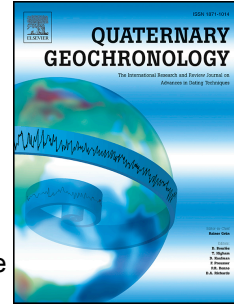


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Charcoal chronology of the Amazon forest: A record of biodiversity preserved by ancient fires

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1 **Charcoal chronology of the Amazon forest:**  
2 **a record of biodiversity preserved by ancient fires**

3  
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## 23 **Abstract**

24 The Amazon region holds a wide variety of ethnic groups and microclimates, enabling  
25 different interactions between humans and environment. To better understand the  
26 evolution of this region, ancient remains need to be analysed by all possible means. In  
27 this context, the study of natural and/or anthropogenic fires through the analysis of  
28 carbonized remains can give information on past climate, species diversity, and human  
29 intervention in forests and landscapes. In the present work, we undertook an  
30 anthracological analysis along with the  $^{14}\text{C}$  dating of charcoal fragments using  
31 accelerator mass spectrometry (AMS). Charcoal samples from forest soils collected  
32 from seven different locations in the Amazon Basin were taxonomically classified and  
33 dated. Out of the 16 groups of charcoal fragments identified, five contained more than  
34 one taxonomic type, with the Fabaceae, Combretaceae and Sapotaceae families having  
35 the highest frequencies.  $^{14}\text{C}$  charcoal dates span ~6000 years (from 6876 to 365 yr BP)  
36 among different families, with the most significant variation observed for two fragments  
37 from the same sampling location (spanning 4000  $^{14}\text{C}$  yr). Some sample sets resulted in  
38 up to five different families. These findings demonstrate the importance of the  
39 association between anthracological identification and radiocarbon dating in the  
40 reconstruction of paleo-forest composition and fire history.

41

42 **Keywords:** Radiocarbon; AMS; Anthracology; ancient fires; charcoal; biodiversity

43

## 44 **Introduction**

45 The rainforests of South America present unique environmental features and their  
46 central role in the regulation of Earth's climate is currently widely recognized (e.g.  
47 Lenton et al. 2008). A complex set of physical, chemical, and environmental

48 characteristics in Amazonia has allowed some of the most diverse forms of life to  
49 flourish. The Amazon Basin has undergone climate variation that has affected forest  
50 extent and composition (Wang et al. 2017).

51 For many years, it was thought that Amazonia was relatively unaffected by humans  
52 prior to the arrival of Europeans (Denevan 1992). However, evidence has emerged  
53 pointing to the existence of complex and sedentary societies in some regions of  
54 Amazonia (Neves 2005). This has led to debate into the onset, extent, and timescale of  
55 the early impact of humans in the area, including the frequency of fire, both as a tool  
56 and applied accidentally (Willis et al. 2004; Heckenberger et al. 2007; Denevan et al.  
57 2011; Levis et al. 2012). In some areas, populations modified their surrounding  
58 environment according to their needs, leaving behind testimonies to their occupation,  
59 such as domesticated species, artefacts, and Anthropogenic Dark Earth soils (*Terra*  
60 *Preta de Índio*) (e.g. Clement 1999; Petersen et al. 2001; Neves et al. 2003; 2004).

61 Fire was the main tool used by indigenous people to transform and manage landscapes  
62 (Erikson 2008). Hence, fire regimes are linked with past human disturbance during the  
63 Holocene and charcoal records have been used to identify such activities (Bush et al.  
64 2008; Mayle and Power 2008). Wildfires may also occur in Amazonian forests in  
65 extremely dry periods (Piperno and Becker 1996; Wang et al. 2017). During fire  
66 episodes, either from natural or anthropogenic origin, common tree species suffer the  
67 greatest total mortality, but rare species are most likely to be locally extirpated  
68 (Cochrane and Schulze 1999; Slik et al. 2002). Wetter forests burn less frequently than  
69 drier forests but are more vulnerable to fire, as trees have thinner protective layers of  
70 bark (Uhl and Kauffman 1990), resulting in higher mortality rates, especially during  
71 drought years.

