The effects of wet wipe pollution on the Asian clam, *Corbicula fluminea* (Mollusca: Bivalvia) in the River Thames, London

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ABSTRACT

The aim of the present study was to evaluate "flushable" and "non-flushable" wet wipes as a source of plastic pollution in the River Thames at Hammersmith, London and the impacts they have on the invasive Asian clam, *Corbicula fluminea*, in this watercourse. Surveys were conducted to assess whether the density of wet wipes along the foreshore upstream of Hammersmith Bridge affected the distribution of *C. fluminea*. High densities of wet wipes were associated with low numbers of clams and vice versa. The maximum wet wipe density recorded was 143 wipes m^{-2} and maximum clam density 151 individuals m^{-2}. Clams adjacent to the wet wipe reefs were found to contain synthetic polymers including polypropylene (57%), polyethylene (9%), polyallomer (8%), nylon (8%) and polyester (3%). Some of these polymers may have originated from the wet wipe reefs.

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37   **1. Introduction**

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39   The working daytime population of central London is ca. 10 million people

40   (Piggott, 2015) and according to Barnes et al. (2009) areas with a higher population

41   density are often more affected by plastic pollution. In London, the high population

42   density and its associated plastic waste (Morritt et al., 2014; McGoran et al., 2017,

43   2018) have significantly polluted the River Thames. Due to the tidal nature of the

44   Thames, downstream (east) of Teddington Lock, plastic debris is able to accumulate

45   along certain reaches of the river on account of it being deposited on the foreshore

46   with tidal cycles (Thompson et al., 2009). Thames21, a charity working in improve

47   the Thames waterways, conduct a biannual 'Big Count' survey to quantify the

48   amount and types of plastics found along the foreshore of the river. Roughly, one-

49   third of the plastic found are toiletry items including wet wipes which comprise 18%

50   of the total litter recorded by Thames21 (2019). Another Thames study (Morritt et

51   al., 2014) found sanitary items ca. 22% of total litter recorded. These toiletry items

52   originate from sewage effluent which overflows into the Thames. The overflows also

53   distribute large numbers of microbeads and synthetic fibres (Horton et al., 2017;

54   Mintenig et al., 2017). While sewage treatment works have the potential to remove

55   ~98% of synthetic fibres, many are still released into the watercourse due to such

56   high population densities (Mintenig et al., 2017; Munoz et al., 2018). This is a

57   particular issue after rainfall when the antique treatment works can only deal with

58   small amounts of precipitation by releasing it and sewage directly into the Thames

59   without processing.

60   Synthetic fibres are the most abundant form of plastic pollution found in

61   marine environments and sediments, in particular, are a sink for microplastics

62   (Thompson et al., 2004; Wright et al., 2013). Previous studies have demonstrated the

63   ingestion of synthetic microfibres by some Thames fish species such as the European

64   smelt, *Osmerus eperlanus*, the European flounder, *Platichthys flesus* and roach

65   *Rutilus rutilus* (Horton et al., 2018; McGoran et al., 2017, 2018). The studies by

66   McGoran et al. (2017, 2018) found that fibres were the most dominant form of

67   ingested microplastic. Of the fish sampled, benthic species ingested more plastics

68   than pelagic. This could potentially be due to their close association with sediment

69 containing microplastics and may be inadvertently consumed when feeding on prey

70 (McGoran et al., 2018). Horton et al. (2018) also found that fibres were the most

71 abundant form of microplastic comprising 75% of all those sampled. All polymers

72 identified were either polyethylene, polypropylene or polyester. These materials are

73 all components of wet wipes; a non-woven cloth that once introduced to waterways

74 can breakdown to release microplastic fibres (Horton et al., 2018; Munoz et al.,

75 2018).

76   The Asian clam, *Corbicula fluminea* has been studied in Chinese rivers to

77 monitor plastic pollution through the ingestion of microplastics, most notably

78 microfibres (Su et al., 2018). These clams are highly efficient filter feeders, filtering

79 up to 1L of water per hour (Silverman et al., 1995) and inhabit superficial

80 sedimentary layers by burrowing using their foot and shell (Baudrimont et al., 1997).

81 These clams are an edible species and, as the soft tissue is consumed whole, they

82 provide a direct pathway for the ingestion of microplastics by humans (Su et al.,

83 2018). Originating from south-eastern Asia, *Corbicula fluminea* is an invasive

84 species in the UK that was first recorded in 1998 (Elliott and zu Ermgassen, 2008). It

85 has previously been identified at 4 locations along the River Thames, in West

86 London (Elliott and zu Ermgassen, 2008) and provides an ideal model to assess the

87 potential impacts of plastic pollution.

88   The current study identified *C. fluminea* at a new location along the tidal

89 Thames on the south bank, just upstream (west) of Hammersmith Bridge, that is

90 impacted by a nearby sewage outlet on the north bank and has high densities of

91 plastic pollution in the form of wet wipes and sanitary towels (Fig. 1). The site is on

92 an inside bend of the river, meaning the downstream water velocity is reduced at this

93 point. Consequently, plastic debris suspended in the water column is deposited on

94 the foreshore and subsequently exposed at low tide (Graf and Blanckaert, 2002). A

95 slipway used by St. Paul's School Rowing Club potentially acts further in slowing

96 water flow around the bend of the river, causing wet wipes to be deposited on the

97 downstream side of the foreshore towards Hammersmith Bridge. Here, an

98 investigation was undertaken to determine the density of wet wipes along the

99 foreshore and to assess whether this affects the distribution of the *C. fluminea*

100 population at this site. This study also examined whether microfibres from the wet

101 wipes or other microplastics were being captured by the clams. Based on preliminary

102 observations, it was expected that there would be a significant reduction in the

103   abundance of *C. fluminea* where densities of wet wipes are higher. From previous

104   literature, including the work of Su et al. (2018), it was also expected that the clams

105   would filter the surrounding Thames water and contain microfibres, similar in

106   polymer composition to that of the wet wipes and sanitary items that are

107   accumulating on the south bank foreshore.

