1	UTI	LITY OF SALT-MARSH FORAMINIFERA, TESTATE AMOEBAE AND BULK-SEDIMENT
2		$\delta^{13}$ C values as sea-level indicators in Newfoundland, Canada
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- 24 ABSTRACT
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We investigated the utility of foraminifera, testate amoebae and bulk-sediment  $\delta^{13}$ C measurements for 26 27 reconstructing Holocene relative sea level from sequences of salt-marsh sediment in Newfoundland, 28 Canada. Modern, surface sediment was collected along transects from low to supra-tidal elevations in 29 eastern (at Placentia) and western (at Hynes Brook and Big River) Newfoundland. Consistent with 30 previous work, low-diversity assemblages of foraminifera display an almost binary division into a higher 31 salt-marsh assemblage dominated by Jadammina macrescens and Balticammina pseudomacrescens and a 32 lower salt-marsh assemblage comprised of Miliammina fusca. This pattern and composition resembles 33 those identified at other high latitude sites with cool climates and confirms that foraminifera are sea-level indicators. The lowest occurrence of testate amoebae was at approximately mean higher high water. The 34 35 composition of high salt-marsh testate amoebae assemblages (*Centropyxis cassis* type, *Trinema* spp., Tracheleuglypha dentata type, and Euglypha spp.) in Newfoundland was similar to elsewhere in the 36 37 North Atlantic, but preservation bias favors removal of species with idiosomic tests over those with 38 xenosomic tests. The mixed high salt-marsh plant community in Newfoundland results in bulk surface-sediment  $\delta^{13}$ C values that are typical of C<sub>3</sub> plants, making them indistinguishable from freshwater 39 40 sediment. Therefore we propose that the utility of this proxy for reconstructing RSL in eastern North 41 America is restricted to the coastline between Chesapeake Bay and southern Nova Scotia. Using a simple, 42 multi-proxy approach to establish that samples in three radiocarbon-dated sediment cores formed between 43 the lowest occurrence of testate amoebae and the highest occurrence of foraminifera, we generated three 44 example late Holocene sea-level index points at Hynes Brook.

#### 45 **1. INTRODUCTION**

46 In eastern North America, quiet-water coastal environments are occupied by salt marshes that have a 47 robust and predictable relationship to tidal datums (e.g., Redfield, 1972). These ecosystems respond to 48 relative sea-level (RSL) rise by accumulating sediment to ensure that the active salt-marsh surface 49 maintains its tidal elevation, while increasing in absolute elevation relative to a fixed point of reference on 50 land. Consequently, buried units or sequences of salt-marsh sediment preserve a history of Holocene RSL 51 changes (e.g., Bloom and Stuiver, 1963, Stuiver and Daddario, 1963). Interrogating this sedimentary 52 archive to reconstruct RSL relies on the use of sea-level indicators to accurately establish the elevation of 53 dated paleomarsh surfaces with respect to contemporary and modern tidal datums (e.g., Woodroffe and 54 Barlow, 2015).

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56 Sea-level indicators are biological assemblages, chemical signatures or physical features with a known

57 relationship to tides. In salt-marshes, the most commonly used sea-level indicators are plants,

assemblages of microfossils such as foraminifera (e.g., Edwards and Wright, 2015, Scott and Medioli,

59 1978), diatoms (e.g., Zong and Horton, 1999, Zong and Sawai, 2015), or testate amoebae (e.g., Barnett et

al., 2016, Charman, 2015) and bulk-sediment geochemistry (e.g., Kemp et al., 2012b, Lamb et al., 2006).

61 The analogy between modern sea-level indicators and their paleo counterparts is the basis for estimating

62 the paleo tidal elevation at which a fossil sample formed and subsequently for reconstructing RSL. This

63 approach is reliant upon the availability of an empirical modern training set comprised of paired

observations that establish and quantify the relationship between a sea-level indicator and tidal datums.

65 Due to the influence of secondary environmental variables such as the prevailing climate, oceanographic

and salinity regimes, it is usually necessary to develop a modern training set in the region that will be

67 targeted for reconstructing RSL.

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69 Since deglaciation of the Laurentide Ice Sheet (by ~7000 years before present; BP, 1950 CE), the

70 principal cause of RSL change along the Atlantic coast of North America was ongoing and

spatially-variable glacio-isostatic adjustment (GIA). Earth-ice models predict that the hinge line between

regions experiencing recent RSL rise/fall from GIA occurs close to the Gulf of St. Lawrence (Figure 1;

e.g., Peltier, 2004). Newfoundland is therefore one of the most northerly regions to preserve a salt-marsh

record of RSL rise since the mid-Holocene, which makes it an important location for investigating drivers

of paleo-RSL change that vary with latitude such as ocean dynamics (Ezer et al., 2013, Levermann et al.,

76 2005, Yin and Goddard, 2013) and the fingerprint of Greenland Ice-Sheet melt (Mitrovica et al., 2011).

77 We build on previous investigations of foraminifera (Daly, 2002, Daly et al., 2007) to explore the utility

of foraminifera, testate amoebae and bulk-sediment  $\delta^{13}$ C values as independent sea-level indicators in

79 Newfoundland and to evaluate their use as part of a multi-proxy approach. This work will support future 80 efforts to reconstruct RSL using salt-marsh sediment. At three sites (Placentia on the Avalon Peninsula 81 and Hynes Brook and Big River on the Port-au-Port Peninsula; Figure 1) we collected surface sediment at 82 regular vertical increments to establish the relationship between each type of potential sea-level indicator 83 and local tidal datums. We demonstrate that for aminifera and testate amoebae are viable sea-level indicators (with some limitations), while bulk-sediment  $\delta^{13}$ C values have little utility in this region. An 84 85 example RSL reconstruction is produced by application of these sea-level indicators to three 86 radiocarbon-dated basal salt-marsh peats from Hynes Brook spanning the last ~2000 years. These 87 reconstructions demonstrate the utility of using a simple, multi-proxy approach to identify material that formed between the lowest occurrence of testate amoebae and the highest occurrence of foraminifera. 88

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#### 2. **REGIONAL SETTING**

91 We studied three sites in Newfoundland (Placentia, Hynes Brook and Big River; Figure 1) that were 92 identified from existing literature as having well-developed modern salt marshes and being underlain by 93 sediment (high salt-marsh peat) that was likely to yield a viable RSL reconstruction (e.g., Bell et al., 94 2005, Brookes et al., 1985, Daly, 2002, Daly et al., 2007, Wright et al., 2011, Wright and van de Plassche, 95 2001). In some cases, these earlier studies also included analysis of surface foraminiferal assemblages. 96 These sites experience a maritime climate characterized by short, cool summers (maximum average 97 monthly warm temperature of  $\sim 20$  °C in July) and winters in which the average high temperature for 98 January is approximately -2 °C (Figure 1). The prevailing oceanographic regime brings cold water (the 99 Labrador Current) to the sites resulting in an average annual sea-surface temperature of  $\sim 1 \degree C$  in the open 100 ocean.

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102 Placentia is located on the Avalon Peninsula in eastern Newfoundland (Figure 1), where salt marshes are 103 rare. The study site lies on the protected side of a progradational barrier system comprised of vegetated ridges and swales. It is indirectly connected to the ocean because the 'south arm' is closed off by a gravel 104 105 barrier and marine water reaches the site having first entered the back-barrier lagoon and fluvial system 106 through the open 'north arm'. This geomorphology results in a great diurnal tidal range (mean lower low 107 water, MLLW to mean higher high water, MHHW) of 0.91 m. The low gravel barrier directly in front of 108 the salt marsh is overwashed by high tides. Low salt-marsh environments are absent at the site, although 109 isolated stands of *Spartina alterniflora* are present and rooted in gravel rather than fine-grained sediment. 110 The high salt-marsh is a diverse, peat-forming community comprised of *Distichlis spicata*, *Spartina* 111 patens, Juncus geradi, Potentilla sp., Plantago maritima, Glaux maritima, and Carex glareosa. Within 112 the high salt-marsh zone these plants form a mosaic pattern and are frequently intermixed, although some 113 mono-specific stands are present. At the upland edge of the salt marsh is a near-vertical slope of

- 114 weathered bedrock that is sparsely vegetated by fir trees, pine trees and brambles. At this site, Daly
- 115 (2002) described for a in six surface sediment samples spanning an elevational range of ~0.25 m.
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117 On the Port-au-Port Peninsula salt marshes occupy three small incised valleys on the sheltered coast of 118 West Bay that are protected by a beach barrier/spit formation at their entrance (Figure 1). We recovered 119 samples from Hynes Brook and Big River, which both display a characteristic division of floral 120 communities into a low salt-marsh zone dominated by Spartina alterniflora (tall form) and a mixed high 121 salt-marsh mosaic comprised of variable combinations of Iris versicolor, Schoeneplectus spp., Spartina patens, Distichlis spicata, Plantago maritima, Triglochin maritima, Glyceria borealis, Glaux maritima, 122 Agrostis stolonifera, and Spartina alterniflora (short form). These species are frequently intermixed, 123 124 although some mono-specific stands are present, for example Iris versicolor is often the only plant present at the upper limits of tidal inundation (e.g., Daly, 2002). The great diurnal tidal range at Hynes Brook and 125 126 Big River is 1.06 m. At Hynes Brook, Daly (2002) described foraminifera in twelve surface sediment 127 samples spanning an elevational range of  $\sim 0.70$  m. Previous studies demonstrated that Hynes Brook is 128 underlain by sequences of high salt-marsh peat spanning the last ~3000 years (Bell et al., 2005, Brookes 129 et al., 1985, Daly, 2002, Daly et al., 2007).

