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Macroplastic litter colonization by stream macroinvertebrates relative to that of plant litter: A meta-analysis $\stackrel{\star}{\sim}$

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Keywords: Anthropogenic litter Freshwater zoobenthos Plastic pollution Plastic colonization Stream macroplastic Systematic review	Environmental pollution by anthropogenic litter is a global concern, but studies specifically addressing the interaction between macroplastics and macroinvertebrates in streams are scarce. However, several studies on plant litter decomposition in streams have also used plastic strips as a methodological approach to assess if macroinvertebrates colonize plant litter mostly as a substrate or a food resource. Looking at these studies from the plastic strips perspective may provide useful information on the interaction between macroplastics and macroinvertebrates in streams. I carried out a meta-analysis of 18 studies that have compared macroinvertebrate colonization of macroplastic litter and plant litter in streams to estimate the overall macroinvertebrate colonization of macroplastic litter version vertebrate in the plant litter relative to plant litter, and identify moderators of this difference. Macroinvertebrate colonization of macroplastic litter was overall lower (by $\sim 40\%$) compared with plant litter. However, differences in macroinvertebrate colonization between macroplastic litter and plant litter were observed when considering leaf litter but not wood litter, which may be a poorer substrate and food resource for macro-invertebrates. Also, differences in macroinvertebrate colonization between macroplastic litter and redelores, but not for grazers that may feed on the biofilm developed on macroplastics. Macroplastic litter supported lower macroinvertebrate diversity on macroplastic litter may have occurred when macroplastics represented more heterogeneous substrates (e.g., mixture of plastic types) than leaf litter (e.g., needles). Differences in macroinvertebrate abundance between macroplastic litter and leaf litter were not significantly affected by plastic type, mesh opening size, plant functional group or plant identify. By testing previously untested hypotheses, this meta-analysis guides future empirical studies. Future studies should also consider the geographical areas most affected by macroplast				

1. Introduction

The global production of plastics reached 390.7 million metric tons in 2021, most being polypropylene (PP, 19.3%), low-density polyethylene (LDPE, 14.4%), polyvinyl chloride (PVC, 12.9%), high-density polyethylene (HDPE, 12.5%), polyethylene terephthalate (PET, 6.2%), polyurethane (PUR, 5.5%), and polystyrene (PS, 5.3%); bio-based plastics (e.g., polylactic acid, PLA) represented only 1.5% of the global plastic production (Plastics Europe, 2022). Considering their application, most plastics were used for packaging (44%; Plastics Europe, 2022), which is the application with the shortest lifetime expectancy (Geyer et al., 2017). Coincidently, an estimation of the fate of all plastic produced up to 2015 (i.e., 8300 million metric tons) revealed that 59% has accumulated on landfills or the environment (Geyer et al., 2017).

Recognizing the environmental threat of plastic pollution, the research on this topic has increased exponentially over the last 20 years. However, in a systematic literature review for the period 1980–2018, Blettler et al. (2018) found that studies addressing plastic pollution focused more on marine environments (87%) than on freshwaters (13%), and more on micro- (<5 mm; 76%) than on meso- (5–25 mm; 5%) or macroplastics (>25 mm; 19%), and that studies focusing on macroplastics derived mostly from Europe, North America, and Asia. Similarly, in a systematic literature review spanning a 9-year time frame (2013–2021), Gallitelli & Scalici (2022) found that studies addressing

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plastic pollution focused more on marine environments (72%) than on freshwater (15%), terrestrial (10%) or atmospheric (3%) environments, and more on micro- (<5 mm; 95%) than on macroplastics (>5 mm; 5%), and that studies on macroplastics were mostly from Asia (43%) and Europe (36%).

The low number of studies addressing plastic pollution in rivers may partially derive from these ecosystems being mostly perceived as pathways linking terrestrial (source) and marine (sink) environments. In fact, a recent modelling exercise estimated that the plastic outflow from 1518 rivers distributed worldwide to the marine environment was 57 000-265 000 metric tons per year (Mai et al., 2020), which is more than the estimated plastic accumulation in surface ocean water globally (6600-35 200 metric tons; Cózar et al., 2014). However, the large amount of plastics in rivers and their interaction with river physical and biological structures provide numerous opportunities for macroplastics to be retained (e.g., buried in the sediment, trapped in log jams, wooded islands, macrophytes or infrastructure), so that rivers can also act as sinks for macroplastic litter (van Emmerik and Schwarz, 2020; van Emmerik et al., 2022). For example, Liro et al. (2022) found that retention of macroplastic litter depended on river retention structures, being higher in wood jams and wooded islands than in exposed sediments and herbaceous vegetation. Distinct retention structures also differed in the type of macroplastics they retained, with wood jams, exposed sediments, and herbaceous vegetation retaining a higher proportion (59%, 67%, 83%, respectively) of light plastic items, such as bags and sheeting (soft polyolefin) and pieces of foam and food boxes (expanded PS), while wooded islands retained mostly (54%) bottles (PET) and other rigid pieces (hard polyolefin) (Liro et al., 2022). In a field experiment, air-filled PET bottles travelled between 321 m and 1.1 km from the release point in 24 h, suggesting that some types of macroplastics are retained shortly after entering the river (Newbould et al., 2021). Additionally, studies comparing the retention of plastic strips and leaves in streams (to assess if plastic strips could be a good surrogate for leaf litter to address stream retention capacity) found that plastic strips show similar, or even shorter, travel distances compared with leaves, and are retained by the same instream retention structures as leaves (Speaker et al., 1988; Larrañaga et al., 2003). Despite this, only about one-third of the studies addressing macroplastic litter in rivers focused on retained macroplastics (e.g., in the sediment, vegetation or riverbank; 36%), while the majority of the studies focused on floating or suspended macroplastics (i.e., on water; 64%) (Al-Zawaidah et al., 2021; Gallitelli & Scalici, 2022). Also, most studies addressing macroplastic litter in rivers were carried out at their lower end (i.e., potamal environment; 80%), while less than 10% took place at their upper end (i.e., rhitral environment) (Gallitelli & Scalici, 2022). However, macroplastic litter is pervasive across the drainage network, including mountain water courses (Honorato-Zimmer et al., 2021; Poletti & Landberg, 2021; Liro et al., 2022).

Once retained by instream structures (e.g., stones, logs) or settled in depositional areas, macroplastics interact with freshwater biota, e.g., by providing a novel substrate for smaller organisms such as microbes and invertebrates (Windsor & al., 2019). In fact, macroplastics seem to become well colonized by freshwater microbes (both heterotrophic and autotrophic), with no strong differences in amount and community structure being found among plastic types (e.g., PVC, LDPE, and PS; biodegradable and non-biodegradable) and between these and hard natural substrates (i.e., tiles) (Chaudhary et al., 2022; Vincent et al., 2022; Laffet et al., 2023). However, gross primary production was higher on hard plastic substrates than on leaf litter (Hoellein et al., 2014), whereas an opposite pattern was found for microbial densities and enzymatic activities (Laffet et al., 2023). Macroplastics are also colonized by freshwater macroinvertebrates (Kennedy & El-Sabaawi, 2018; Artru & Lecerf, 2019; Wilson et al., 2021; Gallitelli et al., 2023), which may use them as a habitat or refuge, but also as a food source in the case of grazers than can feed on the established biofilm (Michler--Kozma et al., 2022). Macroinvertebrate diversity was higher on

macroplastics than on rocks, which was attributed to higher surface complexity and flexibility of the former compared with the latter substrate (Wilson et al., 2021), but it did not differ from that on leaf litter (Kennedy & El-Sabaawi, 2018). In contrast, macroinvertebrate abundance and percentage of shredders were lower on PLA plastics than on leaf litter (Artru & Lecerf, 2019). However, the number of studies specifically addressing the interaction between macroplastics and freshwater fauna remains very low (virtually absent until 2018; Blettler et al., 2018), which limits strong conclusions. In fact, a wide range of toxic compounds have been identified as plastic additives (e.g., antioxidants, flame retardants, plasticizers), many of which are endocrine disruptors (Hermabessiere et al., 2017; Gunaalan et al., 2020), which can confer distinct toxicity to different plastic types. Accordingly, under laboratory conditions, leachates from PVC were more toxic than those of other plastic types to a variety of freshwater and marine organisms (Lithner et al., 2012; Li et al., 2016; Capolupo et al., 2020).

