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**Diploma thesis**

**Preliminary analytical study of two Egyptian  
mummies from Anthropological Museum of Athens**



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INSIDE EVERY  
MUMMY IS A  
STORY WAITING  
TO BE TOLD...



**The current thesis is dedicated to my best friend and colleague Loizou  
Jean and her shiny little daughter Martina...**

**Abstract:** The current thesis presents the preliminary analytical study of two Egyptian mummies from Anthropological Museum of Athens. The mummies were investigated with visual examination, photograph documentation and sampling procedure in order to estimate their preservation state. All the samples were examined macroscopically and stereoscopically. Samples of bandages, tissue, hairs, balms, filling material of plant, two beads and possible gold residues were analyzed; Optic microscopy, SEM/EDEX, ATR/FTIR, and portable XRF analytical techniques were used; Microbiological analysis also was conducted with the use of Optic microscope and fungus cultivation. The results are discussed making the analytical methodology used, capable for the preliminary study of the mummies. As conclusion the mummies are in bad preservation state and immediate conservation plan is required. Future examination aims are included too.



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## **1. Introduction**

All around the world mummified human bodies or partially human remains have been found sporadically (Cockburn, 1980; Lynnerup, 2007). Mummification processes are distinguished as anthropogenic (artificial), spontaneous (natural) or indeterminate (Aufderheide, 2003; Lynnerup, 2007). Treatment of the corpse after death has been performed in different periods and regions; the most popular burial practices worldwide included inhumation, cremation and mummification (Pearson, 1999).

Ancient Egyptian civilization was distinguished for mummification of the dead. It is characterized by a clearly defined belief in an afterlife existence, but this individual immortality was considered to be dependent as part of the preservation of the body in as lifelike a form as possible (David, 1990). Their religious concepts concerning the afterlife made it necessary to preserve the body as a place for the 'soul' to return to (Jansen et al., 2002). This belief came from observations that the dry sand of the desert preserved buried bodies. Such beliefs go back as early as the Neolithic and Predynastic periods of 5000-4000 B.C. An example of the importance of the preservation of the body is seen in the invocation from the ancient Egyptian mortuary texts referred to as the "Book of the Dead" (fig.1): 'My body is everlasting, it will not perish and it will not decay for ages' (Klys et al., 1999). Even though mummification was practiced in Egypt for nearly 3500 years, from the Old Kingdom, ca.2600 BC to the Christian Period, an end was put to this practice only after the Arab conquest of Egypt in the 7<sup>th</sup> century AD (Maurer et al., 2002).

Nowadays the evolution of technology allows the study of human remains with different ways; Human remains' analytical techniques provide important information concerning bioarchaeology and paleopathology. Demography, nutrition and health status of past populations can be investigated. They also provide data about social identity, social organization, ideology, rituals, religion and symbolism (Larsen, 1997; Pearson, 1999; Walker, 2000).

Mummified remains and associated artifacts are fragile and vulnerable to several types of deterioration, including those factors which result from environmental conditions, physical damage and damage caused by previous inadequate conservation and preservation attempts (David, 2001). It is therefore

essential to develop standardized examination and preservation methodologies in order to maintain their value as a source of biological and biomedical evidence.



Fig.1: Ancient Egyptian funerary text  
(<http://www.crystalinks.com/bookofthedead.html>)

## **2. THEORETICAL PART**

### **2.1 Ethics on protection and preservation of mummies**

The conservation of mummies is a politically and socially sensitive issue (Cassman & Odegaard, 2004). Countries such as United States and Australia have important legislation about the protection and preservation of human remains. NAGPA (Native American Graves Protection and Repatriation Act) in United States was performed on 16 November 1990 for treatment on human remains. In Australia, ATSIHPA (Aboriginal and Torres Strait Islander Heritage Protection Act) was enacted in 1984 providing protection and self-determination for indigenous. Both Australians and US Native Americans have great interest in terms of human remains. The fundamental difference is noted to the concept of private property. Australians law involves greater importance on the idea of ancestral claims than it does on property rights, since indigenous human remains found on private property must be given to indigenous authorities for proper treatment (Cassman & Odegaard, 2004).

In Arizona and Alaska states of USA, there are more strict laws than the federal mandate that covers finds on state and/or private property for information and description of on public lands. This means guidelines must be followed for informing, consulting, negotiating and repatriating, with relevant indigenous representatives to determine the ultimate disposition of human remains and their interim treatment (Cassman & Odegaard, 2004).

Other countries and fourth-world people have also understood the connection between politics and human remains. In Latin America, indigenous struggles have begun. For instance, Mapuche Indians are currently battling for indigenous rights in Argentina and Chile and have demanded the return of ancestral remains from museums (Endere, 1999). Peruvian indigenous cultural organizations such as Yachay Wasi are demanding that Inka mummies on display be removed because they are symbols of genocide and should not be used to attract visitors to a museum exhibition (Samuel and Borrero, 2004).

Furthermore, Egyptian antiquities has been illegal since 1983, the Egyptian government has recently requested greater protection of mummified human



remains (now considered important cultural patrimony) and they are not allowed to travel internationally (Cassman & Odegaard, 2004).

As well museums and institutions that include human remains and sensitive ethnographical material in their collection must follow particular standards. ICOM Code of Ethics (ICOM Code 2.5; 3.7; 4.3) covers care and display of human remains (ICOM, 2013).

Great importance has also been given to the concept about investigation and examination of mummified human remains. Mummies should be regarded as valuable information sources especially for addressing societies without historical records. The storage of undisturbed mummies in anthropological collections today will be the key for future development of the field, the advancement in technology will allow the future 'reading' of currently out-of-reach information stored in mummified human remains (Lombardi, 2001). Increasingly the ethics of studying mummies has been a subject of discussion and investigators should be aware of the current thoughts on this, and should take these into account when embarking on the analysis of a body, together with considering how much information is to be gained by such destructive analysis (Aufderheide 2003; Kaufmann and Rühli 2010; Machant 2010; Taylor and Antoine 2014; Fletcher et al., 2014;).

In General, during scientific studies destructive methods should be avoided. Advances in the application of noninvasive techniques to mummified remains have shed new light on past diseases. The virtual inspection of a corpse, which has almost completely replaced classical autopsy, has proven to be important especially when dealing with valuable museum specimens (Moissidou et al, 2015). Minimally-destructive autopsy methods (Alison and Gerszten 1983) could be complemented by sampling small amounts of mummified tissue (Lombardi, 1992). Physical curation of mummies should be dictated by the requirements of each individual mummy, and according to the microenvironmental conditions that originally contributed to the body's preservation in situ (usually dryness and/or coldness). Original soil composition should also be a factor in the design of curation plans. In any case, the main objective would be to minimize the natural process of decay (Lombardi, 2001). The direct application of chemicals or preservatives should be avoided, because

their presence could alter future molecular research or lead to unforeseen preservation problems. It is preferable to use several physical barriers (acid-free paper, bags, boxes, etc.). Mechanical or physical deterioration due to inappropriate storage, movement, handling, and environmental problems (humidity, rain, direct sunlight, etc.), must be avoided. Prevention plans must include contingencies in case of disasters (fire, flood, earthquakes, etc.). Ideally, mummy collections or artifact assemblages should be preserved with their contexts and documents referring to their original archaeological context. Such a database should be enriched after each study of the remains (Lombardi, 2001). The views of the deceased to being party to such an investigation cannot be established, but the least the investigator can do is to treat the body with all due respect (Ikram, 2015).

All above is likely the beginning of a growing trend towards greater respect for human remains in general, and giving attention to the increasing importance of the symbolic link human remains can have in political-religious freedom struggles, especially by minority indigenous populations in their demands for equality (Cassman & Odegaard, 2004).

## **2.2 Mummification types and materials**

According to the Greek historian Herodotus, three main types of mummification were available, and the client chose the method he could afford (David, 2001). The most important elements of mummification, which were crucial to arresting the decomposition of the body, were evisceration and dehydration of the tissues. Some authors (Smith and Dawson, 1924; Lauer and Iskender, 1955; Leek, 1969; Iskander and Shahin, 1973; Hamilton-Paterson and Andrews, 1978; Iskander, 1980; D'Auria, 1988; Taylor, 1955; Ikram and Dodson, 1998; Aufderheide, 2003; Salter-Pedersen, 2004; Taconis, 2005; Sivrev et al., 2005; Dunand and Lichterberg, 2006) have written on the ideal technique and most expensive method of mummification, which involve many stages (Maskoud and Amin, 2011).

In the second method, oil of cedar was injected into the anus, which was plugged to prevent the escape of the liquid, and the body was then treated with natron. Once this was complete, the anal plug was removed and the liquefied stomach and intestines were drained out with the oil (Hamilton-Paterson and

Andrews, 1978; Abdel-Maksoud, 2001). In the third and the cheapest method, the body was purged so that the intestines came away, and the body was then treated with natron (David, 2001).

It can be said that most authors have agreed on the description of three methods of mummification, but they differed among themselves in the description of the mummification materials. Some authors described the mummification materials depending on the description of old references and this may be due to the lack of analysis tools. All these references were in the first half of the twentieth century. Other authors described these materials depending on the analysis and investigation, most analysis has occurred from the end of the twentieth century to present day. Since the end of the twentieth century, archaeologists have long found themselves faced with the difficult problem of identifying unknown materials used with mummies. Most analyses were done on resinous materials. This may be due to: (1) most authors have analyzed wrapped mummies; (2) most resinous materials have been used on the bandages; (3) from a conservation point of view, it was difficult to remove bandages of mummies (to look for other mummification materials) because this will lead to the deterioration of mummies (Maskoud and Amin, 2011).

In recent years, Egyptian mummies have been the subject of a fairly large number of scientific studies (Mauer et al., 2002), but at the same time have always been a matter of controversy. The analyses were based on high performance liquid chromatography (HPLC) and gas chromatography-mass spectrometry (GC-MS). However, most of methods are directed to the identification of few of the substances that are simultaneously present in the sample, and thus only partial information can be obtained (Colombini et al., 2000). Gas chromatography-mass spectrometry (GC/MS) and other analysis studies allowed the elucidation of a great number of clearly separated compounds found in ancient embalming materials. Phenols, guaiacols, naphthalenes, monoterpenes, sesquiterpenoids, oxidized diterpene resin acids and triterpenoids were identified; through these intermediates, the materials used in mummification could be determined. These compounds also have antibacterial and antifungal effects and also prevent against deterioration caused by insects. Meanwhile analytical investigations have revealed a reasonably clear picture of the process of mummification types and to discuss the materials

used in the mummification processes (history, chemical composition, and their effectiveness in the preservation of the body) (Maskoud and Amin, 2011).

### **2.2.1 Types of mummification**

Naturally or artificially preserved bodies, in which desiccation (drying, dehydration) of the tissues has prevented putrefaction, have been discovered in Egypt (David, 2001). Most probably, the natural preservation of the body was noticed by the proto-dynastic people, perhaps when they were burying a new corpse in the sand near a previously buried one and it might have inspired them to believe that the body could be preserved and could more or less retain its human likeness (Iskander, 1980). True mummification (artificial methods) can be identified as a method, which incorporates several sophisticated techniques, making use of chemical and other agents. Many years of experimentation would be required to perfect such methods. The artificial preservation of the corpse was practiced in Egypt from the Old Kingdom to the Christian era (David, 1990). According to Herodotus account, there were a set of men who practiced the true mummification method and made it their business. When a body was brought to them, the embalmers showed the family of the deceased wooden models of corpses, so that they could choose the level of mummification they wanted. The mummification techniques were classified into three types. The first method which was the most expensive was typically reserved for royalty and very wealthy nobles. The second method was inferior to the first and did not include wrapping of the body and the third method was the cheapest of all (Iskander, 1980).

The first method of mummification can be summarized as follows:

- 1- The body was stripped of its garments, laid out on the embalming couch and purified. This was performed in a temporary structure close to the Nile or a canal (Taconis, 2005).
- 2- An incision was then made in the left side of the abdomen with a knife of obsidian or other kind of stone (Lauer and Iskender, 1955; Iskandar and Shahim, 1973). Once the embalmers inserted their hand through the incision and removed the liver, stomach and intestines, they cut the diaphragm and pulled out the lungs (Ikram and Dodson, 1988). The heart wasn't removed as the heart was believed to be weighed in the

afterlife to determine the goodness of the individual (Iskander, 1980). It is thought that the kidneys were also left in the body. The liver, stomach, intestines and lungs were washed and rinsed out with spices and palm wine (Sivrev et al., 2005). The spices were probably used as a deodorant (Hamilton-Paterson and Andrews, 1978), and a sterilizing material (Iskander and Shahin, 1973; Taconis, 2005). Each of these organs was then individually dried, wrapped in linen and placed in a canopic jar. Each jar held a different organ and in later periods, the jar lids were shaped to represent one of the four sons of Horus (Aufderheide, 2003).

- 3- The brain was not believed to have any importance, so it was cut into small pieces to facilitate removal and discarded (D'Auria, 1988). An examination of ancient Egyptian skulls in the Macalister Collection at Cambridge showed that 56 percent had a hole made in the base of the skull through the plate of the ethmoid bone. In 5 percent it had been made through the left nostril, and in 3 percent through the right one. In others the nasal septum had been wholly or partially removed, which resulted in significant perforation to the base of the skull (Leek, 1969). Brain removal was often incomplete, and modern studies provide evidence that some tissue was usually left behind) (Maskoud and Amin, 2011).
- 4- In order to completely dehydrate the body, the body cavities were packed with natron and then the entire corpse was covered in natron and left on a slanting embalming couch (Winlock, 1930). The body was placed in natron for seventy days, but some scholars argue that the body was only dried for forty days. The natron salt was first used in a liquid state but from the Middle Kingdom it was used as solid natron, which resulted in a shorter desiccation process (D'Auria, 1988). The natron salt not only caused the rapid desiccation of the body, thus avoiding the process of decomposition, but also entailed the saponification of the fatty tissues, assuring 'the chemical stability of the mummy' (Dunand and Lichterberg, 2006).
- 5- After the complete dehydration of the body, the thoracic and abdominal cavities were evacuated of their temporary stuffing materials showed that stuffing materials included dry natron powder contained in linen



packets, packs of linen impregnated with gum resin, straw and vegetable remains, and coarse powders containing quartz sand. Such packing would speed the dehydration of the body tissues, prevent the collapse of the abdominal wall, and combat the odor of putrefaction (Lauer and Iskander, 1955; Iskander and Shahin, 1973; Iskander, 1980; David, 1990).

- 6- After the temporary stuffing materials were removed from the body cavities, the body was washed and dried with alcoholic liquid (Iskander, 1980).
- 7- Then embalmers began to pack the body with permanent dry stuffing materials. The cranial cavity was filled with resin (Iskander, 1980; Salter-Pedersen, 2004). The body could be filled with several different materials, including crushed myrrh, cinnamon, frankincense (Ikram and Dodson, 1998), sawdust packets mixed with resin, cassia (Arya et al., 2001), and occasionally one or more onions (Iskander, 1980). Hot liquid resin was also poured over the body, which served to prevent the growth of bacteria and acted as a disinfectant and deodorant. The inside and outside of the body was prepared with all kinds of oils, aromatic resins, unguent and perfumes to prevent the re-entry of moisture and to strengthen the skin (Taconis, 2005). Layers of beeswax were used for covering the mouth, eyes and ears (Ikram and Dodson, 1998).
- 8- Often the embalmers painted the face and sometimes the whole body with ocher: red for men and yellow for women (Hamilton-Paterson and Andrews, 1978). The final touches of verisimilitude come with the elaborate cosmetic detailing of the face and fine coiffures. Hair was often dyed back to its natural color and made thicker with extensions woven onto existing strands (David and Aechbold, 2000). The abdominal incision was normally left open, but in some cases was covered with a plate of wax or copper alloy. Occasionally the incision was sewn shut (Maskoud and Amin, 2011).
- 9- Amulets were essential in transcending to the afterlife. During all steps of the mummification process, amulets were placed in specific locations including around the neck, waist, and limbs, as well as between the layers of the wrappings (Andrews, 1894).

10- Bandaging the mummy: the fingers and toes were individually wrapped, then layer after layer of linen was wound around the limbs and the torso (David and Archbold, 2000). In many cases, a thick ‘carapace’ of resinous paste and linen was applied over the whole body, separating the outer and inner layers of wrapping. After wrapping was a complete, red linen shroud was draped over the body (Taylor, 1995).

**The second method** of mummification was a less expensive method of mummification according to Herodotus. It did not include complete evisceration. In the second mummification method (which was also used in animal mummification) oil of cedar was injected into anus, which was plugged to prevent the escape of the liquid, and the body was then treated with natron. Once this was complete the oil was drained off and the intestines and the stomach come away with the oil; the flesh had also been desiccated, so only the skin and the skeleton remained (D’Auria, 1988; David, 2001; Taconis, 2005).

**In the third method** of mummification, the stomach and internal organs were removed through an abdominal incision on the left side of the body. The body cavities were sterilized by ethyl alcohol. The whole body was then buried in natron salt (Iskender, 1980; David, 2001).

### **2.2.2 Mummification materials used**

The authors divided the materials used in the mummification processes into two groups:

- Materials used based on analyses and investigations. Most of these materials were resinous materials and oils;
- Materials used based on the descriptions of some authors and literatures (Maskoud and Amin, 2011).

### 2.2.3 Materials used based on analyses

#### 2.2.3.1 Natron salt

The first use of the term “natron” appears to be in the glazing of quartz and steatite. The word (in Egyptian hieroglyphics “ntry”) is most likely derived from the root “ntr” indicating its association with religious and funerary rituals (Maskoud and Amin, 2011).

Natron is a white, crystalline, hygroscopic, and natural material mined at Wadi Natrum in the Nile Delta, and it was an essential component in the mummification process in ancient Egypt. Natron was considered one of the most important materials. It was used in the removal of water from the tissues, prior to their treatment with resins, and prevented or retarded the biological deterioration and putrefaction of the body through fungal and bacterial attack (Edwards et al., 2007).

Some analysis investigation had been performed on natron for different purposes. The following analysis had been done:

- Lucas (1914) had performed chemical analysis on natron taken from different mummies to know the main components of natron used especially in the mummification processes; he stated that sodium compounds (chloride, carbonate, bicarbonate and sulphate) were identified (Maskoud and Amin, 2011).
- Sandison (1963) stated by chemical analysis that the essential process in artificial mummification is dehydration of the body, and for this common salt would have proved successful. The choice of natron may have depended on its superior ability to break down body fat. He also stated that the composition of natron varies widely since it is naturally occurring mixture of salts, but it contains sodium carbonate, sodium chloride and sodium sulphate in varying proportions. Sandison (1963) went on to say that this was true of modern natron from Wadi Natrum and El Kab, as well as ancient natron from tombs of the Eleventh and Eighteenth dynasties (Maskoud and Amin, 2011).

- Iskander and Shaheen (1973) performed quantitative chemical analysis on the tree samples taken from pottery jars (one sample taken from Tura El-Asmant excavation 1960, and the two other samples were taken from the Qurna excavation 1960). They identified the chemical composition of natron as the same compounds that were mentioned by Sandison (1963); Ikram and Dodson (1998) mentioned that the chemical composition (as mentioned above) of the various natron samples varies widely (Maskoud and Amin, 2011);
- Abdel-Maskoud (2001) used artificial sodium components (6 parts of sodium chloride, 1 part sodium sulphate and 1 part sodium bicarbonate) in the experimental studies on mummification techniques used in ancient Egypt. He conducted an experimental study on the mummification process applied on rats, and noted that the natron components used were highly effective in the desiccation of the rat's bodies (Maskoud and Amin, 2011).
- Edwards et al (2007) analyzed eight samples by the Raman spectroscopic analyses. The analysis clearly indicates that the natron specimens are of indefinite composition, comprising sodium sulfate and sodium bicarbonate and gypsum. Halite, sodium chloride, does not have a first-order Raman spectrum (Maskoud and Amin, 2011).
- Cosmacini and Piacentini (2008) mentioned that the mixture of natural sodium carbonate and bicarbonate known as natron, were used as desiccating agents in the sophisticated methods of perfect artificial mummification in ancient Egypt (Maskoud and Amin, 2011).

Desiccation with natron has been identified as the seventh stage in a thirteen-stage process used during mummification. During the dehydration process, the body, probably on a slanting bed, was completely covered with natron. This had the effect of removing any remaining body liquid and consequently ensuring against any further putrefaction (Peck, 1980). Some authors argue that the drying-out process lasted 40 days (Peck, 1980; Cockburn et al., 1980) and some authors argue that it lasted 70 days (Sandison, 1963; Hamilton-Paterson &

Adrews, 1978). Abdel-Maskoud (2001) mentioned that Sadison (1968) proved that mummification process did not take forty days, as was mentioned by Herodotus. Complete dehydration of the body could have taken twenty-eight days or less, as this process depended on one or more of the following factors:

- a. **The condition of the body**, which has already begun to exhibit signs of putrefaction may take a long time to achieve a suitable dry condition.
- b. **The composition of the salt mixture** which makes up the natron. A high quality mixture of salt, especially sodium carbonate and bicarbonate, would produce the best quality results.
- c. **The re-use of natron salt for more than one body**. If natron is used for more than one body, the result after the first use will deteriorate.
- d. **The ratio of natron volume to body volume**. Under ideal conditions, the volume of natron used should be at least ten times greater than the body volume.
- e. **The duration of the natron treatment**. It was found that the ideal treatment depends on the climatic conditions (Maskoud and Amin, 2011).

Many authors and scientists varied in their dealing with the use of natron in the mummification process. Lucas (1914) said that natron was used by the ancient Egyptians in connection with their dead, certainly in a solid state, and possibly also as a solution. Abdel-Maksoud (2001) wrote that Smith and Dawson (1924) and David (1978) confirmed that the Egyptians used natron in the solid form and supported their opinions with the following reasons:

It was thought by some scholars that during the mummification process, a solution of liquid natron was applied to the body. However, this is based on an incorrect translation as dry natron (Cockburn et al., 1980).

#### 2.2.3.2 Coniferous resin

The widespread use of plants oils indicates that the embalmers were aware of the special properties of unsaturated oils that allow them to 'dry' or rather to



polymerize spontaneously. This polymerization would have produced a highly crosslinked aliphatic network, which would have stabilized otherwise fragile tissues and/or textile wrappings against degradation by producing a physico-chemical barrier that impedes the activities of microorganisms (Buckley and Evershed, 2001; Davies, 2011).

According to arguments presented by some scientists, the resin used on the human body at the end of the mummification process, was derived from coniferous trees, specifically the cedar, juniper, and pine trees (Klys et al., 1999). It should be noted that coniferous materials had been used in Egyptian mummies, but there had been confusion regarding the actual trees materials used. Lucas (1931) Herodotus, Diodorus and Pliny mentioned in their writings that the resin employed by the Egyptians in mummification was certainly from conifer, although probably never from the true cedar but from the juniper tree instead. The materials mentioned by Pliny as used for mummification were *cedri succus*, the natural resinous product of coniferous tree, probably juniper and cedrium, which contained pyroligneous acid that was composed of a mix of oil of turpentine and wood tar. Bauman (1960) mentioned that throughout ancient times, junipers were confused with cedars. Iskander (1980) mentioned that in the second method of mummification, oil of cedar was injected into the body through the anus, which was afterwards stopped up to prevent the liquid from escaping. Amoros and Vozenin-Serra (1988) mentioned that coniferous material (in the form of sawdust) came from the cedar tree and was found in mummies dating to different periods (Eleventh, and twenty-first Dynasties and the Greco-Roman period). Taconis (2005) noted that in the First Intermediate Period, evisceration was practiced, either by incision of the abdominal wall or by means of an enema of cedar (or more probably juniper) oil (Maskoud and Amin, 2011).

Some analytical techniques have greatly increases the accuracy of the identification of ancient natural materials such as oils or resin. The following analytical techniques have identified the resin material used on mummies as coming from a conifer:

- Proefke and Rinehart (1992) used fast atom bombardment combined with mass spectrometry (FAB/MS), high resolution FAB/MS, FAB

tandem mass spectrometry (MS/MS), and gas chromatography/mass spectrometry (GC/MS), to determine the composition of the resinous material recovered from the wrappings of an Egyptian Greco-Roman mummy dating to approximately 350 A.D. (Maskoud and Amin, 2011).

The tree oxidation products of abietic acid found in the mummy resin clearly indicate that a true conifer resin was used as the base for embalming fluid.

- Wisseman (1992) stated that chemical analysis of the embalming fluids of an Egyptian mummy at the World Heritage Museum, University of Illinois, indicated that coniferous resin was used.
- Amoros and Vozenin-Serra (1998) mentioned that sawdust taken from a Late Period mummy was analyzed by traditional wood anatomy methods and investigated under optical microscope. The microscope investigation revealed that the sawdust packing was comprised of 70% coniferous wood, 10% tamarix species and 20% unidentified vegetal remains, possibly gramineae stems and leaf parts (Maskoud and Amin, 2011).

The radial walls of the larger vertical tracheids of dry untreated sawdust were investigated under high magnification of the optical microscope. In these walls, highly characteristics bordered pits with fringed torus margins were observed, and assigned these woody elements to the *Cedrus* genus (Pinaceae family, Abitoideae sub-family) (Maskoud and Amin, 2011).

Identification of the *Cedrus* remains at species level is difficult in view of the anatomical similarities between *Cedrus atlantica* and *Cedrus libani*. When compared to other samples, it seems that the sample is closer to *Cedrus libani* (Maskoud and Amin, 2011).

- Connan et al. (1999) stated that the analysis of balms from Egyptian mummies (1000 BC to 400 AD) by GC/MS analysis and GC/C-IRMS reveals complex molecular mixtures which are diagnostic of products such as conifer resin and beeswax mixed bitumen to prepare each balm.
- Connan (1999) stated that one of the major conclusions of his study, restricted to mummies dated between 1000BC and 400AD, is that the

molecular signatures are extremely diverse from one sample to another and that conifer resin is the key ingredient from which most balms were prepared.

- Klys et al., (1999) mentioned that different ancient resin specimens have recently been analyzed by infrared spectrometry and gas chromatography/ mass spectrometry (GC/MS). It was supposed that the resins used in ancient Egypt were from coniferous trees (cedar, juniper, firs and pines). He also stated that the analysis of the resin samples was performed by means of physicochemical tests, infrared spectroscopy and spectrographic method. The tests were performed on many fragments from different parts of the mummy of Iset-Iri-Hetes belonging to the collection of the Archaeological Museum in Krakow, Poland. A pine resin was identified by the analysis mentioned above.
- Buckley and Evershed (2001) made chemical investigation to a collection of Egyptian mummies dating from the mid-dynastic period (c. 1.900 yr BC) to the late Roman period (AD 395). This study revealed that coniferous resin clearly increase in its prominence through time, and is found in material taken both directly from the bodies and from the wrappings. Coniferous resin is identified by the presence of both functionalized and defunctionalized diterpenoid components. For example, 7-oxodehydroabietic acid and 15-hydroxy-7-oxodehydroabietic acid were usually the dominant diterpenoid components, and the normally abundant dehydroabietic acid was virtually absent. Buckley and Evershed (2001) also stated that, although coniferous resin were clearly used in the embalming process at least as early as 2,200 yr BC (VI dynasty), their use becomes ost apparent in later periods; both the tissues and the wrappings of mummies from the Roman period (30 yr BC to AD395) contain appreciable quantities (up to 37%) of coniferous diterpenoids. The increasing use of coniferous resin suggests that the embalmers resin suggests that the embalmers may have become aware of the ability of specific natural products to inhibit microbial defradation by means of mechanisms (physic-chemical barriers and antimicrobial action) analogous to their protective roles in the plants from which they derived.

- Maurer et al., (2002) have used gas chromatography and gas chromatography-mass spectrometry (GC-MS) to analyze four samples taken from four Roman mummies (4<sup>th</sup> century AD), which were discovered in the Dakhleh Oasis excavation in the Western Desert of Egypt. The analyses proved that the soluble material on the mummies mainly consisted of plant material. The use of resins of coniferous trees is indicated by the presence of abietic acid related tricyclic diterpenoid acids.
- Koller et al., (2003) stated that mummy 'Saankh-kare', Eighteenth Dynasty, 1500 B.C. was analysed by gas chromatography. The analysis proved the presence of coniferous resin according to the recovery of guaiacols without syringols and methoxyphenol derivatives, which are formed when soft coniferous wood is heated. They also stated that the brown solid resinous material found near the mummy 'Saankh-kare' also contained sequiterpenoid components normally detected in organic solvent extracted wood from *cedar atlantica* called cedar oil which is composed of junipene, cadalene, candinatriene,  $\alpha$ -curcumene, cuparene etc. Koller et al., (2003) stated that the analysis on bone fragments, now deposited in the Naturhistorisches Museum at Viena, and the mummy of Idu II from the Old Kingdom, suggested that diterpenoid resin acids found on the fragments were evidence of the use of pine resin.
- Charrie-Duhaut et al., (2007) analyzed a sample from a canopic jar in the Louvre with gas chromatography-mass spectrometry and liquid chromatography-mass spectrometry (GC-MS and LC-MS) and absolute dating by Carbon-14. The results revealed that the sample, scraped from the interior face of the jar, was identified as an unguent made of coniferous oil and animal fat, dating from the Third Intermediate Period.
- Davies (2011) mentioned that many sources suggest that cedar was used for mummification and ancient texts by the Roman scholar Pliny and Greek historian Herodotus link a wood called 'cedrium' to embalming. Cedar materials were found in unused ancient Egyptian complex of mortuary temples and tombs dating from about 1500 BC. GC-MS analysis revealed the presence of phenols and sequiterpenoids and, importantly, a compound found in wood smoke called guaiacol. The guaiacol was attributed to tar oil produced from cedar wood, known to

be rich in the compound. The embalming material would have had 'powerful' bactericidal and fungicidal activity (Maskoud and Amin, 2011).

Though the previous studies and analytical methods performed, the coniferous oils can be divided into the following:

- I. Pine oil: pine oil comes from the pine tree (Family: Pinaceae, *Pinus* sp.) which reaches a height of over 40m, has evergreen leaves (needles), a straight trunk with a rather spreadind, irregular crown and bark that is scaly and cracked. The leaves set in pairs on the branchlets, are aromatic, needle-like, sheathed and glausous. Medicinally useful parts are gathered in summer (Chiej, 1988).
- II. Cedar wood oil : the famous cedar of Lebanon is a true cedar. For thousands of years its 70-100 foot height and 16-25 foot girth have inspired men with thoughts of strength and solidarity and the trees have always been regarded with what Franklin Lamb calls "sacred awe". Lebanese cedar usually grows in association with pines and firs. The forests were extensive in Biblical times, but only five small groves exist today-about 6000 feet up Mount Lebanon. These are under the care of a Christian sect called the Maronites. The wood is fragrant, insect-repellent, quite durable and rot-resistant. It was highly esteemed by the Egyptians for many kinds of wood-work and very much so for coffin-making. It was first used in coffins sometime around tenth dynasty an-id persisted well into the Ptolemaic period (Baumann, 1960). Sawdust made from this cedar was also used as a body packing for mummies.
- III. Juniper (fig. 2A): A shrub or small tree (Family: Cupressaceae), about 10 feet (3m) in height. The bluish-green leaves are narrow, leathery and very pungent; there is a pale, concave line running the whole length of the upper surface of each leaf. Male and female flowers are born on separate trees. The fruit is a false berry formed by the bracts surrounding the flower. The medicinally useful parts are gathered in the summer and autumn (Chiej, 1988).

Juniper cones (generally being *Juniperus phoenicea* L.) have often been found in ancient Egyptian graves. An account of Coptic burials at the Monastery of Epiphanius in Thebes relates that handfuls of juniper cones and coarse rock salt were put between the legs and over the body of the deceased. Cones of *J. phoenicea* were plentiful enough to be utilized in large amounts as an embalming substance in the burials (Brusell, 2004).



Fig. 2: Mummification materials used: (A) Juniper, (B) Mastic, (C) Myrrh, (D) Cassia (Maskoud and Amin, 2011).

#### 2.2.3.3 Mastic

Mastic is a light yellowish (fig.2B), semi-transparent, natural resin that is exuded from cuttings made on the trunk of mastic trees (*Pistacia lentiscus* var. *Chia*) every year, from July to October. These trees belong to the Anacardiaceae family, which is traditionally cultivated in the south of Chios, a Greek island in the Aegean Sea (Maskoud and Amin, 2011).

This material remains under the trees for many days and coagulates through the local environmental conditions. The coagulated product is then collected and is called mastic gum. Mastic oil is produced by the steam distillation of mastic (Mills and White, 1989; Paraskevopoulou 2009).

The Egyptians used mastic in embalming and in religious capacities. It probably had religious significance even when used for embalming (Baumann, 1960). There are several studies proving the presence of mastic resin in mummification materials. The following analyses have been done:

- Colombini et al., (2000) used gas chromatography-mass spectrometry to analyze Merneith's mummy balms (plant resin, oils and their degradation products). He compared Merneith's unguents with several natural pure substances, which were collected and used as reference materials. He proved that the main components found on the balm of Merneith's mummy was mastic resin.
- Buckley and Evershead (2001) indicated that the former include the isomasticdienonic, masticdienonic, moronic and oleanonic acids that are diagnostic of the presence of *Pistacia* resin and are found in a female mummy of the Third Intermediate Period (XXI to XXV dynasty; 1,069-664 yr BC) (Maskoud and Amin, 2011).

The analyses that were carried out on one of the blue-glazed faience jars with the cartouche of Ramesses II by Charrie-Duhaut et al., (2007), which entered the Louvre in 1905, proved that the substance stored in the jar was likely an embalming substance, made of pure vegetable resin (*Pistacia*) and dating from the Ptolemaic Period.

- Stern et al. (2003) studied resin taken from some bowls found at Amarna. He suggested that on the basis of the molecular composition the resin is from a species of *Pistacia*. He mentioned that there is also some evidence of the use of pistacia resin during mummification, at least as early as the Third Intermediate Period.

#### 2.2.3.4 Myrrh

Myrrh is an oleo-gum resin (fig. 2C), freely discharged from natural fissures or from incisions made to collect this product. It flows as a pale yellow, bitter, odorous gum, which hardens as it dries to yield a reddish-brown or orange irregular mass. In commerce, myrrh is found as a powder or as granular pieces of many. Myrrh resinous exudates are obtained from trees of certain *Commiphora* species of Burseraceae family. Myrrh oils are occasionally used as flavouring agents. Myrrh contains the resin myrrhin (23-40%), the volatile oil myrrhol (2-8%), gum (40-60%) and a bitter unidentified component (Hamm et al., 2003).

Myrrh, *antiyw* in ancient Egyptian, was fragrant gum, essentially resin in the shape of small yellow-red lumps. Myrrh was imported from Somalia and southern Arabia, and it was commonly used during the New Kingdom to stuff and massage on and into mummies. It was mostly valued for the fragrance that it imparted to the corpse. There are some studies that characterize the presence of myrrh in Egyptian mummies. The following analysis had been done:

Hamm et al., (2003) used Headspace SPME coupled with GC-MS to identify the mono-sesqui-, and diterpenes of myrrh, provided that diterpenes like incensole or isoincensole and their oxide or acetate derivatives are characteristic biomarkers of myrrh (Maskoud and Amin, 2011).

#### 2.2.3.5 Beeswax

Beeswax is a type of wax from the honeycomb of the honeybees. It is yellow, brown, or white bleached solid. The normal color of beeswax varies from shades of yellow, orange, red and brown. The color of beeswax changes with age, for example virgin wax is white but darkens rapidly as it ages, often becoming almost black. It has a faint honey odor. It consists largely of myricyl palmitate, cerotic acid and esters, and some high-carbon paraffins. Beeswax is lipid by nature. It has saturated hydrocarbons, acids or hydroxyl-acids, alcohols, pigments, mostly from pollen and propolis, as well as minute traces of brood (Hossain et al., 2009)



Beeswax was utilized for a number of purposes in ancient Egypt. For example, it was often used to model figures and was also employed in the mummification process. Lucas (1962) cited a few examples of Middle Kingdom and New Kingdom date where wax had been used as an adhesive. Connan et al. (1999) stated that the analysis of balms from Egyptian mummies (1000 BC to 400 AD) by GC/MS analysis and GC/C-IRMS revealed complex molecular mixtures which were diagnostic of products such as conifer resin and beeswax mixed with bitumen to prepare each balm. Serpico and White (2000) reported that the use of beeswax in mummy wrapping dating from the Late Period to the Roman Period had been established. GC and Py-MS. Buckley and Evershed (2001) said that beeswax is characterized chemically by alkanes (C25-C33), wax esters (C40-C50) and hydroxyl was esters (C42-C54). They also stated that the first appears of beeswax notably later than coniferous resin, with its positive identification in a resinous coating taken from the chest cavity of a female mummy of the Third Intermediate Period (XXI to XXV dynasty; 1,069-664 yr BC). In a sample taken from 'Pedeamun', a XXVI dynasty (664-525 yr BC) mummy. Goffer (2007) said that the ancient Egyptians used beeswax over 6000 years ago to preserve mummies, by soaking the linen strips used for wrapping in beeswax. They also coated and sealed the coffins with wax (Maskoud and Amin, 2011).

#### 2.2.3.6 Bitumen

Bitumen may be either (a) a mixture of hydrocarbons originating in petroleum found naturally impregnating certain porous rocks, generally limestone, but occasionally sandstone, in various parts of the world or (b) a similar material mixed with varying, proportions of mineral matter found in the form of deposits, as in the well-known "pitch". Pitch may be either natural or artificial. Natural pitch is simply bitumen, which has become solid by exposure, and is found in the neighborhood of the Dead Sea (Lucas, 1914).

The bitumen used as a preservative in some ancient Egyptian mummies was previously thought to come only from the Dead Sea area in Palestine. A closer source of bitumen was investigated at Gebel Zeit on the southwestern shore of Egypt's Gulf of Suez (Harrell, 2002).

The origin of the black color of mummies has always been a subject of debate (Connan, 1999). Lucas (1914) and Hammond (1959) studied the writings of the Egyptian, Arab, Greek and Latin authors who studied mummies and mummification, and concluded that either bitumen or pitch, or both, were extensively employed by the ancient Egyptians in the preservation of the dead. Lucas (1914) however could not find any careful or systematic examination or analysis of the material, and apparently nothing of the sort has been done. It seems then that the recognition of the use of bitumen or pitch, was solely based on the appearance of the material on the mummy, and in a few cases, the behavior of mummified material when burned (Maskoud and Amin, 2011).

Chemical studies were undertaken to find a solution to this controversial problem:

- Connan (1999) stated that gas chromatography, GC/MS and GC/C-IRMS analysis on 20 balms from Egyptian mummies, mainly from the Valley of the Queens and not older than 1000 BC, showed that bitumen found in balms. This was a result of identifying sterane and terpane patterns and comparing them with bitumen found in the Dead Sea region.
- Harrell (2002) stated that five mummies were analyzed using molecular biomarkers derived from gas chromatography/mass spectrometry. It was found that four of the mummies contained Dead Sea bitumen, and the fifth and oldest one, that of the Libyan Pasehor from about 900 BC, had bitumen from Gebel Zeit. This is the first evidence for the use of an indigenous source of bitumen in ancient Egypt.
- Aufderheide (2003) mentioned that chromatographic techniques proved that bitumen from the Dead Sea, instead of the usual resins, was employed by some late Egyptian embalmers.
- Koller et al., (2003) stated that bone fragments, now deposited in the Naturhistorisches Museum at Vienna, Department of Anthropology, were analyzed by GC and GC/MS. There is strong evidence that a bituminous material was applied on the surface of the mummy and parts of it have migrated into the bones. This is because of the detection of pristane and phytane together with the alkanes (Maskoud and Amin, 2011).

## 2.2.4 Materials used based on the description of some authors and literatures

Some references mentioned that the following mummification materials were used for different purposes in the mummification processes. Unfortunately, scientific analysis and investigations have not been done for these materials. The authors explained these materials depending on the descriptions and explanations of archaeologists and scientists (Maskoud and Amin, 2011). These materials are:

### 2.2.4.1 Cassia

*Cinnamomum cassia* (Nees) Nees ex Blunmie and *Cinnawomum zeyla: nicumn* Breyn. These are the sources of cassia and cinnamon respectively. Various parts such as the fruits, oil, inner bark and leafy twigs of cinnamon are used. The inner bark is a pungent, sweet and spicy herb (Baumann, 1960; Ates & Erdogrul, 2003).

During ancient times, cassia (Fig.2D) and cinnamon were both referred to, although to what extent their botanical terminology aligns with that of modern times is unknown. Cassia was mentioned in Chinese herbal texts as early as 4,000 years ago (Hernandez, <http://www.cinamoncassia.com/>).

There is considerable doubt as to whether cinnamon and cassia were used in ancient Egypt (Baumann, 1960). Ancient records pointing to the use of cinnamon and spices date from the Old Kingdom, around 2,600 BC (Harnandez, <http://www.cinnamoncassia.com/>). Baumann (1960) mentioned that in the Karnak Reliefs of the Nineteenth Dynasty, it is written: “I gather together all the countries of Punt, all their tribute, of gum of myrrh, cinnamon...” And in the Harris Papyrus from the Twentieth Dynasty, cinnamon is mentioned four times and cassia once in the lists of tributes (Maskoud and Amin, 2011).

Because of its natural preservative properties and potent scent, Cassia and cinnamon were a part of ancient embalming practices, most notably in Egypt. The art of embalming was often partly medical and partly spiritual practice, and cinnamon played an important role in both spheres. Its chemical properties

make it a practical ingredient in embalming, but its distinctive scent, high price, and vibrant color served symbolic purposes as well (Ikram and Dodson, 1988).

Pettigrew (1834) mentioned that on the surface of Twentieth Dynasty mummy was "... a thick layer of spicery ...(which)... still retains the faint of smell of cinnamon or cassia" (Maskoud and Amin, 2011).

#### 2.2.4.2 Onions

Onion, (*Allium cepa L.*) is one of the most important and oldest vegetable crops grown in Egypt. Although it is primarily grown for food, it is also used as traditional medicine (Hussein et al., 2007).

Onions were found in body cavities from the New Kingdom until the Third Intermediate Period. David and Archbold (2000) mentioned that Ramesses IV had onions placed in his eye sockets and his ears, and a piece of onion skin covered with resin was inserted into each nostril. Sandison (1957) mentioned that this is probably the only instance of the use of onions to simulate the eye (Maskoud and Amin, 2011).

#### 2.2.4.3 Lichen

Lichen often inconspicuous, hardy and adaptive plants are composed of a fungus and a green or blue-green alga. This union or symbiosis produces a long-lived organism that does not look like either the fungal or algal partners. Both partners contribute to the growth of lichen (Maskoud and Amin, 2011).

The alga uses photosynthesis, like other plants, to produce food, while the fungus supplies water and essential minerals producing a structure that protects the alga from extreme environmental conditions. Together they thrive in some of the harshest environments on earth where few other plants and neither partner alone can survive (Hagan, 2004).

Lichen was used to fill out body cavities (Ikram and Donson, 1988), such as the lichen used in the cavities of Ramesses IV (Hamilton-Paterson & Andrews, 1978), and also lichens were inserted under the skin to try to give the body a more 'fleshy' appearance (Knight, 2009).

#### 2.2.4.4 Henna

The henna plant *Lawsonia inermis* Linn is fragrant shrub native to Asia and northern Africa. The species is sometimes referred to as *L.alba* or *L.rubra* and is cultivated in India, the Middle East, Egypt and tropical America (Avijit, 2002). The henna plant is a glabrous, many-branched shrub or quite a small tree with grayish-brown bark. Leaves are opposite, sub sessile, elliptic or broadly lanceolate, entire, acute or obtuse, 2-3cm long and 1-2cm wide. Flowers are numerous, small, white or rose colored and fragrant (Muhammad & Muhammad, 2005). Henna dye is prepared by grinding the fresh leaves of this plant or by powdering the dried leaves and then mixing into a grayish-green paste with water. The resulting brown dye is extensively used as decorative skin paint, for nail coloring and as hair dye and conditioner (Cordeiro et al., 2008). A variety of analyses on Ramesses II's mummy showed that the embalmers dyed the pharaoh's hair, probably with henna (Brier, 1994).

#### 2.2.4.5 Gum Arabic

Gum Arabic is naturally occurring exudates collected from *Acacia Senegal* trees and, to a lesser extent, from *Acacia seyal* trees. It is one of the oldest and most important industrial gums (Yadav et al., 2007). It is a high molecular weight macromolecule that can easily be dissolved and dispersed in water under appropriate conditions. About 70% of the world production of gum Arabic is in Sudan, the rest is in the French-speaking countries of West Africa. Gum Arabic is used in the production of food, pharmaceuticals and cosmetics; it has also seen some use in medicine (Zaied et al., 2007). Gum Arabic is a branched-chain, complex polysaccharide, either neutral or slightly acidic, found as a mixed calcium, magnesium and potassium salt of a polysaccharidic acid (Arabic acid). The backbone is composed of 1,3-linked b-D-galactopyranosyl units (Maskoud and Amin, 2011).

The side chains are composed of two to five 1,3-linked b-D-galactopyranosyl units, joined to the main chain by 1,6-linkages. Both the main and the side chains contain units of a-L-arabinofuranosyl, a-L-rhamnopyranosyl, b-D-glucuronopyranosyl and 4-O-methyl-b-D-glucuronopyranosyl, the last two mostly as end units (Buffo et al., 2001; Ali et al., 2009). Gum Arabic was comprised of 39-42% galactose, 24-27% arabinose, 12-16% rhamnose, 15-16%

glucuronic acid, 1.5-2.6% protein, 0.22-0.39% nitrogen, and 12.5-16.0% moisture (Islam et al., 1997; Zaied et al., 2007). The chemical composition of Gum Arabic can vary with its source, the age of the trees from which it was obtained, climatic conditions and soil environment (Ballal et al., 2005). The Ancient Egyptians used gum Arabic as an adhesive when wrapping mummies (Yadav et al., 2007).

According to Maskoud and Amin (2011), through previous studies, it is apparent that bodies mummified by the third method were more damaged by bacteria, fungi and insects than others mummified by the second and royal methods. The bodies mummified by the most expensive, royal method were the most protected. This can be explained by the use of plant materials in the second and royal methods.

- **Cedar oil** was used in the second method. It was used in the second method. It was injected into the body, and was also used in the ideal method to treat the body cavities after washing by palm wine. It contains essential oil and some essential ingredients ( $\alpha$ -pinene, myrcene, limonene, terpinolene and  $\alpha$ -terpinene), which have a major effect against bacteria, fungi and some insects. This may explain the reason why the mummies were protected (Maskoud and Amin, 2011).
- **Pine oil** was used in the Third Intermediate and Roman periods. Some ingredients of the essential oil ( $\beta$ -thujene,  $\alpha$ -pinene,  $\beta$ -pinene and bornyl acetate) have antibacterial effects against gram-positive and gram-negative, in addition to antifungal effects (Maskoud and Amin, 2011).
- **Juniper** was used in the First Intermediate Period and has the same effects as that conifer oil (Maskoud and Amin, 2011).
- **Mastic** was used in the New Kingdom, Third Intermediate Period and Ptolemaic Period. Some essential oil ingredients (verbenone,  $\alpha$ -terpineol, linalool and pentacyclic triterpenes) have antiseptic and antimicrobial effects (Maskoud and Amin, 2011).
- **Myrrh** was used in the New Kingdom. Some of its essential oil ingredients ( $\alpha$ -pinene, sesquiterpene hydrocarbons,  $\delta$ -elemene and  $\beta$ -bourbonene, furanosesquiterpenes and germacrene-type compounds) are used to kill and repel pests and these compounds are effective arthropod repellents (Maskoud and Amin, 2011).

- **Cassia** was used around 2,600 BC. Some of the essential oil compounds (cinnamaldehyde, linalool, eugenol and 1,8 cineol) have antimicrobial, antiseptic and antifungal effects in addition to a major effect against insects (Maskoud and Amin, 2011).
- **Onions** were used from the New Kingdom until the Third Intermediate Period. Some of the onion compounds (alliin,  $\gamma$ -glutamylcysteins, steroid, saponins and sapogenins) have antimicrobial effects and have a significant effect against UV light (Maskoud and Amin, 2011).
- **Lichen** was used in the New Kingdom. Some of its compounds (usnic acid, spaerophorin, pannarin and paraconic acid) have antibacterial and antifungal effects. Usnic acid enantiomers caused significant antifeedant activity and toxicity towards larva (Maskoud and Amin, 2011).
- **Henna** was used in the new Kingdom. Some of its compounds (lawsone, 2-hydroxy-1,4naphthaquinone) have antibacterial effects (Maskoud and Amin, 2011).

## 2.3 General human anatomy

### 2.3.1 The Locomotor system

**Skeleton:** The skeleton consists of bones and cartilages. A bone is composed of several tissues, predominantly a specialized connective tissue that is, itself, called “bone”. Bones provide a framework of levers, they protect organs such as the brain, the heart, their marrow forms certain blood, cells and they store and exchange calcium and phosphate ions. Cartilage is a tough, resilient connective tissue composed of cells and fibers embedded in a firm, gel-like, intercellular matrix. Cartilage is an integral part of many bones, and some skeletal elements are entirely cartilaginous (O’Rahily et al., 2004).

**Bones:** The skeleton includes the axial skeleton (bones of the head, neck, and trunk) and the appendicular skeleton (bones of limbs). Bone may be present in locations other than in the body skeleton. It often replaces the hyaline cartilage in parts of the laryngeal cartilages. Furthermore, it is sometimes formed in soft tissues, such as scars. Bones may be classified according to shape: long, short, flat, and irregular (O’Rahily et al., 2004).

- **Long bones:** Long bones (fig.3) are those in which the length exceeds the breadth and thickness. They include the clavicle, humerus, radius, ulna, femur, tibia, and fibula, and also the metacarpals, metatarsals, and phalanges. Each long bone has a shaft and two ends or extremities, which are usually articular. The shaft is also known as the diaphysis. The ends of along bone are usually wider than the shaft, and are known as epiphyses. The epiphyses of a growing bone are either entirely cartilaginous or, if epiphysial ossification has begun, are separated from the shaft by cartilaginous epiphysial plates (discs). The shaft of a long bone (diaphysis) is a tube of compact bone (“compacta”). The cavity of which is known as a medullary (marrow) cavity. The cavity contains either red (blood-forming) or yellow (fatty) marrow, or combinations of both. The cavity of the epiphysis and metaphysic contains irregular, anastomosing bars or trabeculae, which form what is known as spongy or cancellous bone. The spaces between the trabeculae are filled with marrow. The bone on the articular surface of the ends is covered by cartilage, which is usually hyaline. The shaft of a long bone is



surrounded by a connective tissue sheath, the periosteum. Periosteum is composed of a tough, outer fibrous layer, which acts as a limiting membrane, and an inner, more cellular osteogenic layer. The inner surface of compact bone is lined by a thin, cellular layer, the endosteum. At the ends of the bone the periosteum is continuous with the joint capsule, but it does not cover the articular cartilage. Periosteum serves for the attachment of muscles and tendons to bone (O’Rahily et al., 2004).

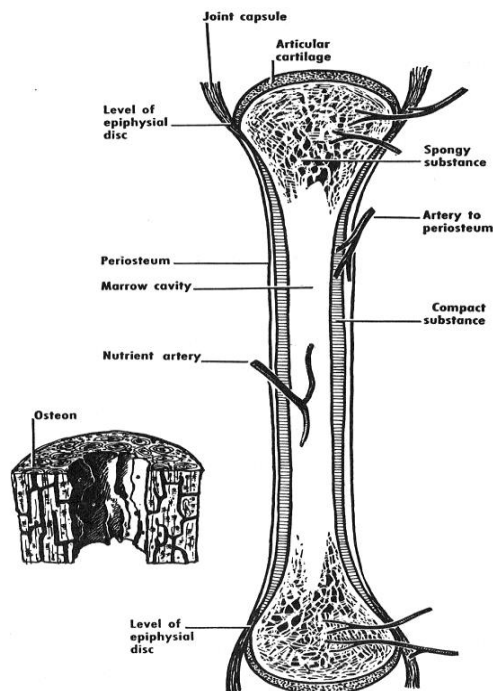


Fig.3: Diagram of a long bone and its blood supply. The inset shows the lamellae of the compacta arranged in osteons, i.e., vascular canals surrounded by concentric layers of bone (<https://www.dartmouth.edu/~humananatomy/>).

- **Short bones:** Short bones occur in the hands and feet and consist of spongy bone and marrow enclosed by a thin layer of compact bone. They are surrounded by periosteum, except on their articular surfaces.
- **Sesamoid bones:** Sesamoid bones are a type of short bone embedded within tendons or joint capsules. These occur mainly in the hands and feet, although the patella represents a particularly large example of a sesamoid bone. They vary in size and number. Some clearly serve to alter the angle of pull of a tendon. Others, however, are so small that they are of scant functional importance (O’Rahily et al., 2004).
- **Accessory bones:** Accessory or supernumerary bones are bones that are not regularly present. They occur chiefly in the hands and feet. They include some sesamoid bones and certain ununited epiphyses in the adult. They are of forensic importance in that, when seen in radiograms, they may be mistaken for fractures. Callus, however, is absent, the bones are smooth, and they are often present bilaterally (O’Rahily et al., 2004).
- **Flat bones:** Flat bones include the ribs, sternum, scapulae, lateral part of the clavicle, and many bones of skull. They consist of two layers of compact bone with intervening spongy bone and marrow. The intervening spongy layer in the bones of the vault of the skull is termed diploe: it contains many venous channels. Some bones, such as the lacrimal and parts of the scapula, are so thin that they consist of only a thin layer of compact bone (O’Rahily et al., 2004).
- **Irregular bones:** Irregular bones are those that do not readily fit into other groups. They include many of the skull bones, the vertebrae, and the hip bones (O’Rahily et al., 2004).

**Muscles:** Movement is carried out by specialized cells called muscle fibers, the latent energy of which can be controlled by the nervous system. Muscle fibers are classified as skeletal (or striated), cardiac, and smooth. Skeletal muscle fibers are long, multinucleated cells having a characteristic crosstriated appearance under the microscope. These cells are supplied by motor fibers from cells in the central nervous system. The muscle of the head is also composed of crosstriated fibers, but its activity is regulated by the autonomic nervous system. The walls of most organs and many blood vessels contain fusiform

(spindle-shaped) muscle fibers that are arranged in sheets, layers, or bundles. These cells lack cross-striations and are therefore called smooth muscle fibers. Their activity is regulated by autonomic nervous system and certain circulating hormones, as well as often reacting to local mechanical factors. They supply the motive power for various aspects of digestion, circulation, secretion, and excretion. Skeletal muscles are sometimes called voluntary muscles, because they can usually be controlled voluntarily. However, many of the actions of skeletal muscles are automatic, and the actions of some of them are reflex and only to a limited extent under voluntary control. Smooth muscle and cardiac muscle are sometimes spoken of as involuntary muscle (O’Rahily et al., 2004).

**Skeletal muscles (general characteristics):** Most muscles are discrete structures that cross one or more joints and, by contracting, can cause movements at these joints. Exceptions are certain subcutaneous muscles (e.g., facial muscles) that move or wrinkle the skin or close orifices, the muscles that move the eyes, and other muscles associated with the respiratory and digestive systems. Each muscle fiber is surrounded by a delicate connective tissue sheath, the endomysium. Muscle fibers are grouped into fasciculi, each of which is enclosed by a connective tissue sheath termed perimysium. A muscle as whole is composed of many fascicule and is surrounded by epimysium, which is closely associated with fascia and is sometimes fused with it. The fibers of a muscle of rectangular or quadrate shape run parallel to the long axis of the muscle. The names of muscle usually indicate some structural or functionan feature. A name may indicate shape, e.g., trapezius, rhomboid, or gracilis. A name may refer to location, e.g., tibialis posterior. The number of heads of origin is indicated by the terms biceps, triceps, and quadriceps. Action is reflected in terms such as levator scapulae and extensor digitorum. Muscles are variable in their attachments: they may be absent, and many supernumerary muscles have been described. Variations of muscles are so numerous that detailed accounts of them are available in special works. Individual muscles are described according to their origin, insertion, and action. Certain features of blood supply are also important (O’Rahily et al., 2004).

### 2.3.2 The nervous system

The nervous system comprises the central nervous system, consisting of the brain and spinal cord, and the peripheral nervous system, consisting of the cranial, spinal, and peripheral nerves, together with their motor and sensory endings (O’Rahily et al., 2004).

**Central nervous system:** The central nervous system is composed of millions of nerve and glial cells, together with blood vessels and a little connective tissue. The nerve cells, or neurons, are characterized by many processes and are specialized for reception and transmission of signals. The glial cells, termed neuroglia, are characterized by short processes that have special relationships to neurons, blood vessels, and connective tissue (O’Rahily et al., 2004).

**Brain:** The brain is the enlarged, head end of the central nervous system; it occupies the cranium, or brain case. The term cerebrum (L., brain; adjective cerebral) generally means brain, but sometimes is used for the forebrain and midbrain only. Encephalon, of Greek origin, is found in such terms as encephalitis, which means inflammation of the brain. The brain presents three main divisions: forebrain (prosencephalon), midbrain (mesencephalon), and hindbrain (rhombencephalon). The forebrain in turn has two subdivisions, telencephalon (endbrain) and diencephalon (interbrain). The hindbrain likewise has two subdivisions, the metencephalon (afterbrain) and the myelencephalon (marrowbrain). The bulk of the brain is formed by two cerebral hemispheres, which are derived from the telencephalon. The hemispheres are distinguished by convolutions, or gyri, which are separated by sulci. The diencephalon lies between the hemispheres. It forms the upper part of the brain stem, an unpaired stalk that descends from the base of the brain. The brain stem is formed by the diencephalon, midbrain, pons, and myelencephalon, or medulla oblongata. The last is continuous with the spinal cord at the foramen magnum. The cerebellum is a fissured mass of gray matter that occupies the posterior cranial fossa and is attached to the brain stem by three pairs of peduncles. Twelve pairs of cranial nerves issue from the base of the brain stem (O’Rahily et al., 2004).

### 2.3.3 The skin, hair and nails

**Layers of skin:** Common integument refers to skin and subcutaneous tissue, hair, nails, and breast. The last-named is described with the upper limb. The skin (cutis) provides a waterproof and protective covering for the body, contains sensory nerve endings, and aids in the regulation of temperature. The skin is important, not only in general medical diagnosis and surgery, but also as the seat of many diseases of its own. The study of these is called dermatology (Gk derma, skin). The area of the body surface is about 2sq m. the temperature of the skin in general is normally about 32 to 36 degrees C. The skin varies in thickness from about 0.5 to 3mm. it is thicker on the dorsal and extensor than on the ventral and flexor aspects of the body. It is thinner in infancy and in old age. The stretching of the abdominal skin during pregnancy may result in red streaks (striae gravidarum), that remains as permanent white lines (lineae albicantes). The skin consists of two quite different layers: 1) the epidermis, a superficial layer of stratified epithelium that develops from ectoderm, and (2) the dermis, or corium, an underlying layer of connective tissue that is largely mesodermal in origin. The dermis makes up the bulk of the skin (O’Rahilly et al., 2004).

**Corium (dermis):** The corium, or dermis, contains downgrowths from the epidermis, such as hair follicles and glands. It presents a superficial papillary layer of loose collagenous and elastic fibers, together with fibroblasts, mast cells, and macrophages. Elevations (papillae) project toward the epidermis. The thicker, deep reticular layer of the dermis consists of dense, coarse bundles of collagenous fibers. Some of the fibers enter the subcutaneous tissue, where they form bundles between lobules of fat. Smooth muscle is found in some regions (areola and nipple, scrotum and penis, and perineum). In some areas, muscle fibers of skeletal type (e.g. platysma) may be inserted into the skin. In tattooing, foreign particles, such as carbon, are introduced into the dermis. The skin lies on the subcutaneous tissue (“superficial fascia”), a layer of fatty areolar tissue that overlies the more densely fibrous fascia. It should be remembered that fat is liquid, or nearly so, at body temperature. The subcutaneous tissue serves as a depot for fat storage and aids in preventing loss of heat. When a pinch of skin is picked up, subcutaneous tissue is included. A hypodermic injection is one given into the subcutaneous tissue (O’Rahilly et al., 2004).

**Epidermis:** The skin is covered by a film of emulsified material produced by glands and by cornification. The epidermis is an avascular layer of stratified squamous epithelium that is thickest on the palms and soles. The epidermis, where it is thick, presents five layers. In the outer layers, which may conveniently be grouped as the horny zone, the cells become converted into soft-keratin flakes that are worn away from the surface continuously. The stratum corneum is tough, resilient, semitransparent cellular membrane that acts as a barrier to water transfer. Under normal conditions, mitotic figures are practically confined to the deepest layer, the stratum basale, which is, therefore, the normal germinative layer of the epidermis. The various layers show the stages through which the basal cells pass before their keratinization and shedding. The cells of the epidermis are replaced approximately once per month. Keratin is a protein that is present throughout the epidermis, perhaps in a modified form. It is readily hydrated-hence the swelling of skin on immersion in water-and dryness of the skin is due chiefly to a lack of water. Human epidermis displays a rhythmic mitotic cycle. Mitosis is more active at night, and it is stimulated by a loss of the superficial, or horny, zone. A part, or the whole thickness, of the epidermis may be raised up in the form of blisters by plasma when the skin is damaged (e.g., by a second-degree burn), and prolonged pressure and friction result in callosities and corns. Several pigments, including melanin, melanoid, carotene, reduced hemoglobin, and oxyhemoglobin, are found in the skin. Melanin, which is situated chiefly in the stratum basale of the epidermis, protects the organism from ultraviolet light. When an area of epidermis, together with the superficial part of the underlying dermis, is destroyed, new epidermis is formed from hair follicles, and also from sudoriferous and sebaceous glands, where these are present. If the injury involves the whole thickness of the dermis (e.g., in deep burn), however, epithelization can take place only by a growing over of the surrounding edge of the epidermis or alternatively by the use of an autograft. Free skin grafts of the epidermis and a part or all of the thickness of the dermis can be applied, and vascularization takes place through connections between the subcutaneous vessels and those in the graft. A defect of the skin that extends into the dermis is termed an ulcer. Lines of thickened epidermis known as papillary ridges form a characteristic pattern on the palmar aspect of the hand and the plantar aspect of the foot. They are concerned with tactile sensation. They contain the

openings of the sweat glands and overlies grooves in the dermis; these grooves are situated typically between rows of double ridges known as dermal ridges. The papillary ridges appear in fetal life in a pattern that remains permanently. They are especially well developed in the pads of the digits, and finger prints in adults and foot prints in infants are used as a means of identification of an individual (O’Rahily et al., 2004).

