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## Improved discrimination of biogenic and diagenetic elements in Palaeolithic mammoth ivory and bone from Hohle Fels Cave in the Swabian Jura of Southwestern Germany

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### ABSTRACT

Mammoth ivory was used by humans to manufacture personal ornaments, sculptures or music instruments during the Upper Palaeolithic. These objects are among the first and most precious witnesses of ancient artistic behaviour. Archaeological ivory, however, has been subjected to complex alteration processes due to exchange with its burial environment over time. Therefore, it is necessary to understand the diagenetic phenomena in order to develop adequate conservation measures for ivory artefacts. The element-analytical study of ivory artefacts can shed light on these processes and can help to track the origin of these objects.

In this work we studied twelve mammoth ivory samples and ten bone samples coming from the Palaeolithic site Hohle Fels located in the Swabian Jura. Mammoth bone samples were included in the study in order to find eventual differences in the diagenetic changes between ivory and bone material. We performed ion beam analysis (PIXE/PIGE) at the microfocus beamline of the accelerator NewAGLAE at C2RMF and micro-X-ray fluorescence (XRF) mappings at the PUMA beamline at synchrotron SOLEIL. The obtained PIXE/PIGE results are consistent with previous studies of ivories from Hohle Fels and other contemporaneous European sites in terms of possible site-specific (Zn, Br, Sr contents) and diagenetic markers (F content). XRF mappings and their semi-quantitative analyses confirm former observations and provide additional criteria for the classification of endogenous and exogenous elements. Based on our results we could not find significant difference in the diagenetic alterations between archaeological ivory and bone samples for the studied archaeological site.

### 1. Introduction

Among prehistoric finds, artefacts made of mammoth ivory play an important role. As a valuable raw material, it was used for the first jewelry and art objects. These objects are closely related to the appearance of modern humans in Europe. During archaeological excavations, numerous objects made of mammoth ivory and bone were discovered in the caves in the Ach and Lone valleys, Swabian Alb, in Aurignacian-era layers (Conard, 2003; Conard and Malina, 2007, 2008; Conard, 2009; Ebinger-Rist et al., 2018). Aurignacian in Swabia is even sometimes referred as “Ivory age” (Conard, 2021, 2018). The Hohle Fels is a karst cave in the Achtal near Schelklingen in Baden-Württemberg. Since the 19th century, prehistoric finds have been discovered in the cave. Very

noteworthy are the female figurine from Hohle Fels brought to light in 2008 in layer V (older Aurignacian, at least 33 ka cal BP) as well as animal figures and musical instruments like flutes made from mammoth ivory (Conard, 2021, 2018; Conard et al., 2004, 2009). The cave is one of the most important sites of the Upper Palaeolithic in Central Europe and was included in the list of UNESCO World Heritage Sites in 2017. It has already been frequented by *Homo sapiens neanderthalensis*, followed by *Homo sapiens sapiens*. Therefore, the artefacts from Hohle Fels represent suitable objects to investigate the chronological variations in the regional and supra-regional use of the raw material mammoth ivory in

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the Aurignacian, and thus the facets of the way of life of early *Homo sapiens sapiens* in Europe.

Mammoth ivory, like elephant ivory and bone, consists of an organic phase, collagen, and a mineral phase, apatite, whose ratio, however, is diagenetically altered in prehistoric artefacts compared to recent ivory and bone depending on the storage conditions (Müller and Reiche, 2011). The aim of the investigations is to use the chemical composition of the Aurignacian mammoth ivory artefacts to identify elements as site-specific markers in the ivory that are related to the habitat of the mammoths (endogenous or biogenic) on the one hand or to the changes in the artefacts over time depending on the local storage conditions (exogenous or diagenetic) on the other.

NewAGLAE and PUMA offer complementary non-invasive methods to study the entire chemical composition of prehistoric ivory and bone at major, minor and trace element levels. Chemical imaging of light to heavy elements at these facilities in Palaeolithic artefacts, makes it possible to determine reliable markers for site-specific and diagenetic characteristics. The data on site-specific elements of the mammoth ivory from the Hohle Fels Cave will be compared with those of other European sites (Reiche et al., 2018) as well as with bone material in the further course of the project in order to be able to draw conclusions about the regional and/or supra-regional use of this symbolic raw material.

This work paves the way to detect new biogenic and diagenetic elemental markers in archaeological ivory and bone.

## 2. Description of ivory and potential markers of biogenic or diagenetic origin

Ivory is generally associated with the dentin part of the exoskeletal incisor teeth or tusks of several mammalian species, mainly elephant and mammoth (*Laxodonta africana*, *Elephas maximus*, *Mammuthus*). Inside the tusk is the pulp corresponding to a soft tissue including blood vessel and nerves. Dentin of tusks is a composite material made up of an organic and inorganic fraction, which are intimately mixed on a nanometre scale. Dentin has basically the same composition as bone but differs in its tubular micromorphology. The organic phase (20 wt.%) is mainly composed of a collagen matrix. Collagen is a protein macromolecule, whose formula can be resumed as  $(\text{Gly-X-Y})_{338 \pm 2}$ , where Gly is the amino acid glycine, present in high amounts, and X and Y are other collagen-characterizing amino acids such as proline and hydroxyproline. The inorganic phase (70 wt.%) corresponds to a poorly crystalline carbonated hydroxyapatite  $(\text{Ca}_{8.3}\square_{1.7}(\text{PO}_4)_{4.3}(\text{HPO}_4, \text{CO}_3)_{1.7}(\text{OH})_{0.3}\square_{1.7}$ , carb. HAP) (Rey et al., 1995) with a hexagonal crystal lattice  $(\text{P6}_3/m)$ . In addition, they contain about 10 wt.% of water. The organic matrix is first formed by odontoblasts and then mineralized during dentinogenesis. The mineralization continues with ages. Therefore, ivory can be considered as a lifelong archive of environmental and climatic conditions of the animal individual. It registers a wealth of information in its chemical and isotopic composition during growth. The elemental composition varies significantly from the central tusk cavity to the cement layer of the tusk. Variations in trace element concentrations (Mg, Zn, Sr) have been correlated to the growth lines in order to derive from it information on the environmental conditions during life time (Müller and Reiche, 2011; Prozesky et al., 1995; Finch and Allison, 2007; Politi et al., 2010). Ivory dentin is rich in Mg (about 6 wt.%) in comparison to other tooth dentin and bone and Mg, being a key component for the stabilization of amorphous carbonate, is supposed to play an important role in biomineralization. Hence, several studies have been focused on this subject (Müller and Reiche, 2011; Prozesky et al., 1995; Finch and Allison, 2007; Politi et al., 2010).

