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THE EFFECT OF AGE ON EXTERNAL BONE MORPHOLOGY PROPERTIES IN ADULTS

ABSTRACT: The analysis of the effect of age on entheseal changes (EC) and bone robusticity is still of anthropologists' interest. Some researchers assert a strong influence of age on bone morphology in adults, underlining the necessity of age control when EC and robusticity are used in activity reconstruction. Others claim the effect of age is not so obvious, noting the "bony self-limiting" process and multifactorial aetiology of bone morphology. The aim of this study is to examine the effect of age on entheseal changes (EC) and long bone robusticity in adults. The bone material used in the study came from a medieval cemetery in Cedynia (Poland). The analyses were performed on humeri on the right side. The sample consists of 59 males (33 young adults, 23 middle adults, and three old adults) and 48 females (26 young adults and 22 middle adults). Pectoralis major and deltoid attachments were observed, and humeral robusticity index was calculated. The effect of age on EC and robusticity was examined. Additionally, the relationship between entheses and humeral robusticity index was analysed. In this study age did not have an influence on ECs or the humeral robusticity index. The relationship between ECs and humeral robusticity was significant (more robust bones have more developed entheses). The obtained results suggest that body size rather than age should be taken into account when ECs and/or bone robusticity are used for activity patterns reconstruction.

KEY WORDS: Entheseal changes – Bone robusticity – Age – Adults

INTRODUCTION

Entheseal changes (entheses, EC) (e.g. Chapman 1997, Eshed *et al*. 2004, Havelková *et al*. 2011, Hawkey, Merbs 1995, Henderson, Alves Cardoso 2013, Molnar 2006, Niinimäki 2011, 2012, Niinimäki, Sotos 2013), robusticity and cross-sectional geometric properties of bones (e.g. Bridges *et al*. 2000, Rhodes, Knüsel 2005, Stock, Pfeiffer 2001, Weiss 2003b, 2005) are used by anthropologists to reconstruct past populations' lifestyles. These studies are based on the assumption that bone changes shape and size in response to environmental stress (biomechanical stimuli, physical activity) to protect against breakage (Ruff *et al*. 2006, Schoenau, Frost 2002). But bone growth, development, modelling and remodelling processes depend not only on

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mechanical loading, but on a complex interaction of genetic (Lovejoy *et al*. 2003) and environmental factors (Daly *et al*. 2004), like body size and shape (Daly *et al*. 2004), hormonal influences (Frost 1999), diet, mechanical stimuli (McGuigan *et al*. 2002), or age (Alves Cardoso, Henderson 2010, Ruff *et al*. 1991). An examination of the contribution of the above factors in the bone remodelling process is a prerequisite for reliable interpretation of past populations' biology and behaviour (Pearson, Lieberman 2004, Ruff *et al*. 2006).

In this regard, the studies on the influence of age on EC (Al-Oumaoui *et al*. 2004, Alves Cardoso, Henderson 2010, Havelková *et al*. 2011, Milella *et al*. 2012, Molnar *et al*. 2011, Niinimäki 2011, 2012, Robb 1998, Villotte *et al*. 2010, Weiss 2003a, 2004, 2010) and long bone robusticity and cross-sectional geometric features (Daly *et al*. 2004, Johannesdottir *et al*. 2012, Klein *et al*. 2002, Pearson, Lieberman 2004, Ruff, Hayes 1983a, 1983b, Ruff *et al*. 1991, 1994, Sumner, Andriacchi 1996, Trinkaus *et al*. 1994, Weiss 2005) are widespread in anthropological science.