**Hairs:** Hairs (or pili; pilus in the singular) are characteristic of mammals. The functions of hair include protection, regulation of body temperature, and facilitation of evaporation of perspiration; hairs also act as sense organs. Hairs develop in the fetus as epidermal downgrowths that invade the underlying dermis. Each downgrowth terminates in an expanded end that becomes invaginated by a mesodermal papilla. The central cells of the downgrowth become keratinized to form a hair, which then grows outward to reach the surface. The hairs first developed constitute the lanugo, or down, which is shed shortly before birth. The fine hairs that develop later constitute the vellus. Although hairs on many portions of the human body are inconspicuous, their actual number per unit area is large. In a few places (such as the palms and the soles and the dorsal aspect of the distal phalanges) the skin is glabrous, that is, devoid of hair. The shaft of a hair consists of a cuticle and a cortex of hard-keratin surrounding, in many hairs, a soft-keratin medulla. Pigmented hairs contain melanin in the cortex and medulla, but pigment is absent from the surrounding sheaths. The color of hair depends mainly on the shade and the amount of pigment in the cortex and, to a lesser extent, on air spaces in the hair. In white hairs pigment is absent from the cortex, and the contained air is responsible for the whiteness; “gray hair” is generally a mixture of white and colored hairs. The root of a hair is situated in an epidermal tube known as the hair follicle, sunken into either the dermis or the subcutaneous tissue. The follicle is dilated at its base to form the bulb (matrix). In the obtuse angle between the root of a hair and the surface of the skin, a bundle of smooth muscle fibers, known as an arrector pili muscle, is usually found. It extends from the deep part of the hair follicle to the papillary layer of the dermis. On contraction it makes the hair erect. The arrectores pilorum are innervated by sympathetic fibers and contract in response to emotion or cold. This results in an unevenness of the surface called “goose pimples” or “goose skin” (O’Rahily et al., 2004).

**Nails:** The nails (or unguis; unguis in the singular) are hardenings of the horny zone of the epidermis. They overlie the dorsal aspect of the distal phalanges. They protect the sensitive tips of the digits and, in the fingers, serve in scratching. Nails develop in the fetus as epidermal thickenings that undercut the skin to form folds from which the horny substance of the nail grows distally. The horny zone of the nails is composed of hard-keratin and has a distal, exposed part, or body, and a proximal, hidden portion, or root. The root is covered by a distalward prolongation of the stratum corneum of the skin. This narrow fold is composed of soft-keratin and is termed the eponychium. Distal to the eponychium is the “half-moon”, or lunula, a part of the horny zone that is opaque to the underlying capillaries. Deep to the distal or free border of the nail, the horny zone of the fingertip is thickened and is frequently termed the hyponychium. The horny zone of the nails is attached to the underlying nail bed. The matrix, or proximal part of the bed, produces hard-keratin. Further distally, however, the bed may also generate nail substance. Moreover, the most superficial layer of the nail may be produced by the epithelium immediately dorsal to the root and proximal to the eponychium. The growth of the nail is affected by nutrition, hormones, and disease. Nail growth involves considerable protein synthesis, as a result of which nonspecific changes occur in the nails in response to various local and systemic disturbances. White spots indicate incomplete keratinization (O’Rahily et al., 2004).



## **2.4 Deterioration of mummies**

### **2.4.1 Factors of mummies' deterioration**

Deterioration in mummies is caused by several factors, including environmental conditions, physical damage, biological damage or damage caused by previous conservation attempts. Sometimes, these factors occur in isolation, but they can also be present in combination (David, 2001)

Insects are considered one of the most serious factors causing damage to Egyptian mummies. Panagiotakopulu (2001) reported that mummies, both human and animal, were highly susceptible to insect attack. The presence of insects depends on three factors (climate, food and competition with other living organisms) (Hill, 1985). Of the direct effects of climate, perhaps the most important, is that it governs the geographical distribution of insects. Some insects thrive in a temperate climate, others in a tropical one. For most insects there is an optimum climate in which we can most easily measure is temperature. Humidity and variation of light intensity or photo-periodicity can also be measured, but all the three are so closely bound together that is difficult to assess the part played by each one, except under controlled condition, as Elton has remarked (Munro, 1966).

The growth of microorganisms in organic materials such as mummies is dependent on the presence of moisture, although, other factors such as temperature should be taken into consideration to understand the biodeterioration mechanism (Valentin, 1996). Many fungal and bacterial species require available moisture for their development on the surface of an object. In this context, scant research has been done to determine the effect of moisture content in a material on the germination of microbial contamination (Valentin, 2001). A temperature range for the growth of microorganisms is 30°C (Valentin, 2002), this temperature keeps reaction rates in check, specifically the denaturing of collagen, the major constituent of mummy skin (Maekawa, 1998), as high relative humidity (RH) that arrives at 65% or higher aids in decomposition of mummies, although there are species of fungi that depend on a temperature range between 4°C to 35°C without need of moisture (Valentin, 2002). We can find other factors such as pH value which aid in the growth of fungi and bacteria, whereas the fungi prefer acidic environments pH

6 is suitable for growth (Abdel-Maskoud, 1995). High temperatures from internal case lighting and windows can also cause the mummified skin to stiffen and become more susceptible to cracking and chemical breakdown. The chemical destruction of mummified tissue can also occur through exposure to air pollution, often high in large cities and industrial areas. Sulfuric acid can result from sulfur emissions combined with fog. Potentially, sulfuric acid produced in the right atmospheric conditions can break down the proteins of the mummified tissue (Maekawa, 1998 & Maier, 2001).

#### **2.4.2 Environmental physicochemical parameters on mummies' decay**

##### 2.4.2.1 The composition of a mummy

According to Capelli (2005) the mummy of an adult maintains the same composition characteristics as a living person, except for the substantial difference of the water content in its tissues. Water represents approximately 60% of the total weight of a living adult man; divided into intracellular water (40%) and extracellular water (20%), of which 15% of the latter is interstitial water while the remaining 5% is plasmatic water.

Water excluded, the human body is 30% composed of soft tissue and the remaining 70% of bony tissue (Netter, 2002). The bony tissue and extracellular matrix is composed of organic components (30-35%) and mineral inorganic components (the remaining 65-70%) (Netter,2002). Samadelli et al., (2013) deduced, therefore, that the organic components of the soft and bony tissues represent circa 60% of a mummy.

Furthermore, Maekawa, (1988) stated that these tissues are composed almost entirely of collagen. Based on these scientific findings, it is imperative to focus on the collagen protein for evaluations and analyses aimed at determining the most efficacious physical-chemical parameters for conservation, while also taking into consideration that Egyptian dynastic mummies are usually wrapped in linen bandage a natural fibre of which 70% is cellulose (Aufderheide, 2003).

##### 2.4.2.2 Main causes of collagen decay

Proteins are above all affected by the following transformation processes: denaturation; proteolysis; putrefaction. Denaturation is the loss of quaternary,

tertiary or secondary structure of proteins with no disintegration of polypeptide chain (primary structure). Denaturation is mainly of a physical-chemical nature, through it can be induced by a microbial process. Among the most influential factors are: temperature, water activity ( $a_w$ ), and ionic concentration (Cheftel and Cheftel, 1988).

The main effects of denaturation are the loss of solubility, with subsequent aggregation and precipitation (coagulation), and the loss of specific chemical-physical and biological properties. Based on this scientific evidence, it is possible to state the principle according to which it is strongly recommended not to subject mummies to excessive lyophilizing treatments, because, during the first desiccation stage almost 95% of water is removed from the tissues, while in the second stage, which takes place at a temperature of circa 60°C, the water bound to the proteins is removed, thus activating the decay process (Jennings, 1999; Oetjen and Haseley, 2004).

Proteolysis and putrefaction are two highly collated processes since the former is the prelude to the second, while they partly overlapped with an ever-increasing intensity of the latter. Proteolysis is the disintegration of the primary structure of protein and results in the creation of smaller polypeptide chains and free aminoacids. Putrefaction is defined as the destruction of amino acids with subsequent development of strongly aromatic compounds derived from nitrogen metabolism, e.g. ammonia, amines, cadaverine and putrescine, and from sulphur metabolism, e.g. hydrogen sulphide or mercaptans (Cheftel and Cheftel, 1988).

A number of biological-molecular studies have demonstrated that the collagen present both in mummy tissues (Janko et al., 2004) and in ancient parchments is an extremely resistant protein which, when conserved in the correct climatic conditions, undergoes only minimum decay even over thousands of years (Sobel and Ajie, 1992).

#### 2.4.2.3 Importance of thermo-hygro-metric environment in mummy decay

The usual procedure of examination of conservation is to define the optimal conditions, i.e. the ranges in temperature and relative humidity, within which the correct conservation of mummies can be ensured. The manifestation and

persistence of temperature and relative humidity values outside those falling within the optimal range cause damage in the long term, but even sudden variations or fluctuation in the short term (9 days if not hours) can induce often irreversible alterations (EN 2010). Any object adapts in time to the environment in which it is placed, reaching equilibrium with its surrounding environment; the nature and speed of the deviation from this equilibrium might accentuate the decay processes (UNI 1999; ASHRAE 1997).

As a general principle, lower air temperatures are preferred because they reduce the probability of decay resulting from chemical-biochemical processes while also inhabiting microbial activity (Aghemo et al., 1997; prEN 2012). The continuing fluctuation in the temperature of the air in contact with the mummy induces, over time, thermal-driven mechanical stress, causing dilations and micro-cracking of the skin. On the basis of these studies on organic materials and collagen carried out in GCI laboratories, the ideal variation in the conservation temperature, in order to reduce biochemical reactions, lies between 10-15 °C (Maekawa, 1998). The temperature is a fundamental factor not only because it determines the type of microbial growth, which can be cryophile, psychrophile, psychrotropic, mesophile or thermophile in nature depending on the temperature range that stimulates their survival and growth (fig.4) (Cheftel and Cheftel, 1988).

#### 2.4.2.4 Photo-oxidation as a cause of decay

Photo-oxidation is a degradation process caused by the combined action of light and oxygen. In order to better understand this phenomenon, it is necessary, firstly, to analyze the mechanism instigated by light alone (photochemical degradation) in the absence of oxygen. The Grotthuss-Draper Law (named after the chemists Theodor Grotthuss and Jhon W. Draper) states that light must be absorbed by a chemical substance in order for a photochemical reaction to take place. Photochemical degradation refers to a set of processes which, following interaction with light radiation, leads to the chemical change of a substance. Chemical reactions take place exclusively when a molecule is provided with enough “activation energy” to cause the breaking of the covalent bonds present along the macro-molecules of natural polymers (Martuscelli, 2006).

These chemical reactions occur within a wide range of electromagnetic radiation starting from the visible spectrum, passing through ultraviolet up to ionizing radiation, which universally has a wavelength ( $\lambda$ ) less than 400 nm (EUR 2003).

International recommendations, in their latest draft form, identify four photosensitivity categories for cultural heritage finds and establish the relevant illuminance maximum values permitted. Mummies belong to “4-very high” category, applied to finds that are extremely sensitive to light (MIBAC 2001). The illuminance maximum value permitted for this category is 50lx and an annual dose of not greater than 15,000 lx. The framework of conservation recommendations and norms in the field of lighting represents a reasonable compromise between the inevitable degradation action caused by exposing mummies to light and museum requirements of putting the mummy on display.

However, it is necessary to pay particular attention to the type of light used for illuminating Egyptian dynastic mummies in oxygen-free environments because, according to scientific studies, the linen bandage they are usually wrapped in is much more sensitive to photo-oxidation (Leary, 1967).

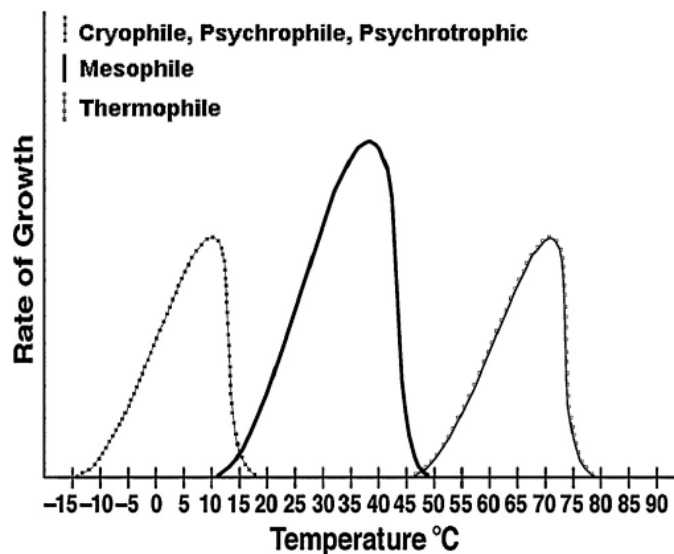


Fig.4: Graph illustrating the effects of temperature on microbial growth (Cheftel and Cheftel, 1988).

Moreover, it is this fundamental principle that provides the guideline for defining the conservation criteria concerning the mummies preserved in storage rooms, while for mummies on display in museums the choice is more dependent on a compromise which allows the ideal conservation temperature to correspond to the display environmental temperature (prEN 2012; Camuffo, 1998).

The relative humidity of air (RH) is defined as the ratio of water vapour pressure to its saturation vapour pressure at a given air temperature ( $t_a$ ). It can be expressed in percentage according to eq. (1) (ASHRAE, 2009).

$$RH = \frac{P_v}{p_{vs}(t_a)} \cdot 100 \quad (1)$$

In normal atmospheric conditions, where the absolute and specific humidity of an air parcel are constant, relative humidity is anticorrelated with respect to the temperature (Camuffo, 1998; EN 2010).

This parameter influences the variations in size and in shape of the tissues constituting the mummy, as well as the chemical and biological processes. In particular, all organic materials that absorb water swell when the relative humidity increases and desorbs when it decreases, with subsequent variations in weight, deformations, fibre fractures and micro-cracking. This effect is technically defined as “a physical deterioration mechanism”. Very often it is hard to discern the difference between the decay caused by temperature and that due to humidity because of the inextricable synergy existing between these two parameters. The ASHRAE psychrometric chart can be used to obtain a graphical evaluation of the relative humidity as a function of the temperature (ASHRAE, 2009; ASHRAE, 1996).

The importance of relative humidity (RH) to conservation is immense (FprEN 2012) and tightly linked to the concept of water activity ( $a_w$ ), the parameter proportional to the escaping tendency of the water molecules present in the tissues constituting the mummy. The water activity ( $a_w$ ) expressed by eq.2 corresponds to the ratio of the partial vapour pressure of water in the tissues

( $P_w$ ) to that of pure water ( $P_w^0$ ) at the surface temperature (Cheftel and Cheftel, 1988, EN 2009).

$$a_w = \frac{P_w}{P_w^0} \quad (2)$$

After a certain time, any substance placed in a sealed environment reaches a “thermal and hygrometric” equilibrium with its surrounding air. Accordingly the moisture content of the substance reaches an equilibrium with the humidity of the environment (EMC). In these conditions, the value of the air relative humidity at equilibrium (ERH) corresponds to the water activity ( $a_w$ ) value of the substance (Cheftel and Cheftel, 1988).

$$a_w = ERH/100$$

The tissues constituting mummies can be considered as more or less concentrated “solutions” and the measurement and limitation of the water activity ( $a_w$ ) are essential to the conservation process because they influence the life of microorganisms and enzyme activity. The greater the water activity ( $a_w$ ) value, the faster the decay of organic tissue, as synthetically represented in the chart (fig.5) (Labuza, 1975).

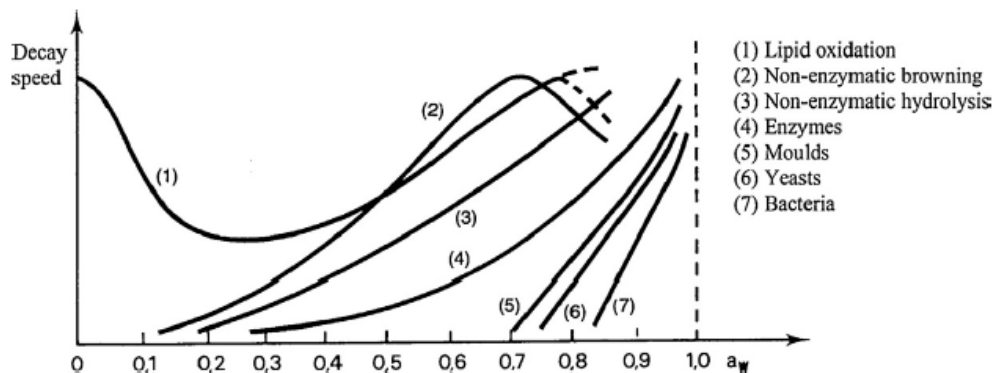


Fig.5: Decay speed of organic tissues in function of water activity (Labuza, 1975).

Table 1 indicates the minimum value of water activity ( $a_w$ ) for the survival and growth of microorganisms (Scott, 1957), it further confirms GCI affirmations, according to which relative humidity values higher than 65%, combined with temperature values higher than 20°C, favour the development of moulds and accelerate the metabolism of many harmful insects (Maekawa, 1998).

Microorganisms	Water activity ( $a_w$ )
Bacteria	0.91
Yeasts	0.88
Moulds	0.80
Halophilic bacteria	0.75
Xerophilic moulds	0.65
Osmophilic yeasts	0.60

Table1: Minimum water activity necessary for growth of microorganisms.

However, it is still very difficult to define the importance of water activity ( $a_w$ ) in organic tissues in relation to lipid oxidation, non-enzymatic browning and enzyme activity, since they are collocated in an extremely dehydrated environment and these types of phenomena have not yet been fully studied nor to understood completely (Cheftel and Cheftel, 1988). Therefore, it is not possible to provide general indications in the absence of data on the exact chemical composition of the mummified tissues.

It is reasonable to assume that with water activity ( $a_w$ ) lower than 0.3 it is possible to inhibit the majority of the causes of biochemical decay in the organic tissues. In particular, for “anthropogenic” mummies, whose moisture content in the tissues is already very low, it should not be difficult to set this environmental parameter. However, attention should be paid to the minimum  $a_w$  threshold value for these types of mummies, which, according to scientific studies, should correspond to the limit set by collagen degeneration, an irreversible process that is instigated when the value of  $a_w$  decreases below 0.25 (Valentin et al., 1990). This limit threshold is reasonable because at  $a_w$  values ranging from 0 to 0.2, water molecules are strongly bound thanks to the creation of hydrogen bonds or electrostatic interactions between the water molecules themselves and highly polar groups (various form of amino, carbonylic or hydroxylic group present in carbohydrates and proteins). In these



conditions, there is no water available to favour microbial development or stimulate the onset of chemical reactions that would result in deterioration on the tissues (Cheftel and Cheftel, 1988).

When evaluating the most effective chemical-physical parameters to be applied for the conservation of mummies, the presence of the linen bandage and its effect on mummy should be taken into account. This consideration is made in the knowledge that fabrics of organic nature maintain their physical integrity at relative humidity rates ranging from 40 and 55%. Below a 40% threshold, elasticity is lost thus leading to an increase of the fragility rate (Aufderheide, 2003).

#### 2.4.2.5 The role of quality of air in the decay of mummies

One of the main decay mechanisms in mummies is defined as “chemical”. It is dependent on the occurrence of chemical reactions and oxidation in particular, but it can include also other undesirable reactions, which could be catalyzed by enzymes, such as proteolysis or lipolysis, isomerization, or photodegradation. The potential to cause irreversible damage depends on the concentration of reagents and on the environment which can favour or inhibit reactions. The elevated presence in the urban air of chemical compounds, such as NO<sub>2</sub>, SO<sub>2</sub> and O<sub>3</sub> and of many volatile organic compounds (VOCs) caused by emissions, which derive from normal anthropogenic activities, into the atmosphere has been known for a long time (Bernardi, 2004), this is true also for acidic and corrosive compounds originating from secondary reaction that result from the contact of these substances with the humidity present in the air. Moreover, the effects of decay caused by these phenomena on cultural assets and on organic materials are indisputable (Schieweck and Salthammer, 2011).

The definition of the chemical composition of the air in which mummies are preserved is essential for their conservation since it influences the growth of microbial flora. Bacteria grow within quite a wide pH range (4.5-11.0) (Table 2), however, the majority of them enjoy strong growth in optimal conditions of neutral pH values (6.5-7.5). Nevertheless, it is known that certain microorganisms, in particular yeasts and moulds, are able to grow at very low pH values as well (Tiecco, 1999).

	Minimum	Optimum	Maximum
Bacteria	4.5	6.5–7.5	1.0
Yeasts	1.5–3.5	4.0–6.5	8.0–11.5
Moulds	1.5–3.5	4.5–6.8	8.0–11.0

Table2: pH maximum and minimum values for various microorganisms.

The inhibiting action of pH, as is the case for any other factor, has a greater effect as the temperature deviates from those values that favour optimal development of the microorganism under analysis. Many studies have demonstrated that the acid tolerability of the organism decreases as the conservation temperature lowers. Furthermore, it should be noted that pH alone can inhibit the development of bacteria with a greater effectiveness, but this effect is diminished for moulds and yeasts, and only the combination with other factors, such as temperature and water activity ( $a_w$ ), results in a significant inhibiting effect on micro-flora. The presence of  $O_2$  in sufficient quantities can favour the development of the microorganisms, which, by metabolizing the acids, causes the pH to shift towards neutral values thus lowering its inhibiting potential. Therefore, should pH be the main conservation factor, it is necessary that mummies are preserved in environments with reduced  $O_2$  concentration (Tiecco, 1999).

The effects of oxidation activity due to the contact of oxygen with any type of material are well known and widely documented (Feller, 1977). It is common knowledge that the nature presence of oxygen in the air, equal to circa 20.9 m/m, is essential for human life, but it is equally essential for any type of aerobic life form, though the oxygen percentage present is lower. In order to improve the conservation of mummies, it would be advisable, as a principle, to completely replace the air within the museum showcases with properly humidified inert gas (e.g. argon or nitrogen), since it has been proved that collagen decay is significantly reduced in oxygen free conditions (Sobel and Hansen, 1989). Moreover, interesting experimental studies carried out in GCI (Maekawa, 1998) laboratories demonstrate the mortality time of many insect species observed in anoxic conditions in function of different thermo-hygrometric conditions (Selwitz and Maekawa, 1998). This is further evidence

that the recommended oxygen threshold for the optimal conservation of mummies should be zero or at least be lower than 0.1% (Maekawa, 1998). Various scientific studies have investigated the possible consequences which could occur to mummified tissues preserved in anoxic environments. In these conditions the collagen from mummified tissues must be kept at a relative humidity between 25% and 40% in order to prevent decay (Hansen et al., 1992).

## 2.5 Methodology of studying mummies

In general the preservation of mummy after study involves:

- (1) Identifying the type of mummification process used and the state of deterioration;
- (2) Selecting an appropriate method of study;
- (3) Individualized restoration; and
- (4) Recommending storage conditions should show the respect that each human body deserves, taking into account the laws and beliefs of each nation (Lombardi, 2001).

### 2.5.1 The different stages of studying a Mummy

The first stage in studying a mummy is a visual examination *in situ*, and thorough documentation. When recording a mummy the need for documentation is paramount, both visual and textual (appendix). This is one of the most important parts of the process. Photographs and videos as well as verbal/written descriptions of what is observed and what happens should be made throughout the mummy's life, from discovery and excavation to removal to storage. Often, despite ones best efforts, mummies do not emerge intact, and thus it is best to record as much information as possible as soon as possible. Of course, when the mummy is *in situ*, only limited amount of information can be extracted from it, especially if it is in a coffin. In the initial phase, one should record whether it is bandaged or not, patterns of bandaging if visible, if flesh, skin, and hair survive or not (Ikram et al., 2015).

After establishing the orientation of the burial in detail, the first challenge is the removal of the mummy (Ikram 2009). How this is accomplished depends on several issues, the most salient of which are: the type of mummification employed the matrix in which the bodies lay, whether the bodies are in a container, and the type of research to be carried out on them. Ideally, a conservator will be on hand to help with the logistics of removing the mummy from its matrix (Garcia et al., 2008). Thus, when a mummy is discovered, prior to its removal, the excavators, together with the conservator, should think of

how best to clear it, lift it, move it, and store it. This planning stage might take more time than the actual removal, but is conducive to a smooth and successful operation. Patience and planning is the key to the success of the endeavor (Ikram et al., 2015).

If relatively intact, the mummy should be extracted as one unit. If it lies straight in the ground, this can be achieved by placing flexible metal sheets (best) or chipboard beneath them, to move them to their study/storage area. Depending on the robusticity of the mummy, even cloth (and wood) stretchers can be used for transportation. Also, bands of cloth can be passed beneath the mummy and these can be used to lift it by two teams on either side, working in unison; this is particularly useful when the mummy is in a coffin. In some instances, in rectangular coffins, and very much depending on the state of conservation, one wall of the coffin can also be temporarily removed and the mummy extracted (Ikram et al., 2015).

In cases where the mummy is in poor condition, cyclododecane can be used to consolidate mummies. This wax-like substance is impermanent; unless sealed with foil or plastic, it evaporates, leaving the object intact. If sampling is to be carried out on the mummy, it is best that this be done before extraction takes place, especially if cyclododecane is being used, although this does not affect all types of tests. Whenever possible, those who actually handle the mummy should use gloves and masks to avoid contamination and to protect the mummy as much as possible (Ikram et al., 2015).

### 2.5.2 Examining the mummy

There are several steps involved in studying a mummy, not all of which can be achieved in the field, or indeed at all, due to financial and legal restrictions that might apply. However, the basics can be carried out, regardless of the location and the budget. The first step is a visual examination accompanied by photo documentation. If possible, once the body has been moved, both the recto and verso should be examined and photographed. This is followed by radiography, ideally both X-rays and CT-scans, and finally by a variety of tests that can be carried out to determine the materials of mummification, diseases that the deceased might have suffered from, genetic information, and possibly studying mummies in the field, and below, there is more detailed version that

this author uses in the field when no unwrapping takes place; it can be adapted for each site and the different situations that are encountered (Ikram et al., 2015).

### 2.5.3 Visual examination

The most basic and important part of examining a mummy is a visual examination. After photography (overall and detailed), this should include measurements of the entire body, or whichever parts are being examined (length, length of arms and/or legs) width at shoulders, width at hips, width at feet, if possible circumference of head), with a notation being made as to whether it is wrapped or unwrapped, details about the bandages (see section on bandages), the presence of bead nets or other adornments of cartonnage, garlands, and amulets if they are visible (Ikram et al., 2015).

### 2.5.4 Examining the bandages/wrappings

Records should be made of any patterns that the bandages follow-or if they follow none. The quality of cloth used should also be noted, and definitions provided for how the quality is being judged. Fineness of the threads and number of threads in the weft and/or warp of one cm<sup>2</sup> is an acceptable way of defining qualities, or clearly illustrated (photos are preferable to drawings) examples of each type can be provided as a guide. If any of the bandages have woven or colored patterns, these should be recorded. A textile specialist is useful for such investigations. The number of layers of bandages should also be noted, as should the average widths of the strips, and the presence of fringed bandages, complete or partial garments used in wrapping should also be recorded when possible. If a shroud is used, its dimensions and measurements, if possible, should be documented. If pads of linen cloth are encountered as part of the wrappings, their dimensions and quality should be registered. The knots used in tying together bandages to secure and lengthen them should also be recorded and photographed. Some bandages bear inscriptions, and all loose mummy bandages should be examined for these; however, in a well-preserved, wrapped mummy these cannot be detected unless they are easily accessible. Unwrapping mummies is not encouraged unless it is disintegrating or is only a partial body, as mentioned above (Ikram et al., 2015).

If one is examining the body where the wrappings have fallen off, one should note the presence of tampons of linen in the nose and ears. The eyes and sometimes the mouth also contain textile, which should be noted (Ikram et al., 2015).

The colors of the linen-particularly relevant in mummies dating from c.500 BC to AD 400-should also be recorded, possibly using a Munsell or paint chart, although the use of the former is still somewhat controversial as different people see the variations in color in a variety of ways. Photographs taken with a spectrum scale should solve this issue. In mummies of an earlier date, colored bands, generally blue, are found forming a border, and these too are noteworthy. Some bandages bear texts, and thus care should be taken to examine each piece of loosen linen carefully (Ikram et al., 2015).

Sometimes cords of linen or even papyrus or halfa grass are used to secure mummies. These should be recoded and studied. Such materials are frequently found in the wrapping of animal mummies, and even within mummies or as garlands. The involvement of an archaeobotanist is important for these identifications, which can provide evidence for the time of year when mummification occurred, and could also hint at the location in which it took place (Ikram et al., 2015).

#### 2.5.5 Examining the body

If the body itself is visible, then it should be described with attention paid to its degree of preservation of skin, nails, and hair: on the head, face, and body. Its overall color and odour should be noted, together with the overall degree of preservation. The nostrils should be examined to see if one can tell if the ethmoid was broken and the brain extracted. Plugs of linen in the nostrils and ears should be recorded. If the neck is visible, one should check to see if excerebration might have occurred via the atlas. The left side of the torso should be checked for the embalming cut and subsequent treatment. If the interior is exposed its packing should be documented. Care should be taken in examining the whole body as sometimes tattoos are found on some of the limbs (Ikram et al., 2015).

The presence of oils is difficult to detect, but stains of oils, resins, or body juices, particularly on the back of the mummy (or where it was positioned after preparation, in the case of animals), should be noted and described. These materials are also found on the body itself, and manifest themselves as dark (black) material. Furthermore, remnants of natron should be recorded. Further information can be extracted from these materials by carrying out tests on them (see section on testing) (Ikram et al., 2015).

Insects or their larvae that are found in the mummy provide information about the making of the mummy-if there is an abundance of fly larvae and poor preservation, clearly there was an insufficiency of natron used during the desiccation process. Using insects, an entomologist (forensic or other) can shed light on the body's exposure to the atmosphere after death (Ikram et al., 2015).

A physical anthropologist can establish the sex, age, and health of the mummy by a visual examination of the bones, and some information can be derived through endoscopy if the mummy is not too tightly wrapped. However, if the mummy is wrapped radiography is needed to study these (Ikram et al., 2015).

#### 2.5.6 Radiographic Imaging examination

In field conditions a portable X-ray machine should be used to image the body without damaging it. These images can provide some information about the wrappings, but mainly show the bones within the wrappings, as well as any amulets on papyri. The bones can show the position of the body, age and sex of an individual and certain aspect of the individual's health (nutrition, broken bones, diseases that are manifest on the bone) can also be revealed. Furthermore, an X-ray is useful as a guide if the mummy is CT-scanned as it allows one to place the slices along the body and to target specific areas for more detailed study (the ideals of imaging in a controlled environment are outlined in Adams and Aslop (2008) and Lynnerup, (2009); it should be noted that these technologies are always advancing) (Ikram et al., 2015).

CT-scans provide a more comprehensive view of the mummy as they consist of several 'slices' of X-rays of an object. These enable one not only to examine the bones, but also the layers of bandages and their contents. However, CT-scanning is expensive and it is not always possible to remove mummies from



the site to take them to a CT-scanner, which makes the X-rays all the more important. It should be noted that if the CT-scans are made and if they are of a high enough quality, these images can be used to create a virtual unwrapping of the mummy, revealing the body and its contents. Facial reconstruction can also be based on the information derived from the scans (Ikram et al., 2015).

#### 2.5.7 Chemical and Other Tests

Several tests can be carried out on the bandages, bodies, and embalming agents to flesh out ones understanding of the mummy. Tissue samples can be used for histological tests, the identification of different diseases, including malaria, leishmaniasis, schistosomiasis, tuberculosis, trichinosis, as well as other diseases (Millet et al., 1980; Miller et al., 1992; Miller et al., 1994; Zink et al., 2006; Sabbahy, 2015). C-14 dating might also be used on flesh, as well as on some of the other materials associated with mummies, such as the bandages, although these might date to considerably earlier than the body itself (Ikram et al., 2015).

Bones and teeth can be subjected to isotope and strontium analysis (Thompson et al., 2005; Buzon et al., 2007; Abbate et al., 2010) in order to establish the origins, diet, and movement of both humans and animals. These anatomical elements can also be used for C-14 dating, as well as for DNA. Although DNA can be extracted, its accuracy remains a vexed question (Pääbo 1985; 1986; Cooper and Poinar, 2000; Zink and Nerlich, 2005; Hawass et al., 2010; Lorenzen and Willerslev, 2010). Nonetheless, it is useful to sample the mummies and to keep these as part of the finds, or in storage bank, either the Manchester mummy Storage facility, or by creating one in Egypt (Ikram et al., 2015).

Additionally, simple tests, such as solubility, or more complex ones, such as gas chromatography, mass spectrometry, and XRF can be carried out on many of the materials used in preparing the mummy, so that they can be identified (Buckley et al., 1999;2004; Buckley and Evershed 2001). Such identifications not only shed light on how a mummy is made, but also about the socio-economic background of the deceased, and the trade networks present at the time that allowed for different materials to be used in mummification (Ikram et al., 2015).

### 2.5.8 Storage

The excavators' and researchers' responsibility to a mummy do not end with documenting and studying it (Gracia et al., 2008). They also need to ensure that they provide a basic level of individual storage to protect the mummy. Thus, planks of wood with straps for securing the mummies, or shallow coffins are good options. The mummy should be protected from dust and dirt by a covering, ideally of acid free paper, but even some kind of textile sheeting is acceptable. Insect threat is a major problem, and insect traps should be placed in strategic locations around the mummy. Camphor and creosote are also possible pest deterrents, but would need to be renewed (Ikram et al., 2015).

In Egypt, the Ministry of Antiquities is responsible for providing and maintaining the storage space. Storage of mummified remains is difficult due to practical issues of space, climate, conservation, finance, and security. An ideal solution would be a room dedicated specifically to mummy storage with climate control, but this is rare and not always feasible. Thus, a room (or tomb) with a relatively constant climate is ideal, with regular checks being made to ensure that there are no leaks or insect infestations (Ikram et al., 2015).

### 2.5.9 Publication

The study of a mummy is not complete until it has been published. After excavation, a basic publication should be made, as soon as possible-without this, the work of the scholars is no different from that of the grave robbers. The initial publication can provide the visual results of the examination if no imaging has taken place. Subsequently, if imaging or testing is carried out on the mummy, the results of these can be published. A useful mode of publication for a group of mummies is illustrated in Raven and Taconis (2005), which has individual entries as well as a table that should be used as a template for mummy publications (Ikram et al., 2015).

### 2.5.10 Final Remarks

Mummies are composite artefacts that give one rare chance to study and come to know individual ancient Egyptians, as well as being a source of evidence that allows one to better understand Egypt's history, culture, religion, economy, and

trade. They unite the research of a variety of scholars; excavators, physical anthropologists, paleopathologists, archaeobotanists, textile specialists, entomologists, chemists, mummy specialists, and Egyptologists. It is hoped that excavators and scholars will use all possible means to study mummies holistically in order to extract the maximum amount of information from them, and will be able to store and protect them for future generations to study as new technologies become available (Ikram et al., 2015).

### **3. PRACTICAL PART**

#### **3.1 Scope of thesis research**

The aim of the thesis is to study the preservation state of two Egyptian mummies from Anthropological museum of Athens by:

1. Identifying the nature of materials found on mummies and investigating their preservation state.
2. Describing the deterioration aspects found on mummies.
3. Defining the factors of deterioration on the specific mummies.

By the completion of this study valuable information will be available in order to:

- a) Propose future research analysis for the integrated examination of the specific mummies.
- b) Investigate the future conservation methods for the preservation of the mummies studied.
- c) Develop the knowledge and improve the correlation between physicochemical and archaeological data on Egyptian mummies' analysis.

## 3.2 Materials and Methods

### 3.2.1 Historical background

The mummies of the two young boys belong to the collection of Anthropological Museum of Athens (fig.6). The museum is housed at the Department of Histology and Embryology, Medical School, National and Kapodistrian University of Athens. It is one of the oldest museums in Europe as it was founded in 1886. It is accessible to the public and many schools visit it annually. The backbone of its scientific collections is a number of skeletal series, covering all the Greek world geographical space and represent people of all chronological and cultural periods of the ancient Greek world.

Little information is available for the historical background of the mummies. So far it has been found that they were donated from a German Institute/museum in Bermi before 1860. After that, they were included for a short term in the collection of the museum of Zoology and Natural History at the University of Athens.

It is estimated they are dated in the first centuries after Christ (A.D.). The deceased were children at the age of 3-5 and 5-7 years old when they died. Due to the elaborate mummification style, most probably they belong to the high society status. The bandages of the younger mummy have been removed allowing the recognition of its sex, male. The other mummy, also male preserves its hair; they are tied at a small braid, a common Egyptian hair combing for the royal young males. Their origin, as well as more background information, is still under investigation, due to missing archives.



Fig.6: The Anthropological Museum of Athens.

### **3.2.2 Visual Examination**

Visual examination and documentation was conducted in collaboration with the Anthropologist Dr D. Moysidou, and the Egyptologist Dr V.Chryssikopoulo. Primarily the mummies were moved from the storage room of the museum to the laboratory of the museum. Protective conservation materials for packing and transportation were used for safe transfer of the bodies. In addition documentation code numbers were given to mummies as M1 and M2 for reference.

The first stage of examination included photograph documentation of the mummies. Full body photographs were taken as well as detailed once with the use of high resolution digital camera Nikon D810 (led: Nikon AF-S, Micro Nikkor 105mm f/2.8G IF-ED VR, Macro:Tamron SP 24-70mm F/2.8 Di VC USD) and the digital camera Olympus SZ-17. Afterwards separate measurements of both bodies were taken (length, length of arms and/or legs, width at shoulders, width at hips, width at feet etc). Also macroscopical examination of the body and the bandages was performed, including recording every stage of the above examination.

### **3.2.3 Sampling procedure**

Fragmented materials of the mummies were collected and digitally recorded. In the case of M1h the sample was retrieved with the use of the scalpel No3; (fig.7) whereas the samples M1h and M1i collected from the already opened thoracic cavity of the mummy's with the use of a tweezers. Subsequently, all the samples of research were stored in polyethylene bags and tagged with the serial code numbers (M1 and M2). At the end all selected samples were stored in a polypropylene box which contained antacid paper and silica gel in order to control the humidity in the container (fig.8).

For the study of deterioration samples were collected by swabbing with dry swabs in tubes (fig.9) and sampling by clear masking tape. Pieces of masking tape were placed on slides for microscopy observation (fig.10). Masking tape was used on white spots that were observed of M1 and M2 bodies and dark spots from the M1's fragmented fabric piece. All the collected samples including the areas that were taking from are presented on the Table 3, 4 and 5.



Fig.7: Retrieval of M1h sample by the use of scalpel.



Fig.8: Stored samples in plastic polypropylene box, with antacid paper and silica gel.



Fig.9-10: Dry swabs (left image) and clear masking tape (right image) for biodeterioration examination.

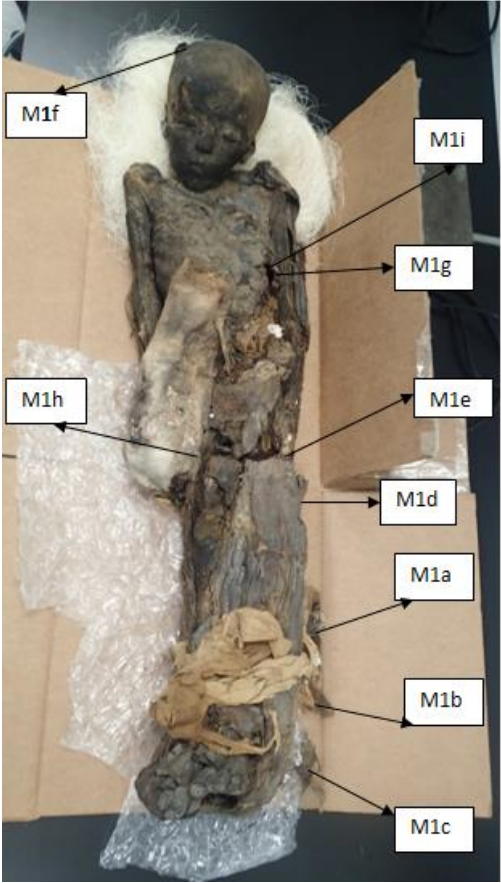
<b>Samples for analysis techniques</b>	
<b>Mummy1</b>	
M1a: fragment of bandage	
M1b: fragment of bandages (with colors)	
M1c: fragment of balm-impregnated bandages	
M1d: fragment of the tissue from the right femur	
M1e: fragment of the bone and the tissue probably from the right femur	
M1f: a bunch of fragmented scalp hairs	
M1g: a piece of balm from the interior of the thorax	
M1h: a piece of the wrapped fabric	
M1i: fragment of the plant from the interior of the thorax	

Table 3



<b>Samples for analysis techniques</b>	
<b>Mummy2</b>	
M2a1: black bead M2a2: yellow bead	
M2b: piece of the (later) textile where the mummy is lied on	
M2c: fragments of tissue from the left area of the thorax	
M2d: fragments of bandages (with color)	
M2e: fragment of bandage	
M2f: fragment of different type of bandage	
M2g: fragments of balm-impregnated tissue	

Table 4

<b>Samples for microbiological analysis</b>		
<b>Mummy1</b>	<b>Mummy2</b>	<b>Method</b>
M1.1: Upper left side of the thorax	M2.1: the pines	Swab & masking tape
M1.2: Fragmented painted textile		Swab & masking tape
M1.3: Left side of the neck		Swab & masking tape

Table 5

### 3.2.4 Stereoscopic examination

Stereoscopic observation was performed in all selected samples to collect information

1. about the nature of materials
2. the preservation state of the materials' surface
3. the products (if any) of deterioration and the deterioration aspects
4. Collect possible information of manufacture technology.

For that purpose OPTICA SZN-1 with binocular zoom stereomicroscope and led cold light generator was used in the Conservation laboratory of Archaeological museum of Pylos, Messenia.

Furthermore, for stereoscopic analysis, USB Microscope X200 KONIG Electronic was also used for the examination of M1f, M1b and M1g samples (which included hair and insects). This examination took place in the University of Peloponnese, Kalamata with the collaboration of the PhD researcher E. Kyriazi.

With regards to hair samples, each ones length (M1f01-M1f10) was counted with the following method: a glass slide was placed gently on top of each hair and with the use of indelible pen the length of the hair was draw on the slide. After that a thread was used to reflect the hairs' curvature. At the end, straightening the thread, the original hair's length was counted by a digital caliper (No.: Z22855, version: 06/2007) (fig.11-Table 11 pp. 154).

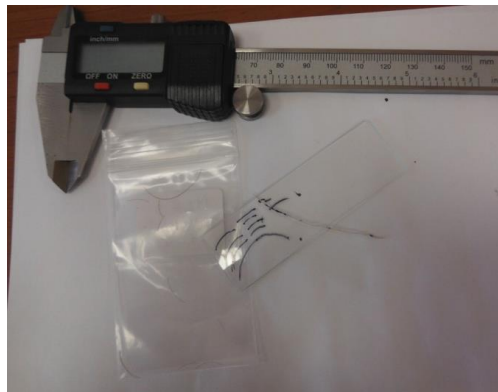


Fig.11: Hairs' length performed by the use of a glass slide, a thread and a digital caliper.

### 3.2.5 Optic microscope examination

Optic microscopy was used in order to:

- Study the preservation state and the coated material of the sample M1f (bunch of scalp hairs). LEICA DM EP microscopy was used at the University of Peloponnese with the collaboration of PhD researcher E. Kyriazi. Each hair was put on the slides with some drops of demineralised water and a coverslip.
- Study the morphology and identify the origin of the sample M1i (a piece of plant). The preparation and observation were conducted by the Prof. C. Faseas, Biologist and the Dr M. Nikolopoulou, Agronomist at the Agriculture University of Athens (fig.12). Pieces of the plant were cut off in sections with the use of a razor and then were located on glass slides with demineralised water and a coverslip (fig.13).



Fig.12: Optic Microscopy at the Agriculture University of Athens.



Fig.13: Sample preparation of M1i for Optic Microscopy observation.

### **3.2.6 Scanning Electron Microscopy (SEM) and Energy-dispersive X-ray spectroscopy (EDX)**

SEM analysis was used on the following samples:

- M1i-fragment of unknown plant for the examination of the preservation state, the morphology of the sections and possible identification of the species of the plant. The operation of the method was conducted in collaboration with the Prof. C. Faseas, Biologist and the Dr. A. Nikolopoulou, Agronomist at the Agriculture University of Athens. JEOL JSM 6360 SEM was used. The sample was cut off in several pieces and sections; then they were coated by gold (fig.14).
- M1a, M1b, M1c, M1h, M2d, and M2f (fragments of bandages) to examine the type of the fibers, the structure of the weaving, the preservation state and the deterioration aspects. Also EDEX was used in order to study the nature of materials that were covering the above samples as well as the ingredients of the color pigments on the sample M1b. The M1b sample was cut off in three small pieces per color (white, red and blue).
- M1d and M2g (tissue) to examine the preservation state and the materials covering the mummified tissue; EDX also was used. Also M1d sample was cut off in three small pieces (according to the basic three layers that it presented).
- M2a1 and M2a2 (black and yellow beads) for the investigation of the the preservation state, information on materials structure and possible manufacturing technology; EDX was also used. The analysis was performed in collaboration with Dr. Y. Fakorellis, Chemist, and Dr., D., Moyssidou, Anthropologist at the Technological Institute of Athens. No preparation of the sample was used. Also M1d sample was cut off in three small pieces (according to the basic three layers that it presented). All the samples were mounted on the metal stub by the use of carbon tape. All the above samples measured using the INKA software, accelerating voltage of 20 keV energy and a beam 40 spot size.
- M1f (bunch of scalp hair) in order to examine the hair type (fine or curl), the preservation state and the coated material. EDX also was used.

The samples measured using the INKA software, accelerating voltage of 20 keV energy and a beam 40 spot size.

The analysis operated in collaboration with the PhD candidate E. Kyriazi, at the University of Peloponnese. Two hairs were selected for investigation (M1f1 and M1f3) (fig.15); they were mounted on the metal stub by the use of carbon tape. Two fresh hairs for male and female (Csm and Csf) also were collected in order to be compared with mummies' hairs.



Fig.14: M1i sample were coated with gold for SEM analysis.



Fig.15: M1h sample. Two hairs were selected for SEM analysis.

### 3.2.7 Attenuated total reflectance (ATR) and Fourier Transform Infrared spectroscopy (FTIR) analysis

ATR and FTIR analysis were applied on the samples in order to reveal any information on possible embalming materials, the preservation state and the nature of deposits. The analysis was conducted in collaboration with Dr. S. Boyatzis, Chemist, at the Technological Institute of Athens.

ATR analysis was used on the samples:

- M1h, M1b, M2d, M2f, (fragments of bandages)
- M1d , M2g (fragments of tissue),
- Mπδ1 (control sample-piece of skin from the Dr. Boyatzis, S. thumb)

For ATR analysis no preparation was needed. In the case of the fragments of bandages, the samples were examined for both sides. Three adjacent points of M1h sample were examined. Similarly three points of M1b sample were analyzed, based on color existence (white, red, and blue); Three fragments of M2f were examined (two sides from “clean” fragment, two sides from “dirty” fragment, and two adjacent points from the fragment with unknown stain). From the M1d sample three adjacent points were also studied and for M2g sample two adjacent points. In addition fresh skin sample selected (from Dr. Boyatzis S. thumb) in order to be compared with the mummified tissue. The spectra were the sum of 20 scans collected from 4000-520 $\text{cm}^{-1}$  at a resolution of 4 $\text{cm}^{-1}$ .

FTIR analysis was used on the samples:

- M1b, M1c, M1h, M2e, M2f (fragments of the bandages)
- M1g (piece of balm)
- M1d (M1d $\mu$ ), M2g, M1f (M1f02) (tissue and hairs)
- M $\pi\delta$ 1 (control sample- fresh tissue from the Dr. Boyatzis, S. thumb)

Small pieces (0.5mg) of the samples were collected and enclosed in glass tubes (fig.16) with 1ml solvent (the type of the solvent was selected accordingly with the investigation interest of the material), (Table 6).

Then the tubes were put in Ultrasonic bath device (ELMASONIC S10H-0.8 lt) for 15 to 30'. Subsequently the solution was extracted by the use of glass pipette on the KBr (anhydrous potassium bromide) pellet (fig.17). For pellet preparation, 100mg of powder KBr were located in the metallic matrix then they were pressed in the hydraulic press at 10t/ $\text{cm}^2$  for 1'.

For the water soluble samples, centrifugation was performed for 2' at 1300 rpm (fig.18). Then the solution was extracted on gold pellet which was heated at 40°C until the evaporation (fig.19). Every pellet was examined at the PERKIN ELMER-spectrum GX FT-IR system Infrared spectrometer. The spectra were the sum of 20 scans collected from 4000 to 370 $\text{cm}^{-1}$  at a resolution of 4 $\text{cm}^{-1}$ .

Sample	Solvent			
	CH <sub>2</sub> Cl <sub>2</sub>	hexane	ethanol	distilled water
M1b	CH <sub>2</sub> Cl <sub>2</sub>			
M1c	CH <sub>2</sub> Cl <sub>2</sub>	hexane		
M1h	CH <sub>2</sub> Cl <sub>2</sub>	hexane		
M2e	CH <sub>2</sub> Cl <sub>2</sub>			
M2f	CH <sub>2</sub> Cl <sub>2</sub>			
M1g	CH <sub>2</sub> Cl <sub>2</sub>		ethanol	
M1d	CH <sub>2</sub> Cl <sub>2</sub>			distilled water
M1d <sub>μ</sub>				distilled water
M1f (M1f02)	CH <sub>2</sub> Cl <sub>2</sub>		ethanol	
M2g	CH <sub>2</sub> Cl <sub>2</sub>			distilled water
Mπδ1				distilled water

Table 6: For FTIR analysis different solvents it was used on the samples.



Fig.16: Small pieces of the samples were located on glass tubes.



Fig.17: The solution was extracted by the use of glass pipette on the KBr pellet.



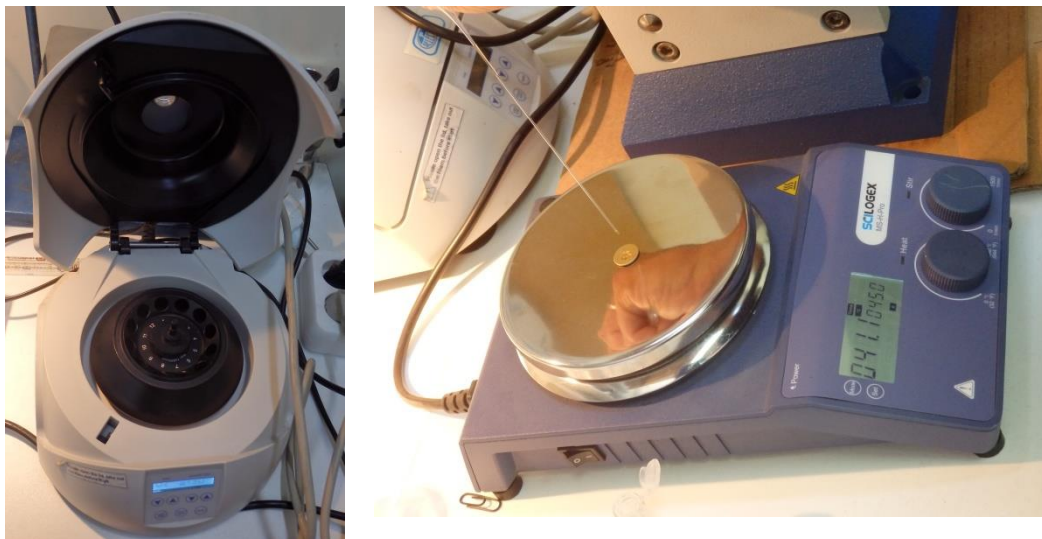


Fig.18-19: For water soluble samples centrifugation and heating was performed.

### 3.2.8 Portable X-Ray Fluorescence (XRF) analysis

XRF analysis was operated on possible gold residues which were retrieved from the surface of the mummified bodies for further investigation. Analysis was also performed on hair to compare with SEM-EDEX data.

Portable XRF detector (Bruker Tracer III SD) with S1PXRF software was used. The settings were as follows; beam diameter: 3mm, collection time: 60 sec., use of AL/Ti filter, accelerating voltage: 40kV and current intensity: 12 $\mu$ A. The operation was conducted in collaboration with Dr V. Kantarelou, Physicist, at the Anthropological museum of Athens. The code number of the spectra and the points of analysis are presented on the Tables 7 and 8.






<b>Mummy1</b>		
<b>Spectra number</b>	<b>Point of analysis</b>	<b>Photo</b>
1280	Right eyelid	
1281	Back side of the skull as control sample for hairs	
1282	hairs	

Table 7: The code numbers of spectra and the points of XRF analysis on Mummy1.





Mummy 2		
Spectra number	Point of analysis	Photo
1275 1276	Left side of the thorax between the leaf axil	
1277	At the middle of forehead	
1278	Right eyelid	
1279	Right foot	

Table 8: The code numbers of spectra and the points of XRF analysis on Mummy2.

### **3.2.9 Microbiological Investigation**

The investigation of microbiological contamination applied with observation of the samples under the optic microscope and the method of fungus cultivation. The samples M1.1, M1.2, M1.3 and M2.1 were hydrated by the use of lactophenol in order to be observed.

The cultivation method was applied by putting the samples in culture broth (sabourad) with chloramphainicol as antibiotic, under sterile conditions. The samples were preserved at room temperature without time limit. Subsequently the material was cultivated in three different solid materials (MEA, PCA, and V8).

The investigation was conducted by the Ass. Prof A. Velegraki, Microbiologist, at the Medical school, University of Athens.

## 4. Results

### 4.1 Visual examination

#### 4.1.1 Mummy 1(M1) (fig.20)

The measurements of M1 are (Table 9):

	<b>General length</b>	<b>Length of arms</b>	<b>Length of legs</b>	<b>Width of shoulders</b>	<b>Width of hips</b>	<b>Width of feet</b>
Mummy1	~101 cm	~40 cm	~50cm	~19cm	~18cm	~15cm

Table 9

M1 is childhood age individual. The body is fairly complete. The posture is presented to have a slope to the right. Both hands lie straight along the torso. The mummy is broken almost in half at the point of the thighs. The bandages have removed inappropriately. The exposed parts of the body include the points from the head until the arms, the abdomen and parts of the feet. The teeth and part of scalp hairs is preserved (fig.21-22). Residues of possible gold are observed on the right side of the eyelid and the right side of the nose (fig.23). Parts of both thighs bones and a small part of the scull bone are visible. Missing and fragmented parts of the flesh are observed from the left side of the burst until the abdomen, where filling materials are exposed (fig.24).

The preserved bandages are around of both legs whereas damaged bandages are peripherally from the shoulders until the thighs (fig.25-26). At the point from the burst until the abdomen there is wrapped fabric, vertically located on the body (fig.27). On the wrapped fabric there is a tag made of thick paper in bad preservation state. Color traces (blue and red) and fragments of plaster are also presented on the fabric. Black areas cover part of the fabric too (fig.28).

Cracking of the skin is observed in almost exposed parts of the body, particularly on the head (fig.29), at the forehead, around the eyes and the cheeks, on the thorax (fig.30) and the abdomen. Whitish spots cover almost all the body's surface whereas (fig.31-32) white spots are observed on the filling materials (fig.33-34) and the wrapped fabric. Holes and bores are visible in several areas of the body (fig.35-36), specifically at the face, the thorax, the

arms, the feet and the lower side of the damaged bandages. Dust and remnants of degradation are all over the body and the holes.



Fig.20: Mummy1-front view.



Fig.21-22: Detail of the head and the face. The scalp hairs and the teeth are preserved.



Fig.23: Detail of the face. Possible gold residues on the right eyelid and the nose.



Fig.24: Detail of the abdomen. Degraded filling materials.





Fig.25: Both legs are covered of the bandages.



Fig.26: Detail of the left shoulder. Damaged bandages around the body.



Fig.27: The wrapped fabric vertically located on the body. Color traces, fragments of plaster and black areas on the wrapped fabric..



Fig.28: The paper tag on the wrapped fabric.



Fig.29: Detail of the face. Cracking of the skin.



Fig.30: Detail of the thorax. Cracking and gaps of the flesh.





Fig.31-32: Detail of the left ear and the right arm. Whitish spots cover the skin surface.



Fig.33-34: Detail of the filling materials. Deposits of white spots.



Fig.35-36: Detail of the right arm and the left foot. Holes and bores.

#### 4.1.2 Mummy 2(M2)(fig.37)

The measurements of M2 are (Table 10):

	<b>General length</b>	<b>Length of arms</b>	<b>Length of legs</b>	<b>Width of shoulders</b>	<b>Width of hips</b>	<b>Width of feet</b>
<b>Mummy2</b>	~83cm	~32cm	~38cm	~17cm	~15cm	~6cm

Table 10

M2 is a male child individual. The mummy lies on a wooden board on a red fabric. The body is fairly complete. Both hands lie straight along the torso. The bandages have removed. The exposed parts of the body include the head, the points from the shoulders until the pelvis, the right foot and part of the left foot. The eyelashes and the scalp hair are preserved (fig.38-39). Two parts of the thorax, the right hand and the right foot are broken (fig.40-41). Missing parts of the body are observed at the abdomen where filling materials are exposed (fig.42). Parts of the ribs and part of the right side of the pelvis bone are visible. A thin layer of bandages covers the points from the left elbow until the left hand, the right hand (fig.43) and both legs (fig.44). Damaged bandages are preserved peripherally of the body. Possible gold residues are detected at the face specifically on the eyelids, the eyelashes and the nose, the left shoulder in the vicinity of the axil, the two broken pieces of the thorax, the right foot and the left toe (fig.45).

Cracking of the skin is observed in almost all exposed parts of the body, particularly the head, under the neck, the shoulders, the left arm and hand, the thorax and the pelvis (fig.46-47). Whitish spots are detected particularly, on the face, the shoulders, the arms, the thorax and the pelvis (fig.48) whereas white spots are seated in the thorax cavity (fig.49). Holes and bores are in several areas of the body notably the face, the right hand, the pelvis, the legs, the feet and the filling materials (fig.50-51.). At the end, dust and residual materials are all over the mummy.



Fig.37: Mummy2, front view.





Fig.38: Detail of M1 head. The eyelashes are preserved.

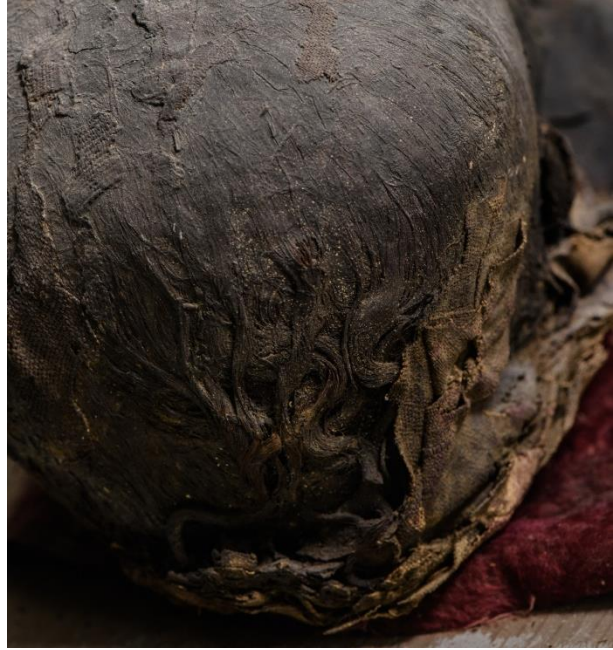


Fig.39: Detail of the back side of the head. The hairs are preserved.



Fig.40: Detail of the broken parts of the thorax.



Fig.41: Detail of the broken right foot.



Fig.42: Detail of the left side of the abdomen. Filling materials are exposed.



Fig.43: Detail of the right hand. The hand is covered with a thin layer of bandages



Fig.44: Detail of the legs covered with a thin layer of bandages.



Fig.45: Detail of the feet. Possible gold residues.





Fig.46: Detail of the face and the thorax. Cracking of the skins, gaps and broken parts of the body.



Fig.47: Detail of the left side of the head. Cracking of the skin.



Fig.48: Detail of the penis. Whitish spots on the skin.



Fig.49: Detail of the thorax cavity. White spots.



Fig.50: Detail of the left leg. Holes and bores at the skin and the bandages.



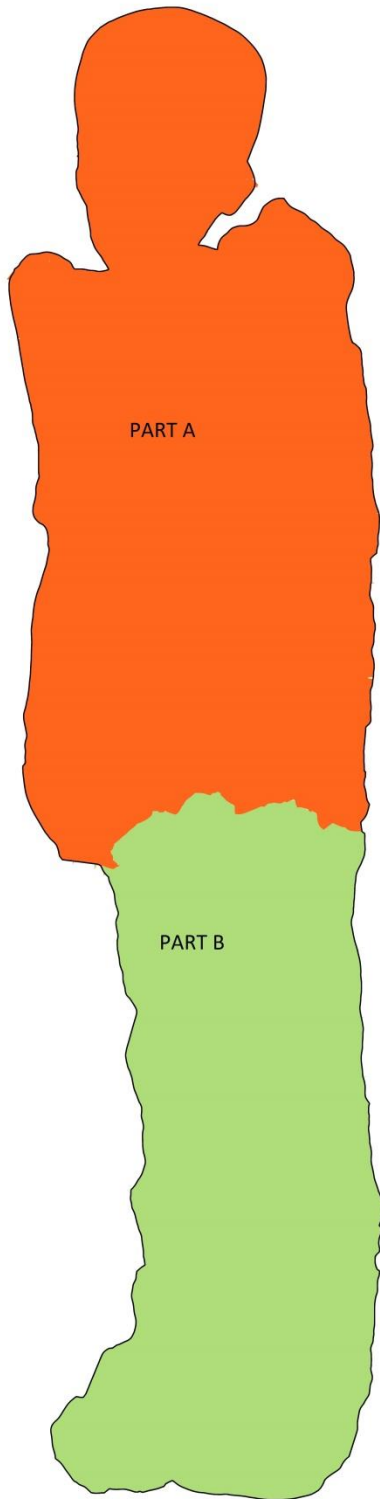
Fig.51: Detail of the feet. Holes and bores at the skin.

