

Isotopic Variations of Oxygen ($\delta^{18}\text{O}$) in Benthic Foraminifera under Anti-estuarine Conditions in the Colorado River Delta

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Abstract

Benthic foraminifera are excellent environmental indicators; CaCO_3 test records the isotopic concentration of their surroundings and can be used to analyze environmental changes that occur during a certain time period. Stable isotopes, particularly those of oxygen ($\delta^{18}\text{O}$), are useful for interpreting ancient environments, given that they are used as “proxy” environmental variables (temperature and salinity). In this study, we provide ranges of isotopic variation in benthic foraminifera communities from the Colorado River delta. Four sampling campaigns were conducted in one year (2009-2010) in the adjacent subtidal zone of Baja California. Four transects with a total of sixteen sample stations were drawn perpendicular to the coastline. Here, we recorded the following *in situ*: sand-type, salinity, and temperature. In the laboratory, 300 individuals per sample were separated, and sub-samples were taken for isotopic analysis. Data was processed using the software’s R 2.12.2, PAST 1.81 and Arc Map 9.3. Forty species were identified in the dead assemblages (Thanatocoenosis), whereas thirteen species were found in the living assemblages (Biocoenosis). The most abundant species in both communities belong to the genera *Ammonia* and *Cibicides*. In the living assemblages, isotopic variation -2.15% to 5.94% within a temperature interval of 11°C , indicated anti-estuarine conditions. In the dead assemblages, isotopic composition -3.04 to -0.74 served as a sign of estuarine conditions prior to damming.

Keywords

Stable Isotopes, Living Assemblages, Dead Assemblages, Upper Gulf of California

1. Introduction

Historically, deltas have played a significant role in human history. The combined presence of fertile soil, water, and river travel favored the development of agriculture and commerce. With the discovery of new energy sources, the economic role of deltas has markedly increased, as they develop in sedimentary basins that favor deposition, maturation, and retention of hydrocarbons. The economic importance of deltas has led to an increase of many geological studies over the past 30 years, favoring a better understanding of sedimentary and stratigraphic processes [1]. These studies have also revealed a noticeable anthropogenic impact, which in some cases have caused an appreciable transformation of the ecosystem: such is the case of the Colorado River delta.

The Colorado River extends for approximately 2334 km from its source in the Rocky Mountains in Colorado, USA to its mouth in the Gulf of California. The delta is located at 31°37' and 31°45' north and 114°10' and 114°52' west (**Figure 1**). Its drainage basin comprises an approximate area of 644,000 km² [2]. Before dam construction, waters at the river mouth would be enriched with abundant suspended material rich in nutrients, giving rise to high benthic activity. This can be demonstrated by the millions of shells that compose the mountain ridges or cheniers located throughout the coast of Baja California [3] [4]. The construction of the Hoover Dam in 1935 and the Glenn Canyon Dam in 1963 achieved ultimate control of the river waters thereby causing obstruction of the sediment supply, and erosion of the delta [5] [6] [7] [8]. In parallel, during and after dam construction throughout the riverbed of the Colorado, decrease of freshwater discharge to the Gulf of California caused drastic environmental changes that intensified after 1970 when the discharges to the gulf were almost completely cut off [9]. Subsequently, the river has flowed towards Mexico only during extraordinarily rainy seasons when the storage limits of the dams are exceeded [10].

The most noticeable ecological impact resulting from the absence of freshwater is the radical transformation of the former estuary into an anti-estuary environment [11] [12]. These authors reported a previously salinity gradient of 12% to 34%, and currently, this gradient can reach up to 36.8% [7] [11], who studied the sediments from the Colorado River delta, confirmed the antiestuarine conditions of the rivers estuary. These antiestuarine conditions are most accentuated in the present day, given that the salinity values can reach up to 48% [13] [14]. As a consequence of environmental deterioration, various species of flora and fauna are seriously threatened and/or in danger of extinction, including the totoaba (*Totoaba macdonaldi*), the vaquita (*Phocoena sinus*), and the Yuma clapper rail (*Rallus longirostris yumanensis*). The Colorado Delta clam (*Mulinia coloradoensis*) is the one species that most dramatically records the magnitude of this impact [15], given that only small populations currently exist in the vicinity of the river mouth when in the recent past they populated the entire area of the delta [16].