72 According to Cochrane (2003), fire susceptibility in tropical forests depends mostly on  
73 moisture stress, with forests becoming potentially flammable during periods of  
74 extensive drought. Transitional deciduous and semi-deciduous forests have longer dry  
75 seasons, greater water stress, more open canopies and greater leaf litter that affect fire  
76 characteristics. Closed-canopy evergreen rainforests, in turn, have short, or no seasonal  
77 moisture deficits, with higher humidity levels preventing sustained combustion, even  
78 after months without precipitation (Uhl et al. 1988). However, both natural and  
79 anthropogenic disturbances of forest canopies increase ground irradiance, leading to  
80 drier necromass and greater vulnerable to fire (Cochrane 2003). Independently of the  
81 cause of the fires in these ecosystems, fire frequency is related to climate, with return  
82 intervals of hundreds or thousands of years (Sanford et al. 1985).

83 The study of ancient fires relies on the analysis of charcoal remains. AMS radiocarbon  
84 dating of charcoal can be used to determine the date that carbon was incorporated into  
85 wood, and for young wood, an approximate date of burning (McFadgen 1982). Over  
86 millennia, charcoal fragments retain the age record of each component of the original  
87 tree.

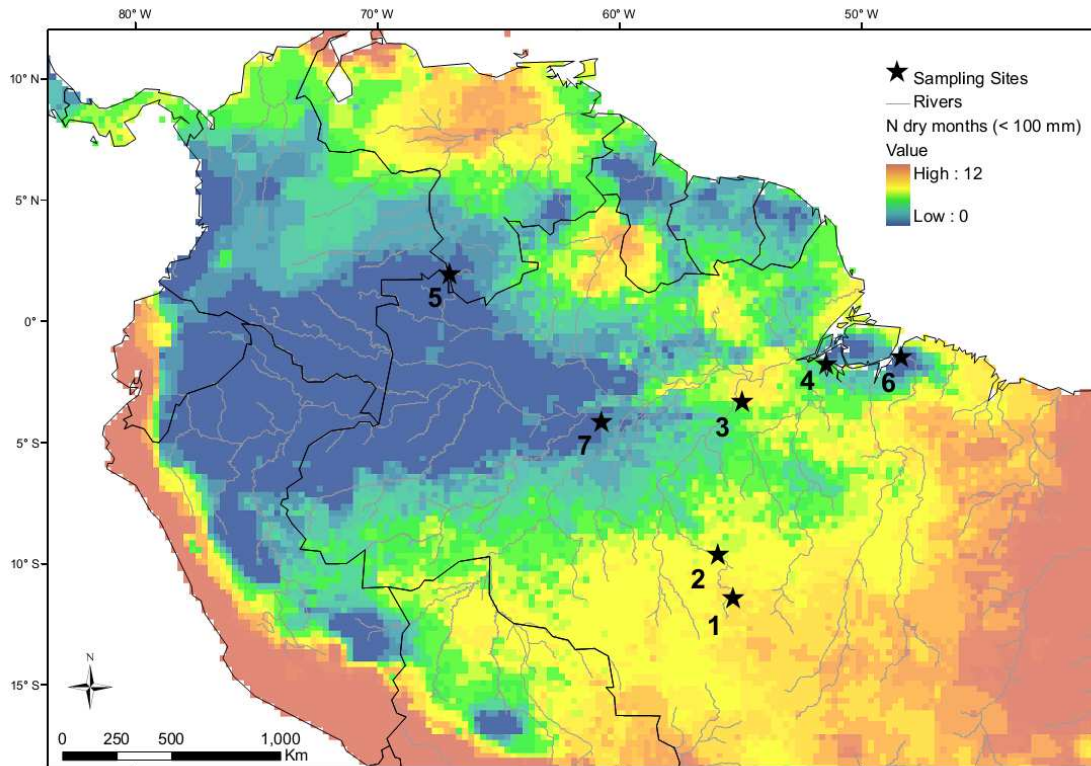
88 Even if the death of the plant is related to the time of burning, growth layers formed  
89 over the life of the tree can add years to age determination (McFadgen  
90 1982). McFadgen (1982) defines the time lapse between the death of a tree and the date  
91 of an event as 'inbuilt age', introducing the concepts of growth and storage ages.  
92 Growth age is due to the heartwood of living trees being composed of old cells that  
93 have lost most of their functions and ceased taking up  $^{14}\text{C}$ . Although important to  
94 provide structural strength, this part of the wood is considered dead and may be  
95 hundreds of years old when the tree dies. The idea of a storage age, in turn, arises from  
96 wood being stored for long periods before decomposing. Thus, inbuilt age is the sum of

97 growth and storage ages. It follows that the effect of the inbuilt age in the dating of the  
98 sample is greater when analyzing long-lived trees (McFadgen 1982). In the case of  
99 natural fires, the inbuilt age of trees may frequently be small due to fire preferentially  
100 killing young, small diameter trees (Cochrane et al. 1999). Therefore, different charcoal  
101 fragments can reflect the ages of different parts of the same tree, different trees in a  
102 single fire, or even different fire episodes.

