

IDENTIFYING AUTOCHTHONOUS VS ALLOCHTHONOUS TERRIGENIOUS ORGANIC MATTER USING DISSOLVED LIGNIN AS A BIOMARKER

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Introduction

Objective

- Develop lignin oxidation and phenol detection method at RSMAS using known material (local red mangrove leaves)
- Compare lignin concentration and composition in local waters of Biscayne Bay and advected waters of the Florida Straits
- Hypothesis: advected waters in Florida Straits contains degraded lignin originating from a distal source
- There will be a lower lignin concentrations and a higher Ad:Al indicative of more advanced degradation, and S:V ratio may be different (indicative of angiosperm vs. gymnosperms)

Background

- Lignin is a large biopolymer sourced from land plants [1]
- Lignin is made up of 3 major types of subunits, of which specific ratios can be used for identifying:
 - Syringyl:vanillyl (S:V)- angiosperm or gymnosperm
 - Cinnamyl:vanillyl (C:V)- woody or non-woody tissue (not useful in marine samples due to other sources of cinnamyl phenols)
 - Acid:aldehyde (Ad:Al)- state of degradation
- Biscayne Bay waters have short residence time with large terrigenous input, while Florida Strait surface waters are older and of Amazonian origin [2,3]

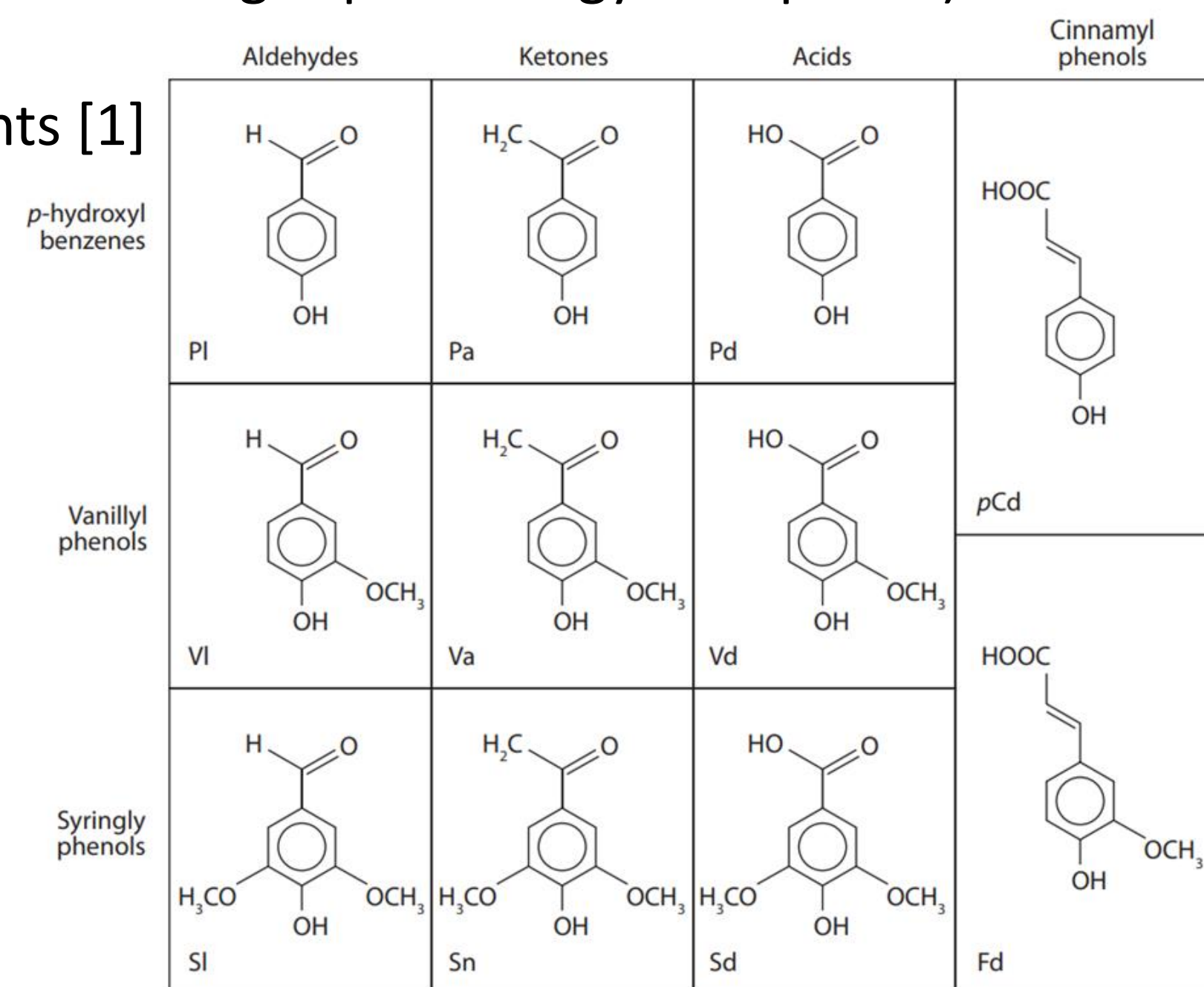


Figure (above) depicts various lignin phenols derived from CuO oxidation [1].

