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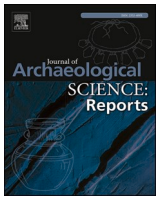

A multidisciplinary investigation of a mummified Egyptian head and analysis of its associated resinous material from the Salinas Regional Archaeological Museum in Palermo (Sicily)

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ABSTRACT

Among the 70 items donated by the abbot Antonio Pietro Paternostro to the former National Museum of Palermo (now Salinas Regional Archaeological Museum) in 1870, an ancient Egyptian mummified human head stands out. In 2022 the finding was submitted for a multidisciplinary investigation that relied upon non-invasive or minimally invasive approaches. Investigations revealed that this is a possible female head, which was likely subjected to trans-nasal craniotomy, and dated to the Egyptian Graeco-Roman period. The head was packed with an abundant amount of resin which was analysed using thermogravimetric analysis, infrared spectroscopy, and gas chromatography with mass spectrometry. The analysis suggested that the resin was most likely comprised of a natural resin, pitch, or tar, from the Pinaceae family of conifers, and mixed with other materials including a fat, oil, or wax. The use of multiple sample preparation techniques for the chromatographic analysis provided a high level of confidence in the identification of a wide variety of compounds, including a range of himachalene derivatives, which indicate the inclusion of cedar tar or oil.

1. Introduction

Among the archaeological collections of the Salinas Regional Archaeological Museum of Palermo, Sicily (Italy), is a mummified human head from Egypt (Salinas, 1882). This finding was donated on 9 September 1870, by the abbot Antonio Pietro Paternostro, the then spiritual director of a historic educational organisation known as the Vittorio Emanuele II Institute. Reportedly, the owner had collected this head, alongside 69 ancient artefacts, during his travels to Africa (L'Amico del Popolo, 1870). However, no further details on the head were disclosed at the time and, in the unpublished list of bequeathed goods, this is simply indicated as a "mummy head" (accession number #14). The box containing the head also held a mummified right hand, though this is not present in the aforementioned list and there is no evidence to suggest the hand and head are from the same individual. In early 2022, one of the authors (DP-M) was invited to investigate the remains and create a multidisciplinary team of scholars which comprised of specialists in archaeology, archaeometry, biological anthropology, chemistry, forensic imaging, and skeletal pathology. Within this framework, a resin sample was provided to the Analytical Methods Laboratory at Staffordshire University (Stoke-on-Trent, UK) and a sample of bandage was provided to the Curt-Engelhorn-Centre of

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Archaeometry (Mannheim, Germany) for radiocarbon dating. Analysis of the samples was conducted over the following months.

2. Aims of this study and ethics statement

The initial aim of this research was to determine whether the mummified head was authentic or a forgery (e.g. Petaros et al., 2015; Szvák et al., 2023). Secondary aims of this analysis were to attempt to gain information about the age, sex, and morphology of the head, establish its chronology, and identify the chemical composition of the resinous sample and, if possible, the original formulation. Permission to investigate the remains was issued by the museum's director on 31 January 2022 (Document #0000500). The remains were handled with care and deference, and samples were already detached from damaged areas; therefore, invasive procedures which may have compromised the integrity of the head were unnecessary (CNR, 2019). This was in keeping with the recent recommendations of the Italian Ministry of Culture (Riga, 2022). Additional ethical approval to conduct this research was granted by Staffordshire University on 6 May 2022 (ref. no. DEF-KES3-015).

3. Materials and methods

The head concerned is represented by a skull partly covered by both soft tissue and resin-soaked bandages (Fig. 1). The right eye is missing but the left is covered by bandaging. Dating to the Graeco-Roman period (332 BCE to 395 CE; Shaw, 2003)[2] is supported by the fact that some of the resin-soaked bandages exhibit gilding (Dunand and Lichtenberg, 2006). Both the bandages and the resin suggest that this is a genuine Egyptian mummy, which underwent an embalming process (Ikram and Dodson, 1998). There was a significant amount of resin within the box containing the head, including large and small fragments, and resin powder. The samples had a black, glossy, resin-like appearance and a woody, camphor-like odour. Some of the larger pieces showed patterns suggesting that the material had previously melted and flowed before hardening, and the flat underside of some fragments showed patterns that appeared to have come from a wooden surface (see, for example, Fig. 2). Samples were brittle and easily ground into a powder-like consistency. Its appearance was similar to material from undated skulls held by the University of Florence (Łucejko et al., 2012), material from the abdomen of Merneith's mummy (26th Dynasty, 664–525 BCE) from Bakenrenef's tomb in Saqqara (Egypt) (Colombini et al., 2000), and to embalming material found on the remains and in the coffin of a noblewoman from the House of Nestawedjat in Thebes (ca. 700 BCE; Vandenbeusch et al., 2021). The remains were safely transported to the Capuchin Catacombs in Palermo, where anthropological and radiological investigations were conducted. This location was selected as the place of analysis as a project using the required equipment was taking place concurrently (Squires et al., 2024). Radiocarbon dating was carried out at the Curt-Engelhorn Centre of Archaeometry. All other procedures, along with comparisons of locally sourced samples of beeswax, fresh Pinaceae (pine) resin, stock chemicals and alkane standards C_8-C_{20} (Fluka) and C_{21}-C_{40} (Fluka), were conducted at the Analytical Methods Laboratory at Staffordshire University. No comparison archaeological resin samples or certified reference standards of other resin-related chemicals were available.

3.1. Biological anthropology

The characteristics of the skull were visually inspected to roughly estimate the age and sex of the individual, bearing in mind that some parts of the head were not directly accessible, such as the cranial sutures. In particular, age could only be inferred by tooth development (AlQahtani et al., 2009) and dental wear (Brothwell, 1981; Lovejoy et al., 1985), while sex estimation was hypothesised based on a qualitative scoring system for sexually dimorphic features of the skull. Each visible trait, as summarised by Schwartz (2007) and White et al. (2012), was scored independently.

3.2. Radiology

Imaging was performed by two highly experienced radiographers working for the company Reveal Imaging Ltd. A radiation risk assessment was completed prior to the analysis in order to minimise hazard. Adjustments made included an assessment of architectural plans for the location where imaging was to take place, the use of warning signs, ensuring people were at a safe distance from the source of radiation, and positioning the beam towards a solid concrete floor. The ALARP (As Low As Reasonably Practicable) principle, limiting radiation dose use, was followed. Radiographs of the cranium and mandible were taken using a portable direct digital X-ray system: the Xograph DR-go system uses a Canon Lumix CDXI 35x43 cm direct digital radiography plate with a Sedecal SP4 4 kW stationary anode X-ray tube and a high frequency generator X-ray machine. A total of six exposures were made. The cranium was examined in the antero-posterior, lateral, and sub-mento vertex projections, and the mandible was examined using postero-anterior and left and right lateral oblique projections. The radiation exposures given followed pre-existing guidelines established by Reveal Imaging for archaeological radiography. The factors were selected relevant to the skeletal element under examination and projection made. For the skull radiographs these were within the range of 60–65 kV and 7–10 mAs at a distance of 1 m. For the mandible radiographs the exposures were in the range of 55–60 kV and 4.5–8 mAs at a distance of 1 m. DICOM image files were viewed and exported using Merge Efilm (v 3.4.0) software. The images were analysed using DICOM Reader software OsirixMD v 13.0.3 (Pixmeo SARL, Switzerland) running on an Apple iMac (Retina 5 k – 2020) using the Ventura operating system.

3.3. Radiocarbon dating

An already detached sample from the bandage was submitted for radiocarbon dating. Firstly, the sample was washed with benzene. As outlined by Aufderheide et al. (2004), this process removes possible contamination of bitumen-based embalming substances. A full description of the applied steps of decontamination, pre-treatment, graphitization, and sample measurement using the MICADAS type accelerator mass spectrometer (AMS) is provided in Kromer et al. (2013) and Lindauer and Kromer (2013). Here, the measurement result is presented in yrs BP as well as the calibrated age (in cal BCE or cal CE). The sample of the mummy was calibrated by using the software OxCal (Bronk Ramsey, 1995) with the INTCAL20 dataset (Reimer et al., 2020).