103 For these reasons, before radiocarbon dating charcoal, a careful structural analysis must  
104 be performed. The present study makes use of cellular wood structures preserved after  
105 carbonization, using anthracological analysis to taxonomically classify charcoal  
106 remains. Such analysis enables the recognition of bark and twigs, which are the younger  
107 parts of the trees and can be therefore more closely associated with time of death.

108 Anthracological analysis involves conducting microscopic observation of preserved  
109 anatomical structures to determine tree taxonomic identity through a comparison  
110 between unknown charcoal fragments against well-identified samples from an  
111 anatomical database. Ideally, after anthracological analysis, each charcoal fragment  
112 should be individually dated so that more precise results can be related to the initial  
113 characteristics of the fragment.

114 In this study, we performed both anthracological analysis and radiocarbon dating in sets  
115 of charcoal samples collected in mature forests with no known history of fire and no  
116 evidence of recent anthropogenic disturbance in several locations within the Amazon  
117 Basin, Our aim was to reconstruct past biodiversity and fire chronology.



118

119 Figure 1: Distribution of sampling sites in the Amazon Basin. These sites were selected  
 120 across a gradient of rainfall seasonality. Number of dry months were calculated by  
 121 maximum cumulative number of months with < 100 mm using data of the Tropical  
 122 Rainfall Measuring Mission (TRMM) satellite product 3B43 V6 at a 0.25° resolution  
 123 (Kummerow et al. 1998). The river network was obtained from the HydroSHEDS  
 124 dataset (Lehner et al. 2008).

125

## 126 **Materials and Methods**

127 The Amazon Basin study sites span a large precipitation seasonality gradient,  
 128 encompassing Southern Venezuela and the Brazilian states of Amazonas, Pará, and  
 129 Mato Grosso (Figure 1). The soils of a total of 7 research areas were sampled. In six  
 130 areas, eight soil pits were excavated to 2 m depth in representative locations for the  
 131 dominant soil and topographic positions. Charcoal found in each soil pit was collected,

132 dried, and stored. At one location, two small pits were excavated to 50 cm in depth at  
133 intervals of 10 cm. Using a Kopecky cylinder (100 cm<sup>3</sup>), horizontal undisturbed soil  
134 samples were collected at each 10 cm depth. The soil was dried and charcoal samples  
135 visible to the naked eye were removed and stored. We define a sample set as a group of  
136 charcoal fragments collected from a single soil pit at a given depth.

137 Anthracological analysis was performed at the National Museum of the Rio de Janeiro  
138 Federal University (UFRJ). To allow wood anatomical investigation, charcoal pieces  
139 were manually broken, exposing transverse, longitudinal-tangential and longitudinal-  
140 radial sections. Each section was examined under a reflected light brightfield/darkfield  
141 microscope and identification was achieved through the comparison to a reference  
142 collection (Charcoal collection from the National Museum, UFRJ – Scheel-Ybert, 2016)  
143 and the use of specialized literature (e.g. Metcalfe and Chalke 1950; Détienne and  
144 Jacquet 1983).

145 Following this analysis, charcoal fragments were prepared and analyzed at the  
146 Radiocarbon Laboratory of the Universidade Federal Fluminense (LAC-UFF). Standard  
147 acid-base-acid (ABA) pretreatment chemistry was employed to decontaminate the  
148 samples before dating. This was undertaken using 1.0M hydrochloric acid (HCl) (2 hr at  
149 90°C) and 1.0M sodium hydroxide (NaOH) (1 hr at 90°C). After pre-treatment, the  
150 samples were combusted in prebaked quartz tubes containing silver powder and cupric  
151 oxide at 900°C for 3 hr in a muffle oven. Carbon dioxide was then purified with dry  
152 ice/ethanol traps in the graphitization line and converted to graphite using the  
153 zinc/titanium hydrate method with an iron catalyst (Xu et al. 2007; Macario et al. 2015).  
154 Individual torch-sealed tubes were heated at 520°C for 7 hr in a muffle oven. Graphite  
155 samples were pressed in aluminum cathodes and measured in a NEC 250kV Single  
156 Stage Accelerator System (SSAMS). The results were corrected for isotopic