Therefore, Mg can be considered as a key chemical element for identifying ivory material with some limitations because Mg concentration decreases during diagenesis (Müller and Reiche, 2011; Rey et al., 2022). Other important biogenic elements are Zn and Sr (Grupe and

Herrmann, 2012; Tütken and Vennemann, 2011; Geigl et al., 2004; Lee-Thorp and Sponheimer, 2006; Zazzo and Saliège, 2011). Elemental and stable isotopic composition of bioapatite (carb. HAP, enamel and dentin) and collagen (dentin) can be investigated to infer palaeoclimatic/palaeoenvironmental conditions as well as species behaviours. The oxygen isotope composition ( $\delta^{18}\text{O}$ ) of phosphates and carbonates of the bioapatite is related to that of local water composition and can thus be used to relate to the climatic condition during life of an animal. Carbon ( $\delta^{13}\text{C}$ ), nitrogen ( $\delta^{15}\text{N}$ ) and sulphur ( $\delta^{34}\text{S}$ ) isotope compositions of collagen can be used to determine feeding behaviours of fossil fauna and identify trophic level (Grupe and Herrmann, 2012; Tütken and Vennemann, 2011; Geigl et al., 2004; Lee-Thorp and Sponheimer, 2006; Zazzo and Saliège, 2011). Carbon isotope ratio ( $\delta^{13}\text{C}$ ) can be obtained from bioapatite. Mobility and provenance can be investigated by strontium isotope composition ( $^{87}\text{Sr}/^{86}\text{Sr}$ ) (Rey et al., 2022). The development of high resolution isotope analysis in dentin and enamel teeth can allow reconstructing modifications of climates or diet, as well as migrations during the life of mammal specimens (Grupe and Herrmann, 2012; Tütken and Vennemann, 2011; Geigl et al., 2004; Lee-Thorp and Sponheimer, 2006; Zazzo and Saliège, 2011). It is thus expected that it is also possible to use specific trace elements for these issues if their biogenic nature can be confirmed.

In previous research (Reiche et al., 2018) it was shown that the evaluation of the state of preservation of mammoth ivory material is crucial for assessing its full informative potential. By analysing mammoth ivory artefacts originating from different key Palaeolithic sites (Abri Castanet, the caves La Verpillière I and II in Germolles and the Vogelherd Cave, etc.) in France and Germany using micro-PIXE analysis, a non-invasive evaluation of the preservation state (via their Mg vs. Ca vs. F contents) and the definition of potential provenance markers contained in the mineral fraction (Sr or Zn vs. Br) of the studied mammoth ivory objects were possible. Other elements such as Mn, Fe and F were found to be uptaken over time in ivory and are typically chemical elements in ivory linked to diagenetic pathways in the burial environment (Reiche et al., 2018; Heckel et al., 2014, 2016). Especially the uptake of F is interesting as it can be considered for relative dating. F uptake in bone and ivory over time is a widespread, fairly consistent, and extensively studied phenomenon. However, previous research (Heckel et al., 2014) on F content in archaeological ivory from Early Aurignacian deposits produced highly variable levels of F in some contexts, and highly uniform patterns in others. Observed variations in F content may be due to localized differences in sediment chemistry and/or hydrology, to the unique structural features of ivory tusks, or to different approaches to ivory procurement (at the time fresh or already aged ivory).

In order to validate the potential provenance markers and for searching new criteria contained in the chemical composition, further analyses and imaging of Palaeolithic bone and ivory samples were performed in this study.

## 3. Material and methods

A selection of Aurignacian-era unworked mammoth ivory and bone artefacts from the Hohle Fels Cave (see Table 1) has been investigated using micro-Proton-induced X-ray Emission and Gamma-ray Emission (micro-PIXE-PIGE) at the microfocus beamline of the NewAGLAE particle accelerator. PIXE and PIGE are ion beam analyses (IBA) that involve the use of high energy ions produced in a particle accelerator to probe in a very sensitive manner the chemical composition at the surface and near-surface layers. The samples were also analysed by means of synchrotron-induced micro-X-ray fluorescence imaging on the PUMA beamline at the synchrotron SOLEIL. Both measuring stations allow non-invasive examinations, with a few exceptions, of the artefacts with high sensitivity and high spatial resolution. We completed these measurements with the analysis of several reference materials (two mammoth ivory samples aged of 10 000 years conserved in permafrost, a modern elephant ivory and a modern elephant bone sample).

**Table 1**

Archaeological references of the mammoth ivory and bone samples from Hohle Fels. The Aurignacian archaeological and “geological” litho-stratigraphic horizons are explained in the following Refs. [Bataille and Conard \(2018\)](#), [Goldberg et al. \(2003\)](#).

