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7 From Sea to Source: The Abundance and Characteristics of Microplastics in Surface8 Water in the Transboundary Ganges River

9

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Abstract

Microplastics (plastic < 5 mm in size) are now known to contaminate riverine systems but understanding about how their concentrations vary spatially and temporally is limited. This information is critical to help identify key sources and pathways of microplastic and develop management interventions. This study provides the first investigation of microplastic abundance, characteristics and temporal variation along the Ganges river; one of the most important catchments of South Asia. From 10 sites along a 2,575 km stretch of the river, 20 water samples (3,600 litres in total) were filtered (60 samples each from pre- and post-monsoon season). Overall, 140 microplastic particles were identified, with higher concentrations found in the pre-monsoon (71.6%) than in post-monsoon (61.6%) samples. The majority of microplastics were fibres (91%) and the remaining were fragments (9%). We estimate that the Ganges, with the combined flows of the Brahmaputra and Meghna rivers (GBM), could release up to 1 - 3 billion (10^9) microplastics into the Bay of Bengal (north-eastern portion of the Indian Ocean) every day. This research provides the first step in understanding microplastic contamination in the Ganges and its contribution to the oceanic microplastic load.

Main finding: We estimate that up to 1 - 3 billion microplastics are released into the Bay of Bengal every day.

1.0 Introduction

The durability, versatility, low cost and wide-scale use of plastic items means that plastic litter is now prevalent worldwide, even in remote areas (Free *et al.*, 2014; Obbard *et al.*, 2014; Geyer *et al.*, 2017; Waller *et al.*, 2017). Accumulation of plastic waste was initially documented in oceanic environments, with the first observation of buoyant plastics in the ocean dating back to 1972 (Carpenter and Smith, 1972;

70 Carpenter *et al.*, 1972). Since then, numerous studies have increased our
71 understanding of oceanic plastic characterisation, transport and accumulation zones
72 (Moore *et al.*, 2001; Thompson *et al.*, 2004; Law *et al.*, 2010; Goldstein *et al.*, 2012;
73 Lebreton *et al.*, 2018). Within the last decade, there has been additional focus on
74 plastic presence in estuarine systems (Bakir *et al.*, 2014; Gallagher *et al.*, 2016),
75 freshwater (Klein *et al.*, 2015; Zhang *et al.*, 2015; Horton, Walton, *et al.*, 2017; Peng
76 *et al.*, 2018; Eo *et al.*, 2019; Mai *et al.*, 2019; Mani *et al.*, 2019; Watkins *et al.*, 2019;
77 Zhao *et al.*, 2019), and terrestrial environments (Rillig, 2012; Horton, Walton, *et al.*,
78 2017; Corradini *et al.*, 2019).

79

80 Plastic in the microplastic size range (0.1 μm –5 mm) is an environmental pollutant of
81 substantial public and scientific concern (Thompson *et al.*, 2004; Paul *et al.*, 2020).
82 Until recently, microplastics have been a largely overlooked part of plastic pollution
83 monitoring but are thought to be ubiquitous within the environment (Rochman, 2018;
84 Napper and Thompson, 2020). Within the last decade, microplastics have received
85 increasing research interest due to the rapid increase in plastic production, the
86 longevity of plastic and the disposable nature of plastic items (Thompson *et al.*,
87 2004); as such, data have been accumulating on the sources, distribution and impact
88 (Andrady, 2011; Ivar Do Sul and Costa, 2014; van Sebille *et al.*, 2015). Due to their
89 small size, microplastics are available for ingestion by a wide range of marine
90 species, from microscopic zooplankton to large vertebrate predators (Botterell *et al.*,
91 2019; Nelms *et al.*, 2019), and can cause negative impacts on biological processes
92 (Lo and Chan, 2018; Messinetti *et al.*, 2018)

93

94 An estimated 80% of plastic pollution in the ocean potentially originates from land-
95 based sources (Jambeck *et al.*, 2015), such as leakage from wastewater treatment
96 plants (WWTPs) (Kay *et al.*, 2018), urban centres and road runoff (Horton,
97 Svendsen, *et al.*, 2017), industry (Lechner and Ramler, 2015), atmospheric pollution
98 (Dris *et al.*, 2015; De Falco *et al.*, 2020) and degradation of larger items of plastic
99 waste (Barnes *et al.*, 2009; Schmidt *et al.*, 2017; Schwarz *et al.*, 2019). Freshwater
100 systems often connect inland and coastal communities to the ocean, and therefore
101 represent a substantial downstream transport pathway for microplastic input (Rech *et al.*
102 *et al.*, 2014; Miller *et al.*, 2017; Schmidt *et al.*, 2017; Seo and Park, 2020; Weideman *et al.*
103 *et al.*, 2020). An estimated 0.4 – 265,000 million tonnes of plastic are released into

104 coastal seas by the world's rivers annually (Wagner *et al.*, 2014; Lebreton *et al.*,
105 2017; Schmidt *et al.*, 2017; Mai *et al.*, 2020).