157 fractionation by measuring the  $\delta^{13}\text{C}$  online in the accelerator. Background was  
158 measured using processed Alfa Aesar graphite with an average  $^{14}\text{C}/^{13}\text{C}$  ratio of  $6 \times 10^{-13}$ .  
159 Average machine background was  $10^{-13}$  for unprocessed graphite. For quality control  
160 both International Atomic Energy Agency (IAEA) reference materials and an internal  
161 charcoal secondary standard were measured. Calibration was performed with the OxCal  
162 v 4.2.4 (Bronk Ramsey 1995) software using the SHCal13 atmospheric curve (Hogg et  
163 al. 2013) for negative latitudes and the IntCal13 curve for positive latitudes (Reimer et  
164 al. 2013).

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## 166 **Results and discussion**

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168 Most of the radiocarbon results are concentrated between 2700 and 500 yr BP (Table 1  
169 and Figure 2). The high density of  $^{14}\text{C}$  dates during this period is consistent with other  
170 datasets from the Amazonian region and with most dates of archaeological charcoal  
171 (e.g. McMichael et al. 2012; Bush et al. 2008; Nevle and Bird 2008). Only 3 of 27 dates  
172 were <500 cal BP and those youngest dates were from the same soil depth (15 cm) at  
173 one study location, suggesting the dates were from the same fire event. Recent fire,  
174 therefore, was rare and geographically restricted in our dataset from mature forests. This  
175 contrasts with McMichael et al. (2017) based on a geospatial analysis of archaeological  
176 evidence, suggesting that mature forest plots such as those of our study may have been  
177 affected by early European settlers.

178 The calibrated dates show varied and asynchronous fire episodes over greater time  
179 periods spanning the >1000 km sampled area and large precipitation seasonality  
180 gradient (Figure 1). For example, the chronology indicates that fire occurred in the  
181 intervals 2700-2400 cal yr BP (positions 1 and 7), 1900-1500 (positions 3,4, 5 and 6),

182 1400-1000 (positions 1, 4 and 6) and 700-500 cal yr BP (positions 3, 4, 5, 6 and 7)  
183 (Figure 3). Coeval episodes in different places may be a sign of a climatic event,  
184 possibly a drier period. The oldest dated ages were in eastern Amazonia (Belterra, Pará)  
185 and the most recent in central Amazonia (Careiro, Amazonas) (Table 1).

186 The fire history of Amazonian forests may be linked with global climate change  
187 records. One important factor related to global climatic changes which should be  
188 considered is the production of cosmogenic nuclides such as  $^{14}\text{C}$  and  $^{10}\text{Be}$ . Since the  
189 formation of these nuclides depends on the arrival of galactic cosmic rays in the  
190 atmosphere, the effect of solar activity deflecting such particles would decrease their  
191 production rate. Therefore, this anti-correlation with  $^{14}\text{C}$  and  $^{10}\text{Be}$  production is used as  
192 a proxy of solar activity, and potentially also an indicator of drought episodes (Stuiver  
193 and Braziunas 1993; Mayeski et al. 2004). According to Stuiver and Braziunas (1993),  
194 variation in  $\Delta^{14}\text{C}$  over time (Fig 3) is tied to either ocean circulation change or solar  
195 modulation of atmospheric  $^{14}\text{C}$  production with the possibility of a simultaneous  
196 contribution by both factors, where presumably solar forcing results in climatic change  
197 which, in turn, causes ocean circulation change. In addition, other factors can be related  
198 to climatic changes such as the presence of greenhouse gases  $\text{CO}_2$  and  $\text{CH}_4$  or increases  
199 in volcanic aerosols. All these factors will contribute to the complex pattern of  
200 thermohaline circulation as its intensity determines poleward heat transport, and  
201 influences global climate (Stuiver and Braziunas, 1993). Climatic changes during the  
202 Holocene were investigated by Mayeski et al. (2004), who identified Rapid Climatic  
203 Changes (RCC) in different parts of the world. The first RCC was recorded between  
204 9000 and 8000 cal yr BP and was considered the last major deglaciating stage affecting  
205 the Northern Hemisphere. It was attributed mainly to reduced oceanic ventilation, since  
206 there was no clear change in cosmogenic isotopes production. Volcanic  $\text{SO}_4$  production

207 was unusually high in the Northern Hemisphere, which could be evidence of intense  
208 volcanic activity, while biogenic CH<sub>4</sub> declined - probably in response to aridity in the  
209 low- to mid-latitudes (Blunier et al. 1995; Mayeski et al. 2004). Fire episodes recorded  
210 in this study may be associated with past climatic changes. Our oldest dated charcoal  
211 (7759-7585 cal yr BP) from Belterra, for instance, corroborates the idea of tropical  
212 aridity at that time. Although severe droughts were documented in Amazonia between  
213 8000 and 4000 cal. yr BP (Mayle and Power 2008), only one charcoal fragment was  
214 dated within this period.