Ivory sample number	Excavated object number	Box grid number	Geological horizon	Archaeological horizon	Year of excavation	Artefact type
AI-HF-01	1685	100	3d	IId	2014	Unworked ivory fragment
AI-HF-02	1743	100	3da	IIda	2014	Unworked ivory fragment
AI-HF-03	1998	100	6a	IIIa	2014	Unworked ivory fragment
AI-HF-04	593	54	7	IV	2013	Unworked ivory fragment
AI-HF-05	551	64	7	IV	2013	Unworked ivory fragment
AI-HF-06	849	27	7a	Va	2013	Unworked ivory fragment
AI-HF-07	1370.1	27	7a	Va	2013	Unworked ivory fragment
AI-HF-08	2119.1	28	7ab	Vab	2014	Unworked ivory fragment
AI-HF-09	2346	31	7aa	Vaa	2016	Unworked ivory fragment
AI-HF-10	1942	28	7aa	Vaa	2014	Unworked ivory fragment
AI-HF-11	2067.1-2	28	7ab	Vab	2014	Unworked ivory fragment
AI-HF-12	1109	10	8	Vb	2007	Unworked ivory fragment
Bone sample number	Excavated object number	Box grid number	Geological horizon	Archaeological horizon	Year of excavation	Artefact type
AB-HF-01	1709	27	7a	Va	2013	Unworked bone fragment
AB-HF-02	1814	27	7a	Va	2013	Unworked bone fragment
AB-HF-03	1969	27	7aa	Vaa	2014	Unworked bone fragment
AB-HF-04	1847	27	3d	IIac	2014	Unworked bone fragment
AB-HF-05	1649	31	7a	Va	2015	Unworked bone fragment
AB-HF-06	1387	30	7aa	Vaa	2016	Unworked bone fragment
AB-HF-07	1520	31	7a	Va	2015	Unworked bone fragment
AB-HF-08	1613	31	7a	Va	2015	Unworked bone fragment
AB-HF-09	1150	101	3d	IId	2016	Unworked bone fragment
AB-HF-10	1362	110	3d	IIac	2015	Unworked bone fragment

By means of the combination of methods, the chemical elements from C to U and their distribution at the major (above 1 wt.%), minor (in the range of 0.1–1 wt.%) and trace (less than 0.1 wt.%) element levels can be determined non-destructively qualitatively and quantitatively. New AGLAE/C2RMF is a 2 MV tandem accelerator (Pelletron 6SDH-2) and enables combined 3 MeV micro-PIXE/PIGE/RBS imaging with a spatial resolution up to  $(6 \times 3) \mu\text{m}^2$  at the microfocus beamline. An area up to  $(20 \times 20) \text{cm}^2$  can be analysed. The measuring head includes five PIXE, one RBS outdoor detector, one PIGE detector and one microscope ([Pichon et al., 2014](#)).

For the investigations at PUMA/SOLEIL, the micro beam structure was used with an incident beam having an energy of 18 keV and a size of  $7 \mu\text{m} \times 5 \mu\text{m}$ . The sample is tilted at  $45^\circ$  to the beam and the SDD detector is placed at an angle of  $90^\circ$  toward the incoming beam. An optical microscope is used to visualize the area probed by the X-ray incident beam. More information on the setup at PUMA beamline can be found in the following references [Tack et al. \(2019\)](#) and [Gianoncelli et al. \(2020\)](#).

A more complete description of the possibilities offered by these two instruments to analyse prehistoric ivory will be presented in a forthcoming paper. Briefly, New AGLAE has a high sensitivity to detect light elements ( $Z \leq 20$ ) making it possible in particular to characterize the lighter elements of the mineral phase of ivory, while PUMA has a high sensitivity to detect heavier elements ( $Z \geq 20$ ) especially at trace level allowing to image them.

#### 4. Data acquisition

A total of 36 XRF and 41 PIXE mappings has been carried out on the archaeological and reference samples. The mapping sizes range from  $0.4 \text{ mm} \times 0.5 \text{ mm}$  to  $1.5 \text{ mm} \times 1.5 \text{ mm}$  for archaeological ivories and from  $0.5 \text{ mm} \times 0.5 \text{ mm}$  to  $1.7 \text{ mm} \times 4 \text{ mm}$  for archaeological bones. Mappings of a few  $\text{mm}^2$  have been carried out for reference samples. XRF mappings have been treated with the PyMCA software ([Solé et al., 2007](#)) and the concentrations of PIXE mappings have been obtained with the GUPIX and TRAUPIX softwares ([Campbell et al., 2000](#); [Pichon et al., 2015](#)).

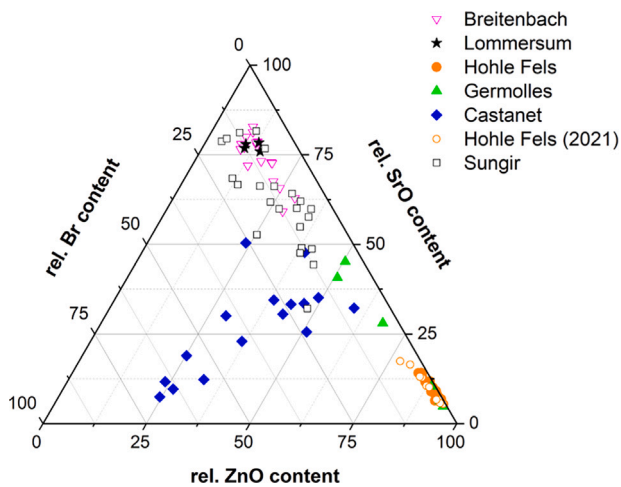


Fig. 1. Comparison of the relative contents of biogenic trace elements (Br and Sr, Zn the latter two expressed as oxides) for ivory samples of Hohle Fels Cave compared with other archaeological sites previously studied (Reiche et al., 2018). The sum of the absolute concentrations of these three elements was normalized to 100% for each sample and the obtained relative amounts were plotted in a ternary diagram. Hohle Fels data 2021 correspond to the ivory samples specifically studied in this paper whose archaeological references are mentioned in Table 1.

