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# POSSIBLE USES OF DEPAS AMPHIKYPELLON FROM KÜLLÜOBA IN WESTERN CENTRAL ANATOLIA THROUGH GC-MS ANALYSIS OF ORGANIC RESIDUES

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## ABSTRACT

The end of the Early Bronze Age, in other words the EBA III, is one of the significant turning points in the cultural history of the Anatolian Peninsula. Metal objects that indicate social class differences, advancements in architecture, and finds that point to connections with distant regions all demonstrate that a political structure began to emerge in Anatolia during this period. Parallel to these developments, especially with the increase in interregional relations, the use of new vessels indicates a novelty in eating and drinking habits. Tankards, bell-shaped cups and depas amphikypellon stand out within this group and are referred to as the new drinking vessels. In addition to these, it is note-worthy that beakers or goblets, and also container amphorae started to become widespread. All these types are considered to be associated with new drinking habits, and the new drink type is suggested to be wine. However, the suggestions proposed regarding the use of these vessels are not supported by any archaeometric studies until now. In this study, we share results from total lipid extraction analyses (TLE) performed by gas chromatography-mass spectrometry (GC-MS) on a group of depa vessels from the archaeological site of Küllüoba located in Eskişehir, Turkey, and re-evaluate at the possible purposes of these vessels. Biomolecules such as fatty acids, salicylic acids, organic acids, hydrocarbons, n-alkanes, and herbal steroids identified as a result of these analyzes provided the first direct evidence of the use of fermented products such as wine and herbal analgesics in Early Bronze Age Anatolia. Additionally, we present the first archaeological data of the period and the region regarding the use of salicylic acid.

**KEYWORDS:** Depa, organic residue analysis, analgesics, wine, Küllüoba, Bronze Age, feasting, salicylic acid

## 1. INTRODUCTION

A condensed review of organic residues analysis, the archaeological/chronological background of the Küllüoba settlement and the Depas Amphikypellon, as well as its use, typology and dispersal, is given first prior to the materials, methods and results.

### 1.1. Organic Residues

Nutrition and health quality are the basic building blocks in the struggle for survival of human beings at individual, biological and social levels. For this reason, the determination of these habits has a great importance in determining the cultural history of human beings. Ceramics made of clay, which are widely found in the archaeological record, can provide important data about the economies, cultures and traditions of the societies that lived in the past. With the appropriate analytical techniques, organic residues (ORs) that are absorbed by ceramics can be analysed and provide valuable information about daily life, trade and rituals of communities in the past (Blanco-Zubiaguirre *et al.*, 2019; Dunne *et al.*, 2019; Pecci *et al.*, 2018; Roffet-Salque *et al.*, 2017; Koh and Betancourt 2010; Chovanec 2022; Kruegerl *et al.* 2018; Mayyas 2018; Mayyas *et al.* 2013).

Visible ORs are encrusted deposits that adhere to the inner or outer surface of a ceramic sample. These remains can be defined by archaeologists as burn-soot residues accumulated by heating the ceramics over fire or as materials used for sealing and adhesive purposes (Roffet-Salque *et al.*, 2017). However, these visible residue types are not common in ceramic samples and the related remains tend to be contaminated or removed during the removal and cleaning of such artifacts. Since it is very difficult to clean soil from the visible ORs on the surface or to remove impurities that penetrated the sample during excavation, such residues are not preferred for analyses (Roffet-Salque *et al.*, 2017). Today, most analyses target absorbed ORs, which usually derive from original food products stored or processed in unglazed ceramics and represent an accumulation of a single use or multiple cooking events over the lifetime of the relevant artifact.

During processes, such as cooking, transfer and storage, the organic compounds interact with the ceramic structure and are absorbed in the pores on the walls of the ceramics. In particular, the inorganic porous matrix of unglazed ceramic vessels can provide a protective environment for the decomposition products and biomolecules of the absorbed organic compounds, and thus they can be preserved for thousands of years. The porous structure of unglazed ceramic vessels allows for deep penetration of the organic compounds, providing strong protection

against the microbial attacks and advanced degradation that may occur due to water seepage in the process in which they are buried (Dudd *et al.*, 1999; Heron *et al.*, 1991). Lipids appear as the largest group of ORs found in archaeological ceramics (Rosiak *et al.*, 2020). Edible oils in the lipid group and especially fatty acids, which are the major compounds of these oils, are considered the most important members of the ORs (Evershed, 2008; Gregg and Slater, 2010; Tite, 1999). It is also possible to determine characteristic compounds that can directly show the materials from which they originate and are defined as biomarkers, in ORs in unglazed ceramics. Succinic, tartaric, malic, and syringic acid as grape-derived biomarkers; fumaric and citric acid are small molecule minor acids that are accepted as biomarkers related to alcoholic fermentation (Barnard *et al.*, 2011; Garnier and Valamoti, 2016; Jerković *et al.*, 2011; Manzano *et al.*, 2016). On the other hand, tartaric (Inserra *et al.*, 2015; Pecci *et al.*, 2017; Zhang *et al.*, 2018) and syringic (Fujii *et al.*, 2019; Pecci *et al.*, 2017) acids detected in unglazed ceramic artifacts is accepted as the most important biomarker showing that these vessels were used to transport or store red wine. It should be considered that these mentioned bioactive compounds may also be present in the soil, and care should be taken when interpreting the results.

Different compounds for medical purposes rather than food were also stored or transported in archaeological ceramics. The most well-known of these compounds are terpenoid acid resins (Colombini *et al.*, 2005; Manzano *et al.*, 2016; Ribechini *et al.*, 2008) and this resin has been used not only as a waterproofing material or sealant, but also for purposes such as medicine, antiseptic or balm in rituals (Colombini *et al.*, 2005). It is known that salicylic acid has been used for a long time as an anti-inflammatory or pain reliever. It is mostly found in the leaves and bark of the willow, and poplar trees (Norn *et al.*, 2009). Salicylic acid has been known for its medicinal properties since ancient times (around 4000 years ago), notably early clay tablets from the Sumerians show that salicylate-rich willow leaves were prescribed for rheumatic diseases (Jack, 1997). Both the ancient Egyptians and Greeks described the use of willow leaves or myrtle for joint pain or inflammation more than 2,000 years ago as well as Hippocrates (460-377 BCE) suggested chewing on willow bark for fever and pain and using a tea brewed from willow bark for pain during childbirth (Goldberg, 2009).

Biomolecules found in ORs are usually found at trace levels and cannot be determined and detected by conventional analytical methods. Therefore, hyphenated chromatographic techniques with mass spectrometers stand out with their high sensitivity and molecular identification. Among them, especially

gas chromatography-mass spectrometry (GC-MS) has been successfully used in studies dealing with ORs from archaeological ceramic samples (Blanco-Zubiaguirre et al., 2019; Liritzis et al., 2020).

The present study describes the molecular composition of ORs from seven depas samples from the site of Küllüoba using GC-MS. Using this technique the first physical evidence of the use of salicylic acid in ancient times is presented in this study.

### 1.2. Archaeological-Chronological Background

The name early bronze age (EBA) for Western Anatolia is first used with the Troy excavations and this period dates to the beginning of the 3rd millennium BC, in parallel with Cilicia, Central and Eastern and southeastern Anatolia. With the publication of the results of the Tarsus-Gözlükule excavations, we use classical terminology to describe the EBA phases in the region, divided into three main phases: EBA I, EBA II and EBA III (Fidan et al., 2015). According to Efe, the transition period to the EBA coincides with the late Chalcolithic period according to the Mesopotamian chronology (Efe and Türkteki, 2011). The following EBA I covers a shorter period of time. EBA II is divided in two or three sub-phases, and EBA III is examined in two sub-phases as early and late. The EBA III coincides with the beginning of the Troy II settlement according to the western Anatolian chronology, and in this period, a significant change was experienced in both architecture and pottery traditions in the settlement. The emergence of these innovations for the first time in Tarsus has been decisive in terms of the chronological correlation of two distant regions. Research on the EBA in Western Anatolia has been going on for a long time. However, the EBA III, in which many innovations emerged, has not been adequately researched. In this context, Limantepe on the Aegean coast, apart from Tarsus and Troy, provided important data in terms of both the understanding of the architectural change and the finds showing the relations with western Anatolia in this period (Şahoğlu, 2008; Erkanal and Şahoğlu, 2016). The early stages of EBA III in Denizli-Beycesultan, which is an important settlement in terms of EBA, are not yet fully known (Lloyd and Mellaart, 1962). On the other hand, EBA III is not represented in Demircihüyük, which is the only settlement providing long-term information on the EBA chronology of the inland western Anatolia (Efe, 1988). In terms of understanding this period in Central Anatolia, Kültepe, with its long-term chronology, offers new information about the EBA III period, (Kulakoğlu, et al., 2020). Recently published C14 results in Kültepe not only contribute to a better dating of the period, but also provide new data in the context of the beginning of the international trade network, which is the main

element of the period, and western Anatolian relations.

Generally, the 3<sup>rd</sup> millennium BC is an important period of change for the Anatolian Peninsula. While small-scale settlements with simpler social hierarchy were seen at the beginning of the EB I in Troy I (Blegen et al., 1950:pl. 436), settlements of larger-scales with a more complex socio-economic system appeared with them in EB II such as in Troy II and Karataş-Semayük (Blegen et al., 1950, pl. 426; Mellink and Angel 1966:247, fig.2). Advancements in defense systems, constructions of monumental structures, and the separation of residential and public areas indicate a gradually increasing social complexity. For instance, during this period (IVC Phase), the monumental public structures at Küllüoba were separated from the other part of the settlement (Efe and Fidan 2008; fig.3). A similar situation is also observed in layers IIa-c of Troy (Blegen, Caskey, and Rawson 1951, pl.453-455). During the same period, influences from Western Anatolia began to be seen in Cilician region of Tarsus, as well. These influences, which manifest in the form of the megaron-shaped structures (Özyar, 2017), votive pits (Türkteki and Başkurt, 2016; Türkteki and Türkteki 2021) and vessel types such as the depas-tankard in Tarsus, point to the relations between Northwestern Anatolia and the Cilicia region.

During EBA III settlements, production of goods, social organization and hierarchical structure change as well as the intraregional and long-distance trade of raw metal materials increases (Massa and Palmisano, 2018). As a result, the political view of Anatolia also changes. The trade route that ran from the Cilicia region to Northwestern Anatolia, and from there to the islands of the east Aegean, Thrace; with depas presence being a ceramic style feature in Cycladic islands, Minoan centers and mainland Greece; all of which have played a significant role in the interregional relations (Efe, 2020, 2007; see also Şahoğlu, 2008; Broodbank, 2010).

These relations, which intensified especially at the end of the EBA III, manifest themselves in novelties such as the widespread use of tin in metallurgy, the production of metal vessels, the use of potter's wheel in pottery production techniques (Türkteki, 2013), and metrology (Rahmstorf, 2006). With the entry of foreign jewelry, weapons, objects made of semi-precious stones, and vessel types such as Syrian bottles and goblets to the region, it is possible to trace the signs of new social classes.