215 In the period from 3500 to 2500 cal yr BP, pronounced aridity was recorded in East  
216 Africa, the Amazon basin, Ecuador and the Caribbean/Bermuda region (Haug et al.  
217 2001). Mayeski et al. (2004) have suggested that solar variability is responsible for such  
218 an event since it coincides with maxima in  $\Delta^{14}\text{C}$  and  $^{10}\text{Be}$ . The period from 1200 to  
219 1000 cal yr BP includes generally dry conditions in Tropical Africa, monsoonal  
220 Pakistan and Ecuador, when there is a slight increase in atmospheric CO<sub>2</sub>, while drought  
221 was linked to solar output in Yucatan (Hodell et al. 1991, 2001). The more recent  
222 episode, starting at 600 cal yr BP, would be related to the fastest and strongest climatic  
223 changes in the Holocene. At this time, there was a drop in CO<sub>2</sub> and a rise in CH<sub>4</sub>,  
224 suggestive of wet conditions. However, this period features a more variable response in  
225 humidity at low latitudes.

226 Fire episodes that happened from 1900 to 1600 cal yr BP do not coincide with those  
227 reported by Bush et al. (2006) or with RRC events described by Mayeski et al. (2004),  
228 and may not be significant in a global scale. The occurrence of fires in positions 3, 4  
229 and 6 may be related to human occupation, since these locations follow the course of the  
230 Amazon River that was densely occupied in pre-Columbian times. From 2500 years BP,  
231 these societies started to settle along the major river bluffs with a significant population

232 growth and the development of a sedentary lifestyle (Neves et al. 2003; Heckenberger  
233 and Neves 2009; Denevan 1996). A peak in pre-Columbian population occurred from  
234 1300 to 500 BP in different regions of Amazonia, associated with the development of  
235 sedentary and complex societies (Heckenberger and Neves 2009; Arroyo-Kalin 2011;  
236 Moraes and Neves 2012; Stenborg 2016). By the shore of the Solimões river, in Central  
237 Amazonia, three main periods of human occupation have been observed (1400-1300 yr  
238 BP, 1200-930 yr BP and 600-350 yr BP), with 1400-1300 yr BP being the less intense  
239 period and 1400-1300 BP the most (Machado 2005). Our results include dates from  
240 these three periods, leading to the possibility of anthropogenic influence in the  
241 occurrence of such fires because of fire management, disturbance, and gap formation  
242 due to human activity.

243 A total of 13 families were identified from the anthracological analysis (Table 1).  
244 Fabaceae, Sapotaceae and Combretaceae families were the most frequent among the  
245 identified fragments. Families identified in four of the seven locations are also  
246 abundant in the present-day. For instance, Fabaceae, which we identified at four of the  
247 seven regions, was also the most abundant and most diverse family of the largest dataset  
248 of Amazonian forest plots representing present-day contemporary forest composition  
249 (Steege et al. 2013). The Goupiaceae family is abundant and has species considered  
250 hyperdominant in the Amazonian tree flora, but has very low diversity (Steege et al.  
251 2013). However, we only identified one charred species of Goupiaceae.

252 Some of the charcoal fragments could not be identified; some were unidentifiable  
253 because they were too damaged (53D and E, 66B, 77C, 79A, B and C), and others  
254 remain indeterminate because they were too small and contained few visible diagnostic  
255 features (52A, 53C, 87B). Most of the reference material is from the Atlantic Forest and  
256 the studied material comes from the Amazon forest. A comparison between these two

257 forested ecosystems reveals that for the same families, ecosystem-specific  
258 characteristics can be distinguished, e.g., vessel diameter was larger in Amazon samples  
259 of the same family due to greater water availability (Alves and Angyalossy 2000). In  
260 tropical forests with high species diversity, identification is often difficult as the  
261 anatomy of many species is still unknown (Wheeler and Baas 1998). Expansion of  
262 charred reference material is therefore a priority to aid identification.

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279 Table 1. Sample identification, present abundant families, and calibrated radiocarbon  
280 ages. Families in bold were both identified through anthracological analysis and are  
281 abundant in present-day composition for the region.

