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Assessing the performance of a foraminifera-based transfer function to estimate sea-level changes in northern Portugal

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ABSTRACT

We assessed the performance of a transfer function model for sea-level studies using salt-marsh foraminifera from two estuaries of northern Portugal. An independent data set of 12 samples and 13 sub-fossil samples from a core were used to evaluate if reconstructions and errors derived from current models are adequate. Initial transfer function models provided very strong results as indicated by cross-validation (component 2; $r^2 = 0.80$–0.82; RMSEP ranged from 10.7 to 12.3 cm) and improved its performance by *ca.* 10% when sample size reached *ca.* 50. Results derived using an independent test data set indicate that cross-validation is a very effective approach and produces conservative errors when compared to observed errors. We additionally explored the possible effect of transforming the concentration data into percent in the error estimations by comparing the results obtained based on the use of both concentration and compositional data. Results indicate that this type of transformation does not affect the performance of the transfer function. Results derived from a reconstruction of sub-fossil samples from a core indicate that high-resolution sea-level reconstructions are possible, but show that depositional environments have to be selected carefully in order to minimize the impact of possible taphonomical loss.

Introduction

Phleger (1965) first suggested the existence of vertical zonation of salt-marsh foraminifera that corresponded to major marsh zones of vascular plants. This zonation indicates a response of foraminifera to strong variations linked to the penetration of tides and to freshwater influx from inland sources. Besides the great environmental variability of marsh environments, marsh surface foraminiferal distributions are roughly similar in all temperate areas: highest tidal levels are dominated by the *Jadammina macrescens*/*Trochammina inflata* assemblage, replaced by *Haplophragmoides* spp and *Miliammina fusca* with decreasing salinity (e.g., Hayward et al., 2004; Fatela et al., 2009). Mid-tidal elevations tend to reflect local variables (e.g., fresh water input and distance to the mouth of the estuary) and calcareous species become more abundant, although dependant on the availability of calcium carbonate (Phleger, 1970; Moreno et al., 2007; Valente et al., 2009).

Marsh foraminifera are considered the most accurate proxies for elevation and able to predict elevations to within ±0.05 m or better (Southall et al., 2006; Kemp et al., 2009) due to the strong correlation of the assemblages of agglutinated marsh foraminifera with elevation above mean tidal level (e.g., Gehrels et al., 2008). Since their introduction by Guilbault et al. (1996), foraminifera-based transfer functions have become a widely used tool in high-resolution sea-level reconstructions (see Discussion section for references). Through the use of transfer functions, the modern relationship between marsh foraminifera and elevation is used to calibrate core sediments in order to reconstruct past tide levels, determining the relative height of marsh sediment in the tidal frame. In fact, estimation of sea-level rise rates based on foraminiferal transfer functions and tide-gauge sea-level estimates are in good agreement for the 20th century (Church et al., 2008).

This research is especially relevant under the present scenario of rapid sea-level rise (19 cm for the 20th century, Jevrejeva et al., 2008) resulting from the global warming (IPCC, 2007). Furthermore, pre-20th century instrumental records are sparse and geographically very limited making difficult to resolve when the global sea-level acceleration started (Jevrejeva et al., 2006; Miller and Douglas, 2007). It is therefore desirable to provide sea-level curves at high resolution that are geographically distributed to supplement existing instrumental data sets and, importantly, extend these data sets back in time in order to accurately resolve the timing of the sea-level acceleration and the causes

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of the present sea-level rise. These records can be used to narrow down the uncertainties of current model predictions of future sea-level rise.

However, in the development of transfer functions, the “predictor” variable in the training sets is “height” (e.g., Kemp et al., 2009; Woodroffe, 2009). However, height is not an ecological or environmental variable. The ecological parameter that directly controls foraminiferal distributions is flooding duration (Gehrels, 2000). Furthermore, transfer functions are almost exclusively based on raw percent data (closed compositional data), despite the limitations that compositional data sets present (Loubere and Qian, 1997; Kucera and Malmgren, 1998). One additional limitation of this method is the lack of independent data sets to test the reliability of the reconstructions and their prediction ability is only assessed by cross-validation methods.

In this work, we present one transfer function model developed on the modern distributions of foraminifera recorded from six transects sampled in the Minho and Lima estuaries, northern Portugal (Fig. 1) and we assess for the first time its applicability using an independent test data set. This area was selected based on the fact that the tidal range is the smallest found in the SW European Atlantic coast (Leorri et al., 2010) and, therefore, sea-level reconstructions will have smaller errors. Also, the current lack of long-term sea-level data from this area and its relevance to adequately reconstruct sea-level trends for the last several centuries at regional scale were considered. We describe the development and application of this transfer function in order to provide a quantitative assessment of the potential of intertidal foraminifera for relative sea-level studies in Portugal, assess the performance of the transfer function model using an independent test data set, and investigate the impact of the use of percent data in the performance of the transfer function evaluated *versus* concentration data. In a final step, we will calibrate sub-fossil samples from a core recovered in the sampling area.

Sites description

Six salt marsh profiles in the Minho and Lima estuaries in northern Portugal were sampled between 2002 and 2006 (Fig. 1). This region has an average annual precipitation of 1300 mm, although during the wet season maximum precipitation often exceeds 2500 mm (Bettencourt et al., 2003).