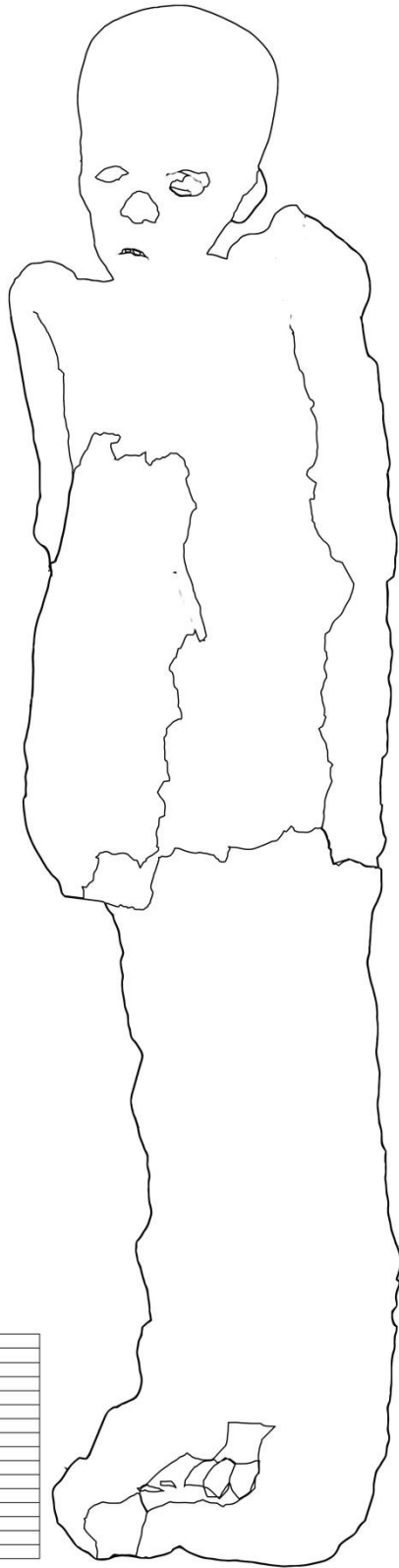


## 4.2 Deterioration ASPECTS

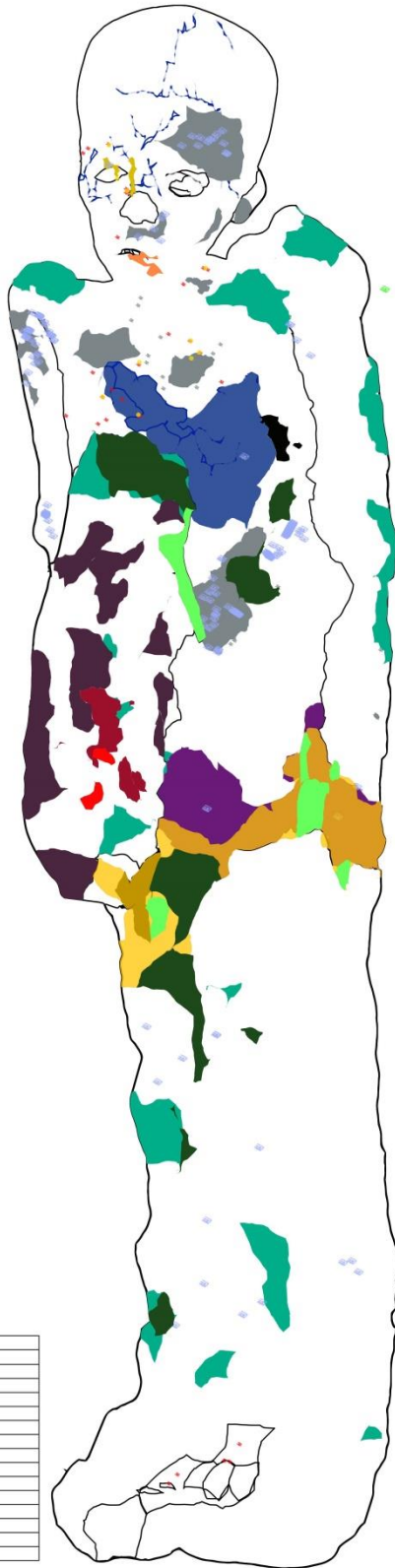
### 4.2.1 Mummy1



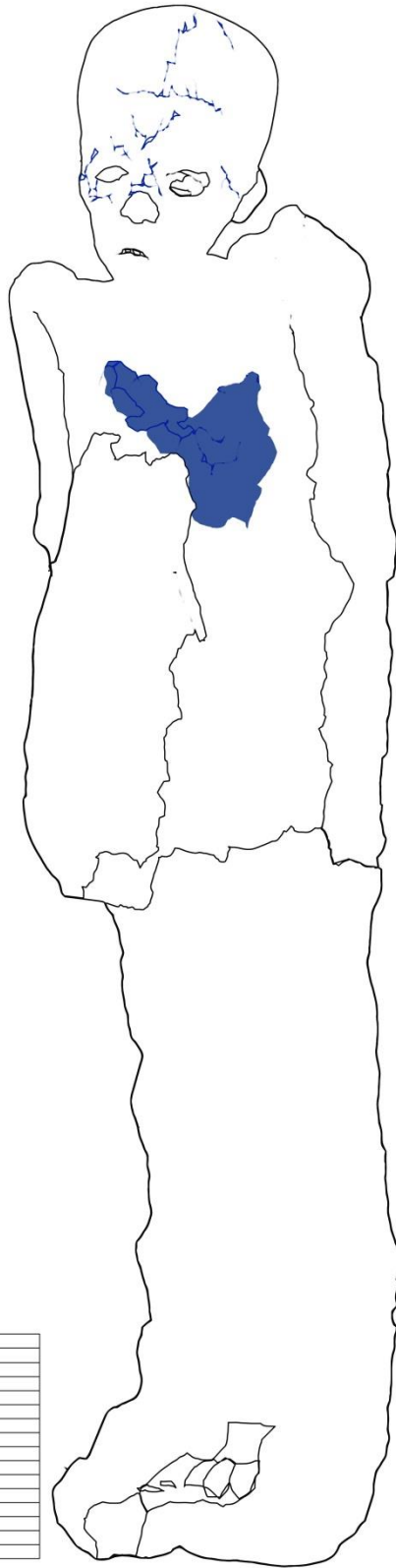




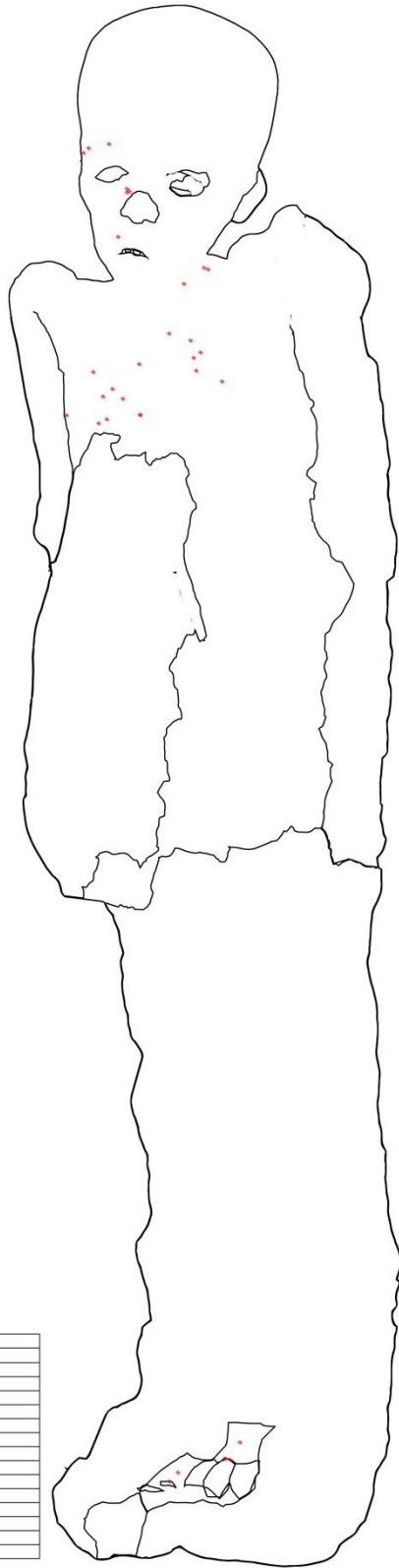
	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bones.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.



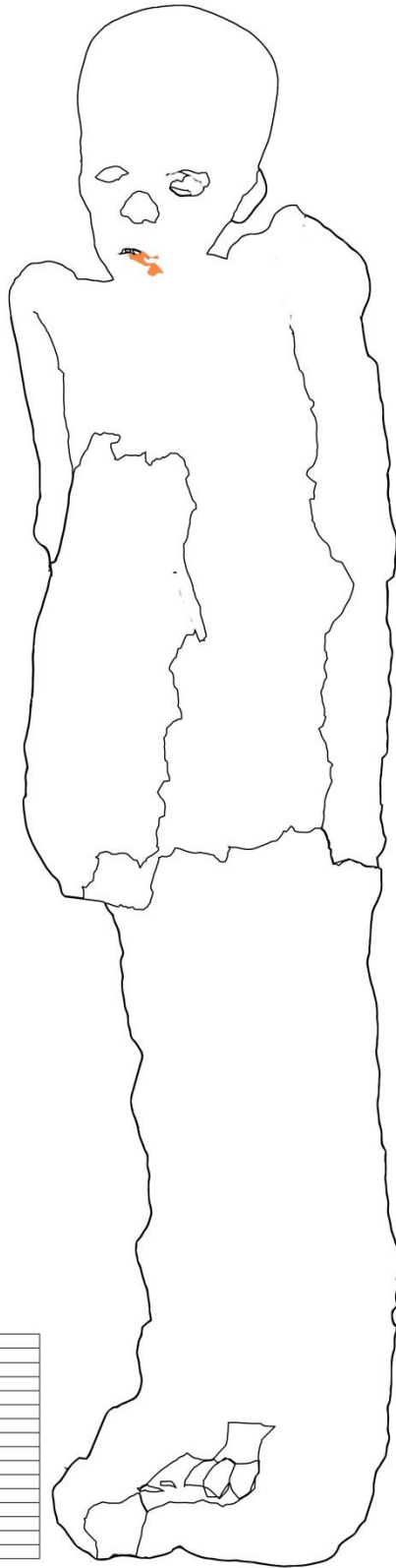
	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.



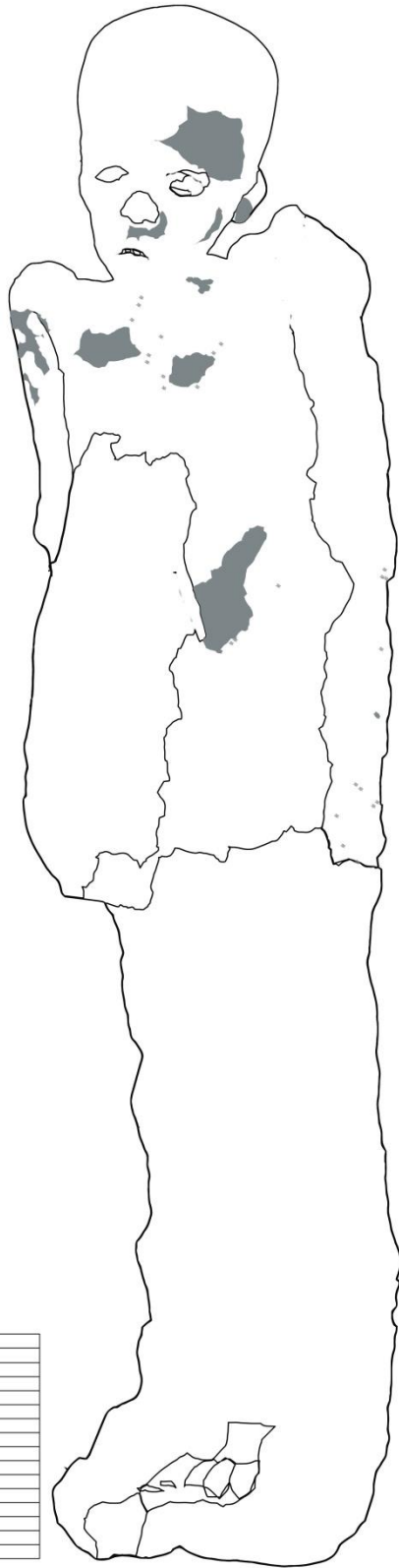
	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bones.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.



	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bones.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.

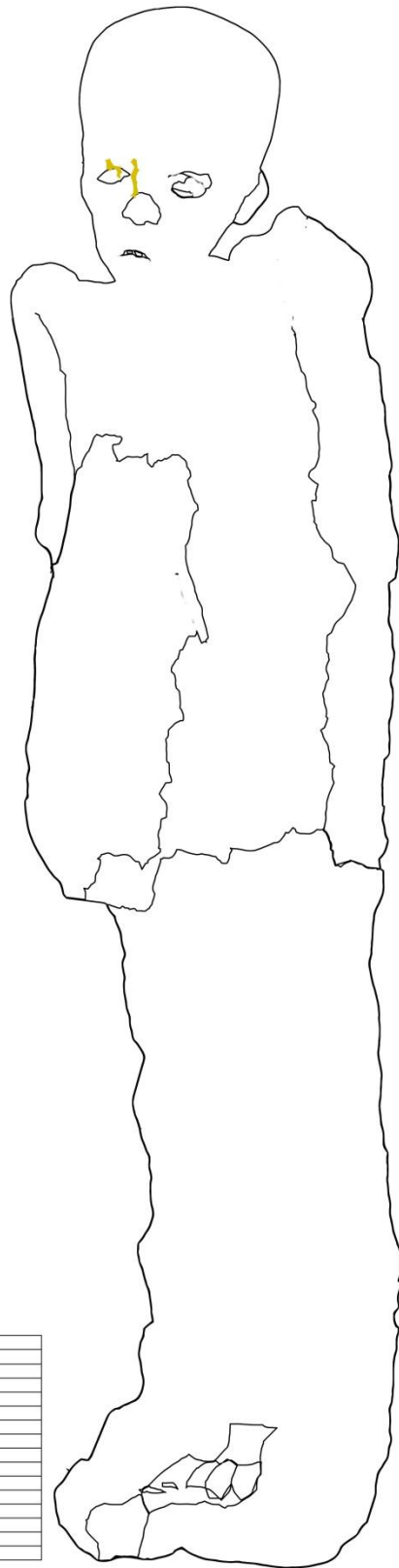


	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin
	Holes and bones
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.

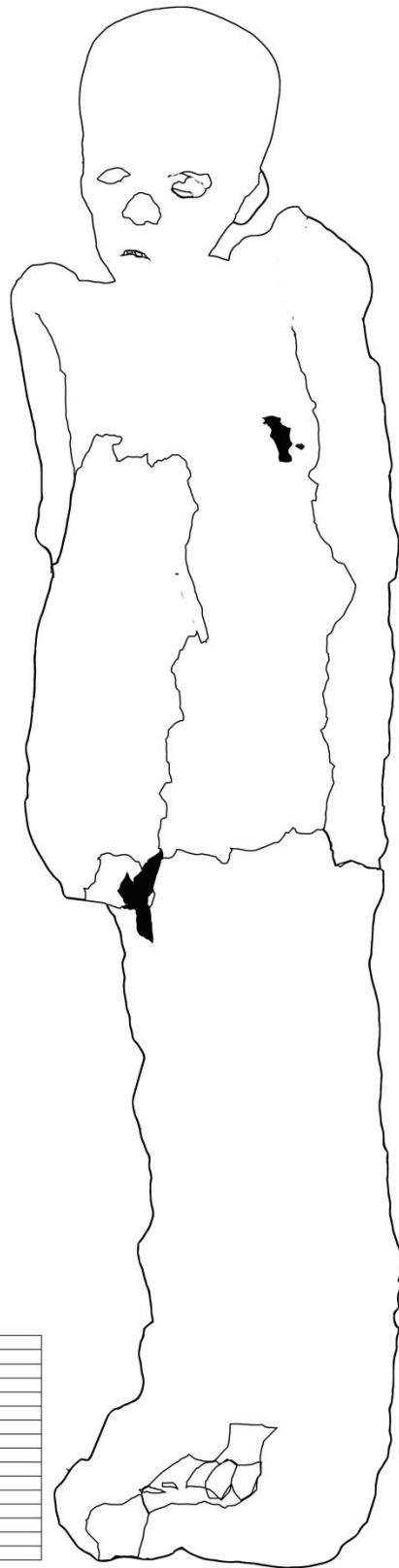


	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bones.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.

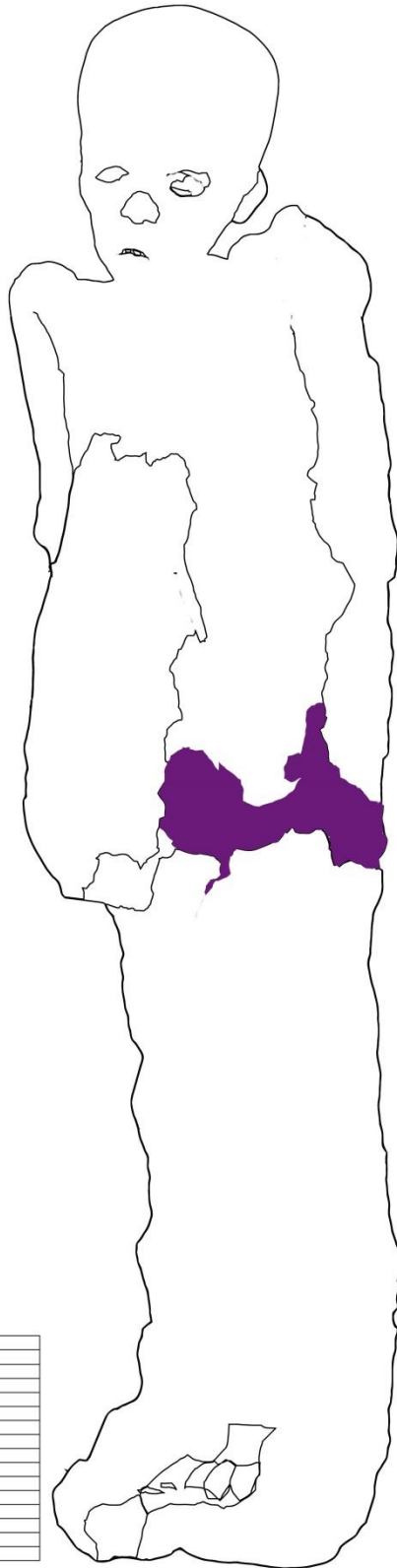




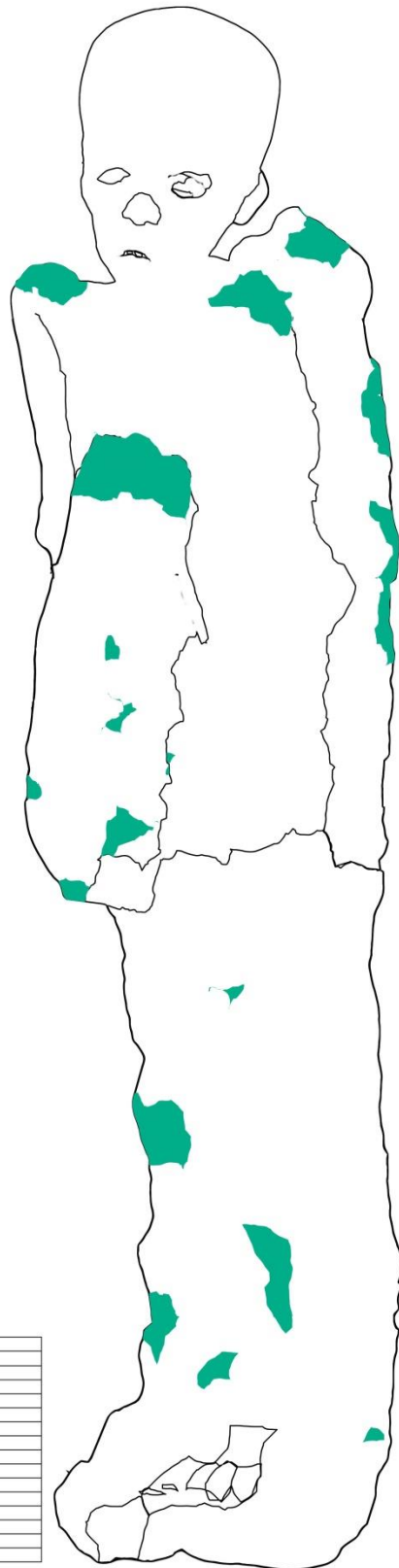
	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.



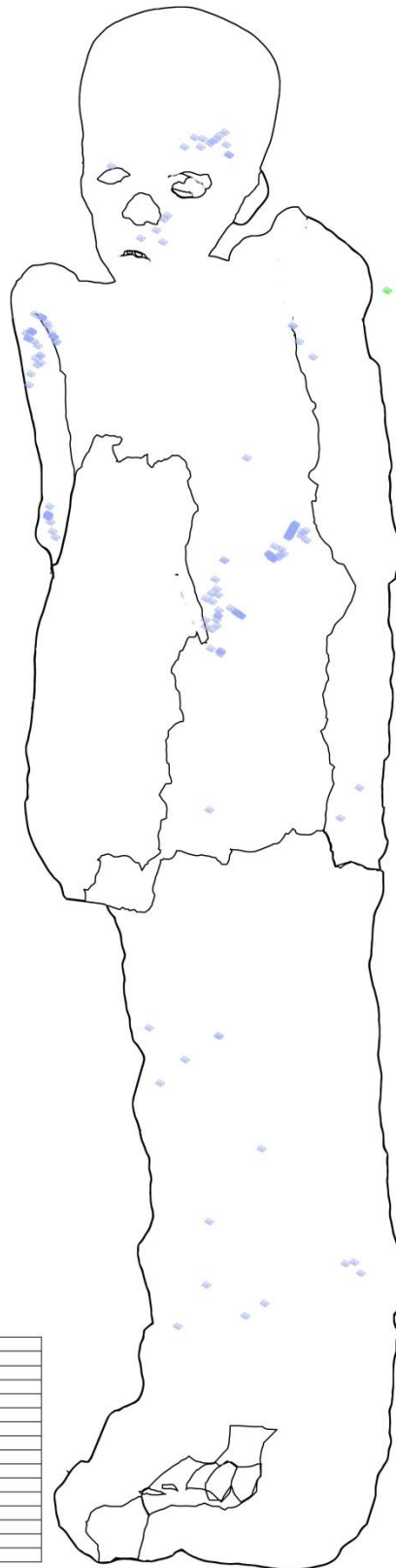
	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.



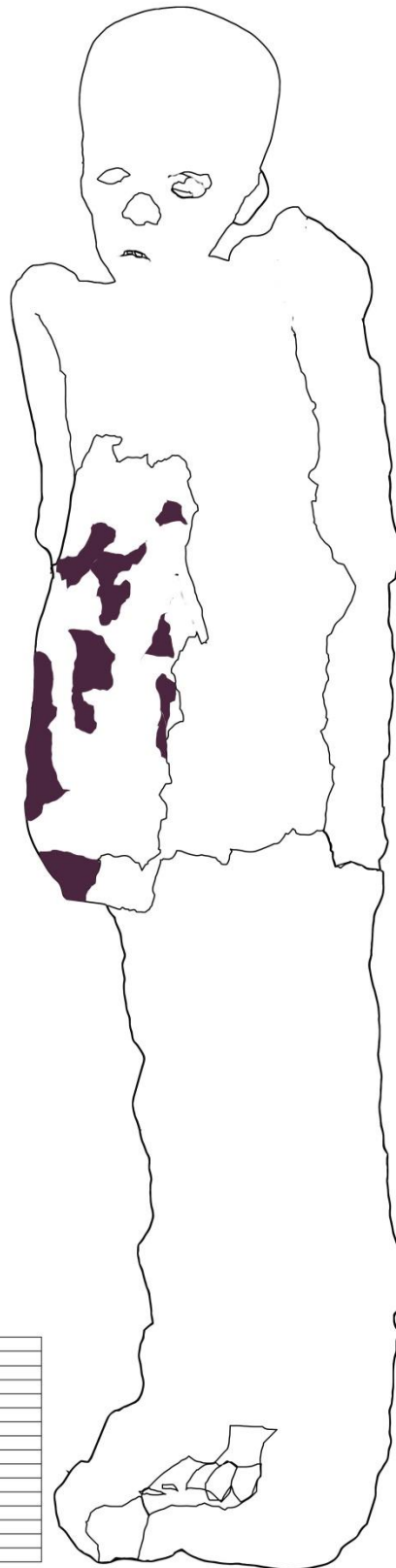
	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bones.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.



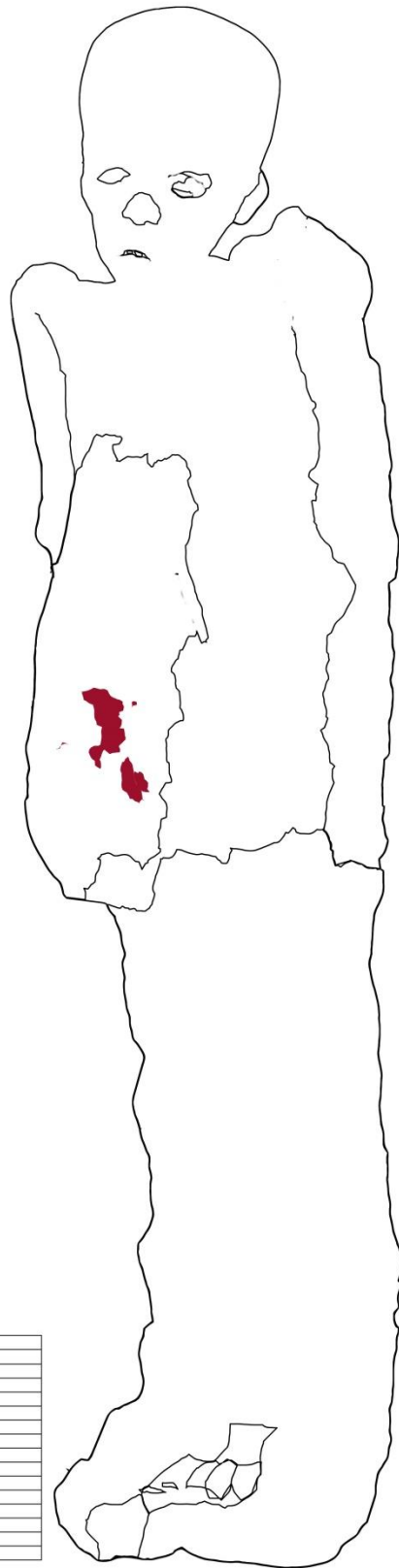
	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.



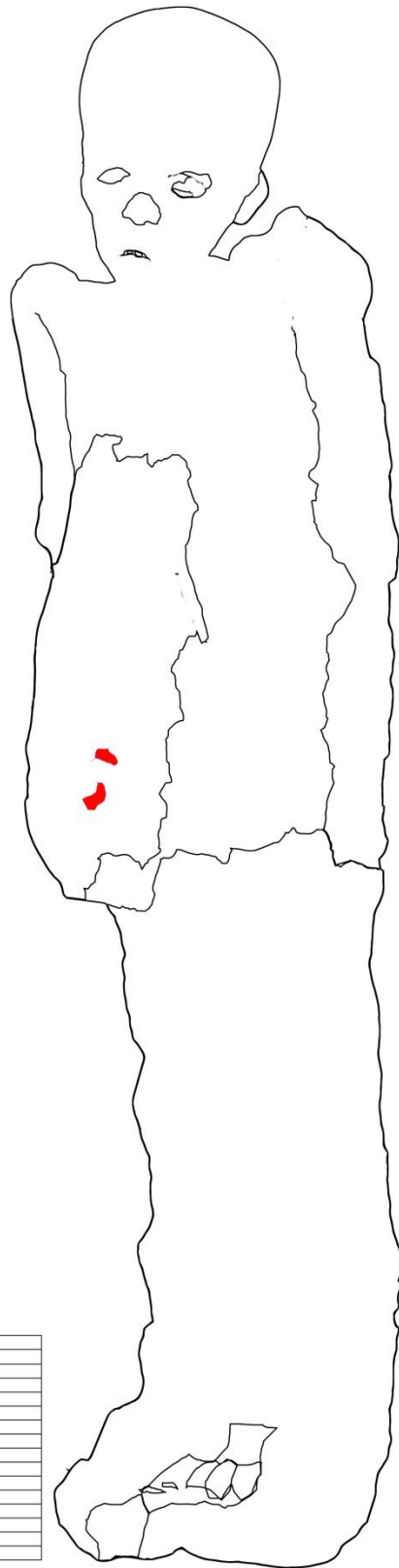
	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.



	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.

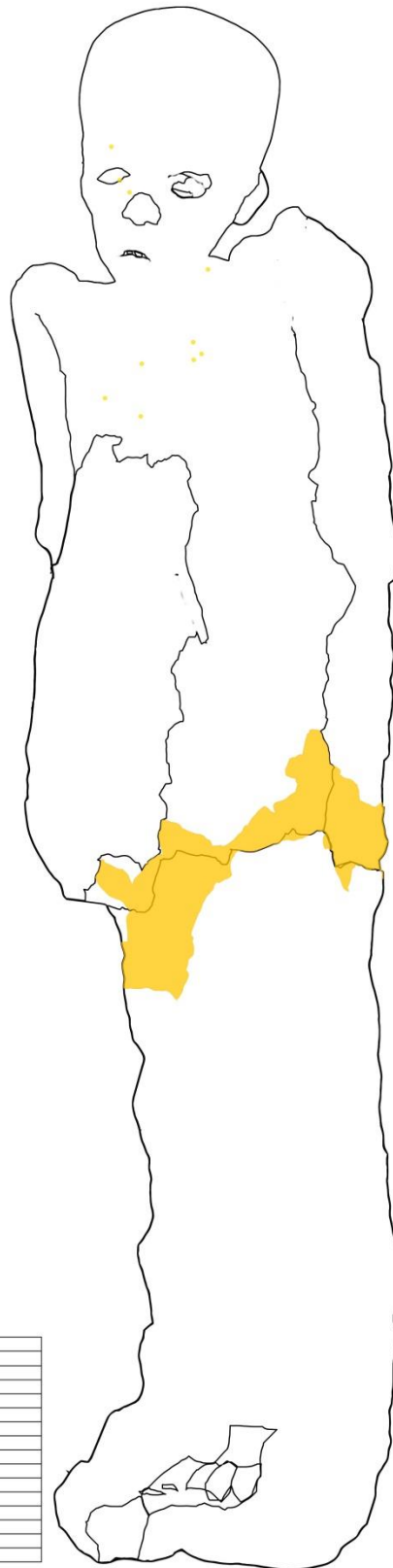


	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.

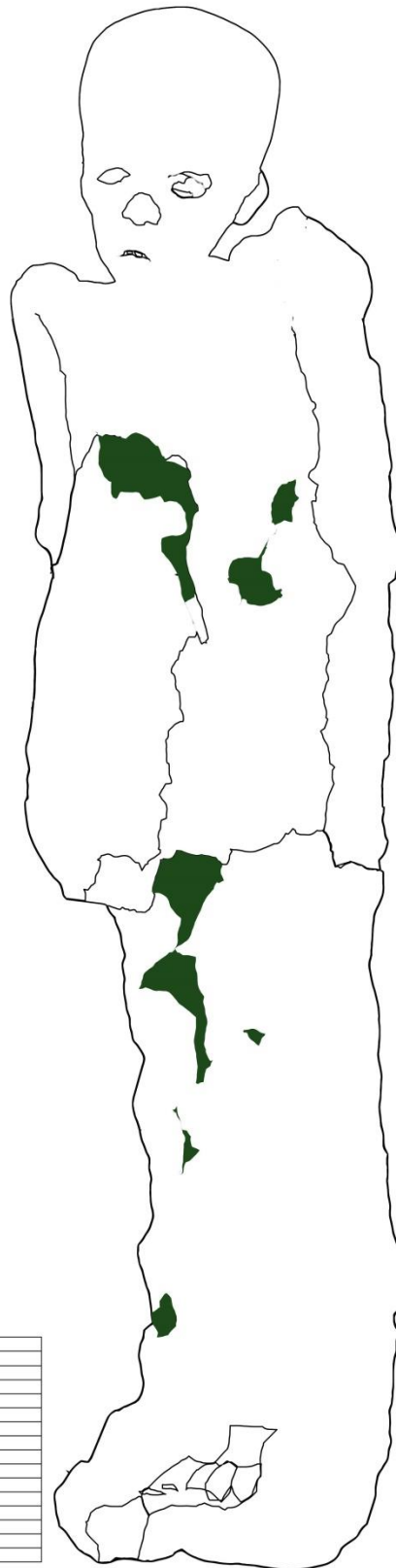


	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.

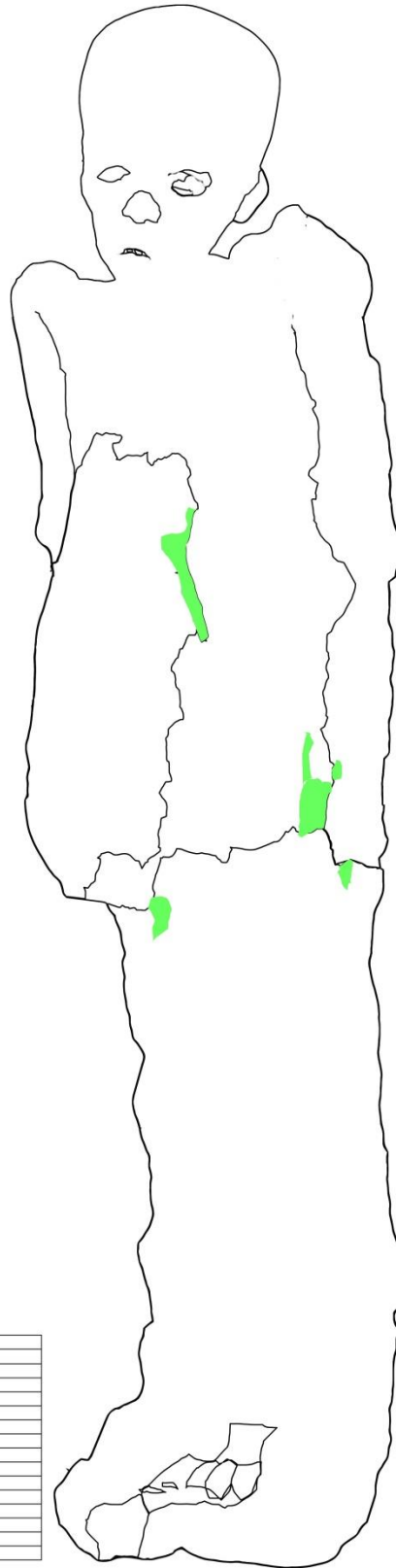




	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.



	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.

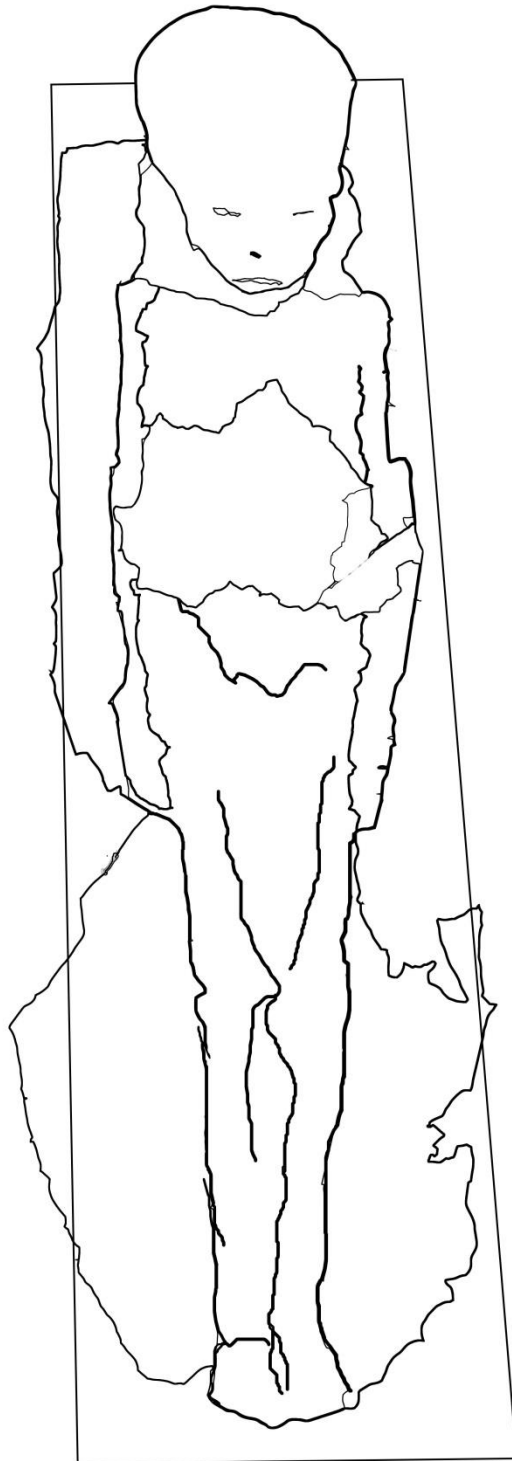









	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps.
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.

	Preserved teeth, scalp hair and eyelashes.
	Cracking of the skin.
	Holes and bores.
	Residual of the balms.
	Whitish spots.
	Gold residues.
	Gaps
	Missing parts of the flesh.
	Damage bandages.
	White spots.
	Black spots.
	Parts of plaster.
	Color traces.
	Dust or remnants of degradation.
	Broken parts of bandages.
	Broken bones.

#### 4.2.1 Mummy2

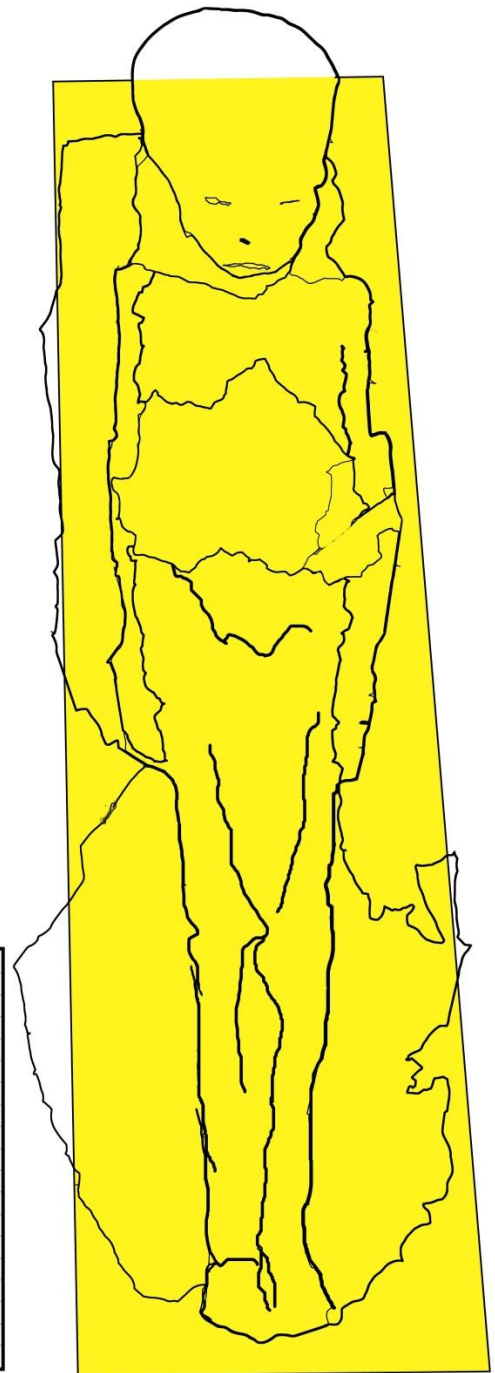






















	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair

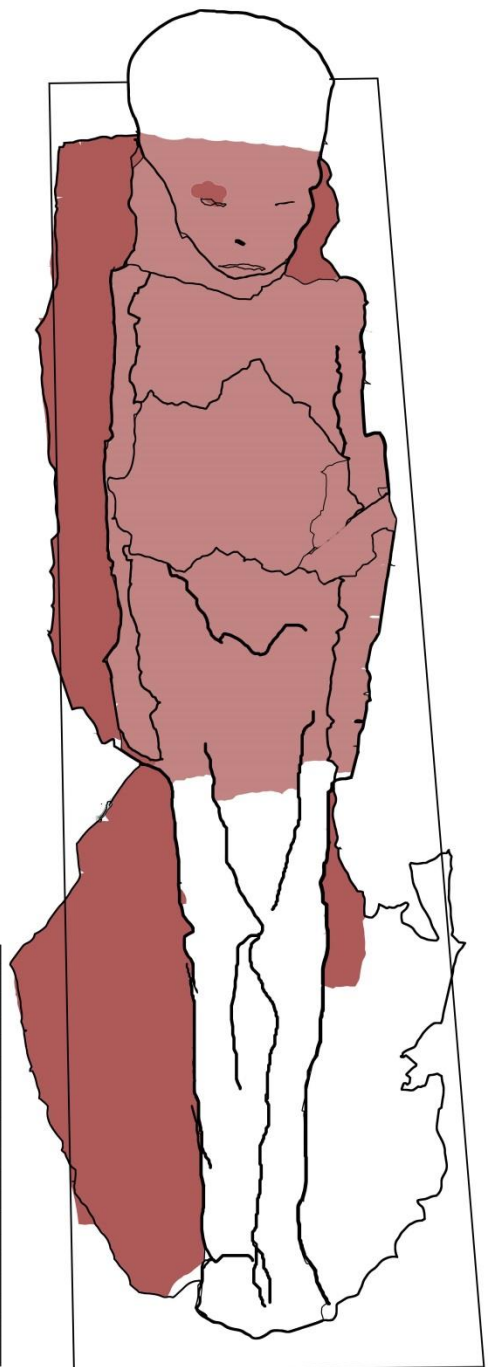




















Yellow	Wooden board
Red	Red textile
Dark red	Visible flesh
Green	Preserved bandages
Red	Broken parts of the body
Light green	Missing parts of the body
Grey	Exposed bones
Light grey	Exposed filling materials
Pink	Fragments of bandages
Brown	Gold residues
Blue	Cracking of the skin
Teal	Whitish spots
Black	Gaps
Dark grey	Dust, residual material, cobweb
Dark green	Degraded filling material
Magenta	Holes and bores
Dark blue	Damaged bandages
White	Preserved eyelashes and scalp hair

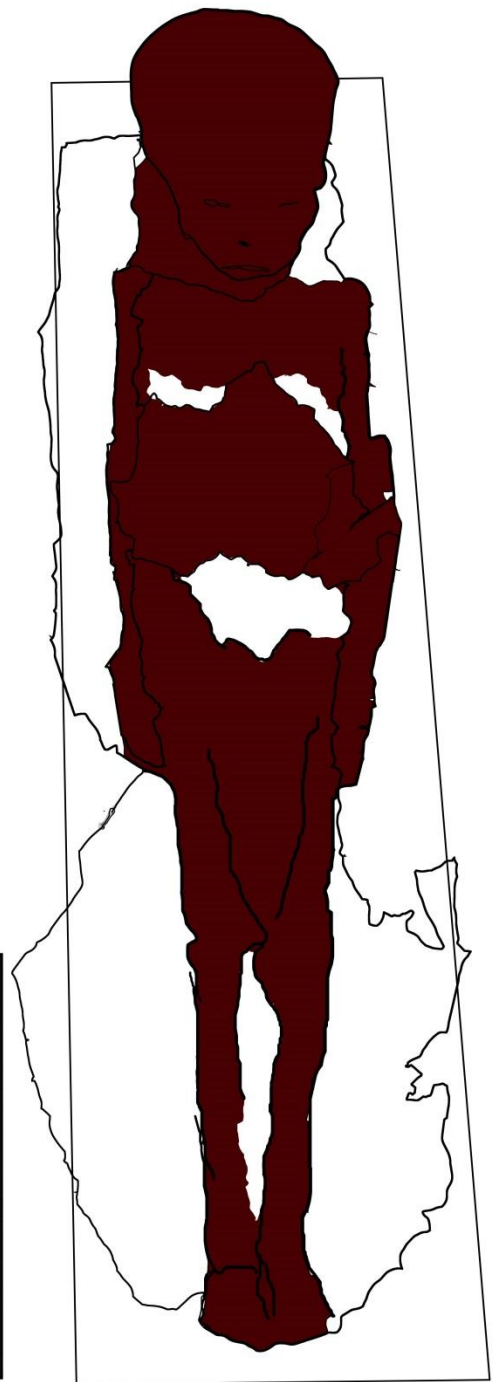





















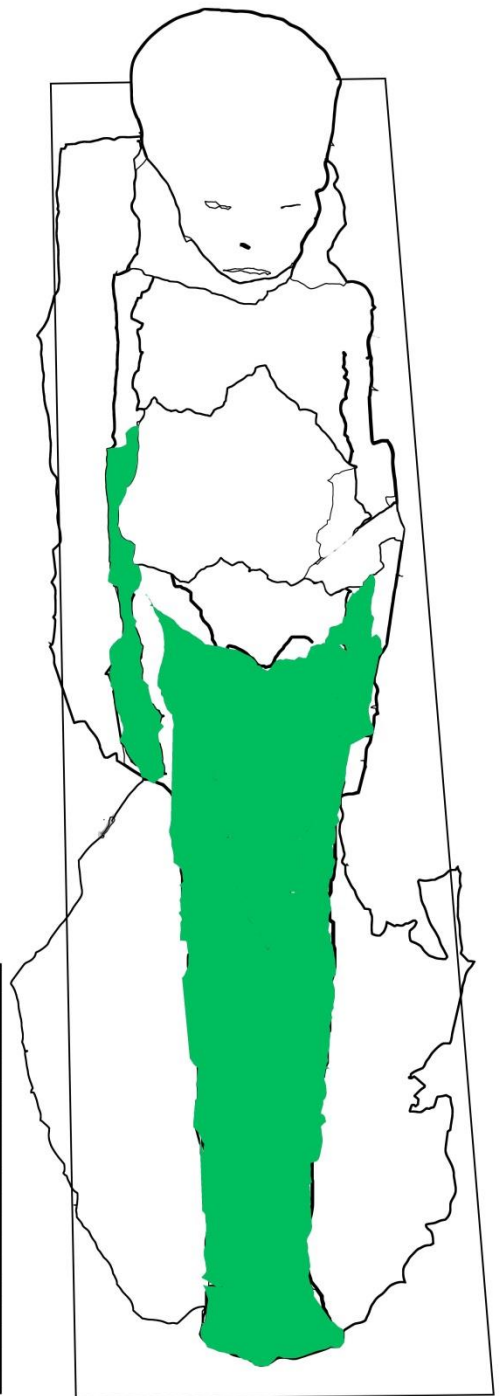
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair





















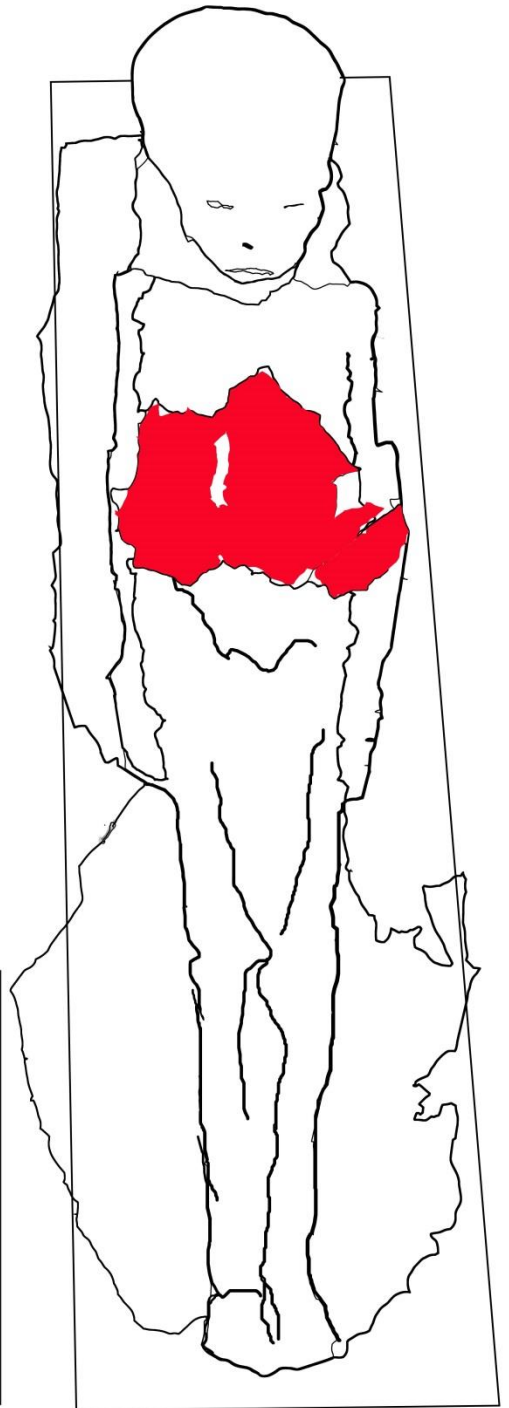
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair





















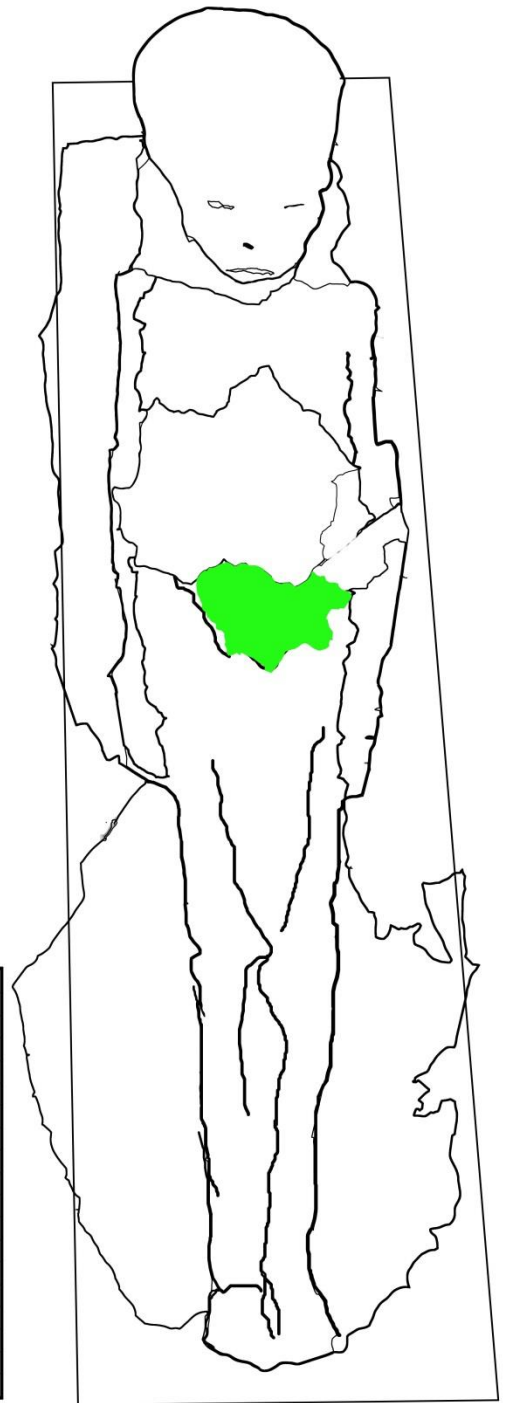
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair



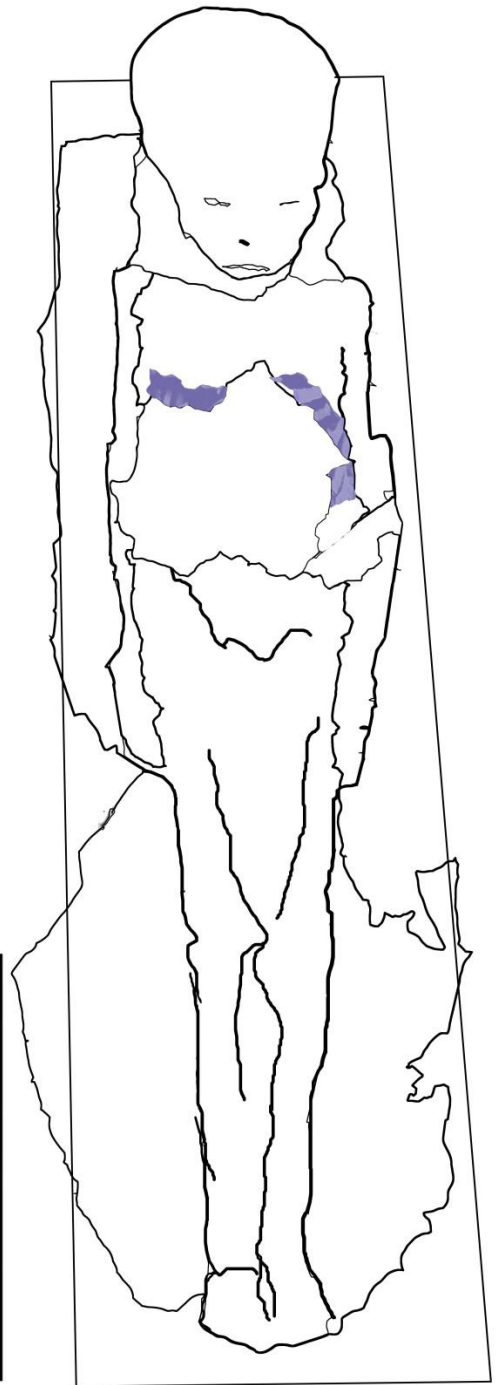
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair




















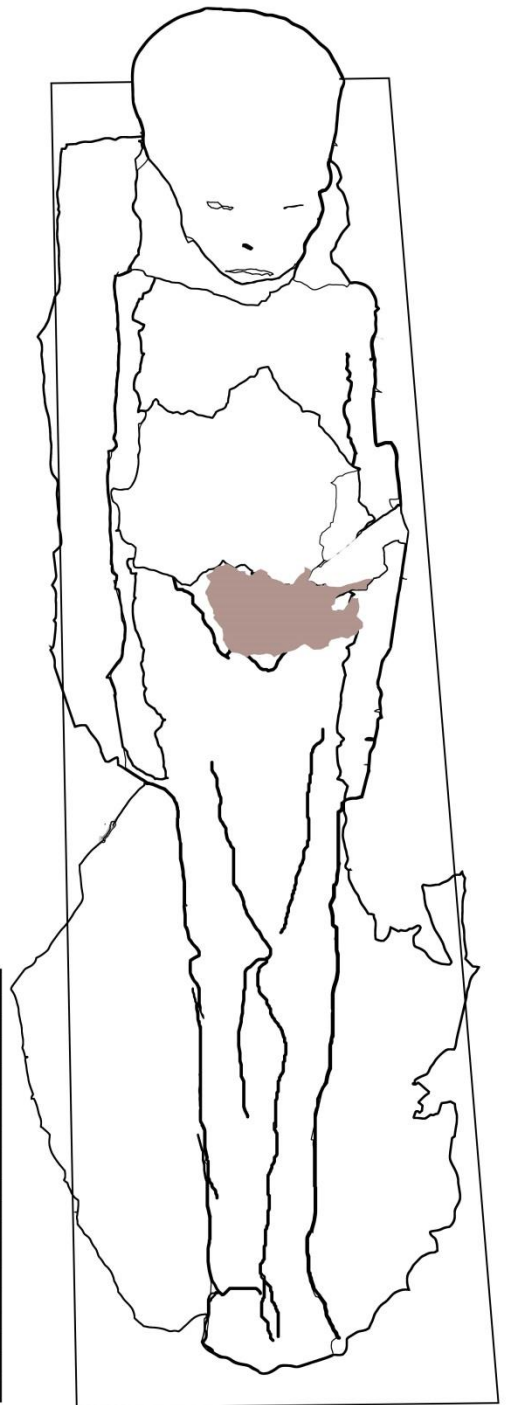
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair












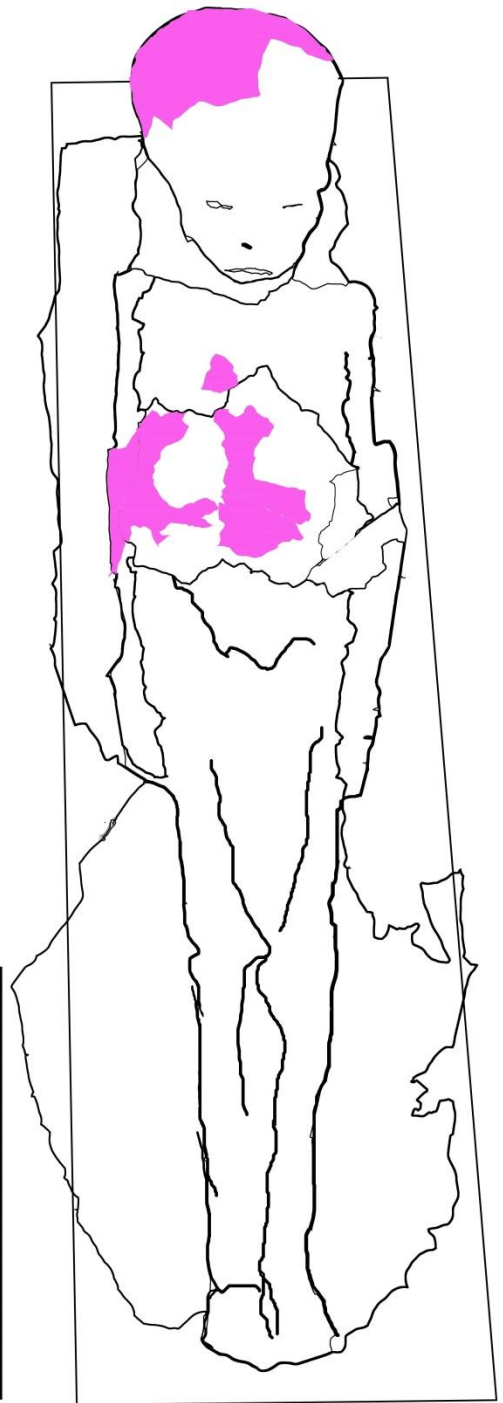
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair





















	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair

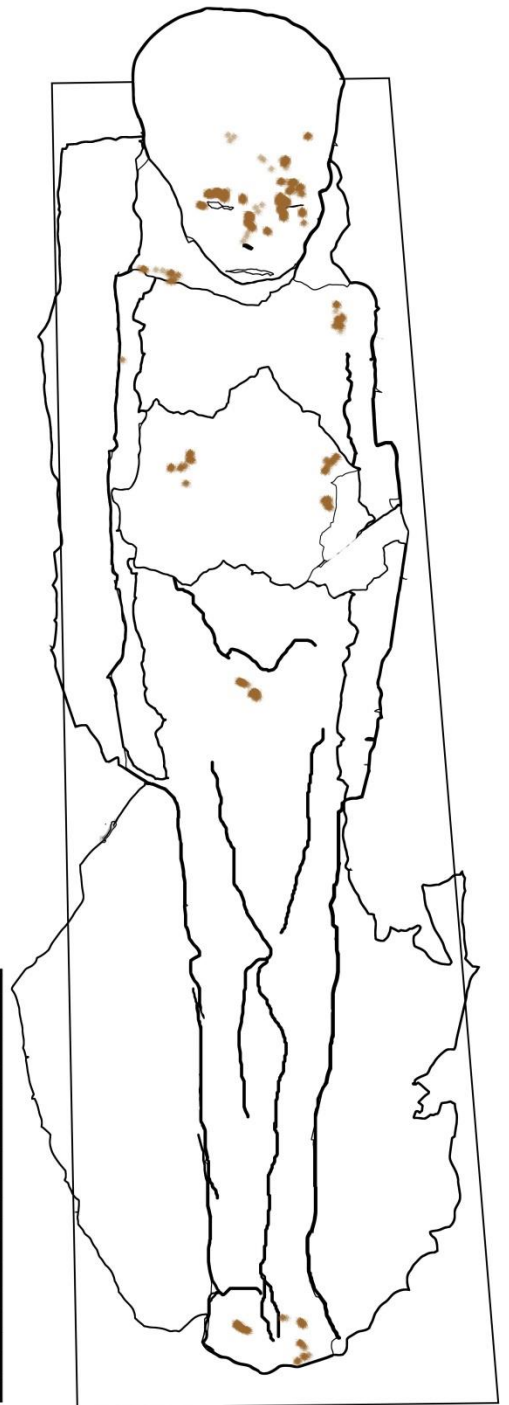



	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair

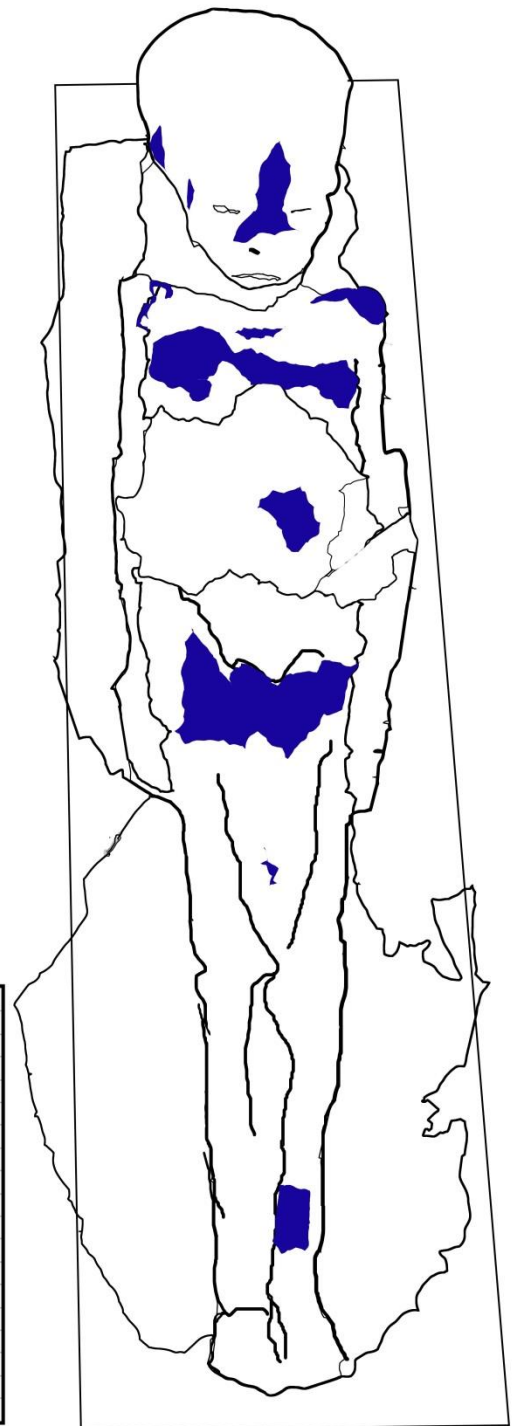




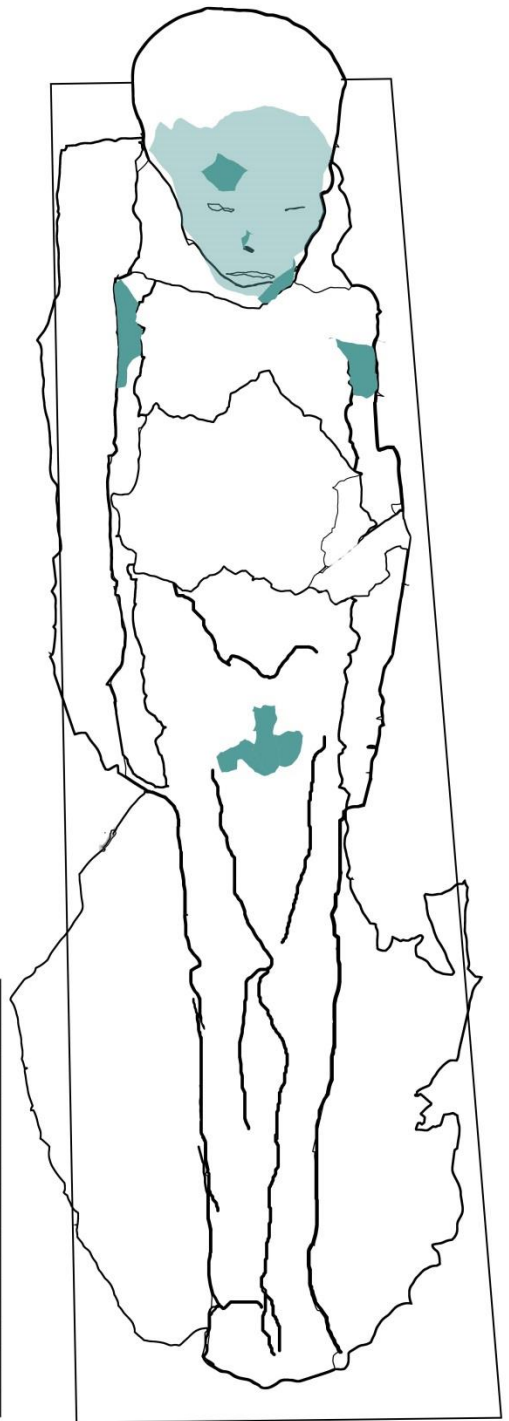
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair




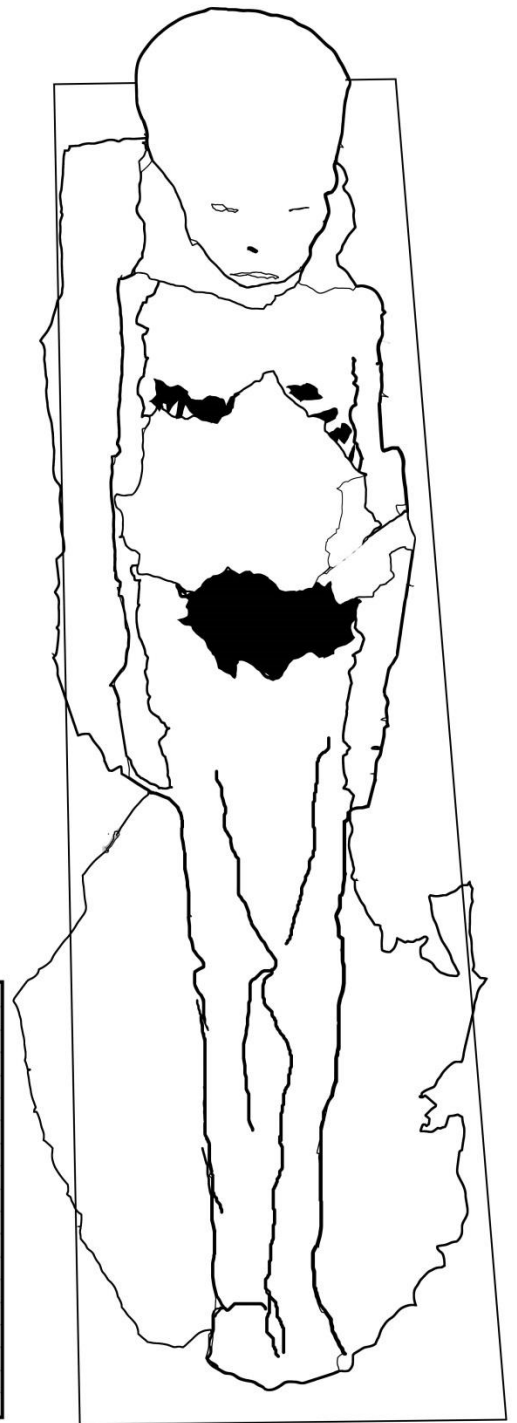
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair





















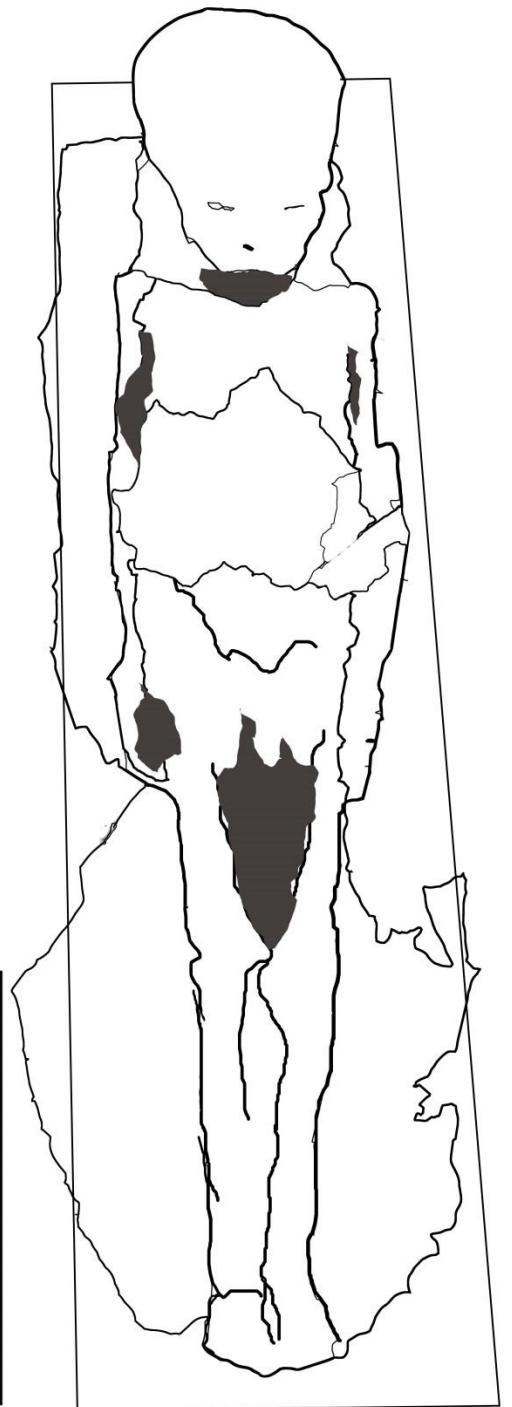
Yellow	Wooden board
Red	Red textile
Brown	Visible flesh
Green	Preserved bandages
Red	Broken parts of the body
Light green	Missing parts of the body
Light blue	Exposed bones
Light purple	Exposed filling materials
Pink	Fragments of bandages
Brown	Gold residues
Dark blue	Cracking of the skin
Light blue	Whitish spots
Black	Gaps
Grey	Dust, residual material, cobweb
Dark green	Degraded filling material
Pink	Holes and bores
Purple	Damaged bandages
White	Preserved eyelashes and scalp hair





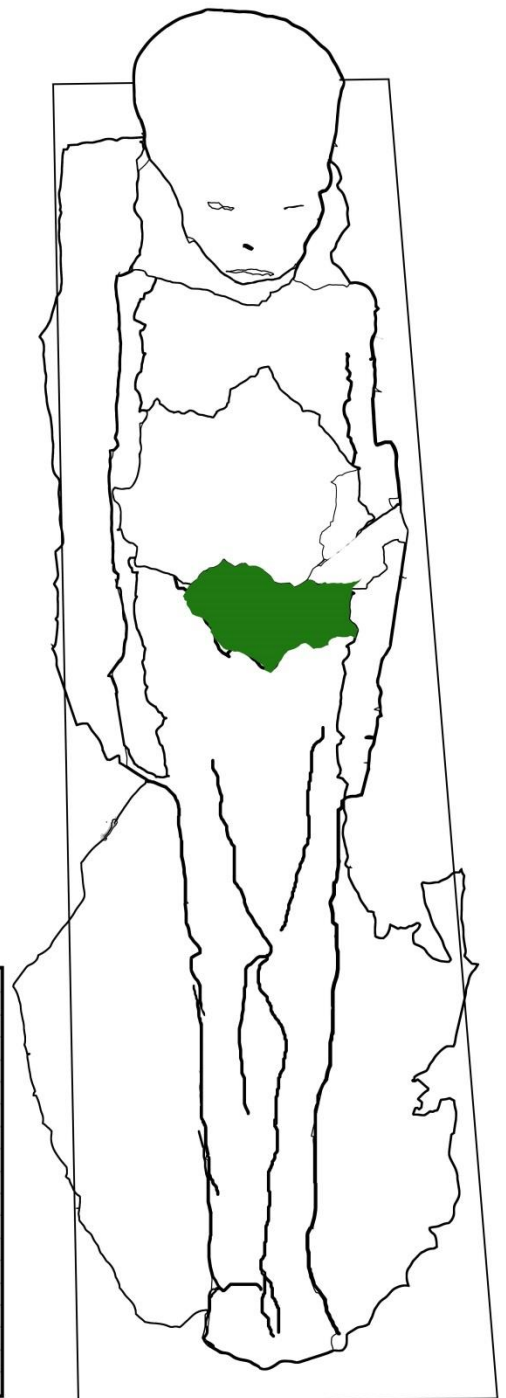
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair




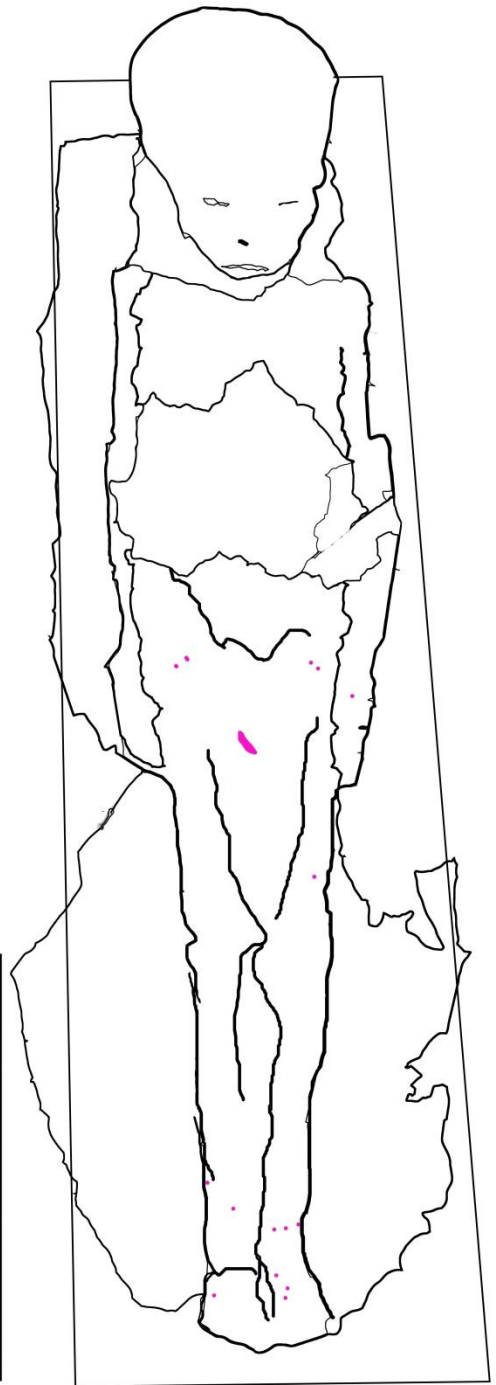
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair



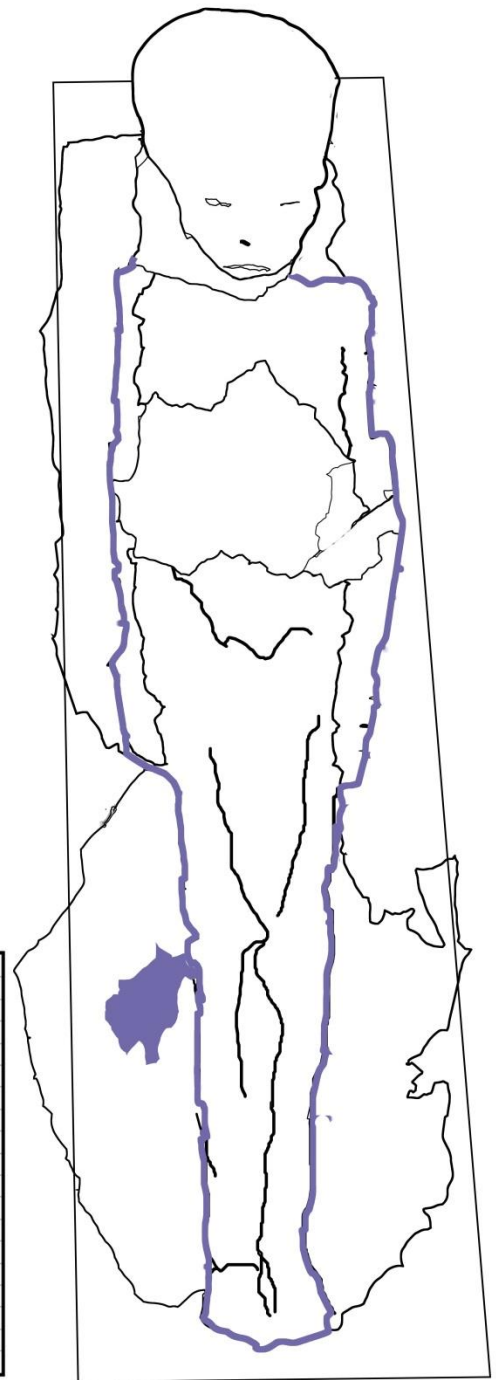
	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair



	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair

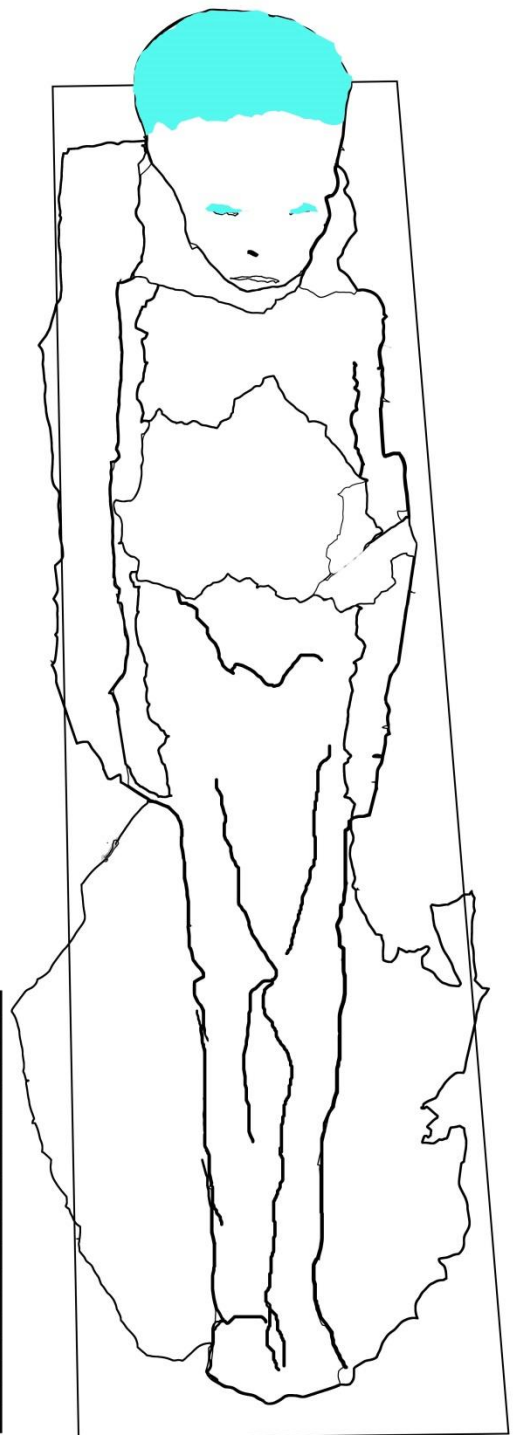




















Yellow	Wooden board
Red	Red textile
Brown	Visible flesh
Green	Preserved bandages
Red	Broken parts of the body
Light green	Missing parts of the body
Grey	Exposed bones
Light blue	Exposed filling materials
Pink	Fragments of bandages
Brown	Gold residues
Dark blue	Cracking of the skin
Light blue	Whitish spots
Black	Gaps
Dark grey	Dust, residual material, cobweb
Dark green	Degraded filling material
Pink	Holes and bores
Dark blue	Damaged bandages
White	Preserved eyelashes and scalp hair




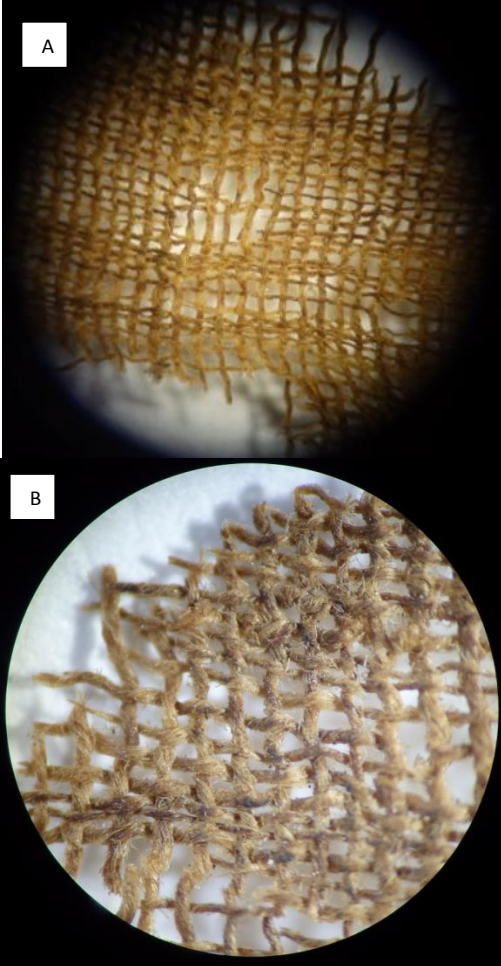


Yellow	Wooden board
Red	Red textile
Dark red	Visible flesh
Green	Preserved bandages
Red	Broken parts of the body
Light green	Missing parts of the body
Grey	Exposed bones
Light brown	Exposed filling materials
Pink	Fragments of bandages
Gold	Gold residues
Blue	Cracking of the skin
Light blue	Whitish spots
Black	Gaps
Dark grey	Dust, residual material, cobweb
Dark green	Degraded filling material
Magenta	Holes and bores
Light purple	Damaged bandages
Cyan	Preserved eyelashes and scalp hair



	Wooden board
	Red textile
	Visible flesh
	Preserved bandages
	Broken parts of the body
	Missing parts of the body
	Exposed bones
	Exposed filling materials
	Fragments of bandages
	Gold residues
	Cracking of the skin
	Whitish spots
	Gaps
	Dust, residual material, cobweb
	Degraded filling material
	Holes and bores
	Damaged bandages
	Preserved eyelashes and scalp hair

### 4.3 Macroscopic and Stereoscopic examination of the samples

M1 samples			
Code name	Macro-image	Stereoscopic image	Description
M1a			<p><b>Fragment of bandages:</b>  <b>Spinning:</b> monofilament S twisting direction  <b>Weave:</b> sparse, simple 1x1  <b>Number of warp and /or weft:</b> ~17/30 per cm<sup>2</sup></p> <p>The fabric preserves its natural properties. Wear and tear, partial loss of the threads and weakening of the weave is observed (A); Dark brown particles and impurities are detected between the threads. Some of these are presented to be in dark brown color (B).</p> <p>A. MAG: x0.6          B. MAG: x2</p>

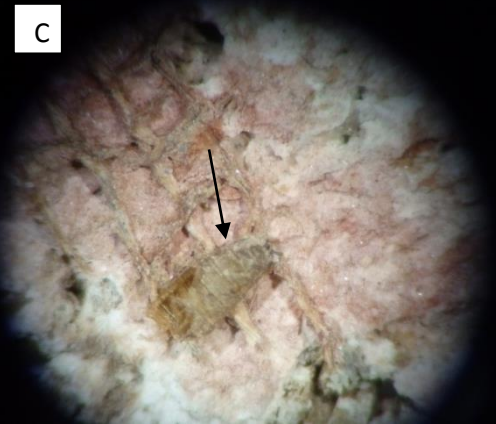
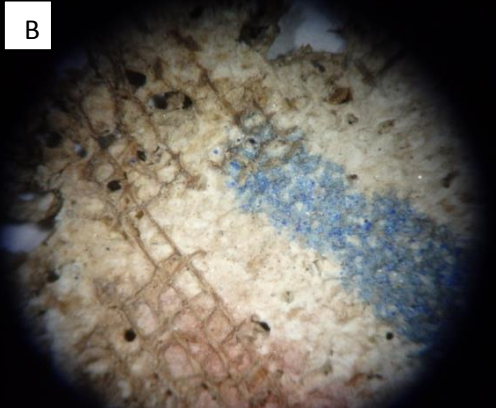
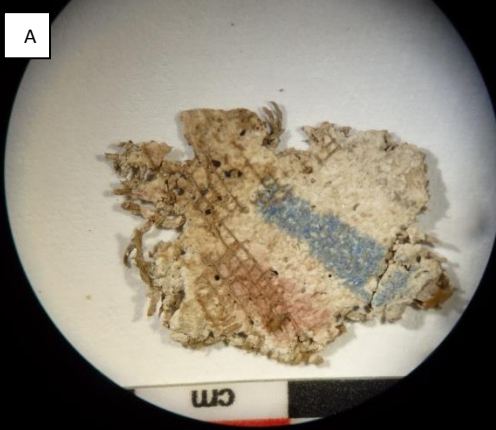
M1b



Front side of M1b



Back side of M1b



**Fragment of bandages:**

**Front side:**

The fragment is consisted from textile and possible plaster in white-beige color. On the plaster are distinguished two bands in blue and red color (A). On the red band are preserved the remnants of another layer of textile (B). Cracking and brittleness of the plaster and the colored areas is observed. Possible holes are detected too. On the red area an insect residue is determined (C, D).

**Back side:**

**Spinning:** monofilament S twisting direction

**Weave:** tight, simple 1x1

**Number of warp and /or weft:** ~10/17 per cm<sup>2</sup>

Partial wear and tear and loss of the threads is observed. Holes are visible. At the middle of the sample the weaving is appeared to be convoluted (F). Several impurities and particles of the plaster and color pigments are seated on the threads (E).

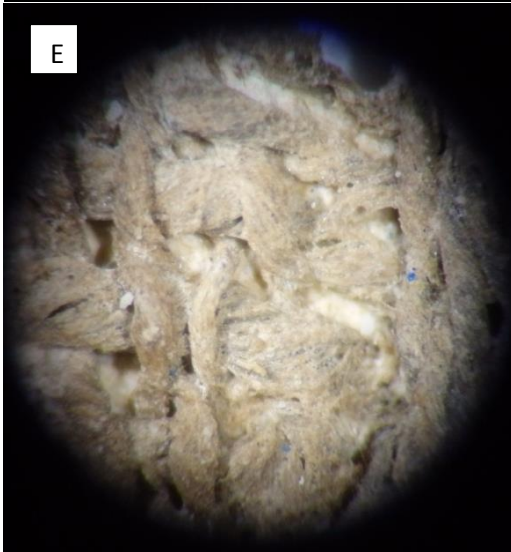
A. MAG: x0,6

B. MAG: x2.2

C. MAG: x2.2 and camera zoom

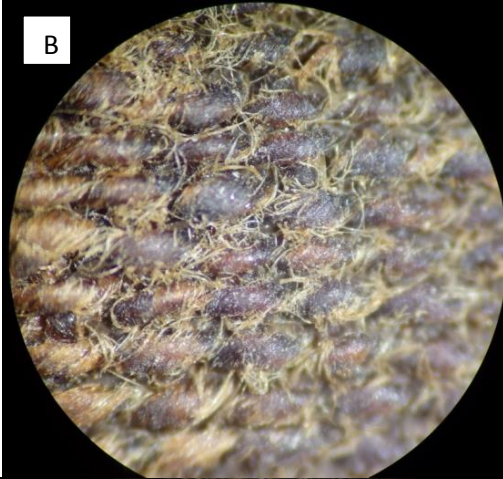
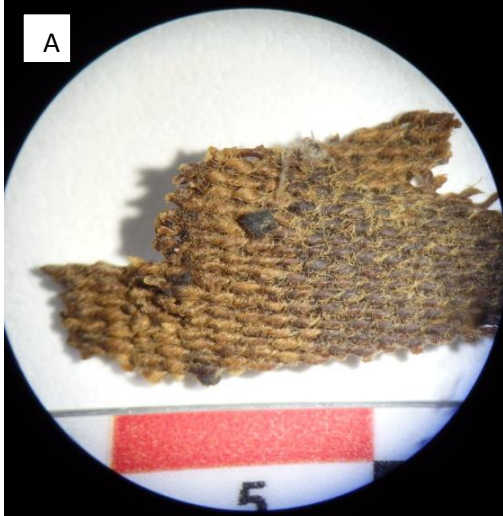


D. MAG: x1.7  
E. MAG: x2 and camera zoom





M1c



**Fragment of balm-impregnated bandage:**  
**Spinning:** monofilament S twisting direction  
**Weave:** tight, simple 1x1  
**Number of warp and /or weft:** ~10/15 per cm<sup>2</sup>

Wear and tear of the treads, soil or dust particles and black piece (A) on the sample is observed. Partial dark brown colored threads are detected (B).

A. MAG: x1  
 B. MAG: x4

M1d



Front side of M1d



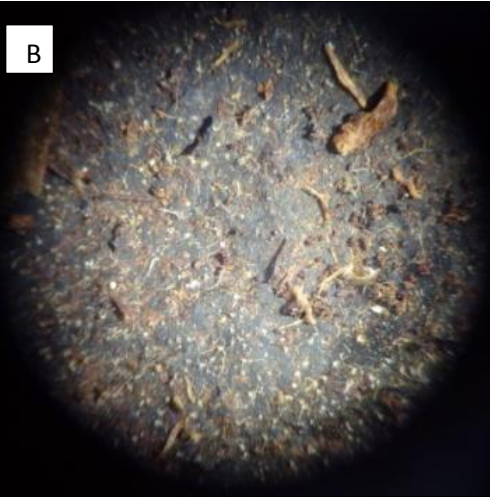
**Fragment of the tissue from the right femur:**

The piece of tissue is presented to be in black color. Exfoliation and cracking of the surface is observed (A). Remnants of the mummification materials and white spots are detected in both sides (A, B).

A. MAG: x0.9



Back side of the M1d



B. MAG: x0.8

M1e



Front side of M1e



Back side of M1e



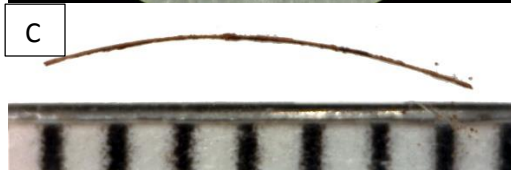
**Fragment of the bone and the tissue probably from the right femur:**

The bone is appeared in brown coloration. Part of tissue and part of spongy bone is preserved (A, B). Exfoliation, cracking and friability of the tissue is observed. White spots are detected in both sides.

A. MAG: x0.65

B. MAG: x0.66

M1f



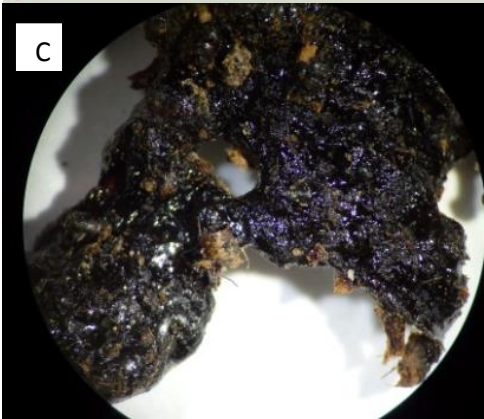
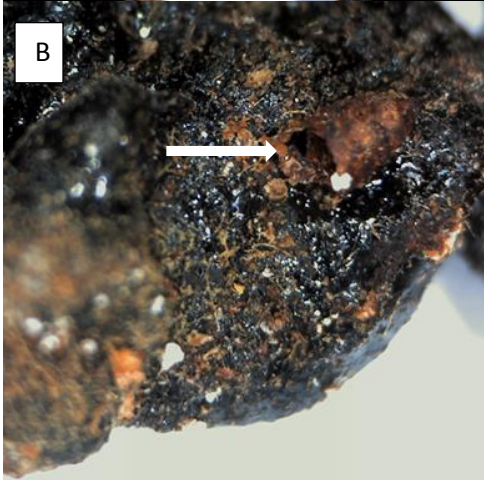
**A bunch of fragmented scalp hairs:**

The hair's appearance is in reddish brown color. A material is detected to coat the hairs (A, B, C). Fragmentation of the material is observed (A).

- A. MAG: x4.5
- B. MAG: x4.5
- C. LED-MICROSCOPE M1f07. MAG: 1280X960



**M1g**

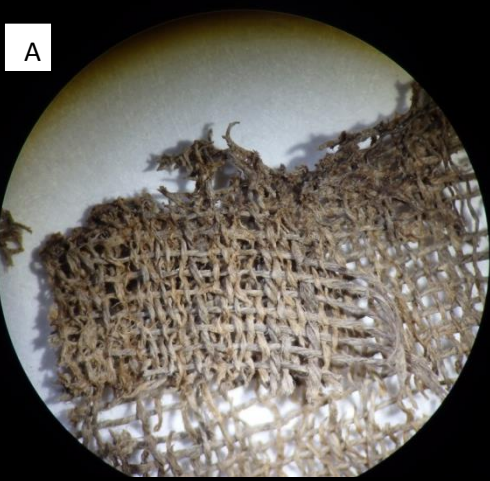


**A piece of balm from the thorax cavity:**

The piece of balm has resinous appearance. An insect residue is detected (A,B). Remnants of mummification materials are observed on the sample.

- A. MAG: x1.4
- B. LED-MICROSCOPE image: Detail of the insect residue.
- C. MAG: x1.4

M1h



**A piece of the wrapped fabric:**

**Spinning:** monofilament S twisting direction

**Weave:** simple 1x1

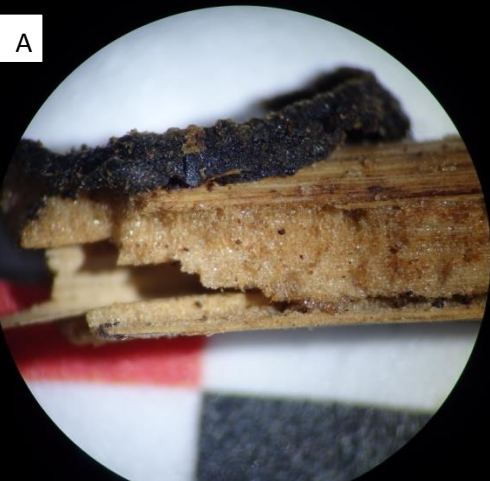
**Number of warp and /or weft:** ~12/15 per cm<sup>2</sup>

The textile is bleached and dried out (A). Wear and tear and crumbling of the threads is observed. At the edges of the bigger part of textile the threads are damaged (B). Soil particles and small blackish pieces are detected between the threads.

A. MAG: x1.4

B. MAG: x3

M1i


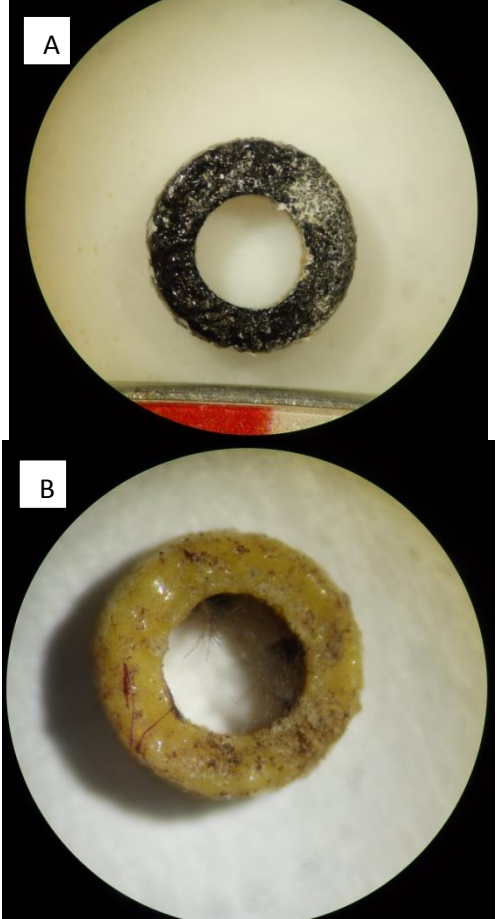


**Fragment of the plant from the thorax cavity:**


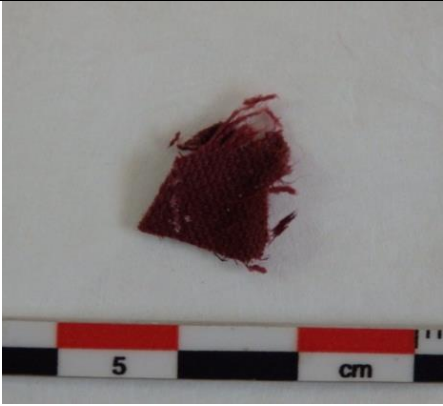
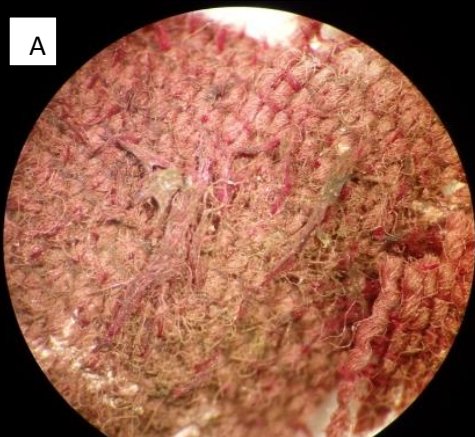

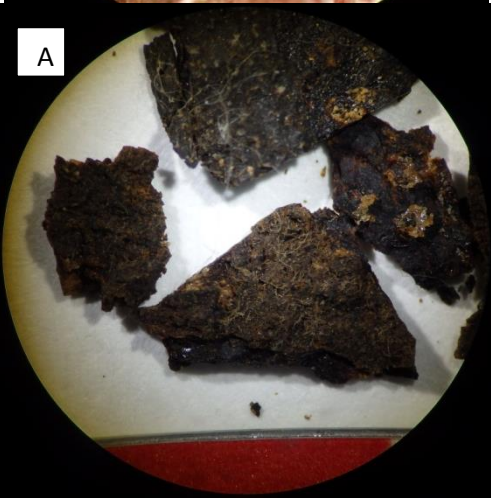
The plant sample is cracked in several areas. A piece of black material is on the edge of the fragment (A). Blackish and brown particles are detected on the hall surface.


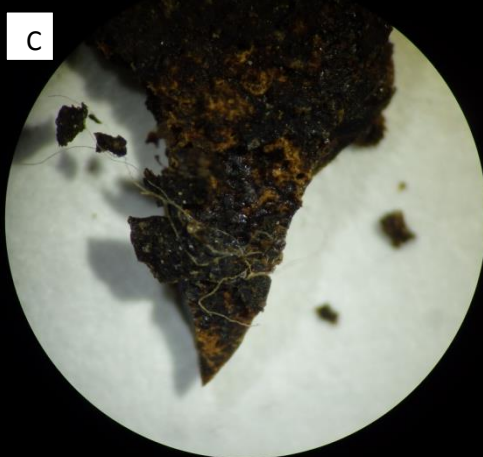


A. MAG: x2

Mummy 2 samples

Code name	Macro-image	Stereoscopic-image	Description
M2a1 black  M2a2 yellow			<p><b>Black and yellow beads:</b></p> <p><u>Black bead M2a1</u> Circle shaped black bead with a hole in the middle. The surface is heterogeneous and glossy. Soiling and Impurities are seated on the bead. During a cleaning test with acetone solvent, a material with fatty appearance was removed (A).</p> <p><u>Yellow bead M2a1</u> Circle shaped yellow bead with a hole in the middle. The surface is heterogeneous and glossy. The sample is partially covered with impurities and soil particles (B). A capillary structure is detected on the point of the inner circle(B, C).</p> <p>A. MAG: x3.9 B. MAG: x3.9 C. MAG: x 3.9</p>

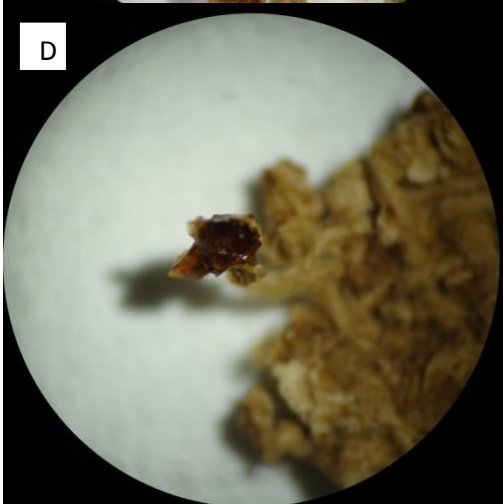
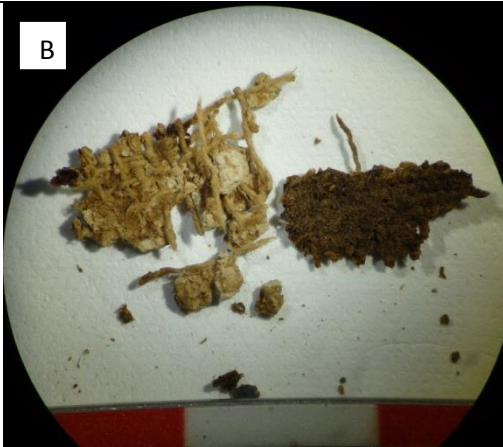


			
M2b			<p><b>Piece of the (later) textile where the mummy lies on:</b></p> <p>Synthetic red color textile. Fragmentation and damaging of the threads is presented. Dust, soiling and possible glue residues are observed (A).</p> <p>A. MAG: x2.3</p>
M2c			<p><b>Fragments of tissue from the left area of the thorax</b></p> <p>Four fragments of tissue are presented in black brown color. Yellow stains, white spots, degraded materials and parts with resinous appearance are observed (A, B). In two fragments of possible textile fibers are detected (C). The skin is cracked and exfoliated.</p>

		 <p data-bbox="669 331 701 361">B</p>  <p data-bbox="669 760 701 789">C</p>	<p data-bbox="1221 310 1425 340">A. MAG: x1.4</p> <p data-bbox="1221 346 1425 375">B. MAG: x1.4</p> <p data-bbox="1221 382 1425 411">C. MAG: x2.8</p>
M2d	 <p data-bbox="263 1627 506 1656">Front side of M2d</p>	 <p data-bbox="669 1228 701 1257">A</p>	<p data-bbox="1172 1207 1572 1270"><b>Fragments of bandages (with color)</b></p> <p data-bbox="1172 1312 1572 1785">Two fragments of bandages with blue color pigments. The bandages present different characteristics of textile used (A, B). The bigger fragment's threads are in light yellow color whereas smaller fragment's threads are in brown color. The weaving of the bigger fragment is sparse and tight for the small fragment (B). White and baize parts of plaster are preserved</p>



Back side of M2d

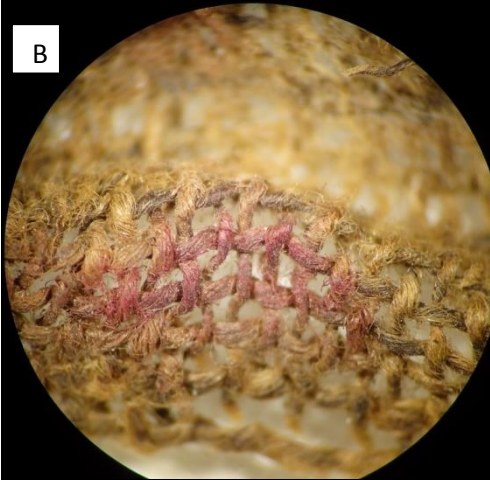
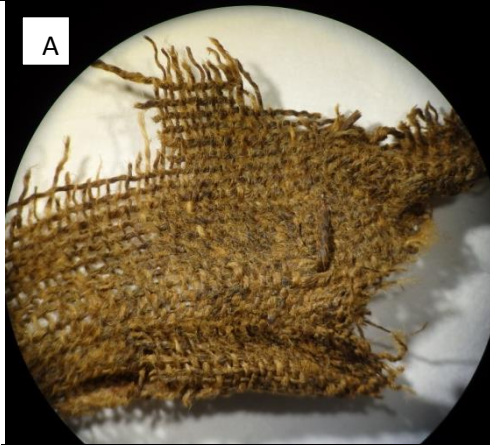


(A, B). Blue colored areas have impurities and deposits (C). Residuals of resinous materials are detected (D). The sample is very fragile. Cracking, brittleness and crumbling of the plaster is observed. Dust and degraded materials are on the threads (A, B, C).

- A. MAG: x 0.67
- B. MAG: x0.9
- C. MAG: x1.6
- D. MAG: x2.9



M2e



### Fragment of bandage

**Spinning:** monofilament S  
twisting direction

**Weave:** simple 1x1

**Number of warp and /or  
weft:** ~10/15 per cm<sup>2</sup>

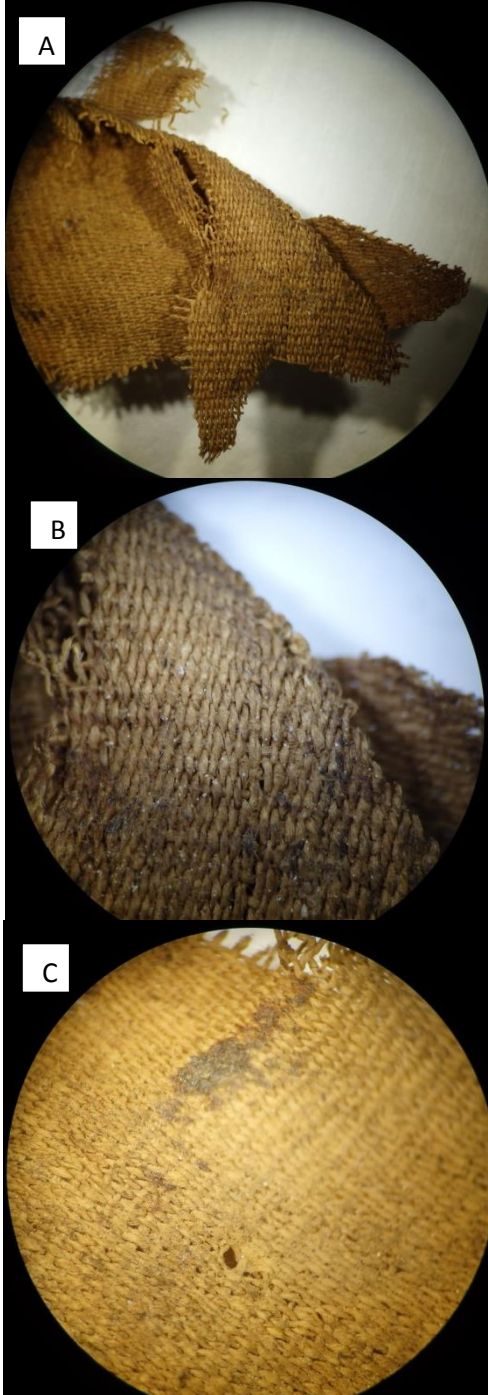
The textile preserves its natural properties. Light yellow and brown threads are observed (A). Bleaching and partial stiff threads is determined (B). Soil particles and impurities are detected between the fibers (B). Parts of the fragment are appeared crumpled or damaged (C).

A. MAG: x0.67

B. MAG: x3

C. MAG: x1

M2f



**Fragment of different type of bandage**

**Spinning:** monofilament S twisting direction

**Weave:** dense, simple 1x1

**Number of warp and /or weft:** ~17/33 per cm<sup>2</sup>

The textile is stiff and crumpled (A). Stains in brown color, white spots and impurities are observed on the threads (B, C). A hole in the middle of the fragment is detected (C).

A. MAG: x0.66

B. MAG: x2

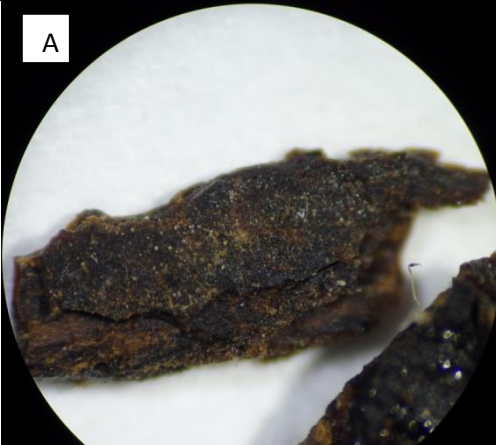
C. MAG: x1.5



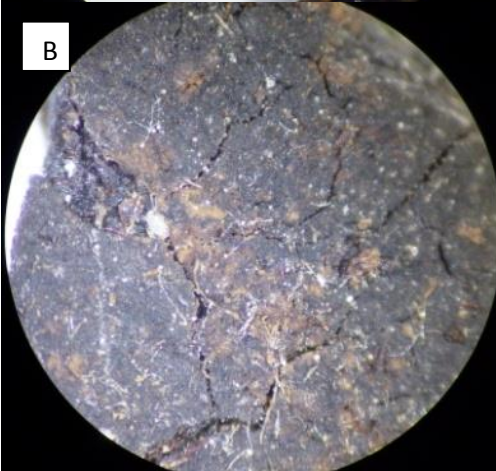
M2g



A



B



C



### Fragments of balm-impregnated tissue

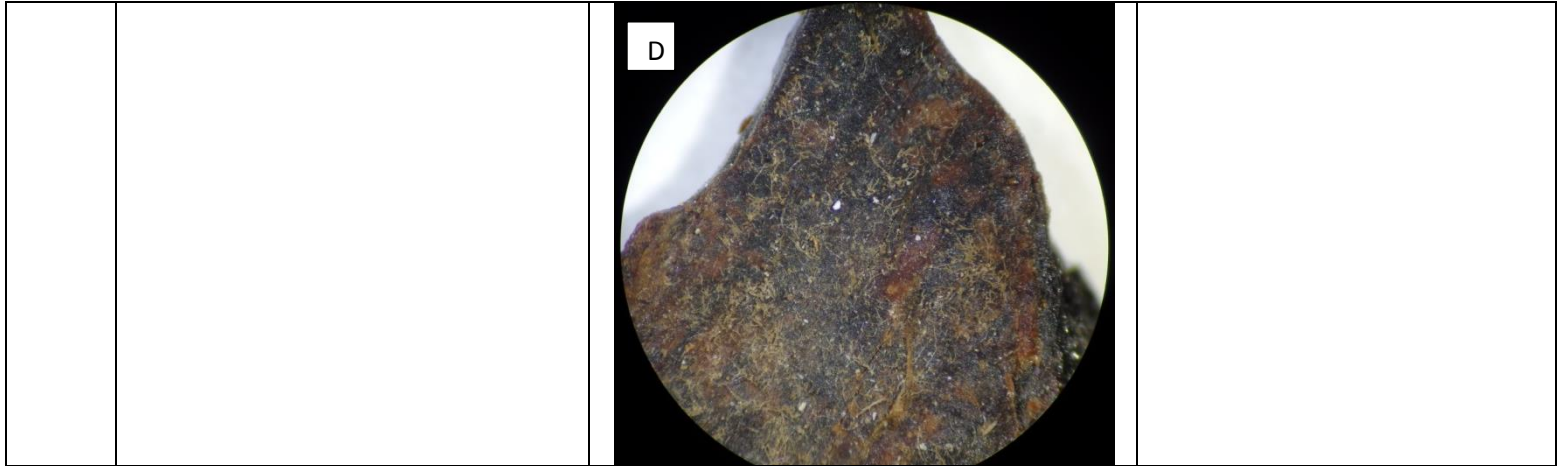
The fragments of tissue are in black-brown color. The skin is presented stiff, exfoliated and cracked (A, B). Degraded materials, impurities, white spots and soil particles are observed in the surface (A, B, D). Balm residues and possible remnants of fibers are determined (C, D).

A. MAG: x4.5

B. MAG: x4.5

C. MAG: x4.5

D. MAG: x4.5



<b>Table 11</b>	
<b>M1 hairs' length (mm)</b>	
M1f01	29,39
M1f02	13,78
M1f03	12,63
M1f04	11,45
M1f05	10,18
M1f06	8,43
M1f07	7,38
M1f08	7,21
M1f09	7,02
M1f10	3,18

## 4.4 Optic Microscope

### 4.4.1 Hairs (M1f)

The hairs under the microscope are presented to be in gold bronze color. Pieces of solid material in brown-orange color are on the hair (fig.50). The coated material is fragile and crackled (fig.51). Vertical slots like breakage are observed in some hairs (fig.52-53). Characteristic structures of microorganisms are also determined (fig.54-56).

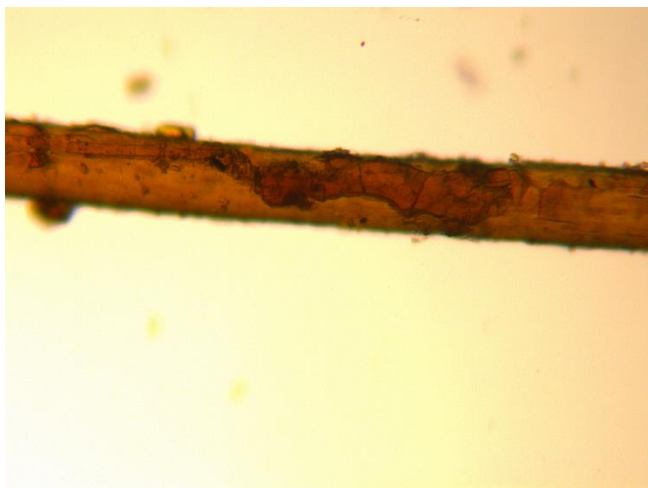


Fig.50: Hairs sample M1f1. The solid material covers part of the hair. MAG: x10

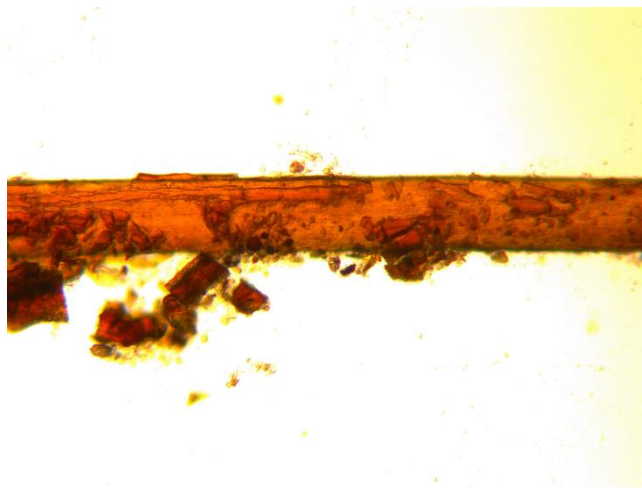


Fig.51: Hair sample M1f10. The coated material is fragile and crackled. MAG: x10



Fig.52: Hair sample M1f4. Vertical slots along the hair. MAG: x10

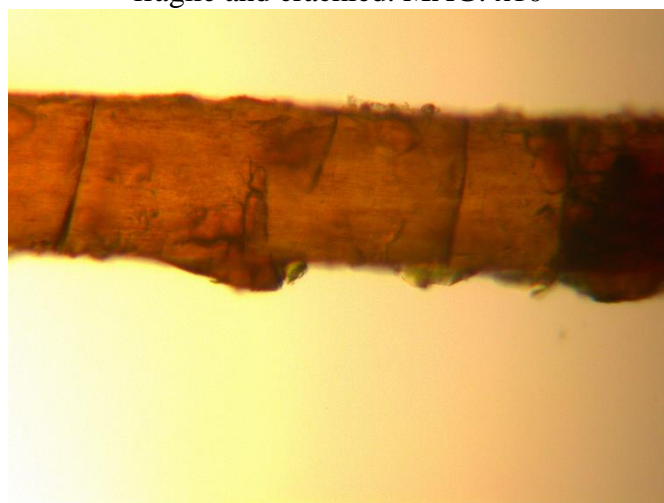


Fig.53: Hair sample M1f4. Detail of the vertical slots. MAG: x20

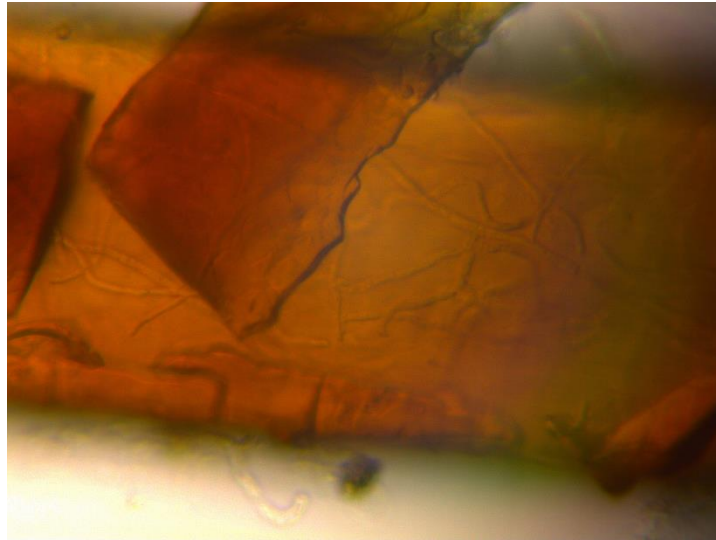


Fig.54: Hair sample M1f1. Detail of part of coated material and hyphae of microorganisms. MAG: x50

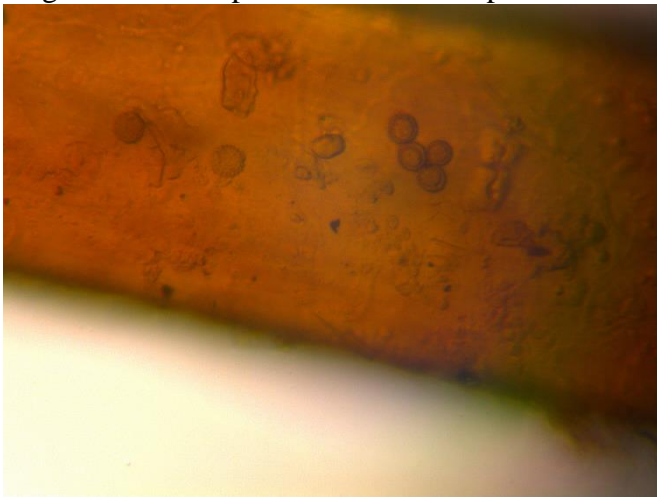


Fig.55: Hair sample M1f1. Detail of. MAG: x50

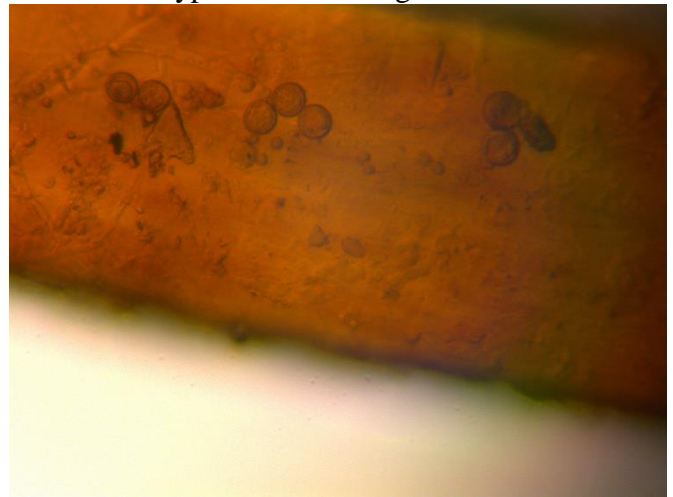


Fig.56: Hair sample M1f1. Detail of microorganisms. MAG: x50



#### 4.4.2 Plant sample (M1i)

By the observation of the cross section of shoot plant, anatomical characteristics were determined. Plant cells and vascular bundles are presented (fig.57-58).

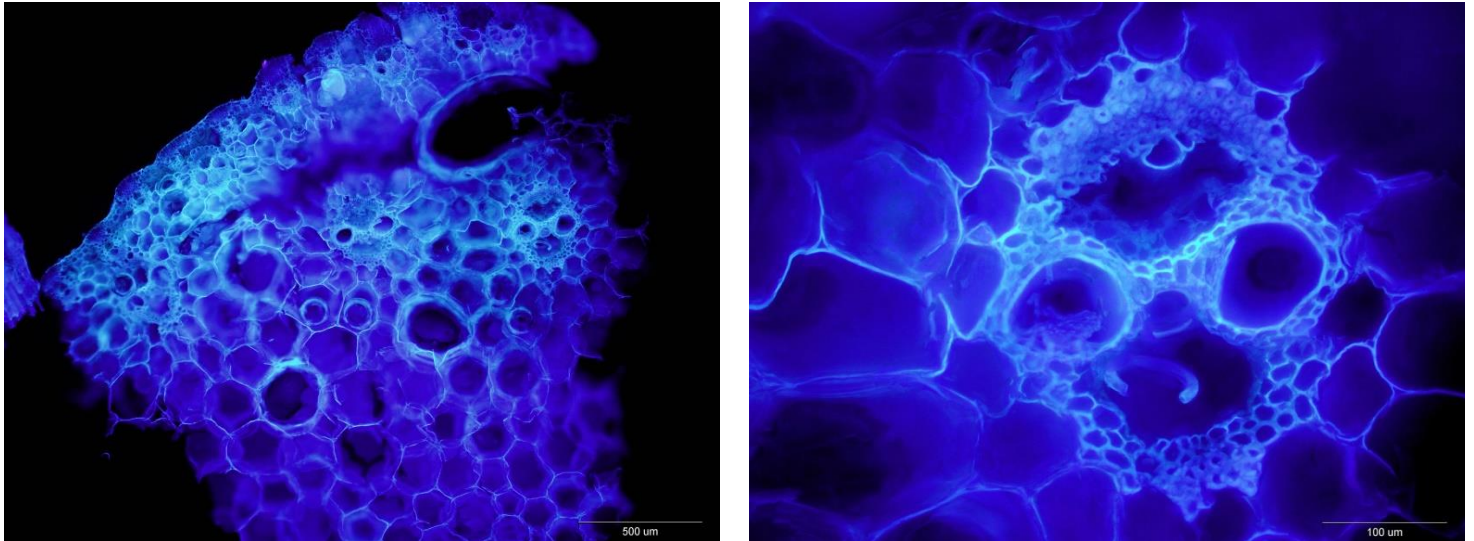
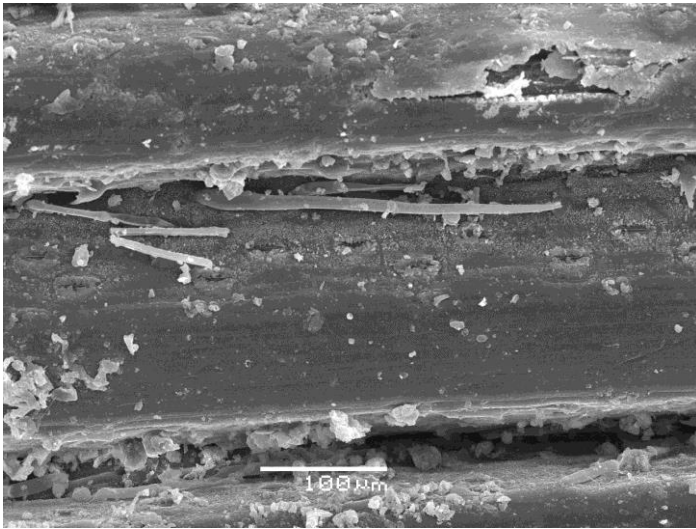


Fig.57-58: Soot of plant M1i: Cross section of plant stem, vascular bundle.

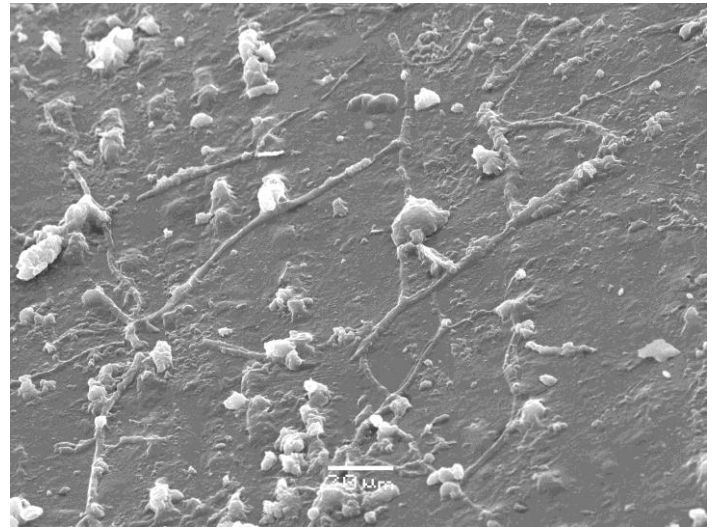
## 4.5 Scanning Electron Microscopy (SEM) and Energy-dispersive X-ray spectroscopy (EDX)

### 4.5.1 Plant sample (M1i)

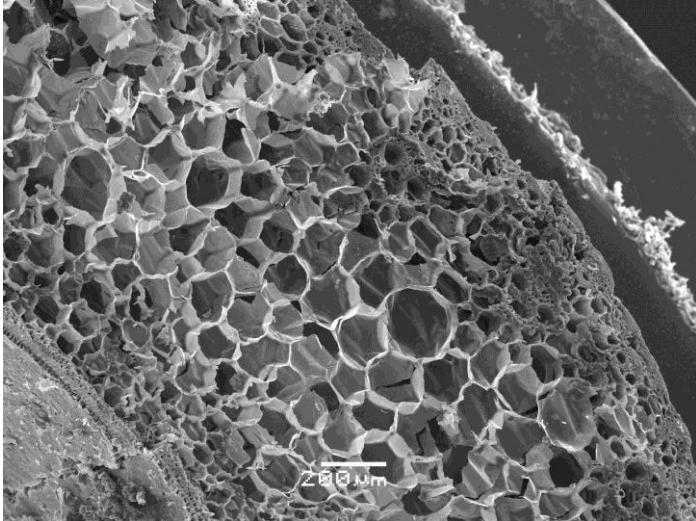
The anatomical characteristics were observed in M1i are; stomata of the epidermis, plant cells and the vascular bundles. Hyphae of microorganisms were also determined.



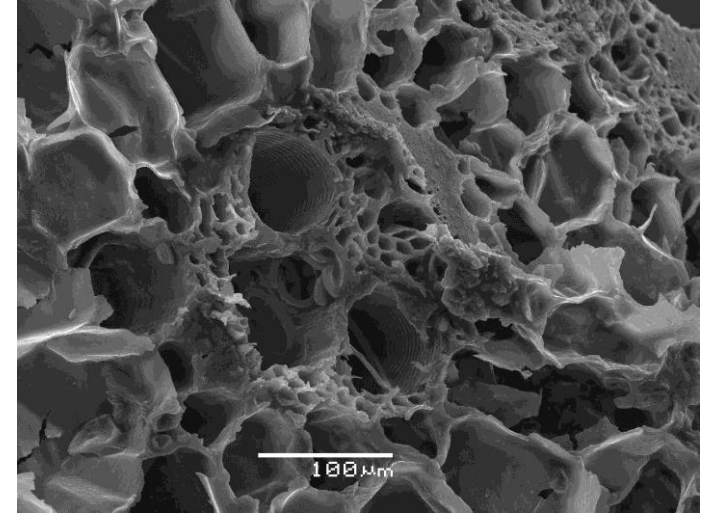
SEM image: Surface section: Epidermis: stomata microscopic anatomical openings. Transporting system.



SEM image: Cross section of epidermis: Hyphae of microorganisms.



SEM image M1i: Plant cells.