In this study, I investigated the colonization of macroplastics by benthic macroinvertebrates in streams (i.e., rhitral environment), in comparison with that of plant litter, by means of a meta-analysis. Only two studies have so far specifically aimed to assess macroplastic litter colonization by benthic macroinvertebrates in streams, in comparison with that of plant litter, in the scope of stream pollution by macroplastics (Kennedy & El-Sabaawi, 2018; Artru & Lecerf, 2019). However, several other studies focusing on plant litter decomposition have, since 1992, compared macroinvertebrate colonization of plant litter and plastic strips in streams as a methodological approach to understand if benthic macroinvertebrates use decomposing plant litter primarily as a substrate (physical habitat, refuge) or as a food resource, by considering plastic strips as a control that would preferentially be used as a substrate compared with plant litter that would be used as both substrate and food (Table S1). In these studies, authors generally aimed for similar surface area or volume between the plastic strips and the plant litter, with care being taken to increase the three-dimensional complexity of the plastic strips (Table S1). Reanalysing these data from a plastic perspective can inform on the potential use of macroplastics by stream benthic macroinvertebrates and advance our understanding about the impacts of stream pollution by macroplastics.

By combining 18 primary studies that have compared macroinvertebrate colonization of macroplastics and plant litter in streams (regardless of the original goal) using meta-analytic techniques, I aimed to identify (i) the overall effect (its magnitude and direction) of stream pollution by macroplastics on benthic macroinvertebrate colonization of macroplastic litter relative to plant litter, (ii) the moderators of this effect, and (iii) future research avenues. My general hypothesis is that macroinvertebrate colonization of macroplastic litter is lower than that of natural plant litter because benthic macroinvertebrates may use the former mostly as a substrate while the latter can be used both as substrate and food. I also hypothesized that the magnitude of the difference in macroinvertebrate colonization between macroplastic litter and plant litter is affected by the type of plant litter, macroinvertebrate functional feeding group, macroinvertebrate metric, plastic type, litter bag mesh opening size, plant functional group, and plant identity. The specific questions posed and hypotheses tested in this review are listed in Table 1.

2. Materials and methods

2.1. Literature search

Studies comparing the colonization of macroplastic litter *and* plant litter by benthic macroinvertebrates in streams, published in English between January 1, 1970 and December 31, 2022, were searched on January 9, 2023 using Web of Science (database: Core Collection; indices: Science Citation Index Expanded, Conference Proceedings Citation Index – Science, Book Citation Index – Science, and Emerging Sources Citation Index). The following search string was applied to the Questions and hypotheses addressed, datasets used and location of the results.

Questions	Hypotheses	Datasets ¹	Results
Q1: Does macroinvertebrate colonization of macroplastic litter differ from that of natural plant litter in streams?	H1: Macroinvertebrate colonization of macroplastic litter is lower than that of natural plant litter because macroinvertebrates may use the	Total macroinvertebrates	Table 2, Fig. 2
inter unier nom unie of natural plant inter in streams.	former mostly as a substrate while the latter can be used both as a substrate and as a food resource.	Total FFG	Table 2, Fig. 3
Q2: Is the difference in macroinvertebrate colonization between macroplastic litter and plant litter affected by the type of plant litter?	H2: The difference in macroinvertebrate colonization is higher when macroplastic litter and leaf litter are compared than when macroplastic litter is compared with wood litter because macroinvertebrate colonization of leaves is higher than that of wood.	Total macroinvertebrates	Table 2, Fig. 2
Q3: Is the difference in macroinvertebrate colonization between macroplastic litter and plant litter affected by the FFG?	Hactonivertebrate colonization of leaves is higher than that of wood. H3: The difference in macroinvertebrate colonization is higher for shredders that use plant litter as both a food resource and as a substrate and can use the macroplastic litter only as a substrate than for grazers that can graze the biofilm from both the plant and the macroplastic litter.	Total FFG	Table 2, Fig. 3
Q4: Is the difference in macroinvertebrate colonization	H4: The difference in macroinvertebrate colonization is higher for	Total macroinvertebrates,	Table 2,
between macroplastic litter and plant litter affected by the	diversity-related variables than for abundance-related variables	Leaves	Fig. 2
macroinvertebrate metric evaluated?	because a higher number of taxa colonizes plant litter (i.e., taxa that	Shredders	Table 2,
	use it as a food resource and taxa that use it as a substrate) than	0.11	Fig. 3
	macroplastic litter (i.e., taxa that use it mostly as a substrate), while	Collectors	Table 2,
	the persistence of macroplastic litter may support high numbers of the fewer taxa that colonize it.		Fig. 3
Q5: Is the difference in macroinvertebrate colonization	H5: The difference in macroinvertebrate colonization is higher when	Total macroinvertebrates	Table 2,
between macroplastic litter and plant litter affected by the	PVC plastics are compared with plant litter as PVC can leach more	abundance, Leaves	Fig. 4
type of plastic?	toxic substances (e.g., metals, dioxins, vinyl chloride) compared with	Total macroinvertebrates	Table 2,
	other plastic types. Difference in macroinvertebrate colonization may also be higher when PET plastics are compared with plant litter as PET can leach antimony trioxide and phthalates.	richness, Leaves	Fig. 4
Q6: Is the difference in macroinvertebrate colonization	H6: The difference in macroinvertebrate colonization is higher for	Total macroinvertebrates	Table 2,
between macroplastic litter and plant litter affected by	larger mesh openings as these facilitate the colonization of plant litter	abundance, Leaves	Fig. 5
mesh opening?	by cased-caddisfly shredders that may be too large to access plant litter enclosed in mesh bags with smaller mesh openings.	Shredder abundance	Table 2, Fig. 6
		Collector abundance	Table 2,
		m 1	Fig. 6
Q7: Is the difference in macroinvertebrate colonization	H7: The difference in macroinvertebrate colonization is higher for leaf	Total macroinvertebrates abundance, Leaves	Table 2,
between macroplastic litter and plant litter affected by the N-fixing capability of the plant species?	litter from N-fixing than from non-N-fixing plant species because the former has higher nutrient concentrations that may attract more	Shredder abundance	Fig. 5 Table 2,
N-fixing capability of the plant species:	macroinvertebrates (e.g., shredders).	Silleddel abuldance	Fig. 6
Q8: Is the difference in macroinvertebrate colonization	H8: The difference in macroinvertebrate colonization is higher for leaf	Total macroinvertebrates	Table 2,
between macroplastic litter and plant litter affected by the	litter from deciduous than from evergreen plant species because the	abundance, Leaves	Fig. 5
deciduousness of the plant species?	former is generally more palatable (e.g., softer, lower concentration of	Collector abundance	Table 2,
	structural and secondary compounds) to macroinvertebrates.		Fig. 6
Q9: Is the difference in macroinvertebrate colonization	H9: The difference in macroinvertebrate colonization is higher for leaf	Total macroinvertebrates	Table 2,
between macroplastic litter and plant litter affected by plant species?	litter from species known to produce more palatable (e.g., alders) than more recalcitrant leaf litter (e.g., oaks) because the former may attract macroinvertebrates either to feed on it or use it as a substrate while the latter will be used more as a substrate at early decomposition stages.	abundance, Leaves	Fig. 5

FFG, Functional feeding group; ¹See Table 2 for details on the datasets used for each analysis.

field 'Topic', which considers titles, abstracts, and key-words (both those provided in the study and those used for indexing purposes): '((stream OR river) AND (plastic strips OR plastic leaves OR artificial detritus))'. The literature search retrieved 331 studies (Fig. S1). Another 6 potentially relevant studies known to the author, but that were not identified in the literature search, were added, making a total of 337 studies (Fig. S1).

