Marine Pollution Bulletin 173 (2021) 113111

Contents lists available at ScienceDirect

Marine Pollution Bulletin

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

Plastic ingestion by green turtles (*Chelonia mydas*) over 33 years along the coast of Texas, USA

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ARTICLE INFO

Keywords:
Chelonia mydas
Anthropogenic marine debris
Debris ingestion
Trash
Gulf of Mexico
Endangered species

ABSTRACT

Despite exponential growth of anthropogenic marine debris in recent decades, plastic ingestion by marine turtles in the Gulf of Mexico is not well understood. Gastrointestinal tracts were examined from 464 green turtles that stranded in Texas between 1987 and 2019, and 226 turtles ingested plastic (48.7%). This number doubled from 32.5% in 1987–1999 to 65.5% in 2019, but mass of ingested items was lowest in 2019. No turtles showed evidence of death directly related to plastic ingestion. Compared to other regions, plastic ingestion was low. Small turtles (<25 cm straight carapace length) ingested plastic more frequently and in greater amounts than larger turtles. Small turtles also ingested more hard plastic while larger turtles ingested more sheet-like and thread-like plastics, which may correspond to size-based habitat shifts. This is among the largest marine turtle ingestion studies to date and demonstrates an increasing prevalence of plastic ingestion.

1. Introduction

Prevalence of anthropogenic debris in the marine environment has grown exponentially in recent decades (Avio et al., 2017). Between 1950 and 1989, annual global plastic production increased exponentially from 1.5 to 100 million metric tons and reached 368 million metric tons by 2019 (PlasticsEurope, 2010, 2020). Plastic and other anthropogenic debris (hereafter, plastic) have become one of the greatest threats to marine fauna and affect a wide range of marine vertebrates, primarily through direct ingestion or entanglement (Derraik, 2002; Galgani et al., 2019; Kühn and Van Franeker, 2020; Nicolau et al., 2016; Schuyler et al., 2014). Among taxa most affected are marine turtles, of which all seven species have been found to ingest plastic (Nelms et al., 2016). Though the number of peer-reviewed studies investigating marine turtle plastic ingestion has grown rapidly over the past decade (Lynch, 2018), the impact of oceanic plastic is not yet well-understood and there remains an urgent need for further study of plastic ingestion by marine turtles, especially in regions and species receiving less research focus (Clukey et al., 2017; Hamann et al., 2010; Nelms et al., 2016; Vegter et al., 2014).

Globally, the majority of plastic ingestion studies on marine turtles have occurred in the Atlantic Ocean, but comparatively little is known about plastic ingestion by marine turtles within the Gulf of Mexico (GoM) (for review, see Lynch, 2018, Nelms et al., 2016). Worldwide, plastic often occurs at higher densities in semi-enclosed seas like the GoM (Barnes et al., 2009; Lebreton et al., 2012), and models suggest moderately high plastic density in the GoM (Eriksen et al., 2014). Further, density of microplastics (<5 mm) within the GoM is among the highest worldwide (Di Mauro et al., 2017). However, except for a few reports between 2000 and 2011, plastic ingestion by marine turtles in the GoM has received little attention since the 1990s (Table S1). These studies have included all five species of marine turtle known to occur in the region, but loggerhead (*Caretta caretta*) and Kemp's ridley (*Lepidochelys kempii*) turtles have received the most focus, and less is known about others, such as green turtles (*Chelonia mydas*). In addition, the primary metric of plastic ingestion reported across these studies was percent frequency of occurrence (the percent of turtles which ingested at least one item [%FO]), which is "essentially meaningless" for small sample sizes (Casale et al., 2016). Despite multiple research efforts, there is little meaningful quantitative information regarding plastic ingestion by green turtles in the GoM.

In addition to geographic information gaps, few studies have quantified temporal trends in marine turtle plastic ingestion, which are vital to assessing changing threats to marine turtle conservation and in tracking the amount of plastic in the marine environment (Lynch, 2018; Nelms et al., 2016). Successful comparisons over time require a long

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https://doi.org/10.1016/j.marpolbul.2021.113111
Received 7 August 2021; Received in revised form 26 October 2021; Accepted 27 October 2021

collection period and adequate sample size (Casale et al., 2016), but most studies have spanned less than five years (Nelms et al., 2016). Further, 27% have included just one turtle while less than 5% have had sample sizes exceeding 150 individuals (Lynch, 2018). Long term studies with large sample sizes may also be important in detecting population-level effects of marine turtle plastic ingestion, for which evidence is inconclusive (Senko et al., 2020). Across species and geographic regions, there is a major need to generate high-quality baseline information about marine turtle plastic ingestion that is informed by large sample sizes.

Accurate and reliable data on plastic ingestion requires standardization in both data collection and reporting, something largely absent from the literature (Casale et al., 2016; Fossi et al., 2018; Lynch, 2018; Provencher et al., 2017; Schuyler et al., 2014). Most studies only report %FO, which is not representative of actual impacts to individuals or populations (Casale et al., 2016; Lynch, 2018; Nelms et al., 2016). Among those reporting a quantity (e.g., number or mass of pieces), most have excluded individuals not ingesting any plastic, which can obscure data interpretation (Lynch, 2018). Recent studies have suggested standardized procedures for data collection and reporting, emphasizing the importance of combining multiple metrics such as number, mass, and size of pieces (Clukey et al., 2017; Lynch, 2018; Matiddi et al., 2019; Nelms et al., 2016; Rizzi et al., 2019). In addition, characterizing ingested items by type (e.g., plastic bags vs. fishing line) and color is important, and a standardized approach has been recommended based on successful methodology used to monitor plastic ingestion by seabirds (Duncan et al., 2019; Matiddi et al., 2019). Global standardization in documenting marine turtle plastic ingestion has been widely called for (Domènech et al., 2019; Duncan et al., 2019; Lynch, 2018; Nelms et al., 2016; Provencher et al., 2017).

Measuring the severity of marine turtle plastic ingestion in specific regions and species is necessary for directing policy initiatives and conservation strategies. However, to date, no study in the GoM has included the sample size or standardized procedure necessary for reliable assessment of the threat of plastic pollution on marine turtles. Therefore, this study examines plastic ingestion by a large number of green turtles found stranded along the Texas coast between 1987 and 2019. Study objectives were to 1) establish standardized baseline information of plastic ingestion by green turtles in the GoM to which future studies can be compared; 2) investigate trends in plastic ingestion over a three-decade period; and 3) determine what factors predict plastic ingestion by green turtles in the northwestern GoM.

2. Materials and methods

2.1. Sample collection

The gastrointestinal tracts (GITs) were examined of 276 green turtles that were incidentally captured (e.g., hook and line, power plant intake) or found stranded (floating or washed ashore either dead or alive) along the GoM shoreline and along bays and estuaries in Texas, USA, from 2007 through 2009 and 2019 (Fig. 1). This data was supplemented by examining dried samples of GIT contents from 188 turtles collected in the same area from 1987 through 2002, yielding a total sample size of 464 individual turtles. A subset of turtles collected between 1987 and 2009 (n = 233) were also examined for foraging habits in Howell and Shaver (2021). Included were turtles found alive but which died during transfer to a rehabilitation center (n = 72), turtles found freshly dead (n = 248), and turtles found with moderate decomposition (n = 144); turtles with severe decomposition were excluded. All turtles were collected by the Texas Sea Turtle Stranding and Salvage Network. Straight carapace length (SCL) was measured to the nearest tenth of a centimeter using metal calipers from the nuchal notch to the posterior tip. After collection or death, all turtles were necropsied immediately or frozen for future necropsy; during necropsy, GITs were separated and frozen for later examination.

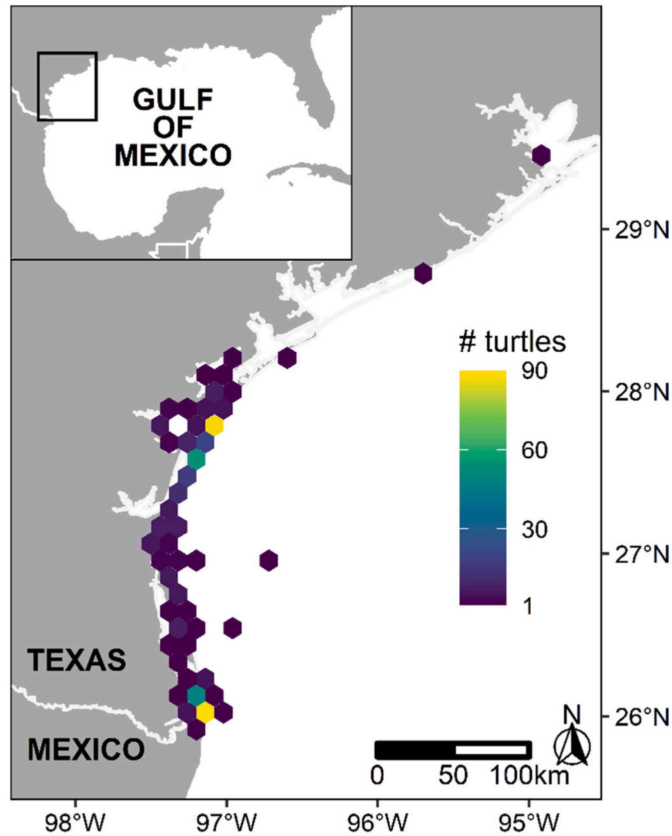

Fig. 1. Study region and collection locations of 461 green turtles (*Chelonia mydas*) found stranded in Texas between 1987 and 2019. Turtles were grouped into 0.12° grid cells. Three additional turtles were included in the project but lacked specific coordinates and are not represented in this map. Two turtles were found offshore, one during an offshore netting project and one found within the stomach of a fish caught by rod and reel.

