"ALEXANDRU IOAN CUZA" UNIVERSITY OF IAȘI
FACULTY OF BIOLOGY
DOCTORAL SCHOOL OF BIOLOGY

Microanalysis of ancient human remains discovered in archaeological sites from North-Eastern Romania

PHD THESIS SUMMARY

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Iași
2023

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Special thanks

The doctoral thesis represents the cumulative result of the scientific activity during the doctoral training, but also of the rigor and experiences acquired as a graduate of the Faculty of Biology, "Alexandru Ioan Cuza" University of Iasi.

First of all, my thanks are addressed to Mrs. University professor hab. PhD Bejenaru Luminița, the supervisor of this doctoral thesis, who supported me throughout the doctoral internship, constantly, with professionalism and a combined seriousness and gentleness, trust and respect, opening many paths for me to carry out the first studies regarding the microanalysis of ancient human remains from our country. Although the road to scientific maturity was not an easy and straight, Mrs. Professor was by my side, thus being able to overcome many key situations on the road to scientific affirmation.

Also, my thanks go to the members of the guidance committee, Ms. Lecturer PhD Anca-Narcisa Neagu, University professor hab. PhD. Lucian Hrițcu and Mr. Associate Professor PhD Ion Cojocaru, for the advice and suggestions offered during the doctoral training.

My sincere thanks are addressed to the Coordinator of the Center for Anthropological Research "Olga Necrasov" of the Romanian Academy Iași Branch, University professor PhD Cătălin Tănase, corresponding member of the Romanian Academy and to my colleagues of the research team, Mrs. Scientific Researcher PhD Mariana Popovici and Mrs. Scientific Researcher III PhD Vasilica-Monica Groza, for the advice and the understanding offered during all these years. Also, my thanks go to the late Academician University Professor PhD Constantin Toma, who at the time of my scientific initiation in the field of physical anthropology showed his encouragement and support for the chosen scientific path. Last but not least, I want to thank Miss Teaching assistant PhD Gabriela-Alina Ștefan, and to my family for the unconditional trust and permanent encouragement offered since the first moment when I chose the path of research.

Special thanks go to my husband, Mr. Scientific Researcher III PhD Mitică Ciorpac, for the constant understanding and encouragement offered during the doctoral training.

Abbreviations list

A	adolescents
AM	adults
AT	young adults
BF	bright field
Cr	crypt
Cv	vascular channels
CVA	canonical variates analysis
DA	discriminant analysis
DIC	differential interference contrast
lp - w	large pits - width
lp - l	large pits - length
sp - w	small pits - width
lp - l	small pits - length
FLUO	epifluorescence microscopy
HE	hematoxylin and eosin
î.e.n	before our era
Lc	cement line
LCI	internal circumferential lamellae
LH	lacune Howship
Lo	bone lamellae
m_{2r}	right mandibular M2 molar
M^{2r}	right Maxillary M2 molar
m_{2l}	left mandibular M2 molar
M^{2l}	light maxillary M2 molar
MANOVA	multivariate analysis of variance
mm	milimeter
N	number
N.lp	number large pits
N.sp	number small pits
N.fs	number fine scratches
N.cs	number coarse scratches
O	osteocyte lacunae
OHI	Oxford histological index
P	primary bone (woven bone)
PCA	principal component analysis

PED	percentage of dentine exposure
POL	polarized light microscopy
fs - w	fine scratches - width
fs - l	fine scratches - length
cs - w	coarse scratches - width
cs - l	coarse scratches - length
SOT	total occlusal area
TOA	total degraded area
TDEA	total dentine exposure area
V	old adults

I. THEORETICAL PART

Introduction

Paleoanthropology is a discipline that approaches the complex past of human existence (Bejenaru, 2014). As an integrated part of physical anthropology, paleoanthropology has a strong interdisciplinary character.

Nowadays, microbiology, archaeogenetics, paleohistology, archaeobotany, archaeoentomology are frequently used (Cerrito *et. al*, 2022; Gismondi *et. al*, 2018; Magni *et. al*, 2023; Tencariu *et. al*, 2022; Weyrich *et. al*, 2017), all being included in bioarchaeology research area.

A special research niche in paleoanthropology is represented by paleohistology, which opens a frontier for the study of human skeletal and dental remains, through the microscopic examination of material preserved in varied conditions and contexts (Thompson and Errickson, 2017).

Today, the histological studies can be divided according to the addressed subject into age-at-death determination studies (Karydi *et. al*, 2022), taphonomic studies (Brönnimann *et. al*, 2018) and the study of past diseases manifested at the skeletal level (Turner-Walker and Mays, 2007). The studies based on the analysis of tooth tissue surfaces (tooth wear) are the subject of the paleodiet evaluations (Bas *et. al*, 2023).

Through the studies included in the doctoral thesis, we aimed to initiate a new paleoanthropological research niche in Romania and to facilitate alignment with international scientific research in the field. The scientific approach of Thesis is based on the microanalysis (optical and electronic microscopy, histology, imaging and statistical analysis) of ancient human remains discovered in archaeological sites in North-Eastern Romania.

The research directions approached in the PhD Thesis refer to dental wear as a bioindicator of diet (1) for two human samples from the medieval period from Iași city and, respectively, from prehistoric sites in the North-East of Romania (Chalcolithic and Bronze Age), the study of the taphonomy degradation of non-skeletal human remains (2) discovered in the Iași city

(17^{th}-19^{th} centuries) and the study of past diseases (3) on three skeletons affected by treponematosis, discovered in Iași city (15^{th}-19^{th} centuries).

The doctoral thesis is composed of a theoretical part and an original part. The first part contains the Motivation, the aim and objectives of the thesis, the limits of the research carried out, the ethical considerations of scientific research, the *Introduction*, and the chapter entitled *Current state of knowledge in the field*. The original part of the doctoral thesis is structured in two chapters and a Conclusions section. The second chapter refers to the *skeletal and non-skeletal material used in the thesis and the applied research methodology*. The third chapter, entitled *Results and discussions*, consists of four sub-chapters that present the results of the own research together with related discussions and refer to: *(i) Dental wear as a diet indicator for the skeletal series discovered in the necropolis of the Royal Court in Iasi (17^{th} century)*, *(ii) Diet-related dental macrowear in archaeological human populations of Chalcolithic and Bronze Age from North-Eastern Romania*, *(iii)* the *Histotaphonomy of human hair fibers discovered in archaeological context (18^{th}-19^{th} centuries) from Iași city*, and the *(iv) Histological characterization of inflammatory reactions at the postcranial skeleton level in three possible cases of treponematosis identified Iasi city (15th-19th centuries)*. The final part of the thesis is represented by the *Conclusions* section followed by the *scientific activity* during the doctoral training.

Motivation, purpose and objectives of PhD Thesis

Motivation. The analysis of biological microstructures of ancient human remains allows modern paleoanthropology to investigate a wide range of subjects, such as paleodiet, histotaphonomy, paleopathology representing at the same time a new research niche in Romania.

The aim and objectives of the study. The aim of the thesis is to contribute with aspects of microscopic analysis of human remains to the knowledge development of the past human populations of North-Eastern Romania, from Prehistory (Chalcolithic and Bronze Age) to the medieval (17th century) and pre-modern (19th century) periods.

The thesis objectives are:

***O_1*. *Paleodiet characterization through dental wear assessment using optical and electronic microscopy and image analysis.*

***O_2*. *Taphonomic assessment of ancient human remains by histological and micrometric methods.*

***O_3*. *Identification and characterization of skeletal pathologies using optical microscopy.*

***O_4*. *Dissemination of the obtained results.*

The limits of the study

The limits of the research carried out in the doctoral thesis are mainly due to the nature of the material studied – ancient human remains (hundreds and thousands of years old), but also the reduced number of scientific articles at the international level, particularly in the field of paleopathology, which limited the comparisons and the discussions.

Ethical considerations in research

Considering the particularities of the research topics, the ethical principles included in the codes of the *American Physical Anthropological Association* and the *American Anthropological Association*, as well as the bioethical ones included in the Belmont report "*Ethical principles and guidelines for the protection of human subjects*" were considered in this thesis.

CH. 1. Current state of knowledge

1.1 Dental wear – a paleodiet marker for past human populations

Dental wear is a normal, non-pathological physiological process characterized by the gradual loss of tooth enamel, usually at the occlusal surface (Smith, 1984). According to Levrini *et. al* (2014), dental wear can be classified into two categories mechanical wear (attrition, abrasion, abfraction) and chemical wear (erosion).

The tooth wear is strongly correlated with the physical properties of the food consumed, therefore the relationship between tooth wear and paleodiet has been addressed and debated in recent years (Peigné and Merceron, 2017). Dental macrowear is the result of a cumulative and a multifactored process which takes place over an individual's life (Fiorenza *et. al*, 2018).

Visible to the naked eye, worn facets of teeth and the presence of dentine are used to provide information regarding diet and masticatory activities, being also correlated with lifestyle, habits and food processing techniques (Dawson and Brown, 2013).

Known as the "Last Supper effect", dental microwear refers to the refers to the microscopic features left on the occlusal and buccal surface of the teeth and reflects the mechanical and abrasive properties of foods consumed during the last weeks of the individual's life (Scott and Halcrow, 2017). There are two main types of dental microwear features: the linear type – characterized by scratches, and the non-linear type – characterized by pits (Schmidt, 2010; Smith *et. al*, 2019).

Usually, plant foods and meat, can often cause parallel scratches on the dental surface (Ungar, 2019). Dental microwear profiles dominated by a few fine scratches are associated with relatively soft food. An abrasive diet leaves greater and more pronounced marks such as wide scratches. Harder food items produce more pits due to both the hard nature of items and the effect of chewing forces during mastication (Petraru *et. al*, 2020a).

1.2 Histotaphonomy of ancient human remains

Histotaphonomy aims to describe, quantify, and interpret the taphonomic processes on microstructural level. Regarding the archaeological human remains, they are represented by bones and dental material but also by non-skeletal elements (Brothwell, 1981).

1.2.1 Histotaphonomy of non-skeletal ancient human remains

Human hairs discovered in archaeological contexts can provide valuable information to both paleoanthropology and forensic science (Robertson 1999). Regarding favoring conditions for preservation, the restriction/inhibition of microbial activity is necessary for morphological and structural hair preservation (Wilson *et. al*, 2007). Hair taphonomy studies focus on two directions: morpho-structural changes of the hair shaft and the structural changes of the hair root (Domzalski, 2004).

Other non-skeletal human remains have been reported in the scientific literature: brain, liver, intestines, muscle tissue, keratinocytes, erythrocytes, and nucleated cells of the hematopoietic tissue (Altinoz *et. al*, 2014; Hess *et. al*, 1998; Setzer *et. al*, 2013).