The Minho estuary is oriented NNE-SSW, with a mean tidal range of 1.81 m (Table 1). Although tides can be measured up to 42 km upstream (Bettencourt et al., 2003), the direct marine influence has been reported between 11 km (Moreno et al., 2005) and 35 km (Bettencourt et al., 2003). The average river discharge is 300 m^{3}s^{-1} (www.maretec.mohid.com). The Minho estuary is very shallow due to widespread siltation resulting in a significant area of the bottom being exposed during low water spring tide, when the connection to the sea

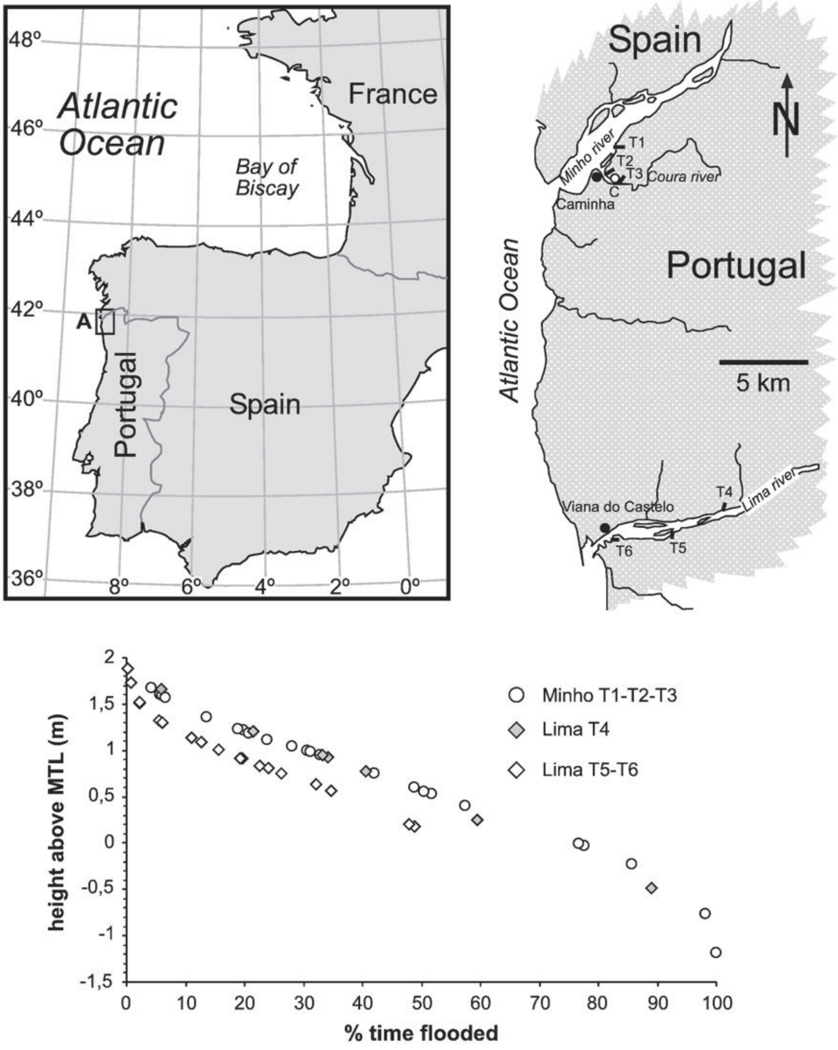

Figure 1. Location map of sampled marsh transects (T) and core (C) in the Minho and Lima estuaries in northern Portugal and relationship between flooding duration and height calculated for each sample included in the training set. MTL=mean tide level.

Table 1
Tidal parameters of the marsh transects studied (m referred to the hydrographical chart datum, 2 m below mean sea level): HAT = Highest Astronomical Tide; MHHW = Mean Highest High Water; MHW = Mean High Water; MTL = Mean Tide Level; and MLW = Mean Low Water.

	HAT	MHHW	MHW	MTL	MLW	Mean tidal range
T1-T3	4.26	3.81	3.47	2.56	1.66	1.81
T4	4.28	3.88	3.53	2.51	1.48	2.05
T5-T6	3.97	3.56	3.20	2.15	1.10	2.10

is restricted to two shallow channels (1 to 2 m below mean tidal level; Alves, 1996).

The Lima estuary is oriented ENE-WSW and is located about 20 km south of the Minho estuary (Fig. 1). The mean tidal range of ~2.10 m effects can be measured 20 km upstream (Table 1; Alves, 2003; Ramos et al., 2006). The mean fluvial flux is 62 m^3s^{-1} (www.maretec.mohid.com). The lower part of this estuary has been strongly modified by anthropogenic activities (installation of industrial and commercial facilities) and is subjected to periodic dredging to maintain channel navigation. The Lima estuary becomes very shallow upstream where salt marsh and several tidal islands develop. These islands are just below low spring water levels (Alves, 2003; Ramos et al., 2006).

Materials and methods

We sampled the uppermost centimeter of surface sediment along six transects that followed the elevational gradients. A total of 78 sediment samples were collected for micropaleontological analysis in the Minho–Lima area. Because the height above mean tide level was of primary interest, we obtained samples from as many elevations as possible. The absolute orthometric height of the marsh transects, tidal elevation and sampling location were obtained in the field using a Zeiss Elta R55 total station from previously selected bench marks. The bench marks were referenced to the national altimetric datum using differential GPS equipment in combination with a regional geoid model (Catalão, 2006) then linked to local chart datum. The regional geoid model was adjusted to local bench marks by using the differential GPS in RTK mode. All data are presented relative to the local hydrographical chart datum of Viana do Castelo which is 2.00 m below the mean sea level at Cascais in 1938.

Since the mean tidal range for the different salt marshes varies between 1.81 m (Minho; Table 1) and 2.05 to 2.10 m (Lima; Table 1) [Fig. 1], the height of surface samples was standardized to a common datum. We used a linear height normalization technique (e.g., Horton et al., 1999; Gehrels et al., 2001; Hamilton and Shennan, 2005; Leorri et al., 2008), here elevations are expressed as a standardized water-level index (SWLI):

$$\mathrm{SWLI}_n = \frac{100(h_n - h_{\mathrm{MTL}})}{h_{\mathrm{MHHW}} - h_{\mathrm{MTL}}} + 100$$

where SWLI_n is the standardized water-level index for sample n, h_n the elevation of sample n (m above local ordnance datum-alod), h_{MTL} the mean tide level elevation (m alod), h_{MHHW} the mean higher high water elevation (m alod). This technique produces a SWLI for each modern sample. An SWLI of 100 represents mean tide level (MTL) and an SWLI of 200 corresponds to mean higher high water (MHHW).

To assess the ability of the transfer function to reconstruct former sea-level changes and to identify suitable areas for high resolution studies (i.e., the depth at which marsh sediments occurred) the micropaleontological content of 21 samples were sampled from the upper 3 m of a borehole drilled in the Minho estuary close to transect 3 in 2003. One sample (2.6 m depth) was analyzed for radiocarbon content at Beta Analytic Inc. (USA) in order to provide a chronological framework.

