

Evidence to support the hypothesis of tuberculosis as a cause of extreme osteonecrosis and osteomyelitis of the mandible in a West African population

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S. M. Khullar, N. Katebi, B. B. Herlofson, D. Tvedt, B. R. Olsen: Evidence to support the hypothesis of tuberculosis as a cause of extreme osteonecrosis and osteomyelitis of the mandible in a West African population. Int. J. Oral Maxillofac. Surg. 2016; xxx: xxx–xxx. © 2016 International Association of Oral and Maxillofacial Surgeons. Published by Elsevier Ltd. All rights reserved.

Abstract. Maxillofacial tuberculosis (TB) is rare. The cases of 19 patients showing extreme bony destruction in the mandible, collected over a 3-month period in West Africa, are presented. Clinical, radiographic, and histological evidence indicated *Mycobacterium tuberculosis* as a possible cause. Further studies are in progress.

Key words: osteonecrosis; tuberculosis; mandible; West Africa.

Accepted for publication 17 June 2016

The authors have previously reported 60 cases of extreme osteonecrosis of the jaw (ONJ) based on sporadically collected records over a 3-year period from the hospital ship m/v Africa Mercy, while it was serving in Liberia, Benin, and Togo in West Africa.¹ Many of the patients showed unilateral facial swelling, persistent sinus tracts, and destruction of the jaw bone. The degree of bony destruction was similar to that reported previously for conditions such as phossy or radium jaw. However, the pattern of destruction seen in the group of 60 patients was not the

same in all patients. Some patients had maxillary lesions, although the preponderance was for mandibular cases. Furthermore, it seemed that the osteonecrosis was of two different types. One type of defect produced extremely necrotic 'greyish' bone sequestra of the type associated with 'phossy jaw'. This manifestation occurred in both the maxilla and mandible. Patients with this type of necrosis did not have gross swelling and sinus tracts. The second type, by far the most common in the cohort of 60 cases, was characterized by unilateral gross facial swelling, frequently with

a draining fistula, either intraorally or extraorally.

In this study, the association between tuberculosis (TB) and the development of necrosis was examined in the mandibular samples of 19 additional patients from Liberia and Sierra Leone. The samples were collected over a 3-month period from outpatients who voluntarily attended the Mercy Ships Dental Clinic while the ship Africa Mercy was stationed in Sierra Leone for a period of 9 months and from outpatients voluntarily attending Trinity Dental Clinic in Liberia. The osteonecrosis

of these additional patients strongly resembled the second type of disease process described for the first group of 60 patients, both clinically and in the pattern of bony destruction of the mandible.

A potential link between TB and destruction of the jaw bones is suggested by three recent case reports describing patients with a tuberculous lesion in the condylar head of the mandible. In one case, a bone scan demonstrated an inflammatory destructive process in the condyle, ascending ramus, and angle.² This pattern of destruction has a notable resemblance to the anatomical location of the ONJ in the 19 patients included in the current study. The second and third case reports presented data in support of the conclusion that the condylar head lesion was a primary tuberculous lesion.^{3,4}

Methods

Patient population

The tissue samples of the 19 participants in this study were collected between March and June of 2011 from patients in Sierra Leone ($n = 17$) and Liberia ($n = 2$). The patients were voluntarily attending dental clinics for the treatment of pain and swelling in the facial region. All of the patients presented with unilateral gross facial swelling, frequently with a draining fistula, either intraorally or extraorally. The patients answered a brief questionnaire regarding their general health and occupation. They were subsequently photographed to document the

clinical appearance at the time of presentation.

Gross bony destruction was demonstrated by panoramic radiography and/or cone beam computed tomography (CBCT) in all but one of the cases. Facilities and logistics at the clinics limited the possibilities of chest X-rays and blood tests.

The patients all underwent a surgical debridement procedure as part of their treatment. After their surgical procedure was complete, and subsequent to obtaining informed consent from the patient, the discarded surgical debris was collected in 10% formaldehyde.

Specimen analysis

Micro-computed tomography (micro-CT)

High-resolution three-dimensional (3D) micro-CT scans (MicroXCT-200; Zeiss) were used to evaluate the degree and pattern of bony destruction in the condylar head of samples from four patients.