1.2.2 Histotaphonomy of skeletal ancient human remains

Postmortem changes appear as the interaction results between the mineralized tissue, and the geochemical and biological factors (Bertoglio *et. al*, 2021). Currently, there are debates regarding the origin of microorganisms that postmortem degrade the bone tissue: (1) intestinal endogenous bacteria, (2) soil microorganisms together with exogenous factors, or the synergistic action of the two mentioned categories (3) (Papakonstantinou *et. al*, 2020). The microstructural changes of bone tissue are divided into two categories: the Wedl tunnels (fungal tunnels) and non-Wedl tunnels (bacterial tunnels). At the tissue level, taphonomic inclusions or infiltrations can be distinguished (Jans, 2021).

1.3 Histological study of skeletal pathologies

The paleohistology studies of archaeological bone evolved with the section obtaining techniques upgrade and the use of various optical or electron microscopy techniques (Turner-Walker and Mays, 2007).

The histological studies of pathological bone tissue can contribute to strengthens in the differential diagnosis or can expose a pathognomonic morphology in several diseases or disorders: Paget's disease (Kesterke and Judd, 2019), hyperparathyroidism (Mays *et. al*, 2007), osteomalacia (Welsh *et. al*, 2020), and osteoporosis (Miszkiewicz *et. al*, 2021).

A topic of interest in the study of ancient human disease is the identification of markers for skeletal infectious disease by means of histology (Schultz, 2001). Skeleton infections are characterized by osteolytic activity and an inflammatory activity of the periosteum, which leads to the formation of new bone tissue - callus (Assis and de Boer, 2022).

The formation of new bone tissue in diseases such as tuberculosis, leprosy and treponematosis has been analyzed histologically in order to identify pathognomonic characters and/or as a complementary method in a differential diagnosis (Assis *et. al*, 2015; Assis and Keenleyside, 2016).

According to the international literature, the specificity of histological features suggestive for treponematosis at the skeletal level has been debated by numerous researchers, some claiming that certain histomorphological aspects are pathognomonic for treponematosis (Schultz, 2001), some who nuanced certain features, and others who refuted their specificity (Assis *et. al*, 2015; Van der Merwe *et. al*, 2010).

At the national level, there is a high deficiency regarding studies that approach the histology of skeletal hyperostosis in treponematoses cases, the present thesis being the first one in our country. The international literature is characterized by a reduced frequency of paleohistology works regarding this subject (Assis *et. al*, 2015; Schultz, 2001; Van der Merwe *et. al*, 2010; Von Hunnius, 2004; Von Hunnius *et. al*, 2006).

II. ORIGINAL PART

CH 2. MATERIAL AND METHODS

2.1 Applied methods for the study of dental wear

2.1.1 Dental macrowear

Archaeological sites. The dental material subjected to macrowear study has been discovered in prehistoric archaeological sites from North-Eastern Romania (Brad, Aldești, Valea Lupului, Holboca, Bârgăuani, Cioinagi, Cândești, Roman, Trușești, Ciritei, Brăești, Doina), dating from Prehistory and from 17th century (Iași).

Material. The dental material comes from multiple archeological sites and was considered as a single sample for Prehistory, including Chalcolithic (5200-3800/3700 BCE) and Bronze Age (3500-1200/1150 BCE).

For the evaluation of dental macrowear in the prehistoric human populations, the second mandibular and maxillary molar, M2, was selected (Table 2.1) according to the criteria stated by Petraru *et. al* (2022).

Table 2.1. The M2 molar teeth considered in the study.

Period	Royal Court from Iași (17th century)					Prehistory (5000-1200/1150 BCE)				
M2 molar type	M^{21}	M^{2r}	m_{21}	m_{2r}	N	M^{21}	M^{2r}	m_{21}	m_{2r}	N
Females	5	6	6	10		7	8	7	10	
Males	21	26	31	37	142	17	14	14	18	100
Undeterminable sex	-	-	-	-		1	2	1	1	

2.1.1.1 Macrowear scoring method.

Macrowear scoring is based on the loss of tooth enamel and the presence of dentin on the occlusal surface of the molars (Scott, 1979). Depending on the degree of dentine exposure, three categories of wear were considered according to methodology proposed by (Tencariu *et. al*, 2022; Tomczyk *et. al*, 2020).

2.1.1.2 Dental macrowear analysis.

Dental macrowear analysis involves recording photographs of the occlusal surfaces of the M2 molars. The quantification of PDE is based on the quantification of total occlusal area (TOA, mm^2), total area of dentine exposure (TADE, mm^2) and the percentage of dentine exposure (PED %) by image analysis (Galbany *et. al*, 2016).

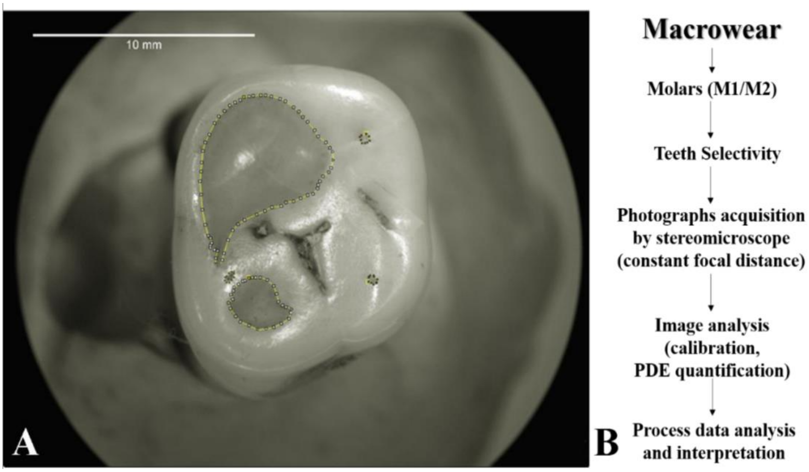

Figure 2.1. A – The M2 molar occlusal surface under stereomicroscope showing areas of exposed dentine. B. Main steps in quantitative dental macrowear analysis (Petraru and Bejenaru, 2019).

In order to assess the variable data sets normality, the Shapiro-Wilk test was applied. The Kruskal-Wallis, Tukey and Mann-Whitney tests were used to compare the PDE values between molar groups. Correlation and linear regression analysis were used to assess the relationship between PDE and the age of individual. Multiple regression analyses were carried out to determine whether there are other variables (age at death, occlusal surface, period) which could have control or influence over the wear. A resampling method was used on the dataset (*bootstrap*) and within the *t*-test or Mann-Whitney or was performed in R (Kohl and Kohl, 2020). Descriptive statistics, regressions and correlations were made using PAST 2.17 and XLSTAT 2019 4.2 version.

2.1.2 Dental microwear analysis

Material. The study material is represented by maxillary and mandibular M2 molars (n=56) belonging to 14 skeletons discovered in the 17th century necropolis at the Royal Court in Iasi.

Quantification of dental microwear. The work methodology refers to: the *preparation of dental material* and the imaging process. This initially involves the application of methodology for the visualizing dental samples with a Tescan Vega II SBH scanning electron microscope, fallowed by the acquisition of 3 non-overlapping photographs at the facet 9 situated at the disto-buccal cusp (for m_2), respectively of the mesio-lingual cusp (for M^2). The photographs were further analyzed using the MicroWeaR (R) software (Strani *et. al*, 2018). It returns data for each microwear type (fine and coarse scratches, small and large pits, Figure 2.2), as well as the mean and standard deviation of the lengths and widths of the identified types of wear.

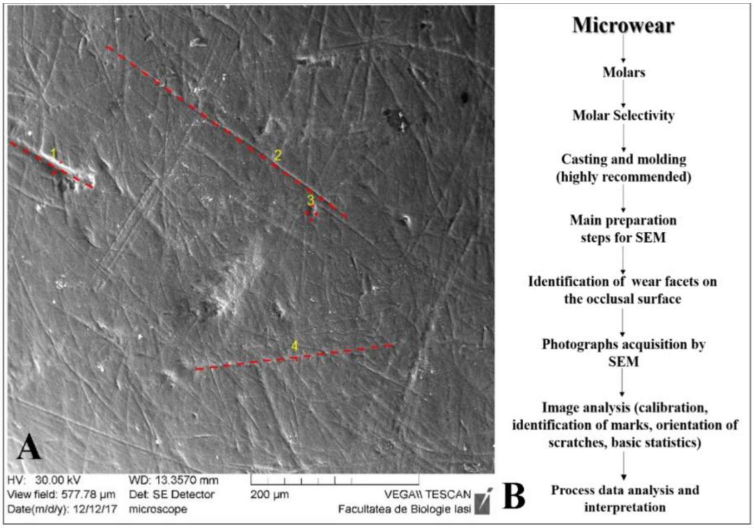

Figure 2.2. A – SEM micrograph showing different types of marks on enamel: 1 – coarse scratch; 2, 4 – fine scratches; 3 – large pit; B. Main steps in quantitative dental microwear analysis (Petraru and Bejenaru, 2019).

Statistical analysis. The following variables were used: sp - l, sp - w, lp - l, lp - w, fs - l, fs - w, cs - l, cs -w. The T and Mann-Whitney U tests were used to determine if there were any differences between maxillary and mandibular molar microwear patterns. The principal component analysis (PCA) was used as an exploratory tool for correlation of the variables underlying microwear patterns and relationships between molars. The variation patterns for both sexes were analyzed with multivariate analysis of variance (MANOVA) and canonical variate analysis (CVA). The statistical tests were performed using XLSTAT, PAST (Hammer *et. al*, 2001) and R (Team, 2013).

2.2 Taphonomic and paleopathological methods

2.2.1 Cryotome sectioning histological method for the non-skeletal human remains (hair fibers)

The study material is represented by human hair fibers which belonged to males with the age at death around 50-60 years old (18^{th}-19th centuries) discovered during the archaeological excavations at "Adormirea Maicii Domnului" Roman-Catholic Cathedral from Iasi. The methodology includes different work stages: material sampling, impregnation in cryoprotective gel, cryo-sectioning, HE staining and mounting (Petraru *et. al*, 2020b).

2.2.2 Polish sectioning histological method for skeletal human remains (bone tissue)

Biological material is represented by bone tissue sampled from the long-bones belonging to three skeletons (M40, R30 and R26) and showing areas of new bone tissue formation (callus) along with a pathognomonic character for treponematosis called "*caries sicca*". The skeletons were discovered during the archaeological excavations at the "Adormirea Maicii Domnului" Roman Catholic Cathedral in Iași and dates from the 15^{th}-19^{th}

centuries. The methodology involves several work stages: tissue sampling using a Dremel multi-tool, embedding sampling in EpoThin2 epoxy resin, polishing and grinding of the embedded samples and mounting.

2.2.3 Microscopy methods and techniques

The microscopic methods and techniques used are corelated with the aim and objectives of the study, in accordance with the world-wide literature, different types of microscopic techniques (optical and electronic) were used to study the biological material, which enhanced the degree of scientific information obtained (Bozzola and Russell, 1999; Wilson *et. al*, 2010).

