



## The Presence of Unusual Foramina in the Femoral Intercondylar and Trochanteric Fossae on Skeletonized Individuals from Tanzania

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### Abstract

According to the standard nomenclature, skeletal land marks are all identified by their given names in the world of human skeletal anatomy. But it is also clear that the discovery of new anatomical features continues because human beings have so many anatomical variations caused by genetics, functions, geographical adaptations, and diseases; they are all described and given names. Therefore, an anatomical feature that is rare and undescribed is certainly a discovery and it deserves to be recorded. The discovery of anatomical features on the epiphyseal ends of the femur reported here adds two new names, namely the **intercondylar fossa foramen** and the **trochanteric fossa foramen** in that long list of human skeletal anatomical structures. The intercondylar fossa foramen is an outlet found in the intercondylar fossa of the distal femur. It passes through the diaphysis of the femur and emerges in the trochanteric fossa of the femoral neck; and vice versa is the trochanteric fossa foramen. This feature was first observed on one skeletonized archaeological individual and later on five cadaveric remains. There seems to be a presence of this anatomical variation within the Tanzanian population. Only a small percentage of people have this unusual anatomical feature of the intercondylar fossa foramen, which might be a developmental ossification failure or a particular genetic-based trait. These features were assessed in the laboratory of the Anatomy Department at the Muhimbili University of Health and Allied Sciences (MUHAS) using a Dino-lite digital microscope and X-ray images.

**Keywords:** Anatomical structures, Femoral foramen, Intercondylar fossa, Trochanteric fossa.

### Introduction

Apart from the nutrient foramina on long bones (Longia et al. 1980, Mysorekar 1967), epiphyseal end foramen rarely occur and have been observed only on humeral distal ends as septal foramen. Hrdlička (1932) detailed the occurrence of septal foramen among white American cadaveric population, particularly in females. Ashley-Montagu (1950) pointed out that septal foramen were developmental (endochondral) ossification failures on distal epiphyseal ends of the humerus. Such anatomical features have so

far not been reported on epiphyseal ends of the femur anywhere in the anatomical literature. The presence of intercondylar fossa foramen in a skeletonized population in Tanzania was first observed in 2018 by a forensic anthropologist and a police officer from the Forensic Bureau of the Tanzania Police Force during an archaeological excavation he was supervising as part of the University of Dodoma forensic anthropology field school. The bones of an adult male individual (age estimated between 42 and 47 years old) with the foramen in the femoral

intercondylar fossa was among the skeletons of 36 individuals exhumed by the forensic anthropology field team of the University of Dodoma, which is held annually as part of the field-based practicals for students. The remains were then taken to the Forensic Field School Laboratory at the College of Natural and Mathematical Science, the University of Dodoma for further investigation. In the laboratory, the femoral bones of an adult male individual with intercondylar fossa foramen were discovered (see Figure 1) thus prompting an extensive investigation. At the time, the outlet (hole or socket) was thought to be probably the result of periosteal bone surface alteration activities, paleopathologically diagnosed and identified to be osteomyelitis in the proximal region (Bintliff 2008, Fornaciari 2018). Given the fact that most of the excavated bones were poorly preserved (highly decomposed), especially in the epiphyseal ends (distal and proximal), it was difficult to discern whether such foramen in other individual skeletal remains were also preserved (Klepinger 2005). Nevertheless, it could not be ruled out the possibilities of other individuals within the assemblage possessing that unusual skeletal feature of the intercondylar fossa foramen. Unusual enough, further examination also revealed the presence of the trochanteric fossa on the same individuals.