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## 3. MATERIALS AND METHODS

#### 3.1 Modern sampling regime

We described the modern distribution of foraminifera, testate amoebae and bulk-sediment  $\delta^{13}$ C values from surface (0–1 cm) sediment samples collected along transects at each of our three study sites (Table 1). Sampling stations were positioned at regular (~5 cm) vertical intervals spanning the full height range occupied by salt-marsh vegetation to ensure even sampling of the environmental gradient of interest (elevation). The uppermost samples of each transect were recovered from supra-tidal environments to capture the highest occurrence of foraminifera (Wright et al., 2011) and the turnover of testate amoebae assemblages across the upper limit of marine influence (e.g., Barnett et al., 2016).

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Sample heights at each site were surveyed with a total station and are expressed relative to mean tide level (MTL). Local tidal datums were determined by comparing data from on-site water-loggers (corrected for the inverse barometer effect) with the closest tide gauge station operated by Fisheries and Oceans Canada. At Placentia, two water-loggers were installed to quantify the distortion of the tidal wave as it propagates into the back-barrier system. One logger measured water depths immediately adjacent to the salt-marsh site, while a second logger was deployed in the open bay beyond the gravel barrier (Figure 1c, d). High 147 and low tides captured by the water-logger in the bay match those measured by the neighboring tide

148 gauge at Argentia in timing and amplitude. We used hourly water-level measurements from Argentia

spanning the current tidal epoch (1983–2001) to define tidal datums following the definitions provided by

the National Ocean and Atmospheric Administration (NOAA). We then used the difference in water-level

151 measurements between the salt-marsh and open-bay water loggers to establish local tidal datums at

152 Placentia by scaling those previously defined for Argentia. For Hynes Brook and Big River, we used the

tidal datums reported in Wright et al. (2011), who employed a similar approach to the one outlined above

using the long-term tide-gauge measurements from Port aux Basques (Figure 1).

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# 3.2 Processing and analysis of surface sediment samples

Samples collected for foraminiferal analysis were placed into vials with buffered ethanol and stained with 157 158 rose Bengal to allow identification of individuals that were living at the time of collection (e.g., Figueira 159 et al., 2012). In the laboratory, each sample was washed over 500 µm and 63 µm sieves to isolate the 160 foraminifera-bearing fraction, from which a minimum of 100 dead individuals were counted in water 161 under a binocular microscope. We determined the highest occurrence of foraminifera to occur where the 162 concentration of dead tests decreased abruptly and above which there were insufficient individuals to 163 reasonably conclude that they represented a viable and *in situ* population. Species were identified through 164 comparison to published literature (e.g., Edwards and Wright, 2015, Scott and Medioli, 1980, Scott et al., 165 1981, Smith et al., 1984, Wright et al., 2011) and type slides from nearby regions. Different taxonomic 166 approaches to the classification of salt-marsh foraminifera exist, reflecting the extent to which 167 morphological variation is viewed as phylogenetic or ecophenotypic. While early work combined a range of forms within the taxon Trochammina macrescens, we differentiate between Jadammina macrescens 168 169 and Balticammina pseudomacrescens following (Daly, 2002), de Rijk (1995), Gehrels and van de Plassche (1999), Wright et al. (2011), in recognition of their spatially distinctive distributions (see 170 171 Edwards and Wright, 2015 for discussion). We also distinguished between Trochammina inflata and

172 Siphotrochammina lobata.

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At each sampling station  $\sim 2 \text{ cm}^3$  of surface sediment was collected and sealed in a bag for subsequent analysis of testate amoebae following the protocol developed for salt-marsh environments (Barnett et al., 2013). For each sample, 1 cm<sup>3</sup> of surface sediment was combined in 100 ml of deionized water with one tablet of *Lycopodium clavatum* to provide an exotic marker of known concentration during counting (Stockmarr, 1971). The material was heated at 80 °C for one hour on a hot plate and regularly stirred to aid disaggregation. After being left to soak for >12 hours, the material was wet sieved and the 300-63 µm fraction retained for counting. Samples were mounted onto glass microscope slides using deionized water and observed under 400x magnification. We counted 100 individual tests per sample. Where test

182 concentrations were <1500 tests per cm<sup>3</sup>, counting ceased after 100 *Lycopodium* spores were counted.

183 This point marked the lowest extent of testate amebae occurrence where counts per *Lycopodium* spore

decreased abruptly and below which there were insufficient individuals to reasonably conclude that they

represented a viable and *in situ* population. Our taxonomy is based on the systematic identification keys

186 of Charman et al. (2000) and Booth and Sullivan (2007). These keys were supplemented by published

187 literature pertaining to salt-marsh and littoral taxa (Cash et al., 1905, 1909, 1915, Charman et al., 2002,

188 Gehrels et al., 2006a, Golemansky, 1974, Golemansky and Todorov, 2004, 2007, Nicholls, 2007, 2009,

- 189 Ooms et al., 2015, Todorov et al., 2009).
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Representative sub samples of the surface material were analyzed in duplicate for  $\delta^{13}$ C,  $\delta^{15}$ N, total organic 191 192 carbon (%), and total nitrogen (%). Sediment samples were treated with 10% HCl to remove carbonate, rinsed to neutral pH with deionized water, and then dried at 50 °C and ground to a fine, homogenized 193 194 powder. The samples were analyzed using a Costech 4010 Elemental Analyzer interfaced with a Thermo 195 Delta V Plus stable ratio mass spectrometer (EA-IRMS) at the University of North Carolina Wilmington. 196 Isotopic composition is reported in standard  $\delta$  -per mil notation (‰) relative to the Vienna-Pee Dee 197 Belemnite (V-PDB) standard. Presented results are the average of the duplicate measurements. Repeated 198 analyses of USGS 40 and 41 glutamic acid standards indicate that the precision of these analyses is better than  $\pm 0.5$  ‰.  $\delta^{15}$ N, total organic carbon (%), and total nitrogen (%) 199

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## 3.3 Relative sea-level reconstruction

To investigate the utility of foraminifera, testate amoebae and bulk-sediment  $\delta^{13}$ C values for 202 reconstructing RSL in Newfoundland, we collected three cores of basal salt-marsh sediment along 203 204 transect HBM-2 at Hynes Brook (Figure 1g). The sediments were recovered using a Russian-type corer to 205 prevent compaction or contamination, and were stored in rigid plastic sleeves, wrapped in plastic and 206 refrigerated until processing. Each core comprised a single, 50-cm long sequence that captured the 207 transition from basal consolidated gray silty clay into an overlying dark-brown to black organic silt. One 208 half of each core was sliced into 1-cm thick contiguous samples for analysis of foraminifera, testate 209 amoebae and bulk-sediment  $\delta^{13}$ C. Processing of these samples followed the methods outlined for surface 210 samples with the exception of adding rose Bengal to the foraminiferal samples.

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212 The other half of each core provided material for radiocarbon dating which was extracted from the

sediment matrix and cleaned following Kemp et al. (2013c). We selected only samples that were

deposited on, or close to, a paleo marsh surface such as small, horizontal twigs and the shallow rhizomes

of identifiable salt-marsh plants. The samples were analyzed at the National Ocean Sciences Accelerator

- 216 Mass Spectrometry facility, where they underwent standard acid-base-acid pretreatment and  $\delta^{13}$ C was
- 217 measured directly on an aliquot of CO<sub>2</sub> collected during sample combustion. Reported radiocarbon ages
- 218 were individually calibrated using the Intcal13 dataset (Reimer et al., 2013) and we used the upper and
- lower  $2\sigma$  calibrated ages as the range of possible ages for the dated sample.
- 220
- 221 The lowest depth with a viable assemblage of foraminifera and material suitable for radiocarbon dating 222 was used to produce a sea-level index point from each core. We counted foraminifera and testate amoebae 223 and measured bulk-sediment  $\delta^{13}$ C values in additional core samples surrounding the dated level to ensure 224 that it was representative of the prevailing environmental conditions at the time of sediment deposition. 225 Relative sea level was calculated by subtracting reference water level (estimated from foraminifera, testate amoebae and bulk-sediment  $\delta^{13}$ C values) from measured sample elevation (e.g., Woodroffe and 226 227 Barlow, 2015). Vertical uncertainty for the reconstruction was calculated following Engelhart and Horton 228 (2012), where sample thickness was 0.01 m and we estimated a leveling error of  $\pm 0.05$  m, a benchmark 229 error of  $\pm 0.1$  m and an angle of coring error that was 1% of depth. Collectively, these cores record the 230 time-dependent transgression of the site by late Holocene RSL rise
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# 3.4 Statistical analysis

233 To objectively distinguish and describe regional microfossil groups using Partitioning Around Medoids 234 (PAM; e.g., Kaufman and Rousseeuw, 1990, Kemp et al., 2012a), we combined the results from all sites 235 into a single dataset of foraminifera and a single dataset of testate amoebae. Species counts were expressed as percentages and samples that yielded zero or low counts were excluded prior to the analysis. 236 We determined the number of distinct assemblages within each dataset using the maximum average 237 silhouette width calculated for 2 to 20 groups. Silhouette widths close to 1 (maximum possible value) 238 239 indicate that a sample was classified appropriately, while values close to -1 (minimum possible value) 240 reflect poor classification. This metric aims to minimize within group variance, while maximizing 241 differences among groups (Rousseeuw, 1987). To investigate the ecological plausibility of our RSL 242 reconstructions, we measured the (Bray-Curtis) dissimilarity between each microfossil assemblage in the 243 core samples to their closest modern analog in the modern training sets of foraminifera and testate amoebae. If the measured dissimilarity was less than the 20<sup>th</sup> percentile of dissimilarity measured among 244 all possible pairings of modern samples then we deemed the core sample to have an acceptable modern 245 246 analog (e.g., Jackson and Williams, 2004, Watcham et al., 2013). Core samples with a measured 247 minimum dissimilarity that exceeded this threshold were deemed to lack a modern analog. 248

To facilitate comparison among sites with different tidal ranges, we standardized elevation using a variant
of the standardized water level index (SWLI) of Horton (1999). We used MTL as the lower datum (SWLI

= 100) and the highest occurrence of foraminifera as the upper datum (SWLI = 200) because Wright et al.