Effect of age on ECs morphology

Entheses (Alves Cardoso, Henderson 2010, Villotte *et al*. 2010) are bone changes manifested as increased complexity of the surface where a muscle, a tendon or a ligament inserts onto the periosteum and into the bony cortex (Benjamin *et al.* 2002, Niinimäki 2011). According to the tissue type present at the attachment site two types of entheses can be distinguished: fibrocartilaginous and fibrous (Benjamin *et al.* 1986, 2002, Villotte, Knüsel 2013). Fibrocartilaginous entheses occur on long bone epiphyses, short bones, and some part of vertebrae. Fibrocartilaginous do not attach to bone via periosteum (Benjamin *et al*. 2002, Jurmain, Villotte 2010). Fibrous entheseal changes occur on long bone diaphyses and attach to bone directly, or indirectly – by periosteum (Benjamin *et al.* 1986, 2002, Jurmain, Villotte 2010).

Age is considered to be a confounding factor in EC development (Havelková *et al*. 2011, Niinimäki 2011, Robb 1998, Stirland 1998, Villotte *et al*. 2010). It is thought that older individuals, where physical activity and therefore mechanical loading have lasted longer, tend to have more developed muscle markers than younger ones (Turner 2000, Weiss 2004, 2007, 2010). The influence of age on EC manifestation was proved by Chapman (1997) in the indigenous population of Pecos Pueblo, by Robb (1998) in the Italian Iron Age material, by Mariotti *et al.* (2004), who investigated male skeletons from Italy, and Molnar (2006), who analysed Neolithic material from Gotland and found age to be a contributing factor to increased ECs, especially in the female group. Wilczak (1998), analysing a skeletal collection from North and Central America found that only in the male group was the effect of age on EC present. Weiss (2003a), who examined ECs of upper limbs in a British Columbia and Quebec sample, found that, although aggregate entheseal changes correlate significantly with age, this relationship does not exist when a single EC is analysed. When the same skeletal sample was examined according to lower limbs, it was found that both, aggregate and single ECs correlate with age (Weiss 2004). Similarly, age was a good predictor of both aggregate and single upper limb entheses in prehistoric central Californian Amerinds (Weiss 2007). The conclusion that aging is a key factor in EC presence can be drawn also from studies by Alves Cardoso and Henderson (2010, 2013), Villotte *et al.* (2010), Niinimäki (2011), or Milella *et al.* (2012).

Conversely, in the study by Havelková *et al.* (2011), although the development of EC was higher among older individuals, when EC were analysed separately, only a part of them were significantly correlated to age. Weiss *et al.* (2012) found in examining entheses from the upper limb bones that not all ECs were significantly correlated with age. Similarly, in Weiss (2012), only few of the examined ECs were significantly affected by age. Niinimäki and Sotos (2013) also observed that only some ECs of the lower limb were affected by age. Henderson and co-workers (2013), analysing the effect of age on fibrocartilaginous entheses, however observed a general trend for ECs increasing with age, but did not note a statistical significance for all features.

In conclusion, the influence of age on entheseal change development seems to be evident, but the existence of exceptions forces us to continue these studies. Furthermore, for the last few years, the interpretation of the effect of age on fibrous and fibrocartilaginous EC has been equivocal. Mariotti *et al.* (2007), Niinimäki (2011) have argued that age is a causative factor of fibrous entheses. Villotte (2009) proved that fibrous entheses are more greatly affected by age than fibrocartilaginous. But Villotte *et al.* (2010) and Alves Cardoso and Henderson (2010) pointed out that age also plays a very important role in the case of fibrocartilaginous EC development. In Weiss (2012), regardless of whether it was fibrocartilaginous or fibrous ECs, only a few of the examined ECs were significantly affected by age, fewer fibrocartilaginous ECs correlated with age than did fibrous. Henderson *et al.* (2013) showed that although in general fibrocartilaginous entheses increase with age the statistical significance was not noticed for all features. These discrepancies between researchers indicate a need for further analyses of this topic.

Effect of age on bone robusticity

Skeletal robusticity refers to the strength of bone in terms of its shape and size (Stock, Shaw 2007). Primarily, the term robusticity refers to diaphyseal thickness standardised to bone length (Martin, Saller 1957, Pearson 2000). Advanced research techniques (e.g. computer tomography and radiographs) allow for detailed measurements of cross-sectional geometry (Ruff 1992, Shaw, Stock 2011, Stock, Pfeiffer 2004, Trinkaus *et al.* 1994). Because the high cost of the methods and the availability of the equipment limits the widespread application of technically advanced methods to the bone robusticity examination, traditional methods (e.g. external bone metrics) are still useful (Stock, Shaw 2007).