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109   **2. Methods & materials**

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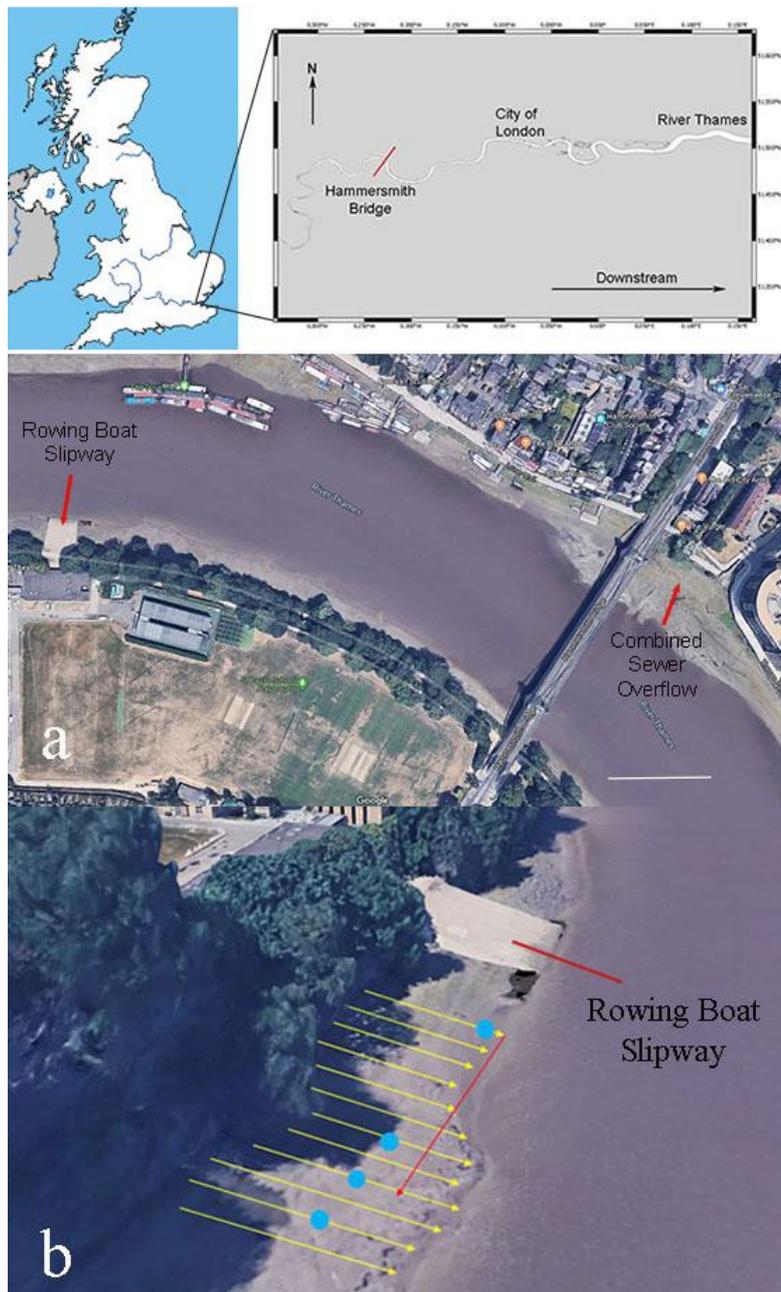
Fig. 1. (a) The sampling site along the south bank of the River Thames at Hammersmith Bridge. To the far right is the rowing boat slipway of St Paul's School that was used as a distance marker for transects which were set out eastward (downstream) of this point towards the bridge. Scale bar = 60m. **(b)** Layout of the 12 transects along Hammersmith south bank foreshore: yellow arrows indicate where wet wipe surveys were conducted (section 2.2), blue dots indicate where clam samples were collected along the same tidal height (section 2.3) and the red arrow indicates where clam densities were measured along the same tidal height (2.4) (Google Earth Pro).

2.1. Survey site

Field surveys were conducted along the foreshore of the south bank, just upstream of Hammersmith Bridge (51˚29'17.574" N 000˚13'55.217" W; Fig. 1a), on the River Thames between February and May 2019. This site is located adjacent to the Hammersmith Pumping Station (HPS) which discharges untreated sewage effluent into the river via the combined sewer overflow, due to a limited sewage capacity when precipitation levels are increased (Fig. 3). Thus the survey site is likely to be exposed to HPS effluent. This section of the foreshore was selected based on Thames21 methodology for their Big Count so that the data collected would be comparable with previous studies. During sampling, mounds of wet wipes and other debris were observed and appeared to alter the topography of the foreshore by forming reefs.
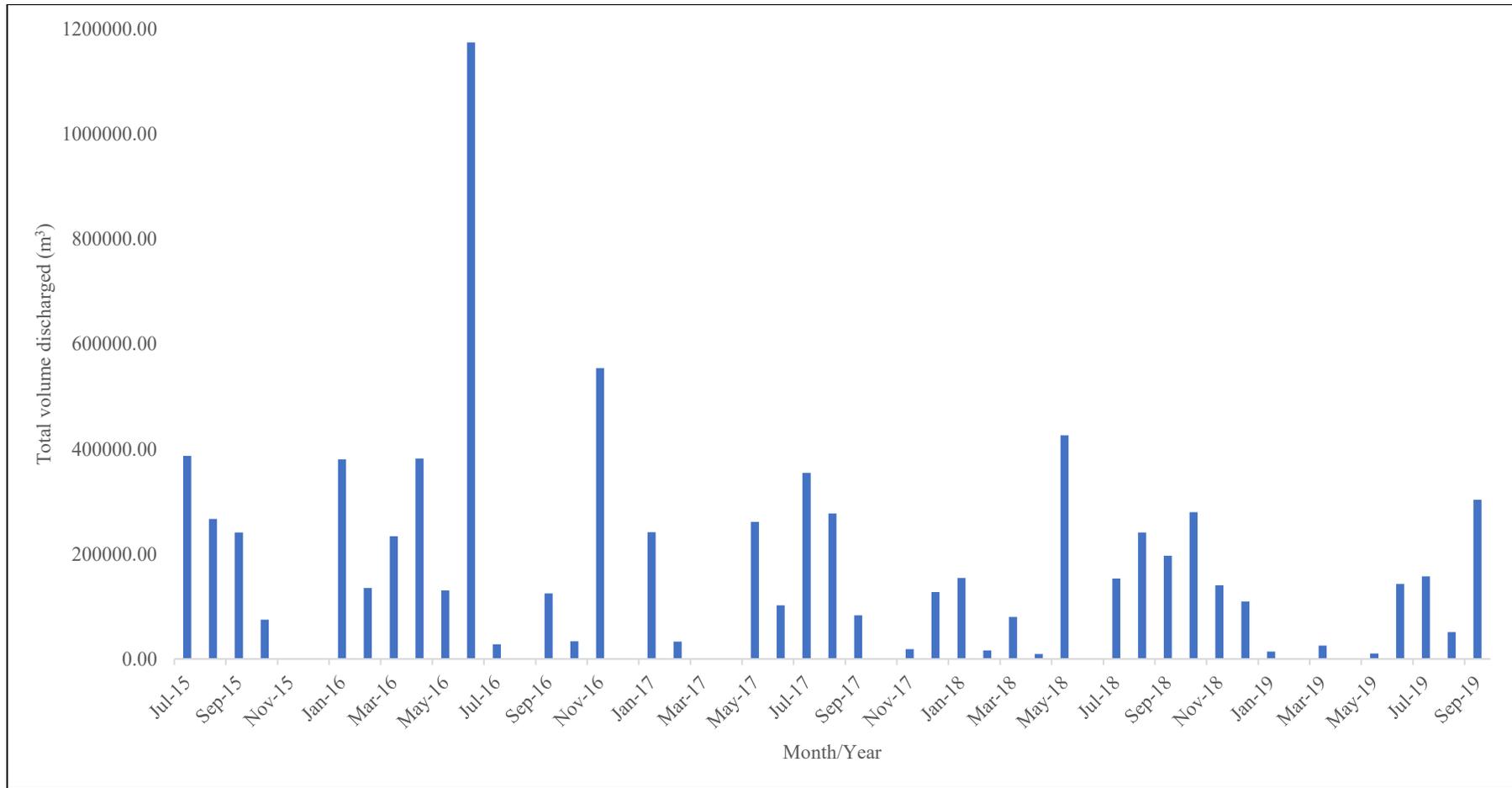
Fig. 2. The amount of sewage discharged (in cubic metres or tonnes) by Hammersmith Pumping Station into the western tideway of the River Thames between July 2015 - September 2019 (Thames Water data).