252 (2011) demonstrated that this approach improved assemblage alignment in higher marsh settings where

- the relationship between elevation and inundation is often nonlinear.
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## **4. Results**

# 4.1 Foraminifera

257 The highest occurrence of foraminifera at Placentia was 0.75 m above MTL (PLA-B station 11) and samples above this elevation on all three transects were devoid of foraminifera (Figure 2). Each of the 258 259 three sampled transects displayed a similar distribution of foraminifera. On transect PLA-A samples from 260 0.41–0.70 m were dominated by Jadammina macrescens and Balticammina pseudomacrescens (39-98% 261 when combined). Trochammina inflata was an important part of the assemblage (45-73%) at 0.22–0.46 m 262 MTL and *Miliammina fusca* characterized samples below 0.30 m MTL (more than 25–50%). On transect 263 PLA-B Jadammina macrescens and Balticammina pseudomacrescens were the dominant species above 264 0.69 m MTL (up to 37% and 62% respectively and up to 96% when combined), while Trochammina inflata comprised 62-77% of individuals at 0.51–0.63 m MTL. Miliammina fusca was rare (<4%), but the 265 266 lowest sample on this transect was above MHHW. On transect PLA-C a single sample at 0.68 m MTL 267 yielded a foraminiferal assemblage that was 34% Jadammina macrescens and 64% Balticammina 268 pseudomacrescens. Trochammina inflata comprised 32–68% of individuals from 0.34 m to 0.55 m MTL. 269 Assemblages from 0.22 m to 0.43 m MTL included an average of 28% Miliammina fusca. 270 Haplophragmoides spp. were absent on all three transects at Placentia. 271 Foraminifera along two transects at Hynes Brook (HBM-0 and HBM-2; Table 1) were described by 272 273 Wright et al. (2011). Results from a third transect (HBM-1) described here for the first time show a very

similar distribution of foraminifera. The highest occurrence of foraminifera among all three transects was

at 0.90 m MTL (HBM-1 station 9; Figure 3), which is 0.05 m higher than estimated by Wright et al.

276 (2011) from the smaller dataset. On transect HBM-1 Jadammina macrescens (up to 63%) and

- 277 Balticammina pseudomacrescens (up to 83%) made up the majority of individuals (57–100% when
- combined) at 0.50–0.90 m MTL, while *Miliammina fusca* dominated (44–86%) samples from 0.15 m to
- 279 0.45 m MTL. *Tiphotrocha comprimata* and *Haplophragmoides manilaensis* were present on HBM-1.
- 280 Trochammina inflata was absent or rare (maximum abundance of 7.8%) on the three transects from
- 281 Hynes Brook.

283 The distribution of foraminifera at Big River is largely consistent among transects and there is also a high 284 degree of similarity to those at Hynes Brook. The highest occurrence of foraminifera among all three 285 transects was 0.88 m MTL (BRM-2 station 7; Figure 3). On transect BRM-0 elevations above MHW 286 (0.31–0.86 m MTL) were dominated by Jadammina macrescens (up 93%) and Balticammina 287 pseudomacrescens (up to44%; 58-100% when combined), while Miliammina fusca was the characteristic 288 species (53-82%) in assemblages at -0.09 m to 0.26 m MTL. *Tiphotrocha comprimata* was present on this 289 transect (up to 39%). On transect BRM-1, assemblages of foraminifera from 0.10 m to 0.85 m MTL were 290 dominated by Jadammina macrescens (up to 40%) and Balticammina pseudomacrescens (up to 85%; 291 49-100% when combined). Miliammina fusca made up 31-66% of individuals at 0.05-0.20 m MTL. On 292 transect BRM-2 elevations from 0.33 m to 0.88 m MTL are characterized by high abundances of 293 Jadammina macrescens (up to 62%) and Balticammina pseudomacrescens (up to 60%; 41-86% when 294 combined). Two samples at 0.23 m and 0.28 m MTL are dominated by Miliammina fusca (81% and 66% 295 respectively). The abundance of *Haplophragmoides manilaensis* at Big River demonstrates variability 296 among transects (maximum abundance of <5% on BRM-0, but up to 49% on BRM-1 and 37% on 297 BRM-2) as does Trochammina inflata (maximum abundance of 29% on BRM-0, 2.5% of BRM-1 and 6% 298 on BRM-2), which may reflect a patchy distribution (e.g., Kemp et al., 2011, Swallow, 2000) and/or 299 within site variability of secondary environmental factors such as salinity or sediment texture that were 300 not measured. Modern foraminifera data are presented in the supporting appendix.

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#### *4.2 Testate amoebae*

At Placentia the lowest occurrence of testate amoebae occurred below MHHW at 0.30 m MTL (PLA-A station 23) with lower samples yielding fewer than 9 individuals after counting a sufficient volume of material to recognize 100 *Lycopodium* spores (Figure 4). Samples above 0.63 m MTL were dominated by *Tracheleuglypha dentata* (average 16%) and *Euglypha rotunda* type (average 26%). Because the highest sample on the transect included abundant testate amoebae, the upper limit of this assemblage cannot be estimated. At 0.30–0.55 m MTL the most abundant species was *Centropyxiella* type (average 33% and up to 63%). Other common species on PLA-A included *Pseudocorythion* type (up to 30%) and

310 *Pseudohyalosphenia* type (up to 22%).

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312 At Hynes Brook testate amoebae were sparse (fewer than 10 individuals per 100 Lycopodium spores) at

- stations 10–15 and the lowest occurrence of testate amoebae was established at 0.70 m above MTL
- (Figure 4), which is slightly above MHHW. At elevations above 0.98 m above MTL, the most common
- species are *Trinema lineare* type (17–26%) and *Trinema enchelys* type (23–34%). The samples between
- 316 0.74 m MTL and 0.93 m MTL were characterized by an increased abundance of *Euglypha rotunda* type

317 (20% in each of the three samples). The lowest viable assemblage of testate amoebae includes more than
318 40% *Centropyxis cassis* type.

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320 At Big River, the lowest occurrence of testate amoebae was recorded at 0.52 m above MTL (0.01 m 321 above MHHW) in the sample from BRM-0b station 11 (Figure 5). At elevations from 0.56 m to 1.51 m 322 MTL, assemblage composition resembles those identified at Hynes Brook, with the most abundant 323 species being Trinema lineare type (up to 25%), Trinema enchelys type (up to 30%), and Euglypha 324 rotunda type (up to 27%). The samples at 0.52 m and 0.57 m MTL are characterized by increased 325 abundance of *Centropyxis cassis* type (>34%) and the appearance of *Arcella catinus* (up to 35%) at the 326 lower limit of testate occurrence. Modern testate amoebae data are presented in the supporting appendix. 327 328 4.3 Regional-scale microfossil distributions

329 We combined the modern foraminifera results from Placentia, Hynes Brook and Big River into a single 330 dataset comprised of 134 samples (Figure 5). Analysis of this dataset using PAM indicates two plausible 331 ways to partition the samples. A maximum average silhouette width is returned by dividing the dataset 332 into two groups based on the relative abundance of Miliammina fusca versus Jadammina macrescens and 333 Balticammina pseudomacrescens. This pattern describes the universal distinction between low and high 334 salt-marsh samples. A more nuanced classification is possible by partitioning the regional-scale dataset 335 into five groups. Group 1 is dominated by Jadammina macrescens with correspondingly low occurrences 336 of Balticammina pseudomacrescens. This group is present at Hynes Brook and Big River, but is absent 337 from Placentia. Group 2 is dominated by Jadammina macrescens and Balticammina pseudomacrescens 338 and was present at all sites. Group 3 is a diverse high salt-marsh assemblage in which *Tiphotrocha* 339 comprimata and Haplophragmoides manilaensis occur alongside Jadammina macrescens and 340 Balticammina pseudomacrescens. This group was present at each of the three study sites. Group 4 is 341 comprised of low salt-marsh assemblages characterized by high abundances of Miliammina fusca and is 342 absent from Placentia due to the lack of a well-developed low salt-marsh environment at the site today. 343 Group 5 is dominated by samples from Placentia and captures the uniquely high relative abundances of 344 Trochammina inflata in these transects. 345

We combined the samples from Placentia, Hynes Brook and Big River to create a regional-scale dataset of testate amoebae from 43 modern samples (Figure 5). Partitioning around medoids showed that the

maximum average silhouette width (0.38) occurred when the dataset was divided into two groups and no

- finer-scale sub-division is warranted. Group 1 (30 samples) was represented by *Trinema lineare* type
- 350 (13%), *Trinema enchelys* type (18%) and *Euglypha rotunda* type (20%). This group was present at all

three sites. In contrast, Group 2 (13 samples) only occurred at Placentia and in a single sample from
Hynes Brook (HBM-0b station 11). It is characterized by *Arecella catinus* (discoides group), *Centropyxiella* type (25%) and *Pseudohyalosphenia* type (Ooms; 22%).

