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Title: Calcified structures associated with human skeletal remains: possible atherosclerosis affecting the population buried at Amara West, Sudan (1300-800BC)

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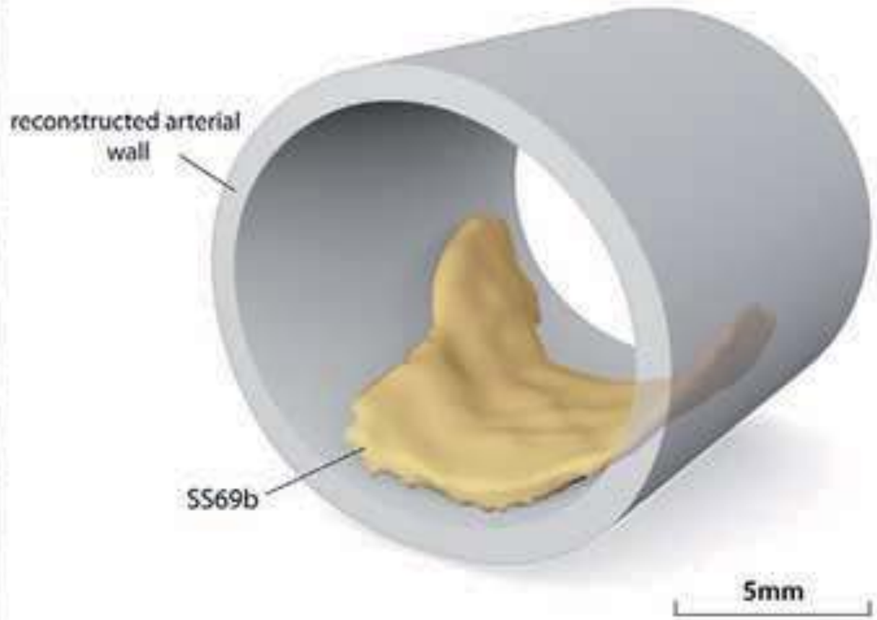
Abstract: Today, cardiovascular diseases (CVDs) are the leading cause of death worldwide. Atherosclerosis, the thickening of the artery wall due to accumulating lipids, is one of the major causes. Generally assumed to be a disease of modern life-style related factors (smoking, obesity, hypertension), its history and epidemiology in the past are virtually unknown. Research on mummies from various geographic locations, time periods and socioeconomic backgrounds has revealed conclusive, albeit scant, evidence that atherosclerosis also affected past human populations. Little is known about the morphology of calcified atherosclerotic plaques that may be associated with human skeletal remains. Therefore, direct evidence of atherosclerosis from skeletal remains is largely absent. This paper presents five possible examples of calcified blood vessels which may represent atherosclerosis recovered from burials at Amara West, Sudan (1300-800BC) and reviews other potential causes of arterial calcification. Calcifications were recovered from the chest area of three middle-adult individuals as well as from the abdominal area and alongside the femur of two more. Based on morphology, anatomical location, scanning electron microscopy and radiography, they are probably calcified arterial plaques. These findings are unique in the bioarchaeological record and indicate that people have experienced atherosclerosis for at least 3000 years.

Graphical Abstract (for review)



Highlights:

- Calcifications were recovered with skeletons from Amara West, Sudan (1300-800BC).
- The structures can possibly be identified as calcified blood vessels.
- The structures may result from calcification of atherosclerotic plaques.
- The calcifications could be evidence of atherosclerosis in skeletal human remains.

Calcified structures associated with human skeletal remains: possible atherosclerosis affecting the population buried at Amara West, Sudan (1300-800BC)

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Abstract

Today, cardiovascular diseases (CVDs) are the leading cause of death worldwide. Atherosclerosis, the thickening of the artery wall due to accumulating lipids, is one of the major causes. Generally assumed to be a disease of modern life-style related factors (smoking, obesity, hypertension), its history and epidemiology in the past are virtually unknown. Research on mummies from various geographic locations, time periods and socioeconomic backgrounds has revealed conclusive, albeit scant, evidence that atherosclerosis also affected past human populations. Little is known about the morphology of calcified atherosclerotic plaques that may be associated with human skeletal remains. Therefore, direct evidence of atherosclerosis from skeletal remains is largely absent. This paper presents five possible examples of calcified blood vessels which may represent atherosclerosis recovered from burials at Amara West, Sudan (1300-800BC) and reviews other potential causes of arterial calcification. Calcifications were recovered from the chest area of three middle-adult individuals as well as from the abdominal area and alongside the femur of two more. Based on morphology, anatomical location, scanning electron microscopy and radiography, they are probably calcified arterial plaques. These findings are unique in the bioarchaeological record and indicate that people have experienced atherosclerosis for at least 3000 years.

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Introduction

During the 21st century, cardiovascular diseases (CVDs), have replaced infectious diseases as the leading cause of death worldwide (WHO, 2013). Calcification or mineralisation of blood vessels is a very common feature of cardiovascular disease. This process can originate in the arterial intima, media, microvessels or valve leaflet (Demer and Tintut, 2008). Clinically, at least four different types of vascular calcification can be distinguished (see Table 1): atherosclerotic calcification, medial vascular calcification, aortic valve calcification, and vascular calciphylaxis (Johnson et al., 2006; Towler, 2008). Detection of arterial calcification in a clinical context is mainly based on plain film radiographs, computed tomographic (CT) scanning or an angiogram.

By far the most common cause of arterial calcification is atherosclerosis (Demer and Tintut, 2008), generally defined as the thickening of the artery wall resulting from an accumulation of lipids in the arterial intima (Lusis, 2000). With increasing size, the accumulated materials gradually form plaques (*atheroma*), initially integrating fibrous tissue. In advanced stages, mineralisation or calcification of plaques through calcium deposition can occur (calcification, Fig. 1). The mechanisms governing plaque calcification are now understood to be similar to ossification of bone (Abedin et al., 2004; Demer and Tintut, 2008). Such calcifications are very common features of atherosclerosis in modern clinical studies and are considered pathognomonic for atherosclerosis (Stary et al., 1995). Atherosclerosis can occur in all major and medium-sized arteries (Lam, 2012).

Early stages of atherosclerosis prior to atheroma formation are considered clinically silent and can already occur in children (Stary et al., 1994). Morbidity and mortality due to ischemia or thrombosis are only linked to the advanced stages and are usually seen from the third decade of life onwards where fibrous or calcified plaques are present (Stary et al., 1995). However, even though age is an important contributory factor, atherosclerosis is not a degenerative disease (Rose, 1991). It has been shown to have a complex, multifactorial aetiology with genetic, environmental and life-style related risk factors (Lusis, 2000; Lusis et al., 2004). The main life-style related influences are a high-fat diet, smoking and lack of physical activity, as well as hypertension and diabetes. In recent years a number of infectious diseases such as *Chlamydia pneumonia*, *Helicobacter pylori* and periodontal disease have also been associated with atherosclerosis (Rosenfeld and Campbell, 2011; Scannapieco and Genco, 1999). Poor maternal health and low birth weight are further suspected to significantly increase the risk of developing atherosclerosis in adult life (Barker, 1998).

Other forms of arterial calcification are generally far more rare (Abedin et al., 2004; Demer and Tintut, 2008). Medial vascular calcification, also known as Mönckeberg's arteriosclerosis, is a common complication of diabetes mellitus and chronic kidney failure as well as being associated with advanced age (Towler, 2008). In contrast to atherosclerotic calcification, calcium deposition starts in the arterial media and causes calcification of the entire arterial circumference (Fig. 1). Medial calcification mainly affects the peripheral arteries of the lower limbs (Sinha et al., 2008), and atherosclerosis usually occurs as a secondary phenomenon (Demer and Tintut, 2008). Metabolic conditions such as uremia and hyperparathyroidism can also lead to non-atherosclerotic calcification of blood vessels affecting the microvessels, even though this is generally very rare (Demer and Tintut, 2008; Towler, 2008). None of these conditions is mutually exclusive and there can be considerable overlap, with atherosclerosis usually developing in due course (Demer and Tintut, 2008).

