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**Wild gudgeons (*Gobio gobio*) from French rivers are contaminated by
microplastics: preliminary study and first evidence**

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Abstract

Marine ecosystem contamination by microplastics is extensively documented. However few data is available on the contamination of continental water bodies and associated fauna. The aim of this study was to address the occurrence of microplastics in digestive tract of gudgeons (*Gobio gobio*) from French rivers. These investigations confirm that continental fish ingested microplastics while 12% of collected fish are contaminated by these small particles. Further works are needed to evaluate the occurrence of this contamination.

Key-words: fish; gudgeon, *Gobio gobio*, rivers, microplastics

In 2012, 280 million tons of plastics were produced around the world and used in many domestic and industrial applications (Rochman et al., 2013). This large production associated to their high durability and to the poor rate of recycling contributes to the introduction and accumulation of plastic debris in the environment and particularly in marine environment where this phenomenon is strongly documented (Moore, 2008; Barnes et al., 2009). These wastes are characterized by a large range of size including meso, macro and mega debris with a size >5, 20 and 100 mm respectively but also particles <5mm named microplastics. In marine ecosystems, these synthetic small particles are available in a wide range of organisms such as fish, aquatic mammals, birds, and are able to induce several effects including physical harm but also adverse effects due to desorption of persistent, bioaccumulating and toxic pollutants adsorbed on microplastics (Cole et al., 2011; Wright et al., 2013). Unlike marine ecosystems, few data is available on the contamination of freshwater environment by microplastics. Only recent studies highlight that continental hydrosystems are also contaminated by microplastics. Indeed, microplastics were found in surface water samples from American Great Lakes (Rios Mendoza and Evans, 2013), Lake Geneva in Switzerland (Faure et al., 2013), Lake Garda in Italy (Imhof et al., 2013) and Jade System in Southern North Sea (Dubai and Liebezeit, 2013). Several sources of microplastics in continental waters are clearly identified. Among them, effluents from urban waste water treatment plants appear as a major source (Dubai and Liebezeit, 2013) but wastewater from domestic washing machines cannot be ignored (Browne et al., 2011). However, to our knowledge, no data is available on the contamination of freshwater wildlife such as fish by microplastics. To bridge this gap, the present study was designed to address occurrence of microplastics in wild fish from French rivers.

For this purpose, the gudgeon (*Gobio gobio*), a sedentary Cyprinid fish living only in freshwater ecosystems during all its life cycle and widely used in ecotoxicology (Sanchez et

al., 2011), was selected as sentinel fish species. In Autumn 2012, 186 adult wild gudgeons (mean length: 114 ± 22 mm; mean weight: 14.1 ± 7.5 g) were electrofished in 11 French streams characterized by various environmental pressures (Table 1). Fish were immediately sacrificed by a blow on the head, measured, weighed and gendered. Digestive tract was removed and stored in 70% ethanol at room temperature prior laboratory investigations. For research of ingested microplastics, digestive tract was dissected and its content was subjected to a direct visual inspection under a dissecting microscope. To prevent sample contamination by microplastics, all dissection tools were cleaned using 1.2 μ m filtered ultra-pure water and observed under a dissecting microscope to confirm the lack of microplastic. Examination of dissection tools stored at ambient air during 1 hour indicated that contamination by airborne particles is negligible. In the present work, observations focused on hard and coloured fibers that are easily identifiable but other kinds of microplastics such as transparent fibers and pellets, were also recorded (Figure 1). For occurrence analysis, three groups of fish were distinguished including fish without observed microplastics, fish with ingested microplastics and fish with suspected microplastics (i.e. abnormal particles were observed but cannot be clearly defined as microplastics).

Results of the present study are presented in the figure 2. 12% of collected fish are contaminated by microplastics. Among 11 investigated stations, microplastics were clearly observed in digestive tracts of gudgeons from 7 sites with an occurrence of contaminated gudgeons between 11 and 26 %. This occurrence was up to 40% if suspected microplastics were considered. Result analysis shows that microplastics were not detected in fish from the low anthropised sites BRA, AUV and OLL located in the upper area of river basins. They are observed on the other hand on all urban rivers (*i.e.* BED, LOI, LOV and HER). This hypothesis is in accordance with the sources of microplastics in continental water bodies such as urban waste water effluents (Dubaish and Liebezeit, 2013). However, further studies are needed to

confirm these trends in a large number of sampling stations. To our knowledge, the present study is the first evidence of freshwater fish contamination by microplastics. In a recent work, Faure et al. (2013) documented Lake Geneva's pollution by microplastics and investigated fish contamination. The authors analyzed gut content of 21 northern pikes (*Esox lucias*), 18 roach (*Rutilus rutilus*) and 2 bream (*Abramis brama*) and no plastic pieces was observed. This result could be explained by the behaviour of gudgeons that eat on substrate and could be more exposed to microplastics than other fish species used by Faure et al. (2013) that eat in water column. This hypothesis is consistent with recent observations performed in the Lake Garda indicating that sediments and associated organisms of this freshwater ecosystem are contaminated by microplastics (Imhof et al., 2013). Compared to marine fish, occurrence of freshwater fish contaminated by microplastics appears as weaknesses. Indeed, occurrence levels between 26 and 52% were reported in various fish species from the English Channel (Lusher et al., 2013). Analysis of the present results shows a lack of relationship between gender and microplastic contamination but also between biometric parameters and occurrence of microplastics (data not presented).