146 *2.2. Wet wipe surveys*

147   A total of 12 transects were arranged along the foreshore at the

148 Hammersmith sampling site during low tide. The transects were laid from the bottom

149 of the bricked bank where it meets the shoreline to the low water line at 5m intervals

150 between 45–100m from the rowing slipway (Fig. 1b). Quadrats ($1m^2$) were then

151 placed along each transect at 5m intervals. At each quadrat, the number of wet wipes

152 present was recorded along with the number of sanitary items and other

153 miscellaneous plastics. Wet wipes were found by counting those present on the

154 surface and by using a hand trowel to make shallow excavations at each quadrat

155 down-shore from the distance mark. All wipes down to a depth of ~4cm inside the

156 $1m^2$ quadrat were pulled out of the sediment and counted as well as any that

157 protruded into the quadrat. Fragmented wet wipes were counted as individuals. Only

158 a limited number of transects were completed within one tidal cycle, so data were

159 collected over 5 days (11th, 18th, 26th February; 5th, 8th March 2019). Differences in

160 tidal heights restricted the number of quadrats that could be completed on each visit,

161 as some days only allowed for measures to be made down to 20m from the

162 riverbank.

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164 *2.3. Corbicula fluminea surveys*

165   The density of wet wipes and live clams (no empty shells) were counted

166 within $0.5m^2$ quadrats. Twenty-four quadrats were placed at ca. 2m intervals

167 between 45m and 85m from the slipway along the same tidal height of the foreshore,

168 and the total number of wet wipes and the total number of clams counted per $0.5m^2$

169 quadrat. Quadrats were placed at a distance of 25.4–30.3m from the bank along the

170 same tidal height. The range of 45–85m was selected as previous surveys showed

171 that this area had the greatest variation in the density of wet wipes. Wet wipes and

172 clams were counted as per the methods outlined in section 2.2.

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174 *2.4. Corbicula fluminea sampling*

175   Asian clams were collected from the survey site in March 2019 and used for

176 laboratory analysis. These were gathered at horizontal distance points 45m, 70m,

177 85m and 95m downstream from the slipway. For each of these 4 stations, a total of 3

178 quadrats were placed at low tide at times of 1000hrs, 1100hrs and 1145hrs (2 hours

179 13 minutes, 1 hour 13 minutes and 28 minutes respectively) before the predicted

180   time of low water at 1213hrs based on Port of London Authority 2019 tide tables. A

181   total of 8 clam samples were collected from the south bank foreshore at

182   Hammersmith Bridge; 1 sample at 45m from the slipway, 2 at 70m, 3 at 85m and 2

183   at 90m. Due to the shape of the river and the foreshore, collecting clams at the same

184   distance measured from the riverbank would have resulted in environmental

185   heterogeneity across samples. Therefore the clams were gathered from the same tidal

186   height, as determined by marking the height of the receding tide with stakes, and

187   consequently, each collection point was comparable.

188   The presence or absence of *C. fluminea* was recorded for each of the 12

189   quadrats in total and if present, samples of clams were collected. Only live clams

190   were counted and collected with empty shells being disregarded. This meant that

191   different numbers of clams were collected from each of the 4 stations due to

192   presence/absence. Where possible, a minimum of 30 individuals were collected per

193   quadrat, washed *in situ* with Thames water to remove sediment using a sieve,

194   transported back to the laboratory in clean plastic bags and stored in a laboratory

195   freezer prior to further analyses. A control sample of 12 clams was collected from

196   Chelsea Embankment (North Bank of Thames, just upstream (west) of Albert

197   Bridge) on the 1st May 2019. This was used a site for comparison, as wet wipes were

198   absent from the foreshore. Due to their smaller population size, it was not possible to

199   collect large quantities of clams at this location.

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201   *2.5. Sample processing*

202   In the laboratory, all clams were again washed with filtered deionized water

203   (10μm) to remove residual sediment and any microplastics adhered on the surface of

204   the shell. The shell width of each clam was recorded to the nearest 0.01mm using

205   digital Vernier callipers. The soft tissue of each bivalve was dissected out of the shell

206   with a new scalpel blade, cleaned once more with filtered water (10μm) to remove

207   microplastics potentially present in the shell. The clam tissue was then dried by

208   blotting with tissue paper, transferred to 50mL Falcon tubes and frozen at -20˚C to

209   await further analysis. The soft tissue wet weight of each individual was weighed to

210   the nearest 0.0001g and digested in 50mL of 10% potassium hydroxide. The

211   solutions were then mixed and kept at 60˚C in an oven for approximately 12hrs.