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110 Most freshwater microplastic studies originate from North America, Western Europe
111 (Horton, Walton, *et al.*, 2017; Peng *et al.*, 2018) and China (Peng *et al.*, 2018; Ding
112 *et al.*, 2019). The Yangtze River was predicted to emit 16–20 trillion microplastic
113 particles into the East China Sea in 2017 (Zhao *et al.*, 2019).

114

115 There are limited studies and empirical field data on plastic pollution from major
116 Asian rivers such as the Ganges and Mekong (Blettler *et al.*, 2018; Chowdhury *et al.*,
117 2020). However, recent research by Duncan *et al.*, (2020) tracked plastic PET
118 bottles through the Ganges river system and into the Bay of Bengal marine system
119 by using both GPS (Global Positioning System) cellular networks and satellite
120 technology and found a maximum distance tracked of 2845 km over a period of 94
121 days. Additionally, Nelms *et al.*, (2020) found that fishing gear is a significant source
122 of plastic pollution in the Ganges River system.

123

124 The Ganges is known as the Ganga in India and the Padma and Meghna in
125 Bangladesh (hereafter known as the Ganges). The combined flows of the Ganges,
126 Brahmaputra and Meghna rivers are the largest in South Asia and form the most
127 populous basin in the world, where over 655 million inhabitants rely on the water it
128 provides (The World Bank, 2014; Rahman *et al.*, 2020).

129

130 There is limited research on microplastic pollution in the Ganges river (Baroth, 2019)
131 and currently no published data are available on microplastic contamination in the
132 freshwater matrix of the Ganges. Additionally, there is limited understanding of how
133 microplastic concentrations vary along a whole river course and over different
134 seasons, particularly considering the major influences of the monsoon on the
135 Ganges (Clift, 2020). It was hypothesized that microplastic abundance would
136 increase as the river travels downstream and that lower microplastic concentrations
137 would be observed during the post-monsoon season due to the high levels of rainfall

138 resulting in increased volume and flow of freshwater; subsequently transporting more
139 microplastics into the marine environment. The aims of this study were to provide the
140 first investigation, over two different seasons, on the abundance, polymer type and
141 characteristics of microplastics in water along the mainstream Ganges river, from
142 sea to source.

143 2.0 Methods

144

145 2.1. Study area

146

147 The Ganges river originates from the Gangotri glacier in the Himalayas (India) at an
148 elevation of nearly 7,010 m and traverses a length of about 2,575 km before it
149 flows south-east, transforming into distributaries and ultimately flowing into the Bay
150 of Bengal (Bangladesh) (Whitehead, 2018; Singh and Singh, 2019). The Ganges
151 river is joined by a number of large and small tributaries. The river joins the
152 Brahmaputra river in Bangladesh as the Padma and further down the combined
153 discharge joins the Meghna river at Chandpur. The combined stream is called the
154 Meghna river, which 90 km further downstream discharges into the Bay of Bengal.
155 The total annual Ganga-Brahmaputra-Meghna (GBM) river basin inflow into
156 Bangladesh from India is 1,110 km³ (FAO, 2012). Over 138 700 m³/s of water flows
157 into the Bay of Bengal during flooding (particularly in the monsoon season) through a
158 single outlet of the GBM river in Bangladesh. This is the largest in the world for a
159 single outlet to the sea and exceeds even that of the Amazon discharge by about 1.5
160 times (Parua, 2001). The Ganges is a transboundary river basin distributed between
161 five countries; India, China, Nepal, Bhutan and Bangladesh. The basin and its
162 tributaries are diverse in social, economic and political terms as well as for water
163 availability and use.

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2.2 Site Selection

In this study, 10 sites in India and Bangladesh were selected along the length of the mainstream river to represent the whole length of the Ganges (Fig.1). In Bangladesh, three sites were included: Bhola (S1), Chandpur (S2), Rajbari (S3); the sample points were from the largest tributary of the Ganges or its confluence into the Meghna river. The other seven sampling sites were in India: Sahibganj (S4), Patna (S5), Varanasi (S6), Kannauj (S7), Anupshahar (S8), Rishikesh (S9) and Harsil (S10). The number signifies the site's position along the river, with S1 being closest to the ocean and S10 closest to the source; the position number will be used predominately in lieu of the sampling site name. The sites were selected on the basis of site characteristics (rural/urban/barrage/river confluence/industry/tourism/religious), primary land use, logistics and possibility/ease of sampling.

2.3 Sampling method

To collect the samples, 30 L water was pumped from 0.5 m below the river surface and immediately filtered through a 330 µm nylon mesh placed across a polypropylene tube, using a hand-operated bilge pump. The pumping volume was adjusted to 30 L considering the target microplastic size range (>300 µm) and logistical challenges/practical challenges during sampling. Each nylon mesh filter was immediately double wrapped in foil and then placed in separate clear polypropylene bags for transportation. The samples were transported to the University of Plymouth (U.K.) for laboratory analysis. Contamination control measures were applied throughout the sample collection and transport process (see Section 2.5).

