ARTICLE

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Detection of microplastics in the human penis

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The proliferation of microplastics (MPs) represents a burgeoning environmental and health crisis. Measuring less than 5 mm in diameter, MPs have infiltrated atmospheric, freshwater, and terrestrial ecosystems, penetrating commonplace consumables like seafood, sea salt, and bottled beverages. Their size and surface area render them susceptible to chemical interactions with physiological fluids and tissues, raising bioaccumulation and toxicity concerns. Human exposure to MPs occurs through ingestion, inhalation, and dermal contact. To date, there is no direct evidence identifying MPs in penile tissue. The objective of this study was to assess for potential aggregation of MPs in penile tissue. Tissue samples were extracted from six individuals who underwent surgery for a multi-component inflatable penile prosthesis (IPP). Samples were obtained from the corpora using Adson forceps before corporotomy dilation and device implantation and placed into cleaned glassware. A control sample was collected and stored in a McKesson specimen plastic container. The tissue fractions were analyzed using the Agilent 8700 Laser Direct Infrared (LDIR) Chemical Imaging System (Agilent Technologies. Moreover, the morphology of the particles was investigated by a Zeiss Merlin Scanning Electron Microscope (SEM), complementing the detection range of LDIR to below 20 μm. MPs via LDIR were identified in 80% of the samples, ranging in size from 20–500 μm. Smaller particles down to 2 μm were detected via SEM. Seven types of MPs were found in the penile tissue, with polyethylene terephthalate (47.8%) and polypropylene (34.7%) being the most prevalent. The detection of MPs in penile tissue raises inquiries on the ramifications of environmental pollutants on sexual health. Our research adds a key dimension to the discussion on man-made pollutants, focusing on MPs in the male reproductive system.

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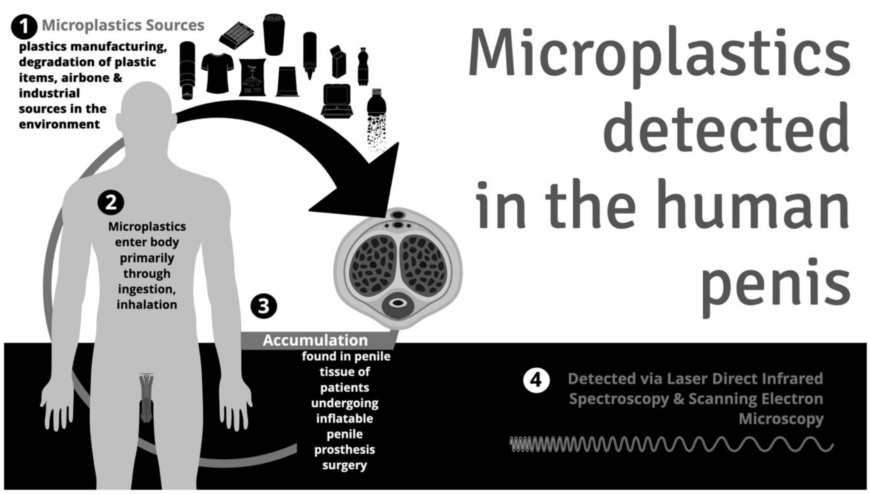
Graphical Abstract



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INTRODUCTION

The pervasive spread of microplastics (MPs), particles less than 5 mm in size, across diverse ecosystems is becoming an alarming environmental and health concern. Recent investigations have shed light on their prevalence in human tissues, such as stool samples, lung, placenta, and cardiac tissue, sparking a crucial discourse about their health repercussions [1–6]. These particles emanate from a plethora of sources and are influenced by various natural factors and their intrinsic physicochemical properties, which dictate their movement and ultimate fate as well as impacts in the environment [7]. These tiny pollutants have permeated freshwater, atmospheric, and terrestrial environments, finding their way into everyday items such as seafood, sea salt, food packaging, and bottled beverages. Moreover, their detection in the gastrointestinal tracts of marine animals points to a broader ecological issue, as these foreign entities can trigger immune responses [3]. MPs, owing to their small size yet large surface area, can engage in chemical interactions with physiological fluids and tissues, raising concerns about their persistence, bioaccumulation, and potential toxicity. Their ability to carry pathogens and pollutants further amplifies these concerns [8, 9].

