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ECOLOGY OF LARGE WOOD IN THE UPPER MISSISSIPPI RIVER

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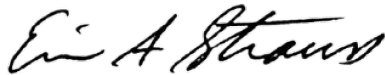
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ECOLOGY OF LARGE WOOD IN THE UPPER MISSISSIPPI RIVER

By: Matthew Chen

We recommend acceptance of this thesis in partial fulfillment of the candidate's requirements for the degree of Biology Aquatic Science Concentration.

The candidate has completed the oral defense of the thesis.



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ABSTRACT

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Wood is recognized as a key habitat structure in riverine ecosystems, yet we know relatively little about its role in large floodplain rivers such as the Upper Mississippi River. Despite its clear ecological importance, wood has actively been removed from large rivers for navigational and recreational purposes. Defining the ecological role of wood in large floodplain rivers, such as the Upper Mississippi River, is an important step towards developing better conservation, restoration, and management practices. Two differing yet interconnected objectives were examined. We surveyed shoreline wood in Pool 8 of the Upper Mississippi River quantify the subsidy and to gather insight into the mobility of wood within the pool. Evidence of wood movement in the main channel was clear, yet such evidence was less apparent in the smaller side channels. Riparian land cover was a significant factor in the accumulation of shoreline wood. Additionally, wood colonizing biofilms were sampled experimentally and from natural wood surfaces to understand how wood may directly support microbial communities. Greater biofilm biomass on wood surfaces, in comparison to inorganic surfaces, was found through in situ experimental deployments. Isotope analyses suggested direct assimilation of $\delta^{15}\text{N}$ & $\delta^{13}\text{C}$ from natural wood surfaces into their colonizing biofilms. Developing both our understanding of the availability of wood and the utilization of this resource by biofilms provides context to the importance of an allochthonous resource in the UMR.

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INTRODUCTION

Wood in Rivers

The input of terrestrial material to river systems plays an important role in their structure and function. Wood subsidies provide numerous functional and structural roles in river ecosystems. Large wood ($\geq 1\text{m}$ long and $\geq 0.1\text{m}$ in diameter) offers important ecosystem functions to rivers ranging from whole-river geomorphic control to locally providing shelter/habitat for a vast range of organisms such as fishes and macroinvertebrates (Triska 1984; Dewey & Jennings 1992; Sparks et al. 1998; Angradi et al. 2010). In small, steep, upland streams wood has significant geomorphic influence on the channel. Single downed trees can widen or narrow streams, create localized deep pools, and trap fine sediments otherwise washed downstream (Nakamura & Swanson 1993; Wohl & Merritts 2007; Swanson et al. 2020). These factors contribute to stream ecosystem function and supporting a diverse aquatic community.

In lower gradient floodplain rivers such as the Upper Mississippi River (UMR), wood is an equally critical structure (Triska 1984; Wohl et al. 2019; Swanson et al. 2020). Structure in these rivers, such as wood, can create biogeochemical hotspots by trapping large quantities of nutrient rich sediments, leading to macrophyte growth and high utilization by microorganisms such as periphyton and bacteria (Triska 1984; Nakamura & Swanson 1993). Additionally, these structures provide much needed refuge for many fishes and invertebrates in an otherwise bare benthic habitat (Dewey & Jennings 1992; Crook & Robertson 1999; Pettit et al. 2013). Wood in rivers can provide a particularly useful and abundant source of carbon to the ecosystem (Swanson et al. 2020). In floodplain rivers, organic material is often transported in irregular pulses and can

sometimes be related to seasonal shifts in precipitation and instream productivity (Vannote et al. 1980; Junk et al. 1989). Buried or permanently lodged wood in these large rivers may prove to be a significant semi-permanent source of allochthonous carbon in systems where transient carbon (DOC, FPOM, etc.) is the typical driver (Vannote et al. 1980; Triska 1984; Junk et al. 1989; Thorp & Delong 2016).

Historical Changes to Wood in Rivers

Despite its ecological importance, large wood has a long history of active removal from rivers around the world (Wohl 2014; Stout et al. 2018). In North America, wood snags (generally immovable collections of large pieces of wood) have been actively removed from river channels for navigational and recreational purposes since the early 1800s (Angradi et al. 2010; Wohl 2014). In addition, the natural flow of wood, or recruitment, into and within large rivers has been altered in many ways. Decreased supplies of new wood because of riparian forest loss and human alteration of flow/flood regimes, which historically transported quantities of wood downstream, are particularly evident (Montgomery et al. 2003; Wohl 2014; Wohl et al. 2019). For example, much of the UMR floodplain forest was removed as the region was settled and the lock and dams were constructed (Nelson et al. 1994; Johnson & Hagerty 2008; Angradi et al. 2010).

Climate change, longer inundation periods, and invasive species (e.g., reed canary grass) continue to make changes to new forest development (De Jager et al. 2019). Further land cover changes such as agricultural development and urban expansion have led to formerly forested landscapes being cleared, and thus largely eliminating the flow of wood from terrestrial ecosystems into smaller tributaries and eventually into larger river systems such as the UMR (Figure 1) (Piégay 2003; Wohl et al. 2019).

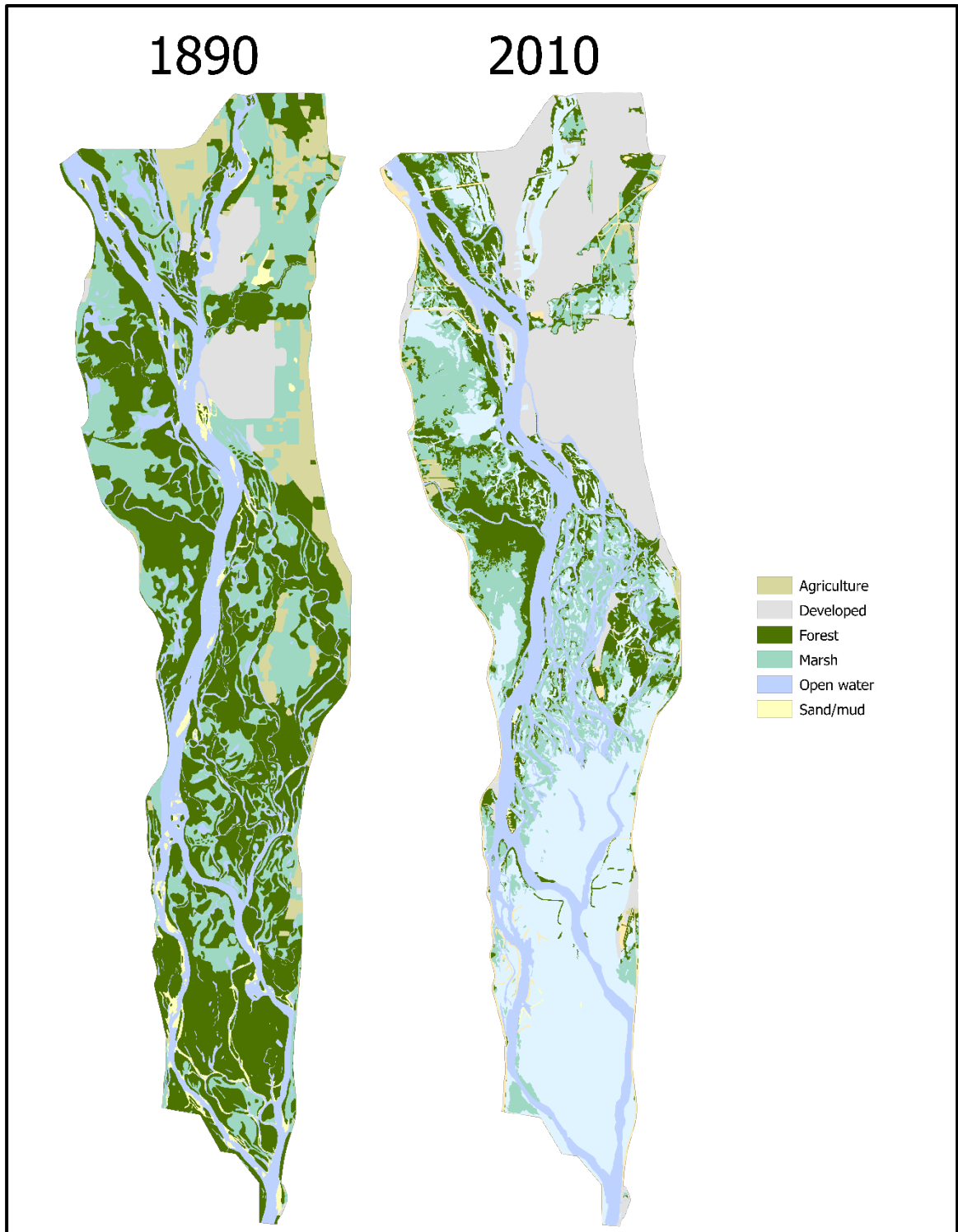


Figure 1: Land cover of UMR Pool 8 in 1890 and 2010 showing long-term changes in the abundance of forested land in the floodplain. Figure adapted from Belby et al. 2019.

Confidently defining the ecological role of wood becomes an important task with critical changes to wood loads in modern rivers (Wohl et al. 2019; Swanson et al. 2021). In recent literature, wood has been identified as a third major “regime” supporting and explaining the ecological integrity of rivers (Wohl et al. 2019). Wohl et al. (2019) explains that wood in rivers is perhaps as equally important as the flow regime and sediment regimes in terms of maintaining ecological and geomorphic diversity within river corridors. The characterization of how wood is recruited, stored, and transported within river systems is of high importance to our understanding of the function and health of a river system. Like many defining factors of rivers, high inter-system variability in the wood regimes of rivers exists around the globe, making the generalization of a wood regime difficult without robust and expansive knowledge across varied systems (Wohl & Merritts 2007; Wohl et al. 2019).

Within the UMR, the lock and dam system has significantly altered the natural state of the ecosystem (Johnson & Hagerty 2008). These alterations have likely affected the direct downstream transportation of wood and potentially disconnected individual pools from this once abundant allochthonous resource (Nelson et al. 1994; Angradi et al. 2010). Although the UMR has widespread ecological significance, our understanding of the natural state of large wood the river is comparatively small. Much of the documentation on the river began after the era of wood removal, potentially leading to modern underestimates of the historical importance of wood to the UMR and its unique habitats. Furthermore, academic research on large rivers has been historically underrepresented due to logistical constraints surrounding necessary studies to describe such large systems (Angradi et al. 2010; Wohl 2017; Swanson et al. 2021). In many

smaller rivers and streams, particularly upland streams, wood is a defining characteristic and thus much of the foundational literature on wood in rivers has been focused within these types of rivers (Nakamura & Swanson 1993; Sabater et al. 1998; Wohl et al. 2019; Swanson et al. 2021). Comparatively little is known about the importance of wood in large, lowland floodplain rivers such as the UMR.

Recruitment and Transience of Wood in the UMR

The flux of wood from the terrestrial ecosystem into the aquatic ecosystems is a complex process often taking place over long periods of time (Wohl et al. 2019). The rate of recruitment of wood is often linked to the surrounding terrestrial area and its relative connectivity to the river. As individuals or stands of trees die, they may become available to enter the river, either direct deposition or through large flood events connecting the floodplain forest to the river. In lowland floodplain rivers like the UMR, wood recruitment from the floodplain is likely driven by heavy spring flood pulses and shoreline erosion (Wohl et al. 2019). Thus, changes to wood availability (forest loss, lack of new forest growth, etc.) have likely altered the natural rates of wood recruitment in the UMR over the past century (De Jager et al. 2018; Belby et al. 2019).

The movement of wood is an important factor to consider when examining wood resources in a river. Understanding the relative availability of mobile and immobile wood is a critical factor in determining the role of wood in a river (Wohl et al. 2019). Transient wood likely offers little to the aquatic biota. However, once immobilized wood can provide many critical ecological benefits to a localized area. Changes to the channel structure in the UMR likely have altered the relative abundances of transient/immobile wood. Alterations to the depth and velocities in the main channel have likely increased

the mobility of wood, allowing for rapid translocation of large quantities of wood during high-flow periods (Johnson and Hagerty 2008, Wohl et al. 2019).

