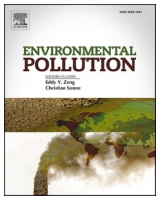
Environmental Pollution 343 (2024) 123233

Contents lists available at ScienceDirect

Environmental Pollution

journal homepage: www.elsevier.com/locate/envpol



Exposure of U.S. adults to microplastics from commonly-consumed proteins☆

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ARTICLE INFO

Keywords:
Food
Plastics
Microplastics
Proteins
Ingestion
Human exposure

ABSTRACT

We investigated microplastic (MP) contamination in 16 commonly-consumed protein products (seafoods, terrestrial meats, and plant-based proteins) purchased in the United States (U.S.) with different levels of processing (unprocessed, minimally-processed, and highly-processed), brands (1 – 4 per product type, depending on availability) and store types (conventional supermarket and grocer featuring mostly natural/organic products). Mean (±stdev) MP contamination per serving among the products was 74 ± 220 particles (ranging from 2 ± 2 particles in chicken breast to 370 ± 580 in breaded shrimp). Concentrations (MPs/g tissue) differed between processing levels, with highly-processed products containing significantly more MPs than minimally-processed products ($p = 0.0049$). There were no significant differences among the same product from different brands or store types. Integrating these results with protein consumption data from the American public, we estimate that the mean annual exposure of adults to MPs in these proteins is 11,000 ± 29,000 particles, with a maximum estimated exposure of 3.8 million MPs/year. These findings further inform estimations of human exposure to MPs, particularly from proteins which are important dietary staples in the U.S. Subsequent research should investigate additional drivers of MPs in the human diet, including other understudied food groups sourced from both within and outside the U.S.

1. Introduction

Microplastics (MPs) are pervasive across global ecosystems, and have been identified in aquatic and terrestrial animals, as well as in numerous organs within the human body (de Sá et al., 2018; Dissanayake et al., 2022; Jenner et al., 2022; Leslie et al., 2022; O'Brien et al., 2023; Prata & Dias-Pereira, 2023; Ragusa et al., 2021). Concerns regarding food security and safety have led to an increasing number of studies documenting MPs in human-consumed foods, particularly finfish, bivalves (mussels and clams), and shrimp, though limited studies have been conducted on other food and beverages (e.g., drinking water, beer, and honey per Akoueson et al., 2020; Andreas Hadibarata et al., 2021; Baechler et al., 2020; Catarino et al., 2018; Curren et al., 2020; Daniel et al., 2020; Danopoulos et al., 2020b; Devriese et al., 2015; El et al., 2020; Fernández Severini et al., 2020; Hossain et al., 2020; Kosuth et al., 2018; Liebezeit & Liebezeit, 2014; Liebezeit & Liebezeit, 2015; Mercogliano et al., 2020; Pivokonsky et al., 2018; Rainieri & Barranco, 2019; Saha et al., 2021; Sarijan et al., 2021; Van Cauwenberghe & Janssen, 2014; Yozukmaz, 2021; C. Zhang et al., 2020). From these studies and other research, several estimates of annual human MP consumption have been published (Cox et al., 2019; Danopoulos et al., 2020a; Domenech & Marcos, 2021; Mohamed Nor et al., 2021); however, such estimates have limitations, as they consider only a small fraction of the human diet due to limited availability of data on the contamination of MPs in many commonly-consumed foods (EFSA Panel on Contaminants in the Food Chain (CONTAM), 2016; Koelmans et al., 2019; Rainieri & Barranco, 2019).

More data are needed to accurately estimate human MP consumption and ultimately to develop risk assessments (Thornton Hampton et al., 2022). For example, little is known about MP burdens in, and subsequent human exposure from, terrestrial meats, dairy products and other common food items such as fruit, vegetables, and grains (Domenech &

☆ This paper has been recommended for acceptance by Eddy Y. Zeng.

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https://doi.org/10.1016/j.envpol.2023.123233
Received 24 October 2023; Received in revised form 20 December 2023; Accepted 24 December 2023
Available online 28 December 2023

Marcos, 2021). While there is clear evidence of MP accumulation in marine and terrestrial food chains, few studies have investigated this type of contamination in land-based proteins like raw meats or meat products (Bilal et al., 2023; Giorgetti et al., 2020; Huang et al., 2020; Huerta Lwanga et al., 2017; Lu et al., 2020; Sun et al., 2020; Wang et al., 2022). As with seafood, small particles can translocate into edible tissues of terrestrial animals following ingestion (Hussain et al., 2001). However, because many of the products we consume are processed and packaged prior to reaching our dinner tables, it is necessary to understand other potential contributors of MP contamination in the food system as well. MP contamination in foods could be the result of airborne particles or those shed from equipment used in the food production and distribution process, additional ingredients, or packaging (Danopoulos et al., 2020a; Du et al., 2020; Eerkes-Medrano et al., 2019; Fadare et al., 2020; Jadhav et al., 2021; Karami et al., 2018; Kedzierski et al., 2020; Kosuth et al., 2018; Kutralam-Muniasamy et al., 2020; Liebezeit & Liebezeit, 2014; Oβmann et al., 2018; Panno et al., 2019; Schymanski et al., 2018; Shruti et al., 2020). Some studies suggest current estimates of MP exposure levels may negatively impact human health, demonstrating an urgent need for further research to understand sources of contamination, and impacts to human health to develop effective strategies to mitigate risk (Danopoulos et al., 2022).

1.1. Study aims and objectives

The objective of this research was to quantify MPs in understudied/unstudied food items, further inform MP exposure estimates from the human diet, and identify potential drivers of MP contamination in foods. Specifically, we aimed to understand MP burdens in various U.S. protein products, including seafoods, terrestrial meats, and plant-based proteins, and the influences of processing level, packaging material, brand and store type on MP contamination. We combined our findings with responses from a recent survey by our research team regarding protein consumption among United States (U.S.) adults to yield estimates of human MP exposure through consumption of these different protein types (Baechler et al., 2024).