3.4. Thermal gravimetric analysis (TGA)

For thermogravimetric analysis, a 4–10 mg fragment of the resinous material was placed into a ceramic crucible (PerkinElmer, N5370464) and analysed using a TGA8000 (PerkinElmer, USA) with Pyris software (v.13.4.0.0047). The analysis was conducted under nitrogen and the sample was heated to 1000.00 °C at a rate of 10.00 °C/min, followed by a hold for 1.0 min at 1000 °C.

3.5. Scanning electron microscopy and energy dispersive X-ray analysis (SEM-EDX)

A small sample of resinous material was prepared for imaging by scanning electron microscopy and energy dispersive X-ray analysis by

[2] Various authors refer to either "the Graeco-Roman Period" or the "Ptolemaic Period' and the "Roman Period" as being separate and distinct. Although the actual dates of rule are easily verified the dating of the cultural distinctions (including mummification techniques) are more vague.

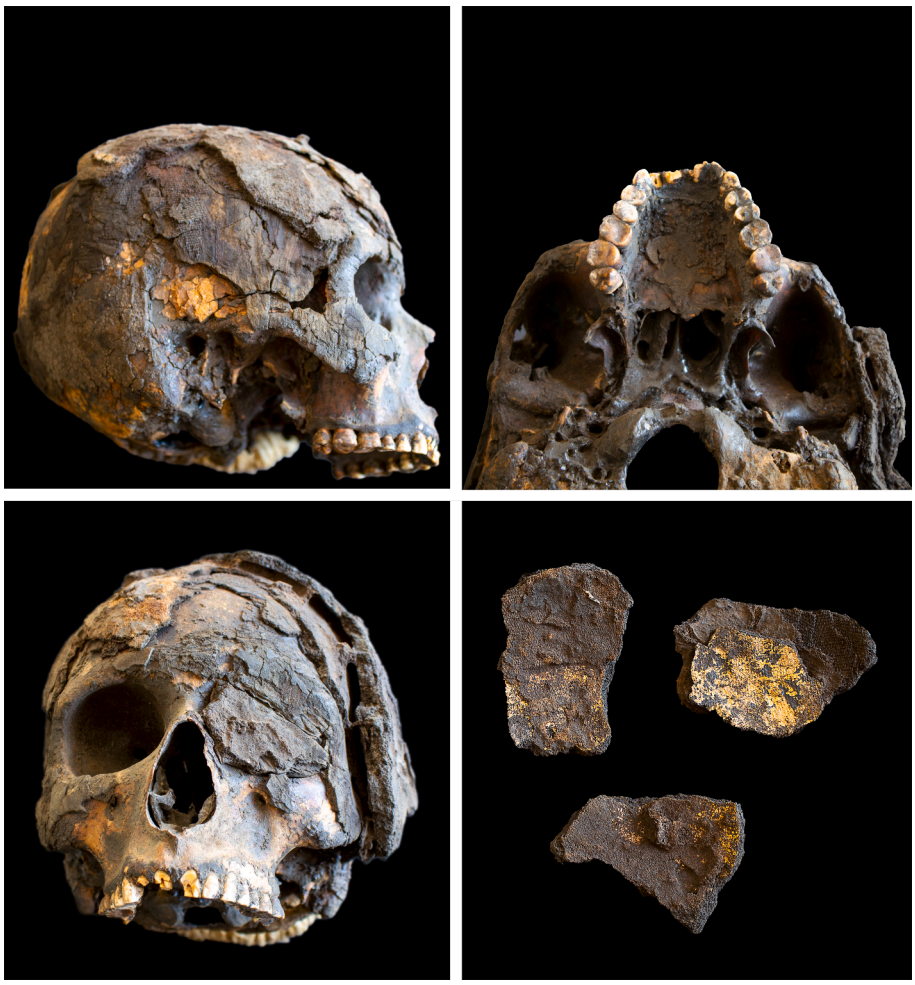

Fig. 1. The Salinas Egyptian head with bandages showing, clockwise from top left: the characteristic gilding, inferior view of the cranium, fragments of the wrappings, anterior view of the cranium. Note, the mandible was present but is not depicted here.

Fig. 2. Resin sample. Left: upper surface showing flow patterns, right: lower surface showing pattern of wooden surface.

splitting a piece of the resin-like material to produce a flat surface. The fragment was mounted on a standard aluminium stub and carbon tab (Agar Analytical) and the analysis was performed using a JEOL 6610-LV with the SEM User Control Interface software (ver. 3.08). The analysis was achieved under low vacuum conditions with the pressure set at 40 Pa. Imaging was conducted using low vacuum secondary electron imaging (LSEI) and backscatter imaging (composite mode). X-ray analysis was performed on six areas using an Oxford Instruments MMax 50 and INCA software (ver. 5.04) with the X-ray collection set for 60 s livetime.

3.6. Fourier transform infrared spectroscopy (FTIR)

Small (approx. 5 mm) flakes of the resinous sample were placed onto the diamond ATR crystal of a Spectrum 2 FTIR universal attenuated total reflectance accessory (UATR) and controlled by Spectrum IR v 10.7.2.1630 software (PerkinElmer, USA). Following collection of a background spectrum, 32 scans were performed from 4000-450 cm^{-1} in reflectance mode with a resolution of 2 cm^{-1} with CO_2 and H_2O correction. A search was performed against available libraries.

3.7. Gas chromatography with mass spectrometry (GC-MS)

GC-MS was performed to investigate the organic chemical composition of the resin-like material. A range of different sample preparation techniques were used. In all cases samples were ground to a powder in an agate pestle and mortar before being weighed into vials for further preparation. GC-MS is an important technique for the analysis of organic mixtures, coupling as it does the separation capability of gas chromatography (GC) along with the identification ability of both the GC retention time and, more powerfully, mass spectrometry. Five different sample preparation methods were used for the GC-MS aspect of this study to ensure a wide range of chemical species were extracted, and to provide additional confidence in the results. Two of the sample preparation methods employed headspace sampling as the material was odorous and therefore contained volatile organic compounds which should be present in the headspace. A dedicated headspace sampler was trialled (as it has not previously been used for this sample type) and compared with solid phase microextraction (SPME). This approach was adopted as Hamm et al. (2004) demonstrated that it was possible to successfully extract over 100 different chemicals from a range of resins and resinous archaeological samples using SPME. Solvent extraction was used for the remaining sample preparation methods as this approach allows a range of different polarity solvents to be used and for derivatisation to be employed. A simple ethanol extraction was trialled for comparison with hexane extraction as Colombini et al. (2000) showed the latter to be successful for the determination of a range of non-polar chemicals in archaeological embalming materials. Derivatisation was also used for those polar compounds which are less amenable to chromatography, such as fatty and terpenoid acids (see e.g. Colombini et al., 2000; Łucejko et al., 2012).

3.7.1. Headspace GC-MS

For headspace GC-MS analysis, 0.25 g of powdered sample was transferred to a 50 mL headspace vial with a screw top cap and PTFE septum (Thames Restek). The sample was placed into the carousel of the headspace sample (HS-40, PerkinElmer) and heated for 30 minutes at 125 °C. The needle temperature was 135 °C and the transfer line 145 °C. The vial was pressurised for two minutes at 15 psi before the sample was directed onto the GC via the inert transfer line. The GC, a Clarus 690 (PerkinElmer), had an injector temperature of 220 °C and the carrier gas was helium (BOC) at a pressure of 10 psi. The separation was performed using an ELITE-5MS column (PerkinElmer) with dimensions 30 m x 0.25 mm x 0.25 μm. The GC temperature programme started at 50 °C with a hold of two minutes before ramping at 6 °C/min to 200 °C where the temperature was held for one minute. A second ramp then took the temperature to 270 °C where it was held for two minutes. The overall run time of the programme was 37 minutes. The MS was a SQ 8 T (PerkinElmer) scanning from 50-500 *m/z*, and the instrument control software employed was Turbomass ver. 6.1.2.2048.