Histology and immunohistochemistry of soft tissue samples

Based on the limited amounts of soft tissue associated with the bony fragments, samples from four patients were selected for paraffin embedding, sectioning, and haematoxylin and eosin (H&E) staining and immunohistochemistry, following standard protocols. Sections of samples from two of the four patients were stained with antibodies against EMR-1 (PA5-33502; Thermo Fisher Scientific, MA, USA), a

marker for mature macrophages. In addition, immunohistochemical staining with antibodies (GWB-EF714E; GenWay Biotech Inc., CA, USA) against purified proteins derived from *Mycobacterium tuberculosis* was used to determine whether mycobacterial proteins were present in the soft tissue samples.

Results

Clinical characteristics

Clinical characteristics are summarized in Table 1. All 19 patients showed destructive lesions in the mandible.

The median age of the patients was 24 years. The oldest was aged 70 years and the youngest was 17 years (Fig. 1).

Seven patients reported pain elsewhere in the body. These areas included the neck, back, and 'body'. One patient reported bilateral hip pain in addition to back pain, and one patient reported chest pain while coughing. Eleven of the patients reported having fever and fatigue.

Nine patients showed lesions on the left side and four showed lesions bilaterally, but none had bilateral condylar lesions. Five patients had right-sided lesions. One patient had a lesion in the anterior body region. Thirteen of the patients had an actively draining extraoral fistula and one patient had a closed extraoral fistula. Thirteen of the patients had condylar lesions. Eight other patients had posterior body/angle/ascending ramus lesions.

Only one patient had an isolated anterior lesion, and further examination of his

Table 1. Characteristics of the patient population.

Patient	Sex	Age, years	Location	Specified location	Discharging extraoral sinus	Exposed bone intraorally	Additional signs and symptoms
1	M	28	Mandible	Bilateral angle/condyle	Yes	No	Multiple extraoral fistulae
2	F	22	Mandible	Left body	Yes	Yes	Large extraoral wound with necrosis
3	F	20	Mandible	Right ramus/condyle	No	No	Discharging intraoral wound
4	M	56	Mandible	Left body/angle/condyle	Yes	No	Multiple discharging extraoral fistulae
5	F	24	Mandible	Left body/angle/condyle	Yes	Yes	Discharging intraoral wound
6	F	23	Mandible	Left angle/condyle	Yes	No	Multiple discharging extraoral fistulae
7	F	18	Mandible	Left angle/condyle	No	No	Discharging intraoral wound
8	M	32	Mandible	Anterior body	No	No	Multiple mobile teeth
9	M	41	Mandible	Right ramus/coronoid	No	No	Closed extraoral sinus
10	M	23	Mandible	Left angle/ramus/condyle	No	No	
11	F	70	Mandible	Left body	No	No	Discharging intraoral wound
12	M	25	Mandible	Left body/angle/condyle	Yes	No	
13	M	42	Mandible	Right ramus/condyle	Yes	No	Discharging intraoral wound
14	F	22	Mandible	Right body/angle/condyle	Yes	No	Discharging intraoral wound
15	M	17	Mandible	Left body/angle/condyle	Yes	No	Multiple discharging extraoral fistulae
16	F	21	Mandible	Bilateral body	Yes	No	
17	F	27	Mandible	Bilateral body/condyle	Yes	No	Pain for 8 years; discharging intraoral wound
18	M	19	Mandible	Bilateral body, left angle/condyle	Yes	No	
19	F	30	Mandible	Right body	Yes	No	Discharging intraoral wound

M, male; F, female.

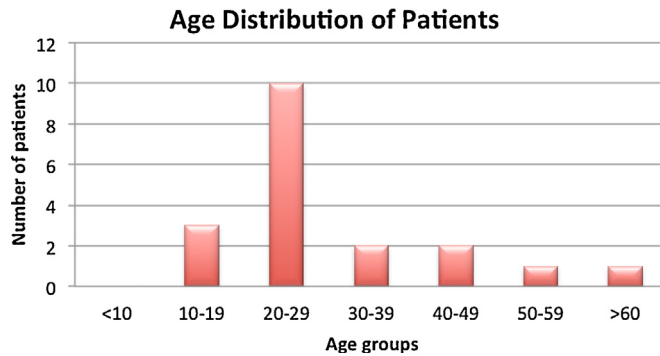


Fig. 1. Age distribution of patients.

clinical photographs and history, with no description of a draining sinus or exposed bone, suggests that the destruction seen on CBCT was related to the severe periodontal condition around his lower incisors. His clinical details suggest that his lesion may represent a different pathology to that of the other patients in the group.