2. Methods

2.1. Sample collection and processing

Protein samples were purchased in April 2022 from two conventional supermarkets and one grocer featuring mostly natural and organic products in the metropolitan area surrounding Portland, Oregon, U.S. All samples were labeled as U.S. products on the exterior packaging. Store-collected samples consisted of 13 different protein types: breaded shrimp, minced pollock fish sticks, white Gulf shrimp (*Litopenaeus setiferus*; headless/shell-on), Key West pink shrimp (*Pandalus borealis*; headless/shell-on), Alaska Pollock (*Gadus chalcogrammus*) fillets (skinless), chicken nuggets, top sirloin steaks, pork loin chops, chicken breasts, plant-based nuggets, plant-based fish sticks, plant-based ground beef, and tofu blocks. One product per bag or package was considered a replicate. Product packaging varied by sample type and brand. In addition to the protein samples obtained from grocery stores, three replicates each of unprocessed, whole Alaska pollock, white Gulf shrimp (head-on, shell-on), and Key West pink shrimp (head-on, shell-on) were obtained in February 2023 from vessels. Samples of each vessel-obtained product type were packaged together in one clear plastic bag. Alaska pollock were dissected to remove one fillet per fish. Each fillet was skinned and rinsed with reverse osmosis (RO) water filtered to 0.45 μm to remove any contamination of gut contents or airborne contamination from the filleting step before it was wrapped in aluminum foil and stored in a Ziploc bag at −20 °C until further processing.

All samples, regardless of origin, were transported to the University of Toronto (Ontario, Canada), where each replicate was removed from its packaging (e.g., a single chicken nugget, a block of tofu, etc.) and weighed, and packaging characteristics (color and rigidity) were noted. Three replicates were processed per product type and brand. Each replicate was wrapped in aluminum foil and stored in an individual Ziploc bag at −20 °C until further processing. For tofu samples, the liquid in each package was poured out and only the block of tofu was weighed and kept.

Each sample was individually chemically digested in a 1 μm pre-filtered 20% potassium hydroxide (KOH) solution for at least 24 h to break down the food material (Munno et al., 2018). After KOH digestion, samples were rinsed using RO water and passed through a 45 μm sieve to remove the KOH. The remaining material in the sieve was poured into individual clean glass beakers. For shell-on shrimp samples, the shell did not break down during the KOH procedure so it was rinsed into the sample beaker using RO water and then discarded. 60 mL of 1 μm pre-filtered 30% hydrogen peroxide solution (H_2O_2) was added to each sample and left for 20 min to break down any remaining food material. Due to some sample loss during processing some products had fewer than three replicates (further details about sample sizes are in Supporting Information Table S1).

A small proportion of samples that were resistant to breaking down (three samples of pork loin chop, four samples of plant-based ground beef, three samples of top sirloin steak, one sample of tofu, and six plant-based fish stick samples) underwent a wet peroxide oxidation (WPO) procedure involving iron sulfate Fe(II)SO_4 as a catalyst and 30% H_2O_2 in a 1:5 ratio. An ice bath was used to maintain a temperature below 50 °C throughout WPO, to minimize MP loss (Munno et al., 2018). Following chemical digestion, samples were run through a 45 μm sieve, and rinsed with RO water into their respective individual beakers to soak for approximately 2 h in a 10% Alcojet detergent solution (1:1 ratio; Decon Conrad 70, Fisher Scientific) to remove any remaining fatty material. Finally, these samples were rinsed through 45 μm and 125 μm sieves to produce two separate particle size fractions (45 μm–125 μm and >125 μm) per sample. A 45 μm minimum particle size was selected due to previously-noted limitations associated with visual identification of microplastics using microscopy. An interlaboratory study assessing the accuracy of methods for microplastic identification found that microscopy can accurately identify plastic particles >50 μm in size (Kotar et al., 2022). Below this size, particles may be missed or incorrectly identified as plastics (i.e., 'false positive results').

2.2. Suspected MP Sorting and Quantification

Samples were examined using a dissecting microscope (10-80 × magnification; Leica S8 APO Stereozoom; Leica Microsystems, Canada) to visually identify suspected MPs (Lusher et al., 2020). The first ten suspected MPs of a particular morphology and color observed per size fraction (e.g., blue fragments in the >125 μm size fraction) were removed using forceps and mounted onto double sided tape in a Petri dish (Fig. S1). The categories used to categorize particles were: fragment, fiber, film, rubber, fiber bundle, sphere, and foam (Lusher et al., 2020). All particles of a particular morphology and color beyond the first ten in each size fraction were tallied to produce a total count of particles, but were not analyzed further. All particles were then photographed and measured for length and width using OMAX Toupview software (version 3.7; ToupTek).

2.3. Chemical confirmation using spectroscopy

For this study, we characterized MPs according to the 2020 California State Water Board's 'Microplastics in drinking water' definition, which is inclusive of any: "… material consisting of one or more solid polymer-containing particles, to which additives or other substances may have been added …" that is "greater than 1 and less than 5000 μm;" however, for this study, particles over 5000 μm in size were also included and reported (Coffin, 2020).

A subset of particles was chemically analyzed using spectroscopy to

determine material type. One suspected MP particle from each test sample and laboratory blank was randomly selected as a representative subsample to accurately determine the proportion of plastic particles present across all protein samples (De Frond et al., 2023). In total, 124 particles were analyzed (13% of all particles in the blank-corrected data set, plus one particle from each blank; 14 blanks). Raman spectroscopy (Horiba Raman XploRA PLUS confocal microscope, Piscataway, NJ, USA) was used for chemical identification using LabSpec6 software and equipped with a charge coupled device detector (−60 °C, 1024x256 pixels Raman spectra were obtained using a 100× LWD objective (NA = 0.8) resulting in laser powers of 15.0 mW and 17.8 mW at 100% filter for the 532 nm and 785 nm lasers, respectively. Spectral dispersion ranged from 1.3 cm^{-1} per pixel (785 nm excitation laser, 600 grooves/mm) to 3.3 cm^{-1} per pixel (532 nm excitation laser, 1200 grooves/mm). Spectra were matched to reference spectra from the KnowItAll Raman Spectral Library (listed in Table S2) as well as the Spectral Library of Plastic Particles (SloPP and SloPP-E) using Wiley KnowItAll and ID Expert spectral matching software (ID Expert version 23.1.45.0) (Munno et al., 2020). Minimal manual corrections to spectra were made; including baseline correction, and vertical clipping.

