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Research Paper

Glass reinforced plastic (GRP) boats and the impact on coastal environment – Evidence of fibreglass ingestion by marine bivalves from natural populations

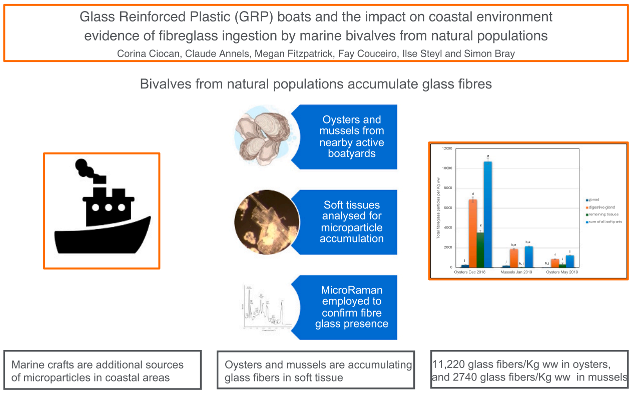
Corina Ciocan [a],[*], Claude Annels [a], Megan Fitzpatrick [a], Fay Couceiro [b], Ilse Steyl [c], Simon Bray [d]

[a] *University of Brighton, School of Applied Sciences, Moulsecoomb Campus, Brighton BN1 4GJ, UK*
[b] *University of Portsmouth, School of Civil Engineering and Surveying, Portsmouth PO1 2UP, UK*
[c] *Aqass Limited, Netley Abbey, Southampton, UK, SO31 5QA*
[d] *School of Biological Sciences, University of Southampton, Life Sciences Building (Building 85), Highfield Campus, Southampton SO17 1BJ, UK*

HIGHLIGHTS

- Potential threat of glass reinforced plastic (GRP) boats to marine environment.
- GRP debris found in oysters & mussels near boatyard on south England coast.
- Filter-feeding bivalves at risk due to particle accumulation.
- Higher levels in winter when boat maintenance increases.
- First study of widespread GRP contamination in bivalves.

GRAPHICAL ABSTRACT



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ABSTRACT

Classified as marine debris, man made materials are polluting the world's oceans. Recently, glass reinforced plastic (GRP) has been shown to degrade and contaminate the coasts. In this pioneering study, fibreglass particles have been detected in the soft parts of oysters and mussels collected from natural populations, in front of an active boatyard. The presence of particulate glass, with concentrations up to 11,220 particles/kg ww in *Ostrea edulis* and 2740 particles/kg ww in *Mytilus edulis,* was confirmed by micro Raman spectroscopy. The results showed higher accumulation during the winter months, when boat maintenance activities are peaking and, through repair work, the release of glass fibres in the environment is more likely. Bivalves are considered high risk species due to their sessile nature and extensive filter feeding behaviour. The microparticle inclusion may contribute to adverse impacts on physiological processes and eventually to a decline in the overall health and subsequent death of the animal. The high costs involved in the proper GRP disposal and the lack of recycling facilities worldwide lead to boat abandonement and further contamination of the coasts. For the first time this

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[*] Correspondence to: University of Brighton, School of Applied Sciences, Moulsecoomb Campus, Lewes Road, BN1 4GJ Brighton, UK.
E-mail address: C.Ciocan@brighton.ac.uk (C. Ciocan).

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study presents the extensive fibreglass contamination of natural bivalve populations, in a popular South England sailing harbour, designated a biological and geological site of specific scientific interest (SSRI).

1. Introduction

The problem of end-of-life Glass reinforced plastic (GRP) boat disposal has recently reached global proportions, particularly for island nations where landfill space is very limited [32]. Efforts are currently being made to identify viable disposal solutions (including pyrolysis, chemical breakdown, and mechanical disintegration) and to halt at sea dumping (scuttling) or onshore burning [27]. Concerns are raised mainly regarding the potential impact of dumped vessels on sensitive habitats, e.g. reefs, seagrasses, coastal communities [29] where physical damage can result through scour due to the motion of vessels, weathering, burning [26].

The use of GRP in sea-faring craft was adopted in the mid 20th century, but truly boomed in the 1960′s, with the whole gamut of boats being manufactured since [30]. Rubino et al. [36] estimates almost 80 % of ship hulls (less than 20 m long) are GRP made.