The analysis on the response of bone robusticity to age has been conducted for years (Garn *et al*. 1969, Ruff *et al*. 1991, 1994, Trinkaus *et al*. 1994, Mays 2001). It has been known that bones adapt their structure to the mechanical loading, and increased mechanical forces lead to greater bone robusticity (see Mays 2001). Thus the cross-sectional properties of long bones are used by anthropologists to reconstruct past people's lifestyles (Bridges *et al*. 2000, Rhodes, Knüsel 2005, Shaw, Stock 2009, Stock, Pfeiffer 2001, Weiss 2003b, 2005). General model proposed by Ruff *et al.* (1994) is that, while juveniles deposit subperiosteal bone, and slow the endosteal resorption in response to strenuous mechanical loading, adults tend to slow the rate of endosteal resorption, but are not able to add substantial amounts of subperiosteal bone, even if they are very active in their adulthood. But the detailed mechanism and factors underlying age-related changes in bone robusticity and geometry is not completely understood (Klein *et al.* 2002, Pearson, Lieberman 2004), and the results of the researches are inconsistent. Increasing in subperiosteal diameter with aging were firstly recorded by Smith and Walker (1964). Similar results were presented by Ruff and Hayes (1982, 1983b) in the Pecos Pueblo skeletal sample, or by Stein *et al.* (1998) for adult cadavers from Australia. The studies by Ruff and Hayes (1988) for the cadaveric skeletal material from the US or by Feik *et al.* (1996) for the modern Australian population confirm endosteal resorption, subperiosteal expansion and apposition, but with a more stable model of bone age-changes in males. Ruff *et al.* (1991, 1994) and Trinkaus *et al.* (1994), who examined age-related changes in bone cross-sectional parameters, revealed that periosteal expansion is small in adults, and mechanical loads during adulthood have little effect on the external dimensions of long bone diaphysis. Niinimäki (2012) showed that the impact of age on humeral cross-sectional properties was also not evident. Because the researches results are not consistent, and the detailed mechanism and factors underlying age-related changes in bone robusticity and geometry is not completely understood (Klein *et al*. 2002, Pearson, Lieberman 2004), the analysis of bone response for aging process should be continued.

The aim of this study is to complete the studies where the effect of age on muscle markers and bone robusticity is analysed. For better understanding of bone remodelling process during life, the dependency between entheseal changes and humerus robusticity index was performed. These analyses are the attempt to better understanding of bone modelling and the remodeling processes during lifetime. Furthermore, it can improve knowledge about past people lifestyle reconstruction.

MATERIALS AND METHODS

The material used in the study came from the medieval cemetery in Cedynia, Poland. It has been dated to the period spanning from the end of the 10th century to the first half of the 14th century (Malinowska-Łazarczyk 1982, Porzeziński 2006).

In order to avoid loading on means, no missing data treatment was undertaken. Thus, only individuals with complete humeri were included in the statistical analysis. The analyses were made for 59 males (33 young adults, 23 middle adults and three old adults) and 48 females (26 young adults and 22 middle adults). Age categories were taken from Buikstra and Ubelaker (1994): young adults (YA, 20–34 years), middle adults (MA, 35–49 years), and old adults (OA, 50+ years). As noted by Mariotti *et al.* (2004), muscle insertion sites are not well developed in young individuals, and many pathological changes on muscle insertion sites are observed over 60 years. Therefore, only individuals older than 20 years and younger than 60 years were included.

