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1 Organic matter decomposition and associated microbial communities in wetlands:
2 insights from tropical and subtropical *Melaleuca* forests in Australia

3

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20

21 Abstract

22 Wetlands are characterised by soils rich in organic matter that accumulate carbon, providing an
23 important pathway for carbon dioxide sequestration. Nevertheless, not all the carbon fixed can
24 be accumulated, and a proportion will decompose through microbial consumption and be partly
25 released into the atmosphere. Rates of organic matter decomposition in tropical wetlands and
26 the factors associated with this process are scarce. We conducted a two-year field study in three
27 *Melaleuca* wetlands in tropical and subtropical Australia using standardised tea litter substrates
28 (green-labile and rooibos-recalcitrant) to measure organic matter decomposition and the
29 microbial communities associated with this process. Decomposition rates were 4-fold higher in
30 labile litter, which was low in carbon: nitrogen, compared to recalcitrant litter. The prokaryotic
31 communities associated with the decomposing litter were unique at each site and different from
32 the soil. They contained taxonomic groups adapted to anaerobic, high temperatures, acidic
33 conditions and suggestive of slow anaerobic turnover. Microbial communities changed as
34 decomposition progressed, with the latter characterised by taxa with cellulose-degrading
35 functions. The decomposition of recalcitrant organic matter within *Melaleuca* soils was
36 relatively slow, with half of the organic matter inputs remaining after two years, supporting
37 long-term carbon sequestration.

38

39 **Keywords:** carbon, decomposition, isotopes, litter, mineralisation, nitrogen

40

41

42 Introduction

43 Coastal wetlands are characterised by soils rich in organic carbon (C) and slow
44 decomposition rates, favouring C sequestration. The rate at which wetlands store soil C is the
45 net sum of litter accumulation and decomposition. Decomposition is variable among wetland
46 types and varies with physicochemical characteristics (e.g. soil characteristics, salinity,
47 nutrients), biological components (plant species, microbial community), environmental setting,
48 disturbances and climate (Spivak et al. 2019). The highest decomposition rates are predicted in
49 wetlands where the temperature and nutrients are high, salinity is low, and the soil has a
50 microbial community capable of degrading compounds from the local litter (Rejmánková and
51 Houdková 2006). However, studies of decomposition rates in tropical wetlands, where many of
52 these conditions are met, are scarce.

53 Tropical wetlands of *Melaleuca* trees, commonly known as "paper bark trees", are
54 widespread in Southeast Asia and Oceania. Only in Australia, *Melaleuca* forests cover an area
55 of 6 million ha (ABARES 2016; Finlayson 2005; Tran et al. 2015). *Melaleuca* wetlands can
56 also be found in the Caribbean, the United States and South America, where species such as *M.*
57 *quinquenervia* have become invasive (Turner et al. 1998). In Australia, *Melaleuca* wetlands
58 have been heavily lost to agriculture and urban development (Johnson et al. 1999). The
59 deforestation of these ecosystems has led to the loss of the diversity and biogeochemical
60 functions that support many ecosystem services they provide, including C storage and water
61 quality improvement (Tran and Dargusch 2016; Adame et al. 2019a).

62 *Melaleuca* wetlands are highly productive (Finlayson et al. 1993) and have anoxic soils
63 rich in C (Adame et al. 2020). Decomposition in *Melaleuca* wetlands varies with inundation
64 frequency (Wallis and Raulings 2011) and litter type (Rayamajhi et al. 2010). However, there
65 is limited understanding of other factors that drive organic matter decomposition in these
66 wetlands, compounded by a fundamental lack of knowledge on their microbial communities
67 and functions.

68 Microbial communities of wetland soils are key for soil organic matter turnover and are
69 variable among wetland types (Spivak et al. 2019). Microbial communities are also temporally
70 variable, with community shifts during decomposition, reflecting changes in metabolic
71 pathways, for instance, fermentation or sulphate reduction (Trevathan-Tackett et al. 2017).
72 Communities also shift depending on the availability of organics, such as simple carbohydrates
73 at the beginning of the decomposition versus lignocelluloses later in the decomposition process
74 (Trevathan-Tackett et al. 2017). Changes in these microbial communities and metabolic
75 pathways can be detected by changes in isotopic values ($\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ of the litter), which
76 change during decomposition (Kelleway et al. 2022).

77 Assessing biochemistry and microbiology responses is important to understand the causal
78 mechanisms that support ecosystem services. For instance, the restoration of *Melaleuca*
79 wetlands in Australia has gained momentum in the past five years, and obtaining carbon credits
80 (ACCUs, Australian Carbon Credit Units) through restoring these wetlands is now possible
81 (Lovelock et al. 2022). Restoration projects will benefit from understanding how the soil and
82 microbial community change before and after restoration (Farrer et al. 2022).

83 In this study, we follow the decomposition process over two years in three *Melaleuca*
84 wetlands in subtropical and tropical Queensland, northeast Australia. We used standardised
85 substrate proxies for labile (green tea) and recalcitrant (rooibos or red tea) organic matter
86 (Keuskamp et al. 2013). We measured changes in biomass, elemental C and nitrogen (N),
87 isotope values ($\delta^{15}\text{N}$ and $\delta^{13}\text{C}$) and the prokaryotic microbiome composition of the
88 decomposing litter. We hypothesised that: (1) rates of organic matter decomposition will be