For humans, the primary pathways for MP exposure are ingestion, inhalation, and dermal contact. Annual intake estimates range from 39,000 to 52,000 MPs per person, predominantly through ingestion [10, 11]. These particles can penetrate the body directly via the atmosphere, drinking water, and sea salt, or indirectly through the food chain. After consumption, only sufficiently small MPs or those forming a biocompatible surface corona can traverse intestinal mucus to reach intestinal cells, potentially entering the circulatory system and accumulating in organs like the gut, liver, and kidneys. Inhalation presents another route, with average daily intake estimated at 272 MPs, which can lead to lung inflammation and other respiratory complications [12]. Dermal contact, though less concerning, is not to be overlooked, especially for particles under 100 nm that can cross the skin barrier [13]. The smallest particles, nanoplastics, may infiltrate cells and disrupt normal cellular functions, but it is unclear if the MP levels in human organs are sufficient enough to impact organ health [14]. The internalization and accumulation of MPs, evidenced by adverse effects in animal models, marine creatures, and human cell lines, pose significant health risks [8, 13, 15, 16]. Notably, recent preliminary studies indicate that MPs can impact fertility and sperm quality, thus threatening reproductive success [17]. This can include live sperm count reduction and morphological abnormalities [18].

While direct evidence linking MPs in penile tissue to erectile dysfunction (ED) is lacking, existing research on environmental factors affecting penile health and erectile function offers valuable insights. Studies such as Sorkhi et al. (2022) on microvascular perfusion, Jung et al. (2014) on neurogenic structures, and works by Jaeger & Walker (2016) and Sopko, Hannan, & Bivalacqua (2014), highlight the multifactorial nature of ED, potentially exacerbated by environmental pollutants such as MPs [19–22].

In this study, we employed LDIR microspectroscopy to detect the existence of MPs in penile tissue of individuals undergoing inflatable penile prosthesis (IPP) placement for the treatment of ED. The detection of MPs in penile tissue would create a new avenue for the environmental impact on sexual health, prompting inquiries about the sources, pathways, and potential ramifications of MPs exposure on erectile function.

METHODS

Participants

The study was approved by the Institutional Review Board of the University of Miami (Study # 20150740) and conducted following the Declaration of Helsinki. Six individuals who underwent surgery for multi-component IPP at the University of Miami, Florida from August 2023 to September 2023 were included in the study. Inclusion criteria were men aged 18 and older, who were diagnosed with ED and were favorable candidates for IPP surgery. There were no exclusion criteria. All patients provided written and informed consent to participate in the study prior to surgery. A sample size of six was chosen, factoring in the significant time investment required to process each sample through LDIR analysis. The demographics of the patients can be seen in Table 1.

Table 1. Demographics.

	Subjects (n = 5)	Control (n = 1)
Age, years		
Mean (SD)	66.6 (12.8)	73
Range	46–81	–
Race		
White	3	1
Black	2	–
Ethnicity		
Hispanic or Latino	3	1
Non-Hispanic or Latino	2	–
Metrics		
Height, meters (SD)	1.7578 (0.09)	1.803
Weight, kg (SD)	87.36 (21.06)	89.4
BMI (SD)	27.91 (4.02)	27.5

SD standard deviation

Sample collection

A single member of the research staff donned synthetic polyisoprene surgical gloves, positioning themselves in proximity to the operating table during the preparation of samples. The operating surgeon exposed the corpora bilaterally with electrocautery. 2-0 polydioxanone PDS suture was utilized for suture retraction on the corpora. Tissue samples were carefully retrieved from the corpora using stainless steel Adson forceps prior to corporotomy dilation and device implantation. An example of tissue sample size can be visualized in Fig. 1C. 25 mL glass Erlenmeyer flasks with glass stoppers were purchased from DWK Life Sciences (Mainz, Germany) to eliminate potential contamination of specimens by storage receptacle. The forceps were then transferred off the field to the research staff member, who promptly opened the glassware tops and deposited the penile corpus tissue samples into the glassware using the forceps. For the one control sample collected, penile corpus tissue was obtained in the same manner and then placed in a McKesson Sterile Specimen Container with translucent polypropylene Base and polyethylene lid.