Methods

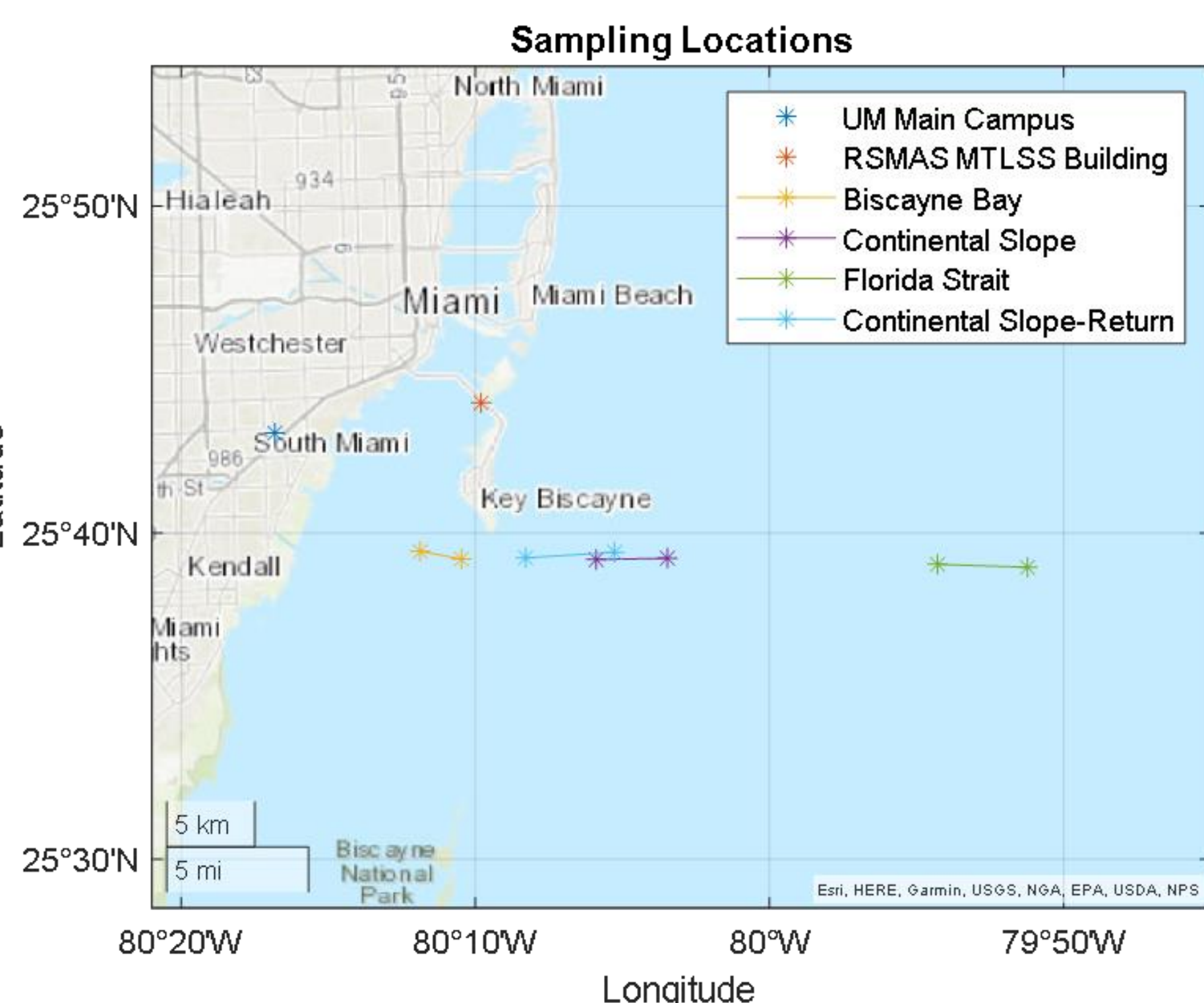
Sample Collection

- Red mangrove (*Rhizophora mangle*) leaves used for method development of cupric oxide (CuO) oxidation and gas chromatography tandem mass spectrometer (GC-MS) analysis
- RSMAS MTLSS building water used for method development of filtration of dissolved organic matter from seawater
- 4 surface water samples collected using underway system during Walton Smith cruise on November 13, 2021, representing Biscayne Bay, Continental Slope, and Florida Strait

Sample Preparation

- Leaves freeze-dried and crushed
- Dissolved organic matter (DOM) filtered from marine samples with Agilent Mega-BE C18 solid state extraction cartridges [4]

- Samples made basic with addition of sodium hydroxide (NaOH)
- Samples oxidized using CuO oxidation method and MARS microwave system [5,6]
- Samples acidified with hydrochloric acid (HCl) and liquid-liquid extraction (LLE) performed with ethyl acetate (EtOAc)
- Organic layer trimethylsilylated (TMS derivatized) with N,O-Bis(trimethylsilyl)trifluoroacetamide (BSTFA) to be amenable to GC-MS analysis
- Full ion scan of mangrove leaves used to identify retention times of each lignin phenol
- Selective Ion Monitoring (SIM) mode used to make GC-MS more sensitive to low concentration of lignin phenols in marine samples