206 The samples were collected during pre-monsoon (May 2019 – June 2019) and post-
207 monsoon (October 2019 – December 2019), to capture temporal variation in
208 microplastic abundance. At each sampling site, a 5 km stretch of river was selected
209 and samples were collected from three points at 2.5 km intervals (0, 2.5, and 5 km)
210 from the centre of the river. Samples were replicated on two consecutive days (n = 6
211 per site). Post-monsoon sampling occurred at the same sites using the GPS
212 locations from pre-monsoon. For tidal sections of the river (sites S1-S3), samples
213 were collected on an ebbing tide to ensure microplastics within the outflowing river
214 water were not those brought inshore from the Bay of Bengal.

215

216 2.4 Laboratory Analysis

217

218 Each mesh and subsequent foil packaging was examined for microplastics using a
219 light microscope (S9E - Leica) and information on the type of particle (i.e. fragment
220 or fibre), dimensions (length and diameter) and colour was recorded. Shape
221 classifications include fragments, films, spherical beads and fibres.

222

223 Suspected microplastics were analysed with Fourier transform infrared spectroscopy
224 (FT-IR) in transmission mode with a Hyperion 1000 microscope coupled to a Vertex
225 70 spectrometer (Bruker). Any spectra were recorded with 32 scans in the region of
226 4000– 600 cm^{-1} . The spectra obtained were compared against a spectral database
227 of synthetic polymers (BPAD polymer and synthetic fibres ATR).

228

229 2.5 Contamination Control

230

231 The water pumping system (bilge pump and polypropylene hose) and polypropylene
232 tube were checked for contamination before and after the expedition. A procedural
233 blank of 30 L of filtered (1.6 μm) distilled (DI) water was used to simulate the
234 sampling process and identify potential sources of contamination from the
235 equipment. No contamination was observed. Additionally, before the expedition each
236 mesh was inspected for contamination using a microscope (S9E - Leica), and any
237 particles removed before use. The mesh was wrapped in two layers of clean foil
238 before and after use to avoid subsequent contamination.

239

240 During field sampling, the pumping system was first flushed with water from the river.
241 The inside of the propylene tube was also rinsed with distilled water (filtered to 330
242 μm) prior to use. To control for airborne microplastic contamination, a damp (300 μm
243 filtered DI water) piece of filter paper (Whatman 47 mm diameter, 0.45 μm glassfiber
244 filter) in an open petri dish was placed nearby while samples were collected. The
245 petri dishes were kept open for the duration of the water pumping so that the blanks
246 and samples were exposed to the same levels of airborne contamination.

247

248 During any laboratory analysis, all steps were conducted in a dedicated clean room
249 for microplastic work, which had a positive pressure air system, limited access and
250 procedural blanks. Cotton laboratory coats and clothes were worn to reduce
251 contamination from synthetic textiles. All laboratory ware used was made of glass or
252 stainless steel and thoroughly rinsed with filtered (1.6 μm) Milli-Q water before use.

253

254 2.6 Statistical analysis

255

256 Zero-Inflated regression analysis was carried out using a Poisson distribution to
257 assess the spatial (site) differences in microplastic abundance, within the R Package
258 'pscl' (Jackman 2020). Microplastic concentrations (MP L^{-1}) were modelled as a
259 function of site for Poisson counts and binomial zeros (Jackman 2020). Further
260 analysis, by zero-inflated regression modelling, was used to assess the differences
261 between sites and polymer types by factor order manipulation. The most
262 parsimonious models were selected by sequential removal of terms and pairwise chi-
263 square comparison using the 'lmtest' package in R (Zeileis and Hothorn, 2002).
264 Thus, non-significant terms were deleted. Data were manipulated and visualized
265 using 'tidyverse' packages within the R computer programming language (R Core
266 Team 2019; Wickham et al. 2019).

267 Linear regression analysis was used to investigate the change in microplastic length
268 (μm) as a function of sampling date and site. To fit model assumptions, lengths were
269 transformed by natural logarithm for analyses. To plot the model predictions the
270 values were converted back to their original scale for plotting using the exponential
271 function.

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276 2.7 Predicted Microplastic Discharge into the Ocean

277

278 To estimate microplastic discharge from the Ganges, the average microplastic
279 concentrations at site 1 (Bhola) were used for both pre- and post-monsoon; 0.051
280 MP L⁻¹ and 0.026 MP L⁻¹, respectively. The rivers in the watershed carry an average
281 water discharge of 29,692 m³/s into the Bay of Bengal, and 6,041 m³/s during the
282 low water season (Khan and Islam, 2008); these flow data were used for post- and
283 pre-monsoon calculations, respectively. Following Miller *et al.*, (2017), flow rate (F_l)
284 was multiplied by the depth proportion and the recorded microplastic concentration
285 (MP_c) to calculate microplastic output at this site (MP_t). This is shown by equation i,
286 where F_l is the flow rate (L s⁻¹); D_i and D_t are the sampling depth and total depth of
287 the river (m) and MP_c is the sampled microplastic concentration (MP L⁻¹). As in Miller
288 *et al.*, (2017), flow rate was assumed to be constant, which is an oversimplification.