Following standardized methodology for examining plastic ingestion by marine turtles, the entire GIT was separated into sections (esophagus/stomach, and small and large intestines), and the contents of each were removed by incision and physical manipulation (Matiddi et al., 2019). GIT contents were then rinsed with running tap water over a 1 mm sieve, and anthropogenic debris was separated from food remains and natural debris (e.g., stones, wood, feathers, etc.) and dried in an oven for 24 h at 50 °C.

The mass (g) of each ingested item was recorded using a digital scale (0.01 g precision) and length and width (mm) were measured using digital calipers (0.01 mm precision). Items were additionally classified by category and color following suggested protocols (Duncan et al., 2019; Matiddi et al., 2019). Categories included industrial plastic (i.e., nurdles), sheet-like plastics, thread-like plastics, foam, hard fragments, other plastics, and non-plastics (for description, see Table 1).

Color was recorded as black, blue, brown, clear, gray, green, orange, pink/purple, red, white, or yellow. Items less than 5 mm in length, or less than 25 mm^2, were not individually measured, as it was assumed these were fragments from larger items within the GIT (Santos et al., 2015; Stahelin et al., 2012). Instead, these items were grouped by category and color, mass was recorded for the group, and the items were counted as one item. However, all industrial plastics were individually measured, regardless of size, as these often exist unfragmented at small sizes. Further, industrial plastics were of special interest due to recent high concentrations of pellets found along the Texas coast (Tunnell et al., 2020).

Table 1
Description of standardized categories used to characterize debris items ingested by marine turtles. Terms are shortened names of each category used in this study. Information adapted from Matiddi et al. (2019).

Category	Term	Definition and Examples
Industrial plastic	Industrial plastic	Industrial grade plastic pellets and granules (also known as "nurdles")
Sheet-like plastic	Sheet	Thin and flexible plastics such as plastic bags, food wrappers, or tape
Thread-like plastic	Thread	Filamentous plastics such as fishing line, plastic ropes, or very thin fragments from larger items (e.g., shreds of woven tarps)
Foamed plastic	Foam	Polystyrene or foamed soft rubber
Hard plastic	Fragment	Rigid, often sharp or jagged plastic pieces that are usually fragments of larger plastic items such as milk cartons or plastic forks
Other plastic	Other plastic	All plastics that do not fit into one of the categories above (e.g., balloons, many dense rubbers)
Other than plastic	Non-plastic	All non-plastic debris items, such as cigarettes, cloth, paper, or metal

2.2. Data analysis

All statistical analyses were performed in R 4.0.2 (R Core Team, 2020). Total number and mass of ingested items have each been used in past studies of marine turtle plastic ingestion (e.g., Domènech et al., 2019; Rizzi et al., 2019), so both metrics were used as response variables. For turtles ingesting a total mass of 0.00 g, values were converted to 0.001 g to avoid underestimating mass of ingested plastics.

Model parameters included time period, season, latitude, inshore vs. offshore, and size class. To minimize potential biases associated with small sample size and year to year variation, turtles were grouped by decade, and turtles from 1987 to 1989 ($n = 25$) were grouped with those from the 1990s, resulting in three periods, 1987–1999 ($n = 169$), 2000–2009 ($n = 112$), and 2019 ($n = 183$). No GITs were included from turtles between 2010 and 2018 as few GITs were collected from turtles found during this period. Based on turtle collection date, season (Northern Hemisphere) of stranding was assigned as spring (March–May), summer (June–August), fall (September–November), or winter (December–February). Each stranding location was classified as inshore (lagoons, bays, channels) or offshore (floating near or stranded on GoM beaches). Size class was assigned based on partitions by Howell et al. (2016): pelagics (<25.0 cm SCL), recruits (25.0–34.9 cm SCL), transitionals (35.0–44.9 cm SCL), and subadults (≥45.0 cm SCL). Only two individuals were considered adults (greater than about 84 cm SCL; Almeida et al., 2011), so these were analyzed together with subadults (one did not ingest any debris items and one ingested an amount similar to other subadults). Measurements for ten turtles which lacked SCL were estimated from curved carapace length (CCL) using an equation derived from turtles used in this study with both measurements ($SCL = 0.561 + 0.924 \times CCL$, $n = 451$, $r^2 = 0.98$, $p < 0.001$).

The general variance of inflation factor (Fox and Monette, 1992) between all parameters was calculated, using a threshold value of 3 (Zuur et al., 2010). No significant correlations were detected, and all parameters were retained. Five turtles which lacked values for size or location were removed from analyses.

The distribution of both total number and mass of plastic items was highly right-skewed and each was 51.4% zeros (Fig. 2). Thus, plastic ingestion was modeled using hurdle (zero-altered) models, which consist of two parts, a Bernoulli distribution to model the presence or absence of ingestion, and a separate distribution to model non-zero data. Zero-altered negative binomial (ZANB) models were used for number of items, as count data were overdispersed, and zero-altered gamma (ZAG) models were used for continuous mass data. For both ZANB and ZAG models, all combinations of the five parameters were fit. ZANB models were fit using the 'pscl' package (Zeileis et al., 2008). For the ZAG models, the Bernoulli and gamma parts were constructed separately using the 'MuMIn' package (Barton, 2020), and then combined following Zuur and Ieno (2016). Models were ranked using AIC, and the models with the lowest AIC values were considered the most parsimonious (Burnham and Anderson, 2002). Model assumptions were verified by plotting residuals against fitted values and against each parameter, and by examining simulated residual plots using the 'DHARMa' package (Hartig, 2020). Marginal effects of model parameters are reported with 95% confidence intervals.

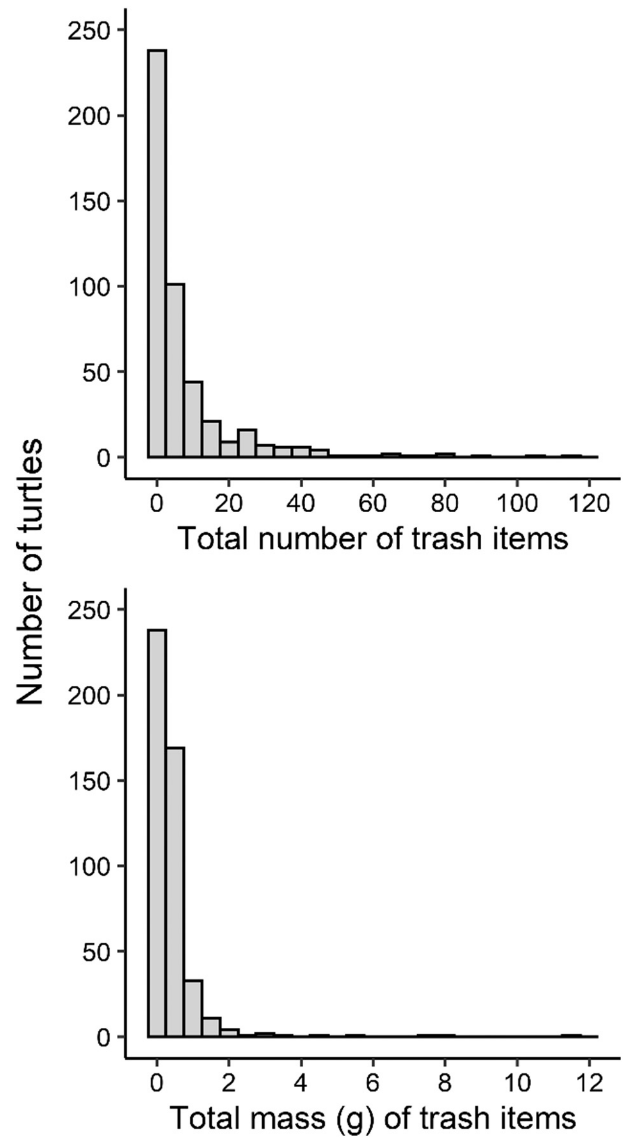

Fig. 2. Frequencies of total number and mass of debris items ingested by 464 green turtles (*Chelonia mydas*) found stranded in Texas between 1987 and 2019.

The mean number and mass of plastic items were calculated for each size class and during each period. Mean surface area was also estimated using the equation for a rectangular prism. Data were not normally distributed, and Kruskal-Wallis tests were used to detect significant differences between periods or size classes. Using the 'FSA' package (Ogle et al., 2020), non-parametric Dunn's tests were used to determine pair-wise significant differences (Dunn, 1964). Kruskal-Wallis and Dunn's tests were also used to determine significant differences in turtle size (SCL) between time periods. Means are reported with one standard deviation unless otherwise noted.

The percent frequency of occurrence (%FO) of plastic ingestion was calculated overall, and per category, color, and GIT section, as the percent of turtles which ingested at least one debris item. The 95% confidence interval of %FO was calculated using Jeffrey's interval (Provencher et al., 2017) in the 'DescTools' package (Signorell et al., 2020). Conditional %FO, defined as the percent of turtles ingesting more than a threshold amount of plastic, was calculated across a range of values for body burden. Body burden was defined as the number or mass of ingested plastic items per cm SCL, which may be a better estimate of body burden than body mass, which can fluctuate with emaciation (Lynch, 2018). Jeffrey's 95% confidence interval was also calculated for conditional %FO.