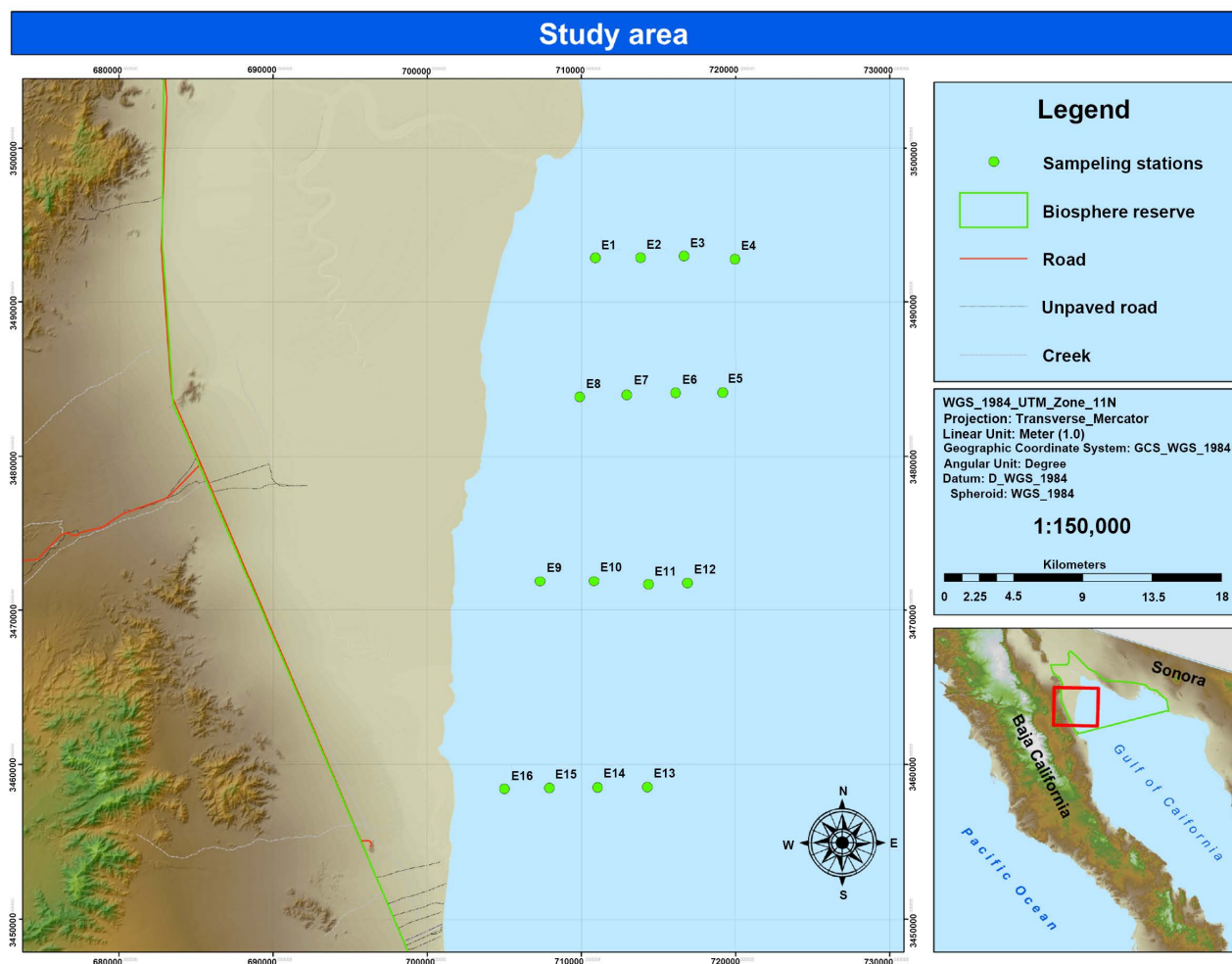


Figure 1. Location of the study area, showing sampling points.

The use of stable isotopes, particularly oxygen isotopes ($\delta^{18}\text{O}$), have been very useful for characterizing and interpreting ancient environments, as they can be used as a proxy for environmental variables such as temperature and salinity [17] [18]. Previous studies of estuarine mollusk shells from the Colorado delta have demonstrated that based on the range of $\delta^{18}\text{O}$ inferred by the comparison of a present-day shell with an ancient one, the $[\delta^{18}\text{O}]$ value tends to be less than -2.5% with more freshwater entering the mixing zone, and the opposite occurring when fresh water input diminishes (*i.e.*, $[\delta^{18}\text{O}]$ values are more positive with less freshwater dilution and higher marine conditions [15]). This finding has also been observed in the bivalve *Protothaca grata* in prehistoric archaeological records that researchers found in a shell midden located at the southern boundary of the Colorado River delta, approximately 65 km from its mouth, showing values between -3.44% and -3.99% , indicating estuarine conditions [19].

An alternative method for independent analysis is benthic foraminifera. They are distributed along, approximately, one-third of the ocean floor, including the adjacent coastal zone, and through their calcium carbonate shells that maintain equilibrium with the physicochemical conditions of the surrounding water;

benthic foraminifera permit the reconstruction of the environmental conditions in which they lived. Other advantages are their sensitivity to environmental changes, and only a small sample size is required.

Little is known about the benthic foraminifera ecology in the Upper Gulf of California. One of the first studies conducted in this field was [20] [21], who studied the intertidal foraminifera. In his study, a low diversity and similarity in the associations between present organisms and fossils were found, providing an indication of similar environmental conditions. [22] found the isotopic composition of fossil foraminifera shells showed significantly more pronounced estuarine conditions during the Pleistocene Epoch where the boundaries of the delta are currently located. [23] compared the isotopic compositions of living and dead assemblages. She found that in the latter, there was a persistent isotopic signal caused by the presence of freshwater in the river delta area due to temporal averaging.

Previous studies (*i.e.* [24]), have been restricted to the intertidal zone. As a result, the composition, structure, and ecology of the benthic foraminifera are unknown. In this study we focus on the analysis of spatial and temporal variations in the isotopic composition of oxygen ($\delta^{18}\text{O}$) in the tests of living and dead assemblages under current antiestuarine conditions. Our goal is to relate $\delta^{18}\text{O}$ values with environmental changes resulting from ecosystem disturbance and to establish a reference marker for future studies of the ecology of the Colorado River delta.

2. Methods and Materials

This study was conducted at the Upper Gulf of California Colorado River Delta Biosphere Reserve, a protected natural area in the subtidal zone adjacent to the coast of Baja California (**Figure 1**). This area has historically shown the most pronounced estuarine conditions [10]. Four sampling campaigns were performed in the course of one year (August-November 2009 and March-June 2010) along four transects perpendicular to the coastline in which a total of 16 sampling stations were located. These stations covered an area extending near the southern boundary of the delta (**Figure 1**). Each station was georeferenced using a Garmin e-Trex global positioning system (GPS) device. The information was later used to create distribution maps of the foraminifera in the study area. At each sampling station, the salinity and temperature were measured using a Vee Gee STX-3 refractometer, and a manual thermometer, respectively. Sediment samples were collected with a “Petite Ponar” dredge with a capacity of 2.4 m³ aboard a 6.7 m-long fishing boat. With the dredge, subsamples of the upper 5 cm of the seabed were collected and represent sediments that are constantly mixing. These subsamples were emptied into plastic bags and fixed with 60% ethanol placing them in refrigeration until subsequent laboratory analysis.

The use of Rose Bengal in micropaleontological analysis in the laboratory was used to distinguish live from dead foraminifera (methodology by [25]). After 48

hours, samples are passed through a wet sieve with a mesh size of 62 μm to eliminate silt and clays. Foraminifera tests were concentrated by filtration through No. 4 Whatman paper with a diameter of 11 cm, then dried in an oven at 40°C. After they were sieved, the fractions between 63 and 180 μm were separated because tests were not found in the fractions above 180 μm . Finally, 300 individuals per sample were separated for identification. This number of individuals is considered to be statistically the minimum sample size to obtain reliable data [26]. Taking into account the density of the living assemblages was very low, the 300 individual contained organisms from both living and dead assemblages were analyzed. The identified species were imaged in a scanning electron microscope (JEOL JSM-35C) at the Earth Sciences Division of the Center for Scientific Research and Higher Education of Ensenada (Ciencias de la Tierra del Centro de Investigación Científica y de Educación Superior de Ensenada, CICESE). The foraminifer were mounted on plates and deposited in the paleontology warehouse of the Department of Marine Sciences of the Autonomous University of Baja California (UABC).

For isotopic analysis, tests from *Ammonia* sp. were utilized because its isotopic compositional data is already available for the area under study [23] [27]. To this end, ten foraminifera from each station were separated from both living and dead assemblages. The isotopic composition was analyzed in the Stable Isotopes Laboratory at the University of Arizona, Tucson, USA. The samples were subjected to a vacuum reaction with 100% anhydrous phosphoric acid at 70°C for 2 hours. The analysis was conducted in a Finnigan MAT 252 mass spectrometer equipped with a Kiel-III automated carbonate sampling device. The $\delta^{18}\text{O}$ values are reported with reference to the V-PDB standard.