2.2. Study selection

Studies were screened at title and abstract level to access their compliance with pre-defined inclusion and exclusion criteria. Studies that reported macroinvertebrate colonization metrics (i.e., abundance, biomass, density, diversity, evenness or richness; in any unit) for both macroplastic litter (>25 mm) and plant litter, enclosed in meshed litter bags or grouped in litter packs and incubated in the benthos of at least one stream, were retained; in case of doubt about compliance with inclusion criteria, the study was retained. In contrast, studies addressing the effects of microplastics or nanoplastics on macroinvertebrates (e.g., López-Rojo et al., 2020; Seena et al., 2022), studies addressing the consumption of macroplastics by macroinvertebrates in the laboratory (e.g., Batista et al., 2022), studies reporting only microbial colonization of macroplastics (e.g., Schlief, 2004; Schlief & Mutz, 2005), and studies

where litter was incubated in lakes (Karádi-Kovács et al., 2015) or in brackish waters (Costa et al., 2021), were excluded. This first level of screening retained 24 studies for full text screening (Fig. S1).

Retained studies were screened at the full text level and studies were selected for inclusion in the database if they reported: (i) macroinvertebrate colonization metrics as means, (ii) variation measures (i.e., standard deviation (SD), standard error (SE) or 95% confidence interval (CI); not mandatory for all studies as it can be imputed from similar cases as described below), and (iii) sample sizes, for both macroplastic litter and plant litter incubated in similar conditions. Six studies were excluded: two studies did not compare macroinvertebrate colonization of macroplastic litter and plant litter (Dobson, 1991; Wilson et al., 2021), one study had the macroplastic litter addition treatment confounded by other manipulations of organic matter (Wallace et al., 2015), one study buried the litter (Fritz & Feminella, 2011), one study did not report macroinvertebrate colonization metrics but showed 2-dimensional representation of community data (Boulton & Foster, 1998), and one study was inaccessible (Robertson & Milner, 2001). After full text screening, 18 studies were included in the database (Fig. S1, Tables S1 and S2).

2.3. Data extraction

2.3.1. Extraction of primary data

Primary data (i.e., means, variation measures, and sample sizes) were extracted regarding the colonization of macroplastic litter and plant litter, enclosed in meshed litter bags or grouped in litter packs and incubated in the stream benthos, by total macroinvertebrates or functional feeding groups (FFG: collectors, shredders, grazers, and predators); data on individual taxa (at any taxonomic level) were not considered because studies differed in the taxonomic level used and in the taxa reported, which would limit data compilations across studies (Table S2). Data were only extracted when macroplastic litter and plant litter being compared were incubated in similar conditions (e.g., same mesh size in litter bags, same environmental conditions) (Table S2); in some studies, the incubation conditions (of both the macroplastic litter and plant litter) differed from reference or control conditions (e.g., streams under urban or agricultural influence; e.g., Dangles et al., 2001; Kennedy & El-Sabaawi, 2018), but the potential impact of these incubation conditions on the results were assessed by sensitivity analyses (see below). Means, variation measures, and sample sizes reported in the text and in tables were extracted directly, data in graphs were extracted with the online open tool WebPlotDigitizer (https://automeris. io/WebPlotDigitizer/), and missing information was requested from the authors.

Most studies reported colonization of macroplastic litter and plant litter by macroinvertebrates at multiple sampling dates, and data were extracted for all dates that were common between macroplastic litter and plant litter (Table S2). In these cases, to mitigate potential problems derived from the non-independence of effect sizes (which would be exacerbated by considering multiple dates for the same treatment), the effects sizes of individual sampling dates were combined within each treatment by subgroup analysis to produce a mean effect size per treatment across sampling dates (see below for information on effect size calculation and subgroup analyses; Table S2). Variation measures were extracted as reported in the studies or as provided by the authors (i.e., SD, SE or 95% CI); SD values were used directly in the calculation of the variance associated with effect sizes, while SE and 95% CI values were first converted into SD values. For two studies (Bird & Kaushik, 1992; Artru & Lecerf, 2019), missing variation measures were imputed from similar cases in the database (i.e., cases reporting values in the same unit) that reported means and associated variation as: $SD_i = \overline{X}_i \times$ $(\Sigma_i^K SD_i / \Sigma_i^K \overline{X}_i)$, where SD_i is the missing SD value, \overline{X}_i is the mean estimate for which SD is missing, K is the number of studies i for which there is information on mean estimate (\overline{X}_i) and associated variation (SD_i) (Lajeunesse, 2013).

2.3.2. Extraction of moderators

Several variables that vary among studies, or even within studies, can affect the magnitude and direction of the difference in macroinvertebrate colonization of macroplastic litter relative to plant litter; these are explanatory variables, also called 'moderators' in metaanalysis. According to the questions and hypotheses outlined in Table 1, information was extracted for several moderators, including: plant litter type (leaves or wood), macroinvertebrate FFG (collectors, shredders, grazers or predators), macroinvertebrate metric (abundance, biomass, density, diversity, evenness or richness), plastic type (several types), mesh opening (1–5 mm, 6–10 mm or >10 mm), plant functional groups (nitrogen (N)-fixing: N-fixer or non-N-fixer; deciduousness: deciduous or evergreen), plant identity (several species), and other factors that could impact litter colonization by macroinvertebrates (several factors, including urbanization, acidification, and agriculture) (Table S4). Information was extracted on other potential explanatory variables, but sample size was too small or there was not enough variation among studies to allow testing hypotheses (e.g., water characteristics were reported for only 5 studies; 15 studies were carried out in temperate climates, while only 2 were done in continental, 1 in dry, and 1 in tropical climates; data not shown).

2.4. Effect sizes

The colonization of macroplastic litter relative to plant litter by macroinvertebrates was assessed using the response ratio R, i.e., the ratio between the mean colonization metric (i.e., abundance, biomass, density, diversity, evenness or richness) on macroplastic litter ($\overline{X}_{plastic}$) and on plant litter (\overline{X}_{plant}); analyses were performed on lnR, i.e., ln ($\overline{X}_{plastic}/\overline{X}_{plant}$) (Hedges et al., 1999). lnR = 0 indicates no difference in macroinvertebrate colonization between macroplastic litter and plant litter, lnR < 0 indicates lower macroinvertebrate colonization and lnR > 0 indicates higher macroinvertebrate colonization of macroplastic litter relative to plant litter (Table S3).