Table 2
Number, mass, and surface area of ingested debris items and frequency of occurrence (%FO) by size class and time period for green turtles (*Chelonia mydas*) in Texas between 1987 and 2019. Mean and one standard deviation (SD) are presented for all turtles (All) and for only turtles which ingested debris (Ing.). Range is given only for turtles which ingested debris. Periods include 1987–1999 (P1), 2000–2009 (P2), and 2019 (P3). Mean and one standard deviation straight carapace length (SCL) is provided by size class and time period. Total sample size (n = 464) is greater than sum of size class subtotals (*n* = 462) because length data was missing for two turtles.

Size class	Period	Sample size	Mean SCL ± SD	%FO (95% CI)	Number			Debris mass (g)			Debris surface area (cm^2)		
					Mean ± SD		Range	Mean ± SD		Range	Mean ± SD		Range
					All	Ing.		All	Ing.		All	Ing.	
Pelagic	P1	18	20.7 ± 4.4	55.6% (33.2–76.3%)	13.7 ± 19.9	24.7 ± 21.0	4–75	0.88 ± 2.00	1.59 ± 2.46	0.04–7.37	74.82 ± 197.44	134.67 ± 249.22	1.02–863.27
Pelagic	P2	10	21.4 ± 4.6	50.0% (22.3–77.6%)	7.8 ± 12.7	15.6 ± 14.2	2–43	0.34 ± 0.55	0.68 ± 0.61	0.08–1.46	20.08 ± 33.78	40.16 ± 38.42	9.33–114.81
Pelagic	P3	31	23.7 ± 1.0	96.8% (85.9–99.6%)	10.6 ± 13.6	10.9 ± 13.7	1–57	0.20 ± 0.35	0.20 ± 0.36	0.00–1.68	34.56 ± 65.00	35.71 ± 65.77	0.18–336.90
Pelagic	All	59	22.4 ± 3.4	76.3% (64.3–85.7%)	11.1 ± 15.8	14.5 ± 16.6	1–75	0.43 ± 1.20	0.57 ± 1.34	0.00–7.37	44.39 ± 121.41	58.20 ± 136.10	0.18–863.27
Recruit	P1	81	30.1 ± 2.7	43.2% (32.8–54.1%)	4.6 ± 10.4	10.6 ± 13.6	1–78	0.20 ± 0.51	0.47 ± 0.70	0.00–3.32	21.18 ± 48.60	49.02 ± 64.05	0.00–299.47
Recruit	P2	56	29.4 ± 2.8	53.6% (40.6–66.2%)	7.3 ± 13.7	13.7 ± 16.2	1–61	0.23 ± 0.48	0.43 ± 0.59	0.00–2.58	23.49 ± 38.37	43.84 ± 43.07	0.42–164.61
Recruit	P3	107	28.2 ± 2.7	69.2% (60.0–77.3%)	8.3 ± 15.4	12.0 ± 17.3	1–114	0.31 ± 0.92	0.45 ± 1.08	0.00–7.83	36.75 ± 77.42	53.14 ± 88.30	0.00–494.43
Recruit	All	244	29.1 ± 2.8	57.0% (50.7–63.1%)	6.9 ± 13.6	12.0 ± 16.3	1–114	0.25 ± 0.72	0.45 ± 0.90	0.00–7.83	28.54 ± 61.68	50.10 ± 74.82	0.00–494.43
Transitional	P1	48	38.5 ± 2.6	14.6% (6.8–26.5%)	1.0 ± 3.5	6.7 ± 6.6	1–22	0.05 ± 0.15	0.31 ± 0.26	0.01–0.64	11.33 ± 43.22	77.68 ± 87.50	5.20–243.50
Transitional	P2	28	39.4 ± 3.1	35.7% (20.1–54.2%)	1.9 ± 3.7	5.3 ± 4.4	1–14	0.05 ± 0.10	0.15 ± 0.11	0.00–0.37	16.94 ± 32.34	47.42 ± 38.50	1.33–113.33
Transitional	P3	22	38.1 ± 2.6	36.4% (18.9–57.1%)	6.3 ± 21.4	17.3 ± 32.8	1–103	0.03 ± 0.07	0.07 ± 0.10	0.00–0.31	8.84 ± 25.99	24.31 ± 38.50	0.11–124.21
Transitional	All	98	38.7 ± 2.8	25.5% (17.7–34.8%)	2.4 ± 10.8	9.5 ± 19.8	1–103	0.04 ± 0.12	0.17 ± 0.19	0.00–0.64	12.37 ± 37.08	48.50 ± 60.31	0.11–243.50
Subadult	P1	20	55.5 ± 11.1	10.0% (2.1–28.4%)	0.3 ± 0.9	2.5 ± 1.5	1–4	0.01 ± 0.02	0.06 ± 0.05	0.01–0.10	1.51 ± 6.46	15.05 ± 14.59	0.46–29.64
Subadult	P2	18	56.5 ± 6.8	33.3% (15.3–56.3%)	6.1 ± 15.9	18.3 ± 23.1	1–65	0.73 ± 2.54	2.18 ± 4.03	0.00–11.11	316.95 ± 1232.24	950.85 ± 1988.09	2.13–5395.26
Subadult	P3	23	55.4 ± 12.1	34.8% (18.0–55.1%)	8.7 ± 22.9	25.0 ± 33.1	1–89	0.13 ± 0.27	0.38 ± 0.34	0.00–0.93	39.96 ± 83.63	114.88 ± 107.24	0.36–276.84
Subadult	All	61	55.8 ± 10.3	26.2% (16.5–38.2%)	5.2 ± 16.9	19.7 ± 28.3	1–89	0.27 ± 1.42	1.01 ± 2.64	0.00–11.11	109.09 ± 684.88	415.89 ± 1288.67	0.36–5395.26
All	P1	169	34.5 ± 10.5	32.5% (25.8–39.9%)	4.0 ± 10.6	12.2 ± 15.6	1–78	0.20 ± 0.79	0.63 ± 1.28	0.00–7.37	21.52 ± 78.77	66.11 ± 126.96	0.00–863.27
All	P2	112	35.6 ± 11.3	45.5% (36.5–54.8%)	5.8 ± 12.5	12.8 ± 16.0	1–65	0.27 ± 1.11	0.60 ± 1.58	0.00–11.11	68.71 ± 506.89	150.89 ± 742.87	0.42–5395.26
All	P3	183	32.0 ± 10.8	65.6% (58.5–72.2%)	8.5 ± 17.1	13.0 ± 19.7	1–114	0.23 ± 0.73	0.36 ± 0.88	0.00–7.83	33.43 ± 72.56	50.98 ± 84.47	0.00–494.43
All	All	464	33.8 ± 10.9	48.7% (44.2–53.2%)	6.2 ± 14.1	12.8 ± 18.0	1–114	0.23 ± 0.86	0.48 ± 1.18	0.00–11.11	37.61 ± 258.25	77.21 ± 365.88	0.00–5395.26

3. Results

3.1. General overview

Debris ingestion was recorded for 238 out of 464 turtles (48.7% [CI: 44.2–53.2%]). Turtle size ranged from 7.3 to 106.5 cm SCL (mean = 33.8 ± 10.7) and differed between time periods ($\chi^2_2 = 23.38$, $p < 0.001$) with significant ($p < 0.001$) pairwise differences between 1987–1999 (mean = 34.5 ± 10.5, $n = 169$) and 2019 (mean = 32.0 ± 10.8, $n = 183$) and between 2000–2009 (mean = 35.6 ± 11.3, $n = 112$) and 2019 (Table 2). In total, turtles ingested 2882 individual plastic items with a mass of 107.94 g. Mean ingested quantities of plastic were 6.2 ± 14.1 items and 0.23 ± 0.86 g, respectively. Excluding turtles which did not ingest any debris, mean quantities were 12.8 ± 18.0 items and 0.48 ± 1.18 g (Fig. 3, Table 2). The highest number of items (114) was found in a recruit (26.0 cm SCL) and the highest mass of items (11.11 g) was found in a subadult (45.8 cm SCL) (Fig. S1). No turtles showed evidence of obstruction or penetration of the GIT or death otherwise related to plastic ingestion.

Most items were found in the intestines (84.1%) (Table 3). The most common plastic category was sheets (37.8%), followed by fragments (27.1%) and threads (22.9%) (Table 3). In total, 16 industrial plastic pellets were found across nine turtles. By mass, fragments were the most common (40.2%), followed by sheets (26.1%) and other plastics (25.7%); by surface area, sheets were dominant (75.0%) (Table 3). Most items were clear (32.7%) or white (23.3%) (Table 3). Mean mass per item was highest for other plastics (0.11 ± 0.19 g) and fragments (0.06 ± 0.17 g), and mean surface area per item was highest for sheets (12.60 ± 37.73 cm^2) and non-plastics (7.78 ± 9.89 cm^2) (Table S2).