Location (Point)	Abundant families at present day	Source	Identified families	Sample	Lab Id	Depth (cm)	<sup>14</sup> C age (yr BP)	Calibrated age (cal BP) 95.4%	
Claudia - MT (1)	Melastomataceae	(KUNZ et al., 2010; IVANAUSKAS, 2002)	Calophyllaceae	14P09A58	LACUFF150361	110	1120±33	1058	928
Novo Mundo - MT (2)	Burseraceae, Moraceae and Fabaceae	(SASAKI et al., 2008)	Fabaceae	14P09A65	LACUFF150319	30	2480±30	2703	2355
			Fabaceae Caesalpinoideae	14P09A66A	LACUFF150330	30	2728±30	2854	2749
			unidentifiable	14P09A66B	LACUFF150321		2685±39	2851	2721
Belterra - PA (3)	Fabaceae, Lecythidaceae and Moraceae	(GUALBERTO et al., 2014; ALMEIDA et al., 2012; ANDRADE, 2011)	Sapotaceae	14P09A51	LACUFF150360	96	637±27	647	541
			indeterminate	14P09A52A	LACUFF150308	30	6876±41	7759	7585
			Combretaceae	14P09A52B	LACUFF150309		1950±31	1927	1747
Melgaço - PA (4)	Fabaceae, Flacourtiaceae, Arecaceae, Goupiaceae and Lecythidaceae.	(ICMBio, 2012; FERREIRA et al., 2015).	Chrysobalanaceae	14P09A53A	LACUFF150310	25	1686±28	1609	1430
			Anacardiaceae	14P09A53B	LACUFF150311		1236±30	1185	985
			indeterminate	14P09A53C	LACUFF150312		1611±31	1534	1377
			unidentifiable	14P09A53D	LACUFF150313		1690±31	1692	1430
			unidentifiable	14P09A53E	LACUFF150314		1748±31	1702	1544
			Goupiaceae	14P09A55	LACUFF150315	15	1134±30	1058	934
			Combretaceae	14P09A56	LACUFF150316	40	1620±32	1539	1378
			Lauraceae	14P09A87A	LACUFF150327	25	783±30	730	577
			indeterminate	14P09A87B	LACUFF150328		1347±29	1294	1180
Rio Negro - AM/VE (5)	Lecythidaceae	(OLIVEIRA et al., 2001)	Melastomataceae	14P09A67	LACUFF150369	54	776±32	729	572
			Combretaceae	14P09A68	LACUFF150364	37	641±29	649	542
			Anacardiaceae	14P09A71	LACUFF150365	78	1975±31	1995	1755
Marituba - PA (6)	Fabaceae and Lecythidaceae	(ALMEIDA et al., 2011; AMARAL et al., 2009; MELO, 2004)	Fabaceae <i>Lonchocarpus</i>	14P09A60	LACUFF150362	-	1604±31	1530	1375
			Myrtaceae	14P09A61	LACUFF150317	-	662±34	655	550
Careiro - AM/BR (7)	Lecythidaceae, Fabaceae and Sapotaceae	(SILVA et al., 2008; OLIVEIRA et al., 2008; OLIVEIRA e AMARAL, 2004)	Melastomataceae	14P09A73	LACUFF150367	5	523±34	548	495
			Rubiaceae	14P09A75A	LACUFF150369	15	758±37	725	566
			Lauraceae	14P09A77B	LACUFF150372	15	480±29	533	456
			unidentifiable	14P09A77C	LACUFF150373		411±29	500	325
			Fabaceae	14P09A78	LACUFF150374	15	365±29	472	311
			unidentifiable	14P09A79	LACUFF150375	15	2431±33	2696	2337

