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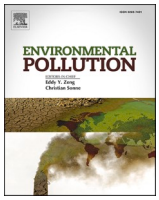

Cellulose-based wet wipes undergo limited degradation in river environments☆

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ABSTRACT

The environmental fate of cellulose-based "biodegradable" wet wipes in freshwater ecosystems remains poorly understood, despite growing market demand and legislative shifts banning plastic-containing alternatives. This study evaluated the degradation behaviour of two commercially available biodegradable wet wipe brands in upland stream mesocosms mimicking real-world river conditions. Using tensile strength loss (TSL) as the primary degradation metric, wipe degradation was compared across varied pH, temperature, nutrient, and light regimes, alongside cotton strip controls. Results revealed that although degradation rates varied by material and environmental context, both wet wipe brands persisted in river systems for 5 weeks, with Brand A degrading ~50 % faster than Brand B and nearly twice as fast as cotton controls. Degradation was significantly influenced by pH, temperature, and total dissolved solids, but not by wipe positioning in the water column (hyporheic, submerged, surface) or microbial biomass alone. Temperature-adjusted TSL (% per degree day) emerged as the most robust degradation metric, suggesting initial physical disintegration preceded microbial breakdown. These findings challenge current biodegradability claims and highlight the need for regulatory testing under environmentally relevant freshwater conditions to ensure truly biodegradable wet wipe products.

1. Introduction

Growing awareness of the environmental impacts of flushing plastic-containing wet wipes has driven the development of bio-based, biodegradable alternatives. However, despite marketing claims, many of these products persist in wastewater and river systems, contributing to environmental pollution (Joksimovic et al., 2020; Ó Briain et al., 2020; Harter et al., 2021; Choudhuri et al., 2024; Kachef, 2024; Bach et al., 2025). This persistence is often linked to low-degradable synthetic fibres within supposedly biodegradable wipes (Khan et al., 2021; Allison et al., 2023). Yet, even fully cellulosic wipes (e.g. viscose, lyocell, or cotton) can persist in aquatic environments and release large quantities of microfibres (Ó Briain et al., 2020; Kwon et al., 2022; Hadley et al., 2023; Allison et al., 2025; Bach et al., 2025). Despite growing evidence of their pollution in aquatic systems, their degradation behaviour and drivers in river environments remain poorly understood. This represents a critical gap, especially in the UK where recent legislation banning plastic-containing wet wipes has led to a rapid market shift toward cellulosic alternatives (DEFRA, 2024).

The lack of a universal, legally binding definition of biodegradability further complicates matters. Existing voluntary standards (e.g., ISO, EN, ASM) vary and typically rely on laboratory tests that fail to reflect real freshwater conditions (Kale et al., 2007; Mitchell, 2019; Zambrano et al., 2020a; Liao and Chen, 2021). Though some wipes meet composting standards like OK compost HOME and ISO 24855, these tests are typically conducted under aerobic composting conditions rather than the rivers where wipes often end up. As a result, their relevance to river biodegradability is uncertain. Clarifying this is crucial to determine whether biodegradable wet wipes truly reduce environmental impact or merely shift the form of pollution.

Cellulose biodegradation is an essential ecological process in rivers for organic matter such as leaf litter, offering insight into the behaviour of cellulosic wipes (Burdon et al., 2020; Carballeira et al., 2020). Microbes initiate and sustain breakdown by colonising surfaces and producing cellulases that hydrolyse cellulose into simpler compounds for further processing by invertebrates (Baudoin et al., 2008; Polman et al.,

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2021). Fungi (e.g., *Trichoderma*, *Penicillium*, *Fusarium* spp.) dominate early stages, while bacteria (e.g., *Bacillus*, *Pseudomonas*, *Streptomyces*, *Clostridium*) become more important as communities diversify (Burdon et al., 2020; Hayer et al., 2022).

Several environmental factors influence cellulose biodegradation in rivers. Warmer temperatures enhance microbial metabolism and enzymatic activity (Yue et al., 2016; Burdon et al., 2020; Li et al., 2023). Circumneutral pH levels optimise cellulolytic (particularly fungal) diversity and cellulase activity, with acidic or alkaline extremes inhibiting these processes (Pye et al., 2012; Li et al., 2023). Adequate oxygen and moderate nutrient levels support microbial biomass and hydrolytic activity, though nutrient extremes can destabilise communities (Tiegs et al., 2019 Chauvet et al., 2016). Light availability supports phototrophic microbial activity (Southwell et al., 2020; Blackman et al., 2024), and interactions among these environmental factors can amplify their effects on cellulose biodegradation. For example, cellulolytic degradation can be accelerated by increased nutrient enrichment and temperatures together (Burdon et al., 2020). Physical fragmentation also enhances colonisation and degradation by increasing surface area (Zambrano et al., 2020a).

Cotton strip bioassays, narrow strips of cotton textile (>95 % cellulose) tested for loss of tensile strength or mass, serve as a useful standardised proxy for studying cellulose degradation in rivers (Tiegs et al., 2009). They share compositional and microbial (e.g., communities) properties with leaf litter and respond similarly to environmental conditions such as pH, temperature, and nutrients (Slocum et al., 2009; Tiegs et al., 2009; Tiegs et al., 2013; Colas et al., 2019). These findings suggest two things: 1) cellulose-based wipes may degrade similarly to cotton strips in rivers, and 2) microbial activity is central to their environmental breakdown.

This study examined the degradation of cellulose-based 'biodegradable' wet wipes in rivers. Based on existing knowledge of cotton strip behaviour, we hypothesised that: 1) greater microbial activity, represented as epilithic microbial biomass, would increase wipe degradation; 2) physico-chemical conditions such as acidity, temperature, light, and nutrient availability, would influence degradation rates; and 3) biological and environmental interactions would shape overall outcomes. To test these hypotheses, experiments were conducted in field-controlled river mesocosms, comparing the behaviour of biodegradable wet wipes to cotton strips.

2. Methods

2.1. Study area

To assess the degradation of cellulosic wet wipes under conditions that closely mimic natural river systems, upland stream mesocosms were utilised at the Llyn Brianne Observatory, Wales, UK, between May and July 2024. Unlike microcosm-based lab experiments, which often lack hydrological complexity and ecological realism (Stewart et al., 2013), these field mesocosms provided a controlled yet dynamic environment. Crucially, these mesocosms were positioned along a natural acidification gradient, allowing for the examination of pH variation effects on degradation while maintaining controlled flow conditions. This design isolated the direct effects of key biological and environmental variables influencing wet wipe degradation within realistic river systems.

The study area, characterised extensively over a 43-year span (Weatherley and Ormerod, 1987; Durance and Durance, 2009; Pye et al., 2023), has a temperate maritime climate with stream temperatures ranging from 0 to 16 °C, a mean annual precipitation of ~1900 mm, and solar radiation levels of 7.85 MJ m^{-2} d^{-1}.