Results

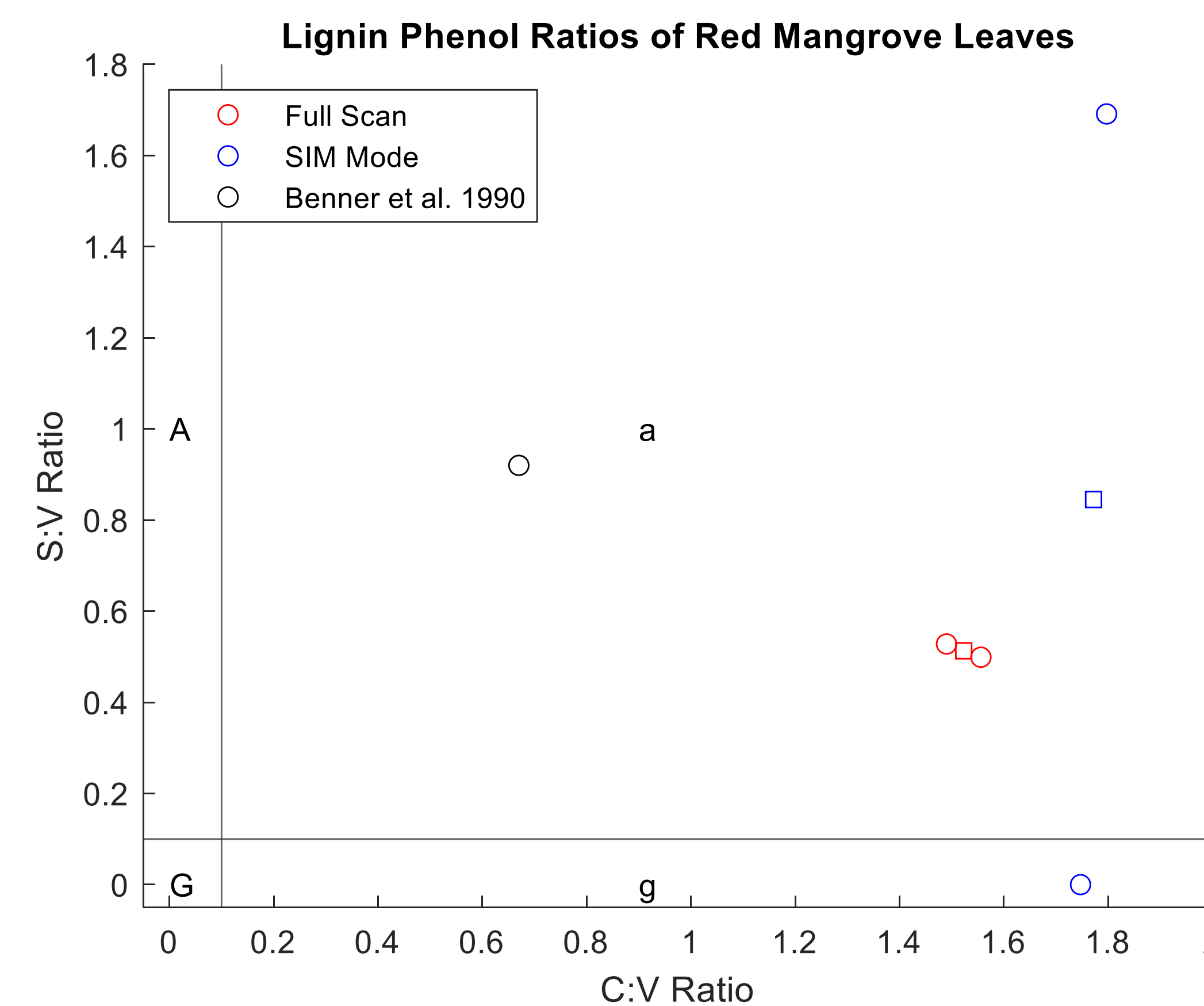