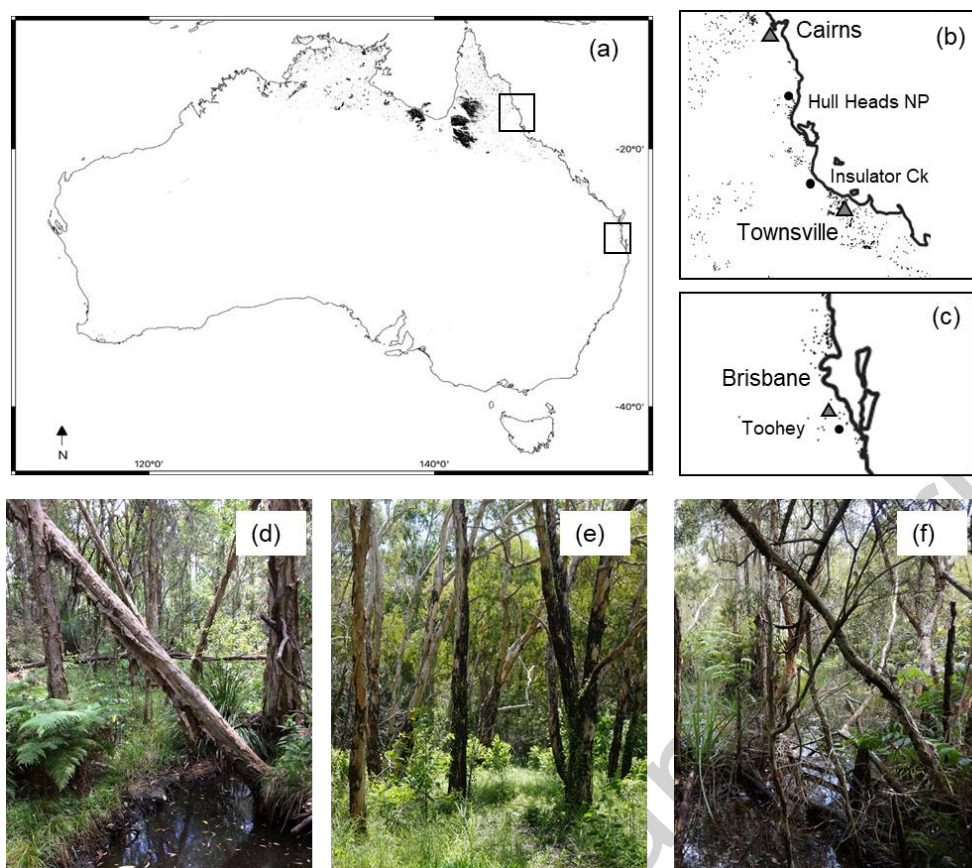
89 faster in labile than recalcitrant litter, (2) changes in C, N, and isotopic values will reflect
90 decomposition, with higher enrichment (increase in values) and an increase in C:N in
91 substrates with faster decomposition (Fry 2006), and (3) microbial communities associated
92 with the tea litter will change as decomposition progresses, reflecting shifts in C and nutrient
93 availability (Trevathan-Tackett et al. 2021). The goals were to fill in a knowledge gap on the
94 biogeochemical cycling of *Melaleuca* wetlands (Adame et al. 2021), to understand soil C
95 accumulation and losses (Wieder et al. 2013) and to provide foundational knowledge on the
96 soil microbiota of these unique ecosystems.

97

98 Materials and Methods

99 Study sites

100 We selected three sites in Queensland, Australia: Hull Heads National Park (HH), Insulator
101 Creek (IC) and Toohey Forest (TF, Fig. 1). The site HH (-18.0013°; 146.0465') has a humid
102 tropical climate with a mean annual precipitation of 4,075 mm, and minimum and maximum
103 mean temperatures of 19 and 28°C (Tully Station, 1925-2021; and Cardwell Marine Parade,
104 1907- 2021, Australian Bureau of Meteorology, ABM, 2021). The site IC (-18.8957°;
105 146.2594') is drier, with mean annual precipitation of 2,145 mm and mean minimum and
106 maximum temperatures of 19.0 and 29.3°C, respectively (1968-2021, Ingham station, ABM,
107 2021). Finally, TF is in the subtropics (-27.5513°; 153.0582') in a cooler and drier climate, with
108 mean annual precipitation of 1,051 mm and 14.5 and 26.4°C, for mean minimum and
109 maximum annual temperatures, respectively (1929-2021, Archerfield Airport, ABM). HH and
110 IC are located within the coastal zone; HH is permanently flooded except during very dry
111 periods (e.g., the dry season of 2019); IC is flooded during the wet season (approximately
112 December to April) and sporadically by high tides. TF is 16 km from the coast, within Mimosa
113 Creek, a perennial freshwater creek. The forest in HH is dominated by trees of *Melaleuca*
114 *viridiflora* mixed with rainforest species, while *M. quinquenervia* dominates IC and TH.



115

116 **Fig 1.** (a) Distribution of *Melaleuca* forests in Australia (wetlands and non-wetlands,
 117 ABARES, 2016); circles indicate the location of study sites in (b) tropical and (c) subtropical
 118 Queensland, Australia; (d) Hull Heads National Park, (e) Insulator Creek, and (f) Toohey
 119 Forest; triangles are major cities within the study area

120

121 Litter decomposition

122 Rates of organic matter decomposition were measured using standardised tea litter bags
 123 from the Global TeaComposition H₂O initiative (Trevathan-Tackett *et al.* 2021), a co-initiative
 124 to the TeaComposition program, with a modified method to the Tea Bag Index technique
 125 (Keuskamp *et al.* 2013; Djukic *et al.* 2018). The technique consists of burying tea bags as
 126 standardised and representative litter within wetlands and estimating exponential decay rates
 127 (k) from their biomass loss over time (Trevathan-Tackett *et al.* 2021). This technique can
 128 overcome natural plant litter variability among sites and provides information comparable to
 129 other ecosystems and studies using the same approach. However, this litter bag technique may
 130 provide rates different from those obtained from local litter and exclude large grazers from
 131 entering the teabag at the cost of retaining small particulate litter within the fine mesh. Thus,
 132 decomposition measured with this method may be underestimated if grazers are common
 133 (Bradford *et al.* 2002).

