastic uptake, translocation and accumulation differ from one plant species to the other, depending on anatomical and physiological characteristics. Li *et al.* (2019) reported polystyrene MPs can be transfer and accumulated in the roots of raw vegetables and transmitted from root to shoot tissues. Polystyrene MPs can be transferred and deposited in the roots of fresh vegetables and they can also be passed from root to shoot tissues. Root and xylem features, growth rate, transpiration, water and lipid fractions, tonoplast and plasma membrane potential, and the pH of vacuoles and cytoplasm are some of the plant characteristics that influence microplastic uptake.

While, pesticides, herbicides and fertilizers are the main sources of microplastics accumulation in plant and soil was reported in Raza Ullah *et al.* (2021). Pietra *et al.* (2019) reported microplastics were present in mineral fertilizer. Allen *et al.* (2019) reported that the treated wastewater and biosolids are applied as fertilizer for irrigation purposes so this is the way also microplastics entry into the plants. Nicolas *et al.*

(2018) have reported organic fertilizer as a vehicle for microplastic entry into the environment, as well as composts, digestates and percolate-leachates from digestion, all of which are used as liquid fertilizer.

Enyoh Christian Ebere *et al.* (2019) have reported humans could consume 80 g of microplastics per day through plants as a food source. Once microplastics penetrate the plants and accumulate, they can serve as a potential route for entering the food chain, where they might bio magnify and cause serious health problems in humans. Zhang *et al.* (2020) have investigated the impact of MPs on agricultural soil and discovered that higher MP concentrations resulted in increased soil mobility, posing potential dangers to crop plants and humans.

5. Conclusion

The present study was mainly focused on microplastic accumulation in edible fruits and vegetables. We investigate the presence of microplastics in vegetables (potato and brinjal) and fruits and to identified and characterized the microplastics group through various analytical methods like FTIR and SEM. This study recommends more studies to examine the risks of human exposure to microplastics due to ingestion of agricultural products. In addition, agricultural impacts of microplastics can be complicated by their ability to absorb other chemicals, and the events of climate change due to anthropogenic factors, which future studies could aim to investigate. Government organisations and health authorities must act quickly to enact and implement environmental rules that will oversee the manufacturing, use, and disposal of plastics.

Declarations

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Tables

Table 1. Spectroscopic bands are the identification of the plastic typologies

S.No.	Sample Name	Bond	Spectral range of the bands (Cm ⁻¹)	Microplastic Group
1.	Grapes	C=O stretching	3436.06 cm ⁻¹ , 1633.82 cm ⁻¹	Nylon
		NH bending	1270.76 cm ⁻¹	
2.	Banana	C=O stretching	3444.68 cm ⁻¹ 1633.55 cm ⁻¹ 1271.cm ⁻¹ 1121 cm ⁻¹ 693 cm ⁻¹	Nylon High density polyethylene terephthalate (HDPET) Polyethylene oxide Polystyrene
3.	Potato	C=O stretching	3435.84 cm-1 1634.11 cm-1 684 cm -1	Nylon
		C=O bending		
4.	Brinjal	C=O stretching	3435.82 cm-1 1120.93 cm-1 696.45 cm-1	Nylon Polyethylene oxide Acrylonitrile butadienestyrene (ABS)