Whilst the material has gained an ever-increasing popularity in manufacturing and construction of all kinds, up to and including wind turbine blades, management practice for composite waste is yet to be addressed worldwide. Landfill is still the most cost-effective way to dispose of waste composites, whilst the incentives for trying to use bio-derived products or recycle materials are hampered by high costs and the lack of standards [23].

The brunt of microplastic contamination in the aquatic environment is well established (e.g. [1,5,6]). GRP refers to composite material comprising of two basic phases, a polymer element (most often a thermoset plastic i.e polyester, vinyl ester or epoxy resin) with covalently bonded molecules providing superior strength [15]. This mechanical durability of the plastic phase is significantly enhanced by the inclusion of glass fibres, historically borosilicate-based E-glass, the tensile strength of which is further improved by coating with organosilane treatments. Coatings, fillers and primers are utilised, the composition of which depends on the end use of the GRP. When moulded into boat hulls, polymeric paint is often applied, which adds further to the microplastic burden in the marine environment [26,44] when the composite breaks down or is damaged through disposal for example. Gaylarde et al. [18] state in their review that the paint microplastics could be more prevalent in the aquatic environment, by a factor of 30, than alternative sources of microplastics. Furthermore, various chemical compounds associated with GRP boats, especially anti-fouling paints, are historically well known to pose serious environmental threats, due to their heavy metal, booster biocide and microplastics components [2,18,1,38,40,9]. Thermoset reinforced glass fibre plastics are a combination of more than just the resin and glass fibres, fillers such as calcium carbonate in the form of limestone are usually added to bulk the material [21,31].

Although anti-fouling paint development and associated environmental implications continue to be reported, there is a recently identified further risk to benthic organisms especially, in the form of glass microfibres [11,17]. The GRP dust, the material resulting from cutting and sanding operations carried out in active boatyards in the maintenance of GRP vessels, was investigated by Ciocan et al. [11], through Raman and infra-red spectroscopies. The corresponding Raman spectrum of the powder showed numbers of sharp, generally high intensity, bands which are assigned to poly diallyl phthalate. Moreover, the report showed clear adverse effects of GRP on laboratory exposed aquatic organisms, mussels and water fleas.

In this study we investigate the presence of GRP fragments in bivalves collected from natural populations, in Chichester Harbour, UK. The main aim of the study is to provide data on a) the accumulation of GRP in the soft parts of oysters (*Ostrea edulis*) and mussels (*Mytilus edulis)* collected from downstream of an active boatyard and b) the chemical composition of the glass fibres using Micro Raman spectroscopy. To our knowledge, this is the first report of GRP contamination of coastal habitats resulting in the bioaccumulation of glass fibres in bivalves collected from natural populations.

2. Material and methods

Oysters were collected in December 2018 and May 2019 from Dell Quay Yard (50.819273,− 0.8161671) and Sophie's Boatyard (50.8196007,− 0.8155556); mussels were sampled in January 2019 from Itchenor Jetty (50.807159888526314,− 0.8663846467151171), Chichester Harbour, South England (Fig. 1). Sampling locations differ in their main activities, the first two are very close to active boatyards and likely impacted by the debris resulting from boathull maintenance or repairs; Itchenor Jetty area is home to thousands of leisure crafts and the associated activities. Sampling was conducted during the winter months, coinciding with the seasonal boat maintenance and then later, in May, at the start of a busy schedule for water sport activities. At low tide, ten oysters (8–10 cm) and ten mussels (6–8 cm) were collected by hand, on each occasion, and immediately placed in cold storage.

All procedures involving bivalves were performed in compliance with the ARRIVE guidelines and were carried out in accordance with the UK Animals (Scientific Procedures) Act 1986 [45] and the EU Directive 2010/63/EU on the protection of animals used for scientific purposes. The procedures had been approved by the Animal Welfare and Ethics Review Bodies (AWERB) at the University of Brighton.