Suspected rubber particles were analyzed using μ-Fourier Transform Infrared (μFTIR) to achieve better chemical identification for this particular morphology. Here, spectra were collected with a Nicolet iN10 infrared microscope (Thermo Fisher Scientific in ATR mode (15 × objective, 0.7 numerical aperture), using a germanium ATR crystal). Spectra were collected at spectral resolution of 16 cm^{-1} and 32 scans (high resolution). Spectra were matched to spectral reference libraries (listed in Table S3) including the μATR-FTIR Spectral Libraries of Plastic Particles (FLOPP and FLOPP-e) and the spectral library developed by Primpke et al. (2018) and De Frond et al. (2021) .

We used the spectral matches obtained using Raman and μFTIR spectroscopy to characterize each particle within the subset of sampled particles as either microplastic, natural, or unknown (Fig. S2a). We used these categorizations of the subset of the particles to spectroscopy-correct the full dataset to only include the proportion of particles that were microplastics. This was achieved by multiplying all reported values by the proportion of particles considered to be microplastics in order to exclude the proportion of particles found to be natural or unknown.

2.4. Blanks, QA/QC and data correction

Throughout all laboratory processing steps, quality assurance/quality control (QA/QC) procedures to both minimize and account for MP contamination in samples were followed. Work was conducted in a laboratory equipped with a HEPA filter and in a clean cabinet when possible. White cotton lab coats were worn by researchers, all glassware was triple-rinsed with reverse osmosis (RO) water prior to use, and all materials were kept covered with aluminum foil when not in use. To quantify contamination, one laboratory blank sample was run per ten test samples ($n = 14$) (Brander et al., 2020). For each laboratory blank, a piece of aluminum foil was laid on the lab bench for the duration of the initial processing and weighing procedure to document any airborne contamination. Each sheet of aluminum foil was folded and stored in a Ziploc bag just like the test samples until further processing steps. Blanks were created when samples were removed from their packaging and weighed for all samples except for whole Alaska pollock (blanks were created during the filleting stage to most accurately reflect potential contamination). Prior to KOH digestion, each piece of aluminum foil was rinsed into a clean sample jar using RO water. Blank samples were processed alongside test samples to document any laboratory contamination from sample processing. Four laboratory blanks of the 14 total were processed using WPO to account for any additional contamination introduced to samples during these steps.

Particle counts within the laboratory blanks were used to calculate a limit of detection for each color and morphology combination within a size fraction (e.g., blue fragments in the >125 μm size fraction) (Table S4). The limit of detection was calculated as the standard deviation multiplied by three plus the mean of all blanks, and was used to exclude particles from samples that were below the limit of detection. As no standard methods for blank subtraction currently exist in MP research, this method of blank subtraction was used to ensure that results were conservative, as has been done in previous research on MP contamination (Brander et al., 2020; Bråte et al., 2018; Hung et al., 2021). For particle types and sizes above the limit of detection, the mean of each particle category found in the blanks were subtracted from particle counts with the same color, morphology and size categorization.

Additionally, three spike and recovery samples were run using the same procedures as the protein samples to measure recovery during the extraction process. All spike and recovery samples underwent both the 30% H_20_2 procedure and the 10% KOH procedure. The spike and recovery samples did not undergo WPO which only a small subset of samples were processed with, and WPO is an established method with reported high recoveries (Herrera et al., 2018; Rodrigues et al., 2018). Ten particles of each of five different MP morphologies/colors were used in each spiked sample: red acrylic microfibers (730–2400 μm), clear polyethylene terephthalate fragments (350–1700 μm), green polypropylene fragments (300–1400 μm), green polyethylene microspheres (63–75 μm), and clear polyethylene microspheres (180–210 μm). For each spike and recovery, a randomly chosen extra sample was used as a matrix to be representative of how the different product types responded to the MP extraction procedures (chicken nugget, breaded shrimp, and tofu).

2.5. Data analysis

To assess trends across sample types, we classified products into three processing levels: ‘unprocessed’ samples were those obtained whole and unmodified from vessels (whole Alaska pollock, head on/shell on white Gulf shrimp, head on/shell on Key West pink shrimp), ‘minimally-processed’ samples were purchased from the grocery store, which were cut and packaged in plastic, but were not modified from their original form (Alaska pollock fillets, headless/shell-on white Gulf shrimp, headless/shell-on Key West pink shrimp, chicken breasts, pork loin chops, top sirloin steaks), and ‘highly-processed’ samples were significantly processed beyond cutting prior to being packaged (minced Alaska pollock fish sticks, breaded shrimp, chicken nuggets, plant-based nuggets, plant-based fish sticks, plant-based ground ‘beef’, tofu block).

All statistical analyses were performed using R statistical software (RStudio version 2022.02.3) with a significance level of $\alpha = 0.05$. Data were found to be not normally distributed using the Shapiro-Wilk test. As such, nonparametric Kruskal−Wallis tests were used to determine differences in MP loadings between products, protein types (seafoods, terrestrial meats, plant-based proteins), brands (between one and four brands per product), and processing levels (unprocessed, minimally-processed, highly-processed). When statistically significant differences were identified, post-hoc Dunn’s tests with Bonferroni corrections were used to determine which pairings differed. To assess differences in particle counts between store types (conventional supermarket, grocer featuring mostly natural and organic products), a nonparametric Mann−Whitney test was used. A nonmetric multidimensional scaling (nMDS) plot was created to examine assemblages of particles across product types and processing levels. Subsequently, PERMANOVAs were run to assess differences between product type and processing level. The nMDS was created with the “metaMDS” function in the vegan community ecology package, and the PERMANOVA with the “adonis2” function in the vegan community ecology package (Oksanen et al., 2022).