3.2. Size class trends

Percent frequency of occurrence was highest for pelagics (76.3% [CI: 64.3–85.7%], $n = 59$), intermediate for recruits (57.0% [CI: 50.7–63.1%], $n = 244$), and lowest for transitionals (25.5% [CI: 17.7–34.8%], $n = 98$) and subadults (26.2% [CI: 16.5–38.2%], $n = 61$). Mean number of ingested items differed between size class ($\chi^2_3 = 57.95$, $p < 0.001$) and was highest for pelagics (11.1 ± 15.8 items), followed by recruits (6.9 ± 13.6 items), subadults (5.2 ± 16.9 items), and transitionals (2.4 ± 10.8 items), with significant pairwise differences between all size classes ($p < 0.01$) except transitionals and subadults. Mean mass differed between size classes ($\chi^2_3 = 52.08$, $p < 0.001$) and was highest for pelagics (0.43 ± 1.20 g), followed by subadults (0.27 ± 1.42 g), recruits (0.25 ± 0.72 g), and transitionals (0.04 ± 0.12 g), with significant pairwise differences between all size classes ($p < 0.05$) except transitionals and subadults. Mean surface area differed between size classes ($\chi^2_3 = 42.88$, $p < 0.001$) and was highest for subadults (109.09 ± 684.88 cm^2), followed by pelagics (44.39 ± 121.41 cm^2), recruits (28.54 ± 61.68 cm^2), and transitionals (12.37 ± 37.08 cm^2), with significant pairwise differences between all size classes ($p < 0.05$) except transitionals and subadults. Excluding turtles which did not ingest debris, there were no significant size class differences for mean number

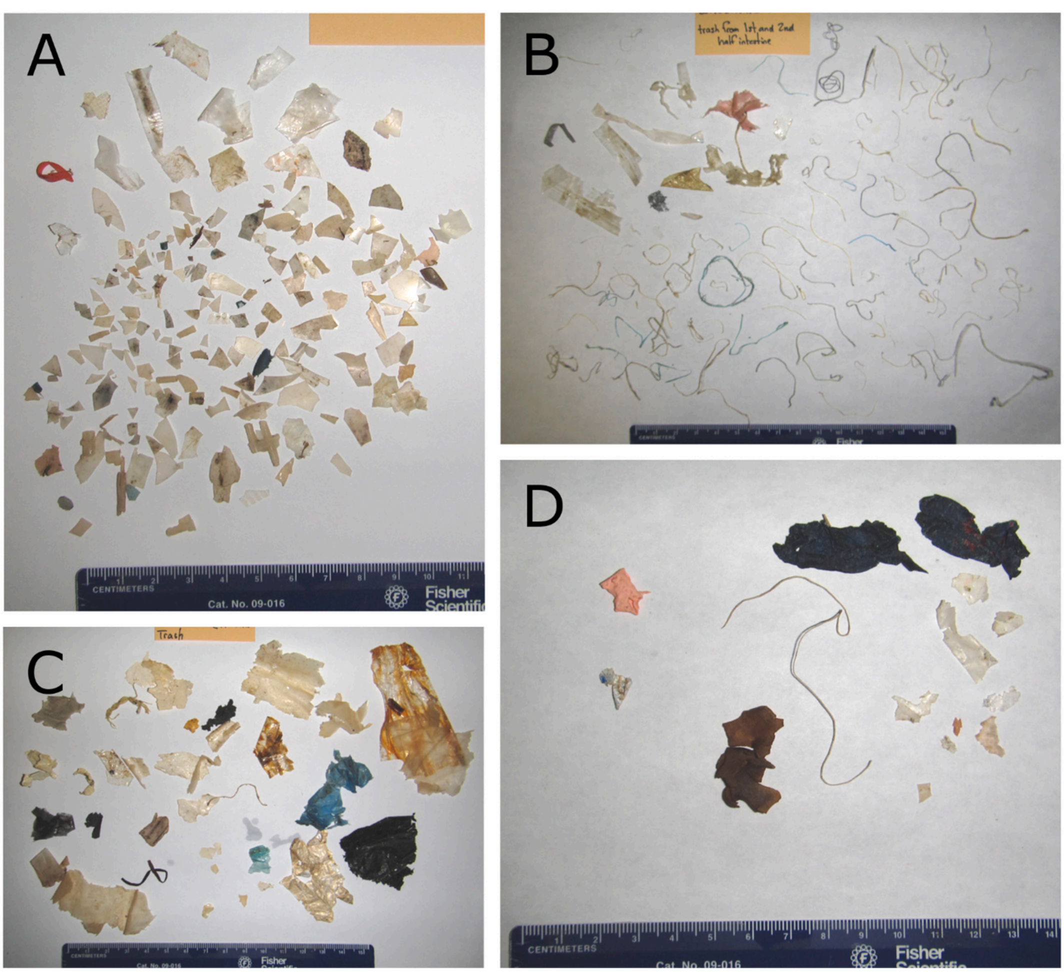

Fig. 3. Representative images of debris ingested by green turtles (*Chelonia mydas*) in Texas between 1987 and 2019. (A) 26.3 cm straight carapace length (SCL) turtle with primarily fragments. (B) 36.1 cm turtle with primarily threads (e.g., fishing line). (C) 27.4 cm SCL turtle with primarily sheets. (D) 27.3 cm SCL turtle with close to the mean number (12.8 items) and mass (0.49 g) of debris items found in all turtles, excluding non-detects. Included for scale is a 15 cm ruler.

Table 3
Section of the gastrointestinal tract (GIT), color, and category of debris items ingested by green turtles (*Chelonia mydas*) found stranded in Texas between 1987 and 2019. Per descriptor, metrics include number, mass (g), and surface area (cm^2) of debris items, and total value and percent of the total. Mean, standard deviation (SD), and frequency of categories (%) are presented for turtles which ingested debris. n is the number of turtles represented by each category. Eso/Stom: esophagus/stomach, Non: non-plastic debris items.

	Number			Mass (g)			Surface Area (cm^2)			n	%FO
	Total	%	Mean ± SD	Total	%	Mean ± SD	Total	%	Mean ± SD		
GIT section											
Intestines	2425	84.14%	10.73 ± 13.24	87.53	86.38%	0.39 ± 1.05	14,214.96	81.47%	62.90 ± 266.32	194	85.84%
Unknown	253	8.78%	1.12 ± 2.92	8.32	8.21%	0.04 ± 0.14	1200.16	6.88%	5.31 ± 22.44	27	11.95%
Eso/Stom	204	7.08%	0.90 ± 3.08	5.48	5.41%	0.02 ± 0.22	2033.83	11.66%	9.00 ± 103.72	54	23.89%
Color											
Clear	941	32.65%	4.16 ± 8.05	31.45	31.04%	0.14 ± 0.58	5774.05	33.09%	25.55 ± 91.46	136	60.18%
White	672	23.32%	2.97 ± 5.04	16.52	16.30%	0.07 ± 0.23	4354.24	24.95%	19.27 ± 136.75	148	65.49%
Brown	293	10.17%	1.30 ± 3.38	18.77	18.52%	0.08 ± 0.40	2565.17	14.70%	11.35 ± 72.50	84	37.17%
Yellow	236	8.19%	1.04 ± 2.92	5.68	5.61%	0.03 ± 0.09	1116.26	6.40%	4.94 ± 18.91	87	38.50%
Green	208	7.22%	0.92 ± 2.00	7.16	7.07%	0.03 ± 0.10	715.08	4.10%	3.16 ± 14.48	84	37.17%
Black	174	6.04%	0.77 ± 1.62	6.17	6.09%	0.03 ± 0.08	1587.73	9.10%	7.03 ± 37.04	74	32.74%
Blue	121	4.20%	0.54 ± 1.33	3.53	3.48%	0.02 ± 0.10	438.62	2.51%	1.94 ± 9.85	59	26.11%
Red	102	3.54%	0.45 ± 1.13	7.47	7.37%	0.03 ± 0.16	333.32	1.91%	1.47 ± 5.56	57	25.22%
Pink/purple	56	1.94%	0.25 ± 0.75	1.93	1.90%	0.01 ± 0.06	213.37	1.22%	0.94 ± 4.25	33	14.60%
Gray	54	1.87%	0.24 ± 0.64	1.74	1.72%	0.01 ± 0.03	247.46	1.42%	1.09 ± 6.68	38	16.81%
Orange	25	0.87%	0.11 ± 0.41	0.91	0.90%	0.00 ± 0.02	103.63	0.59%	0.46 ± 3.33	18	7.96%
Category											
Sheet	1089	37.79%	4.82 ± 7.73	28.09	26.12%	0.12 ± 0.59	13,092.96	75.04%	57.93 ± 352.19	175	77.43%
Fragment	782	27.13%	3.46 ± 9.91	43.25	40.21%	0.19 ± 0.76	1528.43	8.76%	6.76 ± 25.29	89	39.38%
Thread	659	22.87%	2.92 ± 8.10	5.37	4.99%	0.02 ± 0.07	937.69	5.37%	4.15 ± 10.76	139	61.50%
Other	245	8.50%	1.08 ± 2.60	27.60	25.66%	0.12 ± 0.42	1505.42	8.63%	6.66 ± 29.98	90	39.82%
Foam	63	2.19%	0.28 ± 1.17	1.64	1.52%	0.01 ± 0.04	168.81	0.97%	0.75 ± 3.27	23	10.18%
Non	28	0.97%	0.12 ± 0.44	1.15	1.06%	0.01 ± 0.03	202.31	1.16%	0.90 ± 4.85	20	8.85%
Industrial	16	0.56%	0.07 ± 0.44	0.46	0.43%	0.00 ± 0.01	13.35	0.08%	0.06 ± 0.37	9	3.98%
Total	2882	100.00%	12.75 ± 17.99	107.94	100.00%	0.48 ± 1.18	17,448.95	100.00%	77.21 ± 365.88	464	100.00%