Arterial calcifications in the palaeopathological record

Evidence of arterial calcifications, usually diagnosed through the presences of atherosclerotic plaques, has been recognized in ancient human remains since the mid-19th century (Czermak, 1852). Examples almost exclusively derive from studies of mummies. The disease received particular attention during the early days of mummy studies and was detected in a number of individuals from ancient Egypt and Peru (e.g. Long, 1931; Ruffer, 1911; Shattock, 1909; Shaw and Bernard, 1938; Smith, 1912; Williams, 1927). The diagnosis is either based on the detection of calcified atheromatous plaques in artery walls through histological analysis (Zimmerman, 1993) or by radiographic assessment (Allam et al., 2009), even though during early autopsies in some individuals extensive calcifications were readily identifiable macroscopically (Ruffer, 1911; Smith, 1912). Atherosclerosis has been identified in mummies from a wide range of geographical areas and chronological periods ranging from the Neolithic Iceman (Gostner et al., 2011; Keller et al., 2012) to mummies from China (Sakurai et al., 1998), Peru (Williams, 1927) and the Arctic (Zimmerman, 1998). The vast majority of reported examples come from ancient Egypt (e.g. Allam et al., 2009; Ruffer, 1911; Sandison, 1962, 1967; Zink et al., 2011), and the oldest individual with evidence of atherosclerosis dates to the Predynastic period (6000-3100BC) (Moodie, 1931: 26). Most of these mummies had elite or royal backgrounds, which led to the conclusion that in the past atherosclerosis may have been confined to higher social classes with living conditions more similar to those experienced by modern populations (David et al., 2010). However, this assumption has recently been challenged through a comprehensive study of 137 non-elite mummies including Egyptian and Peruvian farming groups, Alaskan hunter-gatherers and a pre-Columbian population from the South-Western United States with a subsistence based on farming and foraging. Evidence for atherosclerosis was found in 34% of the analysed mummies and was present in all four groups (Thompson et al., 2013). Aside from dietary habits, chronic infectious diseases, particularly those of the lungs were identified as the main risk factor for atherosclerosis shared by all four populations. The findings further raise questions about a certain basic genetic predisposition present in some individuals or populations (Thompson et al., 2013).

In contrast to mummified remains, evidence from skeletal remains is only very rarely reported in the palaeopathological literature (Subirana-Domènech et al., 2012) even though one should expect calcified atheromatous plaques to survive burial (Aufderheide and Rodríguez-Martín, 1998: 79). Thus, the macroscopic appearance of these plaques in skeletonised remains is still largely unknown, and may contribute to the lack of recovery during excavation. Reports of other forms of arterial calcification are almost entirely absent from the palaeopathological record. A notable exception is represented by the analysis of an Egyptian mummy where extensive calcification of the femoral, popliteal and tibial arteries was detected by CT scans and attributed to diabetes mellitus (Marx and D'Auria, 1986).

The aim of this paper is to present five skeletonised individuals with associated calcified structures, potentially representing arterial calcifications, from the archaeological site of Amara West in Northern Sudan.

Material and methods

Located on the left bank of the Nile (Fig. 2), Amara West was founded around 1300BC to serve as the administrative capital of the Egyptian province of Upper Nubia during the later phase of the New Kingdom period (1300-1070BC) (Spencer, 2012). Pharaonic control of the region was relinquished near the end of the New Kingdom, around 1070BC. While evidence for continued settlement is still sparse, new evidence from the cemeteries suggests the site continued to be occupied until c. 800BC (Binder, 2011). Previously excavated by the Egypt Exploration Society (Spencer, 1997, 2002), a new research project, led by Neal Spencer of the Department of Ancient Egypt and Sudan at the British Museum, integrates archaeological, bio- and geoarchaeological research on the settlement, associated cemeteries and the surrounding habitat in order to gain a comprehensive understanding of life and living conditions in occupied Nubia, as well as aspects of climate change and its impact on the community at Amara West (Spencer, In Press; Spencer et al., 2012). Archaeological evidence so far indicates a small community with a subsistence largely based on agriculture and live-stock. In addition, due to its function and importance, the settlement may have also housed colonial officials, traders and perhaps military personnel (Binder and Spencer, In Press; Spencer, In Press).

The human remains derive from two separate cemetery areas which were used contemporaneously for the entire time span of occupation of the site (Binder, 2011; Binder et al., 2011). Funerary ritual is quite varied, including both single and multiple burials; the majority of individuals were buried in large underground chamber tombs. The assemblage of articulated skeletal human remains so far comprises 36 New Kingdom and 144 post-New Kingdom individuals (see Table 2). Children are almost absent from the New Kingdom group from the post-New Kingdom group. This is likely to be explained by different funerary customs for infants and children, as has also been observed in other Ancient Egyptian and Nubian cemeteries (Buzon, 2006; Zillhardt, 2009). Amongst the adult individuals, women were slightly over-represented at 46.1% against 35.9% of male individuals, even though this may be biased by the percentage of individuals (18.0%) where sexual dimorphic characteristics were not conclusive or not preserved. Palaeopathological analysis generally suggests challenging environmental conditions acting upon the population of Amara West, manifested through low stature, high levels of dental disease, respiratory infections, new bone formation on the long bones, trauma and osteoarthritis (Binder and Spencer, In Press).

Excavation of the human remains was carried out by trained bioarchaeologists. Analysis of human remains was carried out at the Institute for Bioarchaeology Laboratory of the British Museum using standard bioarchaeological methods, as recommended by Buikstra & Ubelaker (1994) and the British Association for Biological Anthropology and Osteoarchaeology (BABAO) (Brickley and McKinley, 2004). Sex was determined based on morphological features of the pelvis and skull (Bruzek, 2002; Buikstra and Ubelaker, 1994). Age estimation was based on degeneration of the pubic symphysis (Brooks and Suchey, 1990) and auricular surface (Lovejoy et al., 1985). Pathological conditions were assessed macroscopically and through the use of a hand lens (10x magnification) (Ortner, 2003). The calcifications were assigned separate skeleton sample numbers (SS). They were photographed, and SS68 and SS69b were further examined through scanning electron microscopy (Hitachi S-3700N variable pressure, SEM) in order to characterise their surfaces. Material analysis was carried out as part of the SEM examination using Energy-Dispersive X-Ray Spectroscopy (EDS). All structures were further analysed through plain film radiography (Portable GE Medical MPX X-ray unit) and processed digitally using a Kodak Point-of-Care CR120 system. 3D images of SS68 and SS69b were produced using a 3D surface scanner (NextEngine 3D Laser Scanner).

Results (Table 3)

The findings for each skeleton identified with calcified structures are now described.

New Kingdom burials (1300-1070BC)

Sk244-4

Within the heavily fragmented chest cavity of a middle aged adult male individual one large calcified plaque was observed. The plaque is very hard, yellow coloured and of irregular, oval shape (l: 23.0mm t: 0.9mm.) (Fig. 3A). In cross-section, the plaque is clearly of semi-circular outline (**Error! Reference source not found.**B). SEM characterisation shows a very dense, homogenous substance (Fig. 4). EDS was carried out to chemically characterise the material, and confirmed a calcified nature. Radiography revealed a dense internal structure. Digital microscopic imaging (30x) of the surface similarly shows a smooth, homogenous texture. Additional pathologies observed in the individual include remodelled new bone formation on the visceral side of five right ribs (chronic lung disease). In addition there is evidence for moderate periodontal disease.

Sk244-6

Associated with a middle adult male from a non-elite context, five calcified plaques of different sizes were recovered along the lower cervical and upper thoracic spine in the area between C6 and Th2 (Fig. 5) and within the chest cavity. Due to re-use and looting some post-depositional disarticulation in the thorax area occurred, even though the elements were still largely in an anatomically correct position. Therefore, minor displacement of the calcifications is possible. The calcifications have a hard texture and are yellow in colour. The largest (SS69a, Fig. 6) has an irregular elongated outline, 17.4x10mm in length and width, and a thickness of 1.3mm. High magnification (35x) again shows an homogenous structure with smooth edges. The smaller structures recovered from the vicinity of the cervical and upper thoracic spine range in size from between 9.0x12mm (SS69b-e, Fig. 6A and 6B) and 4.4x5.0mm, with an average thickness of 0.6mm. All structures are again semi-circular in cross-section (Fig. 6B). Further pathological changes in this individual include evidence for chronic disease of the lungs, as indicated by remodelled new bone formation on the visceral surfaces of the shafts of three right and four left ribs.

Post-New Kingdom burials (1070-800 BC)

Sk243-3

The middle adult female from a post-New Kingdom non-elite tomb featured a small calcification of semi-circular cross-section (SS67: 14.6 x 8.9mm; 1.1mm in thickness) in upper thoracic area to the right of the spine (Fig. 7and 8). As the chamber was not backfilled after the burial and the individual was not entirely supine, it seems likely that the calcified structure may have been slightly displaced. In addition to a small healed depression fracture on the frontal bone, remodelled new bone formation was observed on the visceral aspect of the shafts of five left and six right ribs. Moderate to severe periodontal disease was also observed.