Biofilms in Rivers

Biofilms are a conglomerate of various microorganisms such as algae, bacteria, and fungi in a porous extra cellular matrix which grows on the surfaces of many hard substrates. In freshwater ecosystems, biofilms can be found on nearly every submerged surface and play an important and irreplaceable role within the ecosystem (Lock et al. 1984; Sinsabaugh et al. 1991). In streams and rivers, biofilms represent a large fraction of nutrient uptake and storage and thus play a significant role in ecosystem-scale cycling of nitrogen, phosphorus, and carbon (Lock et al. 1984; Thorp & Delong 2002; Battin et al. 2016). Uniquely, there is potential for epixylic biofilms (wood colonizing) to be a significant contributor in the cycling of organic material and may play a role in the transfer of energy within aquatic ecosystems (Tank et al. 2010). Wood in aquatic ecosystem is often considered to be a major carbon sink (Hafner et al. 2005; Swanson et al. 2020). However, it is also hypothesized that the microbial decomposition of wood surfaces can directly fuel the transfer of energy within aquatic ecosystems (Sinsabaugh et al. 1991; Sabater et al. 1998). Wood decomposition is primarily driven by microbial activity, as few invertebrate decomposers directly draw nutrients from wood (Eggert & Wallace 2007; Battin et al. 2016). However, wood may provide a long-term and consistent source of carbon to microbial decomposers in the bacterial portion of biofilms in systems and regions where in-stream wood is plentiful (Hladyz et al. 2010; Tank et al. 2010). A large proportion of research on wood in rivers is targeted at its importance as a habitat for macroorganisms such as fish and invertebrates (Swanson et al. 2020). While

this is certainly an important aspect to focus on, it is equally important to understand the mechanisms behind why these structures serve to be great habitat for larger river-dwellers.

Biofilms are an ecosystem cornerstone and a unique aspect in the upward trophic transfer of energy in aquatic food-webs. By providing a plentiful and nutritious food resource for invertebrates and other grazers, biofilms are directly linked to overall ecosystem health (Lock et al. 1984; Battin et al. 2016). By examining microbial communities' potential reliance on woody substrates in the UMR, we hope to build our basal knowledge of how energy is transferred from wood to upper trophic levels in rivers.

Many physical factors such as desiccation, substrate instability, and scouring are known to contribute to large biofilm losses, particularly following extreme disturbance such as flooding. These factors make the abundance of hard substrates a particularly important factor in lowland, floodplain rivers (Biggs 1996; Rodríguez et al. 2006). Light and nutrient availability are often one of the primary driving forces of biofilm growth in freshwater ecosystems, but a stable and abundant substrate is also necessary to support thriving microbial communities (Lock et al. 1984; Rodríguez et al. 2006). Rivers with a history of wood removal, such as the UMR, may have significantly altered the fundamental structure of the ecosystem by removing a formerly abundant substrate for biofilms. In upland rivers, cobbles and exposed bedrock are often a plentiful resource and thus a prime surface for biofilms to colonize (Sabater et al. 1998; Spänhoff & Cleven 2010). In contrast, submerged wood often serves as the primary hard substrate in lowland rivers where benthic regions are dominated by sand and soft sediments prone to frequent scouring (Atkinson et al. 2008).

We know that biofilms represent a main source of production and potentially play a vital role in the cycling of carbon in most aquatic ecosystems, subsequently serving as an important and abundant food source for many invertebrates and fish (Thorp & Delong 2002; Eggert & Wallace 2007; Battin et al. 2016). Furthermore, wood in rivers is known to be a viable substrate for biofilms yet we know relatively little about how or if wood promotes microbial communities in a way that inorganic substrates cannot. Thus, it is unclear how wood-colonizing communities factor into overall ecosystem health and function.

Biofilms develop on many types of surfaces when microorganisms colonize and develop a polysaccharide matrix (Hladysz et al. 2010; Battin et al. 2016; Howard-Parker et al. 2020). It is suggested that autotrophs are typically the initial colonizer, with heterotrophs following (Battin et al. 2016). Autotrophs can rapidly colonize surfaces, such as wood or rocks, assuming the conditions in the water column provide the necessary environment. In contrast, heterotrophic organisms such as bacteria require direct assimilation of organic materials to begin to develop. Interactions between algae and bacteria when related to biofilm development may be described by the priming effect where algae and other autotrophic microbes may act as a stimulant to the heterotrophic decomposition of recalcitrant carbon (Battin et al. 2016; Howard-Parker et al. 2020). Differences in substrate type (organic or inorganic) may play an important role in determining to what extent the priming effect occurs. On an inorganic substrate, there would be minimal recalcitrant carbon to be utilized, which would make the priming effect effectively non-existent. Easily utilized labile carbon would likely be the main source of heterotrophic production. In contrast, on a carbon-rich substrate such as wood, algal

stimulation of the heterotrophic community may unlock a previously unusable source of carbon.

In many large rivers, such as the UMR, much of the naturally occurring substrate has been altered into engineered structures such as rock and concrete. Whether these anthropogenically placed structures foster equal biofilm production as natural wood surfaces has been minimally studied in large rivers. A handful of studies have experimentally found that total biomass, algal biomass, and bacterial biomass are considerably elevated on wood surfaces than on their inorganic counterpart (Sinsabaugh et al. 1991; Sabater et al. 1998; Coe et al. 2009). These apparent differences in community structure between substrate types potentially suggests that wood actively promotes an abundant heterotrophic fraction of the biofilms and requires further study to fully capture the importance of wood for aquatic microbial communities. While it is accepted that biofilms are a critical component of riverine ecosystems, there is a lack of direct knowledge, particularly in large rivers, about the utilization of wood substrates by biofilms and the greater implications of further wood loss (Sabater et al. 1998; Wohl 2017).

OBJECTIVES

I

The first objective of this project was to examine the role of wood substrates in the development of aquatic biofilms, especially to discern differences in algal and bacterial development on wood and inorganic substrata in two UMR habitat types. We hypothesized that wood substrates would prove to be a critical resource for river biofilms and produce higher biofilm biomass compared to their inorganic counterparts.

II

The second objective of this study was to directly examine if and how wood is assimilated into biofilm biomass. The elemental and isotopic signatures of both experimental (Objective 1) and naturally occurring biofilms and their respective substrates were measured to explore trophic relationships. We expected to find evidence of direct substrate assimilation into epixylic biofilms from both the naturally occurring and experimental wood surfaces.

III

The final objective was to broadly understand the distribution of wood in Pool 8 of the Upper Mississippi River. By conducting a survey across the main and side channels we aimed to identify evidence of downstream transportation of large wood. We hypothesized that wood distributions would differ across channel size, with more wood occurring in the smaller, lower flow, channels. More wood was expected to be found on shoreline reaches with heavily forested riparian areas than in areas with development or tree-less landscapes. Additionally, we expected to find strong evidence of the movement of wood from upstream to downstream.

METHODOLOGY

Site Description

The Upper Mississippi River (UMR) is an expansive ecosystem, ranging from its origin as a small stream at the Lake Itasca outflow in northcentral MN to the confluence with the Ohio River at Cairo, IL. Within the UMR there exists a variety of aquatic habitats, all hydrologically connected yet ecologically distinct. Most distinct are the main channel (MC) and backwaters (BW). The main channel, or main navigation channel, is a

fast flowing, often deeply dredged channel maintained to allow for the passage of large shipping vessels. Portions of the MC shoreline are developed and often stabilized with large sections of rip rap or other forms of armament (Figure 1). However, much of the Pool 8 MC shoreline remains as floodplain forest (Figure 1). Backwater regions are hydrologically connected to the main channel but often experience a wider range of flow velocities and depths. Relatively shallow, backwaters often follow strong seasonal variability and are typically highly productive in warm months, often producing stands of macrophytes, floating plants, or algal blooms (Burdis & Hoxmeier 2011).

In 2022, UMR Pool 8 exhibited typical hydrologic patterns (Figure 2). High water periods correspond to spring snowmelt and early spring rains. Water levels consistently drop through the summer to a state of base flow. Summer storms can cause intermittent periods of high flow, but the effects are typically minimal.

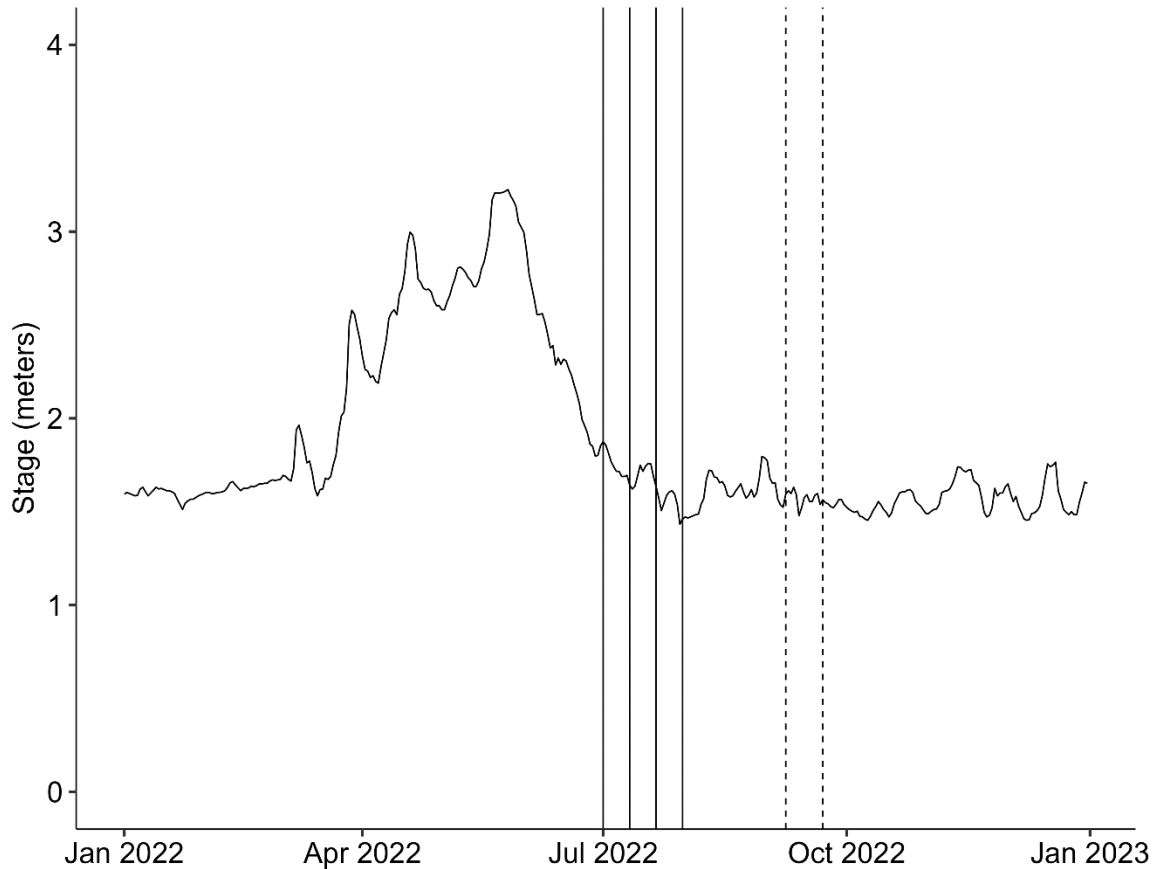


Figure 2: Mean weekly stage height (m) as measured at US Army Corps of Engineers Mississippi River at La Crosse, WI stream gage. Solid vertical lines correspond to biofilm deployment sampling dates. Dashed vertical lines correspond to the start and end of shoreline wood survey.

Experimental Biofilm Deployments

To examine differences in biofilm development across organic and inorganic substrata and habitat type, an in-situ experiment was conducted in Pool 8 of the UMR. Ten sites were randomly selected within an accessible and interconnected stretch of secondary and tertiary channels (Figure 4). Sites were generated with an R script using the packages *sf*, *raster*, and *rgdal*. Five sites were selected along the shoreline of the flowing channels and five sites were selected in backwater reaches. In the field, sites were then adjusted to the nearest downstream location to fulfill criteria to ensure stability

and homogeneity across all sites. Optimal sites were reaches with minimal canopy cover, shallow drop offs to ensure motorboat accessibility, and a minimal depth of 0.6m.

Two substrate types were used: one wood substrate and one inorganic substrate to mimic natural wood and rock substrates found in the UMR. Wood substrates were created by cutting down 0.635m x 1.22m x 2.44m natural Maple plywood sheets into small squares. Inorganic substrates were white ceramic mosaic tiles with a rough glaze. Each inorganic tile had a surface area of 11.93 cm² and each wood tile had a surface area of 9.1 cm².

Deployment structures consisted of a 0.6m x 0.6 clear acrylic plate with four 0.25m x 0.25m acrylic plates glued on with clear aquarium silicone adhesive (Figure 3). On each 0.25m x 0.25m acrylic plate, twelve wood and twelve tile substrates were adhered using the same silicone adhesive. Rebar was attached to each corner using small metal hose clamps which allowed for the structures to be anchored directly to the bed at each site 25-30cm below the surface of the water. HOBO MX Light sensors were attached to each structure and continuously recorded light intensity (lux) for the duration of the experiment (Appendix A).