134 In January 2018, at the beginning of the wet season, 32 pre-weighed labile green and 32
 135 recalcitrant rooibos teabags (hereafter "red tea") were set at each site. The teabags were buried
 136 at 15 cm soil depth in two plots, three meters apart. Within each plot, 16 bags of green tea and
 137 16 of red tea were buried (total $n = 192$ bags); half of the bags were pre-weighed. The tea bags
 138 were retrieved approximately at 3, 6, 12, and 24 months (TF at 97, 194 and 370 days; IC at
 139 154, 281 and 505 days; HH at 154, 281, 505, and 672 days). Differences in sampling times
 140 among sites were due to limited accessibility during the summer when flooding is common and

141 dangerous crocodiles (*Crocodylus porosus*) in IC and HH are present. At least three pre-
 142 weighed green and red bags were retrieved from each plot during each sampling. The bags
 143 were cleaned of soil and in-growth roots, oven-dried (60°C), and weighed to estimate dry
 144 biomass loss. The remaining tea was analysed for elemental C, N, and ¹³δC and ¹⁵δN values
 145 (Elemental analyser coupled with a Mass Spectrometer, Griffith University) to estimate
 146 decomposition as indicated by the increase in C:N, ¹³δC and ¹⁵δN over time (Fry 2006).

147 Additionally, three soil samples at each plot were taken with a graduated mini core at
 148 the beginning of the experiment. The samples of known volume were used to calculate bulk
 149 density. The soil was tested for organic C by adding HCl in five samples per site; none
 150 displayed bubbling, indicating minimal carbonate content in the soil. Consecutively, the soil
 151 was analysed for organic C, N, ¹³δC and ¹⁵δN in non-acidified samples, similar to the tea
 152 (above). From the changes in dry biomass, we calculated exponential decay rates (Wider &
 153 Lang, 1982):

$$W_0/W_t = \exp(-k t),$$

Equation 1

156 Where, W₀ is the initial mass, W_t is mass at time *t* (day), and *k* is the decay rate in proportion
 157 per day (d⁻¹). Decay rates were calculated for each tea type, with all sites combined, since the
 158 variability in sampling among sites would confound inter-site decay rate comparisons.

160 Microbial community

161 Immediately after the tea bags (unweighed) were retrieved, one gram each of green and red
 162 tea and one gram of soil within each plot were transferred to 2 ml vials of Zymo DNA/RNA
 163 Shield. The samples were frozen until transportation to the laboratory and stored at -80 °C until
 164 analyses. The DNA was extracted from each sample with the Zymo Mini Kit as per
 165 manufacturer instructions for soil (ZymoResearch, CA, USA). DNA amplification of the V4
 166 16S rRNA gene region was performed before indexing and sequencing at the Deakin
 167 University Genomics Centre (Trevathan-Tackett et al. 2021). The amplicon sequences were
 168 analysed in the QIIME2 DADA2 pipeline (Callahan et al. 2016; Bolyen et al. 2018). The
 169 resulting amplicon sequence variants (ASVs) were normalised to 18,000 reads per sample and
 170 analysed for Beta diversity (weighted UniFrac). Sequences were classified using the Silva v132
 171 database trained to the V4 hypervariable region at a threshold of 99% homology (Quast et al.
 172 2012). Amplicon data are available at the European Nucleotide Archive under project
 173 accessions PRJEB50314 (tea litter) and PRJEB50315 (bulk soils).

175 Statistical Analyses

176 Differences in Beta diversity were analysed with a three-way multivariate PERMANOVA,
 177 with site, substrate (green and red tea, and soil), and time, as fixed factors. The latter test was
 178 repeated with tea samples only to investigate changes in the microbial community as
 179 decomposition progressed within each site. Since the last sampling (2 years) at HH was about
 180 six months after the last sampling at IC and TF, we limited our statistical analysis interpretation
 181 to (a) the early sampling (3 to 5 months across sites) and (b) shifts through time within sites.
 182 SIMPER analysis was performed using the ASV table filtered at 0.001% and on significant
 183 results from the PERMANOVAs to identify the ASVs driving the differences. For the
 184 PERMANOVA analyses, a Monte Carlo tests (*P*(MC)) were used in cases where permutations
 185 were < 200 (PRIMER + PERMANOVA v7; Anderson et al. 2008).

186

187 Results

188 Soil characteristics

189 The surface soil varied among the sites; IC had higher soil C and N (15.7 and 0.85%,
 190 respectively) compared to TH and HH (< 7 and < 0.3%). HH also differed in that it had higher
 191 C:N (33.8) and considerably lower soil $^{13}\delta\text{C}$ and $^{14}\delta\text{N}$ values (-31.0 and -0.05‰) compared to
 192 the other two sites ($^{13}\delta\text{C}$ of -27.2 to -28.8‰, and $^{14}\delta\text{N}$ of 2.33 and 1.69‰ for IC and TH,
 193 respectively, Table 1).

194

195 **Table 1.** Soil characteristics of three supratidal forested wetlands dominated by *Melaleuca* spp.
 196 in subtropical and tropical Australia, TF= Toohey Forest, IC = Insulator Ck, HH = Heads
 197 National Park, C = carbon, N = nitrogen.