2.2.4. Quantitative, semi-quantitative and qualitative histotaphonomic analysis

The quantitative analysis refers to the serial transversal sections imaging that involves quantifying the degree of internal degradation of the human hairs discovered at the "Adormirea Maicii Domnului" Roman-Catholic Cathedral from Iasi (Petraru et. al, 2020b).

The semi-quantitative analysis refers to the assignment of bone tissue histotaphonomy scores (from 0 to 5) which is based on the Oxford Histological Index (OHI) system (Brönnimann *et. al*, 2018). Microbial degradations, Wedl and non-Wedl tunnels were annotated based on a present/absent scale (Papakonstantinou *et. al*, 2020).

For the non-skeletal human remains – the hair fibers the external taphonomic degradation scoring system is proposed by (Wilson *et. al*, 2010).

CH. 3. RESULTS AND DISCUSSION

3.1 Dental wear as a diet indicator for the skeletal series discovered in the necropolis of the Royal Court from Iași (17th century)

The M2 molars of the skeletal series discovered in the 17th century archaeological site at the Royal Court from Iași were subjected to the dental wear analysis.

The degree of dentin exposure increases with age and is significant in the older age categories ($p < 0.001$), affecting both male and female individuals. When age ranges were pooled, the dental wear differed significantly by sex ($Fs = 46.17$; $p < 0.001$). The dental macrowear results of the M2 molar from the skeletal series discovered at the Royal Court from Iași, differ significantly according to sex, which may suggests that the wear could be associated with differences in diet (Petraru et. al, 2018).

We can assume that a consumption of less abrasive and erosive foods by females could also be associated with differences in food preparation techniques (Petraru *et. al*, 2018).

To confirm this hypothesis, micromorphological microwear features from M2 molars were analyzed by electron microscopy and image analisys. Therefore, 56 M2 molars belonging to 14 individuals were subjected to dental microwear analysis, according to sex, laterality and position in the skull (Petraru *et. al*, 2020a).

All four types of wear were identified at the facet 9 surface of the M2 molars. In the female series (except for the G4M14 skeleton), the distribution of fine scratches were predominant more on the M^{21} and m_{21}. The numeric distribution of microwear features for each individual is shown in Figure 3.1 (Petraru *et. al*, 2020a).

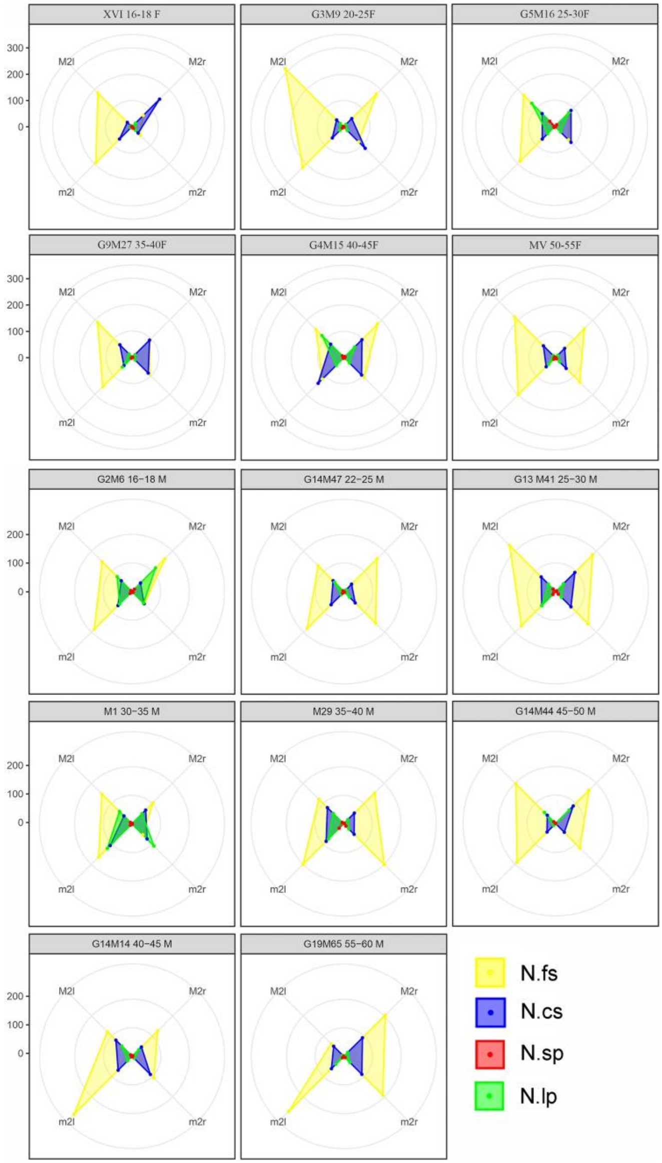

Figure 3.1. Distribution of dental microwear features in each individual at the facet 9 area (N.fs, number of fine scratches; N.cs, number of coarse scratches; N.lp, number of large pits; N.sp, number of small pits, M2r – right maxillary M2 molar, M2l – left maxillary M2 molar, m2r – right mandibular M2 molar, m2l – left mandibular M2 molar (Petraru *et. al*, 2020a).

The microwear data obtained by micrograph imaging have been subjected to DA, using four quantitative variables (N.fs, N.cs, N.lp, N.sp) and two qualitative variables (sex and molar position in the skull).

A major contribution is made by the N.sf variable, which differentiates two microwear patterns, one corresponding to left mandibular molar m_{2l} (where the presence of fine scratches is greater), and one corresponding to the right mandibular molar m_{2r} (where the presence of fine striae is less). These patterns are present, especially in the female series. The total microwear profile was dominated by fine scratches (57.88%), followed by coarse scratches (25.60%). Lower values were registered for nonlinear marks, 1.93% for small pits and 14.57% for large pits.

An attempt to characterize diet based on the number of recorded microwear features is difficult to achieve. Even if the abundance of fine scratches is higher, the number of wider scratches and large pits is also remarkable due to their similar values. According to Sołtysiak (2011), a high proportion of lines and pits are characteristics of an abrasive diet. Moreover, the microwear analysis showed the presence of wider scratches, which may be another indicator of an abrasive diet (Petraru *et. al*, 2020a).

To distinguish a more abrasive diet from a less abrasive one, the “width” of the variables must be taken into consideration (Schmidt, 2010). For a more specific analysis, the data obtained by imaging included for each type of microwear feature another two measurements: length and width, eight variables being included in the flowing analysis: sp - l, sp - w, lp - l, lp - w, fs - l, fs - w, cs - l, cs -w.

Dental microwear analysis for the male series

The results of statistical significance tests (Mann-Whitney U and t-test) suggested that there are no differences between the variables that define the dental microwear profile for upper and lower M2 molars. The paired t-test and the Wilcoxon test showed that there are no differences between the microwear of the right-sided molars compared to the microwear of the left-sided molars, except for the variable sf - l. Thus, the fs - w variable for the right-sided molars is greater than the fs - w on the left-sided molars (t test =

- 3.495; p = 0.01). The metric data related to the eight variables were subjected to principal component analysis (PCA). The variables included in the dental microwear pattern assessment did not suggest particularities (Petraru *et. al*, 2020a).

Dental microwear analysis for the female series

In contrast to the previously obtained results, the paired t-test suggests differences between the dental microwear profile related to the molars on the left side and those on the right side, based on the variables: sp - w ($p = 0.04$), lp - w ($p = 0.02$), fs - w ($p = 0.00$), cs - w ($p = 0.05$).

In the PCA analysis, the lp - l and lp - w variables have a defining role in separating the two tooth microwear profiles, one for the M^{2l} and m_{2l} molars and one for the M^{2r} and m_{2r} molars (Figure 3.2).

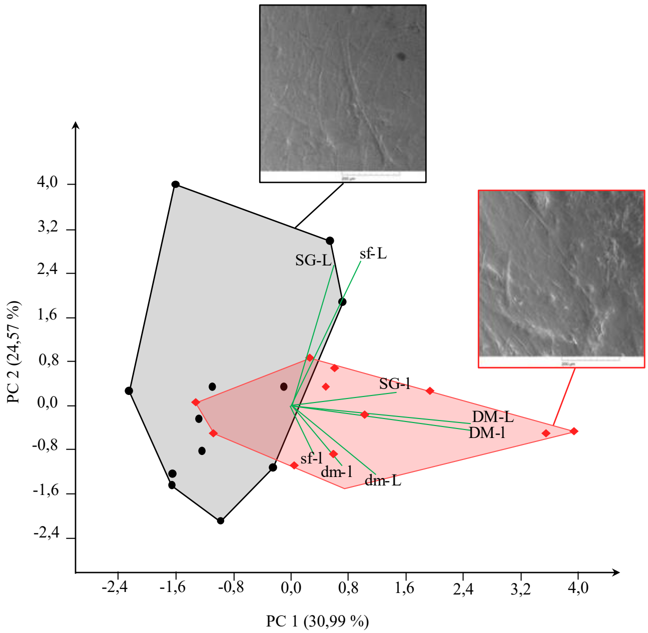

Figure 3.2. Scatterplot of the first two principal PCs of female dental microwear from the 17th century necropolis at the Royal Court in Iași, modified after (Petraru *et. al*, 2020a)

Comparative analysis of the dental microwear profiles

Given the existence of variation in the female sample, our question is whether there are affinities between female and male patterns. Therefore, the eight variables were assessed through MANOVA and CVA.

The results of the multivariate analysis showed a significant difference between microwear patterns (Wilk's Lambda test: Value = 0.536, F =2.09, p < 0.01). A greater affinity in the microwear profiles is highlighted between the male pattern and the right side of female molars (Figure 3.3).