On July 1st, 2022, each of the ten deployment structures was inserted at one of the ten sites. Sampling occurred at regular 10-day intervals on July 11th, July 21st, and July 31st, 2022. Field sampling was done by removing a single 0.25m x 0.25m acrylic sheet (12 wood and 12 tile substrates) from each site and placing the entire sheet into a Ziploc bag. Samples were stored in a cooler on ice until returned to the laboratory. A Hydrolab DS5 multiparameter probe was used to collect temperature, pH, dissolved oxygen (DO), and specific conductivity (SPC) measurements at each site. A HACH FH950 handheld

flow meter was used to record the velocity at the upstream end of each deployment (Table 2).

Upon return to the laboratory, biofilms were carefully scraped off each substrate using sterile razor blades. Ash free dry mass samples were scraped onto pre-combusted glass fiber filters and packaged carefully into aluminum foil packets before being placed into a drying oven. Chlorophyll *a* samples were similarly scraped onto glass fiber filters and packaged in foil packets before being stored in a freezer until further analysis.

Bacterial secondary production (BSP) samples were scraped directly into vials with 10ml of filtered site water and stored in the refrigerator for less than 5 hours before analysis.

Samples for elemental (%C & %N) and isotopic ($\delta^{15}\text{N}$ & $\delta^{13}\text{C}$) analysis were scraped into small aluminum tins and dried at 50°C. Once dried, each sample was ground finely, packaged and weighed in small tin capsules before being sent to the Kansas State University Stable Isotopes and Mass Spectrometry Lab (SIMSL) for further analysis using Elementar EA Vario Pyrocube and Elementar IRMS GeovisION instruments.

Total biomass and algal biomass were processed following methods described in Steinman et al. (2017). Total biomass was measured via combustion, where the dried GF filters were weighed before and after combustion at 550 °C and the difference in weight is the recorded AFDM. Chlorophyll *a* was measured through an acetone extraction and spectrophotometric analysis of pigments using a Thermo Fisher Scientific Evolution 201 UV-Visible Spectrophotometer. Autotrophic index (AI) was calculated as the quotient of Chl *a* and AFDM. Autotrophic index is inversely related to autotrophy, where a high AI suggests a larger heterotrophic fraction. BSP was analyzed and calculated using an adapted method from Benbow et al. 2017, where [^3H] leucine incorporation into bacterial

proteins is measured via liquid scintillation. Bacterial secondary production is reported as bacterial carbon production (BCP, g C/cm²/hr), calculated by converting leucine incorporation to BCP:

[³H]-leucine incorporation (mol/cm²/hr) =

$$\frac{(dpm_{\text{sample}} - dpm_{\text{killed}}) \left(\frac{60 \text{ min}}{1 \text{ hr}}\right) \left(\frac{1000 \text{ mL}}{1 \text{ L}}\right)}{\left(\frac{2.2 \times 10^{12} \text{ dpm}}{Ci}\right) (\text{specific activity}, 0.1 \frac{Ci}{mmol}) (30 \text{ min}) \left(\frac{1000 \text{ mmol}}{1 \text{ mol}}\right) (\text{aliquot})}$$

Bacterial Carbon Production (g C/cm²/hr) =

$$(\text{leucine incorporation}) \left(\frac{100}{\text{mol \% leucine}}\right) (\text{leucine mw})(ID)(ED)(BCP \text{ conversion ratio})$$

Where *leucine mw* = 131.2, *mol % leucine* = 7.3, *intracellular dilution (ID)* = 2, *external dilution (ED)* = 31, and *BCP conversion ratio* = 0.86.

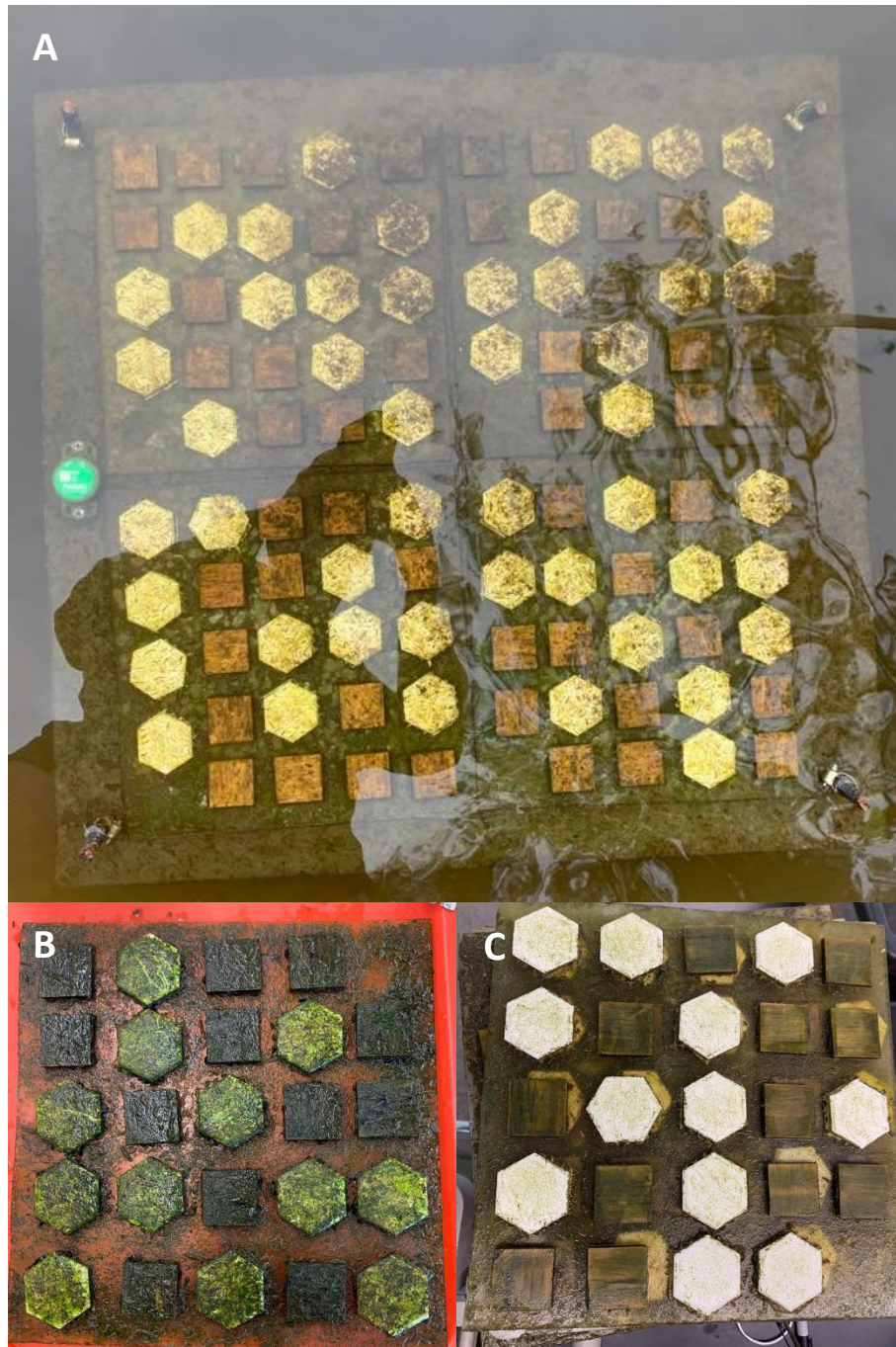


Figure 3: (A) A full biofilm deployment structure as seen on July 11, 2022. Two single plates in the lab before (B) and after (C) biofilm scraping.

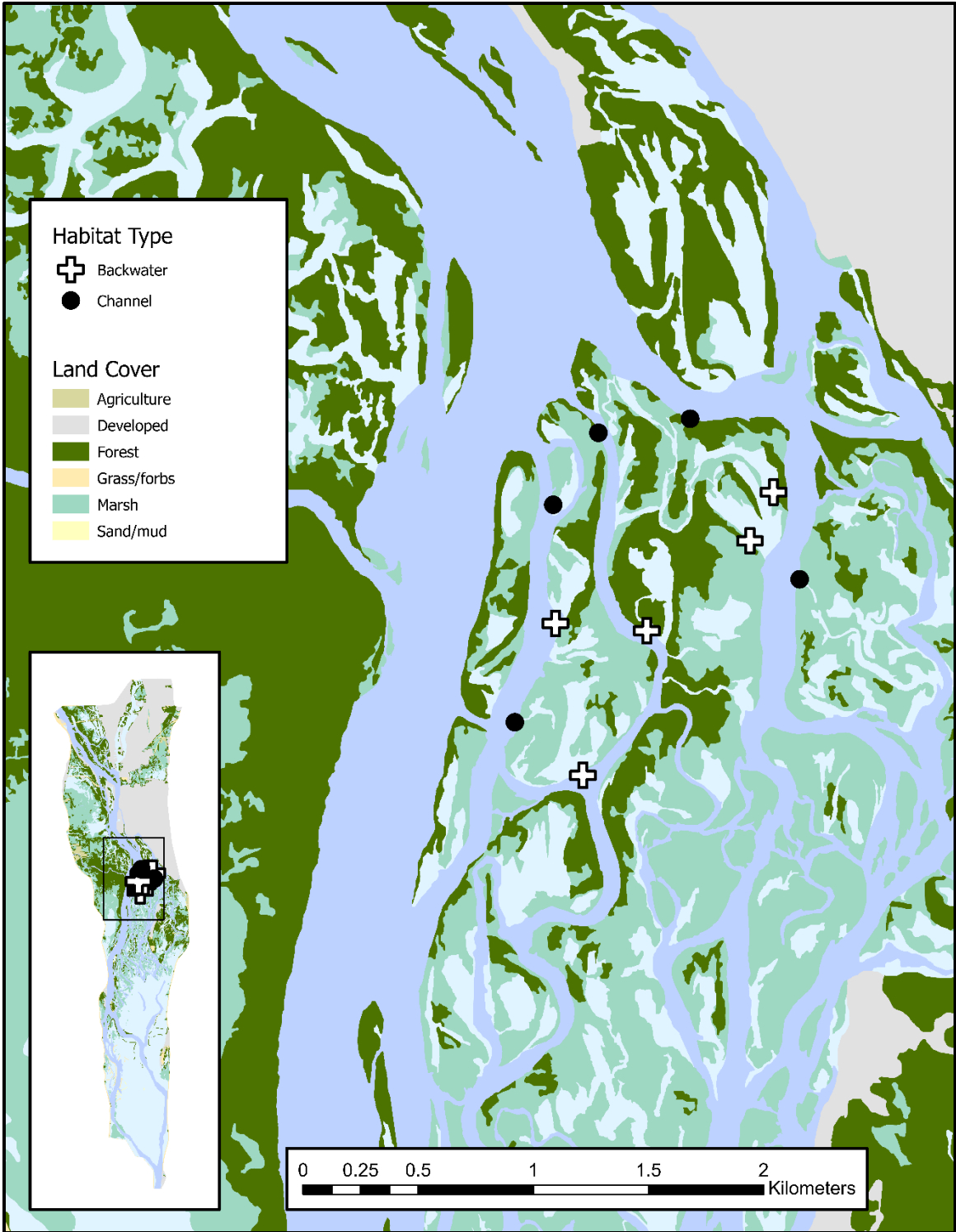


Figure 4: Biofilm study sites in UMR Pool 8. Black dots are sites in the channel shoreline and white crosses are sites in backwater areas.

Natural Biofilms

In addition to the experimental substrate study, biofilms from natural wood and commonly occurring hard inorganic structures were collected to explore the isotopic relationships among materials to infer basal food web patterns. In total, ten sites were sampled. Five sites with wood and five sites with inorganic hard substrates were sampled in May 2022 (Figure 5). While all inorganic substrates sampled were part of man-made stabilization structures, these will be referred to as ‘natural rock’ henceforth, as they likely represent the most common inorganic hard substrate found in the UMR.

Submerged wood ~0.15m in diameter were selectively sampled if there was visible biofilm growth. Biofilms were collected from logs by scraping a ring around the entire diameter and carefully rinsed into a sterilized vial with deionized water. Additionally, samples of the wood tissue were collected. Once the biofilm was removed, wood was shaved down to reveal fresh flesh to avoid any penetrating biofilms and small samples of each log were collected. Samples from the surface of stones were collected using an adapted syringe sampler and similarly rinsed and stored in vials (Steinman et al. 2017). All samples were placed on ice until returned to UWL where they were processed immediately. Processed samples were packaged and shipped to the Kansas State University SIMSL for $\delta^{15}\text{N}$ & $\delta^{13}\text{C}$ isotope analysis as described earlier.