3.7.2. Solid phase microextraction (SPME) and GC-MS analysis

Following the method described by Hamm et al. (2004) and using a 100 μm polydimethylsiloxane fibre (Supleco), 0.05 g of powdered sample was placed into a 6 mL headspace vial (Thames Restek) with a 60 mm thick PTFE/silicone septum (Supelco) and a crimp top (Thames Restek). The resin was then heated in a fan oven (Thermo Heraeus UT12) for 75 minutes at 80 °C with the SMPE fibre exposed to the sample. Akin to the headspace analysis, the GC–MS used here was the Clarus 690 and SQ 8 T. In this instance, the fibre was desorbed in the injector at 250 °C for five minutes. The separation was performed using the same ELITE-5MS column as described above, this time with a temperature programme starting at 40 °C with a one-minute hold, ramping at 9 °C/min to 130 °C, and then at 2 °C/min to 230 °C. The carrier gas was helium with a pressure of 10 psi and no split. The MS scanned from 50-500 *m/z* for the entire runtime.

3.7.3. Ethanol extraction and GC-MS analysis

A simple ethanol extraction was performed on 0.27 g of powdered sample by adding 5 mL of HPLC grade methanol (Fisher) and placing the sample into an ultrasonic bath for 30 minutes. Although the sample appeared to be almost fully dissolved, 4 mL of the solution was extracted and a further 5 mL of methanol was added to the remaining solid which was then sonicated for an additional 15 minutes. The two extracts were combined, filtered through 0.45 μm Polyvinylidene fluoride (PVDF) syringe filters (Whatman Puradisc, 25 mm diameter), and evaporated under nitrogen (BOC) to approximately 5 mL with 1 mL then transferred to an autosampler vial. The GC-MS instrument, column, and software were as described for the headspace analysis. The temperature programme started at 50 °C with a two-minute hold, ramping at 15 °C/min to 300 °C, and then holding for six minutes. Helium was used as the carrier gas with a flow of 1 mL/min and an injector split of 20 mL/min. The injection volume was 1 μL with an injector temperature of 220 °C. The MS was set with a three-minute solvent delay and then scanned from 50-500 *m/z* for the remaining runtime.

3.7.4. Extraction of non-polar compounds and GC-MS analysis

Based on the methods described in Colombini et al. (2000) for non-polar compounds, 0.5 g of ground material was dissolved in 5 mL hexane (Fisher, HPLC grade) and placed in an ultra-sonic bath for 30 minutes. After sonication the sample consisted of an amber-coloured solution and two layers of solid material, a lower layer of black solid (as seen before sonication), and an upper layer of a finer, paler brown solid. The clear solution was transferred into a clean vial while another 5 mL of hexane was added to the solid material; this was then sonicated for another 30 minutes (ensuring the temperature of the bath remained below 40 °C). The second portion of hexane extraction was combined with the first and evaporated to approximately 2 mL. A blank sample of 2 mL hexane was also prepared. Florisil Solid Phase Extraction cartridges (Alltech, 6 mL) were then conditioned with 10 mL of dichloromethane, followed by 10 mL of hexane before adding the extraction solution or hexane blank. The cartridges were eluted with 10 mL of hexane and the eluent was collected. This was then evaporated to approximately 1 mL under nitrogen and transferred to autosampler vials before analysis using the same instruments and conditions as the other solvent extractions.

3.7.5. Derivatisation of polar compounds and GC-MS analysis

A simple derivatisation process was also conducted. Approximately 9 mg of the resin sample was dissolved in 1 mL of dichloromethane (Fisher, HPLC grade) for 15 minutes. The sample was left overnight (and remained in solution) and the following morning 30 μL BSTFA (N,O-bis (trimethylsilyl)trifluoroacetamide with 1 % TCMS trimethylchlorosilane, LiChropur) was added and the sample placed in an oven for one hour at 70 °C. Once cooled, 100 μL isooctane (2,2,4-trimethylpentane, Sigma, anhydrous 99.8 %) was added and the sample was transferred to a small-volume autosampler vial (Fisher) for analysis using the same conditions as the ethanol extractions.

4. Results

4.1. Biological anthropology

Evaluation of the head revealed the presence of permanent teeth,

including the maxillary third molars. There is significant occlusal wear on the teeth, and fractures are noted on the two central maxillary incisors and the right lateral maxillary incisor which has led to the loss of dental crowns. The limited analyses that could be carried out suggested that this was a young-to middle-aged adult (20–50 years) at the time of death. However, it is known that dental abrasion is particularly common among ancient Egyptian populations, which could lead to overestimation of the actual age of this individual (Forshaw, 2009). The sexually dimorphic traits of the skull are compatible with those of a female.

4.2. Radiology

Radiological analysis indicates that there is no evidence of bone or tooth conditions, such as periapical cysts or abscesses. It is not possible to determine the route for excerebration (the removal of brain) from the X-rays, but this procedure has been performed as witnessed by the resin in the cranial cavity and confirmed by the introduction of a wooden stick through the nasal cavity directly into the cranial fossa. This indicates perforation of the cribriform plate. It has been presumed that the resin has "leaked" out of the cavity via the foramen magnum at some point in time as it has pooled in the bottom of the box container. The evidence thus suggests a well-mummified head with evidence of excerebration and packing of the cranial cavity with resin (Fig. 3).

4.3. Radiocarbon dating

The result of radiocarbon dating (commission no. 220755) is shown in Table 1. The 14C measurement result which is not a calendar age is given as BP (before present). The 14C ages must be calibrated to give absolute calendar ages. The results of the calibration are given in the two columns showing calibrated age ranges with a corresponding 68 % probability (1σ) and 95 % probability (2σ). The calibrated ages of around 3000 BCE, indicate an origin of the bandage sample from the Early Dynastic Period (3000–2686 BCE; Shaw, 2003) suggesting that the textiles employed were reused, as they are much older than the expected Graeco-Roman chronology of the mummified head. Reuse and recycling of materials from former burials was a practice applied in all periods of ancient Egyptian history (Taylor, 2001). Even though the pretreatment is well suited to remove remains of bitumen, it cannot be excluded that