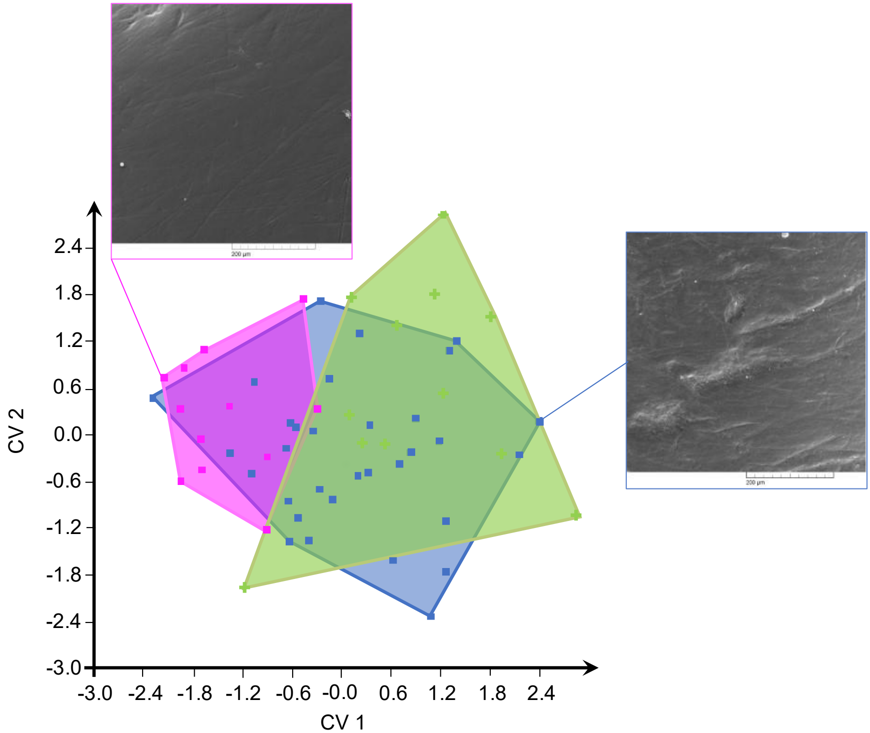

Figure 3.3 Plot of CV1 and CV2 (pink – microwear profile of the left sided M2 molars in females; green - microwear profile of the right sided M2 molars in females; blue – microwear profile of the M2 molars in males) (micrograph scale bar=200µm) (Petraru *et. al*, 2020a)

The fs – w variable has the highest values in male series, being closer to the values from the right side molars of females (Petraru *et. al*, 2020a).

According to a previous study based on macrowear analysis of the M2 molar, it has been hypothesized that the male series would be characterized by a different microwear profile than the female series, and the results of our study only partially confirm the initial assumption (Petraru *et. al*, 2020a). The dental microwear analysis emphasized a hard and abrasive diet in both males and females, although different microwear profiles were distinguished. Dietary abrasiveness may be influenced by the number of abrasive particles ingested with food, especially in agriculture products (Romero *et. al*, 2013). Gügel *et. al* (2001) discovered that different types of cereals can cause varied pit sizes causing a difference in microwear pitting. Also, grit adhering to food can cause micro-scratches (0.1–1 µm in width) comparable to those produced by phytoliths (Ungar, 1995). Also, unwell processed cereals can be linked to a more abrasive microwear profile (Romero *et. al*, 2013).

Archeological and documentary evidences indicate that the Medieval population in Iasi City used rye, barley, buckwheat, and millet in their diet (Bilavschi, 2013). Cereals were substituted with dried bulrush which was ground to obtain an alternative kind of flour. Also, the technique of baking bread directly on the clay oven floor increased the content of abrasive particles in the food (Székely, 2018).

Asymmetry is commonly found in human populations (Thiesen *et. al*, 2016), especially in the craniofacial structures (Primozic *et. al*, 2012). The microwear pattern associated with the right-sided molars in the female dataset suggests a preferred chewing side used to triturate apart harder foods. Apparently, therefore, lateralized chewing behavior can affect the dental microwear pattern (Petraru *et. al*, 2020a).

3.2 Diet-related dental macrowear in archaeological human populations of Chalcolithic and Bronze Age from North-Eastern Romania

The macrowear scoring results show that over 50% of the samples obtained scores for occlusal macrowear between 16 and 24 in both mandibular (M^2) and maxillary molars (m_2) (Figure 3.4). The molars were categorized into three categories: molars with wear facets that are very small (2.94% in m_2), molars with wear facets that are moderately advanced (55.88% in m_2 and 81.25% in M^2), and molars with wear facets that are highly advanced (41.17% in m_2 and 18.75% in M^2)(Figure 3.4, b, d) (Petraru *et. al*, 2022).

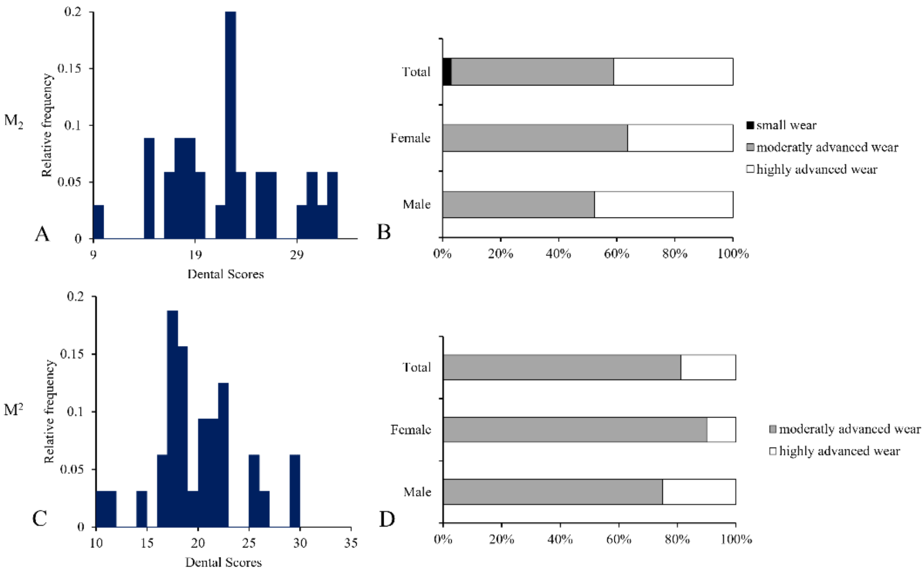

Figure 3.4. Macrowear scoring in the Chalcolithic and Bronze Age samples, from North-Eastern Romania: relative frequency of the dental scores in mandibular (a) and maxillary molars (b); stages of the dental wear in mandibular (c) and maxillary molars (d). (Petraru *et. al*, 2022)0

The percentage the dentine exposure (PDE) values by sex are characterized by a higher mean in males than females for the mandibular m_2 molars (17.01 and 9.62, respectively), and for maxillary M^2 molars (9.33 and 5.89, respectively). The PDE relative frequency of the total sample is shown in Figure 3.5 a. The obtained result showed an insignificant difference between PDE values in female and males in both maxillary molars (t: 0.27, p:0.76, bootstrapped p-value: 0.75) and mandibular (t: 1.10, p: 0.28, bootstrapped p – value: 0.29). When the sex criterion was removed, the PDE by age-ranges (i.e., YA vs. MA) showed statistical differences (m_2, Mann-Whitney, Z: 3.64, p: 0.0002; M^2, t test, t: 2.10, p: 0.04). The obtained results confirmed that wear rates increase with the age-ranges (Figure 3.5b and c).

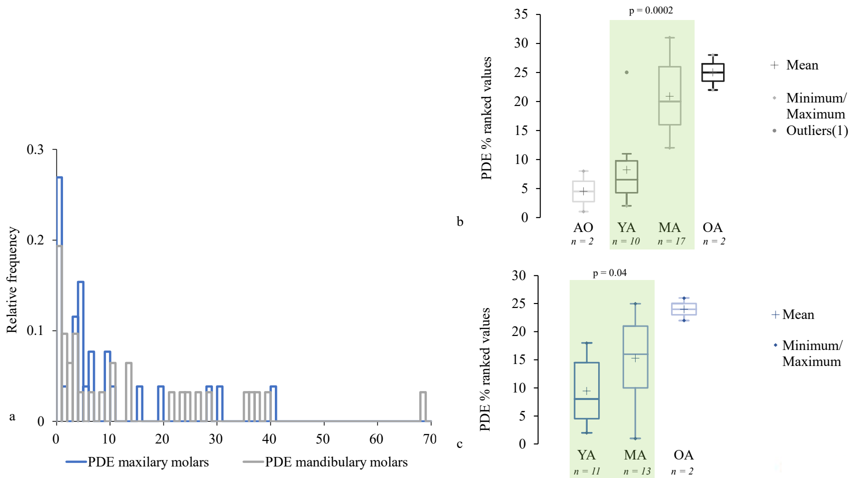

Figure 3.5. Macrowear analysis in the Chalcolithic and Bronze Age overall samples, from North-Eastern Romania: (a) relative frequency of PDE % in the studied sample; (b) distribution of dental wear according to age category for mandibular molars; (c) distribution of dental wear according to age category for maxillary molars. AO – adolescents, YA – young adults, MA – middle adults, OA – old adults (Petraru *et. al*, 2022).

The dental wear was compared in the same age group (i.e., middle adults age group - MA and young adults' group - YA), between males and females no statistical difference was observed in both, mandibular and maxillary molars (M2, YA: t: 0.77, p: 0.46; MA t: 0.009, p: 0.99; M2, MA: t: 1.38, p: 0.19).

Dental wear – age dependent process

When an interval of age was available, the middle value of this was used to perform the regression analysis between the PDE values and the age at death. In the mandibular molar dataset, the two variables were correlated in 31% of cases (R^2: 0.31; $p < 0.0001$) (Figure 3.6a), while for the maxillary molars the PDE and age variables were correlated in 49% of cases (R^2: 0.49; $p < 0.0001$) (Figure 3.6b).

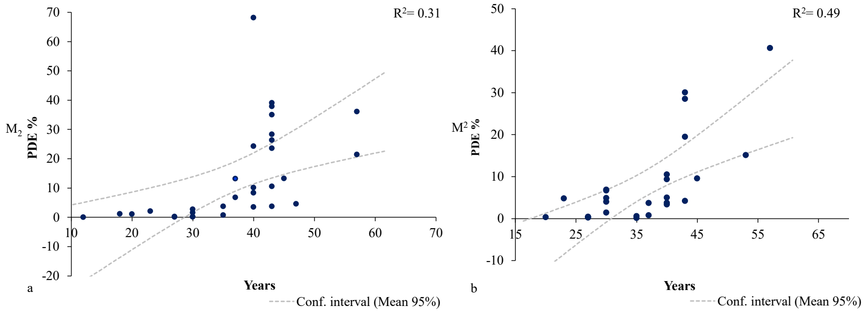

Figure 3.6. Relation between PDE and age at death for the mandibular (a) and maxillary M2 molars (b) in the Chalcolithic and Bronze Age sample from North-Eastern Romania ($p < 0.001$) (Petraru *et. al*, 2022).

Given the obtained results, it can be considered that the age was not the only factor that influenced the dental wear, the diet and food-processing methods are also important factors to be considered when discussing about the loss of tooth tissues in archaeological populations (Petraru *et. al*, 2022).

Occlusal area and prehistoric period influence on dental wear

In order to perform further correlation analysis, we tested if there are differences in the occlusal area and dentine exposure between the Bronze Age molars and those from Chalcolithic period.

Although, the dental material under analysis belongs to populations close in geographical space and time, there is a significative difference between the occlusal area of the molars from Chalcolithic and those from Bronze Age (m^2, Mann-Whitney, Z: 3.04, p: 0.002; M^2, Mann-Whitney, Z: 1.97, p: 0.04). Regarding the dentine exposure area no differences were observed (m_2, t test, t: 0.80, p: 0.30; M^2, t test, t: 0.14, p: 0.88) (Petraru *et. al*, 2022).

For a more specific analysis, a multiple linear regression was performed. Among the variables considered, age at death and occlusal surface have the greatest influence on dental wear ($p = 0.094$), but only age was statistically supported ($p < 0.0001$).