$\delta^{15}\text{N}$ & $\delta^{13}\text{C}$ can help assess broad trophic levels and can help follow energy transfers within an ecosystem (Hershey et al. 2017). Consumers and their food sources will often have more similar isotopic signatures, indicative of enrichment (Hershey et al. 2017). Biplots of $\delta^{15}\text{N}$ & $\delta^{13}\text{C}$ were created to visualize food web relationships among the biofilms and substrates.

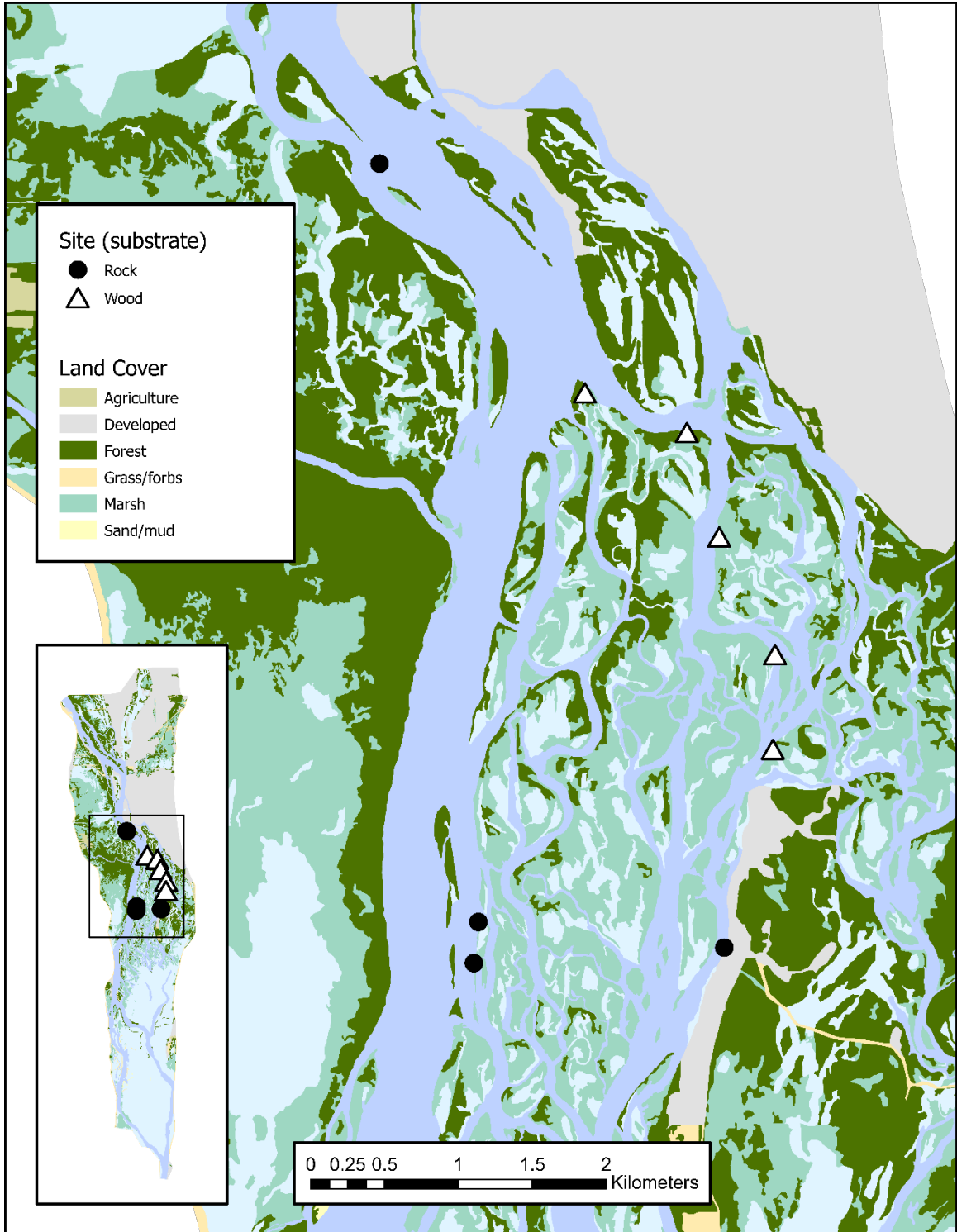


Figure 5: Natural biofilm sampling sites in UMR Pool 8. Black dots are sites where rock samples were collected, and white triangles are where wood samples were collected.

Shoreline Video Survey

To assess the distribution of shoreline large wood within the main and side channels of Pool 8, a probability survey was conducted. Fifty sites were randomly generated within the northern 2/3 of Pool 8. The lower 1/3 of Pool 8 is primarily an impounded area surrounding the main channel and therefore was not sampled due to ecological differences and lack of channel shoreline. Sites were generated using an R script (*sf*, *raster*, and *rgdal*) which randomly selected points along the shoreline of the main channel and secondary/tertiary channels, henceforth referred to as side channels (SC). Sites were equally represented in both the main channel and side channels (25 sites in each). These generated points acted as the upper end of a 100m shoreline transect to be surveyed by motorboat and video between September 8-22, 2022. In total, 47 of the 50 sites were surveyed (Figure 7). Three sites were inaccessible at the time of the study due to low water and connectivity issues. River stage deviated by less than 0.1 meters over the course of the survey with no major precipitation events occurring over the timespan (Figure 2). Within the main channel, forested reaches were the dominant land cover class with 56% of the sites being forested. 30% of MC sites were developed and 14% of sites were open. Open sites dominated the side channels with 79% being open and only 21% being forested. No developed sites were sampled in the side channels.

Sites were accessed via motorboat and video recorded with a GoPro Hero 10 attached to a Zhiyun Crane M2 gimbal to improve video stability. The camera was positioned on the bow of the boat and was oriented perpendicularly to the shoreline (Figure 6). Video recording started from the upstream end of the 100m transect where the entire length of each site was recorded and saved for later analysis. Boat distance from

shoreline varied between 1m-5m depending on channel width and depth. Telemetry data (speed, distance, GPS track) was extracted from each video using *gopro telemetry extractor.com*. Videos were stored locally and analyzed, unedited, using VLC media player. Only wood pieces which were approximated to be > 1m in length were included in the survey. Characteristics indicative of wood mobility were of particular interest and are described in Table 1. Characteristics were adapted from Angradi et al. 2004 & Angradi et al. 2010 and were chosen based on successful implementation in pilot studies conducted in Fall 2022.



Figure 6: Camera, tripod, and gimbal setup with operator on the bow of the boat.

Table 1: Shoreline large wood (LW) parameters assessed via video analysis.

Category	Description
Anchored	LW anchored to the shoreline above the bankfull level or exhibiting other evidence of originating at the current location.
Beached	LW that lacks evidence of having originated at its current location, is situated below the bank full level, and is not inundated. Usually, oriented parallel to shoreline and often has evidence of being transported (bark, small limbs, roots, root wads are missing).
Wet	LW that is at least partly below the wetted edge. Otherwise like beached.
Perpendicular	LW that is approximately perpendicular to shoreline (between 45° - 90°) Typically, also classified as anchored wood.
Parallel	LW that is approximately parallel to shoreline (between 0°-45°). Typically, also classified as beached or wet.
Root wad	LW with rood wad intact. Typically anchored.
Land cover	Forested, developed, open land

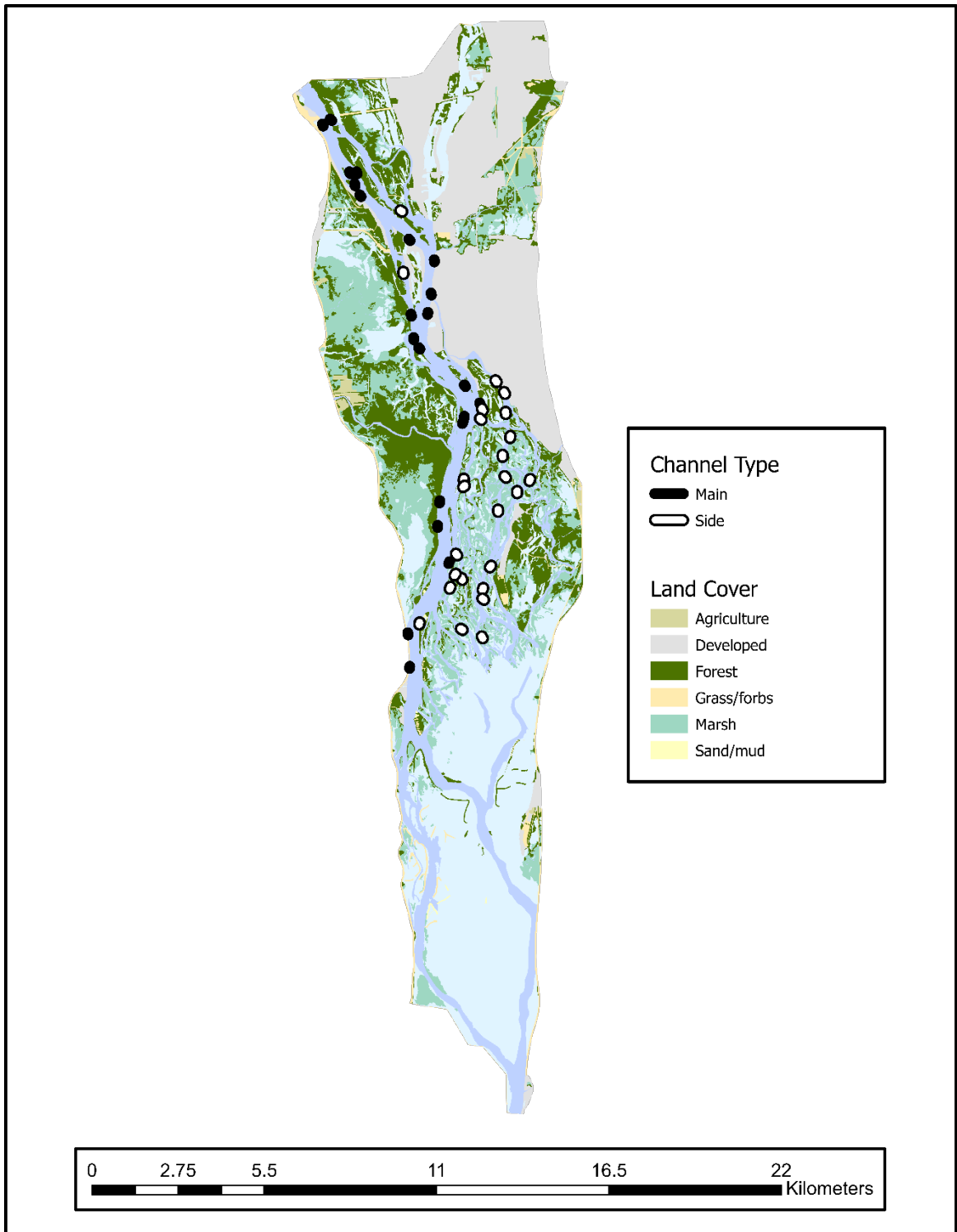


Figure 7: Shoreline survey sampling sites. Black lines represent main channel sites and white lines with black borders represent side channel sites.

Statistical Analyses

All data analysis and visualization were done in R version 4.3.0 using packages *boot*, *forcats*, *ggpubr*, *multcomp*, *RDRR*, *reshape2*, *rstatix*, *tidyverse*, and *zoo*. A split-plot design was used to assess the role of substrate type on biofilm development on the deployed tile and wood substrates between the two habitat types (channel and backwater). To understand the effects of habitat type and substrate type on biofilm development, a series of split-plot ANOVAs were performed on data recorded at peak biomass (i.e., peak AFDM). AFDM increased significantly ($t_{(30)}$, $p < 0.05$) by 69% between day ten and day twenty. Between day twenty and day thirty there were no significant changes to AFDM with only a 6.8% difference. Thus, all statistical analyses presented henceforth are pooled data from period two and three of the experiment.

Analysis of shoreline wood survey data was done using two-way ANOVAs to assess differences in abundance among the two factors, channel type and riparian land cover. Tukey's honest significant difference (Tukey's HSD) was used to examine specific differences among means.

