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## Multiple-stressor effects of sediment, phosphorus and nitrogen on stream macroinvertebrate communities



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### HIGHLIGHTS

- Effect of multiple stressors on invertebrate communities is still poorly understood.
- Sediment, phosphorus and nitrogen were manipulated simultaneously.
- Sediment was the most pervasive stressor particularly at high cover levels.
- Multiple stressors interacted in complex ways for several invertebrate metrics.
- Improving river ecological quality requires improved management of sediment inputs.

### article info abstract

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### GRAPHICAL ABSTRACT



Multiple stressors affect stream ecosystems worldwide and their interactions are of particular concern, with gaps existing in understanding stressor impacts on stream communities. Addressing these knowledge gaps will aid in targeting and designing of appropriate mitigation measures. In this study, the agricultural stressors fine sediment (ambient, low, medium, high), phosphorus (ambient, enriched) and nitrogen (ambient, enriched) were manipulated simultaneously in 64 streamside mesocosms to determine their individual and combined effects on the macroinvertebrate community (benthos and drift). Stressor levels were chosen to reflect those typically observed in European agricultural streams. A 21-day colonisation period was followed by a 14-day manipulative period. Results indicate that added sediment had the most pervasive effects, significantly reducing total macroinvertebrate abundance, total EPT abundance and abundances of three common EPT taxa. The greatest effect was at high sediment cover (90%), with decreasing negative impacts at medium (50%) and low (30%) covers. Added sediment also led to higher drift propensities for nine of the twelve drift variables. The effects of nitrogen and phosphorus were relatively weak compared to sediment. Several complex and unpredictable 2-way or 3 way interactions among stressors were observed. While sediment addition generally reduced total abundance at high levels, this decrease was amplified by P enrichment at low sediment, whereas the opposite effect occurred at medium sediment and little effect at high sediment. These results have direct implications for water management as they highlight the importance of managing sediment inputs while also considering the complex interactions which can occur between sediment and nutrient stressors.

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### 1. Introduction

Anthropogenic activities have had a profound effect on ecosystems worldwide with activities such as urbanisation, forestry and agriculture contributing to the degradation of surface waters throughout the world [\(Sala et al., 2000;](#page--1-0) [Dudgeon et al., 2006\)](#page--1-0). Agriculture has been identified as a predominant driver of ecological change in both terrestrial and freshwater ecosystems [\(MEA, 2005\)](#page--1-0). Pollution from agriculture often represents the main contributor to total pollutant loads in river systems [\(Heathwaite et al., 2005;](#page--1-0) [Stoate et al., 2009](#page--1-0)).

The two most prevalent stressors arising from agricultural activities are deposited fine sediment ([Jones et al., 2012](#page--1-0); [Collins et al., 2018\)](#page--1-0) and elevated nutrient concentrations, namely phosphorus (P) [\(Jordan et al.,](#page--1-0) [2007;](#page--1-0) [Torrent et al., 2007](#page--1-0); [Jarvie et al., 2008;](#page--1-0) [Jarvie et al., 2010\)](#page--1-0) and nitrogen (N) [\(Carpenter et al., 1998](#page--1-0); [Stark and Richards, 2008;](#page--1-0) [Dupas](#page--1-0) [et al., 2015](#page--1-0); [Nhiwatiwa et al., 2017;](#page--1-0) [Gonzales-Inca et al., 2018](#page--1-0); [Zhang](#page--1-0) [et al., 2018\)](#page--1-0). The loss of nutrients from agricultural systems to surface and groundwater receptors has been highlighted as one of the main threats to water quality in the European Union (EU), where it is estimated that agriculture in the EU contributes over 40% of the N and 20–40% of the P entering surface waters [\(OECD, 2012\)](#page--1-0).

The individual impacts of nutrient enrichment and elevated inputs of deposited fine sediment on macroinvertebrate communities are well documented (e.g. [Kreutzweiser et al., 2005](#page--1-0); [Matthaei et al., 2006](#page--1-0); [Wang et al., 2007](#page--1-0); [Jones et al., 2012](#page--1-0)). Elevated nutrient levels can lead to eutrophication which can have indirect impacts on macroinvertebrate communities through degradation of habitat structure and reduced oxygen levels ([Correll, 1998;](#page--1-0) [Smith et al., 1999](#page--1-0); [Cook et al.,](#page--1-0) [2018\)](#page--1-0). At very high concentrations, nutrients have also been shown to have direct toxic effects on aquatic macroinvertebrates [\(Camargo](#page--1-0) [et al., 2005](#page--1-0); [Dalu et al., 2017;](#page--1-0) [Graeber et al., 2017;](#page--1-0) [Zhang et al., 2018](#page--1-0)). Agricultural practices which increase catchment erosion can lead to the deposition of fine sediment in streams [\(Jones et al., 2012\)](#page--1-0). This has been shown to clog interstitial spaces ([Soulsby et al., 2001\)](#page--1-0) and reduce habitat heterogeneity [\(Wood and Armitage, 1997\)](#page--1-0), which can adversely affect crevice-dwelling macroinvertebrates and gravelspawning fish and lead to reduced oxygen availability [\(Pretty et al.,](#page--1-0) [2006](#page--1-0); [Jones et al., 2012\)](#page--1-0). Sediment can also cause burial of biota [\(Wood and Armitage, 1997;](#page--1-0) [Wood et al., 2005;](#page--1-0) [Conroy et al., 2017](#page--1-0)), smother the gills of organisms or clog organs associated with filter feeding [\(Jones et al., 2012\)](#page--1-0). All of these can lead to increases in the propensity of stream organisms to drift ([Larsen and Ormerod, 2010;](#page--1-0) [Conroy](#page--1-0) [et al., 2016](#page--1-0)).