Accordingly, a year after the initial discovery, in 2019 a second case of the presence of intercondylar foramen was observed from the forensic investigation case that involved a criminal suspect in possession of human femoral bones belonging to a person with albinism (Bryceson 2010, Jilala et al. 2021). This bone was taken to the Forensic Bureau of the Tanzania Police Force for examination where it was found to possess both the intercondylar fossa foramen and the trochanteric fossa foramen. These two cases provided ample evidence triggering the description of these new anatomical features that were completely unknown in the standard or general skeletal biology known to many osteologists, particularly bio-archaeologists, forensic anthropologists, and paleopathologists. The

additional discovery raised more questions about the existence and the uniqueness of the morphological features of the foramina on the epiphyseal ends of the thigh bone. It was hypothesized that these features might be unique among people with albinism (Dallapiccola 2005) as a rare genetic inherited skeletal characteristic (Weedn 2007) and it was linked to the frequencies of skeletal parts, especially long bones of people with albinism illegal sold on markets at exorbitant prices (Hong et al. 2006, Mwiba 2018) to people seeking wealth and or political powers (Jilala et al. 2021). Due to the ongoing killings of people with albinism in the Great Lakes region (Bryceson 2010, Burke et al. 2014) in 2019, a comparative study of the skeletal signature on people with albinism against those with melanin for forensic identification purpose was conducted (Jilala et al. 2021). The study assumed that epigenetics, particularly sensitivity to solar radiations, cancers, and responses to albino allele, plus adaptive behaviours of people with albinism would at a molecular-level be manifested within the bones as stress-related genetic markers that contribute to morphological features that can be used for forensic investigation (Dallapiccola 2005, Jilala et al. 2021). In their study, almost complete skeletal remains of 20 individuals were analysed, 10 with melanin and the other 10 with albinism. Coincidentally, five out of the twenty individual skeletal remains possessed the foramen in the intercondylar fossa; all of them (female and male) with melanism. This demonstrated that the presence of the foramen was not at all associated with albinism. However, it further established the existence of this feature beyond reasonable doubt, that such unusual foramina do exist within the population in Tanzania and probably in eastern Africa. Therefore, a series of evidence for the presence of this foramen in the intercondylar fossa led to the preparation of this particular article reporting the presence of the unusual outlet or socket in the thigh bone (Matshes et al. 2004). In making this report more relevant and compelling further evidence were obtained from a skeletal collection of

cadaveric sample from the Anatomy Department at the Muhimbili University of Health and Allied Science, and later other samples were added from various institutions. The samples were examined to confirm the presence of the intercondylar and trochanteric fossa foramen. The foramen was found to be between 3.1 to 3.6 mm in width, the difference in measurement found in small and large openings. So far, it is not yet known exactly what is the function of the foramen. In this paper, several anatomical principles pertaining to the function of the socket are discussed, with a call to speed up the investigation process about these foramina, thus bringing significant benefits, especially in health departments. Specialists in arthrology, osteology, radiology and many other medical specialists will need to know the occurrence of this feature in some populations as an anatomical variation when providing their medical services.