RESULTS

Experimental Biofilm Deployments

The only statistically significant results of substrate type on any of the response variables (AFDM, Chl *a*, AI, BCP) was a significantly higher AFDM on the wood substrates than the tile substrates ($F_{(1,22)} = 15.44$, $p < 0.001$) (Table 3). Across both habitat types, wood substrates had 42.6% greater total biomass than tile substrates (Figure 8). While no significant effects of substrate or habitat type were found to affect algal biomass, AI, or BCP, clear patterns were observed (Figure 8). Across both habitats, autotrophic index and bacterial carbon production were consistently greater on wood than on tile substrata (41% and 21.6% respectively). Algal biomass was the only variable which had an apparent, albeit statistically insignificant, pattern between habitat types. Wood substrates had 26% more algal biomass in the backwater sites but in the channel, sites tile substrates had 25% higher algal biomass (Figure 8).

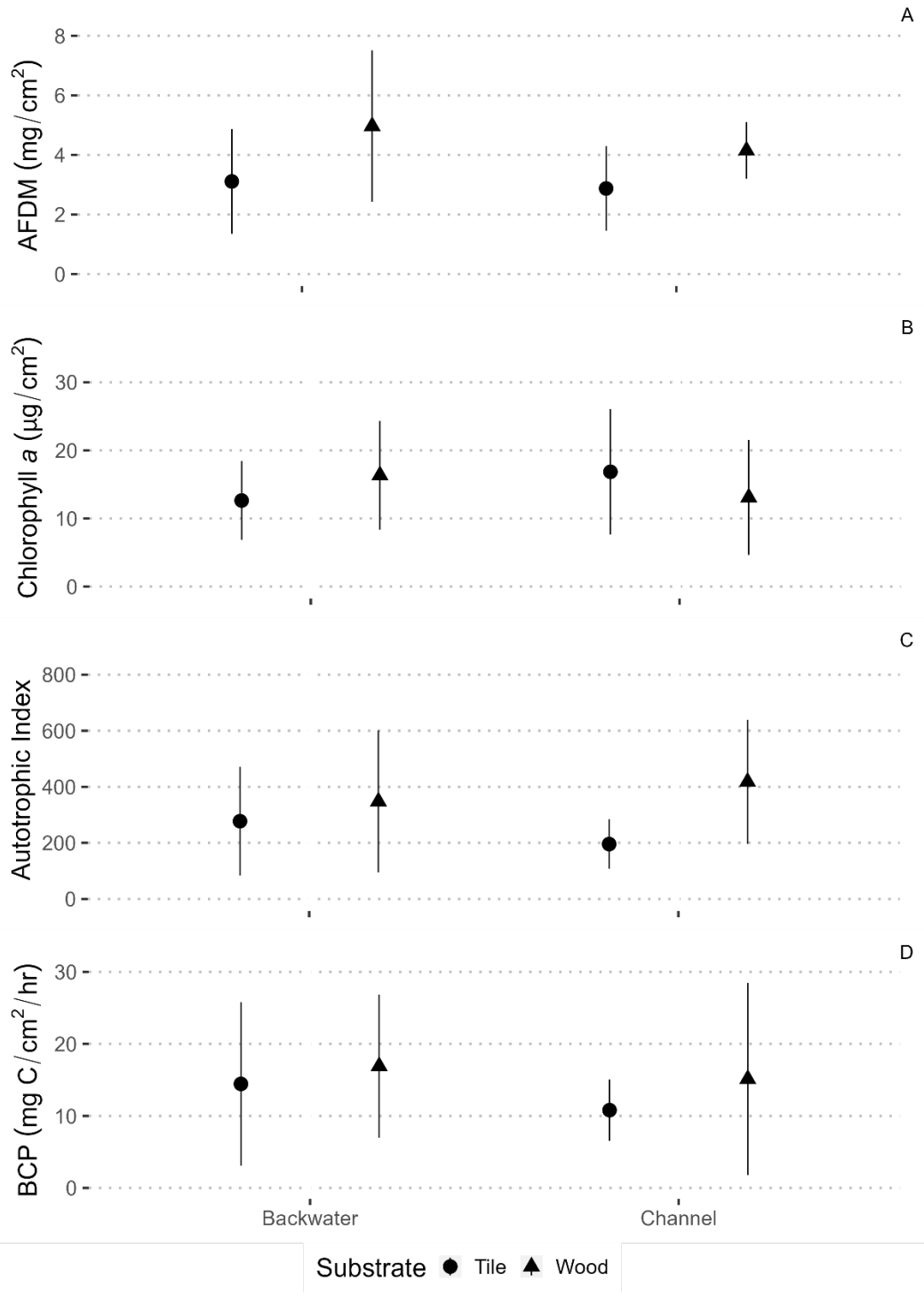


Figure 8: Mean AFDM (A), chlorophyll *a* (B), AI (C), and BCP (D) for wood and tile substrates in backwater and channel sites. Wood and tile are represented with circles and triangles, respectively. Error bars represent standard deviation.

Table 2: Mean values of temperature, pH, dissolved oxygen (DO), specific conductivity (SPC), and water velocity measured at each site over the course of the experiment. BW = backwater, C = channel.

	Temperature (°C)		pH		DO (mg/L)		SPC (µS/cm)		Velocity (m/s)	
	BW	C	BW	C	BW	C	BW	C	BW	C
11-Jul-22	26.25	25.65	8.41	8.52	9.58	9.76	387.78	392.35	0.01	0.05
21-Jul-22	25.60	25.97	8.30	8.54	6.75	8.76	348.78	346.10	0.01	0.05
31-Jul-22	24.78	24.71			10.65	13.75	347.88	345.77	0.01	0.04

Table 3: Split plot ANOVA results for AFDM, Chl *a*, AI, and BCP. An * indicates significant effect where $p < 0.05$.

	AFDM		Chlorophyll <i>a</i>		AI		BCP	
	$F_{(1,6)}$	<i>P</i>	$F_{(1,6)}$	<i>P</i>	$F_{(1,6)}$	<i>P</i>	$F_{(1,6)}$	<i>P</i>
Habitat	0.179	0.687	0.012	0.915	0.004	0.953	0.91	0.377
	$F_{(1,22)}$	<i>P</i>	$F_{(1,22)}$	<i>P</i>	$F_{(1,22)}$	<i>P</i>	$F_{(1,22)}$	<i>P</i>
Substrate	15.44	< 0.001 *	0.179	0.676	3.535	0.0734	0.660	0.425
Habitat:Substrate	0.459	0.505	2.837	0.106	1.177	0.2898	0.054	0.819

Isotope Analysis of Biofilms

Clear separation of the wood substrates (both ply and natural) and their respective biofilm samples can be seen in the elemental analysis (Figure 9A). These observed differences are indicative of suitable sampling and scraping methods of wood surfaces, suggesting that wood particulates were not incorporated into the biofilm samples at a significant level.

$\delta^{15}\text{N}$ of all the biofilms are somewhat similar while the $\delta^{13}\text{C}$ varies largely (Figure 9B). The grouping and evident enrichment of the natural wood biofilms and the corresponding wood substrate can be seen in Figure 9B. The slight upward-right trend from substrate-biofilm is apparent in the isotopic signatures and indicates direct assimilation of nutrients from the natural wood substrate into the biofilm. On average the $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of natural wood biofilms were enriched by +0.94‰ and +0.31‰ respectively. No clear patterns in isotope signatures can be seen across the other three substrate types, including the plywood tiles used in the experimental deployments.

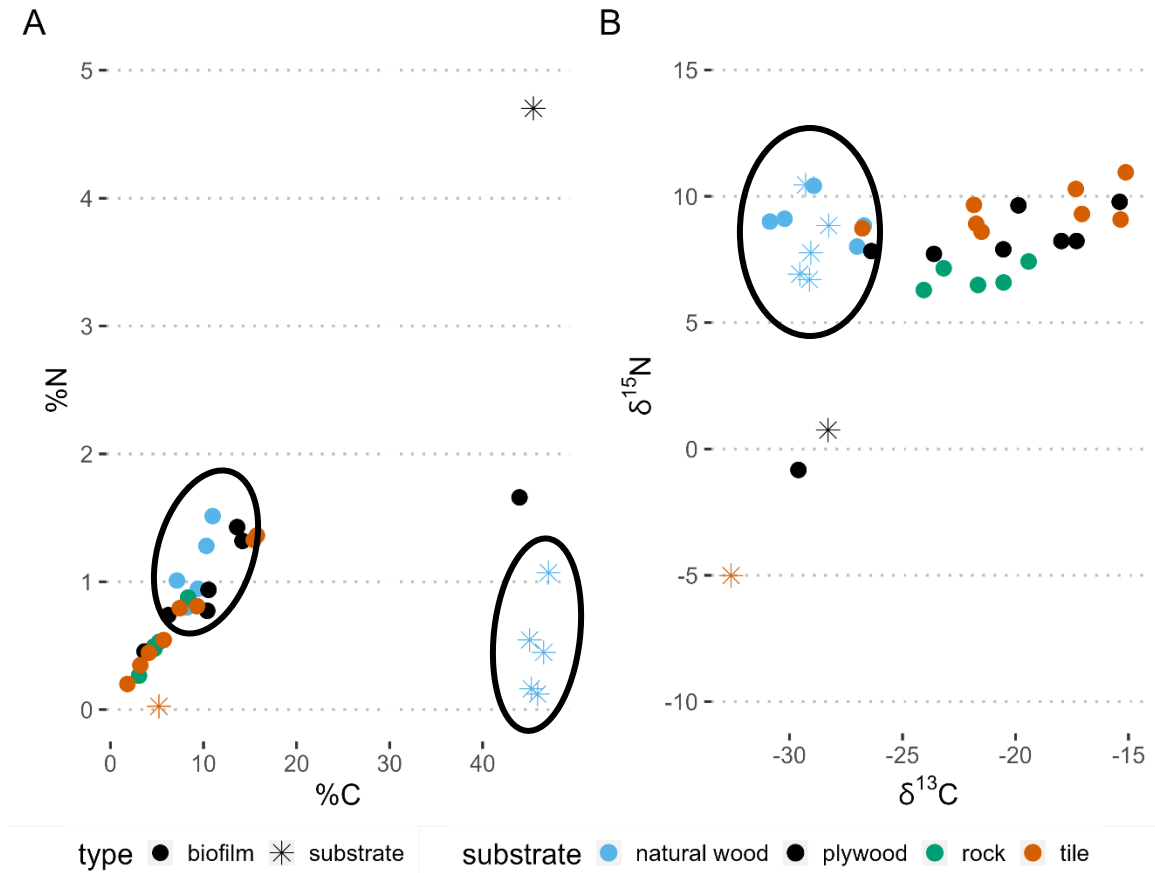


Figure 9: Elemental (A) and stable isotope (B) relationships among the biofilms and substrates collected across two experiments. Black ovals encircle natural wood biofilm and substrate samples.

Shoreline Wood Survey

A total of 125 pieces of shoreline wood were surveyed across 47 sites (2.65 pieces per 100m) (Figure 10 and Table 4). Across 23 main channel sites a total of 81 pieces were surveyed (3.52 pieces per 100m) and across the 24 side channel sites a total of 44 pieces were surveyed (1.83 pieces per 100m). Both channel type and land cover were significant effects on total wood occurrence ($F_{(1, 42)} = 4.56, p < 0.05$ and $F_{(2, 42)} = 7.15, p < 0.005$ respectively), where main channel sites had significantly more wood than side channel sites (Tukey's HSD, $p < 0.01$) and forested sites had significantly more wood than either open or developed sites (Tukey's HSD, $p < 0.05$) (Figure 10).

To examine wood transport, the orientation of wood in relation to the channel border was assessed. In general, parallel orientated wood (i.e., parallel to channel) is recognized as a sign of downstream transportation while a perpendicular orientation usually indicates local depositions (Angradi et al. 2004). Both channel and land cover were found to be significant effects on the occurrence of parallel wood ($F_{(1, 42)} = 10.27, p < 0.01$ and $F_{(2, 42)} = 4.94, p < 0.05$ respectively). Parallel wood occurred more frequently in the main channel sites than in the side channel sites (Tukey's HSD, $p < 0.01$). Forested sites had significantly higher occurrence of parallel wood than developed sites (Tukey's HSD, $p < 0.05$) but not different from open sites (Figure 11). No significant effects were found in relation to perpendicularly oriented wood, indicating similar rates of non-transported wood occurrence across channel types and land covers.

Anchored wood only differed significantly across land cover ($F_{(2, 42)} = 5.14, p < 0.05$), where forested sites had higher occurrence of anchored wood than open sites (Tukey's HSD, $p < 0.05$). No anchored wood was found at developed sites. To further examine the significance of channel and land cover on the transportation potential of wood, beached and wet wood were summed to create a class representing "transported wood". Channel type and land cover class both significantly impacted the occurrence of transported wood ($F_{(1, 42)} = 11.77, p < 0.01$ and $F_{(1, 42)} = 16.16, p < 0.05$, respectfully). Transported wood in the main channel was more common than in the side channels (Tukey's HSD, $p < 0.01$) and forested sites had more transported wood than developed sites (Tukey's HSD, $p < 0.05$) but not more than open sites (Figure 12).