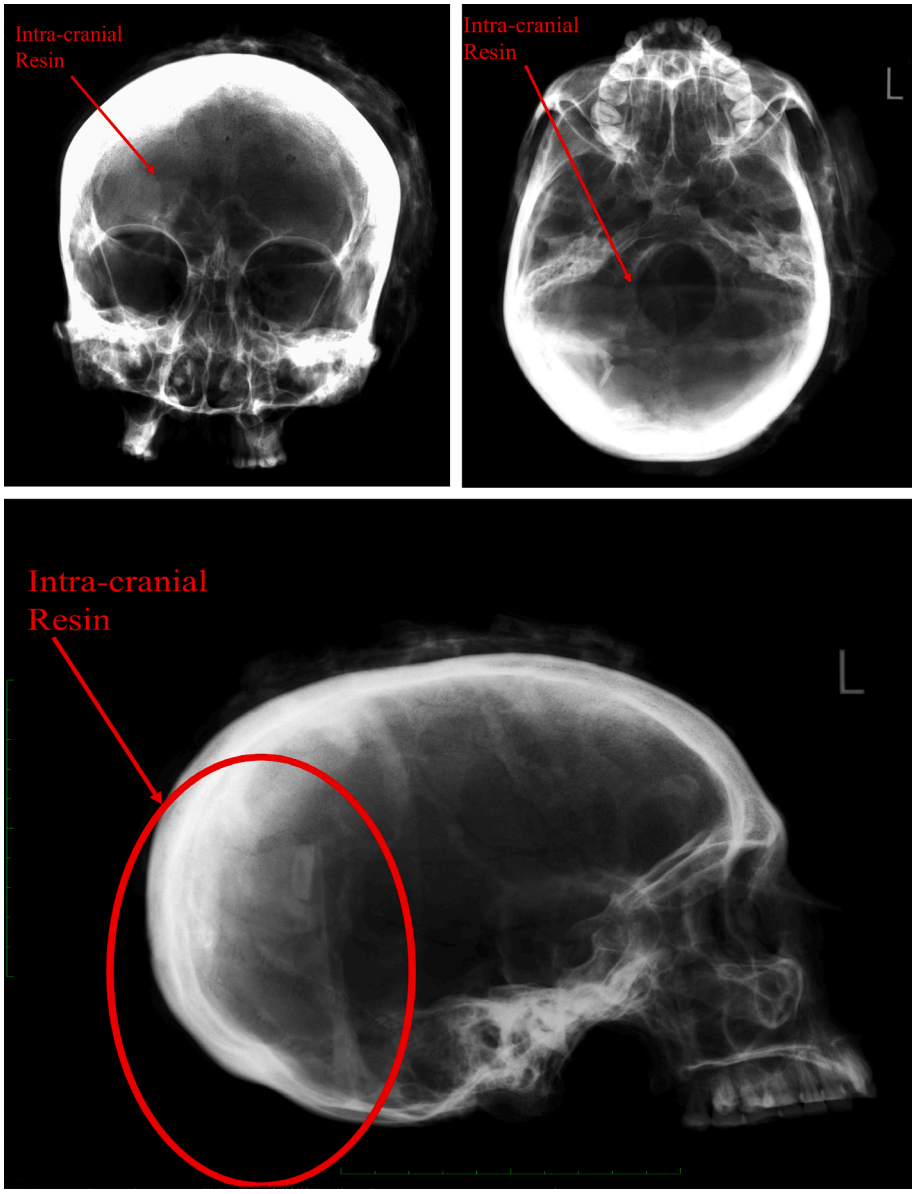

Fig. 3. Radiographs of the cranium. Clockwise from top left: anterior, inferior, and lateral views of the cranium.

Table 1
Radiocarbon dating of a textile fragment from the Salinas mummy head.

Lab No	Sample name	14C age (yr BP)	±	13C AMS (‰)	Calibrated age (68 % probability)	Calibrated age (95 % probability)	C (%)	Material
60,021	GMP 426	4331	24	−18.4	Cal BCE 3008–2900	Cal BCE 3013–2897	57.4	Textile

some contamination is still present. At this point, bitumen is radiocarbon-dead; furthermore, it would take a significant amount of contamination to make the sample's date older than presented in the results (Aufderheide et al., 2004; Quiles et al., 2014).

4.4. Thermal gravimetric analysis (TGA)

The results of the thermogravimetric analysis showed that the majority of the sample mass was organic matter. Here, there was an 84–87 % loss in mass when heated between 100 and 550 °C. The thermogram for one sample is shown in Fig. 4 and, as illustrated clearly by the derivative (dotted black line), the loss of the mass in this temperature range occurred in two phases indicating that the sample was comprised of a mixture of different organic compounds. Of the nine sub-samples analysed, most (n = 7) thereafter showed another 2–3 % loss of mass between 550 and 1000 °C, leaving 10–11 % of inorganic material remaining at the end of the analysis. For two samples however, the thermogram showed a more pronounced slope after 750 °C with another 7–8 % of the mass being lost between 750 and 1000 °C, indicating a higher proportion of inorganic material. For these samples only 2–4 % of the total mass remained after heating to 1000 °C. One of these samples is shown in Fig. 4.

The loss of mass observed for the resinous material from the Salinas sample was similar to that seen from the TGA analysis of 'natural resin' thought to be from a Pinaceae variety, which was sampled and analysed from a coffin in a Ming tomb in Shaanxi (China) (Guo et al., 2021). Although the information gained from our TGA analysis is rather limited as reference samples were not analysed, it does support the FTIR analysis (see below) in demonstrating that the Salinas sample contained very little inorganic material and the results are shared here to aid future researchers who may use this technique.

4.5. SEM-EDX

Secondary electron imaging of the inside surface of the resin-like material showed a smooth surface without any features of interest. X-ray analysis was used to explore whether biomarkers for bitumen (e.g. sulphur, vanadium, nickel, and molybdenum) were detectable in the sample (as per Mikhailenko, 2015; Yatsishina et al., 2019). Sulphur was detected at just over 1 % however, none of the other target elements were detected. Bitumen typically contains between 0.9–6.6 % sulphur (Mikhailenko, 2015), however, the presence of sulphur in the material may also be due to its inclusion in organic chemicals. Overall, the SEM-EDX results showed very little variation across the sample with carbon making up approximately 80 % of the material while 18 % of the resin was composed of oxygen. These findings support the results obtained from TGA analysis, in that the majority of the material is organic.

4.6. FTIR

The FTIR spectrum (Fig. 5) showed peaks due to –OH groups (3299 cm^{-1}), CH_2 and CH_3 aliphatic stretches (2922 and 2852 cm^{-1}), the carbonyl C = O stretch (1703 cm^{-1}), aromatic / olefinic C = C bonds (1611 cm^{-1}), and other peaks generally typical of many types of organic

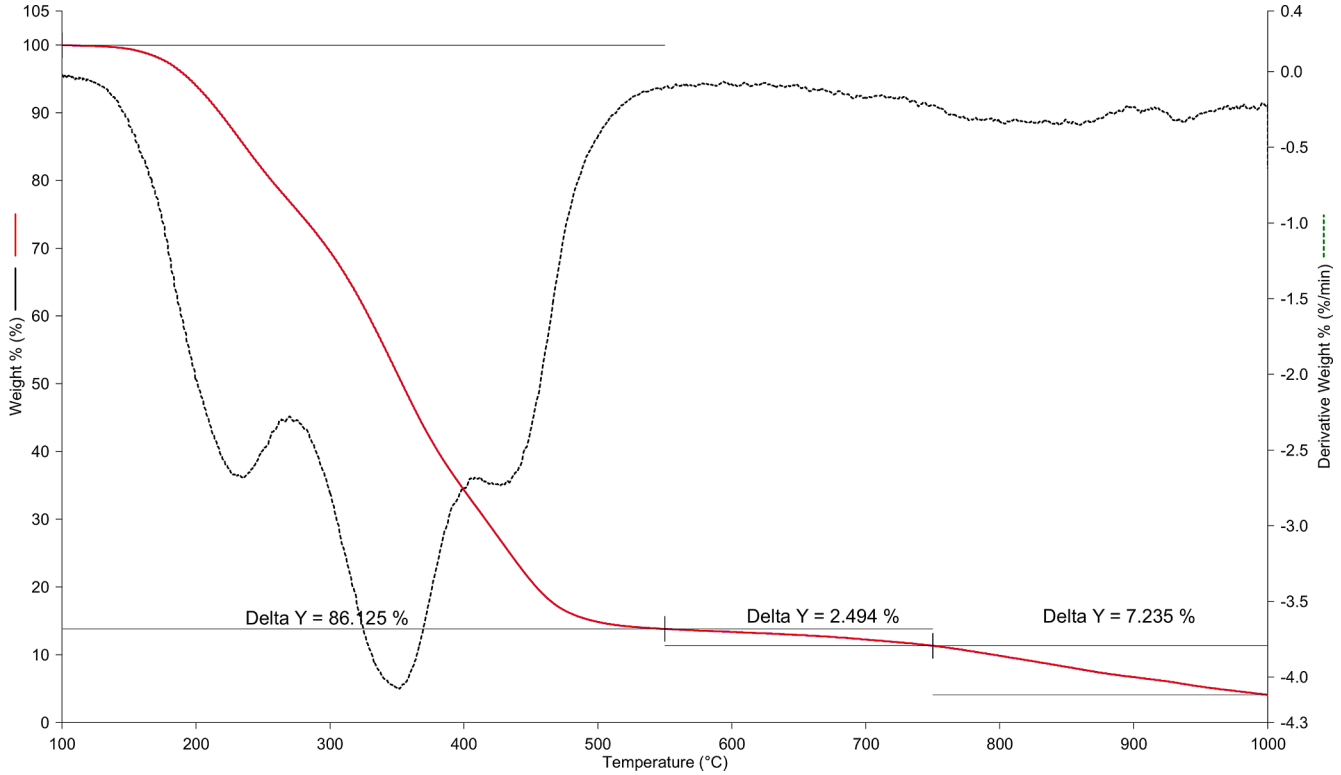

Fig. 4. Thermogram for resin-like sample showing the mass lost (solid red line) and using the derivative (dotted black line) to better visualise the step changes (for interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article).