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# 4.4 Bulk-sediment $\delta^{13}C$ values

Despite differences in floral communities and microfossil assemblages, measured bulk-sediment  $\delta^{13}$ C 356 357 values from each of the three study sites exhibit no consistent trend with station elevation (Figures 2 & 5) 358 and largely fall in the range of -24% to -29% (data are provided in the supporting appendix). The 359 exceptions to this pattern are three stations from the middle of transect PLA-A at Placentia, which 360 returned values of -21% (the maximum typically associated with C<sub>3</sub> vegetation) and -15% to -17%, which is characteristic of  $C_4$  terrestrial plants and also some marine algae (e.g., Lamb et al., 2006). At the 361 362 time of sampling these stations were vegetated predominantly by *Spartina patens* (a  $C_4$  plant) and *Juncus* 363 geradii (a  $C_3$  plant). Despite the presence of  $C_4$  plant species on the salt marshes at each of our study sites (e.g. Spartina patens and Distichlis spicata) the pervasive bulk-sedimentary signatures are those 364 365 reflecting  $C_3$  plant material (Lamb et al., 2006).

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#### 4.5 Radiocarbon dates and microfossil assemblages

368 We collected three cores from Hynes Brook that were positioned to capture the transition from 369 coarse-grained clastic sediment to basal organic sediment at different elevations marking the 370 time-dependent transgression of RSL during the late Holocene (Figure 6). In core HBM-C102 (48.60133 371 °N; 58.94578 °W) a radiocarbon date from 2.11 m yielded a calibrated age of 2718–2489 years BP ( $2\sigma$ 372 range; Table 2). Foraminifera from the dated interval are comprised almost exclusively of Jadammina 373 macrescens and Balticammina pseudomacrescens. The most abundant testate amoebae in these samples were Centropyxis cassis type (63–78%) and Difflugla lucida type (10–12%). In core HBM-C103 374 (48.60135 °N; 58.94575 °W) a radiocarbon date from 0.99 m yielded a calibrated age of 1825–1713 years 375 376 BP ( $2\sigma$  range; Table 2). For a from the dated interval are comprised almost exclusively of 377 Jadammina macrescens and Balticammina pseudomacrescens. The most abundant testate amoebae in 378 these samples were *Centropyxis cassis* type (64–62%) and *Centropyxis platystoma* type (14–34%). In 379 core HBM-C104 (48.60133 °N; 58.94584 °W) a radiocarbon date from 1.28 m yielded a calibrated age of 380 1736–1612 years BP (2σ range; Table 2). Foraminifera from the dated interval are 100% Jadammina 381 macrescens and Balticammina pseudomacrescens. The most abundant testate amoebae in these samples 382 were Centropyxis cassis (54–70%), Centropyxis platystoma (6–14%) and Centropyxis delicatula-ecornis 383 (up to 16%). In each core the consistency of microfossil assemblages indicates that no significant

environmental change took place over the period of time represented by the counted samples.

Bulk-sediment  $\delta^{13}$ C measurements from all of the samples in each core yielded values of approximately – 26 ‰. All assemblages of foraminifera had good modern analogs, while all assemblages of testate amoebae lacked modern analogs due to the high abundance of *Centropyxis cassis* in core samples (54– 78%) compared to the modern dataset (maximum of 43%).

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390 **5. DISCUSSION** 

# 5.1 Distribution of salt-marsh foraminifera

Some of the earliest investigations into the use of salt-marsh foraminifera as sea-level indicators were 392 393 conducted in Nova Scotia (Scott and Medioli, 1978, 1980), where intertidal transects revealed a characteristic vertical zonation of distinct assemblages. Shallow sub-tidal environments included large 394 395 numbers of calcareous species that became scarce in low salt-marsh assemblages dominated by the 396 agglutinated species Miliammina fusca. High salt-marsh environments were dominated by Jadammina 397 macrescens with Trochammina inflata and Tiphotrocha comprimata. A monospecific zone of Jadammina 398 macrescens at the transition from salt marsh to upland occupied the narrowest range of elevation and was 399 consequently deemed to be the assemblage that could produce the most precise RSL reconstruction. Since 400 this work, the utility of salt-marsh (and mangrove) foraminifera as sea-level indicators has been 401 reinforced by recognition of vertically-zoned assemblages at sites around the world under a wide range of 402 climate, salinity and tidal conditions (e.g., Barbosa et al., 2005, Hayward et al., 1999, Horton and 403 Edwards, 2006, Horton et al., 2005, Leorri et al., 2008, Patterson et al., 2005, Scott et al., 1996, Spencer, 404 2000). However, the composition of high salt-marsh assemblages in particular varies among (and within) 405 regions, as do the specific elevation of boundaries between zones (e.g., Edwards and Wright, 2015, Kemp 406 et al., 2009, Wright et al., 2011). This pattern necessitates using an appropriate training set to provide 407 adequate modern analogs for interpreting assemblages preserved in the sedimentary record (e.g., Edwards et al., 2004). Since surface and sub-surface assemblages at a site may differ due to environmental change 408 409 through time, the search for appropriate modern analogs commonly requires compilation of data from 410 multiple locations (e.g., Horton and Edwards, 2005). As part of this process, new insights into the ecology 411 of salt-marsh foraminifera can be gained that may, in turn, improve the quality of foraminifera-based RSL 412 reconstructions.

413

414 To establish the regional-scale relationship between assemblages of salt-marsh foraminifera and tidal

415 elevation in Newfoundland we identified five distinctive groups of foraminifera using PAM and by

416 standardizing sample elevations to account for differences in tidal range among sites (Figures 5 and 7).

417 The major faunal turnover occurs at ~140 SWLI and is characterized by a transition from a low

418 salt-marsh assemblage dominated by *Miliammina fusca* (Group 4; 86–140 SWLI) to spatially-variable

419 high salt-marsh assemblages in which *Jadammina macrescens* (Group 1; 134–200 SWLI), *Balticammina* 

- 420 pseudomacrescens (Group 2; 133–200 SWLI), or Trochammina inflata (Group 5; 129–184 SWLI) is the
- 421 dominant species. Group 3 (111–187 SWLI) straddles the boundary between low and high salt-marsh

422 assemblages and is characterized by greater species diversity including the presence of *Tiphotrocha* 

- 423 *comprimata* (all three sites) and *Haplophragmoides manilaensis* (absent at Placentia and rare in parts of
- 424 Big River).
- 425

426 Significantly, Groups 1 and 2 extend over the same vertical range and either may characterize the 427 uppermost salt-marsh samples. Consequently, when considered as tools for RSL reconstruction, the 428 distinction between *Jadammina macrescens* and *Balticammina pseudomacrescens* provides little or no 429 improvement of precision or accuracy over the 'lumping' of both species into a single taxon. In 430 Newfoundland, since no other groups extend to the upper limit of marine influence, relative abundances 431 of *Jadammina macrescens* and/or *Balticammina pseudomacrescens* greater than ~80% are indicative of 432 accumulation in the highest salt-marsh environment.

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434 Group 5 is dominated by *Trochammina inflata* and (with the exception of a single sample at Big River) is 435 only present at Placentia, suggesting particular suitability to environmental conditions at that site. High 436 abundances of Trochammina inflata coincide with an absence or relative scarcity of Balticammina 437 pseudomacrescens, which may reflect differing environmental preferences or competitive exclusion. 438 Since all of our study sites share a similar atmospheric and oceanographic climate (Figure 1), it is 439 tempting to ascribe the observed distribution of Group 5 to a prevailing regime of lower salinity at 440 Placentia. The geomorphology at Placentia restricts direct exchange of water with the ocean and the site's 441 position in the south arm likely produces a water mass with lower salinity than at Hynes Brook and Big 442 River where there is direct access to ocean water and little fluvial input. Furthermore, the steep slopes at 443 the rear of the Placentia salt marsh cause freshwater to drain directly onto the marsh and at the time of 444 sample collection we observed several springs at the rear of the marsh that serve to further dilute the 445 salinity at higher tidal elevations. However, Trochammina inflata was not noted elsewhere as a 446 low-salinity indicator. In fact, Balticammina pseudomacrescens and Haplophragmoides spp. are more 447 typically linked to low-salinity conditions (de Rijk and Troelstra, 1997, Edwards and Wright, 2015). 448 Therefore further work is necessary to distinguish the relative importance of secondary environmental 449 variables as a cause of among-site and among-transect variability in foraminiferal assemblages. 450

The pattern of zonation that we observed at Hynes Brook and Placentia is similar to that described byDaly (2002), who recognized three groups of foraminifera from a total of 29 surface sediment samples

453 collected at four Newfoundland salt marshes (Hynes Brook, St. Paul's Inlet, Village Cove and Placentia; 454 Figure 1). The three groups of Daly (2002) are all from high salt-marsh environments because sampling 455 did not extend below mean high water (MHW). The dominant species in these groups were Jadammina 456 macrescens with Balticammina pseudomacrescens, Jadammina macrescens and Balticammina 457 pseudomacrescens. Assemblages of salt-marsh foraminifera in Newfoundland closely resemble those 458 described at sites in the Canadian Maritimes including, Prince Edward Island (Scott et al., 1981), Nova 459 Scotia (Scott and Medioli, 1978, 1980, Smith et al., 1984), New Brunswick (Patterson et al., 2004) and 460 the Magdalen Islands (Barnett et al., 2016). A similar distribution is present in Maine, USA (Gehrels, 461 1994). We conclude that salt-marsh foraminifera from our three study sites (and others; e.g., Daly, 2002, Daly et al., 2007) in Newfoundland conform to the pattern of vertical zonation observed in surrounding 462 regions and confirm that they are viable sea-level indicators in this region. A notable characteristic of the 463 464 for a semblages from Newfoundland is their low species diversity (e.g., Daly, 2002; Figure 7), 465 which results in bipartite division of the salt marshes into near-monospecific high and low salt-marsh 466 assemblages. This pattern is also typical at other high latitude sites in the northern hemisphere that 467 experience a cold maritime climate such as arctic Norway, (Barnett et al., 2015), Iceland (Gehrels et al., 468 2006b), the Aleutian Islands of Alaska (Kemp et al., 2013a), Denmark (Gehrels and Newman, 2004) and 469 the White Sea (Russia; Kemp et al., Submitted). These low-diversity assemblages stand in contrast to 470 those from warmer climates along the U.S. Atlantic coast where several studies reported a larger diversity 471 of species at single sites, but also marked variability in the composition of high salt-marsh assemblages 472 among sites (e.g., Goldstein and Frey, 1986, Kemp et al., 2009, Kemp et al., 2013d, Wright et al., 2011). 473 We contend that these differences among regions arise from the climate regime, where cold air and ocean 474 temperatures limit the diversity of high salt-marsh foraminiferal assemblages to a handful of species, 475 namely Jadammina macrescens and Balticammina pseudomacrescens. Additional systematic work is 476 needed to identify the specific ecological conditions (e.g. water temperature, seasonal air temperature, 477 frequency and duration of freezing) that drive this geographic gradient in species distributions. If it can be 478 adequately understood then temporal changes in salt-marsh foraminiferal assemblages may yield insight 479 into past climates as well as RSL change.

