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The plague's impact paleodemographic and genetic measures in 15th to 16th century Gdańsk

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ABSTRACT: Yersinia pestis caused plagues and haunted Gdańsk several times during the 15th and 16th centuries. This study focuses on the following demographic effects: 1/ distributions of deceased by age in a plagued city, 2/ parameters of the life tables, 3/ estimation of the natural increase. To assess genetic effects of the plague, measures of the opportunity for natural selection were considered. Skeletal remains of 283 people from the 15th - 16th century ossuary 3009 from the Dominican Monastery in Gdańsk provided research material. Yersinia pestis DNA in this skeletal material has already been found (Morozowa et al. 2017, 2020). Distributions of the deceased by age in the study sample were compared with those for Gdańsk before the plague and with those for the mass burial of plague victims in the 14th century Lübeck. Neither catastrophic mortality was found in the material studied, nor selective nature of the plague with regard to sex and age had been demonstrated. Using the Weiss method, the rate of natural increase r = -0.005was reconstructed. With the wide dating range of the ossuary and the fact that it contains results of both the epidemic and "normal" mortality, the natural increase value at this level seems justified. There was a deterioration in the values of life tables parameters, especially life expectancy. Newborn life expectancy dropped to 19.5-22.6 years and for a 20-year-old to 17.7 years. The measures of opportunity for natural selection also deteriorated primarily due to child mortality: the biological state index Ibs values were low (within the 0.3-0.4 range) and values of the Im Crow's index about 1.0. Natural selection also acted on adults as evidenced by values of the gross potential reproduction rate Rpot below 0.7.

Demographically the study sample was at the level of the early Middle Ages rather than the Rennaisance.

KEY WORDS: mortality profiles, natural increase, life table, opportunity for natural selection



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Introduction

The plague is an acute infectious disease caused by *Yersinia pestis*, a bacilliform bacterium. It can be spread by various routes of transmission. Certain rodent species, especially rats, are animal reservoir hosts of the plague, and the fleas which live in rat fur are vectors of the disease; other vector species, as has been recently proven, include human ectoparasites such as human fleas and lice (Scott and Duncan 2001; Karpiński 2012; Kacki 2017; Dean et al. 2018; Bosio et al. 2020).

There are three clinical forms of the plague: the most common one is the bubonic plague, which manifests itself in abnormally swollen glands ("buboes") and subcutaneous haemorrhages, which shortly turn dark blue, hence the name "Black Death." Formally speaking, although this variety of the plague was the least deadly one, in the past, when antibiotic therapy was not available, it caused death in 40% to 90% of cases, depending on time and place. Two other forms of the plague: pneumonic plague and septicemic plague were fatal in nearly 100% cases (Margerison and Knüsel 2002; Karpiński 2012; Pechous et al. 2015; Kacki 2017; Dean et al. 2018). This dramatically high mortality and extremely high infectiousness allowed the plague to spread rapidly over the world and repeatedly return in the form of a pandemic, killing millions.

The first large-scale, historically and more recently genetically documented pandemic, was the Plague of Justinian dated to the sixth century AD. However, *Yersinia pestis* is much older and was endemic as early as over 5000 years ago (Rasmussen et al. 2015). The Plague of Justinian probably started in Africa. It came to Europe from Egypt and affected mostly Byzantium with its capital in Constantinople. Its first wave killed 20-30% of the pre-pandemic population (Scott and Duncan 2001; Byrne 2008; Gładykowska-Rzeczycka 2008; Wójcik 2011; Harbeck et al. 2013; Feldman et al. 2016; Keller et al. 2019). The outbreak of the bubonic plague triggered the second pandemic known as the Black Death. It was the largest and most severe pandemic, which spread from the steppes of Asia to North Africa, the Levant and Europe. In Europe, it appeared 1347 and kept recurring for four hundred years until the 18th century. It caused dramatic loss of life; according to various estimates, it killed 1/3 to 2/3 of the continent's population (Scott and Duncan 2001; Byrne 2008; Gładykowska-Rzeczycka 2008;Możejko 2012). It reached Poland as well. The third pandemic started in the 1830s in East Asia, mainly in China, and by 1960 it spread across the globe. Already in its early stages, in China alone, it caused deaths of several million people (Byrne 2008). Today, the world is not yet entirely free from this deadly disease. Yersinia pestis is still endemic in Asia, Africa and even in Americas. Admittedly, in the age of modern medicine this has not led to such death tolls as previously. However, the WHO still considers it a serious public health risk (Byrne 2008).

According to historical sources, the Black Death was brought to Poland in 1348 or 1349 on a sea vessel. It wrought havoc in Gdańsk, Pomerania and Prussia, and went on to infect Lesser Poland and Greater Poland. It returned to Poland many times throughout the 15th century. The city of Gdańsk was the gate through which the plague made its entry.

A port dating back to early Middle Ages, 15th and 16th century Gdańsk was a rapidly growing economic centre. Its lo-

cation on the southern shore of the Baltic Sea (Gdańsk Bay) at the estuary of River Vistula ensured numerous business contacts. Vistula was used to float commodities made in Poland and neighbouring countries down to the Baltic Sea. The sea, in turn, offered trade opportunities with Scandinavian, German and other merchants. Gdańsk's membership in the Hanseatic League, a commercial confederation of market towns, brought enormous profits and an economic boom. Still, the city's geographic and commercial position had both benefits and drawbacks; the latter included constant exposure to epidemics brought by incoming vessels. In the 15th century, there were 6 to 7 outbreaks of the plague in the city (source documents reveal varying numbers): once or twice at the beginning of the century, and every 10 to 14 years by 1450. A substantial decline in population was reported in 1464. In that year, 5800 people, i.e. 19% of the then residents of Gdańsk, died of the plague. Various historical sources contain accounts of that wave of the pandemic. In 1463, before reaching Gdańsk, it struck the Netherlands (towns and villages), Cologne, Braunschweig, and then Salzburg (the Holy Roman Empire). In 1465 it spread to Saxony, Hamburg, Lübeck, followed by Prussia and Gdańsk. The plague returned to Gdańsk 4 to 5 times in the following two centuries. In 1564, it killed more than 23,000 people, which amounted to 30% of the city's population. The last wave of the epidemic in Gdańsk was recorded in 1709, but at that time it took the greatest number of lives in the city's history: Gdańsk lost approximately 40% of its population. Of the 24,535 deaths reported at that time, 90% were due to the plague (Kizik 2012; Możejko 2012; Trzoska 2012).

Many DNA studies have confirmed long-term persistence of *Yersinia pestis* in Europe, along with the pathogen's genetic diversity (Bos et al. 2016; Seifert et al. 2016; Klunk et al. 2019; Morozowa et al. 2020).

In 2017, Morozowa et al. were the first to confirm the presence of the plague caused by $15^{\text{th}}/16^{\text{th}}$ century *Yersinia pestis* bacterium in Gdańsk in DNA tests. Three years later, using ¹⁴C carbon, the dating of bone samples tested for the disease was refined to the years 1425-1469. In addition, rat bones were discovered among plague-altered human remains. DNA tests helped determine that the bones belonged to the black rat species (*Rattus rattus*). In this way, a potential animal reservoir of the plague in late mediaeval Europe was identified (Morozowa et al. 2020).

In the past, a contagious and deadly disease such as the plague was a powerful population size regulator. Historians often emphasize the extremely high mortaliy caused by the plague. Nevertheless, its detailed demographic outcomes have been scantily examined.

Generally speaking, as pointed out by various palaeodemographers, mortality patterns in historical human populations reveal a high degree of uniformity. It is characterised by a remarkably high death rate for small children, especially infants, a clear drop in mortality among adolescents, and a gradual increase in the number of deaths among adults. This type of mortality pattern is referred to as attritional mortality. Its has its reverse in catastrophic mortality, a pattern emerging in the course of sudden, dramatic events such as an epidemic, hunger, natural disaster etc. and manifests itself by a relatively short mortality crisis, in which all age groups are exposed

to elevated risk of death. In a catastrophic mortality profile, the likelihood of death for all individuals is approximately equal. Accordingly, it should reflect the age structure of a living population (Paine 2000; Gowland and Chamberlain 2005; Chamberlain 2006). With regard to the plague, it has been discussed whether it killed people indiscriminately or selectively, depending on age and sex. Attempts were made to attribute such differences to health, as expressed by the severity of various stress markers on bones. However, the results presented by researchers are inconclusive. Some authors report differences in mortality rate for men and women during the plague (Hollingsworth and Hollingsworth 1971; Scott et al. 1996; De Witte 2009; 2010; Curtis and Roosen 2017), while others do not point to such relationships (Waldron 2001-02; Kacki 2017; Bramanti et al. 2018). Certain researchers claim that the plague caused deaths regardless of age (De Witte 2010; Kacki 2017; Bramanti et al. 2018), while others found marked differences in the distribution of mortality between the deceased from epidemic and non-epidemic graveyards, sometimes indicating clearly catastrophic mortality patterns due to the plague (Hollingsworth and Hollingsworth 1971; Prechel 1996; Scott et al. 1996; Margerison and Knusel 2002; Gowland and Chamberlain 2005).