	Bulk density (g cm ⁻³)	%C	%N	C:N	$^{13}\delta\text{C}$ (‰)	$^{14}\delta\text{N}$ (‰)
TH	0.27 ± 0.15	3.8 ± 2.2	0.23 ± 0.13	18.9 ± 0.6	-28.8 ± 0.2	1.69 ± 0.24
IC	0.58 ± 0.04	15.7 ± 1.2	0.85 ± 0.01	21.3 ± 1.1	-27.2 ± 1.5	2.33 ± 0.23
HH	0.84 ± 0.12	6.2 ± 0.8	0.21 ± 0.02	33.8 ± 2.5	-31.0 ± 0.2	-0.05 ± 0.16

198

199

200

201 Litter decomposition: Biomass, C and N losses

202 Despite differences in climate and location of the sampling sites, the trends of biomass,
 203 C and N were similar, with differences mostly explained by litter type (green or red) (Fig. 2).
 204 Biomass decay rates for the green tea litter were 4.3-fold higher ($0.0073 \pm 0.00061 \text{ d}^{-1}$) than for
 205 red tea litter ($0.0017 \pm 0.0001 \text{ d}^{-1}$). As a result of the higher decay rate, green tea samples had
 206 only $9.7 \pm 0.12\%$ of their biomass remaining at the end of the two-year incubation, compared
 207 to $42.7 \pm 1.4\%$ biomass remaining of the red tea.

208 The rates for C decay were $0.0015 \pm 0.00009 \text{ d}^{-1}$ for red tea and $0.0058 \pm 0.0006 \text{ d}^{-1}$ for
 209 green tea. The loss of C was similar among sites, ranging between 54.6 and 61.4% of the initial
 210 C for red tea and 77.6- to 90.9% for the green tea (Table 1S, Fig. 2b). The changes in N were
 211 more variable, as N concentrations increased in the green and red tea in IC and TF from $3.8 \pm$
 212 0.1% to 6.0 ± 0.1 and $6.0 \pm 0.2\%$ for the green and from $1.1 \pm 1.1\%$ to 1.5 ± 0.3 and $1.4 \pm$
 213 0.1% for the red tea (Table 1S).

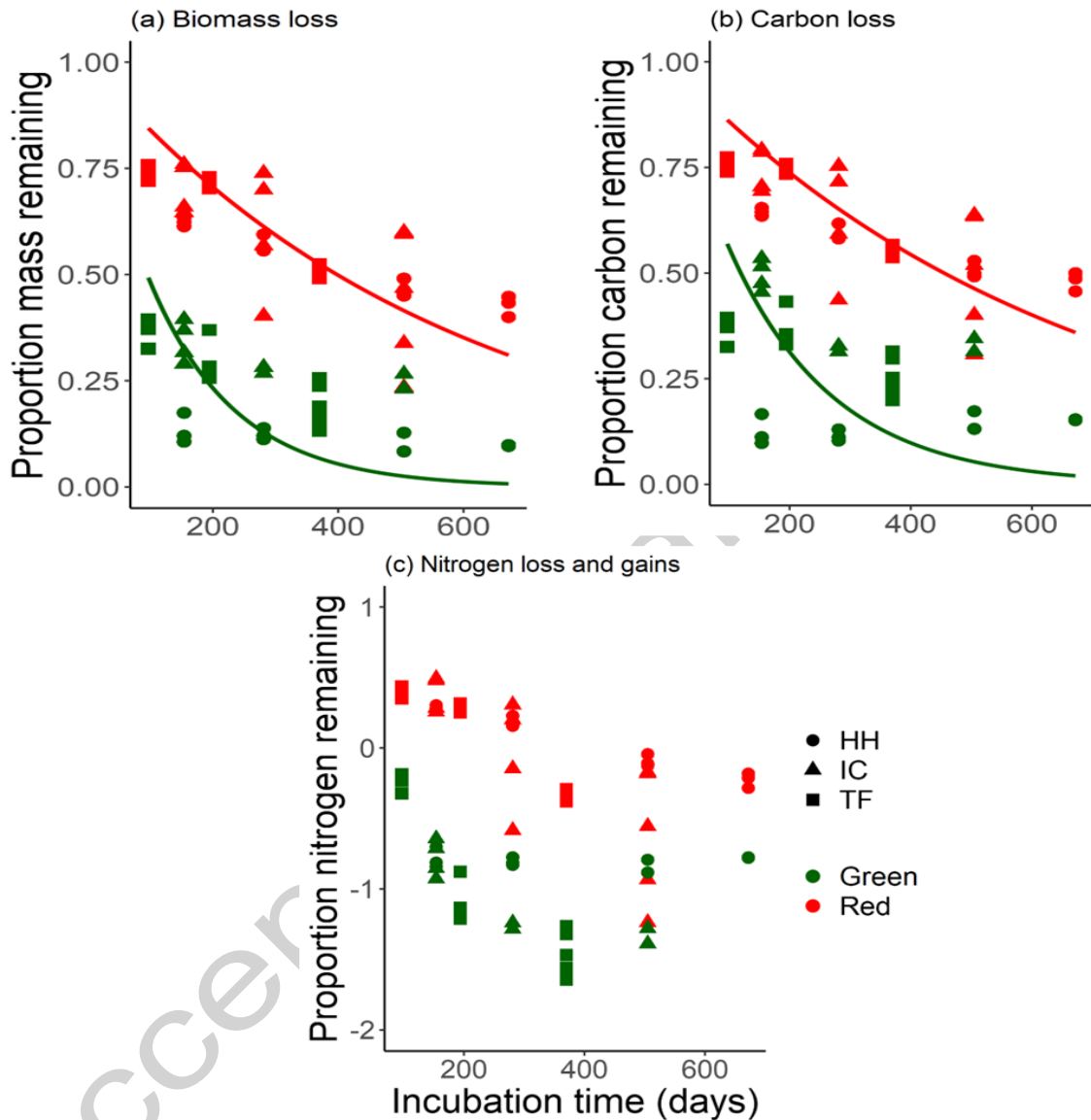
214 The composition of organic matter also changed during the experiment. The C:N ratio
 215 of both teas decreased during decay; however, for the green tea, the decrease was limited to the
 216 first few months (Fig. 3). In comparison, the C:N of the red tea decreased throughout the
 217 incubation, especially in IC (Fig. 3a). For the stable isotopes, initial values were similar
 218 between the tea litter types for $\delta^{15}\text{N}$, while the $\delta^{13}\text{C}$ of red tea was more enriched (higher) than
 219 green tea (Fig. 3b,c). As decomposition progressed, green tea litter became enriched (higher)
 220 for both $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ in TF and IC but not in HH, where $\delta^{13}\text{C}$ remained constant (Fig. 3b,c).
 221 In contrast, the red tea litter became depleted (lower) in $\delta^{13}\text{C}$ and $\delta^{15}\text{N}$ within the first few

222 months, with relatively little change after that, except for a further decrease at two years at HH
 223 (Fig. 3b,c).