To minimise natural variability in degradation, two sets of replicate mesocosms were used (Fig. 1), each fed by first-order streams across an acid-base gradient. Two mesocosms are fed by streams draining sheep-grazed moorland catchments with typical pH levels between 6.8 and 7.2 (L6-Carpenter and L7-Davies), while two are fed by streams draining regularly logged conifer catchments with pH levels estimated between 5.3 and 5.8 (L3-Hanwell and L8-Sidaway). Each mesocosm shares physicochemical and ecological characteristics with its source stream, enabling controlled comparisons that reflect the environmental diversity of the UK uplands (Seymour et al., 2018). Each stream mesocosm consists of three channels (20 m × 0.2 m x 0.2 m) with a mixed gravel

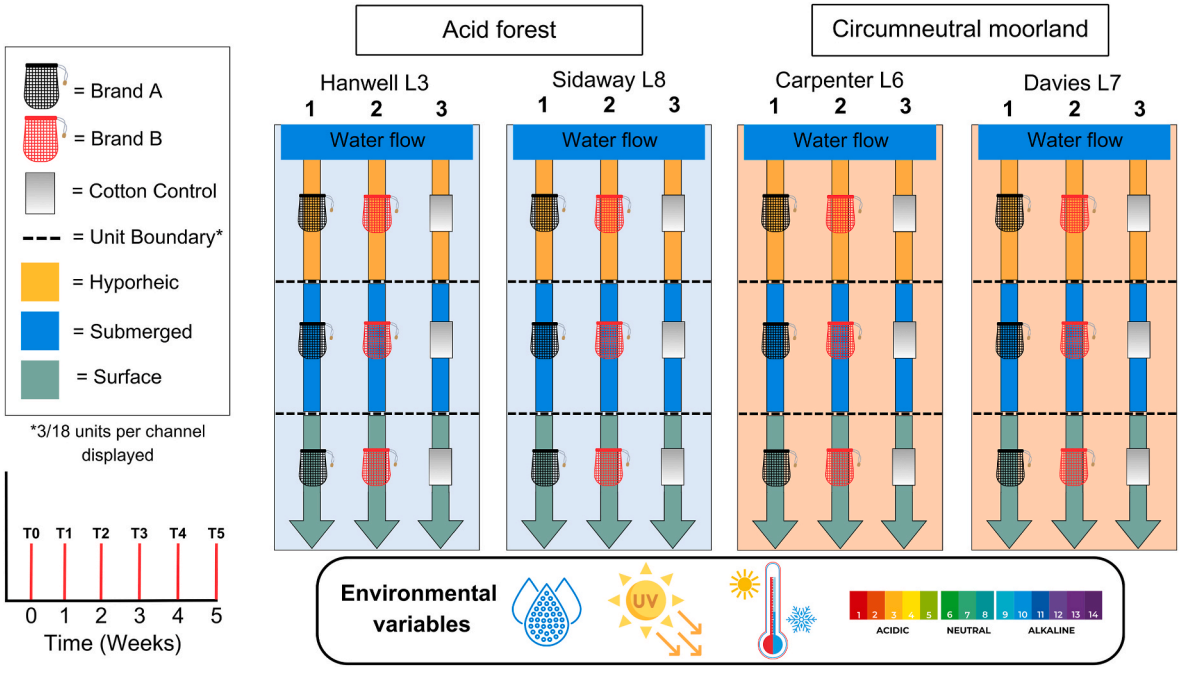

Fig. 1. Overview of the mesocosm design, including environmental variables and weekly subset collection points. Each mesocosm is represented by their associated names. Dashed lines indicate each 1 m channel unit boundary (only the first 3 units are displayed here). Channel colours (yellow, blue, green) represent the different environmental treatments in each unit. Background colours (blue and orange) signify the pH gradient of the mesocosms. (For interpretation of the references to colour in this figure legend, the reader is referred to the Web version of this article.)

and cobble substrate and water sourced directly from adjacent headwater streams.

2.2. Environmental treatments

To simulate typical conditions where improperly disposed wet wipes may end up, three treatments were introduced: hyporheic, submerged, and surface. In the hyporheic treatment, samples were embedded in the channel substrate, replicating wipes buried in riverbeds or riparian zones that experience dynamic exchanges between stream and groundwater. The submerged treatment placed samples fully below the water surface, while the surface treatment held samples just above the water, simulating wipes caught on riparian vegetation with intermittent contact to water. Simultaneously, each treatment represents a distinct ecological niche with unique environmental and hydrological conditions likely to shape microbial community composition, diversity, and functional potential (Ouyang et al., 2020; Wang et al., 2023).

Each channel consists of 18 useable units (only the first three are shown in Fig. 1 for simplicity) and is categorised by its position relative to the headwater source: proximal (channel 1), middle (2), and distal (3). Treatments were systematically applied across these units within each channel, ensuring that each treatment had six replicates distributed along the entire channel length.

To monitor environmental factors affecting degradation, temperature and light was measured hourly using automated data loggers (HOBO Pendant® Temp/Light). Two loggers per channel were positioned randomly using stratified sampling to capture spatial and treatment-based variations (24 loggers across four mesocosms), but due to malfunctions, data was excluded from two of these loggers. Throughout the study period, pH and total dissolved solids (TDS) was monitored weekly with a handheld tester (HANNA instruments® pH/EC/TDS Combo Tester, HI98129). Values were in line with the 40 years of monitoring records at those sites.

2.3. Decomposition bioassays

Wet wipes labelled as biodegradable and composed of 100 % bio-based fibres were selected from two different brands (referred to as Brand A and B) for the degradation analysis. Labelled additive categories in Brand A included moisturisers, anti-inflammatory agents, antimicrobial preservatives, antioxidants, soothing botanical extracts, and pH buffer. Brand B also contained similar ingredients, but included emollients, surfactants and emulsifiers.

Attenuated Total Reflectance-Fourier Transform Infrared (ATR-FTIR) spectroscopy confirmed the bio-based composition of these two wipe brands against cellulose reference spectra. However, decisive classification between cellulose types (e.g., natural vs regenerated) was not possible due to the inherent difficulty of spectral discrimination (Saito et al., 2021), particularly in materials containing various chemical additives. FTIR spectra, and data comparisons from Geminiani et al. (2022), seemed to indicate that Brand A was predominantly (if not completely) natural cellulose-derived, and Brand B a mixed composition of natural and regenerated cellulose. For FTIR analysis, the Happ-Genzel function in absorbance mode enhanced accuracy, and baseline correction improved interpretation of spectral peaks.