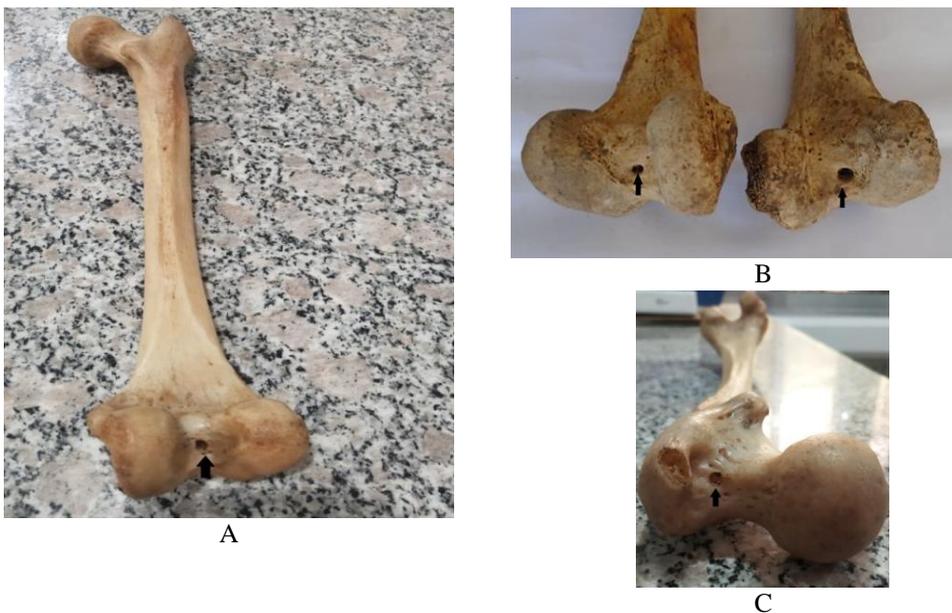
### **Materials and Methods**

A total of 104 femoral bones of the right side were randomly sampled from the skeletal collections at the various institutions in the country: 32 were obtained from the Department of Anatomy at the Muhimbili University of Health and Allied Sciences, 14 from the Department of Anatomy at the Hubert Kairuki Memorial University, 31 from the Forensic Field School Laboratory at the College of Natural and Mathematical Science, University of Dodoma and 17 from Department of Anatomy at the College Health and Allied Sciences of the University of Dodoma, 6 from Mumba Bioarchaeology Field School Laboratory Collection and other 4 from the Forensic Bureau of Tanzania

Police Force (see Table 1). The sampling assumed that the right femur of adults' individuals of any sex would be appropriate to examine the intended skeletal feature (foramen). All sampled skeletons belonged to East Africans (black Africans), specifically Tanzanians (Feldesman and Fountain 1996, Yurka 2014). Aside from careful visual bone surface observations, macro- and microscopic techniques as well as X-ray images were used to detect these unusual foramina (Matshes et al. 2004, Bintliff 2008, Bristow et al. 2011). First, the holes were easily detected because they are visible to the naked eye (Klepinger 2005, Dallapiccola 2005). Thereafter, the Dino-Lite Digital Microscope AD4113T-12V was used in the morphology study of the socket (Saha et al. 2017, Fornaciari 2018, Chakravarthi et al. 2018). Metric and non-metric assessment methods were also employed to determine the depth and width of the socket (fossae foramina) in millimetres (Chowdhury et al. 2013, Dogan et al. 2014, Edwards et al. 2018, Varacallo 2018, Ranaweera et al. 2020). To determine the depth of the foramen a very thin and very smooth probe was inserted into the outlet from the femoral neck to determine its passageway and see if it could protrude into the other side to the intercondylar fossa. Additionally, X-ray images (Phillips Medical System, Digital Diagnostic C50, PMS\_ELEVA\_37.0 Software) were taken to accurately trace the passageway of the foramina, particularly their inlet and outlets. The images were also used in determining whether the passageway was caused by a mechanical object that would have been insert in after the femora were perforated.

**Table 1:** Present material sources and statistical presence of fossae foramina

Institution name	Number of femoral bones obtained	Source	Preservation condition	Sex	Age range	Presence of fossae foramina
Muhimbili University of Health and Allied Sciences	32	Cadaveric	Good	9 female	26–49	3
				23 male	23–45	9
Department of Anatomy at the Hubert Kairuki Memorial University	14	Cadaveric	Good	3 female	25–33	0
				11 male	24–45	1
Forensic Field School Laboratory at the College of Natural and Mathematical Science, University of Dodoma	31	Archaeological	Well preserved	10 female	Adult	0
				21 male	Adult	1
Department of Anatomy at the College Health and Allied Sciences of the University of Dodoma	17	Cadaveric	Good	8 female	28–47	2
				9 male	21–55	1
Mumba Bioarchaeology Field School Laboratory Collection	6	Archaeological	Well preserved	4 female	Adult	1
				2 male	Adult	0
Forensic Bureau of Tanzania Police Force	4	Criminal possession of human remains	Well preserved	4 male	Adult	1



**Figure 1:** (A) and (C) are distal and proximal ends of the femoral bone examined by the Forensic Bureau in 2019, (B) are the distal ends of the left and right femur unearthed 2018 by the forensic team of the University of Dodoma.