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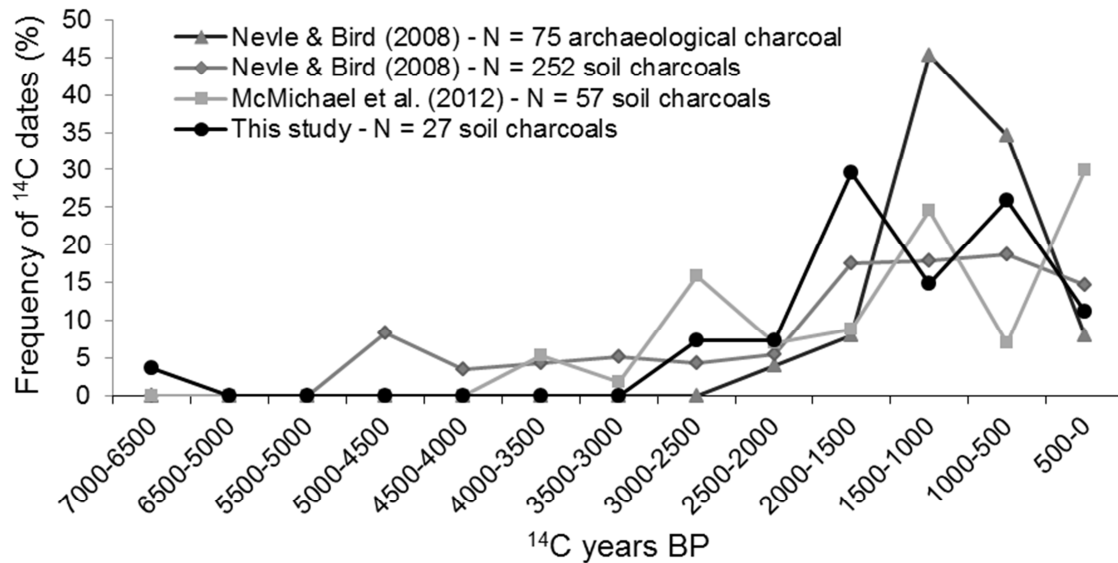
284 Uncertainty in the interpretation of charcoal radiocarbon ages can be introduced, as

285 previously discussed, due to the non-systemic offsets caused by inbuilt age. Long-lived

286 species can be categorized into two successional groups. Late secondary succession  
287 species may have lifespans between 40 and 100 years (Maciel et al. 2003); long-lived  
288 species show slow or very slow growth with long lifespans of more than 100 years  
289 (Maciel et al. 2003). Amongst the families identified in this paper, Fabaceae,  
290 Sapotaceae and Combretaceae include several long lifespan species, although they also  
291 include some short-lived pioneer and early secondary species, while Goupiaceae is  
292 frequently a pioneer.

293 Out of the 16 sets of charcoal fragments identified, five contained more than one  
294 taxonomic type. For the radiocarbon dating of sample sets with more than one family,  
295 some of the dates are statistically similar, such as sample sets 66 and 77. In both cases,  
296 one fragment could not be identified but could be distinguished from the others,  
297 suggesting the fragments belonged to two different families that were contemporaneous.  
298 Other sample sets had differences in age of hundreds of years. In sample set 52 the age  
299 difference is very large and again the oldest fragment remains indeterminate.

300 Differences in dates at a given site might be attributed to inbuilt age. In sample set 87,  
301 only the youngest fragment could be identified, making it difficult to evaluate such a  
302 possibility. Another reason for differences in ages could be if the different fragments  
303 represent different fire episodes. In this case, the different fragments could be disturbed  
304 by the action of bioturbation.