## 5. PIXE/PIGE study of Hohle Fels samples

### 5.1. Comparison with other European sites

In Figs. 1 and 2 the PIXE/PIGE results obtained for all studied ivory samples from Hohle Fels Cave are compared to those obtained in previous studies (Reiche et al., 2018; Heckel et al., 2014, 2016) for ivory samples coming from other European archaeological sites. The relative contents of biogenic trace elements (Sr, Br, Zn) are plotted as oxides except Br in a ternary diagram (Fig. 1). By the way Br was also discussed as a possible bio-essential diet marker in collagen and bones by other scholars (McCall et al., 2014; Kozachuk et al., 2020). Fig. 1 allowed to reveal site-specific patterns of both trace-element composition and dispersion (Reiche et al., 2018). The Hohle Fels objects are characterized by low SrO and Br contents and a high ZnO content and thus differ from other archaeological sites. The F contents and the ratios of MgO to CaO were considered as indicators for diagenetic changes (Müller and Reiche, 2011; Heckel et al., 2014) and are plotted against each other in Fig. 2 for all studied sites. The Hohle Fels samples show relatively low amount of F and similar MgO/CaO regarding to the other sites but low MgO/CaO in comparison with modern ivory (Müller and Reiche, 2011). This indicates that these samples have been modified by diagenesis. The archaeological sites examined here differ primarily in terms of the F content. However, the F-content of the ivory samples, which can be used as an indicator of their relative age, is affected by a number of factors in the depositional environment and cannot be reliably compared between sites in this context. The data can be interpreted for relative dating in terms of dispersion of values within one site but not in terms of absolute values. In the case of Hohle Fels, the F content shows a low dispersion indicating rather the use of fresh ivory (Heckel et al., 2016). The observed differences in the PIXE/PIGE results may be related to the respective type of archaeological site: Hohle Fels and Germolles are cave sites, Castanet is a rock shelter and the others (Breitenbach, Lommersum and Sungir) are open air sites (Reiche et al., 2021). We stated in a previous study that low F content dispersion within one site, as it is the case for Hohle Fels, is a sign that fresh ivory was used at this site during the Aurignacian (Heckel et al., 2016).

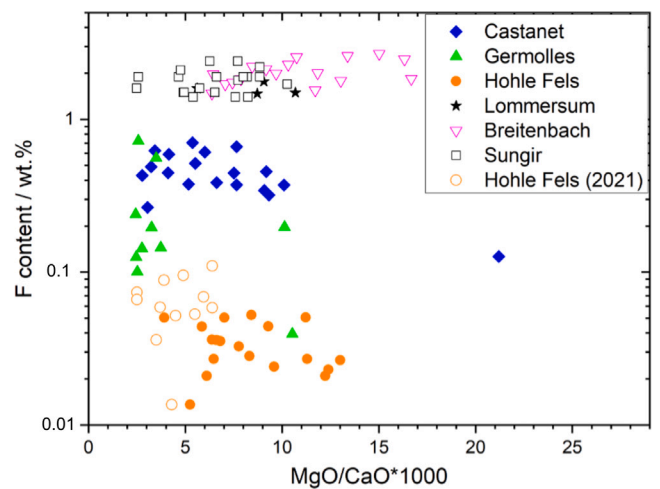


Fig. 2. Plot of the F contents versus the MgO to CaO ratios for all studied archaeological sites (estimated limit of detection for F is 0.01–0.02 wt.%). Data published elsewhere are included in all Figures and the discussion (Reiche et al., 2018; Heckel et al., 2016). For each prehistoric site dispersion of F contents and not their absolute values should be considered for interpretation. Hohle Fels data 2021 correspond to the ivory samples specifically studied in this paper whose archaeological references are mentioned in Table 1.

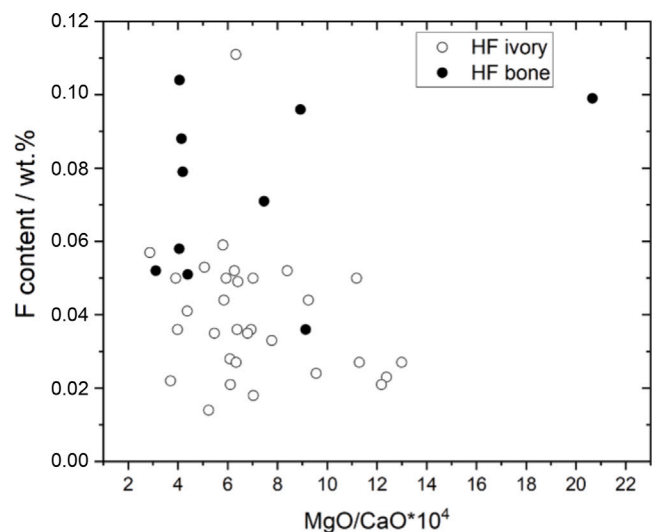
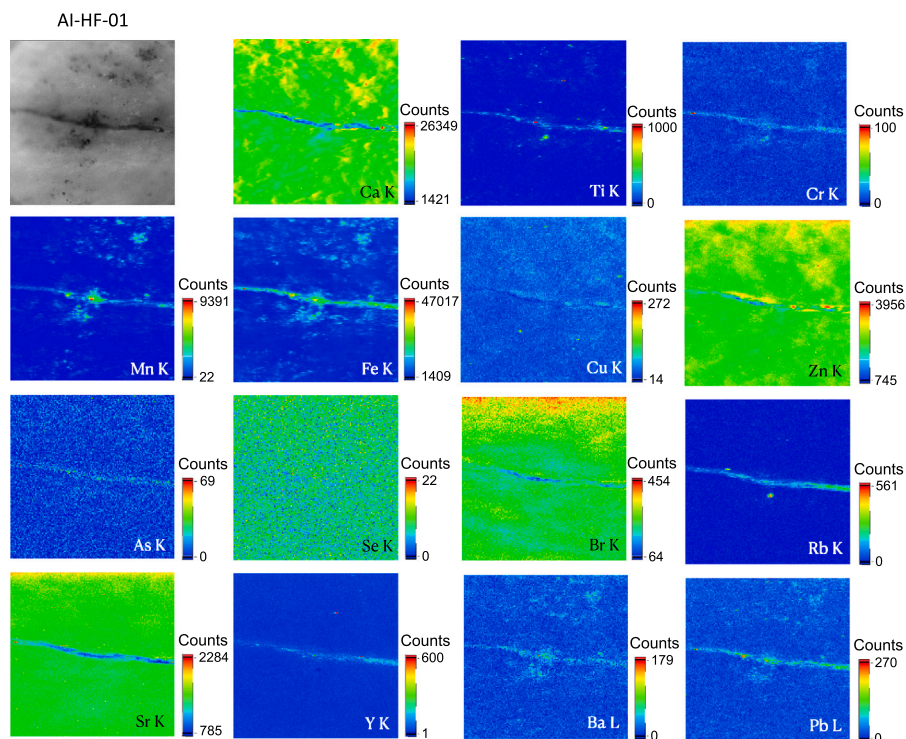