Distinctive swelling, actively discharging fistulae, and scarring in the angle region of the mandible were present in 14 of the 19 patients of which 4 were bilateral (Figs 2A and B, 3A and 4A and B). A characteristic ascending ramus

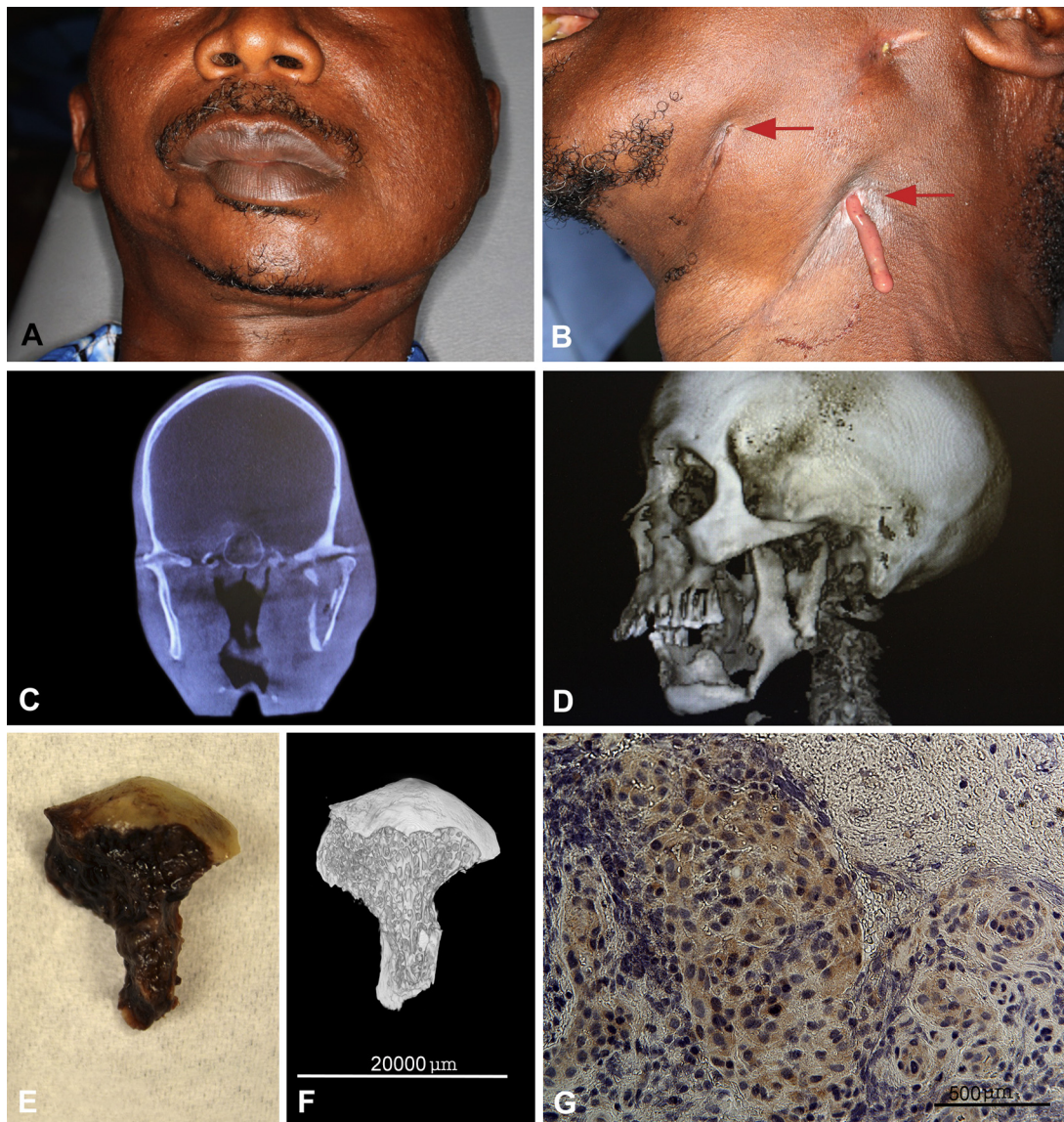


Fig. 2. Adult male with recurrent draining fistulae in the left angle and body region of the mandible. Family members had similar swellings and fistulae. Frontal view showing asymmetrical swelling (A). Lateral view showing scarring from previous fistulae and a fistula actively draining pus (B). 3D reconstructed CBCT showing destruction in the ascending ramus and on the medial aspect of the condylar head extending towards the mandibular angle in coronal view (C) and lateral view (D). Fragment from surgical debridement showing destruction on the medial aspect of the condylar head (E). Micro-CT scan of the fragment showing pitted surface destruction on the articular and medial aspects of the condylar head (F). Tissue section showing macrophages stained with EMR-1 antibody (G) (scale bar 500 μ m).