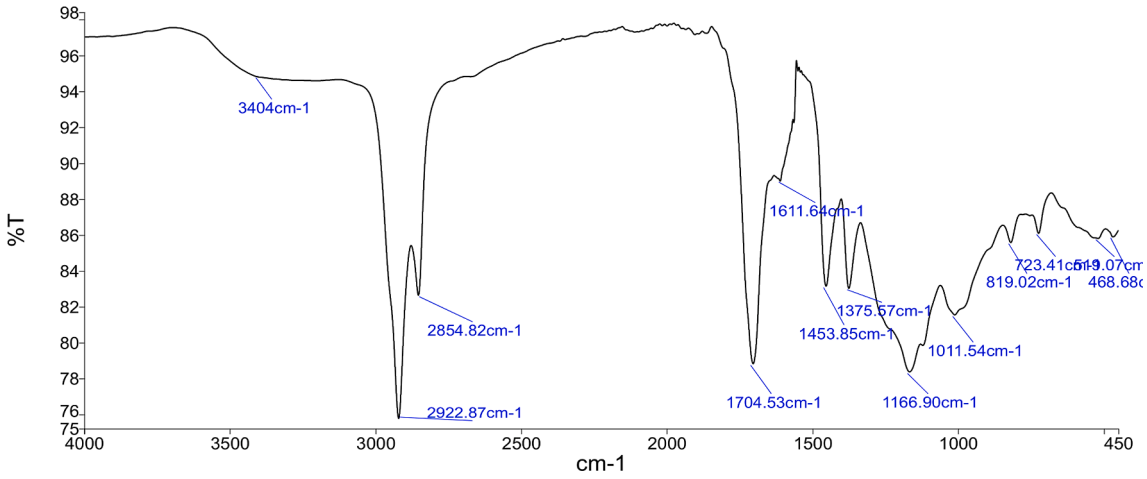

Fig. 5. FTIR spectrum of resin-like material from the Salinas mummy head.

resins and related embalming materials (Łucejko et al., 2012).

The instrument library match software produced an 87 % match to 'pine' resin (as named by the PerkinElmer library), but as illustrated in Fig. 6, some areas of the spectrum do not match. Therefore, pine resin cannot be confirmed by this method, especially as other resinous materials often used in embalming are not included in the library.

The FTIR spectrum of the resin-like material shows that it is a complex organic mixture as demonstrated by the number of peaks, and the presence of the CH_2 and CH_3 aliphatic stretches, and the carbonyl C = O stretch. Comparisons with FTIR spectra of archaeological samples analysed by Łucejko et al. (2012) show that the Salinas sample does not contain any of the inorganic materials identified in that study. While it would be unlikely to find a clear match between samples given the variations in potential embalming recipes (Buckley and Evershed, 2001), the Salinas resin spectrum does appear similar to that recorded by Oras et al. (2020). These authors identified embalming material in the abdominal cavity of the mummy of an older child from the Late/Graeco-Roman Period, which is held in the collections of the University of Tartu Art Museum (Estonia). Interestingly, this mummy was also the subject of a multidisciplinary investigation which concluded that the mummy was Egyptian and from the Late/Graeco-Roman Period.

4.7. GC-MS

Chromatograms were integrated using the Turbomass software (ver. 6.1.2.2048) and spectra were searched against the NIST library (ver. 2.3, 4 May 2017). The Reverse Match score (out of 1000) was used to evaluate the quality of the match. Chemical names and Chemical Abstract Service (CAS) numbers were checked, the latter of which were obtained from online websites (The Good Scents Company, 2021; PubChem, 2022; NIST, 2023). Peak identifications were compared with chemicals identified by researchers, including Colombini et al. (2000), Hamm et al. (2004), Łucejko et al., (2012, 2017), El-Sayed (2021), and Huber et al. (2022). Peak identifications were compared with those noted by Colombini et al. (2000) and Buckley and Evershed (2001). Table 2 was constructed to compare the range of chemicals identified in the various preparations of the Salinas sample with that of the lists provided by Hamm et al. (2004), who used SPME extraction, and Łucejko et al. (2012), who used solvent extraction and derivatisation.

4.7.1. Headspace GC-MS

The headspace analysis, using the HS-40, produced 18 GC peaks. The largest peak areas were identified as camphene (40 %), α-pinene (22 %), α-dehydro-*ar*-himachalene (15 %), and *ar*-himachalene (5 %), each with

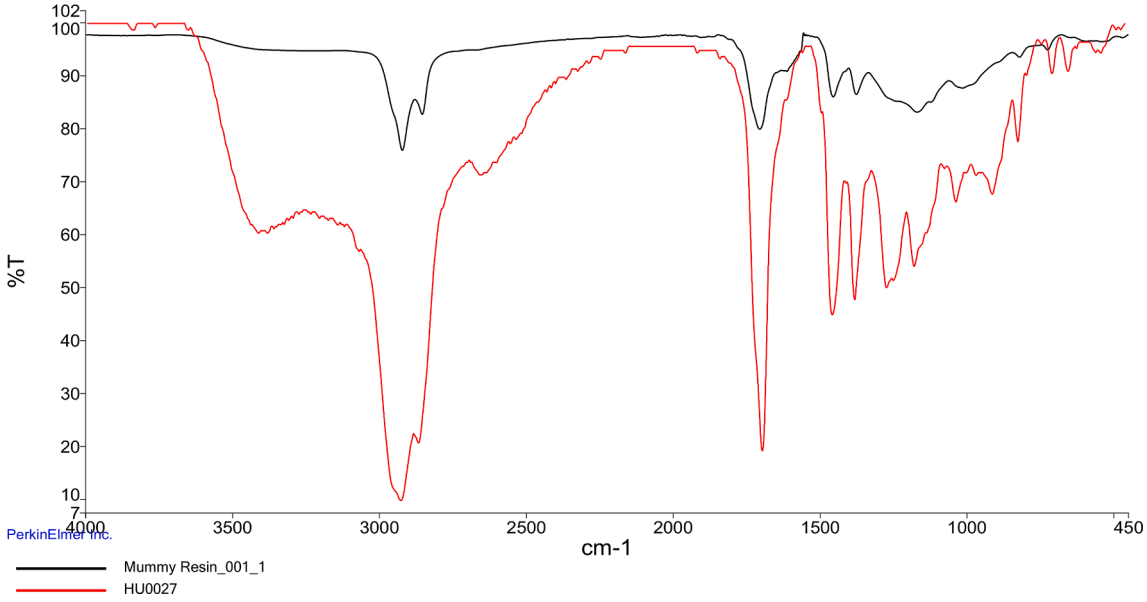

Fig. 6. Library match of 'pine' resin to the resin-like material from the Salinas mummy head.

Table 2
Significant compounds identified by GC–MS (RM: Reverse Match, RT: Retention Time).

