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1     **Benthic Foraminiferal Salinity index in marginal-marine environments: a case study**  
2                                   **from the Holocene Guadalquivir estuary, SW Spain**

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4

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14

15    **Abstract**

16    Here we developed and validated a new Benthic Foraminiferal Salinity (BFS) index from  
17    marginal-marine environments by analysing benthic foraminifera from the Holocene  
18    Guadalquivir estuary sediments (SW Spain). This index is formulated utilising only four  
19    species: *Ammonia tepida* and *Haynesina germanica* with higher tolerance to brackish waters  
20    and indicating lower salinity, and *Elphidium translucens* and *Elphidium granosum* indicative  
21    of greater marine influence and pointing to higher salinity. Thus, the BFS index is calculated  
22    easily and rapidly, and therefore it makes it possible to analyse a higher number of samples in  
23    less time. The BFS index values from the studied cores enabled the detailed description of  
24    subtle changes in the Guadalquivir estuary restriction during the Holocene. For this purpose,  
25    three degrees of salinity, depending on marine influence, were defined: higher (BFS index =

26 0.0-0.4, high marine influence), moderate (BFS index = 0.4-0.7, moderate marine influence),  
27 and lower (BFS index = 0.7-1.0, low marine influence). Before 2000 BCE, the estuary was  
28 moderately open and well-connected to the Atlantic Ocean. From 2000 BCE, the estuary  
29 experienced a greater marine influence, increasing in extension, as a consequence of a sea-  
30 level rise and subsidence. Immediately afterwards, it began to experience restriction  
31 processes due to southward shoreline progradation related to the growth of littoral spits and  
32 sediment supply. From 1400 to 1000 BCE, gradual restriction transformed the open estuary  
33 into a semiclosed estuary. A last phase of estuary restriction occurred from 1000 BCE to the  
34 present day, leading to the lowest salinity and the highest estuary restriction. Finally, the BFS  
35 index was successfully applied in two other marginal-marine environments: a Pleistocene  
36 lagoon in northern Italy, and a Pliocene coastal bay in southeastern Spain. The index allowed  
37 assessment of the degree of restriction in these different environments, supporting its utility  
38 in different regions, environments and timescales.

39

40 **Keywords:** Micropaleontology; Coastal settings; Restriction; Brackish; Quaternary; Europe

41

## 42 **1. Introduction**

43 Marginal-marine environments, including estuaries, deltas, fjords, marshes and littoral  
44 lagoons, are very sensitive to changes in sea level, tide and wave regime, as well as fluvial  
45 dynamics (Chiverrell, 2001; Ybert et al., 2003). These factors play a key role in controlling  
46 coastal geomorphological features (spits, dunes, cheniers, marshes, levees, tidal channels),  
47 and sedimentary features (sediment supply, sedimentary bodies, sediment infilling), which  
48 modify the paleogeography of marginal-marine environments (Gerdes et al., 2003; Durand et  
49 al., 2016). Coastal environments occur at the transition between continental fresh waters and

50 marine normal salinity waters, consequently they are commonly characterised by brackish  
51 waters (Rich and Maier, 2015). In coastal environments, the gradual decrease of seawater  
52 influence reduces salinity (Guelorget and Perthuisot, 1983, 1992; Debenay, 1995). Thus,  
53 marginal-marine environments have lower or higher salinity depending on the lower or  
54 higher influence of marine waters, respectively. Salinity is governed by the geomorphological  
55 restriction of the coastal environment (i.e., degree of connection to the open sea), which is  
56 related to changes in both geomorphological and sedimentary features (Gregoire et al., 2017;  
57 Devillers et al., 2019; Haas et al., 2019). Therefore, lower or higher restriction causes higher  
58 or lower salinity, respectively.

59 Benthic foraminifera are widely used for reconstructing paleoenvironmental changes  
60 from marginal-marine to deep-sea environments since they are very sensitive to variations in  
61 sea level, organic fluxes, oxygen content, salinity and type of substrate (e.g., Alve and  
62 Murray, 1999; Jorissen et al., 2007; Di Bella et al., 2008; Martins et al., 2014; Blázquez et al.,  
63 2017; Pérez-Asensio et al., 2014, 2017, 2020). In coastal settings, it is well known that  
64 benthic foraminiferal distribution and abundance can be influenced by salinity, which is  
65 dependent, among other factors, on the geomorphological restriction (Debenay et al., 2006;  
66 Barbieri and Vaiani, 2018; Blázquez-Morilla et al., 2019). In fact, benthic foraminifera have  
67 been successfully used as proxies of restriction of marginal-marine environments and for  
68 developing salinity indices (Debenay, 1995; Hayward et al., 2004; Debenay and Luan, 2006).  
69 Previously defined salinity indices (Debenay, 1995; Hayward et al., 2004) are based on  
70 complex and time-consuming identification of numerous benthic foraminiferal species, which  
71 requires a profound taxonomic knowledge. Hence, it is necessary to create simpler salinity  
72 indices based on a low number of species, which would be useful for scientists who are not  
73 specialized on benthic foraminiferal taxonomy. Furthermore, identifying fewer species would

74 imply a faster process for obtaining foraminiferal data, making it possible to analyse more  
75 sedimentary records at higher resolution.

76         The principal objectives of this study were to develop a simple Benthic Foraminiferal  
77 Salinity (BFS) index, and to validate it as proxy for different degrees of salinity in marginal-  
78 marine environments. For these purposes, we analysed benthic foraminifera from 8 short  
79 cores (this study) and two deep cores (Rodríguez-Ramírez et al., 2015), which recovered  
80 Holocene sediments from the Guadalquivir estuary (SW Spain) (Figs 1 and 2). The  
81 Guadalquivir estuary is an excellent area to assess the performance of our BFS index because  
82 its paleogeography, and therefore its connection to the open sea (i.e., marine influence),  
83 changed substantially over the Holocene owing to geomorphological and sedimentary  
84 processes (Rodríguez-Ramírez et al., 2019). In addition, the applicability of this BFS index  
85 was tested in two different environments (lagoon, coastal bay) from other regions (northern  
86 Italy, southeastern Spain).

87

## 88 **2. Study area: the Guadalquivir estuary**

89         The Guadalquivir estuary (SW Spain) is located in the Gulf of Cadiz (Atlantic Ocean)  
90 (Fig. 1). It contains a 180,000 ha freshwater marsh including the Doñana National Park  
91 (UNESCOMAB Biosphere Reserve). This estuary has two enclosing spits (Doñana and La  
92 Algaida), which are partly covered by active dunes. Both spits protect a wide marsh area  
93 behind. This marsh grew without sediment input from the Guadalquivir and convergent rivers  
94 because this riverine sediment supply filled in the ancient marine Guadalquivir estuary as  
95 finger deltas in a low-energy setting. The growth of the two large littoral spits isolating the  
96 estuary and the development of a wide chenier plain favoured this sedimentary process  
97 (Rodríguez-Ramírez and Yáñez-Camacho, 2008).

98 Fluvial regime, tidal inflow, wave action, and drift currents controlled the  
99 Guadalquivir estuary hydrodynamics. The Guadalquivir river is the largest river draining SW  
100 Spain and the principal source for fluvial sediments in the southwestern Spanish coast. This  
101 river has a mean annual discharge of 164 m<sup>3</sup>/s, with winter spates that can easily exceed 5000  
102 m<sup>3</sup>/s (Vanney, 1970). At the river mouth in the period from 1997 to 2003, the average tidal  
103 range was 2 m with a maximal tidal range of 3.86 m (Spanish Ministry of Fomento, 2005).  
104 Accordingly, the coastline is mesotidal, semidiurnal.

105 The wave regime is directly related to the prevailing SW winds, with 22.5% of the  
106 days of the year with SW winds (Rodríguez-Ramírez et al., 2003). Overall, the wave regime  
107 has medium-to-low energy, with waves normally <0.6 m high (data of Departamento de  
108 Clima Marítimo). Atlantic cyclones are frequent during winter and they foster strong SW  
109 winds, generating significant erosion in the coastline (Rodríguez-Ramírez et al., 2003). Most  
110 of the wave fronts approach obliquely the coastline generating longshore currents that  
111 transport sand from the Portuguese coast to Spanish nearshore areas.