While new production-related classes, such as metallurgists and potters began to emerge, the elites or rulers who use prestige objects and control both the region and the trade also became more prominent. Amongst these prestige objects, a new group of drinking vessels appeared. Within the group, bell-shaped

cup, tankards, depa and goblets are among the noteworthy vessel types. Although these vessels were occasionally made of metal, ceramic imitations are primarily found in archaeological record (Türktekı, 2021).

### 1.3. *Depas Amphikypellon: Its Use, Typology, and Dispersal*

The emergence of depas, one of the most diagnostic pottery forms, is significant in determining the beginning of the EBA III, which is the last phase of the Early Bronze Age in Western and Central Anatolia. The appearance of this vessel, which was first discovered in phase IIc of Troy and numbered A45 (Blegen et al., 1950), is acknowledged as the beginning of the EBA III in many Western Anatolian settlements. Its examples were initially found during the first excavation season of Troy. Using Homer's *Iliad* as a basis, Henrich Schliemann believed that the depas vessels might be the two handled drinking cups mentioned in the book. Schliemann incorrectly named this vessel type "*Depas Amphikypellon*" However, though incorrect, it entered the archaeological literature under this name (Schliemann, 1874; Schliemann, 1881). In his published study, Spanos states that *depas* is a Greek word

without an etymology and *kypellon* could be translated as a drinking vessel.; Thus, according to him, *depas amphikypellon* roughly means a two handled drinking cup (Spanos, 1972).

Roughly cylindrical in shape, the depa (plural for depas, Betancourt 2007) have pointed or flat bottoms and these vessels cannot stay upright on a flat surface without support (Figure 1a-Type 1-4; Figure 1b). However, among the comparably smaller wheel-made examples of, there are those that have flat-bottoms and pedestals (Figure 1a-Type 5,7). In some of the examples, the body slightly narrows in its middle section. The two handles, placed symmetrically on opposite sides, extend from under the rim all the way to the bottom. The fact that the depa, which have very fragile handles in terms of static, were produced in bright burnished and mostly red and gray tones among the quality ware groups, shows that the ceramic samples of this vessel were produced to imitate metal samples, at least at the beginning. A discovered silver depas, which must be a piece of the Trojan treasure, can be regarded as evidence of this assessment (Sazcı, 2006). For that reason, this type of vessel is also evaluated to be a prestige object, and is proposed to belong to the elite (Şahoğlu, 2014).

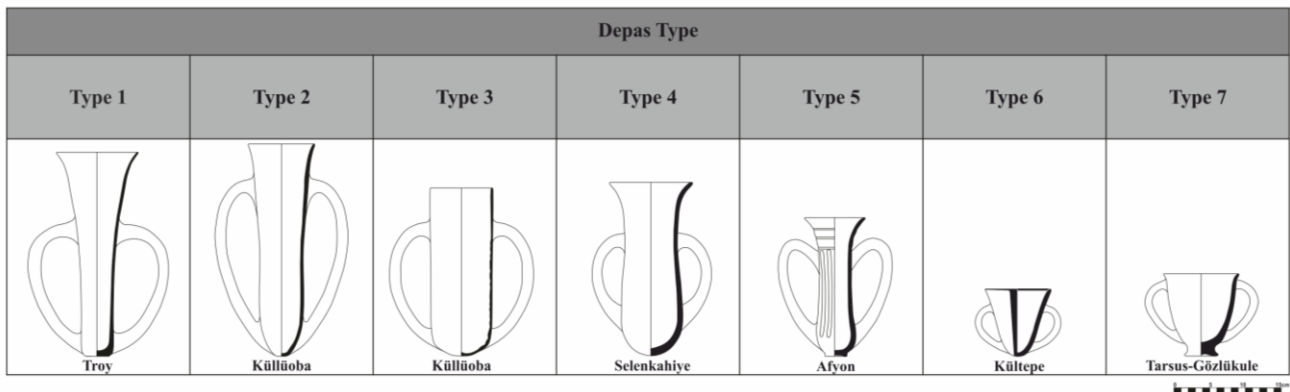


Figure 1a. Typology of depa (After Şahoğlu 2014)



Figure 1b. Whole depa samples from Külliöba



It is stated that examples of depa were recovered from both funerary and domestic settings (Şahoğlu, 2014), and there are various comments on its intended use. Parallel with the emergence of this type, the increasing use of tankards, goblets and amphorae (Türkteki, 2020) indicate that a new drinking culture has become widespread in this period. Thus, the depa are mostly associated with wine consumption. Based on the fact that it is two-handled, this type of vessel, which is thought to be used in ceremonies where especially the elite participated, is considered to be passed from hand to hand while the drink inside is consumed (Eslick, 2009). Also, because the depa usually have pointed bottoms and cannot possibly stay upright by themselves, the drink inside must be completely consumed by the end of the ceremony. The discovery of numerous examples, such as in the temple area of Kültepe, supports the theory that these ceremonies were associated with sacred areas (Şahoğlu, 2014).

Another explanation for the use and spread of depa is that these were private items, and that the elites who were personally involved in trade carried these items around with them. According to this hypothesis, the exaggerated handles must be used as the vessel's attachment points for the purpose of transportation (Şahoğlu, 2014). Therefore, we suggest that the metal examples primarily belonged to elites while other classes who tried to copy the elites might have used their imitations. On the other hand, the fact that the majority of the depa were found in votive pits

brings forward another possibility. This "pit ritual" involves the intentional burial of feasting remains, intact artefacts, and other items. In EB III, the pits seem to be connected with public spaces, as seen in Limantepe, Troy, and Kanlıgeçit. Perhaps this ritual was therefore somehow connected with elites and feasting. Feasting was connected with the consumption of food and alcoholic beverages by the elite or by the entire community (Kouka, 2011) and in EB III some new types of drinking vessels such as depa begin to be used. In order to be thrown into the pit after the rituals, the ceramic examples, which are less costly and particularly breakable, might have been preferred. The discovery of numerous depas examples at Troy, Külliüoba, and Kültepe (Ezer, 2014) shows that the depa were well-liked and accepted.

Named A45 in Troy, this type of vessel initially appears in phase IIc. The wheel-made examples, however, are first found among the gray-black coated wares in phase II d (Blegen et al., 1950). Other than these, red coated ware is the other ware group that wheel-made depa belong to. Found in quite large numbers, all examples of this group are mainly from phase II g. Unlike other types of depa (Blegen et al., 1950) one fluted example recovered from phase II g has a low pedestal. This type, which is mostly represented by red coated ware in Troy III, as well, is more elegant and expands relatively wider from the bottom to the rim during this phase when compared to the previous ones (Blegen et al., 1951).



Figure 2. Sites mentioned in text and distribution of depa drinking vessels.

The depa, which are discovered from Northern Syria to Mainland Greece and Bulgaria, play an important role interregional communication. Despite the fact that this vessel type is widely spread, it also displays regional variations (Figure 2).

As understood from previous evaluations (Aykurt & Kaya, 2005; Bittel, 1942; Blegen *et al.*, 1950; Çalış-Sazcı, 2007; Spanos, 1972; Spanos and Strommenger, 1993; Podzuweit, 1979; Huot, 1982; Mellink, 1989; Efe, 1988; Hüryılmaz, 1995; Schachner and Schachner 1995) of Anatolian depa on the basis of type and ware, it can be said that there are five distinct main groups in terms of geographical regions. These are represented mostly by red and a few gray/black coated (usually with rim slip below the mouths) examples in the Northwestern Anatolia region that have cylindrical bodies, flat or everted rims and egg-shaped bottoms; horizontally or vertically grooved examples (Figure 1-Type 5), also called the Southwestern Anatolian type (Oğuzhanoglu, 2019b), which are red coated and have grey paste; and in Central Anatolia, the examples with an S-profile that have rounded (Figure 1-Type 4) or flat bottoms; and the depa that have short bodies and paint decorations (Figure 1-Type 6) (Ezer, 2014).

According to new studies carried out in the northern sector of Kültepe, the depa first appears in the phase VIII in Kültepe, and this phase is dated to the middle of the 3rd millennium BC (Kulakoğlu *et al.*, 2020). Along with the characteristic types seen in Western Anatolia, the short form type unique to central anatolia (Figure 1-Type 6) and very well-known from Kültepe, is seen together. The emergence of the depa in Kültepe at a stage that can be considered quite early, with its distinctive form features, simultaneously with western Anatolia indicates that there may be different regional production centers in Anatolia (Şahoğlu, 2014).

Lastly, found in Southwestern Anatolia, Cilicia and Southeastern Anatolia, there are depa with pedestals (Figure 1-Type 7). These pedestaled examples, which were mostly discovered in the tombs of Gedikli, are considered to be an influence of the regional pottery on the depas form (Spanos, 1972).

#### 1.4. Küllioba Settlement and Depas Amphikypellon

The settlement of Küllioba is located at the western end of the Sakarya Basin, northeast of the Eskişehir Province-Seyitgazi District. The geographical area where the settlement was established has highly fertile agricultural soils throughout its history. This geographical area is also a natural transportation route that can provide passage between Central Anatolia and the Marmara region. Founded 930 m above sea level on a slight elevation on the northern side of

the Kireçkuyusu Creek, which is completely dried up today, the settlement has an area of 350 x 250 m and is 10 m above the plain level. The mound, inhabited uninterruptedly for 1450 years between 3300-1850 BCE, includes all phases of the Early Bronze Age (Figure 3). It is an important settlement that provides information on the city planning of the EBA II as well as the EBA chronology of Western Anatolia. (Efe and Türkteki, 2011) (Efe and Fidan, 2008) (Fidan *et al.*, 2015).

The architecture of the Early EBA III, in which depa first appeared, is known in Küllioba only from a few rows of stone walls that do not yield a complete plan, in grid-squares AD 18 - AD 19. In grid-square Z 19, levels of Late EBA II and EBA III are layered on top of one another. EBA III includes a Late EBA III phase (Transitional period) with five layers at the top, and below it an Early EBA III phase with at least three layers. The thickness of the cultural fill that contains both phases is almost 5 m (Türkteki, 2013).

KÜLLÜOBA			
Dates	Periods	Eastern Cone	Western Cone
13th-15th Cent. AD.	Islamic Burials	IA	
1 st. Cent. BC.	Late Hellenistic	IB	
-1 st. Cent. AD.	Early Roman		
1950 BC.		IIA	
		IIB	
	Late EB III	IIC	
	Übergangsperiod	IID	
2200 BC.		III	
		IIIA	
		IIIB	
2400 BC.	Early EB III	IIIC	
		IVA	
		IVB	
		IVC	
2800 BC.	EB II	IVD	
		IVE	
		IVF	
		IVG	1
3000 BC.	EB I	VA	
		VB	2
		VC	3
			4
3200 BC.	Transition to the EBA		5
3300 BC	Late Chalcolithic		6

Figure 3. Chronology of Küllioba.

Besides the stratification of the Early EBA III observed in grid square Z 19, other important data come from the votive pits (Türkteki-Başkurt 2017). Belonging to this period, approximately 200 pits were discovered in the mound. Of these, 80 were identified as votive pits. Special-purpose vessels, metal objects, bone idols, and grinding and crushing stones were found in these pits. Also, we determined that, in some pits, animals were sacrificed (Gündem 2020:84-85).