2.6. Annual microplastic exposure estimates among U.S. adults

Protein consumption data were obtained from a nationwide survey of Americans 18 years of age and older conducted by Ocean Conservancy and EDGE research in fall 2021 (Baechler et al., 2024). Survey

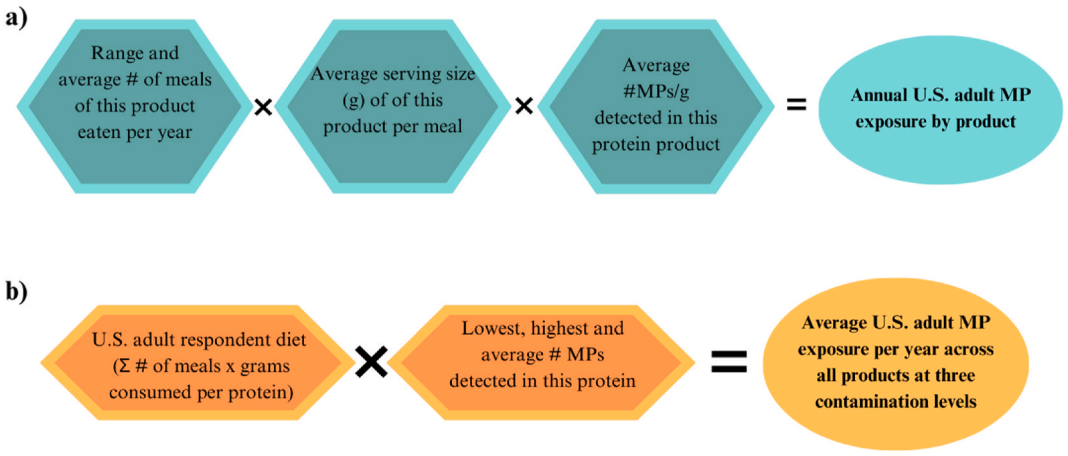

Fig. 1. Schematic showing the calculations performed to calculate: a) product-by-product microplastic (MP) exposure; and b) average U.S. adult exposure to MPs. g = grams.

respondents ($n = 1961$) were recruited using an online, opt-in consumer research panel. Response quotas were managed to ensure the sample was demographically and geographically reflective of the 2020 U.S. Census Bureau population estimates (Bureau, 2020).

Estimates of annual MP exposure for each product type were calculated by multiplying respondents' mean number of annual servings consumed for each product by the mean reported portion size (grams), and mean MP contamination (MPs/g) determined through the present study for that product (Fig. 1a).

To estimate annual MP exposure for all products combined, survey data on respondents' frequency and serving size (grams) of each protein were used to produce a representative distribution of annual U.S. adult consumption for 13 of the 16 protein products studied. Unprocessed Alaska pollock, Gulf shrimp and Florida pink shrimp were excluded, as the survey did not distinguish processing levels for those product types. Thus, store-bought, minimally-processed versions of these products were used for the calculations instead, as we assumed grocery store versions of the products to be most widely available to U.S. consumers. Each survey respondent's annual consumption of the 13 product types was multiplied by the mean contamination (MPs/gram) for each product (Fig. 1b). These 13 product-specific exposure values were summed to yield an annual MP exposure estimate for each survey respondent,

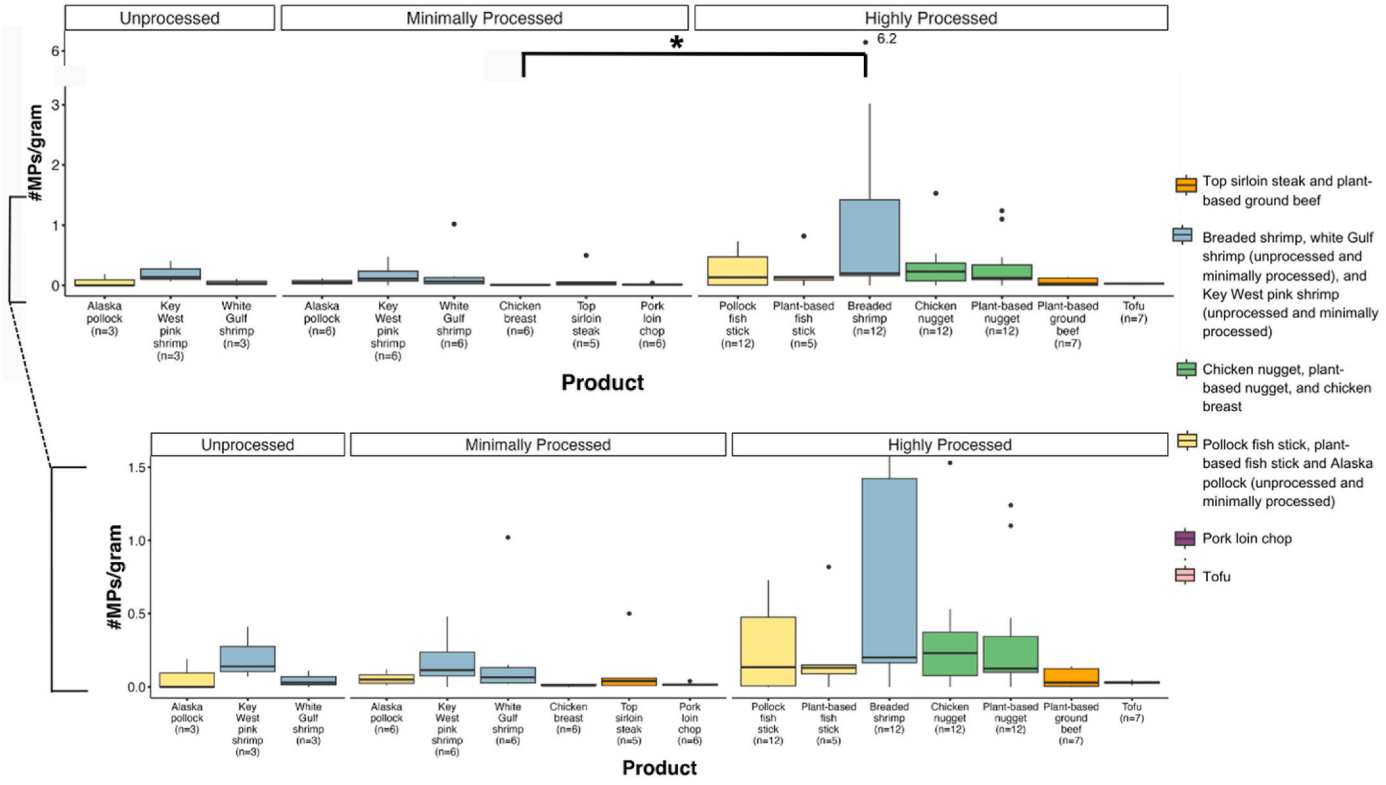

Fig. 2. Boxplot showing counts of microplastics (MPs) standardized per gram (g) of product. Products are organized by relative amounts of processing: unprocessed, minimally-processed, and highly-processed. Products with the same protein base are grouped by color (e.g., pink = pollock based product or its plant-based analogue). * shows statistical significance ($p < 0.05$) between product types, n = sample size. Dots show outliers for each product type. Top panel represents full range of microplastic values per gram (# MPs/gram); bottom panel shows a truncated range of 0–1.5 microplastics per gram (# MPs/gram) to more clearly visualize results within that range. (For interpretation of the references to color in this figure legend, the reader is referred to the Web version of this article.)

