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Endo-Perio Paleopathology – Antropological and Microbiological Evidences

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Diseases of teeth and the surrounding periodontal ligament are important topics in paleopathological investigations because they provide useful information about living conditions and oral health of ancient populations. Since dental diseases can cause periodontal lesions (and vice versa), these two types of pathological conditions are often closely related. In early populations, the main causes of tooth loss were periodontal diseases (periodontitis) and pulpoalveolar diseases (periapical periodontitis), both of which are attributed to oral infections. However, a little is known about endo-perio pathology in ancient time and there is no relevant paleopathological and paleomicrobiological data in the literature. The paper describes two cases (one from Roman period and one from medieval period of Plovdiv) of endo-perio lesions emphasizing on paleopathological and microbiological evidences. The results revealed typical morphological presentation of endo-perio lesions in combination with mixed microbiota, relevant to this pathology.

Key words: endo-perio lesions, paleopathology, paleomicrobiology

Introduction

The prevalence of periodontal diseases in archaeological populations has been a controversial topic in paleoepidemiology [9]. Early studies on periodontal disease supported the idea that ancient populations experienced little periodontal disease, with the prevalence of periodontal disease increasing in populations during recent centuries [2, 3]. More recent studies of periodontal disease have recognized that the prevalence of periodontal disease has been variable between archaeological populations and factors other than diet also influencing the development of periodontal disease [1, 6, 12]. However the tooth-related factors predisposing initiation and further development of periodontal diseases in archaeological populations are poorly understood. The pulp-periodontal interrelationship is a unique one and can consider them as a single continuous system or as one biologic unit in which there are many paths of communication [14]. The interrelationship of these structures influences each other during health, function and disease [14]. They can get affected individually or combined; when both systems are involved they are called true endo-perio lesions. The relationship between the periodontium and the pulp was first discovered by Simring and Goldberg in 1964 and now is considered as a diagnostic and treatment challenge both in endodontics and periodontology [14]. However, little is known about endo-perio pathology in ancient time and there is no relevant paleopathological and paleomicrobiological data in the literature.

This paper describes two cases (one from the Roman period and one from the medieval period) of endo-perio paleopathology emphasizing on anthropological and microbiological evidences.

Material and Methods

The material used in this study belongs to the paleopathological collection of the Medical University of Plovdiv. The collection had been previously catalogued by the Plovdiv Archeological Museum, including burial details and estimates of age and gender.

The data were collected by the authors. All teeth present in their sockets were recorded. Alveolar bone surrounding the teeth on all surfaces was studied carefully for signs of vertical bony lesions. The criteria for these lesions were a clear vertical loss or destruction of the alveolar bone adjacent to a tooth surface. Both sharp edged ragged lesions and more rounded defects with even and smoother bony surfaces were recorded. Periodontal probes UNC 15 with 1 mm gradations were used for the measurements. The measurement was carried out from the deepest part of the lesion within the alveolar bone up to the surrounding bone crest. Assessment for caries lesions was made by dental probe. Furcation's involvement (according to I. Glickman classification), as well as the presence of calculus, visible periapical lesions and other pathologies were also described.

The heavy deposits of dental calculus provided our study with material for paleomicrobiological analysis. Fragments of calculus around the affected teeth were gently detached and prepared for scanning electron microscopy (SEM) evaluation (JEOL 6390, Institute of Physical Chemistry, Laboratory of Electron Microscopy, Bulgarian Academy of Sciences).

Results

Case #1

The first skeleton originates from archeological excavations dated from the Roman period of Plovdiv (1th-3th century AD) and belongs to a female individual approximately 40-45 years of age. The remains were found in a family tomb (archeological site "Medical University", Vasil Aprilov №3) that belongs to the Western necropolis of the ancient city of Philipopolis (Plovdiv). The anthropological examination revealed right mandibular posterior region exhibiting advanced bone loss, furcation involvement and root approximation of tooth 46. Deep mesial-occlusal caries lesion of 46 with pulp camera exposure was identified (**Fig.1A**) Grade III furcation involvement (3.5 mm) was established by measurement with periodontal probe. The bone loss and furcation involvement were confirmed by periapical radiographs. (**Fig. 1B**).