To determine if significant spatial (transects) or temporal (through the annual cycle) differences exist, in both the living and the dead assemblages, analysis of variance (ANOVA) was conducted on the $\delta^{18}\text{O}$ concentration data using the statistics program R version 2.12.2. The normality and homogeneity of the variances in the data were tested utilizing the Shapiro-Wilks statistical test. If this requirement was not fulfilled, the type II ANOVA test was chosen. Subsequently, to obtain precise in the data interpretation, the a posteriori analysis was performed utilizing Tukey test to determine the most distinct or similar seasons.

3. Results and Discussion

Due to logistical and climatic problems occurrences, only transects II and III were sampled during the summer (**Figure 1**). In the following seasons, all transects were completed and samplings were performed during the following time period: August 22 (summer) and November 13 (fall) of 2009 as well as March 20 (winter) and June 18 (spring) of 2010. The temperatures throughout the cycle oscillated between 20°C and 31°C with a salinity range between 34‰ and 49‰ (**Table 1**).

Table 1. Temperature, salinity and depth recorded during the 2009-2010 cycle.

Sampling point	Salinity (%)	Temperature (°C)	Depth (m)
Summer E5	36	31	13
Summer E6	38	30	12.3
Summer E7	38	31	10.6
Summer E8	37	30.5	10.4
Summer E9	35	31	14.4
Summer E10	37	30.5	11.9
Summer E11	38	31	9.2
Summer E12	38	30.5	6.8
Autumn E1	45	21	11.9
Autumn E2	40	22	10.6
Autumn E3	46	21	8.4
Autumn E4	49	20	7.3
Autumn E5	40	22	13
Autumn E6	42	22	12.3
Autumn E7	40	22	10.6
Autumn E8	47	22	10.4
Autumn E9	42	22	14.4
Autumn E10	40	22	11.9
Autumn E11	42	23	9.2
Autumn E12	42	22	6.8
Autumn E13	44	22	10.7
Autumn E14	40	22	9.1
Autumn E15	42	22	6.7
Autumn E16	44	22	4.1
Winter E1	38	21	11.9
Winter E2	38	20	10.6
Winter E3	36	20.5	8.4
Winter E4	36	20.5	7.3
Winter E5	36	20	13
Winter E6	38	20.5	12.3
Winter E7	36	21	10.6
Winter E8	36	22	10.4
Winter E9	37	21	14.4
Winter E10	36	21	11.9
Winter E11	37	21	9.2
Winter E12	37	20	6.8

Continued

Winter E13	36	21	10.7
Winter E14	37	21	9.1
Winter E15	37	21	6.7
Winter E16	37	20.5	4.1
Spring E1	35	29	11.9
Spring E2	36	29	10.6
Spring E3	36	30	8.4
Spring E4	39	30	7.3
Spring E5	36	28	13
Spring E6	35	29	12.3
Spring E7	35	29	10.6
Spring E8	35	29	10.4
Spring E9	35	28	14.4
Spring E10	35	28	11.9
Spring E11	36	29	9.2
Spring E12	35	28	6.8
Spring E13	35	27.5	10.7
Spring E14	35	28	9.1
Spring E15	34	28	6.7
Spring E16	35	28.5	4.1

3.1. Composition and Abundance

Considering all sampling stations, a total of 40 species were identified, with 14 comprising 95% of the total population in dead assemblages and three comprising more than 50% of the total (*Ammonia* sp; *Criboelphidium* sp; and *Rosalina* sp.; **Figure 2**). In living assemblages, 13 species were identified, with the most abundant being *Ammonia* sp and *Criboelphidium* sp. (**Figure 2**). They comprised 86% of the total individuals (**Table 2**). These organisms have also been reported by [23] in the intertidal zone; however, the number of species reported by this author found a 10% increase in the same subtidal zone. The above indicates the intertidal zone is relatively less diverse than the subtidal zone because the latter has conditions with less environmental severity [28].

The stability-time hypothesis, proposes that communities of benthic organisms can be of two types: 1) “physically controlled communities”, which are exposed to severe physico-chemical changes (hypersaline bays, estuaries, and deserts, being among the most important), and 2) “biologically suited communities”, which appear when the physico-chemical conditions are uniform over a long period of time, such that biological interactions and their diversity increase. Based on the above, the subtidal zone, being more stable compared with the intertidal zone, is one of the explanations for the greater relative diversity that was

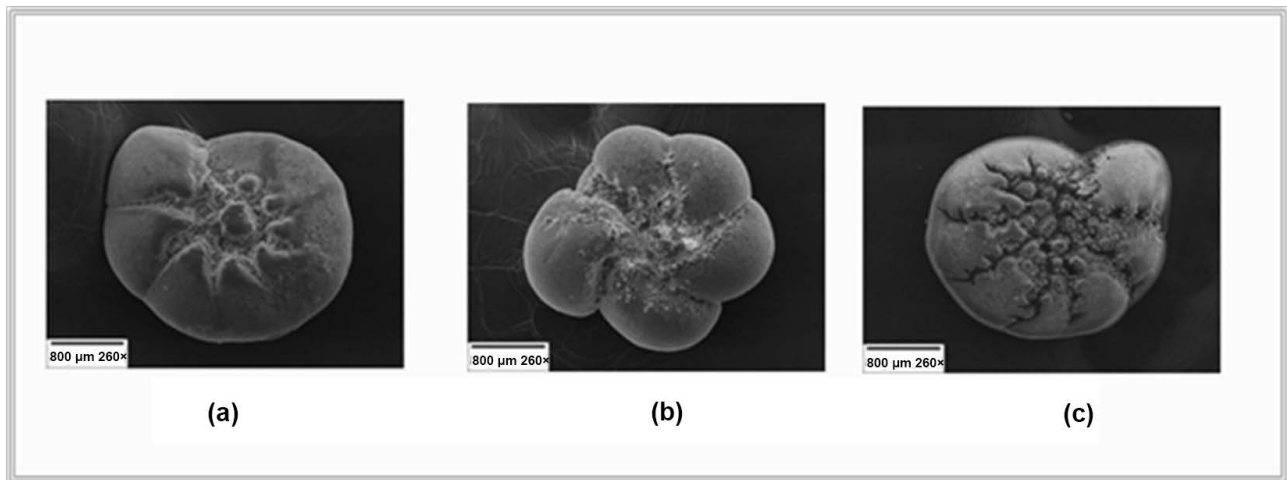


Figure 2. Most abundant species in the Upper Gulf of California. (a) *Ammonia beccarii parkinsoniana*, (b) *Rosalina* sp. and (c) *Criboelphidium excavatum*. Biocenosis (a), (b) Thanatocoenosis (a), (b), (c).

Table 2. List of the most abundant species of foraminifera found in the cycle from 2009 to 2010.