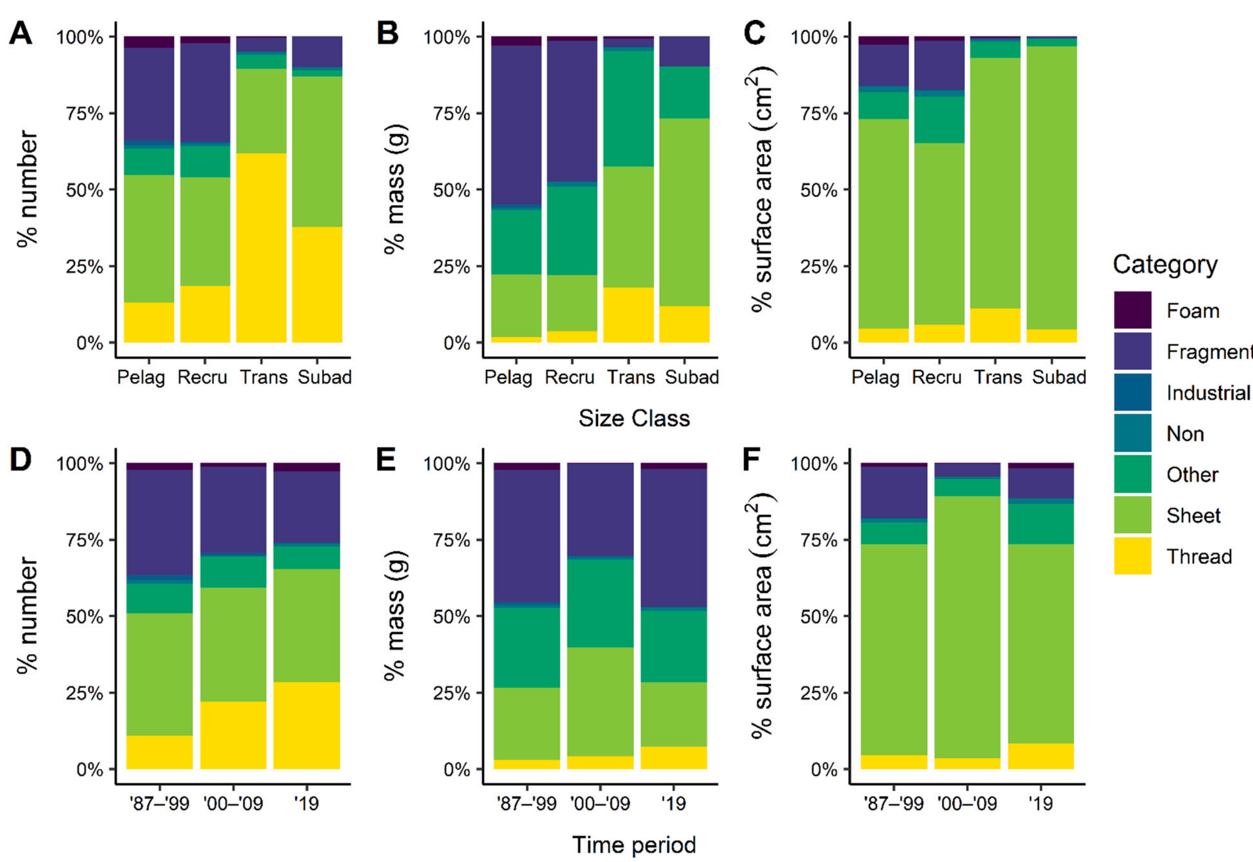

Fig. 4. By size class and time period, percent of total number (A, D), mass (g; B, E), and surface area of items (cm^2; C, F) for each category of debris items ingested by green turtles (*Chelonia mydas*) found stranded in Texas between 1987 and 2019. Pelag: pelagic stage, Recru: recruitment stage, Trans: transitional stage, Subad: subadult stage.

($\chi^2_3 = 3.31$, $p = 0.347$), mass ($\chi^2_3 = 1.24$, $p = 0.745$), or surface area of items ($\chi^2_3 = 3.40$, $p = 0.334$).

Relative abundance of each category of ingested items varied according to size class (Fig. 4). Relative number of fragments was three times higher for pelagics (30.3%) and recruits (32.3%) than transitionals (4.6%) and subadults (10.2%), while number of threads was two to four times lower for pelagics (13.0%) and recruits (18.4%) than transitionals (61.8%) and subadults (37.8%); other categories were more similar across size classes. Relative masses of sheets and threads were similarly greater in transitionals (sheets: 39.6%; threads: 17.9%) and subadults (sheets: 61.2%; threads: 11.9%) than in pelagics (sheets: 20.5%; threads: 1.7%) and recruits (sheets: 18.3%; threads: 3.7%), while relative mass of fragments was about five times greater in pelagics (51.8%) and recruits (46.0%) than transitionals (2.7%) and subadults (2.7%). Relative surface area of fragments was also highest in smaller turtles, while surface area of sheets dominated for all size classes (Fig. 4).

For values of body burden by mass less than 0.02 g/cm, conditional %FO for pelagics was 10–15 percentage points higher than recruits and 20–30 percentage points higher than transitionals and subadults; few turtles of any size class ingested more than 0.04 g/cm ($n = 22$) (Fig. 5). Similarly, for values of body burden by number less than 1 item/cm, conditional %FO for pelagics was 15–20 percentage points higher than recruits and 20–40 percentage points higher than transitionals or subadults; few turtles of any size class ingested more than 2 items/cm ($n = 8$) (Fig. 5).

3.3. Temporal trends

Percent frequency of occurrence was 32.5% (CI: 25.8–39.9%) between 1987–1999 ($n = 169$), 45.5% (CI: 36.5–54.8%) between 2000–2009 ($n = 112$), and 65.6% (CI: 58.5–72.2%) in 2019 ($n = 183$) (Table 2). Mean number of ingested items differed between time periods ($\chi^2_2 = 28.18$, $p < 0.001$) with significant pairwise differences between 1987–1999 (4.0 ± 10.6) and 2019 (8.5 ± 17.1, $p < 0.001$) and between 2000–2009 (5.8 ± 12.5) and 2019 ($p < 0.01$). Mean mass differed between time periods ($\chi^2_2 = 20.65$, $p < 0.001$) with significant pairwise differences between 1987–1999 (0.20 ± 0.79) and 2019 (0.23 ± 0.73, $p < 0.001$) and between 2000–2009 (0.27 ± 1.11) and 2019 ($p < 0.05$). Mean surface area differed between time periods ($\chi^2_2 = 27.39$, $p < 0.001$) with significant ($p < 0.01$) pairwise difference between all periods (21.52 ± 78.77 cm^2 in 1987–1999, 68.71 ± 506.89 cm^2 in 2000–2009, and 33.43 ± 72.56 cm^2 in 2019).

Excluding turtles which did not ingest debris, mean number of items was 12.8 ± 18.0 and did not differ significantly between periods ($\chi^2_2 = 2.00$, $p = 0.369$). Mean mass differed between periods ($\chi^2_2 = 12.99$, $p < 0.01$) with significant pairwise differences between 1987–1999 (0.63 ±