212   After this time, the samples were filtered through individual 10 μm Cyclopore™

213   Polycarbonate Membrane Filters using a Millipore vacuum filtration system. The

214   filters containing organic/synthetic debris were placed in Petri dishes and sealed with

215   Parafilm, prior to further analysis.

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217   *2.6. Polymer identification*

218   The filtered clam digestions were observed under a Nikon stereomicroscope

219   (model C-LEDS) to identify any microparticles including potential microplastics. All

220   particles that appeared to be sediment or chitin films were not counted.

221   Microparticles were counted on the Petri dish and categorised by colour and shape

222   (i.e., fibre/film). Due to their minute size, the length of each microparticle could not

223   be measured. An AutoIMAGE Perkin-Elmer Fourier Transform Infrared

224   Spectroscopy (FTIR) at the Natural History Museum (NHM) was used to analyse a

225   sub-sample of microparticles, including microplastic fibres to identify polymer

226   composition. Approximately 20% of the total counted particles from digestions were

227   separated and analysed as a sub-sample due to handling limitations. These particles

228   were placed under an FTIR microscope and visualised using AutoImage Microscope,

229   the aperture recorded, and the material identified using Spectrum Spectrometer

230   containing a spectra library compiled by NHM staff. The spectrum produced by each

231   particle was compared with that of the library by examining the peaks produced to

232   find the most appropriate matches. The percentage match was recorded along with

233   whether the particle was organic, synthetic or semi-synthetic. Those that did not

234   produce a comparable spectrum were disregarded and recorded as 'n/a'. Particles

235   recorded as semi-synthetic were labelled as such if FTIR identified them as either as

236   viscose, chipboard or cellophane.

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238   *2.7. Wet wipe sampling*

239   Samples of wet wipes and sanitary items were collected from the study site.

240   These items were placed in plastic bags and transported to the laboratory where they

241   were washed with filtered water ($10\mu m$) and stored in bags for later analysis. Clear

242   fibres were extracted from each of these items using tweezers to pick out single

243   fibres in order to analyse the types of polymers present using FTIR. All 10 of the

244   extracted fibres were analysed as per the method outlined in section 2.6.

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2.8. Experimental quality control

In order to remove contamination from all water used, reverse osmosis water was filtered through 10μm Cyclopore™ Polycarbonate Membrane Filters using a Millipore vacuum filtration system. Laboratory coats made of 100% cotton were worn to eliminate polyester fibres as a source of contamination. Eco nitrile gloves were worn at all times in order to prevent any further outside microplastic contamination. All dissections, digestions and filtrations were carried out under a laminar flow hood, Airstream® ESCO Class II Biohazard Safety cabinet, model number AC2-4E1. The 10% KOH solution was filtered through a 10μm polycarbonate membrane in a vacuum filtration system prior to use. An open Petri dish containing a damp polycarbonate filter was placed under the laminar flow hood during filtration as an atmospheric control. These could then later be compared with isolated microplastics from digestions and filtrations. Any control dishes that contained synthetic/semi-synthetic particles were accounted for by subtracting the fraction (if significant i.e., > 0.5) from the total count of that material from the samples.

2.9. Statistical Analysis

All statistical analyses were performed using the software R, version 3.4.1. Using data collected from wet wipe surveys (Table 1), a series of correlation tests were used to assess whether there was a relationship between the distance from the slipway or distance from the riverbank and the average number of wet wipes per m^2. A Shapiro-Wilk test was used on the data for each of the different variables to test whether the data were normally distributed. If data were normally distributed, then a Pearson's Correlation was performed, however, if not then a Spearman's Rank Correlation was used as a non-parametric alternative. This was repeated for 2018 and 2017 data provided by Thames21.

The same process was used to assess whether there was a relationship between the distance from the slipway and the number of Asian clams sampled per $0.5m^2$ and again to test for a relationship between the number of wet wipes and the number of clams per $0.5m^2$. After finding that there was a linear relationship between the number of wet wipes and the density of clams, a linear regression was used to determine the relationship between these variables.

279   Pivot tables were used in Microsoft Excel to compare all data recorded for

280   FTIR analysis of microparticles. This was followed by correlation tests which were

281   conducted to assess the potential relationships between different variables and the

282   distribution of microparticles and microplastics contained in *C. fluminea*. These

283   variables included the dry soft tissue weight (g) of each individual, the distance from

284   the slipway at which the sample was collected and the number of wet wipes present

285   at each distance. A Pearson's correlation was selected for data that were normally

286   distributed and a Spearman's rank used as a non-parametric alternative for non-

287   normal data.

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289   **3. Results**

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Distance from slipway/m	45	50	55	60	65	70	75	80	85	90	95	100
Distance from bank/m												
0	0	0	0	0	0	0	0	0	0	0	0	0
5	5	0	0	0	0	1	0	1	0	0	0	1
10	31	63	23	15	27	14	3	14	6	6	20	12
15	85	118	143	52	90	8	5	9	4	0	31	34
20	143	130	102	62	102	34	0	1	0	0	5	1
25	102	3	44	84	114	1	0	0		0		
30				0	1	0						

Table 1.

The number of wet wipes counted per m^2 along 12 transects of the Hammersmith foreshore, measured from the riverbank down to low water at 5m intervals.

316

3.1. Wet wipe surveys

The counts of wet wipes along 12 transects (Fig. 1b) on the Hammersmith south bank foreshore can be seen in Table 1. These results provide an overview of wet wipe distribution along the foreshore by showing densities indicated as a heat map. A colour scale of red through to green indicates areas of high to low densities of wet wipes accordingly. Table 1 is similar to those produced in both 2017 and 2018 from Thames21 data, showing that the highest densities of wet wipes appear to be closer to the slipway and further down the shore between 15–25m. Further data collected by Thames21 shows that one of the largest wet wipe mounds has in fact increased in height by 0.7m between 2014–2018 and a further 0.7m between September 2018 and May 2019. This increased in spite of efforts by Thames21 to remove thousands of wet wipes during their annual Big Count which saw a total of 23,000 wet wipes collected on 23rd March 2019.