Specie	Relative Abundance in Thanatocoenosis (%)	Number of Individuals	Relative Abundance in Biocenosis (%)	Number of Individuals
<i>Ammonia beccarii parkinsoniana</i> (d'Orbigny, 1826)	28.19	5653	64.5	676
<i>Criboelphidium excavatum</i> (Terquem, 1876)	15.99	3206	21.6	226
<i>Rosalina</i> sp 1 (d'Orbigny, 1826)	14.22	2851	2.3	24
<i>Bucella tenerrima</i> (Bandy, 1950)	6.47	1297	2.4	25
<i>Criboelphidium spinatum</i> var. <i>translucens</i> (Cushman and Brönnimann 1948)	6.36	1276	X	X
<i>Ammonia</i> sp (Linné, 1758)	6.13	1230	X	X
<i>Criboelphidium gunteri</i> (Cushman and Brönnimann 1948)	4.58	919	X	X
<i>Criboelphidium incertum</i> (Cushman and Brönnimann 1948)	3.99	800	X	X
<i>Criboelphidium poeyanum</i> (Cushman and Brönnimann 1948)	2.21	443	X	X
<i>Quinqueloculina</i> sp 1 (d'Orbigny, 1826)	2.09	420	2.3	24
<i>Bolivina</i> (d'Orbigny, 1839)	X	X	1.1	12
<i>Bulimina marginata</i> (d'Orbigny, 1826)	1.37	274	1.7	18
<i>Quinqueloculina</i> sp 3 (d'Orbigny, 1826)	1.12	224	X	X
<i>Nouria</i> (Heron-Allen y Earland, 1914)	X	X	1.6	17
<i>Quinqueloculina</i> sp 2 (d'Orbigny, 1826)	1.08	217	X	X
<i>Miliolinella</i> (Wiesner, 1931)	X	X	0.8	8
<i>Buliminella elegantissima</i> (d'Orbigny, 1826)	1.02	205	0.7	7
<i>Pseudomasselina</i> (Lacroix, 1938)	X	X	0.5	5
<i>Fissurina</i> (Reuss, 1850)	X	X	0.3	3
<i>Reusella</i> (Galloway, 1933)	X	X	0.3	3
<i>Ammoscalaria pseudospiralis</i> (Williamson, 1858)	0.76	152	X	X

observed [28] [29]. Although the environmental severity is still considerable given the current hypersaline conditions, as discussed above, the lack of water supply from the Colorado River has led to a change in the salinity of the study area. This change, in turn, may have resulted in modification of the diversity of the benthic foraminifera species, which live in the Upper Gulf of California.

3.2. Isotopic Composition

Living assemblages—the variability in the $[\delta^{18}\text{O}]$ composition of the foraminifera exhibited two patterns: 1) Winter-Spring, characterized by negative values; and 2) Summer-Fall, in which positive values predominate (**Figure 3**). The variation range during the yearly cycle was 8.09%, with values from -2.15% up to 5.94% (**Figure 3**), which is to be expected given that the area is currently influenced by strong variations in the physico-chemical conditions, principally temperature and salinity. Given that currently there is no freshwater supply to the delta, these isotopic variations can be attributed exclusively to these two variables. The high positive values can be attributed to the fact that the delta is currently an evaporation basin. Therefore, the variation range of the isotopic values indicate, for the benthic foraminifera in the study area, that a change of 11°C corresponds to a variation of 8.09% in the oxygen isotopic concentration, with a minimum negative value of -2.15% , obtained exclusively from the temperature and salinity effect under the current no-flow conditions. Therefore, more negative isotopic values in subtidal benthic foraminifera tests would indicate the supply and/or dilution of isotopically lighter water, such as from the Colorado River.

[15] reported values of 0.79% to -2.48% for the bivalve mollusk *Chione fluctifraga* in the Colorado River delta in the absence of freshwater flow and values of -1% to -6% for fossil shells of the bivalve mollusk *Mulinia coloradoensis*, which live in estuarine conditions when diluted waters of the river extended up to 65 km from the mouth of the river. Outside this geographic range, [19]

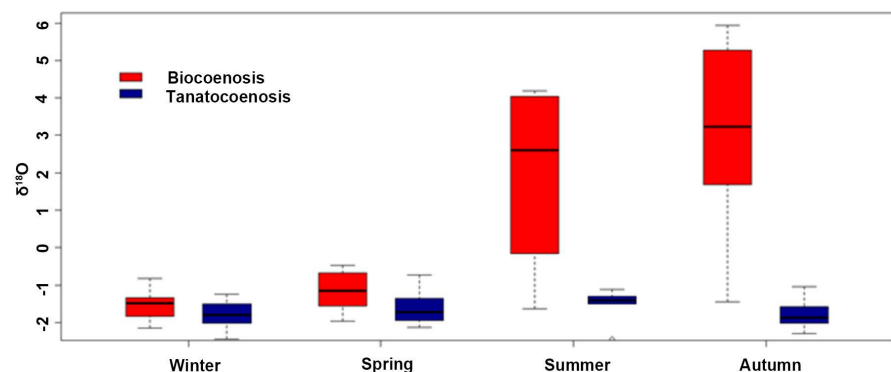


Figure 3. Ranges of variation of the oxygen Isotope composition ($\delta^{18}\text{O}$) in the living and dead benthic foraminifera across the Colorado Delta area during the annual cycle from 2009 to 2010. Which notes the box containing the quantiles of 75% and 25%; the average center line, and the extreme values of 5% and 95%.

reported a minimum value of -2.37% in shells from the bivalve mollusk *Protothaca grata* in an archaeological shell midden in Campo Cristina, approximately 60 km to the south of the delta boundary. The aforementioned findings suggest that isotopic variation ranges related by the changes in temperature are very similar among members of the same taxonomic group. Our data concerning benthic foraminifera exhibit a wider variation range than those reported for bivalves living under the same environmental conditions. These differences can be attributed to the vital effect observed in organisms of different taxa, which respond slightly differently to the same environmental conditions, given that although initially, at the time they form their tests or shells, both organisms fix the oxygen that is found in the surrounding water. The fixation process can differ in organisms from different taxa.

Moreover, [23] reported values of -4.1% in living assemblages of intertidal benthic foraminifera during the winter season on Zacatosa Island, which is located at the mouth of the Colorado River. The author also related this value to possible discharges of freshwater from agricultural drainage from the Valley of Mexicali, and its proximity to the mouth of the river. [30] studied dimorphism in benthic foraminifera from deep waters in the Pescadero basin and reported an isotopic variation between 1.586% and 2.450% in a range from 6°C to 9°C , *i.e.*, 0.864% over 3°C , which implies 0.288% per $^{\circ}\text{C}$. Based upon this work, in the subtidal delta benthic foraminifera, the isotopic variation was 8.09% within a range of 11°C , which corresponds to 0.735% per $^{\circ}\text{C}$. The difference, as previously mentioned, could be due to this vital effect. Despite organisms from the same taxonomic group, they respond differently. The environmental conditions to which they are exposed are completely different from individuals studied by [30] in deep waters, which are at a much lower temperature than that observed in our case in the subtidal zone where the maximum recorded temperature was 31°C .