224

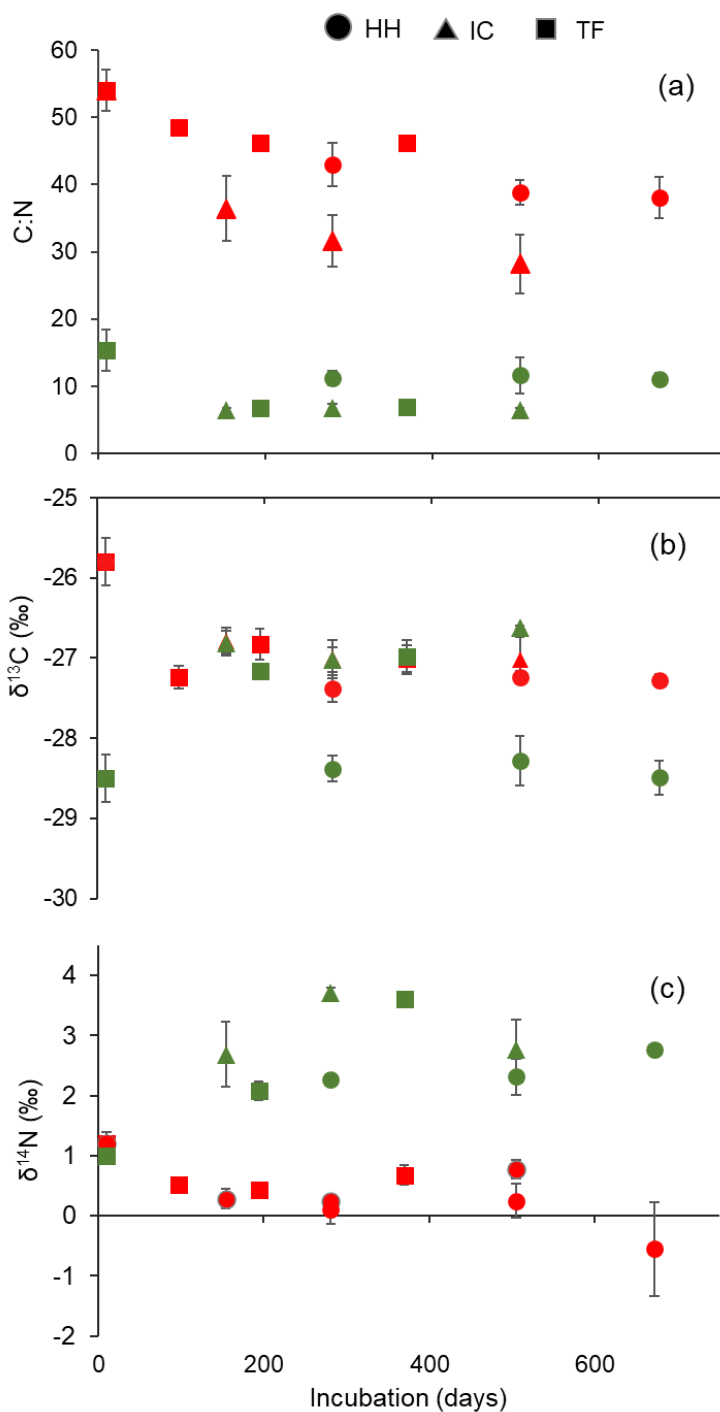
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227

228 **Fig 2.** Proportion of (a) biomass, (b) carbon, and (c) nitrogen of green and red tea litter
 229 remaining after incubation in the soil of three *Melaleuca* wetlands (HH = Hull Heads, IC =
 230 Insulator Creek, TF = Toohey Forest). Negative values in (c) indicate N gains. Biomass loss for
 231 the first 300 days has been previously published as site-level averages in Trevathan-Tackett et
 232 al. (2021)



233

234 **Fig 3.** Trends of red and green tea litter decomposition as indicated by changes in C:N, $^{13}\delta\text{C}$
 235 and $^{14}\delta\text{N}$ in *Melaleuca* wetlands. HH = Hull Heads, IC = Insulator Creek, TF = Toohey Forest)