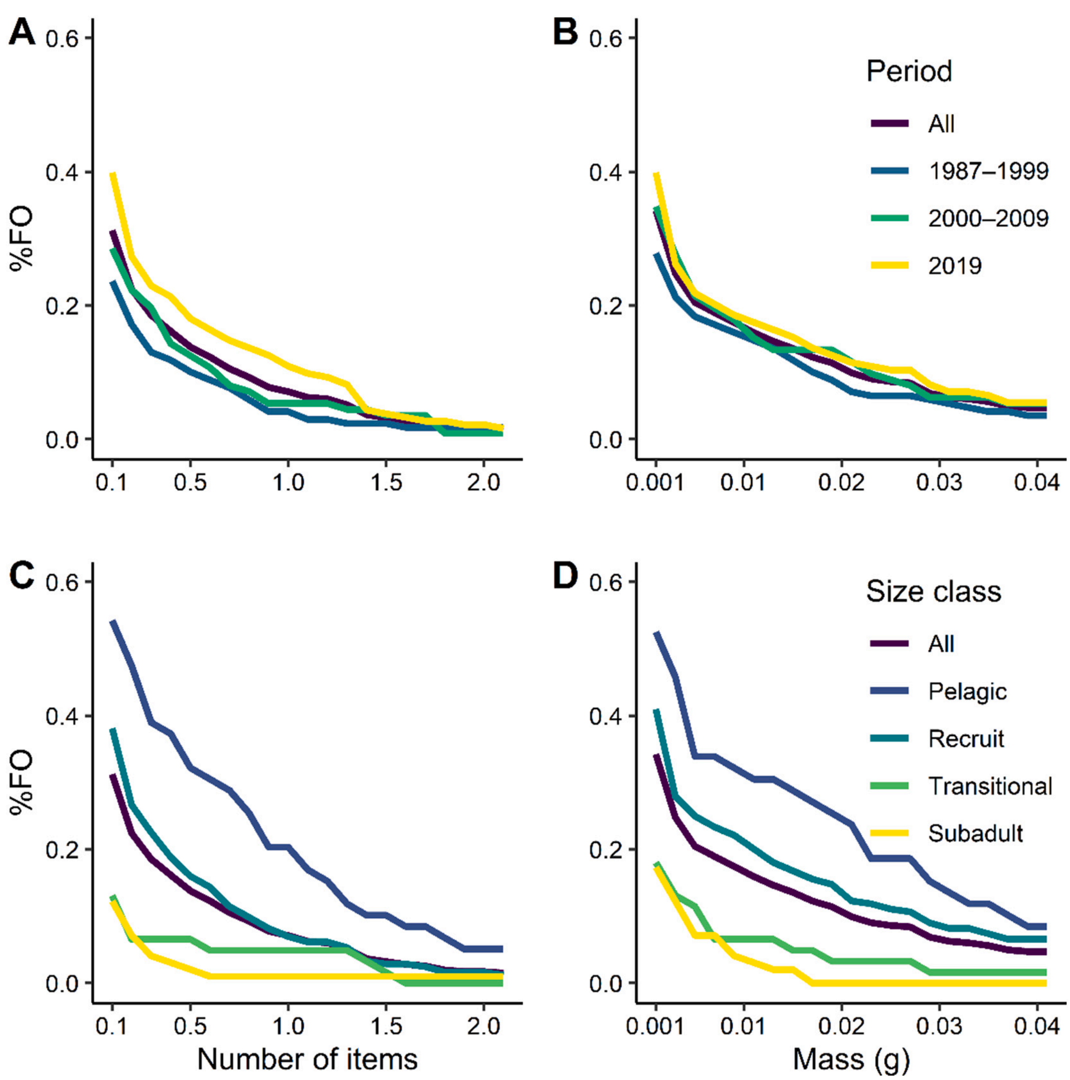

Fig. 5. Conditional frequency of occurrence (%FO) of debris ingestion as a function of body size by green turtles (*Chelonia mydas*) found stranded in Texas between 1987 and 2019. Conditional %FO is the percent of turtles which ingested at least a threshold (x-axis) amount of debris per cm straight carapace length. Values are displayed for number (A, C) and mass (g; B, D) of debris items and are presented by time period and size class.

1.28 g) and 2019 (0.36 ± 0.88 g, $p < 0.01$) and between 2000–2009 (0.60 ± 1.58) and 2019 ($p < 0.05$). Mean surface area was 77.21 ± 365.88 cm^2 overall and did not differ significantly between periods ($\chi^2_2 = 3.80$, $p = 0.149$).

Relative abundance of each category of ingested items varied little over time for most categories (Fig. 4). Relative number of threads increased from 1987–1999 (10.9%) to 2019 (28.5%), but fragments decreased (1987–1999 [34.6%]; 2019 [23.4%]); other categories remained similar.

Relative mass and surface area of sheets were 12–17 percentage points higher in 2000–2009 (mass: 35.7%; surface area: 85.6%) than in other periods, while relative mass and surface area of fragments were 6–13 percentage points lower in 2000–2009 (mass: 30.1%; surface area: 4.1%) than in other periods. Relative mass and surface area of other categories changed little throughout the study period (Fig. 4).

Conditional %FO of body burden by mass (g/cm) did not change over time: the maximum difference between time periods was 5%, occurring at a body burden of 0.02 g/cm or more (8.3% of turtles in 1987–1999, 13.4% in 2000–2009, and 12.0% in 2019) (Fig. 5). By number, conditional %FO of body burden was 5–10 percentage points higher in 2019 than in earlier periods (e.g., in 1987–1999, 10.1% of turtles had a body burden of at least 0.5 items/cm, 12.5% in 2000–2009, and 18.0% in 2019), though values were similar for body burdens of 1.4 items/cm or higher (Fig. 5).

Across both time period and size class, %FO was highest for pelagics from 2019 (96.8% [CI: 85.9–99.6%], $n = 31$) and lowest for subadults from 1987–1999 (10.0% [CI: 2.1–28.4%], $n = 20$) (Table 2).

3.4. *Predictors of plastic ingestion*

One ZAG and three ZANB models were considered competitive (<2 ΔAIC) (Table 4). Each model included all five parameters in the Bernoulli part, and marginal effects indicated rate of plastic ingestion was 2.4 times higher in pelagics (71% [CI: 56–83%]) than in subadults (29% [CI: 17–43%]); 1.4 times higher in turtles found offshore (54% [CI: 47–61%]) than inshore (38% [29–47%]); 1.7 times higher near the northern end of the study area (28° N latitude, 59% [50–67%]) than in the southern end (26°N latitude, 35% [27–45%]); 1.7 times higher in 2019 (59% [CI: 50–68%]) than in 1987–1999 (35% [CI: 27–44%]); and 1.9 times higher during spring (66% [CI: 57–75%]) than in fall (34% [CI: 23–47%]) (Table 5, Fig. 6). All parameter estimates for the negative binomial part of each ZANB hurdle model were not significant.

Marginal effects for the top ZAG model (Table 4) indicated that mean mass of items ingested was 2.5 times greater in subadults (0.94 g [CI: 0.35–2.54 g]) than other size classes; 2.9 times greater in turtles found offshore (0.44 g [CI: 0.33–0.59 g]) than inshore (0.15 g [CI: 0.08–40.26 g]); 2.6 times greater in more southern areas of the study area (26° N latitude, 0.62 g [CI: 0.37–1.06 g]) than the northern end (28° N latitude, 0.24 g [CI: 0.16–0.35 g]); 3.7 times greater in 1987–1999 (0.81 g [CI: 0.47–1.40 g]) than in 2019 (0.22 g [CI: 0.15–0.31 g]); and 2.4 times greater during spring (0.56 g [CI: 0.38–0.81 g]) and fall (0.50 g [CI: 0.22–1.11 g]) than during summer (0.19 g [CI: 0.12–0.30 g]) and winter (0.21 g [CI: 0.10–0.43 g]) (Table 5, Fig. 6).

Table 4
Top (ΔAIC < 2) zero-altered negative binomial (ZANB) and zero-altered gamma (ZAG) models of number of ingested debris items by green turtles (*Chelonia mydas*) found stranded in Texas between 1987 and 2019. Included are degrees of freedom (df), ΔAIC, and model weight (wt). InOff: inshore/offshore.

Bernoulli	Negative binomial (ZANB)	df	ΔAIC	wt
InOff + latitude + period + size class + season	InOff + season	17	0.00	0.28
InOff + latitude + period + size class + season	Season	16	1.24	0.15
InOff + latitude + period + size class + season	InOff + latitude + season	18	1.99	0.10

Bernoulli	Gamma (ZAG)	df	ΔAIC	wt
InOff + latitude + period + size class + season	InOff + latitude + period + size class + season	23	0.00	0.79

Table 5
Parameter estimates (±one standard error) for top (ΔAIC < 2) zero-altered negative binomial (ZANB) and zero-altered gamma (ZAG) models of number and mass of ingested debris items by green turtles (*Chelonia mydas*) found stranded in Texas between 1987 and 2019. Significance codes: *$p \leq 0.05$; **$p \leq 0.01$; ***$p \leq 0.001$. InOff: inshore/offshore. Reference categories are [1]1987–1999, [2]fall, [3]inshore, and [4]pelagic-stage.

Parameter	Bernoulli	Negative binomial			Gamma
	All models	ΔAIC = 0.00	ΔAIC = 1.24	ΔAIC = 1.99	ΔAIC = 0.00
(Intercept)	−13.89 ± 4.21***	1.66 ± 0.43***	2.05 ± 0.39***	2.02 ± 4.22	12.10 ± 5.21*
Period (2000–2009)[1]	0.58 ± 0.30				−0.69 ± 0.39
Period (2019)[1]	1.02 ± 0.28***				−1.32 ± 0.34***
Season (spring)[2]	1.33 ± 0.34***	0.35 ± 0.38	0.33 ± 0.39	0.34 ± 0.38	0.12 ± 0.46
Season (summer)[2]	0.71 ± 0.36*	−0.57 ± 0.39	−0.63 ± 0.40	−0.58 ± 0.40	−0.97 ± 0.49*
Season (winter)[2]	0.01 ± 0.36	−0.26 ± 0.44	−0.28 ± 0.45	−0.27 ± 0.45	−0.87 ± 0.54
InOff (offshore)[3]	0.67 ± 0.25**	0.49 ± 0.26		0.49 ± 0.26	1.10 ± 0.33***
Latitude	0.49 ± 0.15**			−0.01 ± 0.15	−0.48 ± 0.19*
Size class (recruit)[4]	−0.83 ± 0.38*				0.32 ± 0.33
Size class (transitional)[4]	−1.52 ± 0.44***				−0.35 ± 0.50
Size class (subadult)[4]	−1.83 ± 0.49***				1.26 ± 0.59*

4. Discussion

4.1. *Overview*

This study presents one of the largest examinations ($n = 464$) of plastic ingestion by marine turtles worldwide (Table S3). Globally, two studies have been larger, but these used primarily fecal samples without full necropsy or focused more on pathological effects of plastic ingestion than on plastic ingestion itself (Casale et al., 2016; Jerdy et al., 2017). In this study, 48.7% (CI: 44.2–53.2%) of green turtles found stranded or incidentally captured in Texas ingested plastic with a mean number of six items and a mean mass of 0.23 g per turtle. For turtles that ingested at least one item, mean number and mass of items were 13 items and 0.48 g, respectively. Santos et al. (2015) predicted that as little as 0.50 g could cause mortality in juvenile green turtles, and Wilcox et al. (2018) predicted that ingesting 14 items could result in a 50% chance of mortality. However, ingestion-related mortality was not detected in this study, suggesting higher thresholds in Texas. Additionally, it is difficult to interpret the prediction by Wilcox et al. (2018) as size of ingested plastic items can vary drastically regionally and ingesting a few large items could be much more harmful than many small items.