289

290 Equation i: $MP_t = \frac{D_i}{D_t} \times F_l \times MP_c$

291

292 3.0 Results

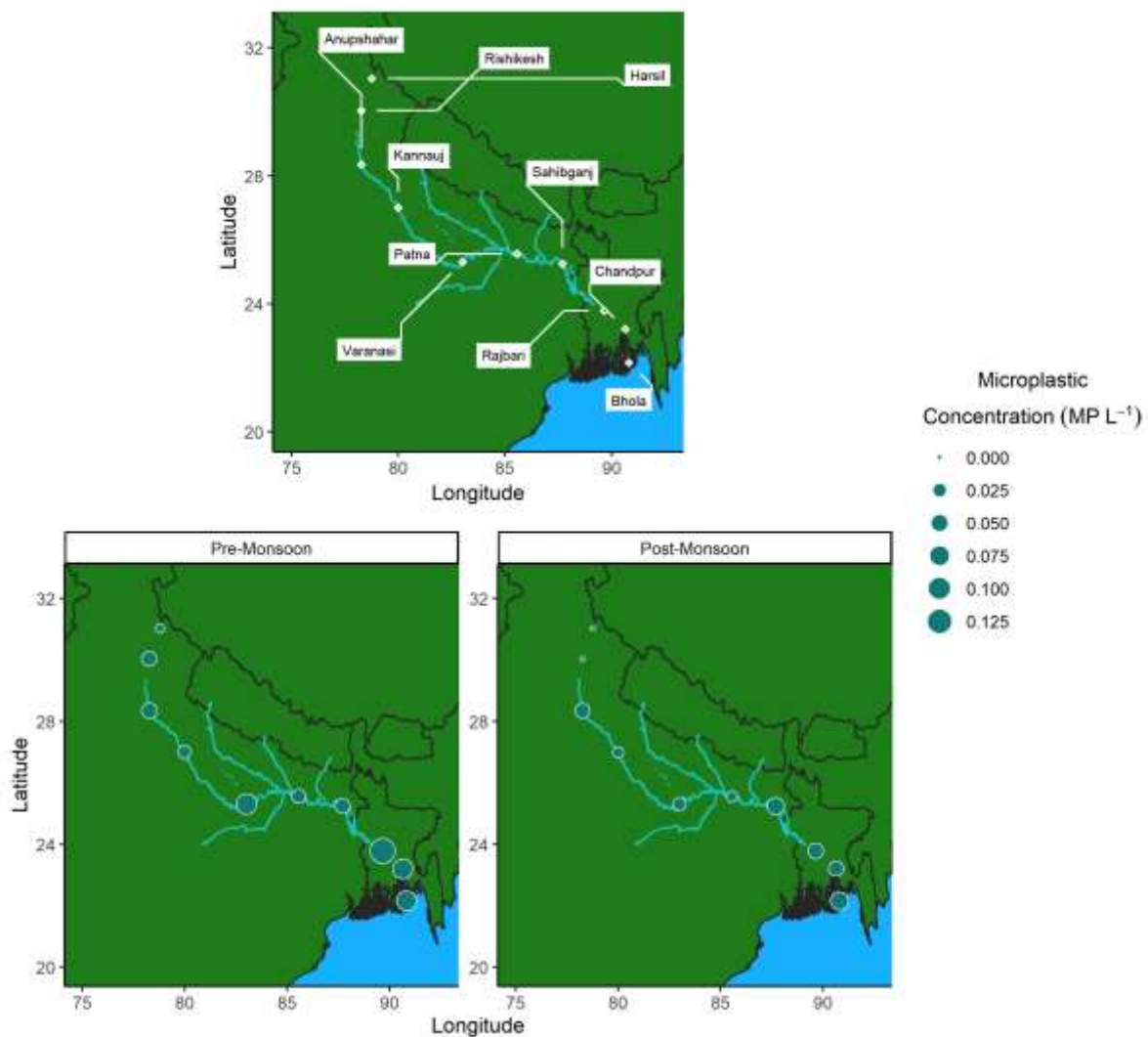
293

294 In total, 120 samples were collected from 10 sites (60 from both pre- and post-
295 monsoon) with a total 3,600 litres of freshwater filtered. Overall, 140 microplastic
296 particles were identified; these were found in 43 (71.6%) of the samples for pre-
297 monsoon, and 37 (61.6%) for post-monsoon. In total, the procedural controls (air
298 blanks) (n = 40) had an average of 0.005 ± 0.002 MP L⁻¹ day⁻¹ (mean ± S.E.). Any
299 contamination observed from a sampling day was subtracted from the microplastic
300 total for each subsequent sample. Microplastic contamination in the lab was minimal;
301 procedural blanks accumulated an average of 0.098 ± 0.042 MP filter⁻¹; only blue
302 microfibrils were found.