Cotton strips served as control assays, chosen for their high cellulosic content (>95 %), sensitivity to environmental conditions, as well as their convenience in assessing microbial activity (Tiegs et al., 2013). Cotton strips were prepared by cutting unprimed, 100 % natural cotton canvas (12 0z heavyweight, Discount Fabrics LTD, UK) into 8 × 2.5 cm rectangles, following established protocols (Tiegs et al., 2013; Colas et al., 2019). Strip edges were frayed (>3 mm) to prevent unravelling and handling was limited to strip edges to preserve tensile strength.

Brand A and B wet wipes and cotton controls were deployed in mesocosms. Wipes were deployed in their original sizes (Brand A = 200 × 160 mm; Brand B = 180 × 165 mm) to replicate natural freshwater pollution, but additional samples were cut to match control strip dimensions for tensile testing. This setup also allowed standardised comparisons across sample types.

Each mesocosm received 18 intact wipes and 18 cut strips from Brands A and B, distributed equally across channels 1 and 2, respectively, with only control strips in channel 3. This ensured exposure across treatments and spatial coverage, deploying a total of 72 intact wipes and 72 strips per brand, alongside 72 controls, for five weeks. Samples were placed in zip-lock mesh bags (11 × 15 cm, 100 μm mesh aperture, iQuatics, https://www.iquaticsonline.co.uk/), a standard method for isolating microbial biodegradation from invertebrate interference (Tiegs et al., 2007; Pye et al., 2012). A specific focus on microbial-driven biodegradation mechanisms in biodegradable wet wipes was allowed by this approach, while potential confounding effects from invertebrate activity were minimised.

Weekly, subsets from each channel were collected, covering all treatment and channel segments. Collected samples were sealed in zip-lock plastic bags (1 L, 24 cm × 17 cm) with 5g desiccant, transported to the lab chilled and in the dark to minimise ambient environmental degradation. In the lab, samples were gently rinsed with deionised water, dried at 40 °C for 24 h, and stored in desiccators until analysis.

2.4. Degradation measurements

Wet wipe degradation was assessed using mass loss and tensile strength loss (TSL), as these have been shown to reliably reflect microbial activity and cellulose breakdown in aquatic environments (Tiegs et al., 2019 Colas et al., 2019; Carballeira et al., 2020; Read et al., 2024). Initial wet and dry masses of reference wipes and strips (n = 20) were recorded to the nearest 0.1 mg (Ohaus Pioneer). For weekly subsets, mass loss (%) was calculated by subtracting each dry sample weight from its corresponding reference weight. Tensile strength was measured for wet wipes and control strips using a Zwick/Roell Z050 tensile testing machine with self-tightening roller grips. Strips were pulled at a fixed rate of 20 cm/min (preload = 1 N, preload speed = 5 cm/min, initial grip-to-grip separation = 11.61 cm), and each strip's maximum tensile strength was recorded.

To quantify TSL over time, the percentage of initial tensile strength lost per day of incubation was calculated, following a linear degradation model from established methods (Tiegs et al., 2013; Colas et al., 2019; Tiegs et al. 2013). TSL was determined using the formula:

$$TSL = \left[1 - \left(\frac{Tensile\ Strength_{treatment\ strips}}{Tensile\ Strength_{reference\ strips}}\right)\right] \times 100 \Big/ Incubation\ Time \quad (1)$$

where Tensile $Strength_{treatment\ strips}$ is the maximum tensile strength of field-incubated strips, Tensile $Strength_{reference\ strips}$ is the mean tensile strength of 10 non-incubated reference strips (per wipe brand and cotton controls), and incubation time is the number of days the strips were exposed in the field. To account for temperature differences, TSL was also expressed per degree-day, with incubation time replaced by cumulative degree-days (Tiegs et al., 2013). Degree-days was calculated by summing mean daily temperatures in each mesocosm channel (>0 °C).

2.5. Microbial biomass

As a recognised proxy for microbial activity, biofilm biomass samples were collected from each mesocosm channel (n = 24) at the end of the 5-week study period. Duplicate unglazed terracotta tiles (15 cm × 25 cm x 5 cm) were randomly placed along each channel to capture spatial variations. Both sides of the tiles were scraped into 50 mL Corning tubes, following established biomass sampling protocols (Steinman et al., 2006), and samples were stored on ice in the dark for transport.

Using the ash-free dry mass (AFDM) method, a robust gravimetric estimate of biomass, microbial biomass was quantified and correlated

with degradation rates to assess microbial colonisation and activity. Following Steinman et al. (2006), tile biomass samples were filtered onto pre-weighed glass microfibre filters (47 mm, VWR, UK) and dried at 80 °C for 24 h to a constant weight. The dried samples were then oxidised in a muffle furnace at 500 °C for 1 h, cooled in a desiccator, and reweighed. Based on Steinman et al. (2006), dry mass and AFDM were calculated as:

$$Dry\ Mass = \left(W_a - W_f\right) / A_t \quad (2)$$

$$AFDM = \left(W_a - W_{ash}\right) / A_t \quad (3)$$

where W_a is the dried biomass weight on filter (mg) before ashing, W_f is the filter weight (mg), W_{ash} is the post-ashing weight on filter (mg), and A_t is the tile area (cm^2).

2.6. Statistical analyses

Statistical analyses were conducted using R (version 4.3.1) (R Core Team, 2021). Metrics of mass loss (%) and tensile strength loss (% per day, % per degree day) were analysed to assess degradation in intact wipes and all strips.

Normality and homoscedasticity were verified through Shapiro-Wilk tests, Q-Q plots, and residual plots. Descriptive statistics (e.g., mean, standard deviation) were calculated with the '*psych*' package (Revelle, 2024). Data manipulation and visualisation were performed using the *tidyverse* package (Wickham et al., 2019), with plots arranged using '*gridExtra*' (Auguie and Antonov 2017), '*ggstatsplot*' (Patil, 2021), and '*patchwork*' (Pedersen, 2024). Model selection followed a backward stepwise approach using Akaike Information Criterion (AIC) to optimise parsimony and fit when required.

To test for differences in material type, treatment type, mesocosms, and time on degradation metrics, Aligned Rank Transformation (ART) from the '*ARTool*' package was used (Kay et al., 2021), with ART post hoc tests for pairwise comparisons. ART was selected over traditional ANOVA or Kruskal-Wallis tests due to its suitability for nonparametric data in factorial designs and its ability to incorporate random effects for hierarchical data.

To further examine relationships between key explanatory variables and degradation metrics correlation analyses were conducted, as were linear mixed models (GLMMs) from the '*glmmTMB*' package (Brooks et al., 2017), and generalised additive mixed models (GAMMs) from the '*mgcv*' package (Wood, 2011) to account for the hierarchical and spatio-temporal structure and complex interactions within the data. A summary of these different mixed model variables is provided (Supplementary Table 1).