Even if the Prehistoric period did not manifest a significant contribution in the variability of dentine exposure in the analysed samples, a new analysis was performed to provide a regression model between the two Bronze Age cultures: Monteoru and Noua. Similar results were obtained, age showing the greatest influence. The role of the occlusal area should not be overlooked especially in the maxillary molars considering the p value was very close to the 0.05 significance threshold ($p=0.054$).

The dental wear is strongly correlated with the physical properties and abrasiveness of the consumed food, with processing techniques and storage of aliments. One of the events with major implications for food was the shift from hunting, gathering and fishing to animal husbandry and plant cultivation. Considering the abrasive character of foods used in Prehistory, the rate of the dental wear is higher in the past human populations compared to recently human populations (Mays, 2002).

The dental wear study regarding prehistoric human populations, requires the integration of the wild and domestic regional food resources in the bioarcheological regional-specific context.

The tooth macrowear among the Chalcolithic and Bronze Age populations from North-Eastern Romania has not been previously documented. The difference between the dentine exposure values in males and female has not statistical significance in the same age at death group (Petraru *et. al*, 2022). The multiple regression analysis shows no differences between Chalcolithic and Bronze Age archaeological populations, as well as between the Monteoru and Noua Cultures.

3.3 Histotahopnomy of human hair fibers discovered in archaeological context (18th-19th centuries) from Iași city

The obtained results refer to the heterogeneous taphonomic degradation of huma hair fibers (18th-19th centuries), discovered in comparable condition of conservation. The non-skeletal human remains belonged to four male skeletons discovered during the archaeological excavation for the rehabilitation of the "Adormirea Maicii Domnului" Roman Catholic Cathedral of Iași (Petraru *et. al*, 2020b).

The first microscopic evaluation of the hair fibers through optical microscopy revealed qualitative features identified previously in the literature, but required for the study (Robertson, 1999). Initially the microscopic observation was based on the histological slide preparation contained on cryosectioning and on the intrinsic autofluorescence properties of keratins (Robertson *et. al*, 2013).

The ellipticity index (%), based on the micrometric measurements of hair serial cross-sections showed an intermediate mean elliptic shape between 63.25% and 66.22% (Petraru *et. al*, 2020b). According to the percentage range proposed by Franbourg *et. al* (2003) the ellipticity index shows that the hair fibers belong to the European type.

The microscopic screening of the hair samples showed the presence of fungal deposits and fungal bloom (Figure 3.7a, b). Although the hairs seem to be macroscopically non-degraded, the presence of circumferential fissures were noticed (Figure 3.7c). The action of keratinolytic micro-organisms could destroy the cuticle and induce the collapse of hair fibers by separation and alteration of the cortical macrofibrils, followed by the break of the hair shaft (Figure 3.7d). In epifluorescence microscopy, due to the autofluorescence proprieties of hairs, external hair lesions such as holes specific to the degradation activity of fungi were observed (Figure 3.7e). In transverse sections, the hair fibers show zones of degraded cortex with a cuticle partially destroyed (Figure 3.7f) (Petraru *et. al*, 2020b).

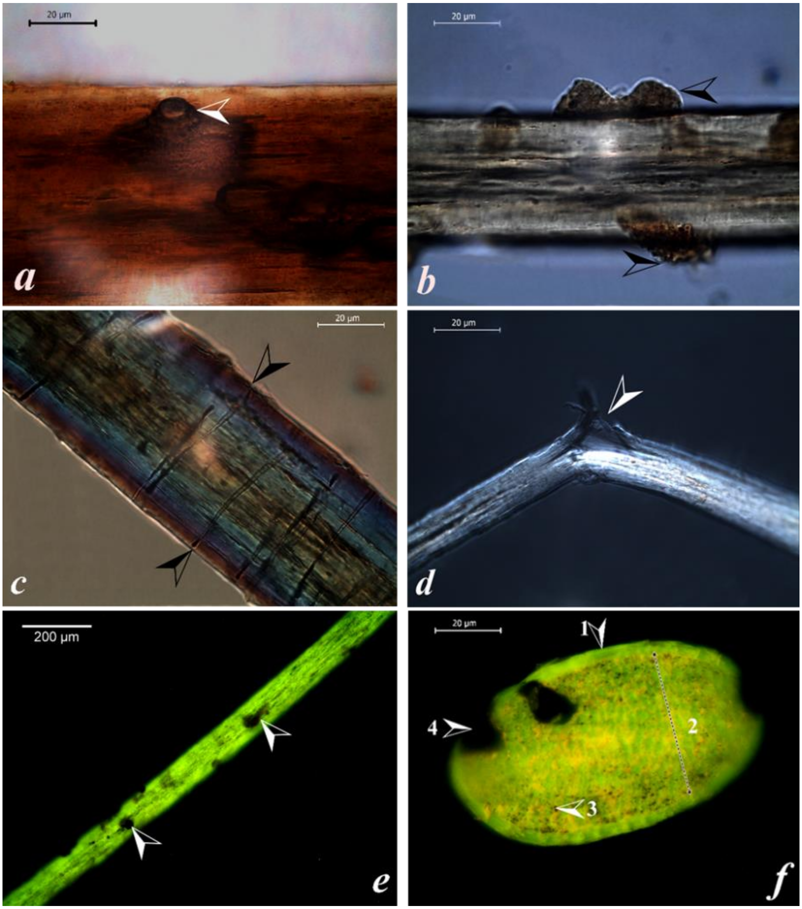

Figure 3.7. Light microscopy images of hair fibers discovered at the Roman Catholic Cathedral of Iași: surface fungal "bloom" (a – DIC); adherent fungal deposits (b – BF); circumferential fissures (c – DIC); hair shaft break (d – DIC); external ovoidal lesions (e – FLUO); and cortical destruction with a partially persisting cuticle - cross-section (f – FLUO): 1 – cuticle; 2 – cortex; and 3 – granule with melanic pigment, 4 – cuticular and cortical destruction (Petraru *et. al*, 2020b).

The scanning electron microscopy screening of human hairs dating from 18th-19th centuries hairs, showed a variable degradation of the scalp hair by micro-organism activity (Figure 3.8a-e). The fibers examined by SEM provided with an intact (Figure 3.8a), partially or completely destroyed cuticle showed alterations initiated by fungi (Figure 3.8b, c), emphasized by the presence of the tunnels within the hair shaft (Figure 3.8d, e). On the surface of the hair shafts, a great number of fungal hyphae and spores were observed (Figure 3.8f, g).

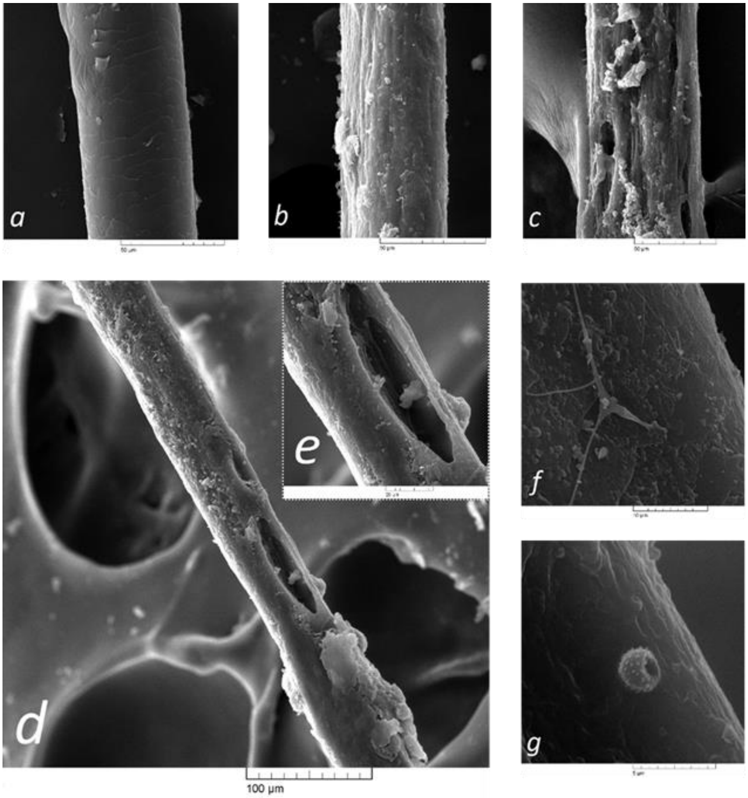

Figure 3.8. Scanning electron microscope (SEM) images of hair fibers showing different degrees of degradation assessed by histological scores: (a) surface morphology of overlapping cuticle cells with minor modifications (score 1); (b) intermediate change with hair cuticle partially destroyed (score 2); (c) severe change with hair cuticle completely destroyed, emphasizing the fibrillation process (score 3); (d) extended surface lesions and fungal tunnelling; (e) detail of fungal tunnelling; (f) fungal hyphae extended on the hair surface; and (g) spore found in a fungal tunnel (Petraru *et. al*, 2020b).

The heterogeneity of the external hair degradation pattern has been achieved by the attribution of histological scores. The results show that the C9b hair sample is the most degraded: 6.66% of the observed hairs was assigned with a score of 1; and 60% with a score of 2; 3.33% with a score of 3 (severe morphological change with total destruction of the cuticle). A score of 0 was assigned to C3 (16.67%) and C9a hairs (13.33%). Further, Tukey's HDS test reveals significant statistical differences between pairs C9b–C3, C9b–C9a ($p< 0.0001$) and M31–C9a, M31–C9b ($p< 0.01$) (Petraru *et. al*, 2020b).

The internal histotaphonomical degradation indicates differences between the hair samples. At the cross-sections of sample C9b, where the cuticle is degraded, the alteration of the internal structure can be observed through the presence of anastomosed microtunnels (Figure 3.9a). In reverse, the M31 cross-sections are characterized by well-bounded tunnels, the pattern being associated with the cuticle presence (Figure 3.9, b). The two degradation patterns may be caused by the action of different types of microorganisms (bacteria or fungi).

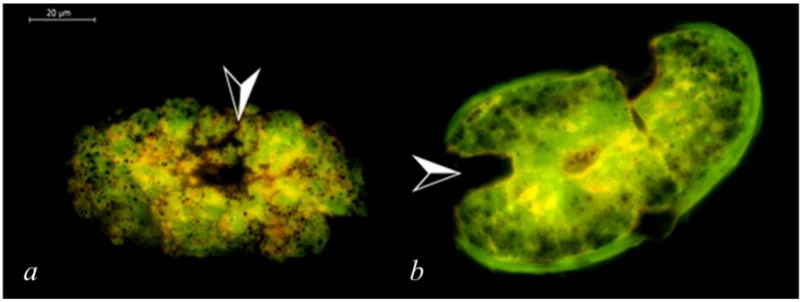

Figure 3.9. Epifluorescence microscopy images showing different alteration patterns of hair fibers (FLUO, cross-sections): (a) hair cuticle completely destroyed associated with microtunnels within the cortex, (b) hair cuticle partially destroyed associated with large tunnels within the cortex, modified after Petraru *et. al* (2020b).