QA/QC. Research focusing on particulate plastics in the micro- and nanometer size range is still in an adolescent stage and related data comes along with significant analytical uncertainties [23, 24]. Effective quality assurance (QA) and quality control (QC) measures are mandatory to support the reliability and relevance of generated data.

Contamination prevention

To prevent contamination from various sources such as sampling equipment, laboratory apparatus, reagents, clothing, and airborne particles, a stringent protocol was adhered to [25, 26]. This involved the meticulous utilization of class 100 clean benches (Spetec GmbH, Erding, Germany) filtration of all reagents (polycarbonate (PC) track-etched membranes (0.4 µm; Whatman GmbH, Dassel, Germany), and employing meticulously cleaned metal or glass laboratory equipment. All glassware was cleaned with Type I reagent grade water, Milli-Q-water (Milli-Q Integral water purification system (Merck-Millipore, Darmstadt, Germany) equipped with a Q-Pod Element system and a 100 nm endfilter) and filtered 96% ethanol (Reag. Ph Eur, Merck KGaA, Darmstadt, Germany), and subsequently heated for 8 h at 250 °C (while covered with Al foil). Procedural blank samples (method blanks) comprising the entire sample processing protocol were rigorously conducted.

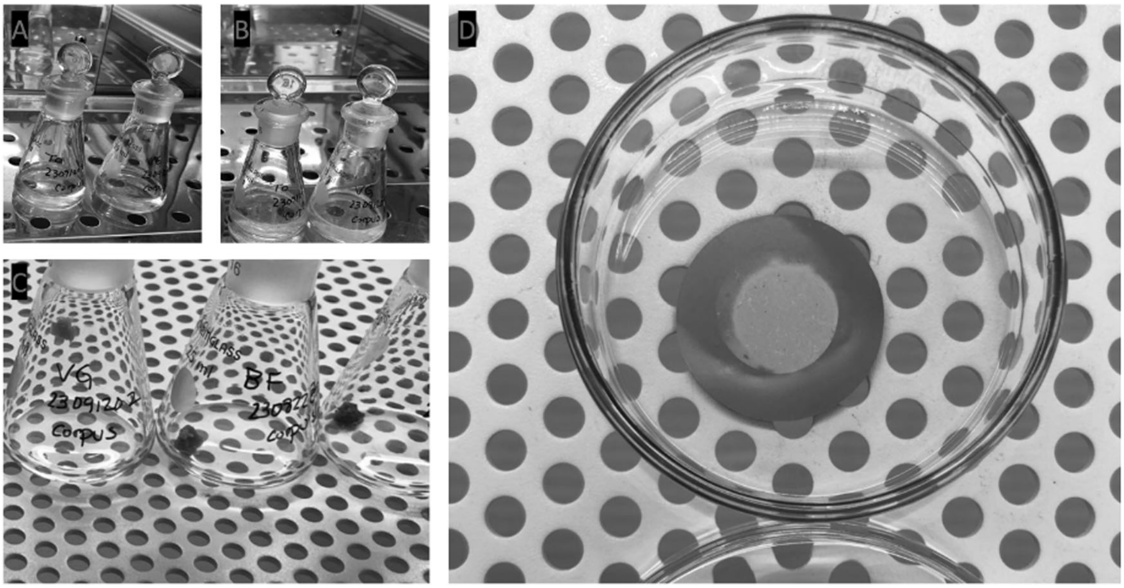

Fig. 1 Sample Preparation. A, **B** Two penile tissue samples at the beginning and end of digestion respectively. **C** Representative size of penile tissue samples collected during surgery. **D** Hot filtration of a tissue sample onto Au-coated filter.