The bivalve samples were stored at − 20° prior to testing and subsequently thawed at room temperature ahead of dissection. Whole flesh from each individual was dissected and separated into digestive gland/hepatopancreas, gonad and remaining tissues. For each group of bivalves (December 2018 oysters, January 2019 mussels, May 2019 oysters), soft tissues from each organism were pooled together into separate samples (gonad, digestive gland and remaining tissues), which were then further divided into 3 aliquots and separately processed. Each aliquot was weighed (wet weight - ww), labelled and subjected to a chemical digestion method adapted from Van Cauwenberghe and Janssen [46]. In brief, organic material was transferred into 500 mL glass beakers with acid solution (a mixture of 30 % H_2O_2 and 65 % HNO_3; 1:3 v/v) and left for 48 h at room temperature. After 48 h, beakers were placed in an orbital shaker for 30 min at 300 rpm. The resulting solutions were filtered through a series of steel sieves (Fisher Scientific, 100 µm, 53 µm and 35 µm). The sieves were then rinsed on the reverse side with RO water and the liquid was transferred into glass Petri dishes. The isolation procedure was repeated three times to ensure all microparticles were captured. Glass petri dishes were dried in an incubator at 60° for 24 h.

Contamination is commonly reported amongst microparticle studies [16], although glass fibres are not usually present in a laboratory environment, so the risk for our study was minimal. However, additional measures were put in place: all equipment and glassware was rinsed three times with filtered RO water (1.0 µm filter, Whatman, glass microfibre) prior to use in a sterile laminar flow cabinet; all of the apparatus used were rinsed three times with filtered tap water. A 100 % cotton lab coat was worn throughout laboratory testing and all windows and doors were closed to prevent increased air circulation. Soda lime glass petri dishes (DURAN™ steriplan, ø = 60 mm, 120 mm) were used only covered in aluminium foil. A blank extraction without tissue was performed simultaneously, to identify and characterise any procedural contamination.

Petri dishes were analysed using a compound binocular microscope with Nikon SLR/DSLR camera adaptor, at 5 ×, 10 × and 20 ×

magnification. Petri dishes (with microscope slide-grids) were examined methodically from left to right along the first row, and then right to left along the second row etc, to prevent double-counting of the glass fibres. Other types of particles (e.g. microplastics, debris etc.) were not identified or counted for the purpose of this study. Some larger fibres were collected using forceps and stored in small glass Petri dishes for later Micro Raman analysis. Final glass fibres concentrations were expressed as particles per Kg of wet tissue (ww).

Samples containing glass fibre particles isolated from oyster and mussel soft tissues were sent for analysis to the University of Portsmouth, UK. A Renishaw InVia Qontor microscope Raman with a 785 nm 300 mW air cooled laser with 1200 lines/mm gratings and a CCD array detector was employed for the analyses. 50 times objective lens was used, and the 785 nm laser at 5 % power for 10 s and 10 accumulations. The Renishaw Polymeric Materials and ST Japan 60,000 libraries were used here for reference spectra.

One-way analysis of variance (ANOVA) was performed, followed by Tukey's post-hoc multiple comparison test, to identify significant differences between the average (mean) levels of glass fibres for different sample groups, with $p < 0.05$.

3. Results

A high incidence of GRP fibres was detected mostly in samples collected in the winter months (reaching a total 11,220 mp/kg ww) and they were least prevalent in May (calculated level of 1380 mp/kg ww). Overall glass fibre concentrations were highest in the oysters (6880 mp/kg ww digestive gland, 3550 mp/kg ww remaining tissues) and in the mussel digestive gland (1890 mp/kg ww).

3.1. Raman and Infra-red analysis of GRP fragments

The spectra for the glass shard show additional peaks that are clearly identified as associated with the powder, which is coating the entirety of the sample. A clear signal in a broad peak 1200–2000 cm $^{-1}$, is recognised as the spectra for glass under a 785 nm laser (Fig. 2). The GRP powder was previously identified from Raman spectra library as aluminosilicate glass (in the form of the fibreglass content) and a binding resin poly diallyl phthalate [11]. Environmental samples analysed herein led to the identification of the glass fibre corresponding peaks matching the reference spectra from the pristine GRP sample and comparing well with the borosilicate fingerprint [21]. Silicate glasses lack the long-range order observed in crystals, hence they have broad and smooth infra-red and Raman spectral features (e.g., [13]). Bands in the 1000–1200 cm^{-1} region are consistent with [Si-O-Si] asymmetric stretches associated with silicate glasses, those in the 600 to 850 cm^{-1} region are generally assigned to [Si-O-Si] symmetric stretches [13].