Table 4: Summary of shoreline survey count data from Pool 8 of the UMR. MC = main channel, SC = side channel. Number of sites in main and side channels are represented in parentheses as such (n = MC, SC).

	Large		Medium		Parallel		Perpendicular		Anchored		Beached		Wet		Total	
	MC	SC	MC	SC	MC	SC	MC	SC	MC	SC	MC	SC	MC	SC	MC	SC
Developed (n = 7, 0)	0	-	8	-	1	-	7	-	0	-	8	-	0	-	8	-
Forested (n = 13, 5)	55	10	14	7	50	11	19	11	19	13	35	2	15	2	69	17
Open (n = 3, 19)	3	24	1	3	0	13	4	13	0	5	1	3	3	9	4	27

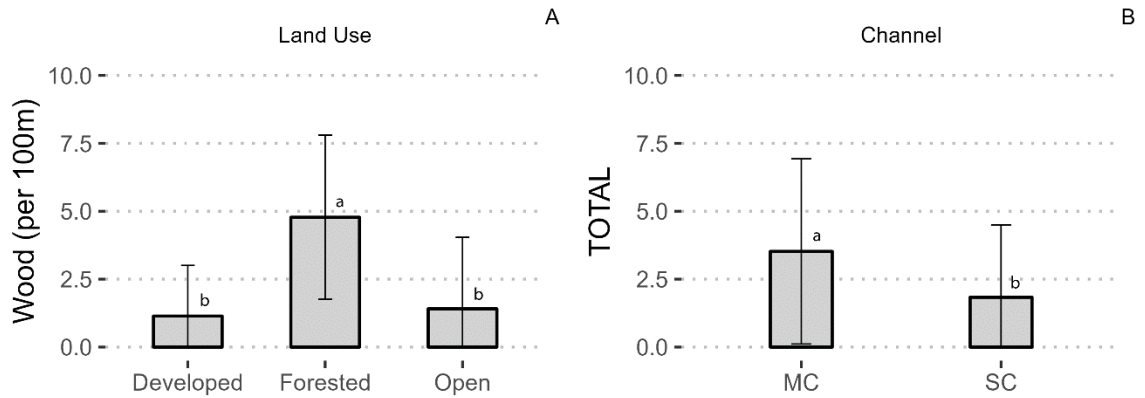


Figure 10: Mean wood occurrence per 100m of shoreline across land cover (A) and in the main channel (MC) and side channels (SC) (B). Error bars represent standard deviation. Different lower-case letters represent significant pairwise comparisons (Tukey's HSD).

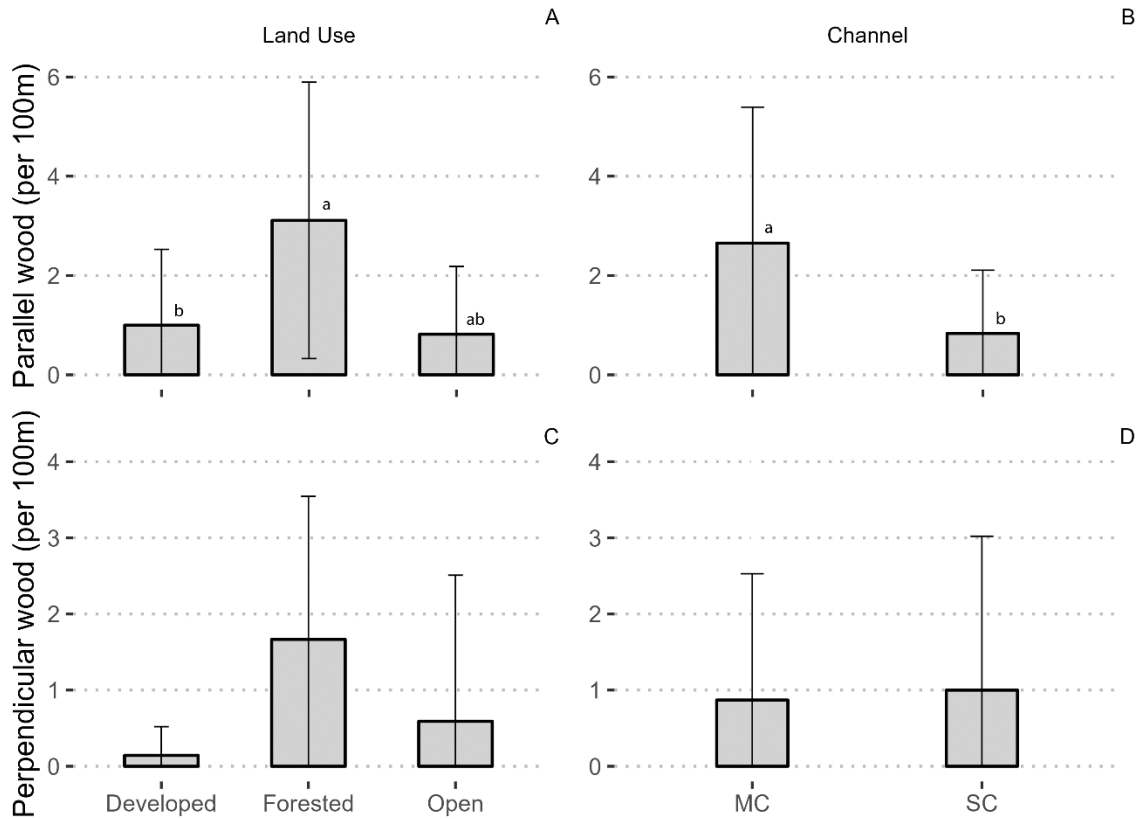


Figure 11: Mean parallel (A, B) and perpendicular (C, D) wood per 100m of shoreline across land cover (A, C) and in the main channel (MC) and side channels (SC) (C, D). Error bars represent standard deviation. Letters represent significant pairwise comparisons (Tukey's HSD).

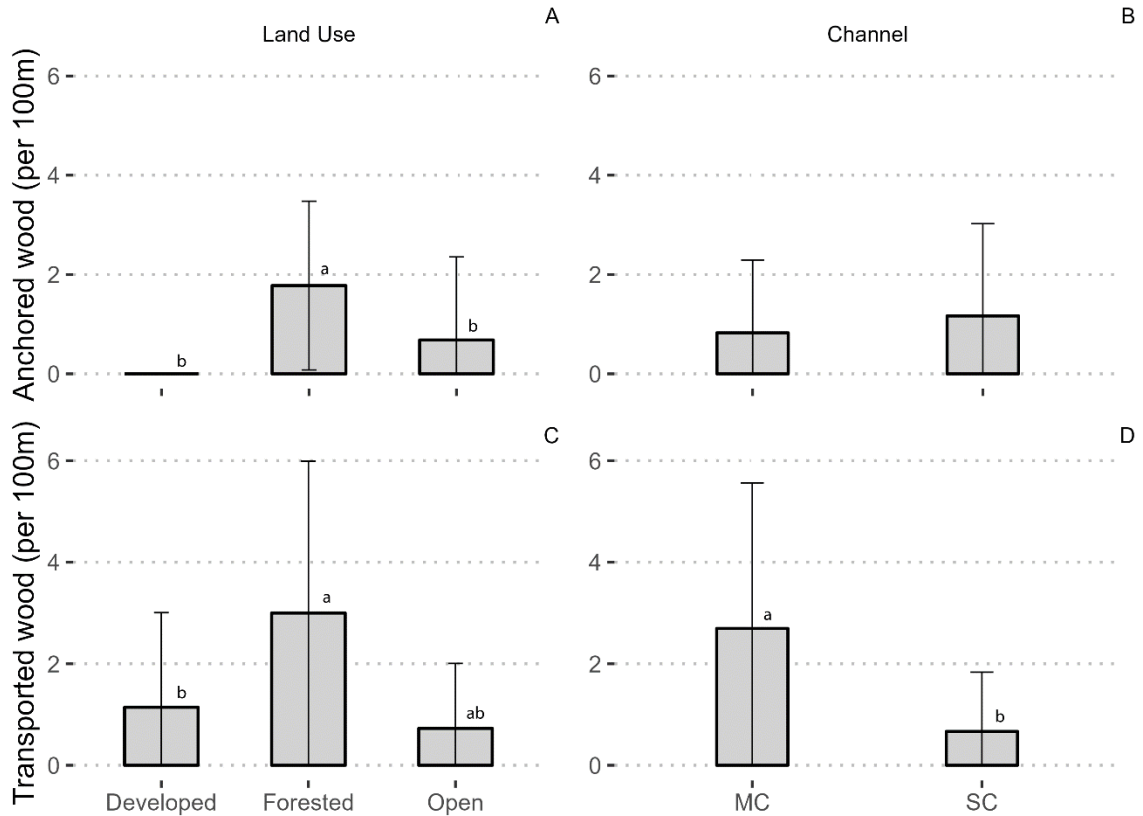


Figure 12: Mean anchored (A, B) and transported (beached + wet) (C, D) and perpendicular wood per 100m of shoreline across land cover (A, C) and in the main channel (MC) and side channels (SC) (C, D). Error bars represent standard deviation. Letters represent significant pairwise comparisons (Tukey's HSD).

DISCUSSION

Biofilm Development

Substrates

Resource availability is the driving force behind strong and productive biofilm communities. Wood is an important habitat element in lotic ecosystems, especially in large rivers where wood is often one of the few hard, stable, substrates suitable for periphyton colonization and growth. The impacts of wood substrates on biofilm development were directly assessed in two habitat types of the UMR.

Our findings suggest that wood substrates may provide heterotrophic organisms (bacteria, fungi, etc.) a better habitat to develop than inorganic surfaces. Higher total biomass and higher AI values suggest relatively low levels of autotrophy, and thus indicate strong heterotrophic activity on wood substrates. Total biomass, BCP, and AI all exhibit higher mean values on wood substrates than on the tile substrates across both backwater and channel sites. Algal biomass was the only measured response which exhibited varying effects of substrate between the two habitat types, suggesting that there is a minimal substrate influence on algal growth. This aligns with literature which suggests that wood subsidies in freshwater ecosystems provide heterotrophic microorganisms a source of high-quality carbon which is not always found within the water column or on inorganic surfaces (Freeman & Lock 1995; Hafner et al. 2005; Seo et al. 2008; Hladyz et al. 2010).

Another possible explanation for observed differences between biofilm growth on each substrate could be differences in the physical microhabitat of the surface. It has been suggested that wood surfaces, often more porous with more ‘topography’, can be more rapidly colonized (Sinsabaugh et al. 1991). It could be possible that our wood substrates simply had a more featured surface which allows for more biomass accumulation per observed surface area. In other words, the true surface area of the wood may be greater than the measured surface area (as calculated by $L \times W$), thus providing more room for biofilm growth. Future studies may be bolstered by utilizing more variety of substrates, both wood and inorganic, to examine the role of local surface habitats. However, even if this is one factor influencing greater AI and biomass on wood surfaces, it remains an important factor as these physical characteristics likely hold true in naturally occurring

substrates. If wood is simply a more hospitable physical substrate than stones, this is still an important ecological phenomenon to consider.

Habitats

Surprisingly, minimal differences were found between biofilm development in channel and backwaters sites. The literature has varied results when relating flow velocities and biofilm biomass accrual, but common theory suggests that flowing waters may provide a more continuous source of nutrients perhaps leading to higher biomass accumulation (Horner & Welch 1981; Ryder et al. 2006; Graba et al. 2013).

In contrast, high velocities are also usually related to scouring of biofilms and have been suggested to limit biofilm growth (Ryder et al. 2006; Atkinson et al. 2008). In either case, differences between the backwater and channel sites were expected (Henderson 2020).

No significant differences were found in the total biomass, algal biomass, or BCP between flowing and non-flowing sites, which could be attributed to generally low velocities at the channelized sites. Our deployments were located near to the shoreline; higher and more impactful velocities would likely be found further into the channel. Additionally, the experiment took place during a low-flow period in the summer and our channel sites averaged a velocity of 0.0458 m³/s, which may have been low enough for there to be indistinguishable differences in biofilm biomass accrual compared to no-flow sites. Associated studies examined far higher velocities than our sites experienced (Ryder et al. 2006; Atkinson et al. 2008). If our deployments did not experience extreme velocities expected to be found in channels, the results presented here may mis-represent the true effect of flowing water on biofilm growth. Future studies to examine the effects of flowing water on biofilm growth may benefit from utilizing sites with higher velocities

found further from the shoreline. However, our results still represent the true environment of biofilms growing on submerged substrates in the littoral zones of side channels.