GLMMs used a Tweedie distribution with a log link function to manage zero-inflated data and overdispersion, while GAMMs used a Gamma distribution and log link due to the positive skew of the degradation metrics. Scales of the environmental variables in the GLMM and GAMM models were standardised to improve model performance and interpretations. Thin-plate regression splines and Restricted Maximum Likelihood (REML) improved GAMM flexibility and performance. Model assumptions in these mixed models were validated using residual diagnostics through the '*DHARMa*' package (Hartig, 2024) and the '*gam.check*' function.

In the dataset, approximately 9 % (18 out of 202) of observations exhibited negative TSL, presenting extreme outliers. A sensitivity analysis was performed comparing GLMMs using the full dataset versus a dataset excluding these negatives. Fixed effects were consistent across models, while the model without negatives improved fit (lower AIC and dispersion). Therefore, negative TSL values were excluded in analyses to improve model accuracy.

3. Results

3.1. Mesocosm characteristics

Environmental variables varied across mesocosm sites and are summarised in Table 1. Acidic forest sites (L3, and L8) had lower mean pH values (6.12 and 5.71) compared to circumneutral moorland sites (L6, L7), which averaged 6.88 and 6.99. Total dissolved solids (TDS) were lowest at L8 (18.8 ppm) and highest at L7 (43 ppm). Biofilm biomass, measured as ash-free dry mass (AFDM), was highest on average in acidic forest sites (~0.03 mg/cm^2 for both) and lowest in L7 (0.014 mg/cm^2). Temperatures ranged from approximately 11.5 °C (L3) to 13 °C (L6), while light intensity showed considerable variability, with L8 recording the highest average (800 lux) and L3 and L7 the lowest (~180 lux). Temperature and light intensity also varied significantly between individual mesocosms channels (see Supplementary Fig. 1). Temperature differences were most notable in L3 ($F_{2,68} = 33.65$, $p < 0.001$) and L7 ($F_{2,68} = 96.99$, $p < 0.001$), particularly in Brand B (L3) and Control (L7) channels. Light variation was even more pronounced in L7 control ($F_{2,68} = 543.92$, $p < 0.001$) and L8 Brand A channels ($F_{2,68} = 88.65$, $p < 0.001$).

3.2. Material degradation

Wet wipe degradation was assessed using tensile strength loss (TSL) and mass loss as proxies. Reference tensile strength (Fmax) values were similar for both Brand A (18.03 N ± 3.35) and Brand B (17.76 N ± 0.89), while cotton strips were significantly stronger (317.21 N ± 9.27). Reference dry masses followed a similar trend, with cotton (0.746 g ± 0.026) > Brand B (0.122 g ± 0.007) > Brand A (0.094 g ± 0.01).

3.2.1. TSL metrics

TSL rates were measured using two metrics: TSL % per day (d^{-1}) and TSL % per degree day (dd^{-1}). Overall TSL rates varied widely across mesocosms, ranging from 0.02 % to 7.18 % d^{-1} and -0.0003 to 0.21 % dd^{-1} after removing extreme outliers primarily characterised by negative percentage values (tensile strength gain). TSL rates varied between material types (d^{-1}, $F_{2,174} = 22.22$, $p = < 0.001$; dd^{-1}, $F_{2,174} = 23.61$, $p < 0.001$; Fig. 2a), with Brand A degrading the fastest for both metrics (ART post-hoc contrast test; $p < 0.001$; mean ± SD: 2.03 ± 1.23 % d^{-1}; 0.09 ± 0.05 % dd^{-1}), followed by Brand B (1.55 ± 1.35 % d^{-1}; 0.07 ± 0.05 % dd^{-1}) and cotton control strips (0.96 ± 0.87 % d^{-1}; 0.05 ± 0.04 % dd^{-1}).

TSL rates differed between mesocosms (d^{-1}, $F_{3,174} = 11.43$, $p < 0.001$; dd^{-1}, $F_{3,174} = 13.47$, $p < 0.001$; Fig. 2b). Specifically, pairwise contrasts revealed that circumneutral moorland sites L6 and L7 generally had higher degradation rates than acidic forest sites L3 and L8 ($p < 0.05$) although L6 and L8 differences were not statistically significant. Overall, L7 had the fastest degradation (1.97 ± 1.22 % d^{-1}; 0.09 ± 0.05 % dd^{-1}) and L3 the slowest (1.01 ± 1.15 % d^{-1}; 0.043 ± 0.038 % dd^{-1}).

To understand wet wipe degradation behaviour in different riverine zones, three experimental treatments were introduced in the mesocosms (hyporheic, submerged, surface). However, treatments did not significantly affect TSL metrics alone or between different material types.

TSL metrics across all materials demonstrated significant temporal trends in generalised linear mixed models (GLMMs). TSL % d^{-1} declined over time ($\beta = -0.023$, $p < 0.01$), whereas TSL % dd^{-1} increased ($\beta = 0.027$, $p < 0.001$). LOESS predictions revealed non-linear temporal patterns - a rapid initial decline in TSL % d^{-1} over the first two weeks followed by stabilisation – while an inverse pattern was observed for TSL % dd^{-1} (Fig. 2c). Generalised additive mixed models (GAMMs) confirmed these non-linear findings for TSL % d^{-1} (edf = 1.9, F = 8.94, $p < 0.001$), however the effect of time became strictly linear and positively associated for TSL % dd^{-1} (edf = 1, F = 7.45, $p < 0.01$), supporting the GLMM findings.

Material-specific GAMMs helped clarify these temporal patterns

Table 1
Environmental and biological characteristics (mean ± SD) of mesocosm sites over the study period. Calculations and definitions of variables are explained in the main text.

Site	Mesocosm	pH	TDS (ppm)	Temperature (°C)	Light (Lux)	Biofilm biomass (mg/cm^2)
Acid Forest						
	L3	6.12 (0.15)	23.3 (2.16)	11.49 (1.49)	179.8 (201.73)	0.026 (0.006)
	L8	5.71 (0.1)	18.8 (1.33)	12.56 (1.70)	800.11 (879.75)	0.029 (0.009)
Circumneutral Moorland						
	L6	6.88 (0.04)	33 (3.74)	12.97 (1.64)	580.18 (382.87)	0.021 (0.002)
	L7	6.99 (0.02)	43 (3.35)	11.77 (1.28)	180.38 (218.61)	0.014 (0.004)

(Fig. 3). In Brand A, TSL % d^{-1} showed a pronounced non-linear decline (edf = 1.9, F = 14.9, $p < 0.001$), mirroring the LOESS predictions, whereas the temperature-adjusted metric declined in a linear fashion but did not reach significance. Brand B also exhibited an early non-linear drop in TSL % d^{-1} (edf = 1.9, F = 5.2, $p < 0.05$), albeit less steep than Brand A, which began to increase again over the last week. However, this shifted to a non-linear increase once temperature was accounted for (edf = 1.7, F = 3.98, $p < 0.05$). By contrast, Cotton Controls showed no significant time effect in TSL % d^{-1} yet demonstrated a modest near-linear rise in the temperature-adjusted measure (edf = 1.0, F = 9.0, $p < 0.05$).