Foraminiferal samples

At each sampling site 10 cm^3 was extruded by pressing down a hard plastic syringe into the surface layer. The top 1 cm of oxygenated sediment was placed in a bottle containing alcohol rose Bengal solution [1 g l^{-1}] (Lutze, 1964). Modern samples were sieved through a 63-micron mesh and washed to remove clay and silt material and the remaining residue was analyzed. Samples were stained using rose Bengal (Walton, 1952; Lutze, 1964; Murray and Bowser, 2000) to identify live specimens at the time of collection. Here, we used the unstained assemblages (dead) since they represent the time-averaged accumulation of foraminiferal tests (Murray, 1991) and are a better analogue for reconstructing past sea level (Horton, 1999; Horton and Edwards, 2003, 2006; Horton et al., 2005; Leorri et al., 2008). Foraminifera were collected using a micropipette and a wet picking procedure under a stereoscopic binocular microscope using reflected light. When possible, at least 100 individuals were counted in each sample from the dead assemblage as recommended by Fatela and Taborda (2002) for these environments. Identification of the foraminifera followed Loeblich and Tappan (1988) generic classification. Core samples were subsampled into 1 cm thick sections and analyzed as described previously but were not stained. In total, 99 samples were analyzed and more than 9300 foraminifera were examined.

Although infaunal populations of agglutinated foraminifera living at depths of 10 cm have been reported from the USA mid-Atlantic and southeast coastal salt marshes (Goldstein et al., 1995; Hippensteel et al., 2000), they are not analyzed here. Our analysis from the Lima estuary (Portugal, unpublished data) and in the northern Iberian peninsula (Leorri et al., 2008) suggests that living individuals are concentrated in the uppermost interval (0–2 cm) and do not significantly modify the assemblage downcore. This observation is also supported by results obtained elsewhere (e.g., Patterson et al., 2004; Culver and Horton, 2005; Tobin et al., 2005).

Statistical analysis

When key environmental factors (i.e., tidal flooding and salinity) are autocorrelated in the training set, they may influence the ability to reconstruct past conditions (Loubere and Qian, 1997). Moreover, positive spatial autocorrelation (the tendency of sites close to each other to resemble one another more than randomly selected sites) is a property of most ecological data that makes the predictive power of some models over-optimistic and misleading. Combined training sets from various sites provide a more realistic analogue for (sub)fossil assemblages than local data sets (Gehrels et al., 2001). We, therefore, selected six transects that represent different estuarine areas, in addition, this minimizes the possible effect of intercorrelation of elevation and salinity and spatial autocorrelation. Furthermore, Telford and Birks (2005) demonstrated that weighted-averaging partial least squares model (WA-PLS) is least sensitive to these effects.

WA-PLS regression (Juggins, 2004) was used as the transfer function model based on results from detrended correspondence analysis (DCA) and detrended canonical correspondence analysis (DCCA). These techniques help detect the species response, i.e., whether unimodal (Gaussian) or linear models are appropriate (Sejrup et al., 2004). DCCA provides an estimate (as the length of DCCA axis 1) of the gradient length in relation to *x* (environmental variable) in standard deviation (SD) units (Birks, 1995; Korsman and Birks, 1996). If the gradient length is longer than 2 SD units, several species will have their optima located within the gradient and then unimodal-based methods of regression and calibration are most appropriate. WA-PLS is an extension of the unimodal method weight averaging (WA), which considers the variance along the environmental variable of interest (i.e., tidal flooding). Other environmental variables (e.g., salinity) influence foraminiferal distributions, and this may distort the microfaunal-environment relationships. In WA-PLS, further components are calculated orthogonal to earlier components and they are obtained as WA of the residuals for the environmental variable, improving the predictions as it considers the combined influence of additional environmental variables (ter Braak and Juggins, 1993). Statistical analyses were performed using CANOCO 4.0 (ter Braak and Smilauer, 1998) and C2 (Juggins, 2004) software. To statistically assess the predictive power of different transfer function models and select the best model, jack-knife and bootstrapping cross-validation statistical tests were performed (Birks et al., 1990; Line et al., 1994; Sejrup et al., 2004; Telford and Birks, 2005). The models were selected based on low root-mean-square of the error of prediction (RMSEP), low maximum bias, high squared correlation (r^2) of observed *versus* predicted values, and the smallest number of "useful" components. The RMSEP indicates the systematic differences in prediction errors, whereas the r^2 measures the strength of the relationship of observed *versus* predicted values. In general terms, we considered a "useful" component when improved the previous component performance by at least 5% of the component 1 (Birks, 1995). In a further step to evaluate the performance of the different models, we semi-randomly selected a subset of samples to use them as an independent test data set (i.e., samples were clustered into three groups representing different elevations and randomly selected within these clusters, so the data set represented a variety of elevations). Removing samples from the original data set reduces the model performance but, in turn, this provides information of the effect in the model performance of increasing the number of samples. We used bootstrapping to derive a standard error of prediction (SEpred; Birks et al., 1990; Line et al., 1994), which varies from sample to sample depending upon the composition of the test set and the presence or absence of taxa with a particularly strong signal for the environmental variable of interest (Birks, 1995). SEpred was estimated using 1000 cycles.

The initial data set was built up using percent data from unscreened samples above mean low high water (MLHW; see Discussion section below). In order to identify the effect of standardizing the data set we created a second data set using concentration data (foraminiferal abundances per unit of sediment volume), comprising all samples above MLHW. In order to assess the performance of these transfer functions, we semi-randomly removed the same twelve samples in both data sets to use them as an independent test data set.

Results

In this study 44 species of foraminifera with an abundance of at least 2% in a single sample were identified. Although in most samples foraminifera were in low abundance, only eight samples were discarded from statistical analysis. Figure 2 and Table 2 summarize the results from all study sites grouped by elevational zones. The relative abundance of the dominant species at each sampling site shows the transition of the different assemblages (Fig. 2B).