Age and sex of the individuals were performed by Nowak, Piontek (2002). The age and sex of the individuals were estimated according to Ferembach *et al.* (1979) and Buikstra and Ubelaker (1994). The skeletal features used for age estimation were the degree of the changes of the symphyseal surface of the pubic bone, cranial suture obliteration, and the degree of dental crown attrition. The changes on the symphyseal surface of the pubic bone were the main skeletal features used for age estimation. The degree of cranial suture obliteration and the degree of dental crown attrition were used only as auxiliary methods. Features of the cranium and pelvis were used for sex estimation (Nowak, Piontek 2002). To avoid under- or overestimation of the results individuals with pathological changes of the skeleton were excluded.

Because fibrous entheses are less understood than fibrocartilaginous (Benjamin *et al.* 2002) and are thought to be less affected by occupation (Villotte 2009), injuries and traumas (Benjamin *et al.* 2002), in this study an examination of the effect of age on fibrous entheses was made. The recorded muscle sites were *m. pectoralis major* (bicipital groove) (H1) and *m. deltoideus* (deltoid tuberosity) (H2). These entheses are analysed because they are fibrous entheses (see above), they are easy to observe, they are well preserved in analysed material, and finally these muscles are mostly engaged in a daily activity. The analyses were carried out based on the entheseal changes variability scale developed by the authors on the basis of the material from Cedynia (Myszka, Piontek 2012). A three-point rating scale was used (1, low; 2, moderate; 3, high degree of the muscle site morphology complexity) (Myszka, Piontek 2012). Only changes of the robusticity type were included when developing the scale (stress lesions and ossification exostoses were not included) (Myszka, Piontek 2012). As noted by Dutour (1986), Galera and Garralda (1993), Mariotti *et al.* (2004), Benjamin *et al.* (2002), and Villotte *et al.* (2010), changes of stress lesion or ossification type are morphological variations of pathological changes in tendon attachments, the so-called enthesopathies. Moreover, osteophytosis is thought to be an indicator of trauma to the insertion site rather than activity (Eshed *et al.* 2004, Hawkey, Merbs 1995, Weiss 2007). While porosity is connected with some inflammatory processes linked to injuries or systemic disease (see Freemont 2002).

According to Weiss (2007) an aggregate humeral score was created by adding the EC ordinal scores. Only those humeri where both entheses were available were included. Humeral robusticity was calculated using index HRI = (M7/M1) × 100; where M1 is the maximum length and M7 is the minimum shaft circumference. The measurements were taken using the techniques proposed by Martin and Saller (1957), thus M7 and M1 refer to original measurements and their abbreviations described in Martin and Saller (1957). All the analyses were made on the right humeri only.

A correlation between aggregate ECs, robusticity indices, and age was analysed with Spearman's rank correlation test. Additionally, to check the influence of body size on EC manifestation the correlation between aggregate EC and HRI was calculated. The analysis was made using Spearman's rank test. The significance of the differences was determined at a probability level of 0.05. All computations and illustrations were made using STATISTICA 6.0 PL software.

TABLE 1. Mean, median and standard deviation (SD) for the humeral entheses by sex.

		Males				Females			
Entheseal changes type	Age group	*N*	Mean	Median	SD	*N*	Mean	Median	SD
H1 Bicipital groove	All groups	59	1.820	2.0	0.693	48	1.542	1.0	0.683
	Young adults	33	1.794	2.0	0.683	26	1.794	1.0	0.647
	Middle adults	23	1.917	2.0	0.717	22	1.852	1.5	0.727
	Old adults	3	1.331	1.0	0.577				
H2 Deltoid tuberosity	All groups	59	2.033	2.0	0.701	48	1.833	2.0	0.783
	Young adults	33	1.971	2.0	0.704	26	1.971	1.0	0.703
	Middle adults	23	2.125	2.0	0.741	22	2.111	2.0	0.774
	Old adults	3	2.000						
EC Aggregate entheseal changes	All groups	59	3.852	4.0	1.205	48	3.375	3.0	1.347
	Young adults	33	3.765	4.0	1.200	26	3.765	2.5	1.216
	Middle adults	23	4.042	4.0	1.268	22	3.963	3.5	1.412
	Old adults	3	3.333	3.0	0.577				

RESULTS

Tables 1 and *2* present descriptive statistics of entheseal changes of humeral bone, bone measurements, and humeral robusticity indices for the individuals form the Cedynia cemetery.