Sk237

Eight round to oval shaped hard, yellow calcifications of varying sizes were recovered from the abdominal area on top of the lumbar vertebrae of a middle adult female individual from a post-New Kingdom, non-elite tomb (Fig. 9). The individual was buried in a supine position, and the calcified structures were orientated parallel to the body axis. The two largest examples are of elongated oval shape (SS37a 18.7mm x 7.3mm, thickness 1.2mm; SS37b 15.5mm x 5.9mm, thickness: 1.0mm) with a curved cross section. The smaller fragments range in size from between 8.6x7.7mm and 6.0x5.0mm (Fig. 10). Additional pathological evidence again includes remodelled new bone formation on the visceral side of four left ribs in the shaft area, fractures of the 4th and 5th lumbar vertebrae, sternum and iliac blade, as well as moderate to severe periodontal disease.

Sk305-4

This female middle adult individual was recovered from a double-chambered tomb in the elite cemetery of Amara West. A "string" of whitish coloured, round calcifications was found running along the medial side of the right femur from the area of the femoral neck inferiorly over a length of 25cms (Fig. 11). The remaining fragments are also of whitish colour, up to 14mm long. The walls of the structures reach a thickness of up to 0.4-0.5mm (Fig. 12). In contrast to the other calcifications, the full circumference was calcified and preserved intact through most of the length of the lesion, with a diameter of 4-5mm. Remodelled new bone formation on the shafts of three right and three left middle ribs was also observed. In addition, the individual suffered from extensive dental disease with the majority of teeth being lost ante-mortem.

Discussion

The calcifications observed in the skeletons from Amara West were identified as calcified atheromata based on the anatomical positions of the calcified structures (Fig. 13), and their morphological appearance and comparison with published clinical studies and analysis of intact mummies (e.g. Allam et al., 2009; Thompson et al., 2013; Towler, 2008). The calcifications recovered in the upper chest area of Sk244-6 (SS69a), Sk244-4 (SS68a) and Sk243-3 (SS67) are round and semi-cylindrical in shape and cross section suggesting they originated from a cylindrical structure with an estimated diameter of c. 2-2.5mm. This conforms to the average diameter of the *aorta descendens* or *aorta abdominalis suprarenalis* (Kahraman et al., 2006). The curvature of the second larger calcification recovered with Sk244-6 (SS69b) is consistent with a diameter of 0.8-0.9cm (Fig. 14). Based on the location of SS69b and some smaller calcified plaques along C6-Th2, as well as their width, the data are consistent with published dimensions in living people (Engelhorn et al., 2006), and an origin in the subclavian artery appears plausible. Calcifications in these vessels have also been reported in Egyptian mummies (Sandison, 1962).

Based on the anatomical position of the calcifications in the lumbar area of Sk237, they are consistent with the location of the iliac artery (Fig. 9). Furthermore, based on the curvature observed in the larger examples, they would have originated from a structure with a cylindrical shape of 0.5 – 0.7mm. This falls well within the standard range of the common iliac artery (Malnar et al., 2010). They further conform to CT-findings in mummified human remains diagnosed with atheromatous plaques in the iliac artery (Allam et al., 2009).

The calcifications associated with Sk305-4 are consistent with the anatomical location of the common femoral artery (Fig. 11). The observed diameter of 0.4-0.5 mms conforms to average values of intact femoral arteries vessels in the living (Sandgren et al., 1999). Therefore, it seems plausible to identify them as calcifications of the femoral artery. In contrast to the other structures, the entire circumference was calcified. Circumferential calcification is a common feature of medial arterial calcification, and the absence of narrowing of the arterial lumen distinguishes it from atherosclerotic calcification (Lehto et al., 1996; Towler, 2008). Medial calcification is particularly common in the uterine, femoral and tibial arteries (Sinha et al., 2008), thus appearing to be a likely cause for the calcifications observed in Sk305-4.

Calcification of soft tissue can occur secondary to a large number of conditions in different parts of the body, for example in the thoracic cavity, and intracranially, in joints, and in the pelvic and abdominal cavities (Baud and Kramar, 1991; Steinbock, 1989). Histomorphologically, these structures are all very similar to each other, and generally similar to bone due to the same underlying pathophysiological mechanisms (Demer and Tintut, 2008; Doherty et al., 2003). Calcifications are only occasionally reported in the palaeopathological record (e.g. Baud and Kramar, 1991; Komar and Buikstra, 2003; Perry et al., 2008; Steinbock, 1989). One of the more commonly reported types of calcifications are those of pleural tissue (Fig. 15) secondary to inflammatory conditions of the lung (e.g. Baud and Kramar, 1991; Donoghue et al., 1998; Roberts and Buikstra, 2003) and could be considered as a potential differential diagnosis for the calcified structures recovered from the chest area. However, as these originate from inflamed pleural tissue, they are characterised by a relatively flat surface and an irregular shape (Light, 2012). Nevertheless, identifying the exact origin of a calcification in association with skeletal human remains is often very difficult due to their non-specific nature. A more secure diagnosis could be based on anatomical position and macroscopic appearance or through histological examination, even though neither necessarily leads to a conclusive answer.

The calcifications observed in the individuals from Amara West do not conform to the appearance known for any other condition. Their semi-cylindrical shape and their relative anatomical positions with the skeletons Sk244-4, Sk244-6, Sk237 and Sk243-3 rather argues for an identification of calcified arterial plaques caused by atherosclerosis. Even though often considered a modern disease, a large number of risk factors leading to advanced atherosclerosis were similarly present in the past. One of the major environmental risk factors is linked to dietary composition including high levels of fat, sugar and protein. Archaeozoological and archaeobotanical evidence indicates that people living at Amara West certainly had access to a diet that included meat, and sugar through dates (Ryan et al., 2012), and thus dietary factors potentially could have contributed to the risk of developing atherosclerosis.

The co-occurrence of new bone formation on the visceral side of the ribs providing evidence of chronic infection of the lungs in all five individuals with calcified atheromatous plaques is also notable. Today, tobacco smoking is seen as the second most important cause of atherosclerosis (Lusis, 2000). Recent clinical studies indicate that habitual exposure to wood smoke can have detrimental effects on health very similar to tobacco smoking (Danielsen et al., 2011). The practice of using hearths and cylindrical clay-lined ovens for cooking, fuelled by wood, charcoal and dung, but also manufacturing of ceramics or metalwork within houses and courtyards, is well evidenced at Amara West, including within small roofed spaces with little ventilation (Spencer forthcoming). Moreover, a link between atherosclerosis and bacterial pneumonia has also been established recently (Rosenfeld and Campbell, 2011). The cause of new bone formation on the visceral surfaces of ribs has been explored by many authors, with varying diagnoses being suggested. Conclusions indicate that a range of chronic pulmonary diseases could be represented, including tuberculosis (TB), lung cancer, chronic bronchitis and pneumonia (Lambert, 2002; Roberts, 1999; Roberts et al., 1994; Santos and Roberts, 2006). The bone changes are not pathognomonic for any one condition. At Amara West, new bone formation on the visceral side of the ribs was found in 60.0% (15/25) of New Kingdom and 54.2% (39/72) of post-New Kingdom individuals, indicating that chronic respiratory disease was a common health problem. Further differential diagnostic features allowing for a more precise diagnosis were not observed. Unambiguous evidence of TB is absent at Amara West, even though its presence in the Nile valley from at least *c.* 3000BC is well established (Buikstra et al., 1993). Therefore, given the geographical context and living environment, with close proximity to animals, the disease may have been present at Amara West too. Pleural calcification can accompany a range of lung diseases, although the appearance of the structures found in the thoracic cavities of skeletons Sk244-4, Sk244-6 and Sk243-3 are different to previously identified calcified pleura, as discussed above. The contribution of dental disease also has to be taken into account. Poor oral health with high degrees of caries, periapical lesion formation and ante-mortem tooth loss is a general characteristic of the Amara West population. Periodontal disease affected 84.6% of the New Kingdom (11/13 individuals) and 85.1% of the post-New Kingdom group (40/43 individuals). All individuals showing evidence for arterial calcification also displayed signs of moderate or severe periodontal disease.