Fig. 3. Plot of the F contents versus the MgO to CaO ratios for all studied ivory and bone samples from Hohle Fels cave (estimated limit of detection for F is 0.01–0.02 wt.%). In comparison, the F-content of well-preserved permafrost mammoth ivory was determined to be about 0.04 wt.%.

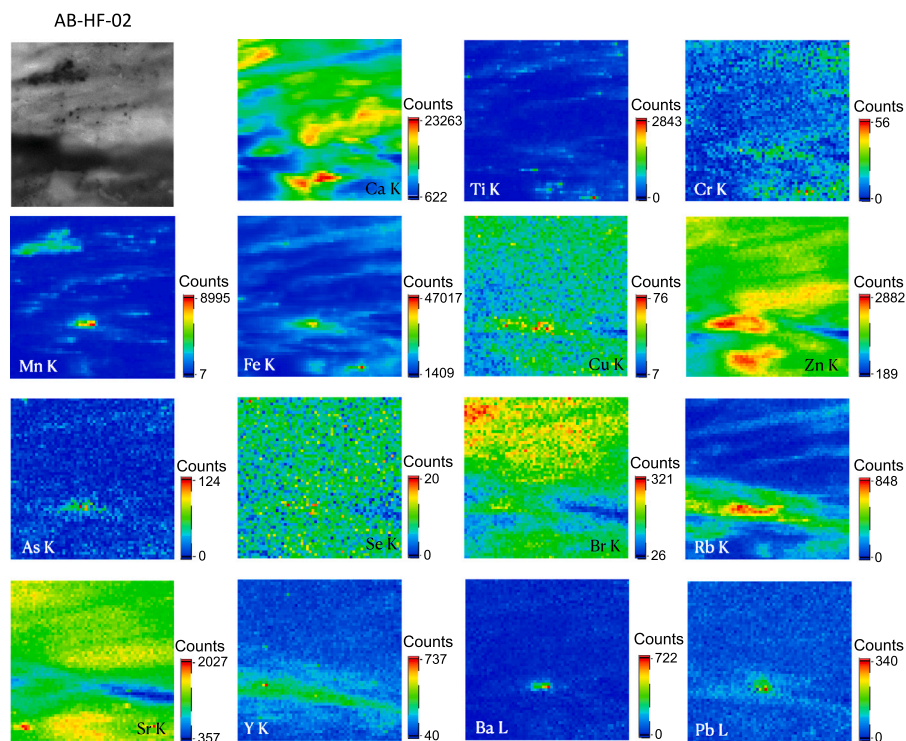
### 5.2. Differences in the diagenetic processes between mammoth ivory and bone

Over time, the chemical compositions of the different kinds of bone material change as result of the diagenetic alteration processes during burial. Dilution of soluble components as magnesium compounds, and uptake of secondary chemical species, e.g. F, Mn and Fe, take place. The extent to which the exchange of chemical species takes place depends on the environmental conditions. For the Hohle Fels Cave, we examined ivory and bone samples to find out whether these diagenetic processes lead to the same chemical changes in ivory and bone. We assume that the environmental conditions within one archaeological site were approximately the same for all findings.





**Fig. 4.** Microscope image of an area of 1.3 mm × 1.3 mm of the archaeological ivory sample AI-HF-01 and XRF mappings of various elements. Mappings correspond to a slightly larger zone (1.5 mm × 1.5 mm) than the zone presented in the microscope image. The spatial resolution is 10 μm and the dwell time per pixel is 0.5 s.



**Fig. 5.** Microscope image of an area of 0.65 mm × 0.65 mm of the archaeological bone sample AB-HF-02 and XRF mappings of various elements. Mappings correspond to a slightly smaller zone (0.6 mm × 0.6 mm) than the zone presented in the microscope image. The spatial resolution is 10 μm and the dwell time per pixel is 0.5 s.

Based on the quantitative results of PIXE measurements, we could not find any significant differences between the chemical compositions of bone and ivory from Hohle Fels. However, a very slight difference in F content could be noticed between both materials (Fig. 3). The mean

F-content of the bone finds is slightly higher in comparison to those of ivory. The latter roughly corresponds to the F-content of well-preserved permafrost mammoth ivory (ca. 0,04 wt.% F). This is consistent with the excellent biomolecular preservation at Hohle Fels, as evidenced by

the detection of ancient mammalian mtDNA in archaeological sediment collected from this site (Massilani et al., 2022). This is probably because of the constant cool temperature and constantly moist sediments.

The new PIXE data for ivory samples from Hohle Fels are consistent with previous analyses and provide reliable information on trace elements whose concentrations are lower than 100 ppm. Synchrotron XRF measurements are a powerful complementary tool to probe these elements, which could be potentially biogenic chemical markers of ivory, in archaeological samples.