The data from each year (2017–2019) were used to assess whether there was a relationship between the average number of wet wipes counted per m^2 and the distance from the rowing slipway. There was a weak negative correlation of in 2017 (Spearman's rank correlation = -0.31; S = 110, p = 0.46). In 2018 there was a stronger negative correlation (95% CI: -0.98 and -0.72, Pearson's correlation = -0.92; t = -7.16, d.f. = 9, p < 0.01). There was also a strong negative correlation in 2019 (Spearman's rank correlation = -0.73; S = 494, p = 0.01). These results indicate that the average density of wet wipes decreases with increasing distance from the slipway (Fig. 3a). Gaps in the data for 2017 may account for the insignificant correlation for this year. There was an outlier at 45m where there was a markedly lower average number of wet wipes than other years which may have skewed the data.

The average number of wet wipes per m^2 was also compared with the measured distance from the riverbank to assess whether there was a correlation between the two variables (Fig. 3b). In 2017 there appeared to be a strong positive correlation (Spearman's rank correlation = 0.82; S = 512.87, p < 0.01). The data for 2018 showed a strong positive correlation (Pearson's correlation = 0.81; t = 4.80, d.f. = 12, p < 0.01). The weak positive correlation found in 2019 was non-significant (Pearson's correlation = 0.35; t = 0.83, d.f. = 5, p = 0.44).
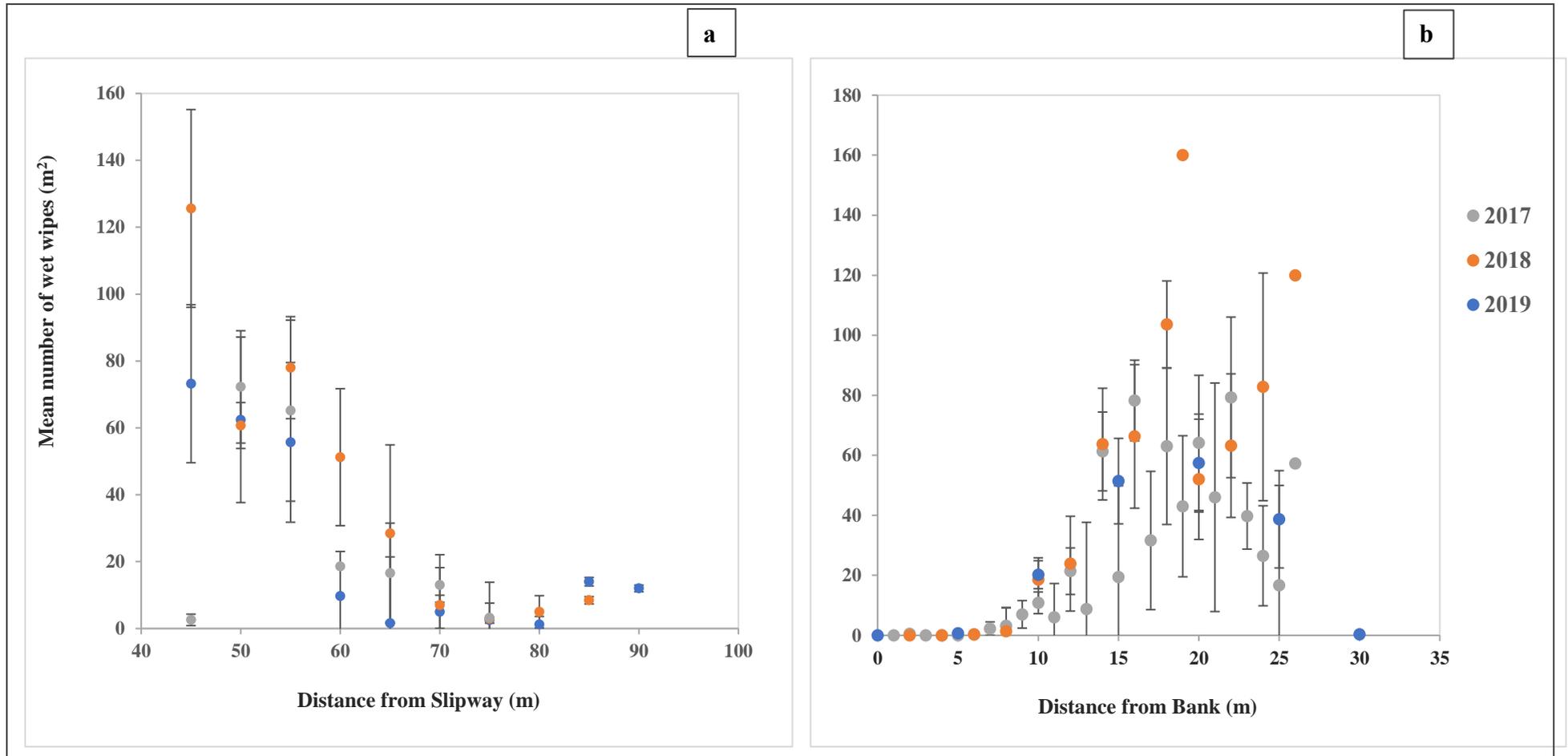
Fig. 3. The average number of wet wipes per m² along 12 transects of the Hammersmith south bank foreshore. **(a)** 5m intervals from the rowing slipway. **(b)** 5m intervals from the bottom of the riverbank down to low water.

378   *3.2. Corbicula fluminea density and distribution*

379   The density of *C. fluminea* appeared to be much higher further down the
380   foreshore towards low water when assessing the presence or absence of them at each
381   quadrat leading to low tide. In addition, there were greater abundances of *C.*
382   *fluminea* further downstream and away from the rowing slipway, where there were
383   fewer wet wipes in general along this part of the foreshore. This was also supported
384   by additional sampling to assess clam densities in the area.

385   There was a significant positive correlation between the approximate distance
386   from the rowing slipway and the number of clams per $0.5m^2$ with the greatest density
387   of clams found at 80m from the slipway with 151 per $0.5m^2$ (Fig. 4; Spearman's rank
388   correlation = 0.56; S = 1020.6, $p < 0.01$). Therefore, the null hypothesis can be
389   rejected. These results parallel those shown in Fig. 3a, which are supported by the
390   negative correlation of -0.73 (section 3.1) between the distance from the rowing
391   slipway and the average number of wet wipes per $0.5m^2$. These data suggest an
392   inverse relationship between the number of wet wipes and the density of clams per
393   quadrat.