By applying type II ANOVA to the isotopic data, the data for different seasons was found to be significantly different (Table 3). However, Tukey's test revealed that the Spring-Winter and Summer-Fall seasons were statistically equal (Table 4) and that another possible combination of seasons would also be significantly different, such as would be the case for Fall-Winter, Summer-Winter, Spring-Fall, or Summer-Spring. Dead assemblages—in contrast to living assemblages, the dead assemblages did not display significant differences in the isotopic composition throughout the year (Figure 3). However, in all stations, the recorded values were consistently negative, resembling more the Winter-Spring conditions of the living assemblages and with even more negative values than those obtained. Because dead assemblages are the accumulation of dead foraminifera shells over the course of time and considering that while performing the analysis to identify communities of living/dead relationship organisms, the unstained (Rose Bengal) adult individuals were separated. All the foraminifera that were separated for analysis had more negative isotopic values than those found in the living assemblages. Thus, taking as a basis the works performed by other researchers in which the isotopic variation ranges are established for the

Table 3. Analysis of variance for biocoenosis in which all simple are compared year over the annual cycle.

Analysis of Variance (ANOVA type II)				
Source of variation	SS	Gl	F	Pr(>F)
Season	158.304	3	24.353	8.77E-09
Residual	78.004	36		

Table 4. Tukey tests for biocoenosis where you can see the difference between each of the seasons analyzed, obtaining that all winter and spring seasons like summer and autumn are statistically equal, while the combination of the other reflects differences between them.

Multiple comparison of Means: Tukey tests					
Season	Std. Estimation	Error	Z value	Pr(> Z)	SC
Autumn – Winter = 0	4.6308	0.6192	7.479	<0.001	***
Spring – Winter = 0	0.3838	0.5774	0.665	0.90826	
Summer – Winter = 0	3.4688	0.8416	4.122	<0.001	***
Spring – Autumn = 0	-4.247	0.6192	-6.859	<0.001	***
Summer – Autumn = 0	-1.162	0.8708	-1.334	0.53446	
Summer – Spring = 0	3.085	0.8416	3.665	0.00123	**

present-day conditions and for the conditions when the waters of the Colorado River freely reached the delta, the dead assemblages are expected to show an isotopic signal of estuarine conditions or of the isotopically lighter water flow that dominated prior to the damming of the river.

The accumulation of generations or temporal averaging represents the average environmental conditions after several generations, which in our case allows us to infer that the environmental conditions in the years prior to the damming of the river were dominated by isotopically lighter water than that of the present day. This finding can be corroborated with various works in the field study of bivalve mollusks, in which the oxygen isotopic ranges have been established before and after the construction of the dams on the upper part of the Colorado River [19] [15] [31].

[23] reported values for isotopic variation in the dead assemblages of intertidal benthic foraminifera from -3.55% to -5.5%, which can be interpreted as indicative of estuarine conditions all the way to “El Faro” beach to the south of San Felipe. However, he attributes to these values a temporal averaging possibly influenced by the mix of fossil foraminifera from a Pleistocene coastal shelf, a site where the delta area extended when the environmental conditions were wetter, and the contributions from the greater river, as suggested by the values of up to -7.36% in fossil foraminifera tests reported by [22].

By comparing the isotopic concentrations for the entire annual cycle for both living and dead assemblages (Figure 3), the isotopic variation can be observed to

range from -0.74% to -3.04% in the dead assemblages, as we have previously discussed. Considering that in the living assemblages the most negative value was -2.15% , the value in our case study represents the boundary of changes exclusively influenced by changes in temperature and salinity. Therefore, the values reported in this work for the dead assemblages are an indicator of the conditions that dominated in the study area before damming the river. Furthermore, based on the work of [19] and [23], who reported values of -3.44% to -3.99% and -3.55% to 5.5% , respectively, we can state that the values reported in this work for the dead assemblages represent, as in the above work, an environment dominated by isotopically lighter waters than the current conditions. In addition, given that we have the record of when the flow of freshwater into the delta began to be blocked, we can state that the isotope concentrations found in the dead assemblages indicate estuarine conditions or isotopically lighter water flows.

The isotopic variations in the living and dead assemblages for each station and season (**Figure 4(a)** and **Figure 4(b)**) demonstrate the existing relationship between temperature and the oxygen isotope concentration. In the living assemblages (**Figure 4(a)**), a higher temperature is expected to be associated with a higher $\delta^{18}\text{O}$ concentration due to the relationship between temperature and evaporation, for which values of up to 0.9 m/year have been reported for the northern end of the Gulf of California [32].

However, by comparing the oxygen isotope concentration with salinity (**Figure 5(a)**), one can see that the relationship between the two variables is much closer than that observed with temperature. This correlation proves, as has already been mentioned, that the higher the temperature, the greater the evaporation, which causes the salinity to increase, although the location of the study area in an evaporation basin must be considered. Therefore, when the temperature increases, evaporation of the light isotope ($\delta^{16}\text{O}$) occurs in turn, which will be eliminated, resulting in an enrichment of the heavy isotope ($\delta^{18}\text{O}$). Simultaneously, this same isotope will be fixed by the foraminifera in their CaCO_3 tests [8].

Nevertheless, the isotopic concentration was observed to vary quite broadly for the same temperature (**Figure 6(a)**), which could be explained by the optimal growth of the analyzed species (*Ammonia* sp.), which lies in a temperature range from 24°C to 30°C [33]. For this reason and given that in the sample area temperatures between 20°C and 31°C were recorded and *Ammonia* sp. would be expected to grow practically throughout the entire year. Therefore, in the living assemblages, what is being recorded is an average of the temperatures and salinities throughout its annual cycle.

It is important to consider that to obtain the isotopic composition, tests were performed without discriminating between juveniles and adults, and based on the results obtained in the isotopic variations, juvenile organisms may represent a better indicator as their tests, unlike those of the adults, have less exposure time to the changes occurring in the surrounding water and are thus useful for obtaining more precise data. In addition, salinity showed a higher statistical