Development of foraminifera-based transfer functions

Table 3 summarizes the results. The application of DCCA to the training sets produced a gradient length of between 2.10 and 2.74 SD, suggesting a unimodal distribution of foraminiferal abundance with respect to SWLI for all data sets. ter Braak et al. (1993) concluded that for compositional data, WA-PLS is commonly a better calibration method than either WA or partial least square. All models use WA-PLS component 2 based on the smallest number of components, low RMSEP and low maximum bias (see Birks, 1995).

Percent data

Model 1 corresponds to the unscreened data set using 49 samples (samples above MHLW) and 44 species (abundances greater than 2% in a single sample) of percent data and displayed a very strong performance. Model 1b replicates Model 1 after semi-randomly removing twelve samples to independently test the model with similarly strong results (see Table 3). Removing the twelve samples reduced the performance *ca.* 10% *versus* the original model. The results derived from the use of the independent data set to test the training set were also very strong ($r^2 = 0.91$; RMSE = 6.8). These models use samples above MLHW (a standardized water-level index of 120) since samples below MLHW are considered less suitable for sea-level studies (Hamilton and Shennan, 2005; Woodroffe, 2009).

Concentration data

Model 2 corresponds to the unscreened data set using 49 samples (samples above MHLW) and 44 species (abundances greater than 2% in a single sample) of concentration data, while Model 2b replicates Model 2 after semi-randomly removing twelve samples to independently test the model. The results were very strong in all cases whether the test was done with cross-validation or using the independent test data set (see Table 3). The twelve samples removed were the same ones used in Model 1b and model performance was also reduced (between 7 and 9%).

Models 1 and 2 perform very strongly (component 2; $r^2 = 0.80$–0.82; RMSEP ranged from 10.7 to 12.3 cm). The results derived from both models were identical. The results obtained from the use of the training set were also very strong. From the twelve samples included in the training set, both models were able to reconstruct within their error range the elevations of ten of them and only two fell outside the error envelope (Fig. 3) by a small amount. In the case of sample 1, it is placed at the lower end of the gradient considered and sample 9 was associated with the presence of *Quinqueloculina* spp. and *Cibicides lobatulus* (see Discussion section below).

Core data

Borehole C was located at 1.505 m above MTL in the higher vegetated sub-environment of the marsh (Fig. 1). The uppermost 2.6 m of the core was highly organic composed of dark brown laminated mud (*ca.* 90% silt) with a small sand content. Below this depth, sediment became sandy with a lower content of organic matter. Our analysis was restricted to the uppermost 2.6 m section.

A micropaleontological study of 21 samples resulted in only 13 samples that could be used in the statistical analysis. Three distinct foraminiferal assemblage zones (FAZ), dominated by five different taxa, were differentiated. The basal 80 cm (FAZ 3, 260–170 cm depth interval) was characterized by a high dominance of *T. inflata* (average 34%) together with *Paratrochammina* cf. *guaratibaensis* (average 11%). The intermediate (FAZ 2, 170–60 cm) section presented very low numbers of foraminifera. The uppermost 60 cm (FAZ 1) were dominated by *H. manilaensis* (average 48%) and *J. macrescens* (average 10%). Figure 4 summarizes micropalaeontological results, bulk density data and transfer function calibration for this core section. The sedimentological and foraminiferal data suggest a salt marsh environment where foraminiferal taxa have optima high in the tidal frame (within FAZs 3 and 1). Thus, Model 1, which consists of modern samples from higher elevations is adequate to calibrate these foraminiferal assemblages. The calibration process assigns a paleomarsh-surface elevation to each core sample together with sample-specific standard errors of prediction for individual core samples. The predicted paleomarsh elevations reflect the changes in foraminiferal assemblages with bootstrapped estimated errors between 12 and 15 cm. Although the dominant species are fairly similar throughout the section analyzed, their relative abundances vary significantly. As a consequence, the transfer function predicts that the marsh surface at the sample site has varied up to 30 cm relative to MSL during the last *ca.* 2500 years.

One radiocarbon analysis (Beta-181635) at 260 cm depth yielded an age of 2345 cal yr BP (cal yr BP 2365 to 2320; 2 sigma).