Selectivity of LDIR imaging

The polymer type identification via LDIR imaging (8700 LDIR Chemical Imaging System; Agilent Technologies Inc., Santa Clara (CA), USA) was validated using different MP reference materials: Artificially aged polyethylene (PE), cryomilled polystyrene (PS), polyethylene (PE), polyvinyl chloride (PVC), polypropylene (PP), polyethylene terephthalate (PET) and artificially aged polyamide (PA) (20–206 µm) from the Federal Institute for Materials Research and Testing (BAM, Berlin, Germany); PE, PET, PS, PP, PVC, and PC (all 50–300 µm) from Chiron AS (Trondheim, Norway); expanded PS (EPS) foam, cellulose acetate powder (CA), acrylonitrile butadiene styrene (ABS) pellets and crumb rubber from used tires from the Polymer KIT 1.0 (Hawaii Pacific University, Hawaii, USA).

Sample preparation

Combinations of potassium hydroxide (KOH) and sodium hypochlorite (NaClO) have proven to be effective in the extraction of MPs from biological tissue [27, 28]. Adding the surfactant Tween-20 to the KOH solution at the beginning of the digestion process prevents the formation of fat layers and significantly accelerates the filtration of digested solution [29]. Thus, 50 g KOH, 160 mL NaOCl solution (13%), 1 mL Tween-20 solution (10%), and 340 mL Milli-Q-water (2× filtered) was the solution utilized for sample digestion. 5 mL of digestion solution was added to each sample. Samples were then incubated at 45 °C for 20 h, and then 60 °C for 4 h. The samples were filtered in a hot state with polyethylene terephthalate glycol (PETG) gold-coated membrane filters (0.8 µm pore size, 100/0 nm coating, 25 mm diameter) from Sterlitech Corp. (Auburn, WA, USA). After drying and storage in pre-cleaned Petri dishes, the filters were mounted on a specialized filter holder for the LDIR (Agilent Technologies Inc) that can carry two filters.

Sample analysis by LDIR

The particles (in terms of size, size number distribution, polymer type) were analyzed using the Agilent 8700 LDIR Chemical Imaging System (Agilent Technologies, Santa Clara, USA). LDIR facilitates the rapid analysis of up to 1000 particles or fibers, with sizes less than 300 µm [25, 30]. The particle analysis workflow of the Clarity Software (Version 1.5.58, Agilent Technologies Inc.) was used for the automated analysis of the entire sample set. The particle size range was set to 20–500 µm and the acquired IR spectra were compared with a specific spectral library (MP starter 2.0, Agilent Technologies). Spectra of all assigned synthetic polymer types were thoroughly checked to identify matrix-related interferences, e.g., from remaining fatty acids. MPs identifications were either accepted or not accepted. Only spectra in conjunction with high hit quality values related to reference spectra (>0.90) were considered for the final statistics without further manual confirmation. If confirmation of any particle was not possible, the respective particles were marked as "unknown". The approach presented can be considered very conservative compared to the literature [31–33].

Sample analysis by SEM

The same filters employed in LDIR analysis were used for an investigation by scanning electron microscopy (SEM). A central piece of the filter was cut out and sputter-coated with 1 nm platinum layer using a CCU-010 coating device (Safematic, Switzerland). The SEM images were recorded on a Merlin (ZEISS, Oberkochen, Germany, software SmartSEM) at an accelerating voltage of 3 kV using a secondary electron detector to highlight the topography. After recording overview images, representative spots with micrometer-sized structures were selected for higher magnifications.

RESULTS

A total of six patients consented to contribute penile tissue samples for this study. Six samples were collected during an eight-week period during surgery for insertion of multi-component IPP. Of the six samples, one was obtained as a control through the application of standard sample collection procedures typically employed in surgical cases. Five were gathered with the specific goal of avoiding cross-contamination with MPs. Three participants, including the control, reported prior Intracavernosal Injection (ICI) use. No patients had prior penile surgery. The clinical characteristics of each patient can be found in Table 2.