112

### 113 **3. Methodology**

114

#### 115 *3.1. Lithostratigraphy*

116 We studied the sedimentary sequences and facies from 8 short cores (<3 m): S6, LB,  
117 AR, AA, VT, VAc, VAli.2 and VAli.1 (Table 1). The cores were drilled with a 4-cm-  
118 diameter Eijkelkamp gouge, an 8-cm-diameter drill, and trenches. We also analysed  
119 previously published sedimentary records from two long cores: S7 and S11 (Table 1)  
120 (Rodríguez-Ramírez et al., 2015). Grain-size analyses were performed using conventional  
121 sieving for fractions >2 mm, and a Malvern Mastersizes 2000 laser diffraction particle

122 analyser for smaller particle sizes between 2 mm and 2  $\mu\text{m}$ . Shepard's sediment classification  
123 (Shepard, 1954) was applied to grain-size results in order to describe sediment texture,  
124 including sand, silt and clay fractions.

125

### 126 *3.2. Radiocarbon dating*

127 Eight new radiocarbon dates were measured on mollusc shells at the laboratories of  
128 Centro Nacional de Aceleradores (Seville, Spain) and Accium BioSciences Accelerator Mass  
129 Spectrometry Lab (Seattle, USA) (Table 2, Fig. 2). Other 14 radiocarbon dates from previous  
130 studies (Rodríguez-Ramírez et al., 2014, 2015, 2016) were used in this work (Table 2, Fig. 2).  
131 Calibration of radiocarbon data was conducted by means of CALIB 7.10 software (Stuiver  
132 and Reimer, 1993) and the calibration dataset of Reimer et al. (2013). Uncertainties of the  
133 calibrated ages are expressed as  $2\sigma$  errors. The reservoir effect was corrected using the  $\Delta R$   
134 values recommended by Soares (2015). This author suggested a  $\Delta R$  value of  $-108 \pm 31$   $^{14}\text{C}$   
135 yr for the late Holocene on the Andalusian coast of the Gulf of Cadiz; excluding the years  
136 4400–4000  $^{14}\text{C}$  yr BP, for which he recommended a  $\Delta R$  value of  $+100 \pm 100$   $^{14}\text{C}$  yr. In the  
137 interval from 4000 to 2000  $^{14}\text{C}$  yr BP, lack of data impedes pinpointing the most recent time  
138 boundary to which the  $+100 \pm 100$   $^{14}\text{C}$  yr  $\Delta R$  value can be extended. In spite of this problem,  
139 we decided to tentatively extend it to the middle year 3000  $^{14}\text{C}$  yr BP.

140

### 141 *3.3. Micropaleontology*

142 A total of 45 sediment samples (~50 g) was wet-sieved over a 63  $\mu\text{m}$  mesh and dried  
143 in an oven at 40°C. Samples were thoroughly split using a microsplitter so as to obtain sub-  
144 samples. Then, these sub-samples were dry-sieved over a 125  $\mu\text{m}$  mesh, and at least 300  
145 benthic foraminifera were carefully counted and identified to the species level (Loeblich and

146 Tappan, 1987; Milker and Schmiedl, 2012; Pérez-Asensio et al., 2012). Raw counts were  
147 transformed into relative abundances (%). Q-mode principal component analysis (PCA) was  
148 carried out on all samples from the short and long cores using the software OriginPro 2020.  
149 All species were included in the analysis. The PCA allowed to establish benthic foraminiferal  
150 assemblages and to infer environmental factors controlling these assemblages (Parker and  
151 Arnold, 1999; Milker et al., 2009) (Table 3, Fig. 3).

152 In this study, we developed a new Benthic Foraminiferal Salinity (BFS) index for  
153 assessing the salinity changes in marginal-marine environments (e.g., estuaries, lagoons,  
154 marshes). This BFS index is based on the distribution of some of the dominant benthic  
155 foraminiferal species found in the Holocene Guadalquivir estuary sediments (see discussion  
156 section 5.2).

157

## 158 **4. Results**

159

### 160 *4.1. Lithostratigraphy and chronology of the deep cores: S7 and S11*

161 The sedimentary sequence and chronology of deep cores S7 and S11 are described in  
162 detail in prior studies (Jiménez-Moreno et al., 2015; Rodríguez-Ramírez et al., 2015).

163 The core S11 is 18 m long and its lower part (18 to 12.5 m) consists of a Pleistocene  
164 marsh sequence of mostly greyish ochre clayey silts (Fig. 2). From 12.5 m to the top, the  
165 Holocene sequence is dominated by grey-greenish clayey silts, with three intercalated sandy  
166 layers at around 12, 9, and 7 m (Fig. 2). These sandy layers have been interpreted as Extreme  
167 Wave Events (EWEs) (Rodríguez-Ramírez et al., 2015) occurring at ca. 2270, 1657, and  
168 1421 BCE, respectively (Fig. 2).



169 The sedimentary sequence in core S7 is 9 m long, beginning with aeolian sands (9-  
170 8.75 m) from the El Abalarío Dune Formation (Fig. 2). Holocene greyish ochre clayey silts  
171 dominate from 8.75 to the core top. Three sandy layers related to EWEs appear at around 8.5,  
172 5.5, and 2.5 m, being dated at ca. 1983, 1657, and 1142 BCE, respectively (Fig. 2).

173

#### 174 *4.2. Lithostratigraphy and chronology of the short cores: S6, LB, AR, AA, VT, VAc, VAl.2* 175 *and VAl.1*

176 The core S6 recovered 1.5 m of sediments, with clayey silts at the lower part and  
177 sands at the upper part (Fig. 2). Sandy facies were dated at 2439 BCE. The core LB is 1.25 m  
178 long and it mostly consists of clayey silts. At around 0.9 m, a shelly layer appears with an age  
179 of 52 ACE. The sediment record of core AR includes 1.25 m of clayey silts with a shelly  
180 layer at around 0.5 m (232 BCE). The core AA has a length of 1.25 m and it is composed of  
181 clayey silts with an intercalated shelly layer at around 0.4 m (304 BCE). In the 2-m long core  
182 VT, clayey silts range from 2 to 1.25 m, and a thick shelly layer, dated ca. 1304-1151 BCE,  
183 occurs towards the top. The core VAc recovered 2 m of sands dated at ca. 1659-1408 BCE.  
184 The VAl.2 core (1.75 m long) shows clayey silts in the lower part (1.75-1 m) and a shelly  
185 layer (ca. 2205-2022 BCE) in the upper part (1-0 m). The VAl.1 core encompasses 1.4 m of  
186 sands dated ca. 2230-2190 BCE.

187

#### 188 *4.3. Benthic foraminiferal data*

189 The Q-mode principal component analyses (PCA) considering all samples from the  
190 studied cores yielded three assemblages (PC-1 to 3), explaining 67.8%, 23.3% and 4.2% of  
191 the total variance, respectively (Table 3). The benthic foraminiferal assemblage from the PC-  
192 1 is dominated by *Ammonia tepida* with *Haynesina germanica*, *Elphidium translucens* and

193 *Elphidium granosum* as secondary species. In the assemblage from the PC-2, *H. germanica* is  
194 the unique and dominant species. The assemblage from the PC-3 has *Triloculina trigonula* as  
195 dominant species, and includes several additional taxa such as *Ammonia beccarii*,  
196 *Quinqueloculina seminula*, *Miliolinella* sp., *Quinqueloculina* sp., *Triloculina* sp.,  
197 *Quinqueloculina vulgaris*, and *Quinqueloculina laevigata*. All taxa from this assemblage  
198 (PC-3) are marine species transported into the estuary by EWEs (Rodríguez-Ramírez et al.,  
199 2015). Consequently, they will not be considered to assess the salinity conditions of the  
200 Guadalquivir estuary.