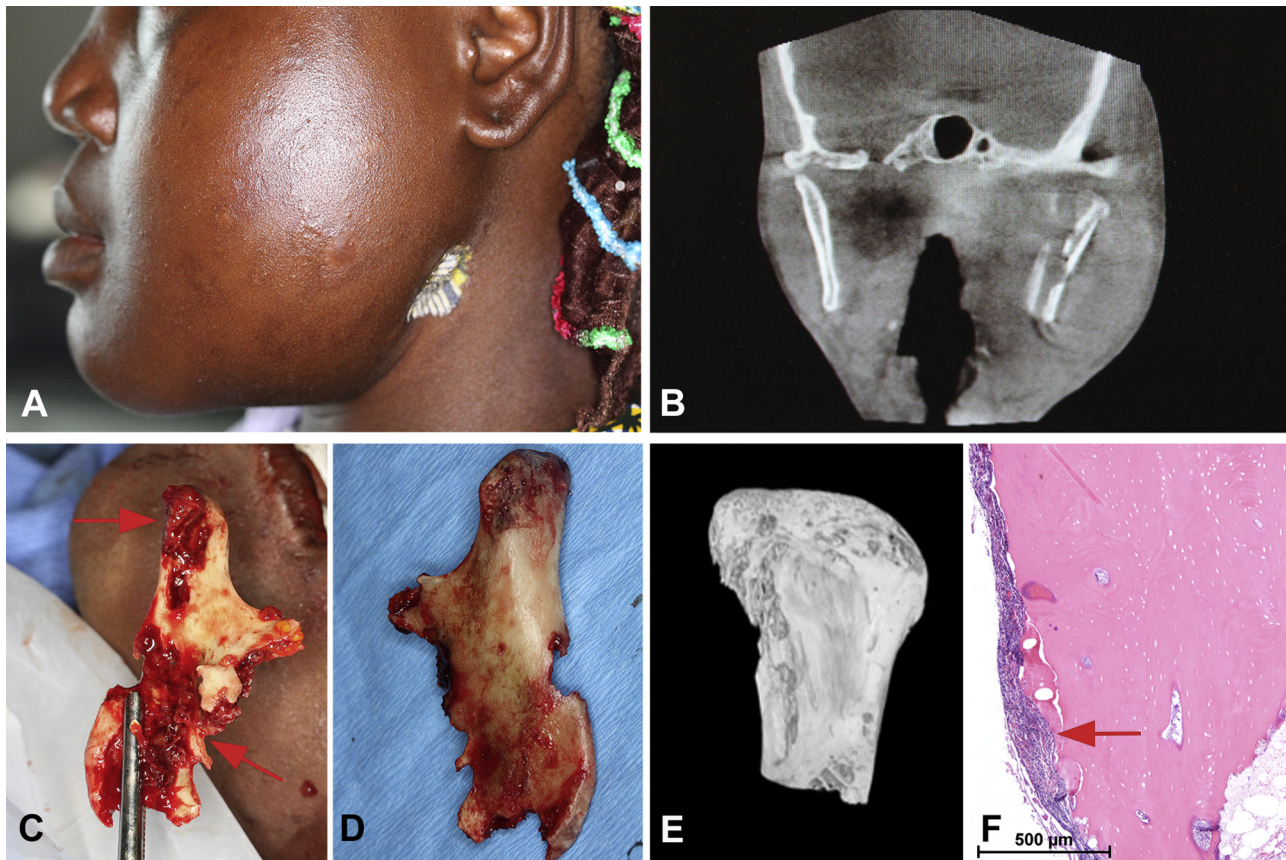


Fig. 3. Adult female with left facial swelling and a draining fistula at the mandibular angle (A). CBCT showing destruction of the medial aspect of the left condylar head and ascending ramus (B). Bony fragment from surgical debridement, with destruction of the medial aspect of the condylar head and ascending ramus (C); lateral view of the fragment (D). Micro-CT showing pitted surface destruction of the condylar medial and articular surfaces (E). H&E stained section of a bony fragment showing a cell-rich periosteum (arrow) adjacent to almost acellular bone (F) (scale bar 500 μm).

split and medial condylar destruction was frequently seen in the 17 CBCTs that were available (Figs 2C–E, 3B–D, 4C and D and 5A–C).