Of particular concern is the fact that stressors rarely act in isolation and co-occurrence of stressors in freshwaters is common ([Ormerod](#page--1-0) [et al., 2010](#page--1-0)). Furthermore, interactions are complex and can be additive, where the response is equal to the sum of the effects of both stressors; synergistic, where the response is greater than the sum of the individual stressors; or antagonistic, where the response is less than the sum of the individual stressors ([Folt et al., 1999\)](#page--1-0). These complex interactions present a challenge for water resource managers to target appropriate and effective measures [\(Dudgeon et al., 2006;](#page--1-0) [Palmer et al., 2009](#page--1-0)).

In recent years, the effects of multiple anthropogenic stressors on stream ecosystems have been investigated in experiments conducted at three different spatial scales (e.g. [Townsend et al., 2008;](#page--1-0) [Matthaei](#page--1-0) [et al., 2010](#page--1-0); [Wagenhoff et al., 2012;](#page--1-0) [Piggott et al., 2015;](#page--1-0) [Beermann](#page--1-0) [et al., 2018](#page--1-0)). All these studies manipulated nutrients (P and N combined) and fine sediment and generally found sediment to be the most pervasive stressor, with nutrient enrichment usually worsening the negative effects of sediment.

The present experiment sought to advance knowledge on the effects of these two key agricultural stressors by including nitrogen and phosphorus as both separate and combined stressors together with fine sediment in a study focusing on stream macroinvertebrate communities. To our knowledge, this is the first experiment in Europe to manipulate N, P and fine sediment simultaneously, and to include a gradient of sediment cover. Based on the related previous studies cited above, we hypothesised that 1) deposited fine sediment will have the most pervasive effects and will result in increasingly negative effects at increasing sediment cover, 2) N and P enrichment will have less pervasive but mainly negative effects due to the high concentrations applied, and 3) effects of N and P enrichment will become increasingly negative when applied in combination with sediment, with the greatest cumulative negative impact at the highest sediment cover.

### 2. Materials and methods

### 2.1. Study site and experimental system

The experiment was undertaken adjacent to the Kildavin River, a first-order stream in County Wexford, Republic of Ireland (N 52° 17′ 24.3″, W 06° 31′02.1″, 22 m a.s.l.). The Kildavin is a relatively natural first-order stream and is typical of small streams in Ireland which are mildly impacted by agriculture. The upper reaches of the Kildavin drain mainly conifer forests and the predominant land use in the area where the experiment was conducted was grassland. Background nutrient concentrations in the Kildavin (measured twice, once before and once during the experiment) were 0.01  $\pm$  0.002 mg L<sup>-1</sup> for dissolved reactive phosphorus (DRP) and  $1.07 \pm 0.012$  mg L<sup>-1</sup> for dissolved inorganic nitrogen (DIN). These concentrations were below the Water Framework Directive (WFD) threshold mean of 0.025 mg  $L^{-1}$  P for high status [\(Irish Government, 2009](#page--1-0)) and below the good status threshold of 1.8 mg L<sup>-1</sup> N but above the high status threshold of 0.9 mg L<sup>-1</sup> N (surrogate mean threshold values defined by the Irish Environmental Protection Agency (EPA) for good and high status ([EPA, 2011\)](#page--1-0)). The experiment spanned 35 days (a 21-day colonisation period followed by a 14-day manipulative period) from 8 October to 11 November 2016.

The experiment consisted of 64 mesocosms and an overview of the setup is provided in [Elbrecht et al. \(2016\)](#page--1-0) and [Beermann et al. \(2018\).](#page--1-0) The system's design was based on the ExStream System which has previously been used in New Zealand (e.g. [Piggott et al., 2015](#page--1-0)) and Germany [\(Elbrecht et al., 2016](#page--1-0)). Two Pedrollo HFm 70C water pumps (Pedrollo, Italy) were used to pump water continuously from the stream (through a 4-mm mesh filter). The water was split evenly between four header tanks, each of which supplied 16 circular mesocosms (external diameter 25 cm, inner outflow diameter 6 cm, volume 3.5 L).

Prior to the colonisation period, each mesocosm received a standard amount of representative substratum to mimic the nearby streambed. This substratum was collected from the stream below the pump intake pipe, air-dried for one week, and sieved before being added to the mesocosms. The substratum added to the mesocosms consisted of 100 mL of sand (grain size 0.5–2 mm), 400 mL of gravel (2–4 mm), 200 mL of large gravel (4–30 mm), 8 medium cobbles (30–60 mm) and 1 larger cobble ( $>60$  mm). Flow through each mesocosm was maintained at 3 L min<sup> $-1$ </sup> and calibrated daily. Water exited each mesocosm through a circular opening in its centre. A nylon net (mesh size 500 μm) was placed on this opening to capture any drifting macroinvertebrates leaving the system for a total of 48 h at the start of the manipulative period (see below). These nets were removed after the first 48 h. This allowed detecting any short-term behavioural responses to the manipulations, especially to fine sediment addition which has been shown to increase macroinvertebrate drift rates within 24 h after addition [\(Larsen and Ormerod, 2010](#page--1-0); [Beermann et al., 2018](#page--1-0)).

Stream biota (macroinvertebrates, algae and microbes) were allowed to naturally colonise the mesocosms for 21 days (i.e. colonisation period). To ensure that macroinvertebrate taxa underrepresented in the drift were also present in the mesocosms, natural colonisation was supplemented with one standard addition of macroinvertebrates. This occurred on the third- and second-last days of the colonisation period. The standardised addition to each mesocosm consisted of 1/8 of a pooled sample comprising four standard Surber samples (sampler area  $33 \times 33$  cm). Samples for seeding were collected from the Bann River,

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