The aim of the present work was to evaluate the biodemographic effects of the plague epidemic on the basis of ossuary material from 15th/16th century Gdańsk. Mortality profiles in the plague-stricken city were compared to non-epidemic mortality in early mediaeval Gdańsk (Pudło 2016) and mortality reported for a mass grave of plague victims in the 14th century Lübeck (Prechel 1996; Pudło 2012). The assessment also included the rate of natural increase and life table parameters. Diseases, especially epidemics such as the plague have been a significant modifier of gene pool changes and the condition of a population, and as such they constitute a crucial (and previously basic) factor in human microevolution. Therefore, this article is also aimed at determining the opportunity for natural selection in the analysed Gdańsk population from the early modern period.

Material and methods

Human bone remains used in this study derived from Ossuary 3009 - one of the ossuaries discovered during archaeological excavations in Dominikański Square in Gdańsk. Work at the site was carried out by the Archaeological Museum in Gdańsk in the years 2009-2011 (Szyszka 2017). The bones collected from the ossuaries were subjected to multi-disciplinary analyses in fields such as archaeology, anthropology and history.1 Ossuary 3009 was related to the the church/monastery compound, which consisted of St. Nicholas's Church and the Dominican Monastery. Both the church and the monastery were built in the mid-14th century. The Dominicans stayed there until 1813, when the monastery was destroyed by Russian artillery fire during Napoleonic wars, and subsequently pulled down. The church func-

¹ Results of anthropological, archaeological and historical studies were analysed in interdisciplinary research under the project co-financed by the Ministry of Culture and National Heritage from the funds of the "Cultural Heritage" Program, Priority: "Protection of archaeological relics" (grant no. 04695/16).

tions to this day (Szyszka 2017). The bones from Ossuary 3009 were deposited in a pit dug in the cloister garth, under the eastern wall of the lavatory. The ossuary was dated to the $15^{th}/16^{th}$ centuries by means of archaeological methods (the period when the lavatory was built, metal artefacts discovered during bone exploration) (Szyszka 2017; Trawicka 2017).

Skeletal material from the ossuary was radically mixed and the bones were laid down without any anatomical order (Figure 1). The ossuary was special in terms of the way in which the bones had been treated. Substantial amounts of lime were found in it, almost entirely covering the upper layer of the bones.



Fig. 1. Ossuary 3009. Photo. M. Szyszka.

As a result, some skeletal material that reacted with lime was severely damaged, which prevented its use in the study. The determination of the exact size of the ossuary proved impossible, as the archaeological survey at the site was performed as a rescue excavation, hence its limited scope. Nevertheless, it provided valuable information on the history of Gdańsk.

Ossuaries offer unique and particularly challenging material for anthropological reconstruction. Due to the accumulation of loose and mixed bones in Ossuary 3009, the first step of anthropological analysis was to determine the number of individuals buried at the site. The details of the analysis are presented in a separate publication (Pudło et al. 2017). All of the bone types unearthed at the site were subjected to in-depth analysis. They were assessed in terms of their original location within the skeleton, body side (right- or left-hand), as well as their condition, size, shape, colour, age and sex of the individual, if permitted by the extant diagnostic features. In addition to main bones (frontal, occipital, temporal, mandibular, humeral, pelvic, femoral, tibial), the analysis involved smaller bones, such as vertebrae, clavicles, forearm bones, fibular bones, sternum, ribs, and bones of the palm and the foot. A total of 11,578 bones were examined, of which 72% were main bones and 38% small bones. Over 90% of them belonged to adult individuals, 9.4% to children, and only 0.5% to adolescents. The right- and left-hand side of the body were represented by similar number of bones. The most common bone fragments present in the material were used to estimate the so-called minimum number of individuals (MNI) (Szczepanek 2013). Since femoral bones are the longest and strongest parts of the skeleton (Bochenek and Reicher 1990; White and Folkens 2005), they are usually best-preserved in the material from excavations. The MNI for Ossuary 3009, estimated on the basis of (left) femoral bones, indicates that it contained the remains of at least 715 individuals. Interestingly, femoral bones were also the largest extant bone group in other ossuaries, providing the basis for determining MNI (Henneberg and Henneberg 2002, 2006; Georges 2007; Rost 2011; Szczepanek 2013; Blanchard et al. 2014).

Sex and age was determined on the basis of several types of bones. For this purpose, we used a set of diagnostic features commonly applied in anthropology. The research methods were discussed in the aforementioned publication, which also cites relevant literature (Pudło et al. 2017). The age of individuals with less precise features were estimated as members of relevant age categories according to mortality sequence. Sex was successfully determined for 140 men and 92 women. The remains of 283 individuals (37 children and 246 adults) were ultimately considered suitable for paleodemographic analyses.

The deceased were grouped by age on the basis of estimated age at death, both for the entire material and separately for adult men and women. Regression curves for changes in death rate across ages (trend lines) were also introduced to emphasize the shape of mortality distributions generated in this manner. In addition, cumulative mortality distributions depending on age at death were also created. In order to find whether they could be considered catastrophic or attritional, distributions for early mediaeval Gdańsk (Pudło 2016) and 14th century Lübeck (Prechel 1996) were prepared on the basis of data available from publications. The early mediaeval gravevard in Gdańsk dated from the mid-10th

century to 1227 represents pre-epidemic mortality profiles. The material from Lübeck includes victims of the Black Death from a mass grave at the city's Holy Spirit Hospital. Differences between distributions were compared by means of the Kolmogorov-Smirnov test (Blalock 1997).

Death rates in successive age categories were also used to create life tables. The tables were used for two model situations: the stationary population model, using the classical Halley method (Acsádi and Nemeskéri 1970) and the stable population model, adjusted for a non-zero value of natural increase (Pressat 1966; Holzer 1999).

Natural increase estimation for pre-historic and early historical populations is very difficult and challenging. The rate of natural increase corresponds to the difference between the number of births and deaths at a given time and place, and is standardised for population size. If death rate is the only reliable indicator, we may merely attempt to reconstruct the value of natural increase. In the present study a number of life tables were created for various model assumptions (different numbers of children per woman at the end of the reproductive period and different natural increase levels). Life tables built in this way were juxtaposed with Weiss's model life tables (1973). The table closest to the model was selected as the most representative one. In this way the most likely value of natural increase was determined.

As previously mentioned, the ossuary contained the remains of very few children. Insufficiency of child bones in skeletal material from excavation sites is a common issue. In this study the probable number of children in the ossuary was reconstructed according to the formula proposed by Henneberg (1977):

$$d_{0-14} = 1 - \left(\frac{2R_0}{R_{pot}U_0}\right)$$

where $d_{0.14}$ is the death rate at a pre-reproductive age, R_0 is the net reproductive rate, R_{pot} is the potential gross reproductive rate, and U_c is the number of children per woman after the reproductive period. It was assumed that an average woman from the analysed population gave birth to 5 or 6 children.

The potential gross reproductive rate $R_{\rm not}$ is one of the measures of the opportunity for natural selection by varied mortality (Henneberg 1975; 1976). It is a combination of death rate among adults from successive age categories (from age 15 up to and including the age of the oldest individual in the group, ω) and the so-call reproductive loss rate s_x . The s_x comes from the "fertility archetype" for non-Malthusian populations, in which reproduction occurs according to the natural fertility regime. It stands for the likelihood of an individual aged x not having the total number of children. The potential gross reproductive rate is expressed by the formula:

$$R_{pot} = 1 - \sum_{x=15}^{\omega} dx \times sx$$

Its value ranges from 0 to 1. It is a component of the biological state index I_{bs} (Henneberg and Piontek, 1975; Henneberg 1976; Stephan and Henneberg 2001):

$$I_{bs} = 1 - \sum_{x=0}^{\omega} dx \times sx$$

The structure of the index is the same as the structure of $R_{not'}$ and its value also

ranges from 0 to 1. However, it includes both adult and child death rates. The intensity of natural selection is inversely proportional to the value of the index.

Last but not least, the intensity of natural selection only among children is measured by Crow's I_m (1958). It stands for the proportion of children who did not survive until the reproductive age (P_d) to the children who reached the reproductive age (P_s) :

$$I_m = \frac{P_d}{P_s}$$

In life table categories, it corresponds to the proportion of the fraction of individuals deceased at a pre-productive age $(d_{0.14})$ to the fraction of individuals who reached the age of 15 (l_{15}) .

All calculations were performed by means of a standard Microsoft Excel 2016 spreadsheet, in which the authors of this study wrote their own subroutines. Life table parameters were computed using proprietary software "Population dynamics modelling for anthropological situations" by Maciej Henneberg and Martyna Steyn, developed for the purposes of their publication (Henneberg and Steyn 1994). The software was also applied to determine errors in life expectancy e_x and 95% confidence intervals for those values.

The significance tests in this study were carried out with the use of one level of significance p=0.05.

Results

Mortality distributions

Figure 2 shows age-at-death structure for the individuals from Ossuary 3009 in early modern Gdańsk in comparison with the corresponding structure from a non-epidemic graveyard in early mediaeval Gdańsk.



Fig. 2. Distributions of deceased by age in Gdańsk in the early Middle Ages and during the plague epidemic (oss. 3009, 15th–16th centuries).

One may easily notice a high degree of similarity between the two distributions, as indicated by the trend lines included. Dominant age categories of the deceased from both populations were as follows: 20–29.9; 30–39.9 and 40–49.9 years. There were few children, individuals in

the *Juvenis* and persons aged above 60. Slight differences between various age categories from both populations were not statistically significant. The equality of distributions was also confirmed by cumulative mortality distributions (Figure 3).