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#### 5.2 Distribution of testate amoebae

Intertidal testate amoebae were initially identified in Atlantic Canada (Medioli and Scott, 1983), often in
samples of salt-marsh sediment being analyzed for foraminifera (Scott et al., 1983, Scott, 1977, Scott and
Martini, 1982), but it was not until the late 1990s that intertidal environments were systematically
sampled for testate amoebae in order to establish their utility as sea-level indicators (Charman et al.,
1998). Subsequent studies demonstrated that the lowest occurrence of testate amoebae in salt marshes

487 typically occurs at, or slightly below, high tidal datums such as mean high water spring tide or MHHW at

- 488 mid-latitude sites in the northern hemisphere (Barnett et al., 2013, Gehrels et al., 2006a, Gehrels et al.,
- 489 2001). Similarly, the concentration of testate amoebae in surface sediment samples from Newfoundland
- 490 decreased abruptly near MHHW. Typical test concentrations above MHHW were ~2000 to ~5000 tests
- 491 per cm<sup>3</sup>, but below MHHW counting statistically significant numbers of testate amoebae became
- 492 unviable. This pattern occurs because few taxa are tolerant of the increasingly brackish conditions
- 493 experienced below MHHW (Patterson and Kumar, 2002). Therefore the presence/absence of viable
- 494 testate amoebae populations is a useful paleo-ecological constraint for establishing if sediment samples
- 495 from a core accumulated above/below the lowest occurrence of testate amoebae that occurs close to
- 496 MHHW, but whose specific elevation must be established from an appropriate modern training set.
- 497

498 Using PAM, we identified two distinct groups of testate amoebae in Newfoundland (Figure 5, 7). Group 1 499 is characterized by high abundances of Trinema enchelys type, Trinema lineare type, Euglypha rotunda 500 type and *Centropyxis cassis* type (Figure 5). This group occurred at all sampled elevations at Hynes 501 Brook and Big River and in the uppermost samples at Placentia. Consequently, it spans a minimum 502 elevational range of 126–269 SWLI because its lower limit is constrained, but its upper limit is not. These 503 taxa also dominated similar environments from the nearby Magdalen Islands in the Gulf of St Lawrence 504 (Barnett et al., 2016) and are widespread in salt marshes throughout the North Atlantic (Barnett et al., 505 2013, Charman et al., 2002, Gehrels et al., 2006a, Gehrels et al., 2001, Ooms et al., 2015), suggesting that 506 certain intertidal taxa may be cosmopolitan in and around salt marshes, although a synthesis of existing 507 studies, datasets and taxonomies would benefit the development of this proxy for future sea-level 508 research. Group 2 occurs at Placentia, but not Hynes Brook or Big River. It is comprised of 509 *Centropyxiella* type, *Pseudocorythion* type, *Pseudohyalosphenia* spp. and *Corythionella* type (Figure 5) and exists at elevations from 140-187 SWLI (its upper and lower limits are constrained by the modern 510 511 dataset). Centropyxiella type was also common at comparable elevations in the Magdalen Islands 512 (Barnett et al., 2016), the UK (Charman et al., 2002) and Norway (Barnett et al., 2013), but sometimes 513 under different names because the nomenclature for salt-marsh testate amoebae is based on a wide diversity of literature and has evolved over recent decades. Differences in the lowest occurrence of testate 514 515 amoebae and assemblage composition between sites in Newfoundland probably occurs in response to 516 secondary environmental variables such as salinity, pH and sediment composition (Barnett et al., 2016, Charman et al., 2002). The geomorphological setting and freshwater input to the marsh at Placentia is the 517 518 most likely cause of the differences between sites. Lower salinity as a result of runoff and reduced 519 penetration of ocean waters at Placentia would provide favorable conditions for testate amoebae at lower

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# 5.3 Bulk-sediment $\delta^{13}C$ values as a sea-level indicator

524 Salt-marsh plants are sea-level indicators because the varied tolerance of species to the frequency and 525 duration of tidal inundation results in a systematic pattern of zonation, where low and high salt-marsh 526 communities at the same site are vegetated by different species (e.g., Eleuterius and Eleuterius, 1979, 527 Johnson and York, 1915, Redfield, 1972). However, the composition of these zones can vary among sites 528 as a result of secondary environmental factors such as climate and salinity. On the U.S. Atlantic coast for 529 example, high salt-marsh environments are typically vegetated by Spartina patens and Distichlis spicata at locations north of Chesapeake Bay, while Juncus roemerianus occupies this ecological position at 530 531 locations to the south (Eleuterius, 1976), until mangroves replace salt marshes in southern Florida (Figure 1).

elevations than at the other sites. Increased runoff can also be an influence on pH and sediment substrate

that may be additional factors influencing the testate amoebae assemblages (e.g., Charman et al., 2002).

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534 As an adaptation to regular submergence, some salt-marsh plants (e.g., Spartina spp., Distichlis spicata) utilize the C<sub>4</sub> (Hatch-Slack) photosynthetic pathway in which fractionation against atmospheric <sup>13</sup>CO<sub>2</sub> in 535 favor of  ${}^{12}CO_2$  is less pronounced than in C<sub>3</sub> (Calvin-Benson) plants such as *Juncus* spp. and the majority 536 537 of terrestrial species that live above the highest reach of tides. As a result,  $\delta^{13}$ C values measured in C<sub>4</sub> 538 plants are usually less depleted/negative than -17% compared to the VPDB standard, while C<sub>3</sub> plants 539 return more depleted/negative values between -21‰ and -32‰ (e.g., Lamb et al., 2006). In the organic 540 salt-marshes of eastern North America, the primary source of sediment is the in situ accumulation of 541 above and below ground biomass from the surface plant community and bulk sediment  $\delta^{13}$ C values reflect the dominant plant community at the time of deposition (e.g., Chmura and Aharon, 1995). 542 543 Post-depositional processes (principally preferential biodegradation of cellulose over lignin) cause 544 bulk-sediment  $\delta^{13}$ C values to differ slightly, but systematically, from the parent plant material (e.g., Benner et al., 1991, Benner et al., 1987, Ember et al., 1987, Haines, 1976), although this change is not 545 546 large enough to prevent distinction between C<sub>3</sub> and C<sub>4</sub> plants. The use of bulk-sediment  $\delta^{13}$ C values as a 547 sea-level indicator is reliant upon salt-marsh environments being dominated by C<sub>4</sub> plants such as *Spartina* 548 patens and Spartina alterniflora, which enables them to be readily and objectively distinguished from the 549 surrounding C<sub>3</sub>-dominated freshwater upland environments. For example, peat-forming communities 550 between MTL and MHHW in New Jersey are vegetated almost exclusively by Spartina spp. and Distichlis spicata resulting in bulk-sediment  $\delta^{13}$ C values that are less depleted than -18.9%, while 551 elevations above MHHW (including those without marine influence) had bulk-sediment  $\delta^{13}$ C values more 552 553 depleted than -22‰ (Kemp et al., 2012b). However, sediment more depleted than -22‰ in which

554 for aminifera are present formed between MHHW and the highest occurrence of for aminifera. This pattern

- enabled Kemp et al. (2013b) to reconstruct RSL using bulk-sediment  $\delta^{13}$ C values, in which the precision
- of a multi-proxy approach employing for a minifera and  $\delta^{13}$ C values was up to 60% better than using
- 557 foraminifera alone (Cahill et al., 2016). In contrast, a similar study in North Carolina concluded that
- 558 bulk-sediment  $\delta^{13}$ C values could not be objectively used as a sea-level indicator because the monospecific
- high salt-marsh zone is vegetated by the C<sub>3</sub> species *Juncus roemerianus*, making it indistinguishable from
- 560 a freshwater upland (Kemp et al., 2010).
- 561