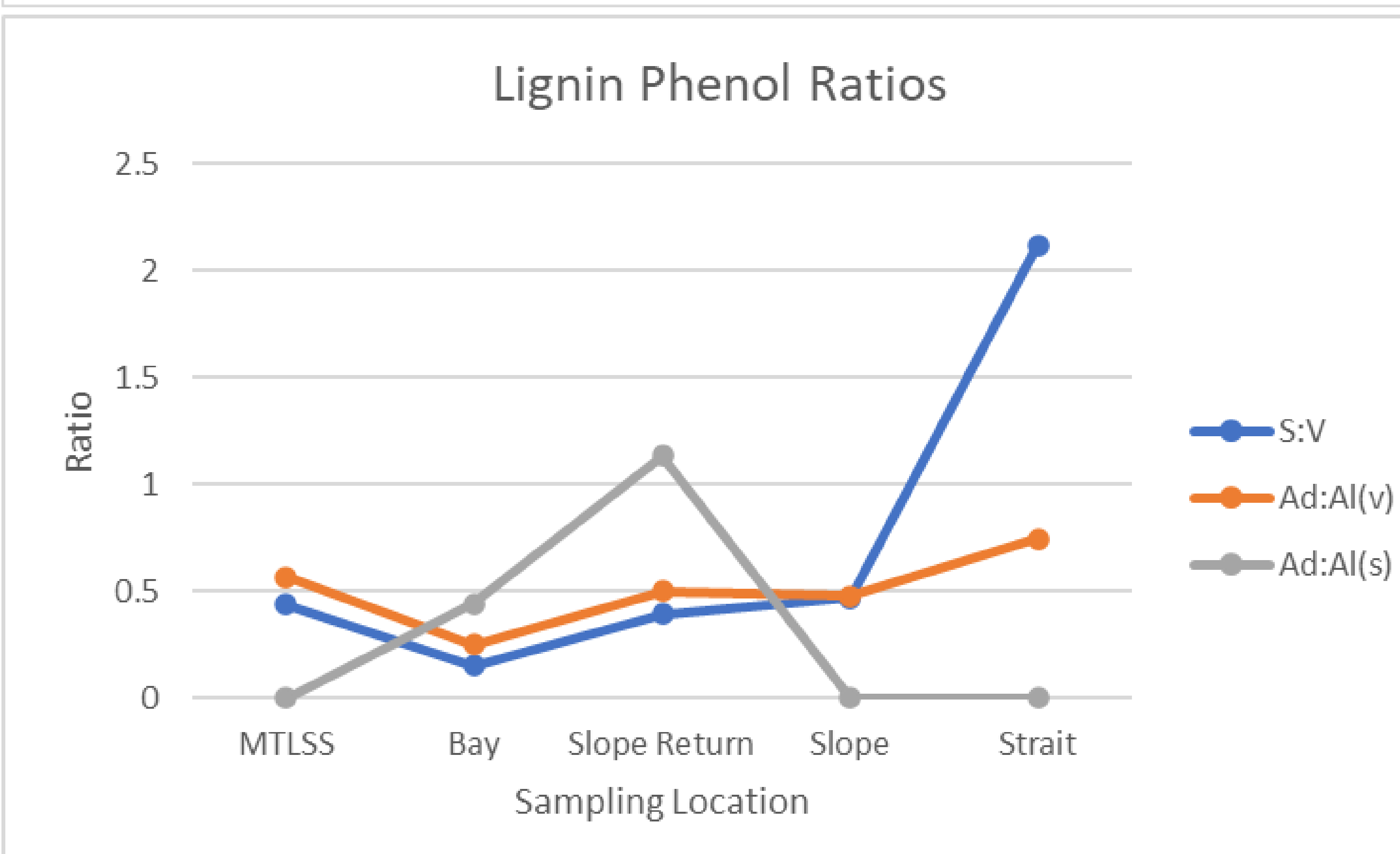