A genetic background also has to be taken into consideration. Recent findings in mummies have led researchers to argue for a genetic background as the main reason for the development of atherosclerosis (Thompson et al., 2013). It has been suggested that ancient Egyptians had a genetic predisposition for the formation of atherosclerosis that accounts for the high prevalence of advanced atherosclerosis in Egyptian mummies (Zink et al., 2011). Direct evidence for a genetic predisposition in past human populations has so far only been identified in the "Iceman" (Keller et al., 2012). Two of the individuals from Amara West with calcified plaques (Sk244-4, Sk244-6) were buried next to each other within the same grave. Even though it is currently impossible to prove familial ties with any certainty, this finding is nevertheless intriguing. While a genetic predisposition to atherosclerosis in Nile valley populations is certainly within reason, much more research into the genetics or epidemiology of atherosclerosis in African countries and in other regions of the world in general, is needed in order to argue for or against this claim.

The calcified structures observed in Sk305-4 rather fit the criteria for medial arterial calcification particularly since, in contrast to the other observed calcifications, the entire circumference is calcified. This type of arterial calcification can be caused by diabetes, end-stage chronic kidney disease or old age (Towler, 2008). However, extensive, "railroad-track like" calcifications in the peripheral arteries of the lower limbs are a common secondary phenomenon in diabetes mellitus (Lehto et al., 1996; Towler, 2008). Due to the fact that this disease does not cause any direct skeletal changes, the antiquity and paleoepidemiology of this disease are unknown (Aufderheide and Rodríguez-Martín, 1998: 343). Literary sources from Egyptian medical papyri have tentatively been interpreted as evidence for diabetes (Loriaux, 2006) and soft tissue changes ascribed to the disease have been noted in an Egyptian mummy (Marx and D'Auria, 1986). Therefore, the possibility that the individual may have suffered from diabetes is not without reason.

Conclusions

Even though direct evidence of atherosclerosis and other forms of arterial calcification is almost absent in association with human skeletal remains, the lack of evidence cannot be seen as evidence of absence in past human populations. To a large degree it may simply be explained by poor recovery strategies in the field and failure to identify calcified structures. The examples from Amara West contribute further to the knowledge of the morphological appearance of calcified arterial plaques and could in future aid to increase the dataset of palaeopathological evidence. Careful excavation of human remains, including sieving (Mays et al., 2012), together with improving the knowledge of excavators about what could be expected, and where, could significantly increase the number of recovered examples. Furthermore, highlighting the significance of arterial calcifications in providing an important contribution to the understanding of disease and living conditions in the past may lead to increasing awareness.

While it will never be possible to gain a full picture of the scale of cardiovascular disease and atherosclerosis in the past, due to lack of preservation and detection of calcified plaques in association with human skeletal remains, it could nevertheless add another important piece of information about morbidity in the past. Archaeological, historical and bioarchaeological evidence suggests that several risk factors that lead to atherosclerosis in modern human populations were also present in the past. Well recorded palaeopathological evidence integrated into a broader archaeological, including socio-cultural, context could significantly contribute to gaining a deeper understanding of the factors leading to atherosclerosis from a pre-modern perspective.

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Captions

Fig. 1: Types of arterial calcification (modified from Towler, 2008)
Fig. 2: Map of Sudan showing location of Amara West
Fig. 3: Calcified structure SS68 (Sk244-4) A:view from both sides, B: SS68 in cross-section (arterial wall dimension based on data in Kahraman et al., 2006)
Fig. 4: SEM image of the surface of SS68
Fig. 5: In-situ position of SS69b and SS69c alongside spine of Sk244-6
Fig. 6: Calcified structures recovered with Sk244-4: A;SS69a-e, B: cross section of SS69b (arterial wall dimension based on data in Engelhorn et al., 2006)
Fig. 7: Location of calcified structure in the chest area of Sk243-3
Fig. 8: Calcified structure, possibly representing calcified atheroma of the aorta, view from both sides (SS67, Sk243-3)
Fig. 9: Possible calcified atheromatous plaques in the abdominal area of Sk237
Fig. 10: Calcified structures associated with Sk237 (SS37a-h)
Fig. 11: Calcified femoral artery in situ (Sk305-4)
Fig. 12: Calcified femoral artery associated with Sk305-4
Fig. 13: Anatomical location of calcified structures recovered at Amara West in relationship to the main arterial system
Fig. 14: 3D scan of SS69b (Drawing: M. Dalton).
Fig. 15: Example of calcified pleura from a historical medical collection

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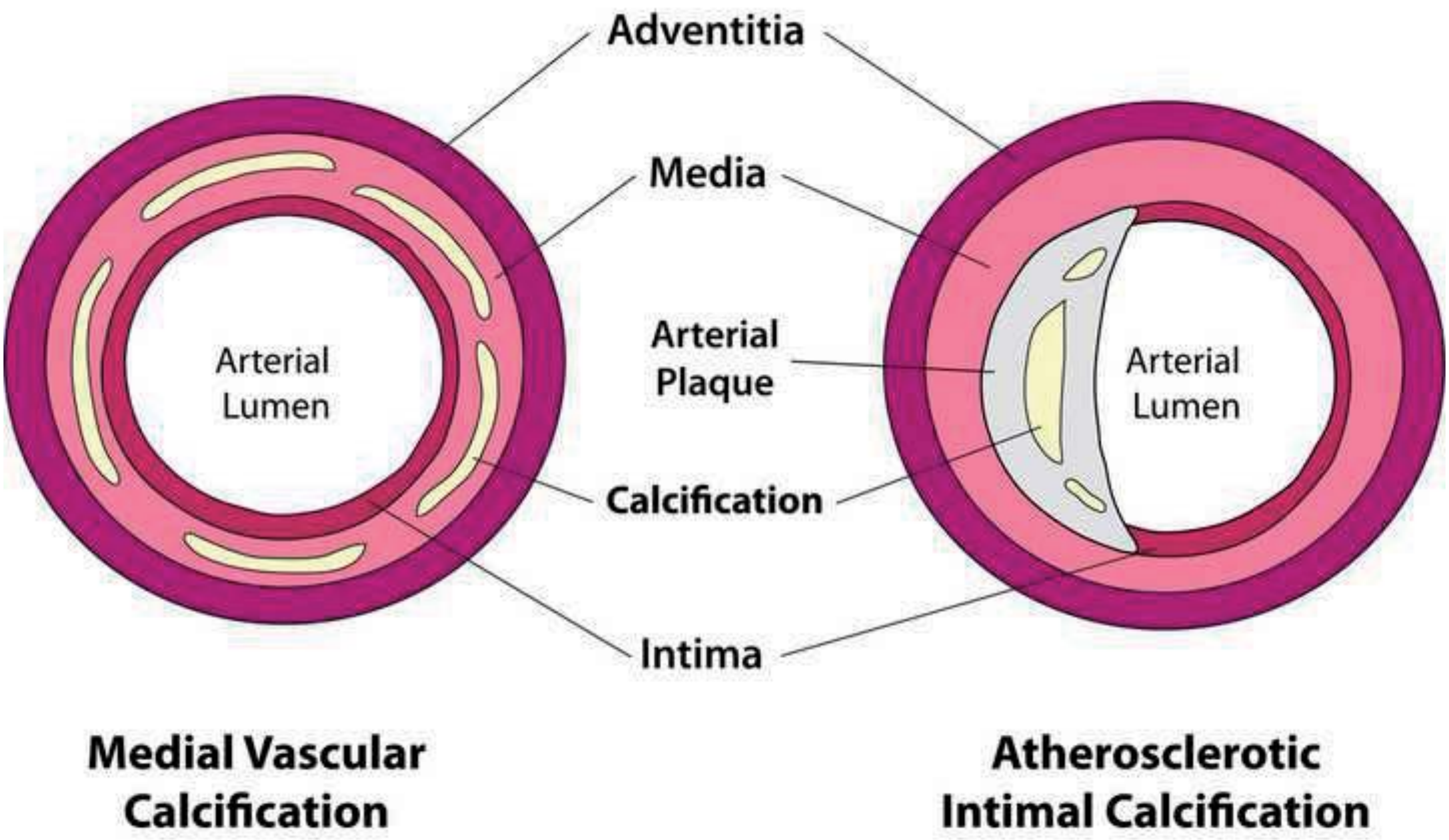