201         The relative abundances of the dominant and secondary taxa from the PC-1 and PC-2  
202 assemblages are shown in Fig. 2. In the S11 core, *H. germanica* shows fairly low values  
203 (<20%) from the base to 4.5 m (Fig. 2). Then, it increases until reaching maximal values  
204 (80%) from 3 to 2 m, and it decreases upwards. *A. tepida* relative abundance rises from the  
205 bottom to 5 m, it diminishes from 5 to 2 m, and it rises towards the core top. *E. translucens*  
206 has higher values in the lower part of the core (12.5-5 m), and shows its highest abundance  
207 (~25%) at 6.5 m (Fig. 2). In the S7 core, *H. germanica* has very low abundance (<10%) from  
208 the core base to 2.5 m, and gradually increasing abundances from 2.5 m towards the core top.  
209 *A. tepida* shows considerably high values (40-60%) from the core base to 3.5 m, and it  
210 declines upwards. Both *E. translucens* and *E. granosum* have higher abundance (10-35%)  
211 from the core bottom to 3.5 m than in the upper part of the core (3.5-1 m), where they  
212 virtually disappear. In core S6, both *A. tepida* and *H. germanica* are very abundant (~50%) in  
213 interval from 1.25 to 0.75 m, whereas *E. translucens* has very low abundance (<6%) in the  
214 same interval. The core LB shows opposite trends for *H. germanica* and *A. tepida*, with  
215 higher values of *H. germanica* (~65%) and lower values of *A. tepida* (~25%) from 1.1 to 0.4  
216 m. In the interval from 0.4 to 0.2 m, *A. tepida* increases while *H. germanica* decreases. *E.*  
217 *translucens* and *E. granosum* have very low abundances (<10%) along the core. In the AR

218 and AA cores, *H. germanica* is very abundant (80-90%), whereas *A. tepida*, *E. translucens*  
219 and *E. granosum* show low abundances (<15%) in both cores. The cores VT and VAc have  
220 high values of *A. tepida* (50-65%), and low values of *H. germanica* (10-20%). *E. translucens*  
221 and *E. granosum* have low values (<15%), decreasing upwards. In the core VAli.2, *A. tepida*  
222 shows higher abundances (40-60%), whereas *H. germanica* shows lower abundances (15-  
223 30%). Both species abundances drop towards the core top. *E. translucens* and *E. granosum*  
224 have very low abundances (<10%). The core VAli.1 has high abundance of *A. tepida* (30-  
225 50%), which diminishes upwards. In this core, *H. germanica* and *E. translucens* have very  
226 low abundances (<10%).

227

## 228 **5. Discussion**

229

### 230 *5.1. Ecology of benthic foraminiferal species from marginal-marine environments*

231 The two most significant assemblages (PC-1 and PC-2) include *Ammonia tepida*,  
232 *Haynesina germanica*, *Elphidium translucens* and *Elphidium granosum* (Table 3). *A. tepida*  
233 is a euryhaline species tolerating brackish waters (Murray, 2006; Blázquez and Usera, 2010;  
234 Pérez-Asensio and Aguirre, 2010). This species can thrive in environments with low oxygen  
235 content and/or high organic matter fluxes (Martins et al., 2013; Wukovits et al., 2018).  
236 According to Jorissen et al. (2018), *A. tepida* is a second-order opportunist, which highly  
237 increases when organic matter supply is maximal. The species *H. germanica* is also a  
238 euryhaline foraminifera resisting a wide range of salinities (Murray, 2006; Blázquez and  
239 Usera, 2010). This species can inhabit low oxygenated settings, and it is abundant in  
240 sediments with highly variable organic matter content (Alve and Murray, 1999; Martins et al.,  
241 2013). *H. germanica* is a third-order opportunist that increases its abundance due to higher

242 organic matter supply, but it is absent when organic matter input is maximal (Jorissen et al.,  
243 2018). *E. translucens* is common in euryhaline marginal-marine environments (Murray,  
244 2006; Pérez-Asensio and Aguirre, 2010). As other *Elphidium* species, it is less tolerant to low  
245 oxygen conditions than species from the *Ammonia* group (Sen Gupta and Platon, 2006). This  
246 species is an indifferent species, which do not increase with organic matter fluxes, and  
247 disappears if organic matter input is high (Jorissen et al., 2018). *E. granosum* is abundant in  
248 marginal-marine environments, and tolerates wide salinity changes (euryhaline) (Vaiani,  
249 2000; Curzi et al., 2006; Pérez-Asensio and Aguirre, 2010). This species can benefit from  
250 high organic matter fluxes as long as oxygen content is not very low (Jorissen, 1988). It is  
251 considered a third-order opportunist, rising its abundance as a response to higher organic  
252 matter supply, yet disappearing with maximum organic matter enrichment (Jorissen et al.,  
253 2018).

254

## 255 5.2. Development of the Benthic Foraminiferal Salinity index

256 Benthic foraminiferal assemblages from the Holocene sediments recovered in the  
257 Guadalquivir estuary area allowed to developed a new Benthic Foraminiferal Salinity (BFS)  
258 index. As mentioned above, the most important species (*A. tepida*, *H. germanica*, *E.*  
259 *translucens*, *E. granosum*) are euryhaline species (i.e., tolerating wide salinity variations).  
260 These species can inhabit marginal-marine environments with brackish waters such as  
261 estuaries, marshes and littoral lagoons (Debenay, 1995; Ruiz et al., 2005; Murray, 2006;  
262 Blázquez and Usera, 2010; Pérez-Asensio and Aguirre, 2010). Marginal-marine  
263 environments can have different degrees of salinity related to higher or lower influence of  
264 marine waters. This depends on their paleogeography, geomorphology, and wave and tide  
265 regimes (Pethick, 1984; Pérez-Ruzafa et al., 2019). The comparison of the Principal

266 Component (PC) scores of species belonging to the euryhaline assemblage PC-1 shows that  
267 *A. tepida* and *H. germanica* present higher scores than *E. translucens* and *E. granosum* (Fig.  
268 3, Table 3). This suggests that higher scores of PC-1 indicate less salinity since *A. tepida* and  
269 *H. germanica* have high tolerance to brackish conditions (i.e., less marine influence), and can  
270 inhabit inner estuaries (Alve and Murray, 1994; Debenay and Guillou, 2002; Ruiz et al.,  
271 2004, 2005; Murray, 2006; Mojtahid et al., 2016). On the contrary, *E. translucens* and *E.*  
272 *granosum*, as other noncarinate elphidiids, occur in outer estuaries with important marine  
273 influence (Ruiz et al., 2005; Mojtahid et al., 2016). Considering the environmental conditions  
274 indicated by *A. tepida*, *H. germanica*, *E. translucens* and *E. granosum* in the Guadalquivir  
275 estuary, a new Benthic Foraminiferal Salinity (BFS) index has been developed for  
276 reconstructing salinity changes within overall hyposaline conditions. Consequently, the BFS  
277 index was established using the relative abundances of *A. tepida* and *H. germanica* as  
278 indicative of lower salinity, and *E. translucens* and *E. granosum* as indicative of higher  
279 salinity. The expression of the BFS index is:

$$\text{BFS index} = (\% A. tepida + H. germanica) / (\% A. tepida + H. germanica + E. translucens + E. granosum)$$

282 The BFS index varies from 0 (higher salinity) to 1 (lower salinity), therefore higher  
283 values of the index indicate lower salinities, and vice versa. We posit that this BFS index may  
284 be a powerful tool to qualitatively reconstruct salinity changes and geomorphological  
285 restriction in present-day and ancient marginal-marine environments such as estuaries,  
286 marshes and lagoons. Similar indices for reconstructing salinity fluctuations were developed  
287 previously (Debenay, 1995; Hayward et al., 2004), but they were based on the relative  
288 abundances of a high number of species from benthic foraminiferal assemblages. The  
289 calculation of these indices is more complex since it is necessary to perform taxonomic  
290 identifications of a high number of species. In contrast, our BFS index requires the

291 identification of a very low number of species, which makes the application of this index easy  
292 and rapid. As a result, a profound taxonomic knowledge is not needed to use the index, and a  
293 higher number of samples can be analysed more rapidly.

294

### 295 *5.3. Holocene paleoenvironmental evolution of the Guadalquivir estuary*

296 The Holocene paleoenvironmental evolution of the Guadalquivir estuary was  
297 unravelled using the new Benthic Foraminiferal Salinity (BFS) index based on dominant  
298 benthic foraminiferal species. In order to assess past paleoenvironmental changes in detail,  
299 we use three degrees of salinity based on Debenay (1995) (Fig. 4): 1) higher salinity (BFS  
300 index = 0.0-0.4, high marine influence), 2) moderate salinity (BFS index = 0.4-0.7, moderate  
301 marine influence), 3) lower salinity (BFS index = 0.7-1.0, low marine influence).

302 In the interval before 2000 BCE, the salinity is higher and moderate in cores at a SW  
303 position, near the Atlantic Ocean (cores S7, S11) (Figs 2 and 4). Therefore, this area was well  
304 connected with the open sea during this interval. The core S6, located north of cores S7 and  
305 S11, records lower salinity, suggesting proximity of paleocoast towards the N. In the eastern  
306 area, the cores V.Ali1 and V.Ali2 show high BFS index values (lower salinity) confirming  
307 that paleocoast was in a northern position. The morphostratigraphic relationship between  
308 V.Ali1, sandy chenier, and V.Ali2, shelly chenier, reflects the progressive restriction of the  
309 estuary in the eastern area, before 2000 BCE. These data indicate an overall wide and  
310 moderately open paleoestuary, with an efficient connection to the Atlantic Ocean, although  
311 the processes of restriction and sedimentary infilling are evident, especially in the east. In this  
312 moderately open estuary, marine influence was high as shown by presence of marine fauna  
313 transported by tidal flows (Ruiz et al., 2005; Rodríguez-Ramírez et al., 2015).