**Figure 2:** Mechanically perforated bones: (A) Femoral head, and; (B) distal condyles with a medio-lateral passageway.

### **Taphonomic observations**

The macro and microscopic bone surface modification assessment was conducted in great detail and found beyond any doubt that there is unusual socket located in the intercondylar and trochanteric fossa (Feneis and Dauber 2000, Sendemir 2017). Comparison with intentionally modified femoral bones (mechanically drilled) was carried out in the laboratory and images were sent to the University of Colorado Denver's Comparative Human Osteology Teaching Lab for verification. The differences between mechanically perforated bones (as part of assembling articulated teaching skeletons) with the collection studied is the nature of the areas where the bones are perforated; on intentional and mechanically modified bones, usually the holes are on the femoral head (with a medio-lateral passageway) and on the distal end, they are on usually perforated on the media and lateral condyles of the femur (Figure 2A & B). Furthermore, mechanical perforations on wet or dry bones usually destroy the periosteum, thus leaving some scarring features. The holes or outlets are usually uniform in diameters (based on the tool used to perforate them), contra to the foramen presented in this study. This assessment has yielded the best results in science of human skeletal anatomy which opens the door to important questions that require solid answers especially in explaining

the functions of these intercondylar and trochanteric fossa foramina.

Perhaps the most basic question that can arise here is how we have been able to identify these outlets as embryological or anatomical features and not artificial. Indeed the characteristics of the features themselves are well expressed and covered with periosteal membrane. The outlets are completely natural anatomical structures and do not resemble any artificial holes in that cortical bones are not mechanically perforated or damaged. The holes are smooth from the perimeter of the socket on the outside to the inside of the walls and their edges. It is clear that the roundedness and the walls are covered with a layer of endosteum which makes the whole canal laminated and well protected. These openings are not uniform in their width due to bone formation or osteogenesis and not artificial perforations. If they were artificial they would be the same width depending on the tip and width of the device used for drilling. Also the artificial openings on the bones tend to have circles and walls that are rough due to the perforation process. Even if they are polished they still cannot be as smooth as the ones observed here for these individuals. Artificial holes contain the signatures of detachments that were demolished during drilling and not covered by the endosteum layer which is by nature the layer that covers the bone canals (see Table 2).

**Table 2:** Present images and summary of differences between anatomical and mechanically perforated bones

Mechanically perforated signatures	Embriological or anatomical perforated signatures
<div style="display: flex; justify-content: space-around;">   </div> <p style="text-align: center;"><b>A</b>                      <b>B</b></p> <ol style="list-style-type: none"> <li>1. These holes are rough.</li> <li>2. Have scars of detach</li> <li>3. Artificial holes-widths depend on devices used for drilling so they match size and shape.</li> </ol>	<div style="display: flex; justify-content: space-around;">   </div> <p style="text-align: center;"><b>C</b>                      <b>D</b></p> <ol style="list-style-type: none"> <li>1. These holes are smooth.</li> <li>2. Presence of endosteum which makes the whole canal laminated and well protected.</li> <li>3. These holes also do not match their widths and shapes.</li> </ol>

*Figures A and B are mechanical perforated holes in the medial and lateral condyles of the femur and then Figures C and D are of the intercondylar and trochanteric fossa foramen from the femur bone as well. The pictures were taken using Dino-Lite Digital Microscope AD4113T-12V*

Second, the history of the relevant skeletal collection used. The skeletal remains are recent and belong to contemporary Tanzanians, most not more than ten years in the collection. We have cleaned ourselves with our own hands and stored them, and we are still developing skeletal dis-articulation from the cadavers brought here to the college for teaching. It is true these skeletal are used for teaching and research work, but have never been drilled and deliberately connected to be used for any exhibition or class lesson. Such evidence of the history of this skeletal collection makes us confident and gives the undisputed explanation that these holes are anatomical features and not artificial.

The third reason is skeletal evidence from the burial context exhumed by Forensic field school team of the University of Dodoma and that of the Forensic Bureau of the Tanzania Police Force found from criminals. The individual exhumed in Dodoma was buried with his body and his bones were never disarticulated by human. Soft tissues decomposed when he was buried and became skeletonized. His bones were found in situ in the primary context of burial with the holes in the femoral trochanteric and condylar fossae.