3.2. GRP in bivalve soft tissue

Under the light microscope, glass fibre fragments in the digested samples were readily identifiable as transparent cylindrical shards, with lengths ranging from 50 to 250 μm (Fig. 3). All glass particles present were counted for each analysed sample. Results were expressed as number of microparticles (mp) per kg wet weight (ww) tissue. Although we only measured a fraction of the glass fibre fragments, there appear to be no significant differences between fragments from the different sample types; average glass fibre fragment lengths were between 50 μm and 250 μm (Fig. 3).

Fig. 1. Sample sites with key location points, within the Chichester Harbour, South of England.

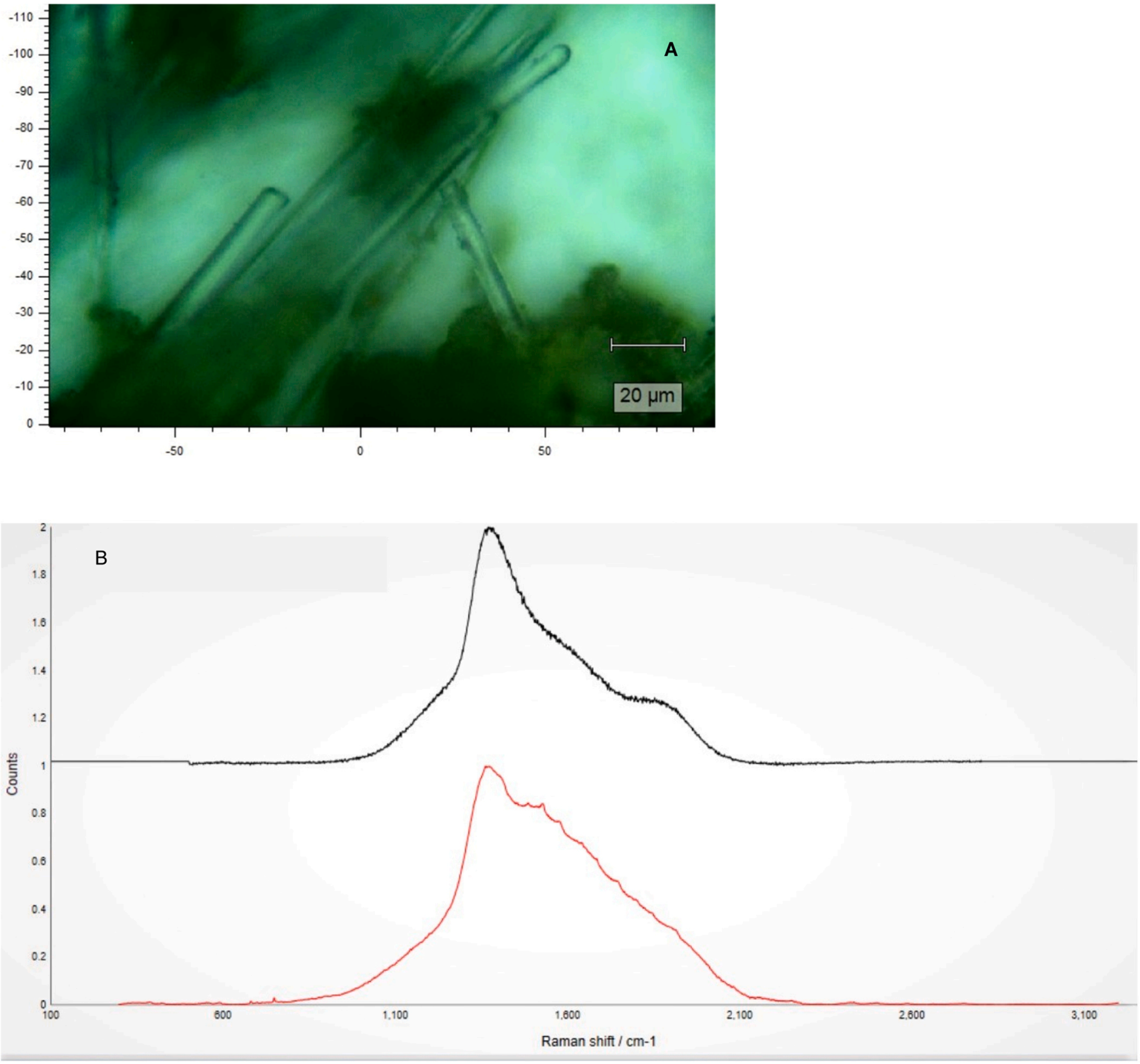

Fig. 2. Raman microscope image of environmental glass shards (A) with associated spectra (B), showing the fit peak resolved band of the fibreglass matching the reference spectra (a standard borosilicate microscope slide used as reference). Sample analysed using Renishaw InVia Qontor microscope Raman at the University of Portsmouth.

A high incidence of glass fibre was detected within our bivalve samples, with the highest oyster levels recorded in December 2018 (Fig. 4).

A one-way ANOVA revealed that there was a statistically significant difference in the glass fibres accumulation in the whole soft body tissues, between all three groups ($p < 0.05$). Tukey's HSD Test for multiple comparisons found that the mean value of glass fibres in the whole soft body of Dec 2018 oysters was significantly higher than the levels in the January 2019 mussels and May 2019 oysters ($p < 0.05$).