The result based on quantitative analysis shows that the percentage of internal degradation varies from one sample to another ($p< 0.0001$) The sample collected from the C9b skeleton was the most degraded due to the complete destruction of the cuticle layer (Petraru *et. al*, 2020b). The exposure of the subjacent cortex is one of the most important factors in hair weathering (Chang *et al.* 2005).

3.4 Histological characterization of inflammatory reactions at the postcranial skeleton level in three possible cases of treponematosis identified Iasi city (15th-19th centuries)

The inflammatory manifestations (periostitis and osteitis) identified at the three human skeletons from the 15^{th}-19^{th} centuries (coded M40, R30 and R26) suspected of treponematosis (presents pathognomonic characters - *sicca caries*) represents the subject of the histological study.

Considering the archaeological provenance of the biological material, a taphonomic evaluation at the microstructural level is necessary.

M40 skeleton (adult female, 30-35 years old)

The bone samples collected from the M40 skeleton showed good preservation (OHI = 4), with few visible areas of destruction, in some places some areas with infiltrations and taphonomic inclusions were observed.

At the left femur, the histological transverse sections suggest the presence of a new bone formation formed by apposition, on the external surface of the bone (Figure 3.10a). Morphologically, the new bone formation is characterized by the presence of some crypts which, however, do not intersect with the lamellar bone tissue that separates the original tissue from the new proliferative bone structure (Figure 3.10a, d).

The new bone tissue formed as the result of an inflammatory process is described by the presence of primary bone tissue characterized by the lack of bone lamellae and the abundance of osteocyte lacune with a rounded appearance (Figure 3.10b, d). Also, the lamellar bone tissue is present, indicating that remodeling process has been already initiated. The proliferative tissue illustrates the presence of vascular channels remnants, as a marker of the inflammatory process. Between the new bone formation and the cortical bone, a discontinuous lamellar structure can be observed (Figure 3.10d). This structure resembles with structured identified by Schultz (2001). Between the two mentioned structures, bone resorption lacunae can be delimitated.

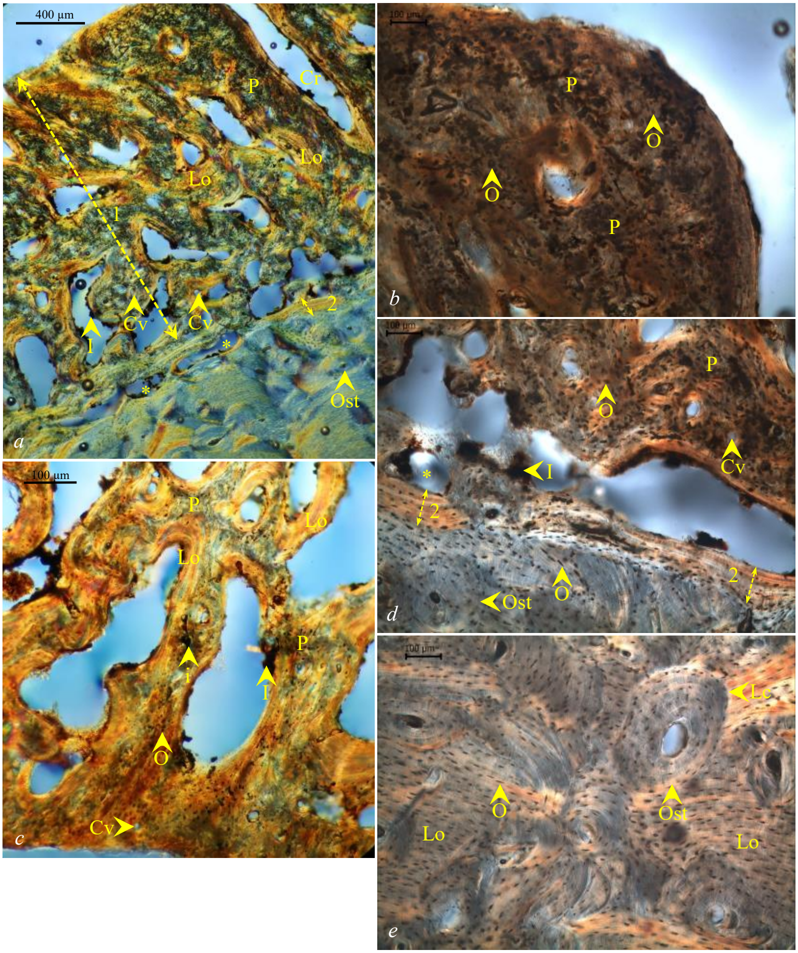

Figure 3.10. Transverse section at the femur hyperostosis belonging to M40 skeleton (OHI = 4): *a* – overview of proliferative bone tissue (1) (POL), *b* – detail of immature bone tissue – callus (DIC), *c* – detail describing the proliferative bone tissue with lamellar bone and immature bone tissue (POL) , *d* – lamellae (2) that separates the cortical bone and the new proliferative bone (DIC), *e* – compact bone tissue unaffected by osteolysis (DIC); Cv – vascular channels , I – taphonomic inclusions, i – taphonomic infiltration, Lo – bone lamellae, P – immature (primary) bone tissue , Cr – crypt, O – osteocyte lacunae, Ost – osteon, Lc – cement line, * – bone resorption lacunae.

The bone sample collected from the right humerus showed a very good preservation, recording only the presence of some inclusions. The periosteal new bone formation showed many cavities, vascular channels, woven and lamellar bone, with a villous morphology. The compact bone tissue revealed many bone resorption lacunae as a result of osteoclastic activity and Howship lacune resulting a special morphology of the affected tissues called "cookie-bite".

R30 skeleton (adult female, 35-40 years old)

The bone samples revealed a good histotaphonomical preservation (OHI = 4), with some deposits in the cavities of the new bone tissue and some isolated areas with inclusions.

The fibula periosteal new bone formation showed different types of morphologies: villous – type and polyp-type (Figure 3.11a, b, c) in comparison with the histoarchitecture described in the M40 skeleton. The bone formed due to the periosteal reaction showed in its structure the presence of cavities (Figure 3.11d). The morphology and the orientation of bone tissue in the polyp-like structure can indicate a recurrent periostitis (Figure 3.11c). A claim at the acute stage of the disease can be suggested by the osteoclastic activity in the cortical area. At this level the presence of numerous bone resorption lacunae near the separation zone between the formed tissue and the cortical area, as well as numerous Howship lacunae, can be noted (Figure 3.11e, f). The osteolytic process can be identified on the whole surface of the cortical bone up to the endosteal surface (Figure 3.11h) where it cand observed several osteons affected by the intense osteoclastic activity.

The new periosteal bone formation at the humerus has a more homogeneous development compared to that of the fibula. The new bone formation collected from the R30 humerus is characterized by the presence of the bone remodeling process and the presence of vascular channels remnants. The cortical bone is characterized by the presence of bone resorption lacunae, Howship lacunae, osteons, and interstitial bone lamellae.

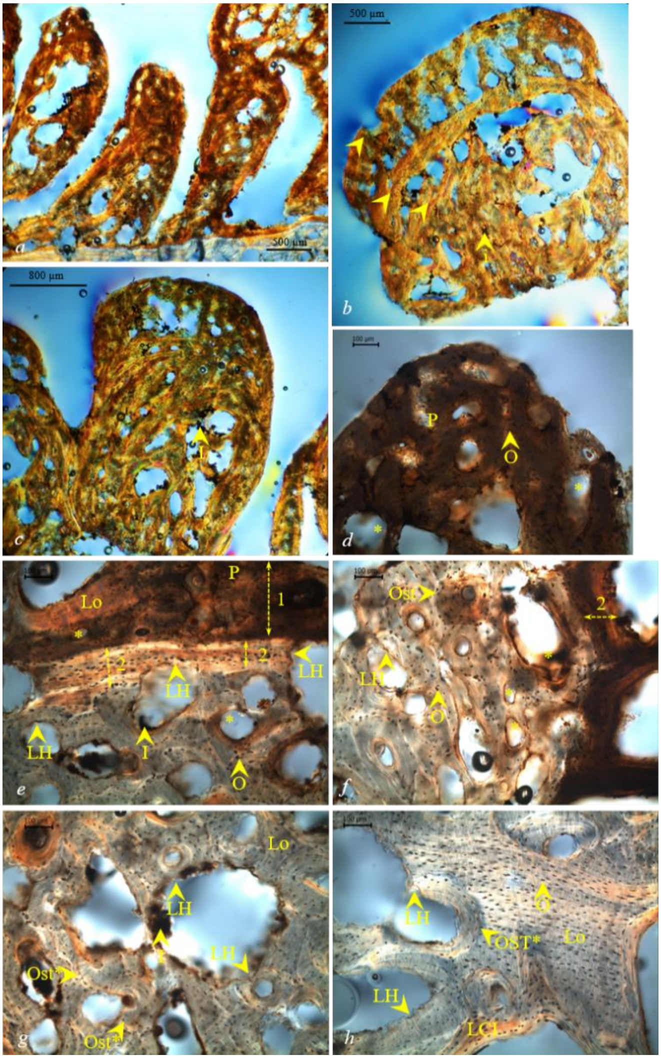

Figure 3.11. Transverse section at the fibula hyperostosis belonging to R30 skeleton: *a, c* – villous-type new periosteal bone formation (POL), *b* –polyp-type new periosteal bone formation (POL), *d* – bifid villous -type bifid of the new periosteal bone formation (POL), *e* – band-like structure that separates the cortical bone from the new proliferative structure (2) (DIC), *f* – compact bone tissue (DIC), *g, h* – cortical bone with several bone resorption lacunae and Howship lacunae (DIC); I – taphonomic inclusions, i – taphonomic infiltration, LH – Howship lacunae, P – woven bone, O – osteocyte lacunae, Ost – osteon, Ost* – osteon affected by osteoclastic activity, LCI – internal circumferential bone lamellae, Lo – bone lamellae, * – bone resorption lacunae.

R26 skeleton (adult male, 35-40 years old)

The bone tissue was sampled only from the right humerus. In comparison with the histological architecture from the previous samples (M40 and R30), the bone sample collected from the R26 skeleton showed particularities at both new bone formation (Figure 3.12a) and cortical zone (Figure 3.12b).

The new bone formed as a result of the inflammatory process of the periosteum is dimensionally reduced. Its dimensions are variable being formed predominant by immature bone with rounded osteocyte lacunae (Figure 3.12a, b, c). In some areas it is formed by bone lamellae (Figure 3.12e). In few areas it can be identified o delimitation between the cortical zone and the new bone formed (Figure 3.12a), in the most cases this lamellar bone structure is missing.