which were then averaged to be nationally representative. Low and high annual exposure estimates were also calculated, by using MP contamination estimates from the sample with the lowest and highest observed MPs/g (instead of the mean value) to produce a range of exposures.

3. Results and discussion

3.1. QA/QC

All numbers and figures reported herein are blank-corrected to conservatively account for any contamination generated from the procedure. Following blank-correction, 66% of suspected MPs (1823 particles) were excluded from the data set prior to data analysis, as they were considered below the detection limit. We predominantly observed blue, black and gray fibers in laboratory blanks, meaning that detection of these morphologies in the samples was limited (for further details on the particles observed in the blanks and the calculations for limits of detection refer to Supporting Information Table S4). From the spike and recovery tests, all recoveries were >76% for the procedure (mean 80% recovery of particles), so extraction procedures were deemed appropriate for use (recoveries are in Supporting Information Table S5). Although the exact cause of particle losses was not identified here, losses can occur during sample transfer between beakers, during the rinsing of sieves, or particles may have been missed during visual identification of spike and recovery samples using microscopy.

From the subset of particles that were chemically analyzed, 94% of particles were classified as MPs, 3% as natural, and 3% unknown materials (Fig. S2a) (Munno et al., 2020). Within MPs, 18% were identified as polymers, including rubber. The most common polymers identified were polyethylene terephthalate/polyester ($n = 8$, 36%), polyethylene ($n = 7$; 32%), and polypropylene ($n = 3$; 14%) (Fig. S2b). The remaining 72% of MPs were fibers made from an unknown or cotton/cellulose base with synthetic additives (e.g., dyes), but still considered MPs per the California State Water Board definition (Coffin, 2020). All of our reported numbers and figures are spectroscopy-corrected to reflect the proportion of particles confirmed to be MPs (94% of particles).

3.2. Microplastics in proteins

Microplastics were present in all 16 protein products and in 88% of all samples tested (98/111 samples). Six different morphologies of MPs were observed: fibers, fiber bundles, fragments, rubber, foams, and films. Fibers were the predominant morphology observed (44% of particles; $n = 418$), followed by fragments (30%; $n = 283$), then rubber (19%; $n = 183$) (Fig. S3). The most commonly observed MP colors were blue (34%, $n = 321$), black (27%, $n = 253$), and gray (12%, $n = 117$) (Fig. S4). MPs ranged in size from 0.04 to 27.3 mm in length, measured as longest dimension, and averaged 1.0 ± 1.7 mm (Fig. S5). Sixteen macroplastic (>5 mm in size) particles (15 fibers, and one fragment) were also identified and included in our analyses. The majority of rubber particles (85% of all rubber particles observed) were extracted from just three samples: two plant-based beef samples contained 40 and 25 rubber particles, respectively, and one top sirloin steak sample which contained 91 rubber particles (Fig. S6). Despite these high concentrations of this specific MP morphology in a few individual samples, we observed no significant differences in groupings of MP morphologies across product types or processing levels when visualized using an nMDS plot and statistically testing them using PERMANOVAs (by product: $p = 0.073$, by processing level: $p = 0.35$) (Fig. S7).

Across our 111 samples, we observed an average of 0.3 ± 0.7 MP/g across all products, with a median of 0.1 MP/g and range of 0–6.2 MP/g (Fig. 2). Product-by-product, mean particle concentrations ranged from a low of 0.01 ± 0.01 MP/g in chicken breasts and pork loin chops, to a high of 1.3 ± 1.9 MP/g in breaded shrimp. Significant differences in MPs/g between individual products were evident ($p = 0.002$); however, a post-hoc test indicated that only one single pair of products were significantly different from one another. MP contamination in breaded shrimp was significantly higher than in chicken breasts (Dunn's test, Bonferroni $P.adj = 0.02$) (Fig. 2).

The number of MPs/g was not significantly different between seafoods, terrestrial meats and plant-based proteins (Kruskal-Wallis test, $p = 0.1$) (Fig. S8). Scaled to average serving size (based on survey results), the mean number of MPs consumed in a single serving for seafood, terrestrial meat, and plant-based protein was 120 ± 320, 32 ± 61, and 40 ± 69 MPs, respectively (Baechler et al., 2024). The ubiquity of MPs across protein types suggests that where proteins originate: ocean, land, and animal production facilities, does not have a clear influence on overall MP contamination.

3.3. Influence of processing level, packaging, brand and grocery store type on microplastic concentrations

Highly-processed products contained significantly more MPs/g than minimally-processed products, but not significantly more than unprocessed products (Kruskal-Wallis test, $p = 0.004$) (Fig. 2). When investigating groupings of similar proteins across processing types (i.e., chicken products, fish products, and shrimp products) we found no significant differences except for chicken products; chicken nuggets (highly-processed) had significantly more MPs than chicken breasts (minimally-processed; Kruskal-Wallis test, $p = 0.004$).

The significance of processing level as a driver of MP contamination in the proteins studied may be because more processed foods are subjected to greater amounts of time in the presence of or in contact with plastic food production equipment (e.g., conveyor belts, and worker clothing) than minimally-processed products (Hamacher, 2020; Ramasamy & Subramanian, 2021). For instance, there can be more than a dozen processing steps during tofu production (categorized as a 'highly processed' protein in our study), with each introducing contact with machinery that may add MP contamination to the final product (Corporation, n.d.). In addition, more processed foods spend more time exposed to airborne contamination like dust (Zhu et al., 2022).