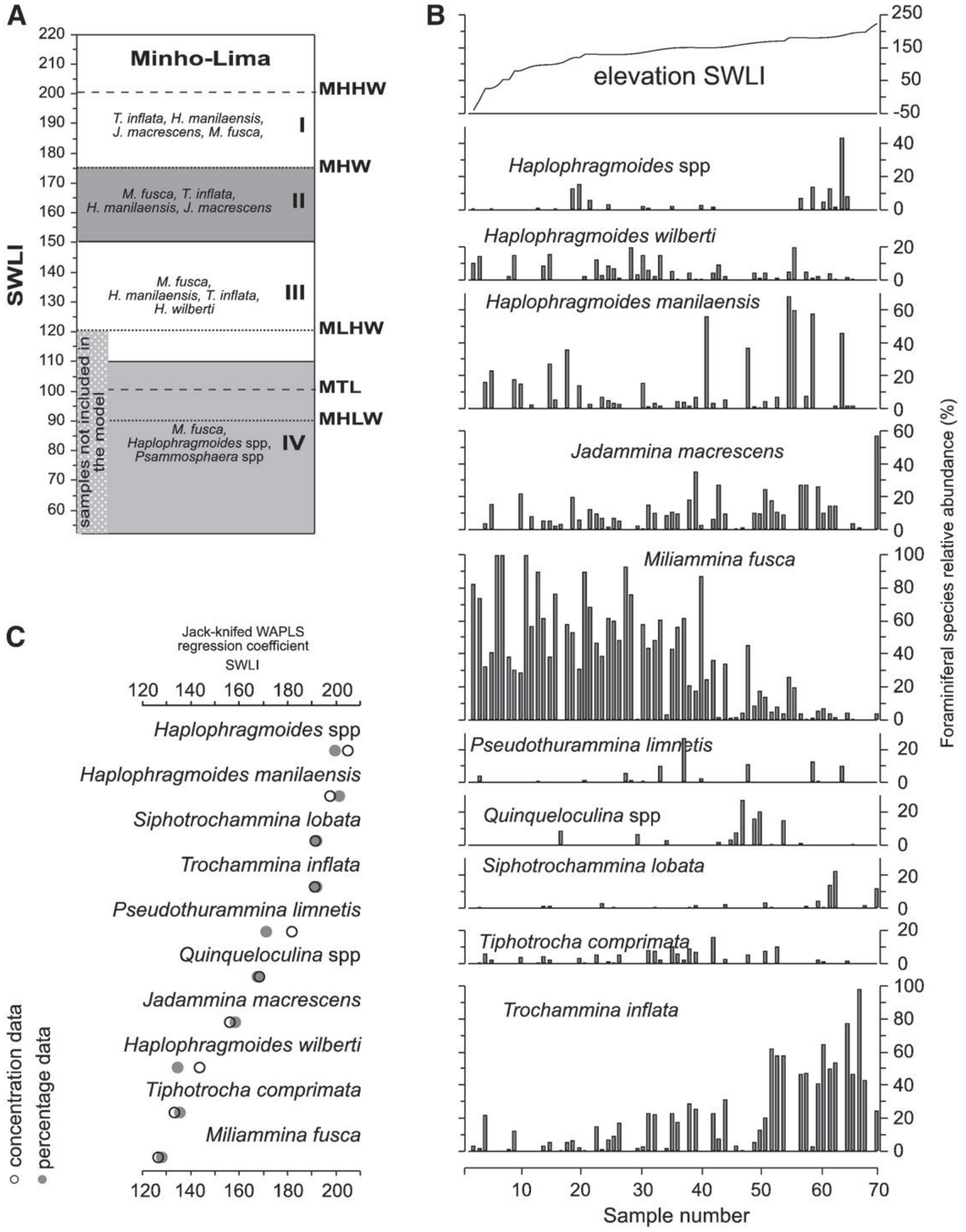

Figure 2. A: Elevational zonation of foraminiferal assemblages across the salt marshes studied on the Lima and Minho estuaries. Dominant species are shown. Samples not included in the models are also indicated. MHHW: mean highest high water; MHW: mean high water; MLHW: mean low high water; MTL: mean tide level; MHLW: mean high low water. SWLI: standardized water-level index. B: Relative abundances of the dominant marsh foraminifera (dead assemblages) from the Lima and Minho estuaries. C: The jack-knifed regression coefficient for the relationship between the SWLI and each dominant foraminiferal species as computed by weight averaging-partial least squared (WA-PLS) regression model. Coefficients are presented for both concentration and percent data. Regression coefficients are similar for all the species except for *Haplophragmoides wilberti*, *Haplophragmoides* spp and *Pseudothurammina limnetis*. Low numbers associated to these species may explain the different coefficients. *Haplophragmoides* spp, *Haplophragmoides manilaensis, Siphotrochammina lobata, P. limnetis, T. inflata* and *Quinqueloculina* spp coefficients indicate that their relative abundance and concentration decrease with decreasing elevation. Species that increase in concentration and relative abundance with decreasing elevation include *M. fusca, J. macrescens, H. wilberti* and *Triphotrocha comprimata.*

Discussion

The microfaunal composition of the dead assemblages presented here reflects the prevailing wet Atlantic climate (Bettencourt et al., 2003). This limits the hydrochemical marine influence within the estuaries on this geographical area as it is recorded by the foraminiferal assemblages, restricting the presence of foraminifera below MHHW and exhibiting a significative presence of *M. fusca* and *Haplophragmoides* spp usually associated with lower salinities. On the other hand, cored material indicated that marsh environments have developed in the area at least during the last 2500 years suggesting that this area might be suitable for high resolution sea-level studies.

Most models for sea-level reconstructions are based on marsh environments in which foraminifera with optimum occurrences in the

Table 2
The most abundant forms found in these marshes clustered by Autochthonous and Allochthonous species and elevational zones (I to IV). See also Figure 2.

	Mean	Range
Autochthonous		
M. fusca	40	0–100
Haplophragmoides spp	17	0–90
T. inflata	12	0–88
J. macrescens	7	0–58
Allochthonous		
Cibicides spp.	6	0–40
Zone I		
T. inflata	32	0–88
H. manilaensis	27	0–68
J. macrescens	7	0–28
M. fusca	7	0–26
Zone II		
M. fusca	25	2–87
T. inflata	17	0–60
H. manilaensis	11	0–56
J. macrescens	10	0–27
Quinqueloculina spp	5	0–20
C. lobatulus	7	0–28
Zone III		
M. fusca	53	1–93
H. manilaensis	6	0–36
T. inflata	6	0–23
H. wilberti	6	0–20
Zone IV		
M. fusca	64	0–100
Haplophragmoides spp	9	0–25
Psammosphaera spp	6	0–53

high part of the flooding duration gradient are dominant, i.e., high marsh assemblages (e.g., Gehrels, 2000). On the contrary, tidal-flat assemblages contain foraminiferal species whose optimum occurs at the bottom end of the gradient, well below the optima of other species. Small changes in the abundance of these latter species considerably affect the predicted indicative meaning of the foraminiferal assemblages and, as a result, the error of the model increases (Gehrels, 2000; Woodroffe, 2009). We, therefore, removed all samples below local MLHW. The performance of the resulting model was very strong.

However, while in the literature transfer functions for sea-level studies are based almost exclusively on height normalization and closed compositional data (percent), these two approaches are not exempt of errors. From an ecological point of view, marsh foraminifera are controlled by sub-aerial exposure, hence, by flooding duration. The relationship between height and flooding duration is non-linear, particularly for the upper marsh surface and, therefore, the time of sub-aerial exposure is very sensitive to small changes in height. This normalization has been recommended as the best option because it produces smaller standard errors for the indicative meaning of foraminiferal assemblages than normalization according to height (Gehrels, 2000). Reconstruction of environmental variables based on transfer functions should be done when these variables are either ecologically important determinants in the system (e.g., flooding time) or linearly related to such determinants (Birks, 1998). This does not rule out the use of height normalization and, in fact, the aim of these transfer functions is to reconstruct the former elevation of the depositional environments, but it should be considered carefully.