314 From 2000 to 1400 BCE approximately, the estuary experimented a greater  
315 connection with the sea with higher marine influence, principally in distal areas, as a result of  
316 subsidence processes, sea-level rise and tsunamis (Rodríguez-Ramírez et al., 2014, 2015).  
317 This is especially visible in the nature of sandy sediments, corresponding to a small spit or  
318 sandy chenier, in the upper part of core S6, although without foraminiferal remains, due to  
319 intense rework of the sands (Rodríguez-Ramírez et al., 2015). At the SW area (cores S7 and  
320 S11), this stronger marine influence is evident by the decrease in BFS index from high to  
321 moderate values until 1400 BCE, approximately (Figs 2 and 4). In the eastern area, the lower  
322 part of cores VT and VAc show high BFS index values (lower salinity) confirming that the  
323 paleocoast was in a more northern position. The absence of sedimentary record at core V.Ali1  
324 might suggest that the NE area was already emerged.

325 In the interval from 1400 to 1000 BCE, the cores S7 and S11 show increasing BFS  
326 index values upwards, and the core S6 could be emerged. This indicates a southward  
327 shoreline progradation in the western area. Cores VT and VAc present high BFS index values  
328 during the entire interval. The locations of cores V.Ali2 and V.Ali1 were probably emerged.  
329 We interpreted the interval from 1400 to 1000 BCE as a phase of estuary restriction leading  
330 to a smaller estuary with less connection to the open sea. This interval coincides with the  
331 transition from an open estuary to a semiclosed estuary dated at 1200 BCE (Rodríguez-  
332 Ramírez et al., 2015). Growth of littoral spits in the river mouth and sedimentary infilling of  
333 the estuary may account for this gradual restriction (Rodríguez-Ramírez and Yáñez-  
334 Camacho, 2008).

335 The interval from 1000 BCE to present day is characterised by high BFS index values  
336 (lower salinity) in the central area (cores LB, AR, AA) (Figs 2 and 4). In the western area,  
337 cores S7 and S11 also show high BFS index values. The location of core S6 was likely  
338 emerged during this interval. The eastern area displays high BFS index values in core VT

339 pointing to high geomorphological restriction for the entire interval. In addition, the locations  
340 of cores VAc, V.Ali2, and V.Ali1 were surely emerged, suggesting a southward migration of  
341 the paleocoast. Therefore, this interval (1000 BCE-present) can be interpreted as a last phase  
342 of estuary restriction representing the lowest salinity and highest estuary restriction in the  
343 Holocene Guadalquivir estuary evolution.

344

#### 345 *5.4. Application of the BFS index to other regions*

346 In order to test the applicability of the Benthic Foraminiferal Salinity (BFS) index to  
347 other regions, we calculated the index in one sediment core (core 223 S12) and one  
348 stratigraphic section (Rambla de la Sepultura (RS) section) from two different regions and  
349 environments (Figs 1 and 5): 1) a Pleistocene lagoon in northern Italy (Barbieri and Vaiani,  
350 2018); and 2) a Pliocene coastal bay in southeastern Spain (Pérez-Asensio and Aguirre,  
351 2010). We selected these two regions because, together with the Guadalquivir estuary, they  
352 are representative of the most common types of marine-marginal environments. The  
353 Pleistocene lagoon from N Italy was a brackish lagoon environment (Barbieri and Vaiani,  
354 2018). Benthic foraminiferal assemblages from lagoonal sediments from core 223 S12 were  
355 dominated by *Ammonia parkinsoniana* and intermediate *Ammonia tepida*–*Ammonia*  
356 *parkinsoniana* forms, with secondary taxa including *Haynesina germanica*, *Aubignyna*  
357 *perlucida*, *Ammonia tepida*, and *Criboelphidium granosum* (Barbieri and Vaiani, 2018). The  
358 Pliocene coastal bay from SE Spain was a restricted coastal bay with coral banks, which was  
359 filled with terrigenous sediments of prograding fan deltas (Pérez-Asensio and Aguirre, 2010).  
360 In the RS section from this sheltered bay, benthic foraminiferal assemblages were mainly  
361 characterised by species from the genus *Ammonia* (*A. beccarii*, *A. tepida*, *A. inflata*) and  
362 noncarinate *Elphidium* species (*E. translucens*, *E. granosum*) (Pérez-Asensio and Aguirre,



2010). BFS index values from these different locations and environments were compared with the BFS index values from core S7 (Fig. 5). In this core from the Holocene Guadalquivir estuary, BFS index increased from low to high values in the lower part of the core, indicating progressive restriction. The BFS index values decreased in the middle part of the core, indicating better connection to the Atlantic Ocean. Finally, values increased from moderate to high values, marking the final phase of estuary restriction. In the core 223 S12 from the Pleistocene lagoon of northern Italy, BFS index was overall high along the core, except for two samples at 163.2 and 161.9 m (Fig. 5). These values suggest that this marginal-marine environment was highly restricted, and only experienced better connection to the sea episodically. This interpretation is consistent with a brackish lagoon environment with minor environmental fluctuations related to both freshwater and marine water inputs (Barbieri and Vaiani, 2018). The BFS index values from the Pliocene coastal bay from southeastern Spain increased gradually from low to high values along the RS section. This upward decrease in salinity is in good agreement with a brackish environment that was progressively restricted due to the progradation of fan deltas (Pérez-Asensio and Aguirre, 2010).

After applying the BFS index to two other regions, we propose that our index can be successfully applied to different regions, environments and timescales. This is very feasible because the four species used to calculate the index are widespread in marginal-marine environments around the world. To calculate the index, it is only necessary the presence of *A. tepida* or *H. germanica*, and *E. translucens* or *E. granosum*. Additionally, if *E. translucens* and *E. granosum* are not found, other noncarinate elphidiids with similar ecological requirements might be used (e.g. *Cribroelphidium excavatum*) (Mojtahid et al., 2016). Accordingly, only two to four species are needed to be identified, making the application of the index to other regions simple and fast.

387

## 388 **6. Conclusions**

389 Benthic foraminiferal distribution and abundance from the Holocene Guadalquivir  
390 estuary are controlled by the degree of salinity. This relationship allowed us to develop a  
391 simple Benthic Foraminiferal Salinity (BFS) index based on four benthic foraminiferal  
392 species (*A. tepida*, *H. germanica*, *E. translucens*, *E. granosum*). Since the BFS index is based  
393 on the identification of a very low species number, it is easy to calculate, even for scientists  
394 lacking a profound knowledge on benthic foraminiferal taxonomy. The low number of  
395 species to be identified also permits to calculate the BFS index rapidly, which is a great  
396 advantage for large-scale studies involving a high number of samples and sites.

397 The BFS index has allowed us to describe reliably the Holocene paleoenvironmental  
398 evolution of the Guadalquivir estuary with great detail, validating the index as proxy for  
399 different degrees of salinity, and therefore marine influence. We used three degrees of  
400 salinity (higher = 0.0-0.4, moderate = 0.4-0.7, lower = 0.7-1.0), which help us to differentiate  
401 subtle changes in geomorphological restriction during the Holocene. According to the BFS  
402 index values, the Holocene paleoenvironmental evolution of the estuary had four distinct  
403 phases: 1) wide and moderately open estuary (> 2000 BCE), with high connection to the  
404 Atlantic Ocean allowing the entrance of transported marine fauna; 2) (2000-1400 BCE)  
405 expansion of estuary due to sea-level rise and subsidence; 3) a phase of estuary restriction  
406 (1400-1000 BCE), with southward shoreline progradation related to spits growth and  
407 sedimentary infilling, which coincides with the transition from open to semiclosed estuary; 4)  
408 last phase of estuary restriction (1000 BCE-present day), with the lowest salinity and highest  
409 estuary restriction.

410 Our BFS index can also be applied to other regions, environments and timescales,  
411 identifying only two to four species (*A. tepida* or *H. germanica* + *E. translucens* or *E.*

412 *granosum*). This means the index is simple and rapid to calculate, and useful for qualitatively  
413 reconstructing salinity changes in worldwide marginal-marine environments from different  
414 time periods.