MPs via LDIR were detected in four of the five samples (80%), within the range of 20–500 µm. Seven types of MPs were identified in the corpus tissue samples (Fig. 2). The most prevalent MPs were PET (47.8%) and PP (34.7%), accounting for about 82% of the total amount of MPs. MPs were found in all but one patient. Additionally, MPs were found in the one control sample, with poly methyl methacrylate being the sole MP specific to this sample.

Figure 2B illustrates the overall distribution of MP diameter in the tissue samples, as measured by LDIR. 84% of samples were in the diameter range of 20–100 µm. Two MP with a diameter in the range of 200–500 µm were found in Sample ID # 1 and Sample ID # 2.

SEM enables the investigation of particles below the 20 µm detection limit applied to LDIR imaging. The images revealed particles with a size of 2–20 µm, some with a spherical shape, some with a polygonal shape and sharp edges. Furthermore, there

Table 2. Subject Clinical Characteristics.

ID	Age, years	Surgery type	Race	Ethnicity	Prior ICI use	Type of ED
Control	73	IMC-IPP	White	Hispanic or Latino	Yes	Erectile dysfunction, unspecified type
1	46	IMC-IPP	Black	Non-Hispanic or Latino	Yes	Erectile dysfunction, organic origin
2	71	IMC-IPP	White	Hispanic or Latino	No	Erectile dysfunction, organic origin
3	81	IMC-IPP	White	Hispanic or Latino	Yes	Erectile dysfunction, organic origin
4	66	IMC-IPP	Black	Non-Hispanic or Latino	No	Erectile dysfunction, post radical prostatectomy
5	69	IMC-IPP	White	Non-Hispanic or Latino	No	Erectile dysfunction, organic origin

IMC-IPP insertion of multi-component inflatable penile prosthesis, *ICI* intracavernosal injection, *ED* erectile dysfunction