Regarding this period, it could be said that the pit tradition was also practiced here, like in all contemporary settlements (Oğuzhanoglu, 2019a). The majority of intact depa found in the settlement came from these pits.

The first depa at Küllioba are hand-made and found in phase IIIC, which is the earliest phase of the EBA III. Of the 9342 hand-made pottery sherds that have been examined from Küllioba, 130 pieces were determined with certainty to be from depas vessels. Along with Troy, this is the highest number of depas sherds found in Western Anatolia. Keeping that in mind, except for certain centers such as Troy,

Küllioba, and Kültepe in Western Anatolia, this type of vessel, which is not intended for daily use, is represented sometimes with only a single example or not encountered at all in other settlements. Thus, the high number of recovered depa indicates that the origin of this vessel type is in Western Anatolia.

The depa discovered in Küllioba are mainly produced within the red coated ware group. The application of a rim slip is highly characteristic particularly in the said ware group. In addition to red coated ware, it is also represented within gray, plain, and wash ware.

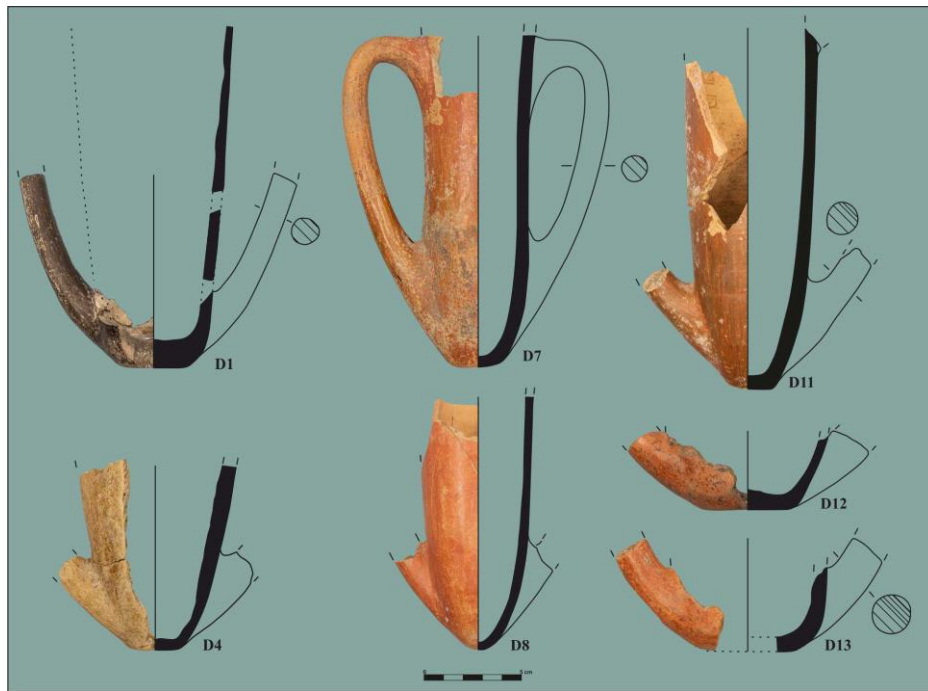


Figure 4. Photo and drawing of analysed depas samples

## 2. MATERIAL AND METHODS

### 2.1. Archaeological depa samples

Seven depas samples of the eleven available artifacts (Figure 4) were selected for OR analysis from Küllioba, Turkey, as part of an ongoing excavation by Bilecik Şeyh Edebali University. The information of the seven samples collected from the excavation

site are provided in Table 1. After removing c. 0.5 mm of each sample to avoid contamination from soil and fingerprints, about a 4 g sample was taken from the rim and body parts of the artifacts, which were more likely to contain ORs (Copley et al., 2003; Olsson and Isaksson, 2008; Charters et al., 1993). The samples were then ground using an agate mortar and prepared for the total lipid extraction (TLE).

Table 1. Catalogue of analysed depas samples

Sample ID	Trench	Unit No	Phase	Context	Base Diameter	Ware	Technique
D1	AD/AE 19	215	III A	Votive pit	3.6 cm	Gray	Wheelmade
D4	AC 26	91	III A	Votive pit	2.5 cm	Plain	Wheelmade
D7	AB 18	106	III A	Layer fill	2.4 cm	Red coated	Wheelmade
D8	AB 16	68	III A	Votive pit		Red coated	Handmade
D11	P 22	31	III A	Votive pit	2.5 cm	Wash ware (Brown)	Wheelmade
D12	AC 26	161	III A	Votive pit		Red slipped	Wheelmade
D13	AC 26	161	III A	Votive pit		Red slipped	Handmade

## 2.2. Extraction procedures

### 2.2.1. Acid-catalyzed extraction

Lipid residues have been extracted in many studies using chloroform/methanol or dichloromethane/methanol mixtures. However, these approaches are effective for the extraction of wax esters and triglycerides on various archaeological ceramics in different geographical and burial conditions, but they are ineffective for the fatty acids in the structure of very well absorbed ceramic pores (Stern *et al.*, 2000; Correa-Ascencio and Evershed, 2014; Hammann *et al.*, 2020), and only measurable amounts of fatty acids can be extracted from a very limited number of ceramics with these methods (Papakosta *et al.*, 2015b). One-step, acid-catalyzed, direct extraction-methylation method, relatively high efficiency for the extraction of low levels of lipid residues in unglazed ceramic artifacts, especially from Southeast Europe and the Middle East, including Turkey (ORs could be detected in approximately 30% of the total samples) appears as an alternative (Papakosta *et al.*, 2015b) and has recently been successfully applied by many researchers examining lipid residues in unglazed ceramics (Leclerc *et al.*, 2018; Mileto *et al.*, 2017; Oras *et al.*, 2017; Papakosta *et al.*, 2015b, 2019). This method, which consists of a one-step extraction for the lipid residues, was used for the ceramics in this study due to its advantages.

Lipid residues were extracted and methylated, following the protocol as described in (Papakosta *et al.*, 2019) with some modifications. According to the method, 2 g of cleaned and crushed sample containing 50  $\mu$ L of *n*-tetratriacontane (1000 mg/L) added as internal standard was heated with 6 mL of a mixture of MeOH and 98% H<sub>2</sub>SO<sub>4</sub> (5:1, v:v) at 70 °C for 4 h and then cooled, and lipids were extracted with *n*-hexane (3 x 2 mL) and separated off after centrifugation (2500 rpm, 3 x 5 min).

### 2.2.2. Base-catalyzed extraction

Since the method used for the extraction of lipid residues is not suitable for more polar molecules, such as wine biomarkers and minor compounds, the method proposed by Pecci *et al.*, (2013) was used with minor modifications. According to the method, 2 g of cleaned and crushed sample was extracted with 1 M KOH (2 x 4 mL) in water in a bath at 70 °C for 120 min. After cooling and centrifugation, the supernatant was acidified until the pH is about 2 with 1 mL of 37% HCl and then 4 mL of ethyl acetate was added to the supernatant, and vortexed for 2 min. The mixture was separated by centrifugation (2500 rpm, 2 x 5 min).

## 2.3. GC-MS Analysis

*n*-Hexane and ethyl acetate phases collected for residue analysis were dried down under a gentle N<sub>2</sub> flow

and further silylated with 100  $\mu$ L of bis(trimethylsilyl)trifluoroacetamide (BSTFA) with 1% trimethylchlorosilane (TMCS) at 70 °C for 1 h. With the silylation process, it was aimed to decrease the polarity of the analyte, increase its stability and improve the chromatographic separation (Moldoveanu and David, 2019). The extract was dissolved in 100  $\mu$ L of *n*-hexane, and filtered through a polytetrafluoroethylene (PTFE) filter and analysed in the GC-MS system. The Agilent (CA, USA) 7890N-5975C GC system was employed consisting of an MS detector and thermostated column oven. The operating conditions of the GC-MS system equipped with Agilent (CA, USA) HP-5MS capillary column (30m x 0.25mm x 0.25 $\mu$ m, (5%-phenyl)-methylpolysiloxane) were as follows: The temperature program of the column oven was held at 50°C for 1 min, then increased at 5 °C/min from 50 °C to 300 °C, and held at 300 °C for 9 min. The temperature of the injection block was 300 °C; the carrier gas was helium; the flow rate was 0.7 mL/min. The injection volume was 5.0  $\mu$ L on splitless mode; the electron ionization mode of MS was 70 eV. The temperatures of source and quadrupole in MS block were 230 °C and 150 °C, respectively. The MS was set to the scan mode between *m/z* 40 and 650, with a scan rate of 1.24 per second. Selected ion monitoring (SIM) mode of the MS was used only for salicylic acid compound, using the ratios of 45, 73, 74, 75, 91, 135, 267, and 268 *m/z* belonging to bis(trimethylsilyl) derivative of salicylic acid. The first 10 minutes each of analysis were defined as the solvent delay time. A blank was performed for each sample using the same protocols, taking great care in order to detect attempts for experimental errors and to avoid misinterpretations. Chromatographic peaks belonging to the sample and similarities to the blank were carefully eliminated. Analyses were performed in triplicates and results were given with standard deviations (SDs). The *m/z* ratios of the chromatographically separated compounds were automatically compared by the MS library data, and the relevant compounds were identified. Compounds identified with a 90% or greater match with the MS library were considered. While the compounds determined with 80-89% match were carefully evaluated, matches below 80% were not considered.

## 3. RESULTS

### 3.1. Acid-catalyzed extraction

Fatty acids are washed away from the ceramic pores to a negligible extent thanks to their hydrophobic structure when they come into contact with water during the burial or post-excavation processes, and these properties make the fatty acids very durable and resistant to environmental factors. In addition, very low levels of fatty acids can be successfully detected with the GC-MS technique. Compared with



other groups of ORs, especially proteins and carbohydrates, fatty acids stand out for their specific stability (Rosiak et al., 2020). It is known that long chain and unsaturated fatty acids are the most sensitive organic compounds to oxidation (Eerkens, 2005). In addition, the long chain fatty acids with more than 18 carbon numbers and short chain fatty acids with less than 14 oxidize faster than medium chain fatty acids (C14-C18) (Eerkens, 2007). As a result of many studies, palmitic and stearic fatty acids are the most frequently

detected fatty acid types in lipid residues in unglazed ceramics due to their stable structure. While the fatty acid and/or related lipid building block molecule ratios in archaeological ceramics obtained by GC-MS cannot provide precise information about the original fatty acid composition due to unpredictable degradation rates, they allow some general interpretations between different food origins due to the varying ratios of certain fatty acids. (Eerkens, 2007).