Studies worldwide have reported amounts of ingested debris far greater than we found in Texas (10–40 times higher, on average) but with infrequent death attributable to plastic ingestion (Lynch, 2018, Table S3). In Brazil, a juvenile (39 cm CCL) green turtle was found that had ingested 3593 items with a total mass of 269.6 g; the turtle died shortly after rescue (Stahelin et al., 2012). This is far greater than the highest mass observed in an individual turtle in the present study (11.1 g) and more than twice the total mass of debris found across all turtles. This suggests a low overall severity of plastic ingestion by green turtles

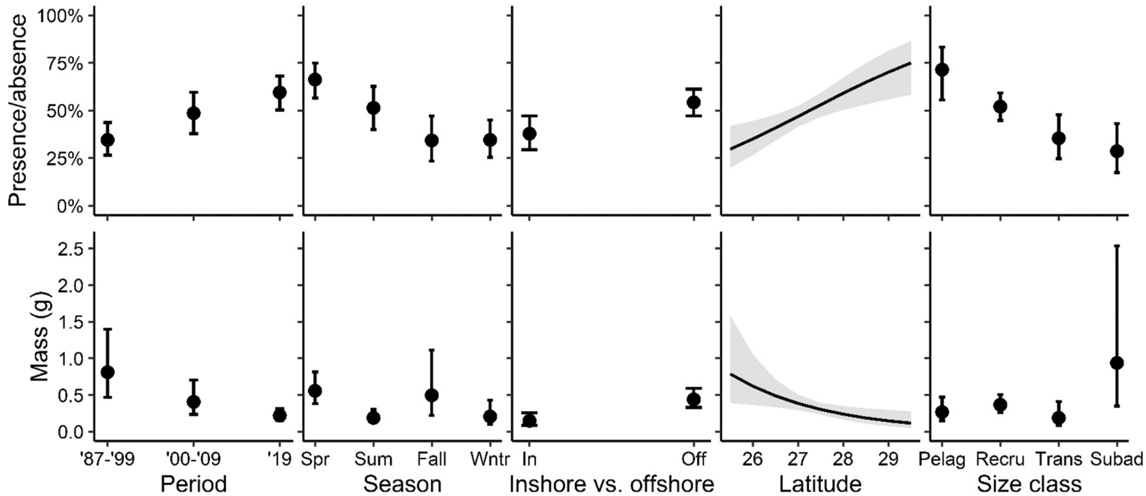

Fig. 6. Marginal effects plots for parameters in the Bernoulli (i.e., presence or absence) and gamma (i.e., mass) parts of the top zero-altered gamma (ZAG) model of debris ingestion by green turtles (*Chelonia mydas*) found stranded in Texas between 1987 and 2019. Shaded regions and error bars represent 95% confidence intervals. Pelag: pelagic stage, Recru: recruitment stage, Trans: transitional stage, Subad: subadult stage.

in Texas, compared to other regions. Indirect effects, such as dietary dilution or chemical absorption, may be of greater conservation concern. For example, emaciated turtles in Brazil were found to ingest much more debris than turtles of normal weight, suggesting that dietary dilution could impact foraging decisions (Santos et al., 2020). Further, ingested debris has been found to effect reproductive output: for loggerhead turtles with GITs containing 3% plastic, predicted seasonal reproductive output declined by 10% (Marn et al., 2020). However, it is currently unclear how indirect effects impact Texas green turtles.

4.2. Comparison to other regions

Percent frequency of occurrence for this study (48.7% [CI: 44.2–53.2%]) was lower than reported elsewhere: half of previous studies on green turtles have reported a %FO of over 70%, including several at 100% (Table S3). Less than 30% of past studies reported %FO values lower than we report here (Table S3). However, %FO can vary substantially depending on differences between study area, turtle collection method, turtle size, and local foraging ecology, so comparing %FO between studies is less meaningful than comparing quantity ingested (Lynch, 2018). This is especially true when small sample sizes create large confidence intervals for %FO, rendering it "essentially meaningless" (Casale et al., 2016). Similarly to %FO, mean number and mass of items ingested per turtle in this study (13 and 0.48 g, respectively) were substantially lower than have been reported for green turtles worldwide (Lynch, 2018). In nearly every study reporting it, mean number of items has been higher than 13 items/turtle, most reporting values greater than 45 items/turtle (Table S3). Most studies have reported a mean mass greater than 7 g/turtle, including three studies reporting 19–40 g/turtle (regions: Uruguay; Hawai'i, USA; and Japan; Table S3). Though likely connected, these differences are not necessarily caused by availability of plastic in the environment. Indeed, a global review found no correlation between ingestion and modeled distributions of oceanic debris (Schuyler et al., 2014). Like %FO, variation in the amount of plastic ingested is likely related to a wide range of variables (e.g., migration, foraging habitats, life-stage, and dietary selectivity), increasing the difficulty of global comparison (this can be remedied in part by reporting ingestion metrics according to specific subsamples of turtles, such as we present for size class) (Duncan et al., 2019; Schuyler et al., 2014; Schuyler et al., 2016). This may explain why we measured low plastic ingestion despite research suggesting moderate to high amounts of microplastics and macroplastics in the GoM (Di Mauro et al., 2017; Eriksen et al., 2014). Regardless, we report substantially lower metrics compared to many other studies, suggesting a true difference between the GoM and other regions.

4.3. Comparison to other species

Green turtles appear to ingest more plastic than most other species within a region, though the few existing studies on hawksbill turtles (*Eretmochelys imbricata*) indicate they may ingest more than green turtles (Lynch, 2018). Global reviews differ in ranking ingestion between different species, but green turtles consistently appear to ingest large amounts compared to other marine turtle species (Balazs, 1984; Lynch, 2018; Schuyler et al., 2014; Schuyler et al., 2016). Scaled to body mass, juvenile green turtles in Hawai'i, USA, ingested more than twice as many grams of debris as loggerheads or olive ridleys (*Lepidochelys olivacea*) (Clukey et al., 2017). In Japan, mass of debris ingested by subadult green turtles was over three times higher than that ingested by loggerheads (Fukuoka et al., 2016). If these global trends are also true in Texas, the low level of plastic ingestion reported here for green turtles also suggests low plastic ingestion by other species in the region. Though this is hopeful, species differences remain unclear and plastic ingestion rates for green turtles should not be applied to other marine turtle species in the GoM. Debris ingestion has been reported for all five species in the GoM, but studies have had small samples sizes or lacked detailed comparison (i.e., beyond %FO) (see supplementary material in Lynch, 2018 and Mrosovsky et al., 2009). Large scale and thorough study of other species, such as critically endangered Kemp's ridley turtles, is essential to assessing the ongoing conservation threat of anthropogenic marine debris on marine turtles in the GoM.

4.4. Size class trends

There was a stark relationship between turtle size and %FO, number of items, and mass of items, with small turtles ingesting plastic more often and in greater amounts than larger turtles. Indeed, an alarming 30 of 31 (96.8% [CI: 85.9–99.6%]) pelagics from 2019 ingested plastic, and across all years, pelagic-stage turtles clearly ingested plastic most frequently (76.2%, about three times that of subadults). Similar findings have been frequently suggested by the literature (Domènech et al., 2019; Lynch, 2018; Nelms et al., 2016; Schuyler et al., 2014, 2016; Yaghmour et al., 2021). Across most marine turtle species, juveniles and post-hatchlings spend their early years foraging in nutrient-rich oceanic convergence zones which also concentrate floating plastic (Barstow, 1983; Carr, 1987; Lebreton et al., 2012; Nelms et al., 2016; Witherington et al., 2012). In the GoM, pelagic green turtles inhabiting these zones forage among large floating mats of *Sargassum*, and GoM diet studies confirm greater debris ingestion in turtles primarily feeding on *Sargassum* (Howell et al., 2016; Witherington et al., 2012). Further, pelagic-stage turtles feed more generally than larger turtles, ingesting diverse food items with little selectivity (Nelms et al., 2016). Many of the plastic items we examined were covered in algal growth and closely resembled prey items (e.g., *Sargassum* and *Sargassum*-related invertebrates), thereby additionally creating a false environmental cue for food availability. Thus, both foraging location and behavior make pelagic-stage turtles especially predisposed to plastic ingestion. This phenomenon has been suggested to represent an evolutionary trap, though additional study is necessary to determine its severity and ecological consequences (Duncan et al., 2021; Santos et al., 2021).

After their pelagic phase, green turtles recruit to nearshore environments in Texas, where they transition again from jetty and channel habitats to lagoons and bays (Howell et al., 2016; Shaver, 1994; Ward, 2017). We found a corresponding shift in the type of ingested plastic: fragments were dominant in pelagics and recruits, but threads and sheets were dominant in transitionals and subadults. This relationship was also reported by Schuyler et al. (2012) and may have important biological effects. For example, hard fragments take up little space compared to a similar mass of sheets but may have sharp edges that could more easily damage the GIT. Conversely, sheets are typically very soft and flexible, but can have a very large surface area and may be more likely to cause GIT impaction. Size and type of plastic can vary geographically, so models linking mortality and plastic ingestion should incorporate plastic type. Simply suggesting a cutoff based on number or mass of items (e.g., Santos et al., 2015; Wilcox et al., 2018) might overlook important nuances in how different types of items interact with the GIT and may lead to either underestimating or overestimating mortality risk. In this study, ingestion rates of all plastic types were low, but if plastic ingestion in Texas continues to increase, size-based variation in the type of ingested plastic and the physical effects of ingestion may have major conservation implications.

