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► **To cite this version:**

Dominique Marguerie, T.R. Krause, C. Whitlock. What was burning in the past? Charcoal identifications supplement an early-Holocene fire-history reconstruction in Yellowstone National Park, USA. *Quaternary International*, 2021, 593-594, pp.256-269. 10.1016/j.quaint.2020.09.033 . hal-03122723

**HAL Id: hal-03122723**

**<https://hal.science/hal-03122723v1>**

Submitted on 16 Feb 2021

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1 What was burning in the past? Charcoal identifications supplement an early-Holocene fire-  
2 history reconstruction in Yellowstone National Park, USA

3  
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10  
11 **Abstract:**

12 Analysis of charcoal particles preserved in lake sediments has been frequently used to  
13 reconstruct fire, vegetation and climate history. Larger macroscopic particles (>125 µm) are  
14 attributed to local fires, whereas microscopic charcoal particles (<125 µm), observed on  
15 pollen slides, are a signal of regional fire. Taxonomic identification of the charcoal particles  
16 adds to the fire-history reconstruction by providing information about fuel composition and  
17 past fire conditions. Identification of charcoal can unravel one of the longstanding questions  
18 regarding past fire regimes, namely what was burning. This paper describes an analysis of  
19 charcoal particles preserved in the sediments of a small closed-basin lake in Yellowstone  
20 National Park. Blacktail Pond (44.954°N, 110.604°W; 2012 m elev) is located in steppe  
21 vegetation surrounded by conifer forest. Previous studies of the site have presented a multi-  
22 proxy environmental history of the last 14,650 years. The focus of this investigation was on  
23 the fire history of the early-Holocene (11,500-9900 cal yr BP). Thirty-nine sediment samples  
24 were taken at 0.5 cm intervals to examine macroscopic charcoal particles, with attention on  
25 two large charcoal peaks at 10,970 and 10,200 cal yr BP that registered large or near-site  
26 fires. Twenty-eight samples contained 243 large-enough particles (180-250 µm) to be  
27 examined with scanning electron microscopy. We were able to identify charcoal of conifers  
28 (*Pinus*, *Picea*, and *Abies* or *Juniperus*), *Artemisia* and unidentified deciduous shrubs, and  
29 unidentified monocotyledons. The first fire burned a mixture of fuels, whereas the second  
30 episode burned *Artemisia*, shrubs and herbs but no conifers. The results are consistent with  
31 previous pollen interpretations at Blacktail Pond that indicate an open mixed-conifer forest in  
32 the early Holocene prior to 10,750 cal yr BP, replaced by *Pinus contorta* forest on the slopes  
33 and steppe in the valley. The relative dominance of *Pinus contorta*-type charcoal points to  
34 the importance of fire as a catalyst for the spread of this species.

35  
36 **Keywords:** charcoal, wood anatomy, fire, early-Holocene, vegetation history, Yellowstone  
37 National Park, USA

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41 **1. Introduction**

42  
43 Charcoal data preserved in lake and wetland sediments are used around the world to  
44 reconstruct fire history, and the interest in fire as an Earth system process has led to the  
45 creation of a publicly available global charcoal database (Marlon et al, 2016). Most fire  
46 history reconstructions are based on changes in charcoal abundance through time, which are  
47 interpreted to reflect variations in fuel biomass, fire intensity, and area burned. Large  
48 charcoal peaks in individual records are attributed to fire episodes near the sites (Whitlock  
49 and Larsen, 2001; Marlon et al., 2006).

50  
51 We suggest that taxonomic identification of the charcoal particles preserved in lake  
52 sediments provides additional information about past fire conditions that cannot be easily  
53 inferred otherwise-namely, what burned? Knowing the fuel type, whether from conifers,  
54 hardwoods, shrubs, and/or herbs, and in some cases, assignment to genus or species help  
55 clarify the characteristics of the fire regime as well as the likely fire source area. This

56 information also provides better understanding of vegetation dynamics during times of  
57 climatic transition.

58

59 Following Jones et al. (1997), the term “charcoal” is assigned here to all burned or  
60 charcoalified (charred) plant remains produced during fires, not only woody fragments.  
61 Charcoal particles are distinguished from other dark particles (especially the mineral ones)  
62 by their black color, total opaqueness, high light reflectivity, and their angular and usually  
63 planar shape with sharp edges. When poked, wood charcoal is easily broken into small  
64 angular fragments that often show plant structures.

65

66 Charcoal can be transported in the atmosphere several meters to tens of kilometers before  
67 final deposition and burial (Garstang et al., 1997; Clark et al., 1998; Pisaric, 2002). The travel  
68 distance is a function of the fire characteristics, the conditions of transport, and the particle  
69 size. Microscopic charcoal particles (<125  $\mu\text{m}$ ) come from both local and distant fires  
70 whereas larger macroscopic fragments (>125  $\mu\text{m}$ ) are transported far less from the source  
71 and thus provide a local fire history (Whitlock and Millspaugh, 1996; Carcaillet et al., 2001,  
72 Whitlock and Larsen, 2001, Olsson et al., 2009).

73

74 The external morphology (size and shape) of macroscopic charcoal has been explored using  
75 automated image analysis under a (stereo)microscope. Umbanhowar and McGrath (1998)  
76 examined the area, length, and width of charcoal particles as an indicator of burned  
77 vegetation type. They experimentally produced charcoal of wood, leaves and grasses by  
78 burning samples in open fires and in a muffle furnace at 350°C. They distinguished these  
79 three types of charcoal based on variations in length (along the major axis,  $\mu\text{m}$ ), width (along  
80 the minor axis,  $\mu\text{m}$ ) and area ( $\mu\text{m}^2$ ) of the particles. The elongation metric R (L/W) enabled  
81 them to separate wood (R=2.13 in furnace and 2.23 in open fires) and leaf (R=1.91 and 2.15)  
82 particles from those of herbs (R=3.62 and 4.83). Grass charcoal was longer than either leaf  
83 or wood charcoal. The anatomical structure of plant and the degree of elongation was  
84 preserved even when the particle was broken, enabling separation of charcoal from  
85 deciduous versus herbaceous taxa. This morphological approach was a reasonable tool for  
86 determining the nature of the burned vegetation.

87

88 Enache and Cumming (2006) analyzed charcoal particles from historic fires dating back to  
89 1919 that occurred within a 20-km radius of Prosser Lake (British Columbia, Canada). The  
90 authors observed the morphology of charcoal particles >150  $\mu\text{m}$  diameter from a sediment  
91 core to infer the fuel type (bark, trunk, branch, leaf), temperature of formation, and  
92 taphonomic modifications of the initial morphology. This work led to the establishment of  
93 seven charcoal morphotypes according to shape, ratio of major to minor axis, and  
94 presence/absence of porosity and initial plant structure. The charcoal morphometric features  
95 suggested different fuels during each fire. Some morphotypes, rich in fibers, originated from  
96 tree bark, whereas other types had parenchyma or vessels that originated from wood. Type  
97 F resulted from fibrous fragments or grass cuticles, and type B represented partially burned  
98 fragments. Another type, M, was characterized by its irregular shape, its fragility and high  
99 porosity, and was attributed to intense crown-fire burning of branches or leaves. Type M  
100 charcoal in the core accurately matched the timing of known fires and was considered the  
101 "most accurate record of fire events". Conversely, other types of charcoal (types B, C, M, P,  
102 S) were thought to have been produced by older fires and redeposited or transported from  
103 distant fires. Both Umbanhowar and McGrath (1998) and Enache and Cumming (2006)  
104 provide general insights about the vegetation burned, but they do not provide specific  
105 taxonomic information.

106

107 Soil charcoal research or pedoanthracology focuses on large burned pieces >0.5 mm  
108 preserved in soils, often in association with archeological sites, to provide taxonomic  
109 information on local vegetation in the context of environmental change and human land use  
110 (Carcaillet and Thion, 1996; Nelle et al., 2013; Robin et al., 2013). Identification rarely

111 includes the charcoal preserved in lakes or wetlands, because these sedimentary particles  
112 are usually small (<0.25 mm). Charcoal particles in lake and wetland deposits are often  
113 identified as conifer, hardwood, shrub, or herb and, in some cases, assigned to genera  
114 (*Quercus* sp., *Pinus sylvestris*-type, *Salix* sp., *Betula* sp., Ericaceae) (Whitlock et al., 2006).  
115 In a study of Holocene fire history, Olsson et al. (2009) identified charcoal particles from a  
116 10,500 yr-long record from a peat and lake sediment deposit at Storasjö and Stavsåkra in  
117 southern Sweden. They suggested that both microscopic and macroscopic charcoal records  
118 represented local fire history and showed three major phases of high fire activity controlled  
119 by climate during the early and middle Holocene and by humans during the late Holocene.  
120 These charcoal identifications offered a partial list of fuels because a high quantity of the  
121 particles were unidentifiable.

122  
123 The objective of this paper is to better understand past vegetation and fire conditions through  
124 the use of charcoal analysis on sediment cores taken from a small lake in northern  
125 Yellowstone National Park, USA. Blacktail Pond (44.954°N, 110.604°W; 2012 m elevation) is  
126 surrounded by steppe with conifer forest growing above the site on adjacent rocky buttes and  
127 at higher elevations. A ca. 14,650-yr-long record of vegetation and fire history was  
128 reconstructed at this site and described in Krause and Whitlock (2013) and Huerta et al.  
129 (2009). Moreover, Krause and Whitlock (2013) present geochemical and isotopic data for the  
130 late-glacial to early Holocene period, as part of a multi-proxy investigation of environmental  
131 history.