We found little evidence to suggest packaging is a major source of contamination in the products studied. Few MPs found in samples matched characteristics of their packaging type (Table S6); we identified only seven total samples of products that contained one or more MPs that matched the properties (morphology and color) of its parent packaging. The largest number of MPs observed in a sample that matched the visual characteristics of their packaging was 11 clear film particles in a grocery-store bought Alaska pollock fillet. The other six samples that had MPs matching their packaging each had less than four particles of that morphology and color. We did not chemically analyze the packaging or the particles from these samples as there was little evidence of contamination directly from packaging; as such, we cannot definitively say these particles actually originated from packaging.

Previous studies have shown variable results with respect to packaging contamination of foods and beverages. For example, Kedzierski et al. (2020) identified polystyrene tray packaging-derived MPs averaging 300–450 μm in size onto packaged poultry products (i.e., chicken breast, turkey escalope) (Kedzierski et al., 2020). Sobhani et al. (2020) reported plastic particles in the size range of 5–20 μm were released from cutting and tearing plastic food packaging. Plastic food containers have also been shown to release millions to billions of sub-micron nanoplastic (NP) particles into the food or liquid it is holding when exposed to high heat in a microwave or similar setting (Deng et al., 2022; K. A. Hussain et al., 2023). Bottled drinking water, a more processed version of tap water, is known to contain higher levels of MP than tap water (Danopoulos et al., 2020b; Gambino et al., 2022). Indeed, MPs have even been detected in bottled drinking water that is packaged in glass, suggesting that while packaging may be a contributor to MPs in water and other foods and beverages, additional contamination sources must also be considered important (Oβmann et al., 2018). The minimum MP particle size analyzed in this study (45 μm) was larger than

packaging particles identified in other research and so it is possible that MP contamination from packaging would be found below this detection limit. Further research on this topic is warranted to better understand packaging's contribution to overall MP contamination in food products, particularly for smaller MPs.

Product brand was not a significant driver of MP contamination for any protein tested in our study, although contamination among products did vary from 0 to 6.2 MP/g. No statistical differences were detected among various brands of the same product type (Kruskal-Wallis tests, $p > 0.05$; Fig. S7). In other studies, variation in MP contamination has been documented among brands, but this is largely dependent on product type. For example, Kutralam-Muniasamy et al. (2020) documented little variation among eight different brands of milk in Mexico (3–11 MP/L) (Kutralam-Muniasamy et al., 2020). Fadare et al. (2020) also found little variation among 23 brands of African table salts (0–1.33 MP/kg). In bottled water, Mason et al. (2018) reported a range of 0 to over 10,000 MP/L, with high variation in particle counts observed even among samples from the same brand. Similarly, we did not observe variation among brands exceeding within brand variation for our study.

We also did not find a significant difference in MP contamination by grocery store type. Our comparison of products obtained from conventional supermarkets versus grocers featuring mostly natural and organic products was not significant (Mann-Whitney U test, $p > 0.05$ for all product pairings tested; Fig. S10, Table S7). As such, natural and organic foods do not appear to be associated with less MP contamination than conventional products. This suggests that, regardless of farming practices, microplastics are present in the atmosphere and soils worldwide (Zhang et al., 2020a,b,c). The lack of significant difference among brands or grocery store types observed in our study indicates that product origin does not significantly impact MP contamination; instead, the processes that occur between harvest and sale may be most likely to introduce MPs into a protein product. However, this research is still nascent and further investigation to determine where along the processing supply chain MP contamination is needed.

3.4. Cumulative annual exposure

We used reported diets of U.S. adults from the national survey reported in Baechler et al. (2024) to determine overall annual exposure to MPs (Table S8). Calculations were conducted for 13 of the 16 product types. Unprocessed versions of Alaska pollock, Gulf shrimp and Florida pink shrimp were excluded from the calculation, as consumption data from Baechler et al. (2024) were not granular enough to distinguish those product types from their minimally-processed counterparts. The minimally-processed versions, purchased at the grocery store were included in the calculation instead. We estimate U.S. adults consume, on average, 11,000 ± 29,000 (median = 4300) MPs/year from a combination of the 13 protein products tested (based on average MP contamination of each of these products; Table S9), with a range of 0–840,000 MP/year. When calculating annual MP exposure based on average reported protein consumption rates, and integrating both the lowest and highest levels of MP contamination found in each individual product type, annual U.S. adult MP exposure ranges between a low of 0 to a high of 3,800,000 MP/year.

3.5. Our findings in the context of previous research

Scientists are increasingly identifying MPs throughout the human body, including our blood (Jenner et al., 2022; ≥700 nm), lungs (Leslie et al., 2022; ≥3 μm), heart (Yang et al., 2023; 20–500 μm), and placentas (Ragusa et al., 2021; 5–10 μm and 20.34–307.29 μm). Our research further elucidates the pathway by which widespread MP contamination within the human food system may be contributing to the MP burdens in our organs and bodies. While current exposure estimates identify inhalation as the likeliest main route of human MP exposure, consumption through food and beverages is another key vector (Cox et al., 2019; Mohamed Nor et al., 2021). It has also become clear through this and other recent works that microplastics are more than just a seafood problem, as has been the preliminary narrative regarding food exposure to date. Based on the present study and others, we are coming to understand that MPs are present within the entire human food system (Zhang et al., 2020b; Danopoulos et al., 2020b; Sewwandi et al., 2023). All 16 protein products included in our study were found to be contaminated with MPs, and are all commonly-consumed by U.S. adults-some more than 10 times per month (Baechler et al., 2024).

Our findings broaden the types and sources of foods in the human diet for which data are now available to inform MP exposure estimates, at least for Americans whose diet contains roughly 15% protein. Previous human MP exposure studies have reviewed the literature on MPs in the eight food and beverage types with the most robust datasets; to date, those have been fish, molluscs, crustaceans, salt, tap water, bottled water, beer, and milk (Danopoulos et al., 2020a; Kosuth et al., 2018; Kutralam-Muniasamy et al., 2020; Liebezeit & Liebezeit, 2014; Vital et al., 2021; Zhang et al., 2020b; Zhang et al., 2023). In aggregate, these foods and beverages represent roughly 20% of the adult diet (Héraud et al., 2013).