The second issue is the effect of closed compositional data. In these transfer functions micropaleontological data are expressed as relative abundances. Transformation of any data into such a form gives rise to the constant-sum constraint, a circumstance violating basic assumptions upon which standard statistical analyses are designated (Kucera and Malmgren, 1998). In fact, Loubere and Qian (1997) demonstrated that the conversion of concentration data to percentages can yield artificial correlations among species. This effect, named as matrix closure, creates linear distortion in the ecological response patterns of the taxa within the data sets (Mekik and Loubere, 1999).

Overall, matrix closure has a homogenizing effect on taxon response by spreading the environmental signals from species that carry a strong environmental signal to those species that do not respond to it, becoming a potential source of error in paleoenvironmental estimations (Mekik and Loubere, 1999). Moreover, the total number of tests per volume unit may provide additional information since foraminifera vanish towards the very top of the intertidal area. This is important because most accurate transfer functions for sea level tend to concentrate on narrow vertical ranges of the highest intertidal zone (e.g., Southall et al., 2006) and, therefore, the information provided by the variance of each individual species is very valuable (i.e., concentration data).

Our aim here is not to provide a new approach to resolve former sea-level changes but instead to assess if the errors calculated using the current models are realistic or some additional sources of error should be considered. Furthermore, Szkornik (2009) proposed to consider errors as much as twice larger than currently used for reconstructions based on diatoms. We, therefore, expected that when comparing Models 1 and 2 based on percent and concentration data respectively, we should obtain errors significantly smaller using Model 1 after reducing the noise of the data set. However, both models provided very similar results using either jack-knife (1% difference) or bootstrapped (3% difference) cross-validation. In order to fully assess their performance, we semi-randomly removed twelve samples from the data set to use it as an independent test data set. To our knowledge, this validation has never been attempted before in sea-level studies. The first direct outcome was the reduction of the transfer function performance between 7 and 9%. Figure 3 summarizes the results of the reconstruction of the test data set *versus* the observed elevation. Results derived from both models were identical, where ten of twelve samples fell within the bootstrapped errors.

Table 3
Statistics summary of the performance of Weighted Averaging-Partial Least Squares (WA-PLS) for the foraminiferal assemblages from the Minho–Lima marshes. Model performance, jack-knife cross-validation, and fall-test set results are presented. Root-Mean Square of the Error Prediction (RMSEP) and maximum bias of cross-validation are also presented as % of the gradient analyzed to allow comparison between the performances of different models. Fall-test results are presented in meters.

		Model performance			Cross-validation						Independent data set	
					Jack-knife			Bootstraping				
Model name	Model	RMSE	r^2	Maximum bias	r^2_{jack}	Maximum $bias_{jack}$	RMSEP	r^2_{boot}	Maximum $bias_{boot}$	RMSEP	r^2	RMSE
Model 1 Standardized data (full data set)	WA-PLS Component 2 (SWLI)	7.19	0.88	10.74	0.81	11.84	9.23	0.82	10.74	10.08		
Model 2 Concentration data (full data set)	WA-PLS Component 2 (SWLI)	7.47	0.87	9.12	0.80	12.75	9.35	0.81	12.26	10.39		
Model 1b Standardized data (partial data set)	WA-PLS Component 2 (SWLI)	7.09	0.89	10.47	0.77	10.44	10.18	0.78	11.12	11.18	0.91	6.8
Model 2b Concentration data (partial data set)	WA-PLS Component 2 (SWLI)	7.61	0.87	8.69	0.78	11.29	9.99	0.78	11.60	11.32	0.90	7.0

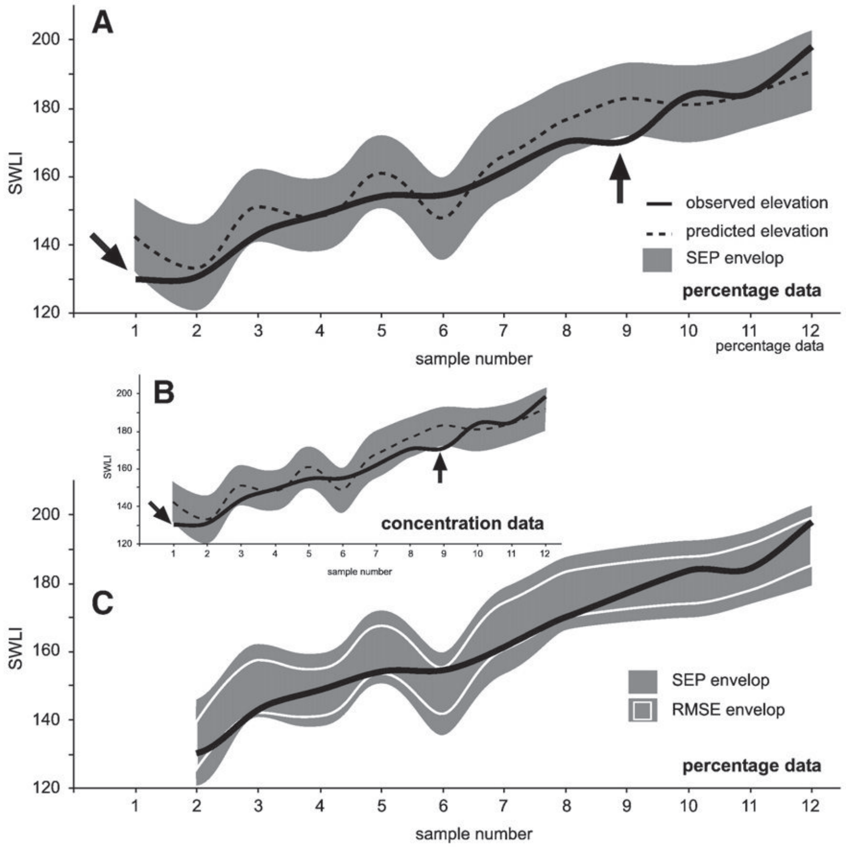

Figure 3. A: Elevational profile of training data set (continuous line) plotted against reconstructed elevations (dashed line) with associated errors (grey envelope) derived from Model 1b (percent data). Arrows indicate the only two samples unable to provide reconstructions within the error range. B: Elevational profile of training data set (continuous line) plotted against reconstructed elevations (dashed line) with associated errors (grey envelope) derived from Model 2b (concentration data). Note the great similarity with Figure 3A. C: Elevational profile of training data set (continuous line) plotted against reconstructed elevations (dashed line) with associated errors (RMSE; grey envelope with white border) derived from Model 1b (percent data) after removing the samples not able to provide reconstructions within the error range as indicated by the arrows in Figures 3A and B. The bootstrapped error it is also indicated (SEP) for comparison.