Table 2. The lipid concentration of seven depa obtained by GC-MS analysis

	D1	D4	D7	D8	D11	D12	D13
Lipid concentration, $\mu\text{g/g} \pm \text{SD}$ (lipid/ceramic powder)	2490.77 $\pm$ 56.53	398.69 $\pm$ 53.29	6286.90 $\pm$ 5.56	227.35 $\pm$ 11.74	213.52 $\pm$ 4.26	97.94 $\pm$ 7.12	441.87 $\pm$ 21.38

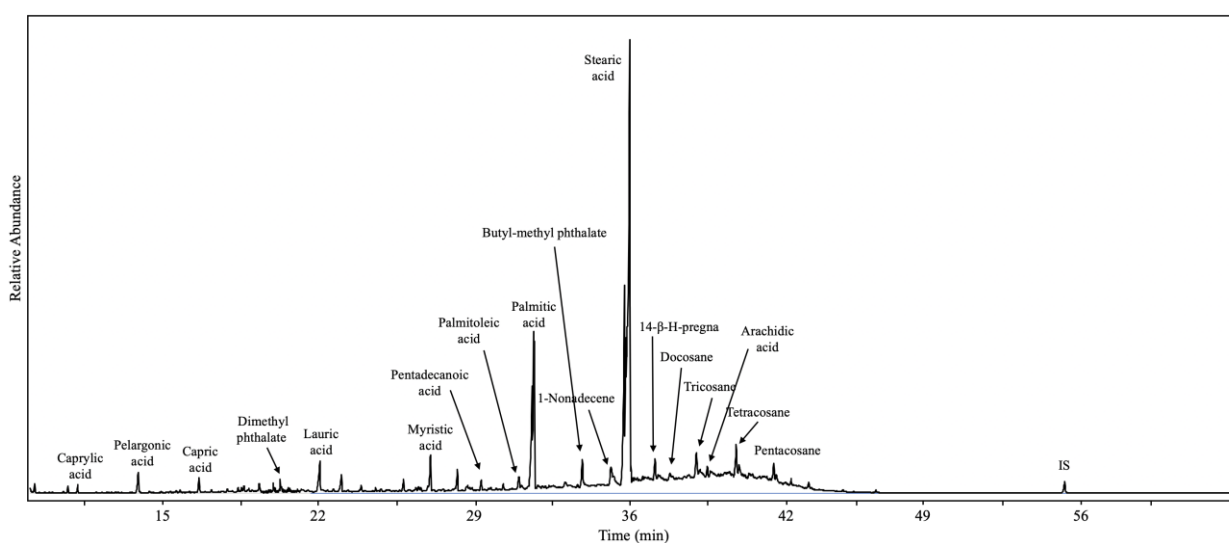


Figure 5. The chromatogram of the composition of D7 obtained as a result of acid-catalyzed extraction.

Table 2 presents the lipid concentration of the seven depa samples obtained by the GC-MS analysis. Lipid residues were detected in all ceramics and one-step, acid-catalyzed, direct extraction-methylation method utilized was found to be a useful approach for TLE of ORs from archaeological ceramics. The amount of the extracted lipid was calculated using the added internal standard. Lipid concentrations well above 5  $\mu\text{g/g}$  were obtained from all ceramics, sufficient to make reliable interpretations (Evershed et al., 2008; Reber et al., 2019). This result shows that the examined ceramics have been exposed to a relatively good preservation period in terms of ORs.

The fatty acid composition of the ceramics is given in Table 3. The results show that palmitic and stearic acids were predominant in the ceramics, with average percentages of 28.34% and 53.16%. The composition of palmitic and stearic fatty acids varied from varied from 7.99% (D8) to 54.63% (D13) and 37.21% (D13) to 80.12 (D11) for palmitic and stearic acids. The total ratio of saturated fatty acids (SFAs) and total ratio of

monounsaturated fatty acids (MUFAs) be highest in D12 (99.97%) and D7 (2.93%), respectively.

ORs in archaeological artifacts are exposed to severe decay conditions over long periods of time. As it is known, the degradation process of fatty acids first starts from double bonds because they have electron density. On the contrary, SFA, which do not contain double bonds, are more stable and most resistant to degradation. When the results in Table 3 are examined, it is seen that most of the fatty acids detected in this study are also SFA, with similar to archaeological ceramics. Only palmitoleic and oleic acids were identified as MUFA structures in some samples. On the other hand, 1-Nonadecene was observed as a monounsaturated hydrocarbon molecule. No fatty acids were detected for the total ratio of polyunsaturated fatty acids (PUFA) formation. The chromatogram of the fatty acid composition of D7 is given in Figure 5. The chromatograms of other ceramics are given in Supplementary Material (at the end).

**Table 3. The organic residue compositions of ceramics obtained as a result of acid-catalyzed extraction**

Compound (RT min)	CX	D1		D4		D7		D8		D11		D12		D13	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
2-ethylhexanoic acid (10)	C8:0											1.97	0.18		
Caprylic (octanoic) acid (13)	C8:0			0.87	0.13	0.35	0.02					2.15	0.17		
Pelargonic (nonanoic) acid (14)	C9:0			4.49	0.68	1.31	0.04	0.93	0.04			2.64	0.06	0.45	0.03
Capric (decanoic) acid (17)	C10:0	0.19	0.01	3.59	0.42	0.80	0.03	0.73	0.05			1.15	0.03	0.70	0.02
Tetradecane (19)	C14													0.41	0.01
Undecanoic acid (20)	C11:0			0.83	0.00										
Dimethyl phthalate (21)	C10	0.18	0.01	0.90	0.01	0.45	0.26								
Lauric (dodecanoic) acid (22)	C12:0	0.34	0.02	3.06	0.50	1.90	0.11							0.75	0.01
Azelaic (nonanedioic) acid (23)	C9:0	0.33	0.02												
Hexadecane (24)	C16							0.50	0.01					0.80	0.03
Heptadecane (26)	C17							0.80	0.28					0.93	0.02
Myristic (tetradecanoic) acid (27)	C14:0	2.64	0.18	6.69	0.40	2.04	0.19	1.56	0.21			2.51	0.38	2.01	0.01
Octadecane (28)	C18							0.29	0.06						
Pentadecanoic acid (29)	C15:0	1.03	0.02	1.53	0.06	0.48	0.02								
Diisobutyl phthalate (30)	C16	0.79	0.02	1.28	0.04										
Palmitoleic (9-hexadecenoic) acid (30)	C16:1	0.77	0.02			1.17	0.16								
Palmitic (hexadecanoic) acid (31)	C16:0	51.51	0.32	30.34	2.10	17.16	0.27	7.99	0.13	12.14	0.40	11.81	0.52	54.63	0.30
Methyl 14-methylhexadecanoate (32)	C18:0														
Eicosane (32)	C20	0.34	0.02												
Margaric (heptadecanoic) acid (33)	C17:0	0.38	0.08												
Butyl methyl phthalate (33)	C13	0.14	0.01			1.89	0.26	1.16	0.37					1.00	0.04
Oleic (9-octadecenoic) acid (34)	C18:1	0.95	0.06	0.79	0.04										
10-octadecenoic acid (34)	C18:1														
12-octadecenoic acid (34)	C18:1														
1-Nonadecene (34)	C19:1					1.18	0.10								
Stearic (octadecanoic) acid (35)	C18:0	38.06	0.32	41.03	1.70	43.36	1.82	79.89	1.07	80.12	1.13	77.74	2.33	37.21	0.32
14-β-H-pregna (36)						1.76	0.10								
Docosane (36)	C22	0.21	0.01			3.70	0.98								
Tricosane (38)	C23	0.16	0.01			4.43	0.75			2.78	1.02				
Arachidic (eicosanoic) acid (38)	C20:0					0.85	0.28	0.75	0.02						
Tetracosane (39)	C24					2.58	0.33			2.34	1.46				
Pentacosane (41)	C25					0.90	0.10			1.82	0.94				
Mono(2-ethylhexyl) phthalate (41)	C16							1.73	0.04						
Other		1.98		4.60		13.69		3.67		0.80		0.03		1.11	
Hydrocarbon saturated		0.70		0.00		11.61		1.59		6.94		0.00		2.14	
Hydrocarbon unsaturated		0.00		0.00		1.18		0.00		0.00		0.00		0.00	
SFA		94.48		92.43		68.25		91.85		92.26		99.97		95.75	
MUFA		1.72		0.00		2.93		0.00		0.00		0.00		0.00	
PUFA		0.00		0.00		0.00		0.00		0.00		0.00		0.00	

Using the fatty acid ratios, very useful interpretations can be made about the lipid residue examined. However, it is known that fatty acids can be formed due to bacteriological deterioration under anaerobic conditions. For this reason, it is necessary to pay attention to other fatty acids and minor compounds, which can impact the interpretations. Ruminant fats can also be distinguished from non-ruminant fats based on the proportions of certain fatty acids. If the stearic acid of the sample is lower than the palmitic acid and especially fatty acids such as pentadecanoic acid, and margaric acid and also oleic acid isomers are present, it has been proposed that the investigated residue may be a dairy product derived from a ruminant animal (Regert, 2011). When Table 3 is examined, it is seen that D1 contains that fatty acid distribution. The fatty acids with odd carbon number are determined in all ceramics, excluding D11 in Table 3. The presence of these fatty acids may also indicate a decomposition for ruminant fat (Evershed et al., 1997).

It is known that azelaic and palmitic acid can be formed from oleic acid due to bacteriological deterioration under anaerobic conditions (Kimpfe et al., 2002). Especially azelaic acid probably indicates the former presence of unsaturated fatty acids with a double bond at the ninth carbon position in the original fatty acids (Hudlicky, 1990), such as oleic acid. Table 3 shows that analysed samples comply with this interpretation and most of them contain azelaic and oleic acid together.

On the other hand, the identification of natural waxes (n-alkanes) provided key indications for the presence of leafy vegetables (Eerkens, 2005) (Kimpfe et al., 2002). Tetradecane, hexadecane, heptadecane, octadecane, eicosane, docosane, tricosane, tetracosane, and pentacosane were the waxes detected in D1, D7,

D8, D11, and D13 (Table 3). These saturated hydrocarbons concentrated in D7 and D11 at 11.61% and 6.94%, respectively. This result suggests that leafy vegetables or food products containing high levels of wax may have been processed in these examined ceramics. Another organic structure, the compound 14- $\beta$ -H-pregna, was detected in D7. This biomolecule is an herbal steroid found in the essential oils and different parts of some plants, including *Urginea indica* Kunth, *Allium rotundum*, *Gundelia tournefortii* L, *Citrus limon*, and *Cenchrus biflorus* and displays antibacterial activity (Zahra et al., 2019).

When a general evaluation is made, it can be said that the lipid residues in the ceramics analysed according to the fatty acid composition results obtained through GC-MS were of both animal and plant origin.

### 3.2. Base-catalyzed extraction

The organic acids residue composition of the seven depa samples obtained by GC-MS analysis is given in Table 4. Benzoic, caprylic, capric, and phthalic acids were found as organic acids present in all samples, varied from 0.59% (D1) to 3.98% (D8), 1.00% (D1) to 4.84% (D8), 0.37 (D13) to 3.08% (D4), and 14.71 (D1) to 3.16 (D12) respectively. Caproic, acetic, oxalic, succinic, pelargonic, glutaric, lauric, and cis-aconitic acids were residues seen in at least 50% of all samples in Table 4. The chromatogram of the composition of D4 is given in Figure 6. No labels were added to the peaks of compounds with an MS match of less than 80%. Peaks that were found to be impurities by blank tests were labeled. The chromatograms of other samples are given in Supplementary Material. Benzoic acid is one of the grape-related or alcoholic fermentation-related product markers detected in most archaeological ceramics (Fujii et al., 2019).