Digestive glands were the main sites of the glass fibres accumulations, with oysters collected in December 2018 showing the highest levels of fibreglass present in their digestive gland compared to the May 2019 oyster and January 2019 mussel counterparts ($p < 0.05$). However, the number of fibreglass detected in the gonads were relatively low for all bivalves, with no statistically significant difference between the gonads of December 2018, May 2019 oysters and January 2019 mussels ($p < 0.05$).

4. Discussion

GRP was patented in 1935 by Owens Corning and in the nautical recreational industry, fiberglass is now the most employed material [30]. First GRP composite boats were produced in the USA in the late 30's and most of them survive to this day. GRP has been intensively used in the construction of numerous craft, including yachts, ships and fast rescue lifeboats, fishing vessels and cruisers. Nowadays some of these crafts are becoming obsolete as repair costs outweigh the vessel value, thus their disposal has became a critical environmental issue [8].

Modern technology has seen the development of fibreglass into such products as cloth, e.g.the E-glass Fiberglass cloth, a lightweight woven composite material commonly used in marine and aerospace applications, including GRP (www.carbonfibreglass.com). It is reasonable to

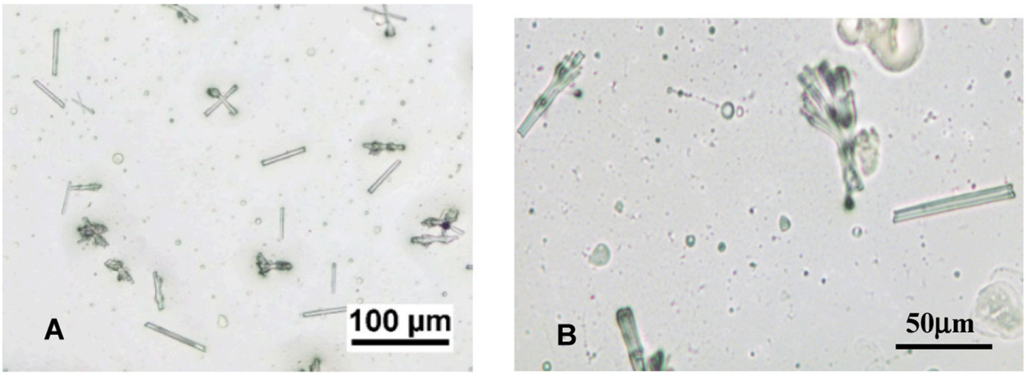

Fig. 3. Glass fibre fragments accumulated in the flesh of bivalves. Shards were visible under the light microscopy after acid digestion of oyster and mussel tissues. A – glass fibre shards present in oyster gonads, December 2018. B – glass fibre shards present in mussel flesh, January 2019.