Correlations between environmental variables and TSL metrics revealed distinct patterns. When all materials were combined, TSL rates increased only slightly with pH ($r = 0.28$, $p < 0.05$). However, material-specific analyses uncovered noticeable differences. For Brand A wet wipes, TSL increased moderately with pH ($r = 0.57$, $p < 0.05$), while temperature-adjusted TSL was also positively linked to pH ($r = 0.41$, $p < 0.05$) but also increased with temperature ($r = 0.51$, $p < 0.05$). In contrast, Brand B wipes exhibited stable degradation with no significant associations, while cotton control strips showed similar trends to Brand A – with TSL increasing with pH ($r = 0.37$, $p < 0.05$) and temperature-adjusted TSL rising with temperature ($r = 0.38$, $p < 0.05$).

Across material types, environmental variables were interrelated. For instance, light intensity increased with temperature ($r = 0.23$, $p < 0.05$) and TDS ($r = 42$, $p < 0.05$), while pH decreased with both light ($r = -0.38$, $p < 0.05$) and biomass ($r = -0.6$, $p < 0.05$). These patterns suggest that co-occurring environmental conditions may indirectly drive degradation or at least set the stage for it.

GLMMs further elucidated the linear effects of environmental factors on degradation over time while accounting for spatial-temporal clustering using a meocosm:day random interaction. In combined-material models, higher pH (d^{-1}, $\beta = 0.29$, $p < 0.001$; dd^{-1}, $\beta = 0.36$, $p < 0.001$) and TDS (d^{-1}, $\beta = 0.15$, $p < 0.05$; dd^{-1}, $\beta = 0.20$, $p < 0.001$) were robust predictors of increased degradation, whereas biomass, temperature and light showed inconsistent effects. Material-specific GLMMs revealed that Brand A was primarily responsible for these trends: its degradation rates increased significantly with pH (d^{-1}, $\beta = 0.40$, $p < 0.05$; dd^{-1}, $\beta = 0.32$, $p < 0.01$) and TDS (d^{-1}, $\beta = 0.29$, $p < 0.01$; dd^{-1}, $\beta = 0.34$, $p < 0.001$), and further increased with temperature ($\beta = 0.32$, $p < 0.001$) when TSL was adjusted for temperature. In contrast, Brand B showed no significant environmental effects on TSL % d^{-1}, while its temperature-adjusted metric greatly increased with TDS ($\beta = 1.32$ $p < 0.05$), with only borderline influences of pH and biomass. Control samples exhibited no significant environmental effects on either metric.

Non-linear relationships were also explored using GAMMs but revealed linear environmental responses (edf = 1). Thus, GAMM results corroborated the GLMM findings.

3.2.2. Mass loss metric

Mass loss (%) was also measured as an additional degradation proxy but proved unreliable due to debris accumulation. Organic matter and fine sediments adhered to both intact and strip samples, and the compact fibre structure of the wipes hindered thorough cleaning with damaging samples. As a result, some samples showed negative mass loss (i.e., mass gain), making the metric inconsistent and unsuitable for main analysis. For reference, debris accumulation was greatest on average for Brand A > Brand B > Cotton, likely reflecting differences in material porosity.

3.2.3. Intact wipe degradation

Over the 5-week study period, both Brand A and B wet wipes (full size and strips) remained intact within the acid forest mesocosms but less so in the circumneutral moorland mesocosms, where both wipe brands starting to fragment and degrade from Week 3 onwards, particularly within L7 (Fig. 4). All wet wipes in contact with river water darkened and collected organic and inorganic materials in their structure over time. The influence of attached debris may have played a role in wet wipe breakdown, particularly in terms of physical fragmentation, which allows for greater degradation from physico-chemical breakdown processes.

4. Discussion

Despite the rising demand for cellulose-based biodegradable wet wipes, their degradation in natural freshwater systems remains poorly understood. This study assessed the degradability of these materials and identified key environmental drivers. The findings help clarify whether such alternatives genuinely reduce pollution or persist similarly to plastic-containing wipes.

4.1. Degradation metrics

Assessing cellulose degradation requires reliable, ecologically relevant metrics. Prior studies using leaf litter and cotton strip bioassays have shown that both mass loss and tensile strength loss (TSL) can effectively quantify cellulose breakdown (Boulton and Boon, 1991; Tiegs et al., 2013; Griffiths and Tiegs, 2016; Tiegs et al. 2013; Blackman et al., 2024). However, in this study, mass loss proved unreliable due to sediment entrapment and organic debris accumulation, likely worsened by the porous fibre structure of wet wipes (Durukan and Karadagli, 2019; Ziklo et al., 2024). Temperature-adjusted TSL (% per degree day) offered more consistent degradation estimates across mesocosms, by controlling for temperature variation, revealing clearer and relationships with degradation drivers. This supports earlier findings that temperature-normalised metrics better capture cellulose breakdown (Mancuso et al., 2023; Blackman et al., 2024). However, TSL reflects total degradation without distinguishing between physical, biological, or other chemical processes.

4.2. Degradation across material types

Degradation rates followed the pattern Brand A > Brand B > Cotton. Brand A degraded ~50 % faster than Brand B and nearly twice as fast as cotton strips. However, despite being marketed as biodegradable, both wet wipe brands remained largely intact after five weeks, with only partial structural degradation observed – especially for Brand A in circumneutral mesocosms.

Wet wipes exhibited an initial rapid decline in raw TSL (% per day), followed by stabilisation, while cotton strips degraded steadily more slowly and steadily. The porous, non-woven structure of wet wipes likely facilitated fibre fragmentation and microbial access (Colas et al., 2019;