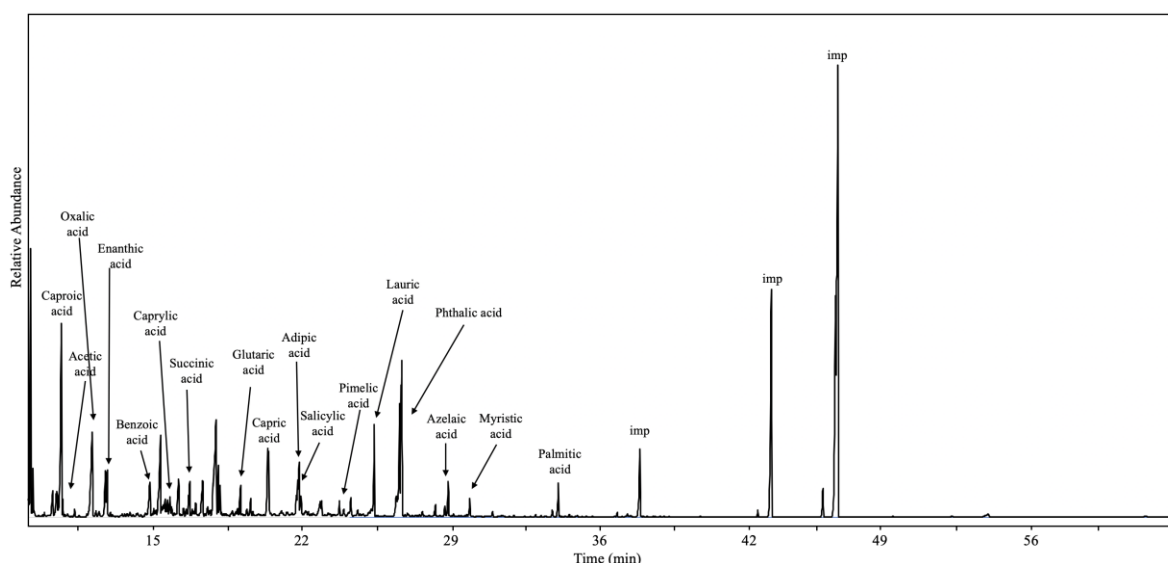


Figure 6. The chromatogram of the composition of D4 obtained as a result of base-catalyzed extraction.

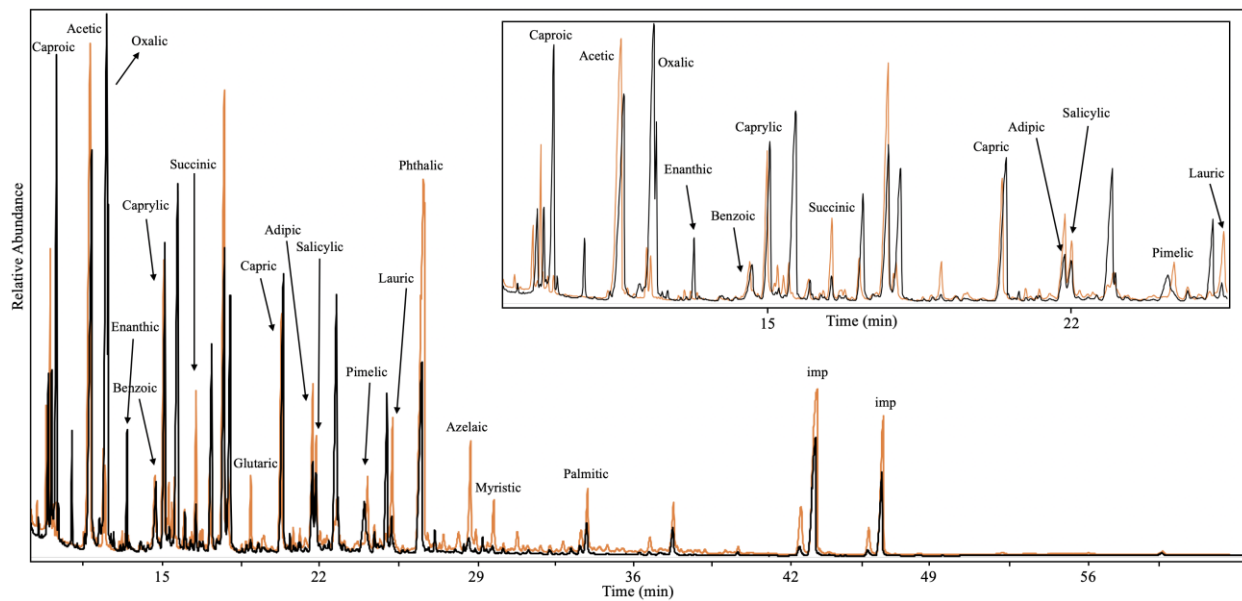


Figure 7. The chromatograms of the organic acids obtained as a result of base-catalyzed extraction by SIM analysis

Other markers from these detected in the analysed ceramics were pyruvic (Garnier and Valamoti, 2016), acetic (Zhang *et al.*, 2018), succinic (Blanco-Zubiaguirre *et al.*, 2019; Fujii *et al.*, 2019; Garnier and Valamoti, 2016; Pecci *et al.*, 2017; Zhang *et al.*, 2018), glutaric (Fujii *et al.*, 2019), malonic (Pecci *et al.*, 2017), and citric acids (Blanco-Zubiaguirre *et al.*, 2019; Manzano *et al.*, 2016; Zhang *et al.*, 2018). When the results in Table 4 are examined, it is seen that especially D1, D4, D7, and D11 contain most of the detected fermentation-related markers. Tartaric (T. Zhang *et al.*, 2018) and syringic acids (Fujii *et al.*, 2019; Pecci *et al.*, 2017), which are accepted as biomarkers of red wine, were not detected. Also, we detected 2-Ethylhexanol, which is found naturally in alcoholic beverages in D1. Therefore, these results indicate that most of these ceramics probably contained wine or different fermented products. Another alcoholic fermentation-related product in ceramics is oxalic acid, possibly derived from calcium oxalate, a byproduct of malt wine, and this was also direct evidence of beer's earliest origins (Zhang *et al.*, 2018). For this reason, especially D1, D4, D8, D11, D12, and D13 may also have been used for this purpose.

In addition, a variety of low-molecular weight aliphatic carboxylic acids such as, butyric, valeric, caproic, adipic, enanthic, pimelic, valproic, pelargonic, azelaic, and lauric and medium-molecular weight aliphatic carboxylic acids such as myristic, palmitic, and stearic acids are also shown in Table 4. Glycerol was also detected as triglyceride degradation products in D1. The samples D7 and D8 contain verbenone, which is naturally found in lavender (Özel, 2019) and pine resin (Jerković *et al.*, 2011) in Table 4, which may be a potential biomarker for consumption of these plants.

As a first time for the analysis of ORs in archaeological ceramics, 0.79% and 0.56% free salicylic acids were detected in D4 and D12, respectively. As mentioned in the introduction, written archaeological sources indicate that salicylic acid has been used since ancient times. However, a study containing physical data on the use of salicylic acid in the past has not been presented to date. For this reason, the base-catalyzed extraction was performed for D4, and D12 and the obtained extracts were analysed by GC-MS device in SIM mode with  $m/z$  ratios selected for free salicylic acid. The chromatograms obtained as a result of SIM analysis are given in Figure 7.

As seen in Figure 5, free salicylic acid peak detected in D4 and D12 intersects. The salicylic acid peak of both samples was 100% confirmed by MS library in the SIM mode. This result shows that the salicylic acid detected in D4 and D12 is in free form. It is important to determine whether the detected salicylic acid is a product that is absorbed during use of the relevant ceramic or is a product formed during the burial process. It is known that eukaryotic and prokaryotic cells including plants can produce salicylic acid as a secondary metabolite (Lefever *et al.*, 2020). In addition to being synthesized from Phe via cinnamic acid by plants (Dempsey *et al.*, 2011), salicylic acid can also be synthesized by some bacteria by isochorismate pathway (Wildermuth *et al.*, 2001). Although salicylic acid can be synthesized in different species, it is known that most of the active salicylic acid molecule in plants is free salicylic acid. (Zhang and Li, 2019). Since the salicylic acid compounds obtained as a result of the GC-MS analyzes are also in free form, the salicylic acid detected in D4 and D12 is thought to be of plant origin rather than bacteria. Since salicylic acid has the

ability to dissolve in lipids, in contrast with the  $\alpha$ -hydroxy acids, it may have been used or stored by mixing with different fats and oils in these analysed ceramics. In the relevant region, the extracts of many plants, which are believed to have many medicinal properties, are mixed with lipid products such as milk and are still consumed traditionally. However, it

should be noted that in order to express an assertive ingredient such as salicylic acid, it is also necessary to simulate the archaeological situation and to examine whether it is likely to survive through burial/leaching experiments. For this purpose, a new project will be started by our research group.

**Table 4. The organic residue compositions of ceramics obtained as a result of base-catalyzed extraction (Results represent peak area percentages)**

Compound (RT min)	Carbon numbers for fatty acids	D1		D4		D7		D8		D11		D12		D13	
		Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD	Mean	SD
Butyric (butanoic) acid (11)	C4:0											1.74	0.11	0.52	0.11
Caproic (hexanoic) acid (11)	C6:0	4.86	1.55	6.45	0.21	8.08	2.12			3.05	1.11	8.22	0.63	1.19	0.02
Acetic acid (12)		0.88	0.20	0.30	0.01	1.02	0.29			0.59	0.10				
Pyruvic (2-propenoic) acid (12)		0.43	0.22												
2-Ethylhexanol (12)		0.30	0.04												
Oxalic (ethanedioic) acid (13)		2.38	0.53	4.44	0.28			5.72	2.68	7.67	0.68	4.78	0.11	1.09	0.06
Acetic acid-2-ethylhexyl ester (13)		0.30	0.01												
Valproic (dipropylacetic) acid (13)								21.94	13.91						
Enanthic (heptanoic) acid (14)	C7:0	0.57	0.13	1.23	0.06										
Valeric (Pentanoic) acid (14)	C5:0											0.61	0.04		
Verbenone (Bicyclo[3.1.1]hept-3-en-2-one) (14)						0.43	0.18	2.55	1.12						
Benzoic acid (15)		0.59	0.02	1.56	0.08	2.11	0.61	3.98	1.67	2.46	0.27	1.60	0.02	1.04	0.06
Caprylic (octanoic) acid (16)	C8:0	1.00	0.05	3.20	0.07	2.62	0.76	4.84	2.08	1.49	0.24	2.40	0.05	0.96	0.08
Glycerol (16)		1.01	0.08												
Succinic (butanedioic) acid (17)		0.90	0.02	1.26	0.06	0.49	0.17	0.50	0.32	1.02	0.08	0.33	0.03		
Pelargonic (nonanoic) acid (18)	C9:0	1.69	0.13			3.79	0.32	6.02	2.77	3.05	0.49	2.68	0.11	1.48	0.09
Malonic (propanedioic) acid (19)						1.39	0.07								
Glutaric (pentanedioic) acid (19)				0.92	0.05	0.16	0.00			0.46	0.08				
Capric (decanoic) acid (21)	C10:0	0.78	0.04	3.08	0.11	0.84	0.06	0.71	0.48	1.07	0.18	0.72	0.03	0.37	0.03
Adipic (hexanedioic) acid (22)				1.81	1.08										
Salicylic acid acid (22)				0.79	0.04							0.56	0.14		
Pimelic (Heptanedioic) acid (24)				0.52	0.00										
Lauric (dodecanoic) acid (25)	C12:0	0.23	0.00	1.86	0.04	0.47	0.18								
Phthalic acid (26)		14.71	1.87	10.91	0.74	5.34	1.32	3.65	1.70	5.08	0.26	3.16	0.19	5.46	0.18
Cis-aconitic (1-Propene-1.2.3-tricarboxylic) acid (28)		1.71	0.06			13.23	2.38			2.11	0.30				
Azelaic acid (28)	C9:0			0.88	0.03										
Myristic (tetradecanoic) acid (29)	C14:0			0.41	0.06										
Citric acid (29)						0.46	0.12								
Palmitic (hexadecanoic) acid (33)	C16:0	3.36	0.33	0.84	0.03	2.56	0.53								
Stearic (octadecanoic) acid (37)	C18:0					4.58	1.45								