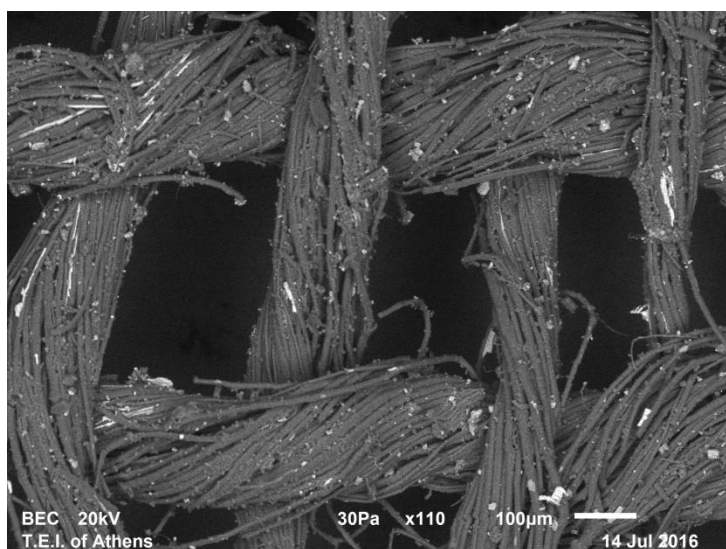
SEM image M1i: Vascular Bands: transporting system. Anatomical characteristics of vascular plant.

#### 4.5.2 Fragments of bandages (M1a, M1b, M1c, M1h, M2e, M2f)

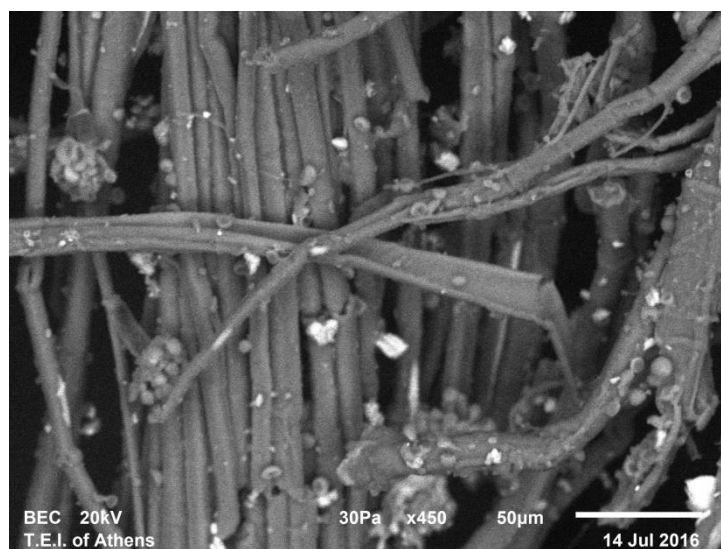
**SEM:** Most of the fragments of bandages were identified made of natural fibers. The weaving (simple) and the texture of the fabric is similar between them expect from M1c where the weaving is presented tight. Broken, missing and damaged fibers were observed in almost all samples. Dust and dirty particles also cover the fibers. In the case of M1a colonized fibers by microorganisms were determined.

**EDEX:** Ca, Si, Cl, and Na are the predominant elements for M1a, M1c, M1h and M2f, fragments of bandages. Less dominant elements are Al, S, Mg and K. Small amounts of Fe and P was determined in M1c, M1h and M2f samples.

For M1b and M2d samples (plaster and color) Ca, and Si were determined in the largest percentages. In blue areas of M1b and M2d, Ca, Cu and Si are the predominant elements. In red area of M1b, Ca, Si and Fe are in the largest amounts. Subsequently less dominant are Cl and Na and Al. Sr was determined in M1b spectra of dark and blue area whereas a small amount of Ti was determined in M2d. The percentages of elements are presented on Table 12.

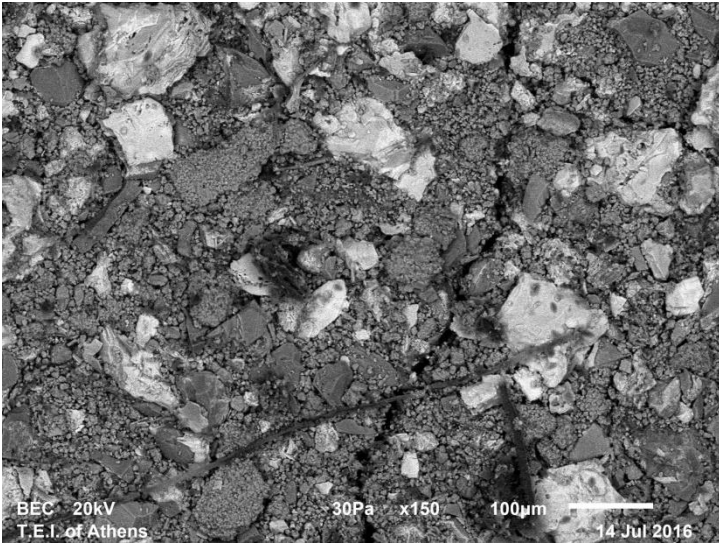


**SEM image M1a:** Detail of threads. Weaving simple, broken fibers, particles (dust and dirty) on the fibers.

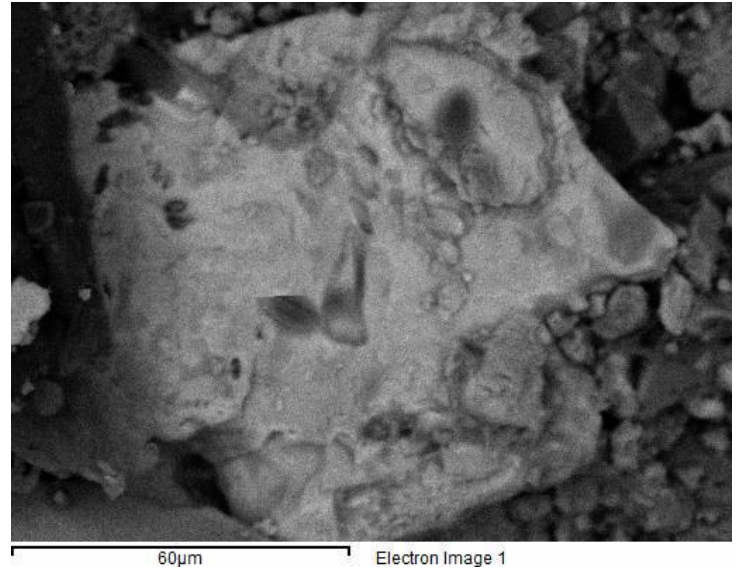


**SEM image M1a:** Detail of the fibers. Anatomical characteristics of natural fiber; Broken fibers. Longitudinal spitting, small scratches, particles (dust and dirty), mass of microorganisms between the fibers.

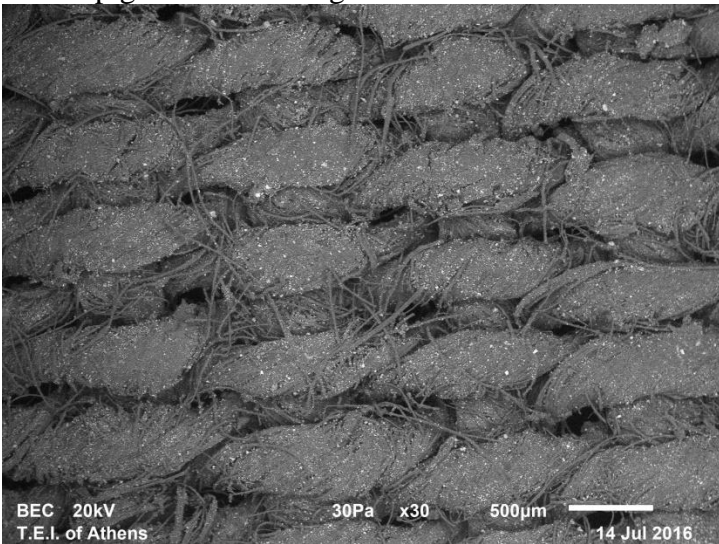




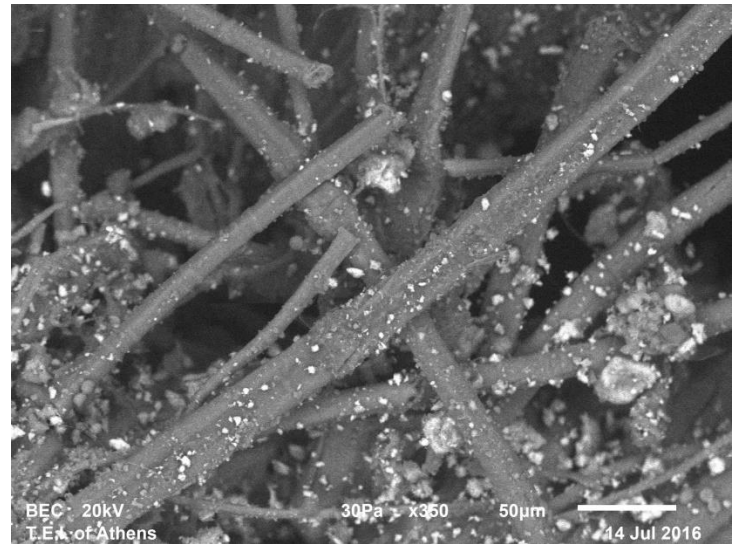
**SEM image M1b:** Detail of area with color. Different size of pigments. Cracking of the surface.



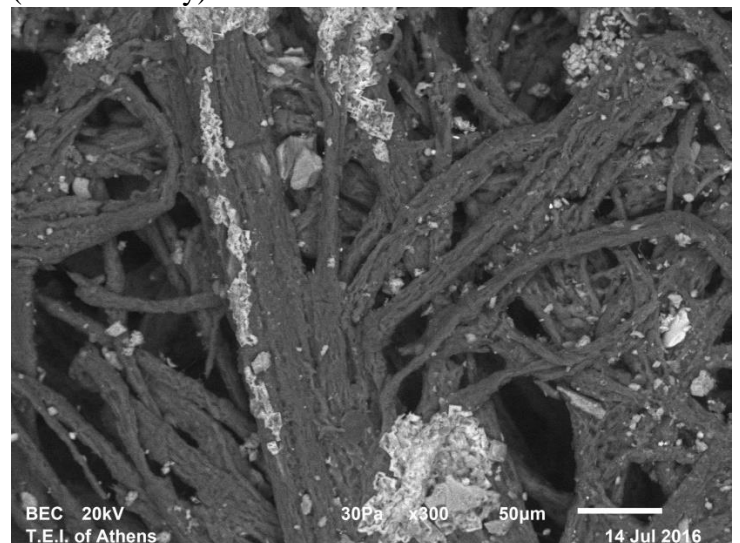
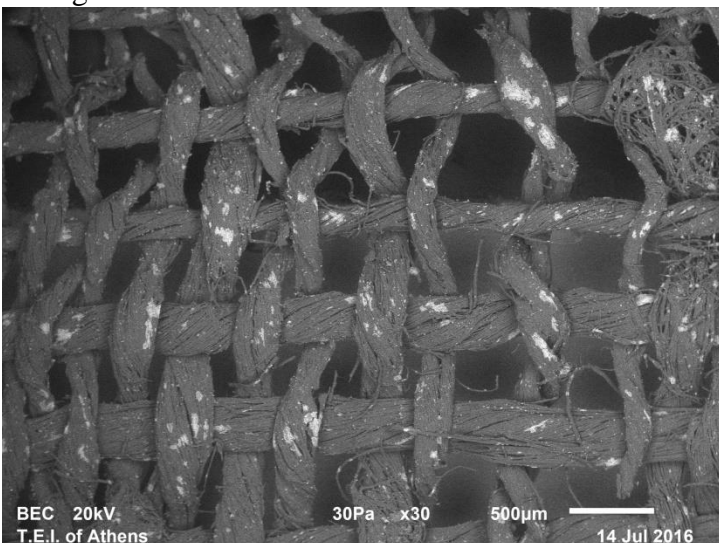
**SEM image M1b:** Detail of the blue grain.



**SEM image M1c:** Detail of the weaving. Simple, weaving, threads impregnated of solid material, damaged fibers.



**SEM image M1c:** Detail of the fibers. Anatomical characteristics of natural fiber; broken fibers, particles (dust and dirty) on the fibers.

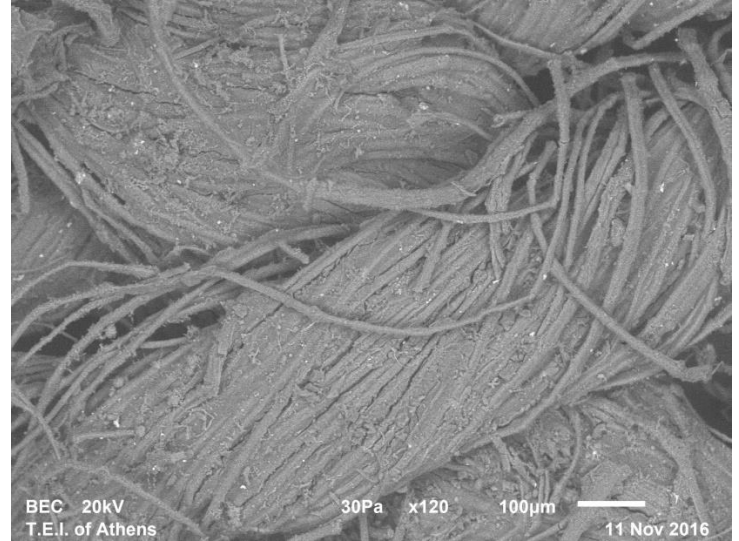




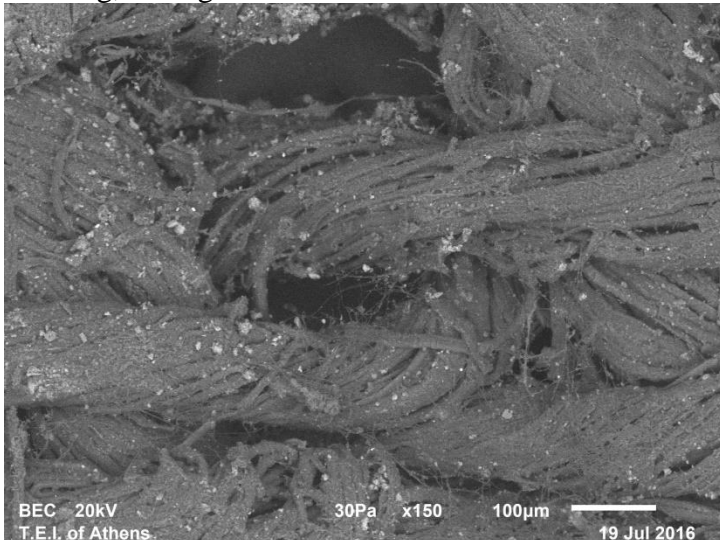
**SEM image M1h:** Detail of the weaving. Simple weaving, broken, convoluted, threads, damaged fibers, particles (of solid material, dust and dirty) on the treads.



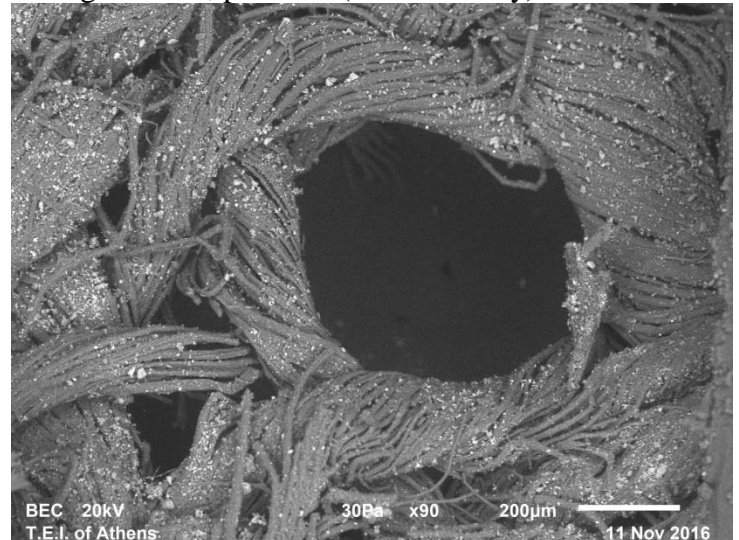
**SEM image M1h:** Detail of the fibers. Pressed, convoluted, damaged fibers, particles (of solid material, dust and dirty) on the fibers.



**SEM image M2e:** Detail of the weaving. Simple weaving, damaged threads and fibers.

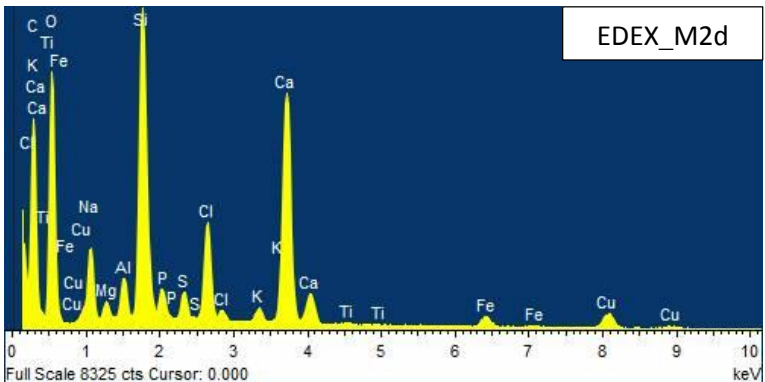
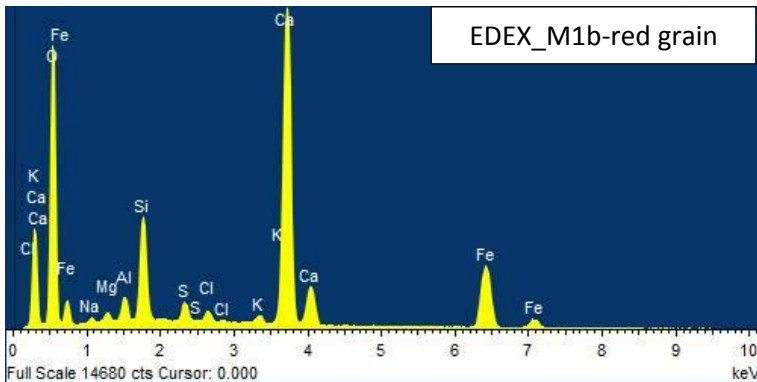
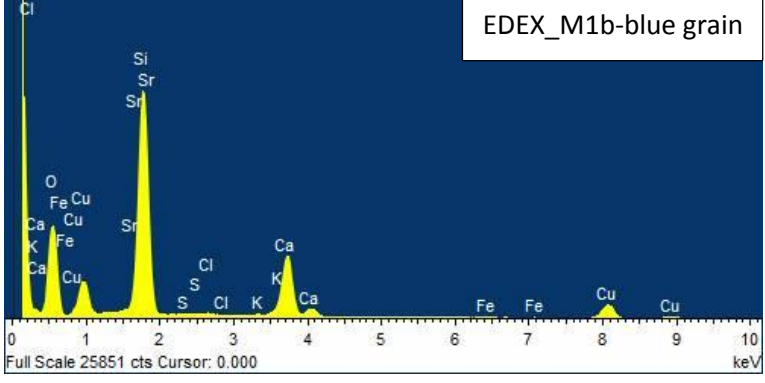
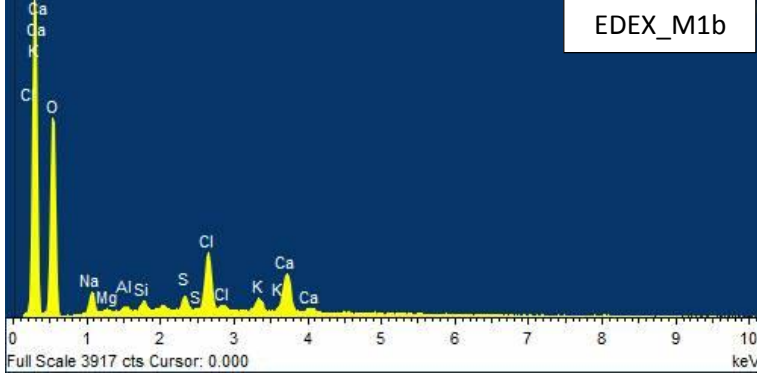
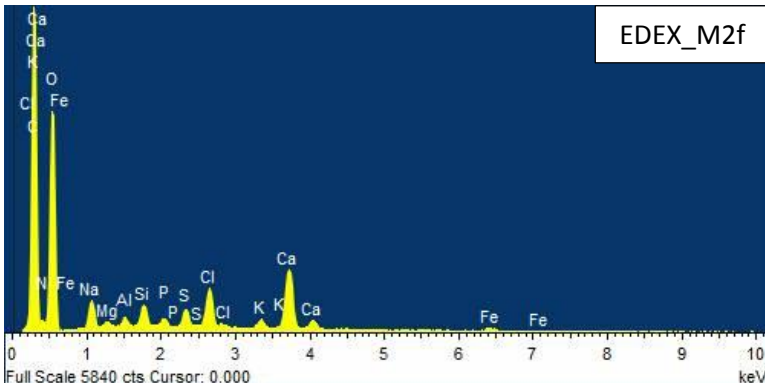
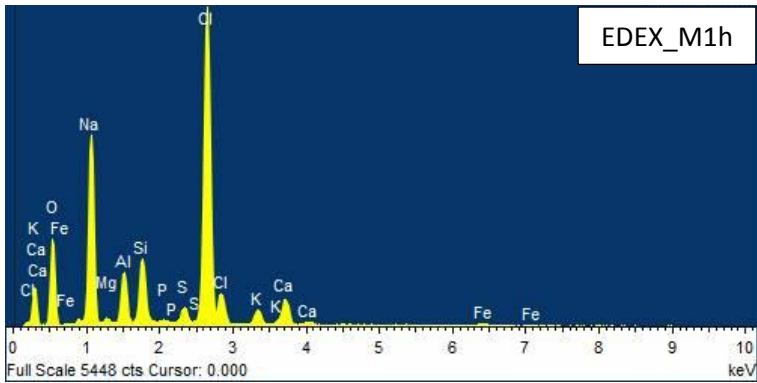
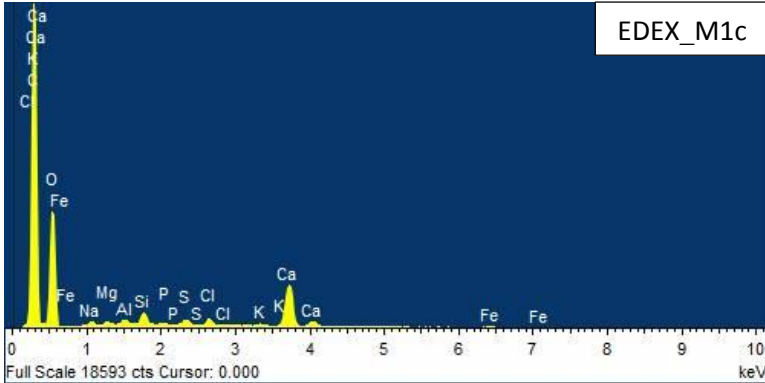
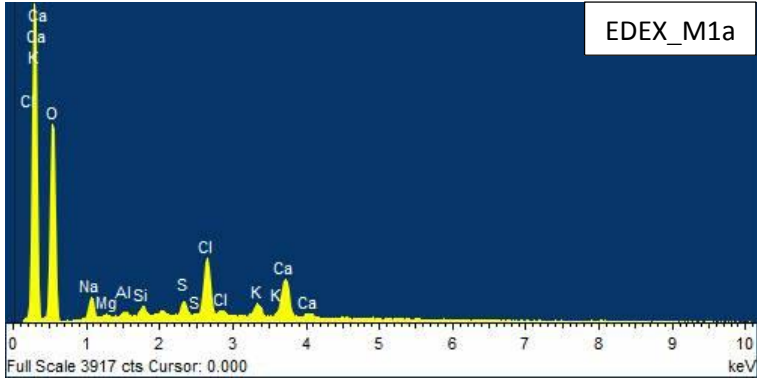


**SEM image M2e:** Detail of the thread. Broken, damaged fibers, particles (dust and dirty) on the fibers.



**SEM image M2f:** Detail of the threads. Damaged threads and fibers, particles (dust and dirty) on the fibers.

**SEM image M2f:** Detail of the hole. Damaged threads, broken fibers, particles (dust and dirty) on the fibers



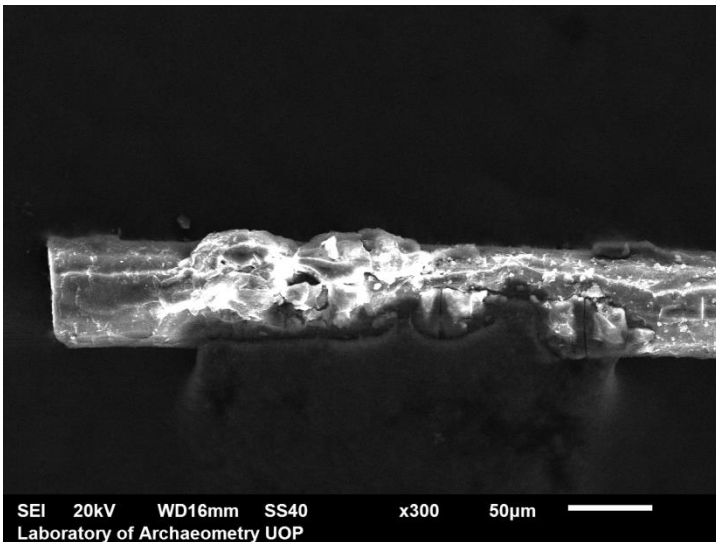
	M1a	M1c	M1h	M2f	M1b(1)	M1b(2)	M1b(3)	M1b_Blue grain	M1b_Red grain	M2d blue fragment
<b>Atomic%</b>										
Cl	4.28	0.13	19.72	0.49	1.08	0.31	0.27	0.14	0.35	
Na	3.42	0.14	18.34	0.67	0.92	0.39	0.24		0.38	1.46
Ca	3.17	0.94	1.82	0.85	15.08	6.68	9.73	5.25	10.96	2.67
S	0.95	0.10	0.85	0.21	0.14	0.13		0.03	0.60	
K	0.99	0.03	1.02	0.10	0.12	0.14		0.01	0.24	0.14
Si	0.70	0.20	3.95	0.25	2.55	4.22	1.83	18.74	3.46	2.96
Al	0.32	0.09	3.21	0.10	0.49	0.38	0.28		0.82	0.41
Mg	0.32	0.07	0.36	0.07	0.33	0.11	0.16		0.44	0.23
Fe		0.04	0.31	0.08	0.31	0.19	0.49		4.50	0.23
Cu						0.54		3.43		0.52
Sr								1.48		
C		66.67		50.79		38.33	33.98	66.67	54.52	
O	85.84	31.57	50.30	36.06						
P		0.03	0.12	0.10						0.35
N				10.21						
Ti										0.03

Table12:   Predominant elements.   Fe   Cu.

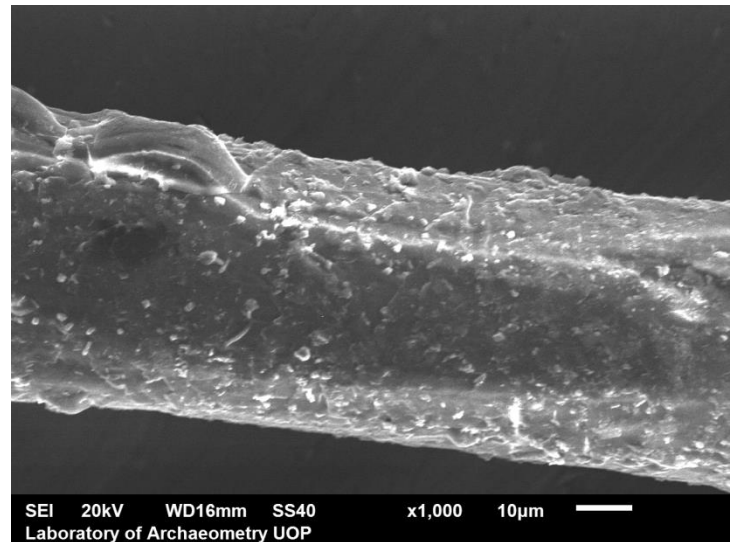
#### 4.5.3 Hairs (M1f1, M1f3) and tissue (M1d, M2g)

**SEM-EDEX Hairs (M1f1, M1f3):** The hairs of M1 are presented deteriorated. Parts of solid material on the hairs are preserved. The material is fragile. Parts of the material were been broken under SEM conditions. The structure of the hair was presented a wave spring shape (M1f3). The scales of the hairs were determined. In one point of M1f3, the outer layer (cuticle) of the hair was damaged. Short distance vertical slits were observed in both samples. Hyphae of fungus were distinct in several points of the hairs. Particles of dust and degraded materials on the hairs were observed.

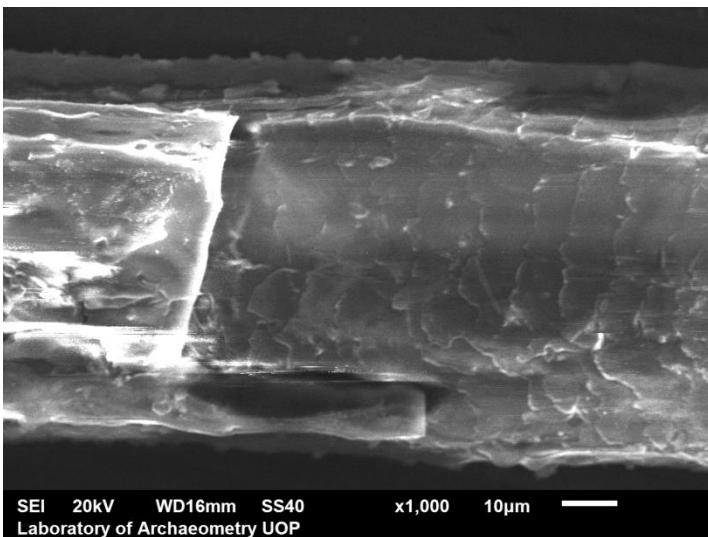
The predominant elements for ancient hairs (M1f1, M1f3) are C, O, S, Al, Ca, and Mg. Less dominant elements are: Si, Na, Cl and K. Small amounts of Ag, Sb, I, S, Fe, Pb, Ti, V, and P were determined in M1f1. In modern hairs S and Al are the predominant elements. Sr and Au were determined in Modern1 hair (male).



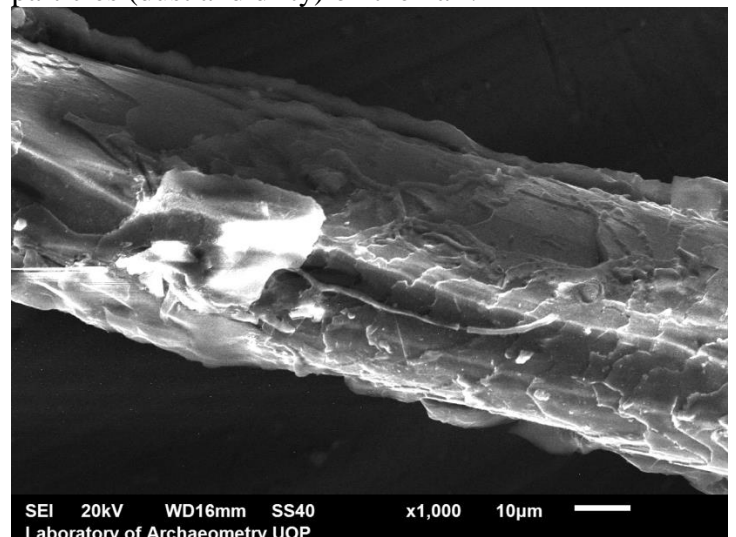
**SEM image M1f3:** Solid material on the hair.



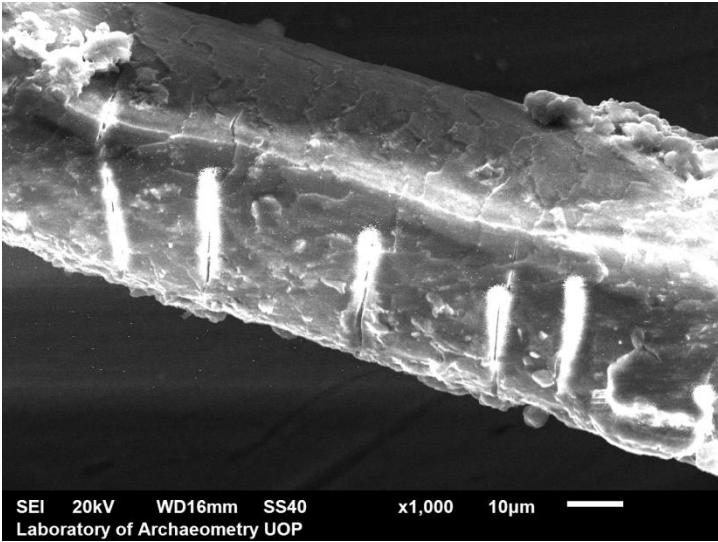
**SEM image M1f3:** Wave spring shape of the structure, particles (dust and dirty) on the hair.



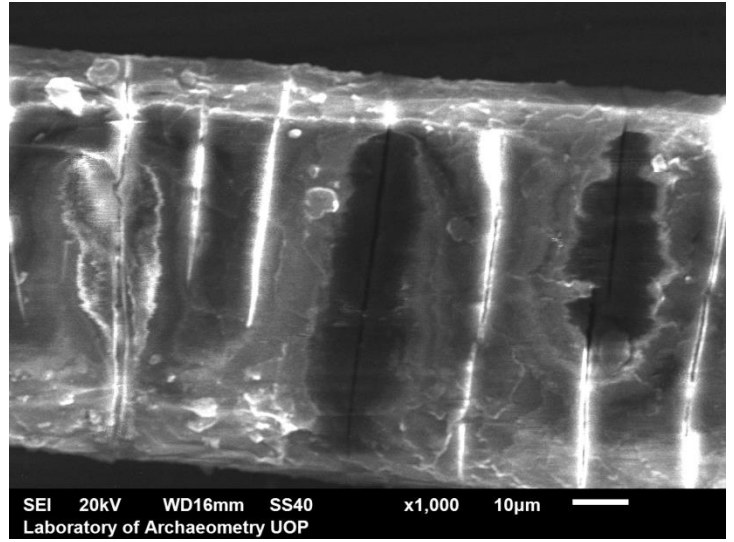
**SEM image M1f1:** The scales are partly covered with solid material.



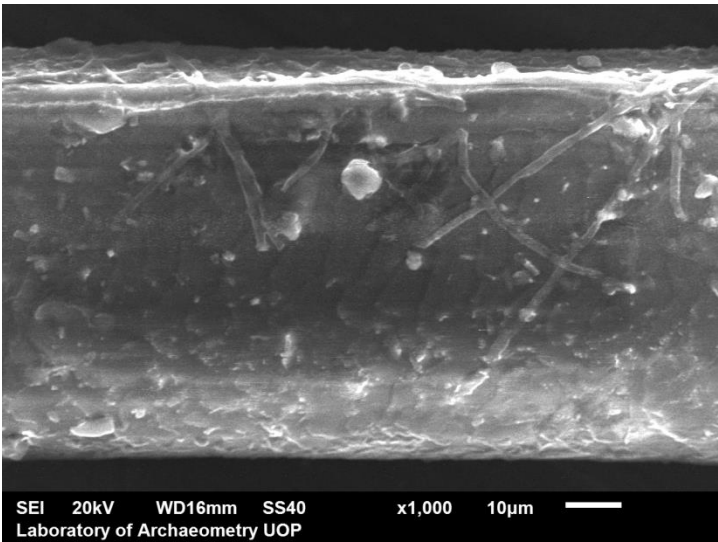
**SEM image M1f3:** The outer layer of the hair (cuticle) is damaged. Fungus hyphae are visible.



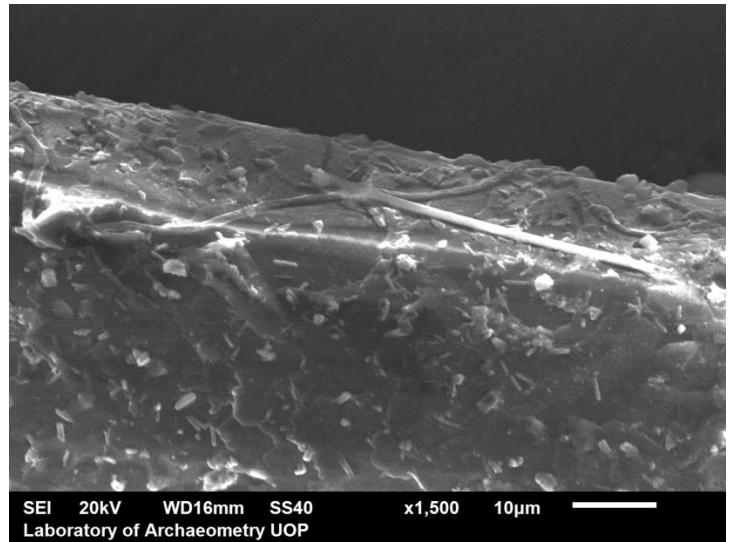
**SEM image M1f3:** vertical slits, parts of solid material.



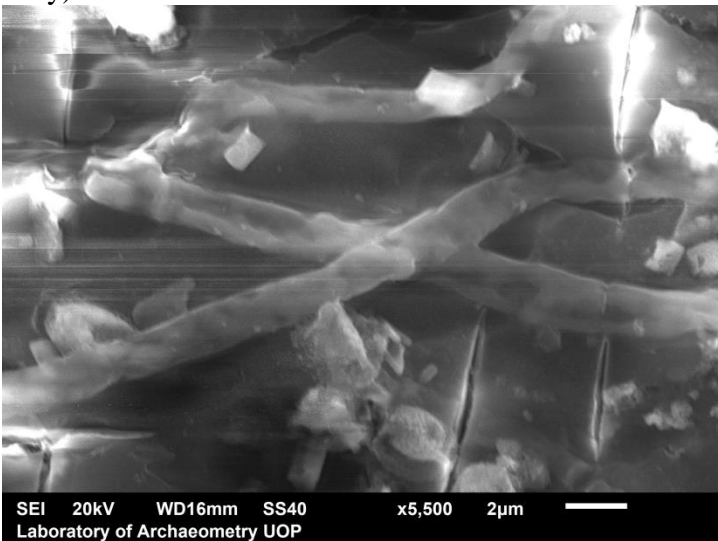
**SEM image M1f1:** Short distance vertical slits.



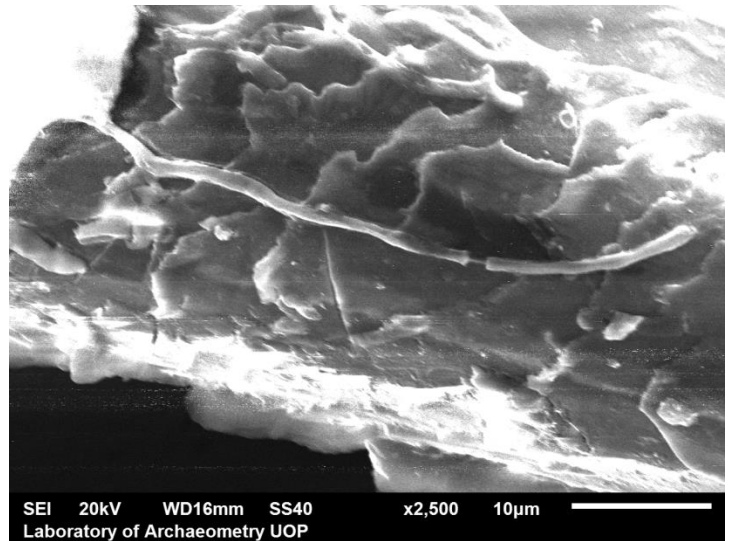
**SEM image M1f1:** fungus hyphae, particles (dust and dirty) on the surface.



**SEM image M1f3:** particles and degraded materials on the hair.

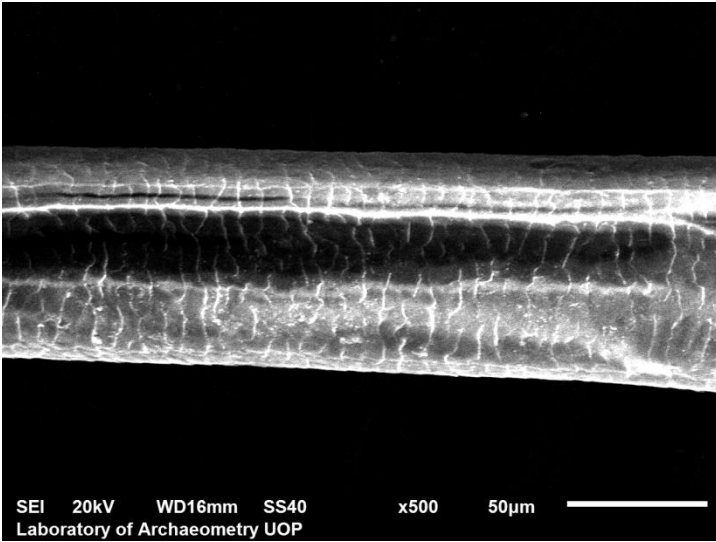


**SEM image M1f1:** fungus hyphae, vertical slits, degraded materials.

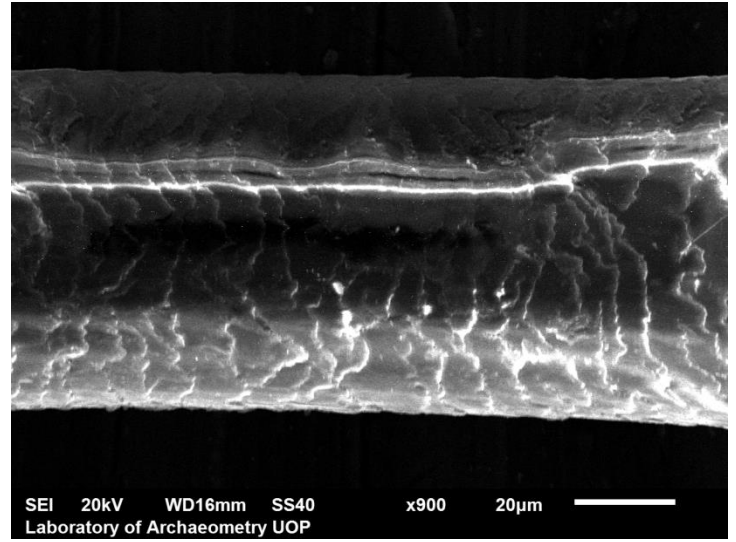


**SEM image M1f3:** fungus hyphae on the hair scales.

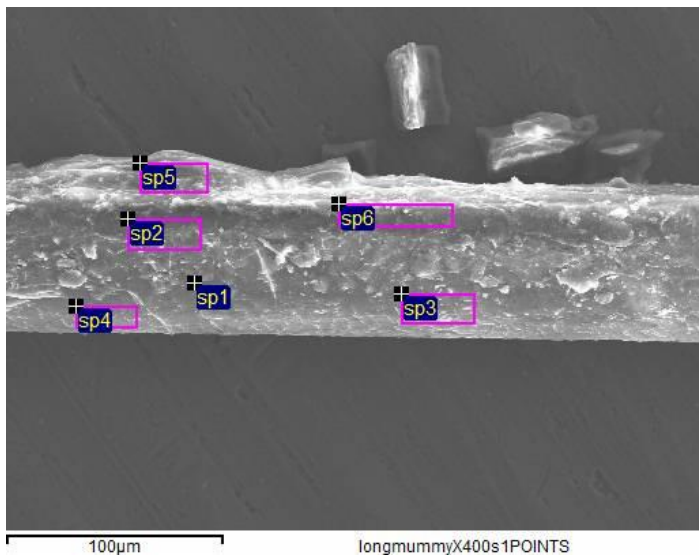




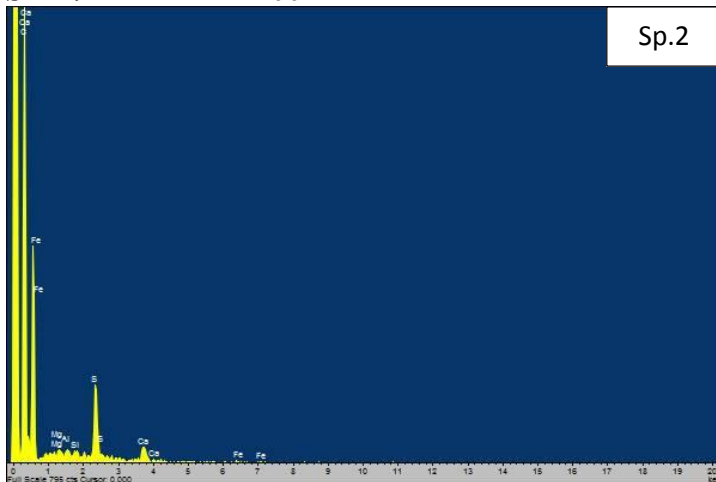
**SEM image of Control sample modern male hair:**  
The hair scales and the structure of fine type hair are visible.



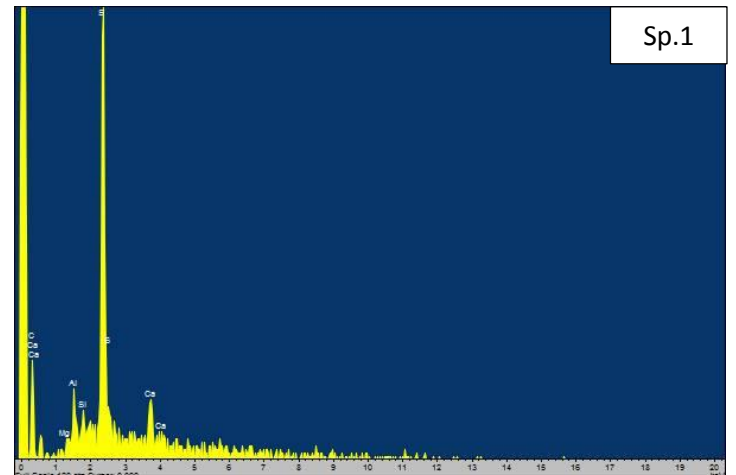
**SEM image of Control sample modern female hair:**  
the scales and the structure of curled type hair are visible.



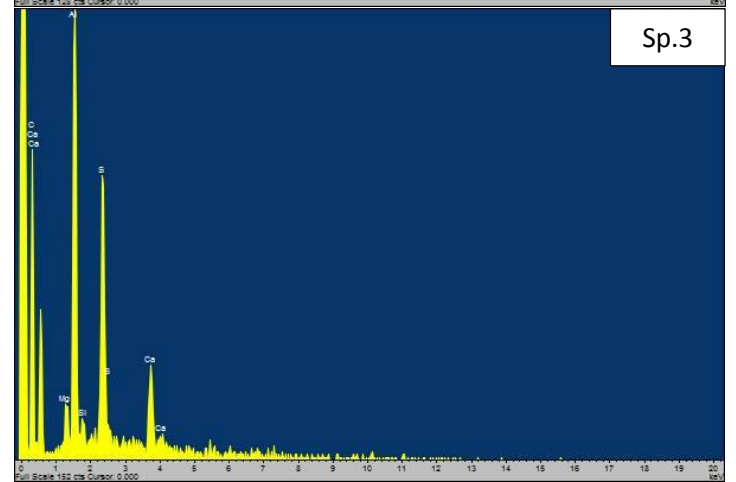
**SEM/EDEX M1fx400**



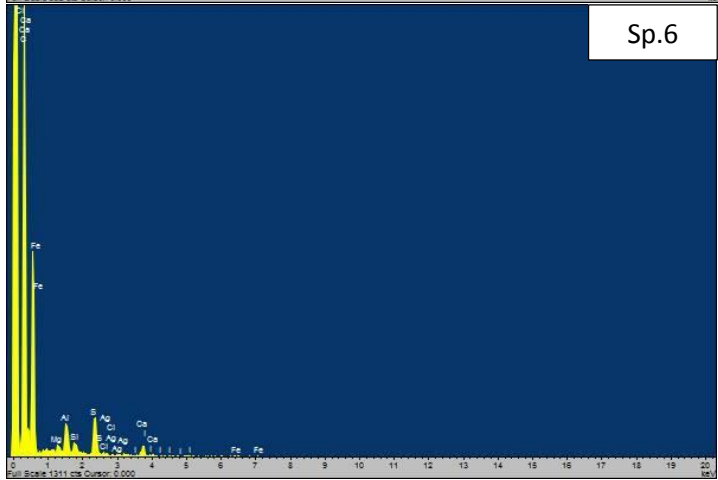
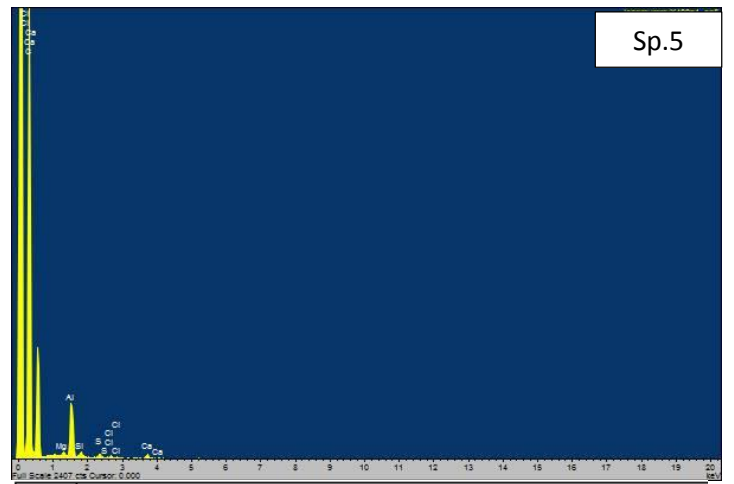
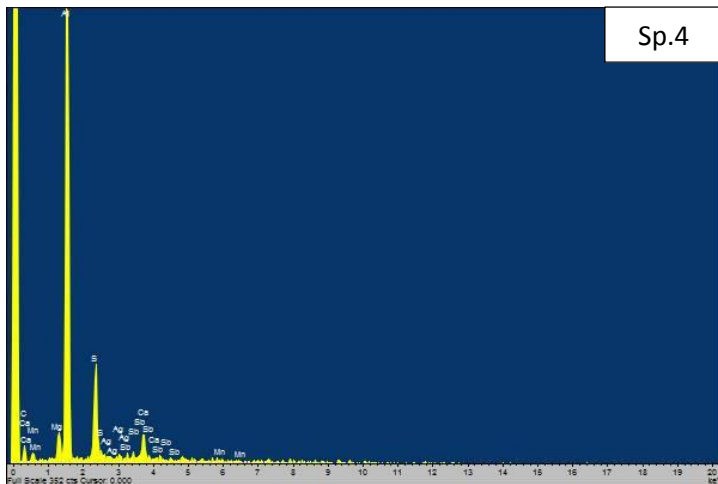
Sp.2



Sp.1

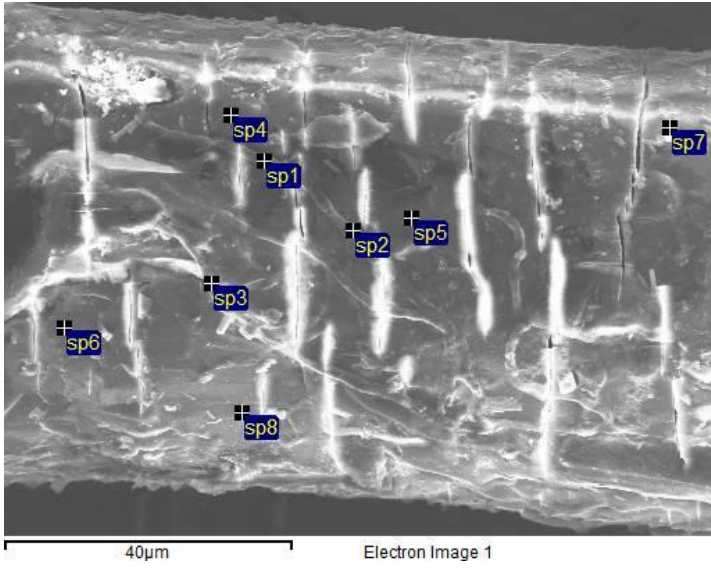


Sp.3

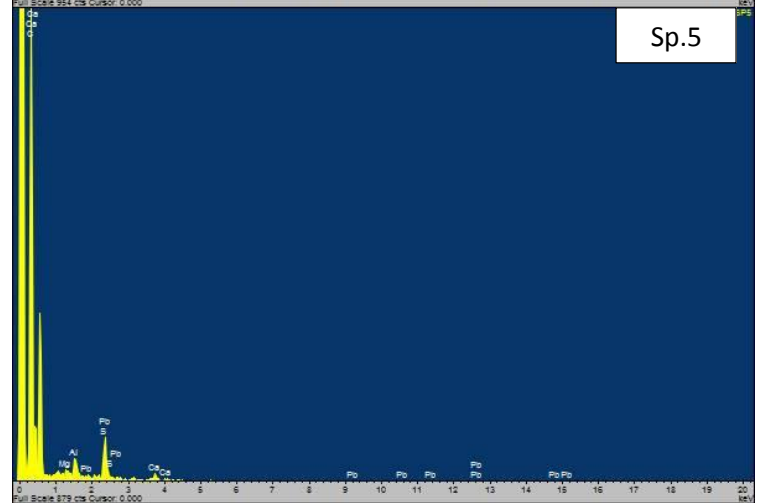
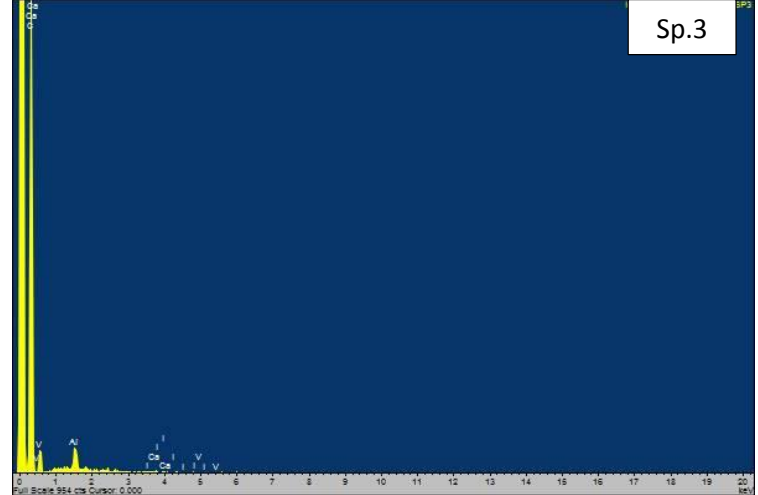
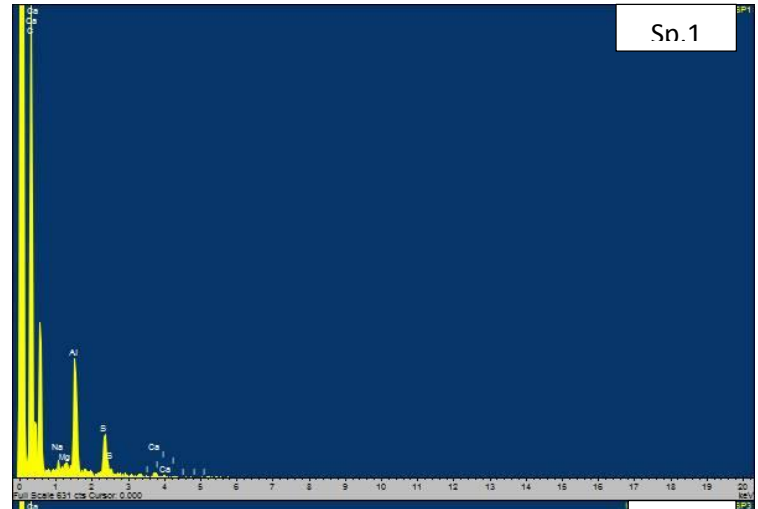
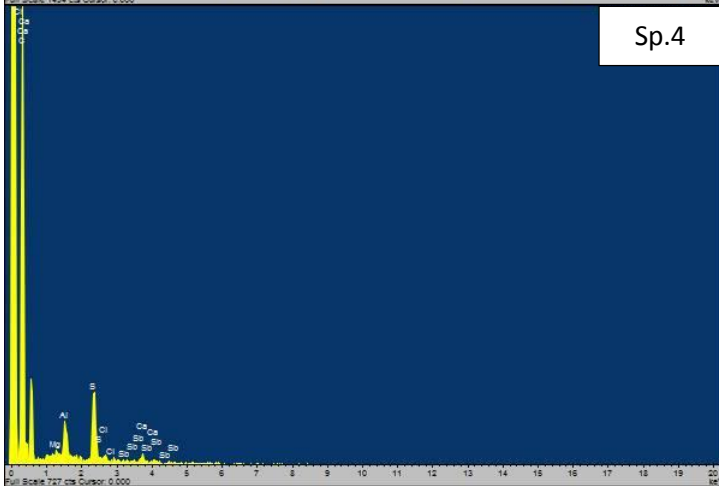
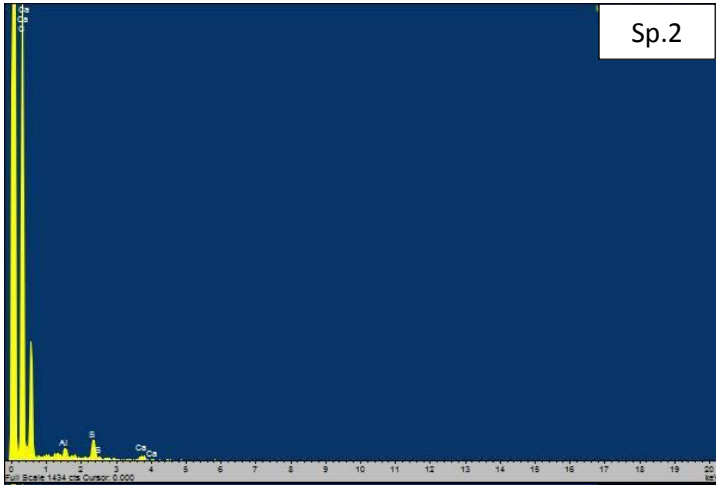


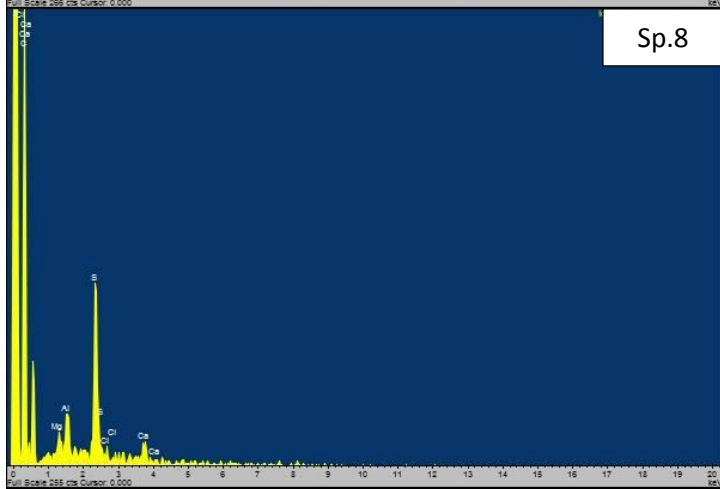
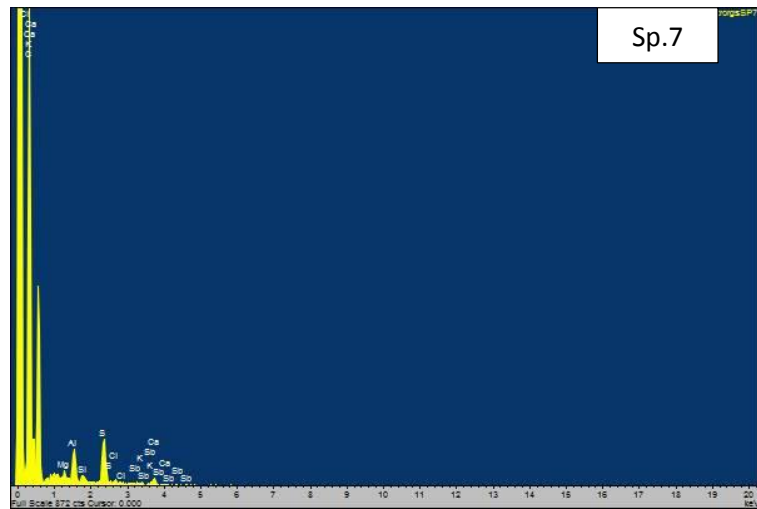
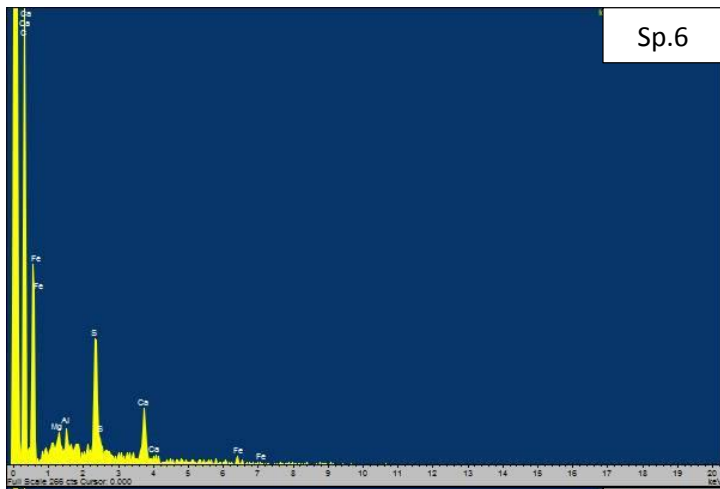
	Mifl					
	X400					
	sp.1	sp.2	sp.3	sp.4	sp.5	sp.6
	Weight %					
<b>C</b>	19.93	26.41	22.12	12.03	26.83	26.53
<b>N</b>						
<b>Na</b>						
<b>Mg</b>	0.20	0.09	0.57	1.41	0.05	0.10
<b>Al</b>	0.67	0.07	4.35	19.04	0.69	0.35
<b>Si</b>	0.41	0.07	0.22	0.03	0.11	0.23
<b>P</b>						
<b>S</b>	8.98	0.95	2.94	4.70	0.04	0.51
<b>Cl</b>					0.03	0.02
<b>Ca</b>	1.48	0.26	1.42	1.65	0.07	0.19
<b>Mn</b>		0.06	0.34			
<b>Ti</b>						
<b>Fe</b>						0.06
<b>Ag</b>				0.74		0.05
<b>Sb</b>				1.95		
<b>I</b>						0.05
<b>O</b>	68.32	72.10	68.38	58.14	72.25	72.04