## 6. Synchrotron XRF study of Hohle Fels samples

### 6.1. Imaging of archaeological ivory and bone

We present the XRF mappings of two samples: an archaeological ivory sample AI-HF-01 (Fig. 4) and an archaeological bone sample AB-HF-02 (Fig. 5) realized at the PUMA beamline. The endogenous elements Ca, Zn, Br and Sr (Reiche et al., 2018) show a homogeneous distribution on the surface and relatively high intensities for both samples. In the case of the ivory, the crack is the area containing the least amount of these elements. In the case of the bone, the distributions of these elements appear heterogeneous at first glance, but this is due to the morphology of the sample. The sample has an intrinsic structure visible in the microscopic image that is reflected in the Ca distribution and Zn, Sr, and Br show very similar patterns. Taking this into account, the distributions of these elements can also be considered homogeneous in the bone. On the contrary exogenous elements are heterogeneously distributed over the sample surface. For AI-HF-01 the crack and the little black dots correspond to the areas rich in Fe and in Mn. We observe the same agreement between the black dots and the Mn and Fe XRF mappings. Concerning the other trace elements detected by XRF, Ti, Cr, Rb, Ba and Pb are mainly located in the crack and in some black dots. Arsenic and Y are mainly located in the crack while Cu tends to have a homogeneous distribution but a very low intensity. The mapping of Se is too noisy to be conclusive. For the bone sample we observe also a heterogeneous distribution for Ti, Cr, As, Rb, Y, Pb and Ba and their mappings show some similarities with those of Mn and Fe. Cu and Se show homogeneous distributions for AB-HF-02 but the intensities are very low making the mapping noisy and difficult to analyse.

### 6.2. Semi-quantitative analysis with XRF

**Normalization.** The quantitative analysis of the XRF mappings depends strongly on several parameters like the incident beam flux, the distance between the detector and the sample, the density and the matrix of the samples. In order to limit the effects of the uncertainties of these different parameters on the quantitative analysis we normalized the weight concentrations obtained with PyMCA by the weight concentration of Sr. We considered only heavier trace elements than Ti because lighter elements are better detected by PIXE. The choice to use Sr to normalize the weight concentrations is driven by two reasons: its spatial distribution is homogeneous in archaeological or modern bone and ivory samples (see Figs. 4 and 5) and the concentration of Sr measured by PIXE showed constant values for the ivory and the bone samples in the whole corpus of Hohle Fels (see Fig. 6). Moreover the concentrations of Sr in archaeological ivory samples from Hohle Fels are similar to those of the reference samples (permafrost mammoth ivory and modern elephant ivory).

**Results.** The normalized concentrations are presented in Fig. 7 for ivory and in Fig. 8 for bone. We can divide the trace elements in three groups. First the elements whose concentrations vary strongly within the corpus of archaeological samples, that is variations larger than one order of magnitude, are Cr, Rb, Ba and Pb. Second the elements with moderately varying concentrations within the corpus but for which the differences between archaeological samples and reference samples

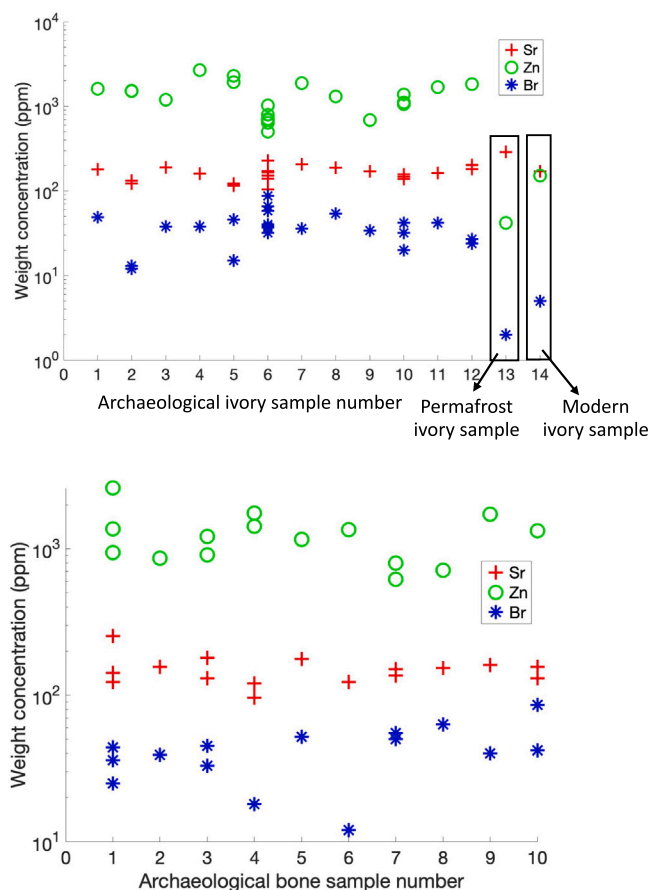


Fig. 6. Weight concentrations of Sr, Zn and Br measured by PIXE for archaeological ivory samples and a permafrost ivory sample and modern ivory sample (upper plot) and for archaeological bone samples (lower plot).

(permafrost ivory, modern ivory, modern bone) are large. This is the case for Ti, Mn, Fe, Zn, Se, Br and Y. Finally the elements whose concentrations are relatively constant for all samples are Cu and As. We propose an interpretation of these three groups in the following section.