#### 4. DISCUSSION

In examples D1, D2, D3 D4, D7, and D11, biomarkers for fermented products were detected. It is possible for alcohol to be considered as a sort of medicine (McGovern 2019). The earliest example in Southwestern Asia that presents the chemical and archaeobotanical evidence of wine consumption is known from the Southern Caucasus (McGovern *et al.*, 2017). Apart from these from the Early Neolithic samples, other examples obtained from studies regarding tartaric acid analyzes are from Hajji Firuz Tepe in Iran (McGovern, 2003), and later from level V of Godin Tepe, dated to 3500-3100 BCE (McGovern, 2009). The earliest data on wine in Anatolia are in written sources recovered from the Assyrian Trade Colonies period at Kültepe (Kanesh) (Barjamovic, 2011). According to the written sources, wine was imported here from Mamma, Tegarama, Urshu and Unibgum, which are known for their sweet wines (Barjamovic, 2011). They also state that there was wine production in the region (Corti, 2017). There are also written documents on wine-making and its consumption from the Old Hittite period (Corti, 2017). During the Hittite period, beer consumption had an important place in ceremonies and religious rites (Brown, 2018). Important analysed samples about ceremonies with wine, beer and mead comes from the Phrygian capital of Gordion (McGovern 1999). Most of the other data on wine consumption is based on archaeobotanical remains (Zettler, R.F; Miller, 1996). The Küllüoba depa are the first to be chemically analysed in Anatolia and the yielded data partially supports previously proposed uses for this vessel type.

Salicylic acid detected in examples D4 and D12 is the first ingredient to be discovered in such vessels until today. It is considered that herbal medicines, analgesics, narcotic substances, and various drinks may be used in connection with ceremonies and rituals since prehistoric times (Samorini, 2019). Although there is very little data available from Southwestern Asia, one of the most significant information on plants that can be used for herbal medicine or analgesic comes from the kitchen area of the Ebla G Palace in Northern Syria, which is contemporary with the period in which the depa were in use. (Peyronel *et al.*, 2014). Wheat, barley, legumes, olives, grapes, and some wild plant residues were found in this area, particularly in the "kitchen room" no L.2890. *Euphorbia* discovered among the wild plant residues and its areas of use point to the possibility that the drink detected inside the Küllüoba depa that contained salicylic acid, in other words willow bark, could have been used similarly in ceremonies and rituals. No written records were found regarding the use of these plants (Peyronel *et al.*, 2014), but there are texts on the

preparation of medicine (Fronzaroli, 1998). There is no example found in Anatolia that can be compared with this example yet. However, it is known that also afterwards, in the Hittite period, willow bark was used in medicinal preparations (Şahinbaş, 1995).

#### 5. CONCLUSIONS

The new vessel forms that emerged during the EBA III period were previously associated with a new drinking culture and feasts. We argue that depas were used in ceremonies involving wine consumption conducted by the elite (Çalış-Sazcı, 2002). This study reveals the first archaeological evidence for the actual use of the depa form beyond theory. Results clearly reveal that there are fermented products in the depa. The results of the study, together with both the context and other related data, explain the role of these vessels in the pit ritual (Warner, 1994; Kouka, 2011; Blegen *et al.*, 1950; Mellink, 1989; Özdoğan and Parzinger, 2012; Oğuzhanoglu, 2019a; Türkteki and Başkurt, 2016). Both at Küllüoba and in settlements where depa have been discovered, the majority of the examples came from these pits, which also contained other special finds. We assert with certainty that alcoholic beverages were consumed in these ceremonies, as was thought before. This substance can be used mixed with alcoholic products, or it can be consumed by mixing with other herbs. In some depa samples, herbal steroids with antibacterial properties were also found, indicating that leafy plants were consumed. In addition, pythalic acid residues that are likely to belong to medicinal aromatic plants can also be evaluated in the same context. Samples of *Erysimum crasipes* preserved in a jar were unearthed in Küllüoba excavations (Çizer, 2015). This data shows that different groups of *Erysimum* species have been used in the field of medicine and that such plants are especially kept in the settlements.

In the context of all these evaluations, the question to be asked here is whether the use of these substances was part of a ceremony for the treatment of a disease or whether the ritual performed was a celebration. On the other hand, the soothing effect of wine or other alcoholic beverages should not be ignored. In this respect, it should be considered that the purpose of the ceremony, which ends with the pit ritual, may not always be a celebration. In this context, considering that some of the pits unearthed in Laodicea-Kandilkırı and Karataş-Semayük may be related to the cemetery (Oğuzhanoglu, 2019a) (Warner, 1994), it is also possible that these substances were used in a post-mortem ceremony to ease the sadness and pain. Although different data are needed to answer this question, it would be appropriate to consider future studies from this perspective.

## AUTHOR CONTRIBUTIONS

*Conceptualization and methodology:* Murat Türkteki and İsmail Tarhan, *formal analysis:* İsmail Tarhan and Hüseyin Kara, *software:* İsmail Tarhan and Hüseyin Kara, *writing -original draft preparation:* Murat Türkteki and İsmail Tarhan, *resources:* Hüseyin Kara, *data curation:* İsmail Tarhan, *writing-review and editing:* Murat Türkteki, *visualization:* Yusuf Tuna, *project administration:* Murat Türkteki, *funding acquisition:* Murat Türkteki and İsmail Tarhan.

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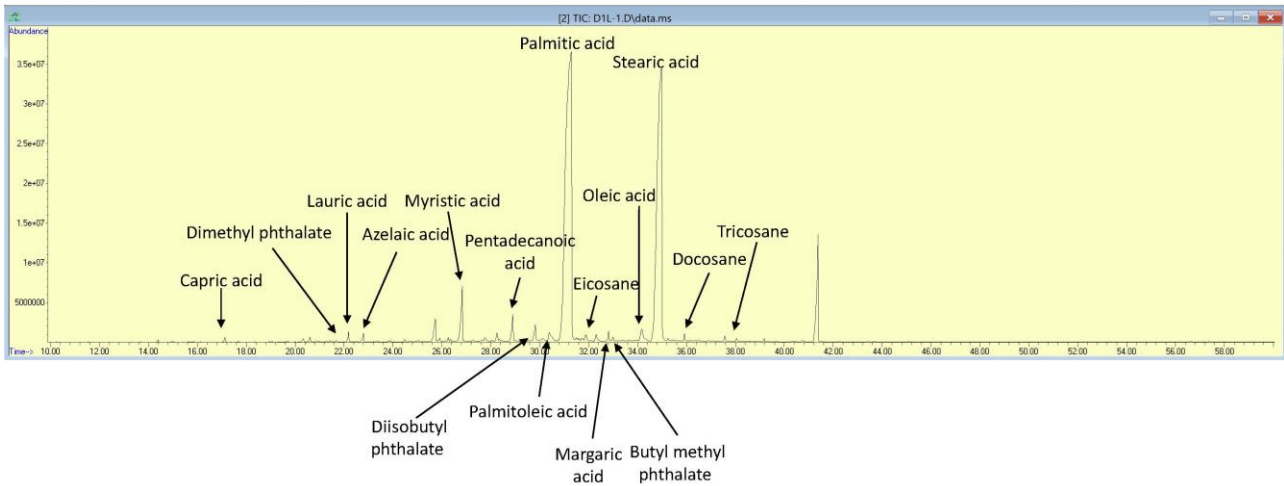


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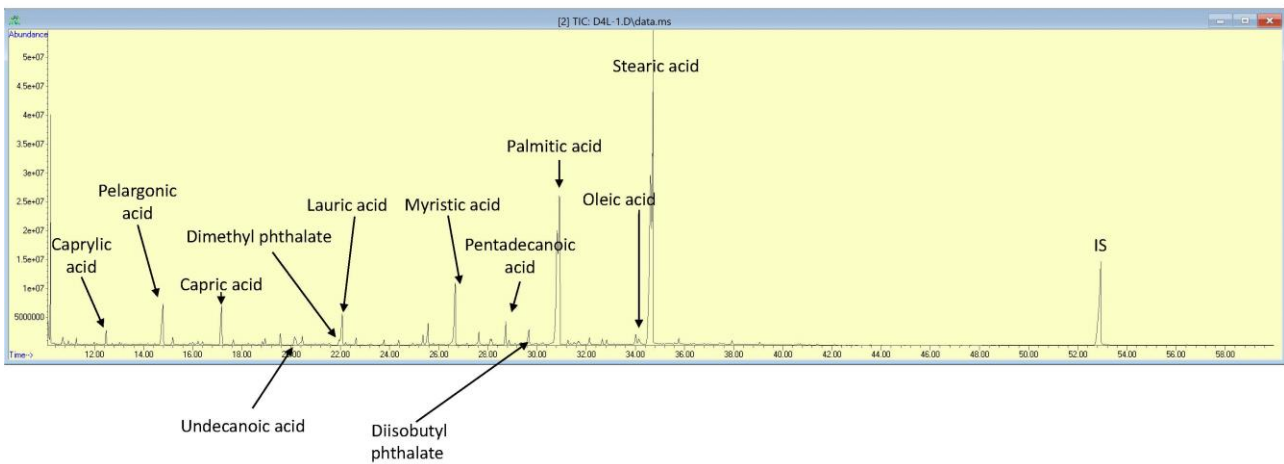
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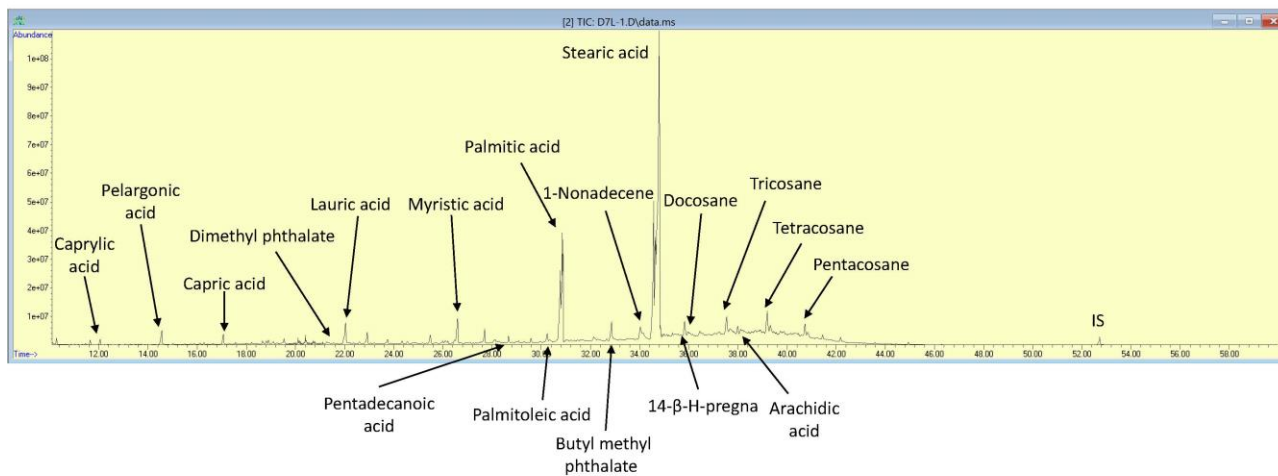
## SUPPLEMENTARY MATERIAL