The variance associated with lnR (V_{lnR}) was calculated using the mean colonization metric, its associated variation, and sample size for the macroplastic litter and the plant litter (Borenstein et al., 2009). The V_{lnR} was needed to define the weight of individual effect sizes in the estimation of the overall effect size; more precise (i.e., with lower V_{lnR}) effect sizes make a larger contribution than less precise (i.e., with higher V_{lnR}) effect sizes to the overall effect size. The V_{lnR} was also needed to determine the 95% CI associated with individual effect sizes, so that lnR associated with 95% CI that do not include 0 are significant (Table S3).

The 18 studies included in the database contributed with 132 individual effect sizes (considering one effect size per treatment, with multiple sampling dates already combined): 88 individual effect sizes for the dataset considering total macroinvertebrate colonization (hereafter referred to as total macroinvertebrate dataset) and 44 individual effect sizes for the dataset considering colonization by FFG (hereafter referred to as FFG dataset) (Table S3). Individual studies contributed with multiple effect sizes (up to 20) to the database, as a result of using multiple incubation conditions such as several streams or incubation treatments (e.g., Dangles et al., 2001; Negishi & Richardson, 2006), multiple plant litter species (e.g., Quinn et al., 2000; Gonçalves et al., 2012; Kennedy & El-Sabaawi, 2018), and multiple colonization metrics (e.g., Bird & Kaushik, 1992). The potential effects of the non-independence of effect sizes on the results were mitigated by using a hierarchical approach to moderator analysis (see below).

2.5. Data analyses

2.5.1. Overall effect size

The overall difference in macroinvertebrate colonization of macroplastic litter relative to plant litter, i.e., the overall effect size, was estimated for the total macroinvertebrate dataset and the FFG dataset using the random effects model of meta-analysis. This model was chosen because studies vary in multiple methodological and ecological aspects, and therefore two sources of variance associated with effect sizes need to be considered: the within-study variance (V_{InR}) and the between-study variance (estimated by the restricted maximum likelihood (REML) method) (Borenstein et al., 2009). Individual effect sizes were weighted by the inverse of their variance (considering both sources), and the overall effect size lnR was considered significant if its 95% CI did not include 0. To facilitate interpretation of results, lnR values were converted back to R (R = exp(lnR)), and comparisons were made against 1 (instead of 0). The contribution of between-study variance to total heterogeneity (I²) was also estimated (Borenstein et al., 2009).

2.5.2. Subgroup analyses

The effects of moderators on the magnitude and direction of the difference in macroinvertebrate colonization of macroplastic litter relative to plant litter, according to pre-defined questions and hypotheses (Table 1), were assessed for subsets of the datasets, depending on available sample size (only moderator levels with at least three

individual effect sizes were considered) and robustness to publication bias (see below). Subgroup analyses (using the random effects model of meta-analysis and the REML method for estimation of between-study variance) were used to estimate mean effect sizes lnR, converted to R (R = exp(lnR)), for moderator levels (subgroups) (Borenstein et al., 2009). Mean effect sizes R for subgroups were considered significant if their 95% CI did not include 1, and mean effect sizes R significantly differed between subgroups if their 95% CI did not overlap (p (Q_M model) < 0.050). To mitigate potential problems derived from non-independence of effect sizes (i.e., multiple effect sizes contributed by individual studies) and avoid moderators confounding each other, moderators were tested sequentially on subsets of the datasets (hierarchical approach; Fig. S2). For example, when considering the total macroinvertebrate dataset, the first tested moderator was 'plant litter type' (2 subgroups: leaves \times wood); as only the mean effect size considering leaves was significant, and had large enough sample size, the next moderator ('metric': density \times biomass \times abundance \times richness × diversity; the evenness subgroup was not considered because N = 2) was tested considering only the subgroup leaves from the previous moderator. There were significant differences for most subgroups within the moderator 'metric', but sample size was large enough only for subgroups 'abundance' and 'richness', and thus the next moderator ('plastic type') was tested considering only the subgroups abundance and richness (separately as they differed significantly) (Fig. S2). When no significant differences were found between subgroups of a moderator, and they showed the same response, they were kept together to test the next moderator (Fig. S2).

2.5.3. Sensitivity analyses

The original datasets included cases where litter was incubated in conditions that were affected by other factors (e.g., agriculture, acidification, intermittency, urbanization; Table S3), which could impact litter colonization by macroinvertebrates. Therefore, datasets without cases where streams were affected by other factors were used to redo the analyses, to the extent possible considering the reduction in sample size (N = 45 for the reduced total macroinvertebrate dataset and N = 20 for the reduced FFG dataset). Considering effect sizes derived from cases affected by these other factors in the original datasets would be a problem if the interpretation of the results changes when considering the reduced datasets.

2.5.4. Publication bias analyses

Evidence of publication bias was assessed for the total macroinvertebrate dataset and the FFG dataset (original datasets and the datasets without cases where streams were affected by other factors that could impact litter colonization by macroinvertebrates) by using funnel plots. Funnel plots are scatter plots with individual effect sizes (lnR) in the x-axis and precision (SE) in the y-axis, and where symmetrical distribution of individual effect sizes around the overall effect size indicates no publication bias. When publication bias was detected, its impact on the overall effect size was assessed by the Duval and Tweedie's trim and fill method, which estimates a new overall effect size by adding the 'missing' individual effect sizes assuming that the funnel plot should be symmetrical (Duval & Tweedie, 2000). Overlap between the original overall effect size and the new overall effect size (considering the 'missing' individual effect sizes) indicates that publication bias is not severely affecting the original overall effect size.

Robustness of subsets used in subgroups analyses to publication bias was assessed by the Rosenberg's fail-safe number (N_{fs}), which gives the number of insignificant effect sizes that would need to be added to the dataset to nullify the mean effect size. N_{fs} > 5 × N + 10 (N = number of effect sizes) indicates that the dataset is robust to publication bias.

Analyses (i.e., overall effect sizes, subgroup analyses, and publication bias analyses) followed standard methods (Borenstein et al., 2009). Estimation of overall effect sizes and subgroup analyses were done on OpenMEE (Wallace et al., 2017), while publication bias analyses were done using the metafor package (Viechtbauer, 2010) in RStudio (RStudio, 2012).

3. Results

3.1. Database and datasets

Studies and effect sizes included in the database were not equally distributed worldwide, but originated mostly from Europe (7 studies and 47 effect sizes) and North America (6 studies and 33 effect sizes), followed by Oceania and South America (each with 2 studies, and 26 and 22 effect sizes, respectively), and Asia (1 study and 4 effect sizes) (Fig. 1). Of the 132 effect sizes, 88 were based on total macro-invertebrates (total macroinvertebrate dataset) and 44 were based on FFG (FFG dataset) (Table S3).

For the total macroinvertebrate dataset, 85 effect sizes contrasted the macroinvertebrate colonization of macroplastic litter with that of leaf litter and 3 with that of wood. Effect sizes contrasting the macro-invertebrate colonization of macroplastic litter with that of leaf litter focused mostly on abundance (40) and richness (27), and less on density (6), diversity (6), biomass (4), and evenness (2) (Table S3). Plastic types most often used were PCV (20 effect sizes), PET (20), and LDPE (16), while PP (8), a mixture of HDPE, PS and PET (8), nylon (4), and PLA (1) were less often used. Effects sizes based on leaf litter were reasonably distributed across mesh opening categories (1–5 cm: 13, 6–10 cm: 32, and >10 cm: 40). Leaf litter (25 species) considered in the total macroinvertebrate dataset were mostly derived from non-N-fixing than from N-fixing species (60 vs. 23 effect sizes), and mostly from deciduous (56) than from evergreen (27) or semi-deciduous (2) species (Table S3).