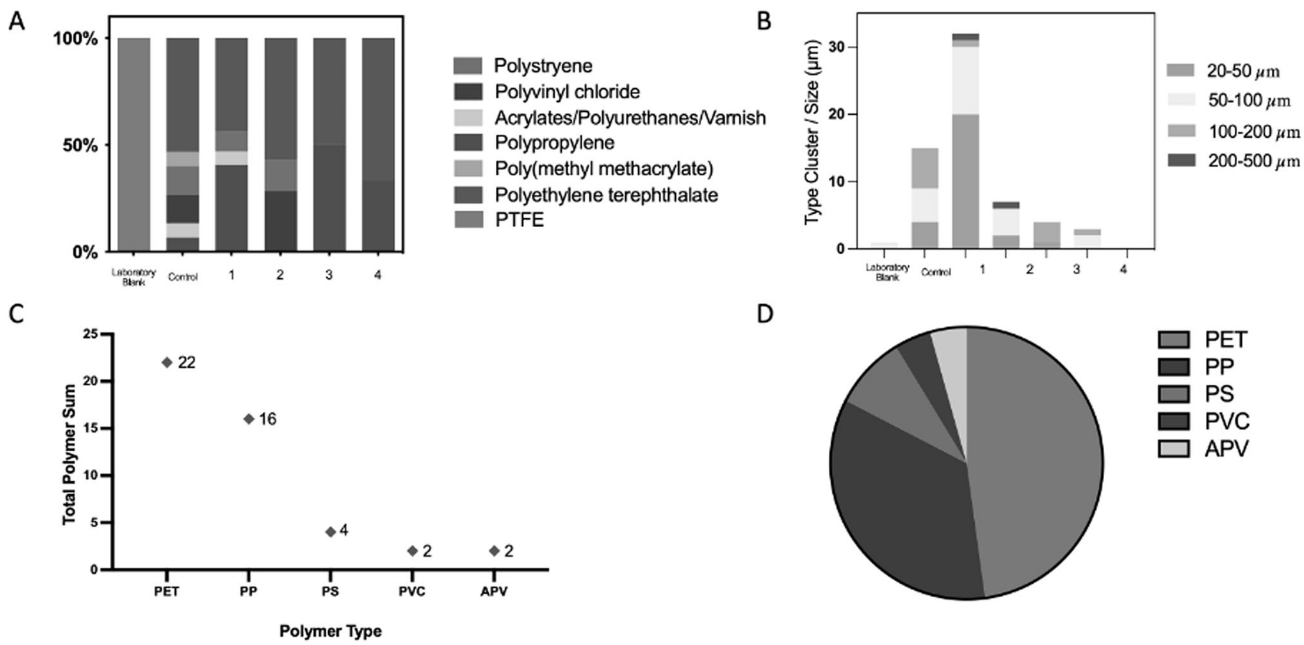

Fig. 2 Characteristics of MPs in Penile Tissue. A Percentage of each type of MP identified by LDIR within each penile tissue sample. **B** Quantity of type cluster/size of MP determined to be within each penile tissue sample by LDIR. **C** Total sum of polymer count by polymer type found in penile tissue of patients. **D** Percentage of each MP in all penile tissue.

are occasional particles of 50 µm length and elongated fibers of more than 100 µm length and 5 µm diameter. All these discernible particles are embedded in an undefined layer of ~1.5 µm thickness, sitting on the filter surface with well-defined pores. SEM images can be seen in Fig. 3.

DISCUSSION

We present the first study to our knowledge to identify the presence of MPs within penile tissue. The detection of MPs in penile tissue opens a novel avenue for understanding the environmental impact on sexual health, raising questions about the sources, pathways, and potential consequences of MP exposure on erectile function.

MPs are commonly thought to enter the human body through ingestion and inhalation. Barboza et al. propose that MPs larger than 150 µm are likely not absorbed, while MPs smaller than 150 µm may have the potential to migrate from the gut cavity to the lymph and circulatory system, leading to systemic exposure [30]. The majority of MPs (84%) in our study were below this threshold. Zhao et al. who identified MP in testes tissue, similarly found that MP ranging from 20–100 µm were most prevalent [34]. Although MPs can pass through the small intestine and be excreted through stool and urine, MP have also been found to accumulate through tissue. More research is required to understand the properties of MP excretion versus accumulation [35].

The predominant types of MP identified were PET and PP. PP and PET are two common non-biodegradable MPs utilized in everyday goods and packaging [36]. PET, derived from petroleum, is commonly used for packaging foods and beverages, including juices and soft drinks. PET is also extensively utilized in healthcare materials due to its desirable properties such as strength and abrasion resistance [37, 38]. PP is also utilized in the same manner as PET, for use in disposable plastic bottles and reusable plastic containers.

Investigations into the role of MPs within the male reproductive system represent a recent area of inquiry, with limited existing literature. Much of the existing research has been predominantly focused on the impact of MPs on male infertility. Several studies have discussed the adverse effects of MPs on sperm production and quality [17, 34, 39–41]. In a study where mice were administered MP (specifically PS) via drinking water, live sperm count within the epididymis was reduced in MP-exposed mice compared to controls [18]. Morphological abnormalities were also observed, alongside increased inflammatory markers (NF-kB, IL-B, IL-6) [18]. However, exploration of MPs impact on erectile function remains notably absent from the literature. During episodes of