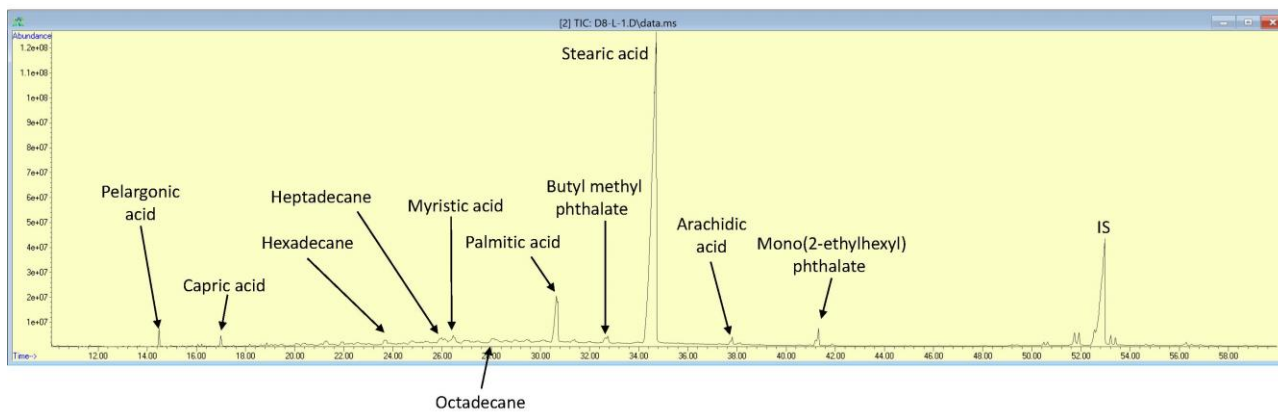
The chromatogram of D1 by acid-catalyzed extraction



The chromatogram of D4 by acid-catalyzed extraction

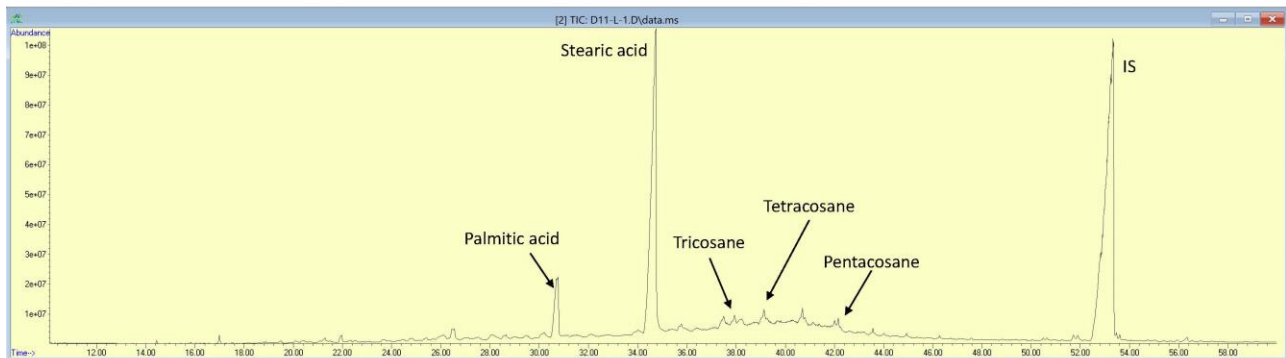


The chromatogram of D7 by acid-catalyzed extraction

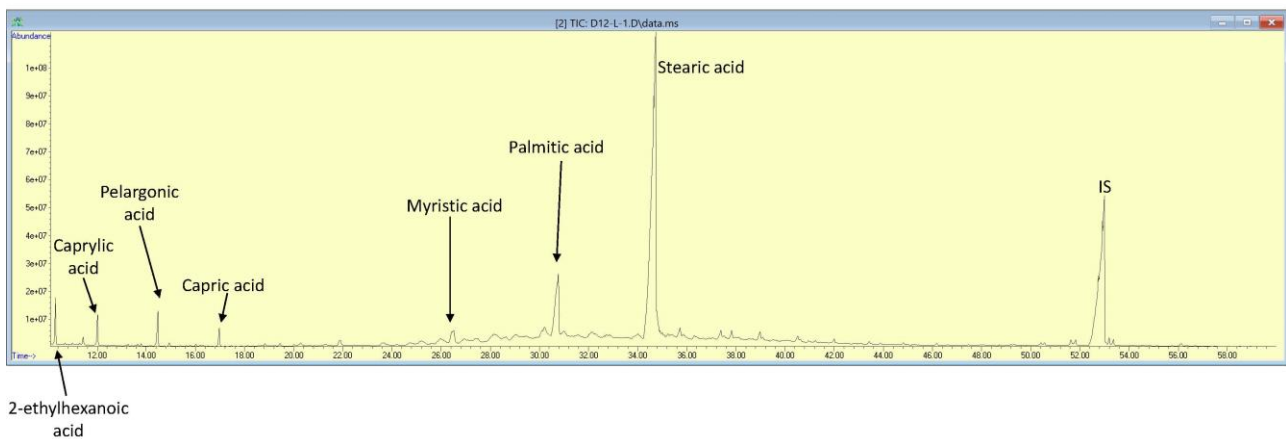


The chromatogram of D8 by acid-catalyzed extraction

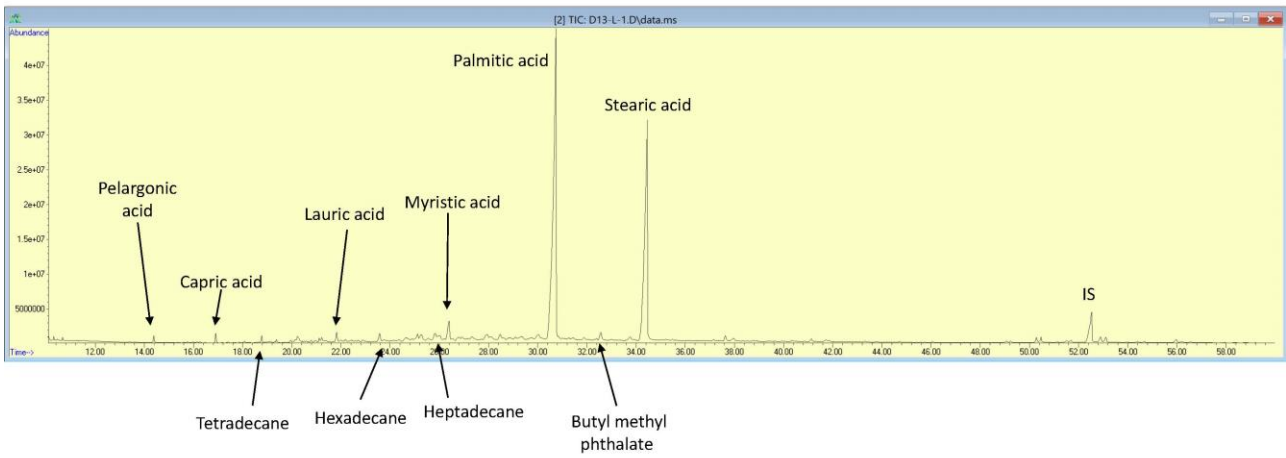




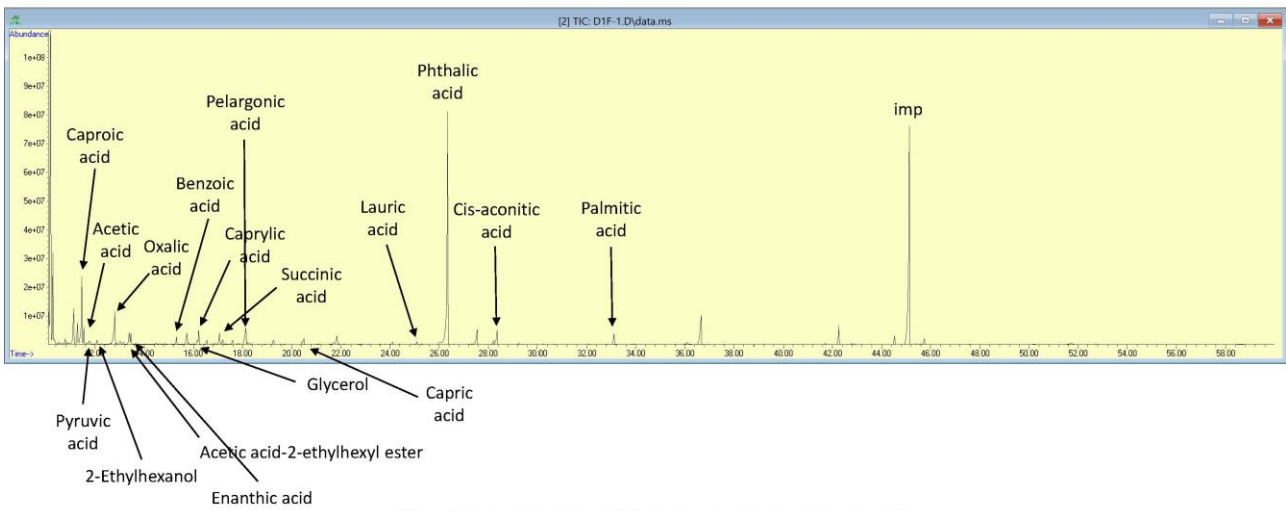
The chromatogram of D11 by acid-catalyzed extraction