132  
133 The period of interest for the present study is the early Holocene from 11,500 to 9900 cal yr  
134 BP at Blacktail Pond when summers were becoming progressively warmer and drier as a  
135 result of increasing summer insolation (Krause and Whitlock, 2013). Early postglacial  
136 warming shifted the vegetation from late-glacial parkland to early-Holocene mixed conifer  
137 forest and then steppe and closed pine forest (Krause and Whitlock, 2013) (Fig. 2). Fire  
138 activity, as evidenced by the charcoal record, was also increasing during this transition in  
139 response to warmer conditions and greater fuel biomass (Fig. 3). Two significant fire  
140 episodes occurred at ca. 10,970 and 10,200 cal yr BP, based on large charcoal peaks  
141 (Krause and Whitlock, 2013). In this paper, we describe charcoal particle identifications that  
142 reveal fuel and fire conditions of the early Holocene and supplement previous understanding  
143 of the vegetation and fire history at Blacktail Pond by answering the question: what was  
144 burning?

## 145 146 **2. Study area**

### 147 148 **2.1. Current vegetation and fire regimes**

149  
150  
151 The vegetation of northern Yellowstone is strongly influenced by elevation, aspect and  
152 geology (Despain, 1990). Today, grassland and steppe communities dominated by *Artemisia*  
153 *tridentata* (big sagebrush), *Festuca idahoensis*, *Ericameria nauseosa* (rabbitbrush),  
154 *Sarcobatus vermiculatus* (greasewood, on saline soils), and various herbs are present  
155 generally below 1700 m elevation. Montane and subalpine forest occurs between 1700 and  
156 2900 m elevation and is replaced by alpine tundra at elevations above 2900 m. Within the  
157 forest zone, *Pinus flexilis* (limber pine) and *Juniperus scopulorum* (Rocky Mountain juniper)  
158 occur at low elevations (1700-1900 m elevation), *Pseudotsuga menziesii* (1900-2000 m  
159 elevation) and *Pinus contorta* (2000-2400 m elevation) are present at middle to high  
160 elevations. *Picea engelmannii* (Engelmann spruce), *Abies lasiocarpa* (subalpine fir), and  
161 *Pinus albicaulis* (whitebark pine) dominate high elevation forests and parkland (2400-2900 m  
162 elevation). *Juniperus communis* is present in the understory at all elevations.

163  
164 Blacktail Pond lies within calcareous glacial outwash and is surrounded by *Artemisia*  
165 *tridentata* steppe, which grows at this elevation on fertile soils. *Pseudotsuga* forest is present

166 on adjacent rocky slopes of basalt and andesite, and *Pinus contorta* forest grows on local  
167 rhyolite exposures and throughout the central part of Yellowstone National Park. Small  
168 populations of *Abies* and *Picea* are found in valleys with cold-air drainage, and stands of  
169 *Populus tremuloides* (quaking aspen) grow on lower slopes in moist areas. *Salix* spp, *Betula*  
170 *occidentalis* (water birch), *Scirpus americanus*, *Carex* and *Typha latifolia* are present along  
171 the lake margin.

172

173 Fires, both lightning and human caused, are common in western U.S. forests, and  
174 Yellowstone National Park became internationally famous for large fires in 1988 that burned  
175 3213 km<sup>2</sup>, or about 40% of the park (Schullery, 1989). Regional studies, based on lake-  
176 sediment studies, indicate that fire activity increased during the late-glacial and early  
177 Holocene periods and then decreased after ca. 9000 and again at 5000 cal yr BP (Iglesias et  
178 al., 2018). The conifers of the region have different adaptations to fire that allow for their  
179 survival. For example, the thick bark of *Pseudotsuga* enables these trees to survive low-  
180 intensity surface fires. Similarly, *Pinus flexilis* and *P. albicaulis* tolerate surface fires as the  
181 trees mature and their bark thickens. *P. contorta* is adapted to high-severity fires, because,  
182 even though the trees are killed, its serotinous cones open immediately and release  
183 thousands of seeds. *Abies* and *Picea* have thin bark with branches to the ground and thus  
184 are highly vulnerable to fires. These conifers grow in wet regions where fires tend to be  
185 infrequent crown fires, and regeneration occurs by seed (Knight et al., 2014).

186

## 187 **2.2. Lithology and chronology**

188

189 This study used core BTP08B of Krause and Whitlock (2013), which was collected at the  
190 western margin of the lake from the fen. The sediment from 5.25 m to 8.10 m depth was the  
191 focus of this study. The chronology for Blacktail Pond record was developed from seven  
192 AMS radiocarbon dates on pollen residues, charcoal, and plant macrofossils, and two known  
193 tephra ages (Krause and Whitlock, 2013). The environmental reconstruction was based on  
194 pollen, geochemical properties (elemental geochemistry, oxygen isotopes from authigenic  
195 carbonates), magnetic susceptibility, and macroscopic charcoal. The pollen and charcoal  
196 data and radiocarbon ages are available at Neotomadb.org.

197

198 The original charcoal analysis focused on particles >125 µm in diameter to capture high-  
199 severity fires occurring within a few kilometers of the lake (Krause and Whitlock, 2013;  
200 Huerta et al., 2009). Charcoal accumulation rates (CHAR; particles cm<sup>-2</sup> yr<sup>-2</sup>) were calculated  
201 using CharAnalysis (Higuera et al., 2008). Charcoal concentrations and deposition times  
202 were interpolated into contiguous 25-yr bins (the median resolution of the record), and CHAR  
203 was determined by dividing the time-interpolated concentrations. Long-term trends in CHAR  
204 (background CHAR or BCHAR) were separated from positive deviations (charcoal peaks),  
205 with a 500-yr loess smoother. BCHAR is considered an indicator of woody fuel biomass  
206 (Marlon et al., 2006). Charcoal peaks were assigned to the positive residuals remaining after  
207 BCHAR is removed from the time series and identified above a 95<sup>th</sup> percentile of the noise  
208 distribution (Higuera et al., 2010). These peaks are interpreted as fire episodes (one or more  
209 fires during the time span of the peak).

210

## 211 **2.3. Previous information on fire and vegetation dynamics**

212

213 Pollen data from Krause and Whitlock (2013) describe the late-glacial to early-Holocene  
214 vegetation history at Blacktail Pond (Fig. 2). The period from ca. 14,650 to 11,500 cal yr BP  
215 was a period of cooler-than-present summer conditions, but temperatures were steadily  
216 increasing with rising summer insolation. The postglacial landscape was initially sparsely  
217 vegetated with little soil development. After ca. 12,900 cal yr BP, conditions were warm  
218 enough for *Picea engelmannii* to form a parkland at the site, and after ca. 12,500 cal yr BP,  
219 *Picea*, *Abies*, and *Pinus* (both *P. contorta* and *P. albicaulis/flexilis* pollen types) were present  
220 in open mixed-conifer forest communities. Beginning at ca. 11,500 cal yr BP, the carbonate

221 d<sup>18</sup>O record from Blacktail Pond indicates a sharp increase in summer temperatures, and the  
222 pollen record suggests a more closed forest at ca. 11,350 cal yr BP. By 9900 cal yr BP,  
223 *Pinus contorta* replaced *Picea engelmannii* as the dominant conifer on hill slopes near  
224 Blacktail Pond. Given soil constraints, *Artemisia* steppe probably still covered the valley floor.  
225

226 The charcoal-based fire history followed the vegetation history closely, showing few fires  
227 before 12,000 cal yr BP when vegetation cover was sparse. This was followed by a steady  
228 CHAR rise, suggesting a maximum in fire activity between ca. 12,000 and 10,500 cal yr BP,  
229 and a slight decline thereafter. Noteworthy are the two large fire episodes at ca. 10,970 cal yr  
230 BP and 10,200 cal yr BP (Fig. 3).  
231

232 It is likely rising fire activity due to increasing summer insolation facilitated the transition from  
233 mixed-conifer forest to *Pinus contorta*, given the fact that *Picea* and *Abies* have little  
234 adaptation to fire. The two prominent fire events may have been important triggers in this  
235 transition. The first peak occurred early in the period of mixed-conifer forest, while the  
236 second peak occurred just before *P. contorta* became the dominant species near Blacktail  
237 Pond at 9900 cal yr BP, where it likely occupied upslope rocky areas and steppe was locally  
238 present (Krause and Whitlock, 2013).  
239

240

### 241 3. Methods

242

243 For this study, a new macroscopic charcoal analysis was undertaken on the sediments  
244 above and below the two charcoal peaks. Twenty-six samples of 5 cm<sup>3</sup> were taken every 0.5  
245 to 1 cm in 0.5-cm-thick sections from 671.5 (ca. 11,510 cal yr BP) to 653.5 cm (ca. 10,620  
246 cal yr BP) depth around the first peak and 13 samples were also collected every 0.5 to 1 cm  
247 in 0.5-cm-thick sections from 643 (ca. 10,230 cal yr BP) to 631 cm (ca. 9910 cal yr BP) depth  
248 around the second peak (Table 3).  
249

250 The sediment samples were disaggregated in 30 ml of 5% sodium metaphosphate for 20  
251 minutes before 30 ml of 6% bleach (sodium hypochlorite, NaClO) was added. The samples  
252 were soaked in this solution for another 24 hours to further deflocculate clays and oxidize  
253 organic matter. This procedure made the samples easier to analyze.  
254

255 The samples were then gently washed through two sieves with mesh sizes of 180 and 250  
256 μm (Whitlock and Larsen, 2001). The residues after sieving were subsequently placed in a  
257 gridded petri dish and examined under a stereomicroscope. Of the 39 samples examined  
258 under a stereomicroscope, 28 contained relatively large charcoal particles (>180 μm in  
259 diameter). A total of 243 particles were mounted on stubs for SEM observation.  
260

#### 261 3.1. Identification

262

263 The classical anatomical criteria used to identify modern wood may also be applied to  
264 charcoal (Marguerie and Hunot, 2007). The identification is based on diagnostic characters  
265 of the wood microstructure and comparison of fossil material with modern reference  
266 collections (Trouy, 2015). Identification of charcoal requires examination of the three-  
267 dimensional arrangement of the cell elements that comprise hardwood or softwood. The  
268 three anatomical planes of the ligneous structure are transverse (Tr), radial (R) and  
269 tangential (Tg). The transverse plane is most easily discovered because of the visibility of the  
270 growth rings. It also is the most important section for observing microscopic features and ring  
271 distinctness. Within a longitudinal section (i.e., the vertical orientation of the cells in tree  
272 trunk) are the radial and tangential planes. The radial plane is a vertical surface running from

273 the pith outwards towards the bark; the tangential plane is perpendicular to the rays, and  
274 tangential to the growth rings.