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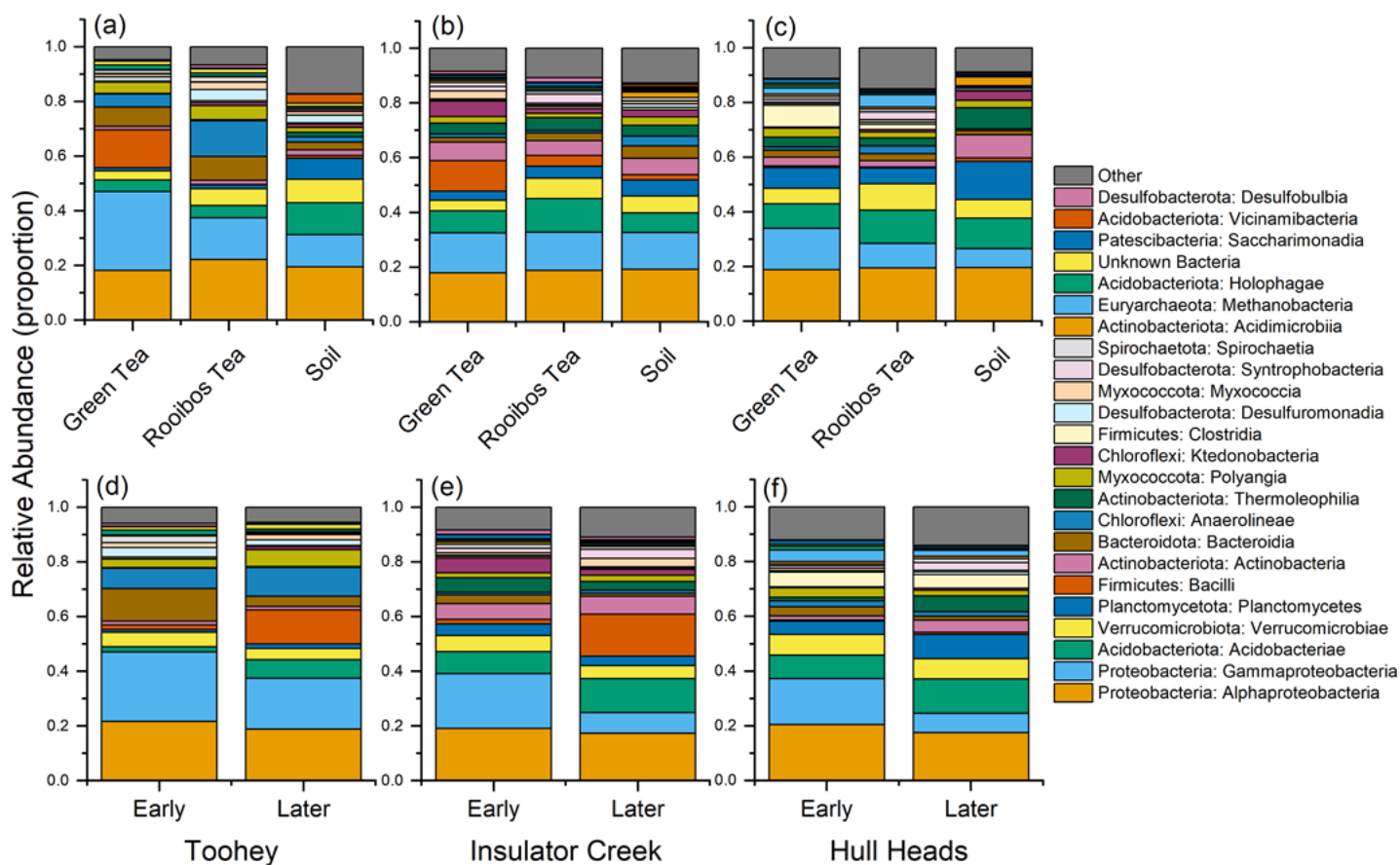
239 Microbial communities

240 Beta diversity or community structure was primarily driven by site and substrate (red,
241 green tea, and soil; 3-way interaction, Psuedo- $F = 2.125$, $P(\text{perm}) = 0.002$; Figs. 4 and S1,
242 Table S2). Only at the early sampling were the tea-associated prokaryotic communities
243 significantly different from the soil microbiome at TF and HH (TF $P(\text{MC}) \leq 0.008$; IC $P(\text{MC})$
244 > 0.05 ; HH $P(\text{MC}) \leq 0.028$; Fig. S1). The tea-associated microbiomes were not significantly
245 different from each other within each site and generally became more similar to the bulk soil
246 microbiome as decomposition progressed (Fig. S1, Table S2).

247 The differences among sites at the early sampling were attributed to TF being enriched
248 in *Bradyrhizobium* sp (ASV11), Candidatus Udaeobacter, and Prosthecobacter (ASV95). In
249 contrast, HH and IC were dominated by the Alpha- and Gammaproteobacteria, Myxococcota,
250 Actinobacteriota and Planctomycetes taxonomic groups. There was a significant shift in the
251 tea-associated microbiome through time within each site (main Site*Time interaction, Psuedo
252 $F = 2.3514$, $P(\text{perm}) = 0.005$, Figs. 4 and 5, Table S2).