The FFG dataset was based entirely on contrasts between the FFG colonization of macroplastic litter with that of leaf litter, with most effect sizes being derived from shredders (23), followed by collectors (13), and less from grazers and predators (4 effect sizes each); abundance was the most often used metric (29 effect sizes), with biomass (8) and richness (7) being used less often (Table S3). The plastic type most often used was LDPE (18 effect sizes), followed by PP (10), nylon (6), PET (2), and PLA (1). Most effect sizes were derived from coarse mesh bags with small openings (1–5 cm: 28) or moderate openings (6–10 cm: 24), and less from bags with large openings or litter packs (>10 cm: 2). Leaf litter (9 species) considered in the FFG dataset were mostly derived from non-N-fixing than from N-fixing species (27 vs. 7 effect sizes), and mostly from deciduous than from evergreen species (30 vs. 14 effect sizes) (Table S3).

3.2. Overall macroinvertebrate colonization of macroplastic litter relative to plant litter

The macroinvertebrate colonization was overall significantly lower on macroplastic litter relative to plant litter. This was visible when considering the total macroinvertebrate dataset (R: 0.62, 95% CI: 0.54-0.71; Fig. 2) and the FFG dataset (R: 0.57; 95% CI: 0.48-0.67; Fig. 3) (Table 2). Datasets were not strongly affected by publication bias since funnel plots deviated from symmetry slightly (Fig. S3) and the new overall effect sizes (including missing effects sizes added by the trim and fill method) overlapped those based on the original datasets. Also, the Rosenberg's fail-safe numbers were above (9× and 8× for the total macroinvertebrate dataset and the FFG dataset, respectively) the threshold for considering the datasets robust to publication bias (Table 2). The contribution of between-study variance to total heterogeneity (I^2) was high (97% and 77% in the total macroinvertebrate dataset and the FFG dataset, respectively), indicating that the difference in the macroinvertebrate colonization of macroplastic litter relative to plant litter depends on study characteristics (moderators).

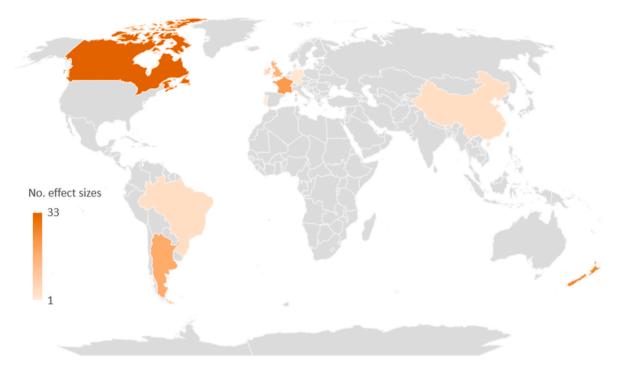


Fig. 1. Global distribution of the 132 effects sizes included in the database, derived from 18 studies published between 1992 and 2019. Please refer to the Web version of this article for the colour scale.

3.3. Effects of moderators on the macroinvertebrate colonization of macroplastic litter relative to plant litter

The difference in total macroinvertebrate colonization of macroplastic litter relative to plant litter did not depend on plant litter type (Table 2), although it was significantly lower on macroplastic litter relative to leaf litter (R: 0.61), but not relative to wood litter (Fig. 2). Macroinvertebrate colonization of macroplastic litter relative to leaf litter did not significantly differ among FFGs (Table 2), although it was significantly lower on macroplastic litter relative to leaf litter for shredders, collectors and predators (R: 0.52–0.62), but not for grazers (Fig. 3).

The macroinvertebrate metric was a significant moderator when considering the total macroinvertebrate dataset (Table 2), with significantly lower macroinvertebrate density, biomass, abundance, and richness (R: 0.31–0.75), but significantly higher diversity (R: 1.94), on macroplastic litter relative to leaf litter (Fig. 2). The macroinvertebrate metric was also a significant moderator when considering colonization by shredders and by collectors (Table 2), with significantly stronger reduction in shredder abundance (R: 0.42) than in shredder richness (R: 0.75), and in collector biomass (R: 0.16) than in collector abundance (R: 0.65), on macroplastic litter relative to leaf litter (Fig. 3).

Plastic type was not a significant moderator of total macroinvertebrate abundance on macroplastic litter relative to leaf litter (Table 2), with similar reduction in macroinvertebrate abundance in PET, LDPE, and PVC litter (R: 0.44–0.61) relative to leaf litter (Fig. 4). In contrast, plastic type affected total macroinvertebrate richness on macroplastic litter relative to leaf litter (Table 2), with significantly lower richness in a mixture of HDPE, PS, and PET litter (R: 0.42) than in PCV litter (R: 0.79) relative to leaf litter (Fig. 4).

When considering total macroinvertebrate abundance, colonization of macroplastic litter relative to leaf litter was not significantly affected by mesh opening, plant N-fixing capability, plant deciduousness, or plant identity (only two species considered) (Table 2), with abundance being significantly and similarly lower on macroplastic litter relative to leaf litter in all subgroups of these moderators (Fig. 5).

When considering shredder abundance, colonization of macroplastic

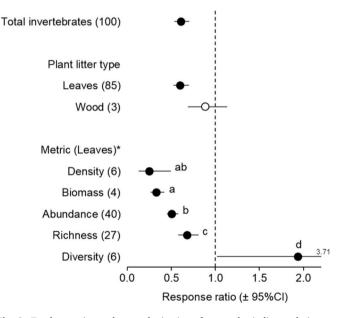


Fig. 2. Total macroinvertebrate colonization of macroplastic litter relative to plant litter in streams, and as a function of plant litter type and of metric (considering leaf litter only); values are response ratios (R; $\pm 95\%$ CI). R = 1 (dashed line) indicates no difference in macroinvertebrate colonization between macroplastic litter and plant litter, R > 1 indicates higher and R < 1 indicates lower colonization of macroplastic litter relative to plant litter. The effect is significant when the 95% CI does not include 1 (black circles). Significant moderators are indicated with an asterisks (*) and subgroups significantly differ if their 95% CI do not overlap (distinct letter). Values in brackets after subgroups indicate sample sizes (subgroups with < 3 effects sizes were not considered; see Table 2).

litter relative to leaf litter was not significantly affected by mesh opening or plant N-fixing capability (Table 2), with shredder abundance being significantly and similarly lower on macroplastic litter relative to leaf V. Ferreira

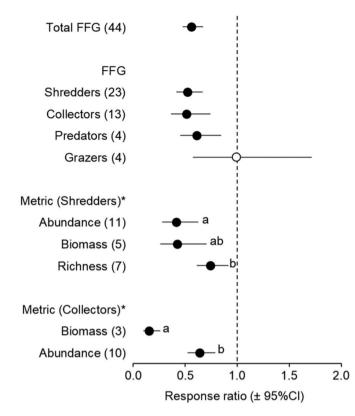


Fig. 3. Total macroinvertebrate functional feeding groups (FFG) colonization of macroplastic litter relative to leaf litter in streams, and as a function of FFG and metric (for shredders and for collectors); values are response ratios (R; \pm 95% CI). R = 1 (dashed line) indicates no difference in macroinvertebrate colonization between macroplastic litter and leaf litter, R > 1 indicates higher and R < 1 indicates lower colonization of macroplastic litter relative to leaf litter. The effect is significant when the 95% CI does not include 1 (black circles). Significant moderators are indicated with an asterisks (*) and subgroups significantly differ if their 95% CI do not overlap (distinct letter). Values in brackets after subgroups indicate sample sizes.

litter in all subgroups for each moderator (Fig. 6). When considering collector abundance, colonization of macroplastic litter relative to leaf litter was not significantly affected by mesh opening or plant deciduousness (Table 2), despite significantly lower collector abundance on macroplastic litter relative to leaf litter for the 6–10 mesh opening (R: 0.60) but not for the 1–5 mesh opening subgroup, and for the deciduous (R: 0.67) but not for the evergreen subgroup (Fig. 6).