Figures

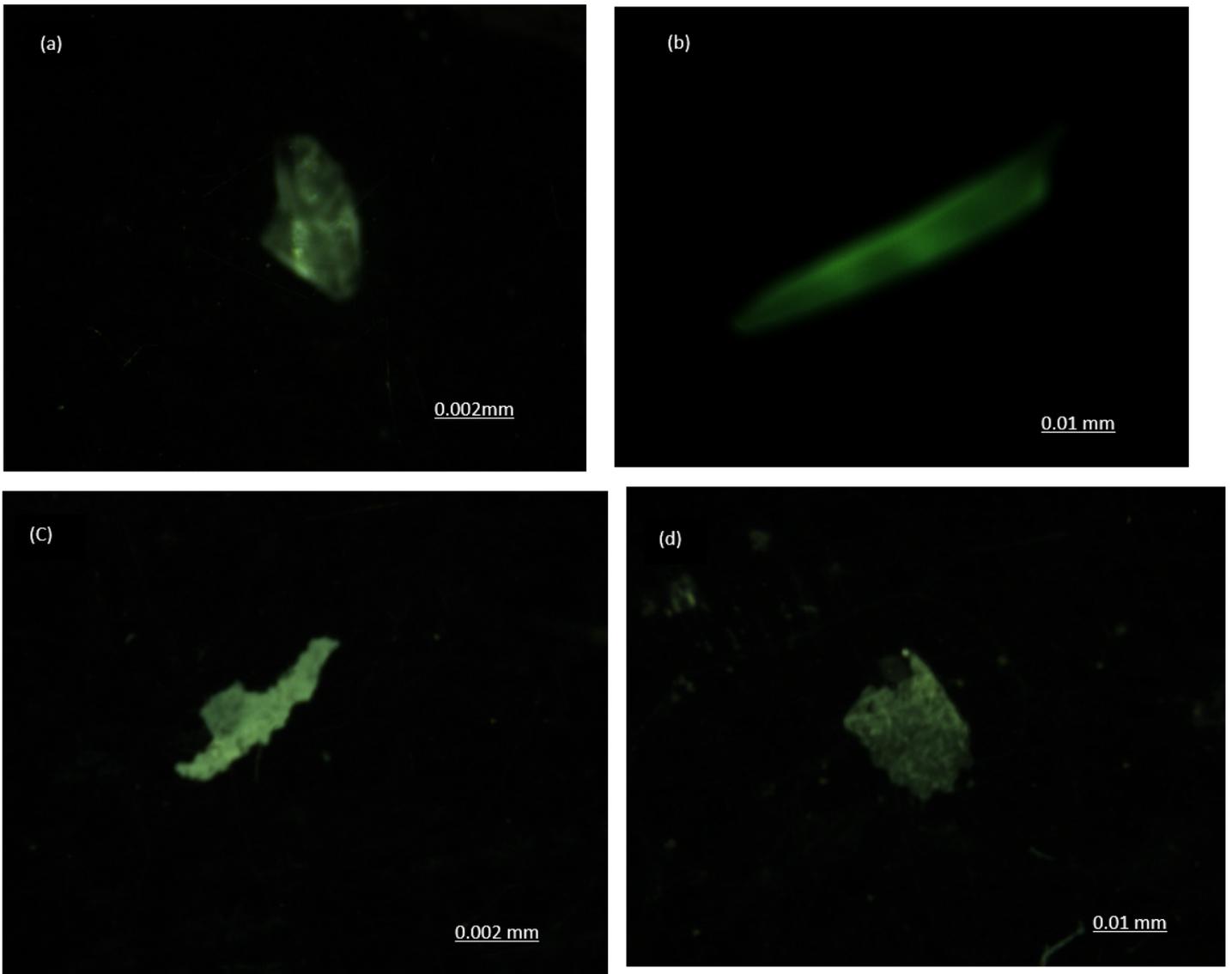


Figure 1

Suspected microplastics images through stereomicroscopical analysis found in fruits and vegetables:(a) Grapes scale bar 0.002mm; (b) Banana scale bar 0.01mm; (c) Potato scale bar 0.002 mm; and (d) Brinjal scale bar 0.01mm.