In recent years, the consumption of plant-based proteins and meat substitutes has risen substantially due to the rise in popularity of vegan and vegetarianism (Sexton et al., 2022). Despite this trend, we did not locate any previous studies to compare to our results for plant-based proteins. Very few studies were also available to compare to our results for other terrestrial meats. One recent study investigating the presence of plastic particles in beef and pork reported MPs in both sample types (53 -7700 μg/g; Van der Veen et al., 2022). Unfortunately, these findings are presented in units of μg/g so they cannot be directly compared to our results using particle counts. Studies have documented MP contamination of chicken breast meats with one estimate of up to 1.19 MP/g (8 - 1455 μm), though cutting boards were confirmed to be the source of these particles (Habib et al., 2022). Expanded polystyrene packaging has been shown to be a significant source of MP contamination to chicken breast (130–450 μm); however, we found little evidence of contamination from packaging (Kedzierski et al., 2020). Our limited ability to compare our results with other studies highlights the need for further research, specifically into MP contamination in plant-based proteins and terrestrial meats (chicken, beef and pork), with harmonized units of measurement to ensure data from different studies can be integrated to further inform human consumption estimates.

Additionally, our work provides MP contamination data for the edible tissues of Alaska pollock and Gulf shrimp, which are arguably some of the most important seafoods in America; Alaska pollock is the top-landed commercial fishery species in the U.S in terms of volume, and shrimp are the top consumed seafood per capita by Americans by mass (U.S. Department of Commerce, 2020). While several species of Gulf shrimp are commercially caught in the U.S. for human consumption, to our knowledge none have been studied for the presence of MPs. A single study on MP in Alaska pollock has recently been published, positively identifying MPs in the gastrointestinal (GI) tracts of 85% of fish sampled (26 - 4479 μm); however, MP in the fillets (edible tissues) of the fish were not quantified (Ding et al., 2023). While MP contamination is often detected in the GI tracts or guts of seafood specimens (e.g., Andreas Hadibarata et al., 2021; ≥47 μm), MP particles can also accumulate within edible muscle and fillet tissues via translocation (e.g., Akhbarizadeh et al., 2020; 10 - 8000 μm; McIlwraith et al., 2021; 12 - >5000 μm). Thus, data like ours on the presence of MPs in human-consumed tissues are needed to better inform estimates of human MP consumption and levels of risk (Coffin et al., 2022). Additional research is vital to better understanding potential threats that MPs pose to marine ecosystems, ocean food webs, ocean-derived food security, human health, and fishery-related jobs.

While the MP contamination observed in breaded shrimp is an outlier among proteins in this study, this result is not an outlier when

Table 1
U.S. adult exposure to microplastics across different protein product types. Mean serving size and mean number of annual servings were calculated using U.S. adult protein consumption data from a 2021 nationwide social survey conducted by our research team (Baechler et al., 2024). For each sample type, exposure was calculated using survey data for the mean number of annual servings ("Avg MPs/year ± SD" column), low number of annual servings (one serving monthly scaled to 12 servings annually), and high number of annual servings (10 servings monthly scaled to 120 servings annually) ("Range of MPs/year" column). For calculation of mean number of servings only nonzero answers were used (i.e., survey respondents that never consumed that certain type of protein were excluded from the analysis). Raw data can be obtained from the supplementary file titled "Sample Sheets Raw." MP = microplastic; SD = standard deviation; # = number; g = gram.

Protein type	MP concentrations and reported protein consumption					Annual U.S. adult exposure	
	Sample type	Avg # MPs/g	Mean serving size (g)	Avg # MPs/ serving ± SD	Mean # servings/yr	Avg # MPs/yr ± SD	Range of MPs/yr ± SD
Seafood	**Breaded shrimp**	**1.2**	**320**	**370 ± 580**	**35**	**13,000 ± 21,000**	**4400 ± 6800 -44,000 ± 68,000**
	Pollock fish stick	**0.26**	**220**	**58 ± 57**	**37**	**2100 ± 2100**	**680 ± 670 -6800 ± 6700**
	White Gulf shrimp (minimally processed)	**0.22**	**240**	**54 ± 87**	**40**	**2100 ± 3700**	**640 ± 1000–6400 ± 10,000**
	Key West pink shrimp (fresh caught)	**0.20**	**240**	**49 ± 36**	**40**	**1900 ± 1500**	**600 ± 420 -6000 ± 4200**
	Key West pink shrimp (minimally processed)	**0.17**	**240**	**42 ± 39**	**40**	**1700 ± 1600**	**500 ± 460 -5000 ± 4500**
	Alaska pollock (fresh caught)	**0.06**	**180**	**11 ± 16**	**36**	**390 ± 590**	**130 ± 190 -1300 ± 1800**
	Alaska pollock (minimally processed)	**0.05**	**180**	**9 ± 7**	**36**	**330 ± 240**	**120 ± 76 -1200 ± 760**
	White Gulf shrimp (fresh caught)	**0.04**	**240**	**10 ± 11**	**40**	**390 ± 460**	**130 ± 130 -1300 ± 1300**
Terrestrial Meat	**Chicken nugget**	**0.31**	**200**	**62 ± 78**	**45**	**2800 ± 3800**	**750 ± 920 -7500 ± 9200**
	Top sirloin steak	**0.12**	**200**	**25 ± 38**	**73**	**1800 ± 3100**	**300 ± 440 -3000 ± 4400**
	Pork loin chop	**0.02**	**190**	**4 ± 2**	**50**	**188 ± 99**	**38 ± 22–380 ± 220**
	Chicken breast	**0.01**	**190**	**2 ± 2**	**75**	**140 ± 130**	**29 ± 18–290 ± 180**
Plant-based protein	**Plant-based nugget**	**0.32**	**230**	**73 ± 90**	**46**	**3300 ± 4400**	**874 ± 1000–8700 ± 11,000**
	Plant-based fish stick	**0.23**	**200**	**46 ± 59**	**47**	**2100 ± 2600**	**560 ± 690 -5600 ± 6900**
	Plant-based ground beef	**0.06**	**170**	**10 ± 10**	**44**	**440 ± 430**	**120 ± 110 -1200 ± 1100**
	Tofu	**0.03**	**230**	**7 ± 3**	**42**	**290 ± 120**	**78 ± 30–780 ± 300**