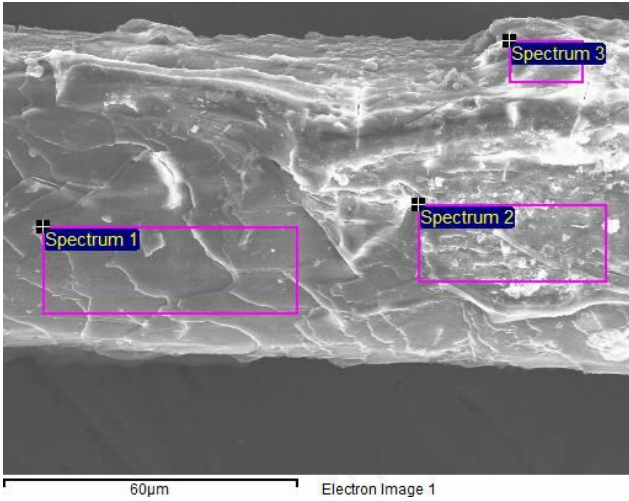


SEM/EDEX M1f1x1300



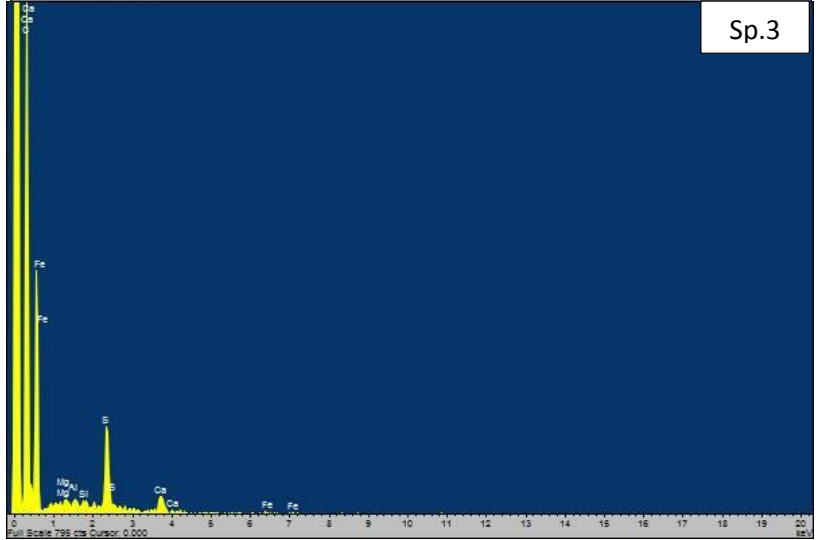
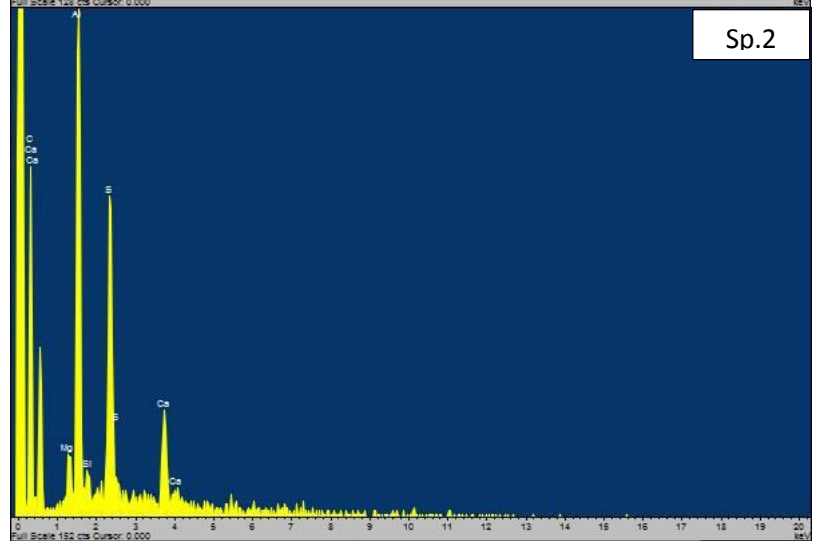
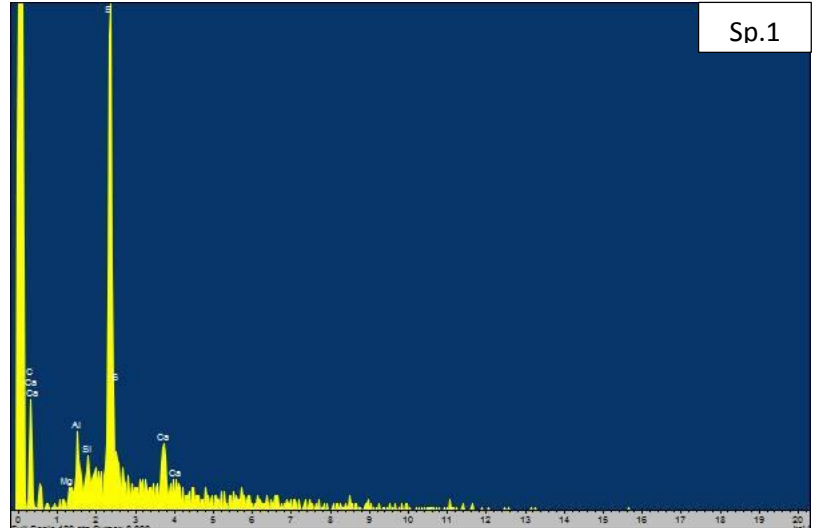


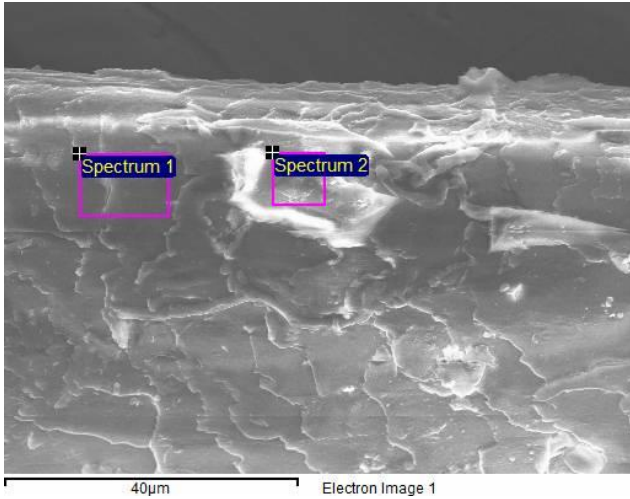
	Mifl							
	X1300							
	sp.1	sp.2	sp.3	sp.4	sp.5	sp.6	sp.7	sp.8
	weight%							
<b>C</b>	26.17	26.35	27.09	26.99	26.71	25.76	26.56	25.80
<b>Na</b>	0.14							
<b>Mg</b>	0.06	0.05			0.06	0.19	0.06	0.13
<b>Al</b>	1.26	0.38	0.30	0.13	0.22	0.16	0.34	0.39
<b>Si</b>							0.06	
<b>P</b>								
<b>S</b>	0.49	0.88		0.31	0.51	1.44	0.58	1.62
<b>Cl</b>			0.09				0.04	0.09
<b>K</b>							0.02	
<b>Ca</b>	0.09	0.14	0.04	0.07	0.11	0.83	0.12	0.25
<b>V</b>			0.03					
<b>Fe</b>							0.18	
<b>Sb</b>		0.12					0.11	
<b>I</b>		0.08		0.06				
<b>Pb</b>					0.17			



SEM/EDEX analysis M1f1x1000

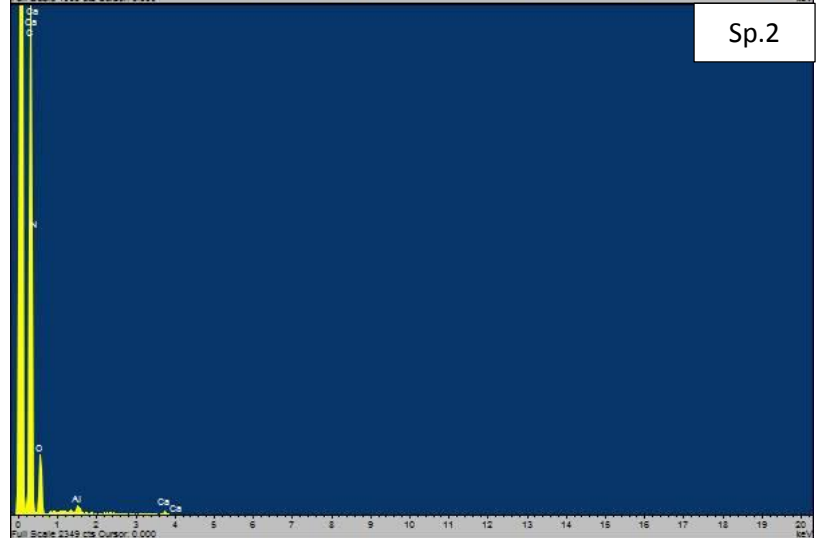
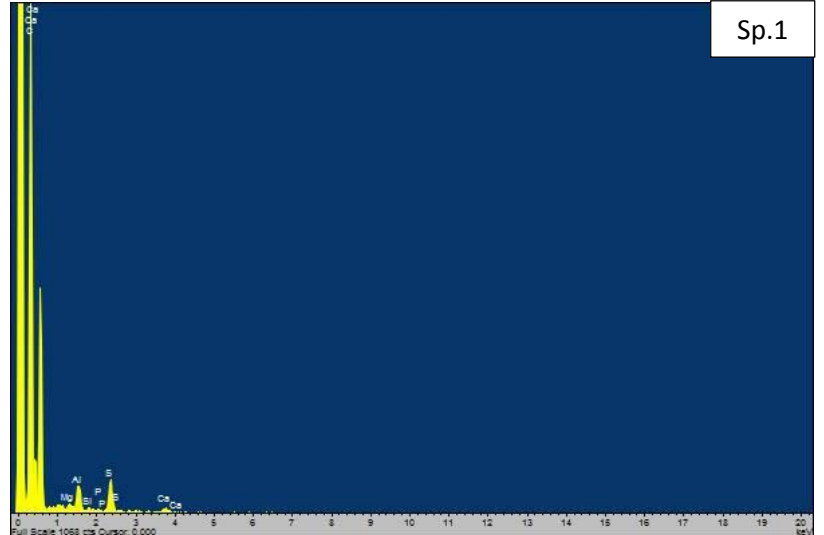
<b>M1f1</b>			
<b>X1000</b>			
	sp.1	sp.2	sp.3
<b>Weight %</b>			
<b>C</b>	24.23	26.41	26.80
<b>N</b>			
<b>Na</b>			0.05
<b>Mg</b>	0.17	0.18	0.06
<b>Al</b>	1.62	0.33	0.66
<b>Si</b>	0.16	0.08	0.12
<b>P</b>			
<b>S</b>	2.78	0.66	0.01
<b>Cl</b>			0.03
<b>Ca</b>	0.41	0.25	0.04
<b>Mn</b>			
<b>Ti</b>		0.02	
<b>Fe</b>			
<b>Ag</b>		0.13	
<b>Sb</b>			
<b>I</b>			
<b>O</b>	70.62	71.96	72.21

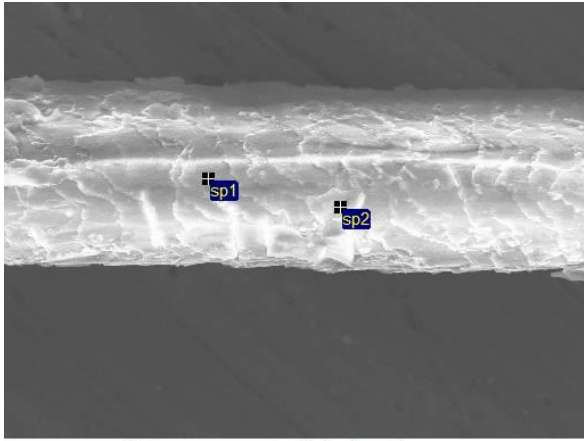




SEM/EDEX M1f1x1500

M1f1		
	X1500	
	sp.1	sp.2
Weight %		
C	26.78	23.15
N		3.89
Na		
Mg	0.07	
Al	0.27	0.07
Si	0.03	
P	0.03	
S	0.41	
Cl		
Ca	0.07	0.05
Mn		
Ti		
Fe		
Ag		
Sb		
I		
O	72.34	72.85

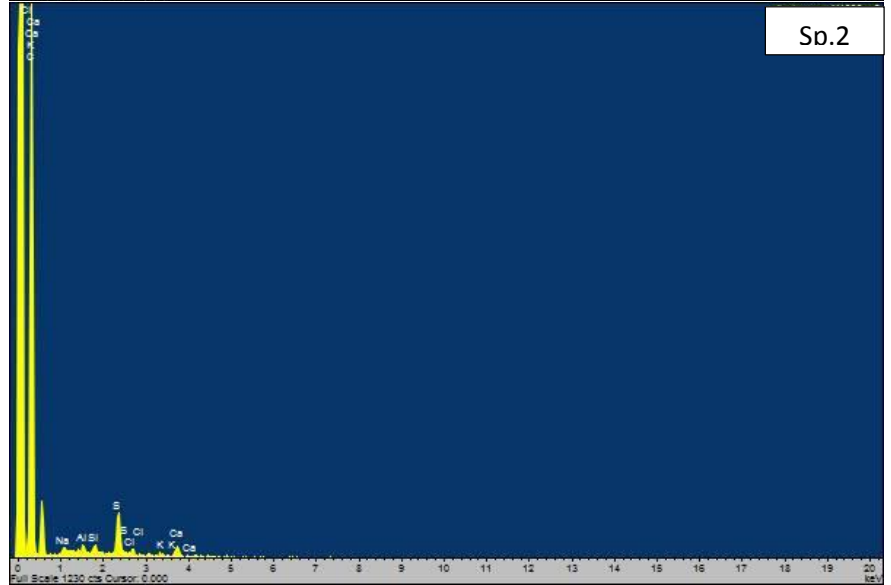
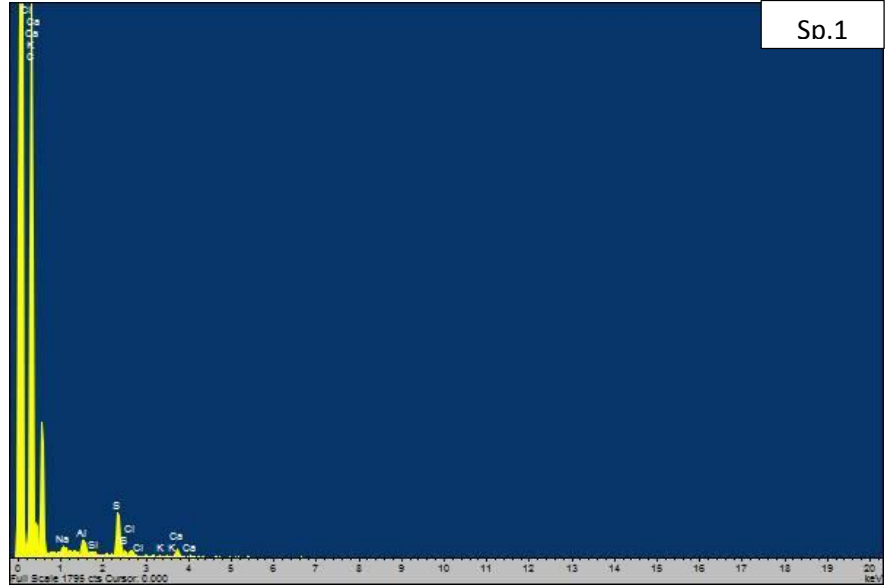


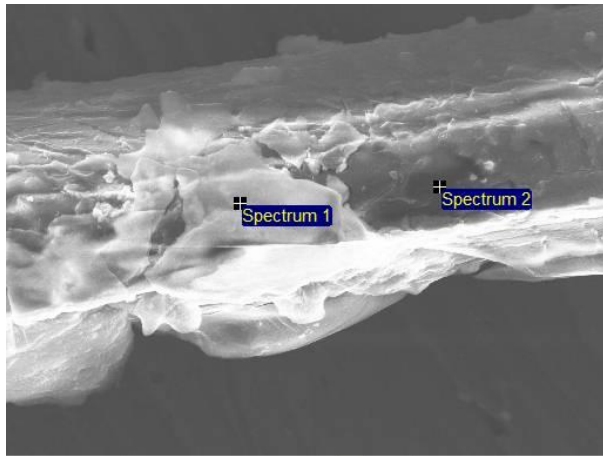


60µm Electron Image 1

**SEM/EDEX M1f3x1000**

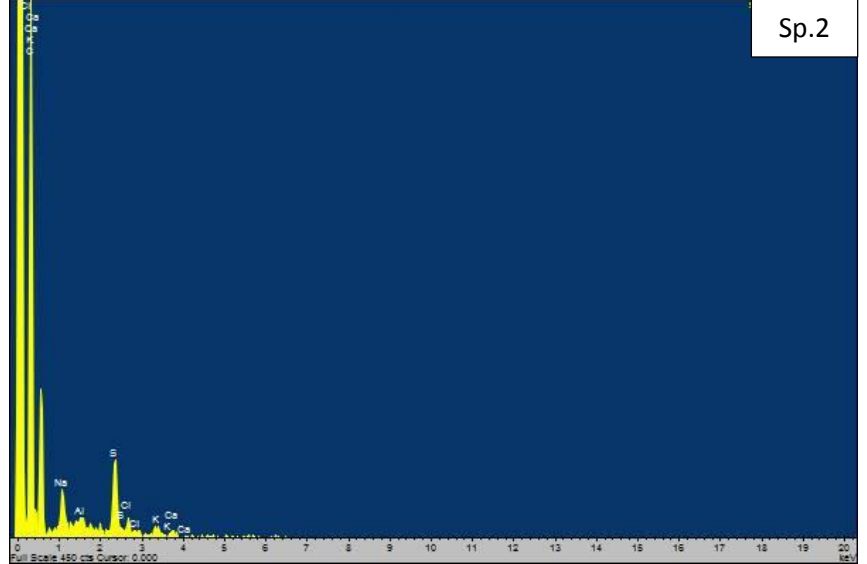
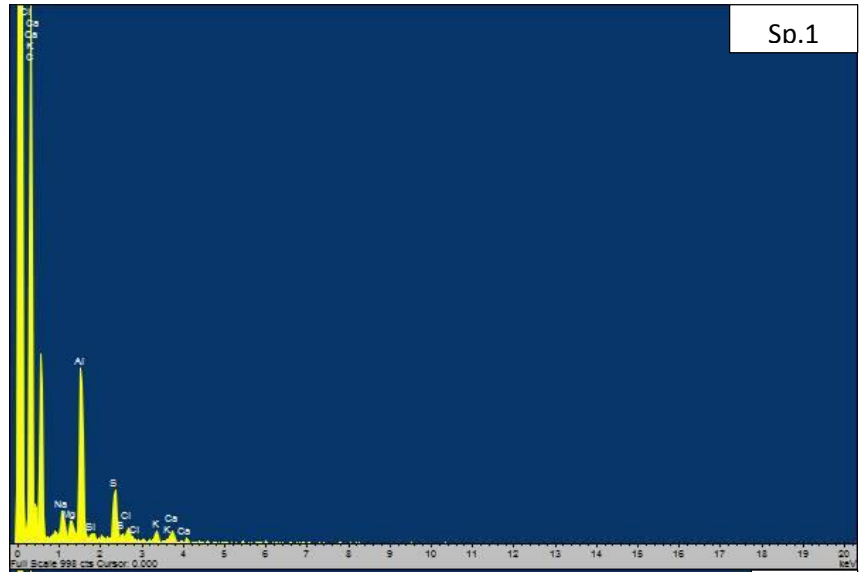
	<b>M1f3</b>	
	<b>X1000</b>	
	<b>sp.1</b>	<b>sp.2</b>
	<b>weight%</b>	
<b>C</b>	26.82	26.81
<b>Na</b>	0.11	0.08
<b>Mg</b>		
<b>Al</b>	0.13	0.06
<b>Si</b>	0.02	0.07
<b>S</b>	0.44	0.45
<b>Cl</b>	0.05	0.07
<b>K</b>	0.01	0.03
<b>Ca</b>	0.10	0.13
<b>O</b>	72.32	72.31

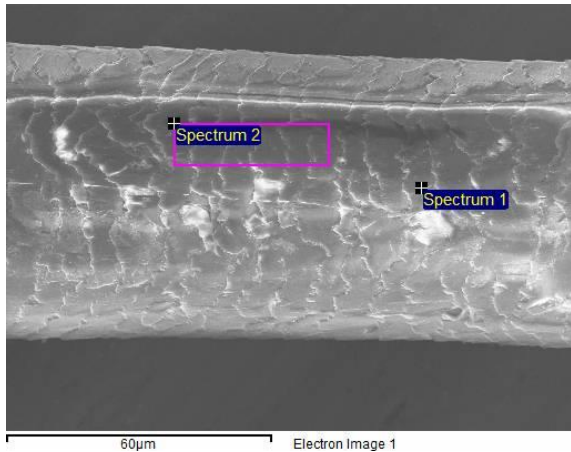




**SEM/EDEX M1f3x1400**

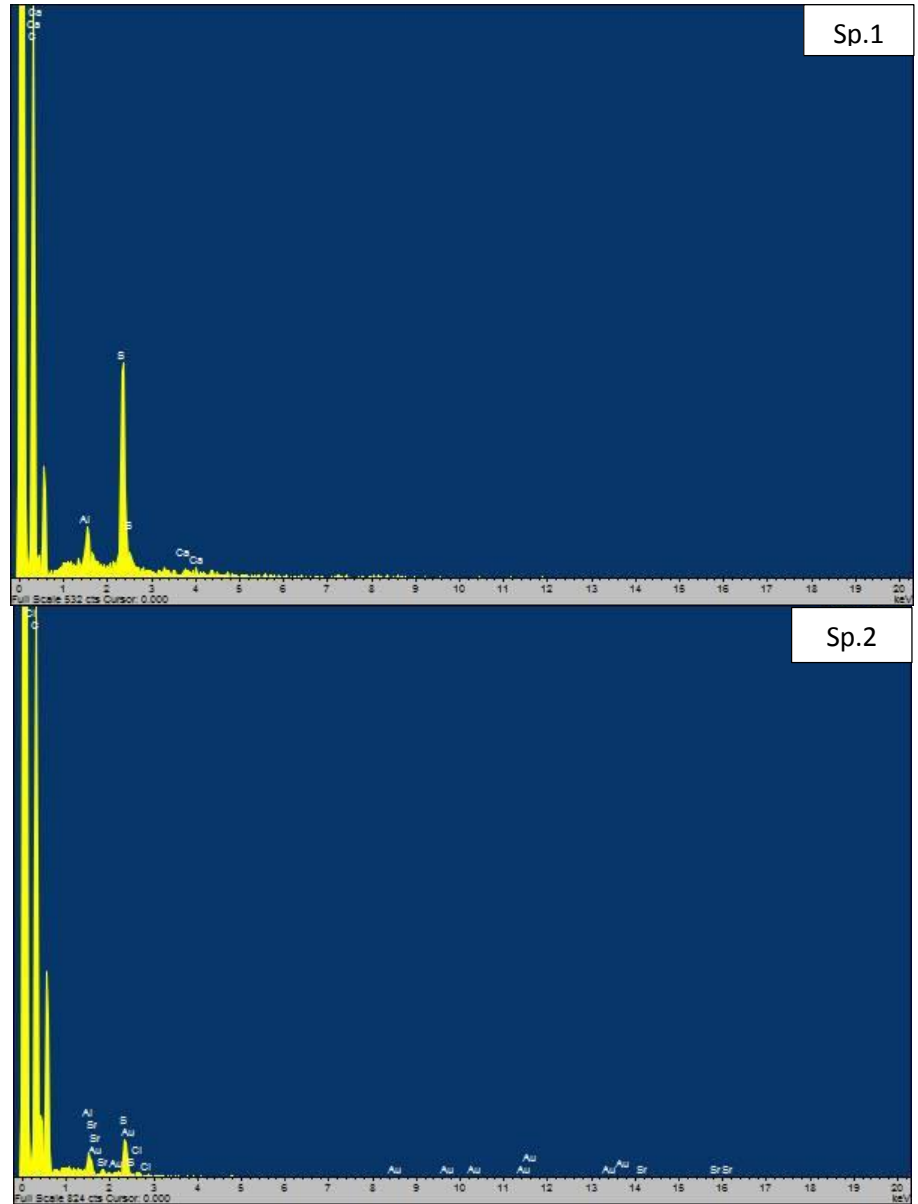
<b>M1f3</b>		
	<b>X1400</b>	
	<b>sp.1</b>	<b>sp.2</b>
	<b>weight%</b>	
<b>C</b>	25.84	26.19
<b>Na</b>	0.56	0.99
<b>Mg</b>	0.14	
<b>Al</b>	1.46	0.09
<b>Si</b>	0.04	
<b>S</b>	0.50	0.97
<b>Cl</b>	0.11	0.19
<b>K</b>	0.11	0.12
<b>Ca</b>	0.15	0.09
<b>O</b>	71.24	71.62



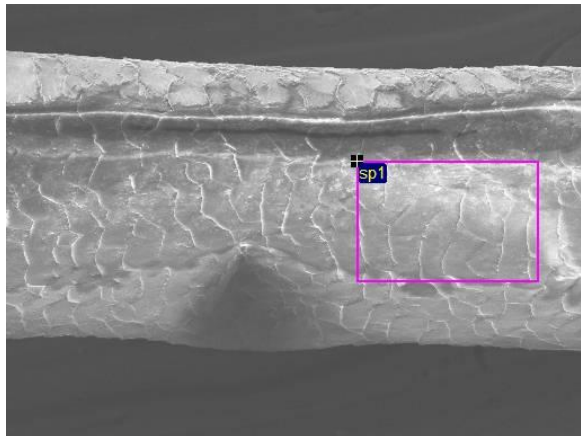


SEM/EDEX modern1 (male) x1100

	Modern1	
	X1100	
	Sp.1	Sp.2
	weight%	
<b>C</b>	25.75	26.74
<b>Al</b>	0.31	0.20
<b>S</b>	2.00	0.53
<b>Cl</b>		0.05
<b>Ca</b>	0.06	
<b>Sr</b>		0.08
<b>Au</b>		0.17
<b>O</b>	71.88	72.23

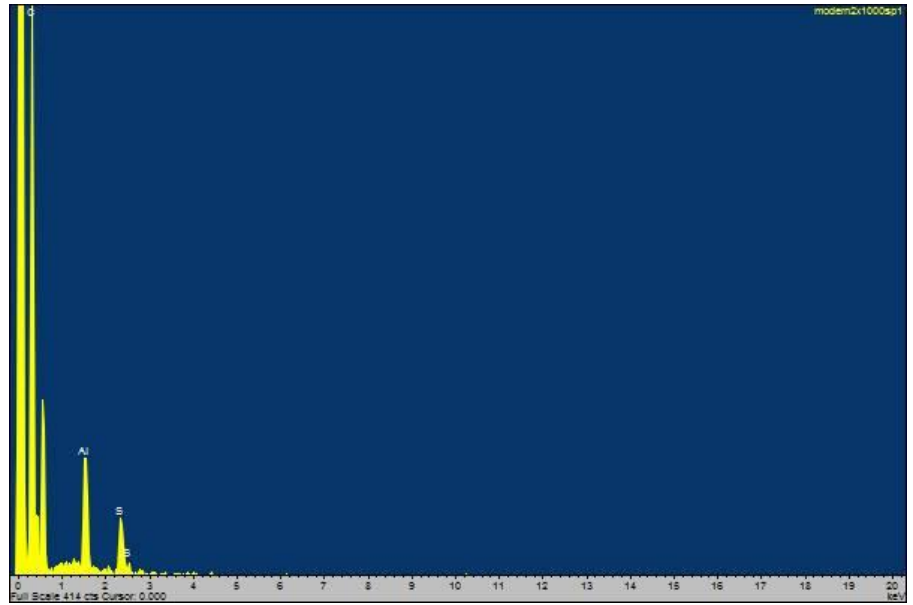






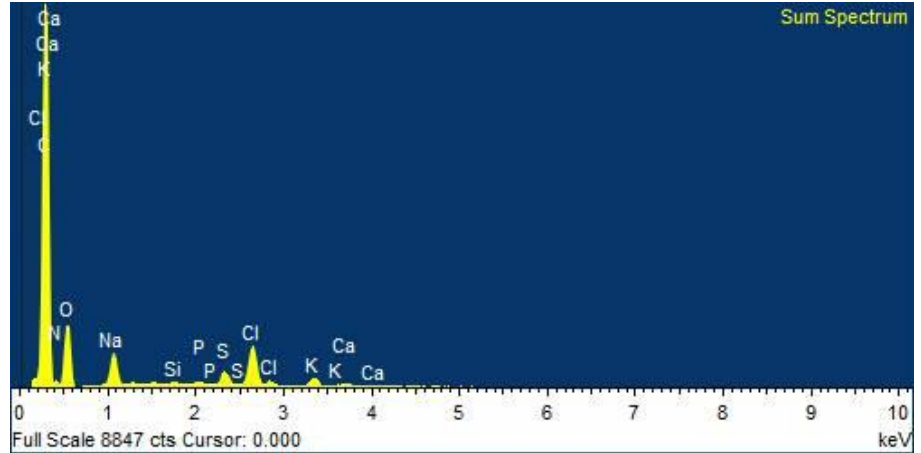
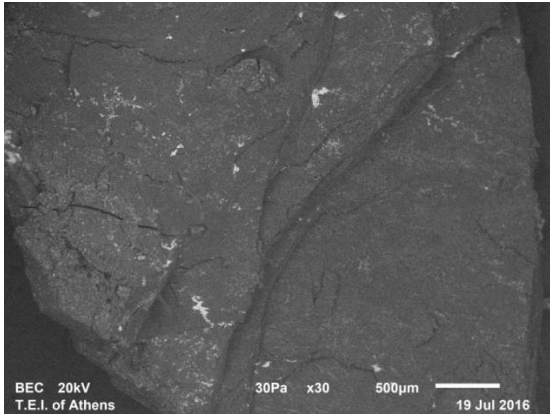
SEM/EDEX Modern2(female)X1000

	<b>Modern 2</b>
	X1000
	Sp.1
	<b>weight%</b>
<b>C</b>	26.28
<b>Al</b>	1.16
<b>S</b>	0.60
<b>Cl</b>	
<b>Ca</b>	
<b>Sr</b>	
<b>Au</b>	
<b>O</b>	71.95

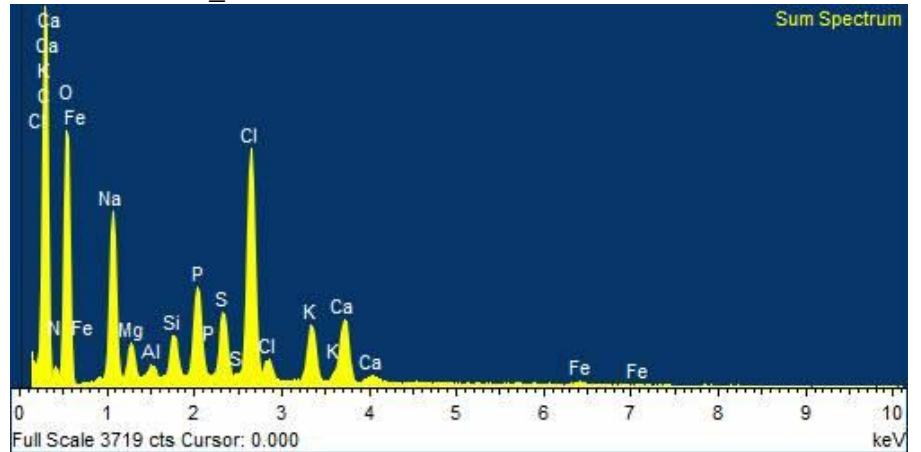
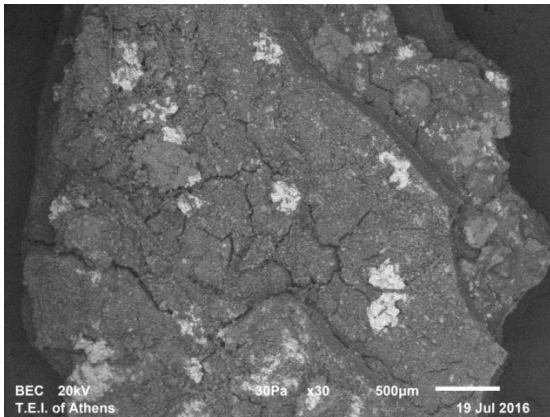


**SEM/EDEX tissue:** The fragments of tissue are presented cracked and exfoliated. Particles of degraded materials dust and dirty are on the surface. The predominant elements were determined are C, N, Na and Cl. Less dominant are Ca, S, P, Mg, Si and K. Small amounts of Al and Fe also was determined. The percentages of the elements are presented on the Table 13.

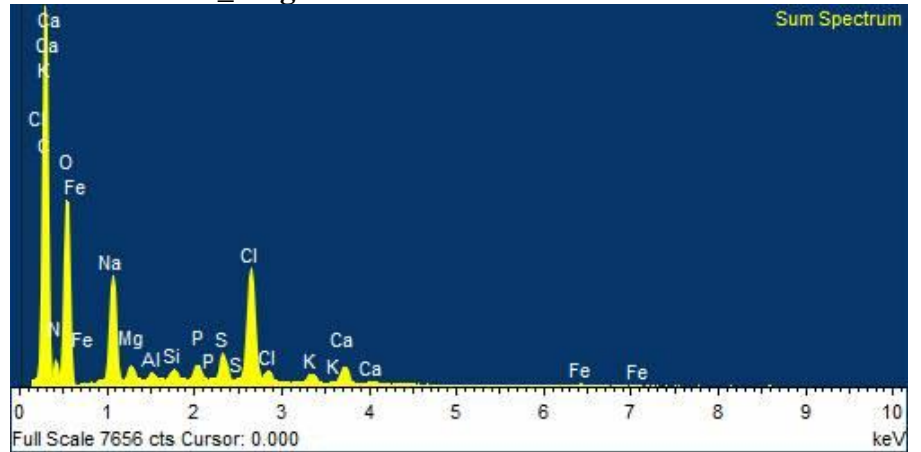
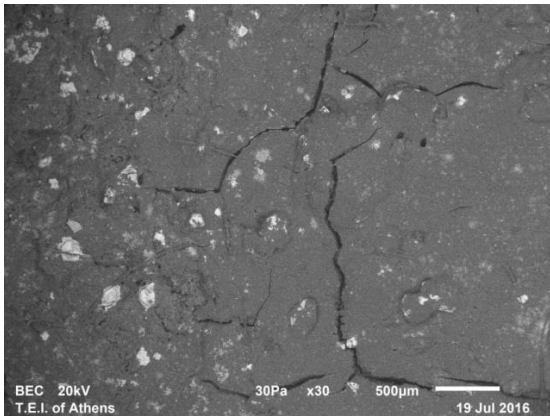
**SEM/EDEX\_M1d1**



**SEM/EDEX\_M1d2**



**SEM/EDEX\_M2g**

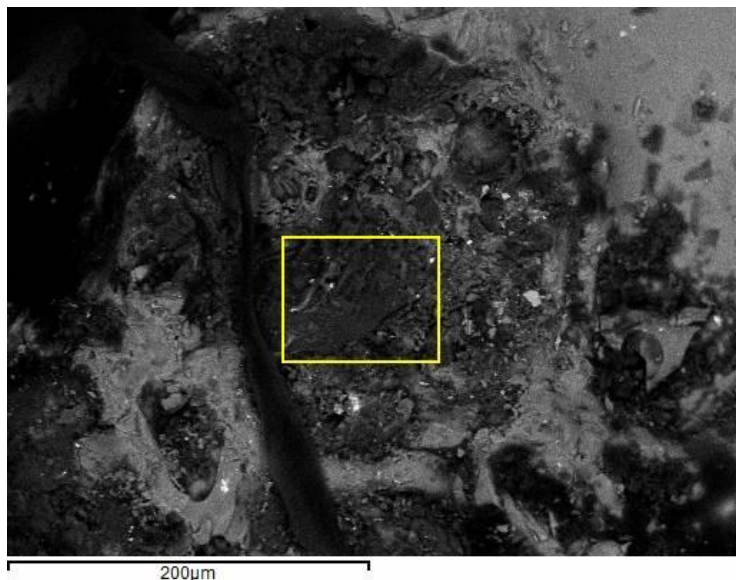


	Tissue		
	M1d bulk1	M1d bulk2	M2g
	<b>Atomic%</b>		
<b>C</b>	69.60	54.89	54.61
<b>N</b>	10.77	10.03	13.87
<b>Na</b>	0.89	2.26	1.91
<b>Mg</b>		0.34	0.16
<b>Al</b>		0.09	0.06
<b>Si</b>	0.02	0.29	0.09
<b>P</b>	0.04	0.70	0.15
<b>S</b>	0.20	0.48	0.24
<b>Cl</b>	0.65	1.78	1.11
<b>K</b>	0.14	0.44	0.10
<b>Ca</b>	0.06	0.54	0.18
<b>Fe</b>		0.05	0.03

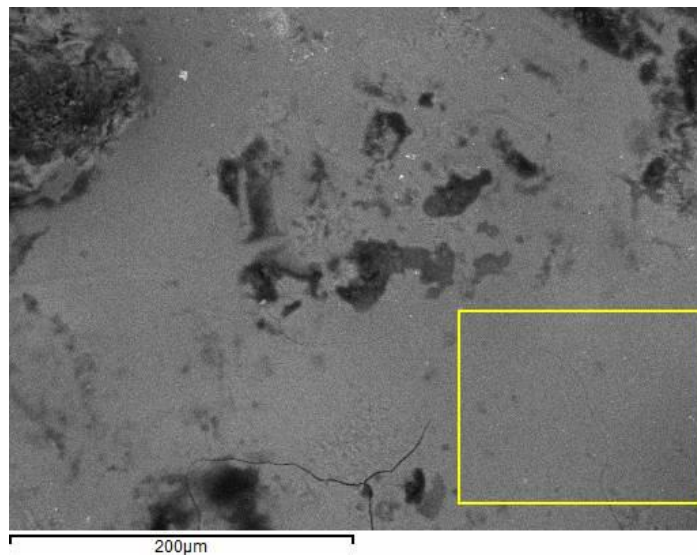
**Table 13**

#### 4.5.4 Black and yellow beads (M2a1, M2a2)

The predominant oxides of both samples are  $\text{SiO}_2$  and  $\text{Na}_2\text{O}$ . Less dominant are  $\text{CaO}$  and  $\text{K}_2\text{O}$ . Amount of  $\text{MnO}$  and  $\text{CuO}_2$  were determined in M2a1 black bead.  $\text{PbO}$  and  $\text{Sb}_2\text{O}_5$  were determined in M2a2 yellow bead. The results of elements are given to the Table 14.



**SEM image: M1a1**



**SEM image: M1a2**

**Table 14**

<b>Sample</b>	<b>M2a1 black bead cleaned surface</b>	<b>M2a1 black bead corroded surface</b>	<b>M2a2yellow bead cleaned surface</b>	<b>M2a2 yellow bead corroded surface</b>
<b>Na<sub>2</sub>O</b>	11.1	6.87	1.98	2.97
<b>MgO</b>	0.08	0.31	0.06	0.29
<b>Al<sub>2</sub>O<sub>3</sub></b>	0.4	1.12	0.54	0.94
<b>SiO<sub>2</sub></b>	69.22	68.60	69.33	82.53
<b>P</b>	n.d.	0.60	n.d.	n.d.
<b>SO<sub>3</sub></b>	0.23	0.76	n.d.	0.97
<b>Cl<sub>2</sub>O</b>	1.45	1.05	2.01	0.87
<b>K<sub>2</sub>O</b>	2.94	2.20	1.52	1.11
<b>CaO</b>	1.17	7.19	0.89	3.90
<b>TiO<sub>2</sub></b>	0.28	n.d.	0.08	0.15
<b>MnO</b>	1.77	1.19	n.d.	n.d.
<b>Fe<sub>2</sub>O<sub>3</sub></b>	0.12	0.71	0.56	1.28
<b>CuO</b>	9.7	7.84	0.14	n.d.
<b>Sb<sub>2</sub>O<sub>5</sub></b>	n.d.	n.d.	0.20	0.98
<b>PbO</b>	1.56	0.42	4.00	4.00
<b>Total</b>	100	100	100	100

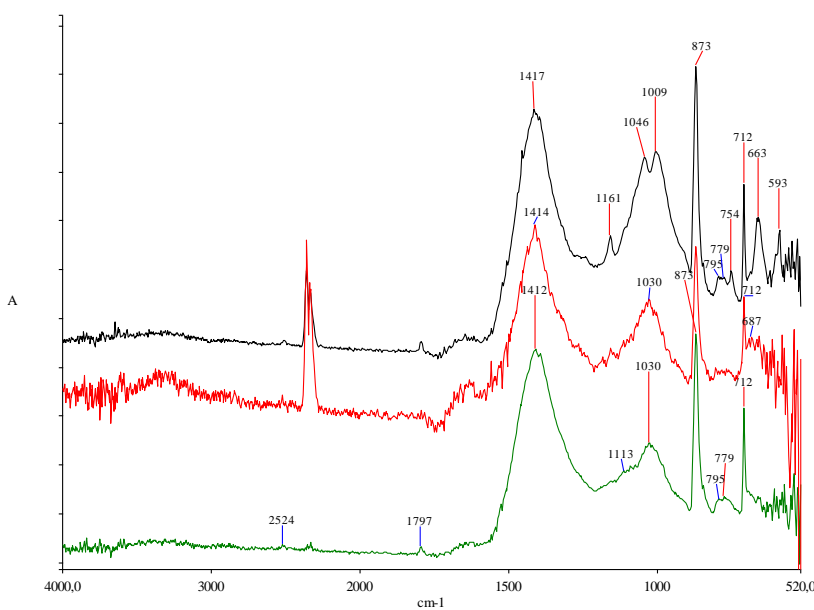
#### **4.6 Attenuated total reflectance (ATR) and Fourier Transform Infrared spectroscopy (FTIR) analysis**

##### **4.6.1 Fragments of the bandages M1b, M1c, M1h, M2e, M2f**

The characteristic bands of cuprorivaite, calcite and silicon oxide were determined in sample M1b (plaster and color). In blue area the peaks 1161, 1050, 1009, 754, 663 and 593 correspond to the stretching bonds Si-O-Si in Cuprorivaite. In the red area, peaks of iron (crystalline) and iron oxides were determined ( $540-550\text{cm}^{-1}$ ,  $540^{-1}$ ). Esters (1387, 1168, 111, 1030) and fatty acids absorbance bands were extracted from M1b in  $\text{Cl}_2\text{Cl}_2$  as binder material. Bands of shelloic and jalaric acid and esters were found in M1c (2927, 2854, 1732, 1715, 1607, 1464, 1379, 1241, 1181) and M2e (2928, 2855, 1715, 1607, 1463, 1380, 1239, 1179). After extraction with hexane of M1c picks of wax compounds were observed. Characteristic bands of cellulosic compounds were determined in samples M1h and M2f. Bands of  $\text{CaCO}_3$  and silicates were determined in M1h. Protein material and fatty material in degraded state (due to 1644) in M2f, were identified.

M1b fragment of bandages (plaster and color)

M1b –  
fragment of  
bandages; area  
with blue and  
white  
pigments;  
**ATR  
spectrum**

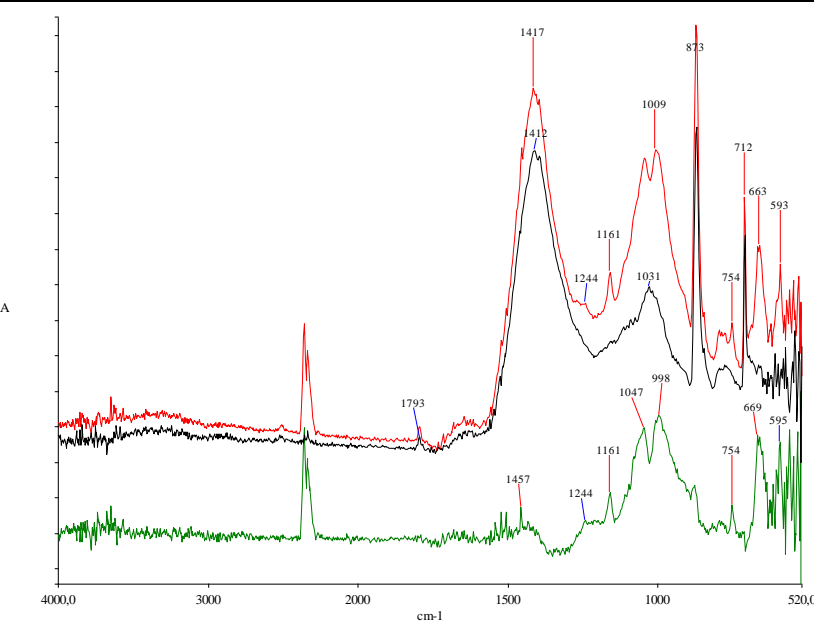


**AK10:** area with blue pigments  
1417, 873, 712 calcite  
1146, 791, 779 silicon oxide  
(quartz or flint)  
1161, 1046, 1010, Cuprorivaite  
663, 593?

**AK13:** area with white pigments  
1412, 873, 712 calcite  
1113  
1030, 791, 779 silicates

**AK14:** area with red pigments  
540-550 $\text{cm}^{-1}$  iron (crystal),  
540 $^{-1}$  iron oxides

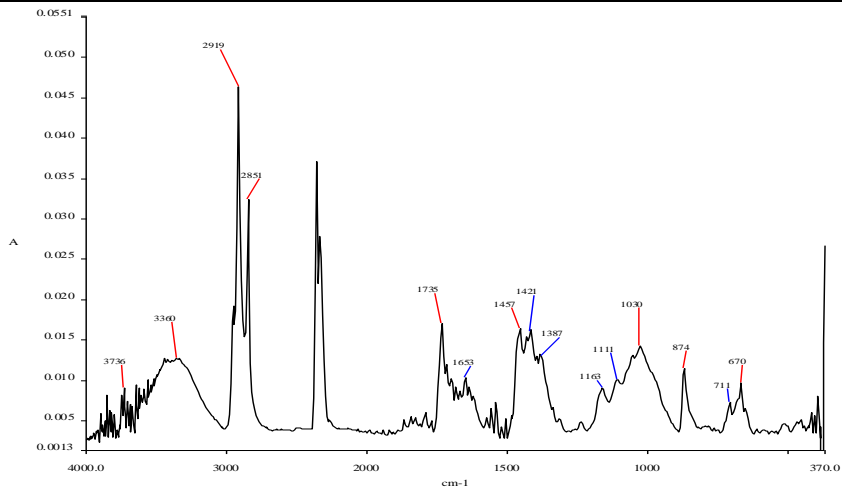
M1b –  
fragment of  
bandages; area  
with blue and  
white  
pigments;  
difference  
spectrum  
**ATR  
spectrum**



**AK13:** area with white pigments  
**AK10** area with blue pigments  
1417, 873, 712 calcite  
1161?  
791, 779 silicon oxide (quartz or  
flint)  
**1161, 1050, 1009**,  $\nu_{\text{asym}}$  Si-O-Si  
in Cuprorivaite (Egyptian Blue)  
**754, 663, 593**  $\nu_{\text{sym}}$  Si-O-Si in  
Cuprorivaite (Egyptian Blue)

**AK10d:** spectrum after  
subtraction: AK10 – AK13  
1244, 1161, 1047, 998  $\nu_{\text{asym}}$  Si-  
O-Si in Cuprorivaite (Egyptian  
Blue)  
754, 669, 595  $\nu_{\text{sym}}$  Si-O-Si in  
Cuprorivaite

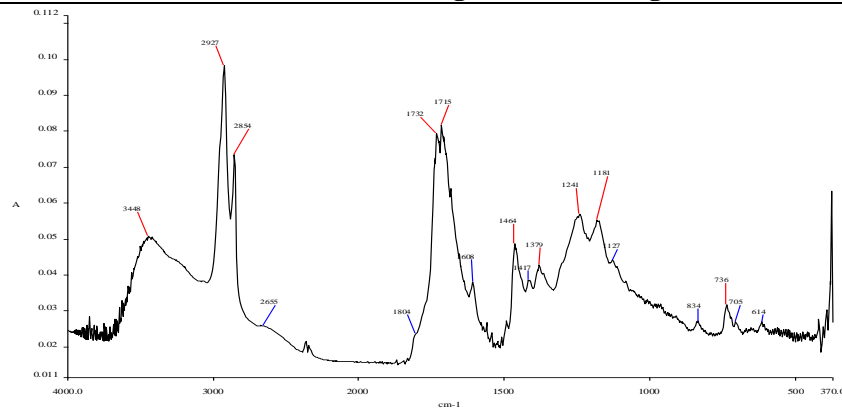
M1b –  
fragment of  
bandages:  
area with  
color;  
checking for  
binder  
through  
extraction  
with CH<sub>2</sub>Cl<sub>2</sub>;  
KBr  
**FTIR  
spectrum**



AK65: area with color  
pigments  
2919, 2851, 1735, 1457,  
1387, 1168, 111, 1030 ester  
1710, 1421 fatty acid  
1653 vC=C double bond

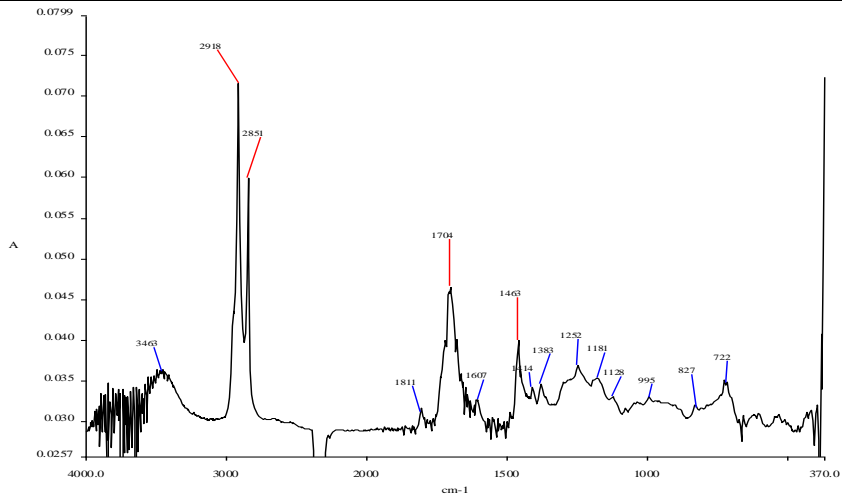
**M1c fragment of bandages**

M1c -  
Extraction in  
CH<sub>2</sub>Cl<sub>2</sub> KBr  
**FTIR  
spectrum**



AK39:  
2927,2854,1732,1715,  
1607,1464,1379,1241,1181  
Shelloic and jalaric acid and  
esters,.

M1c -  
Extraction in  
hexane KBr  
**FTIR  
spectrum**



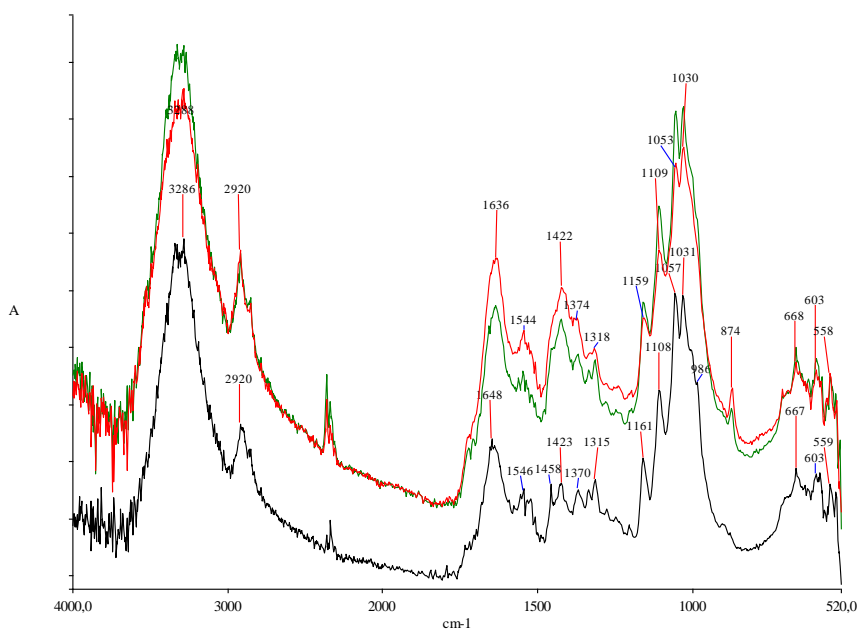
AK70:  
2918, 2881, 1704, 1458,  
1414, 1388, 722 wax



M1h fragment of bandages (wrapped fabric)

M1h –  
fragment of  
bandages

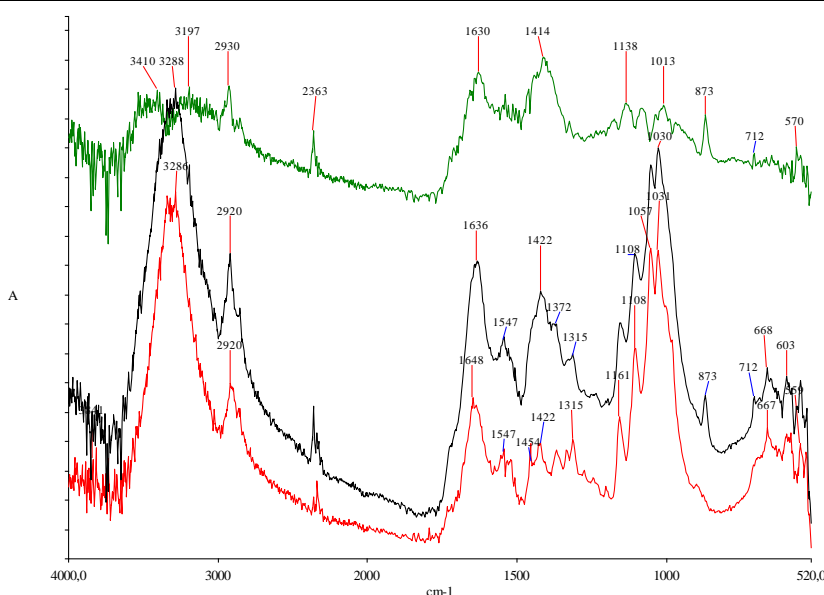
ATR  
spectrum



AK02: face A;  
AK06: face B;  
AK07: face B (different scan)  
Cellulosic textile  
1732  $\nu(\text{C}=\text{O})$  galacturonic esters (in pectin),  
1644, 1624  $\text{H}_2\text{O}$ ,  
1546, 1511  
1423  $\delta_s(\text{CH}_2)$  cellulose,  
1370  $\delta(\text{CH}_2)_w$   
1334, 1315  $\delta_s(\text{CH}_2)$  cellulose  
1264  $\nu(\text{C}-\text{O})$  pectin acids  
1249  $\nu(\text{C}-\text{O})$  pectin acids  
1161  $\nu_{as}(\text{C}-\text{O}-\text{C})$   $\beta(1\rightarrow4)$  glucosidic linkage  
1108  $\nu(\text{C}-\text{O})$  *II*-vibration of 6-membered ring /  $\nu(\text{C}^2-\text{O}-\text{H})$   
**K**  
1057,  $\nu(\text{C}^3-\text{O}-\text{H})$   
1031, 1015, 1000(sh)  $\nu(\text{C}^6-\text{O}-\text{H})$   
989  $\nu(\text{C}-\text{O})$  &  $\nu(\text{C}-\text{C})$  glucose 6-membered ring  
897  $\delta(\text{C}1-\text{H})$  [ $\beta$ -anomeric linkage in cellulose]  
839  $\delta(\text{C}1-\text{H})$  [ $\alpha$ -anomeric linkage in pectin]  
785 breathing vibration of 6-membered ring

M1h –  
fragment of  
bandages

ATR  
spectrum

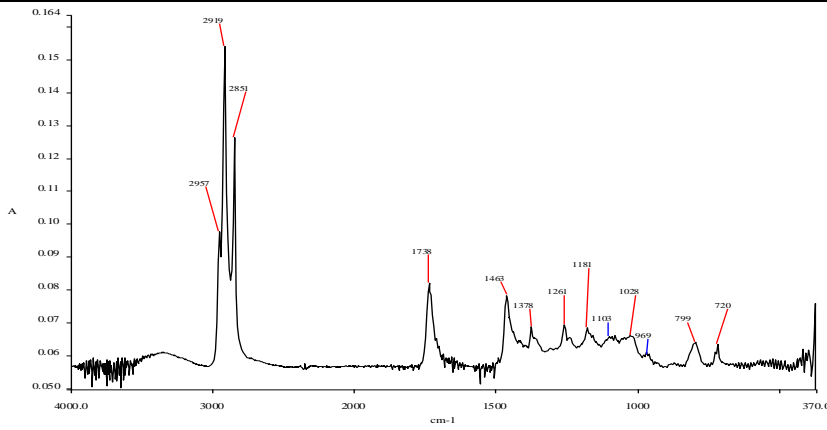


AK02: Face A  
AK06: Face A  
AK06 $\alpha$ : spectrum after subtraction (AK06 – AK02)  
CaCo deposits  
Silicates (soil materials)

M1h –  
fragment of  
bandages

Extraction in  
CH<sub>2</sub>Cl<sub>2</sub> KBr  
(repeated  
extractions to  
increase  
quantity)

**FTIR  
spectrum**

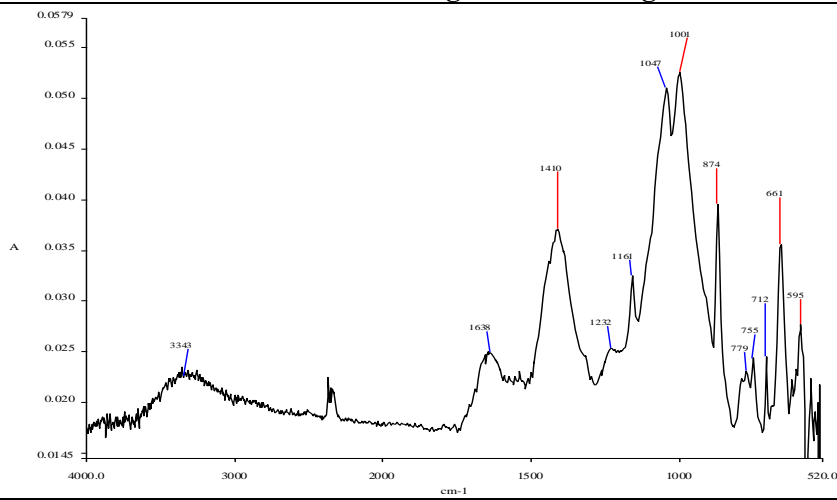


AK38:  
2957, 2919, 2851, 1738,  
1463, 1378, 1261, 1181,  
1108, 799, 720: fatty ester

**M2d fragment of bandages**

M2d-  
Fragment of  
bandages

**ATR-FTIR  
spectrum**



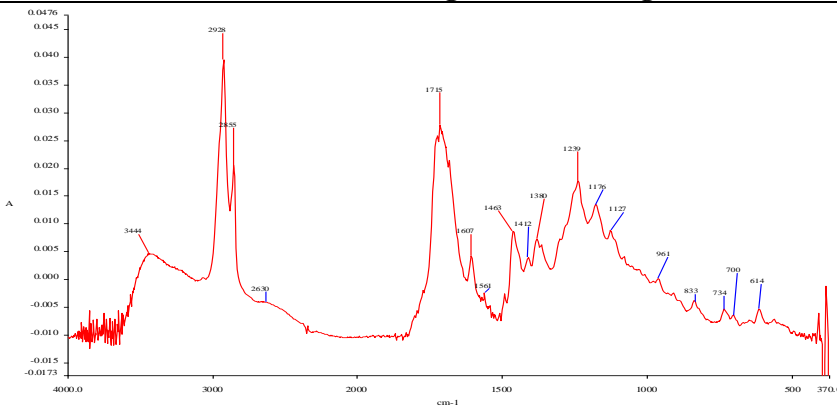
AK54:  
1410, 874, 712 CaCO<sub>3</sub>  
1047, 1001, 661 silicate  
material

**M2e fragment of bandages**

M2e –  
Fragment of  
bandages

Extraction in  
CH<sub>2</sub>Cl<sub>2</sub>

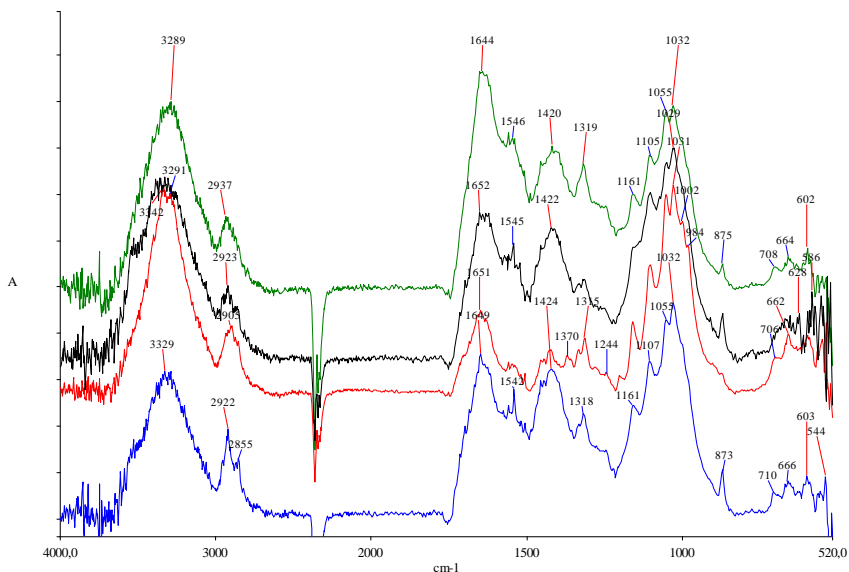
**FTIR  
Spectrum**



AK41:  
2928, 2855, 1715, 1607,  
1463, 1380, 1239, 1179  
Shelloic and jalaric acid and  
esters

**M2f fragment of bandages**

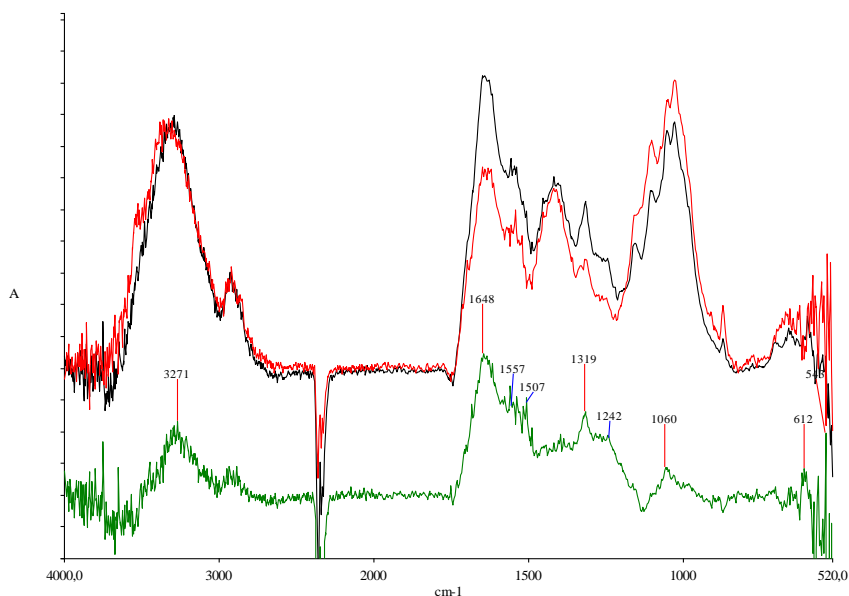
M2f – fragment of bandages  
ATR-FTIR spectrum



**AK22**: main fragment;  
**AK18** :fragment 1;  
**AK19**: fragment 2;  
**AK20** :fragment 2 (backface)

2922, 2855, 1649, 1161, 1105, 1055, 1032 cellulosic material  
1424, 873, 710 CaCO<sub>3</sub>  
3329, 1454, 1318: protein material

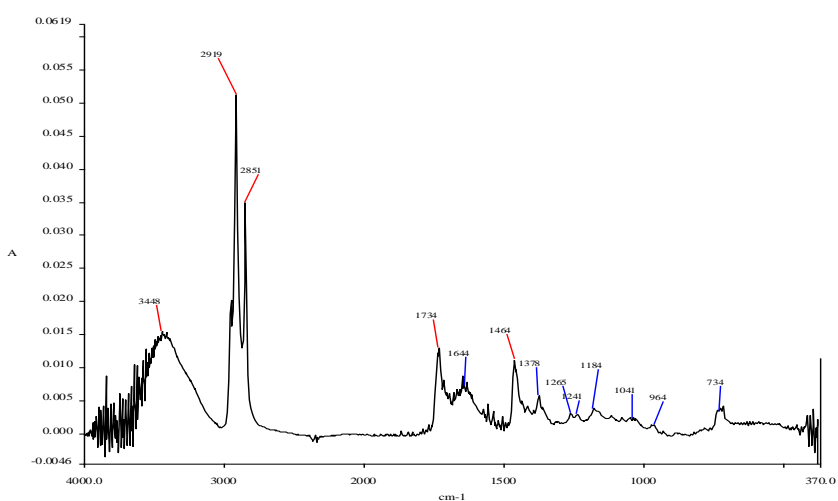
M2f – fragment 2: difference  
ATR-FTIR spectrum



**AK19**: fragment 2  
**AK20** fragment 2 (backface)

**AK20d** difference spectrum  
1648, 1557, 1319, 1242: protein material

M2f – fragment of bandages  
Extraction in CH<sub>2</sub>Cl<sub>2</sub> KBr  
FTIR spectrum



**AK48**:  
2919, 2881, 1734, 1644, 1464, 1378, 1265, 1378, 1184, 734 fatty material in degraded state

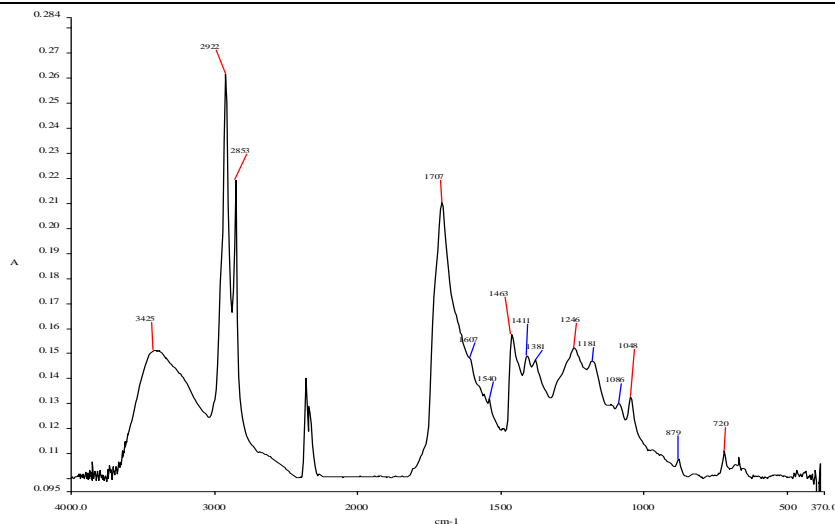
#### 4.6.2 Piece of balm M1g

M1g extraction in ethanol, peaks of mainly carboxylic acids were determined (1707, 1411, 1245). Wax components also were extracted (2922, 2853, 723). The peaks 3425  $\nu$ OH, 1607  $\nu$ C=C, 1468  $\delta$ CH<sub>2</sub>+ $\delta_{as}$ CH<sub>3</sub>, 1381  $\delta_s$ CH<sub>3</sub> were been presented. Ester and alcohols picks, 1181, 1048  $\nu$ C-O-C were also determined.