Fig. 3. Cumulative mortality distributions in Gdańsk in the early Middle Ages and during the plague epidemic (oss. 3009, 15th–16th centuries).

Adult-only mortality distributions were also analysed (Figures 4–7) due to the small size of fractions from both child categories, ranging from 5.6% to 7.5% of all deceased, which may suggest the absence of child remains at burial sites. Since the threshold of adulthood and the start of the reproductive age for historical populations is conventionally set at the age of 15, adult-only distributions charts included the 15–19.9 age category. Also in this case their shapes proved similar. Figures 4 and 5 juxtapose age-at-death distributions for men and women for Ossuary 3009.



Men Women — Wielom. (Men) — Wielom. (Women)

Fig. 4. Distributions of deceased men and women by age in Gdańsk during the plague epidemic (oss. 3009, 15th–16th centuries).



Fig. 5. Cumulative distribution of adult male and female mortality in Gdańsk during the plague epidemic (oss. 3009, 15th–16th centuries).

Peak death rate for both sexes was present in the age category of 30 to 39.9 years. Formally speaking, women prevailed in the higher age category, yet the differences were statistically insignificant (Figure 4). The similarity of the mortality profile for both sexes is also demonstrated by cumulative mortality distributions (Figure 5). No significant differences in mortality for adults from the ossuary and pre-pandemic early mediaeval Gdańsk were observed, either (Figures 6 and 7). In the light of Kolmogorov-Smirnov test, no statistically significant difference between the two distributions were found.



Fig. 6. Distributions of deceased adults by age in Gdańsk in the early Middle Ages and during the plague epidemic (oss. 3009, 15th–16th centuries).



— Gdańsk 15th-16th c., oss.3009 — • Gdańsk 10th-13th c.

Fig. 7. Cumulative distribution of adult mortality in Gdańsk in the early Middle Ages and during the plague epidemic (oss. 3009, 15th–16th centuries).

Nevertheless, such differences were noticed between mortality profiles for the Gdańsk Ossuary 3009 and the 14th century mass grave in Lübeck. This concerns both the entire population and adults (Figures 8–11). All differences between the distributions assessed by the Kolmogorov-Smirnov test proved statistically significant. In Lübeck, the proportions of the deceased in consecutive age categories were more even than in Gdańsk. In comparison to Gdańsk, the number of deaths was higher in age categories such as *Infans I* and *Infans II*, and, above all, in the *Juvenis* category. The Kolmogorov-Smirnov test also showed statistically significant differences across mortality profiles for Lübeck and pre-pandemic Gdańsk (10th to 13th century).



Fig. 8. Distributions of the deceased by age in Gdańsk during the plague epidemic (oss. 3009, 15th–16th centuries) and in the ossuary of plague victims in Lübeck from the 14th century.



Fig. 9. Cumulative mortality distribution in Gdańsk during the plague pandemic in the 15th–16th centuries (oss. 3009) and in the ossuary of plague victims in Lübeck in the 14th century.



Fig. 10. Distributions of the deceased adults by age in Gdańsk during the plague epidemic (oss. 3009, 15th–16th centuries) and in the ossuary of plague victims in Lübeck from the 14th century.



Fig. 11. Cumulative adults mortality distribution in Gdańsk during the plague pandemic in the 15th–16th centuries (oss. 3009) and in the ossuary of plague victims in Lübeck in the 14th century.

Natural increase reconstruction and life tables

Life tables were created based on the mortality sequence discovered in the analysed material. Table 1 contains base table parameters calculated for initial data assuming the stationary population model. One may easily notice that in the two first age categories death rates for children are implausibly low and amount to only 13%. This translates into overly "optimistic" values of other parameters in the life table. For example, the percentage of individuals who survived until the age of 7 exceeded 94%, and reached almost 87% for those who attained 15 years of age. Life expectancy for a newborn e_0 was 33.6 years. We shall return to the problem of insufficient child remains in excavation materials later on.

Age	D_x	d_{x}	l_x	q_x (per year)	L_x	T_x	$e_{_{x}}$	C _x
0	16	5.65	100.00	0.01	680.21	3358.73	33.59	20.25
7	21	7.42	94.35	0.01	725.09	2678.52	28.39	21.59
15	10	3.53	86.93	0.01	425.80	1953.43	22.47	12.68
20	63.29	22.36	83.39	0.03	722.10	1527.63	18.32	21.50
30	82.60	29.19	61.03	0.05	464.35	805.53	13.20	13.83
40	48.27	17.06	31.84	0.05	233.13	341.18	10.72	6.94
50	32.18	11.37	14.78	0.08	90.99	108.06	7.31	2.71
60-x	9.66	3.41	3.41	0.10	17.07	17.07	5.00	0.51

Table 1. Parameters of the basic life table for Gdańsk from the 15th-16th centuries (oss. 3009)

The next step involved the reconstruction of the low number of children in the ossuary. In this step, two following scenarios were assumed: 1/ women from the analysed population during their reproductive period gave birth to six children on average ($U_c=6$), 2/ because of the pandemic, reproductive dynamics was lower and after the reproductive period, a woman had on average five children ($U_c=5$). The first scenario was already discussed in a different study (Budnik, Pudło 2017). Table 2 presents values of life table parameters re-calculated for new age category durations. Table 3 contains the values of life table parameters calculated in the present study for $U_c = 5$. We can observe that the values of life table parameters changed considerably after the adjustment for under-representation of children. Life expectancy values for children e_0 fell by a statistically significant amount. Percentages of individuals surviving until their adulthood dropped, and infant mortality rate increased (Tables 2 and 3).

Table 2. Life table parameters for Gdańsk from the 15th–16th centuries (oss. 3009); stationary population model, Uc = 6

Age	D_x	d_x	l_x	q_x (per year)	L _x	T_x	e _x	C _x
0	184	36.29	100.00	0.05	572.98	2112.27	21.12	27.13
7	77	15.19	63.71	0.03	448.92	1539.29	24.16	21.25
15	10	1.97	48.52	0.01	237.67	1090.37	22.47	11.25
20	63.29	12.48	46.55	0.03	403.07	852.70	18.32	19.08
30	82.60	16.29	34.07	0.05	259.19	449.64	13.20	12.27
40	48.27	9.52	17.77	0.05	130.13	190.44	10.72	6.16
50	32.18	6.35	8.25	0.08	50.79	60.32	7.31	2.40
60-x	9.66	1.91	1.91	0.10	9.53	9.53	5.00	0.45

Age	D_x	d_x	l_x	q_x (per year)	L_{x}	T_{x}	$e_{_{X}}$	C _x
0	120.76	28.57	100.00	0.04	600.00	2426.42	24.26	24.73
7	55.92	13.23	71.43	0.02	518.52	1826.41	25.57	21.37
15	10	2.37	58.20	0.01	285.09	1307.89	22.47	11.75
20	63.29	14.97	55.83	0.03	483.47	1022.81	18.32	19.93
30	82.6	19.54	40.86	0.05	310.90	539.33	13.20	12.81
40	48.27	11.42	21.32	0.05	156.09	228.44	10.72	6.43
50	32.18	7.61	9.90	0.08	60.92	72.35	7.31	2.51
60-x	9.66	2.29	2.29	0.10	11.43	11.43	5.00	0.47

Table 3. Life table parameters for Gdańsk from the 15th-16th centuries (oss. 3009); stationary population model, $U_c = 5$

Estimated natural increase value (r) was determined by the juxtaposition of life tables prepared by the authors with model tables by Weiss (1973) as well as related biometric functions. The best fit was obtained for negative value r=-0.005 both for $U_c=5$ and $U_c=6$. This natural increase level corresponded to model tables MT:20.0–55.0 and MT:20.0–60.0,

assuming that $U_c=5$; and MT:20.0– 50.0, assuming that $U_c=6$. Figures below present values of life expectancy (e_x) , percentages of individuals surviving until consecutive age categories (l_x) and percentages of individuals living in consecutive age categories (c_x) against the values from model tables (Figures 12, 13 and 14).



Fig. 12. Life expectancy values e_x in Gdańsk from the 15th–16th centuries (oss. 3009) assuming r=-0.005 for $U_c=5$ (A) and $U_c=6$ (B) against the model Weiss curves.



Fig. 13. The survival curves I_x in Gdańsk from the 15th–16th centuries (oss. 3009) assuming r=-0.005 for $U_c=5$ (A) and $U_c=6$ (B) against the model Weiss curves.



Figure 14. Age structure of the living population c_x for Gdańsk from the 15th–16th century (oss. 3009) assuming r=-0.005 for $U_c=5$ (A) and $U_c=6$ (B) against the model Weiss curves.

Table 4 lists life table parameters for the stable condition of population due to the reconstructed value of natural increase (r=-0.005) for $U_c=5$, in Table 5 for $U_c=6$. Adjustments for a negative value of r led to decreased values of I_x . This is particularly noticeable where adjustment was made for the number of children at the level of $U_c=6$ (Table 5). In this situation even 45% individuals did not survive until adulthood. Life expectancy for a newborn did not reach 20 years. The decline in e_x in both child age categories proved statistically significant. Statistically significant were also differences in e_0 and e_7 obtained for both model scenarios, with the assumption that a woman gave birth to five or six children on average. Other e_x values did not vary.