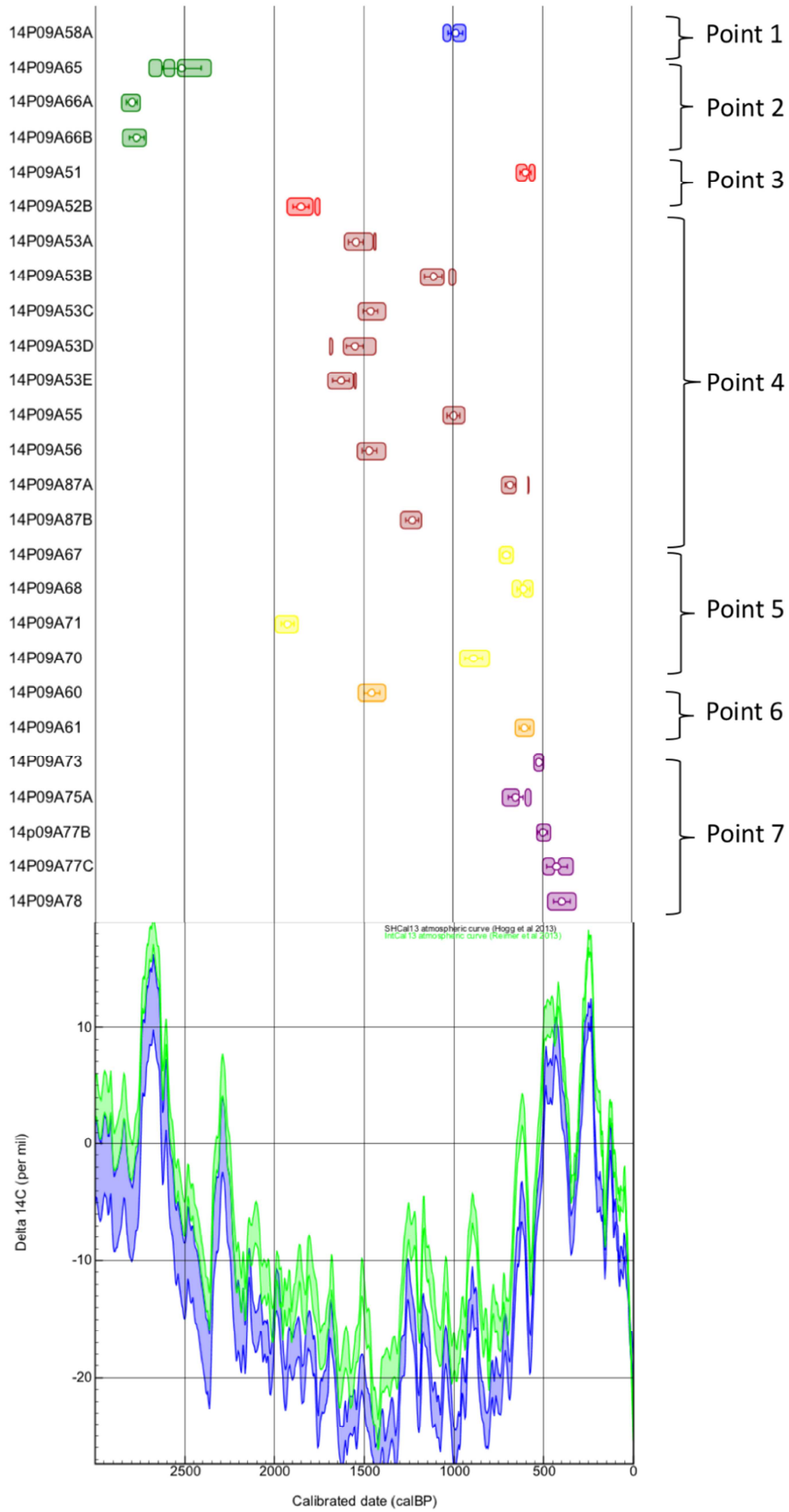


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306 Figure 2. Frequency of charcoal samples in percentage per period of time before present  
 307 (BP). Figure adapted from Neve and Bird (2008). We added dates of soil charcoal  
 308 presented in McMichael et al. (2012) and dates from this study. Charcoal dates are  
 309 shown in age classes of 500 <sup>14</sup>C years. All dates presented in the figure are from  
 310 charcoal collected in soils.

311 Another interesting result is the case of sample set 53, with five different families  
 312 identified. Four of the individual samples (53A, C, D and E) yielded statistically similar  
 313 results, while sample 53B was dated 400 years younger. Moreover, this sample was also  
 314 the only in the group to present convergent rays, which are a sign of young ramification  
 315 (Marguerie and Hunot 2007). Therefore, either the fire occurred about 1000 yr BP and  
 316 most of the trees were already centenary, or the set of samples represents different fire  
 317 events.





319 Figure 3. Calibrated ages of samples in the range from 3 to 0 kyr cal BP. and  
320 atmospheric  $\Delta^{14}\text{C}$  values from curves SHCal13 (Hogg et al. 2013) and IntCal13 (Reimer  
321 et al. 2013).

322

### 323 **Conclusions**

324 Our analysis of the calibrated ages of the charcoal dates shows that the chronology  
325 coincides with both climatic events that occurred in the Holocene and with the inferred  
326 timing of human occupation in the Amazon region. Most of the dates are consistent with  
327 other datasets and with reported climatic events around the world. The higher density of  
328 results in our study corresponds to periods of more intense human occupation, with little  
329 evidence of recent fire in mature forests.

330 A total of 13 different families were identified, suggesting that a wide diversity of  
331 species was burned in the past. The presence of potentially long-lived species amongst  
332 charcoal samples indicates that chronology should be analysed with care. The variety of  
333 species within a single set of charcoal fragments suggests a need for multiple  
334 radiocarbon dates to be undertaken. Comparison of dating results from charcoal  
335 fragments belonging to the same sample set revealed large variation in dates which may  
336 reflect different fire episodes, inbuilt age, or the action of bioturbation.

337

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353

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