**Chapter 1: Temporal and spatial carbon dynamic in Flatwood, Blackwater Streams: the Chimney-Reactor Pendulum**

Streams link terrestrial and marine environments, transporting, storing, and transforming terrestrial carbon before it reaches the world’s oceans. Terrestrial litterfall and debris enters small, low-order streams and accumulate in large, high-order rivers before eventually discharging into coastal marshes. In the “conventional carbon cycle” (Battin et al., 2009), this transport from low to high-order streams and rivers was viewed as passive, with minimal biogeochemical activity. Currently, streams and rivers are understood to play an active role in global carbon cycling. Less than half of terrestrial carbon inputs reach the oceans; the rest is mineralized or stored as water flows toward the coast. Despite only half of terrestrial carbon entering streams, CO2 emissions from global streams are equivalent to terrestrial net ecosystem productivity, and total stream carbon is often greater per unit area than the surrounding terrestrial landscape. Lotic carbon outputs are greater than inputs, creating uncertainties in regional and global carbon budgets. The “carbon gap” is debated in the literature but largely attributed to CO2-rich groundwater degassing from the stream channel. However, other sources, such as wetland or internal production, are insufficiently explored and constrained.

An inadequately researched but increasingly important area of stream carbon cycling is the contribution of low-order, headwater streams. Small streams, though covering less than 1% of Earth's area (Battin et al., 2009), constitute the largest portion of all rivers and streams, drain approximately 75% of all watersheds, and are hypothesized to play a disproportionate active role in global carbon cycling (Marx et al., 2017). Headwater catchments have higher DOC concentrations than high-order, downstream waters, and 36% of stream CO2 emissions is predicted to originate from small streams (0.93 Pg-C/yr). However, estimates of small stream carbon fluxes and the processes driving the high biogeochemical activity are relatively few and largely speculative (Drake et al., 2018; Marx et al., 2017). Current research predominantly investigates carbon dynamics in stream orders four or higher, overlooking first, second, and third order, often perennial, streams include Laurewald 2015 (Cole et al., 2007; Drake et al., 2018). Numerous models have predicted a negative relationship between gas transfer velocity and stream order, estimating CO2 emissions from first to third-order streams contribute three times the global stream average, suggesting that global budgets underestimate global stream emissions (Raymond et al., 2013). This knowledge gap is partially due to the location of small, headwater streams, which are typically in remote, undeveloped areas, making remote sensing delineation challenging and field access energy-intensive. Additionally, scaling limitations, especially for gas exchange rates and discharge estimates, in “infinitely small” perennial streams, create inaccuracies that hinder comprehensive global estimates Laurewald (Battin 2009(Marx et al., 2017)).

What is understood, but nonetheless poorly constrained, is that stream carbon is sourced from two pathways: the chimney or reactor pathway (Bernal et al., 2022; Hotchkiss et al., 2015). The chimney pathway is the passive transport of externally sourced carbon (predominantly from soil, groundwater, or the riparian zone) that degasses from streams with minimal downstream transport. In this pathway, the stream serves as a “chimney,” or a vector for atmospheric exchange. In contrast, the reactor pathway involves the mineralization of organic carbon through respiration or anaerobic processes, producing CO2 as a byproduct. In the reactor pathway, carbon is actively transformed from organic to inorganic forms. As mentioned, more CO2 is degassed from streams than the sum of terrestrial inputs and internal production (the reactor pathway) can account for. This “gap” is attributed to “chimney” carbon, assumed to be sourced from groundwater seepage. However, the reactor pathway, the internal production of carbon via mineralization, is itself poorly constrained (Drake et al., 2018), with publications attributing anywhere from 12% to 40% of total stream carbon to respiration. Current estimates do not incorporate temporal or spatial changes that could impact stream carbon sources. Seasonality affects temperature and precipitation, which in turn influences biogeochemical reaction rates and flow regimes, impacting residence times and affecting the stream's ability to process carbon. Additionally, landscape slope, soil permeability, and wetland area impact lateral, overland, and subsurface carbon export, modulating the influence of the reactor pathway. Streams “swing” between chimney and reactor states, dictated by temporal and spatial fluctuations in the landscape hydrology (Zarnetske et al., 2018). Although the chimney pathway may dominate in some streams, it does not apply to all flowing waters year-round. The same stream may exhibit a more prominent reactor pathway when flow is equal to processing time (low residence times) (Bernhardt et al., 2017; Hall et al., 2016; Zarnetske et al., 2018), while receiving increased chimney carbon during flooding in nearby wetlands. Ignoring spatial and temporal lotic dynamics undermines the concept of “active pipes”—streams as active components in global carbon cycling.