4.5. Temporal trends

Percent frequency of occurrence doubled between 1987–1999 and 2019. In contrast, mass of ingested items (excluding turtles which did not ingest debris) decreased significantly from 0.63 g in 1987–1999 to 0.36 g in 2019. When mass was scaled to body size, conditional %FO was identical for all time periods (Fig. 5). For loggerheads in the Mediterranean, there was a similar increase in %FO but decrease in mass between 1995–2004 and 2005–2016 (Domènech et al., 2019). In that study, it was not clear if the difference in mean mass reflected changing environmental debris availability, turtle cause of death (bycatch from pelagic longline vs. bottom trawling), or turtle size (Domènech et al., 2019). For green turtles in Brazil, there was no clear pattern in %FO or mass between 1997 and 2017, but this could be attributed to small sample sizes and lack of standardization, which made it difficult to detect differences between years (Rizzi et al., 2019). The increasing %FO we observed matches global trends for green turtles (Schuyler et al., 2014). Over recent decades, juvenile green turtle abundance has increased greatly along the Texas coast (Shaver et al., 2017), so the higher %FO observed in 2019 may partially reflect the higher %FO typical of smaller turtles, and not solely an increase in ingestion frequency itself. Additionally, turtle collection (e.g., spatial coverage and effort) was inconsistent throughout the study period and %FO between time periods was likely also influenced by factors including beach visitor use, cold stunning events, weather, and season. However, %FO increased over time within some size classes, even though mean turtle size decreased (Table 2). Although changes in %FO are influenced by many factors, our findings support a true increase in the frequency of plastic ingestion among green turtles in Texas.

4.6. Characteristics of ingested plastic items

Ingested debris items found in green turtles stranded in Texas were primarily plastic (99.0%), as has been reported worldwide (Schuyler et al., 2014). Sheets, fragments, and threads were dominant, which also follows global trends and likely reflects the high availability of these items in the environment (Schuyler et al., 2012, 2014). Indeed, plastics (including rubber) constituted 96.5% of debris items found during beach surveys in the GoM (Wessel et al., 2019). Industrial plastic pellets, however, are rarely found in ingestion studies, even in regions with very high ingestion rates (Casale et al., 2016; Domènech et al., 2019; Matiddi et al., 2017). However, in 2018 high numbers of industrial pellets appeared along the Texas coast, attributed to a transportation spill (Tunnell et al., 2020). Sixteen industrial pellets were found in this study but only two were found in 2019, indicating the 2018 spill likely did not adversely impact green turtles in the study region.

By color, clear and white items were most common, as has been reported elsewhere (Schuyler et al., 2012; Tourinho et al., 2010). Loggerhead and green turtles have been directly observed actively selecting clear or white plastic sheets (e.g., shopping bags) in a similar manner to their pursuit of gelatinous prey (Fukuoka et al., 2016), lending support to the jellyfish hypothesis which has been frequently cited as a driving factor in marine turtle plastic ingestion (Hoarau et al., 2014; Nelms et al., 2016; Rizzi et al., 2019; Schuyler et al., 2012). However, this should be applied with caution to Texas turtles. First, clear and white items are much more prevalent in the marine environment overall, so high %FO should be expected (specific studies on the types of plastic in the GoM are not available) (Marti et al., 2020). Second, plastic sheets in this study were often small and fragmented, and in this form unlikely resembled gelatinous prey; though it is possible that some plastic sheets were fragmented during ingestion. Finally, juvenile turtles in Texas infrequently ingest gelatinous prey, suggesting frequency of clear and white items may be more related to environmental availability (Howell et al., 2016). More generally, discussion of the importance of color in marine turtle plastic ingestion should consider that items may appear differently when first ingested than when examined by the researcher. In this study, many plastic items of all categories, colors, and shapes were covered with marine algae or dirt, and closely resembled seagrasses, *Sargassum*, and *Sargassum*-related invertebrates, which are common food resources for juvenile green turtles (Duncan et al., 2019; Ward, 2017). Sometimes, color of items was not evident until they were cleaned, and it is possible that item color was further obscured before traveling through the GIT where algal growth may have been digested. Though color selectivity has indeed been detected in varying degrees (Duncan et al., 2019; Santos et al., 2016), the mechanisms for this selection should be further examined, especially for different species, size classes, and feeding environments.

Most plastic was found in the intestines (84.1%) and little was detected in the esophagus or stomach, similar to other studies (Camedda et al., 2014; Clukey et al., 2017; Jerdy et al., 2017; Yaghmour et al., 2018). Diet studies frequently sample only the esophagus and stomach, but this can lead to gross underestimation of both %FO and quantity of ingested plastic (Bjorndal et al., 1994). For example, if only the esophagus and stomach were considered in this study, %FO would drop from 49% to 12%. Fecal-only studies also underestimate plastic ingestion because plastic can remain in the GIT for long periods of time (Casale et al., 2016). Thus, studies should be wary of comparing results of different sampling techniques.

4.7. Season, latitude, and inshore vs. offshore

The leading ZAG model predicted that likelihood of ingestion is highest for turtles stranded during spring, which is when the amount of debris along the Texas coast is greatest (Duronslet et al., 1991; Wessel et al., 2019). In spring, onshore winds strengthen substantially and, combined with existing currents, may drive increased amounts of debris to Texas shorelines (Hardesty et al., 2017; Ribic et al., 2011; Wessel et al., 2019). During spring, large *Sargassum* mats may also wash ashore (Gower et al., 2006), followed by pelagic green turtles recruiting to nearshore environments (Donna Shaver, unpublished data). Ingested plastic can remain in the GIT for multiple months (Casale et al., 2016; Hoarau et al., 2014; Lutz, 1990), so high spring ingestion rates may represent a combination of 1) turtles that recently recruited from offshore *Sargassum* habitats where plastic is more prevalent, and 2) a higher amount of plastic in nearshore environments in the spring. Attributing high spring ingestion to recent recruitment is supported by the higher ingestion likelihood and predicted mass of offshore turtles, compared to inshore turtles. Along much of the Texas coast, debris primarily washes ashore from the ocean, while a much smaller proportion originates from freshwater influx as is more common in the northern GoM (Wessel et al., 2019). Juvenile green turtles in the GoM occupy different geographic areas seasonally (Witherington et al., 2012), so high springtime ingestion rates could also be related to spatial variation of plastic in the environment. There was also a positive relationship between latitude and ingestion likelihood in this study (though mass was inversely related to latitude), but surveys of barrier island beaches showed little latitudinal variation in the amount of debris along the Texas coast (Wessel et al., 2019). However, shoreline surveys do not necessarily represent availability of debris in ocean waters where many turtles forage (Ribic et al., 2011). Additionally, how far a turtle drifts between death and stranding is variable: modeled carcasses of Kemp's ridley turtles in the northern GoM were likely to drift for five days before beaching, and drift-time was related to water temperature and winds (Nero et al., 2013). Hence, observed latitudinal variation may be at least partly a function of prevailing currents and seasonality.

5. Conclusion

This study was among the largest completed to date and presented the first standardized and thorough investigation of plastic ingestion by marine turtles in the GoM. Partial data reporting has limited previous work, but this study provided important baseline information regarding amount, size, and category of plastic items. Our findings indicate plastic ingestion frequency has increased over time in Texas, even given the increasing population of juvenile green turtles, which are more likely to ingest plastic. By %FO, number, and mass, pelagic-stage turtles ingested more plastic than large turtles, suggesting feeding ecology is important in understanding plastic ingestion by green turtles. Though there was no evidence of death directly related to plastic ingestion, possibility of harmful indirect effects (e.g., chemical absorption), remains unclear. Future studies should investigate how plastic ingestion may indirectly impact the survival and success of the expanding juvenile green turtle population in Texas. Plastic selectivity between size classes was also evident, but an accurate understanding of selectivity will require further measurement of environmental plastic availability, especially in *Sargassum* habitats where pelagic-stage turtles forage. Future studies must adhere to standardized data collection and reporting (Duncan et al., 2019; Matiddi et al., 2019; Provencher et al., 2017). Specifically, reporting both mass and number of items is necessary in assessing the biological impact of plastic ingestion. In addition, it may be helpful to report conditional %FO by body burden, as this assisted in comparing the overall severity of plastic ingestion between time periods and size classes. Though it is encouraging that mass of ingested plastic by Texas green turtles is among the lowest worldwide, thorough study of plastic ingestion among other threatened species in the GoM is vital to responsible marine turtle conservation.

Funding statement

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

CRediT authorship contribution statement

Daniel Y. Choi: Methodology, Validation, Formal analysis, Investigation, Data curation, Visualization, Writing – original draft, Conceptualization, Writing – review & editing. **Christian Gredzens:** Methodology, Data curation, Project administration, Funding acquisition, Supervision, Conceptualization, Writing – review & editing. **Donna J. Shaver:** Resources, Project administration, Funding acquisition, Supervision, Conceptualization, 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.

Acknowledgements

Daniel Choi was supported during this project by the Scientists in Parks program, a collaborative program between the United States National Park Service and Ecological Society of America. Data collection would not have been possible without Texas Sea Turtle Stranding and Salvage Network participants, Michelle Knuepfer, Rosalin Wilkin, Cody Hoff, and others who assisted in the tedious process of individually measuring each piece of plastic. Heather Wilson patiently answered a continual barrage of questions and helped in thinking through many aspects of the methodology and findings. J Shelby Walker provided recommendations that enhanced this manuscript.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.marpolbul.2021.113111.

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