Chemical	CAS No.	HS		SPME		Ethanol ext.		Hexane-SPE		Hamm et al. (2004)	Łucejko et al. (2012)
		RM	RT	RM	RT	RM	RT	RM	RT		
α-pinene	80-56-8	933	6.76	919	6.43					x	
camphene	79-92-5	962	7.18	972	6.75					x	
camphor	464-48-2			937	10.48					x	
fenchol	22627-95-8	719	11.69	863	9.95					x	
borneol	464-45-9			795	10.93					x	
verbenone	1196-01-6			895	11.57					x	
bornyl chloride	464-41-5	848	13.01	971	10.89						
α-terpineol	98-55-5	786	13.69	942	11.31					x	
longicyclene (+)-	1137-12-8			881	15.56					x	
longifolene	475-20-7			896	16.61	897	10.35	886	10.36	x	
γ-himachalene	53111-25-4			817	18.78						
α-dehydro-*ar*-himachalene	78204-62-3	891	21.22	952	19.90	943	11.14	947	11.15		
calamene	483-77-2			835	20.22					x	x
γ-dehydro-*ar*-himachalene	51766-65-5	811	21.56	914	20.42	929	11.28	953	11.28		
ar-himachalene	19419-67-1	883	21.74	938	20.74	936	11.33	952	11.34		
guaiazulene	489-84-9	731	22.55	841	22.22	855	11.65	855	11.66		
dihydro-*ar*-turmerone	30666-87-6	708	22.87	912	22.90	855	11.72	854	11.73		x
caryophyllene oxide	1139-30-6	675	23.58					820	12.07	x	
allohimachalol	19435-77-9			802	26.09						
cadalene	483-78-3			849	26.34	874	12.40	857	12.41		x
palmitic acid	57-10-3					893	14.28	898	14.32		x
10,18-bisnorabieta-5,7,9(10),11,13-pentaene	6566-19-4							849	15.29	x	
octadecenoic acid isomer*						934	15.39	926	15.43		
stearic acid	57-11-4					900	15.52	881	15.57		x
retene	483-65-8					879	16.02	876	16.03		
methyl dehydroabietate	1235-74-1					911	16.65	936	16.66	x	x
pentacosane	629-99-2					863	18.40	886	20.83		
α-himachalene (−)-	3853-83-6					824	21.83	844	21.88		

* Note: This compound was listed by the NIST library with the top hit listed as *cis*-13-octadecenoic acid. Other isomers, including *cis*-9-octadecenoic acid (oleic acid), were observed in the top five hits. As standards of the two compounds could not be analysed during this research project, the identification cannot be confirmed.

NIST reverse match values above 800 (out of 1000), indicating a good match. Significant peaks are presented in Table 2.

4.7.2. Solid phase microextraction (SPME) and GC-MS analysis

The SPME analysis produced 45 peaks with many of the same chemicals presented in the headspace GC-MS results (Table 2). However, in this case the largest peak was identified as α-dehydro-*ar*-himachalene (26 %), while other abundant chemicals, including longifolene (7 %) were also recorded.

4.7.3. Ethanol extraction and GC-MS analysis

The ethanol solvent extraction was comprehensive and produced more than 50 integrated peaks. Table 2 illustrates the most abundant peaks from the ethanol extraction were palmitic acid (16 %), an octadecenoic acid isomer (13 %), and stearic acid (7 %). The octadecenoic acid isomer was listed by the NIST library with the top hit given as *cis*-13-octadecenoic acid, followed by other isomers, including *cis*-9-octadecenoic acid (oleic acid). As standards of the two compounds could not be analysed during this research project, the identification cannot be confirmed.

4.7.4. Hexane extraction with SPE of non-polar compounds and GC-MS analysis

The hexane extraction with SPE was also successful, producing more than 50 integrated peaks. The largest peaks were α-dehydro-*ar*-himachalene (7 %), an octadecenoic acid isomer (7 %; though there remain the same identification issues as described above), and palmitic acid (6 %). Retene was also found with a peak area just under 4 %. Table 2 shows that the chemicals seen in the ethanol were represented in the hexane extraction, along with caryophyllene oxide and 10,18-bisnorabieta-5,7,9(10),11,13-pentaene (1,2,3,4-tetrahydroretene).

4.7.5. Derivatisation of polar compounds and GC-MS analysis

Although the derivatisation produced a limited number of peaks, those peaks which were identified to a high degree of confidence by the NIST library as trimethylsilyl (TMS) derivatives of significant chemicals are listed in Table 3. The fatty acids which were found could be from beeswax, vegetable oil, animal fats, or potentially adipocere of human or animal lipids (Colombini et al., 2000; Buckley and Evershed, 2001; Łucejko et al., 2017). Isopimaric acid and dehydroabietic acid have been reported in fresh resins from *Pinus pinea* and *Pinus sylvester* by Colombini et al. (2000) and 7-oxodehydroabietic acid in 'ancient pine resin' (Colombini et al., 2000).

In order to explore the likely source of the main resinous ingredient, chromatograms were extracted for the mass/charge values characteristic of the diterpenoids routinely found in conifer resin (*m/z* 241 and 257) and those characteristic of the triterpenoid compounds (*m/z* 189, 203, 410, 424, and 426) which are marker compounds for mastic resin (Colombini et al., 2000). Fig. 7 shows the extracted mass chromatogram for diterpenoids. This reveals a number of abundant peaks, while an extracted mass chromatogram for triterpenoid compounds (not shown) did not show any significant peaks. These results indicate that mastic resin was not an ingredient of the embalming mixture but, instead, some

Table 3
Derivatised chemicals identified in the Salinas resin sample.

RT	PA %	RM	Chemical
13.452	2.816	858	myristic acid, TMS
14.102	0.875	886	pentadecanoic acid, TMS
14.607	1.561	943	palmitelaidic acid, TMS
14.748	17.907	913	palmitic acid, TMS
15.161	0.668	922	heptadecanoic acid, TMS
15.778	16.686	941	oleic acid, TMS
15.920	11.557	926	stearic acid, TMS
16.587	0.940	900	isopimaric acid, TMS
16.816	4.546	763	dehydroabietic acid, TMS
17.917	0.766	860	7-oxodehydroabietic acid, TMS

RT = Retention Time, PA% = Peak Area %, RM = Reverse Match, TMS = Trimethylsilyl.