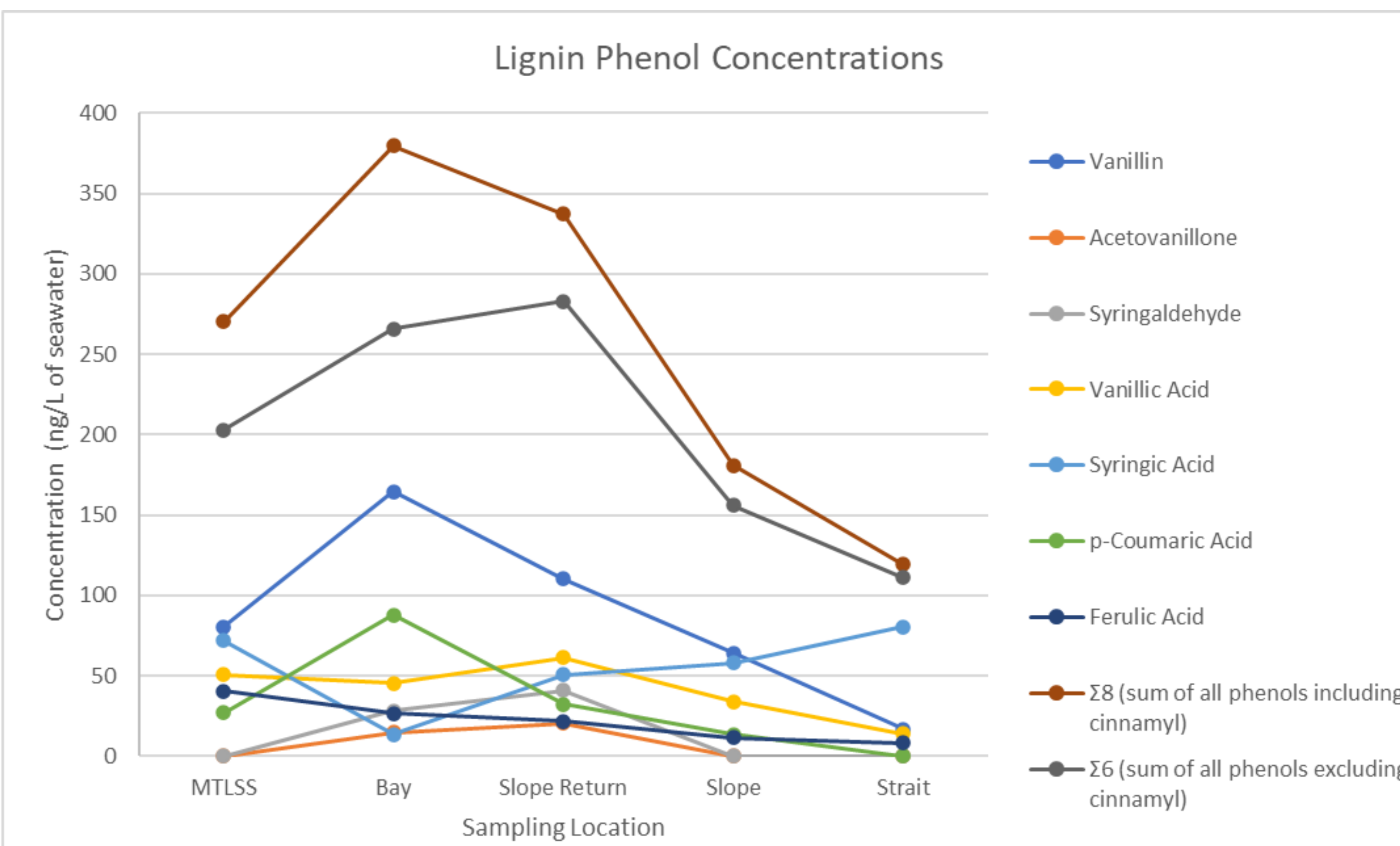