TABLE 2. Means and standard deviations (SD) for the humeral measurements by sex.

		Males			Females		
Measurement	Age group	*N*	Mean	SD	*N*	Mean	SD
M1 Maximum length	All groups	59	336.3	12.36	48	303.2	15.44
	Young adults	33	337.4	13.03	26	304.7	16.14
	Middle adults	23	334.0	11.76	22	301.6	14.76
	Old adults	3	342.0	11.53			
M7 Minimum shaft circumference	All groups	59	64.3	4.04	48	56.6	5.82
	Young adults	33	64.4	4.00	26	55.6	6.50
	Middle adults	23	63.9	4.45	22	58.3	4.61
	Old adults	3	64.7	1.53			
HRI Humeral robusticity index	All groups	59	19.12	1.145	48	18.76	1.776
	Young adults	33	19.10	1.080	26	18.23	1.783
	Middle adults	23	19.15	1.296	22	19.37	1.593
	Old adults	3	18.93	1.077			

TABLE 3. Spearman's correlation coefficients (r_S) between age and humeral robusticity index (HRI) and humeral aggregate entheseal changes (EC) by sex.

	Males			Females		
	N	r_s	*P*-value	*N*	r_s	*P*-value
HRI / Humeral robusticity index	59	0.072	0.607	48	0.271	0.068
EC / Aggregate entheseal changes	59	0.061	0.646	48	0.284	0.055

Correlations between age and humeral robusticity indices and aggregate entheseal changes are shown in *Table 3*. Age did not affect EC and humeral robusticity indices in our sample. But in females, the correlation is marginally insignificant (EC, $r_S = 0.284$, $P = 0.055$; HRI, $r_S = 0.271$, $P = 0.068$) (*Table 3*).

Table 4 presents the correlations between humeral robusticity indices and aggregate humeral entheseal changes. When the analysis was made for all age groups, the correlation between humeral robusticity index (HRI) and aggregate EC is statistically significant for both males and females (*Table 4*). In young adults humeral robusticity index (HRI YA) and aggregate EC (EC YA) were also significantly correlated. In each case the correlation is positive – more robust humerus has more developed EC. In middle adults group the relationship between muscle markers and humeral robusticity index is positive but statistically insignificant (*Table 4*).

DISCUSSION

Entheses and age

In the skeletal population from Cedynia, age is not a dominant factor influencing entheseal changes (*Table 3*). It is not consistent with the majority of previous studies, which demonstrate an increase of ECs with age (e.g. Al-Oumaoui *et al*. 2004, Alves Cardoso, Henderson 2010, 2013, Churchill, Morris 1998, Havelková *et al*. 2011, Mariotti *et al*. 2004, Milella *et al*. 2012, Molnar 2006, Niinimäki 2011, Robb 1998, Villotte *et al*. 2010, Weiss 2003a, 2004, 2007). Some studies confirm that, although a general trend for increase of ECs

TABLE 4. Spearman's correlation coefficients (r_s) between humeral robusticity index (HRI) and humeral aggregate entheseal changes (EC) by sex and age group.

		EC / Aggregate entheseal changes								
		All groups			Young adults			Middle adults		
		N	r_s	*P*-value	*N*	r_s	*P*-value	*N*	r_s	*P*-value
Males										
HRI Humeral robusticity index	All groups	56	0.384*	0.003						
	Young adults				33	0.498*	0.003			
	Middle adults							23	0.360	0.092
Females										
HRI Humeral robusticity index	All groups	48	0.417*	0.003						
	Young adults				26	0.538*	0.005			
	Middle adults							22	0.208	0.352

* Statistically significant $P \leq 0.05$

with age is observed, when single entheses are analysed, not all of them are affected by age (Havelková *et al.* 2011, Niinimäki 2011, Niinimäki, Sotos 2013, Weiss 2003a, 2012, Weiss *et al.* 2012). It suggests that the problem of the real participation of the effect of age in ECs development in adults is not resolved yet, and many aspects should be considered.