This work provides first evidence that freshwater fish ingests microplastics. This result confirms also that freshwater ecosystems are contaminated by these small particles and represent a source of contamination for estuarine and marine ecosystems (Dantas et al., 2012). Now, further researches are required i) to characterize and quantify this phenomenon using other fish species with various trophic levels but also various freshwater organisms such as molluscs and crustaceans but also ii) to assess the impact of microplastic ingestion on fish health. In the future, this work could support the integration of microplastic occurrence in the definition of the good status of continental water bodies similarly to the marine water.

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Table 1. Description of sampling sites and number of collected gudgeons (*Gobio gobio*).

River (code)	GPS coordinates	Environmental pressure ^a	Number of gudgeons (M/F)	Fish length (mm)	Fish weight (g)
Bramerit (BRA)	45°51'55'' N 0°34'11'' E	Low anthropised site	8 (4/4)	108 ± 7	11 ± 3.2
Auvézère (AUV)	45°25'43'' N 1°18'27'' E	Low anthropised site, Agriculture	19 (8/11)	121 ± 29	19.4 ± 8
Yerres (YER)	48°39'04'' N 2°54'12'' E	Agriculture (cereal)	20 (4/16)	104 ± 11	9.4 ± 3.6
Jouanne (JOU)	48°02'01'' N 0°42'20'' E	Industry (Plastic industry)	19 (8/11)	93 ± 4	5.9 ± 0.9
Dore (OLL)	45°40'31'' N 3°37'47'' E	Low anthropised site, Pharmaceutical industry	20 (6/14)	114 ± 22	14 ± 7.5
Risle (RIS)	48°50'36'' N 0°43'21'' E	Industry (Mixed)	7 (2/5)	121 ± 20	16.5 ± 8.5
Bedat (BED)	45°54'03'' N 3°17'42'' E	Urban (500 PE)	20 (13/7)	122 ± 14	15.7 ± 5
Loire (LOV)	45°33'45'' N 4°16'16'' E	Urban (>10.000 PE)	20 (10/10)	113 ± 25	14.8 ± 2.8
Loire (LOI)	46°08'47'' N 4°05'49'' E	Urban (1000 PE)	19 (9/10)	114 ± 12	12.2 ± 4.3
Chée (CHE)	48°46'01'' N 4°40'47'' E	Agriculture (Vineyard, cereal)	20 (8/12)	123 ± 13	14.9 ± 4.8
Hers-mort (HER)	43°44'59'' N 1°22'47'' E	Urban (1000 PE)	14 (7/7)	84 ± 10	6.5 ± 2.6

^a Major environmental pressure is presented for each site. For industrial pressure, the type of industry is indicated between brackets. For urban pressure, the capacity of waste water treatment plants located near the site is presented between brackets and expressed as population equivalent (PE).

Figure caption

Figure 1. Examples of microplastics observed in the digestive tract of gudgeons (*Gobio gogio*).

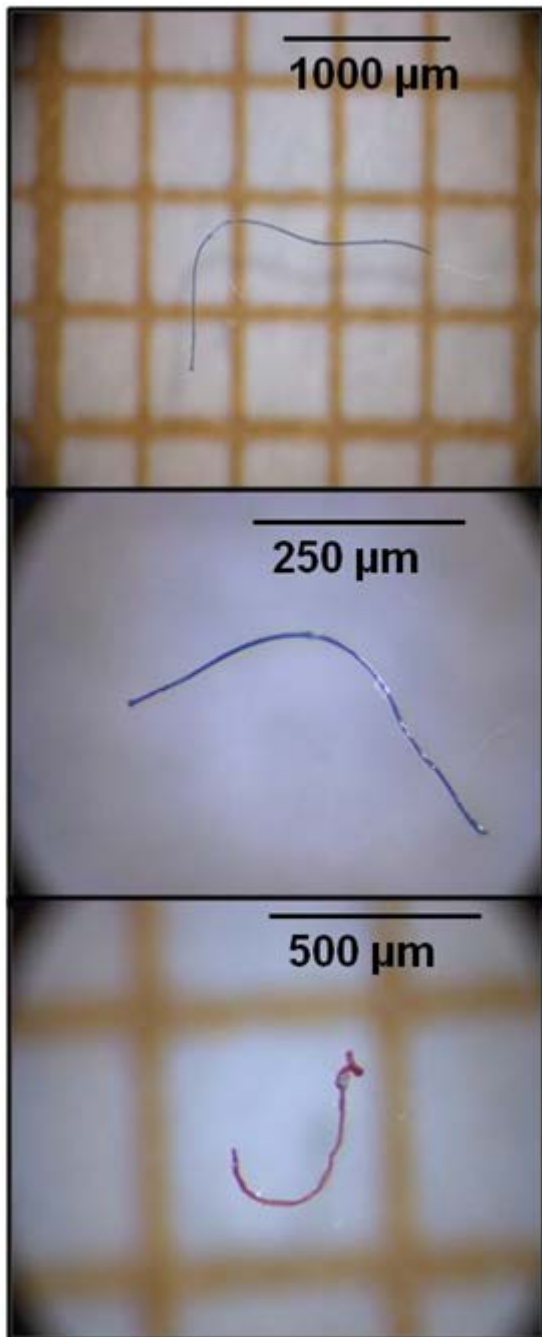


Figure 2. Occurrence of gudgeons (*Gobio gobio*) containing microplastics or suspected microplastics in their digestive tract. Site codes are indicated in the table 1.