Case #2

The second skeleton originates from archeological excavations dated from the medieval period of Plovdiv (11th-13th century AD) and belongs to a female individual approximately 35-40 years of age. The remains were found in a nomad necropolis (archeological site "Antique Forum - West"). The anthropological examination revealed asymmetrical deposition of calculus on left side (both upper and lower quadrants) –

(**Fig. 2A, B**). The meticulous checkup revealed deep mesial-occlusal caries lesion of tooth 36 with pulp camera exposure. (**Fig. 2C**) Grade II furcation involvement (1.2 mm) was established by measurement with periodontal probe. The bone loss and furcation involvement were confirmed by periapical radiographs. (**Fig. 2D**).

The SEM observations of detached calculus at different magnifications revealed fossilized microorganisms – cocci in chain, cocci in clusters and bacilli (rods), respectively (**Fig. 3A, B**). The morphological identification was performed in the Department of microbiology using standard SEM pictures of oral pathogens [13]. The morphological analysis revealed Streptococcus mutans, Streptococcus gordonii, Actinomyces naeslundii, Porphyromonas gingivalis and Tannerella forsythia.

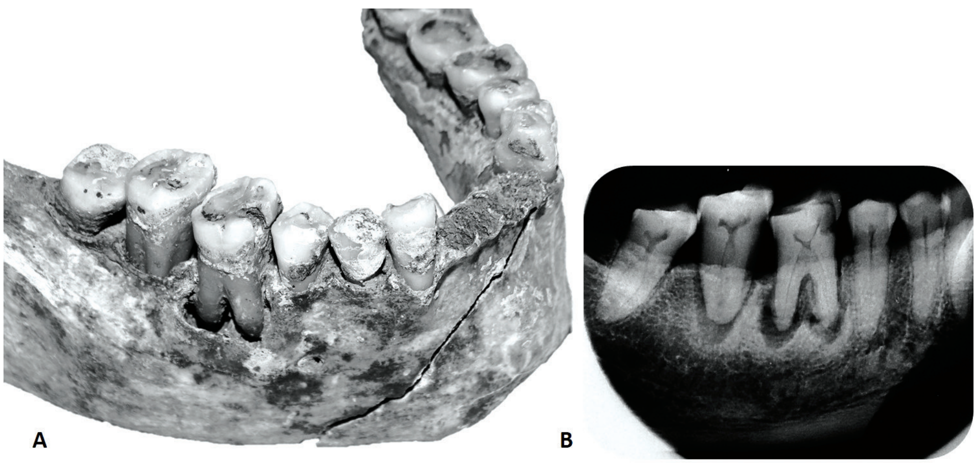

Fig. 1. 40-45 year-old female, buried around 250 AD. Pulpal exposure as a result of deep caries. Note resultant extensive bone loss and full exposure of both mesial and distal roots (**A**). Large apical bony defects on both mesial and distal roots and furcation involvement were verified on X-ray image (**B**).

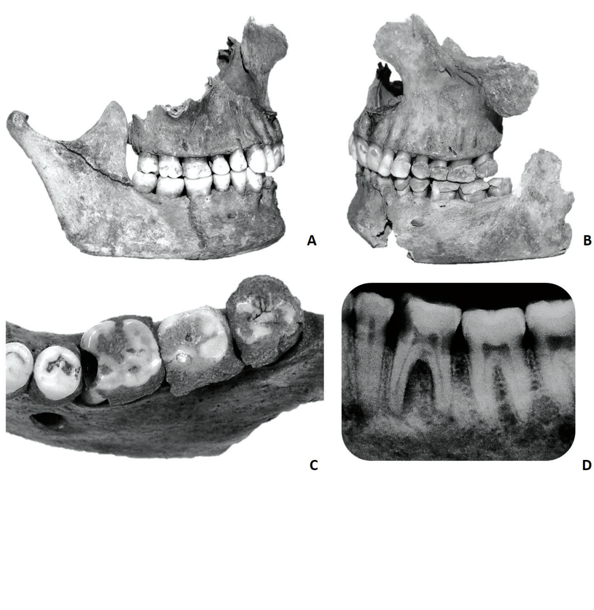

Fig. 2. Right side (**A**) and the affected left side (**B**), showing asymmetrical deposit of calculus. Tooth 36 showed deep MO-carious defect (**C**) X-ray image confirmation of existing endo-periodontal lesion of tooth 36 (**D**).