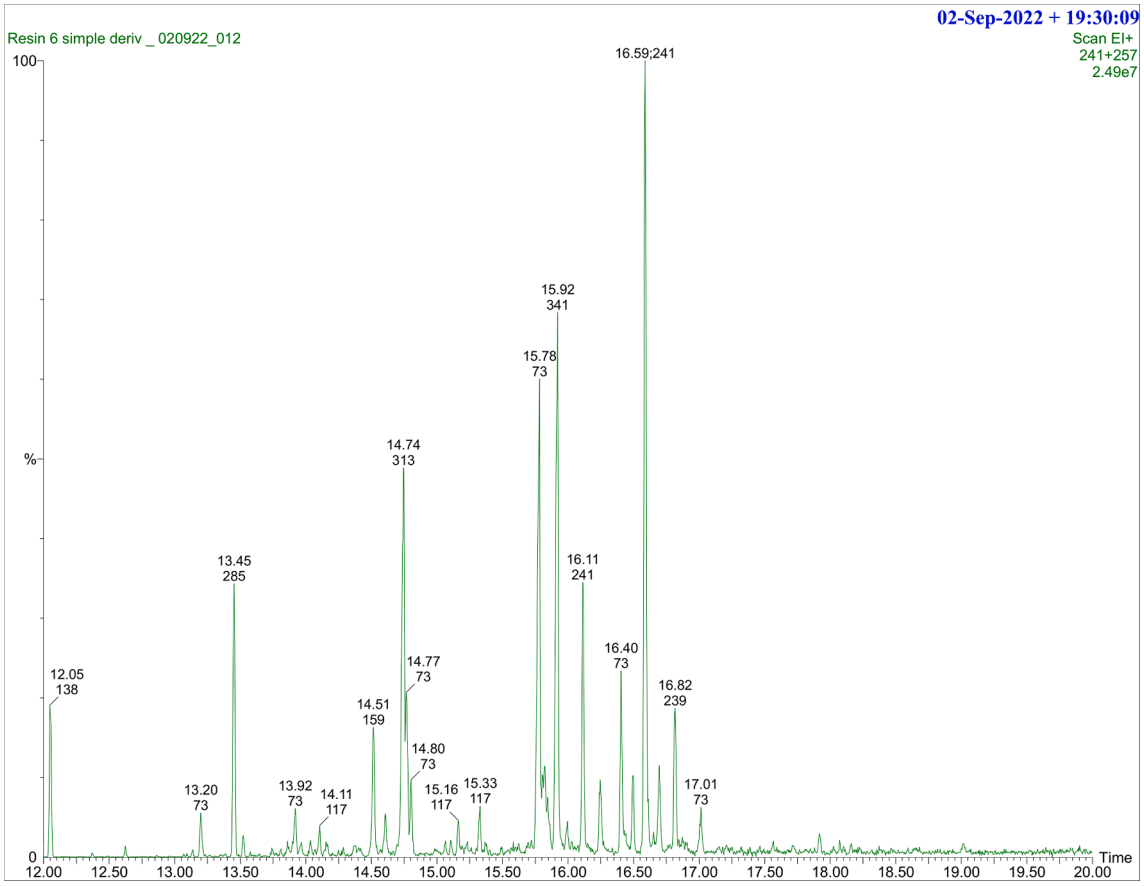

Fig. 7. Extracted chromatogram using characteristic masses for derivatised diterpenoids.

type of Pinaceae (conifer resin) was used.

5. Discussion

The mummified head described in this paper is of particular historic interest. Following Napoleon's campaign in Egypt (1798–1801 CE), there was growing interest in Ancient Egypt in Sicily and further afield (Militello, 2014). Consequently, Egyptian antiquities were collected and displayed, which ultimately paved the way for notable research and studies (Raffiotta, 2020; Raffiotta and Messina, 2020; Nuzzolo, 2021). An examination of the Salinas mummified head revealed a young-to-middle-aged adult female, who was afforded an elaborate embalming process characterised by excerebration followed by the abundant use of resins, which, along with gilding of the bandages, is consistent with funerary treatment during the Graeco-Roman period.

The FTIR and TGA analysis demonstrated that the Salinas sample was a complex mixture, comprised almost entirely of organic material. Although these techniques are unable to identify specific ingredients in the balm, a library of such results may provide a future resource for researchers who would benefit from using these simple, quick, and relatively inexpensive analytical techniques. The SEM analysis not only confirmed the organic nature of the material, and the absence of metals associated with bitumen, but also identified the presence of sulphur which was not seen using the other techniques.

The various GC-MS analyses of the resinous matter proved to be particularly informative. The use of multiple extraction methods provided a high degree of confidence in the identification of the 28 chemicals (Table 2), with the derivatisation providing high match scores for the seven fatty acids and three diterpenes (Table 3). The significance of the chemicals detected was further considered using one or more of the following criteria: their appearance in relevant literature, their occurrence in multiple samples using different extraction methods, and their high reverse match score from the NIST library. Despite the value of adopting GC-MS analysis in this study, the majority of chemical identifications are tentative as reference standards were not available. Additionally, the sample preparation methods used were insufficient to determine the presence of critical wax esters and related compounds and this should be the focus of further chemical analysis of the Salinas resinous material.

The two main components that are most reported in samples of embalming materials are mastic resin from *Pistacia* trees and pine resin from the coniferous Pinaceae species (e.g. Colombini et al., 2000; Buckley and Evershed, 2001; Łucejko et al., 2017). To distinguish between these two materials derivatisation and GC-MS analysis of the sample is recommended, with diterpenoids cited as key marker compounds for coniferous resins and tars, and triterpenoids cited as characteristic of mastic resin (e.g. Colombini et al., 2000). The Salinas sample derivatisation produced good evidence for the presence of a coniferous resin. Dehydroabietic acid and 7-oxodehydroabietic acid have been reported by Colombini et al. (2000) as the main diterpenoid acids found in ancient pine resin, isopimaric acid was seen in fresh pine resin by Colombini et al. (2000), and both 7-oxodehydroabietic acid and isopimaric acid were also observed in fresh pine resin samples analysed at Staffordshire University in the present investigation. Jones et al. (2014) reported all three chemicals in funerary wrappings from bodies in Predynastic period tombs (pit graves) at Mostagedda in the Badari region (Upper Egypt) from the Badarian (Late Neolithic) and Predynastic (Chalcolithic) periods (ca. 4500–3350 BCE), indicating the extended use of these materials in funerary rites.

Hjulström et al. (2006) and Jones et al. (2014) discuss the presence and relative abundance of retene and methyl dehydroabietate in Pinaceae resin and tar. Łucejko et al. (2017) also note that methyl

dehydroabietate derives from the reaction of dehydroabietic acid which occurs during the dry distillation of resinous wood, although as shown by Hjulström et al. (2006), methyl dehydroabietate can occur in unheated resin with lower abundance. In the present study, retene and methyl dehydroabietate were detected, and although quantification was not performed, the relative abundance of these two compounds indicates that some heat-processing may have taken place (Jones et al., 2014). Hamm et al. (2004) also reported retene, camphene, methyl dehydroabietate, and 10,18-bisnorabieta-5,7,9(10),11,13-pentaene (1,2,3,4-tetrahydroretene) in their SPME extractions of pine pitch – all of which were identified by NIST as present in the Salinas sample. As a further indication of the presence of a wood tar, several phenanthrenes (as discussed by Łucejko et al., 2012, 2017) were tentatively identified in the Salinas sample. It should, however, be noted that Hamm et al. (2004) warn that the identification of resinous substances present in archaeological samples of embalming material is sometimes impossible because of the chemical transformation that terpenes undergo during ageing.

In trying to establish the source material for the resin-like sample, it was noted that α-pinene is not very helpful for identifying the source material as it appears in a wide range of resins (Colombini et al., 2000; Hamm et al., 2004). However, longifolene was also present in our sample and is considered characteristic of a conifer resin, having been identified in the resin of *Pinus pinea* by Hamm et al. (2004). Longifolene, together with calamene and cadalene (likewise found in our SPME extract), have similarly been reported in oils from the Atlas cedar (*Cedrus atlantica*), a member of the pine family Pinaceae (Łucejko et al., 2012; Huber et al., 2022).

The presence of various himachalenes in the Salinas samples appear to be significant even though they are rarely mentioned in the literature concerning embalming materials. Huber et al. (2022) propose that α- and γ-dehydro-*ar*-himachalene and *ar*-himachalene, in particular, might be useful biomarkers for material originating from the Atlas cedar species, as they appear to be very stable with aging and are rarely found in other plant extracts. Vandenbeusch et al. (2021) found *ar*-himachalene in resinous material from the mummified remains of a noblewoman from the House of Nestawedjat in Thebes (Egypt; ca. 700 BCE), along with dihydro-*ar*-turmerone, which was also found in the Salinas sample. Yet, Vandenbeusch et al. (2021) also found cuparene and aristolone, though these were not found in the present study. Significantly, Rageot et al. (2023) similarly found vessels at the 26th Dynasty Saqqara embalming facility (Egypt) which they reported as containing oils or tars of cedar, as identified by the presence of γ-dehydro-*ar*-himachalene, *ar*-himachalene, and α-dehydro-*ar*-himachalene.

In trying to further confirm the most likely resin material in the present sample, information from Evershed et al. (1997), Hamm et al. (2004), and Huber et al. (2023) would suggest that the following resins are *not* ingredients in the Salinas sample: olibanum (due to the absence of cembrenes and boswellic acids, along with their O-acetyl derivatives); myrrh (due to the absence of curzerene and lindestrene); galbanum (due to the absence of β-pinene, fenchyl acetate, and guaiol); labdanum (due to the absence of dehydrocinammic acid, aromdendr-1-ene, and ledene); larch wood resin (due to the absence of larixol); and dammar resin (due to the absence of dammernolic acid). Additionally, sandarac, a resin from *Tetraclinus articulata* trees from the cypress family Cupressaceae (Colombini et al., 2000), is unlikely to be present as none of the key biomarkers were found. The chemicals that would be expected if sandarac was present include 12-acetoxysandaracopimaric acid, deemed by Mills and White (1977) to be almost unique to sandarac, and sandaracopimaric acid, considered characteristic for sandarac by Colombini et al. (2000). Communic acid has also been reported by Azémard et al. (2017) but exposure to light polymerises this compound over time (Mills and White, 1977; Azémard et al., 2017) and therefore this is not a suitable marker for archaeological samples unless pyrolysis GC-MS is employed for the analysis. Of course, the absence of these compounds is by no means conclusive in archaeological samples.