562 In Newfoundland bulk-sediment  $\delta^{13}$ C values cannot objectively distinguish between material deposited in 563 a salt-marsh environment and material from the surrounding freshwater uplands that does not have a systematic relationship to tidal datums (Figures 2 and 4). We therefore conclude that bulk-sediment  $\delta^{13}$ C 564 565 values have no utility as sea-level indicators in Newfoundland. Although this is a negative finding for the 566 potential to reconstruct RSL in our study region, it does help to refine the geographic range of locations where bulk-sediment  $\delta^{13}$ C values could be utilized. Eleuterius (1976) found that 49–77% of salt-marsh 567 568 area in North Carolina was vegetated by Juncus roemerianus ( $C_3$  species) compared to <10% in Virginia 569 and Maryland and <0.1% in Delaware, where the high salt-marsh zone is almost exclusively comprised of 570 Spartina patens, Spartina alterniflora (short form) and Distichlis spicata (all C<sub>4</sub> species). Therefore we 571 propose that the southern limit for using bulk-sediment  $\delta^{13}$ C values as a sea-level indicator on the Atlantic 572 coast of North America is close to Chesapeake Bay (Figure 1). On the U.S. Atlantic coast most high 573 salt-marsh environments in New England are solely vegetated by C<sub>4</sub> plants (e.g., Niering and Warren, 574 1980, Redfield, 1972). Although Juncus geradii becomes a common occurrence in Maine (e.g., Gehrels, 575 1994, Johnson et al., 2007) and southern Nova Scotia (e.g., Gordon Jr et al., 1985, Pielou and Routledge, 576 1976, Scott and Medioli, 1980), large areas of high salt marsh are comprised of Spartina patens 577 meadows, suggesting that bulk-sediment  $\delta^{13}$ C values are likely to remain useful sea-level indicators in 578 this region. The increased floral diversity of high salt marshes in New Brunswick (e.g., Gehrels et al., 579 2006a, Magenheimer et al., 1996), Prince Edward Island (e.g., Scott et al., 1981) and Newfoundland (e.g., 580 Brookes et al., 1985; this study) results in a mosaic structure with increasing contributions to the surface 581 sediment from  $C_3$  species such as Juncus spp., Triglochin maritima and Plantago maritima. Despite the 582 observed presence of Spartina patens and Distichlis spicata, we conclude that this diversity and structure results in bulk-sediment  $\delta^{13}$ C values which are characteristic of C<sub>3</sub> plants. Occasional samples with less 583 depleted values (e.g., stations 16 and 17 on PLA-A) likely represent persistent, but patchy stands of C<sub>4</sub> 584 plants. Therefore the utility of bulk-sediment  $\delta^{13}$ C values is restricted on the Atlantic coast of North 585 586 America to regions from Chesapeake Bay to southern Nova Scotia (Figure 1), although it is necessary to

recognize that geographic migration of floral zones may have occurred in response to Holocene climate changes, which should be considered when choosing whether or not to measure  $\delta^{13}$ C in core material.

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### 590

# 5.4 Reconstructing late Holocene relative sea level in Newfoundland

591 We reconstructed RSL at Hynes Brook from three cores of basal sediment by combining the 592 radiocarbon-dated age of each sample with an estimate of paleo tidal elevation inferred from microfossil 593 assemblages preserved in core samples (Figure 8). This approach generated one sea-level index point 594 from each core. Paleo-tidal elevation was estimated from the combined presence of high salt-marsh 595 foraminifera (Jadammina macrescens and Balticammina pseudomacrescens) and testate amoebae. Since the assemblages of testate amoebae lack modern analogs, we conservatively estimate that the dated 596 597 samples formed between the lowest occurrence of testate amoebae (0.52 m MTL at Hynes Brook) and the 598 highest occurrence of foraminifera (0.90 m MTL at Hynes Brook). Each dated sample came from within 5 599 cm of the basal contact and therefore experienced no, or minimal, post-depositional lowering due to

- sediment compaction (Horton and Shennan, 2009).
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602 The three new sea-level index points were combined with existing and standardized data from southwest 603 Newfoundland (Love et al., 2016; Figure 8). There is good agreement between these datasets, which 604 indicate that RSL rose in this region from approximately -3.5 m at 2900 years BP to present. The average 605 vertical and chronological uncertainty is smaller for the three new sea-level index points than for those in 606 the existing database. This pattern occurs because the age of our samples was established by radiocarbon 607 dating of single macrofossils, while some of the existing dates relied on thick sections of bulk sediment 608 that inherently included material spanning a range of ages. The vertical uncertainty in some of the 609 sea-level index points from the database is large because information provided in the original publication 610 was lacking and the data were standardized in a conservative manner (e.g., Engelhart et al., 2011, 611 Shennan and Horton, 2002). However, our results indicate that a simple multi-proxy approach using the 612 presence of high salt-marsh foraminifera and testate amoebae can produce precise RSL reconstructions. 613

- The ecological plausibility of any paleoenvironmental reconstruction generated through reasoning by
- analogy relies on their being an appropriate degree of similarity between modern and fossil assemblages
- 616 (e.g., Jackson and Williams, 2004). A long-running debate surrounding the use of microfossil groups to
- 617 reconstruct RSL is whether to use a local- or regional-scale modern training set to quantitatively interpret
- assemblages preserved core samples (e.g., Horton and Edwards, 2005, Kemp and Telford, 2015,
- 619 Watcham et al., 2013). Local-scale datasets typically produce more precise reconstructions since the
- 620 effect of secondary environmental variables is minimized, while regional-scale datasets provide a broader

621 suite of analogs for generating reconstructions from sites where environmental conditions in the past may 622 not be the same as those today. The distribution of foraminifera in Newfoundland highlights the necessity 623 of compiling a regional-scale training set where the goal is to capture the principal surface assemblages 624 that are present on salt marshes in the region. For example, Placentia possess distinctive microfossil 625 assemblages (foraminifera Group 5 and testate amoebae Group 2) that are absent from Hynes Brook and 626 Big River. Although this diversity of modern assemblages was not necessary to accurately reconstruct 627 RSL from the Hynes Brook cores, it is likely that investigations spanning longer periods of time (and/or from other sites) will require that a regional-scale training set be available. We conclude that a 628 629 regional-scale training set is necessary to capture the range of assemblages of foraminifera and testate 630 amoebae that are present on modern Newfoundland salt marshes.

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632 All assemblages of testate amoebae in the three cores from Hynes Brook lacked modern analogs (Figure 633 6). The broad, biogeographical similarities between modern intertidal assemblages found in 634 Newfoundland (this study) and elsewhere in North America (Barnett et al., 2016, Gehrels et al., 2006a) 635 and Europe (Barnett et al., 2013, Charman et al., 2002, Ooms et al., 2015) suggests that this dissimilarity 636 between modern and fossil assemblages did not arise from temporal changes in the testate amoebae 637 population at the study site, unless ecological conditions differed so significantly in the past that Hynes 638 Brook supported an assemblage that is yet to be observed in modern North Atlantic salt marshes. 639 Alternatively, the lack of analogy between core and modern samples was caused by preferential 640 preservation where certain taxa are more or less likely to be lost through time due to differences in test 641 composition. This hypothesis is supported by comparison of the contemporary and fossil assemblages 642 from Newfoundland (Figure 9). The modern data contains an abundance of idiosomic genera (tests 643 composed of proteinaceous secretion and siliceous plates) such as Arcella, Euglypha, Pseudocorythion, 644 Pseudohyalosphenia, Tracheleuglypha and Trinema. In contrast, these genera are rarely encountered in 645 the fossil samples, where xenosomic genera (tests formed from agglutinated particles) represent the vast 646 majority of taxa (e.g., *Centropyxis*, *Cyclopyxis* and *Difflugia*). Ratios of idiosomic : xenosomic tests are 647 consistently two or three orders of magnitude greater in the surface assemblages than in fossil assemblages (Figure 9), suggesting that idiosomic taxa were preferentially lost over time as evidenced 648 649 (for example) by the uncharacteristically extreme dominance of *Centropyxsis cassis* type (xenosomic test) 650 in the core samples. Analyses of coastal sediments from the UK reported good preservation of testate 651 amoebae in supratidal deposits (e.g., raised bogs and lakes), but poor preservation in mid-Holocene 652 salt-marsh sediments, including a lack of idiosomic genera (Lloyd, 2000, Roe et al., 2002). In contrast, 653 two short (~100 year) cores of salt-marsh sediment collected in Maine and Nova Scotia to reconstruct 654 RSL contained preserved idiosomic genera such as Arcella, Euglypha, Tracheleuglypha and Trinema

655 (Charman et al., 2010). Conditions in organic salt marshes (e.g., dissolved oxygen concentrations and pH) 656 are likely to promote loss of idiosomic tests (e.g. through dissolution). For example, Swindles and Roe 657 (2007) demonstrated experimentally that idiosomic tests were preferentially removed from testate 658 amoebae assemblages in low pH environments. While idiosomic tests may be preserved for several 659 hundred years, they can decay over longer time periods. Systematic studies that encompass a 660 comprehensive range of salt marsh and estuarine environments will greatly elucidate the viability of 661 testate amoebae as sea-level indicators from a preservation potential perspective. However, the simple 662 classification of presence or absence used in this study is unlikely to be unduly influenced by preservation 663 bias between idisomic and xenosomic tests.

664 665

#### 6. CONCLUSIONS

Newfoundland is one of the most northern sites at which cores of salt-marsh sediment can yield detailed RSL reconstructions to investigate physical processes that vary by latitude such as ocean circulation changes (e.g., Levermann et al., 2005) and the fingerprint of Greenland Ice Sheet melt (Mitrovica et al., 2011). In support of this work we investigated the utility of foraminifera, testate amoebae and bulk-sediment  $\delta^{13}$ C values as sea-level indicators in Newfoundland using modern (surface) sediment samples collected along intertidal transects at Placentia (eastern Newfoundland) and Hynes Brook/Big River (western Newfoundland).