### 6.3. Distinction of exogenous and endogenous elements

Based on the XRF mappings (Figs. 4 and 5) and the normalized elemental concentrations (Figs. 7 and 8) we discuss the differentiation of exogenous and endogenous elements. Previous PIXE studies (Reiche et al., 1999, 2018) showed that Zn, Br and Sr were endogenous elements and Mn and Fe were exogenous elements. However, the use of synchrotron radiation offers a higher sensitivity than ion beam analysis (IBA) for heavy elements. At the PUMA beamline trace elements down to a few ppm can be detected for elements between Ti and Y and to a few tens of ppm for Ba and Pb (Tack et al., 2019). This could make possible to detect additional trace elements in ivory or bone and to discuss their potential use as new chemical markers. The normalized concentrations of the elements Cr, Rb, Ba and Pb show strong variations among the archaeological samples and their XRF mappings present strong heterogeneities. They are clearly exogenous elements. The elements Ti, Mn, Fe and Y present lower variations in the normalized concentrations among the archaeological samples (ivory or bone). However, their spatial distribution is very heterogeneous. In the case of ivory, they are mainly located in the crack, which suggests that they are exogenous elements. Moreover, normalized concentrations of these elements differ strongly between the archaeological samples and

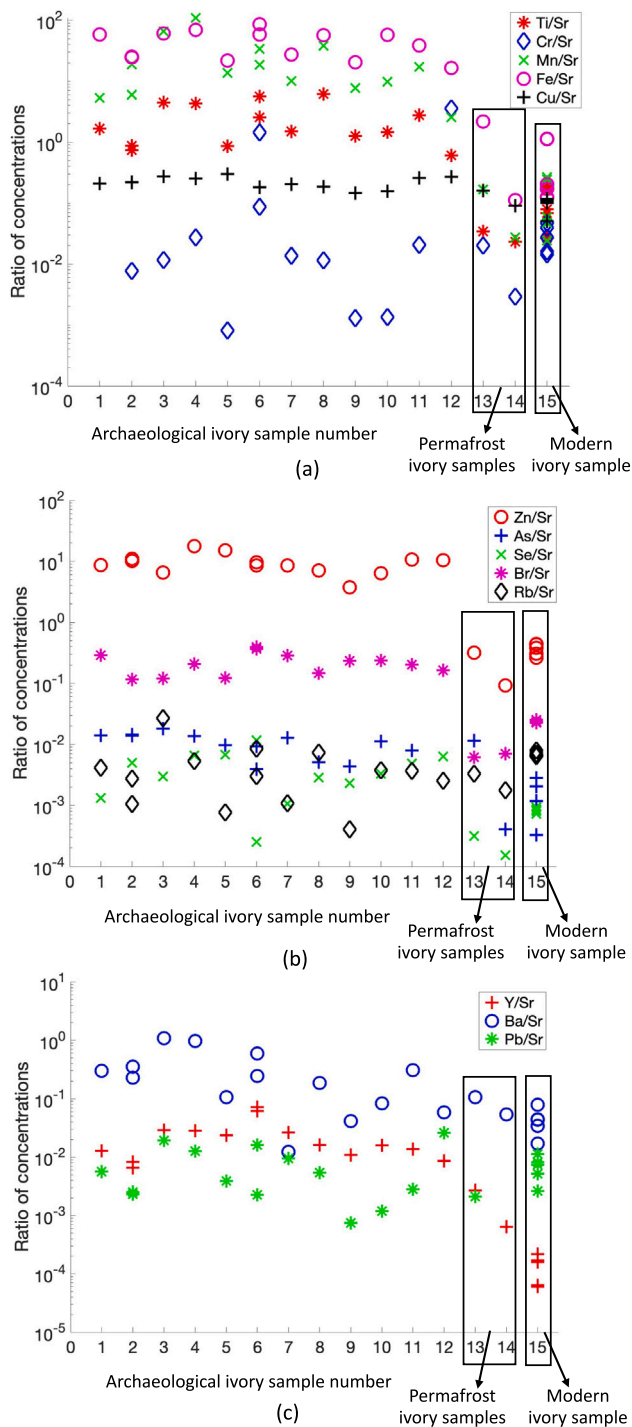


Fig. 7. Normalized concentrations of trace elements regarding to Sr for the 12 archaeological ivory samples, two permafrost ivory samples and a modern ivory sample. The plot (a) shows the results for Ti, Cr, Mn, Fe and Cu, the plot (b) for Zn, As, Se, Br and Rb and the plot (c) for Y, Ba and Pb. Results of trace elements are classified by increasing atomic number.

the reference samples. The concentrations are significantly higher in the archaeological samples because they have obviously been enriched in these elements due to exchange processes with the burial sediment. The case of Zn and Br is interesting because the concentrations of these two elements do not vary significantly among the corpus and their spatial distribution is homogeneous. These observations support their assignment to endogenous elements. The Zn and Br concentrations