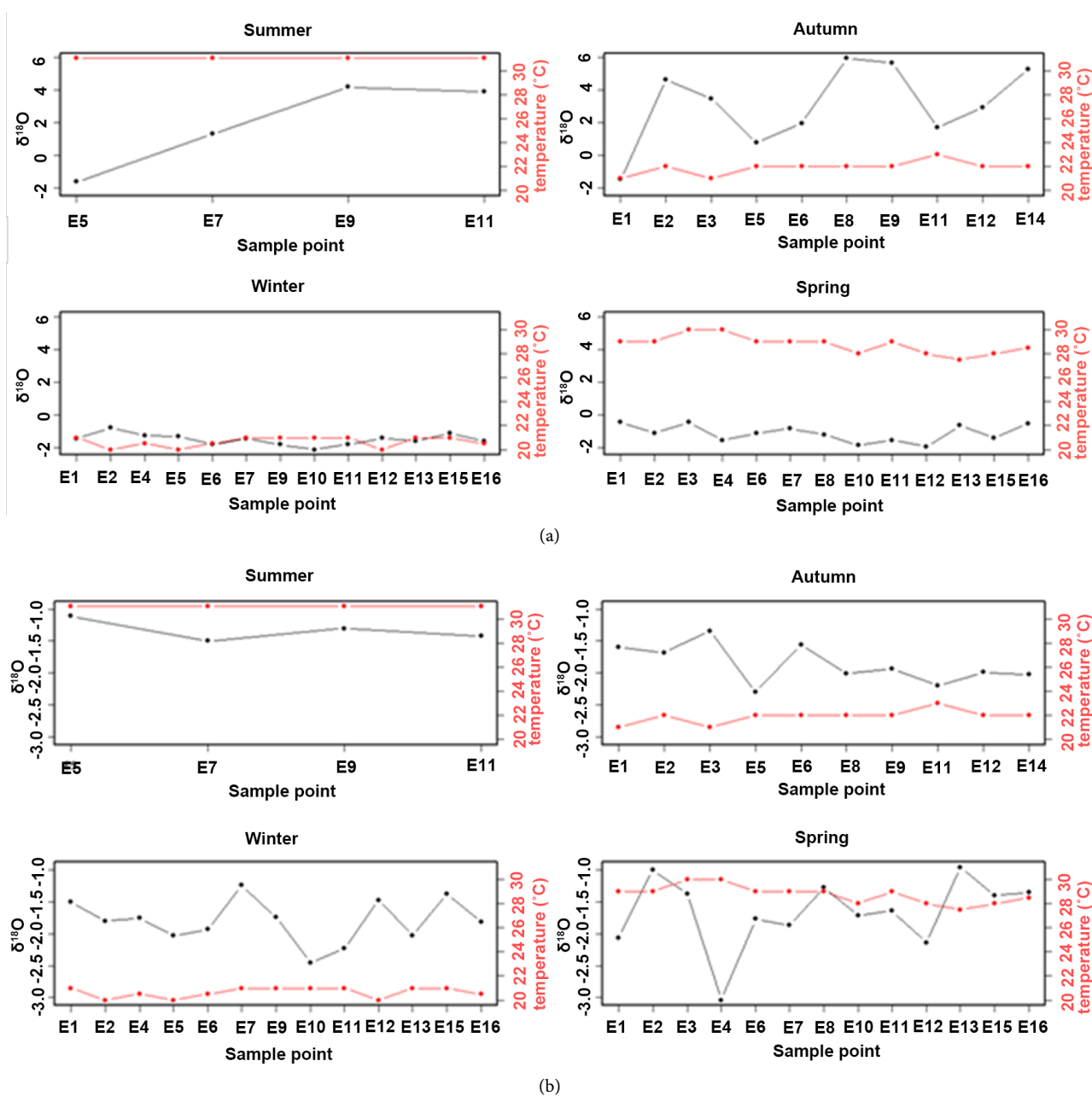
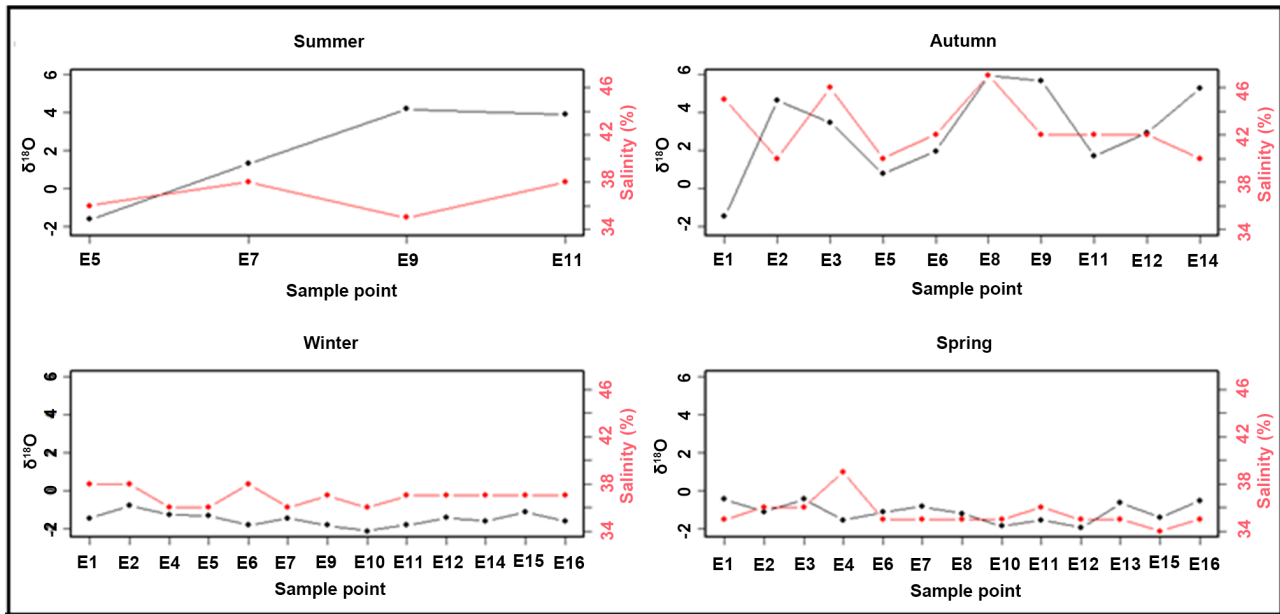
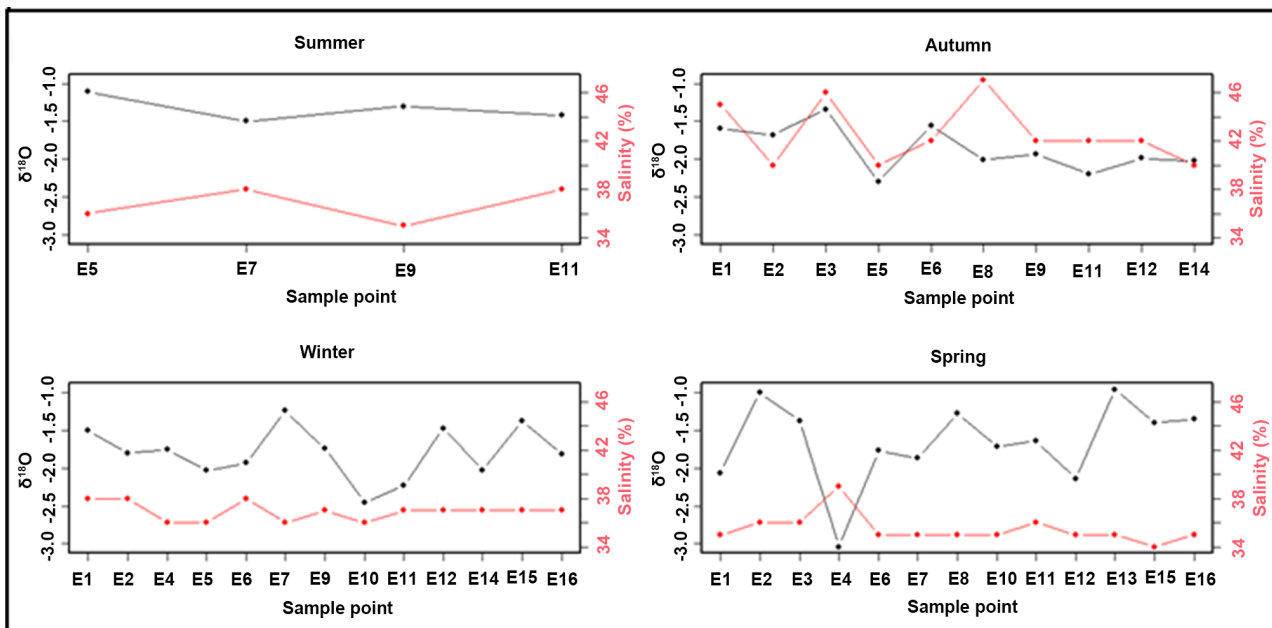