It is claimed that more pronounced muscle markers in older individuals are the result of more stressful activity (Weiss 2004, 2007, 2010), and the accumulation of microtraumatic stress on entheseal surfaces are due to daily activity (Milella *et al.* 2012, Molnar 2006). But some researchers claim that more developed ECs are indeed the result of continued muscle use in repetitive tasks, however "use" should start in childhood and continue through adulthood (Robb 1998, Weiss 2007, Wilczak 1998). Moreover, while children and adolescents exhibit strong bone remodelling responses to mechanical loading, adults exhibit very little or no response to changes in loading (Pearson, Lieberman 2004, Ruff *et al.* 1991, 1994). The same conclusion was drawn by Henderson *et al.* (1995), who found that muscle strength increases during growth. It plateaus between 25–30 years of age, to decline after that time (Henderson *et al.* 1995). Similarly, Robb (1998) found that entheses increase with age, from maturity to 40–50 years. After that age the process levels off (Niinimäki 2011, Robb 1998). The same results were obtained by Klein *et al.* (2002), Niinimäki (2011), Milella *et al.* (2012). This could be due to physical activity decreasing along with advancing age (Milella *et al.* 2012), changing activity regime, or finally it may be that in old age bone may have reached its limits to react to biomechanical stress and therefore the effect of age is levelled off (Niinimäki 2011, Robb 1998). The "levelling off" process could explain the lack of age effect on entheseal changes development in the material from Cedynia, and in other series, where the impact of age was not so evident (see above). Although without more complex analysis of "levelling off" process, this simple explanation can be treated as an assumption only. But, it can be concluded from the above that the existence of the "levelling off" process must be considered when developing ECs scales and interpreting the results.

The methods used to assess changes at the entheses can influence the final results. In the present study the scale of ECs development does not include pathological (degenerative) changes occurred in entheses (osteophytosis, porosity) (Hawkey, Merbs 1995, Mariotti *et al.* 2004). According to Alves Cardoso and Henderson (2010), Milella *et al.* (2012), Havelková *et al.* (2011), and Villotte *et al.* (2010) degenerative processes within an enthesis tend to advance with age. According to this older individuals should have more "pathological" ECs. In this study, as mentioned before, we excluded from the analysis ECs with osteophytosis and porosity, which are treated by many researchers as pathological changes (Benjamin *et al.* 2002, Dutour 1986, Galera, Garralda 1993, Mariotti *et al.* 2004, Villotte *et al.* 2010). Does it influence the results? In the analysis of Alves Cardoso and Henderson (2010), where only robusticity scores were used, correlation between age and EC was evident.

On the contrary, in the study by Niinimäki and Sotos (2013), where only robusticity scores were considered, age did not affect all the studied entheses. Although, in number of studies, where osteophytosis and/or porosity of ECs were compiled in analyses, a strong influence of age on ECs development was found (Havelková *et al*. 2011, Hawkey, Merbs 1995, Milella *et al*. 2012, Molnar 2006, Niinimäki 2011, 2012, Villotte *et al*. 2010, Weiss 2003a, 2004, 2007, Weiss *et al*. 2012). But, it should be pointed out, some of these studies show that not all of examined entheses were influenced by age (Havelková *et al*. 2011, Weiss *et al*. 2012). The inconsistency of the results show that more detailed debate about using pathological and non-pathological bone changes on entheseal sites must be taken. It is necessary firstly for the development of the recording methods for entheses, secondly for the reliable interpretation of the results.