compared to the broader suite of human-consumed foods and beverages that have previously been investigated for MPs; in fact, the concentration of MPs/g in the U.S. shrimp products included in this study (White Gulf shrimp, Key West pink shrimp, breaded shrimp) all fall below MP values for shrimp species in different locations from other studies (Table 1) (Cox et al., 2019; Fernández Severini et al., 2020; Keshavarzifard et al., 2021; Valencia-Castañeda et al., 2022). It should also be noted that the results presented in this study are likely conservative compared to other works due to our methods including both blank and spectroscopy correction of the data. Additionally, while we intentionally focused on U.S.-sourced products for this study, the specific origin and species of the shrimp contained in the breaded shrimp products was unclear. We know that both the White Gulf shrimp and Key West pink shrimp were wild-caught within U.S. waters; however, while the breaded shrimp were labeled as being a U.S. product, we do not know the species, country of origin, or whether the shrimp were farmed or wild-caught. With Americans consuming over 5 pounds of shrimp per capita as of 2021, (National Fisheries Institute, 2021) further research is needed to understand relative MP contamination between farmed and wild-caught shrimp and other seafood, including potential differences between domestic and imported sources and how this may relate to seafood handling and processing.

3.6. Study limitations

While our study significantly advances the understanding of MP prevalence in the human food system, there are some inherent limitations to our findings. Due to the limit of detection associated with counting and identifying suspected MP particles under a microscope, our results pertain only to microplastics 45 μm and larger. This means that nanoplastics (NPs) and any MPs <45 μm are not included in our results. Generally, an increase in MP particle counts is observed with decreasing particle size (Kooi & Koelmans, 2019); therefore, our reported numbers are likely an underestimate of overall NP and MP contamination in these samples, and an underestimate of human microplastic ingestion from the protein types studied in this work. The majority of studies on the impacts of NP and MPs on human health focus on smaller particles <20 μm (Yee et al., 2021). Still, the size-range measured here is relevant to human health as particles >45 μm have been observed to translocate to the tissue and organs of fish (McIlwraith et al., 2021; Collard et al., 2017), and articles <150 μm can translocate across the gut epithelium following ingestion, causing systemic exposure and hazards to human health (EFSA Panel on Contaminants in the Food Chain (CONTAM), 2016). Moreover, we may have missed particles from packaging degradation due to our method. Evidence of MP contamination from opening plastic packaging reported particles in the range of 5–20 μm (Sobhani et al., 2020). Therefore, we may not have observed contamination from plastic packaging in our results, as they may have been below the detection limit of this study.

In addition, we were only able to test a limited number of samples per product during this study. Sample numbers ranged from 3 to 12 per product, depending on processing level and thus availability of different brands. Our limited sample size, combined with high variability in MP concentrations between and among our samples (and many outliers), may have limited the power in some of our statistical tests to detect statistically significant differences (e.g., between brands and stores). This may be one of the reasons we did not see many statistical differences between product types. In addition, a few samples were unusable because of sample destruction or equipment breakage during processing. This reduced sample size even further for some product types and

may have impacted the power of the ensuing statistical tests to identify differences.

3.7. Exposure of U.S. Adults to microplastics, per serving and per year

Based on MP counts and survey data on U.S. adult protein consumption, we estimate that, for the 16 protein types studied, American adults consume an average of 74 ± 220 MPs per serving of protein (Baechler et al., 2024). Scaled to annual consumption, when considering consumption of only a single protein type, the mean exposure is 1500 ± 5000 MPs annually. Average annual consumption among these products ranges from 140 ± 130 MP/year for chicken breast (product with the lowest average MP concentration of those studied), to 13,000 ± 21,000 for breaded shrimp (product with the highest average MP concentration of those studied). Additional annual MP consumption estimates by product can be found in Table 1 and Fig. S10.

3.8. Relevance of findings

The goal of this study was to contribute to a more holistic understanding of human MP exposure from the foods we eat and to foster a greater awareness about MP contamination within our food system. Our findings broaden overall understanding of human exposure to MPs through foods, specifically for adults in the U.S. who consume U.S.-based proteins. However, due to the lack of a risk assessment framework for human health, no conclusions can be made at present about the effects of direct consumption of the MPs identified in the protein products we tested (Coffin et al., 2022). Future work should investigate additional foods consumed in the U.S. that are important constituents of the American diet (e.g., dairy products, grains, fruits, vegetables). More broadly, terrestrial meats and plant-based proteins (along with aquatic-derived farmed and wild-caught proteins) are key protein sources globally – yet they remain understudied in MPs research. Future studies should investigate MPs in a range of products, sourced from both within and outside the U.S., to better distinguish how contamination may vary globally and further understand its drivers. These studies should also attempt to address the critical gaps that limit our current understanding of MP toxicity, whether these materials are intentionally ingested by humans or otherwise consumed (Coffin et al., 2022). Additional data on MPs in commonly-consumed but otherwise unstudied foods are needed to more accurately estimate human MP exposure and ultimately, to determine MP risk thresholds for human health.

CRediT authorship contribution statement

Madeleine H. Milne: Data curation, Formal analysis, Investigation, Visualization, Writing – original draft, Writing – review & editing. **Hannah De Frond:** Writing – original draft, Writing – review & editing. **Chelsea M. Rochman:** Methodology, Project administration, Writing – review & editing. **Nicholas J. Mallos:** Conceptualization, Methodology, Writing – review & editing. **George H. Leonard:** Conceptualization, Methodology, Writing – review & editing. **Britta R. Baechler:** Conceptualization, Methodology, Project administration, Writing – original draft, Writing – review & editing.

Declaration of competing 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.

Data availability

The raw data has been included as an Excel file.

Acknowledgements

Authors would like to thank the McConnell Family Foundation for their generous financial support of this research. Authors would also like to thank the three anonymous reviewers and journal editor for providing useful feedback that improved the quality of this work.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2023.123233.

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