These holes were first discovered from that individual. Furthermore, this evidence got support from Mumba Bioarchaeology Field School Laboratory Collection where also a female skeletonized individual with holes in the fossae was exhumed in 2019 with a team of archaeologists from University of Dar es Salaam. The second time was a femur bone that was linked to albino murder case because of superstitious beliefs. The bone is the most recent of which was actually also found with these holes. Additional evidence have been obtained from the Department of Anatomy at the Hubert Kairuki Memorial University and University of Dodoma. Such endowed historical and scientific evidence of the intercondylar and trochanteric fossa foramina removes all doubts that the foramina were found in a skeletal collection of Muhimbili University of Health and Allied Sciences were drilled or artificial. A very special thing quite recently when this paper is written there is another cadaver which was disarticulated proved to have those holes. Apart from the fact that the University of Dodoma was the first to detect the presence of these sockets in 2018, it was also the first to produce a poster with the title "unusual foramen" showing the

presence of these holes in the femoral bone. The poster was presented as a field school project at the University of Dodoma College of Natural and Mathematical Science research day of 2018. The poster was prepared by students who participated in that year's Field school with the assistance of their instructors Mr. Isaac Onoka and Vincent Jilala; the poster is still displayed in the Chemistry Department at the University of Dodoma. The department of anatomy at the Muhimbili University of Health and Allied Sciences officially sat down to discuss this discovery on 21/06/2021 with the author and then recognized it and decided to further investigate the reported foramina in order to establish their functions. A similar study was initiated in the Department of Anatomy at the Hubert Kairuki Memorial University by Dr. Lufukuja in collaboration with the author of this manuscript.

The trochanteric fossa is the groove of attachment of four femoral muscles, specifically tendon of the obturator externus, obturator internus, inferior gemellus and superior gemellus (Feneis and Dauber 2000, Saha et al. 2017, Ranaweera et al. 2020). So the discovery of trochanteric fossa foramen in the femoral trench has become something new in the standard known skeletal anatomy. The same function is performed by the intercondylar fossa as well. The distal femoral groove is home to the anterior and posterior cruciate ligaments that connect on the intercondylar fossa's right and left walls (Feneis and Dauber 2000, Calais-Germain 2004). As with the trochanteric fossa foramen, the presence of the intercondylar

fossa foramen has become a new discovery in human skeletal landmarks.

## Results

In a total of 104 right femurs taken randomly, 19 were found to have this unusual foramen, equivalent to 18.2% of all the bones (population) sampled. Using a Dino-lite digital microscope, the widths of the sockets were measured, both on the femoral neck and intercondylar fossa its width is 3.1 to 3.6 mm. The range was obtained from the most miniature socket and the largest one out of all 64 holes examined. Each bone was assessed proximally and distally to detect the size of the intercondylar fossa foramen and trochanteric fossa foramen. It was also found that the width of the femoral neck is almost the same as that of the intercondylar fossa, although there is a minor degree of variation in some bones. The depth of the foramen was detected by passing a thin and very smooth probe that crossed in the middle of the thigh bone length from the neck to the other side in the intercondylar fossa. The probe penetrated from the trochanteric fossa foramen and protruded into the intercondylar fossa foramen indicates great association of the two foramina anatomical, a physiological function that is still unknown. Part of the diaphysis was also assessed and found to contain nutrient foramen as usual. The anatomical location of the intercondylar fossa foramen (Figure 3) is situated in the middle of the intercondylar groove, while the trochanteric fossa foremen are placed in the femoral neck inside the trochanteric notch (Figure 4).



**Figure 3:** The arrows show the position of the *intercondylar fossa foramen* located at the distal end of the femur in the intercondylar fossa.



**Figure 4:** Showing the femoral neck *trochanteric fossa foramen* located in the femoral neck inside the trochanteric notch.

The output of the inserted stick is clearly visible in the photo (see Figure 5). It indicates the intercondylar fossa socket is associated directly with trochanteric fossa socket found on the neck of the femur. That is to say that everyone found to have an unusual intercondylar fossa foramen distal groove

also had another unusual trochanteric fossa foramen in the proximal notch of the femoral neck. Everyone who was found to have no intercondylar fossa socket also did not have a trochanteric fossa socket; that also exhibited the high probability of these opening being closely related in their anatomical function.