303

304 The average number of microplastics collected was 0.038 ± 0.004 MP L⁻¹. There
 305 were significant differences in microplastic concentration found among the sites ($p =$
 306 <0.001). The Bangladesh sites (S1, S2 and S3) had significantly more microplastics
 307 than Indian sites S5, S7, S9 and S10. There were also significantly greater
 308 concentrations of microplastics found at Indian sites S4, S6 and S8 compared to site
 309 S10. Additionally, site S6 had a greater concentration compared to site S7 and S9
 310 (Fig. 1) ($p = <0.05$ for all significant interactions). Combining predicted microplastic
 311 concentration at the mouth of the river (S1; Bhola) with the discharge of the river, we
 312 estimate that 1- 3 billion (10^9) microplastics are released from the Ganges into the
 313 Bay of Bengal every day.

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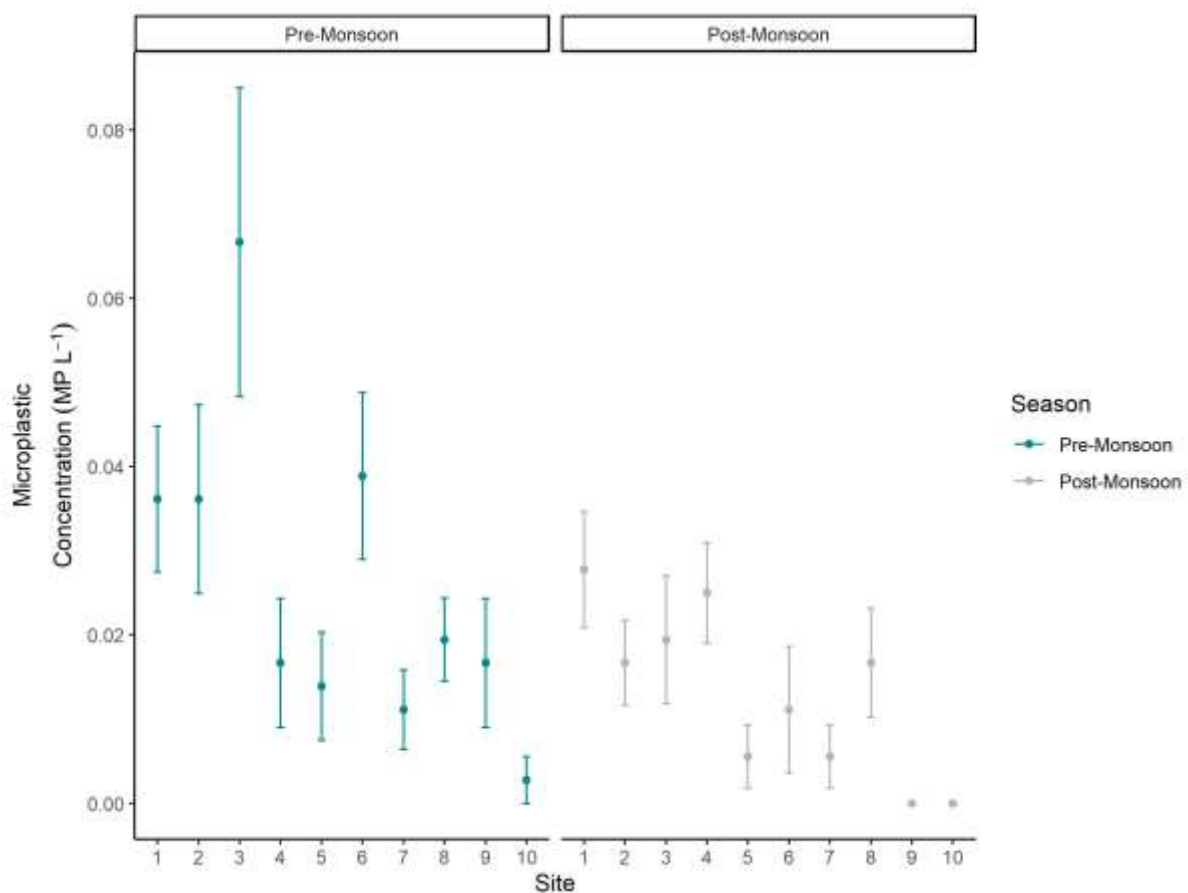
317 **Figure 1.** Sample Sites along the Ganges, point size is relative to microplastic
 318 concentration pre and post-monsoon: a – sample site; b – pre-monsoon microplastic
 319 concentration; c – post-monsoon microplastic concentration. Point size indicates
 320 relative concentration ($MP L^{-1}$).

321

322 There were a greater number of microplastics found on the pre-monsoon sampling
 323 occasions ($0.051 \pm 0.007 MP L^{-1}$) than post-monsoon ($0.026 \pm 0.004 MP L^{-1}$).

324 Furthermore, microplastic concentration at the river mouth (S1; Bhola) had
 325 quadrupled compared to source concentrations (S10; Harsil) in pre-monsoon and
 326 doubled in post-monsoon seasons.

