

## Quaternary environmental evolution of the Argentinean Pampa Deprimida based on mollusc and ostracod analysis

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Multiproxy analyses are efficient approaches in palaeoenvironmental reconstructions since they contemplate the different responses of various taxonomic groups to external changes. In this study, we analyse changes in molluscs and ostracod palaeocommunities to reconstruct the environments and their Holocene evolution in the Salado River, Pampa Deprimida of Argentina. We assessed the temporal and spatial variation of the assemblages from the Alberti-Pla, Ruta 30, and Vientos de Mar localities through various analytical techniques, including analysis of similarity (ANOSIM) and ordination methods such as non-metric multidimensional scaling (NMDS), detrended correspondence analysis (DCA), and principal component analysis (PCA). Eleven species of gastropods were identified, among which Heleobia parchappii, Biomphalaria peregrina, and Succinea meridionalis stand out for their abundance. Nine species of ostracods were recognised, with Limnocythere cusminskyae, Cypridopsis vidua, Heterocypris similis, and Cyprideis salebrosa being the most important. The changes in the assemblages allowed us to infer the paleoenvironmental evolution in the area. During the Late Pleistocene and Early Holocene, drier conditions led to ephemeral, marshy freshwater environments with fluctuating water levels and salinities. In the Late Holocene, increased precipitation caused water levels to rise, resulting in more stable and interconnected freshwater environments. [] Holocene, Argentine Pampas, calcareous microfossils, continental palaeoenvironments, multiproxy analyses.

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The Holocene is an epoch characterized by its climatic variability and instability, with episodes of rapid and significant global changes (Mayewski et al. 2004) that have influenced the development of different environments, mainly those where exogenous processes were the modellers of the landscape. Within tropical latitudes in South America, the Early and Middle-Holocene were characterized by more seasonal climate variability, generally warmer and drier, due to a decrease in summer insolation between 12000 to 5000 years BP and an increase from 5000 years BP (Apaéstegui et al. 2014). The low insolation during the austral summer in the Middle Holocene would have reduced the temperature contrast between land and sea, weakened the South American monsoon system's circulation, and decreased precipitation over the South Atlantic Convergence Zone (Prado et al. 2013). These dry episodes would have been interspersed with other wet ones. Since the increase in insolation led to the global temperature increase during the climatic optimum, this allows us to infer the wide climatic variability that prevailed during the Middle Holocene. In the Late Holocene, climatic conditions were established that were similar to those of today (Hermanowski *et al.* 2012).

For the continental environments of the Pampean region, palaeoclimatic reconstructions were carried out using different proxies (e.g. isotopes, pollen, molluscs, ostracods, mammals) that allowed the development of local and regional models (e.g. Prieto *et al.* 2004; Mancini *et al.* 2005; Villanova *et al.* 2010; García Morato *et al.* 2021; Sanz Pérez *et al.* 2022). Climatic alternations were reflected by the development of deposits and geoforms of varied origin (e.g. eolian, fluvial, marine), such as the presence of palaeosols, and their palaeocommunities that inhabited (Iriondo & García 1993; Fucks *et al.* 2012; 2015; Pommarés *et al.* 2021). In the Salado River Basin, the alternation of wet and dry periods produced an excess or a deficit of water, causing either flooding events or the

interruption of the fluvial network and the drying of shallow lakes (Pommarés *et al.* 2021).

Non-marine molluscs and ostracods have successfully colonized a wide variety of freshwater environments (e.g. rivers, streams, lakes, marshes, temporary ponds), and even land, in the case of molluscs (Dillon 2000; Meish 2000). They are among the dominant groups in non-marine Quaternary deposits, together with diatoms, sporomorphs, and charophytes (e.g. Torres Saldarriaga & Martínez 2010; Laprida et al. 2009; Tietze & De Francesco 2012; De Francesco et al. 2013; Tuncer et al. 2019). Moreover, they are sensitive to changes in the environmental parameters, such as temperature, pH, alkalinity, salinity, and fluctuations in water levels (e.g. Adams et al. 2002; Dillon 2000; Mezquita et al. 2005; Cusminsky et al. 2011; Ruiz et al. 2013; Mohammed et al. 2018). Therefore, they are reliable as palaeoenvironmental indicators. In addition, multi-proxy analyses are preferred since organisms respond differently to external changes in their ability to react and recover (Holmes et al. 2007; Stutz et al. 2014). With these approaches, it is possible to obtain independent information from each proxy and make consistent palaeoenvironmental reconstructions through their integration (Birks & Birks 2006; Stutz et al. 2014; Coviaga et al. 2017). Given that how molluscs and ostracod communities responded to environmental variations throughout the Holocene was not established for the upper Salado River basin, and no previous research has analysed these proxies together in Argentina, the objectives of the present study are to: (1) evaluate changes in the composition of molluscs and ostracods assemblages; and (2) reconstruct the environments and their evolution throughout the Holocene in the study area. To determine the conservation status and the environmental reliability of the assemblages (autochthonous vs allochthonous), they were previously taphonomically analysed (Pisano et al. 2022).

### Study area and geological setting

The Salado River Basin, located in the northeastern region of Buenos Aires Province, belongs to the Pampa Deprimida ('Depressed Pampa') region; Fidalgo (1992; Fig. 1 A), and it is the main watershed of the province. This river originates in Santa Fe Province and flows southeastward into Samborombón Bay. It meanders through an area with a low gradient (0.107 m km-1, Bazzuri *et al.* 2018) and contains Late Quaternary aeolian accumulation geoforms (Iriondo & Kröhling 2007) that affect its hydrological dynamics.

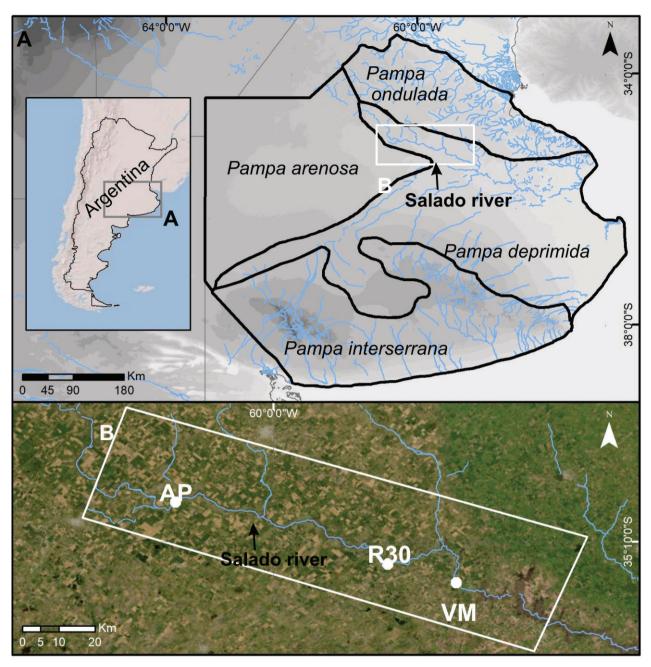
Historically, the area has been affected by alternating wet and dry periods (Iriondo & Kröhling 2007). The periodic floods cause large-scale losses in agricultural production, roads, and urban infrastructure. Conversely, droughts cause a substantial decrease in flow, leading to a total interruption of the river's course and the drying up of the shallow lakes in the area. In particular, the study area is in the uppermedium reaches of the Salado River Basin (Fig. 1 A), where the river has a braided design, with numerous active channels, internal bars, and a wide floodplain that fluctuates between 1-2.5 km in width, with a gentle longitudinal eastward slope, and levees limiting the main course.