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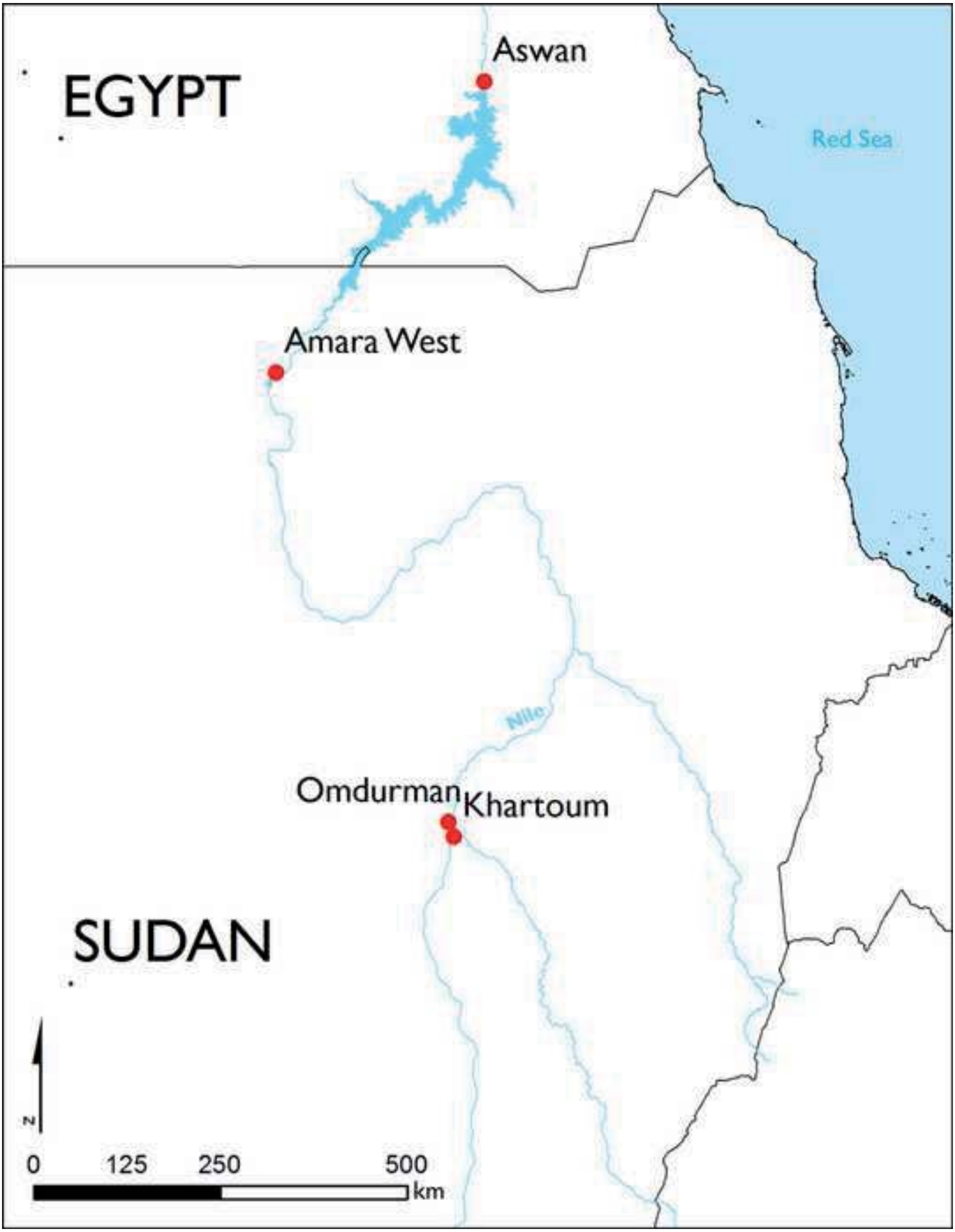

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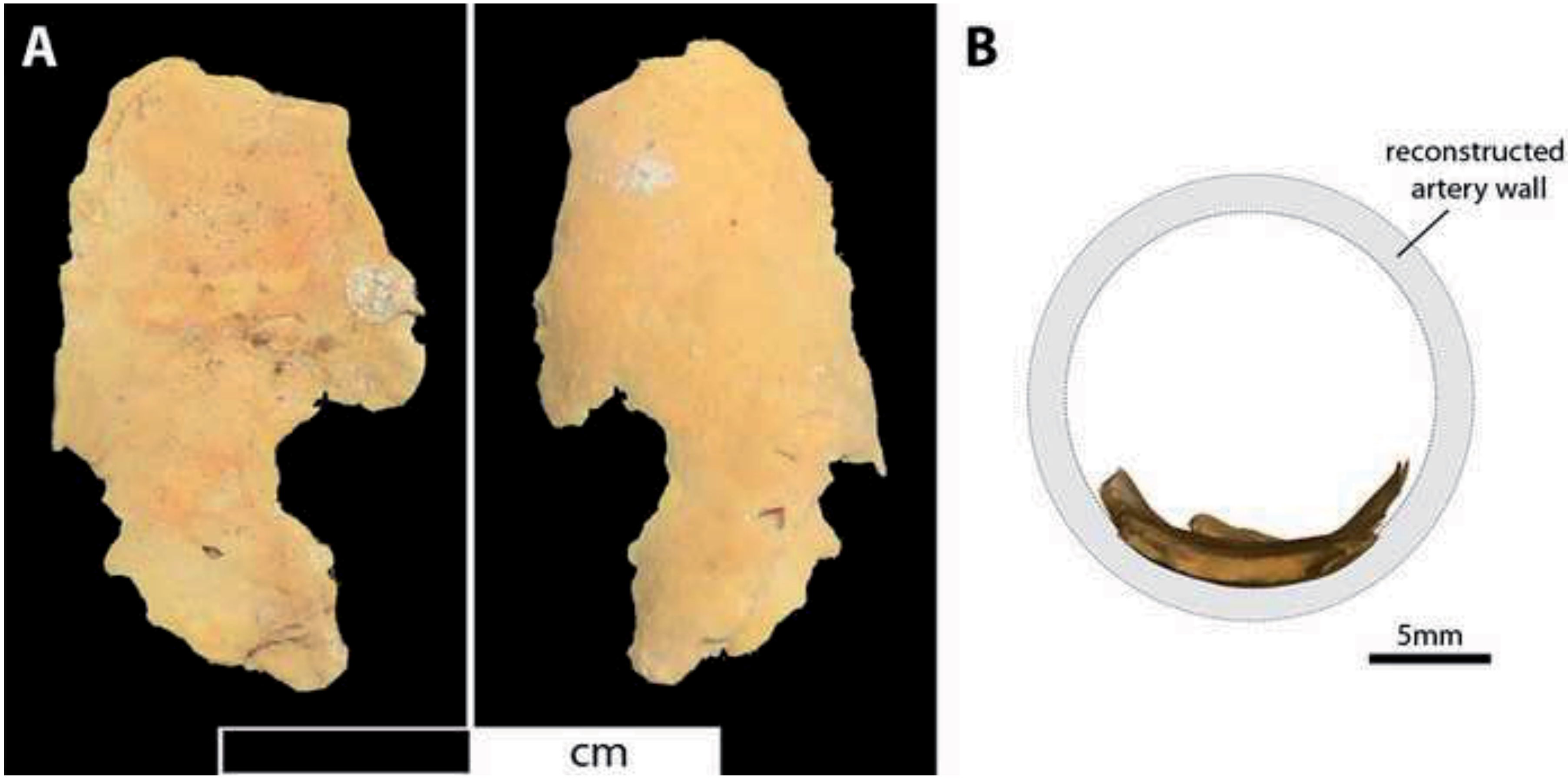