While AI, total biomass, algal biomass, and BCP did not significantly differ between habitats, there does still appear to be a pattern in AI between wood and tile substrates between the two habitats. AI in the backwater sites differed only 22.5% between the two substrates, while it differs 72.4% in the channel sites. This difference suggests that wood substrates may be more critical to heterotrophic organisms in areas with flowing water. Backwater sites may see higher accumulation rates of dissolved organic carbon and fine particulate organic matter, allowing for strong communities to develop on both inorganic and organic substrata (Thorp & Delong 2002). In contrast, carbon may be more limiting in flowing waters. Heterotrophic communities in flowing channels may rely upon carbon in the substrate far more than those growing in carbon rich backwater sites.

Isotopes

Isotopic analyses are useful for determining the upward trophic transfer of resources in a food web, where consumers will have higher $\delta^{15}\text{N}$ values than their food source (Hladyz et al. 2011; Hershey et al. 2017). $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ are processed and moved at varying speeds through the trophic levels, allowing for clear identification and tracking of the movements of energy in a food web (Hershey et al. 2017). In general, slight increases in the $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ signatures from food source to consumer indicates assimilation (Hershey et al. 2017). Literature suggests enrichments of +3.4‰ for N and +0.4‰ for C (Hershey et al. 2017). This trend is observed in the natural wood biofilm and substrate samples (Figure 9B). Clustering and upward directional shift in the isotopic

signatures of natural epixylic biofilms and the wood substrate itself indicates some level of heterotrophic assimilation of the wood substrate directly into the biofilm. This finding strongly supports existing literature (e.g. Sinsabaugh et al. 1991; Sabater et al. 1998; Coe et al. 2009).

Biofilms found on environmentally derived rock and the experimental tiles and wood substrates had similar $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ signatures, indicating that they all have similar sources of nutrients and are not deriving nutrients from their substrata (Battin et al. 2016). Despite the evidence of substrate differences discussed in the previous section, the biofilms from the plywood experimental substrate did not show any signs of substrate assimilation. This may suggest that any differences seen in the biomass of the experimental biofilms were a result of physical differences in the substrate types (Sinsabaugh et al. 1991).

Our results suggest that the plywood used in our experiment may not be a good surrogate for wood found in the environment. While maple is a common tree found in the UMR floodplain, future studies may still find it pragmatic to try and match experimental wood species to other wood types found in the floodplain forest. Glues and other chemicals found in the binding or treatment agents of the plywood may have caused unexpected adverse conditions for the biofilms.

Another potential explanation for the differences seen between the two experiments is the quality of wood and its decay state. Natural wood found in the river was often far along in the decay process, with a more broken-down outer layer for biofilms to accumulate and penetrate, allowing for more rapid decomposition of the wood (Laiho & Prescott 1999; Kumar et al. 2018). The plywood tiles were submerged for a

maximum of 30 days by the end of the experiment, which is a relatively short amount of time in relation to the natural lifecycle of a wood in aquatic systems. It is unlikely that the wood used in the experimental deployments could get to a similar decay stage to logs found naturally in the environment, and thus the assimilation of carbon and other nutrients directly from the surface may have been lessened. Future experimental studies may benefit from using wood collected and processed from the river to mimic the natural state as closely as possible.

The elemental analysis not only serves as an important primary biological analysis, but it can also be used as a method of validating the results from our experiment. One major concern when sampling biofilms from wood is elevated AFDM (and AI) because of accidental scraping of wood particles into the biofilm sample during sample processing. While it may seem logical to question whether the differences in our response variables exhibited between substrate type could be caused by wood itself being measured, this isotopic analysis pacifies these concerns. Figure 9A shows the % C and % N makeup of each biofilm and substrate sample. If significant amounts of wood from the substrate were incorporated into each biofilm sample, thus significantly affecting the biological analyses, we might expect there to be more variation across biofilm compositions. Additionally, a few clear outliers can be seen (natural wood and plywood biofilm samples). These outliers likely represent biofilms samples with excessive amounts of wood incorporated into the sample itself.

Shoreline Wood

Across all sites, wood occurred at a rate of 2.65 pieces per 100m of shoreline. More wood was found in the main channel, with nearly twice the rate of occurrence (3.52

vs 1.83 pieces per 100m). The main channel occurrence rate of 3.52 pieces per 100m in our study parallels the rate (3.31 pieces per 100m) found in Angradi et al. (2010). In comparison to our 23 main channel sites, Angradi et al. (2010) sampled 144 sites across all 26 pools of the UMR, with ~5 sites per pool. This likely suggests heterogeneity in the abundance of shoreline wood across most UMR pools. A bootstrapped ($n = 3000$), rolling 95% confidence interval was constructed to understand how many sites would be needed to confidently describe the shoreline abundance of wood in Pool 8 (Figure 12). We suggest that $n = 23$ is nearing the lower limit of the necessary sites, however additional sites may be useful to gain slightly higher confidence.

In the main channel, forested shorelines were the most common with thirteen of the twenty-three sites being classified as such. In the side channels, open sites (grass/forbs/wetland/etc.) dominated with nineteen of the twenty-four sites being open. Seven developed sites were sampled in the main channel and there were zero in the side channels. As expected, wood was most abundant in sites with forested shorelines. Wood occurred at a rate of 4.77 pieces per 100m (MC = 5.3, SC = 3.4) at forested sites and only 1.4 pieces per 100m (MC = 1.33, SC = 1.42) along open shoreline and slightly less in main channel developed sites (1.14 pieces per 100m). Once again, these findings parallel the reported values from Angradi et al. (2010), where they found forested shorelines to produce the highest rate of wood.

In forested reaches it is likely that there is a mix of locally deposited wood and transported wood accumulating along the shoreline. In contrast, open or developed sites likely see little local deposition and thus, rely more heavily on transported wood. This

seems to be the case, as all anchored wood in the main channel and over 72% of anchored wood in the side channels was found in the forested reaches.

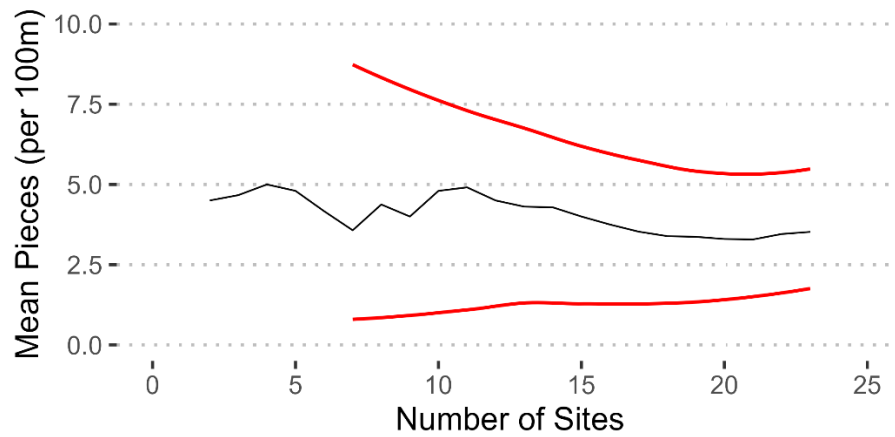


Figure 13: Bootstrap ($n = 3000$) rolling mean and 95% confidence interval (CI) of main channel large wood. The black line is the mean, and the red lines are the 95% CI.

Transport

Wood is often thought to enter a river as a fallen tree, or an anchored piece of wood (Triska 1984; Wohl et al. 2019). As the fallen tree experiences high flows, flood conditions, ice flows, and other forces of nature, branches likely fracture and become transported downstream. Wood in streams and rivers is typically not transported long distances, however the relative size of the river and its flow conditions may be proportional to the downstream movement of wood (Webster et al. 1994). Understanding the potential for downstream transport of wood is of much interest, particularly in heavily altered and managed systems such as the UMR (Swanson et al. 2020).

As suggested, reaches with minimal riparian woody vegetation lack the ability to locally produce wood and evidently rely on the transport of wood to produce in-stream structure. We propose that while there is evidence of significant longitudinal movement of wood within the main channel, side channels are largely lacking in a similar

mechanism. Our results provide evidence of significant downstream transport of wood within the main channel, while far less wood appears to be entering and moving within the side channels. Main channel forested sites were shown to have significantly more transported wood (beached + wet) and parallel orientated wood than any other land cover class across both channels, including side channel forested sites. A few key factors are likely able to explain the differences in transport between the channels.

First, there were far more forested sites in the main channel than in the side channel (13 vs 5). If more trees are available to enter the river in the main channels, it seems logical to assume that more wood would be found on the shorelines of these reaches. Another potential influencing factor is differences in flow magnitude between the larger main channel and smaller side channels. Higher discharges erode shoreline at a higher rate, potentially producing more downed trees and thus higher accumulations of shoreline wood in these areas (Bilby & Ward 1989). Additionally, higher discharges can both break apart and move larger pieces of wood quicker than low magnitudes (Wohl et al. 2019). This theory is supported by a relatively high rate of anchored wood in the side channels, indicating that fallen trees tend to remain intact rather than being broken down and transported downstream.

Finally, there may be methodological concerns which may result in discrepancies. The main channel in Pool 8, and most other UMR pools alike, is highly channelized, generally deep, and fast flowing (Johnson & Hagerty 2008). This may result in pipe-like movement of wood during flood events following periods of high precipitation or spring snowmelt. In contrast, the side channels of Pool 8 are far more prone to overbank flooding, with many wet marshes and floodplain forest still existing in the middle portion

of the pool (Figures 1 & 3). Overbank floods of high magnitude may be able to deposit large wood deep onto the floodplain, likely snagged quickly once entering the floodplain, making it potentially difficult to identify via our videographic method. Exploration of this hypothesis could be tested via transect-based sampling as used in vegetation surveys, or remote sensing with lidar and drone imagery.

Connections

Two complimentary approaches to categorizing the ecological importance of wood have been examined: biofilm experiments and shoreline surveys. These two components can be linked together to build our understanding of the role of wood in the UMR. Our findings that wood supports biofilm biomass is further confirmation of many studies which have suggested that wood can directly fuel the aquatic microbial communities found on wood surfaces (Sinsabaugh et al. 1991; Sabater et al. 1998; Coe et al. 2009). To conceptually link biofilms to the transience of wood, it is useful to reflect upon the critical changes that have occurred to the Upper Mississippi River and its surrounding floodplain over the past century. Climate change, invasive species such as reed canary grass, and management changes to the UMR have permanently altered the state of the floodplain, reduced floodplain connectivity, and significantly decreased the once widespread coverage of floodplain forest (Nelson et al. 1994; Johnson & Hagerty 2008; Belby et al. 2019; De Jager et al. 2018).

It is likely that the abundance of wood, both anchored and movement-prone, was far higher prior to human influence. Wood in rivers supports life across all trophic levels, and thus with a declining source of this important resource we postulate that the ecosystem health of the UMR has been greatly impacted by the loss of wood substrates in

rivers. We found that forested shorelines are critical to both the existence and accumulation of wood, and that within the smaller side channels local deposits of wood are vastly important. Additionally, we provided evidence supporting the hypothesis that wood substrates may be a better substrate for biofilm communities than inorganic substrates. Biofilms are important drivers of production and can improve overall ecosystem health of large rivers such as the UMR (Lock et al. 1984; Freeman & Lock 1995; Battin et al. 2016; Howard-Parker et al. 2020). We propose that UMR Pool 8, particularly in the side channels, has a reduced level of biofilm productivity as a direct result of multi-century declines in shoreline and in-stream wood subsidies.

Towards the Future

Biofilms

The difference in isotopic signature between natural biofilms and those biofilms grown on deployed wood needs to be investigated further. Experimentally deployed substrates allow for multi-effect studies to be easily replicated. However, ensuring that these biofilms represent naturally occurring biofilms well is critical to truly understanding the role of wood as a resource for microorganisms. Using multiple types of more representative wood or the use of natural cut logs, should be explored in future studies. Furthermore, a more comprehensive examination of natural wood and their related biofilm communities is recommended. Confirming the findings of direct incorporation of wood into biofilms would be a key discovery and may support future management decisions.

Survey

The survey presented here provides many insights into the general distribution of wood in the river. We suggest that repeated surveying of the same sites would provide information regarding the transport of wood in the UMR. Our study took a snapshot during low-water conditions. Seasonal returns to the 47 sites presented here would provide key information on the timing of wood-moving events in the UMR. Annual returns may inform on the broad understanding of the wood budget within the pool (Wohl et al. 2017; Wohl et al. 2019).