673

674 Foraminifera are divided into five distinct groups which primarily discriminate low salt-marsh 675 environments dominated by Miliammina fusca (Group 4) from more diverse high salt-marsh zones with 676 assemblages dominated by Jadammina macrescens (Group 1), Balticammina pseudomacrescens (Group 677 2), or *Trochammina inflata* (Group 5). Group 3 spans the transition between the high and low marsh groupings and comprises variable abundances of the secondary salt marsh taxa Tiphotrocha comprimata 678 679 and Haplophragmoides manilaensis. Group 5 is only present at Placentia, indicating the potential need to 680 compile regional-scale modern training sets to capture natural variability within a study region and 681 maximize the analogy between modern and fossil assemblages. Overall, the distribution of foraminifera 682 that we observed is similar to previous results from Newfoundland (e.g., Daly, 2002) and more generally 683 to other sites in the Canadian Maritimes and other high-latitude regions.

684

685 The lowest occurrence of testate amoebae at Hynes Brook and Big River occurs close to MHHW, but is

slightly lower at Placentia reflecting local environmental or taphonomic influences. Idiosomic taxa (e.g.,

687 *Trinema* spp., *Tracheleuglypha dentate* type, and *Euglypha* spp.), which characterize upper salt-marsh

688 environments along the east coast of North America were abundant in Newfoundland. However, the

689 difference in the ratio of idisomic and xenomic tests between modern and fossil samples suggests

690 assemblages of testate amoebae in salt marshes experience preservation bias in which idisomic tests are

691 preferentially removed. This preservation bias results in a lack of analogy between modern and fossil692 assemblages.

693

Bulk-sediment  $\delta^{13}$ C values cannot be used as sea-level indicators in Newfoundand because salt marsh and freshwater upland environments yield values that are characteristic of C<sub>3</sub> plants and that cannot be objectively distinguished from one another. We propose that the utility of this proxy for reconstructing RSL in eastern North America is restricted to the coastline between Chesapeake Bay and southern Nova Scotia.

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We further tested the utility of these sea-level indicators by applying them to three radiocarbon-dated cores of basal sediment from Hynes Brook. A simple, multi-proxy classification approach recognized this sediment as having been deposited between the lowest occurrence of testate amoebae and the highest occurrence of foraminifera. This interpretation is not effected by possible preservation bias of testate amoebae in salt marshes. The three new sea-level index points are in agreement with those in an existing database, but have smaller vertical ( $\pm$  0.22 m) and temporal uncertainties ( $\sim$  $\pm$  80 years).

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719 **Table 1**: Details of modern transects used to characterize the distribution of foraminifera, testate and

Site	Transect	Year	Analysis	No. S	Range (m MTL)	Reference	
Placentia	PLA-A	2015	F, T, $\delta^{13}$ C	25	0.30 to 1.22	This study	
Placentia	PLA-B	2015	F, δ <sup>13</sup> C	15	0.51 to 1.26	This study	
Placentia	PLA-C	2015	F, δ <sup>13</sup> C	10	0.23 to 1.21	This study	
Hynes Brook	HBM-0	1999	F	25	-0.13 to 0.83	Wright et al. (2011)	
Hynes Brook	HBM-0b	2015	Τ, δ <sup>13</sup> C	15	0.24 to 2.16	This study	
Hynes Brook	HBM-1	2001	F	24	0.15 to 1.28	This study	
Hynes Brook	HBM-2	2001	F	22	0.12 to 1.15	Wright et al. (2011)	
Big River	BRM-0	1999	F	25	-0.14 to 1.04	This study	
Big River	BRM-1	2001	F	25	0.05 to 1.23	This study	
Big River	BRM-1b	2015	Τ, δ <sup>13</sup> C	13	0.42 to 1.51	This study	
Big River	BRM-2	2001	F	20	0.23 to 1.16	This study	

720 bulk-sediment  $\delta 13C$  values in Newfoundland.

F = samples analyzed for foraminifera; T = samples analyzed for testate amoebae;  $\delta^{13}$ C = samples

analyzed for bulk-sediment  $\delta^{13}$ C values. No. S = total number of samples on transect (including those in

724 which foraminifera were absent).

- 725 **Table 2:** Radiocarbon ages and relative sea level (RSL) reconstruction from three cores of basal organic
- sediment at Hynes Brook.

Core	Sample Elevation (m, MTL)	Sample ID	<sup>14</sup> C Age	<sup>14</sup> C Error	Indicative Meaning	Sampling Error (m)	RSL (m)
HBM C102	-1.38	OS-123524	2,490	20	LOT-HOF	0.17	$-2.09\pm0.22$
HBM C103	-0.25	OS-123656	1,840	20	LOT-HOF	0.17	$-0.96\pm0.22$
HBM C104	-0.55	OS-123525	1,770	20	LOT-HOF	0.17	$-1.26\pm0.22$

Sample ID is the unique identifier assigned to each reported radiocarbon age by the National Ocean 728 729 Sciences Accelerator Mass Spectrometry facility. Indicative meaning is the tidal elevation at which each 730 sample was judged to have formed at based on the presence of both foraminifera (HOF = highest 731 occurrence of foraminifera) and testate amoebae (LOT = lowest occurrence of testate amoebae) in and 732 around the dated sample. At Hynes Brook and Big River, HOF is at 0.90 m MTL and LOT is at 0.52 m MTL, resulting in a reference water level of 0.71 m above mean tide level and an indicative range of  $\pm$ 733 0.19 m. The sampling error for each sea-level index point is the sum of estimated uncertainties for 734 735 leveling ( $\pm 0.05$  m), benchmarks ( $\pm 0.1$  m), sample thickness ( $\pm 0.01$  m) and core angle (1% of depth 736 equating to  $\pm 0.01$  m for these samples). The RSL error was calculated following Engelhart and Horton 737 (2012).

#### 739 FIGURE CAPTIONS

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741 Figure 1: Location of study sites in Newfoundland, Canada. (A) Approximate geographic boundaries 742 between regions on the Atlantic coast of North America where high salt-marsh environments are 743 vegetated predominately by C<sub>3</sub> (e.g. Juncus spp.) and C<sub>4</sub> species (e.g. Spartina patens, Distichlis spicata), 744 where salt marshes replace mangroves and regions experiencing uplift or subsidence as a consequence of ongoing glacio-isostatic adjustment. Number sites are locations discussed in text (Daly, 2002; 1 = St. 745 746 Paul's Inlet, 2 = Village Cove (**B-D**) Study site at Placentia, close to the Argentia tide gauge. The site 747 experiences tidal inundation because the open North Arm is connected to the South Arm at the eastern edge of the Placentia peninsula. The gravel barrier in front of the salt marsh is overwashed by high tides. 748 749 (E-H) Study sites at Hynes Brook and Big River. The location of the Port aux Basques tide gauge is 750 shown. Foraminifera from transects HBM-0 and HBM-2 were originally presented by Wright et al. 751 (2011). Three cores of basal sediment were collected along HBM-2. (I) Monthly average high (H) and 752 low (L) air temperature at Stephenville (1942–2014) and Argentia (1945–2007), calculated from 753 Government of Canada historic climate data. 754

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for aminifera, which is the single highest sample from all three modern transects at Placentia to provide a robust and *in situ* assemblage of for aminifera. MHHW = mean higher high water; MHW = mean high water; MTL = mean tide level. Measured, bulk-sediment  $\delta^{13}$ C values are expressed relative to the Vienna Pee Dee Belemnite (VPDB) standard. Shaded intervals represent values that are typical of salt-marsh

**Figure 2:** Distribution of foraminifera and  $\delta^{13}$ C values measured in samples of modern salt-marsh

sediment collected along three transects at Placentia, Newfoundland. HOF = highest occurrence of

sediment from floral zones dominated by  $C_3$  and  $C_4$  plant species.

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Figure 3: Distribution of foraminifera in samples of modern salt-marsh sediment collected along three
transects at Big River and one transect at Hynes Brook in Newfoundland. Foraminifera along two other
transects at Hynes Brook (HBM-T0 and HBM-T2) were published by Wright et al. (2011) and are used in
our regional compilation of data, but are not presented in detail here. HOF = highest occurrence of
foraminifera, which is the single highest sample from the six modern transects at Big River and Hynes
Brook to provide a robust and *in situ* assemblage of foraminifera. MHHW = mean higher high water;
MHW = mean high water; MTL = mean tide level.

Figure 4: Distribution of testate amoebae in samples of modern salt-marsh sediment collected along
transect HBM-0b at Hynes Brook, BRM-1b at Big River and PLA-A at Placentia. Panels in the top row

- show the position of sampling stations along each transect; note that the elevation axes differ among sites.
- HOF = highest occurrence of foraminifera; LOT = lowest occurrence of testate amoebae (the sample
- below which test abundance was insufficient to represent a viable and *in-situ* assemblage); MHHW =
- mean higher high water; MTL = mean tide level. The concentration of testate amoebae per counted spore
- of the exotic pollen *Lycopodium clavatum* (row 2) was used to determine LOT. Typically there was an
- easily recognizable and abrupt decrease in test concentration at this point from  $\sim 0.2$  to < 0.1. Rows 3–6
- show the distribution of the five most abundant species of testate amoebae on each transect, note that the
- abundance axes are the same within each transect, but differ among them. The lowest row of panels
- 781 presents measurements of bulk-sediment  $\delta^{13}$ C values. The profile for Placentia is presented in Figure 2.
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**Figure 5:** Regional-scale modern datasets of foraminifera and testate amoebae from Hynes Brook, Big

784 River and Placentia in Newfoundland, Canada. (A) Average silhouette width calculated by partitioning

- around medoids of the foraminifera dataset from which we recognized five distinct groups. (B)
- Foraminifera dataset divided into five groups and colored by site. Hm = Haplophragmoides manilaensis;
- 787 Tc = *Tiphotrocha comprimata*. (C) Average silhouette width calculated by partitioning around medoids
- of the testate amoebae dataset. These results demonstrated that two distinct groups should be recognized.
- 789 (D) Testate amoebae dataset divided into two groups and colored by site. Po = *Pseudohyalosphenia* type
  790 (Ooms).
- 791