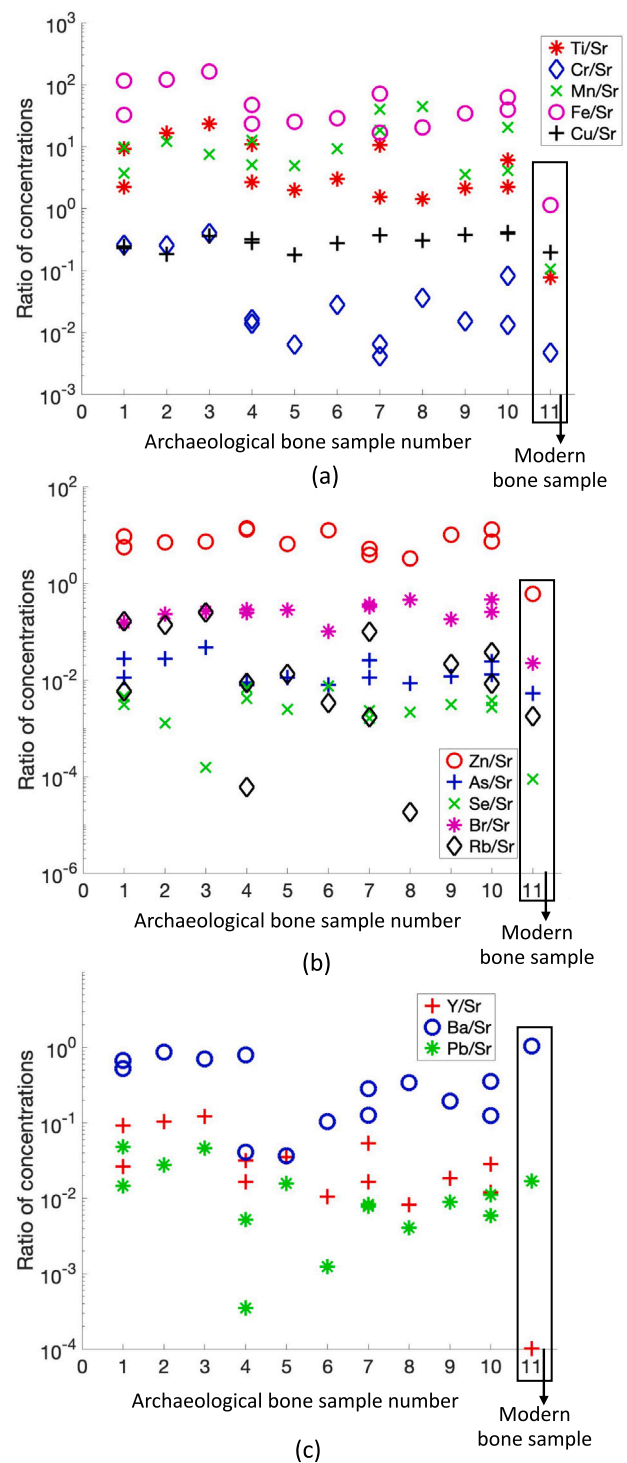


Fig. 8. Normalized concentrations of trace elements regarding to Sr for the 10 archaeological bone samples and a modern bone sample. The plot (a) shows the results for Ti, Cr, Mn, Fe and Cu, the plot (b) for Zn, As, Se, Br and Rb and the plot (c) for Y, Ba and Pb. Results of trace elements are classified by increasing atomic number.

are increased with respect to the here analysed reference samples. However, in previous studies we found relatively strong variations of Zn contents for different kinds of modern bone and ivory samples with various origins (Reiche et al., 1999). The Br content was not considered at that time because the sensitivity of the method was not yet sufficient to reliably detect it.



Finally, the results for the elements Cu, As and Se are difficult to interpret. The normalized concentrations of Se vary among the corpus of archaeological samples and do not differ from the reference samples. The XRF mappings of Se are not conclusive for the ivory and tend to show a homogeneous spatial distribution for the bone sample, but the Se signal is too low to draw conclusions (less than 20 counts in both XRF mappings). The normalized concentrations of Cu, and to a lesser extent As, are remarkably constant over the corpus and the reference samples. The XRF mappings show a homogeneous spatial distribution of Cu and a heterogeneous distribution of As for both samples. The concentrations of Cu and As are relatively low (a few tens of ppm for Cu and a few ppm for As). Therefore, PIXE mappings of Cu and As were impossible but could be realized by synchrotron XRF. The XRF results for Cu suggest that this element is of endogenous nature and may possibly be considered as origin marker in future studies.

## 7. Conclusion

The comparative and multi-method analysis of archaeological ivory and bone samples from the Hohle Fels Cave enabled the comprehensive characterization of the elemental compositions and element distributions of the objects examined. Several trace elements (Cr, Cu, As, Se, Rb, Y, Ba and Pb) present in ivory and bone could not be detected using PIXE due to their very low concentrations. In contrast, synchrotron-based XRF imaging enabled the detection of the trace elements mentioned because it has a higher sensitivity for these elements. Based on the XRF results, the endogenous/exogenous properties of these elements could be discussed, with Cu in particular emerging as a promising candidate for a marker of origin. We conclude that Ti, Cr, Mn, Fe, Rb, Y, Ba and Pb are clearly exogenous elements whereas Zn, Br, Sr and probably also Cu are endogenous elements.

The PIXE/PIGE analysis of ivory samples from Hohle Fels and from other European archaeological sites reveal that ivory from Hohle Fels can be differentiated from other prehistoric sites based on their relative contents and dispersion of biogenic trace elements (Sr, Br and Zn) and indicators for diagenetic changes (F contents). The results combined PIXE/PIGE and XRF study of archaeological ivory and bone from the Hohle Fels Cave did not provide any hints of specific differences in the diagenetic alterations between these two materials. A slightly increased F content in bone with respect to ivory was the only small variation we detected based on PIGE results. Further measurements would be necessary to investigate this in more detail.

## CRedit authorship contribution statement

**L. Tranchant:** Software, Validation, Formal analysis, Investigation, Data curation, Writing – original draft, Visualization. **K. Müller:** Conceptualization, Methodology, Validation, Formal analysis, Investigation, Data curation, Writing – original draft, Writing – review & editing, Visualization, Supervision. **Q. Lemasson:** Software, Validation, Formal analysis, Investigation, Data curation. **L. Pichon:** Software, Validation, Formal analysis, Investigation, Data curation. **S. Schöder:** Methodology, Software, Validation, Formal analysis, Investigation, Resources, Data curation, Writing – review & editing, Supervision, Funding acquisition. **N.J. Conard:** Resources, Writing – review & editing, Project administration, Funding acquisition, Heading the excavations at Hohle Fels. **I. Reiche:** Conceptualization, Methodology, Validation, Formal analysis, Investigation, Resources, Data curation, Writing – original draft, Writing – review & editing, Visualization, Supervision, Project administration, Funding acquisition.

## Declaration of competing interest

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

## Data availability

Research data from this study are available on request ([ina.reiche@chimieparistech.psl.eu](mailto:ina.reiche@chimieparistech.psl.eu)).

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