For my first chapter, I will investigate the temporal and spatial dynamics of carbon within low-order, flatwood streams over multi-annual time scales, focusing on the response to flow extremes, seasonal fluctuations, and the surrounding landscape. To explore these dynamics, I have selected nine remote, flatwood streams within the Bradford Experimental Forest (BEF) and their higher-order receiving river, the Sampson River, to deploy high-frequency, long-term sensor packages containing CO2, dissolved oxygen (DO), and methane sensors. In addition to high-frequency observations, I will collect monthly samples for dissolved organic carbon (DOC), fluorescent dissolved organic matter (FDOM), dissolved inorganic carbon (DIC), and particulate organic carbon (POC). I aim to parse stream carbon fluxes into the reactor and chimney pathways, a perspective rarely explored in current literature. I hypothesize:

1. The chimney pathway dominates in flatwood streams, but the reactor pathway becomes more prominent during baseflow conditions when residence times are longer and external contributions are minimal.
2. I also expect streams in basins with greater wetland areas to have more influential chimney pathways, whereas streams in basins with less wetland area exhibit a more prominent reactor pathway.
3. Lastly, I hypothesize that low-order streams will have greater carbon concentrations compared to their higher-order receiving river, with DIC being the most concentrated, followed by DOC, and then POC, even through flow regime fluctuations.

My objective for this chapter is to provide greater insight into headwater, low-order stream contributions to regional and global carbon cycling and to demonstrate how landscape hydrology can influence stream carbon dynamics.

Methods:

*Sites:*

The BEF is a contiguous pine flatwoods landscape situated above the Hawthorne Formation clay bed, which confines the principal aquifer, the Upper Floridan Aquifer (UFA) (Hensley & Cohen, 2017). This area, characterized by low-relief topography, features numerous depressional basin wetlands typical of North Florida flatwoods. These wetlands, both isolated and riparian, support a shallow surficial aquifer that acts as a transport network for nutrients. The land is primarily managed for silviculture and is largely owned by the Rayonier Corporation, with only a few residential homes and businesses present.

Streams within the BEF exhibit typical characteristics of blackwater systems: they are tannic, rich in dissolved organic carbon (DOC), have low pH levels, and contain high concentrations of carbon dioxide (CO2) (~20,000 ppm). These streams, both permanent and intermittent, drain the landscape before discharging into the Sampson River (at the southern extent) or Sampson Lake (at the northern extent). Nine streams (Stream IDs: 3, 5, 5a, 6, 6a, 7, 9, 13, and 15) across nine delineated basins were chosen for long-term observations and monthly sampling. Each stream displays distinct characteristics owing to each basin’s specific features (FIGURE #), such as wetland area and groundwater influence.

*Long-term, high-frequency observations:*

Each stream will be equipped with a sensor package that tracks hourly changes in water quality. This package includes sensors for dissolved oxygen (DO), pH, CO2, and specific conductivity (SpC), as well as a pressure transducer (PT) to measure depth. In streams 5 and 6a, located in the northern and southern regions respectively, a PT will be deployed in ambient air for accurate water depth calculations. DO, temperature and depth readings will be used to estimate stream metabolism (see below). Temperature, CO2, and pH will be used to interpolate continuous concentrations of HCO3 -and CO32- from Bjerrum equations (see below). Each sensor package will be serviced, and data offloaded once a month.

*Monthly Sampling*:

By sampling for each carbon species, I will estimate total stream carbon and observe how the proportions of these species change with flow and season. Particulate organic carbon (POC) remains largely unexplored due to the nature of POC fluxes, which are storm-driven and therefore challenging to capture (Marx et al., 2017). However, in these low order, blackwater systems, notable POC concentrations are expected. Dissolved organic carbon (DOC) is derived from the weathering of POC and powers respiration. DOC has the potential to alter energy dynamics within lotic ecosystems, serving as the preferred electron acceptor in aerobic and anaerobic biogeochemical reactions (Zarnetske et al., 2018). Furthermore, DOC concentrations have been linked to flow, with high discharge rates positively correlated with DOC concentrations (Marx et al., 2017). Dissolved inorganic carbon (DIC) includes mineral carbon and CO2. On average, the pH of BEF streams is less than 5, so substantial concentrations of DIC are not expected. Nonetheless, given that Florida is essentially a massive limestone deposit, it is necessary to measure DIC. Fluorescent dissolved organic matter (FDOM) will be used to extrapolate carbon quality and assess differences in quality across seasons, and between the Sampson River and its headwater streams.