The recent debate about the effect of age on fibrous and fibrocartilaginous EC has provided no consensus. It is claimed that age-related changes in fibrous are the result of cumulative long-term activity, that leads to rougher ECs, while changes in fibrocartilaginous are connected with the decreasing stiffness of tendons (Molnar 2010, Nagy 1998). It is claimed that fibrous entheses are more greatly affected by age than fibrocartilaginous (Villotte 2009). In the present study, both analysed fibrous entheses are not affected by age (*Table 3*). In Weiss study (2012), only 39% of examined fibrous sites were significantly affected by age. Similarly, Al-Oumaoui *et al.* (2004) found the effect of age only in some of the analysed fibrous EC. Wilczak (1998) observed younger individuals to have more developed fibrous entheses but the relationship concerned males only. But numerous studies show that age is a causative factor of fibrous entheses (Alves Cardoso, Henderson 2010, Mariotti *et al*. 2007, Niinimäki 2011, Niinimäki, Sotos 2013, Villotte 2009, Villotte *et al*. 2010, Weiss 2007). Nagy (1998) and Molnar (2010) suggested that age correlation can reflect the influence of activity on EC morphology (older individuals accumulate more microtrauma during life, than their younger counterparts). Alves Cardoso and Henderson (2010), Niinimäki (2011) underlined that age in and of itself is a cause of entheseal changes development. Taking into account that fibrous entheses are not well understood yet, that their morphology shows a large variation (Benjamin *et al*. 2002), and the results for the effect of age on fibrous morphology are inconsistent, it can be claimed that further, more detailed studies of fibrous entheses are needed.

The problem with age-at-death determination of bioarchaeological samples is a significant limitation in a reliable interpretation of the results (see Alves Cardoso, Henderson 2010). It is not possible to give the precise age-at-death of individuals. Researchers are dealing with biological age only, the assessment process is usually based on damaged, and incomplete material. Thus, in the majority cases it is only possible to give age ranges. That is why the simplistic interpretations of the association between age and EC seems to be inappropriate (Alves Cardoso, Henderson 2013), especially in the case of undocumented series. This problem does not seems to exist in the studies where identified (of known age-at-death, sex, occupation) skeletal collections were investigated (e.g. Alves Cardoso, Henderson 2010, Milella *et al*. 2012, Niinimäki 2011, Niinimäki, Sotos 2013, Perréard Lopreno *et* al. 2013, Villotte *et al.* 2010), and in the majority of such studies, age was the main factor in ECs aetiology. But Alves Cardoso and Henderson (2013) pointed out that even in well documented material the existence of the correlation between age and ECs should be treated with a caution, and multifactorial aetiology of entheses must be considered.

The above requires caution and prudence when interpreting the effect of age on ECs development, and indicates the necessity of the discussion of the other factors which can influence the final results.

Bone robusticity and age

In this work the effect of age on bone robusticity was not observed (*Table 3*). These results seem not to be surprising since we consider that mechanical stimuli during adulthood have little effect on the external dimensions of long bones diaphyses. While juveniles deposit subperiosteal bone in response to strenuous mechanical stimuli, adults are not able to add a substantial amounts of subperiosteal bone (Garn *et al*. 1969, Pearson, Lieberman 2004, Ruff *et al*. 1994). It may indicate that the cross-sectional properties of adult bones are affected by physical activity performed earlier in life, and differences in cross-sectional properties among individuals (samples) could be due to differences in activity before skeletal maturity (for detailed discussion, see Bice 2003). But the final conclusion should not be drawn without taking some limitations.

Firstly, as it was pointed out by Mays (2001), it is difficult to demonstrate continued subperiosteal apposition in skeletal material. Secondly, small sample size and problems with precise age-at-death and sex determination (Mays 2001) must be also considered when the effect of age on bone morphology is examined. Finally, as was underlined before, not only does mechanical loading play important role in bone development, but so do many other factors (including genetics, hormonal, diet, health status, etc.) (Alves Cardoso, Henderson 2010, Daly *et al*. 2004, Frost 1999, Lovejoy *et al*. 2003, McGuigan *et al*. 2002, Ruff *et al*. 1991).