Another characteristic of embalming materials is the presence of fats or waxes of vegetable or animal origin, including beeswax (Buckley et al., 1999; Colombini et al., 2000; Łucejko et al., 2017; Vandenbeusch et al., 2021). Beeswax is characterised by the presence of long-chain fatty acids (Vandenbeusch et al., 2021), with the most abundant being lignoceric acid (C24:0) (Łucejko et al., 2017). A sample of modern commercial beeswax was analysed as part of this study and lignoceric acid was seen in abundance in the derivatised sample (along with other fatty acids up to C26:0 cerotic acid), however it was not detected in the derivatised Salinas sample, and neither were any fatty acids above C18:0 (stearic acid). Beeswax is characterised chemically by alkanes C25 to C33, maximising at C27 (Buckley and Evershed, 2001; Chasan et al., 2021) and while these were clearly identified in the modern beeswax sample, in the hexane extraction of the Salinas sample we tentatively identified the alkanes C25 (0.5 % peak area) and C27 (2 % peak area). Buckley and Evershed (2001) also state that beeswax can be characterised by the presence of wax esters (C40-C50), but these chemical species were not detected even in the modern beeswax due to the GC conditions used. If beeswax had been part of the balm mixture, then it is possible that a significant amount of hydrolysis had occurred, but we would expect to see palmitic acid (C16:0) and long chain alcohols from the wax esters (Łucejko et al., 2012; Chasan et al., 2021; Vandenbeusch et al., 2021). While palmitic acid was detected, the expected alcohols were not found in the Salinas sample. We can therefore say that while there is still a possibility that a small amount of beeswax was present in the Salinas sample, it was certainly not present in any abundance, which is somewhat unusual given its popularity as an embalming ingredient during the Graeco-Roman period (Buckley and Evershed, 2001).

The Salinas sample appeared to contain sizable amounts of palmitic (C16:0) and stearic (C18:0) acids which were detected in the ethanol extract, the hexane extract and the derivatised extract. In each case the peak area of the palmitic acid was significantly higher than that of the stearic acid which *may* indicate the source is vegetable rather than animal (Vandenbeusch et al., 2021). That being said, all fatty acids should be considered and quantification of these acids was not carried out. Additionally, while the hydrolysis of beeswax can produce palmitic acid, there was no evidence that beeswax was present in any abundance in the Salinas sample. Ricinoleic acid was not detected in our sample, so it is unlikely that castor oil is the source (Łucejko et al., 2017).

Pentadecanoic acid (C15:0) and heptadecanoic acid (C17:0) were found by Buckley et al. (1999) in samples of resin-like material from the mummified remains of Khnum-Nakht and Horemkenesi (from the 12th and 21st Dynasty, respectively) and in various samples from Egyptian mummification balms and organic residues from storage jars with a range of dates from the Old Kingdom through to the Copto-Byzantine period (Łucejko et al., 2017). They were also observed in the Salinas sample and, although they can be found in plant oils in low abundance, they are potentially indicative of the presence of animal fats or dairy products (Vandenbeusch et al., 2021). However, determination of the presence of branched-chain isomers of these fatty acids is needed to understand fat source used in the balm (Evershed et al., 1997). Notably, the short-chain fatty acids found by Vandenbeusch et al. (2021) which would be expected from the inclusion of dairy fats, were not detected in the Salinas sample.

Myristic acid (C14:0) was also found in the derivatised samples in the present study, although in smaller amounts. This fatty acid, together with larger amounts of palmitic and stearic acids, were found by Colombini et al. (2000) in samples of resin from the skull of Merneith (26th Dynasty) from Bakenrenef's tomb in Saqqara (Egypt). The same fatty acids were found in resin-like samples from the mummies of Khnum-Nakht and Horemkenesi; Buckley et al. (1999) considered the possibility that their origin was adipose tissue, particularly as they also found the steroidal compounds cholesterol and cholesta-3,5-dien-7-one in their samples. Nonetheless, these steroids were not found in the resin from the Salinas mummy.

A third, often discussed component of embalming resin is bitumen

(Buckley et al., 1999; Colombini et al., 2000; Jones et al., 2014; Łucejko et al., 2017; Vandenbeusch et al., 2021; Ferrant et al., 2022). If bitumen was present in the Salinas sample in abundance, the hexane extraction performed for the non-polar compounds should have detected the presence of hopanes and steranes (Colombini et al., 2000), which it did not. However, the absence of bitumen is not unusual as many balms did not actually include this material (Buckley and Evershed, 2001).

6. Conclusion

Examination of the ancient Egyptian mummified human head held by the Salinas Regional Archaeological Museum in Sicily has revealed that the remains are most likely those of a female aged between 20–50 years at the time of death. The trans-nasal craniotomy combined with the packing of the cranial cavity with abundant resin suggests a late date, possibly during the Ptolemaic or Roman Period. This dating is further supported by the presence of resin-soaked bandages with gilding (Dunand and Lichtenberg, 2006). In this investigation, the period of origin could not be confirmed by radiocarbon dating as the textile sample which was analysed produced a much older chronology (ca. 3000 BCE) than the mummified head.

The black colour and glossy, resin-like texture and solubility of material recovered from the mummified skull, appeared consistent with other archaeological examples (Łucejko et al., 2012; Vandenbeusch et al., 2021). Chemical analysis by various techniques indicated the presence of a natural resin, pitch, or tar, probably from the Pinaceae family of conifers. The detection of a variety of himachalene chemical species was particularly notable and indicates the use of a cedar oil or tar. The resinous material had clearly been mixed with other materials including a fat, oil, or wax but the nature of this material cannot be determined without further study. The inclusion of bitumen is unlikely given the results of the chemical analysis. Many of the chemicals identified in the resin collected from the Salinas mummy have also been noted in samples of embalming material from graves and mummies ranging from the Late Neolithic through to the time of the 26th Dynasty, and in materials recovered from storage jars with dates ranging from the Old Kingdom to the Copto-Byzantine period.

The anthropological and radiological assessment of the mummified head, the chemical analysis of the resin, and the presence of associated textile material support the conclusion that this is a genuine Egyptian mummy, which underwent an embalming process in order to be preserved. This research has shown that a multidisciplinary approach, using minimally invasive techniques, can help build a ‘paleobiography’ (White et al., 2023) of the deceased, revealing a picture of the mummified individual’s life and death as well as the story of their remains after death.

CRediT authorship contribution statement

Kirsty Squires: Writing – review & editing, Writing – original draft, Project administration, Methodology, Investigation, Formal analysis. **Alison Davidson:** Investigation, Formal analysis, Methodology, Writing – original draft, Writing – review & editing. **Simon Cooper:** Writing – original draft, Writing – review & editing, Formal analysis. **Mark Viner:** Formal analysis, Methodology, Writing – original draft, Writing – review & editing. **Wayne Hoban:** Formal analysis, Methodology. **Robert Loynes:** Writing – review & editing, Writing – original draft, Formal analysis. **Stephanie Zesch:** Writing – original draft, Formal analysis, Writing – review & editing. **Wilfried Rosendahl:** Formal analysis, Writing – original draft, Writing – review & editing. **Susanne Lindauer:** Formal analysis, Methodology, Writing – original draft, Writing – review & editing. **Caterina Greco:** Conceptualization. **Dario Piombino-Mascali:** Conceptualization, Formal analysis, Investigation, Methodology, Writing – original draft, Writing – review & editing.

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

Data will be made available on request.

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