Micro-CT and soft tissue staining

Bony destruction seen on micro-CT scans (Figs 2F, 3E and 5D) showed a distinct pitted cortical surface destruction of the condylar heads.

Samples from four patients had some soft tissue available for histology and immunohistochemistry. Histological analysis of sections from these samples showed inflammation with the presence of multiple blood vessels, macrophages, and extravasated erythrocytes (Fig. 4E and F). An area of acellular bone, but with a cell-rich periosteum, can be seen in Fig. 3F. Inflammatory tissue with ring-like endothelial structures containing small blood vessels and macrophages can be seen in Fig. 5E. The lamellar bone in Fig. 5E also shows a few remaining osteocytes. A corrugated front of bony destruction adjacent to

necrotic tissue can be seen in Fig. 5F. In sections stained with EMR-1 antibody for the detection of macrophages, large numbers of positive cells were seen (Figs 2G and 4G and H). Staining with antibodies against *M. tuberculosis* antigens demonstrated intense granular staining of macrophage-rich regions with multiple positive round and rod-like structures within and between cells (Fig. 4I).

Discussion

The World Health Organization (WHO) Global Report for 2015 covers 99% of the world population and examines the prevalence of TB in 205 countries.⁵ In 2014, the African region had 28% of the world TB cases, with an incidence of 281 cases for every 100,000 individuals. The global incidence for TB was 133 cases for every 100,000 individuals.

During the 10.6 weeks that 17 of the 19 patients studied here were treated in Sierra Leone, altogether 3085 patients were treated at the Mercy Ships Dental Clinic. As

all of the patients chose to come to the clinic for the treatment of some form of oral or maxillofacial pathology, this was not a random selection of the population in Sierra Leone. In this specific patient group, the incidence of the clinical symptoms described in this paper was 551 for every 100,000 patients treated.

Pulmonary TB is the most common form of the disease; however, extrapulmonary TB comprises 20% of cases.⁶ TB can affect different systems of the body, including the skeleton, central nervous system, kidneys, and the lymph nodes and lymphatics.⁷ Bone and joint TB may account for up to 35% of all extrapulmonary TB. Skeletal TB most frequently involves the thoracic spine, followed by TB arthritis in weight-bearing joints.⁷

Chest X-rays show pulmonary disease symptoms in about 50% of patients with osteoarticular TB, but active pulmonary disease is uncommon.⁸ Andrade and Mhatre in 2012 reported 46 cases of orofacial TB seen over a 16-year period; 33 of these