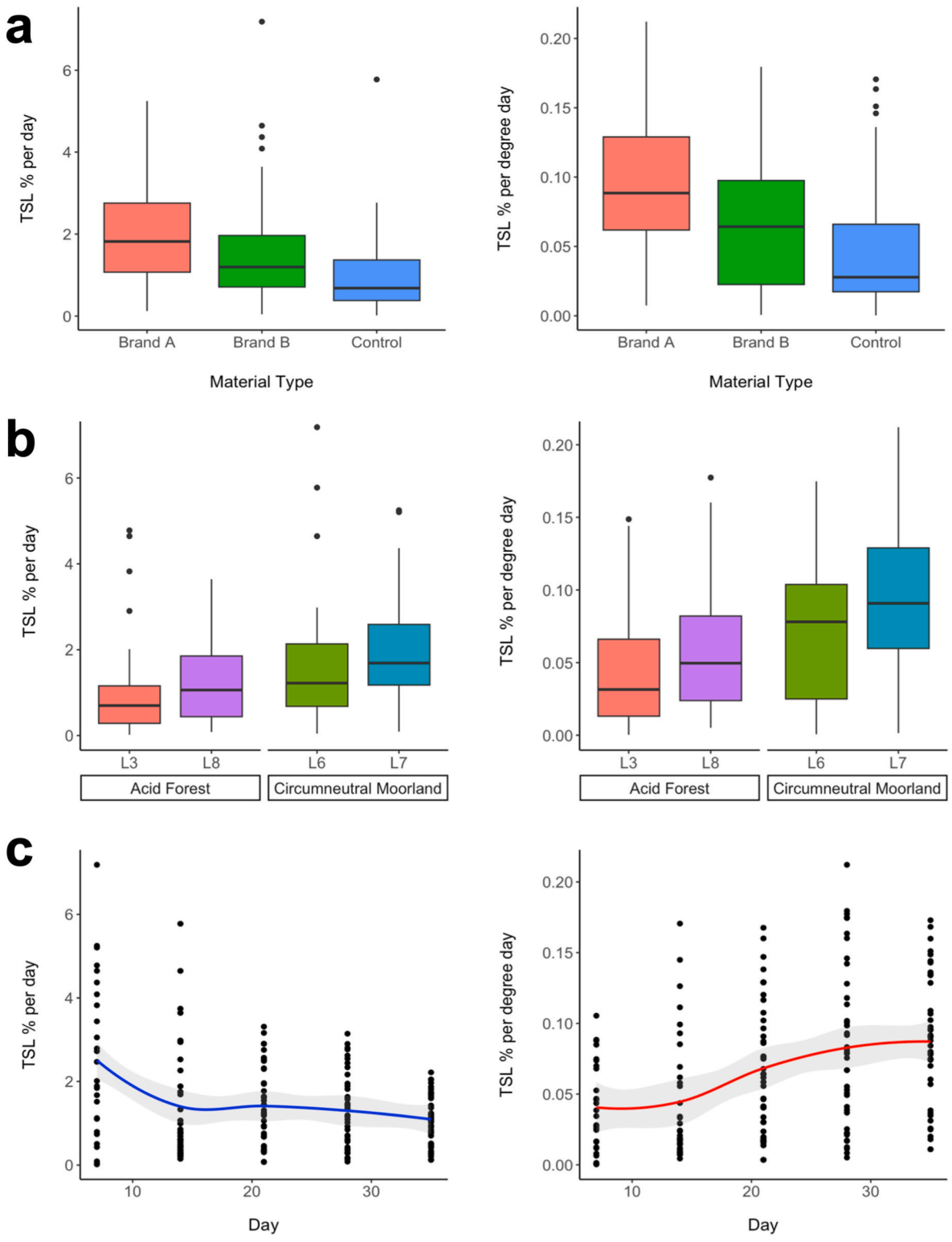

Fig. 2. TSL degradation metrics (TSL % per day and TSL % per degree day) for wet wipe and cotton control strips within the mesocosms. Boxplots of TSL metrics by a) material type and b) mesocosms, while c) shows non-linear LOESS regression trends of TSL rates over time.

Kwon et al., 2022). When adjusted for temperature, however, Brand A's degradation rate flattened, indicating that early breakdown was mainly physical. Brand B's temperature-adjusted TSL increased over time, suggesting temperature-sensitive degradation. Cotton showed minimal raw TSL loss but a steady temperature-adjusted increase, consistent with gradual microbial degradation.

FTIR results suggested that Brand A contained mostly natural cellulose, while Brand B included both natural and regenerated fibres. Material differences likely explain varying degradation patterns. Although regenerated cellulose is generally more biodegradable, natural fibres' ribbon like-structure may have enhanced microbial colonisation via increased surface area (Park et al., 2004; Zambrano et al., 2020b; You et al., 2021; Ziklo et al., 2024). Consistently, previous studies show that natural fibre-containing wipes and textiles exhibit higher TSL and

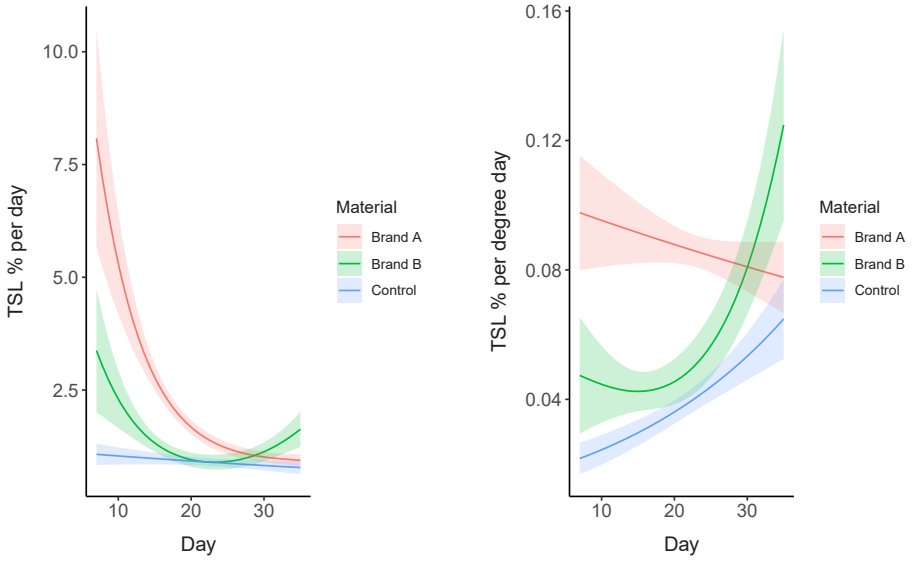

Fig. 3. GAMM predictions for material-specific degradation metrics (TSL % per day and TSL % per degree day) over time including standard errors.