A

B

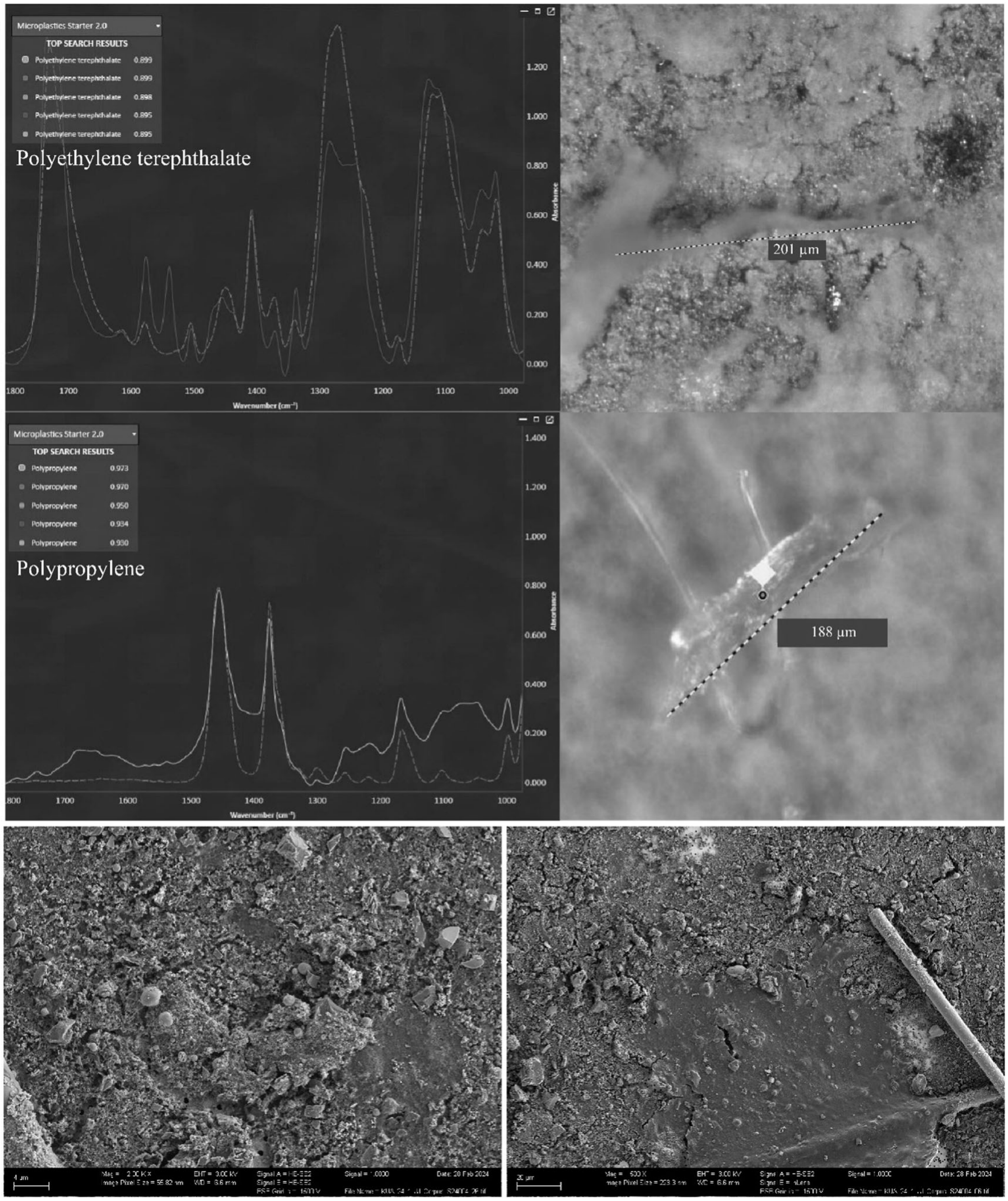

Fig. 3 LDIR Spectra Analysis & SEM Imaging of MPs. A LDIR spectra analysis demonstrating signals matching polyethylene terephthalate and polypropylene in penile tissue. Representative statistics such as size and ID of respective MP particles are also shown. **B** Representative SEM images of the MP particles on filters after digestion of the penile tissue. The images include scale bars of 20 μm (left) and 4 μm (right).

tumescence, characterized by increased blood flow to the penis, the corpus cavernosum and associated tissues undergo expansion. This physiological process includes the dilation of blood vessels, creating an environment where circulating MPs may interact with penile tissue, potentially resulting in its bioaccumulation. The engorgement of the corpus cavernosum with blood offers an expanded surface area for potential interactions between circulating MPs and the surrounding tissue.