Near the new formed tissue, the presence of several cavities with taphonomic inclusion were identified (Figure 3.12b, c). The cortical zone is characterized by a reduced number of osteons (Figure 3.12a).

The compact bone tissue morphology is probably correlated with the increase of porosity produced not by an osteoclastic activity determined by a periostitis, but by an inflammatory result of the compact bone (osteitis). The sample collected from skeleton R26 reflects a more accentuated postmortem degradation (OHI = 3). Areas of bacterial focal destruction (non-Wedl tunnels) were identified (Figure 3.12f), which, however, does not completely obscure the identification of lamellar bone tissue features.

Comparative analysis of the three possible cases of treponematosis

In the three human skeletons, the right humerus was affected, and bone tissue sampling was performed from the distal part of the diaphysis of this bone to ensure anatomical and biomechanical similarity.

In all three individuals, *caries sicca* was identified in the skull, the human skeletons being in the tertiary stage of the disease. Based on the obtained transverse section, two patterns of the periosteal new bone formation were identified, one for the M40 and R30 skeletons, and one for the R26 skeleton.

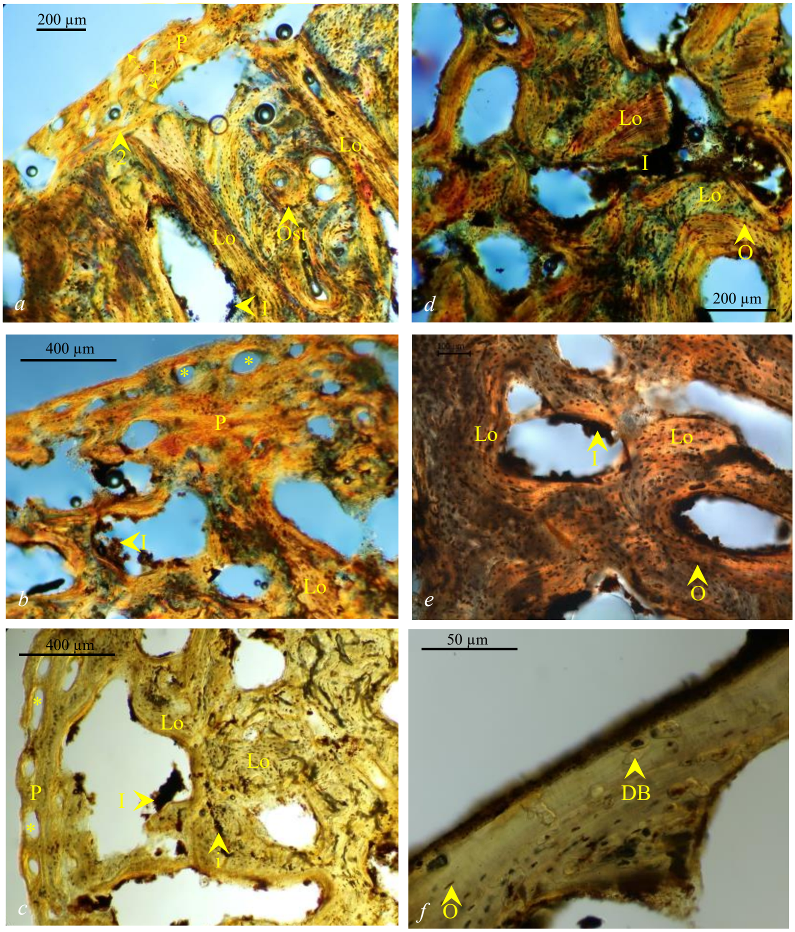

Figure 3.12. Transverse section at the humerus hyperostosis belonging to R26 skeleton: *a, b, c* – new bone formation and cortical zone (a, b – POL, c – BF), *d* – compact bone tissue with a trabeculae aspect- overview (POL), *e* – detail of trabeculae (DIC), *f* – detail bacterial postmortem degradation (BF); DB – bacterial taphonomic degradation, I – taphonomic inclusions, taphonomic infiltration, LH – Howship lacunae, P – woven bone, O – osteocyte lacunae, Ost – osteon, Lo – bone lamellae, * – bone resorption lacunae, 1 – new bone formation, 2 – band-like structure that separates the cortical bone from the new proliferative structure.

The identified pattern at M40 and R30 individuals is characterized by several bone resorption lacunae, having a more accentuated development in comparison with the pattern identified at R26 skeleton. Furthermore, the pattern is present on the entire histological section, while in the case of the R26 pattern the new formed tissue is discontinuous.

Between the new formed tissue on the outer surface of the bone and the cortical bone, a well-individualized lamellar structure was observed in the case of the model identified in M40 and R30 skeletons, while in the case of the R26 skeleton, the lamellar structure is discontinuous on the surface of the section, being present only in certain areas.

The cortical area of the samples collected from M40 and R30 individuals has a similar structure, in which are present osteons, interstitial lamellae, areas of bone resorption and, in some places, Howship lacunae, as a sign of osteoclastic activity. In comparison, the cortical bone of the R26 skeleton has a different morphology with an extremely reduced number of osteons, many cavities at which no Howship lacunae were identified, and an orientation of bone lamellae that suggests a plexiform morphology.

The histological analysis of the samples collected from post-cranial bones suggests acute, chronic, hemorrhagic, proliferative (osteoblastic) and destructive or osteolytic (osteoclastic) episodes that may be suggestive of treponematosis.

The pathognomonic character of these histological structures for treponematosis (newly formed tissue, "grenzlinie", bone resorption lacunae) is debatable, more studies being necessary in this direction. The paleohistological studies contribute to complete paleopathological overview, completing and providing new information regarding the acute or chronic stage of the infection, the time of manifestation of the treponemal infection at bone level captured at the time of the death of the human individual.

Conclusions

1. *This thesis highlights the importance of microscopic analyzes applied in paleoanthropology, as it emerges from the international scientific literature, but also from our own research carried out until now.*

2. *The dental macrowear study of the Royal Court skeletal series could suggest a sex differentiated diet, which represent the hypothesis for dental wear approach at the microscopic level.*
   - ✓ Dental macrowear of the M2 molar for the skeletal series from the Royal Court of Iași affects the male series with a greater intensity.
   - ✓ The semi-quantitative results, correlated with the quantitative ones, can be indicators for a differentiated diet, constituting the hypothesis for the dental wear approach at the microscopic level.

3. *The study of the dental microwear of the 17th century skeletal series discovered in Iași city suggests an abrasive wear for both female and male series, but with different profiles of dental wear, one more homogeneous for the male series and two depending of the laterality of the M2 molars for the female series.*
   - ✓ The dental microwear analysis at the occlusal level, shows a heterogeneous distribution of microwear types on the M2 molars within each skeleton.
   - ✓ We can use the term "abrasive" to characterize the diet of the analyzed human population, due to the large proportion of coarse scratches and large pits.
   - ✓ Within the female series, two models/patterns of dental microwear can be distinguished based on the laterality of the molars, suggesting the predominant use of the right dental hemiarches to triturate hard foods.

- ✓ The dental microwear profile for the male series is more homogeneous, resembling the dental microwear pattern for the molars located on the right hemiarches of the female series.

4. *Dental macrowear in the prehistoric human population from North-East Romania is correlated with age, in 31% of cases for mandibular molars and 49% for maxillary ones, which may suggest that an abrasive diet, poor processing and storage techniques of food could considerably influence the loss of dental tissues.*
   - ✓ Dental macrowear of the prehistoric human population from the North-East of Romania does not differ by sex, but increases with age, being correlated with it in a percentage of 31% for the mandibular molars and 49% for the maxillary ones.
   - ✓ Age, occlusal surface and period, as well as exposed dentin surface are correlated in 38% of cases for mandibular molars and 52% for maxillary molars.
   - ✓ A diet based on cereals consumption and their poor processing and storage methods can considerably influence the loss of dental tissues in the case of prehistoric human populations.

5. *The histotaphonomic study of human hairs discovered in Iași (18th-19th centuries) under similar preservation conditions, illustrates heterogeneous degradation correlated with distinct degradation patterns, being probably the result of the synergistic action of intrinsic biological factors, but also of environmental conditions.*
   - ✓ The hairs discovered and collected from an archaeological site of the 18th-19th centuries belonged to four males, with the age of 50-60 years, and their preservation was favored by the fact that they did not come into direct contact with the soil.
   - ✓ The external taphonomic degradation degree of the samples evaluated by scanning electron microscopy and the histological scores suggest a statistically significant heterogeneous degradation.

- ✓ The quantitative analysis of internal degradation of hairs, based on cross-sections, was correlated with external degradation.
- ✓ Serial cross-sections showed two distinct patterns of degradation that may be the result of the action of different types of microorganisms (bacteria or fungi).
- ✓ Although some preservation conditions (coffin type, burial depth, soil type, estimated age at death and sex) were similar, the hair fibers showed significant heterogeneous degradation. We assume that this variation is most likely caused by the synergistic action of intrinsic biological factors, but also by the environmental conditions involved (burial season, specific burial conditions).

6. *The histological approach to the inflammatory reactions at the postcranial level of the skeletons suspected of treponematosis discovered in Iasi (15th-19th centuries) indicates acute and chronic, relapsing, osteoblastic and osteoclastic episodes that can have a different evolution on anatomical segments within the same individual.*
   - ✓ The good preservation of the skeletons at the macroscopic and microscopic level in archaeological context allowed the description and identification of the structural elements at the histological level. At the level of the R26 skeleton, however, forms of bacterial histotaphonomic degradation were identified.
   - ✓ The histological architecture of the samples collected from the three subjects suspected of treponematosis show acute, chronic, hemorrhagic, osteoblastic, and osteoclastic episodes that may be suggestive of treponematosis. Histologically treponemal infection suggests both periostitis and osteitis.
   - ✓ The morphology of the new formed tissue, as a result of the inflammation of the periosteum, can be an indicator of the recurrence of the treponemal infection. The new formed tissue can have a different structure and morphology (homogenous, polyp-type, villous-type), depending on the acute (inflammatory) or chronic episode of treponemal infection.

- ✓ The manifestation of treponemal infection can have a different evolution within the same individual in different anatomical segments.

Perspectives for further research

The research carried out as part of the doctoral thesis, regarding the microscopic analysis of ancient human remains discovered in archaeological sites in the North-East of Romania, is the first in our country, opening new perspectives for knowledge of the ancient human populations that lived in these territories in a macroregional context.

The directions for further research are multiple and mainly refer to:

- ✓ The approach of new dental samples from different periods - prehistoric, protohistoric, historical and even modern for the study of dental wear.
- ✓ Integration of dental wear results with results regarding dental pathologies, stable isotopes analysis and phytoliths in order to characterize the paleodiet.
- ✓ Knowledge development of past diseases manifested at the skeleton level through histological methods.
- ✓ Integration of histological results with radiographic and computer tomographic results, as well as with medical data for outlining a broad pathological picture.
- ✓ Diversification of age at death methods at human skeletons discovered in archaeological context through cento-chronology studies.