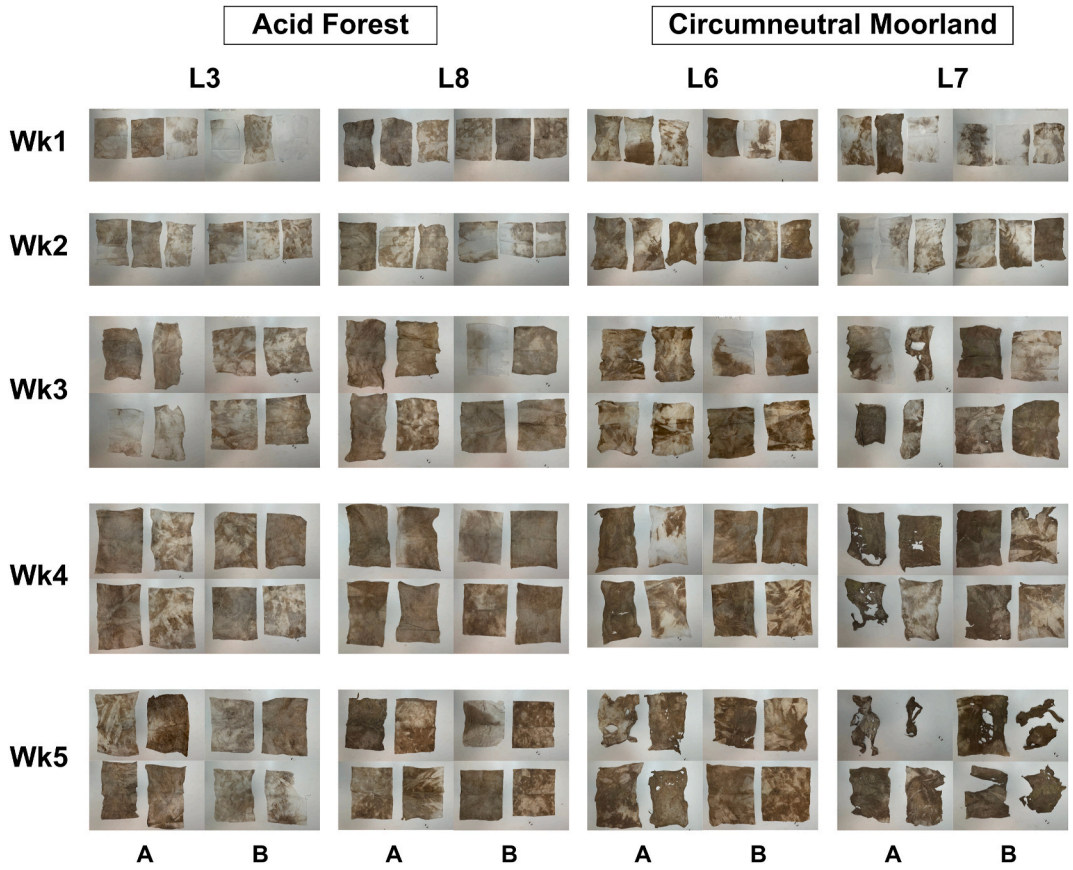

Fig. 4. Visual appearance of intact (Brand A and B) wet wipe degradation in each mesocosm across all treatment types. These intact wipe findings are provided for illustration only, but mirror patterns found for the tensile strength loss analyses of their strip-sized versions.

aquatic biodegradation rates than regenerated cellulose alternatives (Durukan and Karadagli, 2019; Zambrano et al., 2020a; Kwon et al., 2023; Smith et al., 2024).

Microfibre shedding also likely influenced degradation patterns. Natural fibres, with weaker inter-fibre bonding, irregular fibre morphology, and smaller size, tend to shed more microfibres than regenerated ones (Kwon et al., 2022; Li et al., 2022; Allison et al., 2025). This could explain Brand A's steep early raw TSL, largely independent of temperature. As labile natural fibres were lost, remaining material resisted further degradation. Brand B's gradual TSL decline suggests lower initial fibre loss and a different fibre composition, likely with more regenerated cellulose content.

Both wet wipe brands contained chemical additives such as antimicrobial preservatives (e.g., benzoic acid, phenoxyethanol, potassium

sorbate, sodium benzoate) that may have initially suppressed microbial activity(Windler et al., 2013; Tawiah et al., 2016; Malis et al., 2019). These loosely bound compounds likely leached out soon after immersion, reducing their long-term impact on degradation rates (Sait et al., 2021)). For instance, the delayed degradation of Brand B may reflect initial inhibition followed by increased microbial breakdown.

4.3. Environmental and biological drivers of degradation

Higher TSL occurred in circumneutral mesocosms (L6, L7), consistent with literature showing that acidic conditions inhibit cellulolytic microbes (Dangles and Chauvet, 2003; Dangles et al., 2004; Pye et al., 2012; Ferreira and Guérold, 2017; Colas et al., 2019). TDS was also positively correlated with degradation, suggesting greater ionic and organic availability supported microbial activity (Boulton and Quinn, 2000; Gulis et al., 2006; Ferreira and Guérold, 2017). This effect may reflect the relatively low-nutrient conditions of upland streams, as excessive nutrient enrichment – common in agricultural or urban waters - can disrupt microbial communities and inhibit degradation (Woodward et al., 2012; Colas et al., 2019).

Although temperature differences across mesocosms were small (11.5–13 °C), cumulative exposure influenced degradation over time. Brand A and cotton showed stronger temperature sensitivity, with temperature-adjusted TSL increasing with temperature, suggesting that thermal input enhanced degradation, likely via greater cellulolytic microbial availability which is known to accelerate with rising stream temperature (Griffiths and Tiegs, 2016; Yue et al., 2016). Brand B's raw TSL showed no clear correlation with temperature, but its temperature-adjusted TSL increased over time and was positively associated with TDS. This could suggest that chemical conditions, rather than temperature alone, played a greater role in its degradation. However, given the lack of a temperature effect on raw TSL, the observed increase in TSL % dd^{-1} may also reflect artefacts introduced by adjusting for temperature, rather than a true material-specific response.

The increase in temperature-adjusted TSL over time, particularly for Brand B, suggests a shift from early fibre loss to more temperature-dependent degradation processes, such as enzymatic or hydrolytic degradation. If microbial or molecular processes had driven early degradation, temperature-adjusted TSL would have initially declined rather than remain stable. Instead, as raw TSL stabilised and temperature-adjusted TSL increased, molecular degradation likely played a greater role. Temperature effects may have been more pronounced over seasonal and broader spatial gradients (Blackman et al., 2024), with a longer study duration - particularly through peak summer conditions - likely revealing stronger direct and indirect temperature-driven degradation effects (Mancuso et al., 2023).

Overall biofilm biomass, a proxy for biological activity, across sites was low (0.014–0.029 mg/cm^2 on average), reflecting the cool, nutrient-limited nature of upland streams (Anderson-Glenna et al., 2008; Zancarini et al., 2017). Biofilm biomass was higher in acid forest sites than in circumneutral moorland sites. This may reflect two known interacting mechanisms: 1) algal adaptations to acid, low-nutrient conditions, leading to their dominance in epilithic biofilm communities (Winterbourn et al., 1992); and 2) reduced invertebrate grazing pressure in acid sites, allowing greater accumulation of algal and microbial biomass compared to circumneutral sites, where invertebrate densities are typically higher (Ledger and Hildrew, 2008). Given these overall low biomass values, it is unsurprising that our AFDM-based biomass estimates did not emerge as a significant predictor of degradation. This result likely reflects the limited sensitivity of AFDM in headwater streams and does not imply that microbial activity is unimportant for cellulose degradation.