792 Figure 6: Samples of basal salt-marsh sediment from Hynes Brook that were analyzed to reconstruct late 793 Holocene relative sea-level. For each of the three cores (C102, top row; C103, middle row; C104 bottom 794 row), a lithology column shows the position of the basal contact and radiocarbon date with its unique 795 identifier from the National Ocean Sciences Accelerator Mass Spectrometry facility. Core top elevations 796 with respect to modern, local mean tide level (MTL) are listed. Abundance of Jadammina macrescens 797 (Jm) and *Balticammina pesudomacrescens* (Bp) are shown in the first column of panels (blue bars). 798 Dissimilarity between each core sample and its closest modern analog based on foraminifera are shown in 799 the second column of panels (dashed lines represent values for percentiles of dissimilarity measured in 800 pairings of modern samples). Abundance of the two most common species of testate amoebae are shown 801 in the third and fourth columns (red bars; DL= *Difflugia lucida* type; CP = *Centropyxis platystoma* type; 802 CD = *Centropyxis delicatula-ecornis* type). Note that the second most abundant species (after *Centropyxis* 803 *cassis* type) varies among cores. The concentration of testate amoebae is expressed relative to the 804 frequency of Lycopodium spores in the fifth column of panels. The dashed line represents the lowest 805 concentration in a modern sample that was considered to have a viable and *in-situ* assemblage. Values 806 exceeding this threshold indicate deposition above the lowest occurrence of testate amoebae.

807 Dissimilarity between each core sample and its closest modern analog based on testate amoebae are

- shown in the fifth column of panels (dashed lines represent values for percentiles of dissimilarity
- 809 measured in pairings of modern samples). Measured, bulk-sediment  $\delta^{13}$ C values with respect to the
- 810 Vienna Pee Dee Belemnite (VPDB) standard are presented in the final column and the shaded regions
- 811 denote the approximate range of  $C_3$  and  $C_4$  plants.
- 812

813 Figure 7: Relationship between species of modern (A) foraminifera and (B) testate amoebae and tidal 814 elevation in Newfoundland. Symbol color denotes group membership established by partitioning around 815 medoids applied separately to the regional modern datasets of each microfossil group. Colored bars at right show the range of elevations over which each group was found. Arrows indicate groups for which 816 817 upper and/or lower limits could not be reliably established. Note that an anomalously low sample from 818 for a group 5 was discounted in estimating the range of this group. Elevation is expressed as a 819 standardized water level index (SWLI), where a value of 200 is the highest occurrence of foraminifera 820 and 100 is mean tide level.

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Figure 8: Late Holocene relative sea-level history of southwestern Newfoundland produced using a
standardized database of sea-level index points (Love et al., 2016; open rectangles) and the three new
basal sea-level index points from Hynes Brook (this study; filled rectangles labeled to denote the core
from which each new index point was produced). The study of Love et al. (2016) utilized several earlier
datasets to produce standardized sea-level index points (see references therein).
Figure 9: Ratio of testate amoebae with idiosomic (composed of proteinaceous secretion and siliceous
plates) and xenosomic (formed from agglutinated particles) tests in modern and fossil sediment samples

830 in Newfoundland. Bars represent modern samples (colored by site), while the blue, shaded region

- represents the range of values encountered in core samples from Hynes Brook. The apparent loss of
- 832 idiosomic tests is interpreted as preservation bias.

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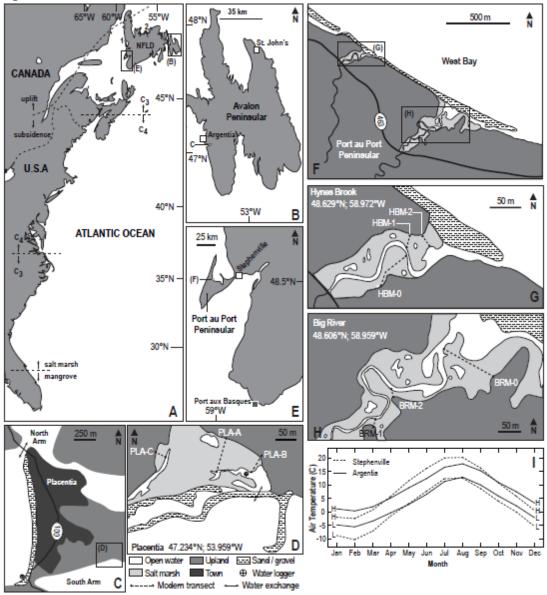
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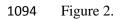
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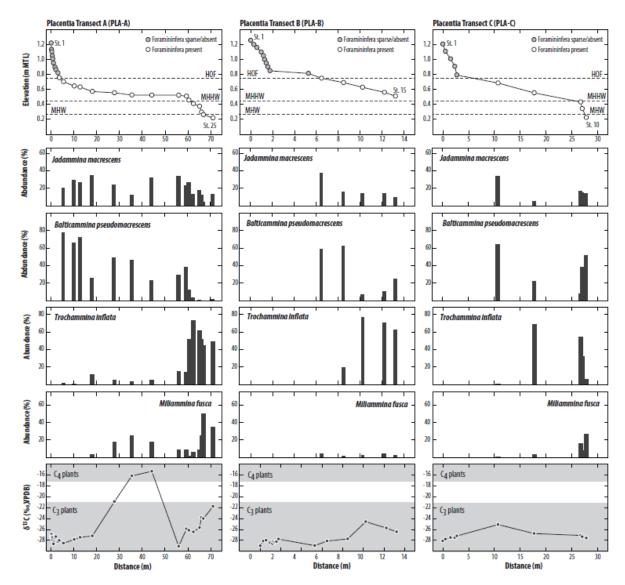
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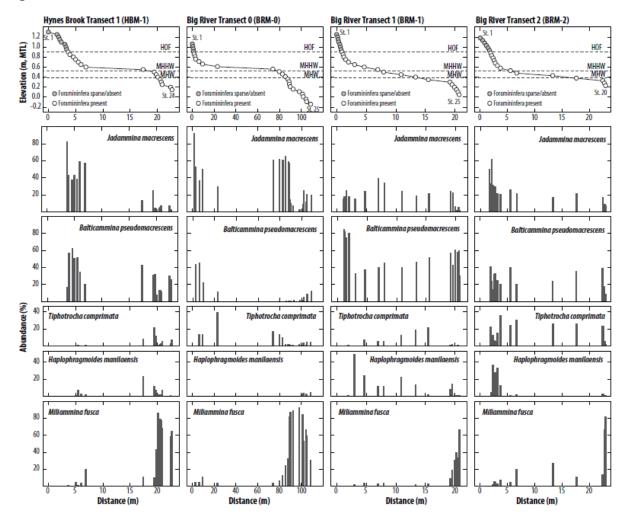




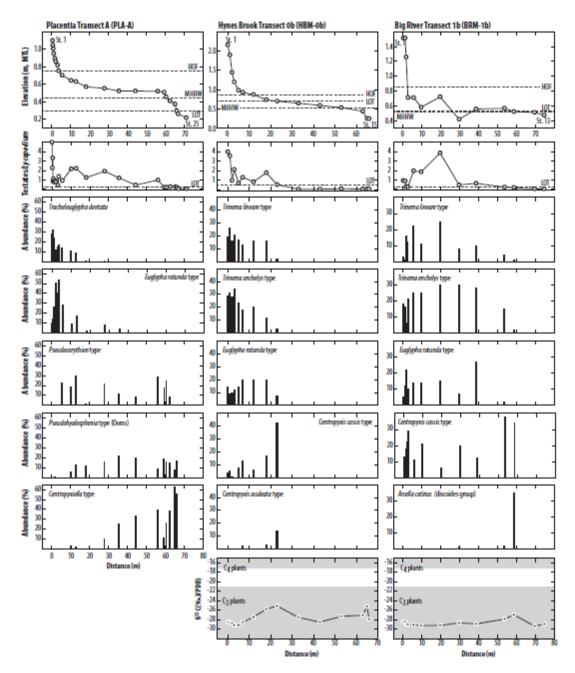


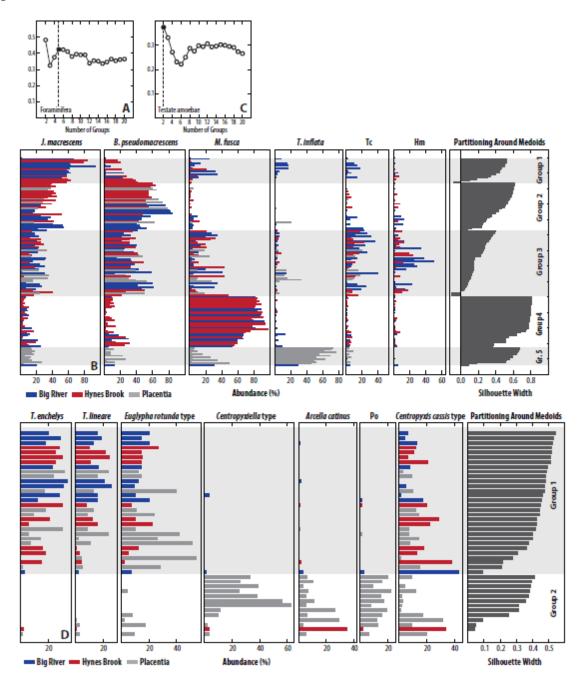


1097 Figure 3.









# 1106 Figure 6.