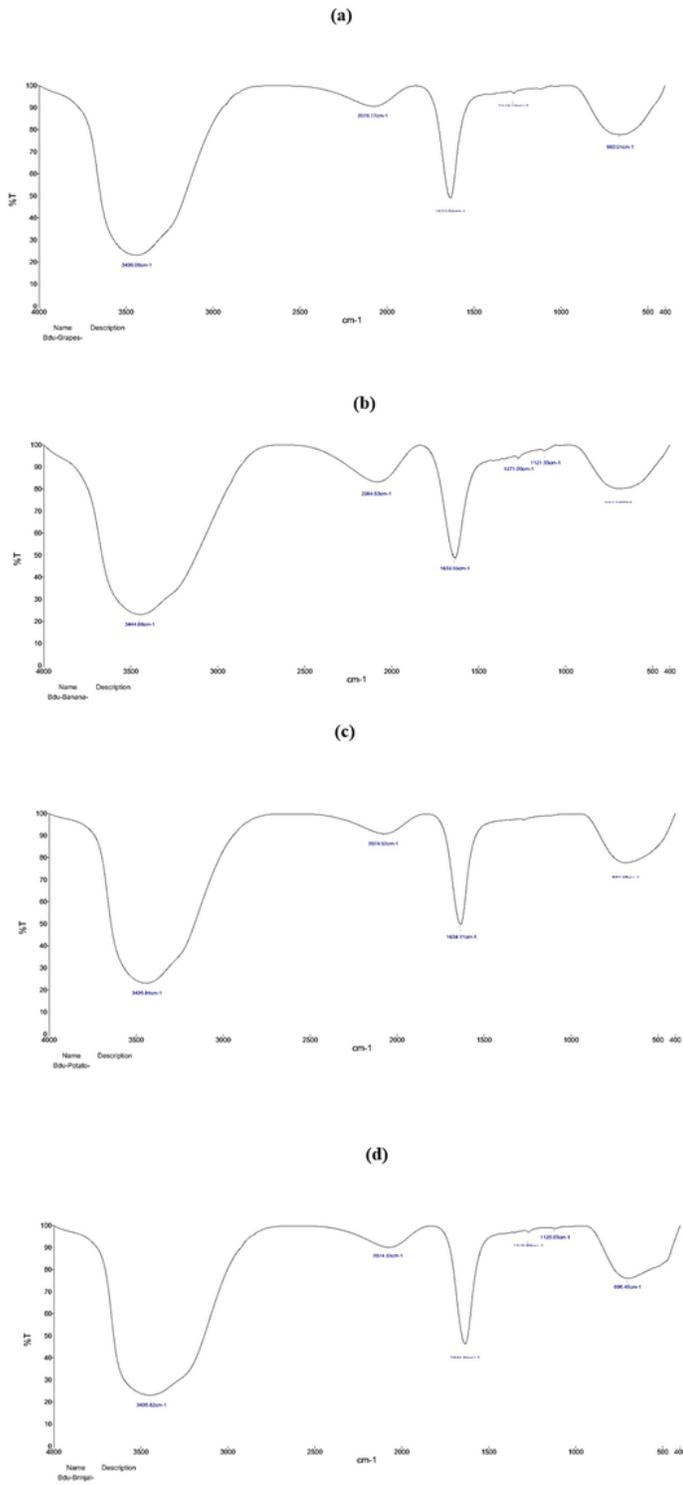


Figure 2

FT-IR spectra showing the presence of microplastic groups in fruits and vegetables: (a) Grapes; (b) Banana; (c) Potato; and (d) Brinjal.

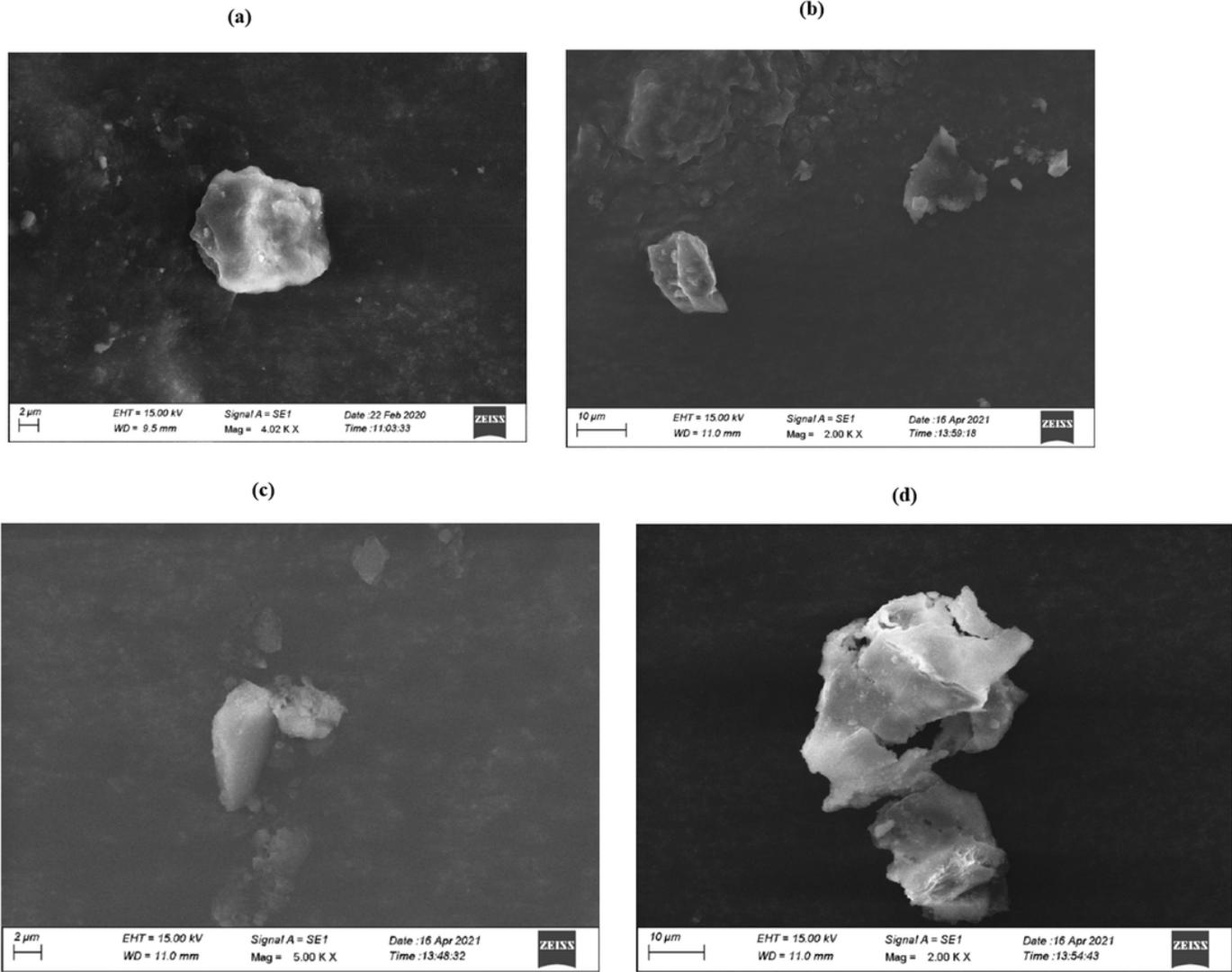


Figure 3

SEM images show the microplastics presence in fruits and vegetables:

(a) Grapes; (2 μm); (b) Banana (10μm); (c) Potato (2 μm) ; and (d) Brinjal (10μm).