275  
276 Given the small size of the charcoal particles (about 180-250  $\mu\text{m}$  in diameter) preserved in  
277 the Blacktail Pond core, usual identification criteria were not systematically possible. The first  
278 step was to determine if a specimen was ligneous (woody) or not. This identification was  
279 done from exposed planar sections produced by natural breakage. Generally, the charcoal  
280 particles offered only one or two anatomical sections visible along the radial or tangential  
281 planes, and it was impossible to split the particle along a new surface. We had to work with  
282 the existing fractures to observe all the aspects of the wood microstructure.

283  
284 Identification keys were consulted for the details of charcoal particle characteristics (e.g.,  
285 pits, perforation plates). The more fragmented the particle was, the more difficult it was to  
286 identify or even find diagnostic features for identification. In contrast, large "mesoscopic"  
287 charcoal particles (180-500  $\mu\text{m}$ ) often contain two anatomical sections allowing distinction of  
288 monocotyledons, dicotyledons and gymnosperms (Marquer et al., 2012). These longitudinal  
289 sections (either radial or tangential) are particularly informative for identifying of some conifer  
290 genera.

291  
292 In addition, processes related to transport and deposition of charcoal in the lake lead to  
293 fractures, deformities, and impurities that often obscure anatomical features. At Blacktail  
294 Pond, a few mesoscopic charcoal particles were recovered, including isolated wood fibers  
295 and vascular elements that enabled conifer identification. Attention to the anatomical criteria  
296 preserved on the longitudinal planes was critical, even when only a few bundles of fibers  
297 were preserved. In conifers and broad-leaved trees and shrubs, axial tracheids and vessels  
298 form the main part of the wood tissue and these groups cannot be separated. However,  
299 longitudinal sections reveal different combinations of pits that are diagnostic.

300  
301 All wood anatomical features preserved in the charcoal particles at Blacktail Pond were  
302 recorded according to the IAWA list of microscopic features for hardwood and softwood  
303 identification (Wheeler et al., 1989; Richter et al., 2004). Taxonomic identification relied on  
304 identification keys and illustrated atlases of microscopic thin sections (e.g. Schweingruber,  
305 1990). To observe cross-field pitting in the radial section required high magnification, and the  
306 charcoal particles were examined through a SEM using magnifications of up to 5000x to view  
307 these very fine anatomical characters. The SEM sessions took place at the Montana State  
308 University (USA) and the University of Rennes 1 (France). Wood anatomical features were  
309 compared with genera in the charcoal reference collection (Archeosciences Laboratory,  
310 CReAAH, CNRS, University of Rennes 1) and several wood anatomical atlases.

311  
312 The low number of identified charcoal particles at Blacktail Pond (243 particles) did not allow  
313 a quantitative reconstruction, but rather documented the presence of some taxa and the type  
314 of vegetation burned. Charcoal accumulation rates (CHAR) were calculated for particles  
315  $>125$  in diameter and ranged from 0.2 to 2.6 particles  $\text{cm}^{-2} \text{yr}^{-1}$  (Krause and Whitlock, 2013).  
316 There were few particles in the  $>180 \mu\text{m}$  size fraction.

### 317 318 **3.2. Collection of fresh wood samples around the site**

319  
320 The characteristics of the fossil wood anatomy were checked against modern, charred, and  
321 wood specimens. In addition, a reference collection was also made of wood from living trees  
322 and shrubs in the vicinity of Blacktail Pond. Wood samples from several plants of each  
323 species were charred in an open furnace after drying at 50°C for 30 hours. After  
324 carbonization (at 350°C with the time depending on the size of the wood), three anatomical  
325 sections were obtained by scalpel or simply by breaking the charcoal by hand. The  
326 fragments of charred tissue were mounted on metal stubs for SEM examination.

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## 4. Results and Discussion

### 4.1 Wood anatomy of living trees and shrubs around Blacktail Pond

We collected modern wood specimens from three pines and four broad-leaved shrubs from the northern Yellowstone region: *Pinus albicaulis*, *P. contorta*, *P. flexilis*, *Artemisia tridentata*, and *Sarcobatus vermiculatus*. For these taxa, wood identifications were based on burned living woody plants. Other conifers were not sampled: *Juniperus scopulorum*, *Picea engelmannii*, and *Abies lasiocarpa*. Information about the wood anatomy of the living conifers at the species or genus level, especially the unsampled taxa, was obtained from several atlases (Panshin and de Zeeuw, 1980; Barefoot and Hankins, 1982; Hoadley, 1990). In addition, we made use of the atlases of Schweingruber (1990) and Vernet et al. (2001), although neither deals with western U.S. trees. The European collection of wood at the laboratory of Rennes, France was also examined for this project. Two papers on identification of North American wood species (Kukachka, 1960; Minnis, 1987) and a special issue of IAWA journal (Richter et al., 2004) were also consulted. For each charcoal type, descriptions were made in accordance with the on-line InsideWood database (IAWA Committee, 1989; InsideWood, 2012).

#### 4.1.1. Wood anatomy of the conifers

The anatomical distinction between conifer (softwood) and angiosperm (hardwood) is straightforward. Conifer wood is less complex because tracheids function for both conduction and support, and their shape is rather different from that of hardwood vessels. Identification of different conifer taxa, however, is generally more difficult than for hardwoods (Kukachka, 1960). Wood anatomy criteria allow conifer identification to genus, but species-level identification is difficult or even impossible.

Among the conifer genera likely to be identified in the Blacktail Pond core, the primary diagnostic features was the presence or not of resin ducts, the height of rays, and the nature of cross-field pitting (i.e., pits intersecting walls of an axial tracheid and a ray parenchyma cell) (Table 1).

Five types of pits occur in the earlywood or springwood and vary in terms of frequency, arrangement, form and size (Ritcher et al., 2004; Schweingruber, 1990, 2011). The types range from a single large pit that occupies most of the area of the cross-field, as in the case of some pines, to very small pits that occupy only a small portion of the cross-field areas, as in *Picea*.

In the case of small charcoal particles where it is impossible to observe the three anatomical sections, the radial plane is often the most obvious one. It is also the crucial section for discerning decisive features for the identification of conifer wood. For example, the characteristics used to identify small fragments of *Abies* are the absence of resin canals, transversal tracheids, nodular ray parenchyma, and the presence of two to four taxodioid pits per cross-field. Crystals appearing in the form of regular prisms, cubes, or octahedra are commonly found in *Abies* species (Schweingruber, 1990). In *Juniperus* wood, resin canals and transversal tracheids are also absent, and key features are ray parenchyma that have abundantly nodular ends walls and cupressoid cross-field pitting. In *Picea*, resin canals are surrounded by rather thick-walled epithelial cells, the ray tracheids are present with a smooth and wavy wall, ray parenchyma cell walls are thick, and cross-field pitting is piceoid-type (Marguerie et al., 2000; Schweingruber, 1990). Crystals in ray cells occur occasionally in *Picea* wood. Finally, small fragments of *Pinus* wood in radial section show abundant and large resin canals with thin-walled epithelium cells, ray tracheids that are dentate to reticulate, and fenestriform or pinoid pits in the cross-fields (Table 1).



383  
384 Species identification of *Pinus* is difficult or impossible. However, the genus can be  
385 subdivided into two groups on the basis of anatomical traits like ray cells, ray tracheids and  
386 the type of cross-field pitting, all of which can be discerned under high magnification (Table  
387 2). *Pinus contorta* wood has rays of medium height in tangential section (1 to 12 cells, mainly  
388 5) (Fig. 4b), ray tracheids that are distinctly dentate to reticulate walls, and one to four, but  
389 generally two, pinoid ray pits per cross-field. Each pit has an irregular shape and size (Fig. 4c  
390 and 4d). *Pinus albicaulis* wood has short rays with only one to ten cells (although sometimes  
391 more), with an average height of three to four cells (Fig. 4f). It also generally has one large,  
392 simple, fenestriform (window-like) ray pit per cross-field, rarely two (Fig. 4g and 4h). *Pinus*  
393 *flexilis* wood has short rays in tangential section with one to eight cells (three to four on  
394 average) (Fig. 4j). Cross-field pitting is also fenestriform, with on average one or two pits per  
395 earlywood cross-field (Fig. 4k and 4l).