Samples 1 and 9, however, fall outside the error range. Sample 1 is located in the lower end of the elevations considered in our Model and this strongly limits the predictive ability. Basically, it is related to the adequacy of these models to reconstruct sample elevation at the edges of the sampling area (see Hamilton and Shennan, 2005).

In the case of sample 9, the offset is associated with the unusual presence of *Quinqueloculina* spp. and *C. lobatulus* in high marsh. Since this is a modern sample, we related this anomaly with the current presence of a sand treatment plant nearby that might release some of the treated sands that are deposited in the marsh surface artificially. Furthermore, Leorri et al. (2008) rejected the reconstructions produced in a core that presented calcareous foraminifera due to the significant increase of the reconstruction errors. We, therefore, removed this sample from the independent training set in Figure 3C

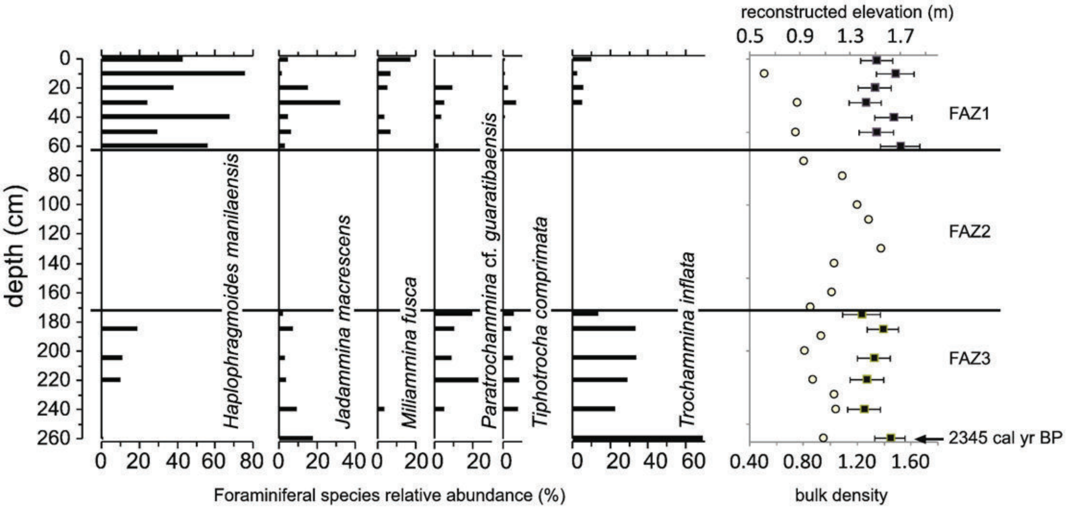

Figure 4. Relative abundance of main foraminiferal species, predicted paleoelevation produced by the transfer function, bulk density (dots), foraminiferal assemblage zones (FAZs) and radiocarbon dating of the uppermost section of borehole C.

since fossil samples including these taxa should be avoided in this type of studies.

However, the poor performance could also respond to the lack of adequacy in the training data set. In the case of cored samples, it could be a response to either, an environmental change or taphonomical problems (see further discussion).

The results obtained strongly support the use of standardized data since they do not seem to affect model performance significantly. Using the independent test data set we can also explore the effect of bootstrapping on each individual sample. We mentioned previously that the overall performance of the transfer function was affected by reducing its predictive ability *ca.* 10%. This is a result of the larger individual SEP errors that on average increased 10%. We need to understand, however, how these errors are generated. Sample-specific errors can be calculated using two components. The first part (v_{i1}; as noted by Birks et al., 1990) represents the effect that the variability of the taxa has on the inferred environmental parameter for each sample. This part of the error tends to decrease in magnitude as the size of the training set increases, and in samples dominated by taxa that are frequent and abundant in the training set. The second part (v_{i2}) includes the error caused by imperfections in the calibration function and model specification error (see Birks et al., 1990 for further discussion). The second part can be only calculated in the training set. In our analysis, v_{i2} only increased by 6% between Model 1 and Model 1b as a result of removing the 12 samples, while the v_{i1} increased by 19%. More interesting is that average SEP of the independent test data set increased by 13% *versus* average SEP obtained from the same samples when included in Model 1. Considering that v_{i2} cannot be calculated for the independent test data set (or sub-fossil samples) since observed values are not available, this part is calculated as the mean across the training data set (Birks et al., 1990) which only increased by 6%. Therefore, the remaining 7% increase is related to an increase of 36% in v_{i1}, associated with variability of the taxon parameters. This increase is apparently too large if we consider that ten of twelve samples from the independent test data set are within ±6.8 SWLIs *versus* the ±11.3 SWLIs average calculated from bootstrapped methods (Fig. 3C). Although we are aware that the independent test data set is small, we are limited by the possible sampling areas. As most transfer functions for sea-level studies are limited to 30–50 samples, we consider that these results are really promising, showing that bootstrapped errors provide a high confident range that overcomes the possible limitations of using percent data and elevation instead of flooding frequency, and that the matrix closure does not affect the performance of these models. On this basis, we can assume that SEP is a conservative error.

On the other hand, these results can help to identify suitable sampling strategies for sea-level studies. Considering that a large part of the error is associated with variability of the taxon parameters and tends to decrease with increasing training data set size, large training sets restricted to the highest high marsh where the number and variability of species is in the minimum should provide a smaller SEP error. This effect is also apparent in the Sado estuary where Leorri et al. (2010) produced a transfer function based on 20 samples from a single transect limited to elevations above MHW. Only eight species were present with an abundance of >2% and only two species were above 10%. Besides the low number of samples studied, jack-knife errors were very small (6.27 in SWLIs units) and it could be anticipated that increasing the number of samples would reduce the error even more.