M1g –  
Piece of balm

Extraction in  
ethanol

**FTIR  
spectrum**



AK59:

2922, 2858, 1707, 1607(sh),  
1463, 1411, 1381

3425  $\nu$ OH

2922, 2853  $\nu$ C-H (mainly in  
wax component)

2640  $\nu$ OH (O-H ... O-H in  
dimeric carboxylic acids,  
very weak in extract)

1707  $\nu$ C=O carboxylic acids  
(main peak due to shelloic  
ester hydrolysis)

1607  $\nu$ C=C

1468  $\delta$ CH<sub>2</sub>+ $\delta_{as}$ CH<sub>3</sub>

1411  $\delta$ C-O-H (carboxylic  
acids)

1381  $\delta_s$ CH<sub>3</sub>

1245  $\nu$ (C-O),  $\delta_{i,p}$ (C-O-H)  
(carboxylic acids)

1181, 1048  $\nu$ C-O-C in ester  
links and alcohols (due to  
ester hydrolysis)

942  $\delta_{o.o.p.}$  C-O-H (O-H ... O-  
H) of dimeric carboxylic  
acids (very weak in extract)

723  $\rho$ CH<sub>2</sub> (long hydrocarbon  
chains in wax component)

#### 4.6.3 Tissue M1d, (M1d $\mu$ ), M2g and hairs M1f (M1f02), control sample-modern tissue M $\pi\delta$ 1

**Tissue:** Fatty acids picks were mainly determined in M1d and M2g ATIR spectrum. Absorbance picks of 1625 amide I ( $\nu$ C=O with small contributions from  $\nu$ C-N and  $\delta$ N-H), 1545 amide II ( $\nu$ C-N with contributions from  $\delta_{ip}$ N-H), 1454  $\nu$ CH<sub>2</sub>, 1379  $\delta$ NH<sub>2</sub> and/or  $\delta$ COOH (glutamic), and 1231 amide III ( $\nu$ C-N +  $\delta$ N-H with contributions from  $\delta$ C-H), were determined. Free fatty acids (2925, 2858, 1710, 1465, 1411, 1375, 722), and fatty esters (2925, 2858, 1736, 1462, 1382, 722) picks were been presented in M1d extraction with Cl<sub>2</sub>Cl<sub>2</sub>. Possibly natural resin peaks (2927, 2855, 1726, 1719, 1463, 1380, 1246, 1175, 721) were determined in M2g extraction with Cl<sub>2</sub>Cl<sub>2</sub>. Protein material (3239, 2932, 1665 (amide I), 1572 (amide II, shoulder), 1457, 1405, 1257) in M2g extraction with H<sub>2</sub>O was distinct. Traces of cellulosic material peaks were also observed in both ancient samples (1155, 1109, 1070, 1027 and 1072, 1032).

ATR spectrum of modern tissue M1 $\pi\delta$ , peaks 3282 amide A ( $\nu$ N-H), 3080 amide B (Second component of  $\nu$ N-H in Fermi resonance with the amide II overtone), 1732 fatty ester, 1625 amide I ( $\nu$ C=O with small contributions from  $\nu$ C-N and  $\delta$ N-H), 1545 amide II ( $\nu$ C-N with contributions from  $\delta_{ip}$ N-H), 1454  $\nu$ CH<sub>2</sub>, 1379,  $\delta$ NH<sub>2</sub> and/or  $\delta$ COOH (glutamic), 1231 amide III ( $\nu$ C-N +  $\delta$ N-H with contributions from  $\delta$ C-H), were determined. The peaks 3430  $\nu$ O-H (fatty acid), 1600  $\nu$ C=O (carboxylic salt or amide I of short chain peptide), 1525 amide II (short chain peptide), 1411  $\delta$ C-O-H (carboxylic acids), 1348  $\delta$ NH<sub>2</sub>, 1128, and 1048  $\nu$ C-O were observed in extraction with H<sub>2</sub>O.

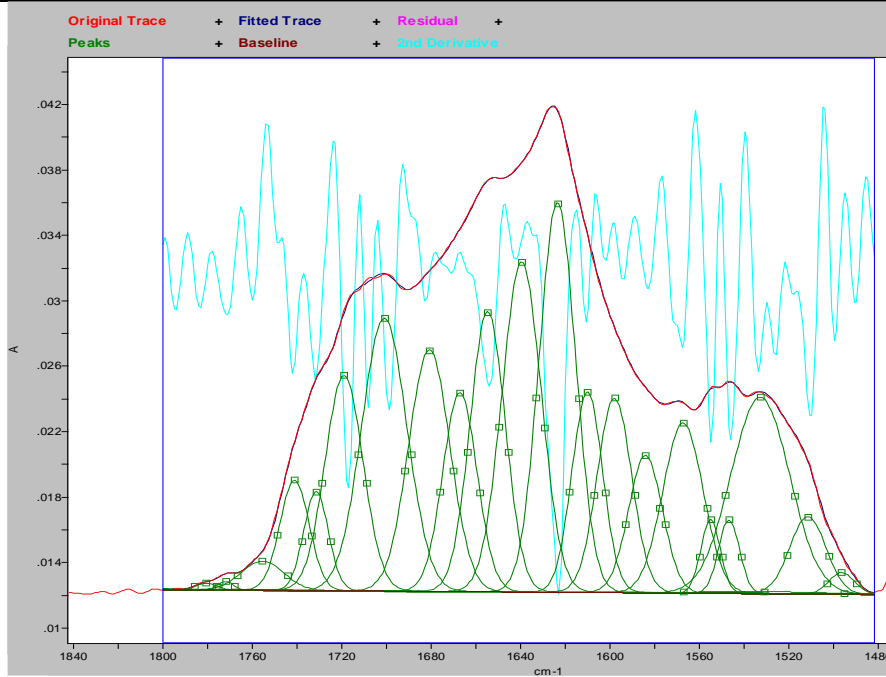
**Hairs:** Extraction with H<sub>2</sub>O of M1f, peaks 2958, 2919, 2851  $\nu$ C-H (hydrocarbon chains), 1734  $\nu$ C=O (ester carbonyl), 1464  $\delta$ CH<sub>2</sub>, 1379  $\delta$ CH<sub>3</sub> (umbrella-type symmetric bending), 1238, 1179  $\nu$ C-O (ester links), and 722  $\rho$ CH<sub>2</sub>, were determined. The extraction with ethanol, peaks, 3452  $\nu$ OH (alcohol), 1601 (possibly carboxylic mineral salt), 2958, 2920, 2851  $\nu$ C-H (hydrocarbon chains), 1737  $\nu$ C=O (ester carbonyl), 1463  $\delta$ CH<sub>2</sub>, 1353  $\delta$ CH<sub>3</sub> (umbrella-type symmetric bending), 3452  $\nu$ O-H, 1048  $\nu$ C-O (alcohol), and 722  $\rho$ CH<sub>2</sub> were given.

<b>M1d tissue</b>		
<p>M1d-Tissue</p> <p><b>ATR spectrum</b></p>		<p><b>AK23: Skin tissue</b>  3275 vN-H,  1701 fatty acid (possibly due to side chains in Glutamic and Aspartic acid, and/or decomposition of protein backbone)  1625 amide I (vC=O with small contributions from vC-N and δN-H)  1545 amide II (vC-N with contributions from δ<sub>ip</sub>N-H)  1454, vCH<sub>2</sub>  1379, δNH<sub>2</sub> and/or δCOOH (glutamic)  1231 amide III (vC-N + δN-H with contributions from δC-H)</p> <p><b>AK27:</b> skin tissue, nearby spot  3259, 1624, 1554, 1379 protein  <b>Additional peaks:</b> 1155, 1109, 1070, 1027 traces of textile tissue</p>
<p>M1d – Tissue extraction in CH<sub>2</sub>Cl<sub>2</sub></p> <p><b>FTIR spectrum</b></p>		<p><b>AK44:</b>  2925, 2858, 1710, 1465, 1411, 1375, 722 are basically assigned to free fatty acid, fatty material</p>
<p>M1d – Tissue extraction in CH<sub>2</sub>Cl<sub>2</sub></p> <p><b>FTIR spectrum</b></p>		<p><b>AK45:</b>  1710 1411, fatty acid 2925, 2858, 1736, 1462, 1382, 722 fatty ester  Spectrum in AK45 is similar to AK44</p>

M1d-  
Tissue

Deconvolution  
of amide I and  
II region

ATR FTIR  
spectrum

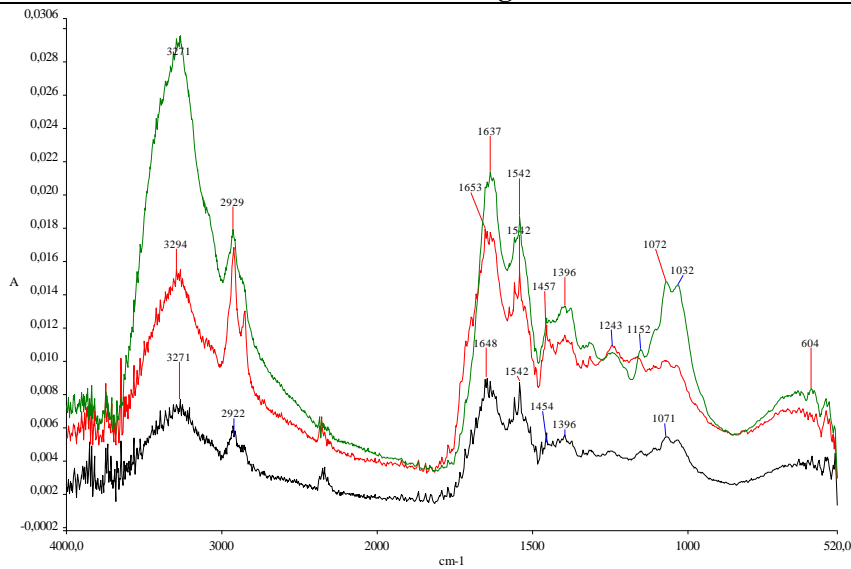


AK27:  
Deconvolution and peak  
fitting of the 1800 – 1480  
cm<sup>-1</sup> region

M2g tissue

M2g-  
Tissue

ATR-FTIR  
spectrum 1



AK29: tissue  
AK30: tissue (nearby area)  
AK31: tissue-FTIR  
Rich in fatty material

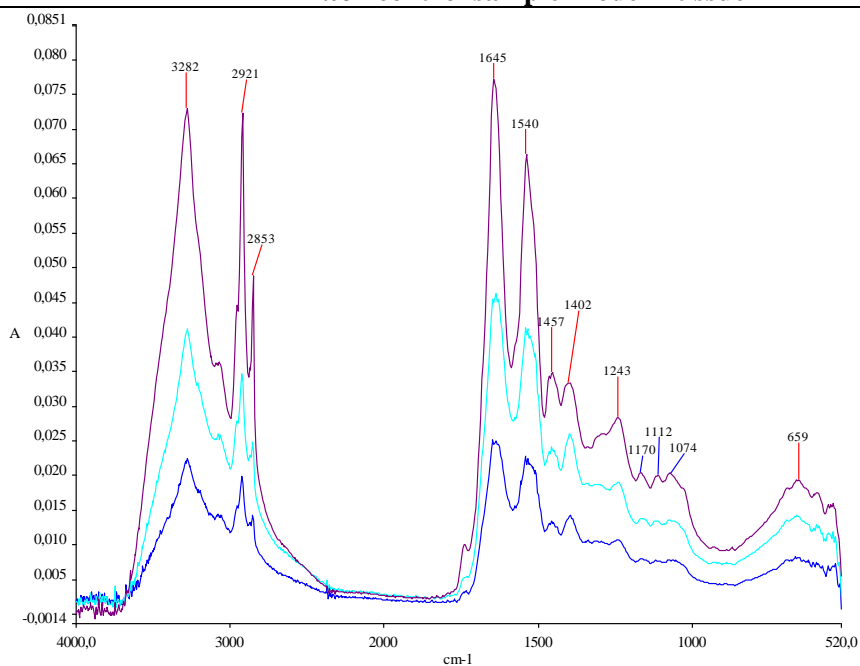
1653-1637 amide I,  
1542 amide II  
1457  $\nu$ CH<sub>2</sub> (mainly Proline  
ring)  
1396  $\delta$ C-O-H (residual  
acidic chains from Glutamic  
and Aspartic acids and/or  
fatty components)  
1243 amide III  
1072, 1032 cellulosic  
material  
1740 cm<sup>-1</sup> fatty esters



<p>M2g-Tissue</p> <p><b>ATR spectrum</b></p>		<p>AK30: tissue (nearby area)  AK31: <b>tissue FTIR</b>  Fatty material  AK31d: spectrum after subtraction (fatty material)</p>
<p>M2g-Tissue</p> <p>Extraction in <math>Cl_2Cl_2</math></p> <p><b>FTIR spectrum</b></p>		<p>AK46:  2927, 2855, 1726, 1719, 1463, 1380, 1246, 1175, 721</p>
<p>M2g-Tissue</p> <p>Extraction in <math>H_2O</math></p> <p><b>FTIR spectrum</b></p>		<p>AK72:  3239, 2932, 1665 (amide I), 1572 (amide II, shoulder), 1457, 1405, 1257: protein material</p>

**Mπδ1 control sample-modern tissue**

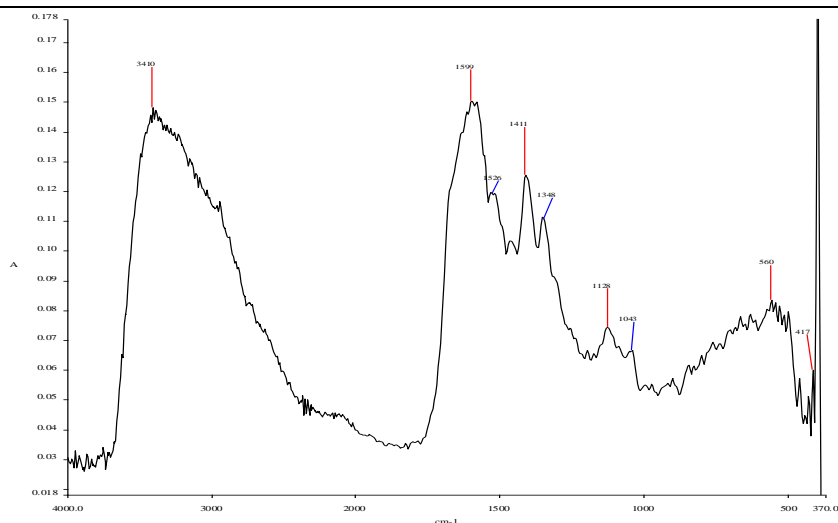
Mπδ1-  
Modern tissue  
**ATR  
spectrum**



AK34: scan 1  
AK35: scan 2  
AK36: scan 3

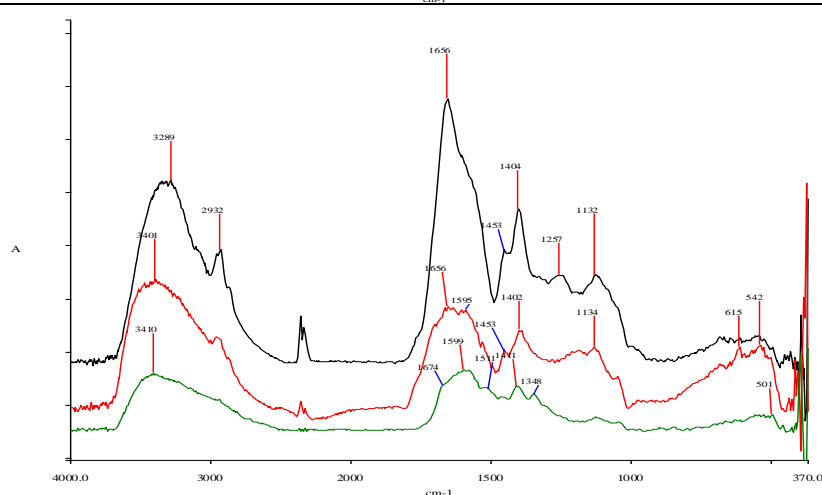
3282 amide A (νN-H)  
3080 amide B (Second component of νN-H in Fermi resonance with the amide II overtone)  
1732 fatty ester (fatty component in skin; more intense in scan 3)  
1625 amide I (νC=O with small contributions from νC-N and δN-H)  
1545 amide II (νC-N with contributions from δ<sub>ip</sub>N-H)  
1454, νCH<sub>2</sub>  
1379, δNH<sub>2</sub> and/or δCOOH (glutamic)  
1231 amide III (νC-N + δN-H with contributions from δC-H)

Mπδ1-  
Modern tissue  
Extraction in water  
**FTIR  
spectrum**



AK94:  
3430 νO-H from fatty acid  
1600 νC=O from carboxylic salt or amide I of short chain peptide)  
1525 amide II of short chain peptide  
1411 δC-O-H (carboxylic acids)  
1348 δNH<sub>2</sub>  
1128, 1048 νC-O

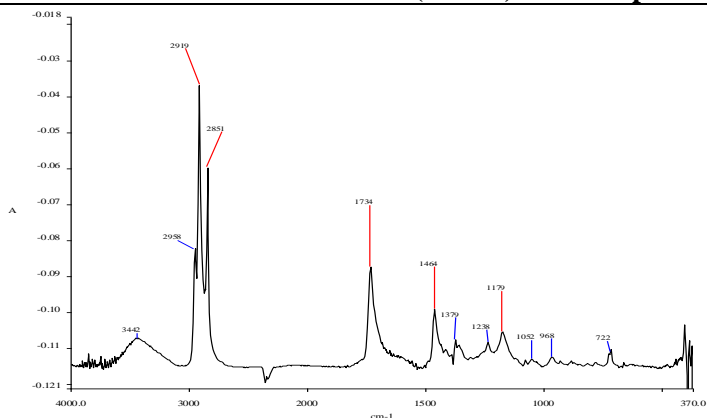
M1d-  
M2g-  
Mπδ1-  
Modern tissue  
comparison of solvent skin  
tissue extracts



AK90: M1d  
AK72: M2g  
AK94 : Mπδ1-  
Modern tissue

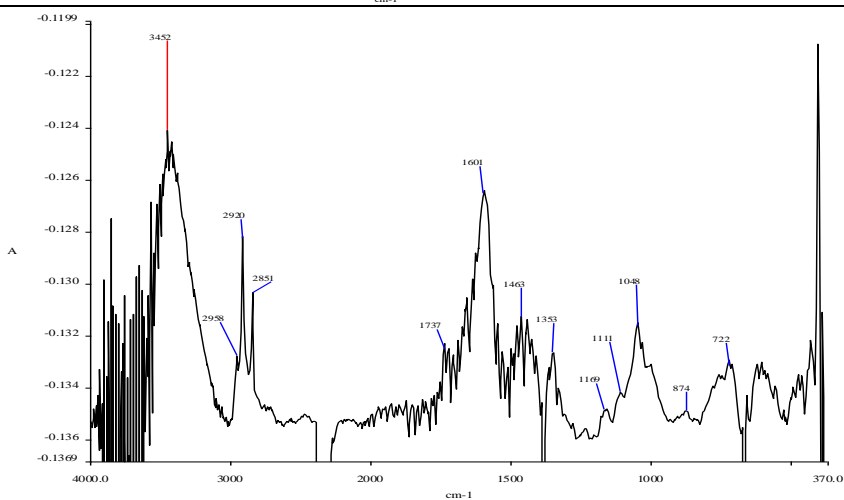
**M1f (M1f02) hair sample**

M1f-  
Hairs  
Extraction in  
Cl<sub>2</sub>Cl<sub>2</sub>  
  
**FTIR  
spectrum**



**AK57:**  
2958, 2919, 2851  $\nu$ C-H  
(hydrocarbon chains)  
1734  $\nu$ C=O (ester carbonyl)  
1464  $\delta$ CH<sub>2</sub>  
1379  $\delta$ CH<sub>3</sub> (umbrella-type  
symmetric bending)  
1238, 1179  $\nu$ C-O (ester  
links)  
722  $\rho$ CH<sub>2</sub>

M1f-  
Hairs  
Extraction in  
Ethanol  
  
**FTIR  
spectrum**

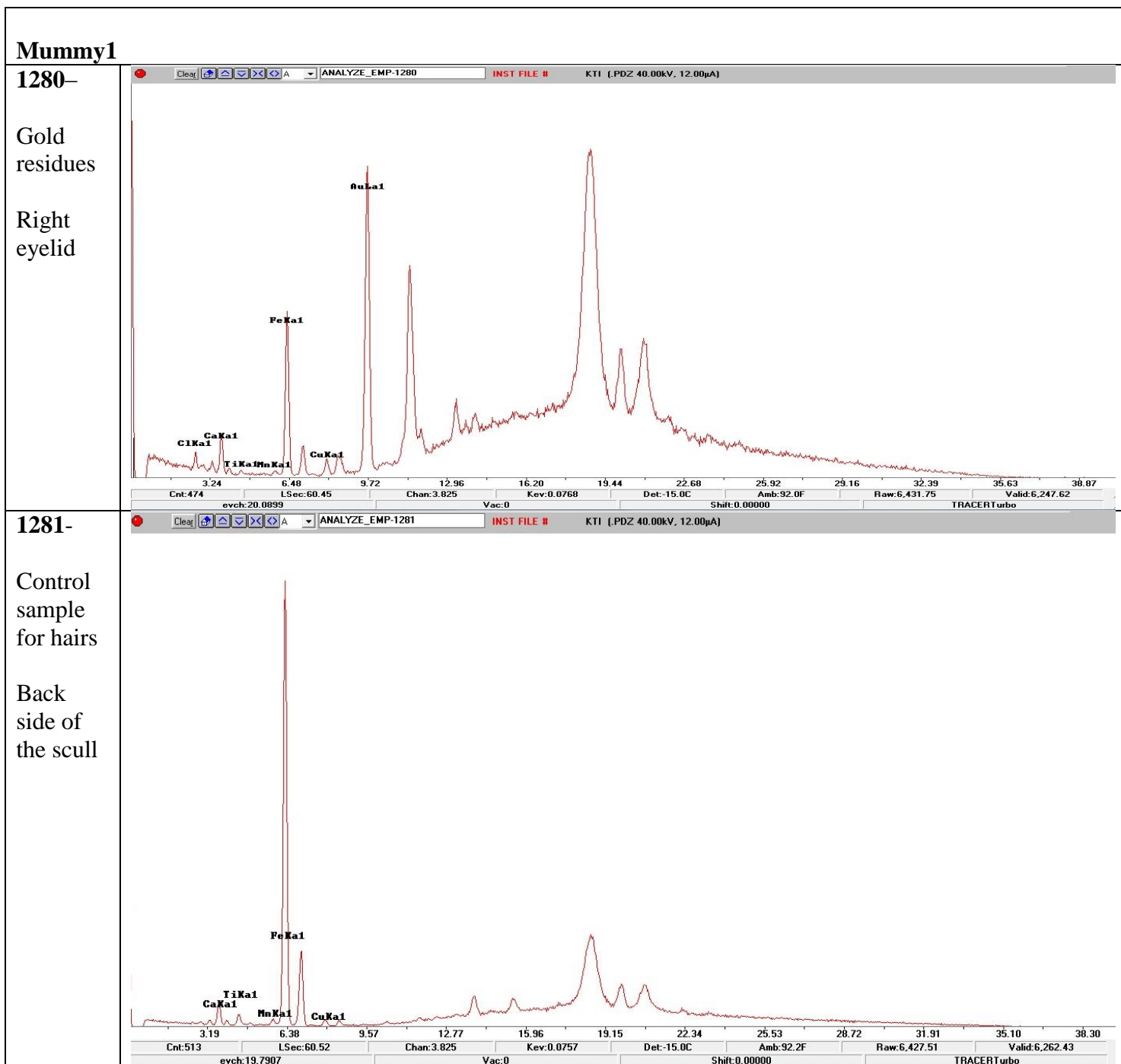


**AK61:**  
3452  $\nu$ OH (alcohol)  
1601  
  
2958, 2920, 2851  $\nu$ C-H  
(hydrocarbon chains)  
1737  $\nu$ C=O (ester carbonyl)  
1463  $\delta$ CH<sub>2</sub>  
1353  $\delta$ CH<sub>3</sub> (umbrella-type  
symmetric bending)  
722  $\rho$ CH<sub>2</sub>  
  
3452  $\nu$ O-H  
1048  $\nu$ C-O (alcohol)

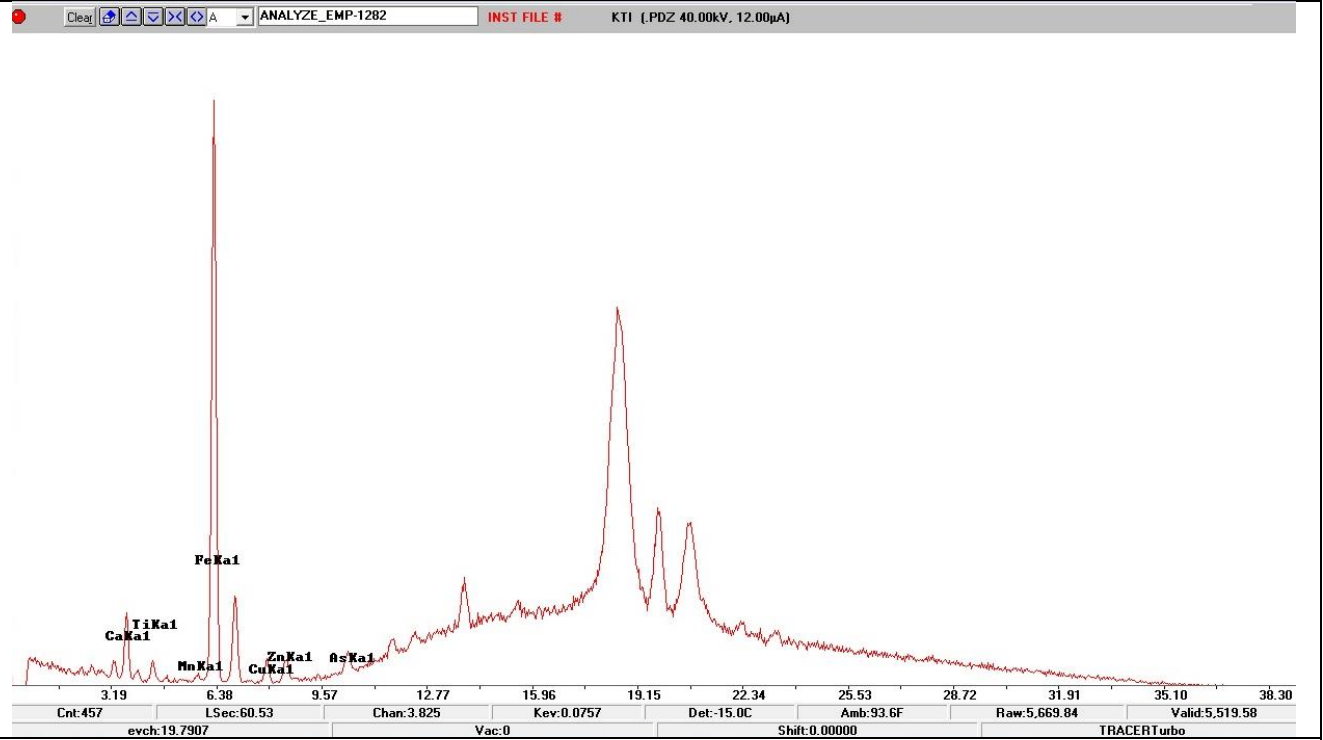
#### 4.7 Portable X-Ray Fluorescence (XRF) analysis

The qualitative analysis of gold residues in both mummies showed similar spectra. In 1275, 1276, 1277, 1278, 1279 and 1280 spectra, the element Au and Fe are presented with the higher peak. The elements Ca and Cu and Ti are presented in all spectra with low peaks. The Cl element was determined in 1280 spectrum.

In 1281 and 1282 spectrum, analysis of hairs, Fe is the higher peak whereas low peaks of Ca, Ti, Mn, Cu and were determined. Zn and As elements were determined in 1282 spectrum.

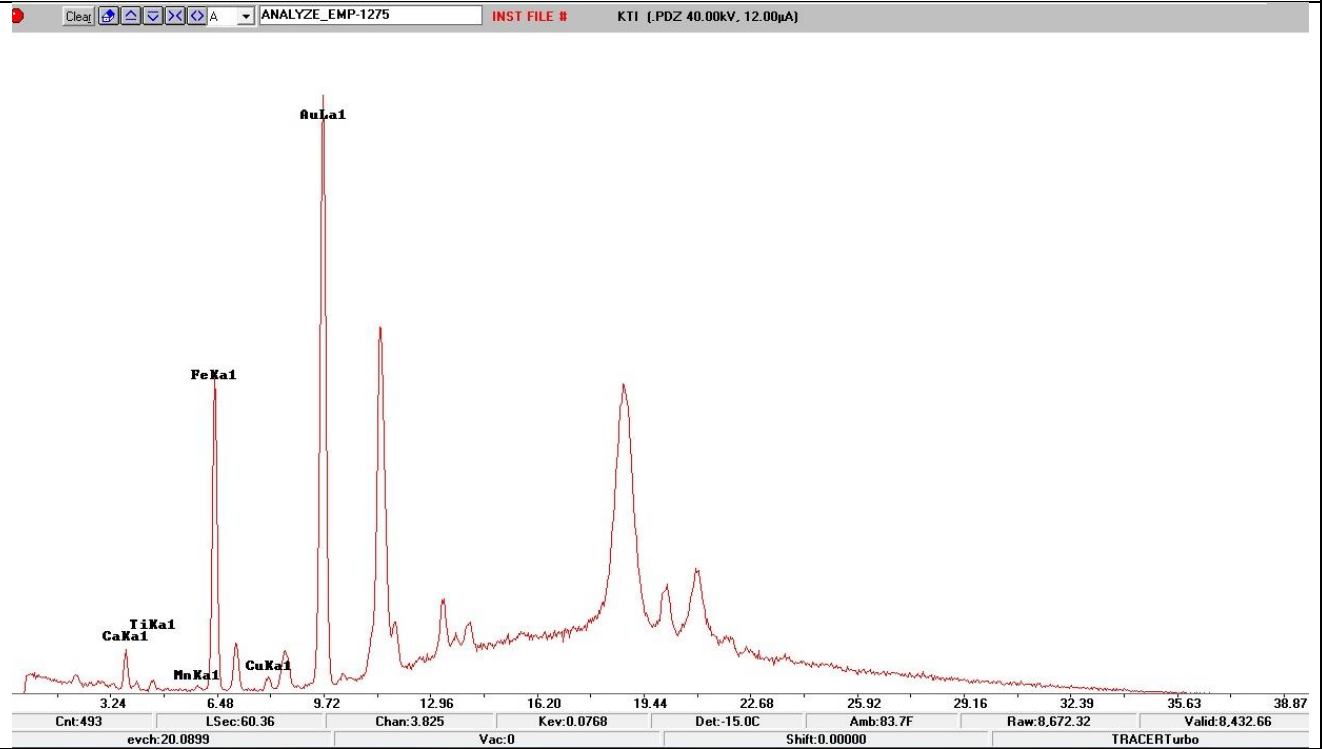


1282-  
Hairs



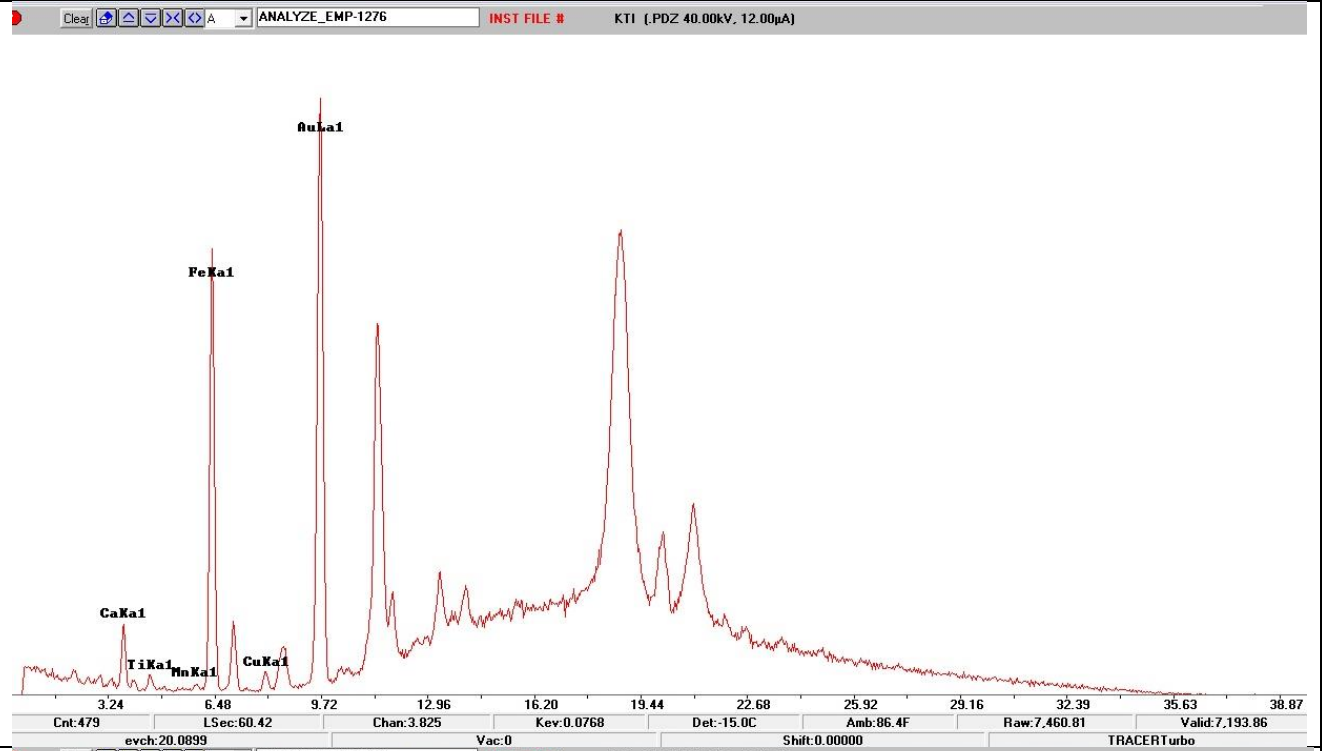
Mummy2

1275-  
Gold  
residues  
  
Left side  
of the  
thorax



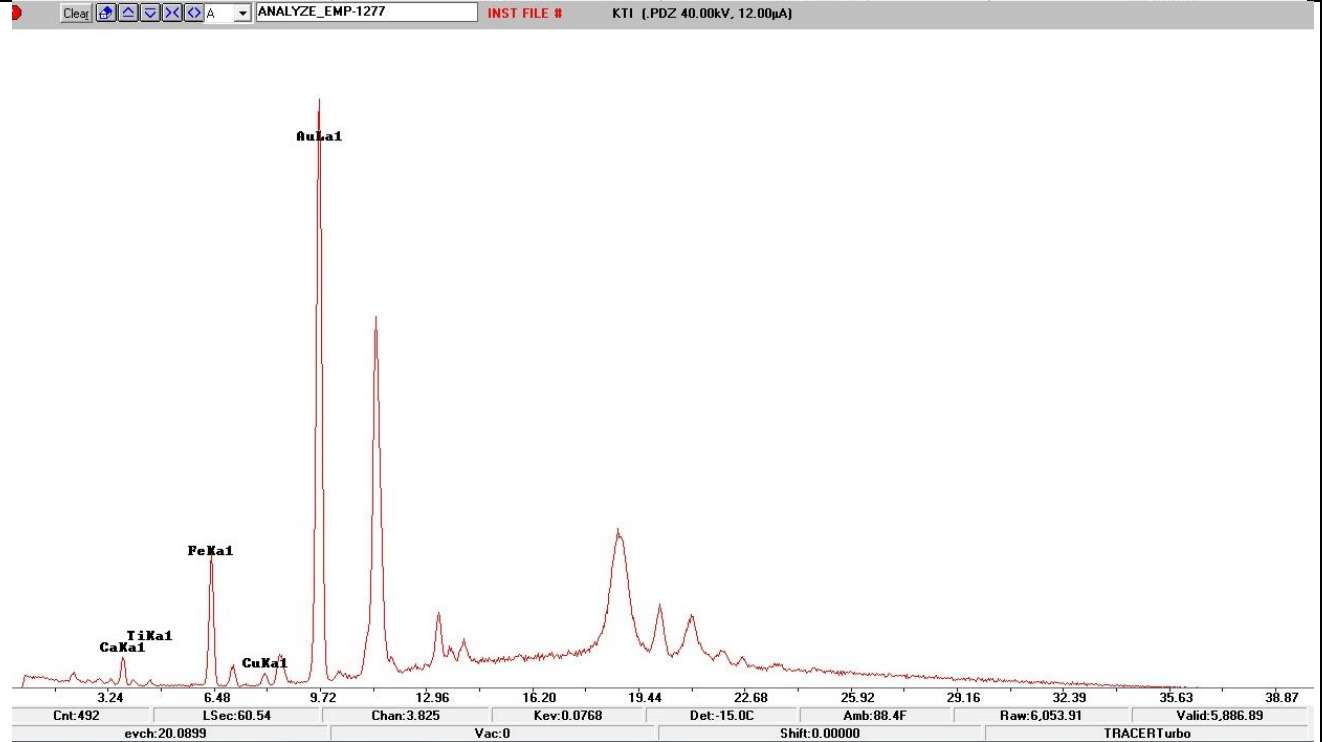
1276-  
Gold  
residues

Left side  
of the  
thorax



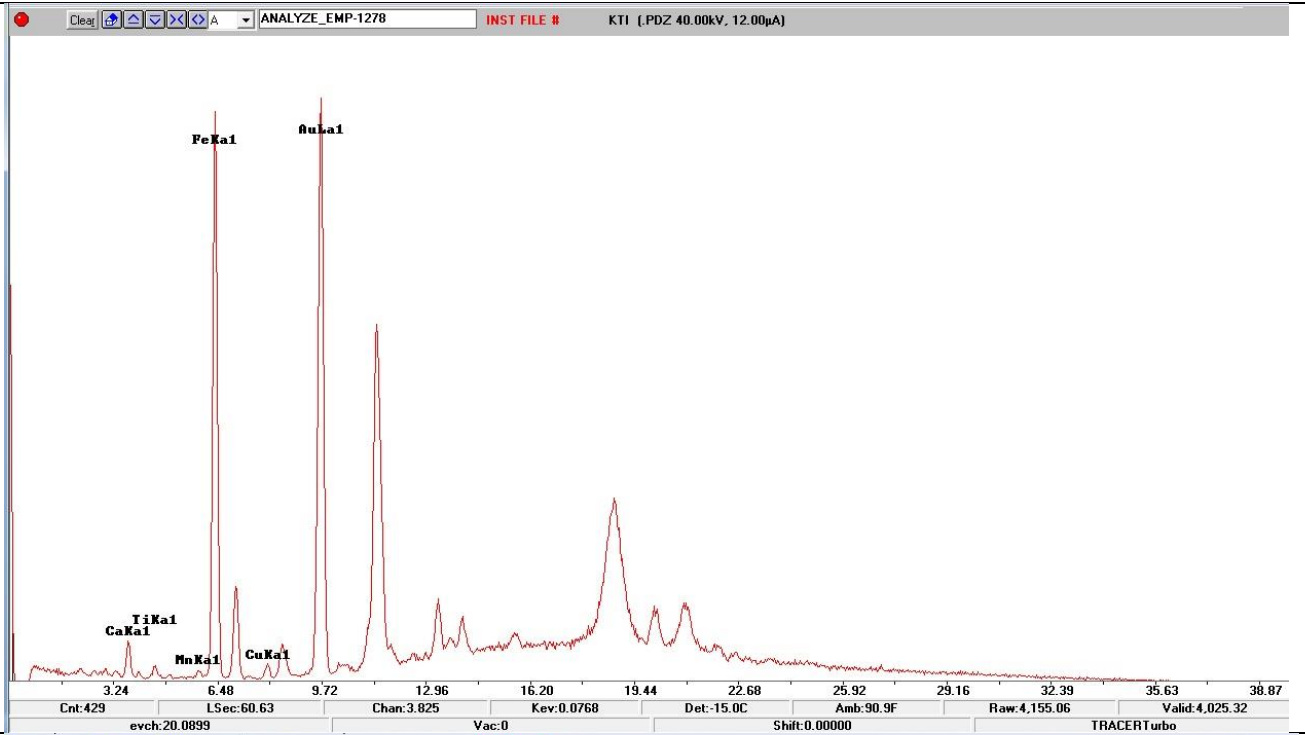
1277-  
Gold  
residues

forehead



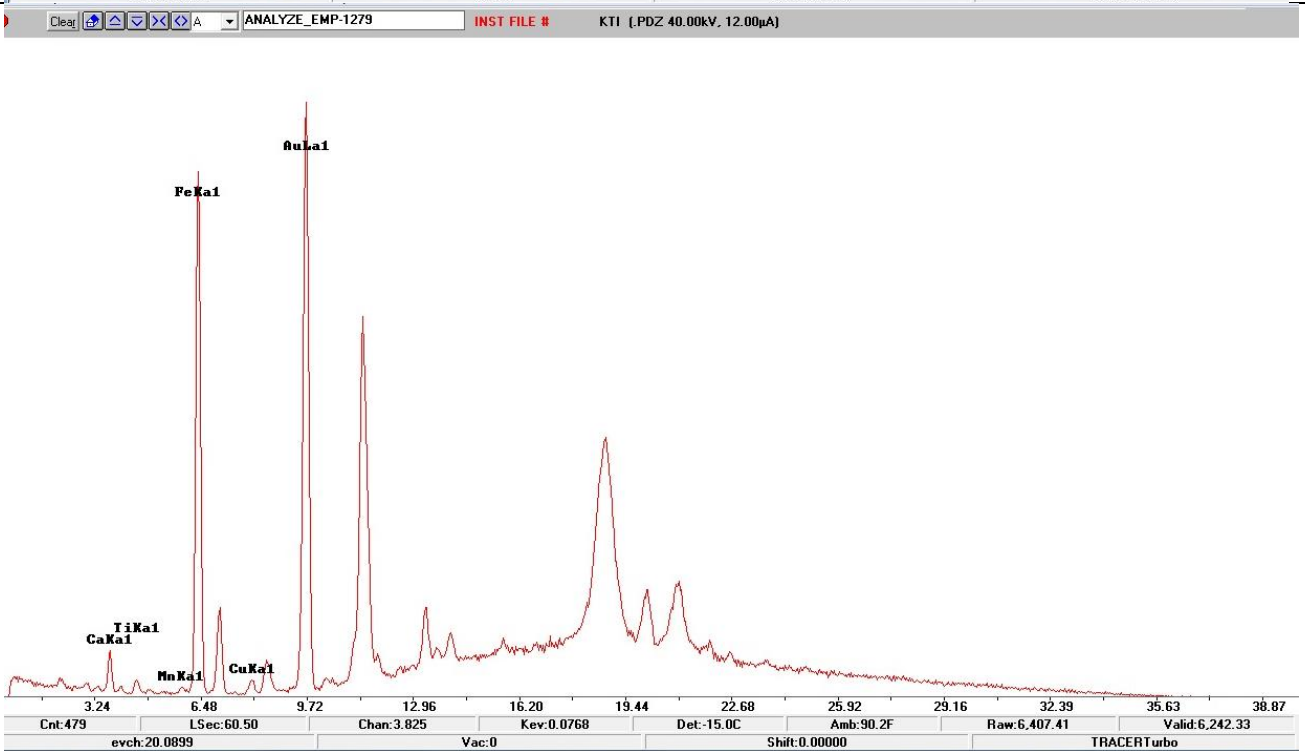
1278-  
Gold  
residues

Right  
eyelid



1279-  
Gold  
residues

Foot





#### 4.8 Microbiological Investigation

Fungi hyphae and spores were determined with optical microscope in both mummies (fig.59-62). During the cultivation, fungus growth was determined on wooden sticks of swabs and broths of M1 and M2 (fig.63-66).

##### Mummy1 (M1)

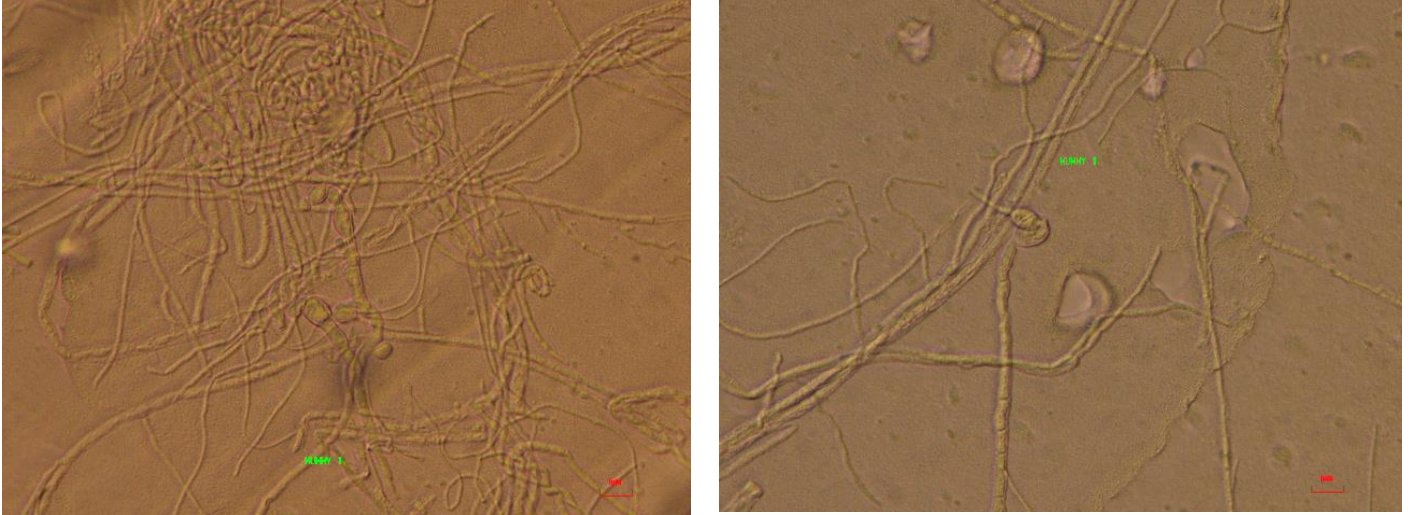


Fig. 59-60: Optic microscope observation on cultivation. Fungus hyphae and spores were determined on the samples of M1.

##### Mummy2 (M2)

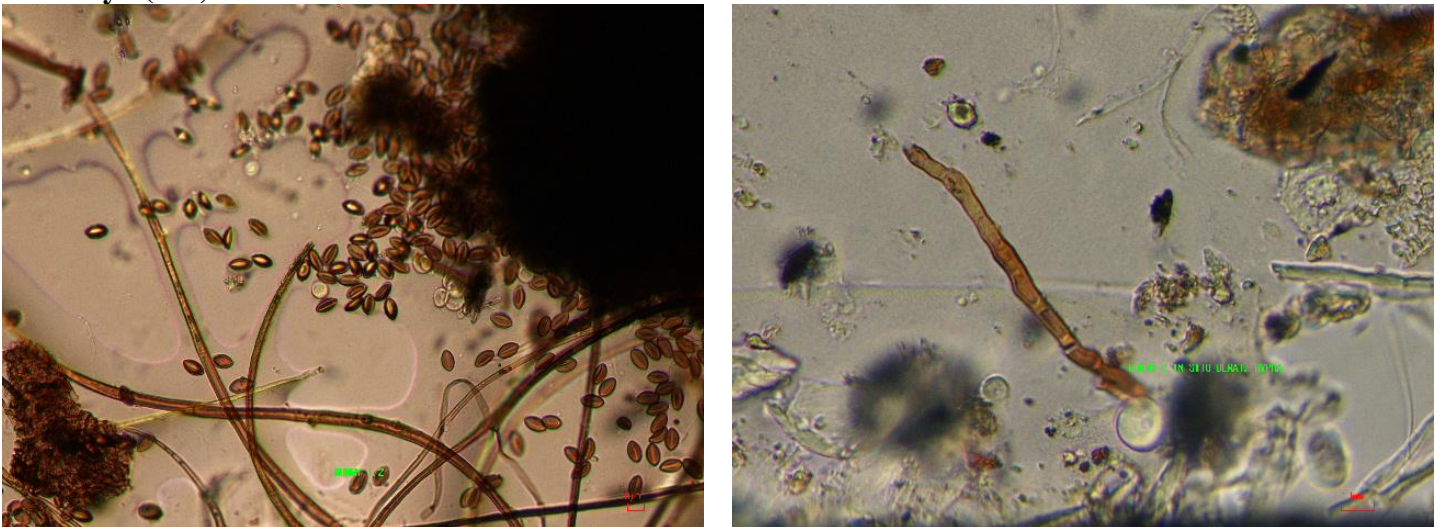


Fig.61-62: Optic microscope observation on masking tapes' samples. Fungus hyphae and spores were determined on the samples of M2.

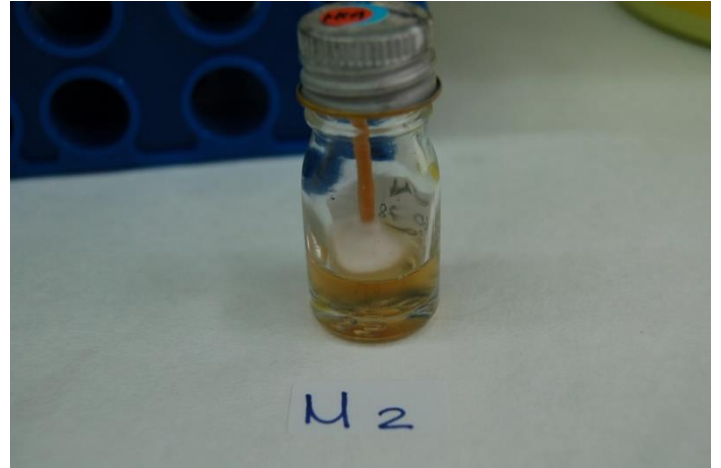
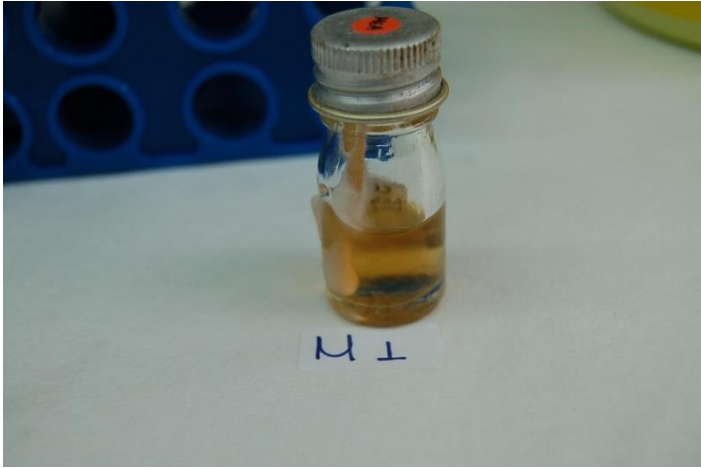


Fig.63-64: Fungus growth on the wooden stics of swabs.



Fig.65-66: Fungus growth in broth cultivation.

## 5. Discussion

The preliminary results of the above study have led to interesting findings. More precisely, these findings give us elementary information both on the conservation status of the mummies, as well as a good hint on the methods and procedures that can be used in the near future for the additional study.

### *Estimation of mummies 'preservation state*

- The broken parts of the bodies, the inappropriate removal of the bandages, the numerous degraded materials' fragments, the dust and the cobwebs that were observed by visual examination and documented, are strong indications of their bad storage conditions and mishandling.

### *Examination of the bandages*

- In both mummies, most parts of the bandages are in light brown color, and impregnated in balms. They mainly cover parts of the bodies. Few bright yellow bandages with no traces of balms are observed as filling materials in the abdomen. A thinner bright yellow textile was identified to be laid on M1's legs (fig.67-68).
- Similarly two types of bandages were determined in M2 samples. The M2f piece differs in weaving (dense), texture and color in comparison with the most bandages in both mummies, macroscopically and stereoscopically. In addition M2f presents an interest as the hole was examined with SEM has not signs of insect attack. This fact can create the hypothesis that it could have been opened for purpose.

Likewise, M1c presents differences with other bandages but it cannot be told for sure if it is of a different type. The samples analyzed are broken fragments from around/on the mummies and their original location remains unknown.



Fig.67-68: The fragments of bright yellow textile found on M1's legs.

### *Plant fibers in bandages*

- There is strong evidence that the type of the fibers is linen. Linen is made of the plant Flax (*Linum Usitatissimum*). It was the basic material of textile production in Ancient Egypt. It was used in clothing for people, in



mummification process for the dead, for bedding, furnishing and sailcloth in everyday life (Riggs, C. 2014; Nicholson. T.P., Shaw, 2000).

The SEM images of M1a and M1c present the characteristic nodes and longitudinal striations of linen fibers (fig.. Also the ATR-FTIR results of M1h and M2f, showed a chemical composition mainly of polysaccharides. The reference spectrum of linen (fig.70) presents similar peaks in comparison with M1h and M2f spectra. Polysaccharides (>80%) are the common composition of cellulose material existing in cell walls of plants; small proportion of lignin, proteins, pigments, waxes and minerals are included too (May and Hones 2006). Cellulose compounds were also detected in M1d and M2g samples of tissue. They probably refer to the use of plants and textiles residues as filling materials that detected in M1 and are described in literature (Abdel-Maskoud and El-Amin 2011).

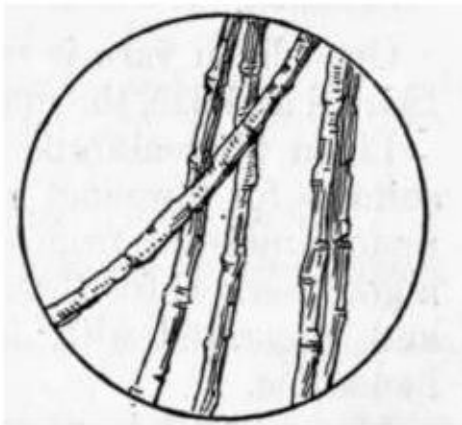


Fig.69: The characteristic anatomy of linen fiber. (<http://chestofbooks.com/food/household/A-Manual-Of-Home-Making/Linen.html>)

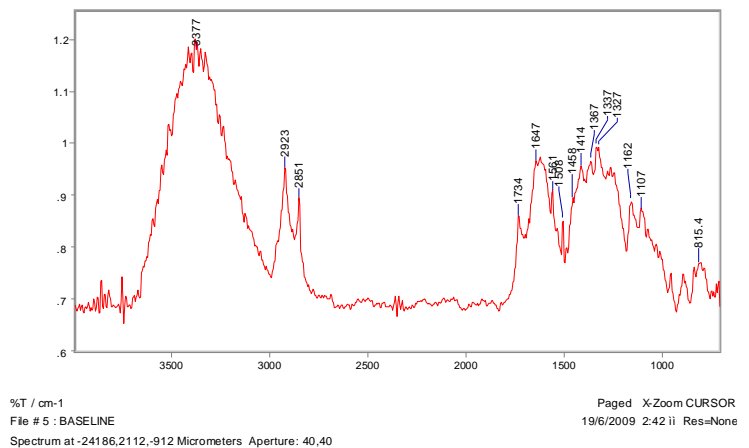


Fig.70: ATR-FTIR spectrum: Linen textile reference sample.

### “Cartonnage” material in bandages

- M1b and M2d samples show decorative motifs with plaster and colors. Residues of plaster and colors were observed too, on the wrapped fabric of M1. The particular decoration technique on bandages refers to as “cartonnage”. It was used in funeral rituals as covering of the deceased’s’ body. The covers were decorated with colorful symbols and inscriptions. According to SEM/EDEX and ATIR-FTIR analysis the white plaster of M1b is mainly made of CaCO<sub>3</sub>, and small amounts of quartz SiO<sub>2</sub>. Similar results have been found in previous investigation of cartonnage plaster of Greek Roman period (Affifi 2011). The pigments of the red color are made of iron oxides, Hematite (Green, 2001, Uda et al 2000), whereas the blue color is possibly Egyptian blue (personal communication with Ass. Prof. Y. Fakorellis and, Ass. Prof. S. Boyatzis). In addition, the presence of cuprorivate and

quartz are strong evidence of the first synthetic color made in Ancient Egypt known as “Egyptian blue” (Mirti et al 1999, Mazzocchin et al 2004). The intermediate binder of the cartonnage has not been defined yet. Evidence of oily binder is also shown on AK65 FTIR spectra but it has not been determined yet.

#### *Shellac material in the bandages*

- According to the FTIR results, components of shellac wax in good preservation state were identified in M1c, and M2e samples of bandages (AK 39, AK41, and AK70 spectrum) (the technique was performed by Ass. Prof. S. Boyatzis, who confirmed the results). Peaks of shellac also were determined in M2g sample of tissue (AK46 sp.). The composition of shelloic, jalaric acids and esters are significant clues for shellac presence (Fagag 2010; Galatis et al 2012). The particular results are confirmed with the shellac reference FTIR spectrum (fig.71).

Shellac is natural resin extracted by the lac insect (*Laccifer Iacca* Kerr) in the forests of India and Thailand. Shellac as conservation product (adhesive or polish) has been used in several materials since 20<sup>th</sup> century. There is little information in literature about the use of shellac on conservation of mummies. Oddy (1992) states the following: “Early restoration treatments of human tissue were robust and intrusive. Those seeking to market ancient bodies would make good any losses or disjointed areas using locally available repair materials such as animal glue and plaster of Paris, sometimes in combination with textile bindings and/or reinforcing strips. In the restoration of a mummy and coffin ensemble (which includes a broad range of original components) one might find any of the following: beeswax, paraffin wax, flour paste, cardboard, wood, shellac and variants, natural gums and resins, and a wide range of fillers. These were combined in various ways often designed to obscure the fact of the item’s restoration” (Harris and Douny 2014).

Considering the above, the intense amount of glossy appearance, resinous material on the right cheek of M1 (fig.72) and on the left leg of M2 (fig.73) or/and the existence of bright yellow bandages with no traces of balms and significant ageing signs, could be evidence for past restoration treatments. Further investigation is recommended for more information.

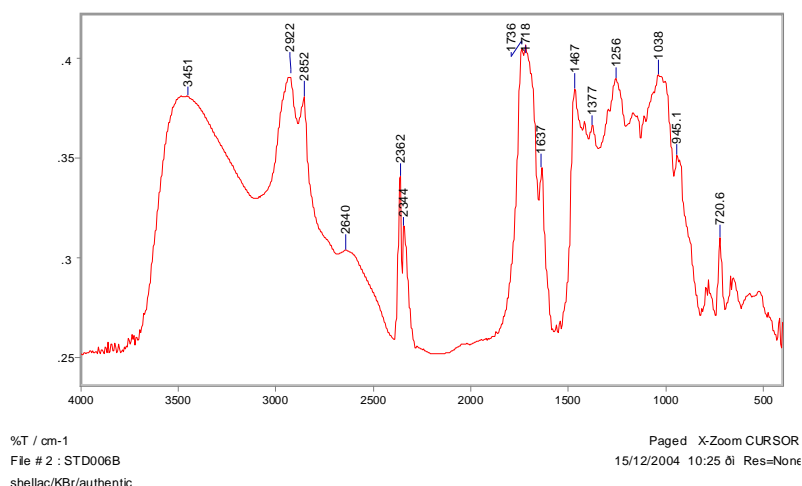


Fig.71: ATR-FTIR spectrum: Fresh Shellac reference sample([http://lisa.chem.ut.ee/IR\\_spectra/coating\\_materials/shellac/](http://lisa.chem.ut.ee/IR_spectra/coating_materials/shellac/)).