396  
397 Using these criteria, we were able to identify *Pinus*, *Picea*, and *Juniperus* or *Abies* genera in  
398 the charcoal particles at Blacktail Pond. It was also possible to separate *Pinus contorta* from  
399 *P. albicaulis* and/or *P. flexilis* on the basis of the cross-field pitting and the walls of ray  
400 tracheids.

#### 401 402 4.1.2. Wood anatomy of the broad-leaved shrubs

403  
404 In contrast to gymnosperms, the xylem of angiosperm woods contains vessels and is thus  
405 easy to distinguish from conifers by its more complex wood anatomy. Four common shrubs  
406 found in the steppe vegetation of northern Yellowstone National Park were examined.

407  
408 The wood of *Artemisia tridentata*, an Asteraceae shrub, is semi-ring porous (Fig. 5a and 5b)  
409 and contains vessels with spiral thickenings and simple perforations. Rays are large (6-8  
410 cells) and heterogenous (Fig. 5c and 5d). In the case of small charcoal particles in the  
411 sedimentary record, the key feature was vessel walls: numerous, big, lentoid, with lined-up  
412 pits (Fig. 5e).

413  
414 *Ericameria nauseosa*, another Asteraceae, has ring porous wood with large (5 to 11 cells)  
415 (Fig. 5f, 5g and 5h), heterogenous (square cells) rays (Fig. 5j), and simple perforations in the  
416 vessels (Fig. 5i). Vessel walls are adorned by rounded pits (Fig. 5i).

417  
418 *Purshia tridentata*, a Rosaceae shrub, has a semi-ring porous wood (Fig. 5k and 5l) with thin  
419 (1-3 cells) homogenous rays (Fig. 5m and 5n). Perforations inside the vessels are simple and  
420 inter-vessel walls bear medium sized and few pits (Fig. 5m and 5n).

421  
422 *Sarcobatus vermiculatus* (greasewood), an Amaranthaceae shrub, has semi-ring porous  
423 wood (Fig. 5o), thin rays (1 cell) (Fig. 5p), simple perforations in vessels (Fig. 5r and 5s), and  
424 rounded pits frequently organized into lenticular openings (Fig. 5q, 5r and 5s).

425  
426 So, in the case of the wood of broad-leaved shrubs near Blacktail Pond, the density,  
427 arrangement, shape, and size of vessel pits were diagnostic features for identification.

#### 428 429 4.2. Charcoal particle analysis in the Blacktail Pond sediment core

430  
431 Charcoal >180 µm in diameter was present in low amounts and variable condition and  
432 comprised 28 of the 243 samples. Despite the small size of most particles, wood anatomy  
433 was often well preserved. Most of the charcoal particles were pieces of burned wood (67.1%  
434 of wood + herb), and some could be identified to genus (11.4%). Samples from the two large  
435 CHAR fire peaks had the most identifiable particles and also were the most taxonomically  
436 diverse. These two fire episodes suggest large fires or fires near the lake over a short period

437 of time. Samples from the period before the first peak that dated from ca. 11,350 to 11,170  
438 cal yr BP also contained abundant and diverse wood (Table 3).

439

#### 440 4.2.1. Remains of monocotyledons

441

442 Charred remains of herbs, including parenchymatous organs that resembled stems, were  
443 commonly observed in three-dimensional views under high-magnification SEM (Fig. 6a to 6f)  
444 (Hather, 1993). Some elements were identified as stems when viewed in trilobal cross-  
445 section (Fig. 6b) and laterally (Fig. 6a and 6d).

446

447 The vascular tissue or xylem of herbs was preserved, almost intact and without serious  
448 distortion in some specimens. Phloem was also recognized by its position relative to the  
449 xylem. In Figure 6b, the phloem is internal and forms a cylinder surrounded by a crown of  
450 xylem. The xylem elements have a polygonal transversal section (Fig. 6c). Sometimes,  
451 deformation of the xylem revealed its staged elements laterally (Fig. 6e). Figure 6d shows  
452 stomata in the lower epidermis of an aerial stem.

453

#### 454 4.2.2. Remains of broad-leaved taxa

455

456 Among the burned wood of broad-leaved shrubs in the Blacktail Pond record, two specimens  
457 (at ca. 10,970 and 10,185 cal yr BP) had well preserved vessels and wall ornamentation  
458 similar to the pitting structure observed in *Artemisia tridentata* wood (Fig. 5e). Numerous  
459 lined-up, lentoid pits were easily distinguished from other plant tissue (Fig. 6g, 6h, 6i).

460

461 Several vessel wall pits were very simple and could not be identified (Fig. 6j, 6k, 6l).

462

#### 463 4.2.3. Remains of conifers

464

465 The presence of vertical or longitudinal tracheids with uniseriate (very common) or biseriate  
466 opposite big bordered pits was characteristic of conifer wood (Fig. 7a to 7e). Small charcoal  
467 particles with tangential sections showed resin canals with epithelial cells and high rays (Fig.  
468 7f, 7g). Some tracheids had helical cracks in their walls, which is a common feature of  
469 compression wood in conifers.

470

471 The radial section was best for distinguishing among conifers. This plane is achieved through  
472 natural breakage along the rays and was found in several charcoal fragments from Blacktail  
473 Pond. *Pinus* species were identified by the cross-field ray pits. Two types of pits were  
474 observed in the *Pinus* wood. The first type was fenestriform pitting, which is window-like, with  
475 a large, simple ray pit occupying almost the entire cross-field. Rarely do wood fragments  
476 have two fenestriform pits per cross-field (Fig. 7h, 7i, 7j). However, in the Yellowstone region  
477 *Pinus albicaulis* and/or *P. flexilis* share this feature (Fig. 4g, 4h, 4k, 4l). The second type,  
478 pinoid pitting, is made by ray pits that are simple and more or less large. Usually two to four  
479 pinoid ray pits can be observed per cross-field. This type of pitting is characteristic of *Pinus*  
480 *contorta* (Fig. 7k, 7l, 7m).

481

482 Wood fragments with ray tracheids and without fenestriform or pinoid pits in the cross-fields  
483 were assigned to *Picea* (Fig. 7d, 7n).

484

485 A third type of cross-field ray pits was taxodioid: large pit aperture with narrow border. When  
486 charcoal particles had such pits and no tracheids, we assigned them to either *Abies* or  
487 *Juniperus* (Fig. 7o to 7r).

488

489 In conifer tracheids, two types of cell contents were present, one of which forms pyramidal  
490 crystals or druses (Fig. 7s, 7t). This common feature was not diagnostic.

491

#### 492 4.2.4. Unidentifiable charcoal

493  
494 16.5% of charcoal particles in the large size range (180 - 250  $\mu\text{m}$ ) lacked any discernible  
495 cellular structure. Remains of leaves were often found as part of charred plant tissues  
496 (11.5%), and hairs (trichomes) and stomata were also observed (Fig. 6m, 6n, 6o). Similarly,  
497 small fragments of vesicular charcoal were noted and could have come from any  
498 parenchymous tissue.

499

500

### 501 4.3. Charcoal analysis results

502

503 The charcoal particles that could be identified include *Pinus*; *Picea*; *Abies* or *Juniperus*; a  
504 deciduous shrub, most likely *Artemisia*; and unidentifiable monocotyledons. *Pinus albicaulis*  
505 and/or *P. flexilis* (these species cannot be distinguished by their pollen) was present in the  
506 vegetation at ca. 11,350 cal yr BP, during a time of mixed-conifer forest (Table 3). *P.*  
507 *albicaulis* and/or *P. flexilis* charcoal was also noted at ca. 9980 cal yr BP, when these taxa  
508 were less abundant locally and may have come from more distant fires. The presence of  
509 *Pinus contorta* charcoal at ca. 11,180 cal yr BP aligns well its rising abundance in the pollen  
510 record. *P. contorta* and/or *Picea* were also identified in three charcoal samples dating to ca.  
511 10,780 cal yr BP. *Artemisia* charcoal was identified at ca. 10,990, 10,180, and tentatively at  
512 9920 cal yr BP, but shrub charcoal was present throughout the record, as was charcoal from  
513 herbaceous taxa. The presence of both hardwood and softwood charcoal suggests that most  
514 fires burned both forest and steppe. However, remains of hardwood and monocotyledons  
515 comprised most of the charcoal from two samples dating to ca. 10,650 cal yr BP, suggesting  
516 that steppe was the main fuel at that time.

517

518 It is interesting that the first large fire episode at 10,970 cal yr BP consisted of burned  
519 conifers (*Picea*, *Abies* and/or *Juniperus*), shrubs and herbs, but no *Pinus* charcoal was  
520 identified. This assemblage is consistent with an open mixed-conifer forest, in which pine  
521 was not dominant. The second fire episode at 10,200 cal yr BP contained shrub and herb  
522 charcoal and no conifer charcoal, implying a local fire that primarily burned steppe vegetation  
523 around the lake (Table 3).

524

525 Two centuries before the first fire peak (from ca. 11,150 to 10,970 cal yr BP) and one century  
526 after the second fire peak (from ca. 10,200 to 10,085 cal yr BP), the cores contained no  
527 charred macroscopic plant remains.