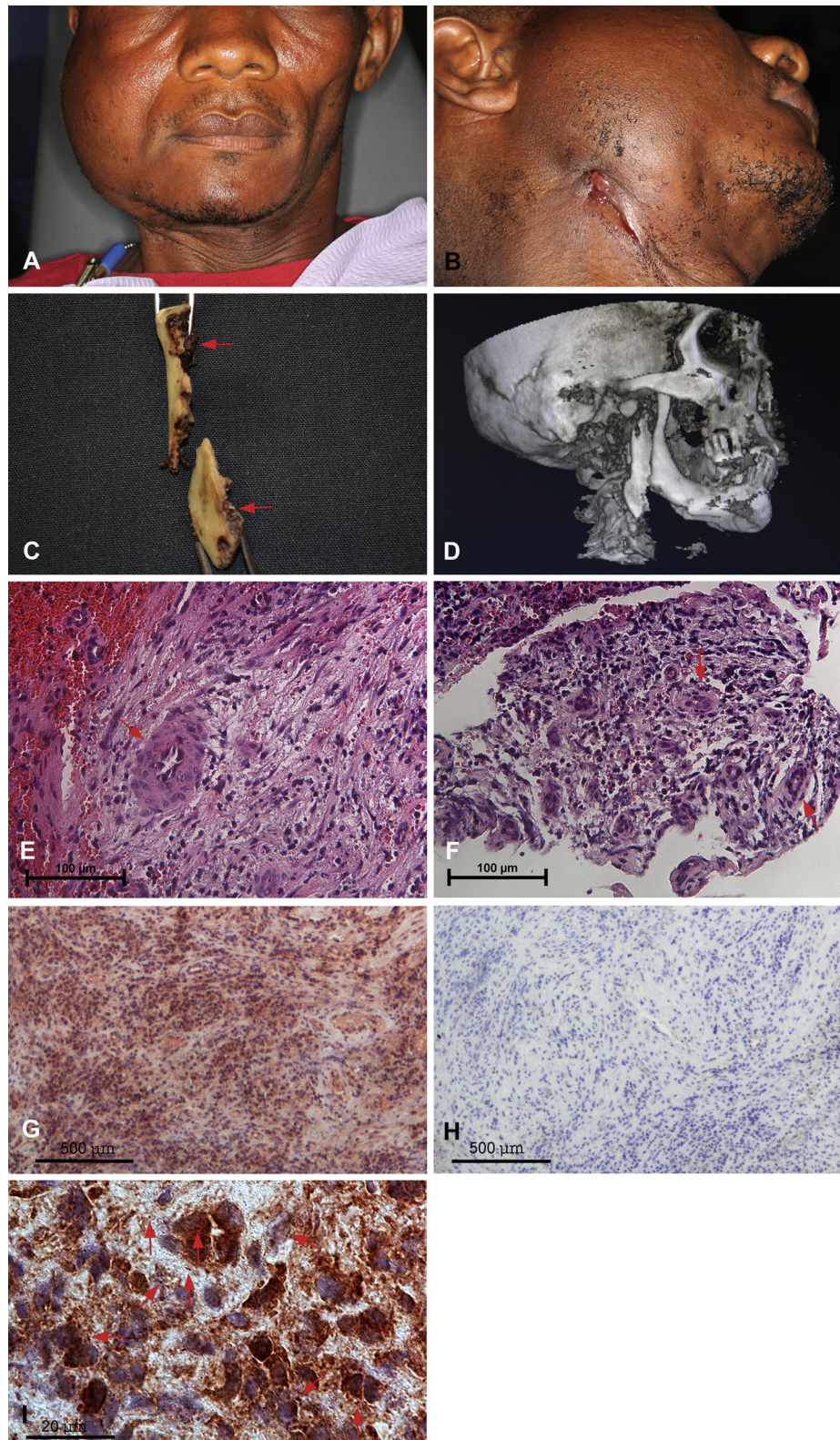


Fig. 4. Adult male with unilateral facial swelling (A), and a fistula at the mandibular angle (B). Lesions developed after he removed an infected tooth himself. Formalin-preserved fragment showing medial destruction of the condylar head and ascending ramus (C). CBCT showing destruction of the condylar head and ascending ramus (D). H&E stained section showing a thick walled blood vessel (arrow) and extravasation of red blood cells in the surrounding tissues (E) (scale bar 100 μm). H&E stained section showing inflammatory tissue with macrophages, multiple thick walled vessels, and red blood cells (F) (scale bar 100 μm). Tissue section showing large numbers of EMR-1 antibody-positive macrophages (G), compared with negative control antibody (H). Tissue section stained with antibody against mycobacterial proteins showing large numbers of round or elongated structures (arrows) within and around macrophages (I) (scale bar 20 μm).