Fig.72-73: Evidence for past restoration treatments (?).The glossy appearance of resinous material in the crack of M1's cheek and the left leg of M2.

#### *Presence of natural resin in the piece of balm*

- FTIR analysis on M1g piece of balm detected organic material (due to the intense peak at 1707 and 1441), hydrocarbon fraction (possibly wax, due to the peaks at 2922, 2858, 1463) along with carbonates, that they could possibly belong to embalming material. According to Dr. Boyatzis it could be assigned either to mastic or to propolis; mastic however, is unlikely, as the expected intense peaks at 1455 and 1380 (in M1g) do not show at the same positions, and also their intensities are not strong. Therefore, it can tentatively be assigned to propolis, on the basis of its historic use, and also that wax (which is also identified in M1g) has been part of the originally applied material, and not added by embalmers as a separate ingredient. The composition of propolis varies significantly, depending on geographical origin, vegetation-considered as nutrients, etc.); but generally it is mainly composed of terpenic acids and succinates (see reference propolis fig.74-75) Further analysis is required in order to identify the actual components and the material itself.

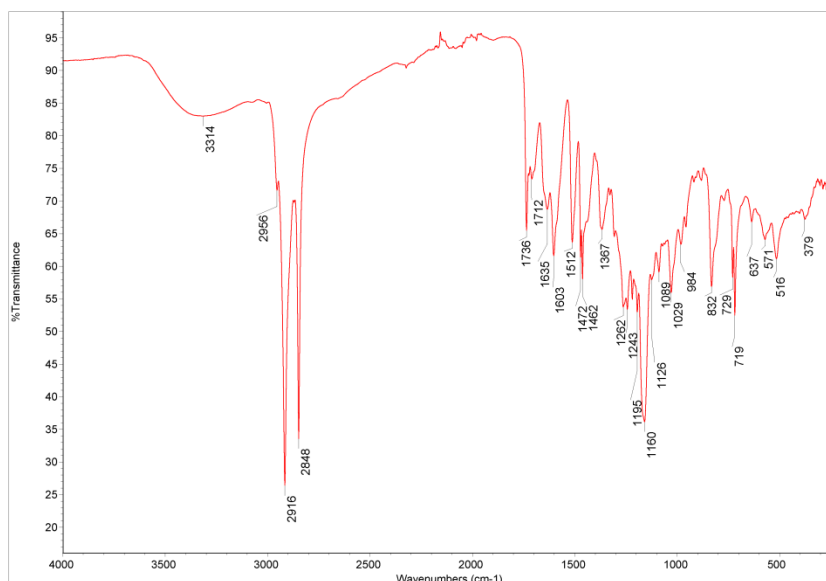


Fig.74: ATR-FT-IR spectrum of Propolis (4000 – 225  $\text{cm}^{-1}$ )

[[http://lisa.chem.ut.ee/IR\\_spectra/paint/binders/propolis/](http://lisa.chem.ut.ee/IR_spectra/paint/binders/propolis/)]

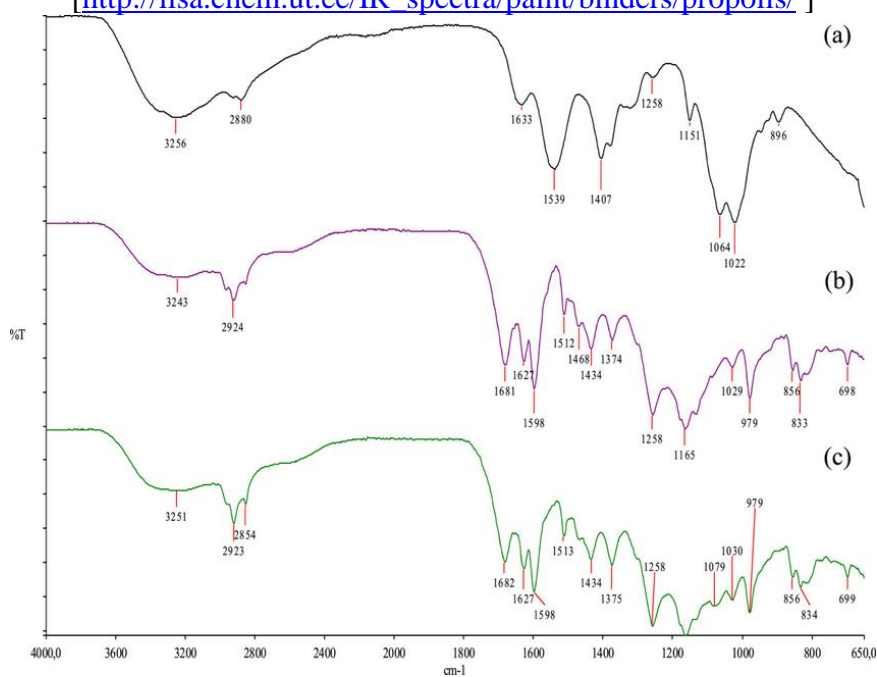


Fig.75: ATR-FTIR spectrum of **a**, blank varnish (CHV); **b**, Ethanollic Propolis extract (EPE) and **c**, Propolis varnish containing 15% of dried extract (PCV 15%). From reference (Kuhnen 2012)

### *The examination of the two beads of M2*

- The examination of the beads has showed that they are made of glass. The contamination mainly in Calcium and Sodium with SEM/EDEX analysis is strong evidence for glass materials (Freestone 1991, Davinson 2003). It would be interesting to study the contamination of Manganese and Copper as colorant in the M2a1. Also, Antimonite in M2a1 probably was used for the yellow coloration.



### *XRF results on gold residues of M1 and M2*

- The presence of gold Au element in all spectra of M1 and M2 gives hints about the good quality of gold mineral as Cu peaks were low enough. Further investigation must be done because the quantification analysis of the spectra could not be obtained. The reasons are probably the thin layer of the sample, the numerous layers under the gold as skin is composed, and the anatomical curvature of the body areas that were applied.

### *Presence of insect damage*

- The numerous of holes on the skin of M1 and M2 and the presence of insects remains in M1b and M1g are strong evidence of insect deterioration. However, further investigation is required for their identification.

### *Degraded oil and fatty material on bandages*

- Protein and fatty materials in degradation state were determined with FTIR in M2f (1184, 734) and M1b (1653  $\nu$ C=C) as deposits or absorbance materials on bandages. In the double bonds, which are referred to 1653 and 1184, 734 peaks are obtained in particular organic compounds as cause of oxidation or hydrolysis reactions. Those reactions led to the weakening and embrittlement of the material.

### *Microbial contamination*

- According to Ass. Prof. A. Velegraki, hyphae of fungus and spores were identified on M1f1, M1f3 sample of hairs with Optical and S.E. microscope. There is a possibility that the vertical slits, which were observed on the hairs with SEM and optical microscope, are the results of fungus attack. In previous investigation on biodegradation of archaeological mammalian hairs, similar micro-scratches have been observed (Silvana et al 2014). Masses of fungus were also determined on M1a sample of bandage, SEM image. The results of cultivation led to the identification of fungus which fed with cellulose compounds as the linen is composed. These species generally come from the ground and they grow in environment with high levels of moisture. In addition the spores were estimated to be in active state as they looked hydrated under the optic microscope.

Bio-deterioration processes are the main factors of cellulosic fibers/materials' degradation. Results of bio-deterioration are the weakening of materials and changes in appearance due to oxidation state, degree of polymerization and breakdown of molecular structure (Szostak-Kotowa 2004).

### *Skin deterioration*

- As FTIR analysis is capable to determine alterations in ancient skin tissue, in our case of M1d, possible collagen proteins were determined. Fatty acids, amide I, II, III which consist proteins of collagen were obtained (Brodsky and Persikov 2005; Payne and Veis1988) In the deconvolution and peak fitting of M1d (AK27) and M2g (AK29,30) it was shown that the amide I and amide II region bands were deconvoluted in many components. This can be attributed due to the existence of low and medium MW peptides showing variable amide I absorptions, which is evidence for the degradation of protein material (Mühlen-Axelsson et al 2014)

According to FTIR results, oily and fatty materials in serious hydrolytic degradation state were determined in M1d (personal communication with Ass. Prof. S. Boyatzis). Only a small percent of fatty material exists in its original form (ester). By speculation, the origin of the fatty substance can be assigned to the skin components; however cosmetic oily substances cannot be ruled out.

### *Hair composition*

- In the terms of raw elements, on average, hair is composed of 50.65% carbon, 20.85% oxygen, 17.14% nitrogen, 6.36% hydrogen, and 5.0% sulfur. Hairs also contains amounts of magnesium, arsenic, iron, chromium and others metals and minerals (Fuchs 1995; Yu et al, 1993). The mineral content of human hair fibers is generally very low (<1%), and it is difficult to determine whether this inorganic matter is derived from an extraneous source (which much of it is) or whether it arises during fiber synthesis (Robbins 2012).

The elements investigated according to the literature in M1f1 and M1f3 sample were: Ca, Mg, Al, Na, K, Zn, Cu, Fe, Ag, Au, Pb, Sb, Ti, Mo I, P, and Se. The B, W and V elements were not investigated; The reason is that the B element is light for EDEX analysis, while the W element causes noise because of SEM chamber is made of wolfram; The V element also causes noise and is convoluted with others peaks.

SEM/EDEX is considered a valid analysis for the elemental contamination of ancient hair, as eighteen elements were determined taking twenty three spectra. In contrast, in XRF analysis only seven elements were determined. However, the Zn and As elements were determined only with XRF analysis. Further investigation with quantification analysis of SEM/EDEX and XRF could provide more information about the capabilities of the equipment and the elemental preservation in ancient hairs' analysis.

### *Ca, Si, Cl and Na elements*

- In all samples, serious amounts of Ca and Si were detected with SEM and FTIR analysis. Cl and Na were also present in all samples with the biggest amount in M1d and M2g samples of tissue. Ca and Si may represent deposits

on samples from the ground or from the decomposition of the material in general. However the idea that mummies have been in touch with ground soil is confirmed by the origin of fungus come from the ground. The elements Cl and Na are possibly referred to the *natron*, Sodium Chloride NaCl that it was used in mummification process (Abdel-Maskoud and El-Amin 2011).

*The plant sample of M1i as monocot*

- The information derived of M1i section via optical and S.E. microscopy is that vascular bundle anatomy along with the stomata parallel arrangement on epidermis, prove that the plant species used is a monocot (fig.76 reference image). The cell walls of the plant are in good preservation state, most probably of the balm impregnation used in mummification process (Pr. C. Faseas personal communication).

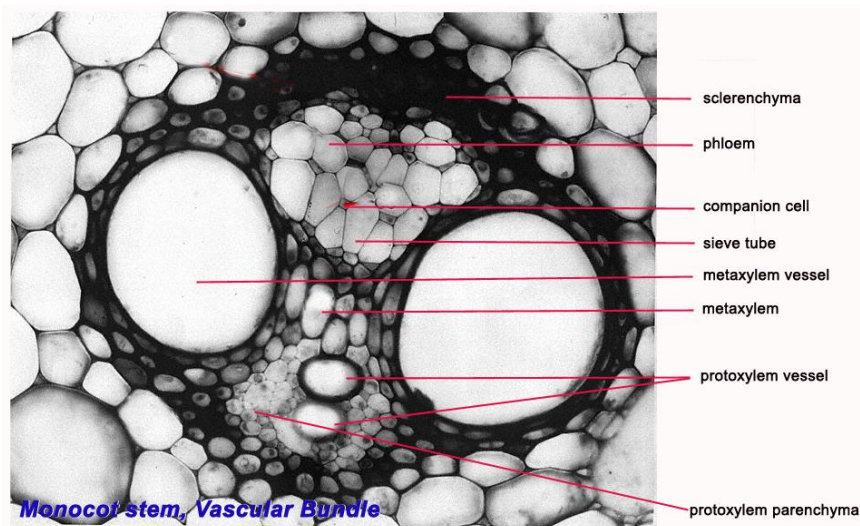


Fig.76: Monocot stem, Vascular bundles

[http://bugs.bio.usyd.edu.au/learning/resources/plant\\_form\\_function1/plant\\_form/anatomy\\_project.html](http://bugs.bio.usyd.edu.au/learning/resources/plant_form_function1/plant_form/anatomy_project.html)

Monocotyledon (monocot) is one of the two great groups of flowering plant, or angiosperms. The other is the dicotyledons (dicots). There are approximately 60.000 species of monocots including the most economically of all plant families *Poaceae* and the largest of all plant families, *Orchidaceae*. Other prominent monocot families include *Liliaceae*, *Arecaceae* and *Iridaceae*. Most of them are distinguished by the presence of only one seed leaf, or cotyledon, in the embryo contained in the seed. Dicotyledon in contrast ordinarily has two cotyledons (Bewley and Black 2006).

Further investigation will provide detailed information about the plant identity or evidence that was used in past restoration treatments as referred above.

## **6. Conclusions**

According to above both mummies are composed of mainly organic (bones, skin, linen, balms) and inorganic materials (bone, natron, gold, cartonnage). The degradation was determined is mainly caused by physical, chemical and biochemical processes. These processes are obtained due to the presence of moisture, oxygen, ultra-violet light and environmental pollutant. As conclusion the mummies are in bad preservation state and immediate conservation plan is required.

## **7. Further examination aims**

- Estimation of preservation state in general CT-scan
- Identification of balms and materials of past treatments GC/MS
- Preservation state of skin histological, m-FTIR analysis
- Composition of hairs m-XRF, and SEM/EDEX analysis
- Identification of plant SEM analysis
- Composition of gold residues m-XRF analysis
- Corrosion patterns of glass beads with Micro-topography and SEM/EDS
- Dating with  $^{14}\text{C}$

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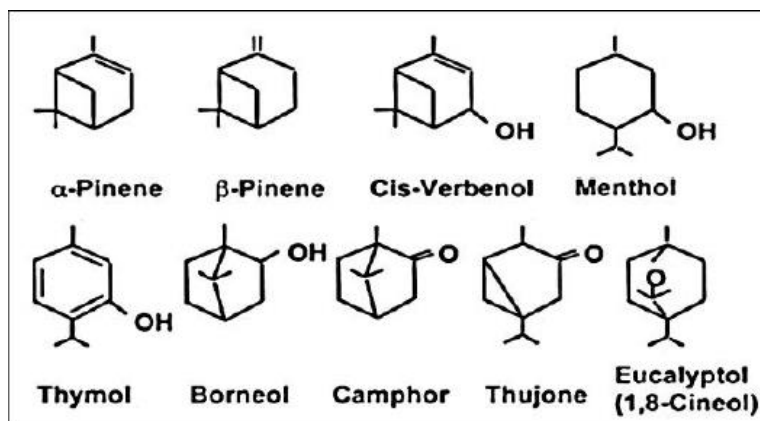
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## **APPENDIX**



## APPENDIX 1: Chemical structures: mummification materials



Structural formulas of the monoterpenes, the major components of essential oils (Mühlbauer et al., 2003).

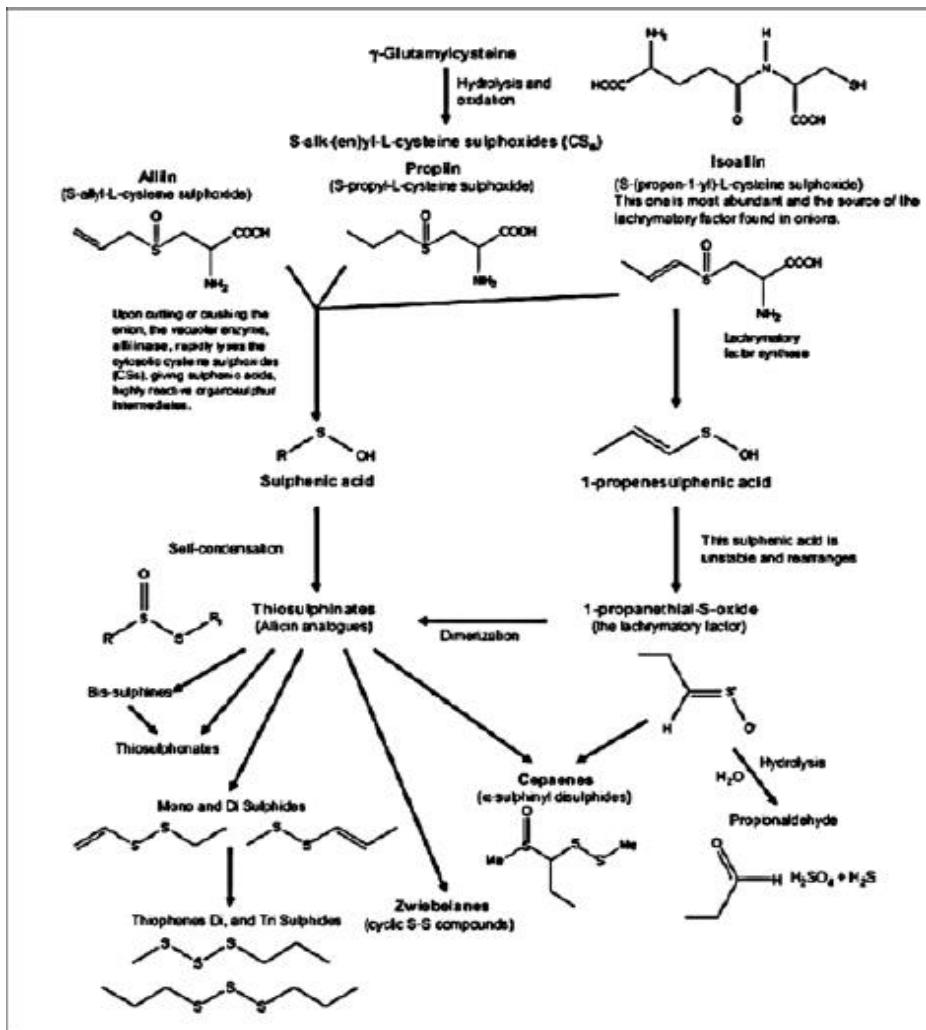
	$\alpha$ -Pinene	$\beta$ -Pinene	Camphene	Limonene	Myrcene	$\beta$ -Caryophyllene
Chemical formula	$C_{10}H_{16}$	$C_{10}H_{16}$	$C_{10}H_{16}$	$C_{10}H_{16}$	$C_{10}H_{16}$	$C_{15}H_{24}$
Structural formula						
Boiling point (°C)	156	166	160	176	167	264
Log P	4.83	4.16	4.35	4.37	4.17	6.30
Vapour pressure (mmHg) at 25 °C	4.75	2.93	2.51	1.98	2.01	-
Solubility in water (mg/l) at 25 °C	2.49	4.05	4.60	13.80	5.50	<1 mg/ml

**A**

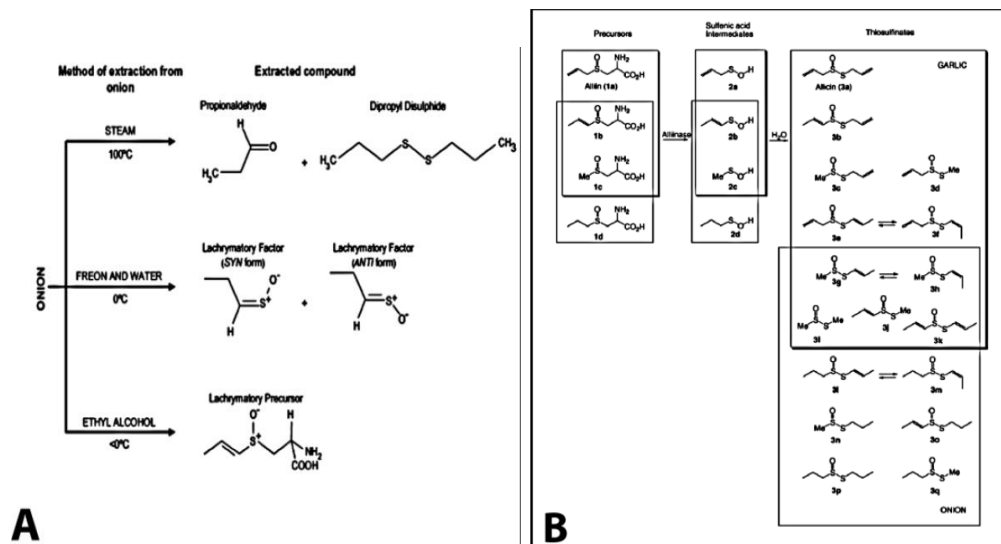
	Oleanolic acid (3 $\beta$ -hydroxy-olean-12-en-28-oic acid)	Ursolic acid (3 $\beta$ -hydroxy-urs-12-en-28-oic acid)
Structural formula		

**B**

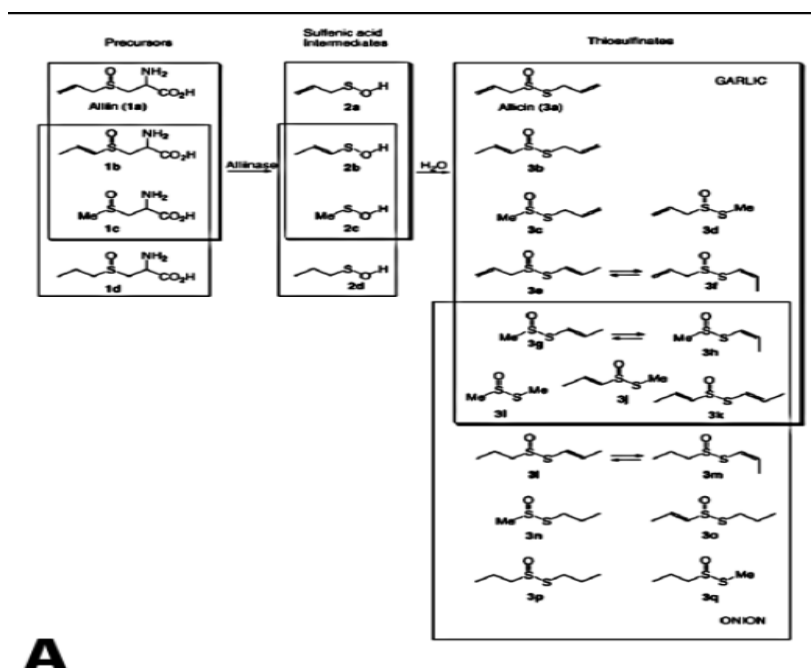
Mastic gum: (A) Physicochemical characteristics of aroma compounds (B) Chemical structure of the triterpenes (Assimopoulou et al., 2005).

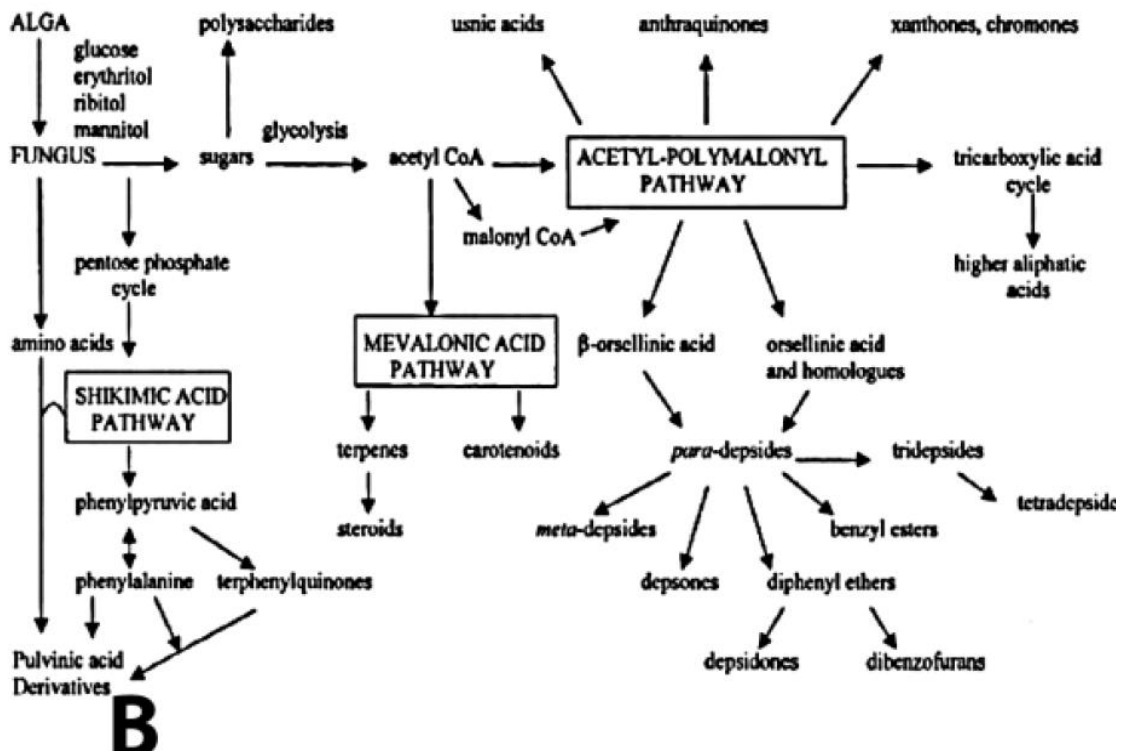


Formation of organo-sulphur compounds during metabolic pathways in processed onion (Corzo-Martinez et al., 2007).

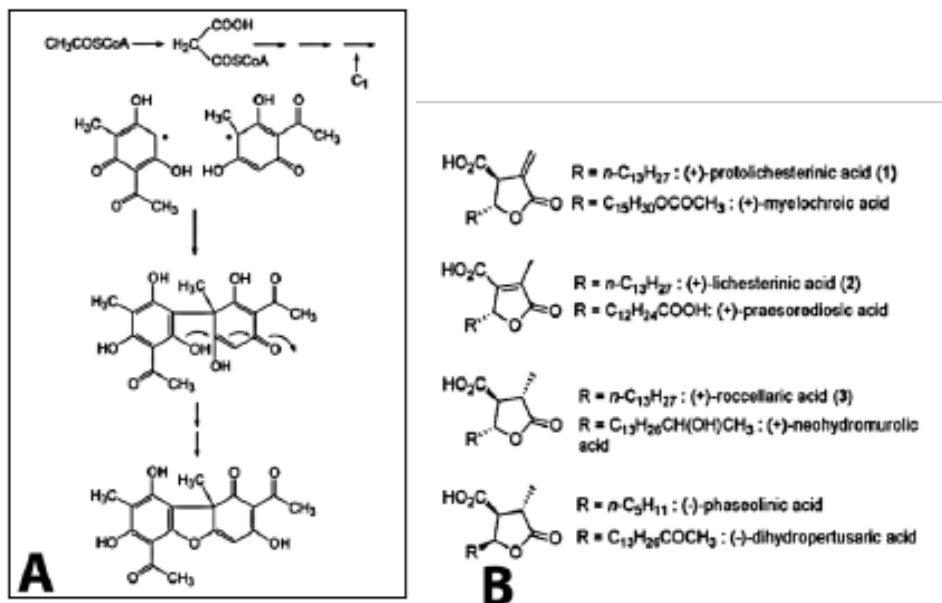


Onion: (A) Major organo-sulphur compounds present in different onion preparations based on the extraction method (Corzo-Martinez et al., 2007); (B) Biosynthetic pathway of thiosulfinates (Lanzotti, 2006).





Lichen: (A) Probable pathways leading to the major groups of lichen products (Edwards et al., 2003), (B) Proposed biosynthetic route for usnic acid (Ingoldfottir, 2002).



Lichen: (A) Structure of various paraconic acids (Horhant et al., 2007), (B) Structure of lichen compounds, sphaerophorin (depside) and pannarin (depsidome) (Russo et al., 2008).

## APPENDIX 2 Basic human anatomy

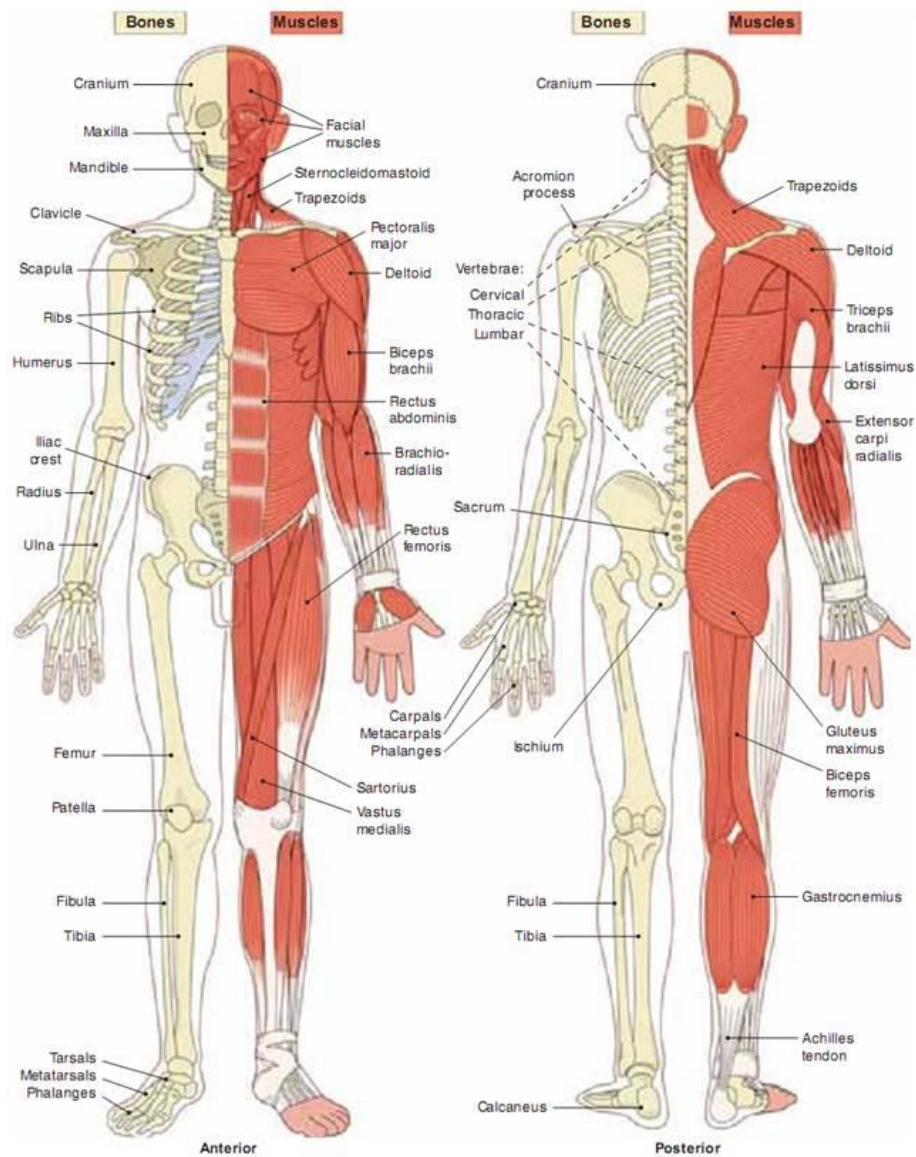
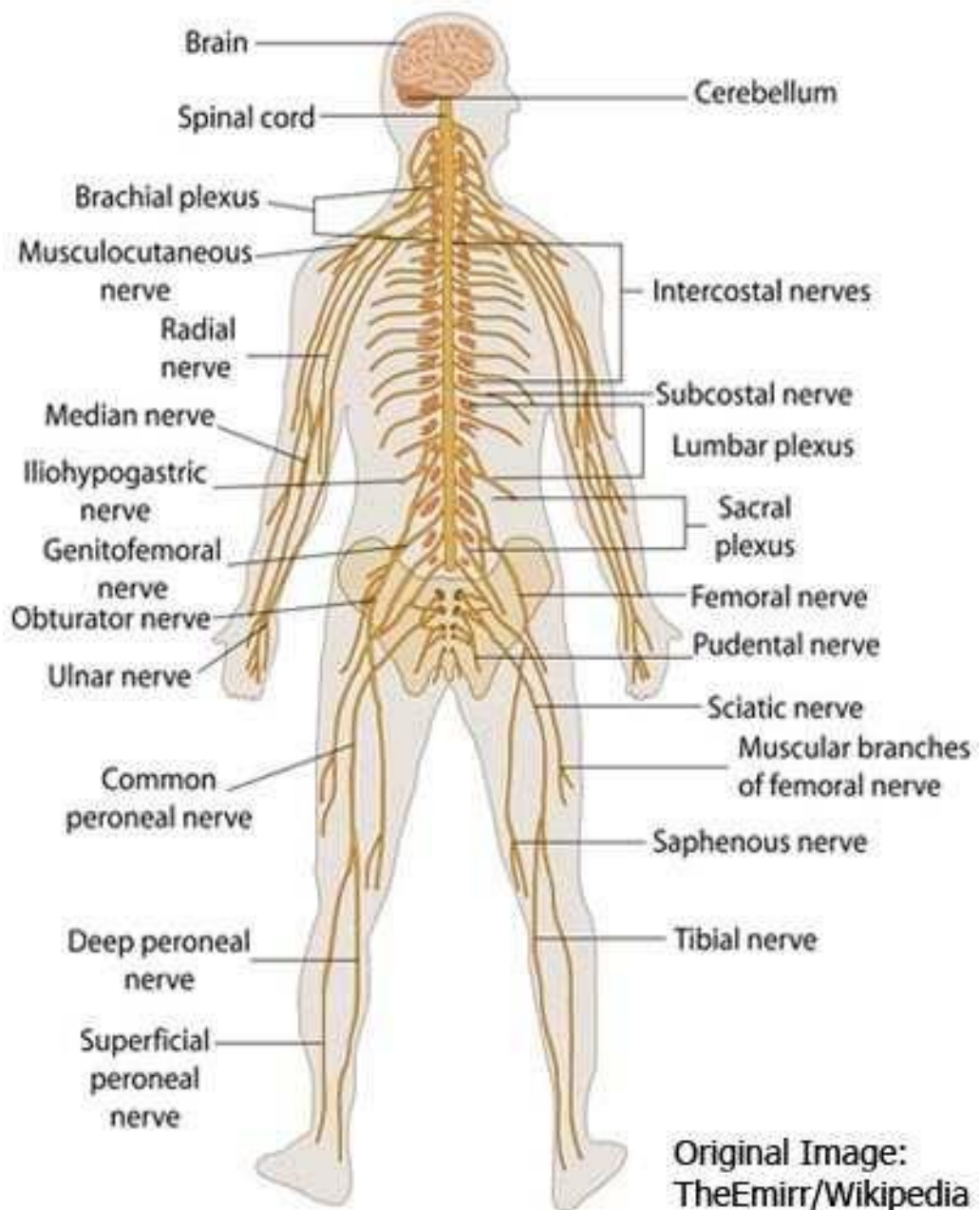
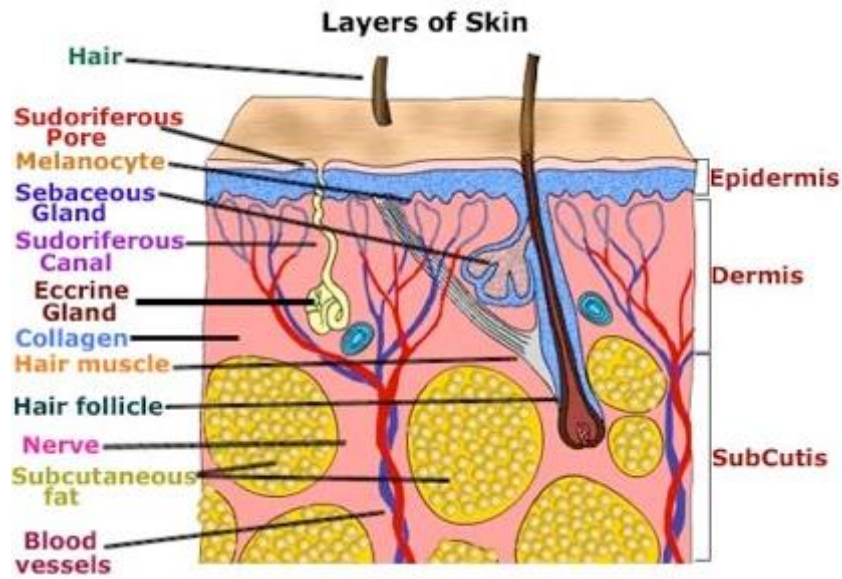


FIGURE 20.1. Cross-section of musculoskeletal system.

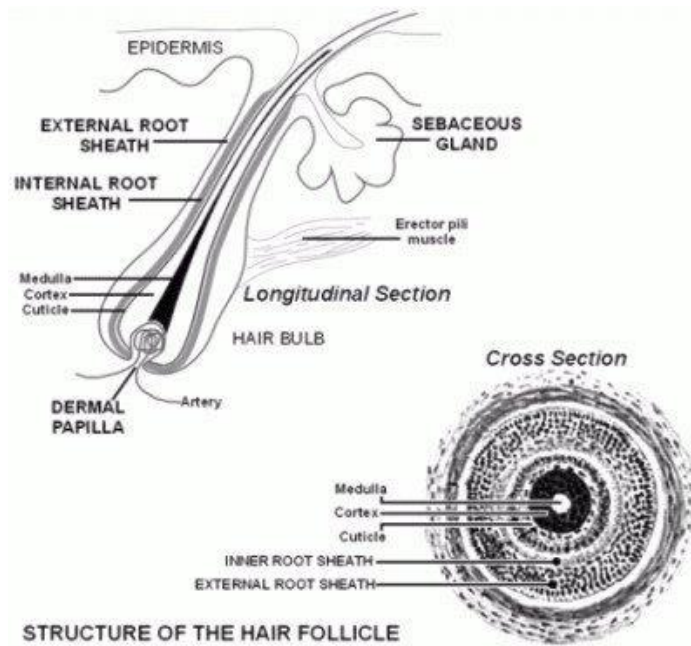
### The Locomotor system



The Nervous system

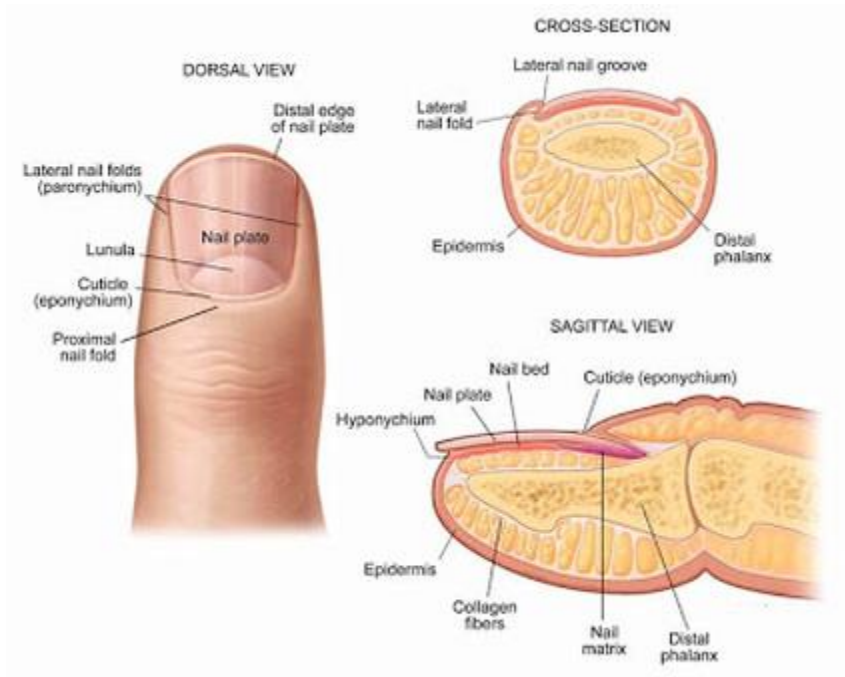


The structure of the skin (<http://sciencewithme.com/learn-about-the-skin/>)



The structure of the hair (<http://www.desalinatedwater.info/study.php>)





The structure of the nails (<http://www.medthical.com/nail-disorders.html>)

**APPENDIX 3 Examination example of mummies' study**

Site Name		Date	
Investigator(s)			
Area	Tomb No.	Mummy No.	
Photograph Nos:			
Drawing Nos:			
Body orientation			
Condition			
<b>Length</b>	<b>Width</b>	<b>Other</b>	
Wrapped	Yes__	No__	Partial__
<b>Wrappings</b>			
General description			
Colour			
Length(s)			
Width(s)			
Knots			
Hands			
Feet			
Nose plugs	Y__ N__	Ear plugs	Y__ N__
Eyes	Y__ N__	Mouth	Y__ N__
Other observations:			
<b>Body</b>			
General Description			
Position			
Colour			
Resinous materials			
Length			
Width			
Insects	Y__ N__		
Insect types			
Evisceration			
Evisceration cut			
Excerebration			
Natron			
Hair (head)	Y__ N__	Hair (body)	Y__ N__
Hair (face)	Y__ N__	Nails	Y__ N__

(Ikram, 2015)

			Head__
Gilding	Y__ N__	Location:	Nails__
Penis	Y__ N__	Circumcised	Y__ N__
Other body treatment			
Species (for animal mummy)			
Age (and basis)			
Sex (and basis)	M__	F__	Unknown__
Pathologies			
Other observations			
X-rays			
Samples			
Other			
Plant material			
Amulets & Jewellery			
Cartonnage			

(Ikram, 2015)

## APPENDIX 4 Example of Risk Assessment Form (British Museum)

### Severity

Likelihood	Low	Medium	High
Low	Low	Low	Low
Medium	Low	Medium	Medium
High	Low	Medium	High

Likelihood (L) x Severity (S) = Risk (R) (high, medium, low)

This RA is specifically designed for the Senior Clothworkers Fellowship Project: Safeguarding a body of evidence: researching and conserving a group of exceptional naturally mummified Nilotic human remains. It is adapted from the General / Job or / Area specific Assessment for controlling the risks of infection at work from human remains, carried out by Claire Messenger and Tania Watkins (AES) with specific advice from Betina Jakob, Physical Anthropologist, University of Durham. A 2007 review carried out by Sherry Doyal and Clare Ward based on advice sought from: Bill White, Curator Centre for Human Bioarchaeology MOL; Natasha Powers, Head of Osteology, MoLA; Martyn Cooke, Head of Conservation, the Royal College of Surgeons of England; Pat Potter, British School of Leather Technology, 2004. Also referencing 'Controlling the risks of infection at work from human remains' HSE 2005; 'Anthrax and historic plaster-technical advice note' English Heritage 1999 *Crypt Archaeology: an Approach*: Institute of Field Archaeologists Paper no. 3; [www.bt.cdc.gov/agent/smallpox/vaccination/faq.asp](http://www.bt.cdc.gov/agent/smallpox/vaccination/faq.asp). Further information is derived from attending a Hazards in Museum Collections course 17/11/11. Advisor was Elise Bourguignon, H&S specialist, Conservation and Scientific Research. Also [http://www.who.int/csr/resources/publications/anthrax\\_webs.pdf](http://www.who.int/csr/resources/publications/anthrax_webs.pdf), chapter 4.2 Susceptibility: data for risk assessments; WHO publication 2008, p37. Also <http://www.hsc.gov.uk/pubns/web01.pdf>: Controlling the risks of infection at work from Human Remains (Health and Safety executive).

TASK/ ACTIVITY AREA	HAZARDS	WHO'S AT RISK	CONTROLS IN PLACE	L	S	R	FURTHER CONTROLS NEEDED	WHO TO ACTION	COMPLETION DATE
The cleaning, support, sampling and study of c. 40 desiccated, recently excavated and naturally-mummified bodies from the Fourth Cataract region of the Nile. Housed in room 60. Fellowship runs over a period of 2 years (part time).  Date range 8th to 15th C AD.	Infectious disease (microbial agents or bacterial) from bodies. Risks posed by anthrax and smallpox  Soil and other material (textiles, jewellery etc.) associated with bodies.	All staff in contact with material; primarily B Wills and any co-workers.	Likelihood of disease very small ('very remote risk') because spores or infectious agents are unlikely to remain viable after 100 years, and less so in a desiccated context. People in contact should be advised of the degree of danger from infection.  Personal hygiene of great importance. Keep hands and fingernails clean, avoid hand-to-mouth contact. Wash hands and face after each episode of contact. No consumption of food and drink in proximity. Cover any cuts, wounds and abrasions with waterproof dressings.  Personal protective clothing should be routinely worn such as disposable apron, and Tyvec sleeves. Suggest disposable nitrile gloves; dust mask type FFP3.  Remove any contaminated PPE when leaving the area.  Keep surrounding areas clean and dust free.  Waste, including PPE and vacuum bags, should be disposed of correctly by incineration.  Ensure that relevant personnel are informed of the risk and risk management strategy.	L	H	L			

TASK/ ACTIVITY AREA	HAZARDS	WHO'S AT RISK	CONTROLS IN PLACE	L	S	R	FURTHER CONTROLS NEEDED	WHO TO ACTION	COMPLETION DATE
As above, focusing on removal of small area of mould on arm of skeleton 353 (3-J-23 Gr 120)	Fungal growth and spores; may be aerated by process of cleaning/ removal.	As above	Refer to mould removal RA. PPE and disposal as above. Minimise aerosol mould: experiment with the use of Groomstick (molecular trap putty) and compare with brush/ vacuum cleaner removal.  Fluctuations of humidity in storage/work area have been reduced by boarding up windows that had failed to close. This reduces risk of future mould outbreaks	L	H	L	Check environmental conditions on a regular basis.		
Removing dust and soil.	Inhalation of particles while removing dust from remains.	All staff in contact area.	Raised dust (with brush or photographers bulb puff ball) should be caught with a vacuum fitted with a high efficiency particulate air filter (HEPA filter).  As above PPE and disposal. Clean area regularly.	L	H	L	Be aware it is important to ensure that work areas are also kept clean. While removing dust from the find please be aware of where it is going in case it could pose a risk to others.		
Removing dust and soil.  Treating bodies as above	Note that at greater risk are those with compromised immunity such as pregnant women, HIV positives and those receiving chemotherapy.	All staff in contact areas who are identified as at higher risk.	All staff have a responsibility to either identify themselves as at risk or to absent themselves from the project.  As above regarding PPE and disposal. Clean area regularly. Check health status of any co-workers/students.	L	H	L	No one should assume that colleagues wish to make personal medical detail public. Make sure the risk is clearly posted to allow colleagues to absent themselves.		


Removing dust and soil.  Treating bodies as above	Note that at greater risk are those with compromised immunity such as pregnant women, HIV positives and those receiving chemotherapy.	All staff in contact areas who are identified as at higher risk.	All staff have a responsibility to either identify themselves as at risk or to absent themselves from the project.  As above regarding PPE and disposal. Clean area regularly. Check health status of any co-workers/students.	L	H	L	No one should assume that colleagues wish to make personal medical detail public. Make sure the risk is clearly posted to allow colleagues to absent themselves.		
Observing physical work with and on human remains	Psychological stress	Staff passing through room 50.  Staff in OIII.	Work area is screened off and not easily visible. The entrance to the work area clearly warns those entering, denoting the nature of the work and presence of human material.  Issues (ethical and practical) should be fully discussed if requested.  Persons should be sure to treat any excavated human remains with appropriate respect, and take steps to ensure other staff or visitors to the department are not offended	M	H	M	Check regularly with staff in OIII that controls are working.		
Physical work with and on human remains	Psychological stress	B Wills	Regular breaks, including informal discussion, should be taken to allow human responses to emotive situations. Human remains treated with respect as above.	M	H	M			

TASK/ACTIVITY AREA	HAZARDS	WHO'S AT RISK	CONTROLS IN PLACE	L	S	R	FURTHER CONTROLS NEEDED	WHO TO ACTION	COMPLETION DATE
Physical work with and on human remains		Students/ co-workers	All students etc. to be to be fully briefed on the overall project rationale and recruited carefully. To be made fully aware of the task they will be expected to undertake.  Human remains treated with respect as above. Issues (ethical and practical) should be fully discussed if requested. Regular breaks and discussions as above.	M	H	M			

## APPENDIX 5 Safety Chemical Data

<b>DICHLOROMETHANE</b>		<b>ICSC: 0058</b> Peer-Review Status: 08.06.2012 Validated
Methylene chloride DCM		
CAS #: 75-09-2 RTECS #: PA8050000 UN #: 1593 EC #: 602-004-00-3 EINECS #: 200-838-9	Formula: CH <sub>2</sub> Cl <sub>2</sub> Molecular mass: 84.9	

TYPES OF HAZARD / EXPOSURE	ACUTE HAZARDS / SYMPTOMS	PREVENTION	FIRST AID / FIRE-FIGHTING
<b>FIRE</b>	Combustible under specific conditions. Gives off irritating or toxic fumes (or gases) in a fire.		In case of fire in the surroundings, use appropriate extinguishing media.
<b>EXPLOSION</b>	Heating will cause rise in pressure with risk of bursting. Risk of fire and explosion. See Chemical Dangers.	Prevent build-up of electrostatic charges (e.g., by grounding).	In case of fire: keep drums, etc., cool by spraying with water.
<b>EXPOSURE</b>		<b>PREVENT GENERATION OF MISTS! STRICT HYGIENE!</b>	
<b>Inhalation</b>	Dizziness. Drowsiness. Headache. Nausea. Weakness. Unconsciousness. Death	Use ventilation, local exhaust or breathing protection.	Fresh air, rest. Artificial respiration may be needed. Refer immediately for medical attention.
<b>Skin</b>	Dry skin. Redness. Burning sensation.	Protective gloves. Protective clothing.	Remove contaminated clothes. Rinse and then wash skin with water and soap.
<b>Eyes</b>	Pain. Redness.	Wear safety spectacles or eye protection in combination with breathing protection.	First rinse with plenty of water for several minutes (remove contact lenses if easily possible), then refer for medical attention.
<b>Ingestion</b>	Abdominal pain. Further see Inhalation.	Do not eat, drink, or smoke during work.	Rinse mouth. Do NOT induce vomiting. Refer for medical attention.

SPILLAGE DISPOSAL	PACKAGING & LABELLING
Personal protection: self-contained breathing apparatus. Ventilation. Collect leaking and spilled liquid in sealable containers as far as possible. Absorb remaining liquid in sand or inert absorbent. Then store and dispose of according to local regulations.	Do not transport with food and feedstuffs. <b>EC Classification</b> Symbol: Xn, R: 40; S: (2)-23-24/25-36/37 <b>UN Classification</b> UN Hazard Class: 6.1; UN Pack Group: III <b>GHS Classification</b> Signal: Danger Harmful if swallowed Fatal if inhaled Causes skin and eye irritation Suspected of causing cancer Causes damage to central nervous system, blood, liver, the heart and lungs May cause drowsiness or dizziness Causes damage to the central nervous system through prolonged or repeated exposure if inhaled May be harmful if swallowed and enters airways 

EMERGENCY RESPONSE	SAFE STORAGE
NFPA Code: H2; F1; R0.	Separated from strong oxidants, strong bases, metals and food and feedstuffs. See Chemical Dangers. Cool. Well closed. Ventilation along the floor.




IMPORTANT DATA	
<p><b>Physical State; Appearance</b> COLOURLESS LIQUID WITH CHARACTERISTIC ODOUR.</p> <p><b>Physical dangers</b> The vapour is heavier than air. As a result of flow, agitation, etc., electrostatic charges can be generated.</p> <p><b>Chemical dangers</b> Decomposes on heating or on burning and on contact with hot surfaces. This produces toxic and corrosive fumes including hydrogen chloride (see ICSC 0163), phosgene (see ICSC 0007) and carbon monoxide (see ICSC 0023). Reacts violently with strong oxidants, strong bases and metals such as aluminium powder and magnesium powder. This generates fire and explosion hazard. Attacks some forms of plastic, rubber and coatings.</p> <p><b>Occupational exposure limits</b> TLV: 50ppm as TWA; A3 (confirmed animal carcinogen with unknown relevance to humans); BEI issued; (ACGIH 2011). MAK: Carcinogen category: 3A; (DFG 2011).</p>	<p><b>Routes of exposure</b> The substance can be absorbed into the body by inhalation, by ingestion and through the skin.</p> <p><b>Inhalation risk</b> A harmful contamination of the air can be reached very quickly on evaporation of this substance at 20°C.</p> <p><b>Effects of short-term exposure</b> The substance is irritating to the eyes, skin and respiratory tract. The substance may cause effects on the central nervous system, blood, liver, heart and lungs. Exposure could cause carbon monoxide poisoning. This may result in impaired functions. Exposure at high concentrations could cause lowering of consciousness and death. The effects may be delayed. If swallowed the substance may cause vomiting and could result in aspiration pneumonitis.</p> <p><b>Effects of long-term or repeated exposure</b> Repeated or prolonged contact with skin may cause dermatitis. The substance may have effects on the central nervous system. This substance is possibly carcinogenic to humans.</p>



PHYSICAL PROPERTIES	ENVIRONMENTAL DATA
Boiling point: 40°C Melting point: -95,1°C Relative density (water = 1): 1.3 Solubility in water, g/100ml at 20°C: 1.3 (moderate) Vapour pressure, kPa at 20°C: 47.4 Relative vapour density (air = 1): 2.9 Relative density of the vapour/air-mixture at 20°C (air = 1): 1.9 Auto-ignition temperature: 605°C Explosive limits, vol% in air: 12-25 Octanol/water partition coefficient as log Pow: 1.25 Viscosity: 0.15 mm <sup>2</sup> /s at 20°C	

NOTES
Addition of small amounts of a flammable substance or an increase in the oxygen content of the air strongly enhances combustibility. Depending on the degree of exposure, periodic medical examination is suggested. The odour warning when the exposure limit value is exceeded is insufficient. Do NOT use in the vicinity of a fire or a hot surface, or during welding.

ADDITIONAL INFORMATION

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**n-HEXANE** **ICSC: 0279**  
Peer-Review Status: 13.04.2000 Validated

Hexyl hydride

CAS #: 110-54-3 RTECS #: MN9275000 Formula: C<sub>6</sub>H<sub>14</sub>  
 UN #: 1208 Molecular mass: 86.2  
 EC #: 601-037-00-0  
 EINECS #: 203-777-6

TYPES OF HAZARD / EXPOSURE	ACUTE HAZARDS / SYMPTOMS	PREVENTION	FIRST AID / FIRE-FIGHTING
<b>FIRE</b>	Highly flammable.	NO open flames, NO sparks and NO smoking.	Use powder, AFFF, foam, carbon dioxide.
<b>EXPLOSION</b>	Vapour/air mixtures are explosive.	Closed system, ventilation, explosion-proof electrical equipment and lighting. Do NOT use compressed air for filling, discharging, or handling. Use non-sparking handtools.	In case of fire: keep drums, etc., cool by spraying with water.
<b>EXPOSURE</b>			
<b>Inhalation</b>	Dizziness. Drowsiness. Lethargy. Headache. Nausea. Weakness. Unconsciousness.	Use ventilation, local exhaust or breathing protection.	Fresh air, rest. Refer for medical attention.
<b>Skin</b>	Dry skin. Redness. Pain.	Protective gloves.	Remove contaminated clothes. Rinse and then wash skin with water and soap. Refer for medical attention.
<b>Eyes</b>	Redness. Pain.	Wear safety goggles, face shield or eye protection in combination with breathing protection.	First rinse with plenty of water for several minutes (remove contact lenses if easily possible), then refer for medical attention.
<b>Ingestion</b>	Abdominal pain. Further see Inhalation.	Do not eat, drink, or smoke during work.	Rinse mouth. Do NOT induce vomiting. Rest. Refer for medical attention.

SPILLAGE DISPOSAL	PACKAGING & LABELLING
Consult an expert! Personal protection: filter respirator for organic gases and vapours adapted to the airborne concentration of the substance. Remove all ignition sources. Do NOT wash away into sewer. Do NOT let this chemical enter the environment. Collect leaking and spilled liquid in sealable containers as far as possible. Absorb remaining liquid in sand or inert absorbent. Then store and dispose of according to local regulations.	<b>EC Classification</b> Symbol: F, Xn, N; R: 11-38-48/20-62-65-67-51/53; S: (2)-9-16-29-33-36/37-61-62 <b>UN Classification</b> UN Hazard Class: 3; UN Pack Group: II <b>GHS Classification</b>




EMERGENCY RESPONSE	SAFE STORAGE
Transport Emergency Card: TEC (R)-30S1208. NFPA Code: H1; F3; R0.	Fireproof. Separated from strong oxidants. Well closed.

IMPORTANT DATA	
<p><b>Physical State; Appearance</b> VOLATILE COLOURLESS LIQUID WITH CHARACTERISTIC ODOUR.</p> <p><b>Physical dangers</b> The vapour is heavier than air and may travel along the ground; distant ignition possible.</p> <p><b>Chemical dangers</b> Reacts with strong oxidants. This generates fire and explosion hazard. Attacks some plastics, rubber and coatings.</p> <p><b>Occupational exposure limits</b> TLV: 50ppm, 176mg/m<sup>3</sup> as TWA; (skin); BEI issued; (ACGIH 2004). MAK: 50 ppm, 180 mg/m<sup>3</sup>; Peak limitation category: II(8); Pregnancy risk group: C; (DFG 2004).</p>	<p><b>Routes of exposure</b> The substance can be absorbed into the body by inhalation of its vapour and by ingestion.</p> <p><b>Inhalation risk</b> A harmful contamination of the air can be reached rather quickly on evaporation of this substance at 20°C.</p> <p><b>Effects of short-term exposure</b> The substance is irritating to the skin. If this liquid is swallowed, aspiration into the lungs may result in chemical pneumonitis. Exposure at high levels could cause lowering of consciousness.</p> <p><b>Effects of long-term or repeated exposure</b> Repeated or prolonged contact with skin may cause dermatitis. The substance may have effects on the central nervous system and peripheral nervous system. This may result in polyneuropathy. Animal tests show that this substance possibly causes toxic effects upon human reproduction.</p>

PHYSICAL PROPERTIES	ENVIRONMENTAL DATA
Boiling point: 69°C Melting point: -95°C Relative density (water = 1): 0.7 Solubility in water, g/100ml at 20°C: 0.0013 Vapour pressure, kPa at 20°C: 17 Relative vapour density (air = 1): 3.0 Relative density of the vapour/air-mixture at 20°C (air = 1): 1.3 Flash point: -22°C c.c. Auto-ignition temperature: 225°C Explosive limits, vol% in air: 1.1-7.5 Octanol/water partition coefficient as log Pow: 3.9	The substance is toxic to aquatic organisms.

NOTES
Depending on the degree of exposure, periodic medical examination is suggested. Card has been partly updated in October 2004. See sections Occupational Exposure Limits, EU classification, Emergency Response.

ADDITIONAL INFORMATION

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**ETHANOL (ANHYDROUS)** **ICSC: 0044**  
Peer-Review Status: 10.02.2000 Validated

Ethyl alcohol

CAS #: 64-17-5 RTECS #: KQ6300000      Formula: CH<sub>3</sub>CH<sub>2</sub>OH / C<sub>2</sub>H<sub>6</sub>O  
 UN #: 1170      Molecular mass: 46.1  
 EC #: 603-002-00-5  
 EINECS #: 200-578-6

TYPES OF HAZARD / EXPOSURE	ACUTE HAZARDS / SYMPTOMS	PREVENTION	FIRST AID / FIRE-FIGHTING
<b>FIRE</b>	Highly flammable.	NO open flames, NO sparks and NO smoking. NO contact with strong oxidizing agents.	Use water in large amounts, powder, alcohol-resistant foam, carbon dioxide.
<b>EXPLOSION</b>	Vapour/air mixtures are explosive.	Closed system, ventilation, explosion-proof electrical equipment and lighting. Do NOT use compressed air for filling, discharging, or handling.	In case of fire: keep drums, etc., cool by spraying with water.
<b>EXPOSURE</b>			
<b>Inhalation</b>	Cough. Headache. Fatigue. Drowsiness.	Use ventilation, local exhaust or breathing protection.	Fresh air, rest.
<b>Skin</b>	Dry skin.	Protective gloves.	Remove contaminated clothes. Rinse and then wash skin with water and soap.
<b>Eyes</b>	Redness. Pain. Burning sensation.	Wear safety goggles.	First rinse with plenty of water for several minutes (remove contact lenses if easily possible), then refer for medical attention.
<b>Ingestion</b>	Burning sensation. Headache. Confusion. Dizziness. Unconsciousness.	Do not eat, drink, or smoke during work.	Rinse mouth. Refer for medical attention.

SPILLAGE DISPOSAL	PACKAGING & LABELLING
Ventilation. Remove all ignition sources. Collect leaking and spilled liquid in sealable containers as far as possible. Wash away remainder with plenty of water.	<b>EC Classification</b> Symbol: F; R: 11; S: (2)-7-16 <b>UN Classification</b> UN Hazard Class: 3; UN Pack Group: II <b>GHS Classification</b>




EMERGENCY RESPONSE	SAFE STORAGE
Transport Emergency Card: TEC (R)-30S1170. NFPA Code: H0, F3, R0.	Fireproof. Separated from strong oxidants.

IMPORTANT DATA	
<b>Physical State; Appearance</b> COLOURLESS LIQUID WITH CHARACTERISTIC ODOUR.  <b>Physical dangers</b> The vapour mixes well with air, explosive mixtures are easily formed.  <b>Chemical dangers</b> Reacts slowly with calcium hypochlorite, silver oxide and ammonia. This generates fire and explosion hazard. Reacts violently with strong oxidants such as nitric acid, silver nitrate, mercuric nitrate and magnesium perchlorate. This generates fire and explosion hazard.  <b>Occupational exposure limits</b> TLV: 1000ppm as TWA; A4 (not classifiable as a human carcinogen); (ACGIH 2004). MAK: 500 ppm, 960 mg/m <sup>3</sup> ; Carcinogen category: 5; Peak limitation category: II(2); Pregnancy risk group: C; (DFG 2004).	<b>Routes of exposure</b> The substance can be absorbed into the body by inhalation of its vapour and by ingestion.  <b>Inhalation risk</b> A harmful contamination of the air will be reached rather slowly on evaporation of this substance at 20°C.  <b>Effects of short-term exposure</b> The substance is irritating to the eyes. Inhalation of high concentrations of the vapour may cause irritation of the eyes and respiratory tract. The substance may cause effects on the central nervous system.  <b>Effects of long-term or repeated exposure</b> The substance defats the skin, which may cause dryness or cracking. The substance may have effects on the upper respiratory tract and central nervous system. This may result in irritation, headache, fatigue and lack of concentration. See Notes.

PHYSICAL PROPERTIES	ENVIRONMENTAL DATA
Boiling point: 79°C Melting point: -117°C Relative density (water = 1): 0.8 Solubility in water: miscible Vapour pressure, kPa at 20°C: 5.8 Relative vapour density (air = 1): 1.6 Relative density of the vapour/air-mixture at 20°C (air = 1): 1.03 Flash point: 13°C c.c. Auto-ignition temperature: 363°C Explosive limits, vol% in air: 3.3-19 Octanol/water partition coefficient as log Pow: -0.32	

NOTES
Ethanol consumption during pregnancy may adversely affect the unborn child. Chronic ingestion of ethanol may cause liver cirrhosis. The flash point of 50% water solution is 24°C. Card has been partially updated in April 2005: see Occupational Exposure Limits.

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