#### 415 **Declaration of competing interest**

416 The authors state that they have no competing financial or personal interests that  
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418

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432

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646

## 647 **Figure captions**

648 **Figure 1.** Study area and location of the short and deep cores. The Spanish local term ‘caño’  
649 refers to a relict, fully filled-in tidal-fluvial channel. Upper map created with GeoMapApp  
650 (<http://www.geomapapp.org/>). (For interpretation of the references to colour in this figure  
651 legend, the reader is referred to the Web version of this article).

652

653 **Figure 2.** Lithostratigraphy, chronology and benthic foraminiferal data (dominant species %,  
654 BFS index) of the short and deep cores. (For interpretation of the references to colour in this  
655 figure legend, the reader is referred to the Web version of this article).

656

657 **Figure 3.** Principal Component (PC) scores of the dominant euryhaline benthic foraminiferal  
658 species of the short and deep cores. (For interpretation of the references to colour in this  
659 figure legend, the reader is referred to the Web version of this article).

660

661 **Figure 4.** Benthic foraminiferal Salinity index values (BFS index) of the short and deep cores  
662 showing the Holocene paleoenvironmental evolution of the Guadalquivir estuary. (For  
663 interpretation of the references to colour in this figure legend, the reader is referred to the  
664 Web version of this article).

665

666 **Figure 5.** Comparison of the Benthic Foraminiferal Salinity index values (BFS index) of  
667 three marginal-marine environments from different regions and timescales: core S7 (this  
668 study), core 223 S12 (Barbieri and Vaiani, 2018), and Rambla de la Sepultura (RS) section  
669 (Pérez-Asensio and Aguirre, 2010) (For interpretation of the references to colour in this  
670 figure legend, the reader is referred to the Web version of this article).

671

## 672 **Table captions**

673 **Table 1.** Location (latitude, longitude) of the studied short and long cores (S6, LB, AR, AA,  
674 VT, VAc, VAli.2 VAli.1, S7, S11).

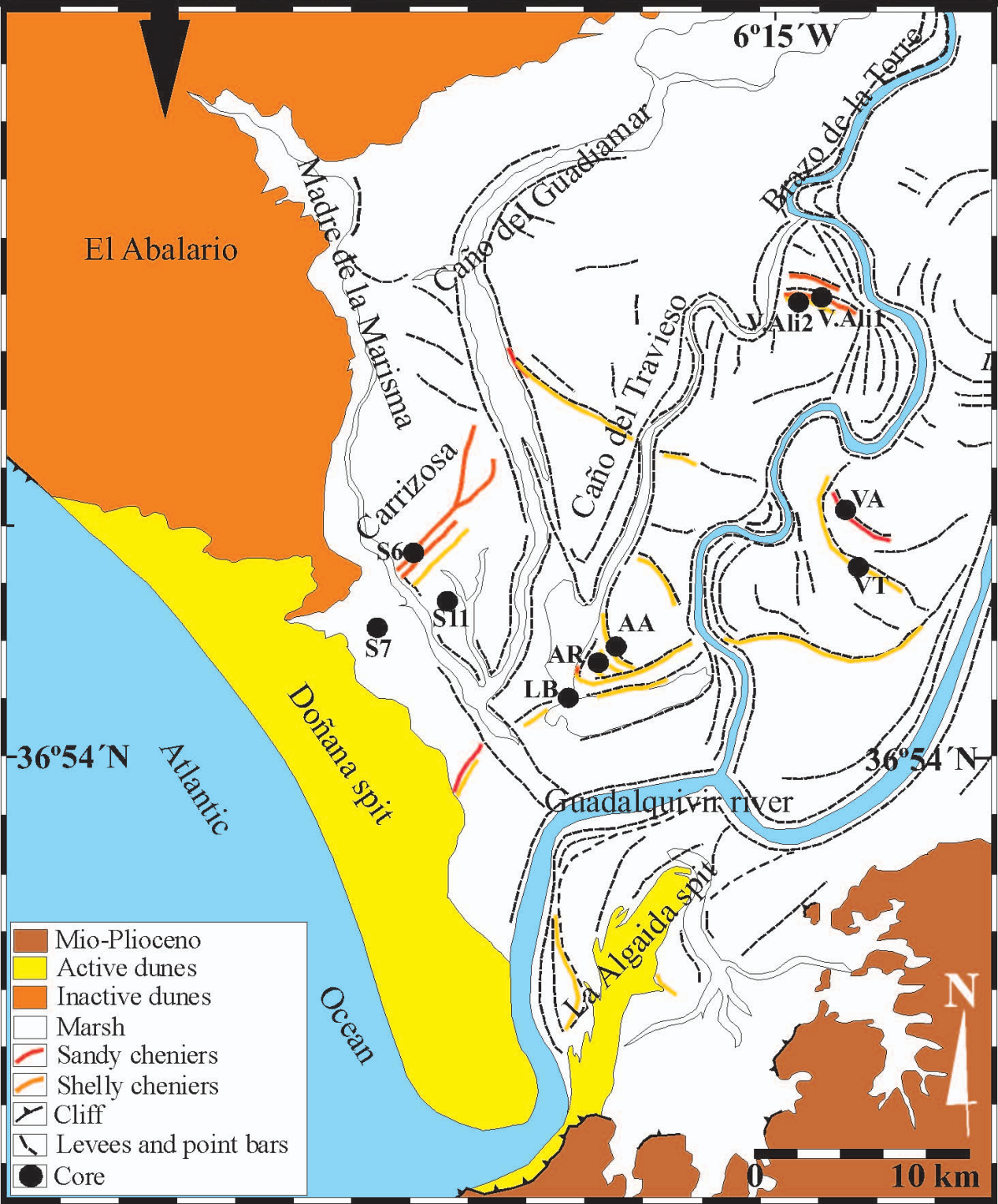
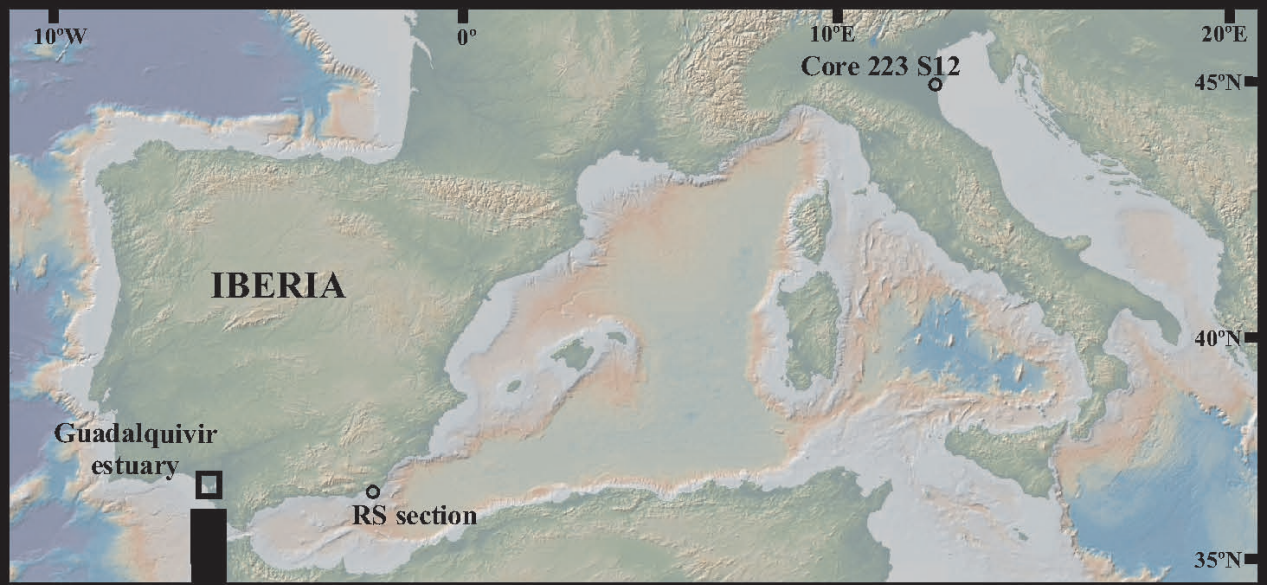
675

676 **Table 2.** Radiocarbon  $^{14}\text{C}$  data using the CALIB 7.10 software (Stuiver and Reimer, 1993),  
677 the calibration dataset of Reimer et al. (2013), and a  $\Delta R$  value of  $-108 \pm 31$   $^{14}\text{C}$  yr (Soares,  
678 2015). Uncertainties of the calibrated ages are expressed as  $2\sigma$  errors. B.—Beta Analytic  
679 Laboratory (Miami, USA). CNA.—Centro Nacional de Aceleradores (Seville, Spain).  
680 DAMS.—Accium BioSciences Accelerator Mass Spectrometry Lab (Seattle, USA). CX.—  
681 Geochron Laboratories, Krueger Enterprises, Inc. (Cambridge, USA). (1) Rodríguez-Ramírez  
682 et al. (2014). (2) Rodríguez-Ramírez et al. (2015). (3) Rodríguez-Ramírez et al. (2016).