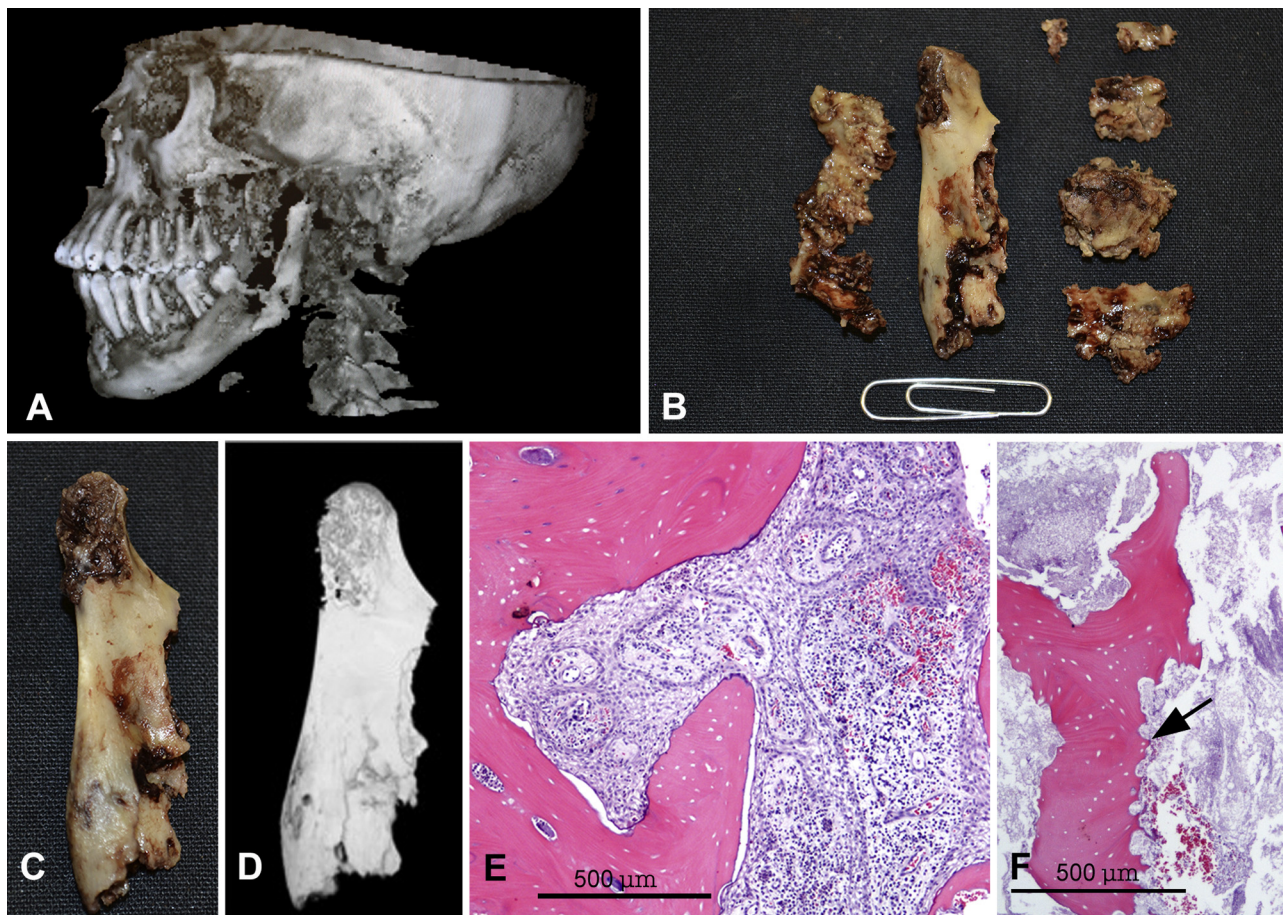


Fig. 5. A young adult male who developed an extraoral fistula about 3 months prior to seeking treatment after dental pain for about 1 year. CBCT showing destruction of the left ascending ramus and condylar head (A). Formalin-preserved fragment of the condylar head, distal border of the ascending ramus, and coronoid process (B). Medial destruction of the left condylar head (C). Micro-CT showing 'pitted' bony destruction on the medial side of the condyle (D). H&E stained section showing inflammatory tissue with extravasation of blood cells and ring-like endothelial structures; lamellar bone showing a few remaining osteocytes (E) (scale bar 500 μm). H&E stained section showing a corrugated front of bony destruction (arrow) (F) (scale bar 500 μm).

patients did not have evidence of pulmonary TB.⁹ A retrospective study of 42 cases of oral TB by Mignogna et al., reported that only 13 of the 42 cases were associated with primary oral infection.¹⁰ Secondary infection in the maxillofacial region can occur by hematogenous spread of the bacilli from a primary focus elsewhere in the body.⁹

The patients with tuberculous lesions of the condylar head described by Patel et al. and Hebling et al. resided in Western Europe (UK and Switzerland), but had emigrated to Europe from Africa.^{2,3} A case of primary tuberculous condylar destruction reported by Ranganathan et al. was from northern India.⁴ The pattern of destruction in the edentate areas of the mandible in the current study resembles those seen in the single case reports by Patel et al.,² Hebling et al.,³ and Ranganathan et al.⁴

Several patients in the current study reported that family members and neighbours in their villages had similar lesions.