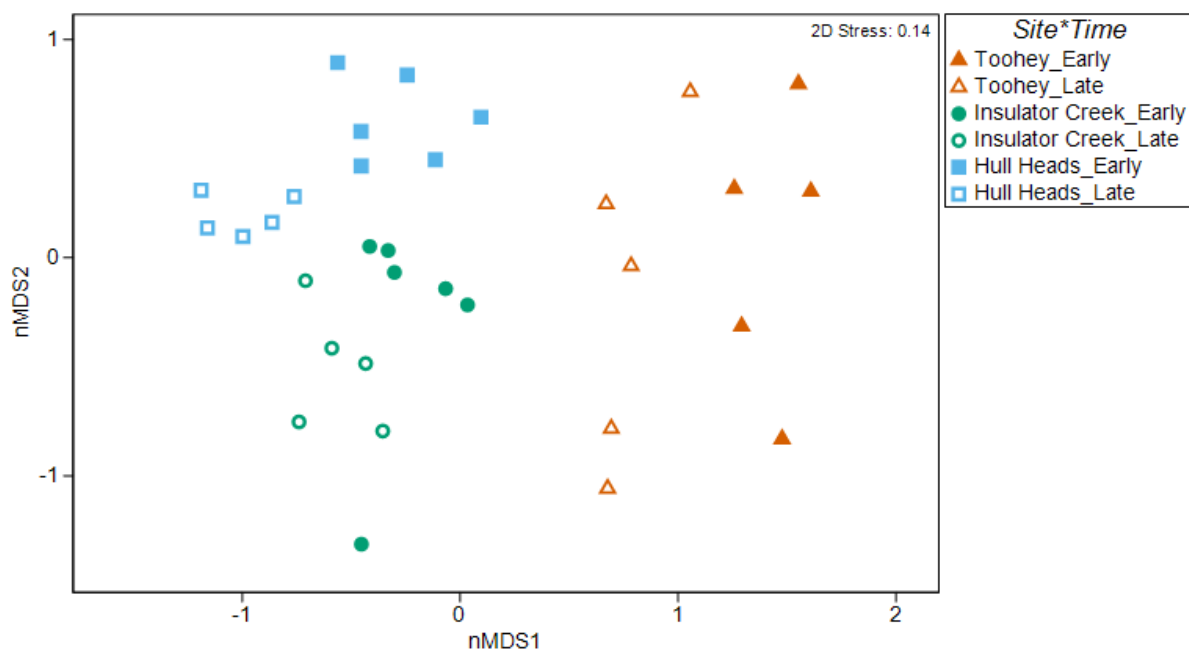
253 At TF, early communities were characterised by ASVs from the genus Bacteroidales,
254 *Citri fermentans* (Geobacter), Burkholderiales and Rhizobiaceae. In contrast, later communities
255 were dominated by Anaerolineae groups SBR1031 and A4b, *Paenibacillus* (Firmicutes) and a
256 *Bradyrhizobium* ASVs ($P(\text{MC}) = 0.031$). In IC, early communities were dominated by the
257 Burkholderia-Caballeronia-Paraburkholderia group, *Dyella* sp., and Ktedonobacteraceae
258 groups, and later communities by Paenibacillaceae (mostly *Paenibacillus* sp.), *Myxococcus* sp.,
259 Xanthobacteraceae, and Streptomyces ($P(\text{perm}) = 0.004$). Finally, at HH, the early and later
260 communities had common orders, such as Acidobacteriales and Clostridiales, but different
261 ASVs within these groups shifted over time ($P(\text{perm}) = 0.003$). Early communities included
262 *Methanobacterium*, *Methanosarcina*, the Burkholderia-Caballeronia-Paraburkholderia genus
263 and *Nevskia*, while later communities comprised Rhodanobacteraceae, Rhizobiales,
264 *Syntrophobacter*, Solirubrobacterales and WPS-2 ASVs.

265



267 **Fig 4.** Class-level changes in community composition across substrate type (green and
 268 rooibos/red tea, and soil, a-c) and time (early and late in the experiment; d-f) for Toohy Forest
 269 (a,d), Insulator Creek (b,e) and Hull Heads (c,f). Time points for Toohy Forests and Insulator
 270 Creek were three months (early) and 12 months (later), while for Hull Heads, the time points
 271 represent five months (early) and 22 months (later). Phylum is included in legend before class.
 272 Values are means

273



275 **Fig 5.** Non-metric multidimensional scaling of tea-associated microbial communities. The
 276 green and red teas decomposing in the soil were collected at 'early' (3 to 5 months) or 'late' (12-
 277 15 months) incubation intervals in three *Melaleuca* wetlands.

278

279

280 Discussion

281 This long-term decomposition study provided insight into organic matter turnover in
 282 widespread tropical *Melaleuca* wetlands. The two standardised tea types roughly represent end-
 283 members for different types of litter (Trevathan-Tackett et al. 2021). The green tea litter,
 284 representing labile organic matter with low C:N (15), was mainly decomposed by the end of
 285 two years. Comparatively, red tea, representing recalcitrant material (C:N of 54) preserved
 286 half of its biomass at the end of the experiment. Thus, depending on the litter type, soil
 287 decomposition and storage pathways will differ vastly. For instance, the leaves and bark of
 288 *Melaleuca* trees are high in lignocellulose and C:N (> 50; Adame & Reef, 2020; Tjokorda et
 289 al. 2021). Thus, the red tea decay trends observed in this study indicate the likely fate of
 290 *Melaleuca* litter. Comparatively, the litter of grasses, typical of the *Melaleuca* forest understory
 291 (Fig. 1), or labile organic matter carried by floods, is likely to have a similar fate as green tea,
 292 with rapid decomposition, subsequent microbial colonisation and N immobilisation. These
 293 results are similar to those for mangrove forests, which found that litter composition, not
 294 climate, were the most important drivers of organic matter decomposition (Simpson et al.
 295 2023).

296 The decay rates for red tea (mean $k = 0.0017 \text{ d}^{-1}$) were within the lower range of
 297 intertidal wetlands, such as mangroves, seagrass, and marshes, whose k values range between
 298 0.001 and 0.005 d^{-1} during the first year of decomposition (Trevathan-Tackett et al. 2021).
 299 Slow decay rates in leaves of other *Melaleuca* species have previously been reported, with *M.*
 300 *ericifolia*, having a $k = 0.001 - 0.002$ (Wallis & Raulings, 2011) and *M. argentea*, *M.*
 301 *leucadendra*, with $k = 0.005$ and 0.007 , respectively (Pettit et al. 2012). Biomass decay rates of
 302 red tea were similar to those of C decay, with a relatively rapid loss within the first months and
 303 a slow decline until the end of the two-year incubation. Similarly, in red tea, $\delta^{13}\text{C}$ and C:N

304 values changed within the first three months and stayed relatively constant throughout the rest
 305 of the experiment. Overall, this study supports the idea that *Melaleuca* litter and soils have
 306 conditions that support long-term soil C storage (Tran & Dargusch, 2016).

307 On the contrary, green tea was rapidly lost, probably due to the fast leaching of labile C,
 308 especially in sites with frequent flooding (Wallis & Raulings, 2011). HH is usually flooded,
 309 and green tea at this site was lost the fastest within the first months of incubation. Our results
 310 are consistent with other long-term tea litter studies showing that after the first few months, the
 311 green tea is exhausted of most of its organic resources, and what remains is likely inaccessible
 312 to further breakdown (Duddigan et al. 2020; Trevathan-Tackett et al. 2021). As green tea
 313 biomass was rapidly lost, the elemental N content of litter increased, and the $^{15}\delta\text{N}$ increased
 314 after the first year of incubation. The changes in N are the net balance between ammonification
 315 (breakdown of organic N to ammonium) and immobilisation (transformation of inorganic N
 316 into organic through microbial consumption; Reddy & DeLaune, 2008). Our results suggest
 317 that N was immobilised in green tea due to microbial colonisation, results previously observed
 318 in other coastal wetlands (Benner et al. 1991; Macko et al. 1994).