394   Where there are higher counts of wet wipes there a few or no clams. In
395   comparison, clams occurred in numbers at locations with few or no wet wipes. There
396   was negative correlation between the number of wet wipes and the number of clams
397   present per $0.5m^2$ quadrat (Fig. 4b; Spearman's rank correlation = -0.76; S =
398   4052.50, $p < 0.01$). The retention time of wet wipes on the foreshore may also
399   influence the apparent correlation however, this could not be determined.
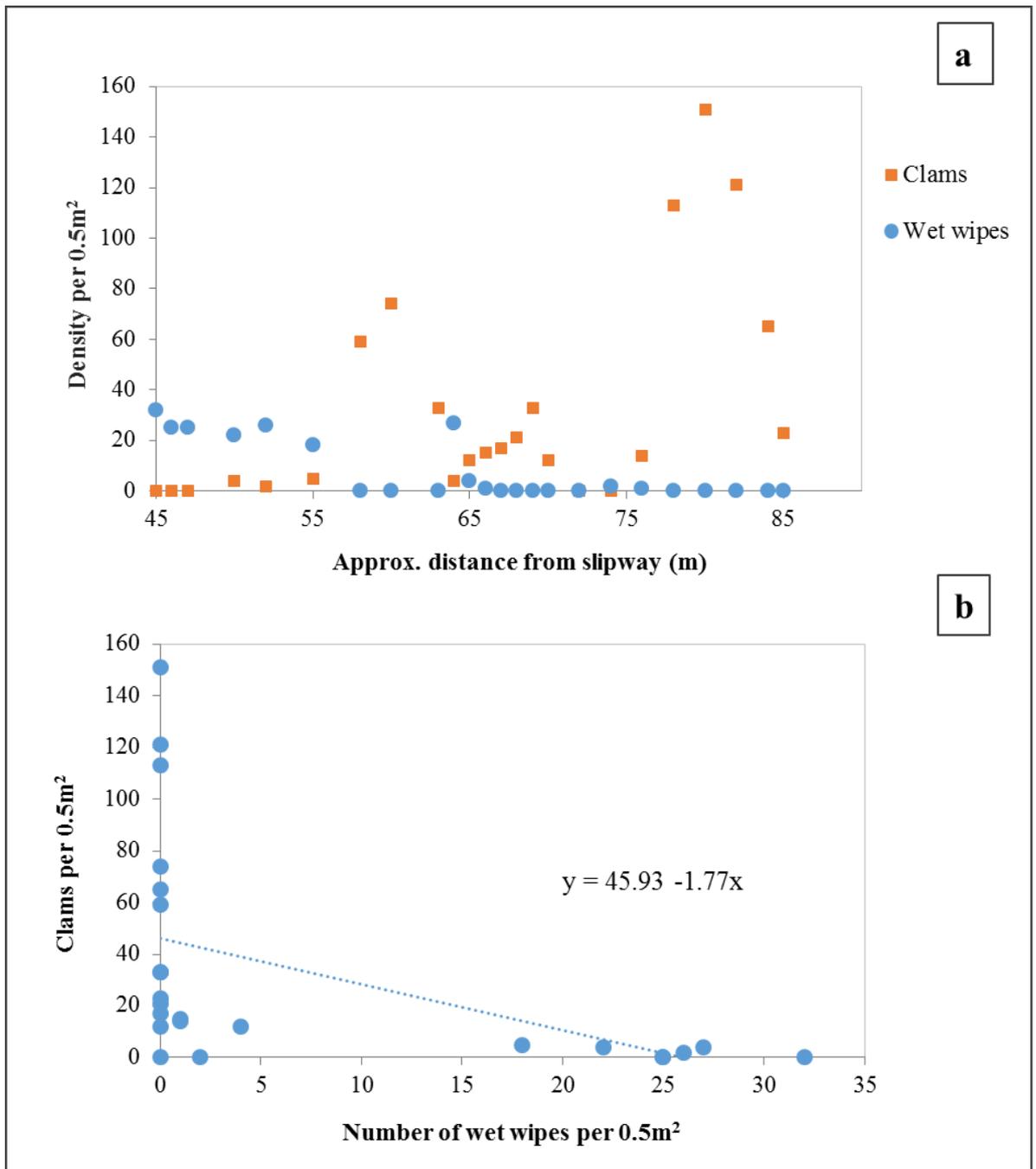
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413 **Fig. 4.** The number of *C. fluminea* and wet wipes per $0.5m^2$ surveyed in 2019 **(a)**
414 with increasing distance from the rowing slipway at the same tidal height (between
415 25.4m at 85m from the slipway and 30.3m at 45m from the slipway) along the
416 Hammersmith foreshore. **(b)** with linear regression fitted trendline (adjusted R^2 =
417 0.2).
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3.3. Polymer identification
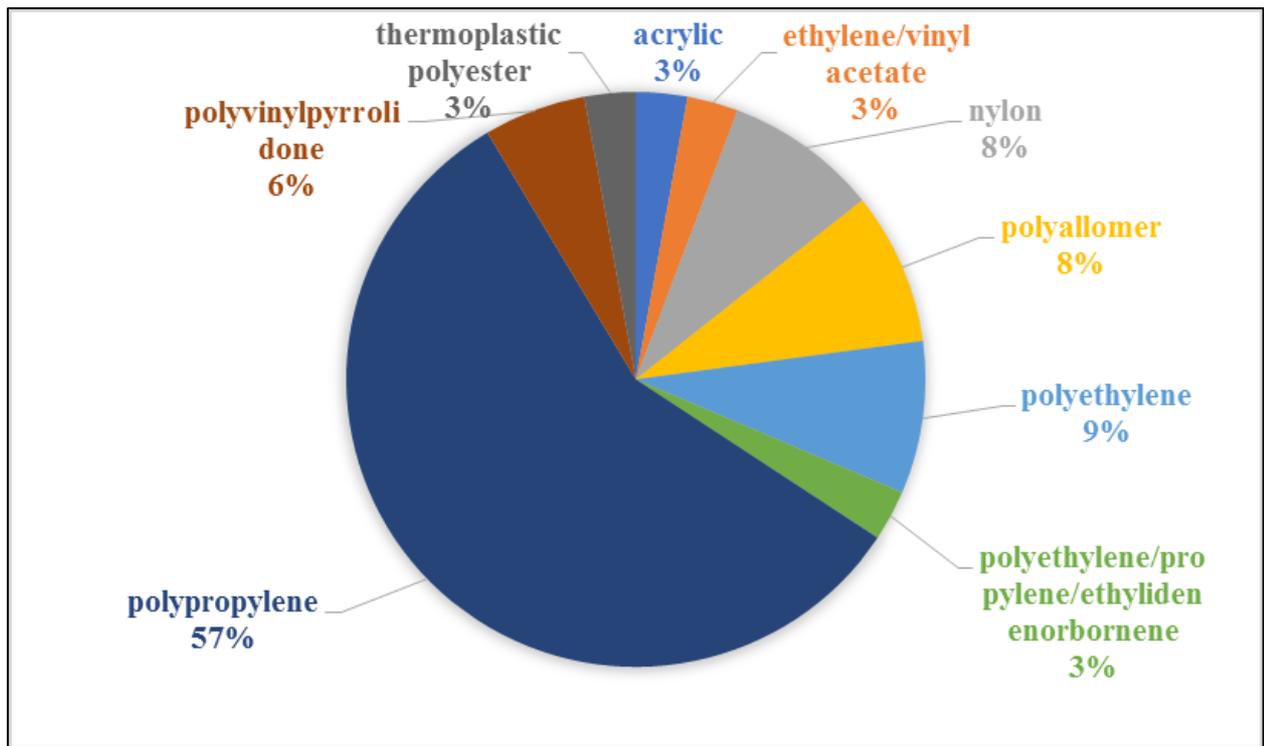
Fig. 5. The types of polymers found from a sample of N=35 synthetic fibres/fragments identified using FTIR spectroscopy of 281 microparticles extracted from the digested soft tissue of *C. fluminea*.