Figure 4. (a) and (b) distribution of temperature and oxygen isotopic composition ($\delta^{18}\text{O}$) of biocoenosis (a) and Thanatocoenosis (b) for the area sampled. The stations correspond to those shown in **Figure 1**. (Not all of the features of sampling stations, because it showed low values of voltage, which does not reliable for proper data interpretation).

correlation with the oxygen isotope, with a value of $r = 0.62$ (**Figure 6(b)**). This statistical test was applied to both variables (salinity and temperature) as an additional tool to visualize the relationship exhibited by benthic foraminifera from the Colorado River delta with the environmental conditions in which they live, and likewise, the results prove that the vital effect in organisms is different depending on the taxa to which they belong because individuals subjected to the same environmental rigors but from different taxa respond differently to the environmental conditions that occur in the surrounding water.



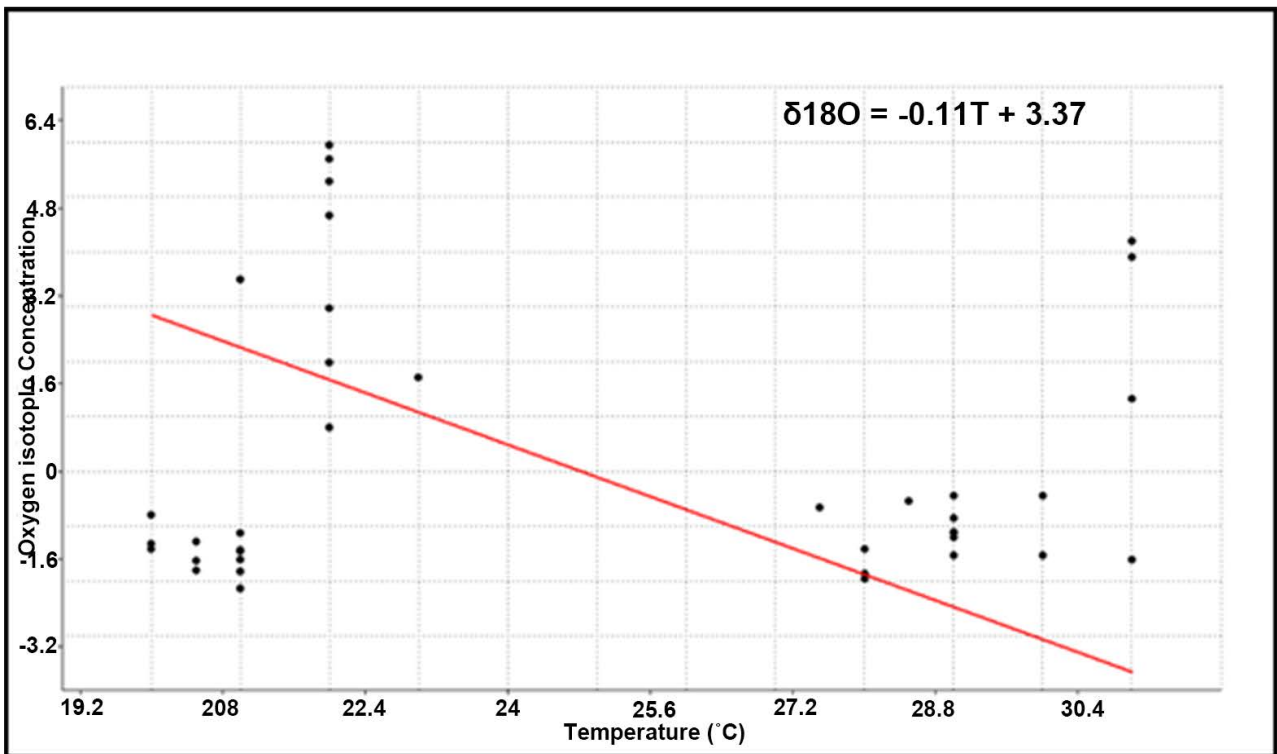
(a)