A



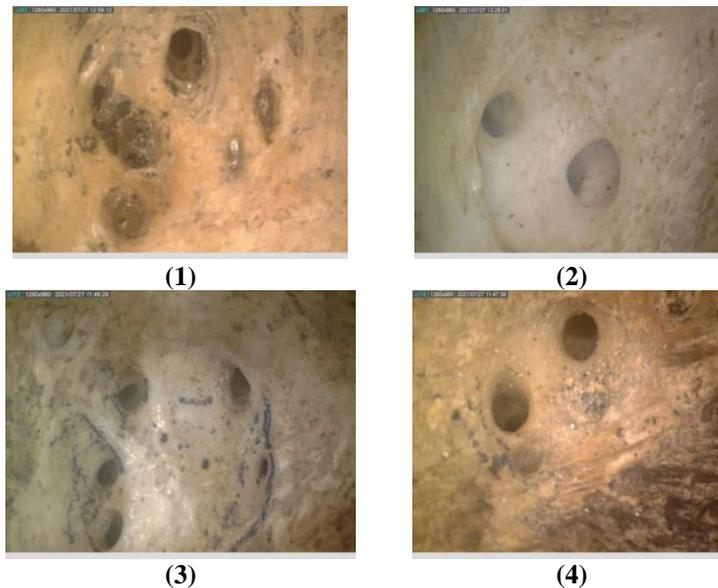
B

**Figure 5:** Foramina inserted stick to show the direct association between the two fossae sockets.

**Variant traits of intercondylar foramina**

As in the diaphysis nutrients foramen that some individuals have more than one hole (Pereira et al. 2011, Chowdhury et al. 2013) even in the intercondylar fossa foramen has been proven so. Some people have been diagnosed with two, three, four and more, all of which are located in the core centre of the

intercondylar fossa. As they become more than one they become smaller in size (widths) compared to those that are one in each intercondylar fossa. In this study, four individuals were found to have more than one intercondylar foramen as shown in Figure 6. These characteristics may be variant traits within this group with foramina in the fossae.



**Figure 6:** Variant traits of intercondylar fossae foramina. Appearance of multiple intercondylar fossa foramina found on femoral bones of four individuals. (These pictures of unusual intercondylar foramina were taken using Dino-Lite Digital Microscope AD4113T-12V).

### Discussion

In skeletal anatomy foramen defined as an opening, passage or hole that allows nerves, blood vessels and other structures to connect one part of the body to another (Feneis and Dauber 2000, Calais-Germain 2004, Sendemir 2017). From that point of view in human skeletal biology these holes found in the intercondylar and trochanteric fossae are not in any doubt whether they are foramina. Based on the surface morphology of the outlets, it is very clear that these features are indeed foramina and the important question is about their functions: Do they afford a passage for nutrient blood vessels and nerves or do they have other functions that require further investigations? What needs to be studied in more detail is the anatomical function of those sockets (Calais-Germain 2004). That can be done through a cadaver dissection during medical training or purposely for particular research that will examine the functions of these sockets (Tumbes 2007). CT Scan technology can be applied in examining what penetrates these passageways where 3D models of the inner surfaces can be created (Matsuo et al. 2019).

It should also be noted that the presence of these features in a population is still very low and that not every individual will have these foramina. So it should be remembered that such research will have to have a self-sufficient population sample size.

The intercondylar fossa foramen identified is distally directed. At the same time, there are also small intercondylar foramina that support nerves and blood vessels at the proximal end of the tibia (Matsuo et al. 2019), which if you think about it in detail, you can agree with the anatomical hypothesis that they may twist and pass through this socket (Feneis and Dauber 2000). Then make contact with the tibia and femur proximally direct to the cranial body parts. The variation perhaps lies in the osteogenesis of these foramina from one person to another (Fazekas and Kosa 1978, Alexander 2009). In most skeletal, the centre in the intercondylar groove or fossa has many small holes (foramina) instead of having one large socket (intercondylar fossa foramen). For numerous individuals, the area is seal with no foramen at all. The morphological variations may be genetically

based (Weedn 2007, Fornaciari 2018). The anterior and posterior cruciate ligaments articulate on the right and left walls within the intercondylar fossa (Feneis and Dauber 2000, Calais-Germain 2004), leaving the centre area intercondylar fossa exposed for other uses. The nerves and blood vessels that occur in the intercondylar surfaces of the proximal tibia (Matsuo et al. 2019) tend to penetrate here into the intercondylar fossa foramen to connect the femur and tibia.