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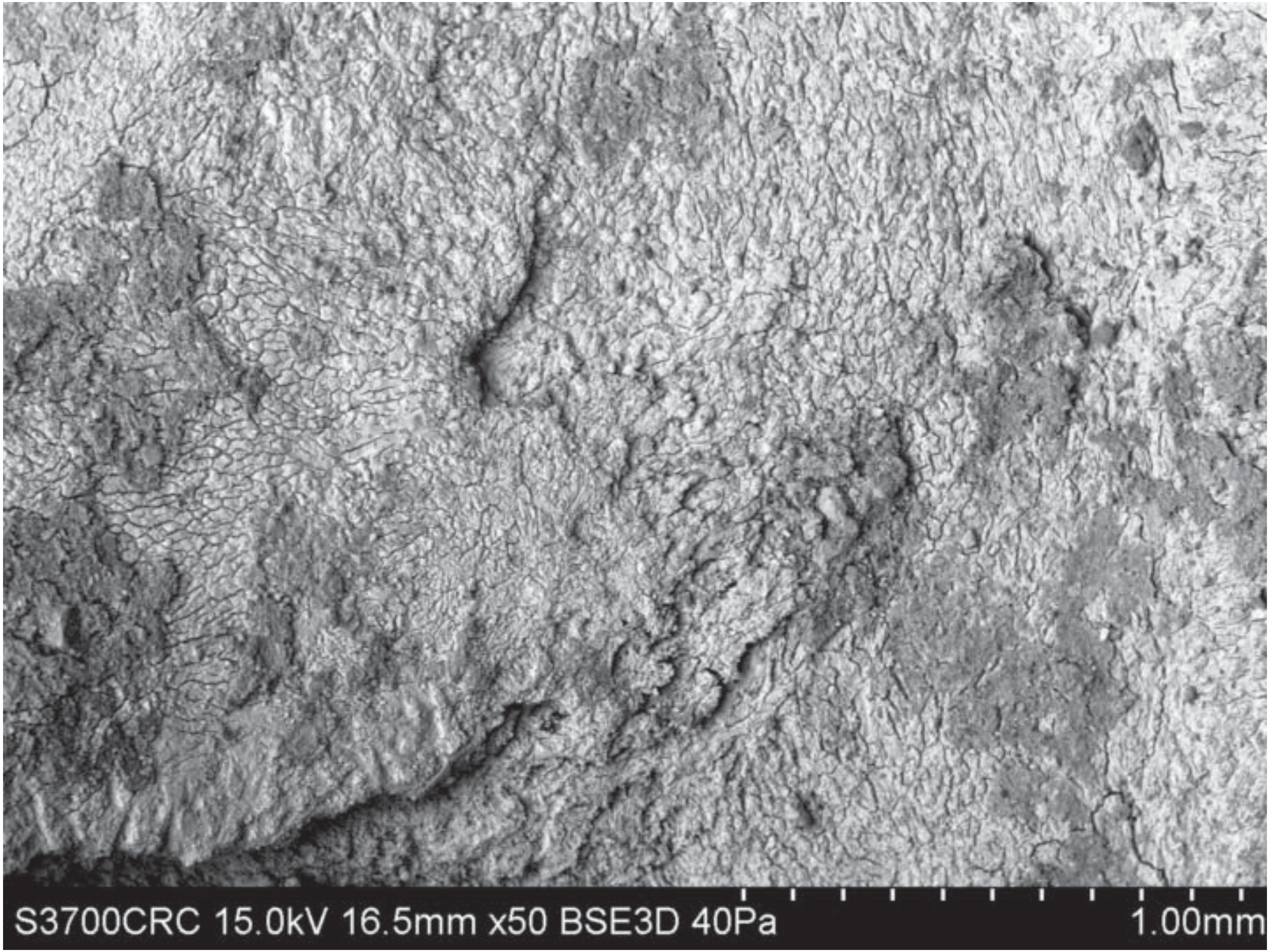

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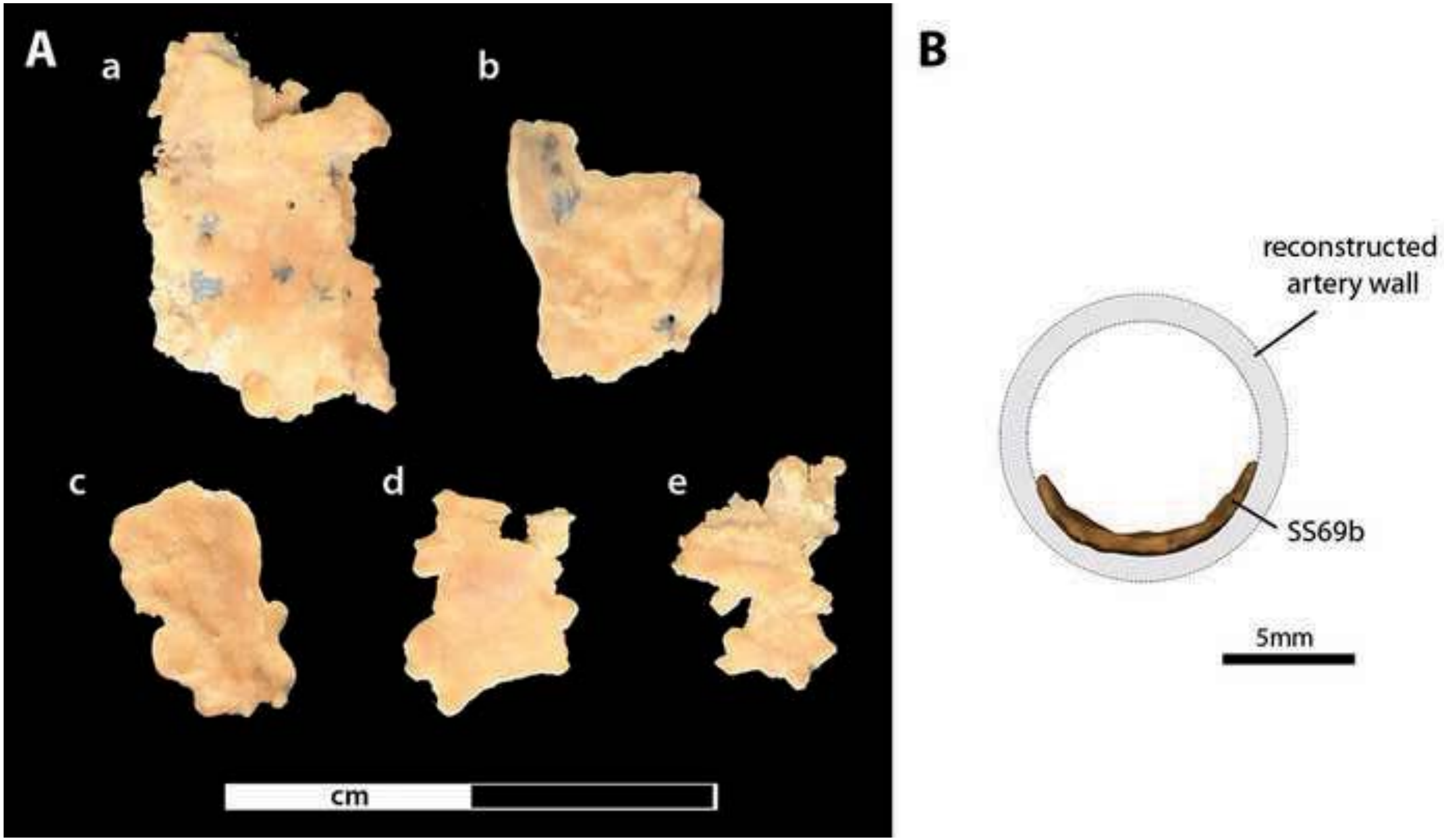