528

529

### 530 5. Conclusion and perspectives

531

532 The Blacktail Pond study contributes new information to our understanding of the fire history  
533 of northern Yellowstone, and the development of vegetation in the northern Rocky Mountains  
534 during the late-glacial and early-Holocene periods. The pollen and charcoal records show  
535 that increased fire activity played a significant role in the transition from late-glacial *Picea*  
536 parkland to early-Holocene vegetation of *Pinus contorta* forest and steppe. The charcoal  
537 identifications enrich this reconstruction by showing that large fire episodes may have  
538 facilitated the transition. In the case of the fire at ca. 10,970 BP, conifers and shrubs were  
539 both fuel types near the lake. The second fire episode at ca. 10,200 cal yr BP, the presence  
540 of hardwood charcoal and no softwoods implies the development of steppe vegetation near  
541 the site with conifers restricted to more distant settings on rocky buttes and at higher  
542 elevations. The second fire must have been fairly local but probably intense to produce so  
543 much charcoal. Another interesting insight is the presence of *Pinus contorta* charcoal as  
544 early as ca. 11,180 cal yr BP, even before it became the dominant vegetation type. Fire and  
545 *P. contorta* are closely linked at present, given its ability to rapidly regenerate after fire. The

546 record at Blacktail Pond suggests that fire facilitated its increasing dominance in the early  
547 Holocene.

548

549 This study shows that it is possible to identify a part of the very small charcoal remains  
550 preserved in the lake sediments with SEM identification. The results provide unique insights  
551 as to the fuel composition. The next phase of research will focus on examining charcoal from  
552 other sites in the Yellowstone region to see if comparable shifts in fuel were occurring at  
553 times of changing climate, vegetation, and fire activity.

554

### 555 **5.1. Perspectives on the identification of very small fragments of burnt wood**

556

557 Identification of charcoal fragments is a valuable tool in reconstructing past environments  
558 and determining disturbances. This paper illustrates the potential for the identification of  
559 charcoal particles to be part of paleoenvironmental reconstructions. We believe that this  
560 approach deserves consideration in other settings.

561

562 The analysis of charcoal in lake or wetland sediment cores is challenged by the smallness  
563 and relative scarcity of suitable material as well as preservation issues related to charcoal  
564 transport and deposition into such sites. These data complement other quantitative proxies in  
565 building an environmental reconstruction (e.g., pollen, macroscopic charcoal, lithologic and  
566 geochemical data).

567

568 Other techniques can improve the results in this study. Reflected Light Microscopy (RLM) is  
569 used to identify sizable and numerous charcoal fragments by studying fresh break surfaces  
570 (Marguerie and Hunot, 2007). In case of very small fragments, Scanning Electron  
571 Microscopy (SEM) is best suited for detailed observation of small irregular surfaces with high  
572 relief. The increasing number of tabletop SEMs in laboratories now enables this type of  
573 analysis at high magnifications and great depth of field.

574

575 Because it is difficult to make transversal or longitudinal sections by breaking, splitting or  
576 slicing to observe wood anatomical features on small charcoal particles, High-Throughput X-  
577 ray Computed Tomography (HT- $\mu$ CT) also opens up new opportunities to reveal fine  
578 anatomical characters. It has been shown to be effective in imaging anatomical features that  
579 cannot be observed on exposed surfaces (Haneca et al., 2012). The ability to yield images of  
580 internal wood structure without having to break or cut the sample is a powerful tool to identify  
581 diagnostic features (3D observation) (Haneca et al., 2012). The technique allows the  
582 production of unlimited virtual sections and clean digital cuts in any direction on very small  
583 samples (Van den Bulcke et al., 2009; Hubau et al., 2013). The resolution obtained is about  
584 2  $\mu$ m, which is sufficient to reveal many features of interest for wood anatomy (Mayo et al.,  
585 2010). However, the resolution of HT- $\mu$ CT is not as high as with SEM, and fine details in the  
586 vessels and fibres, and pitting may not be visible (Hubau et al., 2013). Another related  
587 technique, nanotomography or X-ray nano Computed Tomography (nanoCT), offers higher  
588 resolution and is also excellent for seeing features that are not clearly visible under SEM  
589 because they are concealed by coatings or mineral deposits (Van den Bulcke et al., 2009;  
590 Hubau et al., 2013).

591

592

593 -----

594

### 595 **Acknowledgements**

596

597 SEM sessions took place in the Imaging and Chemical Analysis Laboratory (ICAL) at  
598 Montana State University, Bozeman, thanks to Nancy Equall and the Centre de Microscopie  
599 Electronique à Balayage et de microAnalyse (CMEBA) of the University of Rennes 1, thanks  
600 of Francis Gouttefangeas and Loïc Joanny. Yann Rantier carried out systematic quality

601 adjustments of SEM images at the ECOBIO research unit of the CNRS and University of  
602 Rennes 1. We also acknowledge Peter Kováčik from the PaleoResearch Institute at Golden,  
603 Colorado (USA) and Alan Crivellaro from the Department Land, Environment, Agriculture and  
604 Forestry (TESAF), University of Padova (Italy) for their advice and help on problematic  
605 specimens. The MSU research was supported by NSF grant (0966472) to CW. We thank two  
606 anonymous reviewers for their helpful comments on the manuscript.

607 -----

608

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730 examination of modern charcoal accumulation in Yellowstone National Park, USA. *The*  
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733 -----

734  
735 **Figure captions**

- 736 Figure 1. Location of Blacktail Pond.  
737 Figure 2. Pollen diagram from Blacktail Pond (ex. Krause and Whitlock, 2013)  
738 Figure 3. Fire proxies at Blacktail Pond (ex. Krause and Whitlock, 2013)

739 Figure 4. Scanning electron microphotographs of burnt wood of three pine species in the  
740 northern Rocky Mountains: a. *Pinus contorta*, transverse section; b. *P. contorta*, tangential  
741 section; c, d. *P. contorta*, radial section; e. *P. albicaulis*, transverse section; f. *P. albicaulis*,  
742 tangential section; g, h. *P. albicaulis*, radial section; i. *P. flexilis*, transverse section; j. *P.*  
743 *flexilis*, tangential section; k, l. *P. flexilis*, radial section

744 Figure 5. Scanning electron microphotographs of burnt wood of four shrubs in the northern  
745 Rocky Mountains: a, b. *Artemisia tridentata*, transverse section; c. *A. tridentata*, tangential  
746 section; d, e. *A. tridentata*, radial section; f, g. *Ericameria nauseosa*, transverse section; h. *E.*  
747 *nauseosa*, tangential section; i, j. *E. nauseosa*, radial section; k, l. *Purshia tridentata*,  
748 transverse section; m. *P. tridentata*, tangential section; n. *P. tridentata*, radial section; o.  
749 *Sarcobatus vermiculatus*, transverse section; p, q, r. *S. vermiculatus*, tangential section; s. *S.*  
750 *vermiculatus*, radial section

751 Figure 6. Charred remains of monocotyledons, broadleaf woods and leafs from Blacktail  
752 Pond by a scanning electron microscope: a. Stem of monocotyledon; b, c. Trilobal cross-  
753 section stem of monocotyledon with visible phloem and xylem; d, e. Details of xylem  
754 elements of stem; f. Xylem and phloem in longitudinal section; g, h, i. Particular pits of the  
755 vessel walls *Artemisia* sp.; j, k, l. Simple pits of the vessel walls in unidentifiable broadleaf; m,  
756 n, o. Fragments of leaves with hairs (trichomes) and stomata

757 Figure 7. Charred remains of conifers from Blacktail Pond  
758 a, b, c, d, e. Woody fragments of conifers; d, n. wood fragment of conifer with ray tracheids; f.  
759 Tangential section showing low rays of conifer; g. Possible resin canal and rays in tangential  
760 section; h, i, j. *Pinus* with fenestriform cross-field pitting; k, l, m. *Pinus* with pinoid cross-field  
761 pitting; o, p, q, r. Conifer with taxodioid cross-field pits; s. Crystals in tracheids; t. Other type  
762 of crystals in tracheids

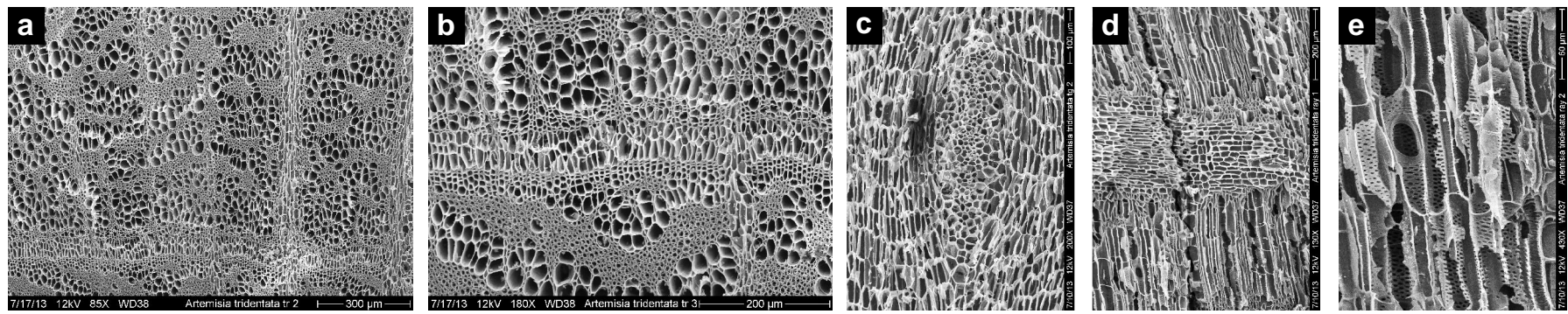
763 Table 1. Main wood anatomical features of the common genera of conifers of the northern  
764 Rocky Mountains around Blacktail Pond