Figure (left) depicts S:V and C:V ratios of red mangrove leaf replicates and average (denoted by ■) for each ion acquisition method compared to Benner et al. [7]. A denotes woody angiosperm, G denotes woody gymnosperm, a denotes non-woody angiosperm, and g denotes non-woody gymnosperm.



Conclusion and Discussion

- Successfully performed lignin oxidation and detected lignin subunits
- Lignin phenol subunit yields were comparable or higher than to published data on similar materials (open-ocean marine DOM and mangrove leaves) [4,7]
- S:V and C:V ratio of leaves put them in range of non-woody angiosperm, which is accurate to material
- Exception is one replicate sampled in SIM mode, which may be resolved by sampling more replicates and optimizing dwell times for SIM mode
- Marine samples show general trend of decreasing lignin phenol concentration from Biscayne Bay to Florida Straits
- Large S:V ratio in Florida Strait compared to other marine samples suggest large local gymnosperm input into Biscayne Bay
- C4 plant material, which are all angiosperms, is transported greater distances due to association with smaller soil grain sizes [1]
- Increasing Ad:Al(v) and syringic acid concentration suggest more degraded lignin along transects, which was expected
- Overall, results appear to support hypothesis in suggesting terrigenous DOM in Florida Strait surface waters are allochthonous compared to being autochthonous in Biscayne Bay
- However, more replicates needed to properly determine if results are statistically significant

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