Primary tuberculous osteomyelitis of the mandible is extremely rare. It may occur through a carious tooth or in an area of gingivitis.^{2,11,12} Andrade and Mhatre collected 46 cases of orofacial TB over a 16-year period, in an area where TB is endemic (Mumbai, India), consistent with the rarity of orofacial TB.⁹ These authors found the same male to female ratio and age distribution as was found in the present study. Twenty-two of the 46 cases presented with a lesion in the mandibular angle. The clinical photographs presented in the article by Andrade and Mhatre resemble those obtained in the present study, showing unilateral facial swelling in the posterior mandibular region. Fourteen of the patients in the study of Andrade and Mhatre also presented with evidence of extraoral fistulae,⁹ as did 14 of the patients in the present study.

Interestingly, Mignogna et al. reported 42 cases of oral TB, but only 21.4% of these cases had any bony involvement.¹⁰

This is in contrast with the present study, in which all 19 patients had some degree of bony involvement, and with the study by Andrade and Mhatre.⁹ The reasons for this difference in presentation are not known; further studies on the mycobacterial strains involved may be useful.

Osteonecrosis of the jaws (ONJ) refers to a condition in which the maxilla and mandible become necrotic with or without exposure. The causes may be related to metabolic defects, infection, radiation, systemic medication, direct chemical toxicity, or trauma.¹³ The development of ONJ may also be due to some factor that compromises the blood supply to the jaw bones resulting in local ischaemic tissue death.¹⁴

When trying to elucidate the cause of the bony destruction seen in the patients described in this study, the histological findings provide important clues. Histology indicated an intense vascular inflammatory response, with massive cellular

infiltration that includes large numbers of EMR-1-positive macrophages, extravasation of erythrocytes, signs of intense osteoclastic activity, and cell-rich periosteal regions. These features are consistent with a response to microbial invasion. In contrast, the histology associated with bisphosphonate or osteoradionecrotic ONJ reflects the toxic chemical and radiation effects, such as empty avascular marrow spaces and no signs of osteoclastic activity. In osteoradionecrosis the periosteal tissues are also viable.¹⁵

Macrophages play a specific, central role in the development of ONJ¹⁶ and are the major target of invasion by *M. tuberculosis*.¹⁷ Infection triggers massive monocyte recruitment from the blood to affected areas, followed by differentiation into macrophages¹⁸ and osteoclasts in periosteal regions of bones. When activated, tumour necrosis factor (TNF)- α expressing macrophages show increased membrane ‘ruffling’ and develop phagolysosomes, compartments of acidic hydrolases able to cause bone resorption.¹⁹ The pattern of macrophage-based destruction of cortical bone surfaces, as opposed to osteoclastic resorption of trabecular bone starting in the bone marrow,²⁰ is remarkably similar to the ‘pock marked/resorption pit’ pattern on the bony surfaces of the condyles seen in the patients studied here.

In addition to the positive staining with antibodies against *M. tuberculosis* proteins, a positive amplification of portions of the IS6110 and MPB64 marker genes by polymerase chain reaction with DNA isolated from some samples (data not shown) supports the conclusion that the ONJ in this cohort of patients was associated with a mycobacterial infection. To further validate this conclusion and characterize the bacterial strains involved, bacterial genome sequencing with DNA from patients with the characteristic pathology described here will be required.

In conclusion, many of the patients in the current study showed an almost identical clinical presentation to case reports documenting tuberculous lesions of the condylar head.^{2–4} The presence of antigenic material derived from *M. tuberculosis* in macrophages was demonstrated in soft tissues clinging to the bony sequestra. These activated EMR-1-positive macrophages, in conjunction with osteoclasts, may have been responsible for the pattern of bony destruction observed by micro-CT imaging of the condylar head. Several factors suggest that there may be an association between *M. tuberculosis* infection and the pattern of maxillofacial

destruction seen in many of the patients in this group.

Funding

None.

Competing interests

None.

Ethical approval

Ethical approval regarding the use of patient material was negotiated by Mercy Ships with the respective host countries. Harvard University Faculty of Medicine, Committee on Human Studies: ethical exemption for laboratory examination of tissue samples was given (CHS study number M21590-101).

Patient consent

Informed, signed patient consent was obtained for all individuals participating in this study.

Acknowledgements. The authors would like to thank Dr. Keith Chapman for his help in clinical sample collection. The authors would like to thank Mr. Haakon Stormer for his expertise in preparing the figures.

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