765 Table 2. Main key characteristics of microscopic anatomical features in the wood of three  
766 northern Rocky Mountains pines around Blacktail Pond

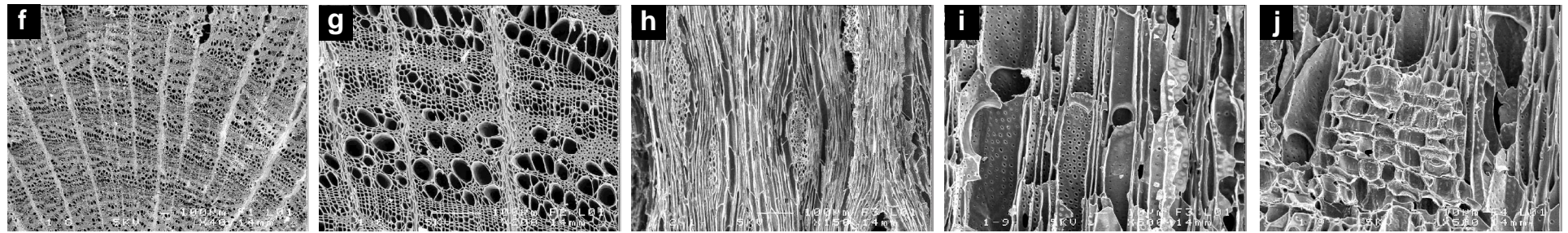
767 Table 3. Results from charcoal analyses of the sediment of Blacktail Pond