Table 4. Life table parameters for Gdańsk from the 15th–16th centuries (oss. 3009); stable population model r=-0.005), U_c =5

Age	D_x	d_x	l_x	q_x (per year)	L_x	T_{x}	$e_{_{X}}$	C_x
0	118.66	31.57	100.00	0.05	589.49	2264.38	22.64	26.03
7	52.92	14.08	68.43	0.03	491.08	1674.89	24.48	21.69
15	9.16	2.44	54.34	0.01	265.62	1183.81	21.78	11.73
20	55.84	14.86	51.91	0.03	444.77	918.19	17.69	19.64
30	69.31	18.44	37.05	0.05	278.27	473.41	12.78	12.29
40	38.52	10.25	18.61	0.06	134.81	195.14	10.49	5.95
50	24.43	6.50	8.36	0.08	51.06	60.33	7.22	2.25
60-x	6.97	1.86	1.86	0.10	9.28	9.28	5.00	0.41

Table 5. Life table parameters for Gdańsk from the 15th–16th centuries (oss. 3009); stable population model (r=-0.005), $U_c=6$

Age	D_{x}	d _x	<i>l</i> _x	q_x (per year)	L_x	T_{x}	ex	C _x
0	180.80	39.48	100.00	0.06	561.80	1953.85	19.54	28.75
7	72.87	15.91	60.52	0.03	420.47	1392.05	23.00	21.52
15	9.16	2.00	44.60	0.01	218.00	971.58	21.78	11.16
20	55.84	12.19	42.60	0.03	365.04	753.58	17.69	18.68
30	69.31	15.14	30.41	0.05	228.39	388.54	12.78	11.69
40	38.52	8.41	15.27	0.06	110.64	160.16	10.49	5.66
50	24.43	5.33	6.86	0.08	41.90	49.52	7.22	2.14
60-x	6.97	1.52	1.52	0.10	7.62	7.62	5.00	0.39

Due to the lack of possibility to determine sex in the case of many child remains, sex-dependent differences in survivability were characterised only for adult individuals. Tables below present life table parameters for men (Tables 6 and 8) and women (Tables 7 and 9). With the exception of the survivability of 40-year-olds, no statistically significant differences in values of life expectancy were found. Value e_{40} was significantly greater in men than in women, both in stationary and stable population model for r=-0.005.

Age	D_x	d_x	l_x	q_x (per year)	L_{x}	T_{x}	e _x	C _x
15	6	4.29	100.00	0.01	489.29	2269.07	22.69	12.98
20	35.59	25.42	95.71	0.03	830.04	1779.79	18.59	22.02
30	49.2	35.14	70.29	0.05	527.21	949.75	13.51	13.99
40	20.94	14.96	35.15	0.04	276.71	422.54	12.02	7.34
50	21.99	15.71	20.19	0.08	123.39	145.82	7.22	3.27
60-x	6.28	4.49	4.49	0.10	22.43	22.43	5.00	0.60

Table 6. Parameters of life table for men from Gdańsk from the 15th–16th century (oss. 3009); stationary population model

Table 7. Parameters of life table for women from Gdańsk from the 15th–16th century (oss. 3009); stationary population model

Age	D_{x}	d_{x}	l_x	q_x (per year)	L_x	$T_{_{X}}$	$e_{_{X}}$	C _x
15	3	3.26	100.00	0.01	491.85	2204.67	22.05	13.28
20	24.08	26.17	96.74	0.03	836.52	1712.83	17.71	22.58
30	29.32	31.87	70.57	0.05	546.30	876.30	12.42	14.75
40	26.18	28.46	38.70	0.07	244.67	330.00	8.53	6.60
50	6.28	6.83	10.24	0.07	68.26	85.33	8.33	1.84
60-x	3.14	3.41	3.41	0.10	17.07	17.07	5.00	0.46

Table 8. Parameters of life table for men from Gdańsk from the 15th–16th century (oss. 3009); stable population model (r=-0.005)

Age	D_x	d_{x}	l_x	q_x (per year)	L_x	T_x	$e_{_{X}}$	C _x
15	5.50	4.73	100.00	0.01	488.17	2195.32	21.95	13.21
20	31.40	27.04	95.27	0.03	817.46	1707.15	17.92	22.12
30	41.28	35.55	68.23	0.05	504.49	889.69	13.04	13.65
40	16.71	14.39	32.67	0.04	254.76	385.20	11.79	6.89
50	16.69	14.38	18.28	0.08	110.92	130.44	7.14	3.00
60-x	4.53	3.90	3.90	0.10	19.52	19.52	5.00	0.53

Table 9. Parameters of life table for women from Gdańsk from the 15th–16th century (oss. 3009); stable population model (r=–0.005)

Age	D_x	d_{x}	l_x	q_x (per year)	L_x	T_x	$e_{_{X}}$	C _x
15	2.75	3.59	100.00	0.01	491.02	2146.03	21.46	13.47
20	21.24	27.76	96.41	0.03	825.28	1655.01	17.17	22.63
30	24.60	32.15	68.65	0.05	525.71	829.74	12.09	14.42
40	20.89	27.30	36.50	0.07	228.44	304.02	8.33	6.27
50	4.77	6.23	9.19	0.07	60.77	75.58	8.22	1.67
60-x	2.27	2.96	2.96	0.10	14.81	14.81	5.00	0.41

The structure of the living population was also reconstructed on the basis of the number of deceased from Ossuary 3009. Percentages of individuals living in consecutive age categories (c_x) are contained in Tables 1–9. The youngest children were the most numerous fraction among the living, followed by children from the 7–14.9 years of age category. Individuals aged 20–29.9 were the most sizeable category among adults.

Opportunity for natural selection through varied mortality

Values of natural selection measures were calculated for each model situation in this study (Tables 10).

Table 10. Measures of opportunity for natural selection through differential mortality in Gdańsk from 15th-16th century (oss. 3009)

	I_m	I_{bs}	R_{pot}
Stationary population model			
$U_c = 5$	0.718	0.400	0.687
$U_c = 6$	1.061	0.334	0.687
Stable population model ($r=-0.005$)			
$U_c = 5$	0.840	0.366	0.673
$U_c = 6$	1.242	0.300	0.673

The introduction of a negative natural increase value brought about a slight drop in biological state index $I_{bs'}$ gross reproductive potential ratio $R_{pot'}$ and an increase in Crow's I_m . Values of Crow's I_m fluctuated around 1, which suggests considerable force of natural selection against children. Quite striking are also low I_{bs} levels, ranging from only 0.3 to 0.4, and reduced R_{pot} values, which did not reach 0.7.

Discussion

This study analysed biodemographic effects of the plague pandemic found in the ossuary material from 15th and 16th century Gdańsk. As previously mentioned, the fact that the plague occurred is confirmed not only on the basis of historical accounts, bone remains dating and a unique type of burial, in which the remains were covered with a thick layer of lime, but also based on genetic tests.

The latter revealed the presence of the DNA of the plague bacterium, i.e. *Yersinia pestis*, in bone remains. What is more, partial reconstruction of the genome of *Yersinia pestis* from the remains of the rat discovered in the ossuary among human bones supplied novel information on natural sources of the disease and the way in which it spread over Europe (Morozowa et al. 2017; 2020).

A dangerous, acute and extremely contagious disease with high death rate, the plague was capable of causing catastrophic mortality. Catastrophic mortality profiles have been previously reported by many authors. For example, the graveyard of the victims of the Black Death at Royal Mint in London, dating to 1349. Age-at-death distributions for the deceased from the graveyard were starkly different from the distributions from various non-epidemic graveyards and model profiles for attritional mortality (Margerison and Knüsel 2002: Gowland and Chamberlain 2005). In addition, Gowland and Chamberlain (2005) confirmed the catastrophic distribution for the remains from Royal Mint using Bayesian statistics. They analysed mortality distributions according to the age of adult individuals and concluded that all age groups had been equally affected by the plague. The Penrith graveyard in England, where distributions from the plague period in 1597 and 1598 were compared with pre- and post-pandemic mortality distribution, could also be described as catastrophic. A dramatic rise in mortality rate in all age categories was reported during the epidemic, especially in children and individuals aged 15 to 44 (Scott et al. 1996; Scott and Duncan 2001). Finally, in a cluster of Black Death victims in mid-14th century Lübeck, a clear peak in the number of catastrophic deaths among children, adolescents and adults aged 20 to 30 was discovered (Prechel 1996). Although recent aDNA studies of some bones from this archaeological site suggest an outbreak of entheric paratyphoid fever rather than plague (Haller et al. 2021), the fact is that the plague was affecting Lübeck at that time and the mortality distribution there is clearly catastrophic.