The precision of the transfer function obtained here, expressed as a percentage of the range of the modern environmental gradient sampled, is comparable to other foraminifera-based transfer functions developed from the northern Atlantic Ocean. These models have a precision that ranges between ±11.5% and ±12.6%. These values are in the low range of error compared with several transfer function approaches (e.g., bottom-water summer-salinity, pH, etc.; see Sejrup et al., 2004) including other sea-level transfer functions (see Leorri et al., 2008) which typically range between 8 and 20% of the gradient analyzed. Although some transfer functions perform better [e.g., ~4% for bottom waters summer temperature (Sejrup et al., 2004) and ~5% for tide level (Massey et al., 2006)], in the case of estuarine transfer functions for sea level, the improvement of the performance responds to the inclusion in the models of lower elevation samples which significantly reduce their reconstruction capabilities (Gehrels, 2000; Woodroffe, 2009). When the errors are presented as height, our results (*ca.* ±0.12 m) are also in the lower range of error compared with other foraminifera-based transfer functions for sea level. Errors of *ca.* ±0.10 m have been reported from the southwestern European Atlantic coast (Leorri and Cearreta, 2008; Leorri et al., 2010), and errors ranging from ±0.12 m (in SWLI units; Horton et al., 1999; Horton and Edwards, 2006), between ±0.18 and ±0.29 m (Gehrels et al., 2001) to ±0.29 m (Massey et al., 2006) have been reported in other areas from the northeastern Atlantic Ocean. On the northwestern Atlantic Ocean the errors reported ranged from ±0.18 m to ±0.25 m (Edwards et al., 2004 and Gehrels, 2000, respectively). Smaller errors (*ca.* ±0.05 m) have been only reported from microtidal areas (e.g., Gehrels et al., 2005; Southall et al., 2006; Kemp et al., 2009).

To illustrate fully the potential application of the transfer function technique, we used the transfer function to calibrate the cored samples. The transfer function provided estimates around MHW (Fig. 4) as expected, since the dominant species have their optima in the higher elevations (Fig. 2C). Estimations were between 166 and 191 SWLIs and fall well within the sampled elevational range with reconstructing errors below 15 cm. These results indicate that high resolution sea-level studies could be performed in this area and that the transfer function technique is a suitable methodology, at least in this area. We cannot, however, attempt to reconstruct sea-level changes from this core due to a very poor chronology and a high degree of sediment compaction that would significantly affect the reconstruction (Fig. 4).

This core reconstruction provides some additional and significant information. Within this core there are two species with a statistically significant presence in relation to the training data set. Those are *P.* cf. *guaratibaensis and M. fusca*. In the training data set, *P.* cf. *guaratibaensis* is secondary (<9% in any single sample) and associated to *M. fusca* and *T. inflata* at high elevations. This is similar to the findings reported by Debenay et al. (2002) who reported this assemblage at the transition to the dry fields and in agreement with the general distribution of these species (e.g., Hayward and Hollis, 1994). In two core samples, however, reaches values *ca.* 20%. This could affect the transfer function performance providing unreliable reconstructions. In order to assess that, we artificially manipulated the test data set increasing *P.* cf. *guaratibaensis* values (that range from 0 to 8.5%) to 20% for every sample and performed the reconstruction again. The reconstructed values presented no significant differences when compared to the original reconstruction using Model 1b. This might suggest that this particular species has not strong influence in sea-level reconstructions and the signal is carried by the remaining species.

On the other hand, *M. fusca* values decrease downcore, becoming a secondary species while it is highly dominant in the training data set. This could represent the current environment where the core was recovered, within the Zone I (see Table 2 and Fig. 2). This Zone I presents an average value of 5% of *M. fusca,* similar to the results found downcore. However, it cannot be fully discarded a possible taphonomical loss. In fact, *M. fusca* has been suggested to be susceptible to taphonomic degradation (Goldstein et al., 1995; Hippensteel et al., 2000). In order to assess the possible taphonomical effect in the transfer function performance, we followed a similar approach to that indicated previously. We artificially modified the relative values of *M. fusca* in the test data set. If we completely remove *M. fusca* from the test data set, five sample reconstructions fell outside the error ranges. These samples are associated with *M. fusca* values of >30%. Moreover, the transfer function performance will be significantly affected when we reduced *M. fusca* values between 25 and 38%. Considering the fact that we aimed the highest marsh elevation where *M. fusca* is less dominant (mean value 5%) and that high-resolution sea-level studies will be restricted to the uppermost 0.5 to 1 m, we expect taphonomical problems to be minimized.

Conclusions

This study was aimed to assess the performance of transfer functions for sea-level studies in marsh sediments using salt marsh foraminifera. The analysis performed here indicates that elevational and sampling errors, the loss of information by using height as "predictor" variable and the limitations that compositional data sets present are well captured by cross-validation methods. Furthermore, reconstructions are very precise (observed error values are below 7 cm) and RMSEP are conservative values compared with the difference between the predicted and the observed value. This fact, confirms the usefulness of salt-marsh foraminifera as sea-level proxies in the Atlantic Iberian coast and the temporal extent of these environments back in time up to at least *ca.* 2500 years. Although salinity is one of the most important environmental factors controlling the foraminiferal distribution at regional level, tidal flooding (and, hence, height above local mean tide level) is the main control in the marsh environment. In fact, foraminifera-based transfer functions offer a quantitative and robust methodology to reconstruct former sea levels from salt-marsh sediments.

The strong correlation of the agglutinated salt-marsh foraminifera assemblages with elevation above mean tidal level has been successfully used to reconstruct cored samples. Furthermore, our findings also suggest that taphonomic loss could affect downcore reconstructions if the depositional environment is dominated by delicate taxa such as *M. fusca*.

Therefore, it is suggested that modern transects should be collected from the same environment aimed to be reconstructed, including *ca.* 50 samples, and focusing mainly in high marsh areas dominated by few species that are also more resistant to degradation and less prone to be biased.

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