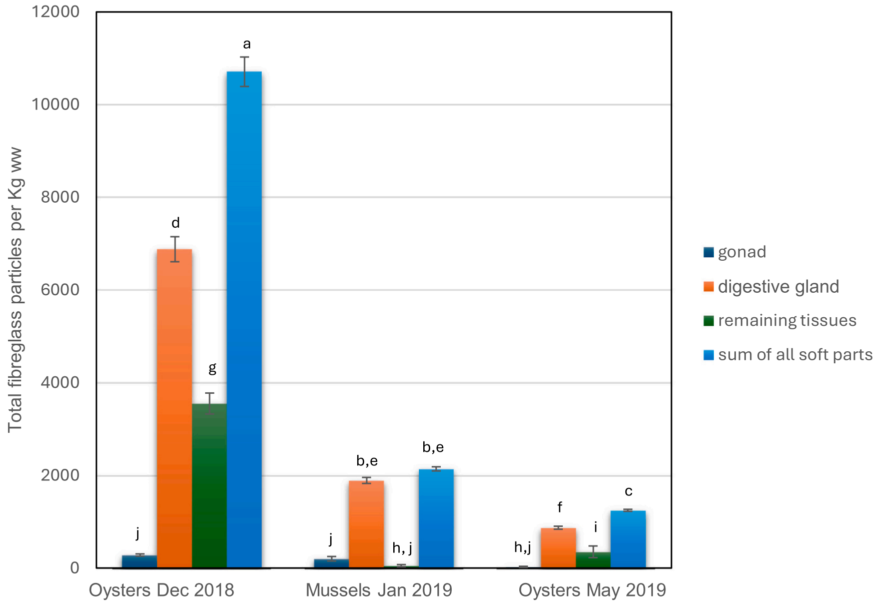

Fig. 4. Levels of glass fibre fragments identified in biota, Chichester Harbour. Vertical bars represent levels calculated as number of particles per Kg wet weight tissue ± SD. n = 10 bivalves per group. Different letters represent statistically significant differences between groups (one way ANOVA, followed by Tukey's post hoc test, $p < 0.05$).

assume that GRP can break down to microplastics and fibre glass [21]; moreover, microparticles can be released when the boats are crushed, dismantled or just repaired [24]. In theory, the GRP fragments have higher density than sea water and will tend to concentrate nearshore, close to boatyards or where vessels are abandoned, disposed of or wrecked [23].

Mussels and oysters are commonly used to study microparticle contamination of the aquatic environment and their consequent biological effects. Field investigations show that microparticle contaminants are detected in marine bivalves worldwide [19,28,49] including polar regions [39,43]. A recent review published by Bom and Sa [4] reported a total of 93 studies on microplastics accumulation in bivalves, with work carried out in 36 countries on 70 species, out of which *Mytilus* sp. and *Crassostrea* sp. were the most common. Oysters and mussels exhibit an equivalent performance as sentinel organisms in a study on microplastics pollution in Brazil [35], showing similar levels of microparticle accumulation, and reflecting the environmental gradient, similar to the findings in our study. However, is it difficult to assess the reliability of the data on microparticle accumulation, and especially on glass fibres, due to the lack of validated and comparable methods for the isolation, chemical identification and quantification. Laboratory studies have also highlighted adverse effects of microparticles in mussels and oysters, including impact on the antioxidant and immune systems, physiological responses, histological changes, energy alteration, genotoxicity and transcriptional responses, and neurotoxic effects (reviewed in Hamm and Lenz [20]; and Ribeiro et al. [35]).

Boating and shipping activities have long since been known to affect environmental ecosystems (e.g. noise pollution, wash impacts, mooring/anchoring), causing habitat loss, ballast/bilge/sewage disposal, macro & microplastic littering and fauna collisions. Moreover, their associated anti-fouling paints leading to toxin release in the environment raises concerns regarding microplastics loss from epoxy based coatings [24,25,37,7]. Antifouling paint particles act as continuous and localised source of heavy metals, biocides and microplastics to aquatic species [33,40,41] and are able to fundamentally alter sediment microbial communities after 30 days exposure [42].