In order to determine the character of mortality in the analysed osteological material, mortality distributions of individuals buried at the ossuary were evaluated. The intention was to ascertain whether the plague was selective in terms of age and sex. It has been repeatedly suggested in the literature of the subject that the plague did not kill indiscriminately; rather, it affected people at a specific age or of a given sex to a greater extent. As observed by Sharon De Witte (2010), most contemporary populations display differences in incidence and mortality rate depending on sex, and those differences tend to favour women. De Witte examined material from the East Smithfield Black Death cemetery in London and found that the proportion of deceased men to deceased women was 1.37:1.00. which means that most victims were male. Higher male mortality rate during the following plague event in London in 1603 was also reported by Holingsworth and Holingsworth (1971). In their study, the men-to-women ratio was 1.29 to 1.00 (0.89 to 1.00 before the plague). Holingsworth and Holingsworth concluded that excessive male mortality during the pandemic was attributable to lifestyle and social habits, greater mobility and lower standards of hygiene of the body and attire. In contrast, Curtis and Roosen (2017), studying plague victim records in southern Netherlands, found that both the epidemic from 1349-1351 and subsequent recurrent epidemic events until 1450 killed more women than men. In non-epidemic years, both in cities and villages, male deaths prevailed: male to female death ratio was respectively 1.20:1.00 and 1.17:1.00. In the years of the epidemic it dropped to 0.89:1.00 in cities and 0.95:1.00 in villages. Female deaths were also dominant in Penrith (England). The proportion between the sexes among plague victims was 1.37:1.00 to women's disadvantage, whereas before and after the epidemic it corresponded to a generic 1.00:1.00 (Scott et al. 1996).

There were more adult men than women in our material, with the proportion of 1.52:1.00. Still, this does not have to be indicative of selective mortality due to the plague. Note that the proportion of men to women had already been strongly distorted in pre-pandemic Gdańsk and significantly diverged from the normal, given as 1.00:1.00. It was 1.70:1.00 in the early Middle Ages, and as much as 2.47:1.00 in the late Middle Ages (Pudło 2016). It seems logical that a harbour and commercial city like Gdańsk attracted crowds of men from Poland and overseas. Female and male mortality distributions (Figures 4 and 5) obtained in this study do not differ to a statistically significant extent. They also do not vary significantly from adults mortality distributions in pre-pandemic Gdańsk (Figures 6 and 7). Although formally the percentage of women who died at the age of 40 to 49.9 years was larger than men, such excessive mortality may have been due to complications during pregnancy, birth and confinement in the final phase of the reproductive period. As late as the 19th century, in rural Greater Poland nearly 1/4 of women in the reproductive age died after childbirth, and the percentage increased with age (Budnik 2005). The equality of mortality distributions between individuals from Ossuary 3009 and early mediaeval Gdańsk was also obtained for the entire population (Figures 2 and 3). The extremely low percentage of child deaths is a striking phenomenon in both groups. Underrepresentation of child remains in osteological material is a quite frequent phenomenon, due to various reasons. A key role is played by taphonomic processes, leading to quick decomposition of delicate bones due to their chemical structure (considerable organic content). Inhumation rites and the manner in which bones are excavated are also of significance (Krenz-Niedbała 2008; Budnik and Henneberg 2009; Pudło 2016; Budnik and Pudło 2017). The two last reasons could be vital with regard to the material analysed in this study. Generally, ossuaries are secondary mass graves, in which human remains transferred from another location were placed. Some bones could have been easily "lost" in the course

of such activities, involving intentional selection due to limited burial space or by accident. In any case, one cannot see any excess infant mortality like the one in the Black Death ossuary in Lübeck (Prechel 1996) or other burial sites representative of catastrophic mortality (e.g. Gowland and Chamberlain 1996; Scott et al. 1996; Margerison and Knüsel 2002).

Certain researchers suggest that the plague was selective for health. Excess mortality caused by the plague was attributed to certain physiological stress markers in bones. It was reported that previous stress increased risk of death during the pandemic (De Witte 2009, 2010a; Crtis and Roosen 2017). It seems that this does not hold true for the analysed ossuary. Kozłowski et al. (2017) concluded that in the analysed ossuary non-specific stress markers such as cribra orbitalia occurred less frequently than in other, later Gdańsk ossuaries. Moreover, there were fewer cases of tooth decay, degenerative alterations in the spine and joints. Based on palaeopathological studies, the authors made a careful suggestion that the material was characterised by a relatively low frequency of most of the analysed pathological alterations. Few alterations were also reported for pre-pandemic Gdańsk: not only was the level of physiological stress markers such as cribra orbitalia or enamel hypoplasia low, but the degree of sexual dimorphism in stature and other morphological traits was high, indicating absence of intense environmental and cultural stressors. In addition, if we consider no significant differences in male and female diets and lack of severe lesions in the mastication apparatus and other systems, the claim that living conditions and health of the inhabitants of mediaeval Gdańsk were relatively good compared to other mediaeval populations seems justified (Pudło 2016).

This would partly explain why the mortality profile reported for the 15th/16th century ossuary was attritional rather than catastrophic. This explanation is confirmed by the absence of significant differences in mortality distributions for the ossuary and for population from early mediaeval Gdańsk, as well as the absence of such differences in comparison to mortality distributions from Lübeck (Figures 8–11). Some mortality distributions for the plague epidemic obtained by other authors seem to confirm that a non-catastrophic mortality model in the case of the plague is possible. De Witte (2010 b) concludes that in spite of its destructive nature the disease generated mortality patterns which were not entirely different from those characteristic of normal, non-epidemic mortality in the Middle Ages. This finding is in line with Kacki (2017) and Bramanti et al. (2018), who did not observe any selectiveness for various biological traits in the plague's victims, which means that the plague killed indiscriminately, regardless of age, sex and health.

It also seems that the attritional mortality profile of Ossuary 3009 may be due to other factors. The ossuary has a wide dating range. Although ¹⁴C dating of bone samples from Gdańsk examined for plague traces was refined to the period from 1425 to 1469 (Morozowa et al. 2020), it relates to only some bones. Archaeological dating is very wide and includes, as previously mentioned, the 15th and the 16th centuries. We may, thus, reasonably expect that the ossuary may have contained bones of plague victims, as well as individuals deceased in non-epidemic periods.

This scenario would account for other results obtained in biodemographic and genetic analyses. Since mortality profiles

found for early modern Gdańsk were not catastrophic, it seemed advisable to reconstruct the number of children missing from the ossuary. The reconstruction method applied was based on mortality distribution for adults, reproduction patterns characteristic of non-Malthusian populations, reproductive rate and the number of children per each woman after the end of the reproductive period (Henneberg 1977). Following the adjustment, the probable number of children in the ossuary should range from 41.80% to 51.48% of all deceased individuals, depending on whether we assume that an average woman who survived until the end of her reproductive period had five or six children $(U_e=5 \text{ or } U_e=6)$. Based on this reconstruction of the number of the deceased, life tables were created for the stationary population model and stable population model due to a non-zero natural increase value. Determining natural increase solely on the basis of the number of deceased individuals from consecutive age categories is considerably difficult, often impossible. For this purpose Weiss's (1973) model tables were used in this study. As previously mentioned, the best fit between the authors' life tables and model tables was obtained for natural increase r = -0.005. Here, a remarkably good fit was obtained with Weiss's MT 20.0-55.0 and MT 20.0-60.0, assuming that $U_c=5$, and MT 20.0-50.0, assuming that $U_c = 6$ (Figures 12-14). A slightly negative value of natural increase in epidemic conditions may be surprising. Note, however that the value probably results from natural movement during epidemic and non-epidemic periods. Also, we do not know which type of the plague was prevalent in the population. As mentioned in the introduction, not every form of the disease was equally virulent, yet, overall, in the age of insufficient medical knowledge and no antibiotics, all of its forms were dangerous. Furthermore, some authors pointed to the possibility of achieving a certain degree of immunity by population who had previous contact with the disease, as well as potentially reduced virulence due to changes occurring in the pathogen over time (Ampel 1991; Gowland and Chamberlain 2005). Historical accounts reveal that the plague struck Gdańsk as early as the mid-14th century and kept returning in later periods (Kizik 2012; Możejko 2012; Trzoska 2012).

Negative natural increase translated into altered life table values. In particular, life expectancies e_{y} dropped. For example, a newborn's life expectancy e_0 decreased to 22.64 or even 19.54 depending on the model assumptions (U = 5 orU = 6). Mean life expectancy of a 20-yearold was 17.7 years. The conventional threshold of adulthood (fifteenth year of life) was reached by 44.6% to 54.3% of individuals, and only slightly over 7% of people survived until their 50th year of life (Tables 4 and 5). Such low natural increase led to the situation in which all e_{y} values, except for e_0 and e_7 , proved statistically insignificant regardless of the assumed model scenario. Finally, the authors' analyses reveal a population which could be placed within the lower limits of many mediaeval populations (Gejval 1960; Henneberg and Strzałko 1975; Henneberg and Puch 1989; Kaźmierowska 1989 [1990]; Budnik et al. 2004; Budnik, Fiszer, Białas 2009; Budnik and Henneberg 2009; Kozłowski 2012; Pudło 2016). Life expectancy for a newborn in early mediaeval Gdańsk was slightly over 22 years, and less than 17 years for an adult individual in the Adultus age category (Pudło 2016). In early mediaeval Espenfeld (Germany), a newborn could be expected to live 19 years on average, and a 20-year-old only less than 15.5 years (Bach and Bach – qtd. in: Henneberg and Strzałko 1975). For the Swedish Westerhus graveyard dated to the 11th/mid-14th century, corresponding values amounted to $e_0=19.9$ years, and $e_{20}=21.1$ years (Gejval 1960). For the early mediaeval site Dziekanowice 22, assuming the stationary population model ($e_0=19.9$ years; $e_{20}=17.4$ years), albeit after adjustment for positive natural increase, the values rose by several years ($e_0=25.4$ years; $e_{20}=19.4$ years; Budnik et al. 2004).