This study balanced a large spatial scale and field sampling ease with relatively low-resolution data collection. The rather coarse nature of the data collected, while informative, may also be refined in future studies. Although a more field-intensive method, actively walking the shoreline of each transect may provide more reliable counts and classifications. Wood not able to be seen via video recordings such as pieces deeply deposited onto the floodplain, covered by vegetation, or partially buried, may be identified via terrestrial transect-based studies, and could be incorporated as additional data to these studies.

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APPENDIX A

TIMESERIES LIGHT DATA

Light sensors logged at five-minute intervals for the duration of each 10-day period. At each sampling date, sensors were typically covered in biofilm. Sensors were wiped clean from biofilm at the beginning of each sampling period, however growth occurs rapidly and led to (likely) inaccurate light readings by day two of each deployment.

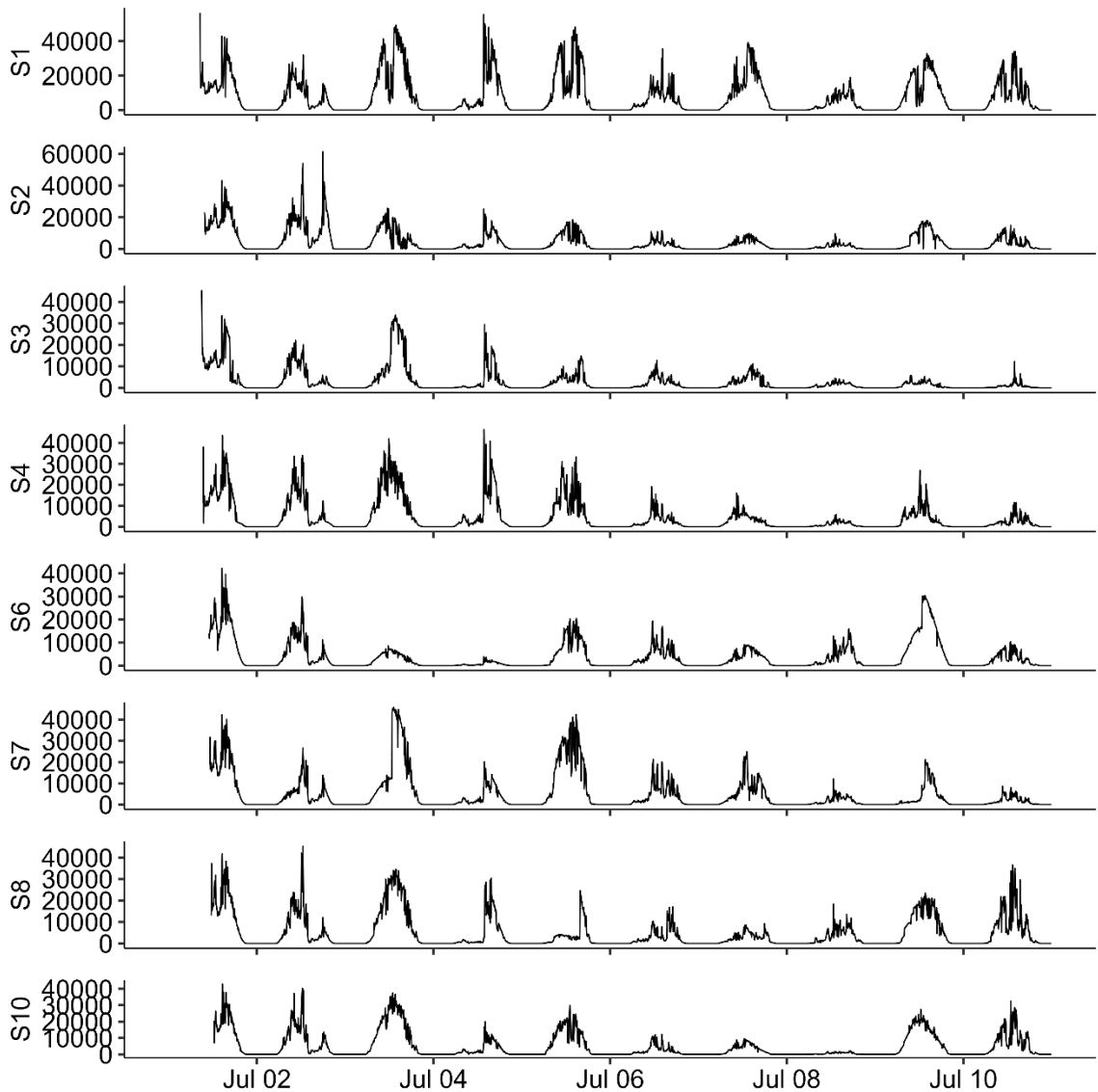


Figure A1: Timeseries light (lux) for all sites between July 2nd-11th, 2022.

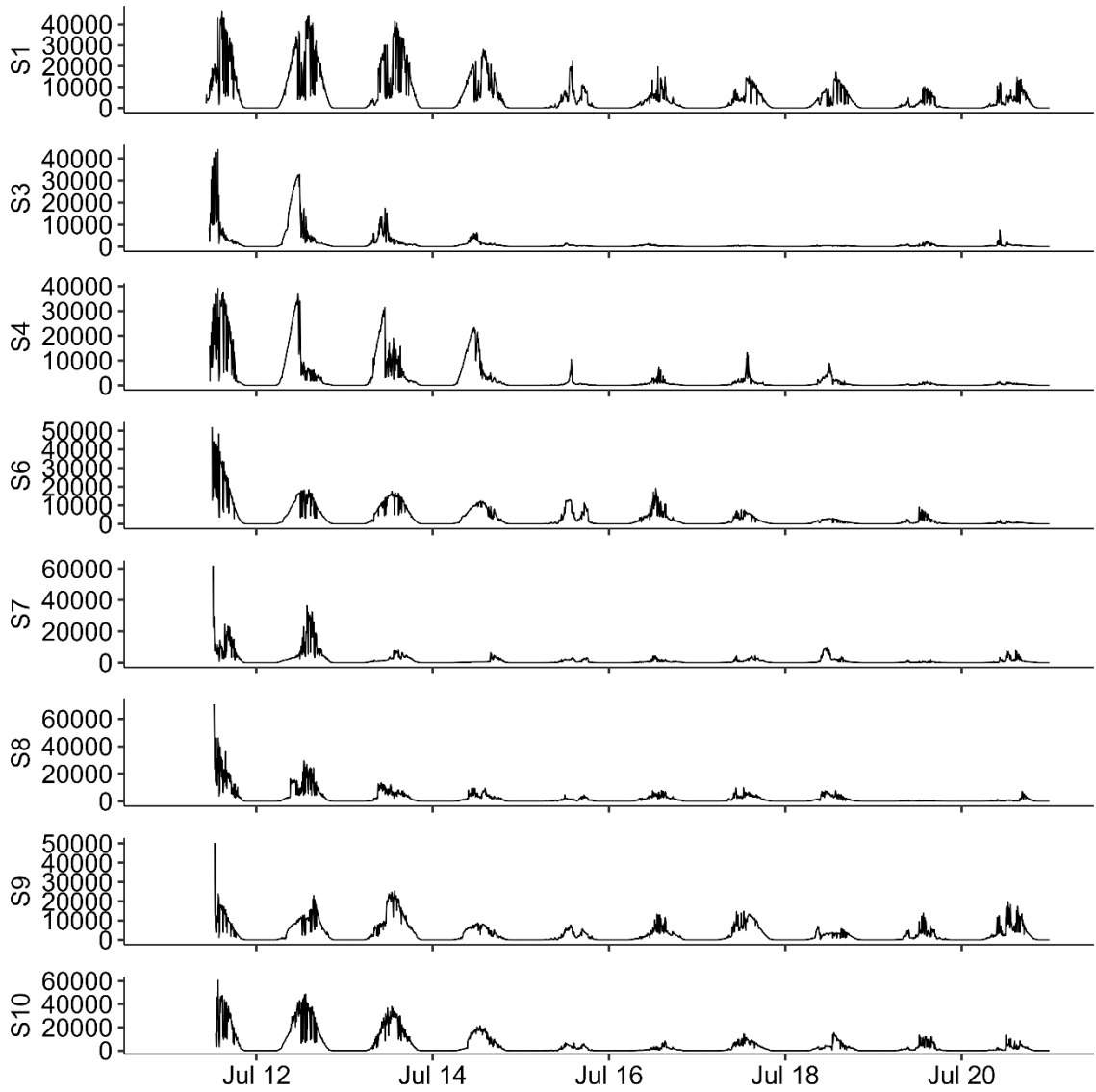


Figure A3: Timeseries light (lux) for all sites between July 12th-21st, 2022.

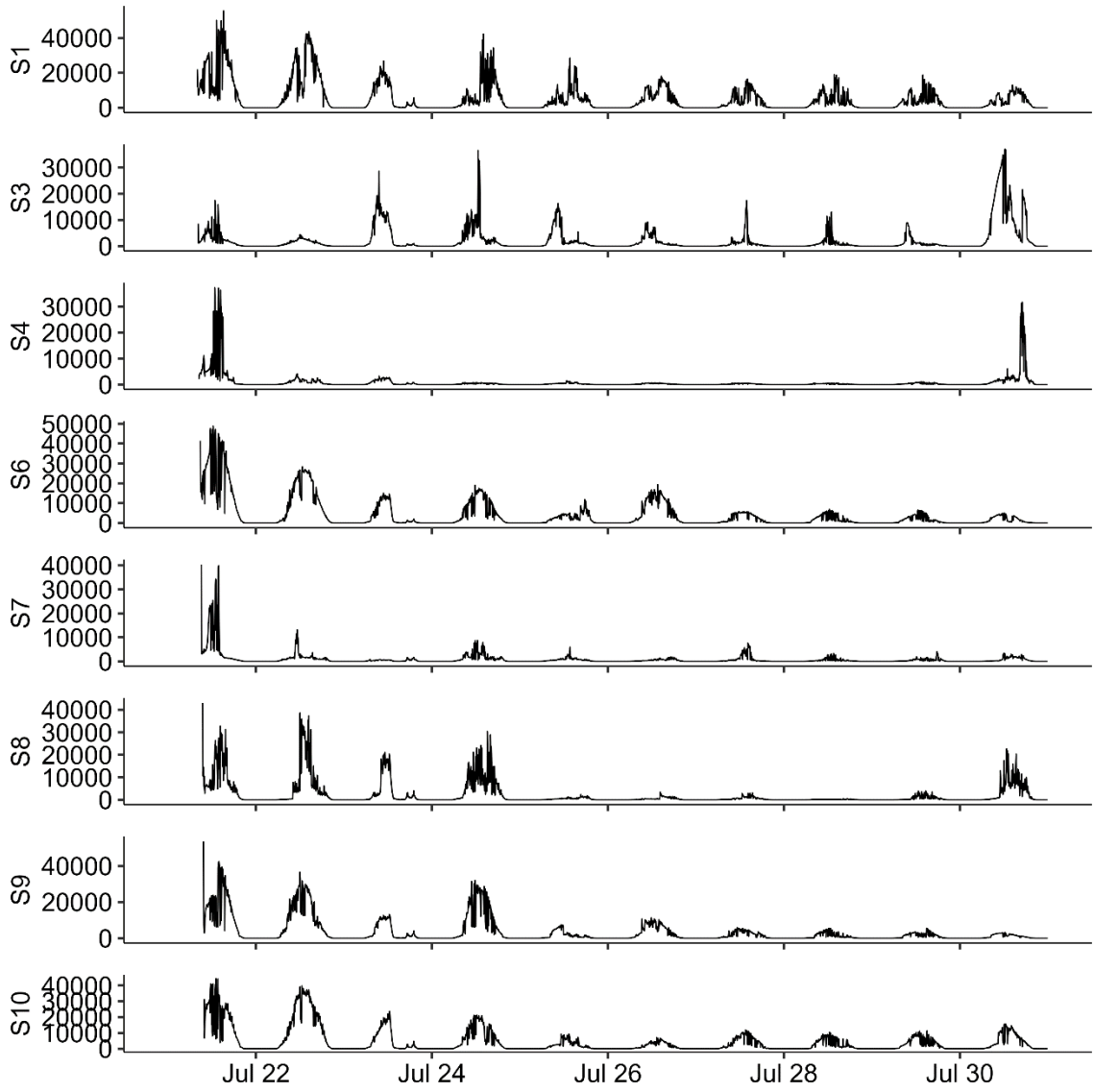


Figure A3: Timeseries light (lux) for all sites between July 22nd-31st, 2022.