3.4. Sensitivity analyses

In some studies, streams and stream sites were affected by factors (e. g., acidification, intermittency, urbanization) that could impact litter colonization by macroinvertebrates, besides the tested moderators. Not considering effect sizes derived from these conditions, however, would not strongly change the results reported above in terms of direction and significance as the overall effect sizes derived from the total macro-invertebrate dataset (R: 0.58, 95% CI: 0.50–0.68) and the FFG dataset (R: 0.39; 95% CI: 0.29–0.54), and the mean effect sizes for subgroups analyses (Table S5), overlapped with those found when considering the original datasets (Figs. 2–6).

4. Discussion

Macroplastics, being artificial substrates, may constitute novel habitats for freshwater biota (Windsor et al., 2019). However, few studies have specifically addressed the interaction between macroplastics and freshwater macroinvertebrates (Blettler et al., 2018). Therefore, studies focusing on plant litter decomposition in streams that compare the colonization of plant litter and plastic strips by benthic macroinvertebrates can provide relevant information about the interaction between macroplastic litter and stream macroinvertebrates. Macroplastic litter in streams was indeed colonized by benthic macroinvertebrates, but this meta-analysis showed that colonization of macroplastic litter was overall impaired between 38% (considering the total macroinvertebrate dataset) and 43% (considering the FFG dataset) compared with that of plant litter. Although impairment of macroinvertebrate colonization of macroplastics was prevalent, there were specific conditions in which macroinvertebrate colonization of macroplastic did not significantly differ from that of plant litter, or was even higher.

4.1. Lower overall macroinvertebrate colonization of macroplastic litter relative to plant litter

As hypothesized (hypothesis H1), overall macroinvertebrate colonization of macroplastic litter was lower than that of plant litter. This difference may have resulted from macroplastics being used mostly as a substrate, while plant litter can be used as a substrate but also as a food resource, especially by shredders. Caddisfly shredders have been shown to fragment plastics and use the resulting fragments in their cases under laboratory conditions (Valentine et al., 2022). In the field, however, shredders most likely collect existing micro- and mesoplastics from the stream bed to incorporate into their cases instead of spending energy chewing macroplastics into smaller pieces (Ehlers et al., 2019). The difference in macroinvertebrate colonization between macroplastic litter and plant litter could also reflect differences in physical structure (i.e., in area and volume) between these substrates. Leaf litter can be assumed to keep its three-dimensional form for some time after submersion and, therefore, to have greater surface area and volume compared with macroplastic litter. However, authors of primary studies aimed for similar area or volume between the plastic litter and the leaf litter (e.g., Dobson et al., 1992; Dangles et al., 2001; Negishi & Richardson, 2006; Márquez et al., 2017), and often report that macroplastic litter was crumpled to provide it a heterogeneous shape (Dobson et al., 1992; Dudgeon & Wu, 1999; Murphy & Giller, 2001) or that macroplastic litter pieces were arranged so that they did not cling together (Richardson, 1992; Graça & Pereira, 1995). Nevertheless, despite these measures to ensure high spatial heterogeneity of macroplastic litter, some authors indeed reported that macroplastic strips "piled upon each other" (Graça & Pereira, 1995).

4.2. Effects of moderators on the macroinvertebrate colonization of macroplastic litter relative to plant litter

The mean effect size did not significantly differ when macroinvertebrate colonization of macroplastic litter was compared with that of leaf litter and that of wood litter, but macroinvertebrate colonization of macroplastic litter was significantly impaired only when compared with that of leaf litter (by 39%), in line with what was expected (hypothesis H2). Leaf litter generally supports higher macroinvertebrate colonization than wood litter because the former substrate has higher surface area-to-mass ratio and is more flexible and palatable than the latter substrate, thus making a better habitat/refuge and food resource for macroinvertebrates (Arroita et al., 2012). This difference in macroinvertebrate colonization between leaf litter and wood litter may explain the higher difference in macroinvertebrate colonization between macroplastic litter and leaf litter than between macroplastic litter and wood litter. Still, the number of effect sizes contrasting colonization of macroplastic litter and wood litter was low (N = 3), and therefore results need to be considered carefully.

Macroinvertebrate colonization was lower on macroplastic litter relative to leaf litter (by 38%–48%) for most functional feeding groups

Table 2

Datasets, moderators, and subgroups tested in the analyses (subgroups with <3 effects sizes were not considered; see footnotes), sample size of the dataset (N) and Rosenberg's fail-safe number (N_{fs}; a dataset is robust to publication bias if N_{fs} > 5 × N + 10), test of heterogeneity between subgroups (Q_M), degrees of freedom (df), and p-values testing the hypothesis that Q_M > df (subgroups significantly differ if p-value <0.050, in bold). The figures (Fig.) showing the results are also indicated.

Datasets	Moderators	Subgroups	Total N	Rosenberg N _{fs}	Q _M	df	p-value	Fig.
Total macroinvertebrates	-	-	88	4024	6916.653	87	< 0.0001	2
Total macroinvertebrates	Plant litter type	2: Leaves \times Wood	88	4024	1.111	1	0.292	2
Total macroinvertebrates, Leaves ¹	Metric	5: Density \times Biomass \times Abundance \times Richness \times Diversity	83	4554	63.112	4	< 0.001	2
Total FFG	-	-	44	1895	153.929	43	< 0.0001	3
Total FFG	FFG	4: Shredders \times Collectors \times Predators \times Grazers	44	1895	4.230	3	0.238	3
Shredders	Metric	3: Abundance \times Biomass \times Richness	23	682	6.352	2	0.042	3
Collectors	Metric	2: Abundance × Biomass	13	158	23.809	1	< 0.001	3
Total macroinvertebrate abundance, Leaves ²	Plastic type	3: PET \times LDPE \times PVC	37	3634	4.780	2	0.092	4
Total macroinvertebrate richness, Leaves ³	Plastic type	2: PVC \times mixture	14	9669	5.774	1	0.016	4
Total macroinvertebrate abundance, Leaves ²	Mesh opening	3: 1-5 \times 6-10 \times >10	37	3634	2.066	2	0.356	5
Total macroinvertebrate abundance, Leaves ^{2,4}	N-fixing	2: N-fixer \times Non-N-fixer	35	3395	2.513	1	0.113	5
Total macroinvertebrate abundance, Leaves ^{2,5}	Deciduousness	2: Deciduous \times Evergreen	36	3486	0.948	1	0.330	5
Total macroinvertebrate abundance, Leaves ^{2,6}	Plant identity	3: Acer saccharum \times Alnus rubra	19	1412	0.150	1	0.698	5
Shredder abundance ⁷	Mesh opening	2: 1-5 × 6-10	9	169		1		6
Shredder abundance ⁴	N-fixing	2: N-fixer \times Non-N-fixer	9	218	1.393	1	0.238	6
Collector abundance	Mesh opening	2: 1-5 × 6-10	10	57	0.826	1	0.363	6
Collector abundance	Deciduousness	2: Deciduous \times Evergreen	10	57	0.079	1	0.778	6

FFG, Functional feeding groups; PET, Polyethylene terephthalate; LDPE, Low-density polyethylene; PVC, Polyvinyl chloride; Mixture, PET + HDPE, High-density polyethylene + PS, polystyrene; ¹Without Metric: Evenness as N = 2; ²Without Plastic type: PP as N = 2 and Plastic type: PLA as N = 1; ³Without Plastic type: LDPE and Plastic type: PET as N = 1 each and without Plastic type: Nylon and Plastic type: PP as N = 2 each; ⁴Without N-fixing: N-fixer + Non-N-fixer as N = 2; ⁵Without Plant deciduousness functional group: Semi-deciduous as N = 1; ⁶Without many species as N < 3; ⁷Without Mesh opening: >10 as N = 2.