In modern times we often notice an extension of human life expectancy. This does not concern only the values of newborn life expectancy, which for a long time, due to immense infant mortality, were incredibly low. Nevertheless, successive e, values increased markedly. Although no clear improvement can be seen in 18th century Gdańsk ossuaries (a newborn had 22 years of life remaining, and a 20-year-old nearly 17.5 to 20.5 years; Budnik, Pudło 2017), examples such as Posada Rybotycka from 17th/18th century reveal e₂₀ rising to 24.5 years, Jaksice from the 15th/18th century show that the value went up to reach 25.9 years, and in Słaboszewo, dated to the period from the second half of the 14th century to the first half of the 17th century, the value increased to 27 years (authors' own calculations based on: Piontek and Malinowski 1976; Piontek 1977; Piontek 1981). Therefore, the impact of the plague was also visible in our results for Ossuary 3009. We may speculate whether low values reported for the Gdańsk ossuaries from the 18th century were affected by the last wave of the pandemic, which reached Gdańsk in 1709.

A similar picture emerges from the analysis of opportunity for natural selection (Table 10). A striking feature are extremely low values of biological state index I_{hsl} ranging from 0.3 to 0.4. This means that only 30% to 40% individuals in the analysed population were well-adapted to the environmental and cultural conditions and reached reproductive success, passing on their genes to the next generation's gene pool. As many as 60% to 70% individuals did not have this opportunity; they had been eliminated along with their genetic outfit by natural selection. Low I_{bs} values were caused by high child mortality rate. Intensive natural selection among children is confirmed by relatively high Crow's I_m values. However, natural selection worked not only against children, but also, to a lesser extent, against adults. Values of potential gross reproductive rate R_{pot} demonstrate that reproductive success was achieved by less than 70% adults, and 30% were eliminated by natural selection. The situation observed in Ossuary 3009 was typical of human populations for an exceedingly long time (Piontek 1979; Budnik and Henneberg 2009). Undoubtedly, epidemic infectious diseases which troubled humans could have been a key selective factor. For 15th to 16th century Gdańsk, this was the plague.

Conclusions

The conclusions arising from the above analyses could be summarised as follows:

1. Mortality profiles in the material did not reveal a clearly catastrophic character. Age-at-death distributions approximate those for pre-pandemic Gdańsk and do not differ statistically from the latter. However, they do differ significantly from catastrophic distributions for plague-stricken 14^{th} century Lübeck. The authors found no selectiveness of the disease for sex or age of the victims. The shape of distributions, approximating that of non-epidemic ones, were probably due to a wide period of time to which the ossuary was dated (15^{th} to 16^{th} century). As a consequence, apart from individuals who were clearly plague victims, as confirmed by DNA tests, the ossuary could have also contained remains of people buried in non-pandemic periods.

- 2. The mixed epidemic/non-epidemic character of the ossuary seems to be confirmed by the value of natural increase. It was reconstructed at r=-0.005 according to Weiss's model tables. The value is only slightly negative, yet not exceptionally low, which could be expected from an epidemic-only ossuary given the plague's notorious virulence and high death rate.
- 3. Negative natural increase had its effect in slightly worsened life table parameters, especially with regard to life expectancy e_x . They approached values typical of the Middle Ages rather than the early modern period. Note, however, that the adjustment for natural increase only resulted in radically lower e_x values only for children. Differences in other e_x values proved statistically insignificant. In the analysed material, the effect of the plague, although undoubtedly marked, was not dramatic.
- 4. Measures of opportunity for natural selection calculated in this study dropped slightly as a result of negative natural increase. However, they were generally contained within the limits typical of early historical populations. The edge of natural selection affected mostly children. This is confirmed by

low values of biological state index I_{bs} and increased levels of Crow's I_m . Natural selection worked against adults, too, although to a smaller extent. This finding is supported by reduced values of potential gross reproductive rate R_{pot} . The above picture is common for many historical populations. Undoubtedly, the plague acted as an important pool gene regulator, a tool of natural selection in the analysed group. Still, one must not exclude various socio-cultural factors related to the capabilities, organisation and living conditions related to life in a rapidly growing early modern harbour city.

The Author's contribution

AB was the originator of the work and the main contractor of the project. She participated in the elaboration of osteological materials, and performed all paleodemographic, genetic and statistical analyzes. Moreover, she made the diagrams and wrote a draft and final version of the work. AP was the contractor for the project. She elaborated a large part of the osteological material and provided pictures of the bones. She participated in writing some fragments of the draft of the work. Both authors carefully read and approved the final version of the manuscript.

Conflict of interest

The authors declare that there is no conflict of interest.

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Usefulness of the analysis of the average ridge width of fingerprints in archaeological research

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ABSTRACT: Skin ridges (dermatoglyphs or fingerprints) are a characteristic pattern of sulci on the skin of primates which appear on the entire hand palm and on the soles of the feet. Fingerprints are unique, irremovable and invariable which allows bio-identification of specific individuals. The aim of the study was to investigate the usefulness of the analysis of the average width of the skin ridges in archaeological research by analyzing the foot and hand prints found on 7 artifacts dating from the Middle Ages.

An attempt was made to describe the preserved skin slate prints, as well as details of the construction of the prints. The fingers used in forming the pottery were recognized. Attempts were made to read the context of individual impressions. In the study, the following features were analyzed: legibility and suitability of the left imprint for the analysis, the possibility of determining the type of figures (whorls, loops, archs), the possibility of determining the minutiae, the density of the skin ridges left on the surface of the ceramics and the width of the skin ridges left on the surface of the ceramics. The classification of fingerprint minutiae proposed by Czesław Grzeszyk (1970) was used as well as classification of epidermal ridges peoposed by Lestrange (1953) and modified by Bochenska (1964) and Rogucka (1968).

Analysis of the material allowed to conclude that imprints left on building ceramics can be as useful as those imprinted on utilitarian ceramics, despite differences in the composition of the raw material used in production. However, they require more skill to examine, as the impressions are often incomplete which may be related to the fillers added to the mass. Minutiae were evident on the impressions examined, and single bifurcation was the most common form. The tactile figure most commonly found on the fingertip impressions was the loop. It was not possible to delineate Galton lines due to the wiping of the triple ray. Due to the high illegibility of the prints, it was assumed with a high degree of uncertainty that the fingerprints belonged to adults, but the gender could not be determined. Noteworthy were the prints printed on the brick belonging to children. On the mentioned artifact, there is a footprint of a child aged 1–3 years and a handprint of a crawling infant, which allowed us to conclude that the children were under the care of craftsmen.

KEY WORDS: average ridge width, epidermal ridges, archeology, fingerprints, dermatoglyphs, derma



Introduction

Epidermal ridges, also known as dermatoglyphs or fingerprints, are a characteristic pattern of sulcion the skin of mammals belonging to the group of Primates. They appear on the fingertips, as well as on the entire palmar surface or on the soles of the feet. Human fingerprints are located on the largest organ of the body – the skin (Latin: cutis, Greek: derma). Montagna and Parakkal (1974) define human skin as "the monumental façade of the human body." The skin is also an organ which possesses a significant diversification of morphological structures (including epidermal ridges). This in turn, enables bio-identification (Montagna and Parakkal 1974).

The aim of the research was to see if footprints and hand prints left on medieval building ceramics might be used to analyze the organization and working conditions of craftspeople. There was an attempt to describe preserved imprints of the skin ridges, as well as the details of the prints' structure. Then, based on the prints left, efforts were made to determine the sex and age of the fingerprint owner. First, the paper provides the basic information essential for futher understanding of the analysis of epidermal ridges. Some basic information about the structure of the skin are provided along with short historical overview of the research on dermatoglyphs. The use of dermatoglyphs in dactyloscopy is also described, taking into consideration the quantitative features of the dermatoglyphs and their role in determining sex and age of particular individuals. The secoond part of the paper provides information on the research on footprints and hand prints

left on medieval building ceramics. The aim was to determine how they might be used to analyze the organization and working conditions of craftspeople and how the analysis of the average ridge width of fingerprints may be used to determine sex and age of the owner of the imprints.

Structure of the skin

Human skin is made up of different types of cells that perform different functions. The skin consists of three main layers. The epidermis is on the outer surface, and the dermis underneath it. Below the dermis there is the third major layer, a fatty layer of varying thickness, beneath which there is a discontinuous flat flap of skeletal muscle that separates the rest of the body's tissues from the integument (Wooi and Lau 2015) (Fig. 1). In turn, the epidermis itself can be further subdivided from the outside to the inside into the horny layer (stratum corneum), granular layer (stratum granulosum), clear layer (stratum lucidum), prickle cell layer (stratum spinosum) and basal layer (stratum basale, also known as stratum germinativum) (Wooi and Lau 2015). The reproductive layer (stratum germinativum) is responsible for the constant renewal of the epidermis, as this is the place where the cells replicate (Moszczyński 1997).

It is on the surface of the hands and feet that the characteristic sulcis, associated with the presence of fingerprints, are distinguishable. The part of the skin where the fingerprints are visible is called friction skin. The dermis of the hands and feet have "prominences in the form of double rows of pimple-like forms" (Moszczyński 1997). These protrusions are skin papillas, and each double row of these is located under a furrow that is found between two fingerprints on the epidermis. On the surface of the fingerprints, there are also pores, i.e. numerous holes through which sweat escapes. The height of the fingerprints varies from 0.1 to 0.4 mm and depends, among other things, on sex, age, height and body composition. In turn, the width of these lines ranges from 0.2 to 0.7 mm (Moszczyński 1997).