768

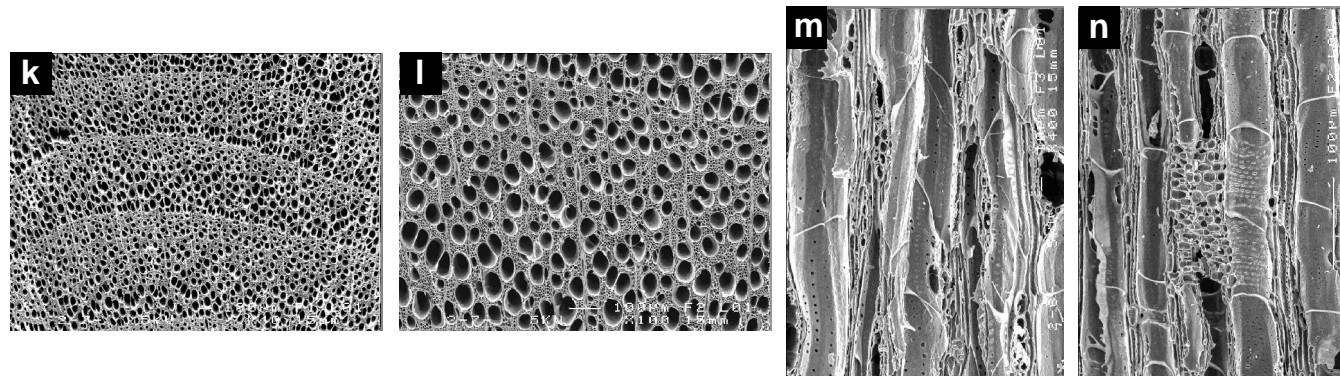




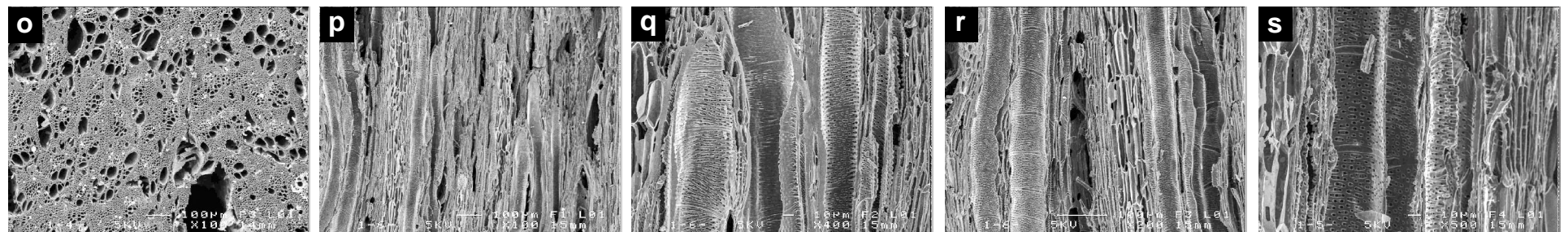
*Artemisia tridentata*



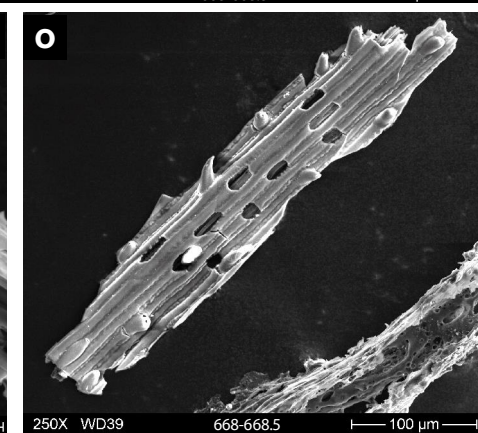
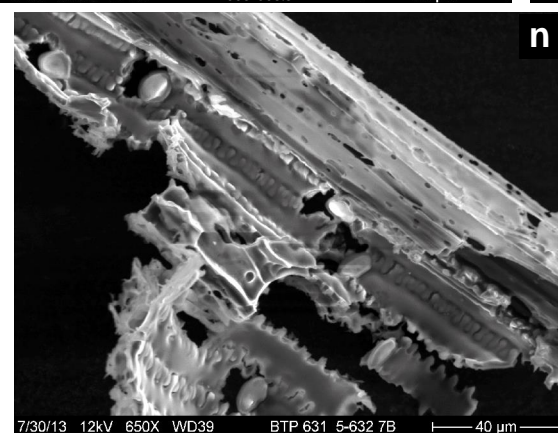
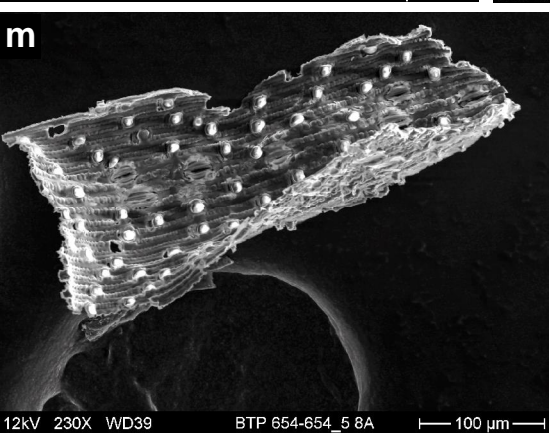
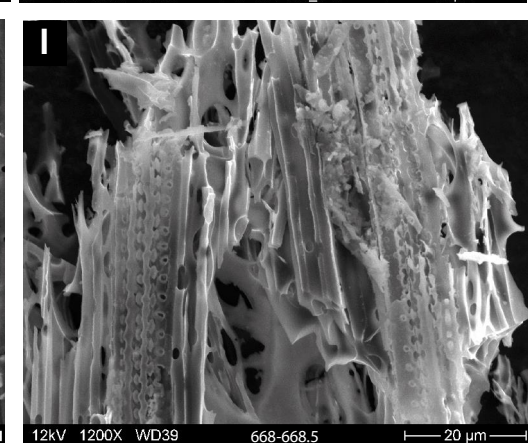
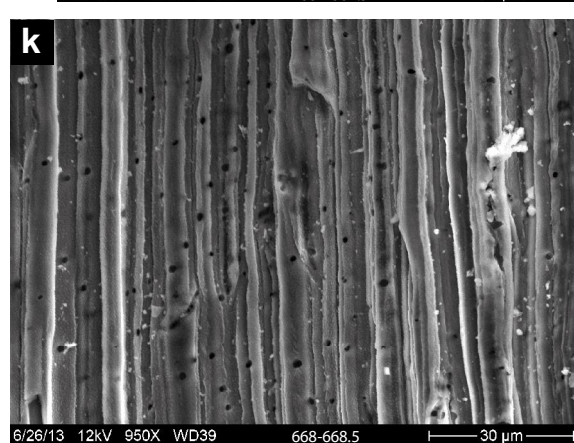
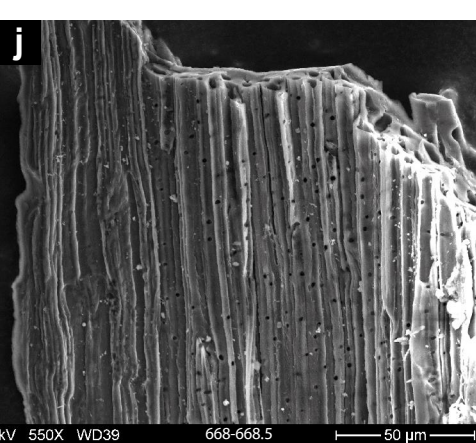
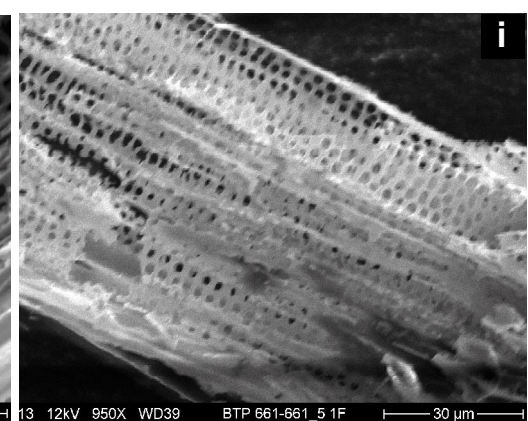
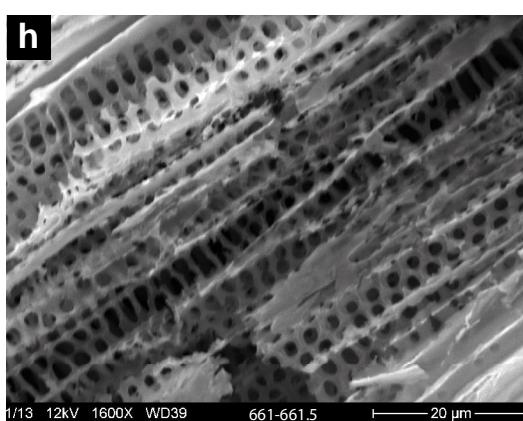
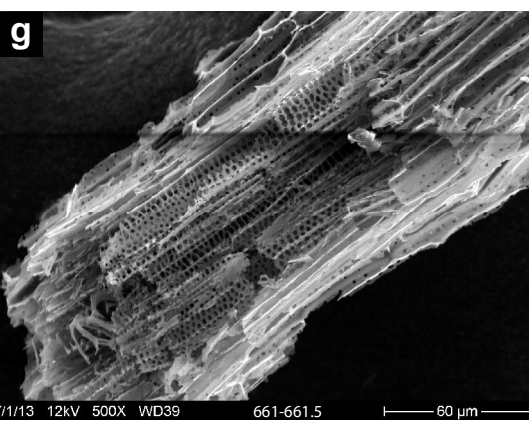
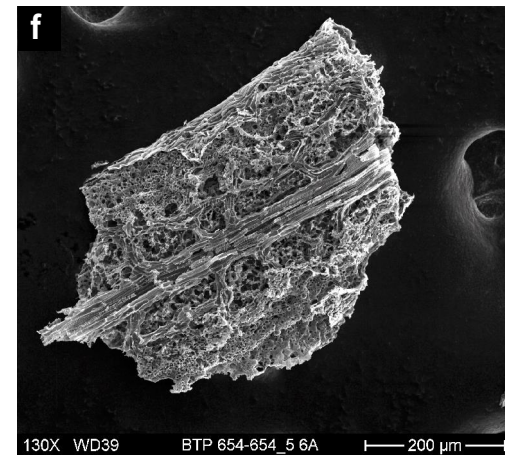
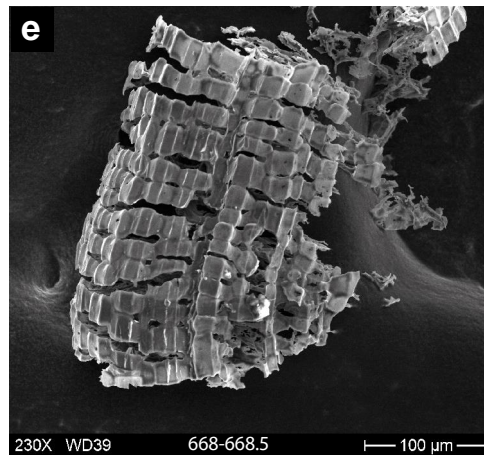
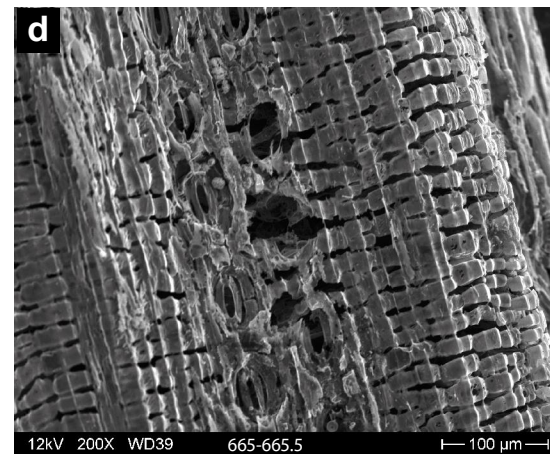
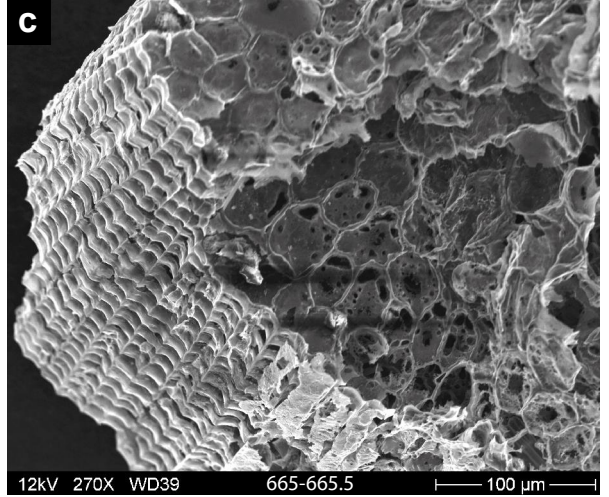
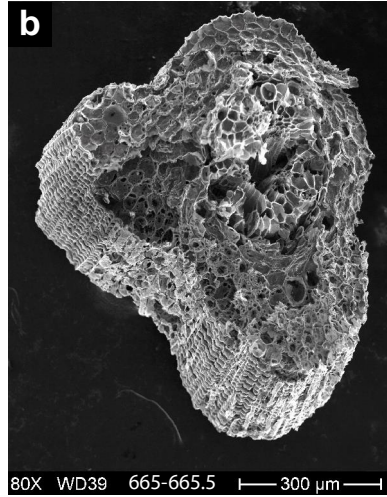
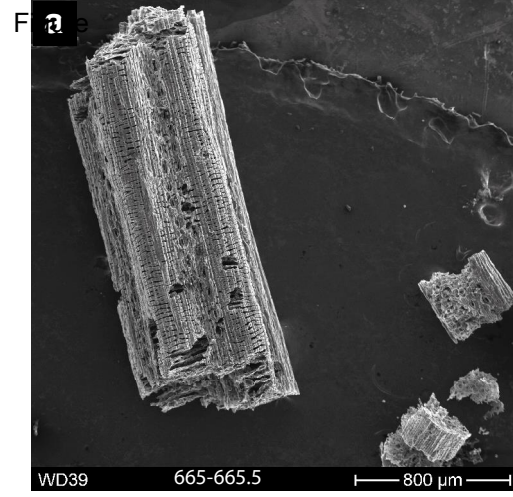
*Ericameria nauseosa*