A total of 1404 microparticles were identified from the 9 samples of digested clams (N = 227) with a range of 0-24 particles counted per individual clam (an average of 6.40 particles per individual discounting control samples). All particles were classified as fibres, apart from two that were considered to be film. Of these, 281 were analysed using FTIR spectroscopy. All particles were categorised as either synthetic, semi-synthetic, organic or n/a (see above) with 12% identified as synthetic, 38% as semi-synthetic, 40% as organic and 10% as undescribed due to inability to identify them. Nine different types of synthetic polymer (N = 35) were identified from the samples analysed as shown in Fig. 5. There was an average of 0.77 synthetic particles per individual.

3.4 Polymer content

442   The soft tissue weight of each clam was compared with the total number of

443   microparticles that were extracted after filtration. The test indicated a weak, but

444   significant correlation of (Spearman's rank correlation = 0.27; S = 12083, p < 0.01).

445   The weak correlation may be explained by the characteristically small size range of

446   *C. fluminea* resulting in a range in soft tissue weight of 0.1126g to 2.6748g.

447   There were no significant correlations between the distance from the slipway with

448   the total number of particles for each of the 4 stations; with the proportion of

449   synthetic particles for each of the 4 stations; and with the average number of

450   particles per individual clam per station (Pearson's correlation, p >0.05 in all cases).

451   All 5 of the clear fibres from the wet wipes were identified as polyester and

452   the 5 clear from the sanitary items were identified as polypropylene or

453   polyallomer/polypropylene. Polyethylene/propylene is found in the top sheets of

454   sanitary pads, and polyethylene is also found in the back sheets (Woeller and

455   Hochwalt, 2015). These results, along with those shown in Fig. 5, would suggest that

456   the clams at this site predominantly contain fibres which potentially originate from

457   sanitary items as opposed to wet wipes, among other possible sources.

458   A total of 7 microparticles were analysed from the Chelsea Embankment

459   control sample using FTIR. None were identified as synthetic. This is in keeping

460   with the lack of wet wipes at this site, so it was not expected that any fibres

461   identified were of potential wet wipe origin. Four of these fibres, however, were

462   identified as viscose (a cellulose-based material), which is commonly found in

463   sanitary items, another major contributor to pollution on the Hammersmith foreshore

464   (Always, 2019; Woeller and Hochwalt, 2015). Out of 8 control Petri dishes used

465   during sampling, only two were found to contain microparticles and each one only

466   contained 1 particle per Petri dish. One of these particles was too small for FTIR

467   analysis, the other was identified as viscose. The control dish containing the viscose

468   fibre was used during digestions of two samples. Of those samples, 24 other viscose

469   fibres were identified, leaving a ratio of 1 in 24 (0.04) as a potential source of

470   contamination. This ratio was considered insignificant as a source of contamination

471   and therefore was not deducted from the final count.

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473   **4. Discussion**

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475   *4.1. Wet wipe surveys*

476   The distance of the transect downstream from the rowing slipway appeared to
477   negatively affect the number of wet wipes counted, with higher numbers being found
478   nearer the rowing slipway. The reason for higher levels of deposition at this section
479   of the foreshore is most likely due to the reduced velocity on the inside bend causing
480   deposition at the apex of the bend on this inner side (Graf and Blanckaert, 2002).

481   The positive correlation between the distance from the riverbank and the
482   abundance of wet wipes is most likely explained by variations in the sediment down
483   the foreshore. At the top of the foreshore near the bank, there was much more
484   shingle and drier sediment, due to reduced tidal immersion at this height. This means
485   that wet wipes are unlikely to be deposited in this area. Wipes are therefore
486   deposited further down the foreshore at ~15–25m, where they combine with other
487   objects such as leaf litter and anthropogenic litter in general to form these reefs.
488   There were fewer wet wipes after 25m down the foreshore, potentially due to a much
489   higher immersion time. The weak correlation found for the two variables in 2019
490   may be explained by a reduced number of replicate quadrats down the foreshore as
491   they were only placed every 5m.

492   The methods used in 2019 were an adaptation of Thames21 methodology
493   from previous years. Previous years data employed a less systematic approach, with
494   different numbers of transects and quadrats laid out at uneven intervals. This was so
495   surveys could target areas with the highest densities in order to provide a better
496   estimate of the total number of wet wipes. The sampling method in 2019 used
497   transects set out at 5m intervals along the foreshore and sampling quadrats at 5m
498   intervals from the bank down to the low water line, effectively a grid. Seasonality
499   may also account for variations in the data, as levels of rainfall may determine the
500   number of wet wipes that are deposited along the foreshore due to flooding or
501   changes in the river's current speed.