Fig. 1. A cross-sectional representation of human skin (Shier et al. 2010).

Epidermal ridges start to be formed around the seventeenth week of gestational age. In the later stages of life, they may increase in size, but their shape remains unchanged. Importantly, however, fingerprints may be deformed or rubbed off under the influence of various factors, such as physical work, local lesions such as clavi or malignancies. These factors can lead to slight blurring of the ridges and minutiae¹ (Grzeszyk 1992). An example of this is by Grzeszyk (1992) who describes the research of Welker, who took his fingerprints and repeated this procedure after 41 years. This experiment showed that the prints, after such a period of time, differed only in the average width of the ridges, while the overall texture did not change. This confirms that the ridges themselves are immutable, while their width may actually alter over the years (Fig. 2).

¹ Major features of a fingerprint.



Fig. 2. Imprint of Walker's left hand at the age of 34 (1856) and at the age of 75 (1897) (Grzeszczyk 1992).

Historical overview

Identifying individuals by fingerprinting has become common practice today and serves as an invaluable tool for identifying people all over the world. This applies both to law enforcement, forensics and archaeological research, but also to large-scale automatic fingerprint identification systems such as devices (e.g. smartphones) or services with access control (e.g. electronic banking). However, it is worth noting that fingerprints and handprints were used as means of identification already in early cultures (Szczepański 2019).

The first observed examples of dermatoglyphs date back to the Neolithic period. The oldest discovered prints are petroglyphs – prehistoric paintings carved in the rocks (Szczepański 2019). An example of such a petroglyph is a drawing of a hand with sulci-like lines found on the shores of a lake in Nova Scotia. Fingerprints can also be found on ceramic products from excavations. These types of imprints are formed accidentally during the formation of these vessels. It has been identified that fingerprints are well preserved on soft clay (Szczepański 2019).

Fingerprints were also intentionally used in the Far East. Fingerprint imprinting on contracts or other legal documents was aimed at identifying individuals and was equivalent to the current use signatures or seals (Szczepański 2019). The unique fingerprint pattern in the form of an imprint on a clay seal, along with the author's name, served to authenticate documents written on bamboo rolls in China during Qin and the Han Dynasties from 221 BC to 220 AD. The earliest example of the use of handprints as evidence in a detective investigation is the examination of a burglary site described in a document from the Qin Dynasty (221-206 BC) (Szczepański 2019).

Initially, scientists interested in fingerprints focused more on their structure than on the analysis of possible identification functions. Although many researchers have described the structure of fingerprints, the work of Johannes Evangelista Purkinje, professor of the University of Wrocław, Commentatio de Examinae Physiologico Organi Visus er SystematisCutanei (1823), should be particularly emphasized. He was the first who not only described fingerprints but created their scientific classification by distinguishing nine types and patterns of fingerprints existing on fingertips: arches, tents, two types of loops and five types of whorls (Moszczyński 1997). The very foundations of modern dactyloscopy, i.e. the science of dermatoglyphs, began to be developed only during the last two decades of the nineteenth century.

A person who had a great influence on the development of the research on fingerprints was Henry Faulds. Faulds who inspired by prints on prehistoric ceramics, began exploring fingerprints to a large extent. His main work was the discovery of the indestructibility of fingerprints – damaged or removed epidermis, after regeneration, has the same shape of fingerprints as the epidermis prior to being damaged. Currently, the feature of indestructibility, apart from individuality and invariability, is one of the basic features of dermatoglyphs (Faulds 1923).

An important figure in the development of dactyloscopy was Francis Galton. In his research, he proposed a system for classifying fingerprint patterns, dividing them into arches, loops and whorls (Moszczyński 1997). His greatest achievement, however, was demonstrating the durability of fingerprints and proving that changes in their course arise only through damage to the reproductive layer of the epidermis. However, an equally important result of his research was to show that there cannot exist two individuals with an identical pattern of fingerprints, which Galton supported with a mathematical formula. The segment

connecting the center of the fingerprint pattern with the delta is now named after him (Galton 1893).

The use of dermatoglyphs in dactyloscopy

Dactyloscopy studies the skin texture on palms and soles. A fingerprint consists of a set of parallel lines that form a unique pattern for each individual. When observing a imprint, we can distinguish ridges (i.e. lines touching the surface) and valleys (these are the spaces between two ridges) (Machhout 2017). Each fingerprint also has a set of singular (centers and deltas) and local points. While the centers correspond to the locations of the ridges convergence, the deltas correspond to the locations of their divergence (Machhout 2017). From a variety of continuous line layouts, including their beginnings, endings, segments and dots the so-called minutiae are formed, which are also characteristic features of the structure of fingerprints (Machhout 2017). Fingerprint identification and examination itself are based on three basic principles:

Individuality (uniqueness) - fingerprints are unique for each individual. Therefore, it can be assumed that no two individuals have the same fingerprint pattern. According to Wójcik et al. (2014), fingerprint studies together with statistical studies show that the probability of the appearance of two identical dermatoglyphs is 1 to 64 trillion. Moreover, fingerprints may also differ in individual fingers of the same person. It is this uniqueness that allows for the identification of individual people. For this purpose, the following are used: minutiae (characteristic features of the structure of fingerprints), general arrangement of fingerprints, distribution and shape of pores, irregular shape of the edges of fingerprints (Moszczyński 1997).

- Indestructibility (irremovability) the durability of fingerprints is ensured by the regenerative and replication capacity of cells. This allows the regeneration of fingerprints in exactly the same pattern, despite various types of damage to the epidermis (Moszczyński 1997).
- Invariability epidermal ridges formed up to the sixth month of life remain unchanged until the death of the individual (Moszczyński 1997).

Despite the fact that fingerprints are characterized by a diverse and unique structure, they can be classified according to specific patterns and configurations. Each area has unique regional and individual structural differences that are not found anywhere else for the same or any other person (Montagna and Parakkal 1974). However, the patterns of the configuration of the ridges and sulci can be grouped according to some common characteristics. For example, it is important to remember that ridges are wider in males than in females (Montagnaand Parakkal 1974). Fingerprints also create recognizable patterns that can be divided into three groups according to their combinations: arches, tent arch patterns (tents), right loop patterns (right loops), left loop patterns (left loops) and whorl patterns (whorls) (Fig. 3) (Stolarek 2008). Thus, dermatoglyphs constitute a peculiar unique personal identification pattern used for identification in dactyloscopy (Fig. 4).



Fig. 3. Major patterns of dermatoglyphics: (A) whorl, (B) arch, (C) loop, (D) combined form (Montagna and Parakkal 1974).



Fig. 4. Fingerprints; the imprints of dermatoglyphics (Montagna and Parakkal 1974).

The quantitative features of the dermatoglyphs are based on the average width and density of the ridges. Both the average width of the skin ridges and their average density depend on the sex and age of the individual. These features are crucial in identifying dermatoglyphs in relation to individuals, as they assist in determining a person's sex and age. Apert from the aging process, fingerprints undergo visual and chemical changes that affect the morphology of the skin ridges (Montagna and Parakkal 1974). The technique of capturing differences between males and females, both in terms of the thickness of the ridges and the distribution of minutiae itself, is morphometry. In the context of research on skin ridges, this technique includes: shape and size

analysis, and can be useful for assessing the age of an individual from fingerprints, in particular by identifying and analyzing the average width of ridges that change over the course of life. During adolescence, the ridges expand, which is caused by the increase in body size during progressive ontogenesis. In early adulthood, quantitative characteristics do not increase significantly. Under the influence of involution processes, these averages begin to change again (Alcaraz-Fossoul 2018). Research shows that aging causes narrowing of the ridges and loss of their continuity, while their density increases. In addition, due to aging, the ridges blur and become less visible, which may also be influenced by environmental factors such as the degree of exposure to natural light, the type of sweat, temperature and humidity (Alcaraz-Fossoul 2018).

When measuring the width and density of the ridges, the length of the previously determined and measured section and the number of ridges passing through this section is taken into account. On the fingertip, this segment is known as Galton's line which is drawn from the center of the pattern to the triple fork. In the case of whorl patterns, it is usually the line that has the greater number of ridges that is selected, while in the case of an arch pattern, it is a randomly defined section running perpendicular to their course, e.g. 2 cm long. In the case of measurements on the sole, several sections of a fixed length are marked - e.g. 2 cm in a few places. These can be the heel surface of the foot and the toe surface of field IV (the surface of the foot place under toes four and five). In the case of measuring the average width of the ridges of the skin, the quotient of the number of ridges and the length of the measured section, given in centimeters, is calculated. However, in measuring the average density of the skin ridges, the quotient of the length of the measured section (given in millimeters) and the number of ridges is used. These measurements are used to determine the sex and age of a person, which is useful in forensic examinations. They can also be useful for archaeological research in order to access more information about the person who made an artefact.

Materials and methods

Imprints left on building ceramics can tell researchers a lot about the production process itself, as well as about the producers themselves and the organization of their workplace. However, there are not many studies focusing on the analysis of fingerprints left on building materials (Wałecki 2019). The use of the average width of the ridges has been based on research on imprints left on ceramics. The aim of the study was to determine the usefulness of footprints and hand prints left on medieval construction ceramics in the assessment of the organization and conditions of craft work. An attempt was made to describe the preserved imprints of the skin ridges, as well as the details of the structure of the prints. Then, on the basis of the prints left, efforts were made to assess the sex and age of the owner of the fingerprints.