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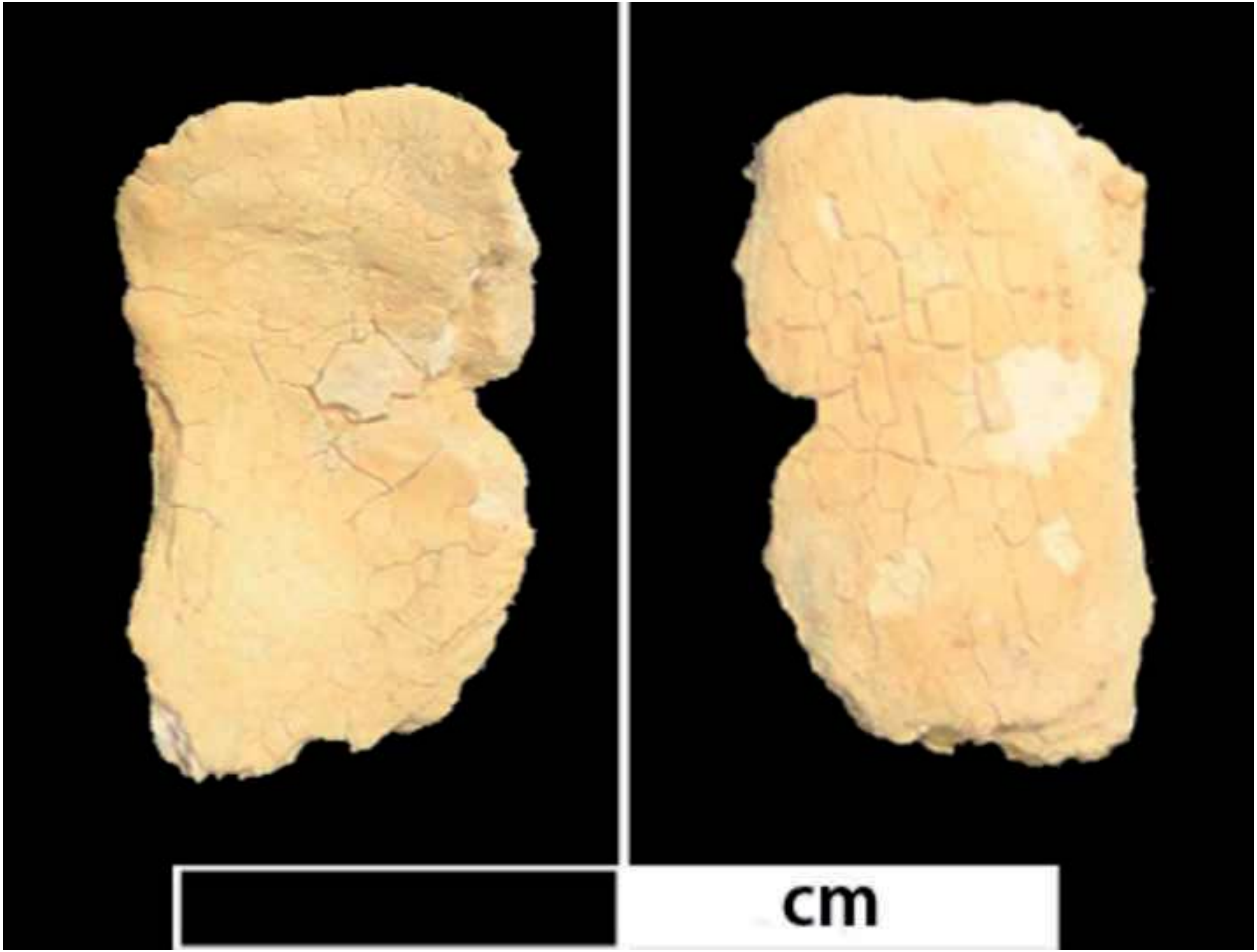

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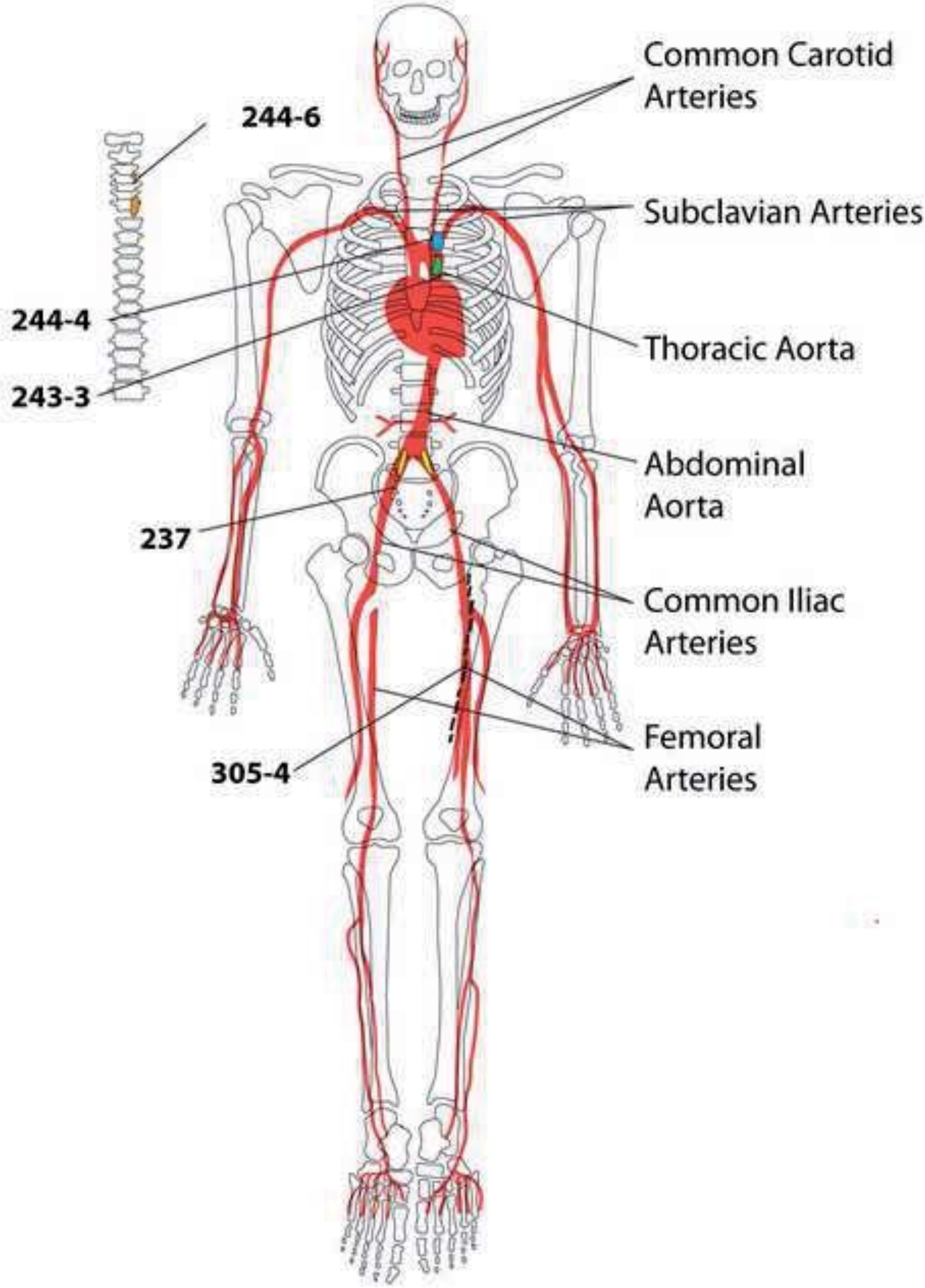

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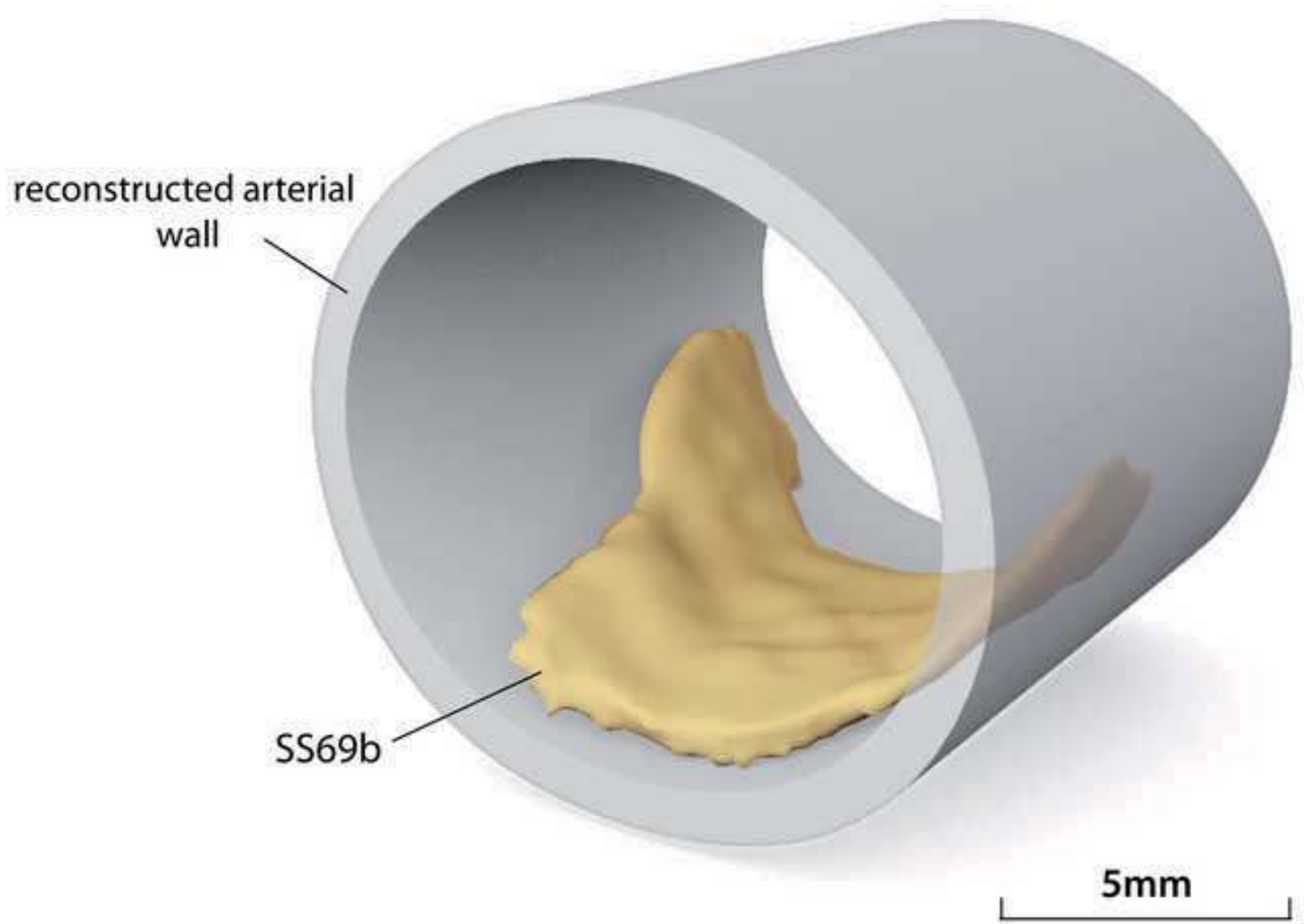