Scientific activity

1. Published articles from the subject of the doctoral thesis

1.1 *Articles published in ISI indexed journals:*

1.1.1 **Petraru, O.-M.,** Groza, V. M., Lobiuc, A., Bejenaru, L., Popovici, M. (2020). Dental microwear as a diet indicator in the seventeenth-century human population from Iasi City, Romania. *Archaeological and Anthropological Sciences*, 12(8), 1-13 (Q2- AIS: 0,7; IF: 2.1).

1.1.2 **Petraru, O.-M.,** Groza, V.-M., Neagu, A.-N., Bejenaru, L. (2020). Archaeological human hair in a burial context: Microscopical evaluation of samples from Iași, Romania (18^{th}–19^{th} centuries). *Archaeometry*, 62(2), 395-409 (Q1-AIS: 0,64, IF: 1,87).

1.1.3 **Petraru, O.-M.,** Bejenaru, L., Popovici, M. (2022). Diet-related dental wear in archaeological human populations of Chalcolithic and Bronze Age from North-Eastern Romania, *HOMO - Journal of Comparative Human Biology*, 73/1, 77–92 (Q4-AIS: 0,26, IF: 0,73).

1.2 *Articles published in BDI indexed journals:*
*(**EBSCO**, ERIH PLUS, ANTHROLIT, INFOBASE INDEX):*

1.2.1 **Petraru, O.-M.,** Groza, V.-M., Bejenaru, L., (2018). Dental Macrowear as Marker of Diet: Considerations on the Skeletal Sample from the 17^{th} Century Necropolis of Iaşi (Iaşi County, Romania). *Annuaire Roumain d'Anthropologie*, 55, 45-54.

1.2.2 **Petraru, O.-M.,** Bejenaru, L., (2019). Overview on Microscopic Methods for Dental Wear Evaluation in Paleodiet Studies. *Annuaire Roumain d'Anthropologie*, 56, 5-14.

1.2.3 **Petraru O.-M.,** Bejenaru L., (2020). Microtaphonomy of archaeological non-skeletal human remains – mini review of microscopic methods. *Annuaire Roumaine d'Anthropologie,* 57: 15-24.

2. Articles published during doctoral studies

2.1 *Articles published in ISI indexed journals:*

2.1.1 **Petraru, O.-M.,** Groza, Bejenaru, L., Popovici, M. (2022). Dimension variability of the M2 human molar teeth: comparisons between prehistoric and medieval samples. *European Journal of Anatomy 26 (4), 371- 386.*

2.1.2 Popovici, M., Groza, V-M., Bejenaru, L., **Petraru, O.-M.,** (2022). Geometric morphometrics of the second molar teeth within the human population from the late medieval city of Iași, Romania. *Archaeometry,* 64(6), 1479-1498 (Q1-AIS: 0,64, IF: 1,96).

2.1.3 Popovici, M., Groza, V. M., Bejenaru, L., **Petraru, O.-M.** (2023). Dental morphological variation in Chalcolithic and Bronze Age human populations from North-Eastern Romania. *Annals of Anatomy Anatomischer Anzeiger,* 245 – 152015. (Q1 – AIS: 0,53, IF: 2,90).

2.1.4 Tencariu, F. A., Asăndulesei, A., Simalcsik, A., Brașoveanu, C. M., **Petraru, O. - M.,** Bejenaru, L., ... Balaur, R. S. (2022). A bolt from the blue: investigations of a singular Bronze Age grave from the Chalcolithic site Ruginoasa (north-eastern Romania). *Archaeological and Anthropological Sciences*, 14(5), 1-18, (Q1- AIS: 0,77; IF: 2,21).

3. Participări la manifestări științifice din subiectul tezei de doctorat

3.1 *Participation at scientific events in the subject of the doctoral thesis:*

- Everyday life in the East-European space from antiquity to the present: interdisciplinary approaches:

3.1.1 **Petraru, O.-M.,** Groza, V.-M., Popovici, M., Bejenaru, L., (2018, October). *Bioindicator of palaeodiet: dental wear in a skeletal sample of 17th century from Iași (Iași County, Romania), Chisinau, Republic of Moldova – oral presentation.*

- The Annual Meeting of the European Association of Archaeologists, EAA:

3.1.2 **Petraru, O.-M.,** Groza, V.-M., Popovici, M., Bejenaru, L., (2019, September). *Dental microwear analysis as an indicator of the diet: case study of a 17th century necropolis from Iași, Romania.* Bern, Switzerland – poster.

3.1.3 **Petraru, O.-M.,** Popovici, M., Groza, V.-M., Bejenaru, L., (2020, August). *Diet-related tooth wear in a human skeletal sample from medieval city of Iași (Romania).* Virtual Networking – poster.

3.1.4 **Petraru, O.-M.,** Groza, V.-M., Neagu, A.-N., Bejenaru, L., (2020, August). *Taphonomic structural changes of archaeological human hairs discovered in Iași (Romania): a microscopic assessment*. Virtual Networking – poster.

3.1.5 **Petraru, O.-M.,** Groza, V.-M., Popovici, M., Bejenaru, L., (2021, September). *Dental macrowear variations in Chalcolithic and Bronze age populations from Northeastern Romania.* Widening horizons, (virtual meeting) – poster.

3.1.6 **Petraru, O.-M.,** Bejenaru, L., Simalcsik, A., Tencariu, F-A., (2021, September). *Preliminary taphonomic evaluation of a Bronze age human skeleton discovered at Ruginoasa (Iasi County, Romania).* Widening horizons, (virtual meeting) – poster.

- The Bioarchaeology Early Career Conference, UK:

3.1.7 **Petraru, O.-M.,** Groza, V.-M., Popovici, M., Bejenaru, L., (2021, March). *Human dental macro-wear during Prehistory: preliminary study on Chalcolithic and Bronze age samples from Nord-Est Romania,* (online) – poster.

- International Colloquium „Prehistoric societies in the Carpathian – Danubian area: environments, technical systems, interactions", Bucharest:

3.1.8 **Petraru, O.-M.,** (2019, Iunie). *Microanalysis of ancient human remains: a doctoral project concerning archaeological sites from east Romania*, București – *oral presentation*.

- International Scientific Conference of Young Researchers, Enhancement of Ethnocultural Heritage in the Education of Young People and Civil Society, 4th edition, *Chisinau, Republic of Moldova:*

3.1.9 **Petraru, O.-M.,** Bejenaru, L., (2019, October). *Paleohistologic approach of archaeologic human remains – a PhD project – oral presentation.*

- International Scientific Conference, Cultural Heritage: Research, Valorization, Promotion, Edition XI: From knowledge to safeguarding and conserving, *Chisinau, Republic of Moldova:*

3.1.10 **Petraru, O.-M.,** Groza, V.-M., Popovici, M., Bejenaru, L., (2019, October). *Microscopic methods for dental wear evaluation in paleodiet studies: case study – necropolis of 17th century in Iași, Romania – oral presentation.*

- The 7th edition of the Landscape Archaeology Conference (LAC), Iași, Romania:

3.1.11 **Petraru, O.-M.,** Popovici, M., Groza, V.-M., Bejenaru, L., (2022, September). *Diet-related wear of the mandibular M2 molar: comparison between Prehistory and 17th century dental samples from North-Eastern Romania –* poster.

3.2 Participation in national scientific events:

- Symposium "ArcheoVest: Interdisciplinarity in Archeology 7th edition: In Honorem Prof. Univ. Dr. Sabin Adrian Luca, Timișoara:

3.2.1 **Petraru, O.-M.,** Popovici, M., Groza, V.-M., Bejenaru, L. (2019, November). *The microwear variation of the M2 molar tooth in a sample of human skeletons from medieval Iași – oral presentation.*

- National Conference of Doctoral Schools from the University Consortium, Second Edition, Timișoara:

3.2.2 **Petraru, O-M**., Groza, V-M., Neagu, A-N., Bejenaru, L., (2019, November). *The histotaphonomic evaluation of ancient human hairs (18th-19th centuries),* *discovered in an archaeological context at the Roman Catholic Cathedral "Adormirea Maicii Domnului" in the city of Iași – oral presentation.*

- The annual scientific session "Anthropology an interdisciplinary approach", Iași Academic Days, 36 edition, "Olga Necrasov" Anthropological Research Center, Romanian Academy - Iaşi Branch, (virtual meeting):

3.2.3 **Petraru, O.-M.,** Groza, V.-M., Popovici, M., Bejenaru, L., (2020, October). *Analysis of dental microwear – a method of intrapopulational diet differentiation – oral presentation.*

3.2.4 **Petraru, O.-M.,** Groza, V.-M., Popovici, M., Bejenaru, L., (2020, October). *Dental macrowear evaluation as an indicator of paleodiet – preliminary study on Bronze Age samples from northeastern Romania – oral presentation.*

- Annual scientific session "Interdisciplinary Anthropology", Iași Academic Days 36/37 edition, Anthropological Research Center "Olga Necrasov", Romanian Academy - Iaşi Branch:

3.2.5 **Petraru, O.-M.,** Groza, V.-M., Bejenaru, L., (2021, October). *Research perspectives in paleohistology: adapting the methodology for obtaining histological preparations from human skeletal remains – oral presentation.*

3.2.6 **Petraru, O.-M.,** Popovici, M., Bejenaru, L., (2021, October). *The relationship between dental wear and diet in ancient Chalcolithic and Bronze Age human populations from North-Eastern Romania* – oral presentation.

3.2.7 **Petraru O.-M.,** Groza V.-M., Neagu, A.-N., Bejenaru, L., (2022, October). *Paleohistological characterization of post-cranial bone lesions: case study of the M40 skeleton discovered at the Roman Catholic Cathedral in Iasi (18th-19th centuries) – oral presentation.*

- Scientific Session of the Faculty of Biology "Trends in Biology: from molecules to complex systems", Iasi, 2021/2022.

3.2.8 **Petraru, O.-M.,** Popovici, M., Groza, V.-M., Bejenaru, L., (2021, October). *Dental microwear patterns – evidence in a human population of 17th century from Iasi city (Romania) – oral presentation.*

3.2.9 **Petraru, O.-M.,** Popovici, M., Neagu, A.-N., Groza, V.-M., Bejenaru, L., (2022, Octombrie). *Histomorphology of bone lesions: evidence of a skeletal treponematosis from 15th-19th centuries, in Iasi, Romania – oral presentation.*

- Annual session of the "Vasile Pârvan" Institute of Archeology of the Romanian Academy "Method, theory and practice in contemporary archaeology", Bucharest 2023.

3.2.10 **Ciorpac-Petraru, O.-M.,** Groza, V.-M., Popovici, M., Neagu, A.-N., Bejenaru, L., (2023 April). *Histological methods applied in paleoanthropology: analysis of ancient skeletal and non-skeletal human tissue – oral presentation.*

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