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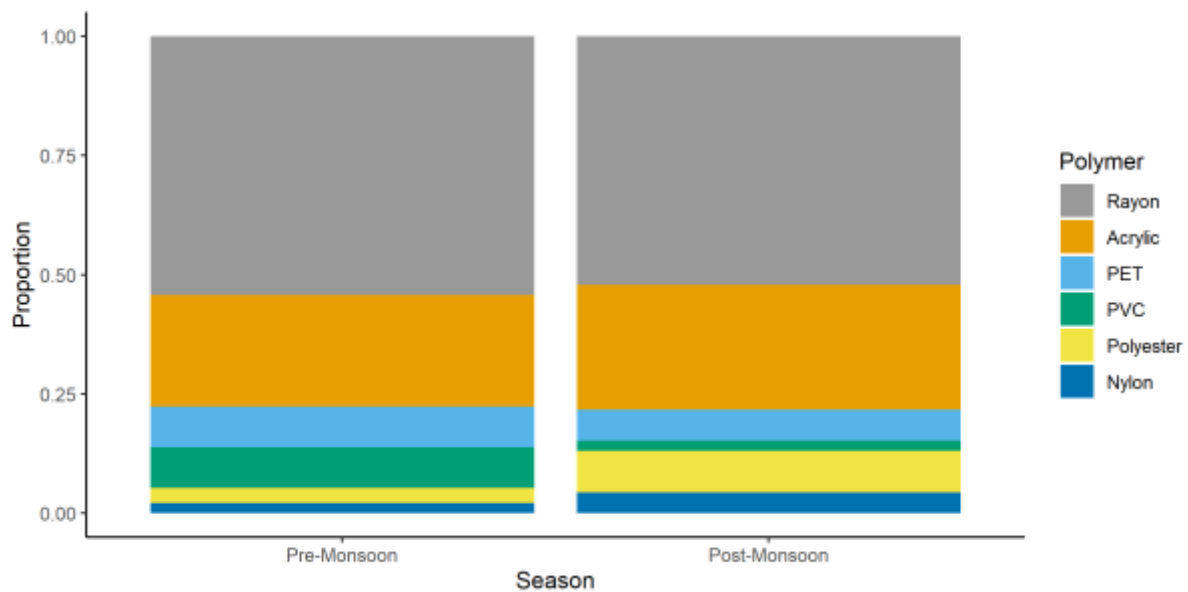


328

329 **Figure 2.** Mean microplastic concentration ($MP L^{-1}$) for both pre and post-monsoon
 330 across all sites. Sites are in order travelling upstream ($n = 6 (30 L), \pm S.E.$).

331

332 In terms of microplastic types, across all sites 91% were fibres and 9% were
 333 fragments. Of the 140 suspected microplastics, FT-IR spectroscopy revealed rayon
 334 (synthetically altered cellulose) as the dominant polymer (54%), followed by acrylic
 335 (24%), PET (8%), PVC (6%), polyester (5%) and nylon (3%; Fig. 3). Considering all
 336 microplastics, the most dominant colour found was blue (74%), followed by black
 337 (11%), red (6%), purple (4%), and brown (2%). Green, yellow and clear particles
 338 each represented 1% of the sample total.
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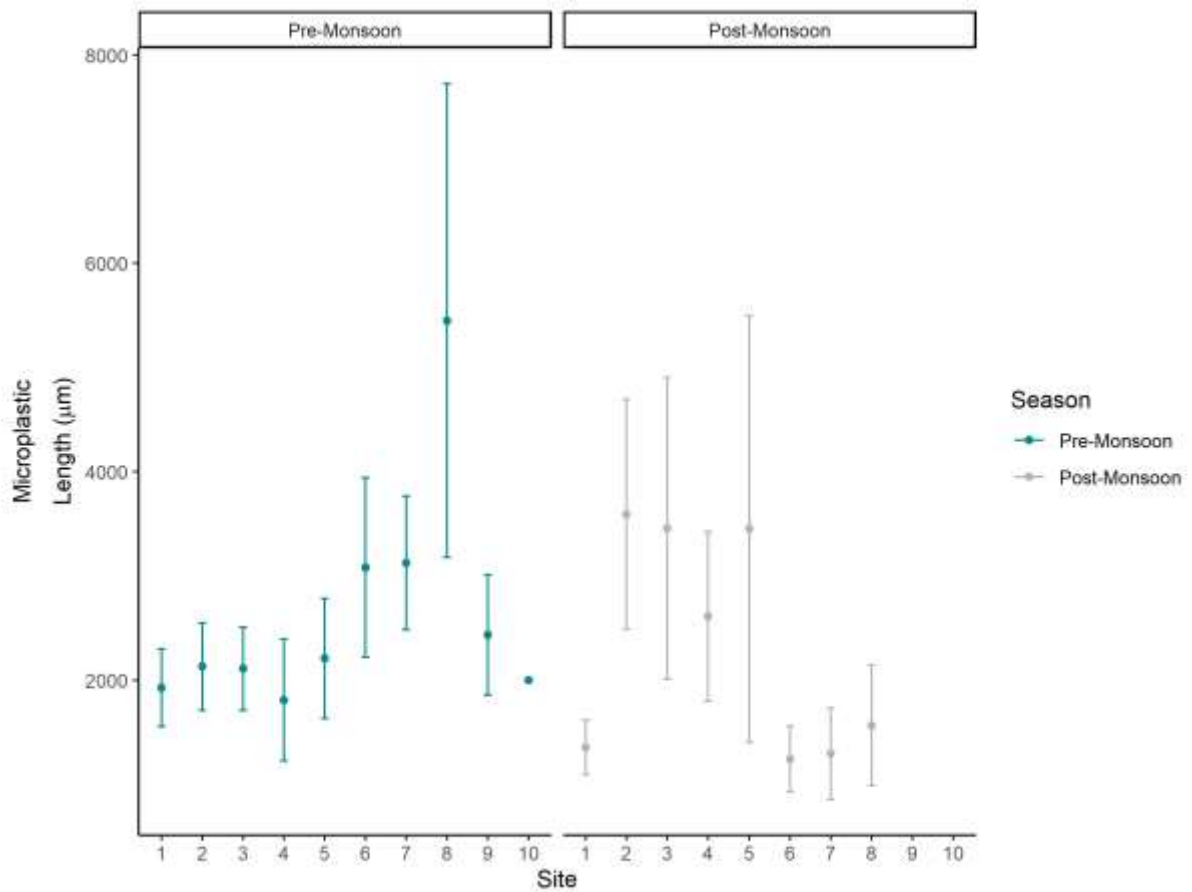
342 **Figure 3.** Proportion of microplastic polymers for pre- and post-monsoon seasons,
 343 across 10 sites along the Ganges river ($n = 6$ (30 L)).