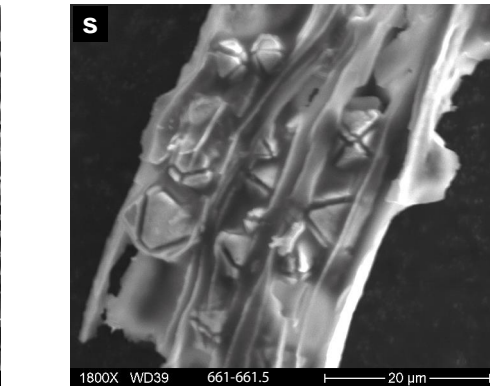
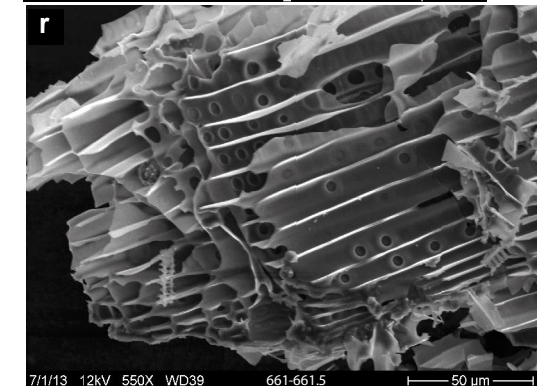
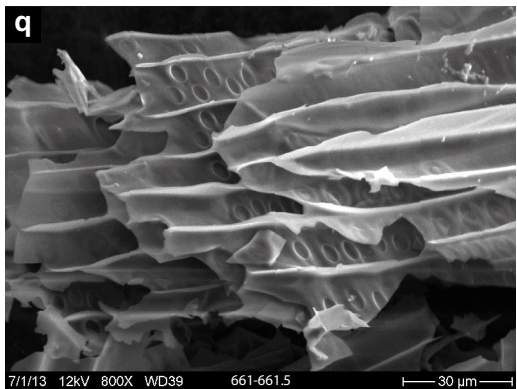
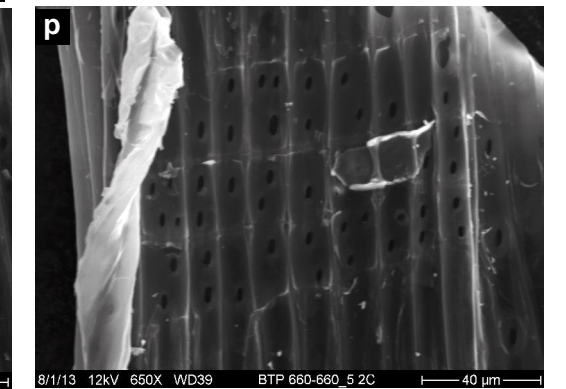
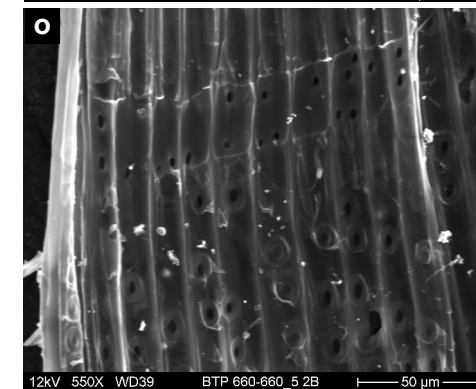
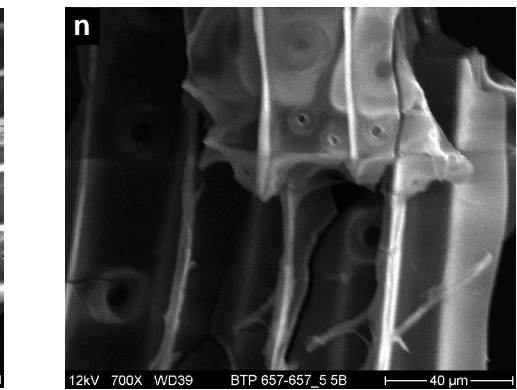
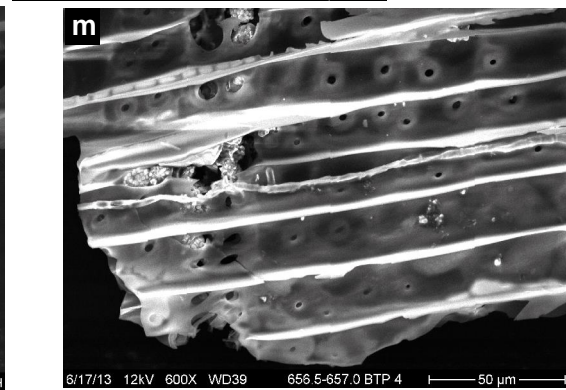
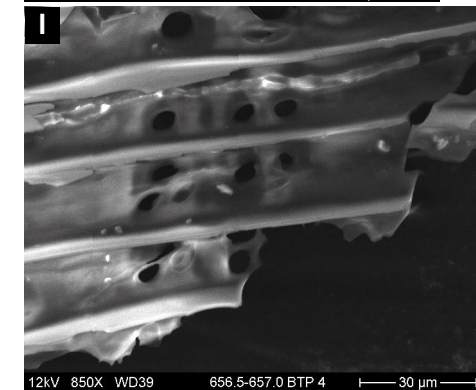
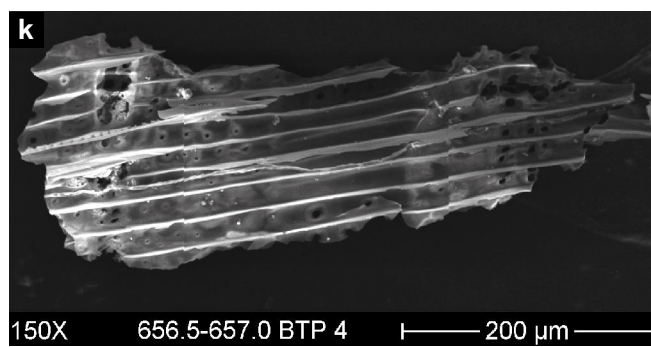
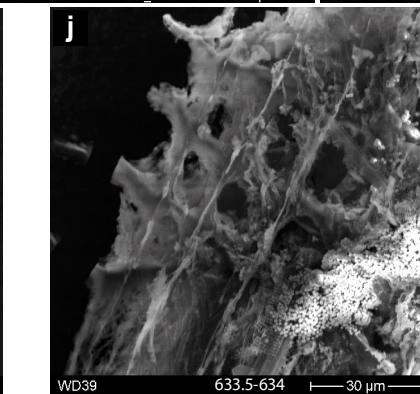
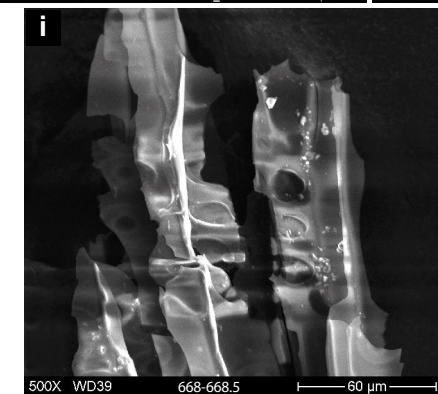
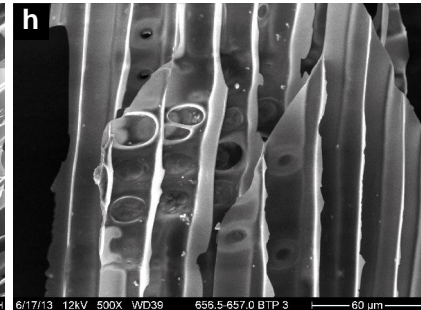
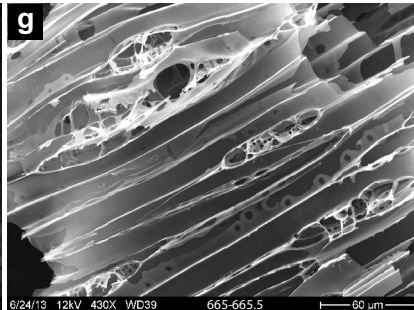
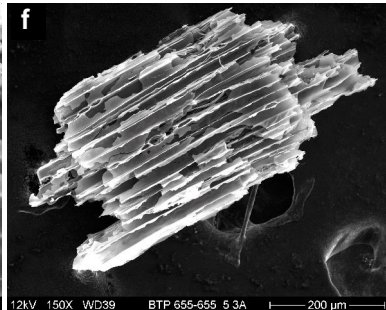
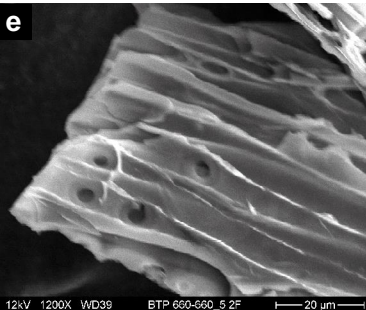
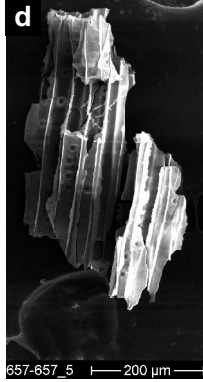
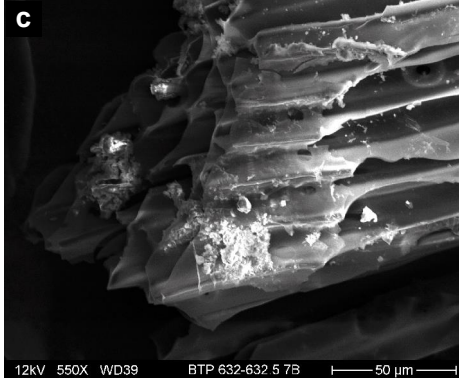
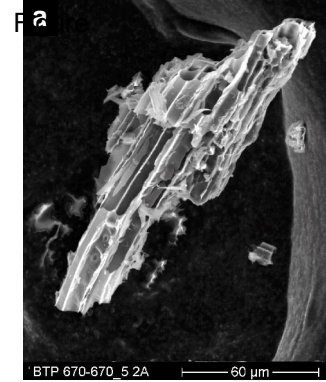


*Purshia tridentata*



*Sarcobatus vermiculatus*





1 No conflict of interest regarding this work on “What was burning in the past? Charcoal  
2 identification supplements early-Holocene fire-history reconstruction in Yellowstone National  
3 Park, USA”.

4

5 Dr. Dominique Marguerie, Dr. Teresa Krause, Dr. Cathy Whitlock

6

1 All the authors signing this paper “Charcoal identifications supplement an early-Holocene  
2 fire-history reconstruction in Yellowstone National Park, USA” declare that they approve the  
3 submission of their work to *Quaternary International*.

4

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8

Wood anatomical features		<i>Abies sp.</i>	<i>Juniperus sp.</i>	<i>Picea sp.</i>	<i>Pinus sp.</i>
Resin canals	Presence	No	No	Yes surrounded by rather thick-walled epithelial cells	Yes with thin-walled epithelium cells
Rays	Tracheids			with smooth, wavy walls	with dentate to reticulate walls
	Height (nb of cells)	15 to 25	2 to 5 (less than 15)	10 -15 cells, upper limit 25	8 to 15 cells
Cross-field pitting	Fenestriform or "window like"				X
	Pinoid				X
	Piceoid			X	
	Cupressoid		X		
	Taxodioid	2 to 4	X ?		

Wood anatomical features		<i>Pinus albicaulis</i>	<i>P. contorta</i>	<i>P. flexilis</i>
Ray cells	Height (nb of cells)	1 to 10 (average 3-4)	1 to 12 (average 5)	1 to 8 (average 3-4)
Ray tracheids		Yes	Yes with dentate to reticulate walls	Yes
Cross-field pitting	Fenestriform or "window like"	1 rarely 2		1 or 2 frequently 2
	Pinoid		1 to 4 frequently 2	







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**Table**

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Whitlock\_Revised.xlsx





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**Table**

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**Table**

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