This study's identification of the presence of MPs in penile tissue raises compelling questions regarding their potential influence on erection function, considering their known association with adverse localized tissue effects. Further investigation into this unexplored area is warranted to better comprehend the impact of MP exposure on male sexual health.

One of the notable strengths of this study lies in its comprehensive methodology, incorporating both quantitative and qualitative approaches. By utilizing both LDIR for qualitative data in conjunction with SEM for quantitative information, a greater understanding of the various permutations and characteristics of MPs in the penile tissue samples was achieved. Additionally, sample collection with a single surgeon reduced inter-operator variability and enhanced consistency of sample collection from the same location with each patient. There are several limitations in our study, notably the potential for MP contamination during surgery, despite the predominance of stainless-steel surgical tools in direct contact with the surgical field and tissue collected in this study. Plastic-containing materials are widely present in operating rooms and introduce the possibility of MP contaminants. ICI, a medical procedure where medication is directly injected into the corpus cavernosum, could potentially represent another avenue for MP uptake and distribution in penile tissue. The injection site provides a localized entry point for substances into the erectile tissue. The fate of MPs in this context would depend on various factors, including their size, composition, and surface properties, as well as the dynamics of blood flow in the penile vasculature. Three participants, including the control, reported prior ICI use. However, we did not identify a correlation between prior ICI use and the amount or type of MP found in the tissue samples. Our study also did not explore occupational and lifestyle exposure to MPs in participants, and future studies should include these metrics where possible to further characterize the impact of MP on human health.

The primary aim of this study was to identify MPs within penile tissue of patients undergoing IPP placement for ED. MPs pervade our environment and are here to stay for the foreseeable future. Therefore, it is imperative to understand how they interact with the human body to grasp their potential implications on human health and physiology. While the results do not establish a correlation between MP presence and ED, they do offer valuable insights that enrich the ongoing discussions on the intricate interactions between MP and human tissues. Further research is needed to elucidate the consequences of MPs in vivo, given their capacity to act as a vector for pathogens, as well as induce and be influenced by oxidative stress, inflammation, and immune response [8, 9, 42, 43].

CONCLUSION

Our study presents a groundbreaking investigation into the presence of MPs in penile tissue. We have identified MPs, primarily PET and PP, in the majority of the penile tissue samples examined. By shedding light on the presence of MPs in human tissues, our research adds a crucial dimension to the ongoing discourse about the effects of environmental pollutants on human health, with a specific focus on male sexual health.

DATA AVAILABILITY

All relevant data to the current study that was generated and analyzed is available upon reasonable request from the corresponding author.

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AUTHOR CONTRIBUTIONS

Jason Codrington—conceptualization, methodology, investigation, project administration, data curation, visualization, writing—original draft, editing. Alexandra Aponte Varnum—investigation, writing—original draft, editing, data curation, visualization. Lars Hildebrandt—investigation, writing—original draft, validation, resources. Daniel Pröfrock—investigation, editing, validation, resources. Joginder Bidhan—resources, writing—original draft. Kajal Khodamoradi—project administration, resources. Anke-Lisa Höhme—investigation, visualization. Martin Held—writing—original draft, editing. Aymara Evans—writing—original draft. David Velasquez—writing—original draft. Christina C. Yarborough—writing—original draft. Bahareh Ghane-Motlagh—investigation. Ashutosh Agarwal—investigation. Justin Achua—writing—original draft. Edoardo Pozzi—editing. Francesco Mesquita—editing. Francis Petrella—writing—review. David Miller—writing—review. Ranjith Ramasamy—conceptualization, methodology, project administration, resources, supervision, editing, funding acquisition

COMPETING INTERESTS

Dr. Edoardo Pozzi is currently an Associate Editor for the International Journal of Impotence Research.

ETHICS APPROVAL

The study was approved by the Institutional Review Board of the University of Miami (Study # 20150740) and conducted following the Declaration of Helsinki. All patients provided written and informed consent to participate in the study.

ADDITIONAL INFORMATION

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