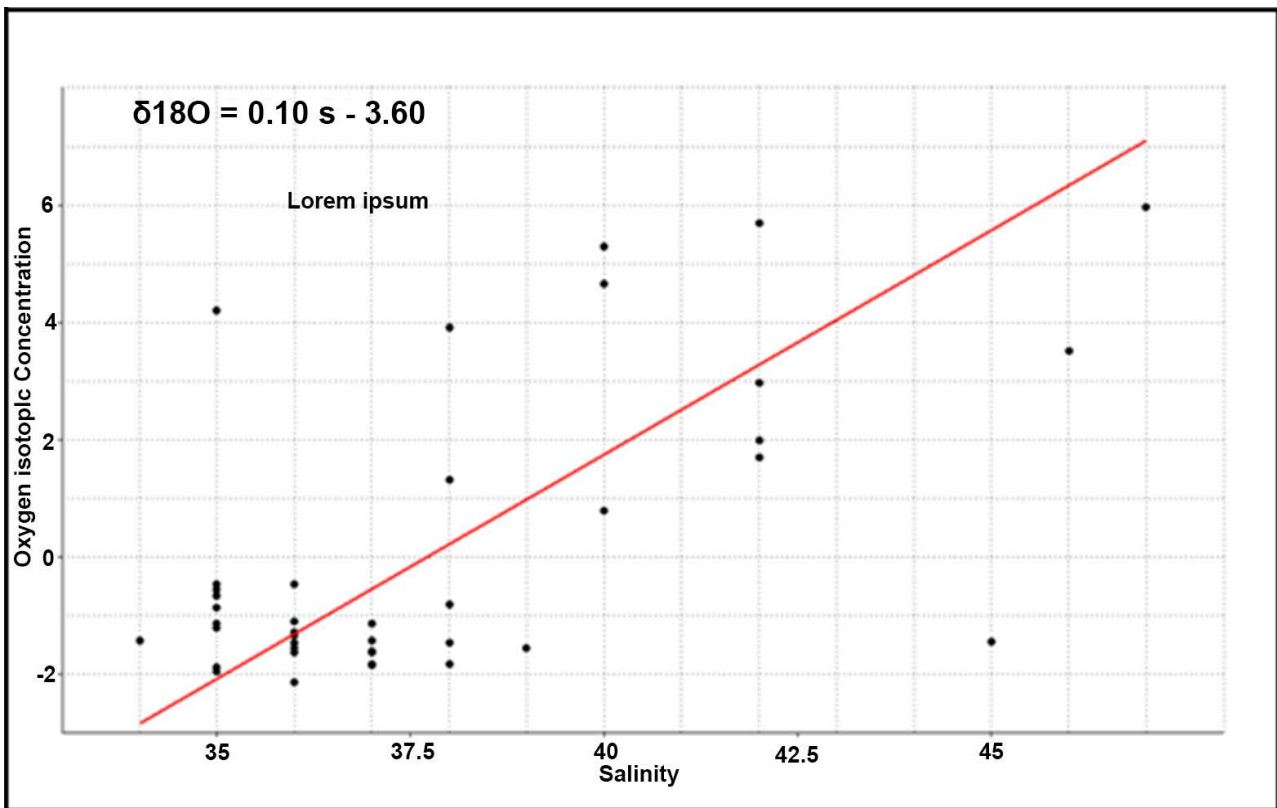
(b)

Figure 5. (a) and (b) distribution of salinity and oxygen isotopic composition ($\delta^{18}\text{O}$) of biocoenosis (a) and thanatocoenosis (b) for the area sampled. The stations correspond to those shown in **Figure 1**. (Not all of the features of sampling stations, because it showed low values of voltage, which does not reliable for proper data interpretation).

This effect can be verified with the analyses performed by [19], who proposed the relationship between isotopic variation and temperature for bivalve mollusks, and by [30], who studied deep-water benthic foraminifera and found a relationship between the temperature and the oxygen isotope concentration without taking salinity into account. We also found that this relationship was present between the variables of salinity and [$\delta^{18}\text{O}$], more so than with temperature.



(a)



(b)

Figure 6. Prediction model between the salinity and the ($\delta^{18}\text{O}$), where one can observe the variation range of the isotope concentration at the same salinity, the value of correlation found is $r = 0.62$.

However, it should be made clear that salinity is in some way linked to temperature changes based on the principle that the higher the temperature, the greater the evaporation and, thus, the greater the salinity.

In contrast, the isotopic concentration of [$\delta^{18}\text{O}$] the dead assemblages (**Figure 4(b)**) did not display a pattern coincident with the variation in temperature or salinity (**Figure 5(b)**). These results can be explained by the fact that the foraminifer tests from this community fixed [$\delta^{18}\text{O}$] at temperatures and salinities different from those found at the time of sampling and that likely represent the present values. Considering that before the construction of dams on the upper part of the Colorado River, approximately 80 years ago, the flow of freshwater to the delta was continuous, one could possibly expect that the isotopic concentrations of these foraminifera dead assemblages reflect the estuarine conditions or conditions predominantly influenced by isotopically lighter water from the said river in the years prior to damming. However, these concentrations could also be due to an extraordinary season of flow, as discussed by [10]; for example, in 1993, rains led to the dam storage limits being exceeded, resulting in the flood-gates being opened and the water flow that reached the Mexican side being much greater than normal, thereby leading to a possible discharge down to the delta.

The dead assemblages reported herein could possibly show diluted isotopic conditions from this time period. However, for future analyses, more precise dating of the tests is recommended to determine the time that is being represented. At the moment, because the dating of the dead assemblages is not available and knowing the expected isotopic values for the flow conditions reported by [15] [23] and [19], it is possible to discern the potential environmental conditions in the case of continuous freshwater flow to the delta.

The preceding facts were proven by performing a statistical test in which homogeneity in the variances was found (**Table 5**), indicating that these seasons do not show significant differences among themselves. This result is interpreted to represent the environmental conditions that prevailed in the study area prior to the damming of the Colorado River because the foraminifera tests fix [$\delta^{18}\text{O}$] from the surrounding water. Thus, the tests reflect the isotopic composition of the water in which they lived [19].

As was previously discussed, this fixation of isotopes in the tests can be affected or influenced by the salinity concentration, which according to bibliographic records for the periods prior to damming was between 35.2 and 35.7 ppm [34]. These values are similar to those that were recorded for the spring season, leaving the remainder of the seasons (Summer, Fall, and Winter) with values above that recorded before the dams.

4. Conclusions

With no current supply of isotopically lighter water from the Colorado River, the isotopic variations found in the tests of foraminifera from the living assemblages are related to changes in temperature and salinity.

Table 5. Analysis of variance for thanatocoenosis.

Analysis of Variance (ANOVA Type I)						
Source of Variation	Gl	Sum Sq	Mean Sq	F value	Pr(>F)	SC
Season	3	0.9009	0.30031	2.6934	0.06314	.
Residual	31	3.4564	0.1115			

Given that the dead assemblages did not fix current isotopic signals, they represent a temporal average by the mixing of generations existing from before the damming of the waters of the Colorado River up to the present.

The variation range of $[\delta^{18}\text{O}]$ in the living assemblages was 8% for a temperature range of 11°C, whereas that in the dead assemblages was 2.3% for the same temperature.

Based on our data and under the same conditions at salinity of 14%, one would expect a $[\delta^{18}\text{O}]$ variation of 8% in live foraminifera tests.

The living assemblages showed a relationship between the oxygen isotope concentration and salinity. The latter could be the cause of the isotopic variations together with the temperature. For the dead assemblages, isotopic variations linked to temperature and salinity changes were not observed.

The isotopic data allowed us to discriminate between two environmental conditions: hot seasons (Summer-Fall) and cold seasons (Winter-Spring). Meanwhile, in the dead assemblages, significant changes were not detected over the course of the year due to temporal averaging.

In the living assemblages, a direct relationship was not registered between the temperature and the oxygen isotope concentration because the data reflected the annual average temperature.

For future analyses, we recommend distinguishing between juveniles and adults, given that the mixing of generations can produce a bias with regard to data interpretation as the adult organisms represent an annual average of temperatures, whereas the juvenile individuals, having a shorter exposure time, would reflect the precise conditions of the environment in which they developed.

For a more exact interpretation of past environmental conditions, we recommend performing more precise dating analyses of the foraminifera tests that compose the dead assemblages, with the objective of understanding to which time or era those tests correspond.

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