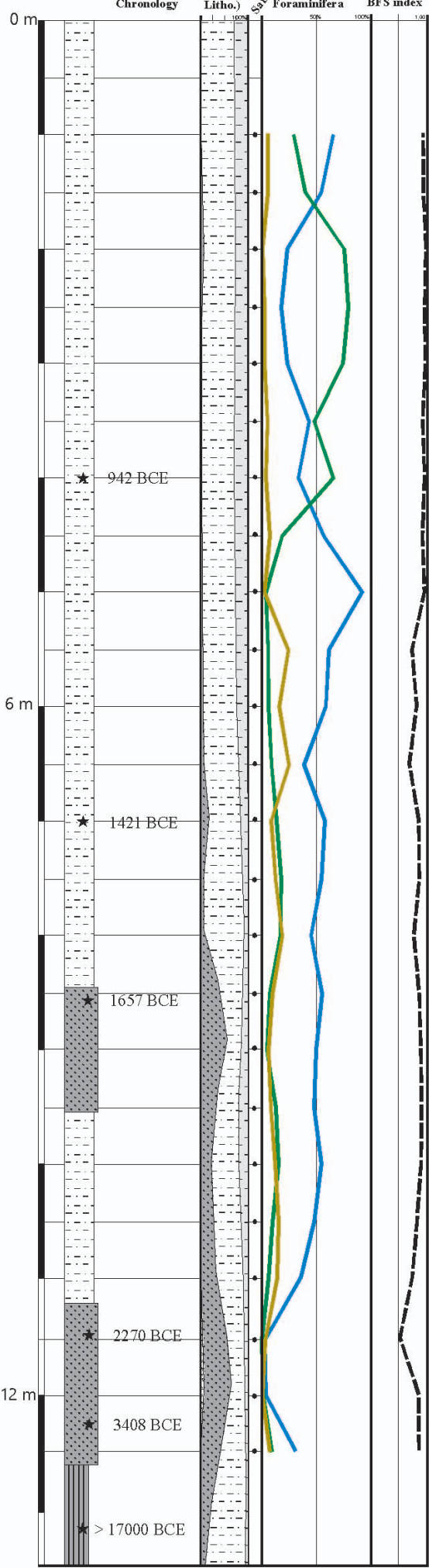
683

684 **Table 3.** Q-mode principal component analysis results from the short and deep cores.  
685 Explained variance (%) of each PC axis (assemblage), and the dominant and secondary  
686 species are indicated.

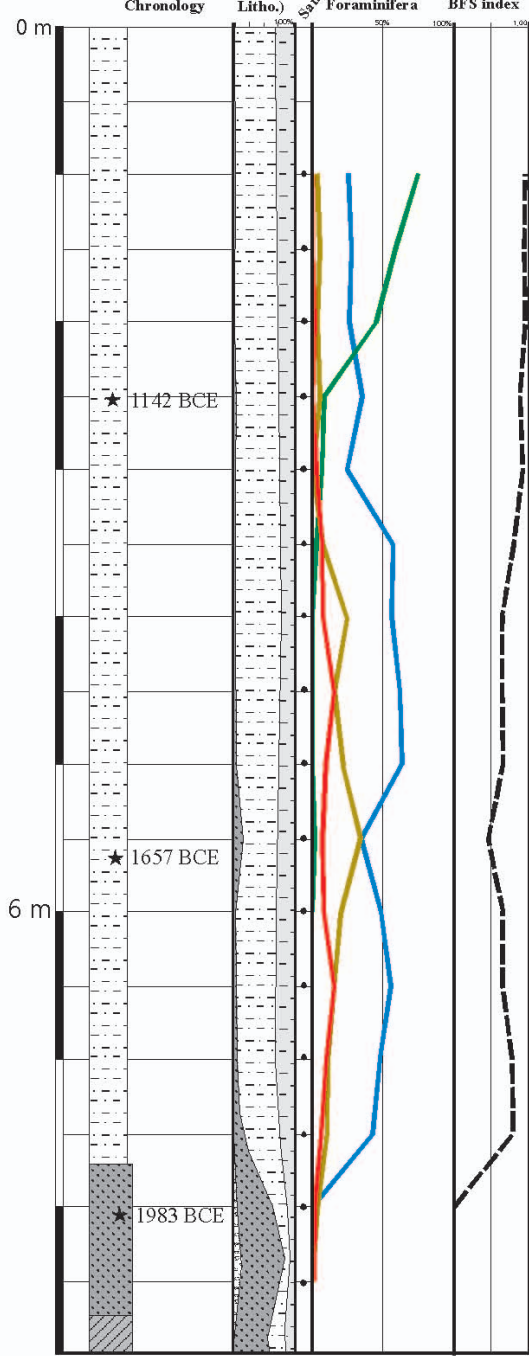
687



# S11

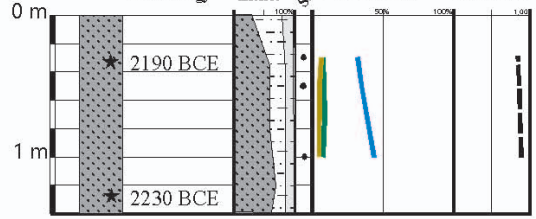


# S7

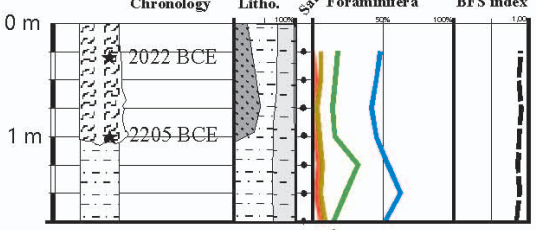


- Clay
- Silt
- Sand
- Gravel
- Shells
- Abalarío Dune systems
- Pleistocene Marshland
- Haynesina germanica*
- Ammonia tepida*
- Elphidium granosum*
- Elphidium translucens*