344

345 The average size of microplastic was $2459 \pm 209 \mu\text{m}$. The average size was $2529 \pm$
 346 $263 \mu\text{m}$ in pre-monsoon season and $2317 \pm 341 \mu\text{m}$ in post-monsoon. There was no
 347 significant difference between sites ($p = 0.16$) or sampling occasions ($p = 0.55$).

348 Although not significant, a steady decrease in microplastic size was seen pre-
 349 monsoon when travelling downstream of the river from site 10 (Harsil) to site 1
 350 (Bhola) (Fig. 4).

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354 **Figure 4.** Mean length of microplastic collected along the Ganges pre and post -
355 monsoon seasons and across 10 sites; sites are in order travelling upstream. (n = 6
356 (30 L), ± S.E.).

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369 4.0 Discussion

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371 It is widely reported that the large quantities of plastic entering coastal seas from
372 land-based sources are transported by rivers, (Eerkes-Medrano *et al.*, 2015; Horton,
373 Walton, *et al.*, 2017; Lebreton *et al.*, 2017; Schmidt *et al.*, 2017; Ding *et al.*, 2019).
374 Yet there has been little empirical data from rivers themselves, and limited research
375 understanding microplastic within the Ganges. The river faces a number of
376 biophysical and socio-economic challenges (e.g. pollution, flooding, erosion,
377 cyclones, salinization and water logging) which are increasing alongside the
378 changing climate and anthropogenic developments (Rahman *et al.*, 2020; Sah *et*
379 *al.*, 2020).

380

381 In our study, an average of 0.038 MP L⁻¹ were detected in the surface water of the
382 Ganges and microfibrils were the prevalent microplastic shape (91%). In
383 comparison, research on the Orange-Vaal river system (South Africa) found >99%
384 microfibrils, and a greater average concentration of 1.70 MP L⁻¹; this was conducted
385 with 10 L replicates of freshwater gravity filtered through a 25 µm mesh. Miller *et al.*,
386 (2017) also studied microfibre contamination in the surface water of the Hudson river
387 (U.S.A.) and found an average of 0.98 microfibrils L⁻¹; this was conducted with 3 L
388 replicates of freshwater vacuum filtered through a 0.45 µm mesh

389

390 Our lower concentrations of microplastic may be due to the volume of water sampled
391 (30 L), filtration size (300 µm) or sampling method. Differences in filtration size has
392 been shown to have a substantial effect for concentration estimations. A 2.5-fold
393 increase in microplastics has been reported for a 100 µm net compared to 333 µm
394 net (Lindeque *et al.*, 2020). Research by Song *et al.*, (2014) also identified
395 substantial differences in abundance of microplastics when different sampling
396 methods are used; such as surface microlayer (SML) sampling (16,272 ± 13,457
397 particles/m³) > hand net (50 µm mesh) (1143 ± 3353 particles/m³) > bulk water (213
398 ± 141 particles/m³) > Manta trawl (330 µm mesh) (47 ± 192 particles/m³).

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401

402 The large river systems of the Ganges, Brahmaputra, and Meghna (GBM) combine
403 to create the GBM delta, which is the third largest in the world in terms of water
404 discharge (FAO, 2012; IUCN BRIDGE, 2018). Combining predicted microplastic
405 concentration at the mouth of the river (S1; Bhola) with the discharge of the river, we
406 estimate that 1- 3 billion (10^9) microplastics are released from the Ganges into the
407 Bay of Bengal every day. As above, this is likely to be an under-estimation due to the
408 sampling method. To put this into perspective against other riverine systems,
409 research from Miller *et al.*, (2017) estimated that the Hudson River's watershed
410 drainage area could contribute an average 300 million anthropogenic microfibrils into
411 the Atlantic Ocean per day. Globally, it has been estimated that 60 billion pieces of
412 plastic are discharged into the ocean from rivers worldwide each day (GESAMP,
413 2016), with a global estimate for oceanic microplastic estimated at 4.85 trillion
414 (Eriksen *et al.*, 2014). The results presented here confirm that microplastic discharge
415 from rivers is a substantial vector of microplastics into the marine environment.