The lithostratigraphical scheme of the *Pampa Deprimida* was established by Fidalgo *et al.* (1973), who defined the Luján Formation including fluvial Quaternary sediments. Later, Fucks *et al.* (2015) redesigned and reorganized this scheme, identifying the La Chumbiada, Gorch, and Puente Las Gaviotas members within the Lujan Formation. Recently, Pommarés *et al.* (2021) identified distinctive sedimentary morphologies and deposits of aeolian and fluvial origins in association with climate changes during the Late Pleistocene-Holocene.

In our study, we followed the stratigraphical framework proposed by Fucks et al. (2015). The sediments of the La Chumbiada Member were deposited during the Late Pleistocene (14,040 ± 50 and 12,100 ± 100 14C years BP; Fucks et al. 2015; Scanferla et al. 2013), whereas the Gorch Member was accumulated in the Early Holocene (11,690 ± 110 - 8,640 14C years BP; Fucks et al. 2015; Pommáres et al. 2021; Pisano et al. 2022). The Puente Las Gaviotas Member, on the other hand, was deposited during the Late Holocene  $(3002 \pm 40 \text{ years BP to the present; Fucks et al. 2012;}$ 2015; Mari et al. 2013). The recently deposited materials that make the uppermost sediment layer gradually accumulated and underwent pedogenesis. This stratum exhibits a dark grey to black colouration, abundant decomposing organic matter, roots, seeds, saline efflorescence on the surface, and anthropogenic materials.

## Material and methods

We analysed three locations along the Salado River (Fig. 1 B): Alberti-Pla (AP, 35° 4' 9.60" S/ 60° 14' 32.70" W), Ruta 30 (R30, 35° 13' 20.80" S/ 59° 43' 4.00" W), and Vientos de Mar (VM, 35° 16' 6.10" S /59° 32' 54.20" W), which were previously examined from a taphonomic perspective by Pisano *et al.* (2022). In each locality, we extracted sediment cores



*Fig. 1.* General location of the study area (A) and sampling sites (B). Abbrieviations: AP: Alberti-Pla, R30: Ruta 30, and VM: Vientos del Mar. Sources: (A) Base map, Esri, Maxar, Earthstar Geographics, GIS user community; Province and hydrography: Instituto Geográfico Militar; Dem: Copernicus 90 and Agencia Espacial Europea; (B), Google Earth, DigitalGlobe.

of 184 cm, 155 cm, and about 200 cm in length respectively, using an Edelman manual auger (Eijkelkamp) for undisturbed sediment. In the laboratory, cores were subsampled at intervals of approximately six centimetres. We washed the samples under running water using two sieves of 500  $\mu$ m to recover molluscs and 63  $\mu$ m for ostracods and dried them at room temperature. Shells, carapaces, and valves were recovered

by manual picking under a stereoscopic microscope. We identified and quantified the specimens recovered from a fixed weight of sediment of 100 grams for molluscs and 5 grams for ostracods to determine the absolute abundance of species (Bernasconi *et al.* 2019; Calvo Marcilese *et al.* 2019; Parras *et al.* 2020).

Given that the recovered molluscs were all gastropods, for quantification we considered the presence of a specimen only if the apex was present in the shell. For ostracods, we calculated the minimum number of individuals (NMI) by adding the most abundant shell (either right or left) to the number of carapaces. The NMI was calculated for each species in each sample. Whenever possible, we identified the specimens to the species level following Fernández & Castellanos (1973), Castellanos & Fernández (1976), Gaillard & Castellanos (1976), Fernández (1981), Rumi (1991), Miquel & Aguirre (2011) for gastropods, and Ramirez (1967), Cusminsky & Whatley (1996), Meisch (2000), Laprida (2006), and Ramon Mercau *et al.* (2014) for ostracods.

#### Numerical analyses

We performed a Similarity analysis (ANOSIM) to evaluate the temporal variation in the assemblages. The original abundance matrices were transformed to log (X+1), which balances species contribution when one is very abundant, and were then converted into similarity matrices using the Bray-Curtis index. These matrices were then compared using localities as a grouping factor to analyse the variation in assemblage composition from a regional perspective. In cases where differences among groups were observed, we conducted post-hoc pairwise comparisons. The statistic R obtained reflects the degree of separation between groups based on their specific composition, R near 1 indicates that the groups are different, whereas R near 0 indicates that the groups are similar. We also employed non-metric multidimensional scaling (NMDS) to analyse the spatial variation in species composition. NMDS is a multivariate ordination technique that visualizes the proximity between a set of samples in a two-dimensional space based on the Bray-Curtis similarity index using data on species abundances. Additionally, we calculated the similarity percentage (SIMPER) to identify the discriminant species that explain the observed similarities and differences within and between groups. Finally, to compare the responses exhibited by molluscs and ostracods, we reduced the variability of each bioindicator to a single variable, following the methodology used by Hassan (2013) and De Francesco et al. (2022). The species abundance data were square root transformed to reduce the asymmetry of their variances (Legendre & Birks 2010), and we conducted a detrended correspondence analysis (DCA) on each set of biological data to determine whether to use linear- or unimodal-based numerical approaches (Bigler et al. 2006).

The unimodal method was applied when the gradient length exceeded 2.5 SD, exclusively for ostracods from VM (SD = 3.74) within this study. These values indicate a relatively long gradient and make the DCA approach appropriate. On the contrary, the linear method was employed when the gradient lengths were shorter than 2.5 SD, specifically in all molluscs and ostracods from PA and R30 (SD ranging between 1.20 and 1.67). This choice renders the linear approach of principal component analysis (PCA) suitable for the analysis Legendre & Birks 2010; Birks 2012). Finally, the scores of the first ordination axis were plotted against the depth of stratigraphic profiles. These analyses were performed using the 'vegan' package (Oksanen *et al.* 2013) within the statistical program R version 3.0.1 (R Core Team 2013) and PRIMER version 5 (Clarke & Gorley 2001).

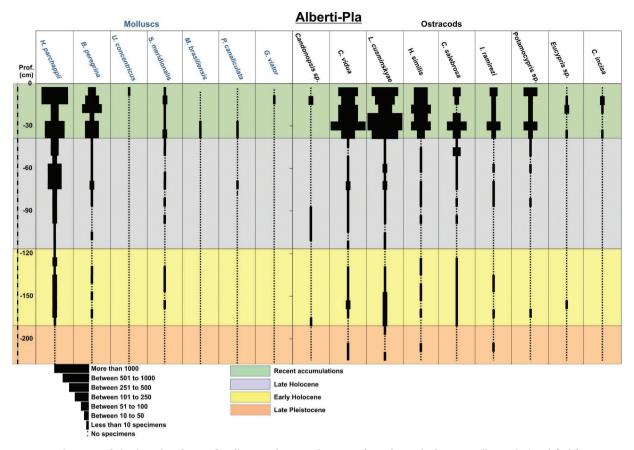
## Results

#### General characteristics of the assemblages

A total of 11 species of molluscs were recorded, all of them gastropods: Heleobia parchappii (d'Orbigny 1835), Biomphalaria peregrina (d'Orbigny 1835), Drepanotrema heloicum (d'Orbigny 1835), Drepanotrema kermatoides (d'Orbigny 1835), Uncancylus concentricus (d'Orbigny 1835), Galba viator (d'Orbigny 1835), Antillorbis nordestensis (Lucena 1954), Pomacea canaliculata (Lamarck 1822), Succinea meridionalis d'Orbigny 1846, Miradiscops brasiliensis (Thiele 1927), and Gastrocopta nodosaria (d'Orbigny 1835). Nine species of ostracods were found: Limnocythere cusminskyae Ramon Mercau et al. 2014, Cypridopsis vidua (Müller 176), Heterocypris similis (Wierzejski in Ramírez 1967), Cyprideis salebrosa Van den Bold 1963, Potamocypris sp. Brady 1870, Ilyocypris ramirezi Cusminsky & Whatley 1996, Eucypris sp. Vavra 1891, Chlamydotheca incisa (Claus 1893), and Candonopsis sp. Vavra 1891.

# *Assemblages changes throughout the Late Pleistocene-Holocene*

Late Pleistocene.—The gastropod assemblages presented a low species richness and abundance in this unit. No specimens were recovered at the AP locality (Fig. 2), while only 1 to 3, belonging to *H. parchappii* and *S. meridionalis*, were found at R30 and VM (Fig. 3 and 4). We found six species of ostracods in AP and R30 (Fig. 2 and 3), *L. cusminskyae*, *C. vidua*, *H. similis*, *C. salebrosa*, *I. ramirezi*, and *Candonopsis* sp., and only *Candonopsis* sp. in VM (Fig. 4). The maximum abundance was ten individuals for the first two species and only 1 or 2 specimens in the others. Anosim analysis



*Fig. 2.* Distribution and absolute abundance of molluscs and ostracods species from the studied core in Alberti Pla (modified from Pisano *et al.* 2022).

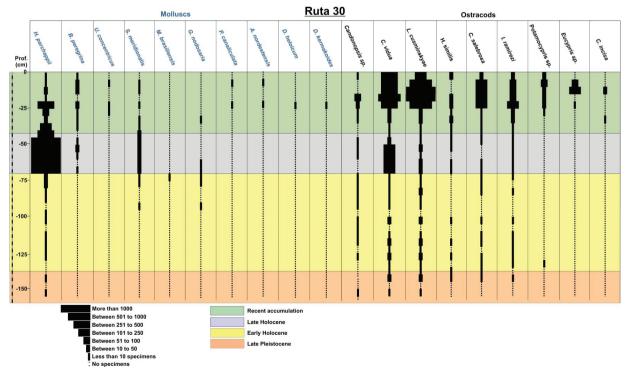
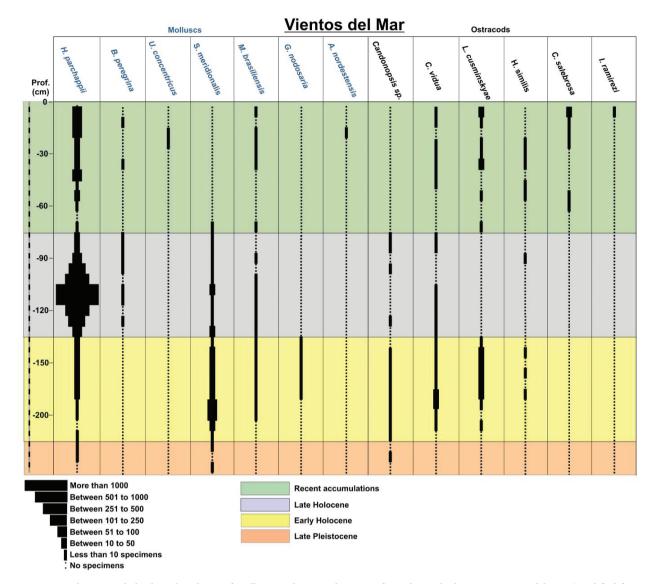


Fig. 3. Distribution and absolute abundance of molluscs and ostracods species from the studied core in Ruta 30 (modified from Pisano et al. 2022).



*Fig. 4.* Distribution and absolute abundance of molluscs and ostracods species from the studied core in Vientos del Mar (modified from Pisano *et al.* 2022).

(Table 1) was performed in this unit considering the two groups together due to the low abundance of individuals. Anosim showed no differences (R = 0.4, p = 0.04) between the localities, while in the NMDS the samples were dispersed without any particular grouping (Fig. 5. A).

*Early Holocene.*—We recorded gastropods at all localities studied (Figs. 2, 3, and 4). With the addition of *B. peregrina*, *M. brasiliensis*, and *G. nodosaria* to *H. parchappii* and *S. meridionalis*, the association shows an increase in species richness compared to the Late Pleistocene assemblages. The Anosim analysis detected differences between VM and the other localities (AP and R30; R = 0.48, p = 0.001, Table 1),

which allows the differentiation of the groups EHg1 and EHg2 in the NMDS analysis (Fig. 5. B). In EHg1, the samples from AP (SP = 63.17) and R30 (SP = 48.14) were clustered. The similar abundances of *H. parchappii*, the dominant specie, which contributed more than 90% in both localities (Table 2), defines the identity of this group even shaping, monospecific assemblages. However, within EHg1, it was possible to identify two subgroups by the changes in abundance of the accompanying species. In AP, *B. peregrina* was more abundant and ubiquitous than *S. meridionalis*, while in R30 the opposite occurred (Figs. 3, 4). In EHg2, the majority of VM samples were found (SP = 67.50; Fig. 5), with *S. meridionalis* being more abundant than *H. parchappii*. Together, these

*Table 1.* Results of the similarity analysis (ANOSIM) and paired comparisons between units, raw data transformed log (x+1), 999 computed permutations. In bold, the statistically significant results (p < 0.005). Abbreviations: AP: Alberti-Pla, R30: Ruta 30, VM: Vientos del Mar.

	Comparison	Statistical R	p-value	
	Molluscs + Ostracods	0.4	0.04	
Late	AP vs. R30	0.037	0.04	
Pleistocene	AP vs. VM	0.67	0.1	
	R30 vs. VM	0.57	0.1	
Early Holocene	Molluscs	0.48	0.001	
	AP vs. R30	0.232	0.013	
	AP vs. VM	0.73	0.001	
	R30 vs. VM	0.56	0.001	
	Ostracods	0.30	0.001	
	AP vs. R30	0.29	0.002	
	AP vs. VM	0.42	0.001	
	R30 vs. VM	0.24	0.011	
Late Holocene	Molluscs	0.24	0.003	
	AP vs. R30	0.33	0.016	
	AP vs. VM	0.18	0.013	
	R30 vs. VM	0.32	0.011	
	Ostracods	0.33	0.001	
	AP vs. R30	0.07	0.22	
	AP vs. VM	0.49	0.001	
	R30 vs. VM	0.31	<b>0.001</b> 0.017	
	Molluscs	0.46	0.001	
	AP vs. R30	0.21	0.025	
	AP vs. VM	0.82	0.001	
Recent	R30 vs. VM	0.34	0.003	
accumulations	Ostracods	0.41	0.001	
	AP vs. R30	0.078	0.14	
	AP vs. VM	0.62	0.001	
	R30 vs. VM	0.49	0.001	

two species accounted for 85% of the group's composition, as shown in Table 2. In addition, *M. brasiliensis* was present in all assemblages, and the specimens of *G. nodosaria*. Finally, the ungrouped samples in the upper right corner of the graph are those with a very low abundance of individuals.

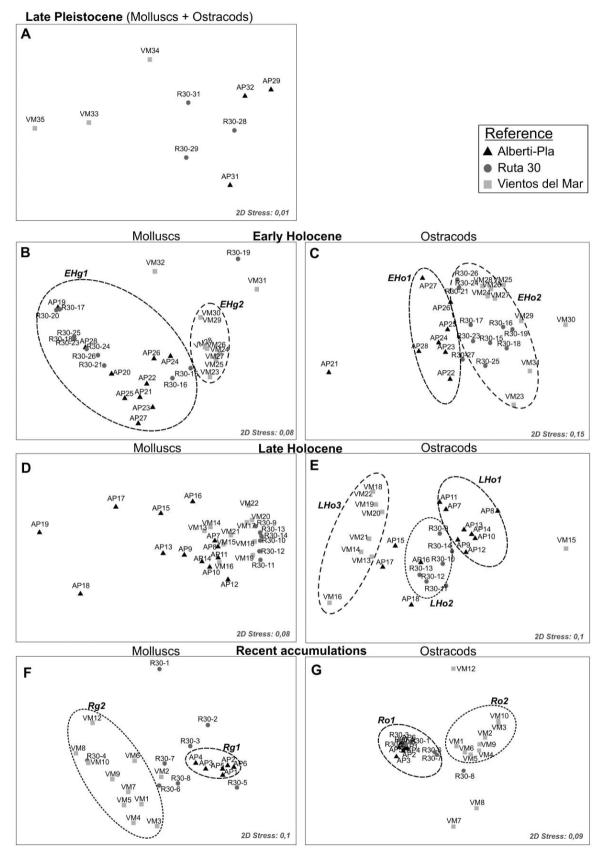
In the ostracod assemblages, we found eight species, commonly *L. cusminskyae*, *C. vidua*, *H. similis*, *C. salebrosa*, *I. ramirezi*, and *Candonopsis* sp., and very occasionally *Potamocypris* sp. and *Eucypris* sp. All these species were present at the AP locality, seven at R30, and only four at VM (Figs. 2–4) The Anosim analysis revealed significant differences (R = 0.30,

p = 0.001), specifically between AP and the other localities (Table 1), and in the NMDS analysis (Fig. 5. C), we can recognize two groups. The main factors explaining the differences between localities were the decrease in diversity from left to right of the graph, the increase in abundance in the opposite direction, and the varying contributions of species. The samples belonging to AP were grouped in EHo1 (SP = 56.82). Limnocythere cusminskyae, C. vidua, C. salebrosa, and H. similis were the species that contribute more than 98% to the similarity of this group (Table 2). In EHo2 there were the R30 and VM assemblages where, in addition to L. cusminskyae and C. vidua, the presence of Candonopsis sp. and I. ramirezi (only in R30) stands out as distinctive species of this group (Table 2). Finally, in the upper right corner of the graph, the ungrouped samples are those with very low abundances.

At the AP and R30 localities, both proxies exhibited similar trends (Fig. 6), showing minimal rhythmic variation attributed to low abundance assemblages and no significant changes in species replacement. Conversely, in VM, a steady increase was consistently observed throughout this unit.

Late Holocene.—The gastropod assemblages within this unit exhibited a stable composition across all three localities, with H. parchappii as the dominant species and variations in the co-dominant species, namely B. peregrina at AP and either S. meridionalis at R30 and VM. In addition, the assemblages were completed with G. nodosaria at R30, M. brasiliensis at VM, and a specimen of *P. canaliculata* at AP (Figs. 2, 3, 4). Therefore, Anosim analysis detected no differences between locations (R = 0.13, p = 0.06%, Table 1), and the NMDS shows that the arrangement of the samples can be explained by the increase in abundance of *H. parchappii*, from left to right of the graph (Fig. 5. D). Heleobia parchappii was the most important species and contributed more than 65% to the similarity of the samples from the different localities (Table 2). In addition, in both R30 and VM, this species reached its maximum abundance levels in the assemblages of this age, and this is reflected in the trends observed in these localities (Fig. 6). Biomphalaria peregrina recorded an increase, both in abundance and in the number of assemblages, with respect to the Early Holocene.

The assemblages observed in AP exhibited the broadest dispersion (SP = 57.84, Table 2), covering a wide range on the graph and indicating an enrichment in abundance towards more recent samples (corresponding to the upper part of the profiles). So, the assemblages of the lower levels (between -110 to



*Fig.* 5. Non-Metric Multidimensional Scaling (NMDS) ordination plot of the samples considering the abundance of gastropod and ostracod species that compose the assemblages in the different localities. Species abundances were log (x+1) transformed and the ordination was performed with the Bray-Curtis dissimilarity matrix.

*Table 2.* Characteristic species that define the similarity of each locality. Results obtained through SIMPER routine. Abbreviations: AA = average abundance, CTB% = percentage contribution, ACM % = accumulative percentage, species = Hp: *Heleobia parchappii*, Bp: *Biomphalaria peregrina*, Sm: *Succinea meridionalis*, Mb: *Miradiscops brasiliensis*, Can: *Candonopsis* sp., Cv: *Cypridopsis vidua*, Lc: *Limnocythere cusminsk-yae*, Hs: *Heterocypris similis*, Cs: *Cyprideis salebrosa*, Ir: *Ilyocypris ramirezi*, Pot: *Potamocypris* sp.

_		Molluscs				Ostracods			
	Species	AA	СТВ%	ACM%	Species	AA	CTB%	ACM%	
	Alber	rti-Pla: averag	ge similarity = 6	59.73	Alber	rti-Pla: averaş	ge similarity = 5	56.82	
	Нр	2.44	90.54	90.54	Lc	2.04	39.39	39.39	
					Cs	1.22	30.88	70.27	
					Cv	1.17	17.24	87.51	
					Hs	0.76	10.74	98.25	
	Ru	ta 30: average	similarity = 54	.29	Ruta 30: average similarity = 68.78				
Early	Нр	1.50	97.28	97.28	Lc	1.99	35.72	35.72	
Holocene	тŗ	1.00	<i>)1.20</i>	<i>)</i> ,. <u>2</u> 0	Cv	1.90	34.80	70.52	
					Ily	0.02	12.25	82.77	
					Can	0.83	8.53	91.3	
	Vientos	del Mar: avei	rage similarity	= 67.50			rage similarity		
	Sm	3.09	54.35	54.35	Cv	1.91	43.46	43.46	
	Нр	2.26	29.29	83.64	Lc	2.14	37.81	81.27	
	Mb	1.07	12.02	95.66	Can	0.82	17.47	98.73	
			$\frac{12.02}{\text{ge similarity}} = 5$				ge similarity = 4		
	Нр	3.28	80.79	80.79	Lc	1.45	37.29	34.53	
	Вр	1.07	15.10	95.89	Cv	1.43	28.98	66.27	
	Ър	1.07	15.10	93.09	Cs	1.14	14.17	80.44	
					Hs	0.83	14.17	91.64	
	Ruta 30: average similarity = 90.48				Ruta 30: average similarity = 52.12				
Late	Нр	7.00	68.20	68.20	Cv	3.15	53.06	53.06	
Holocene	Sm	2.96	28.64	96.4	Lc	1.63	21.52	74.58	
	5111	2.70	20.04	50.4	Ily	0.88	10.97	85.55	
					Can	0.65	6.71	92.26	
	Vientos del Mar: average similarity = 76.14				Vientos del Mar: average similarity = 39.21				
	Нр	5.11	65.98	65.98	Cv	0.79	73.35	73.35	
	Sm	1.78	21.39	87.37	Can	0.51	24.65	100	
	Вр	1.01	7.58	94.95	Guil	0101	21100	100	
	Alberti-Pla: average similarity = 82.92				Alberti-Pla: average similarity = 82.13				
	Нр	5.60	45.69	45.69	Lc	6.75	22.72	22.72	
	Вр	4.77	39.47	85.16	Cv	5.84	20.26	42.98	
	Sm	2.04	14.47	99.63	Hs	5.23	18.66	61.64	
	om	2.01	11.17	· · · · · ·	Pot	4.10	13.97	75.61	
					Ir	3.73	12.26	87.87	
					Cs	3.90	10.39	98.26	
	<i>Ruta 30: average similarity = 51.81</i>				Ruta 30: average similarity = $60.36$				
Recent	Нр	3.27	61.28	61.28	Cv	4.77	24.92	24.92	
accumulations	Вр	1.88	27.21	88.48	Lc	4.94	23.55	48.47	
	Sm	0.96	9.07	97.56	Ir	3.45	17.8	66.27	
					Cs	3.49	16.18	82.45	
					Pot	2.29	7.49	89.94	
					Hs	1.70	5.67	95.61	
	Vientos del Mar: average similarity = 62.86				Vientos del Mar: average similarity = $41.33$				
	Нр	3.11	92.73	92.73	Cs	1.55	54.27	54.27	
	r				Lc	0.94	18.6	72.87	
					Cv	0.63	17.67	90.54	

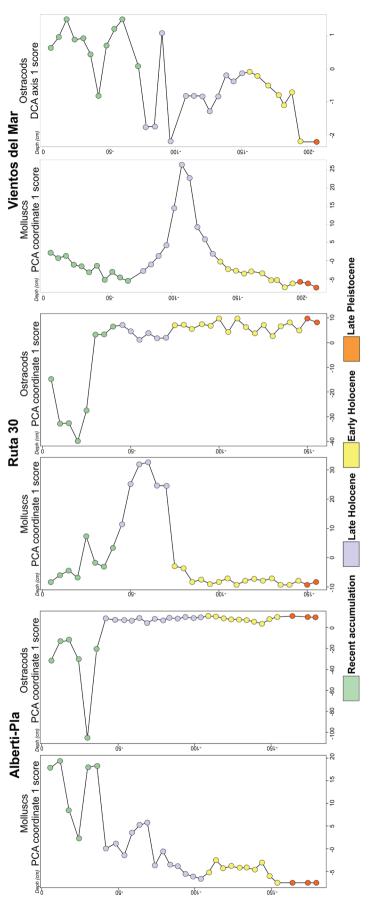


Fig. 6. Scores of the first ordination axis of PCA for Molluscs and Ostracods (from AP and R30), and DCA for Ostracods (from VM) vs Depth in Alberti-Pla, Ruta 30 and Vientos del Mar cores.

-74 cm, from AP 19 to AP 13) showed a very low abundance of individuals, mainly of *H. parchappii*, even forming monospecific assemblages. Progressively, the abundance increased towards the top of the sequence, and *B. peregrina* was more common. In contrast to AP, the assemblages recovered from R30 and VM showed high similarity values within localities (SP<sub>R30</sub> = 90.48, SP<sub>VM</sub> = 76.14) and remained stable across sequences.

*Limnocythere cusminskyae*, *C. vidua*, *H. similis*, *C. salebrosa*, *I. ramirezi*, *Candonopsis* sp., and *Potamocypris* sp. were present in the ostracod assemblages. All the species occur in AP (Fig. 2), then the species richness decreased in the other localities, with six in R30 and only three in VM (Figs. 3, 4). Anosim analysis reflected a significate difference between AP and VM (R = 0.49, p = 0.001, Table 1), and we identified three groups in the NMDS (Fig. 5. E, Table 2).

The AP samples (SP = 45.25) were clustered in LHo1, where C. salebrosa was the most abundant species, but C. vidua and L. cusminskyae were those with continuous records throughout these assemblages. The Simper analysis showed that these three species contribute 80% to the similarity of the group (Table 2). An important change occurs from -84 cm (AP 14) when Candonopsis sp. disappears, L. cusminskyae increases its abundance, and H. similis and C. salebrosa become more frequent and abundant. The R30 samples were grouped in LHo2 (SP = 52.12), where C. vidua was the most abundant species, contributing 53% to the overall group similarity (table2), and accompanied by L. cusminskyae, I. ramirezi, and Candonopsis sp. Finally, LHo3 groups the VM samples (SP = 39.21) characterized by a very low abundance of C. vidua and Candonopsis sp. individuals.

*Recent assemblages.*—The gastropod assemblages recovered from this level presented the highest richness with 11 species (Fig. 6). In addition to those mentioned above, there were present *U. concentricus*, *A. nordestensis*, *D. heloicum*, *D. kermatoides*, and with very low abundances *G. viator* (Figs. 2, 3, 4). Anosim analysis (Table 1) showed differences (R = 0.46, p = 0.001) between VM and the other localities (VM-AP: R = 0.82, p = 0.001; VM-R30: R = 0.34, p = 0.003).

All samples from AP were clustered within Rg1 (SP = 82.45) (Fig. 5F; Table 2). These samples exhibited assemblages characterized by the highest abundances of *H. parchappii* and *B. peregrina*, along with *S. meridionalis*, although to a lesser extent. *Pomacea canaliculata* and *M. brasiliensis* were occasionally recorded in these assemblages, along with the first specimens of *U. concentricus* and *G. viator*.

The samples from R30 (SP = 51.43) were located in the centre of the graph. They exhibited the same species as AP, which explains the similarity between localities and the same abundance ratio (Hp>Bp>Sm). *Heleobia parchappii* presented a progressive increase in abundance in the lower assemblages, reaching a maximum at -25 cm (R30-5) as occurs with *B. peregrina*. In addition, at this level, we found the only records of *D. heloicum*, *D. kermatoides*, and *A. nordestensis*, together with *U. concentricus*. From -20 cm (R30-4), a low abundance and monospecific level of *H. parchappii* begins. The upper assemblages presented a lower abundance of *H. parchappii*, *B. peregrina*, and *S. meridionalis*, and species richness with occasionally recorded of *A. nordestensis*, *U. concentricus*, and *P. canaliculata*.

Finally, Rg2 is composed of the VM samples (SP = 62.86). These assemblages were characterized by a low abundance of individuals, and except for *H. parchappii*, which was present in all assemblages; the remaining species were recorded sporadically.

The AP and R30 ostracod assemblages recorded an increase in species richness (Fig. 6), and abundances of individuals were the highest in AP and R30  $(n_{AP} = 1931, n_{R30} = 2250)$ , and considerably lower in VM (n = 33). So, as for the gastropods, the Anosim analysis identified differences (R = 0.42, p = 0.001, Table 1) between the VM and the other localities analysed (R<sub>VM-AP</sub> = 0.62, p = 0.001; R<sub>VM-R30</sub> = 0.49, p = 0.001), which visualized in the NMDS (Fig. 5. G). Ro1 groups the samples from AP and R30 ( $SP_{AP}$  = 82.13 and  $SP_{R30} = 60.36$ ,  $DP_{AP-R30} = 33.81$ ) in which *L. cusminskyae* and *C. vidua* were the most abundant species; H. similis, Potamocypris sp., I. ramirezi, and C. salebrosa completed these assemblages. The different abundances between the species are the ones that mark the differences between localities (Table 2). In AP, H. similis (Cont.% 18.66) and Potamocypris sp. (Cont.% 13.97) were the most contributing, while in R30 were I. ramirezi (Cont.% 17.80) and C. salebrosa (Cont.% 16.18). In Ro2 are samples from VM (SP = 42.53) with a low abundance of individuals. In this group, C. salebrosa, L. cusminskyae, and C. vidua are the species that contribute most to the identity of the group (Cont. % 54.27, 18.60 and 17.67, respectively).

## Interpretation and discussion

#### General composition of the assemblages

The eleven gastropod species were previously recorded from Quaternary sediments of the middle and lower Salado River Basin (Pisano & Fucks 2016; Pisano *et al.* 2022) and other areas of the Pampean

region (e.g. Rumi *et al.* 2006; 2008; Miquel & Aguirre 2011; Nuñez *et al.* 2010; Tietze *et al.* 2011).

Heleobia parchappii is the dominant species of the assemblages with values of 90-100%, which coincides with data obtained for the lower basin of the Salado River (Pisano & Fucks 2016) and other areas of the Pampean region (De Francesco & Zárate 1999; Prieto et al. 2004; De Francesco et al. 2013). It inhabits lentic and lotic environments, both freshwater and brackish (Tietze et al. 2011; De Francesco et al. 2013). Monospecific assemblages of H. parchappii have been used as indicators of high-salinity environments, in which this species can maintain stable populations (De Francesco et al. 2022). Other gastropod species change prominence over time. For intence, B. peregrina and S. meridionalis alternate in co-dominance, carrying interesting palaeoenvironmental implications. Biomphalaria peregrina inhabits oligohaline lentic environments (lakes and ponds) dominated by submerged aquatic vegetation (Rumi 1991; De Francesco et al. 2013; Steffan et al. 2014). By comparison, S. meridionalis is a hygrophilous species that inhabits terrestrial environments associated with bodies of water and emergent macrophytes (De Francesco et al. 2007; Tietze & De Francesco 2012). This specie can be found in flooded, humid terrain or under rocks in the littoral zones of lentic and lotic bodies (Miquel & Aguirre 2011). It can also inhabit areas with high salinity levels (De Francesco et al. 2007). Among the terrestrial or geophilic species were also found M. brasiliensis, which lives on the vegetation cover of humid environments or under rotting plants (Steffan et al. 2014), and G. nodosaria, whose ecological requirements are poorly known, but it also inhabits the vegetation cover and under plant debris and rocks (Aguirre et al. 2011; Miquel & Aguirre 2011).

The other species found have freshwater habits. Pomacea canaliculata lives in different lentic environments and reservoirs within the floodplain where water accumulates due to rainfall or overflows, and can even be found in still or low-flowing streams (Castellanos & Fernandez 1976; Martin et al. 2001; Martinez & Rojas 2004). Pomacea canaliculata is amphibious, breathing using gills in water, but can also survive desiccation and unstable conditions due to the presence of an operculum (De Francesco et al. 2013). Uncancylus concentricus lives in the hard substrate of lentic or lotic environments of low energy and waters with abundant organic matter (Dos Santos 2003). It usually inhabits the underside of leaves of aquatic plants, partially submerged stems or on stones (Fernandez 1981; Steffan et al. 2014). Galba viator is a pulmonate gastropod that lives in shallow areas of different lentic and lotic environments of clear water and silty bottoms, preferably without turbulence and with aquatic vegetation (Castellanos & Landoni 1981; Scheibler & Ciocco 2011). Finally, representatives of the genus *Drepanotrema* live in bodies of water with rooted vegetation and floating or submerged leaves (Rumi 1991). Also, *A. nordestensis* inhabits a similar environment in streams with stagnant water (Tietze & De Francesco 2010).

The ostracod species encountered were previously recorded in both modern and quaternary sediments of the Pampean Region (e.g. Bertels & Laprida 1998; Laprida 2006; Laprida *et al.* 2009; 2014; Kinh *et al.* 2017; Ramos *et al.* 2019). Until now, studies on ostracods in the *Pampa Deprimida* have primarily focused on examining lacustrine and littoral environments, rather than fluvial environments like the one investigated in this study.

Regarding the ecological requirements of the species, L. cusminskyae inhabits permanent lakes and lotic environments, with oligo to mesohaline waters. It is used as an indicator of shallow brackish environments with high alkalinity and high bicarbonate and sodium content (Ferrero 1996; Laprida 2006; Ramón Mercau et al. 2014), but it lives in modern estuarine environments too (Flores et al. 2021). Cypridopsis vidua is a cosmopolitan species that inhabits different types of environments, including hypohaline and oligohaline, lentic and low energy lotic, both permanent and temporary, mainly associated with vegetated littoral zones (i.e. Meish 2000; Smith et al. 2003; Laprida 2006; Laprida & Valero Garcés 2009; Coviaga et al. 2018; Cusminsky et al. 2020). Danielopol (1991) considered that C. vidua has a low tolerance to poorly oxygenated waters, but Martins et al. (2010) have shown that they can live even under these conditions. Cyprideis salebrosa inhabits both permanent oligohaline (Laprida 2006) as unstable mesohaline and high salinity environments (e.g., marine-influenced or saline-enriched waters) where it is usually the dominant species (Coimbra et al. 2007; Laprida & Valero Garcés 2009; Márquez et al. 2016; Kinh et al. 2017; Flores et al. 2021). It lives associated with littoral vegetation in low and moderate-energy environments, such as streams and canals.

*Heterocypris similis* inhabits littoral zones of small permanent or temporary water bodies. It prefers environments with clay substrates devoid of vegetation or streams with silty sandy substrates, eutrophic to hypertrophic (Laprida 2006; Laprida & Valero Garcés 2009; D'Ambrosio 2014; Kihn *et al.* 2017). It is also considered indicative of hypohaline waters (Laprida 2006).

Illyocypris ramirezi inhabits a wide variety of environments, from permanent coastal lakes to lotic

bodies (Laprida 2006). Although it is euryhaline, it prefers a low-salinity environment (Laprida 2006; Ramón Mercau *et al.* 2012). It is dominant in lotic environments (rivers, streams, creeks, and springs) or lentic environments fed by streams or springs (Cusminsky *et al.* 2005; 2020; D'Ambrosio 2014; Coviaga *et al.* 2017). *Candonopsis* species are usually associated with hypo or oligohaline temporary shallow lakes. They are common in aqueous microenvironments retained in leaf litter with muddy bottoms (Laprida 2006; Moguilevsky & Whatley 1995). Finally, *C. incisa* lives in shallow temporarily vegetated environments (Laprida 2006).

## Changes in assemblages and palaeoenvironmental inferences

Late Pleistocene.—Between  $14040 \pm 50$  to  $12100 \pm 100$ 14C years BP, a low number of specimens belonging to *H. parchappii* and *S. meridionalis* made up the assemblages, and this coincides with previous results obtained in other sectors of the Salado River (Pisano & Fucks 2016). In the ostracod assemblages, six species were recognized with a low abundance of individuals.

During this period, the sediments of La Chumbiada Member began to accumulate, indicating the onset of fluvial sedimentation from pre-existing aeolian sediments. These sediments are associated with salt precipitation, including gypsum and calcium carbonate rosettes (Pommarés *et al.* 2021). The low diversity of gastropods and ostracods suggests the initial stage of a freshwater community establishment. This coincides with the first stage in the conformation of the Salado River, which emerged as a channel linking different deflation basins (Pommarés *et al.* 2021).

*Early Holocene.*—In this interval, the presence of *H. parchappii* (which tolerates wide salinity variations), *B. peregrina*, and hydrophilic species, such as *S. meridionalis* and *G. nodosaria*, allows us to infer the development of vegetated environments with fluctuating water levels in the floodplain of the Salado River. In other words, shallow flooding events occurred on the floodplain, and subsequently, its desiccation led to increased salinity. These were most notable in R30, where monospecific assemblages were numerous and covered a large part of the record. The dominance of *S. meridionalis* at the Vientos del Mar, along with *H. parchappii*, *G. nodosaria*, and *M. brasiliensis*, leads us to infer the presence of shallow flooded environments, with a preference for swamp terrestrial ecosystems.

The ostracod assemblages recorded a slight increase in the number of individuals in comparison to the Late Pleistocene assemblages. In Alberti-Pla, the abundance of individuals decreases towards the upper part of this unit, which ends with a barren level. The presence of *L. cusminskyae*, the most abundant species, together with *C. salebrosa*, *C. vidua*, and *H. similis* enables us to deduce the development of shallow meso- to oligohaline lakes or low energy streams with fluctuating salinities. These eventually dried up, as reflected in the barren level.

In the Ruta 30 locality, the sequence begins with levels where the abundances of dominant species, C. vidua and L. cusminskyae, fluctuated from low to slightly higher and formed autochthonous assemblages (low energy thanatocoenosis, Pisano et al. 2022), and ending with a barren level. From -95 cm, the assemblages begin to recompose with low densities of individuals until reaching the top of the sequence. This alternation allows us to infer the development of unstable environments subjected to variations in their water level, including their drying out and subsequent recovery. The almost continuous record of I. ramirezi indicates the presence of water flows, such as small streams or courses, over the alluvial plain, which may have connected different oligohaline and mesohaline environments, some even vegetated.

The Vientos del Mar assemblages were dominated by autochthonous assemblages of *L. cusminskyae. C. vidua, Candonopsis* sp., and *H. similis* could be associated with small oligo- to mesohaline water bodies. The presence of *Candonopsis* sp. further supports the predominance of ephemeral environments in the floodplains of this locality.

Late Holocene.—The gastropod assemblages displayed no significant differences, suggesting the presence of similar environmental conditions across the analyzed localities during this period. *Heleobia parchappii*, the dominant species, was mainly accompanied by *B. peregrina* in Alberti-Pla, and *S. meridionalis* in Ruta 30 and Vientos del Mar. This similarity in composition allows us to infer the development of homogeneous environments capable of sustaining more numerous populations. Either progressively more stable bodies of water, as in AP, or shallow flooded terrains associated with overflows of the Salado River in the floodplain at R30 and VM.

The ostracod assemblages showed changes in diversity and abundance that determined differences between AP and VM assemblages. In AP, the presence of *Candonopsis* sp. and *C. vidua* suggests that the early environments were hypo- to oligohaline, temporary, and shallow. Towards the middle section, the increase of *L. cusminskyae* and the disappearance of *Candonopsis* sp. indicate the development of stable environments that may have undergone salinity

variations that intensified towards the top of the section with the increase of *C. salebrosa*.

In R30, C. vidua dominates the assemblages with the presence of carapace of adult individuals (Pisano et al. 2022). Articulated carapaces are indicators of mass mortality events caused by unfavourable environmental changes (drying of water bodies, extreme decrease in dissolved oxygen, anoxia, hypersalinity, or sudden increase in temperature), followed by a rapid burial occurred after death to avoid disarticulation (Whatley 1988; Palacios-Fest et al. 1994; Boomer et al. 2003). Moreover, these levels coincide with assemblages which a high abundance of *H. parchappii*. Both species, H. parchappii and C. vidua, are eurytopic and can live in different types of environments; therefore this increase allows us to infer the establishment of water bodies subjected to abrupt changes such as desiccation and salinity increases.

In VM, the scarcity of individuals hinders the generation of reliable palaeoenvironmental inferences, potentially indicating the limited development of conducive freshwater environments or lack of preservation in the geological record.

*Recent Assemblages.*—Although there are no radiocarbon dates, we found remains of anthropic material (fragments of plastics, glass, and garbage in general) that confirm its modern age of this level.

The gastropod assemblages, with 11 species, had the highest species richness. Changes in the abundances of the majority species (H. parchappii, B. peregrina, and S. meridionalis) determined the environmental similarity between Alberti-Pla and Ruta30, and conversely the differences with Vientos del Mar. Therefore, in R30 a low-energy, shallow environment with abundant vegetation developed and dried out, causing the death of the organisms, except for H. parchappii. Subsequent floods allowed the restoration of the communities, although with less abundance. These environments were similar to those recognized in AP, but perhaps with fewer fluctuations in water levels since the populations remained stable throughout the sequence. In the lower assemblages of Vientos del Mar, H. parchappii was the only species present with low abundances, indicating higher salinity conditions or ephemeral environments. However, towards the middle and terminal sector, the freshwater communities are enriched by the sporadic presence of B. peregrina, A. nordestensis, and U. concentricus, indicating the development of unstable flooded areas in the floodplain with established vegetation, which would also explain the presence of M. brasiliensis.

The ostracod assemblages showed the same specific richness (nine species) as in Early Holocene assemblages but with maximum abundances in both AP and R30, while a few specimens were recovered in VM. Considering the species differential abundance and the taphonomic characteristics (Pisano et al. 2022) in the assemblages, it was possible to recognize heterogeneity of subenvironments in the floodplain in the Alberti-Pla and Ruta 30 localities. The autochthonous assemblages dominated by L. cusminskyae, along with H. similis and C. vidua, indicate the establishment of permanent oligohaline shallow lakes featuring vegetated littoral zones. These lakes experienced salinity increases, as evidenced by the expansion pulses of C. salebrosa. Additionally, there was the development of temporary water bodies, where Eucypris sp., Candona sp., and C. incisa thrived. The record of H. similis is associated with the presence of overflow flows or water currents, which may have connected the different temporary and permanent water bodies.

In Vientos de Mar, the assemblages registered low to very low abundance of individuals, where *C. salebrosa*, *L. cusminskyae*, and *C. vidua* presented the most continuous record. The population structure, reconstructed in a few assemblages, was dominated by adult individuals with incomplete population structure or by juveniles, forming taphocoenoses (Pisano *et al.* 2022). The species indicate the presence of oligo- to mesohaline water bodies, with salinity changes. However, these implications are speculative because they are transported assemblages, so they are not good environmental indicators because they may be out of their natural context.

#### *Palaeoenvironmental evolution of the Pampa Deprimida from a regional perspective*

Different biological and geological proxies allowed palaeoenvironmental and palaeoclimatic reconstruction in the Pampean plain along the Holocene (i.e., Stutz *et al.* 2014; De Francesco *et al.* 2022; Vilanova *et al.* 2022). In this paper, we analysed the response of molluscs and ostracods as a function of palaeoenvironmental changes and related them with previous regional analyses.

Pommarés *et al.* (2021) interpreted wet conditions associated with greater water availability, which allowed the accumulation of fluvial sediments in a low-energy environment during the Early Holocene (between 10000–8000 years BP) in the Salado River Basin. In addition, during the Middle Holocene (between 8000 and 3000 years BP), the authors identified a period of water deficit coinciding with the thermal maximum. Streams and shallow lakes experienced a reduction in flow, associated with intense evaporation (due to low precipitation and/or increased temperature) that caused the precipitation of gypsum and carbonates characteristic of Gorch Mb. In addition, a loessic sequence was observed in the middle Salado River Basin from 6.5 ka years BP (Ramos et al. 2018) that would confirm this arid period. These dry conditions favoured dune formation by mobilization of pre-existing eolian and fluvial sediments to the west of the study area. In addition, during this period, there was a depositional hiatus in the Salado River Basin since there are no ages 8 to 3 ka BP, despite the numerous chronological dating made (Mari et al. 2013; Fucks et al. 2015; Pommarés et al. 2021). The cause of this hiatus remains unknown, and it is unclear if it resulted from a disruption in fluvial sedimentation due to a drop in water levels, erosion, or extreme/adverse environmental conditions that hindered the development of freshwater communities in the floodplain.

In other sectors of the Pampean plain (Figure 1. A), such as the Luján River (northeastern Buenos Aires, Pampa Ondulada), Prieto et al. (2004) recognized a decrease in water levels alternating with periods of flooding, between 10500 to 9000 years BP, and a greater reduction in flow between 9000 to 7000 years BP. In the Pampa Interserrana, Steffan et al. (2014) recorded the development and desiccation of marsh environments between 10000 to 6500 years BP. Prieto (1996) identified a period of non-deposition between 7000 to 3000 yr BP in the Tapalqué Stream, similar to what occurs in the Salado River, associated with an erosive period. While in shallow lakes, dry climatic conditions were established during the Early and Middle Holocene, resulting in unstable environments and periods of total desiccation (Stutz et al. 2014; De Francesco et al. 2022). Based on the gastropod and ostracod assemblages, this study's environmental reconstructions are consistent with previous interpretations. They suggest the development of restricted ecosystems characterized by fluctuations in water levels, which led to the desiccation or retraction of shallow lakes and the disruption of fluvial sedimentation in the Salado River during the Early Holocene.

The transition between the Middle and Late Holocene presented a climatic scenario associated with climatic regionalization, resulting in greater climatic heterogeneity during this period (Stutz *et al.* 2014). These changes led to the formation of climatic gradients of temperature and precipitation in the Pampean region, which caused different responses in the proxies used for palaeoenvironmental reconstructions (De Francesco *et al.* 2022). It has been observed that the southeastern of the Pampean plain experienced higher relative aridity between 5000 and 4000 years BP (Zarate *et al.* 1998; Prieto *et al.* 2004). On

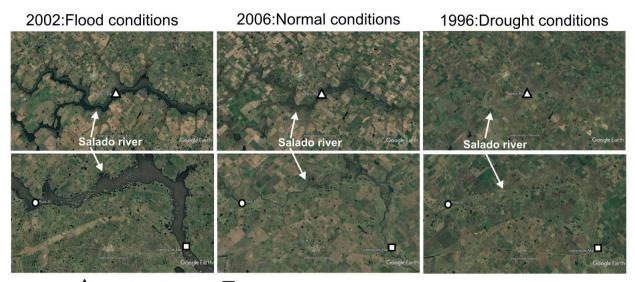
the other hand, Steffan *et al.* (2014) noted an increase in freshwater mollusc diversity and occurrences of flooding events or increased humidity between 5 and 3.5 ka in the *Pampa Interserrana*.

From 3 ka to the present, Pommarés et al. (2021) recognized a gradual return to humid conditions, manifested by the presence of fluvial deposits of the Puente Las Gaviotas Mb. This increase in humidity in the study area during the Late Holocene was documented in other zones of the Pampean region, too (Stutz et al. 2014; De Francesco et al. 2013; 2022). From the use of different proxies, there is a consensus regarding a regional increase in precipitation that favoured the installation of a humid-subhumid climate in various sectors of the Pampean region during the Late Holocene (Stutz et al. 2014; De Francesco et al. 2013; 2022) and that promoted the development of different freshwater environments. In the study area, we recorded an increase in the abundance and diversity of freshwater gastropods and ostracods during the Late Holocene, which are associated with the development of different unstable aquatic environments that favoured the proliferation of assemblages.

Iriondo & Kröhling (2007) estimated that from 1850 onwards, an excess water period began to dominate the Pampean Region. In the studied area, this coincides with the process of pedogenesis that develops in modern sediments, which accumulate on the floodplain during flood events and then remain in the open during periods of normal or low flow. Furthermore, this is consistent with species enrichment and variations in assemblage composition associated with still waters vegetated environments that reflect the establishment of current climatic conditions. Therefore, changes in rainfall regimes originated the heterogeneity of environments that develop in the floodplain of the Salado River and neighbouring shallows lakes during extreme flood and drought events (Figure 7).

From the analysis of mollusc and ostracod assemblage trends and the changes experienced by different environments, we can infer that fluctuations in precipitation directly impact water levels, giving rise to cycles of drought and flooding in the Pampean Region (Quirós *et al.* 2002; Bohn *et al.* 2016) and the Salado River Basin (Pisano *et al.* 2019), thereby affecting both the main course and the various subenvironments within the area.

With the analysis of various parameters in the populations of gastropods and ostracods, we were able to discern the development of multiple environments arising from changes in precipitation regimes. During periods of water deficit, fluctuations and increases in salinity occurred, brought about by the recession of water bodies or the expansion of hydrophilic and



Reference: Alberti Pla ORuta 30 Uvientos del Mar

*Fig. 7.* Changes in the main course of the Salado River and its floodplain under three water situations: during an excess event (e.g. flood, 2002), normal conditions (e.g. 2006), and with a deficit (e.g. drought, 1996) of precipitation in the area. The upper images correspond to Alberti Pla (triangle), and the lower ones to Ruta 30 (circle) and Vientos del Mar (square). Source: Google Earth, DigitalGlobe.

terrestrial environments in the floodplain, as observed during the Late Pleistocene and Early Holocene. This phenomenon is related to the patterns identified in the assemblages, where rhythmic variations were detected, attributed to a limited substitution rate due to the low number of freshwater species and the increased abundance of hydrophilic individuals.

From the Late Holocene onwards, the increment in precipitation led to a rise in water levels in the main course, greater heterogeneity environmental, and enhanced interconnectivity among them, as currently observed. It is reflected in an increased species turnover, as the assemblages were predominantly enriched with freshwater species and experienced an increase in individual abundance.

#### Conclusions

The analysis of gastropods and ostracods provided insights into the characteristics and evolution of diverse environments in the Salado River Basin throughout the Holocene. The microfossil record began incipiently in the area during the Late Pleistocene, underwent changes in composition and abundance during the Holocene, and acquired similar characteristics to the present during the Late Holocene. In the case of gastropods, the dominance of *H. parchappii* strongly influenced the structure of the assemblages. In the ostracod assemblages, although *L. cusminskyae* was mainly dominant, the contributions of other species also played an important role.

The rainfall regime, which influenced the different freshwater environments of the region and directly affected water levels in the Salado River and the shallow lakes formed in its floodplain during the Holocene, influenced the development of different paleoenvironments in the Pampean region. During periods of water scarcity (due to low precipitation and/or increased temperature), such as the Late Pleistocene and Early Holocene, we identified the presence of unstable, ephemeral, swampy, or terrestrial freshwater environments, experiencing episodes of desiccation and fluctuations in salinity. This is directly related to the trends observed in the assemblages, where a slight rhythmic variation was observed, attributed to low abundance and low species replacement.

Starting in the Late Holocene, the rise in precipitation unveiled a heightened diversity of environments, encompassing both permanent and ephemeral habitats, and facilitated increased interconnectivity among them. This diversity of environments led to an abrupt increase in the number of gastropod species, with a rising trend in recent assemblages of both proxies. Therefore, more favourable conditions have developed for the proliferation of more abundant and diverse populations of gastropods and ostracods.

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