Light availability had no direct effect on degradation but likely influenced environmental conditions. Notably, light was positively correlated with temperature and varied between mesocosms, reflecting shading effects from surrounding vegetation and hillside cover, particularly in the acid forest sites. Light was also positively associated with TDS, possibly reflecting increased primary production and organic matter exudation under higher light conditions (Isles et al., 2021). While lower light availability in forested streams has been previously linked to slower decomposition rates (Mancuso et al., 2023), its effect here was likely overshadowed by stronger environmental drivers such as pH and TDS.

Cotton strip TSL rates (~0.96 % per day and 0.05 % per degree day) were generally slower than those reported in other field studies (Tiegs et al., 2013; Griffiths and Tiegs, 2016; Jabiol et al., 2020; Hill et al., 2022). Notably, Griffiths and Tiegs (2016) and Jabiol et al. (2020) conducted their assays in headwater streams, indicating that our slower degradation rates likely reflect more constrained conditions specific to the mesocosm environment. Unlike dynamic downstream or urban river systems, where higher flow, nutrient inputs, and microbial diversity enhance cellulose breakdown, these mesocosms reflected upland stream conditions, characterised by lower nutrient availability, acid waters, and lower organic matter inputs. This is reflected in the low AFDM values in this study, which indicate reduced biological activity across all sites and suggest nutrient limitation as a key constraint on biodegradation.

4.4. Mesocosm treatment effects on degradation

Three treatments – surface, submerged, and hyporheic – were tested to simulate different wet wipe accumulation zones. Microbial biodegradation is often slower in riparian zones due to reduced moisture availability (Mancuso et al., 2023), while submerged conditions can enhance abiotic hydrolysis – a molecular degradation process via moisture interactions (Allison et al., 2023). However, contrary to expectations, no significant treatment-driven differences in TSL were observed across mesocosms or material types. Several factors may explain this result:.

1. Hyporheic conditions may have closely resembled submerged ones due to shallow, permeable substrates allowing high oxygen flow and similar cellulosic degradation rates (Boulton and Quinn, 2000; Burrows et al., 2017).
2. Early degradation was likely dominated by physical fragmentation rather than microbial breakdown (Fig. 5), and a longer duration may have been required to detect microbial-driven differences across treatments.
3. Surface-exposed wipes may have remained moist enough to allow microbial activity and fibre loss similar to the other treatments (Lee et al., 2021; Kwon et al., 2022). Fibre shedding may also have been exacerbated by intermittent water contact and increased wetting and drying cycles (Li et al., 2022).

5. Conclusion

This study is the first to systematically evaluate the degradation of cellulose-based biodegradable wet wipes in freshwater systems, using replicated, near-natural river mesocosms to identify key physicochemical drivers. Our findings show that biodegradable wipes can persist in river systems for over a month, posing aquatic pollution risks. Tensile strength loss proved to be a reliable proxy for their environmental degradation. Degradation was primarily shaped by environmental conditions, with warmer, circumneutral streams and higher total dissolved solids accelerating breakdown. These results raise important considerations for product design, labelling and biodegradability standards. Real-world aquatic testing is essential to ensure biodegradable wipes degrade in the environments where they commonly accumulate, and prominent 'Do Not Flush' labelling and better consumer education designs are needed to reduce improper disposal. Early-stage fibre shedding also underscores the need to balance durability with degradation to minimise microfibre pollution.

Understanding the factors influencing degradation can support more

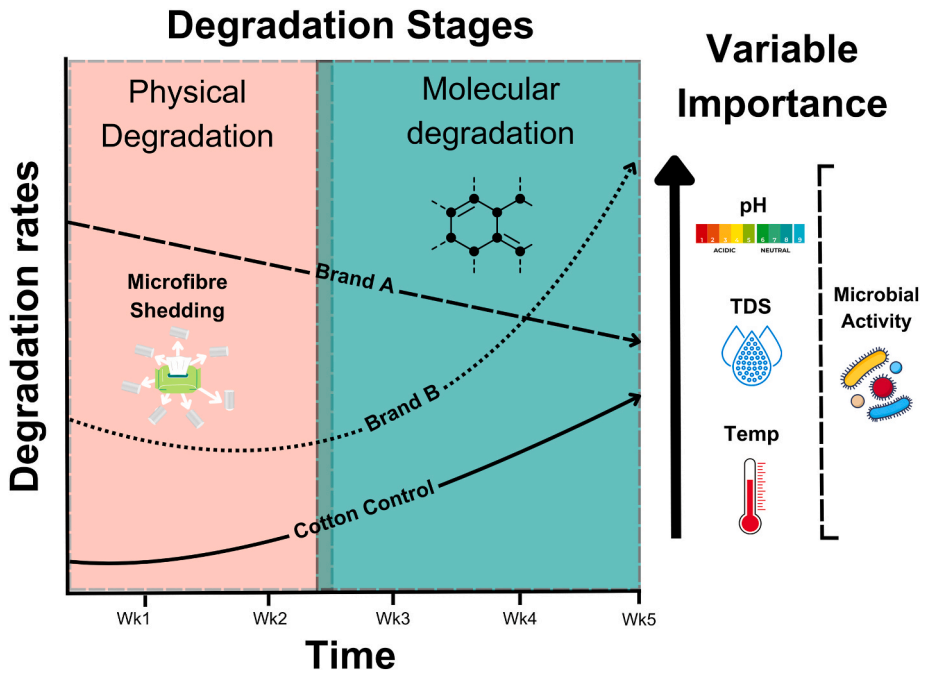

Fig. 5. Conceptual diagram highlighting observed degradation rates for each material type over time, including the likely degradation stages and the most important environmental variables influencing molecular degradation, all in which are indirectly associated with microbial availability and activity.

effective regulation, guide material development, and improve waste management strategies. Future research should extend to longer timeframes, a broader range of environmental conditions, including evaluations of biodegradation within wastewater treatment plants where flushed wipes first enter and where microbial communities and physicochemical conditions differ markedly from rivers - and integrate deeper chemical analyses to better distinguish physical from molecular degradation mechanisms.

CRediT authorship contribution statement

Thomas Allison: Writing – review & editing, Writing – original draft, Visualization, Software, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Benjamin D. Ward:** Writing – review & editing, Writing – original draft, Supervision, Methodology, Funding acquisition, Conceptualization. **Michael Harbottle:** Writing – review & editing, Writing – original draft, Supervision, Methodology, Funding acquisition, Conceptualization. **Isabelle Durance:** 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.

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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.2025.126971.

Data availability

Data used for the research described in this article are available here: https://doi.org/10.17035/cardiff.28937564.v1

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