416

417 Microplastics were also shown to increase in abundance downstream. Microplastics
418 can be transported long distances in aquatic environments and have been found in
419 remote areas far from large cities (Free *et al.*, 2014; Lusher *et al.*, 2015; Zhang *et al.*,
420 2016). However, the distances over which plastics of different sizes are transported
421 to the sea are poorly known (Horton *et al.*, 2017) because most rivers support
422 human populations along their length, and it is hard to differentiate local plastics from
423 those transported from up-river. Understanding the distance travelled and distribution
424 of microplastics in a river is of importance and requires further research. As shown
425 here, a proportion of microplastics are predicted to follow the flow of the river out
426 towards the coast, but research has also reported that microplastic can sink and
427 collect in riverbed sediments; a maximum concentration of approximately 517,000
428 MP m⁻² of river sediment in the U.K. was found in research by Hurley *et al.*, (2018)
429 and approximately 40 microfibrils per 250 g dry weight in Baltic Sea marine sediment
430 (Zobkov and Esiukova, 2017).

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435

436 Half of the microplastics were identified as rayon (regenerated cellulose fibres),
437 followed by acrylic. Rayon is often reported as a common polymer type for
438 microplastics in both freshwater and marine samples (Lindeque *et al.*, 2020; Nan *et*
439 *al.*, 2020; Park *et al.*, 2020). Rayon and acrylic fibres are mainly used in clothing
440 (Frias *et al.*, 2016; Comnea-Stancu *et al.*, 2017).

441

442 Increased rainfall, associated with monsoons, will impact estimated monthly river
443 plastic inputs into the ocean. Research by Weideman *et al.*, (2020) found
444 microplastic concentration averaged slightly higher in the wet ($2.1 \pm 6.9 \text{ MP L}^{-1}$)
445 than dry season ($1.3 \pm 2.5 \text{ MP L}^{-1}$). On the contrary, our research reports that the
446 pre-monsoon (dry) season had a higher concentration of microplastic compared to
447 post-monsoon (wet). This could be due to the 'flushing' mechanism/role that the river
448 plays during the monsoon season. However, the post-monsoon season did result in
449 a greater microplastic discharge into the Bay of Bengal ($>3 \text{ billion MP day}^{-1}$) due to
450 the increased flow rate and increased volume of water in the wet season. It has been
451 observed that the flooding events during the monsoon months are mostly
452 experienced in the lower-middle and lower part of the Ganges basin thus increasing
453 the discharge rate of water and other associated materials like silt, nutrients and
454 subsequently microplastic. It has been previously estimated that the top 20 plastic-
455 polluting rivers contribute over 74% of annual riverine plastic from May-October,
456 according to a worldwide model (Lebreton *et al.*, 2017).

457

458 There was no significant difference between sites or sampling occasions for
459 microplastic size, but some sites showed substantially higher microplastic
460 concentrations. Notably in Rajbari (S3) and Varanasi (S6) during pre-monsoon
461 season. Sampling in Rajbari occurred the day after (23/05/2019) a large storm with
462 substantial rainfall (20 mm). Rain and storm events have been suggested to increase
463 microplastic contamination, with abundance up to 40-fold higher during a storm
464 compared to before (Hitchcock, 2020); this is likely to have influenced the Rajbari
465 data. Additionally, Rajbari is situated at the confluence of Jamuna-Ganges, which
466 means Jamuna also contributes to the microplastic concentration. Another site
467 where this was observed, Varanasi, is one of seven holy cities in India; it has around
468 84 ghats along the Ganges where over 60,000 people gather daily for holy ritual
469 bathing (Kumar *et al.*, 2012). It is also heavily populated with a density of about

470 14,656 persons km² (Geetika Verma and Shrivastav, 2018). The higher tourist
471 footfall also contributes to increased consumption and dumping of plastic at
472 Varanasi. Therefore, it is suspected that the larger population density and waste
473 generation from cities in India and Bangladesh would increase the quantity of
474 microplastics in nearby river water.

475

476 Conclusion

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478 A substantial quantity of microplastic (1- 3 billion pieces) is estimated to be
479 discharged into the Bay of Bengal on a daily basis, with microplastic concentration
480 increasing from source to sea. Understanding of how microplastic concentrations
481 vary along a river's course is lacking but is critical to help identify key sources and
482 pathways of microplastic and develop management interventions. This research
483 provides the first step in understanding how the Ganges, as well as other major
484 rivers, may contribute to oceanic microplastic. The study will also help provide a
485 global context to plastic pollution in the Ganges when compared with similar studies
486 in other rivers across the globe.

487

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