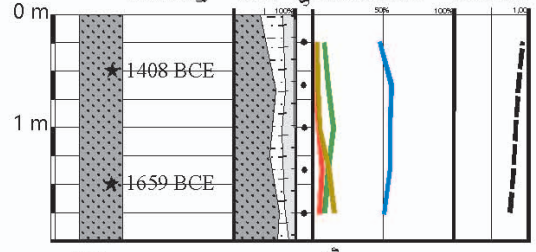
# VAl1.1



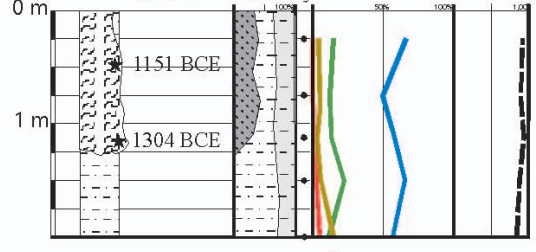
# VAl1.2



# VAc



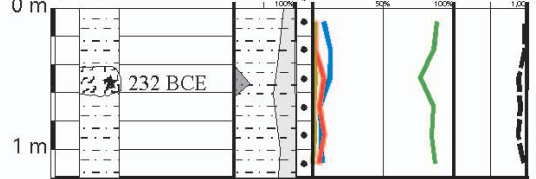
# VT



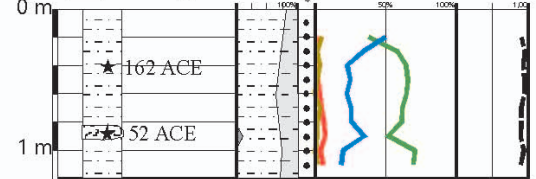
# AA



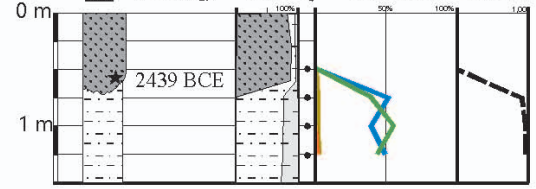
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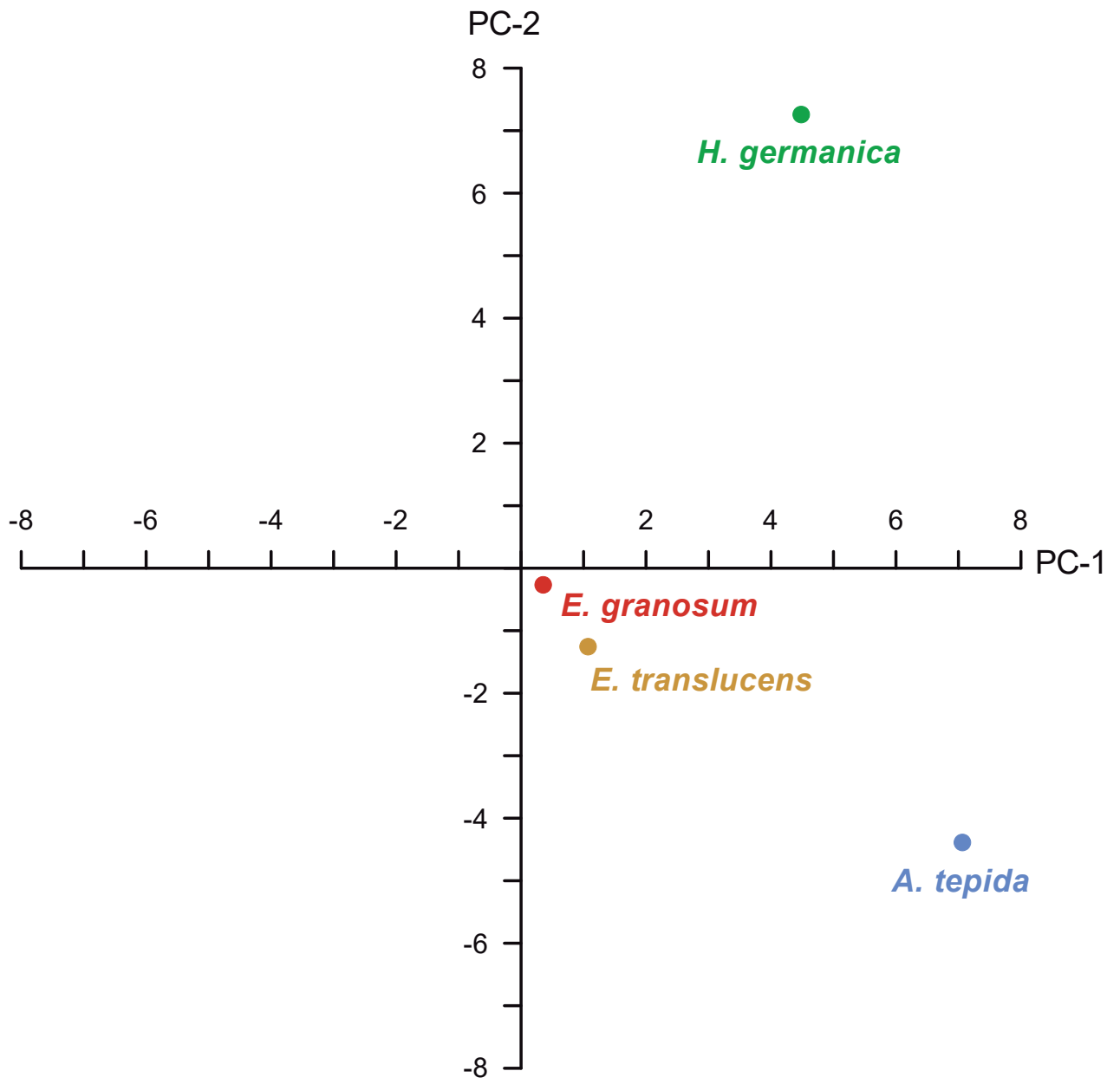
# LB