Analyzing DIC is notoriously challenging due to its tendency to degas from the sample causing concentrations to be underestimated. To minimize error, acid-washed Shimadzu sample vials are used in the field to sample DIC, avoiding any potential degassing during decanting. In the field, vials are filled to maximum capacity to prevent head-space equilibration. After collection, all samples are stored in the fridge and analyzed within 48 hours.

Both DIC and DOC analyses will be conducted using the Shimadzu TOC-L analyzer, employing the total organic carbon (TOC) measurement method. Although FDOM is more stable than DIC, it can degrade after extended periods in storage, or denatured if frozen. Therefore, FDOM samples are stored in the fridge in opaque, dark bottles and analyzed on the Horiba Aqualog within three weeks of collection. POC samples are collected using 500mL to 1L Nalgene bottles, submerged midway in the water column and capped underwater. The samples are then filtered using 0.45-micron filters. These filters are then processed utilizing the ash-free dry weight method.

*Stream Metabolism:*

To estimate the significance of the reactor pathway in flatwood streams, stream metabolism—the rates of gross primary productivity (GPP) and ecosystem respiration (ER)—will be modeled. Stream metabolism serves as a holistic, in-situ measure of stream carbon cycling and organic carbon mineralization. ER represents the total respiration (oxygen consumption) and is the dominant pathway for internal CO2 generation, while GPP represents the total productivity (photosynthesis; oxygen production) within the ecosystem. One-station stream metabolism methods estimates GPP and ER by tracking continuous fluctuations in dissolved oxygen (DO) concentrations (mg/L) over time (hr^-1) and depth (m^-3), calculating an oxygen flux (g O2 m^-3 hr^-1). Assuming the respiration ratio is 1:1 for CO2 and DO (where every mole of DO consumed results in one mole of CO2), the difference between observed CO2, and CO2 estimated by ER, indicates allochthonous CO2 from the chimney pathway. Stream metabolism modeling will be performed using StreamMetabolizer (Appling et al. 2020), an open-source R package that integrates principles from Odum’s *Primary Production in Flowing Waters* and Bayesian modeling to estimate GPP and ER.

*Estimating Gas Exchange*

To determine air-water gas exchange rates—an essential parameter for stream metabolism modeling—the reaeration coefficient (K600) will be field-estimated using gas dome methods. During each monthly field visit, K600 will be empirically estimated using a floating dome, an inverted plastic container with a headspace volume of 15.5 L. Within the dome, a high-frequency CO2 sensor will be placed in the headspace, sampling at 1 sample per 10 seconds for approximately 25 minutes. Given that stream CO2 concentrations often exceed 20,000 ppm, the diffusion of CO2 from the water column to the headspace will be monitored to calculate the rate of air-water equilibration. The gas exchange velocity, K (m d^-1), which represents diffusion between the water column and atmosphere (Hall and Ulseth, 2019), and subsequently the reaeration coefficient, k (d^-1) (calculated as K divided by stream depth), were predicted following methodologies outlined by Khadka et al. (2014) and McDowell & Johnson (2018).

Using the field-measured K600 values and discharge data, a rating curve will be developed to enable continuous estimation of K600. This rating curve will be integrated into *StreamMetabolizer* to refine K600 estimates as a function of discharge to improve accuracy. Discharge measurements will be obtained through periodic dilution gauging. During monthly field visits, streams with observable flow will undergo dilution gauging using salt dilution techniques and slug-injection methods. By correlating depth at the time of dilution gauging with discharge rates, a rating curve will be constructed to maintain continuous discharge estimates.

*DIC Interpolation*:

Using the Bjerrum equation, continuous bicarbonate (HCO3^-) concentrations will be interpolated from temperature, pressure (PT), pH, and CO2 sensor measurements. The R package *seacarb* facilitates estimation of the first and second dissociation constants of carbonic acid based on temperature and water pressure, streamlining data manipulation.

*Statistical analysis:*

To test hypothesis 1, the reactor pathway becomes more prominent during baseflow conditions, a linear regression analysis between respiration derived CO2 and discharge will be employed. Similarly, for hypothesis 2 (greater wetland-area contributes more chimney carbon) a linear regression between wetland-basin area and respiration derived CO2 will interpolate landscape influence on stream carbon dynamics.

As for hypothesis 3, low-order streams will have significant carbon concentrations, ternary plots (DIC vs DOC vs POC) exploring carbon species proportions and how carbon alters with fluctuating discharge will be use detect temporal dynamics in stream carbon.

Preliminary Results: