Journal of Archaeological Science: Reports 57 (2024) 104636

Contents lists available at ScienceDirect

Journal of Archaeological Science: Reports

journal homepage: www.elsevier.com/locate/jasrep

New archaeochemical insights into Roman wine from Baetica

Daniel Cosano [a], Juan Manuel Román [b], Dolores Esquivel [a], Fernando Lafont [c], José Rafael Ruiz Arrebola [a,*]

[a] Organic Chemistry Department, Instituto Química para la Energía y el Medioambiente (IQUEMA), Sciences Faculty, Patricia Unit for R&D in Cultural Heritage, University of Cordoba, Spain

[b] Delegation of Historical Heritage, Museum of the City, Carmona, Spain

[c] Centralized Research Support Service (SCAI), Mass Spectrometry and Chromatography Unit, University of Cordoba, Spain

ARTICLE INFO

Keywords:
Roman wine
ICP-MS
HPLC-MS
Polyphenols

ABSTRACT

Although ancient wines adsorbed on vessel walls or their remains can be identified with the help of specific biomarkers, no analysis of such wines in the liquid state appears to have been reported to date. Therefore, the 2019 finding in a Roman mausoleum in Carmona, southern Spain, of an ash urn roughly 2000 years old, containing a reddish liquid, was rather exceptional and unexpected. An archaeochemical study of the liquid allowed it to be deemed the oldest ancient wine conserved in the liquid state. The study used inductively coupled plasma mass spectrometry (ICP-MS) to determine the chemical elements in the mineral salts of the wine, and high-performance liquid chromatography-mass spectrometry (HPLC-MS) to identify the polyphenols it contained. The mineral salt profile and, especially, the detection and quantification of some typical polyphenols, allowed the liquid to be identified as white wine.

1. Introduction

Rehabilitation work in a house in Carmona, southern Spain, in 2019 unearthed a collective tomb belonging to the western necropolis of Carmo, an ancient Roman city in the Baetic region. The enclosure, 3.29 m long × 1.73 m wide, was probably a family tomb. The maximum height of the vaulted ceiling was 2.41 m (Fig. 1a and b). The mausoleum was dated back to the early 1st century CE (Roman et al., 2019). The left and right wall of the entrance had eight loculi (niches) in all. Two of them were empty while the other six each contained an ash urn, the urns holding cremation remains and various objects typically used in burial rituals and offerings. The urn in niche 7 contained an unguentarium and several amber beads that were examined in previous work (Cosano et al., 2003a; Cosano et al., 2023b). On the other hand, the urn in niche 8, preserved in excellent condition, was a glass *olla ossuaria* with M−shaped handles (Fig. 1c). This urn was inside an ovoid lead case with the flat lid bulged in the middle (Fig. 1d). Inside the urn was a reddish liquid (a total of 5 L, Fig. 1e), which was thought to be part of the original contents, together with cremated bone remains. Given the religious significance of wine in the ancient Roman world, where it was highly symbolic and closely related to burial rituals, it is unsurprising to find vessels that might have originally contained wines among burial furnishings. Consequently, the reddish liquid found in the urn might be wine or vestiges of wine decomposed over time. In fact, according to Vaquerizo Gil (2023), wine was usually placed together with water and foods such as honey among burial furnishing to accompany the deceased in their transition to a better world.

In Roman times, preventing wine decay was one of the greatest problems faced by winemakers. However, they succeeded in extending the useful lifetime of wine by using various additives; one of the most commonly used of which in the Baetic region was gypsum (calcium sulphate dihydrate, $CaSO_4 \cdot 2H_2O$). Another way of extending the lifetime of wine in Roman times was by adding cooked musts containing large amounts of sugars to increase the alcohol content. Alternatively, the wine was supplied with sodium chloride, possibly to enhance its taste. Salt is also an effective preservative and stabiliser for wine. Fino wines currently produced in the Jerez designation of origin are probably the most like those originally obtained in Roman *Baetica*.

So far, all studies aimed at the chemical characterisation of Roman wines —or ancient wines in general— have relied on analyses of absorbed remains (carboxylic acids and polyphenols, mainly) in various types of vessels (Garnier et al., 2003; Blanco-Zubiaguirre et al., 2019; Briggs et al., 2022), but never on liquids. In any case, assuring that a sample is an ancient wine requires identifying specific biomarkers,

* Corresponding author.
E-mail address: qo1ruarj@uco.es (J.R. Ruiz Arrebola).

https://doi.org/10.1016/j.jasrep.2024.104636
Received 2 April 2024; Received in revised form 28 May 2024; Accepted 10 June 2024
Available online 16 June 2024
2352-409X/© 2024 The Author(s). Published by Elsevier Ltd. This is an open access article under the CC BY license (http://creativecommons.org/licenses/by/4.0/).

including polyphenols. The presumed oldest wine preserved in the liquid state is 'Speyer wine bottle', which is thought to be about 1700 years old. It is held in a stoppered bottle in the Historiche Museum der Pfaltz (https://museum.speyer.es.de/startseite) and was found in a tomb excavated in Speyer, a German city, in 1867. The bottle is suspected to contain wine from the year 325 CE, but this assumption has never been confirmed by chemical analysis.

In addition to water and ethanol, wine contains organic substances such as carboxylic acids, sugars, polyphenols, and various aromatic compounds. Wine also contains mineral salts, which are highly influential on its quality. Given that the liquid contained in the ash urn was about 2000 years old, determining its elemental composition was of utmost importance because any organic matter in it should either be present in very low contents or have disappeared altogether. Therefore, the primary aim of this work was to ascertain whether the liquid in the urn was wine or decayed wine. For this purpose, we determined the polyphenolic markers and mineral salts for comparison with the composition of current wines. We also sought ethanol, which might still be present in the wine given the good condition of the tomb and its contents.

2. Archaeological context

Carmona is a city in the Guadalquivir valley, Western Andalusia, 30 km west of Seville. Under Roman domination in the 1st and 2nd centuries A.D., the city became an important municipality (the sixth largest Baetic location in terms of population and land area) (Caballos, 2021) enjoying abundant wheat and olive oil resources. Carmona still retains some buildings from that period, including the Cordoba and Seville gates, an amphitheatre, and a necropolis that is the largest and best conserved on the Iberian Peninsula.

Although a large portion of the ancient necropolis of Carmo lies within the archaeological ensemble of Carmona, the Roman cemetery was larger, so it is not unusual to uncover burial complexes while excavating buildings for construction work in the area. In 2019, rehabilitation work at 53 Sevilla St. exposed the access shaft to an underground enclosure. Preliminary inspection confirmed that it was the chamber of an unlooted Roman mausoleum that had undergone little alteration since it was built. The chamber was topped by a well-preserved vault of voussoir stones and decorated with painted geometric motifs. There were eight carved niches in the walls: six contained ash urns and funerary objects, including a glass mosaic bowl in perfect condition.

The access shaft, which was rectangular and 1.03 m × 0.98 m, was

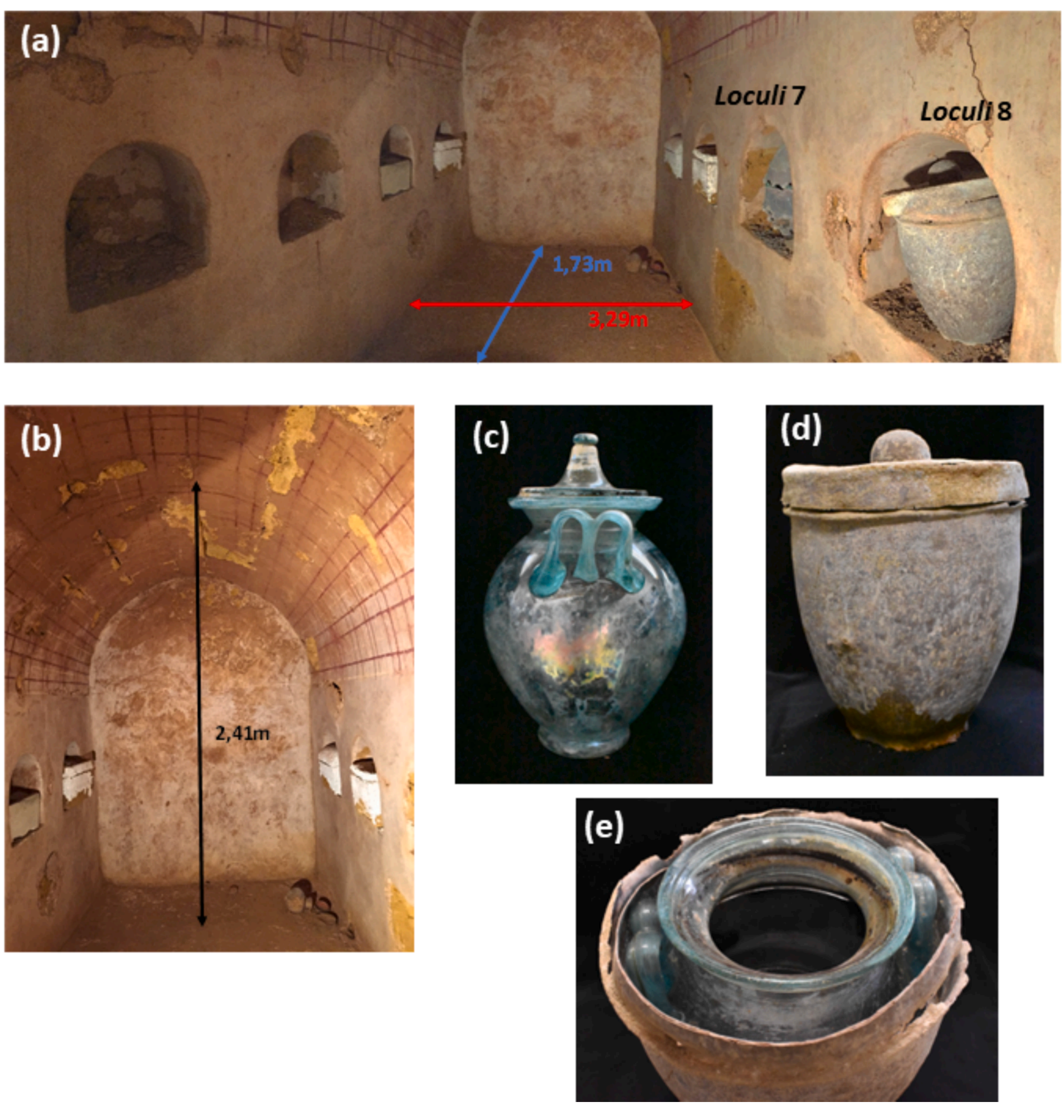


Fig. 1. (a), (b) Funeral chamber. (c) Urn in niche 8. (d) Lead case containing the urn. (e) Reddish liquid contained in the urn.

excavated in the rock and heightened with an ashlar ring reaching ground level at the top (Fig. 2). The doorway leading into the structure was opened in the northern wall; it was 1.80 m high and finished in the manner of a semicircular arch. The doorway was connected to the chamber by a walkway 1.46 m long × 0.70 m wide, also excavated in the rock that was intended to strengthen the zone where the burial monument, possibly a tower, was erected on the underground section.

The chamber was also rectangular (3.29 m long × 1.73 m wide), and the ceiling was a canyon vault with a maximum span of 2.41 m (Fig. 1a). The western and eastern walls contained four niches each. All surfaces inside the chamber were coated; the floor and walls with *opus signinum*, and the vault with reddish lime. The coatings were decorated with a series of geometric motifs that was seemingly left unfinished but has remained in good condition up to the ceiling. The vault was painted with red and ochre interwoven lines forming a grid containing empty inner spaces. The top of the northern wall was decorated with a single, lobulated motif in red that was only partially conserved. Unlike the shaft and the walkway, which were carved into the rock, the presence of chipping suggested that the chamber walls were built with ashlars and the vault was constructed with large, dry-stacked voussoirs of alcoriza stone (a local sandstone variety). This construction method must have required the prior digging of a large open-air pit to erect the burial structure.

Two of the eight niches found in the chamber (L-1 and L-2) were empty, probably because they were placed there, whereas the other six contained one urn each (Fig. 3). The urn found in L-3 was a limestone box while those in L-4, L-5, and L-6 were carved in local sandstone. The urns in L-7 and L-8 were made from glass and were housed in lead cases. All urns contained cremated bone remains from a single individual (a female in L-3, L-5, and L-7, and a male in L-4, L-6, and L-8). The urns in L-4, L-5, and L-6 were partially coated with a gypsum layer and the gypsum in the former two was used to carve the names of the deceased: *Hispanae* and *Senicio* (Limón & Román, 2022).

Fig. 3. The eight niches found in the chamber.

Fig. 2. Access to the tomb.

In addition to urns and bone remains, the chamber contained other objects used in burial rituals, some in the urns themselves. Thus, the lid to the urn in L-3 had been deliberately used to break a glass vessel, possibly an unguentarium, seemingly as part of a ritual that has never been documented elsewhere. Inside that urn were an iron ring and a glass unguentarium. Between the female bone remains in urns in L-3, L-5, and L-7 were burnt fragments of ivory sheets that might have coated small boxes accompanying the deceased in the pyre. On the chamber ground under urn L-5 were a ceramic jar, a thin-walled ceramic bowl, and a peculiar glass mosaic plate that must have contained food and drink offerings. Next to the urn in L-7 was a glass bowl containing a bag made of plant-based fabric (possibly flax or hemp) on the bony remains. The bowl contained three round pieces of Baltic amber (Cosano et al., 2023b). Next to the bag was a tightly sealed unguentarium made of rock crystal (hyaline quartz) carved like an amphorisk that contained solidified remains of the perfume it once held. The unguentarium stopper was sealed with pitch or tar and the solidified remains allowed, for the first time, the chemical composition of a Roman perfume to be established: patchouli, an essential oil (Cosano et al., 2023a). The fact that the burial chamber was almost tightly sealed allowed other organic materials such as fabric vestiges stuck to various objects and other remains to be well-preserved (Gleba and Román, 2024). On a large portion of the urn in L-7 and its lid was stuck a fabric film suggesting that the vessel was wrapped in cloth before it was placed in its protective lead case. Finally, the urn in L-8 not only contained bone remains and a gold ring carved with Jano Bifronte, but it was also filled to the brim with a reddish liquid. Despite the initial surprise, we immediately concluded that the liquid could not have reached the inside of the urn through flooding or leakage in the burial chamber, nor through condensation, especially when the inside of the urn in the adjacent niche, L-7, was under identical environmental conditions but completely dry.

The exceptionally good conservation conditions of the ensemble allowed a valuable trousseau to be recovered. The trousseau was comprised of high-quality pieces typical of high-standing owners; organic materials such as liquids or solidified remains from a perfume; fabrics and amber. All these elements are rarely preserved, so they offered a unique opportunity for study. Based on the materials it contained, and structural similarities with other burial places, the tomb was probably used during the first half of the 1st century CE.

3. Materials and methods

The analysis for mineral salts was performed on a PerkinElmer NexION 350X inductively coupled plasma mass spectrometer, using samples that were previously dissolved in milliQ water containing nitric acid. Elements were quantified relative to the internal standard with the nearest atomic weight in the mix supplied by the instrument's manufacturer.

Phenolic compounds were determined by using an HPLC/MS instrument equipped with a Sciex 7500 QTrap Triple Quadrupole analyser. Samples were diluted 25:1 with HPLC mobile phase, filtered through paper of 0.22 μm pore size and directly injected onto the HPLC/MS instrument in 2 μL aliquots. The HPLC conditions were as follows: compounds were separated on a C18 phase column (10 cm long × 2.1 mm i.d., 1.6 μm particle size), using a mobile phase consisting of A (water containing 0.05 % formic acid) and B (methanol containing 0.05 % formic acid), and was used in the following gradient: t = 0 min, 95 % A; t = 12 min, 100 % B; and t = 15 min, 100 % B. The mobile phase flow-rate was 0.40 mL/min and the column temperature 40 °C. Mass detection was done in the negative electrospray MRM mode, using a minimum of two MRM transitions for each compound. Compounds were quantified by external calibration against standards containing concentrations from 0.001 to 0.5 mg/L (linearity criteria: $r^2 > 0.99$ and individual RSD < 15 %).

Ethanol was determined by GC/FID on a PerkinElmer 560 GC/FID instrument. The sample was diluted 1:1 with water/0.01 % n-propanol and directly analysed by GC on a Supelcowax capillary column (60 m × 0.32 mm i.d.). The injected volume was 1 μL and used in the split mode (1:20). The column temperature was isocratic (40 °C) and the gas (hydrogen) passed at a rate of 2 mL/min. Ethanol was quantified against 1-propanol as internal standard.

4. Results and discussion

4.1. Analysis for elements and mineral salts in the reddish liquid

The reddish liquid in the urn had a pH of 7.5, which is much higher than that of fino wines currently produced in Montilla-Moriles and Jerez (two designations of origins coinciding with the former Baetic region), with pH values ranging from 3.0 to 3.5. The high pH of the liquid was suggestive of strong decay from the potential wine it once was. Also, the proportions of carbon, nitrogen, and sulphur (0.46 %, 0.21 %, and 0.0037 %, respectively) suggested the presence of little organic matter, which was quite plausible for strongly decayed wine by effect of the mineralisation of organic compounds.

Mineral salts in wines come largely from the soil where vines are grown and reach wine through grapes. Salt concentrations are typical of each wine (Kment et al., 2005). The presence of certain metals in the salts can result from impurities reaching the wine during production but particularly through anthropogenic contamination (Pohl, 2007). Table 1 shows the results of the multi-element analysis conducted by ICP-MS. The elements found were classified into three groups according to concentration, namely: (a) elements at > 0.4 g/L. (viz., Na, K, Mg, and Ca; G-I in Table 1); (b) elements spanning the concentration range 0.1 − 75 mg/L (B, Al, Si, Ti, Fe, Cu, Zn, Rb, Sr, Br, and Pb; G-II in Table 1); and (c) trace elements (Li, P, V, Cr, Mn, Co, Se, Zr, Nb, Pd, Sb and others, at even lower concentrations; G-III in Table 1). The elements in group G-I are typically found in current wines, albeit at higher concentrations (Kunkee & Eschnauer, 2003). Also, K is the element present at the highest levels, whereas Ca and Mg occur at much lower, but similar, levels, and Na has a concentration lower than one-half that of Mg. Because they influence its sensory properties, all metals in wine play a crucial role (Pohl, 2007). There are few reported mineralogical composition data for archaeological wines. Arobba et al. (2014) examined the contents of an amphora found among the remains of a shipwreck in the Tirreno sea occurring in the 1st century BCE The amphora was intact, and isotopic and palaeobotanical analysis allowed the authors to establish that the original content was an oenological product produced using ancient techniques. The study was conducted on a liquid sample extracted from the amphora, which was filled with seawater. Therefore, the organic matter of the residue was highly deteriorated. The elemental analysis conducted by these authors was like that obtained by our study, with low carbon and nitrogen contents, indicative of the mineralisation of organic products. The analysis of the mineral salts reveals a composition of many elements similar to that of our sample. The elements found at g/L levels are the same, except for potassium, which in our case appears at a higher concentration. The differences in the concentrations of elements found at levels of mg/L or μg/L can be correlated with the leaching occurring from the containers, one ceramic and one glass, or the contribution of seawater, or the remains contained in the cinerary urn.

The wines produced in Roman Baetica were made as described by Columela in Book XII of his *De Re rustica* (Columela, 1824). As concluded by Tchernia and Brun (1999) from the wine they made in strict accordance with Roman tradition, the most similarly obtained among current wines are probably fino wines from the Jerez designation of origin. For this reason, and based on geographic location, the mineral profile of the reddish liquid is comparable to that of current sherry wines from Jerez, fino wines from Condado de Huelva (López-Artíguez et al., 1996; Paneque et al., 2009; Álvarez et al., 2012), and fino wines from Montilla-Moriles − a designation of origin not far from Carmona (Álvarez et al., 2007a; Álvarez et al., 2007b). The major elements in the wines of today were also present in the reddish liquid. The most abundant element in fino wines is K, which was present in concentrations 2 − 3 times higher in the reddish liquid but is rarely more than 30 % greater in current wines. This high amount could be related to the presence of cremation remains in the urn. The potassium content in an adult person ranges between 120 and 140 g (John, 2015). This element is non-volatile and remains in various salt forms in the ashes after cremation. The high solubility in water, or in wine, of potassium salts could justify this elevated value. The Ca, Mg and Na concentrations in the reddish liquid also exceeded those of today's wines. In addition, the elements typically present at levels of a few milligrams per litre in current wines (B, Ti, Fe, Rb, Ba, Cu, Zn, and Pb) were found at similar levels in the target liquid. On the other hand, the Sr, Al and, especially, Si levels were higher in the liquid. The previous differences can be ascribed to leaching from the urn glass as well as the cremated bones also contained in the urn. As a rule, Roman glass contained around 70 % SiO_2, 15 − 20 % Na_2O, and CaO and Al_2O_3 in proportions from 2 % to 9 %, in addition to other metal oxides

Table 1
Mineral salts composition of the reddish liquid contained in the ash urn.

Element (G-I)	Concentration (g/L)	Element (G-II)	Concentration (mg/L)	Element (G-III)	Concentration (μg/L)*
Na	0,43	B	3,00	Li	14
K	3,28	Al	12,41	P	14
Mg	0,94	Si	70,43	V	43
Ca	1,30	Ti	1,07	Cr	23
		Fe	2,45	Mn	94
		Cu	0,12	Co	51
		Zn	0,29	Se	14
		Rb	2,51	Zr	26
		Sr	9,32	Nb	17
		Ba	0,13	Pd	17
		Pb	0,14	Sb	16

* Other elements (concentration < 0,1 μ/L): Ga, As, Y, Mo, Cs, La, Ce, Nd, Eu, W, Pt, Au, Th).

such as MgO, P_2O_5, K_2O, Fe_2O_3, and MnO at levels around 1 % or lower (Velo-Gala et al., 2019; Zanini et al., 2019). Therefore, the high concentrations of Si, Na, and Al in the reddish liquid were probably due to leaching from the urn glass over 2000 years of contact.

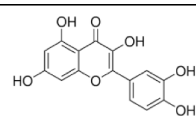
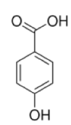
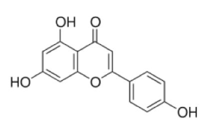
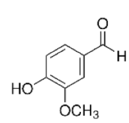
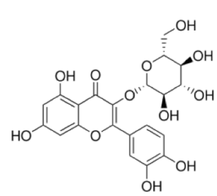
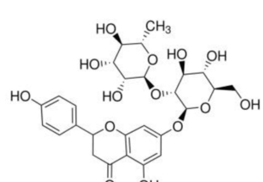
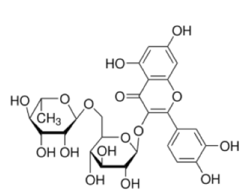
4.2. Analysis of the reddish liquid for biomarkers

Polyphenols are secondary metabolites of plants encompassing a large variety of chemical compounds with widely variable structures that contain at least one benzene ring holding one or more hydroxyl substituents in addition to a side chain. Grapes contain polyphenols in amounts dependent on environmental factors such as climate and soil nature, and grape variety, ripeness stage, and condition (Garrido and Borges, 2013). The polyphenol composition of wine is closely related to that of the grapes, but it additionally depends on the winemaking method used (Fang et al., 2008). Several polyphenols are wine biomarkers, so their presence in a sample confirms whether it is wine or decayed wine. In fact, such biomarkers are currently used to authenticate wines (Álvarez et al., 2007b). This led us to examine the reddish liquid in the ash urn in order to ascertain whether it contained polyphenols. To date, few studies have been conducted on polyphenols in archaeological wine remains, and all have examined the vessels that contained them rather than a liquid (Garnier et al., 2003; Petit-Dominguez et al., 2003; Blanco-Zubiaguirre et al., 2019).

On average, grape pulp has a polyphenol content of 20 – 170 mg/g, most of it in the form of phenolic acids. Anthocyans are the most concentrated polyphenols in grape skin (500 – 3,000 mg/kg), which additionally contains tannins and benzoic acids at the milligram per kilogram level. On the other hand, grape seeds only contain tannins, at concentrations from 1,000 to 6,000 mg/kg [25]. Wine contains polyphenols at much lower, widely variable concentrations. For example, the polyphenol concentrations in Jerez and Montilla-Moriles wines are usually as low as a few milligrams per litre or even lower (Benítez et al., 2003).

HPLC-MS analysis allowed some polyphenol biomarkers to be identified in the reddish liquid, which suggests that it was originally wine but was now highly decayed. Table 2 shows the specific polyphenols found and their concentrations. Five were flavonoids that are present in both white and red wines. The two most concentrated flavonoids in the liquid were quercetin and apigenin, which are also those found in the highest concentrations in today's wines (Flamini et al., 2013). White and red wines additionally contain substantial amounts of naringin and rutin (Artiushenko & Zaitsev, 2023). The fifth flavonoid, quercetin-3-glucoside, is a structural derivative of quercetin present in most wines

Table 2
Polyphenols composition of the reddish liquid contained in the ash urn and chemical group to which they belong.

Compound	Formula	Group	Concentration (mg/L)
Quercetin		Flavanoids	0,09
4-Hydroxibenzoic acid		Phenolic acids	0,08
Apigenin		Flavonoids	0,05
Vanillin		Phenolic acids	0,04
Quercetin-3-glucoside		Flavonoids	0,02
Naringin		Flavonoids	0,01
Rutin		Flavonoids	0,01

(Simonetti et al., 2022). Conversely, 4-hydroxybenzoic acid and vanillin are two benzoic acids found in all types of wine (Garrido and Borges, 2013).

The previous results, which suggest that the liquid in the ash urn was decayed wine, were compared with the composition of current wines. Thus, we determined the polyphenol composition of a fino wine from the Montilla-Moriles designation of origin produced in Doña Mencía, a location in southern Cordoba near major Roman archaeological sites such as Almedinilla, Priego de Córdoba, or Torreparedones. We also analysed two other fino wines from Sanlúcar de Barrameda and Jerez. Table 3 shows selected polyphenols found. All seven polyphenols detected in the reddish liquid were also present in the wine from Doña Mencía; however, rutin was not present in those from Sanlúcar de Barrameda and Jerez, and quercetin-3-glucoside was also absent from the latter. Although, as noted earlier, the polyphenols present in wine ultimately depend on the grape variety and winemaking method used, most of the polyphenols found in the current wines examined – or even all, in some cases – were also present in the reddish liquid.

The colour of an ancient wine can be determined via another polyphenol: syringic acid. This acid forms by decomposition of the main pigment in red wines, namely, the anthocyanin malvidin-3-glucoside, which has been found in many remains from amphorae that once held red wine (Guasch-Jané et al., 2004, Pecci et al., 2017; Fujii et al., 2019). Its absence from the reddish liquid indicates that it was white wine (Guasch-Jané et al., 2006), which is consistent with the writings of Columela about white wine production in the Baetic region. However, the colour of Roman wine is a topic that has been discussed in the literature (Tchernia & Brun, 1999; Thurmond, 2017). In fact, in his *Natural Historia*, Plinio (2010), Pliny distinguishes up to four types of wine based on their colour: *albus* (pale white), *fulvus* (reddish-yellow), *sanguineus* (blood red), and *niger* (black). The wine acquires these colours after the fermentation process and through its storage. Thus, over time, wine becomes darker due to oxidation reactions (Tchernia & Brun, 1999). Some varieties of ancient red grapes produced a dark-coloured must, and these could be the origin of the black wines mentioned by ancient authors (Brun, 2004). In general, the red colour of a wine comes from the maceration of the must with the skin and other solid residues of the grape, due to the release of colouring compounds called anthocyanins, which belong to the tannin family. The objective of maceration in modern winemaking is to achieve a red hue in the wine. The maceration time can last from a few hours to several days or weeks, depending on the tannin content of the grapes and the desired final colour (Robinson, 2006). This modern concept of maceration does not appear in classical sources. This, coupled with the recommendation of agronomists of the time, such as Columella, to transfer the must to dolia immediately after pressing, has generated the idea that Roman wines were essentially white (Brun, 2003; Harutyunyan & Malfeito-Ferreira, 2022; Aguilera Martín et al., 2023). However, according to van Limbergen & Komar (2024), this interpretation is subject to current standards of differentiation between red and white wines, something that did not exist in Roman times. Ancient sources also do not explicitly state the need to remove solid remnants after grape pressing. This fact, coupled with the identification of *Vitis* pollen by some authors (Arobba et al., 2014), could indicate a certain level of maceration in some wines. In Baetica region, wine production followed the guidelines set by Columella, who, as mentioned previously, recommended immediately transferring the must without allowing for maceration. This fact, coupled with the absence of syringic acid in our reddish liquid, makes it plausible that the wine contained in the urn was white. Chemical, physicochemical, or leaching processes from the solid residues contained in the urn may be responsible for the final reddish colour.

5. Conclusions

The exceptional finding in an unlooted Roman mausoleum in Carmona, southern Spain, of an ash urn containing cremated human remains and a reddish liquid that had remained intact for about 2000 years was a unique opportunity to examine the chemical composition of the liquid to ascertain whether it was the oldest wine in the world. The mineral salt composition of the liquid was quite similar to fino wines currently produced in the former Baetic region. The presence in the liquid of increased concentrations of some chemical elements can be ascribed to leaching from the urn glass and cremated remains and suggests that the liquid might be decayed wine. In fact, its elemental analysis revealed a carbon content of only 0.46 %, which suggests strong mineralisation of the original organic compounds. Analysing the liquid for polyphenols typically present in current wines allowed further insight into the identity of the liquid. The results confirmed with a high certainty that the liquid was wine and, more specifically, white wine, an assumption strengthened by the presence of ethanol at very low concentration. However surprising, this result is consistent with the very good preservation condition of the studied mausoleum. The use of wine in Roman burial rituals is well-known and documented. Therefore, once the cremated remains were placed in it, the urn must have been filled with wine in a sort of libation ritual in the burial ceremony or as part of the burial rite to help the deceased in their transition to a better world. The results obtained in this work strongly suggest that the reddish liquid in the ash urn was originally wine that decayed with time, and that it was about 2000 years old, and hence the oldest wine found to date.

CRediT authorship contribution statement

Daniel Cosano: Methodology, Investigation. **Juan Manuel Román:** Writing – review & editing, Writing – original draft, Conceptualization.

Table 3
Polyphenols determined in current wines. All values are expressed in ppm.

Polyphenol	Montilla-Moriles	Sanlúcar de Barrameda	Jerez
Apigenin*	0,04	0,01	0,01
Caffeic acid	0,06	0,92	0,14
Caftaric acid	0,31	24,69	21,23
Catechin	0,07	0,06	0,05
Ferulic acid	0,04	0,08	–
Gallic acid	3,36	2,61	3,63
4-Hydroxybenzaldehyde	0,05	0,17	0,21
4-Hydroxybenzoic acid*	0,63	0,37	0,58
Vanillin*	–	0,06	0,04
Hydroxytyrosol	4,07	4,22	3,37
Quercetin*	2,54	0,13	0,06
Quercetin-3-glucoside*	0,02	0,02	–
Rutin*	0,01	–	–
Vanillic acid	1,48	1,18	1,23
Naringin*	0,02	0,01	–

* * denotes the polyphenols detected in our reddish liquid.

Dolores Esquivel: Writing – original draft, Investigation. **Fernando Lafont:** Writing – original draft, Methodology, Investigation, Conceptualization. **José Rafael Ruiz Arrebola:** Writing – review & editing, Supervision, Investigation, Conceptualization.

Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Data availability

No data was used for the research described in the article.

Acknowledgements

The authors are grateful to Research Group PAI FQM-346, IQUEMA and the Central Research Support Service (SCAI) of the University of Córdoba for their help with the experimental part. They are also grateful to the archaeologists Jacobo Vázquez Paz and Adrián Santos Allely, co-directors of the excavation; Research Group PAI HUM-650 of the University of Seville; and the owners of the excavated building (María García and José Avilés). D. C. acknowledges the FEDER funds for Programa Operativo Fondo Social Europeo (FSE) de Andalucía 2014-2020 (DOC_01376) and Project (PP2F_L1_07).

References

Aguilera Martín, M., Sunyer Sunyer, M., Vaquer Llop, J.M., Gómez Sánchez, J., 2023. "*Vinum Mulsum*. La recuperación experimental del vino romano más exclusivo" in De luxuria propagata romana aetate. Roman luxury in its many forms. Arqueopress Publishing Ltd., Oxford, pp. 368–383.

Álvarez M, Moreno IM. Jos A, Cameán AM, González AG (2007b) Study of mineral profile of Montilla-Moriles "fino" wines using inductively coupled plasma atomic emission spectrometry methods. J Food Comp Anal 20:391-395.

Álvarez, M., Moreno, I.M., Jos, A., Cameán, A.M., González, A.G., 2007a. Differentation of two Andalusian DO "fino" wines according to their metal content from ICP-OES by using supervised pattern recognition methods. Microchem J 87, 72–76.

Álvarez, M., Moreno, I.M., Pichardo, S., Cameán, A.M., González, A.G., 2012. Mineral profile of "fino" wines using inductively coupled plasma optical emission spectrometry methods. Food Chem. 135, 309–313.

Arobba, D., Bulgarelli, F., Camin, F., Caramiello, R., Larchar, R., Martinelli, L., 2014. Palaeobotanical, chemical and physical investigation of the content of an ancient wine amphora from the northern Tyrrehenian sea in Italy. J Archaeol Sci 45, 226–233.

Artiushenko, O., Zaitsev, V., 2023. Competing ligand exchange-solid phase extraction method of polyphenols from wine. Microchem. J. 191, 108917.

Benítez, P., Castro, R., Garcia Barroso, C., 2003. Changes in the polyphenolic and volatile contents of "fino" sherry wine exposed to ultraviolet and visible raditation during storage. J. Agric. Food Chem. 51, 6482–6487.

Blanco-Zubiaguirre, L., Olivares, M., Casto, K., Carrero, J.A., García-Benito, C., García Serrano, J.A., Pérez-Pérez, J., Pérez-Arantegui, J., 2019. Wine markers in archeological potteries: Detection by GC-MS at ultratrace levels. Anal. Bioanal. Chem. 411, 6711–6722.

Briggs, L., Demesticha, S., Katzev, S., Swiny, H.W., Craig, O.E., Drieu, L., 2022. There's more to a vessel than meets the eye: Organic residue analysis of 'wine' containers from shipwrecks and settlements of ancient Cyprus (4th–1st century bce). Archaeometry 64, 779–797.

Brun, J.P., 2003. Le vin et l'huile dans la Méditerranée antique: viticulture, oléiculture et proceeds de fabrication. Errance, Paris.

Brun, J.P., 2004. Archéologie du vin et de ĺhuile: de la prehistoire a ́lepoque hellenestique. Errance, Paris.

Caballos, A. (2021): "La paulatina integración de *Carmo* en la Romanidad", in *Actas del II Congreso de Historia de Carmona, Carmona Romana,* A. *Caballos (ed.).* Universidad de Sevilla y Ayuntamiento de Carmona, pp. 3-17.

Columela, L.J.M., 1824. Los doce libre de agricultura. Álvarez de Sotomayor y Rubio (Translator), Madrid.

Cosano, D., Román, J.M., Lafont, F., Ruiz Arrebola, J.R., 2023a. Archaeometric Identification of a Perfume from Roman Times. Heritage 6, 4472–4491.

Cosano, D., Esqyivel, D., Román, J.M., Lafont, F., Ruiz Arrebola, J.R., 2023b. Spectroscopic identification of amber and fabric in a Roman burial (Carmona, Spain). Vib. Spectrosc. 127, 103557.

Fang, F., Li, J.M., Zhang, P., Tang, K., Wang, W., Pan, Q.H., Huang, W.D., 2008. Effects of grape variety, harvest date, fermentation vessel and wine ageing on flavonoid concentration in red wines. Food Res. Inter. 41, 53–60.

Flamini, R., Mattivi, F., De Rosso, M., Arapitsas, P., Bavaresco, L., 2013. Advanced knowledge of three important classes of grape phenolics: Anthocyanins, stilbenes and flavonols. Int. J. Mol. Sci. 14, 19651–19669.

Fujii, H., Mazzitelli, J.B., Adilbekov, D., Olmer, F., Mathe, C., Vieillescazes, C., 2019. FT-IR and GC–MS analyses of Dressel IA amphorae from the Grand Congloué 2 wreck. J. Archaeol. Sci. Reports 28, 102007.

Garnier, N., Richardin, P., Cheynier, V., Regert, M., 2003. Characterization of thermally assisted hydrolysis and methylation products of polyphenols from modern and archaeological vine derivatives using gas chromatography–mass spectrometry. Anal. Bioanal. Chem. 493, 137–157.

Garrido, J., Borges, F., 2013. Wine and grape polyphenols - A chemical perspective. Food Res. Inter. 54, 1844–1858.

Gleba, M, Román, JM. (2024) Textiles from a recently excavated early Roman tomb in Carmona (Sevilla, Spain), in S. Spandidaki, C. Margaritis, A. Iancu (eds), Purpureae Vestes VIII.

Guasch-Jané, M.R., Ibern-Gómez, M., Andrés-Lacueva, C., Jáuregui, O., Lamuela-Raventós, R.M., 2004. Liquid chromatography with mass spectrometry in tandem mode applied for the identification of wine markers in residues from ancient Egyptian vessels. Anal. Chem. 76, 1672–1677.

Guasch-Jané, M.R., Andrés-Lacueva, C., Jáuregui, O., Lamuela-Raventós, R.M., 2006. First evidence of white wine in ancient Egypt from Tutankhamuńs tomb. J. Archaeol. Sci. 33, 1075–1080.

Harutyunyan, M., Malfeito-Ferreira, M., 2022. Historical and heritage sustainability for the revival of ancient wine-making techniques and wine styles. Beverages 8, 10.

John, E., 2015. Guyton and Hall Textbook of Medical Physiology, 13th Ed.

Kment, P., Mihaljevic, M., Ettler, V., Sebek, O., Strnad, L., Rohlova, L., 2005. Differentiation of Czech wines using multielement composition - A comparison with vineyard soil. Food Chem. 91, 157–165.

Kunkee RE, Eschnauer HR (2003) Wine, 6th Ed. Ullmanńs Encyclopedia for Industrial Chemistry. Vol. 39, Wiley-VCH. Weinheim, Germany pp. 393-431.

Limón, M., Román, J.M., 2022. Dos inscripciones inéditas en urnas procedentes de Carmona (Sevilla). Epigraphica 84, 609–620.

López-Artíguez, M., Cameán, A.M., Repetto, M., 1996. Determination of nine elements in sherry wine by inductively coupled plasma-atomic emission spectrometry. J. AOAC Inter. 79, 1191–1197.

Paneque, P., Alvárez-Sotomayor, M.T., Gómez, I.A., 2009. Metal contents in "oloroso" sherry wines and their classification according to provenance. Food Chem 117, 302–305.

Pecci, A., Clarke, J., Thomas, M., Muslin, J., van der Graaff, I., Toniolo, L., Miriello, D., Crisci, G.M., Buonincontri, M., Di Pasquale, G., 2017. Use and reuse of amphorae. Wine residues in Dressel 2–4 amphorae from Oplontis Villa B (Torre Annunziata, Italy). J. Archaeol. Sci. Rep. 12, 515–521.

Petit-Domínguez, M.D., García-Jiménez, R., Rucandio, M.I., 2003. Chemical characterization of Iberian amphorae and tannin determination as indicative of amphora contents. Microchim. Acta 141, 63–68.

Plinio (2010) Historia Natural. Libros XII-XVI; Manzanero Cano F, García Arribas I, Arribas Hernáez ML, Moure Casas AM, Sancho Bermejo JL, Translators; Biblioteca Clásica Gredos. Madrid.

Pohl, P., 2007. What do metals tell us about wine? Trends Anal. Chem. 26, 941–949.

Robinson, J., 2006. The Oxford companion to wine, Third edition. Oxford University Press, Oxford.

Simonetti, G., Buiarelli, F., Bernardini, F., Riccardi, C., Pomata, D., 2022. Profile of free and conjugated quercetin content in different Italian wines. Food Chem. 382, 132377.

Tchernia, A., Brun, J.P., 1999. Le vin romain antique. Editions Glénat Livres, Grenoble, France.

Van Limbergen, P., Komar, P., 2024. Making wine in earthenware vessels: a comparative approach to Roman vinification. Antiquity 98, 1–17.

Vaquerizo Gil, D., 2023. Necropolis, rites and funerary world in Roman Hispania: Reflections, trends and proposals. Vinc. Hist. 12, 40–64.

Velo-Gala, I., García-Heras, M., Orfila, M., 2019. Roman windows glass in *Hispania Baetica*: Glass origin and manufacture study through electro microprobe analysis. J. Archaeol. Sci.: Rep. 24, 526–538.

Zanini, R., Moro, G., Orsega, E.F., Panighello, S., Selih, V.S., Jacinovic, R., van Elteren, J. T., Mandruzzato, L., Moretto, L.M., Traviglia, A., 2019. Insights into the secondary glass production in Roma Aquileia: A preliminary study. J. Archaeol. Sci. Reports 50, 104067.