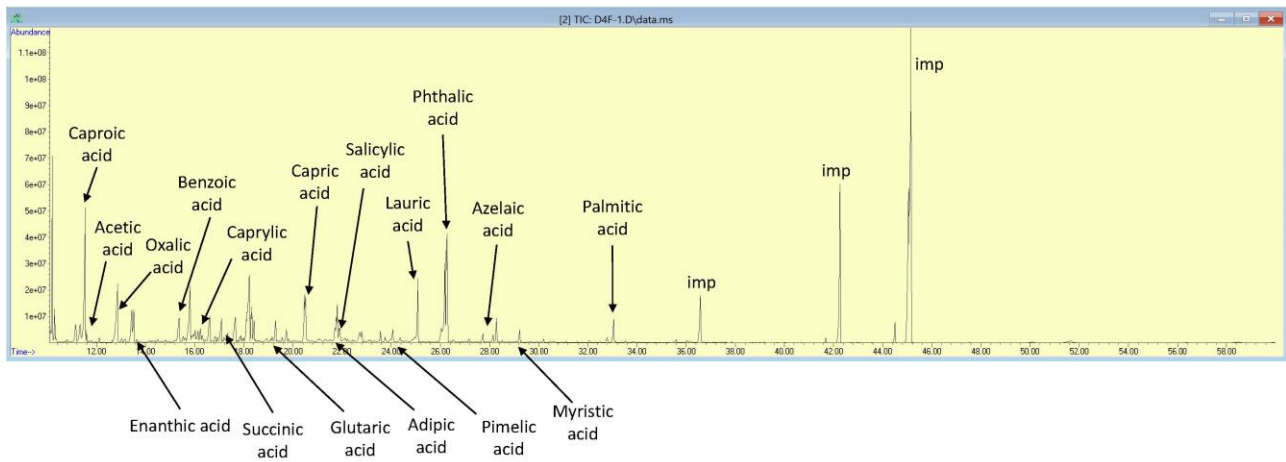
The chromatogram of D12 by acid-catalyzed extraction



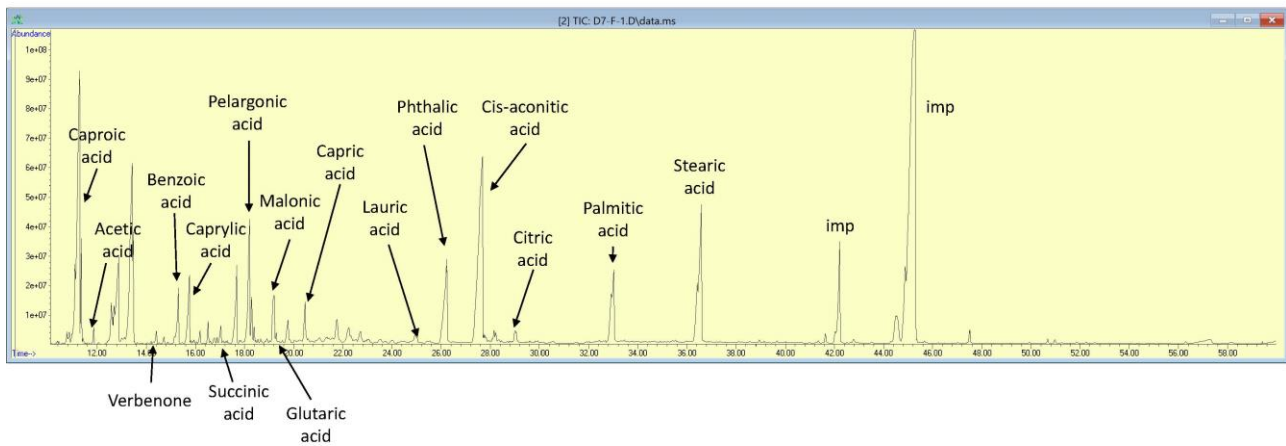
The chromatogram of D13 by acid-catalyzed extraction



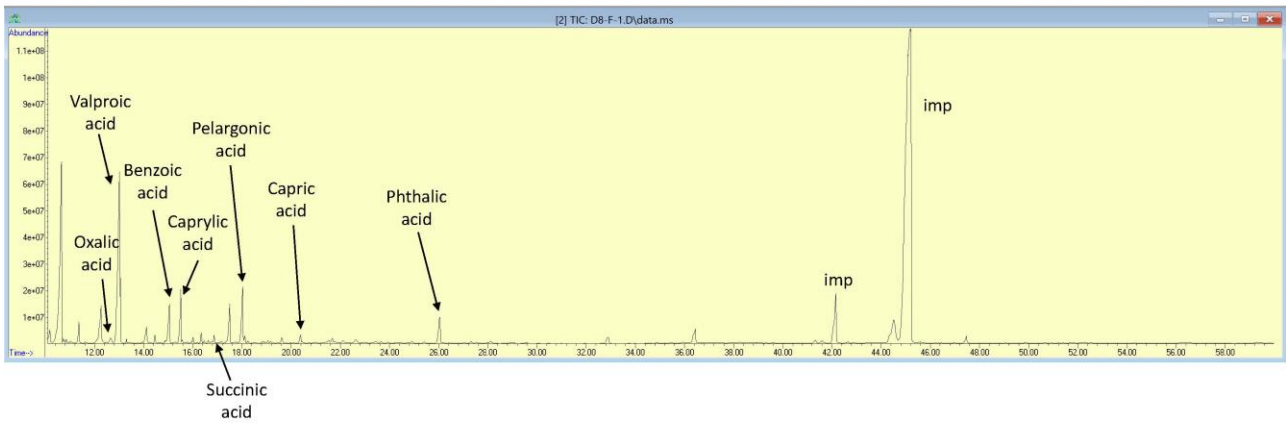
The chromatogram of D1 by base-catalyzed extraction



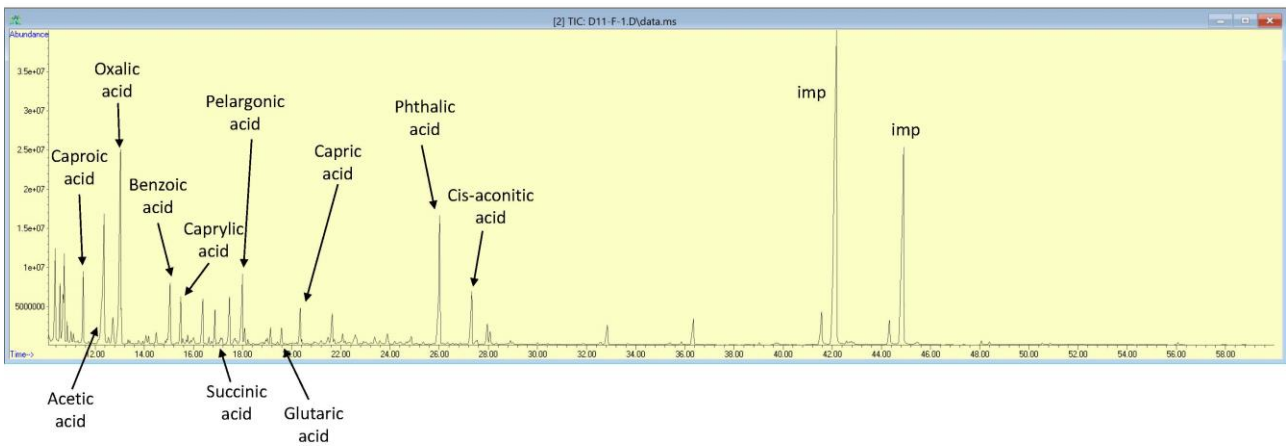
The chromatogram of D4 by base-catalyzed extraction



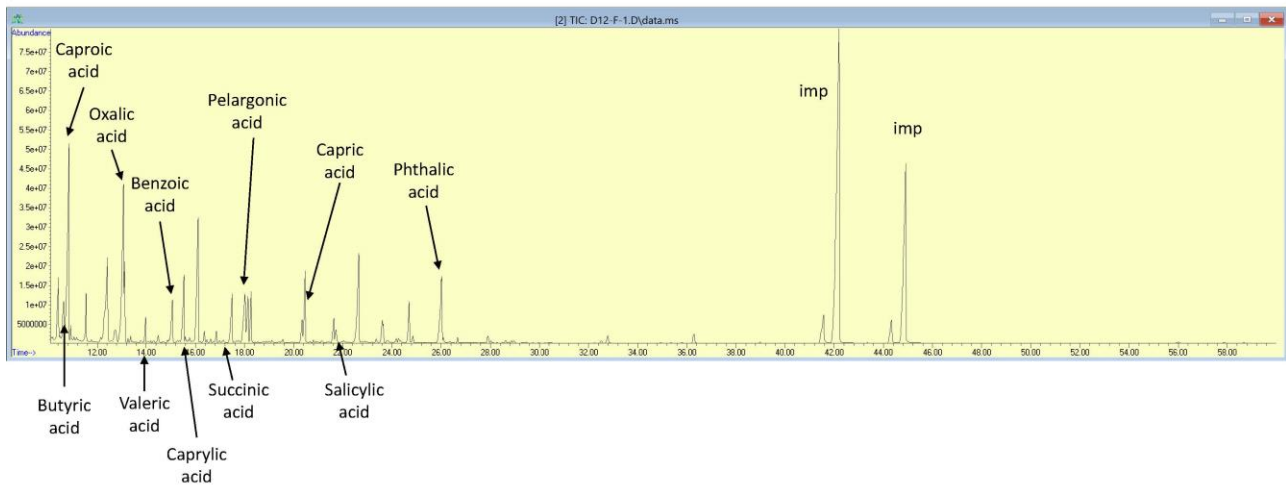
The chromatogram of D7 by base-catalyzed extraction



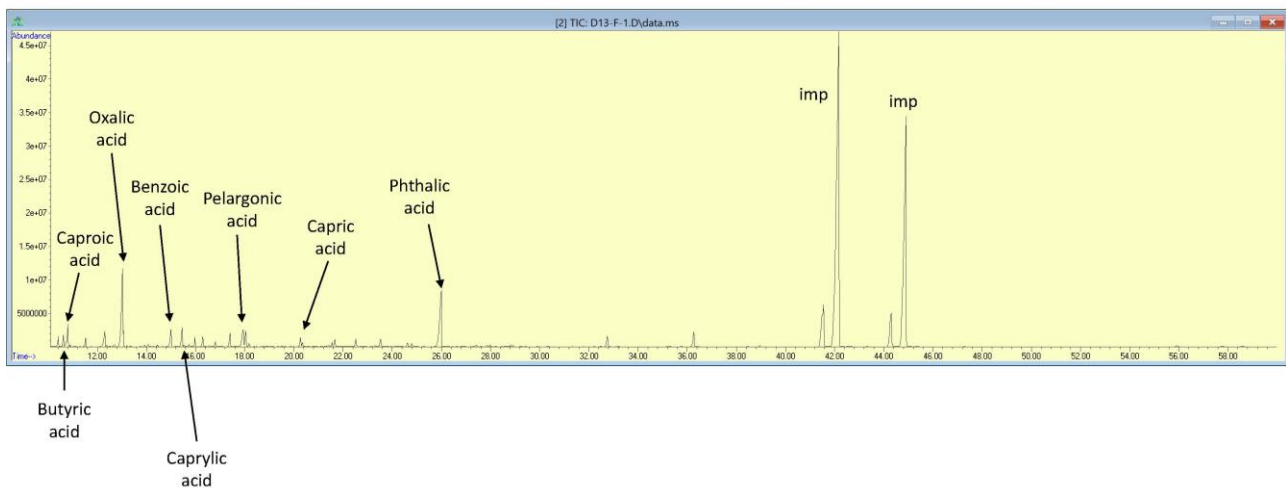
The chromatogram of D8 by base-catalyzed extraction



The chromatogram of D11 by base-catalyzed extraction



The chromatogram of D12 by base-catalyzed extraction



The chromatogram of D13 by base-catalyzed extraction