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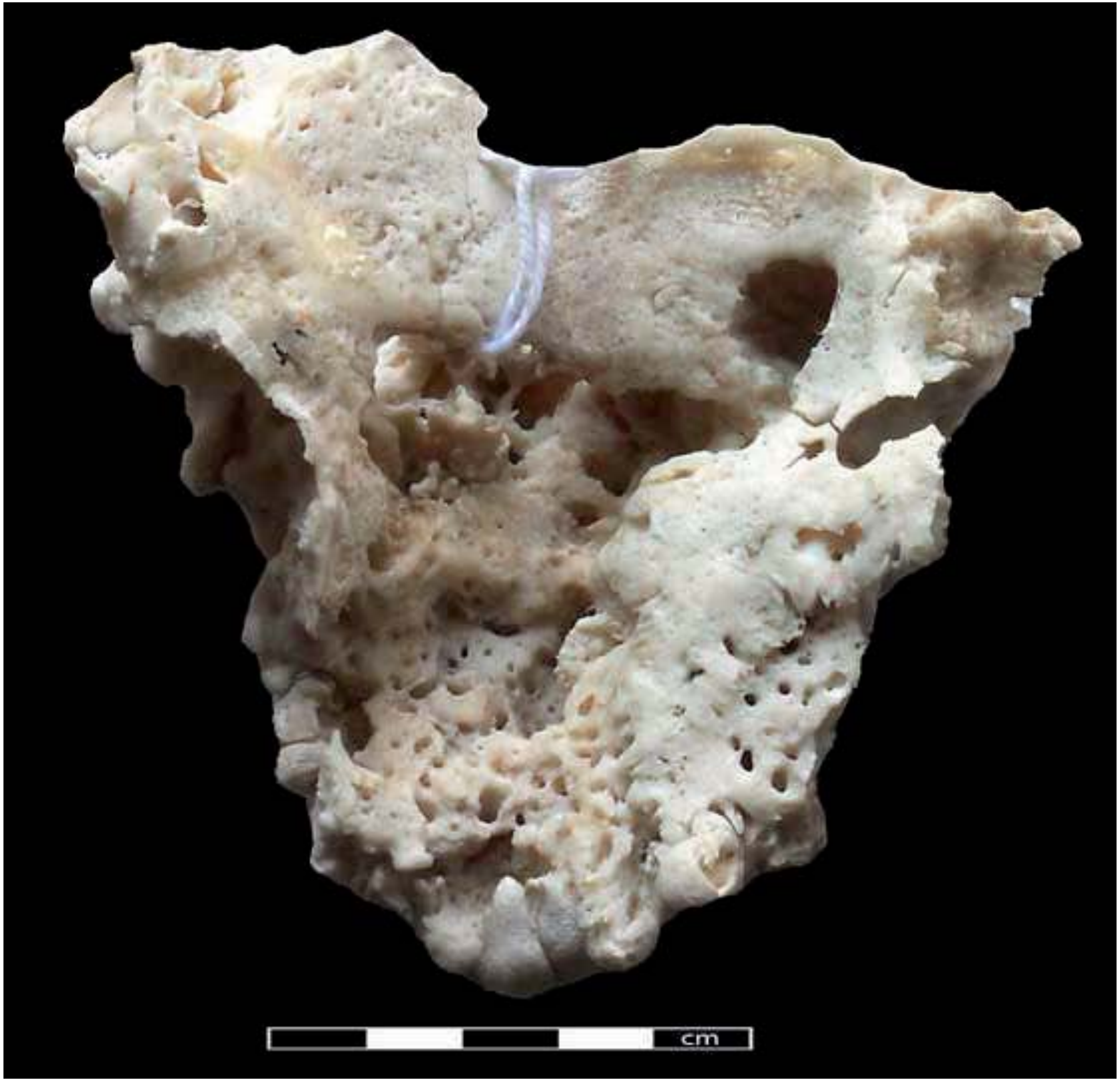

Table 1

Type of calcification	**Location**	**Associated conditions**
Atherosclerotic calcification	Intima	Atherosclerosis, hyperlipidemia, hypertension, inflammation
Medial vascular calcification	Tunica media	Type 2 diabetes mellitus, end-stage renal disease, advanced age,
Aortic valve calcification	Aortic face of the leaflets	Hyperlipidemia; congenital bicuspid valve; rheumatic heart disease
Vascular calciphylaxis (calcific uremic arteriolopathy)	Microvessels	Type II diabetes, Hyperthyroidism, End-stage renal disease, warfarin(?)

Table 1: Types of vascular calcification (Demer and Tintut, 2008; Johnson et al., 2006; Towler, 2008)

Table 2

	New Kingdom		Post-New Kingdom		Total	
	n	%	n	%	n	%
<1	0	-	2	1.4	2	1.1
0-1 yrs	0	-	9	6.3	9	5.0
1-5 yrs	0	-	17	11.8	17	9.4
6-12 yrs	1	2.8	13	9.0	14	7.8
13-20 yrs	0	-	10	6.9	10	5.6
21-35 yrs	14	38.9	39	27.1	53	29.4
36-50 yrs	9	25.0	25	17.4	34	18.9
>50 yrs	2	5.6	9	6.3	11	6.1
Adult indet	10	27.8	20	13.9	30	16.7
Total	**36**		**144**		**180**	

Table 2: Demographic profile of the Amara West population (n=number of individuals)

Table 3

Skeleton number	Sex	Age	Location of calcified tissue	Dimensions (in mm)	Likely blood vessel involved	Additional pathologies	Analytical methods
New Kingdom period (1300-1070BC)							
Sk244-4	M	36-50 yrs	chest	SS68: 2.3x1.5, t: 0.9	Thoracic aorta	New bone formation on the visceral side of the ribs, fractures of Th4, 5 and ribs, periodontal disease	Radiography SEM (**Error! Reference source not found.**) EDS Digital microscopy
Sk244-6	M	36-50 yrs	chest	SS69a: 17.4x10.0, t: 1.3	Thoracic aorta	New bone formation on visceral side of ribs, periodontal disease	Radiography SEM Digital microscopy
				SS69b: 9.0x11.0, t: 0,6	Subclavian artery		Radiography
Post-New Kingdom period (1070-800BC)							
Sk243-3	F	36-50 yrs	chest	SS67: 14.6 x 8.9, t: 1.1	Aorta	New bone formation on visceral side of the ribs, depression fracture on the frontal bone, periodontal disease	Radiography
Sk237	F	36-50 yrs	abdominal area (L3-S1)	SS37a 18.7 x 7.3, t 1.2 SS37b 15. x 5.9, t: 1.0	Iliac artery	New bone formation on visceral side of the ribs, fractures of the left ilium, L4, L5, sternum, ribs and humerus, periodontal disease	Radiography
Sk305-4	F	36-50 yrs	left femur	SS5: l: 2-14, t: 0.4-0.5, d: 0.4-0.5	Femoral artery	New bone formation on tibiae and visceral side of ribs, fracture Th3, periodontal disease	Radiography

Table 3: Calcifications recovered at Amara West (dimensions given as length x breath, t=thickness, d=diameter)