The abandonment of boats no longer fit for use has similarly been noted for some time [29,44], but the scale of this problem is becoming increasingly apparent [26] with particular regard to disposal leading to habitat damage and pollution issues.

The International Council of Marine Industry Associations (ICOMIA) has estimated that there are more than 6 million recreational craft in Europe alone of which 1–2 % are reaching the end of life each year, with only 2.5 % of them (approximately 80,000) disassembled for recycling/

repurposing [14].

Glass fibre reinforced plastics are easily released during cutting or sanding of the boats or other structures (pipes, outdoor furniture, etc) and through normal aging processes [21]. The adverse effects of fibreglass ingestion and accumulation in benthic organisms are yet to be deciphered. Very few studies have investigated the uptake and accumulation of fibreglass in aquatic organisms: e.g Galimany et al. [17], and more recently Ciocan et al. [11] reported the clear uptake of the material, both plastic and fibre glass by blue mussels and water fleas.

Previous studies have shown that microparticles, especially plastics, accumulate in wild or cultured bivalves, with noticeable deposits observed in the digestive tract and gills [12], also in the soft tissues [46]. Negative impacts of microparticles in marine bivalves, largely resulting from ingestion, can influence feeding, growth, reproduction, and animal survival [49]. Interestingly, bivalves are quite selective and particles of different sizes have different ingestion and retention rates, therefore inducing various biological responses [3].

Similar to the Galimany et al. [17] study, we do not know why our bivalves ingested fiberglass, other than because of the possible presence/availability of this contaminant in the environment (Fig. 3). In our study, the quantity of fiberglass found in the digestive gland was notably highest in the winter months (Fig. 4), when boat maintenance activities are peaking and, through repair work, the release of GRP to the environment is more likely. The study shows a significant quantity of GRP particles accumulated in bivalve digestive glands, raising concerns regarding impaired digestive ability. Pedersen [34] found that filtration rate in freshwater mussels declined even in the second day of microplastics exposure, suggesting that consumption of microplastics resulted in enhanced, although false, satiation. Wang et al. [49] suggested that tissue damage associated with microplastic inclusion may contribute to adverse impacts on physiological processes and eventually to a decline in overall health. More specifically, our previous study [11] showed that particulate glass and plastics detected in mussels digestive tubules and gills, led to a suite of inflammatory features observed in all examined organs such as gonad and mantle.

Evidence presented here suggests that marine craft are additional sources of microparticulate contaminants in the aquatic environment though quantifying this is problematic and more research is required [24]. Among them, the fragmented alkaline hygroscopic silicate fibres with aspect ratios comparable to commercial asbestos have the capacity to accumulate in marine bivalves, at very high levels of concentration. Our results highlight the need for better regulation of public access to slipways, commercial boat maintenance facilities and create a better ethos of end of life boat management in general, in order to minimise further exposure and spread of GRP/microplastic contaminants in aquatic environments.

Glass reinforced plastic has been previously compared to asbestos [17,22]. The noxious effects of asbestos in humans have long been established [10,47], although these effects occurred after long periods of ingesting the fibres.

To our knowledge, this is the first report of the introduction of the GRP in a natural food web. Further studies on the potential transfer up the food chain and likely consequences for human health should be prioritised.

Statement of environmental implications

Glass reinforced plastics (GRP) is a composite material consisting of fine strands of glass embedded in a resinous matrix (polyester, vinyl ester, phenolic and epoxy compounds) to form a strong, hydrophobic, and flexible structure.

We previously reported the degradation of the resin and accelerated fibre breakage in the aquatic environment and the release of microplastics and asbestiform like silicate fibres. These microparticles negatively impact the physiology and behaviour of aquatic organisms. Given the recent sales spikes in recreational boats and the lack of recycling solutions, it is imperative to determine the scale of the GRP contamination and the biological impact.

CRediT authorship contribution statement

Claude Annels: Writing – original draft, Investigation. **Fay Couceiro:** Investigation. **Megan Fitzpatrick:** Investigation, Formal analysis. **Simon Bray:** Writing – review & editing. **Ilse Steyl:** Visualization. **CORINA CIOCAN:** Writing – review & editing, Writing – original draft, Supervision, Methodology, Funding acquisition, Conceptualization.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

Data will be made available on request.

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