502

503   *4.2. Bivalve density and distribution*

504   The presence and absence of *C. fluminea* in relation to the distance from the
505   riverbank indicated a wider distribution of clams at heights further down the
506   foreshore at ca. 30–35m. This was the lowest height for low water sampled, with
507   greater tidal exposure, and most likely had a higher abundance of clams due to a
508   greater immersion time needed for their characteristic filter feeding. The results
509   show that there were greater densities of *C. fluminea* the further away from the

510 slipway which is paralleled by low densities of wet wipes further from the slipway,
511 thus supporting the hypothesis that the density of clams is higher where wet wipe
512 densities are lower. In addition, there were almost no *C. fluminea* present in areas
513 with a large abundance of wet wipes. This suggests that the wet wipes are causing a
514 physical disturbance to the distribution of *C. fluminea*, as has been demonstrated in
515 other studies (Aloy et al., 2011; Green et al., 2015; Richards and Beger, 2011). This
516 interaction is not surprising because wet wipes cause a smothering effect as outlined
517 by Goldberg (1997). The accumulation of plastics covering sediments creates a
518 blanketing effect which can result in anoxia by inhibiting gas exchange/redox
519 potential between river water and sediment pore water (Goldberg, 1997; Green et al.,
520 2015). This has been observed when studying the surface sediments beneath wet
521 wipe accumulations where a black layer of anoxic sediment can be observed. Clams
522 inhabit areas near the surface of sediment in order to filter feed from the water
523 column (Sousa et al., 2008). Therefore, the anoxic layer produced by wet wipe
524 smothering makes this area uninhabitable for the clams.
525
526 *4.3. Polymer content*
527   The results from FTIR analysis indicate that *C. fluminea* along the foreshore
528 at Hammersmith contain small quantities of synthetic fibres with an apparent lack of
529 wet wipe-related polymers. No significant relationship was found between the
530 distance from the slipway and the proportion of synthetic particles in each of the 4
531 stations although this may be, in part, due to the limited number of samples used in
532 these analyses.
533   There were few *C. fluminea* individuals present where wet wipe abundance
534 was high, almost a presence or absence distribution. Interestingly there was a higher
535 number of particles where there were fewer wet wipes. The absence of wet wipes
536 and subsequently the lack of synthetic fibres in clams from the Chelsea Embankment
537 control site, support the hypothesis that the wet wipes and other plastic debris found
538 at Hammersmith are the possible sources of synthetic polymers found in *C. fluminea*.
539 It was expected that the soft tissue weight of the individual clams would be
540 positively correlated with the number of particles (Welden et al., 2018), but this was
541 not the case in the present study.
542   Both 'flushable' and 'non-flushable' wet wipes have been found to contain
543 polyester (PET). In addition, 'flushable' wipes are known to contain many more

544 synthetic fibres such as high-density polyethylene (HDPE), polyethylene/vinyl

545 acetate (PEVA/EVA), polypropylene (PP), low-density polyethylene (LDPE),

546 expanded polystyrene (EPS) and polyurethane (PU; Munoz et al., 2018). Therefore,

547 it was expected that some, if not all, of these polymers, would be identified from the

548 wet wipes forming the reefs at Hammersmith. The samples of wet wipes and sanitary

549 towels collected from the Hammersmith foreshore only appeared to contain PET and

550 PP fibres respectively, although other polymers being present cannot be discounted.

551 Flushable and non-flushable wet wipes both contain PET and flushable wipes also

552 contain PP, HDPE and PEVA/EVA all of which were captured by the clams (Munoz

553 et al., 2018). The term captured is used, as this study did not isolate the gut of the

554 organisms sampled, and therefore it is uncertain whether the route of microplastic

555 entry was ingestion. This would suggest that the fibres being captured by the clams

556 have potentially been released by wet wipes on the foreshore and in the water

557 column. PP and viscose fibres/particles were two of the most abundant fibres

558 identified. PP, which accounted for 57% of all synthetic fibres/particles were also

559 identified in every fibre analysed from the sample of sanitary items. Another

560 potential source of fibres is from clothing in domestic washing machines which feed

561 directly into waterways, and in a single wash, a garment can shed over 1900 fibres

562 (Browne et al., 2011).

563   Variations in the amounts of particles contained by individual *C. fluminea*

564 may be explained by a number of factors. Some of the clams may have fed more

565 recently in relation to when they were sampled meaning their gut contents were

566 likely to include more microplastics than those that have had time for gut depuration.

567 Su et al., (2018) found *C. fluminea* showed relatively high retention of microplastic

568 fibres. Future research would benefit from depuration studies to assess gut retention

569 times of microplastics in *C. fluminea*. Seasonality may affect the number of

570 microplastics captured by the clams. Studies have demonstrated that marine species

571 such as *Lepidorhumbos boscii* and *Nephrops norvegicus* were found to ingest fewer

572 plastic items in winter (Welden and Cowie, 2016; Vassilopoulou and Haralabous,

573 2008). Bivalves are potentially one of the groups most impacted by microplastic

574 pollution and have therefore been used widely as bioindicators (Li et al., 2019; Ward

575 et al., 2019). Clams have a wide distribution, are easily accessible, in abundance and

576 are sessile (Li et al., 2019). Therefore, *C. fluminea* is a valuable indicator of

577 freshwater plastic pollution due to their inability to discriminate between

microplastics and other particles during feeding activities and within the digestive system (Su et al., 2018). The effects of capturing and potentially ingesting synthetic polymers have not been demonstrated in the present study, as was the case for Su et al. (2018), but this species is relatively understudied. More research would be needed to assess the potential physiological effects of microplastic ingestion by *C. fluminea*.

4.4. Wider implications

While there were only small amounts of captured microplastics observed in *C. fluminea* examined for the present study there is evidence for a clear physical effect of wet wipe accumulation on the distribution of the clams. Both fibre capture and physical impact on the habitat have potential implications for other species, including native species, that are of a greater conservation concern such as the depressed river mussel *Pseudanodonta complanata* and the pearl mussel *Margaritifera margaritifera* (ZSL, 2018).

5. Conclusions

This study is the first to demonstrate how wet wipes, as a form of plastic pollution, can affect organisms in the River Thames. Accumulations of wet wipes have the potential to affect the distribution of aquatic biota most likely due to the physical disturbance to the environment. This disturbance also conceivably reduces the feeding activity of *C. fluminea*. Furthermore, this invasive species may act as a valuable indicator of plastic pollution in the Thames and by implication the potential impacts on other Thames biota. As such the study provides valuable information for the conservation of biodiversity in the river. Research conducted by Thames21 and Thames Tideway suggests that the problem is increasing in severity, with one of the largest wet wipe mounds showing a height increase of 1.4m since 2014. Our results provide further evidence for the environmental impacts caused by the inappropriate disposal of wet wipes and similar products and the need for greater public awareness of these impacts, improved labelling on packages and appropriate legislation.

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