The lack of an age effect on bone robusticity and entheses in a contemporary study and in another ones (see above) might suggest a similar relationship of EC and robusticity to physical activity. Niinimäki (2012) and Schoenau and Frost (2002) argued that stress directed by muscles results in bone remodeling to both, the overall shaft (and diameters) and specific location. Rhodes and Knüsel (2005) indicated that limb robustness is correlated with overall limb function. According to that, it can be hypothesised that adults from Cedynia were not engaged in strenuous activity during their adulthood and, therefore the features of bones (robusticity, EC) were not able to increase with age. On the other side, heavy labour can also distort the results, since Niinimäki (2011) proved that bone is unable to react to continuous heavy loading and the increase of bones parameters may slow down after a certain level of loading is reached. Therefore, what was pointed out before, other skeletal features (e.g. osteoarthritis), biological, and archaeological sources should be also considered when EC and skeletal robusticity are used to examine habitual activity patterns of past groups.

In this study individuals with more massive humeral bones have more developed entheses (*Table 4*). This relationship can indicate the similar aetiology of bone response to mechanical stimulus. When we take the fact that juvenile individuals, who are engaged in strenuous physical activity, build considerably more bone than their non-exercising peers (Pearson, Lieberman 2004, Ruff *et al*. 1994), and the assumption of Schoenau and Frost (2002), Rhodes and Knüsel (2005), or Niinimäki (2011) that use of muscles results in bone remodelling as a whole (overall shaft and specific location), we could claim that the period before the end of bone growth seems to be essential in achieving the shape and size of bone. A number of studies confirm that individuals with larger bones have more developed ECs (Berget, Churchill 1994, Myszka, Piontek 2011, Niinimäki 2012, Stirland 1998, Weiss 2003a, 2004, 2007, 2012, Weiss *et al*. 2012). But some of them show that only when ECs are treated as a whole, the correlation between EC and bone robusticity is significant, while this relationship doe not always exist when single entheses are considered (Myszka, Piontek 2011, Stirland 1998, Weiss 2012, Weiss *et al*. 2012). Bridges (1997) explained the lack of the correlations as an evidence for the independent nature of the processes controlling the formation of bone robusticity and the size of ECs. According to Bridges (1997) the shape, size and robusticity of bones are affected by factors such as the so-called "overall load" (resultant of body weight and physical activity) placed on the skeleton, while the complexity of the muscle attachment sites is caused by the "specific load", resulting from a muscle (or a group of muscles) being engaged in a physical activity.

It could be hypothesised from the above that the shape and size of adult bones depends mostly from the "overall load" (including muscle use, loading connected with body size) during childhood and adolescence and that the bone changes are very dynamic. While in adulthood external bone features are not so plastic (especially robusticity), and their "development" is a result of specific loading. But, bearing in mind that the mechanism of skeletal changes according to age is not well understood yet, and, what was underlined before, bone growth, development, modelling, and remodelling depends on many other than physical activity factors (Daly *et al*. 2004, Frost 1999, Lovejoy *et al*. 2003, McGuigan *et al*. 2002, Ruff 2003, Ruff *et al*. 1994), this assumption must be treated with a caution.

SUMMARY

In this study, we examined the effect of age on entheseal changes (EC) and long bone robusticity in adults. Two fibrous humeral entheses and humeral robusticity index were scored in a medieval population from Poland. Additionally, the relationship between entheses and humeral robusticity index was analysed. In this study age did not influence either ECs or the humeral robusticity index, but more robust bones have more developed entheses. The results suggest that body size rather than age should be taken into account when entheses are used in relation to activity patterns reconstruction.

But this conclusion must be treated with a caution because, although the present results are consistent with some data, they are not in agreement with others. Inconsistency of the researchers should prompt continued analysis of skeletal morphology changes according to age, especially in adults. This knowledge is an essential condition for the reliable interpreting of lifetime, biology and behaviour of past populations.

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