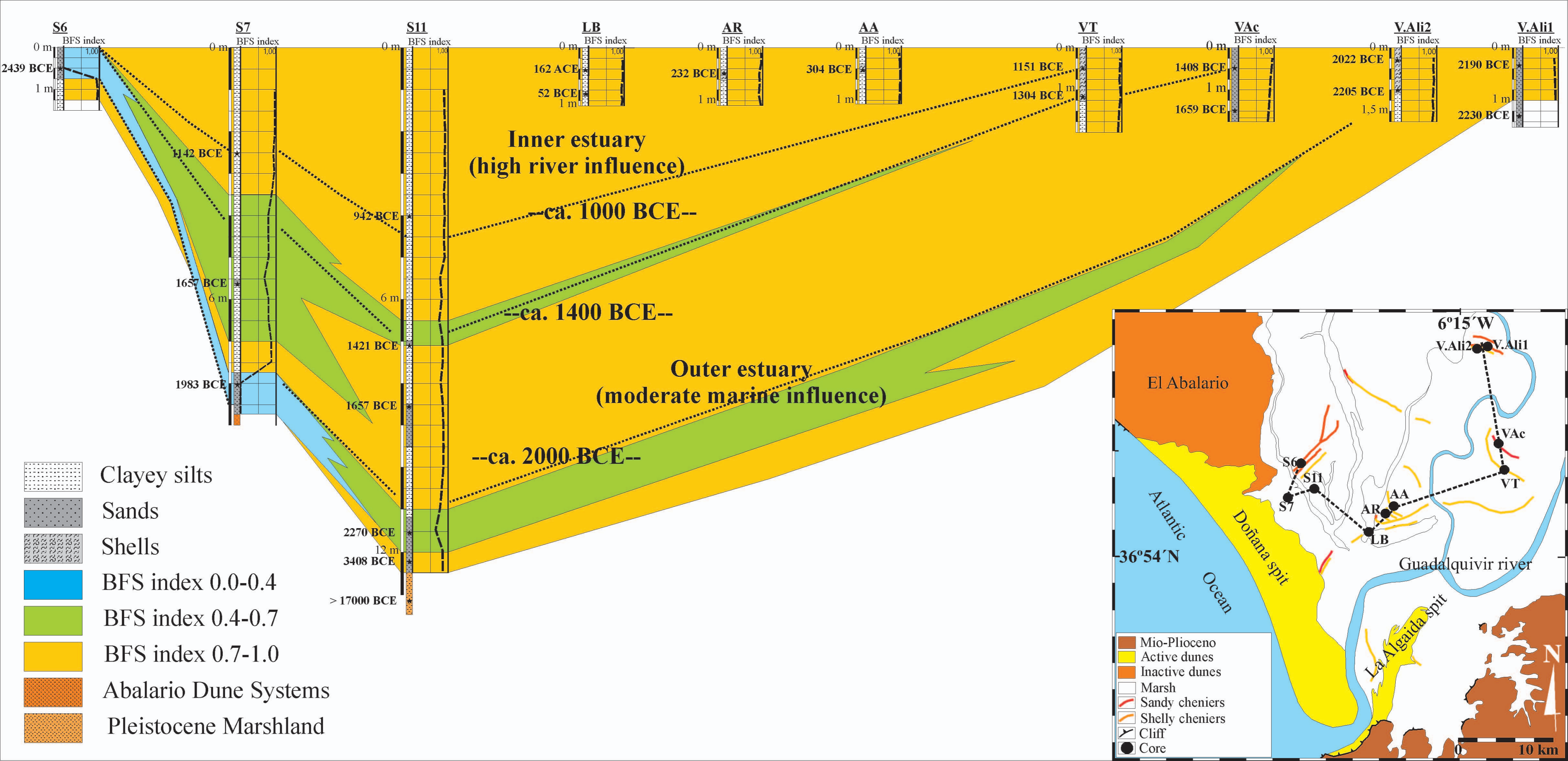


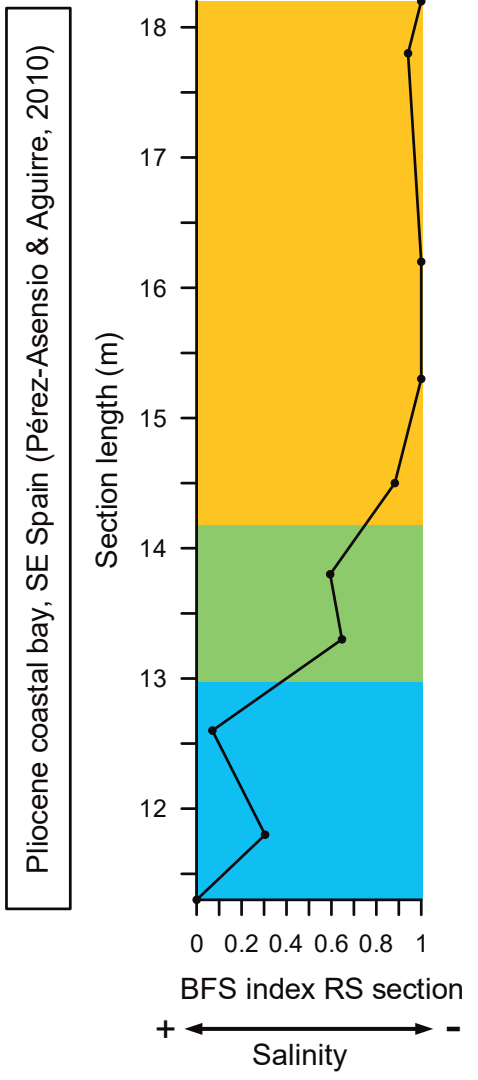
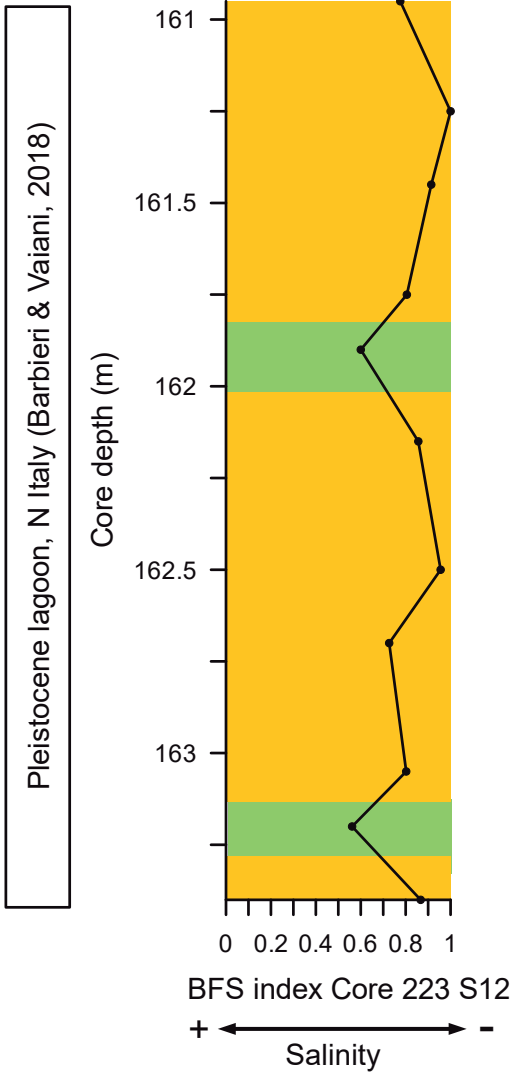
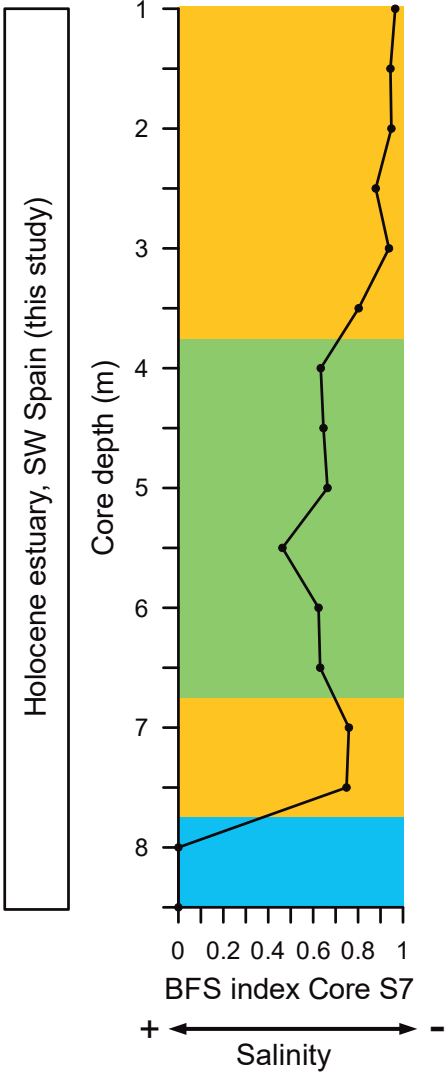
# S6











BFS index 0.0-0.4    BFS index 0.4-0.7    BFS index 0.7-1.0

Core	Latitude	Longitude
S6	36°58'35.90"N	6°24'39.08"W
LB	36°56'24.95"N	6°20'04.55"W
AR	36°56'47.99"N	6°19'15.98"W
AA	36°56'56.67"N	6°19'05.50"W
VT	36°58'00.78"N	6°13'04.52"W
VAc	36°59'38.31"N	6°13'07.99"W
VAl.2	37°04'13.89"N	6°14'19.84"W
VAl.1	37°04'13.37"N	6°14'27.25"W
S7	36°56'27.79"N	6°24'46.46"W
S11	36°58'04.93"N	6°23'47.38"W

Core	Lab. Ref.	Depth (m)	<sup>14</sup> C age (BP)	<sup>14</sup> C cal yr 2σ (BCE-ACE)
S6	B-287646 <sup>(1)</sup>	-0.5	4370±40	2757 BCE-(2439BCE)-2122 BCE
S7	B-285000 <sup>(2)</sup>	-2.5	3370±40	1409BCE-(1142BCE)-876BCE
S7	B-285001 <sup>(2)</sup>	-5.5	3780±40	1917BCE-(1657BCE)-1397BCE
S7	B-285002 <sup>(2)</sup>	-8	4040±40	2281BCE-(1983BCE)-1685BCE
S11	B-285006 <sup>(2)</sup>	-4	3190±40	1244BCE-(942BCE)-722BCE
S11	D-AMS 002422 <sup>(2)</sup>	-7	3596±60	1719BCE-(1421BCE)-1123BCE
S11	D-AMS 001537 <sup>(2)</sup>	-8.5	3781±33	1912BCE-(1657BCE)-1402BCE
S11	D-AMS 001538 <sup>(2)</sup>	-11.5	4260±30	2560BCE -(2270BCE)-1981BCE
S11	B-285007 <sup>(2)</sup>	-12.5	4860±40	3511BCE-(3408BCE)-3305BCE
S11	B-285008 <sup>(2)</sup>	-13.5	19360±80	Uncalibrated
VAl.1	D-AMS 030336	-1.3	4226±68	2557BCE-(2230BCE)-1904BCE
VAl.1	D-AMS 013505	0.4	4200±27	2470BCE-(2190BCE)-1910BCE
VAl.2	D-AMS 013507	0.5	4067±27	2314 BCE-(2022BCE)-1730 BCE
VAl.2	D-AMS 013506	-0.75	4215±24	2486 BCE-(2205BCE)-1925 BCE
VAc	CNA1118	-0.5	3590±80	1737 BCE-(1408BCE)-1079 BCE
VAc	DAMS-002424 <sup>(2)</sup>	-1.5	3778±54	1937 BCE-(1659BCE)-1382 BCE
VT	D-AMS 030340	-0.5	3384±64	1442 BCE-(1151BCE)-860 BCE
VT	CNA1117 <sup>(2)</sup>	-1.1	3504±48	1589 BCE-(1304BCE)-1019 BCE
AA	D-AMS 008483 <sup>(3)</sup>	-0.5	2494± 23	413 BCE-(304BCE)-196 BCE
AR	D-AMS 008481 <sup>(3)</sup>	-0.5	2404± 27	351 BCE-(232BCE)-113 BCE
LB	D-AMS 030339	-0.5	1981±60	113ACE-(162ACE)-437ACE
LB	D-AMS 008482	-0.8	2267± 26	166 BCE-(52ACE)-62 ACE

PC	Variance (%)	Species	Score
1	67.8	<i>Ammonia tepida</i>	7.06
		<i>Haynesina germanica</i>	4.48
		<i>Elphidium translucens</i>	1.07
		<i>Elphidium granosum</i>	0.35
2	23.3	<i>Haynesina germanica</i>	7.26
3	4.2	<i>Triloculina trigonula</i>	7.48
		<i>Ammonia beccarii</i>	2.76
		<i>Quinqueloculina seminula</i>	1.58
		<i>Miliolinella</i> sp.	1.14
		<i>Quinqueloculina</i> sp.	1.12
		<i>Triloculina</i> sp.	0.76
		<i>Quinqueloculina vulgaris</i>	0.53
		<i>Quinqueloculina laevigata</i>	0.45