319 Most prokaryotic groups associated with the tea litter and soil were characteristic of
 320 freshwater ecosystems or terrestrial forests, with significant differences among sites. While we
 321 only characterised the community profile, we could use the assigned taxonomy to infer putative
 322 functions. Many of the abundant taxa were adapted to anaerobic and acidic conditions, such as
 323 those in these wetlands where organic matter is abundant, and soils are waterlogged (Finlayson,
 324 2005). The conditions at TF were different due to its location upstream with no tidal influence
 325 and in a subtropical climate. In contrast, HH and IC are on the tropical coast and experiencing
 326 sporadic tidal influence. Within the TF site, two groups were dominant, the freshwater
 327 decomposer *Prosthecobacter* (Staley, 1981) and Candidatus Udaeobacter, a common bacterium
 328 in acidic soils (Willms et al. 2021). The genera *Bradyrhizobium*, a common root symbiont
 329 known to survive harsh soil environments, was also abundant (Vaninsberghe et al. 2015).

330 Comparatively, HH was the site most regularly flooded and had the highest soil C:N,
 331 which is a key driver of microbial community composition (Peralta et al. 2013). Thus, this site
 332 had a unique microbial community dominated by two groups within the Actinobacteriota
 333 phylum: *Acidothermus* sp., tolerant to acidic conditions, and Thermoleophilia ASVs, tolerant to
 334 very high temperatures ($> 60\text{ }^{\circ}\text{C}$; Kristjansson & Alfredsson, 1981), suggesting soil
 335 communities enriched in taxa tolerant to hot and low redox and pH conditions. Finally, IC was
 336 dominated by alpha- and gammaproteobacterial groups, typical soil families. These include
 337 Burkholderiaceae, associated with roots in forest soils (Zwetsloot et al. 2020) and
 338 Rhizobiaceae, important for denitrification and N fixation (Wang et al. 2012).

339 Temporal changes in microbial composition varied among sites. Early in the study,
 340 fermenters Citrifermantans, Burkholderiaceae, and Rhizobiaceae were common in TF. While
 341 later, methane oxidisers and cellulolytic fermenters (Anaerolineae groups SBR1031; Xia et al.
 342 2016), *Bradyrhizobium*, and *Paenibacillus* (Firmicutes), a potential cellulose degrader in
 343 tropical freshwater swamps (Kanchanadumkerng et al. 2017) dominated. Similarly, in HH,
 344 *Methanobacterium*, *Methanosarcina*, prokaryotes involved in the methane cycle, the
 345 Burkholderia-Caballeronia-Paraburkholderia genus (Burkholderiaceae family), and *Nevskia*,
 346 common in stagnant freshwater (Cypionka et al. 1981), were common. Later in the study,
 347 dominant groups included Rhodanobacteraceae, Rhizobiales, Syntrophobacter, an anaerobic
 348 group with fermentative metabolism, and Solirubrobacterales, which includes bacteria that
 349 break down leaves in tropical forests (Donald et al. 2020). Finally, in IC, early communities
 350 were found of *Dyella* sp, which can degrade aromatic compounds (Kong et al. 2013), and the
 351 aerobic Ktedonobacteraceae. While later in the incubation, *Paenibacillus* (Firmicutes), a

352 potential cellulose degrader in tropical soils, *Myxococcus* sp. ASVs and *Streptomyces* ASV,
353 essential for cellulose and chitin breakdown (Chater, 2016), were common. While these
354 microbial communities differed in the taxa associated with the tea litter within each site, their
355 putative abilities to break down recalcitrant organic matter (cellulose) and use methane and
356 fermentation pathways suggest a degree of functional redundancy.

357

358 Conclusions:

359 The fate of organic matter in *Melaleuca* wetlands will significantly differ depending on the
360 type of litter that enters the soil. Local *Melaleuca* litter (leaves, barks and roots), which have
361 high lignocellulose content and high C:N, will follow a similar trend as red tea, with low
362 decomposition rates, with half of the C remaining in the soil after two years of incubation.
363 Comparatively, grass, shrubs and labile organic matter transported during floods may follow a
364 similar pattern as green tea. The biomass and C of litter low in C:N will be consumed through
365 colonising microbial communities, immobilising N. The microbial community was
366 characterised by groups of taxa common to freshwater ecosystems or terrestrial forests, with
367 many groups adapted to anaerobic, high-temperature and acidic conditions. There was an
368 evident change in these communities as decomposition progressed, with communities at later
369 experiment stages characterised by cellulose degraders. The microbiome data also suggested
370 metabolic pathways, such as the slow anaerobic turnover of organic C. Overall this study has
371 shown that *Melaleuca* wetlands have conditions that support microbial degradation of organic
372 matter along with long-term sequestration of recalcitrant carbon.

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380

381 Statements and Declarations

382 Conflict of Interests

383 The authors declare no conflict of interests

384

385 Data Availability

386 Amplicon data are available at the European Nucleotide Archive under project accessions
387 PRJEB50314 (tea litter) and PRJEB50315 (bulk soils). Soil physical and chemical properties
388 and organic matter decomposition data will be available open access at Griffith Data
389 Repository (<https://research-repository.griffith.edu.au>) upon acceptance of the manuscript.

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393 Author contributions

394 MFA, SMTT and PM contributed to the conception and design of the study; MFA and NI
395 conducted fieldwork and laboratory work, MFA, NI and STT analysed the data, MFA wrote
396 the first draft and all co-authors contributed to the writing of the final draft of the manuscript.

397

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