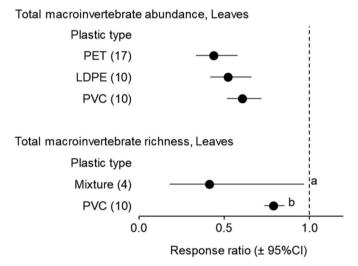
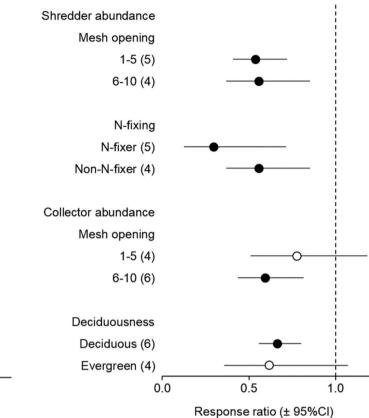


Fig. 4. Total macroinvertebrate abundance and richness on macroplastic litter relative to leaf litter in streams as a function of plastic type; values are response ratios (R; \pm 95% CI). R = 1 (dashed line) indicates no difference in macro-invertebrate abundance between macroplastic litter and leaf litter, R > 1 indicates higher and R < 1 indicates lower abundance on macroplastic litter relative to leaf litter. The effect is significant when the 95% CI does not include 1 (black circles). Significant moderators are indicated with an asterisks (*) and subgroups significantly differ if their 95% CI do not overlap (distinct letter). Values in brackets after subgroups indicate sample sizes (subgroups with < 3 effects sizes were not considered; see Table 2). PET, Polyethylene terephthalate; LDPE, Low-density polyethylene; PVC, Polyvinyl chloride; Mixture, PET + HDPE, High-density polyethylene + PS, polystyrene.

(i.e., shredders, collectors, and predators), but not for grazers, in line with hypothesis H3. Grazers may be less sensitive to the nature of the substrate provided that it allows for the development of a biofilm on which they can feed (Michler-Kozma et al., 2022). In fact, macroplastics can support a well-developed biofilm (Chaudhary et al., 2022; Vincent et al., 2022; Laffet et al., 2023) and gross primary production can be higher on hard plastic substrates than on leaf litter (Hoellein et al., 2014). Additionally, as macroplastics are a more persistent substrate than leaf litter (Kennedy & El-Sabaawi, 2018; Artru & Lecerf, 2019), they may provide a more stable substrate for biofilm development and consequently for long-term grazer colonization. For example, Gallitelli et al. (2023) found that gastropods (grazers) represented most of the invertebrates associated with PET bottles collected from an urban river in Italy. In contrast, shredders and collectors (i.e., detritivores) are more dependent on leaf litter as a food resource. While shredders feed directly on coarse organic particles (i.e., large leaf pieces), and some caddisfly shredders also use leaf litter for their cases, collectors benefit from the fine organic particles released as a result from shredders activities on the leaf litter packs (i.e., fragmentation, faecal production) (Marks, 2019). Therefore, as expected (hypothesis H3), colonization by shredders and collectors was lower on macroplastic litter than on leaf litter. Predators likely followed their prey and preferentially colonized the litter type (i. e., leaf litter) where more of them gathered.

The macroinvertebrate metric played an important role in determining the magnitude and direction of the effect size, but not as anticipated (hypothesis H4). Surprisingly, macroinvertebrate diversity was higher on macroplastic litter than on leaf litter (by 94%). The higher macroinvertebrate diversity on macroplastics may reflect higher evenness (not tested due sample size < 3) since both abundance and richness were reduced on macroplastic litter compared with leaf litter. This implies that macroplastic litter holds lower numbers of individuals and of taxa but individuals are more evenly distributed among the taxa, which deserves empirical testing. Nevertheless, sample size was low (N = 6)



Total macroinvertebrate abundance, Leaves Mesh opening 1-5 (4) 6-10 (7) >10 (26) N-fixing N-fixer (17) Non-N-fixer (18) Deciduousness Deciduous (29) Evergreen (7) Plant identity Acer saccharum (4) Alnus rubra (15) 0.5 0.0 1.0 Response ratio (± 95%CI)

Fig. 5. Total macroinvertebrate abundance on macroplastic litter relative to leaf litter in streams as a function of mesh opening, plant functional type (N-fixing and deciduousness), and plant identity; values are response ratios (R; \pm 95% CI). R = 1 (dashed line) indicates no difference in macroinvertebrate abundance between macroplastic litter and leaf litter, R > 1 indicates higher and R < 1 indicates lower abundance on macroplastic litter relative to leaf litter. The effect is significant when the 95% CI does not include 1 (black circles). There were no significant moderators. Values in brackets after subgroups indicate sample sizes (subgroups with < 3 effects sizes were not considered; see Table 2).

and was derived only from the studies by Kennedy & El-Sabaawi (2018; N = 4), which used a mixture of macroplastic litter (HDPE + PS + PET), and by Márquez et al. (2017; N = 2), which used conifer needles as leaf litter. In these scenarios, the comparison between macroplastic litter and leaf litter may have been favourable towards macroplastic litter that may have offered a more heterogeneous and stable substrate to macroinvertebrates than leaf litter. Therefore, the higher macroinvertebrate diversity associated with macroplastic litter than leaf litter needs to be considered carefully as it may occur under very particular circumstances only, i.e., when macroplastics provide a more heterogenous substrate than leaf litter. In contrast, macroinvertebrate density (-75%), biomass (- 67%), abundance (- 49%), and richness (- 31%) were lower on macroplastic litter than on leaf litter, but differences between litter types were weaker for richness, which contrasts with our hypothesis (hypothesis H4). Maybe, differences between macroplastic litter and leaf litter can be attributed to differences in substrate heterogeneity. In fact, research carried out in different contexts have shown higher biological colonization of more than less heterogenous substrates due to more heterogenous substrates, e.g., supporting higher niche diversity that can sustain higher species richness, having greater surface area for microbial and macroinvertebrate colonization, decreasing the interaction rate between predator and prey (Kovalenko et al., 2012).