It is correspondingly possible that both unusual femoral intercondylar fossa foramen and trochanteric fossa foramen are derived from the vascular system in its dispersion (Feneis and Dauber 2000). In the femoral neck, trochanteric fossa foramen may perhaps have blood vessels arising from the ascending branches of the lateral circumflex femoral artery or deep vein (Pereira et al. 2011, De Boo et al. 2020). Occurring in the foramen of the femoral neck and passes directly into the femoral metaphysis and diaphysis while supplying in the small vessels and then arises in the femoral intercondylar fossa, which is why the foramen in the trochanteric fossa has a direct connection with that in the intercondylar fossa. If you detect closely at the femoral vascular system and the presence unusual foramina, especially by focusing on the profound femoris artery until it scopes the popliteal artery and all its sub-branches (De Boo et al. 2020), it is clear that there is a high possibility the foramina or sockets are resulting from vascular system.

Numerous studies have examined the presence of additional foramen in the midshaft of femoral bones (Pereira et al. 2011, Saha et al. 2017). Most common reported are the nutrient foramen in the diaphysis region more than other foramina. Most of the researchers in human skeletal biology focus on the number, location and morphology of these nutrients foramina (Sendemir 2017, Saha et al. 2017, Ranaweera et al. 2020). Reports show that the number of nutrient foramina in the femoral bone varies from one person to another. Some have one nutrient foramen, two, and up to three (Chowdhury et al. 2013, Saha et al. 2017), but there are also those who do not have a

single visible nutrient foramen (Sendemir 2017). Most nutrient foramina are directed proximally, and their location is around the left or right side of the linea aspera on the midshaft of the femoral bone (Pereira et al. 2011). The majority of these foramina are 1.0–1.5mm wide ( Saha et al. 2017). In this new discovery, it is clear the foramen located in the intercondylar fossa, extends distally; and is associated with another unusual foramen located on the femoral neck with its average width ranging between 3.1 and 3.6 mm. That means correspondingly is the largest and broadest foramen of all the foramina in the thigh bone. The locations and directions of these intercondylar and trochanteric fossa foramina make this discovery very paramount. Further research is needed to understand the functions of these foramina.

The skeletal anatomy of the individuals from East Africa has not been well established yet. The lack of enormous human skeletal collections in medical and anthropological research institutions has made much of the unknown about the human skeletal variations of the East African population. There is a great need to address this issue as soon as possible by establishing a centre of excellence for human skeletal study in this region. All researches related to human skeletal anatomy can be carried out efficiently. The opportunity we have here in East Africa is that human cadavers are still available for training. It is financially strong, and a little training support is needed to facilitate the optimal process of preparing the skeletal bodies that are so important to anatomy and particularly osteological researches (paleopathology, bioarchaeology and forensic anthropology). The human skeletal research of these fields contributes significantly to the discovery and development of health, especially the science of anatomy and pathology, where many skeletal signatures are discovered because they are plentiful visible on skeletonized human remains than if covered by soft tissues.

## Conclusions

The assessment carried out in this work has established beyond doubt that these newly discovered foramina are not the consequence of a periosteal reaction on femoral bones as previously thought. Again it is not a sex skeletal characteristic; hence it happens to both males and females. It may be a genetic inherited skeletal phenotype that needs to be subjected to an extensive scientific investigation. First, to find out its functions. Second, possible side effects, if any. This will greatly help to know more about anatomical variants present in both knee and hip joints. It will also help specialists who provide services for hip fractures, dislocations, arthritis and other related health conditions, and even specialists in arthroscopy and arthrology anatomists, in general, will benefit significantly from such discovery, which is a continuation of this study.

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and it is impossible to mention them all, but they are all highly appreciated for their valuable assistance.

## Ethical Considerations

The ethical clearance was obtained from the Senate Research and Publications Committee of the Muhimbili University of Health and Allied Sciences (MUHAS) Ref. No. DA 282/298/01.C/ MUHAS-REC-07-2020-300.

## Competing Interests

Authors declare that there is no any conflict of interest regarding this work.

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### Appendices

The following pictures (A1–A7) are attached as supplementary materials for further reference and are useful for supporting the details of this paper.