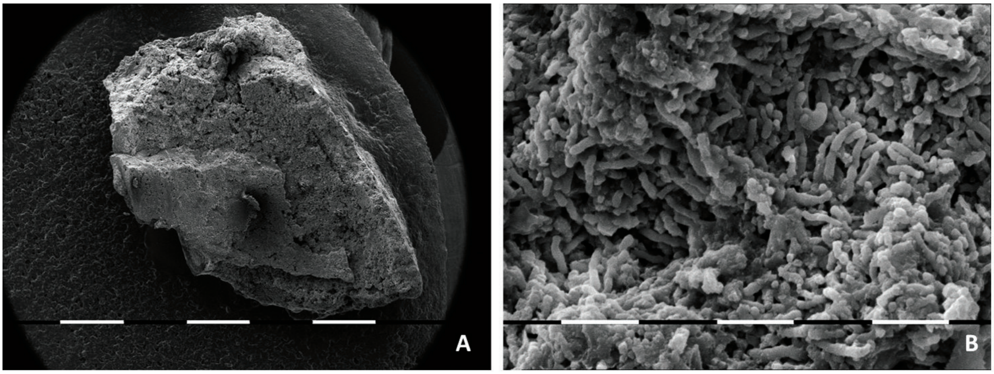

Fig. 3. Dental calculus observed by SEM at different magnification - 40x (**A**) and 200x (**B**) revealed fossilized microorganisms, morphologically identified as oral pathogens.

Discussion

Our findings revealed the typical morphological presentation of the endo-perio lesions in combination with mixed microbiota, relevant to this pathology. Despite the accuracy of anthropological evaluation in cases when the lesions are connected to infection, the microbiological evidences are crucial for the final diagnosis. Nowadays there is a growing recognition of the importance of archaeological dental calculus as a source of oral health information. Early studies of archaeological dental calculus can be traced back nearly a century [7], but it was not until the 1960s and 1970s that dental calculus began to receive serious attention by archaeologists, dental anthropologists and dentists, who described its occurrence in human remains and determined its mineral composition [5, 11]. In addition to macroscopic analysis, scanning electron microscopy (SEM) investigation of dental calculus also greatly advanced during the late 1980s and 1990s. Pioneering work by K. Dobney and D. Brothwell proved a great diversity of well-preserved bacterial forms within dental calculus [4]. The first biomolecular investigation of dental calculus was conducted in 1996 and aimed to identify the oral pathogen Streptococcus mutans (*a causative agent of dental caries!*) through immunohistochemical analysis [8]. In 2011, the preservation of bacterial DNA within dental calculus was confirmed by gold-labelled antibody transmission electron microscopy [10], and this was followed in 2012 by targeted PCR-based genetic approaches, which identified Streptococcus mutans and additional oral taxa, including Fusobacterium nucleatum, Actinomyces israeli, Porphyromonas gingivalis and others.

Microbiology of endo-perio lesions includes bacterial species like Aggregatibacter actinomycetemcomitans, Tannerella forsythia, Eikenella corrodens, Fusobacterium nucleatum, Porphyrominas gingivalis, Prevotella intermedia [14]. Considering the limitation of this study (morphological identification, without genetic confirmation) the microbiota found in the dental calculus (Streptococcus mutans, Streptococcus gordonii, Actinomyces naeslundii, Porphyromonas gingivalis and Tannerella forsythia) confirmed the mixed microbiota involved in endo-perio lesions. Our findings are relevant to data reported by other authors for present populations [14].

Conclusions

This is the first paleopathological report on endo-perio lesions supported by anthropological and paleomicrobiological evidences. Despite the limitations of this study the results revealed typical morphological presentation of endo-perio lesions in combination with mixed microbiota, relevant to this pathology. In ancient times when pulpitis rapidly progressed to periodontitis, and without adequate treatment, every necrotic tooth has been a potential source of oral infection and eventually endo-perio lesion. For this reason, the proper understanding of these coexisting pathologies and the related microbiological status as well as their identification is of primarily importance for human bioarcheology.

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