Contrary to hypothesized (hypothesis H5), plastic type did not affect macroinvertebrate abundance on macroplastic litter (PET, LDPE or PVC) relative to leaf litter. Although PVC leachates are generally more toxic

Fig. 6. Shredder and collector abundance on macroplastic litter relative to leaf litter in streams as a function of mesh opening and plant functional type (N-fixing for shredders and deciduousness for collectors); values are response ratios (R; \pm 95% CI). R = 1 (dashed line) indicates no difference in abundance between macroplastic litter and leaf litter, R > 1 indicates higher and R < 1 indicates lower abundance on macroplastic litter relative to leaf litter. The effect is significant when the 95% CI does not include 1 (black circles). There are no significant moderators, but different subgroups within several moderators are distinctly significant (see the text for details). Values in brackets after subgroups indicate sample sizes (subgroups with < 3 effects sizes were not considered; see Table 2).

that leachates from other plastic types to aquatic organisms under laboratory conditions (Lithner et al., 2012; Li et al., 2016; Capolupo et al., 2020), in the stream (i.e., flowing water) leachates probably have a weaker effect on macroinvertebrates. However, the difference in macroinvertebrate richness between macroplastic litter and leaf litter was significantly higher when considering the mixture of HDPE + PS + PET (Kennedy & El-Sabaawi, 2018; impairment by 58%) than when considering PVC (impairment by 21%), but sample size was low (N (mixture) = 4) and variation was high (95% CI: 0.18–0.97) to allow strong conclusions about effects of plastic type on macroinvertebrate richness.

Stronger differences in macroinvertebrate abundance between macroplastic litter and leaf litter were expected when considering larger than smaller mesh openings (hypothesis H6), but the difference among mesh openings was non-significant. As most studies were carried out in temperate regions, where plant litter macroconsumers (e.g., decapods) are rare, mesh opening (minimum of 3 mm) may not have been a factor limiting macroinvertebrate access to litter.

Plant N-fixing capability and deciduousness also did not significantly affect the difference in macroinvertebrate abundance between macroplastic litter and leaf litter, contrary to expectations (hypotheses H7 and H8). This suggests that these plant traits are not important factors determining macroinvertebrates colonization of macroplastic litter and that macroinvertebrate abundance could be similarly reduced on macroplastics compared with leaf litter in streams flowing through different types of riparian vegetations (e.g., conifer forest, broadleaf forest).

Palatable leaf litter was expected to attract more macroinvertebrates than recalcitrant leaf litter, as the former could act as both a substrate and a food resource while the latter would primarily act as a substrate, especially at early decomposition stages (hypothesis H9) (Abelho, 2008; Ferreira et al., 2012; Monroy et al., 2016). However, the plant species contrasted following the hierarchical approach, i.e., *Alnus rubra* (red alder) and *Acer saccharum* (sugar maple), produce palatable leaf litter (Triska & Sedell, 1976; Ostrofsky, 1997), which explains that macro-invertebrate abundance was similarly lower on macroplastic litter than on leaf litter from either plant species (– 59% and – 62%, respectively).

5. Conclusion

- (1) This meta-analysis contributed to the assessment of the interactions between macroplastics and freshwater macro-invertebrates by reanalysing data derived from plant litter decomposition studies, which allowed comparison of macro-plastic colonization by stream benthic macroinvertebrates relative to that of plant litter. This revisited data indeed showed that stream macroinvertebrates interact with macroplastics, but colonization of macroplastic litter is overall impaired compared with that of plant litter. Although macroplastics may contribute to increased stream environmental heterogeneity and provide additional substrate for biofilm development (Wilson et al., 2021; Michler-Kozma et al., 2022), their low nutritional value makes it a non-useful food resource for shredders, which are especially abundant in streams (Boyero et al., 2021).
- (2) This meta-analysis also suggested that different macroinvertebrate metrics should be used to assess macroinvertebrate interaction with macroplastic litter since different metrics may respond with different magnitudes, and even in opposite directions. In particular, it suggested that assessing the components of macroinvertebrate diversity (i.e., abundance, richness, and evenness) may be more useful than assessing diversity *per se* in shedding light on the interactions between macroinvertebrate and macroplastic litter.
- (3) Additionally, this meta-analysis suggested specific conditions in which colonization of macroplastic litter by stream benthic macroinvertebrates is similar to that of plant litter, generally when the contrasting plant litter is a poor food resource (e.g., wood) or when the macroinvertebrates feed on biofilm (i.e., grazers).
- (4) However, most of the evidence reported by this meta-analysis is review-generated evidence, it derived from variation across primary studies that did not themselves test the hypotheses of interest (Table 1), as opposed to study-generated evidence that would be derived from primary studies addressing the hypotheses of interest, and therefore needs to be confirmed by empirical research.
- (5) Moreover, the studies included in this review had a biased geographical distribution that should be overcome in future studies. In particular, empirical studies should cover regions most strongly affected by plastic pollution; for example, the 10 rivers contributing the highest annual plastic outflows to the ocean are located in Asia (8 out of 10), South America (the Amazon) and Africa (the Congo) (Mai et al., 2020).
- (6) Also, while the studies included in this review used relatively small pieces of macroplastics to mimic the size of plant litter, future studies should consider the type and form of the macroplastics found in streams (Liro et al., 2022), as well as their mixture (Kennedy & El-Sabaawi, 2018), to more realistically evaluate the interaction between macroplastics and stream macroinvertebrates.

- (7) Primary studies addressing litter decomposition also used new macroplastics (which had not previously been used for any other purpose), which were incubated in streams for a relatively short period of time (maximum of 14–160 days across studies), while macroplastics in the environment are generally exposed to terrestrial conditions (e.g., retained by the riparian vegetation; Honorato-Zimmer et al., 2021; Cesarini & Scalici, 2022), and endure abrasion, photodegradation, and other forms of deterioration, before entering the streams. Therefore, it is important that future studies on the interaction between macroplastics and stream macroinvertebrates consider the degree of plastic degradation as a variable of interest. Indeed, a recent study found a positive correlation between taxon abundance and the degree of plastic bottle degradation in an urban river (Gallitelli et al., 2023).
- (8) Future studies should also consider the interaction between macroinvertebrates and other types of anthropogenic litter (e.g., paper, metal, glass) that are retained in stream banks and channels (Honorato-Zimmer et al., 2021; Poletti & Landberg, 2021; Wilson et al., 2021; Cesarini & Scalici, 2022). Similarly, comparisons with natural substrates should consider mineral substrates (e.g., rocks, sand) in addition to plant litter. Consideration of the variety of anthropogenic and natural substrates present in streams is important because differences in macroinvertebrate colonization between substrates may relate to substrate characteristics (e.g., heterogeneity, flexibility). For example, a recent study found that macroinvertebrate communities differed among different types of anthropogenic litter and between these and rocky substrates in an urban stream, and that macroinvertebrate diversity (but not density) was higher on anthropogenic litter (all types combined) than on rocky substrates (Wilson et al., 2021).
- (9) Finally, while the available studies allowed the assessment of macroplastic litter colonization by stream macroinvertebrates relative to that of plant litter, future studies should also assess how macroplastic presence may affect macroinvertebrate interactions with plant litter. For example, if grazers are using macroplastic litter as new feeding grounds, they may reduce their grazing activity on plant litter with negative consequences for plant litter decomposition (Schaller, 2013; Xiang et al., 2019). Although the amount of macroplastic litter in forest streams should remain low compared with that of plant litter, urban streams can accumulate large amounts of macroplastic litter that could compete with plant litter as feeding grounds for grazers (Hoellein et al., 2014; Poletti & Landberg, 2021).

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CRediT authorship contribution statement

Verónica Ferreira: Writing - review & editing, Writing - original draft, Visualization, Resources, Project administration, Methodology, Investigation, Funding acquisition, Formal analysis, Data curation, Conceptualization.

Declaration of competing interest

The author declares that she has no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

The matix used in the meta-analysis is made available in the supplementary material.

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Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.envpol.2023.123108.

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