The gathered research material contained seven fragments selected from a collection of medieval bricks. The fragments which were selected contained clear traces of human handprints and footprints. The material was excavated from debris from the former Gomółka Hill in Wrocław, now known as the Anders Hill. The material was cataloged and analyzed at the turn of May and June 2019 (Wałecki 2019).

During the examination of the fingerprints, the following features were analyzed: legibility and suitability of the left imprint for the analysis, the possibility of determining the type of figures (whorls, loops, archs), the possibility of determining the minutiae, the density of the skin ridges left on the surface of the ceramics and the width of the skin ridges left on the surface of the ceramics. The classification of fingerprint minutiae proposed by Czesław Grzeszyk (1970) was used as well as classification of epidermal ridges proposed by Lestrange (1953) and modified by Bochenska (1964) and Rogucka (1968) (Fig. 5, Fig. 6).

The analysis showed that on artifact no. 1 there are: fragments of fingerprint II, III and IV (Fig. 7). Artifact no. 2 shows fragments of fingerprints II, III and IV (Fig. 8). Artifact no. 3 contained traces of fingers II, III and IV (Fig. 9), while artifact no. 4 of fingers II and III (Fig. 10). Artifact no. 5 shows most probably a partial fingerprint of the finger I of the left hand (Fig. 11), while artifact no. 6 shows an imprint of the child's left foot (Fig. 12) (Wałecki 2019).

NAZWA MINUCJI		SYMBOL	SYMBOL WZÓR		
w języku polskim w języku łacińskim					
Początek	Initium	I	1)))	
Zakończenie	Terminatio	Т	\gtrsim		
Rozwidlenie pojedyncze	Bifurcatio simplex	B1	\sim	\sim	
Rozwidlenie podwójne	Bifurcatio duplex	B2	-00	X	
Rozwidlenie potrójne	Bifurcatio triplex	B3	~		
Złączenie pojedyncze	Iunctio simplex	In1	>	5	
Złączenie podwójne	Iunctio duplex	In2	3	\rightarrow	
Złączenie potrójne	Iunctio triplex	In3	27		
Haczyk	Unculus	U		~~	
Oczko pojedyncze	Ocellus simplex	01	-0-	-0-	
Oczko podwójne	Ocellus duplex	02	-00-	->	
Mostek pojedynczy	Ponticulus simplex	P1	T	H	
Mostek bliźniaczy	Ponticulus gemellus	Pq	Ŧ	Ŧ	
Punkt	Punctum	Pm			
Odcinek	Segmentum	S			
Styk boczny	Iunctura laterialis	Ilat			
Linia przechodząca	Linea intermittens	Li	=/=		
Skrzyżowanie	Decussatio	D	\sim	\sim	
Trójnóg	Tripus	Tr	\prec	-E	
Linia szczątkowa	Linea rudimentalis	Lr	\sim	~	
Minucja typu "M" "m"	Minutia "M" formis	М		2	

Fig. 5. Types of epidermal ridges minutiae on fingertips proposed by Grzeszyk (Moszczyński 1997).



Fig. 6.: Classification of epidermal ridges on fingertips (according to Lestrange 1953 – as modified by Bochenska 1964 and Rogucka 1968) (Grzeszyk 1992).



Fig. 7. Image of the artifact no. 1 (Wałecki 2019).



Fig. 8. Image of the artifact no. 2 (Wałecki 2019).



Fig. 9. Image of the artifact no. 3 (Wałecki 2019).



Fig. 10. Image of the artifact no. 4 (Wałecki 2019).



Fig. 11. Image of the artifact no. 5 (Wałecki 2019).



Fig. 12. Image of the artifact no. 6 (Wałecki 2019).



Fig. 13. Image of the artifact no. 7 (Wałecki 2019).

Results

In the above-mentioned research, only three of the imprints were found to have a 5 mm section with an undamaged surface of the imprint. On artifact no. 2, fingerprint III, more precisely the fingerprint of the fingertip segment, had 10 skin ridges per 5 mm segment. Artifact no. 4 and fingerprint III had 9 skin ridges by 5 mm and artifact no. 5, most likely showing fingerprint I, had 9 skin ridges by 5 mm (Table 1). The degree of material shrinkage during drying and firing was not known due to the chemical composition. No information was found on the shrinkage of building ceramics during its manufacture, therefore, it was difficult to determine the age of the craftsmen. If there had been an artifact with several distinct fingerprints from one craftsman, the likelihood of determining the age and sex of the owner of the print would improve. There were too few prints in the study, which severely limited the accurate estimation of the craftsman's sex and age (Wałecki 2019).

artifact number	type of the imprint	number of skin ridges over 5mm section
artifact no. 1	fingerprint II	It is not possible to determine the density or width of epider- mal ridges due to the deficiencies.
artifact no. 1	fingerprint III	It is not possible to determine the density or width of epider- mal ridges due to the deficiencies
artifact no. 1	fingerprint IV	It is not possible to determine the density or width of epider- mal ridges due to the deficiencies
artifact no. 2	fingerprint II	It is not possible to determine the density or width of epider- mal ridges due to the deficiencies
artifact no. 2	fingerprint III	10 skin ridges
artifact no. 2	fingerprint IV	The fingerprint is unreadable, was preserved during a move- ment, making it impossible to read the fingerprint.
artifact no. 3	fingerprint II, III, IV	The imprint represents movement. There is some mortar residue in the center of the imprint.
artifact no. 4	fingerprint II	6 skin ridges
artifact no. 4	fingerprint III	9 skin ridges
artifact no. 5	fingerprint I	9 skin ridges
artifact no. 6	left foot imprint	no traces of skin ridges
artifact no. 7	hand imprint	no traces of skin ridges

Table 1. Analysis of artifacts

The analysis showed the following results: While fingerprints of II, III and IV fingers were the most common, it was not found that they belonged to the same person. The most common minutiae in fingerprints was single bifurcation. They were located on fingerprint II and IV on artifact no. 1, fingerprint II on artifact no. 2, fingerprint II and III on artifact no. 4. The prints on artifacts 2, 4 and 5 were assumed to belong to an adult which was estimated on the basis of the average width of epidermal ridges. It can be concluded that the width of the skin ridges and their distinct fragments probably derived from an adult. From the collected material, it was not possible to unequivocally determine the sex and age of the owners of the prints. The print on artifact number 6 belonged to a child aged 1-3 years. The trace

visible on artifact number 7 belonged to a crawling child. The imprint showed the marks made when the hand was torn off. The depth and uneven distribution of the mark indicated that the infant was already able to crawl, which allowed us to assume that the child was between 6 and 12 months old. Artifacts 6 and 7 had prints from two different children, as evidenced by the fact that an imprint on artifact 6 was left by a walking child walking, while imprint on artifact 7 was left by younger, crawling child. Artifact 6 had an imprint of a walking child. Most likely the child was from 1 to 3 years old. The beginning of the transverse arch of the foot is visible, as well as a slight supination (Assuming the supine position or the state of being supine. It was assessed on the basic of the imprint of the foot). This may indicate the correct development of the foot and the difficulty of the terrain in which the child had to move in. Artifact 7, on the other hand, contained an imprint of the palm, which was left when the brick was very wet. This is evidenced by the characteristic patterns formed on wet clay immediately after pressing the hand.

In the above study, an important element of the analysis was the average width of the ridges. Even though it was not possible to determine age and sex of the owners of the prints, with the help of these measurements, it was possible to try to determine the age and sex of an individual who left a mark on a given vessel or tool. Moreover, it is definitely possible to state that the analysis of the width of epidermal ridges may be beneficial while trying to discover age and sex of the individual to whom the prints belong to.

In this particular research, there were too few prints which limited the accurate estimate of the sex and age of the individual. Nevertheless, it can be concluded that the thickness of the skin ridges and their clear fragments may indicate an adult. Still, prints on artifacts 2, 4 and 5 pose a problem in sex identification. It should be noted that the interpretation of artifacts is subjective, and the material analyzed due to the fragmented state of preservation makes it difficult to make an unambiguous and certain statement. More materials with better preserved imprints and the examination of the chemical composition of the ceramics would enable a more detailed analysis. The average width of the ridges is in fact a measurement often used in identification and dactyloscopic research in archeology. In scientific works, attention is paid to the correlation between the width of the ridges and the age of individuals.

Discussion

In 1987, Franaszek and Grzeszyk examined 879 volunteers in terms of a correlation between age and the width of the skin ridges. 444 men and 435 women participated in the study. The respondents were divided according to their age into 10 groups from 7 to 25 years of age and above. It was found that over a distance of 5 mm, the mean numbers of ridges start to differ between women and men from 13 to 14 years of age, and the mean width of the ridges increases with age (Franaszek and Grzeszyk 1987). However, the authors noticed that taller and more strongly built people have wider ridges than shorter people of weaker body composition.

Along with the human growth process which leads to an increase in body size, the skin ridges increase their dimensions (height, length, width), while maintaining their numbers. Depending on the finger we are considering, the width of the ridges will be different. It was found that on the thumbs and on the fingers of the right hand, the skin ridges were wider than on the left hand. On the other hand, women have narrower skin ridges than men. The changes taking place on the ridges of children are most intense during their adolescence, and with the completion of the growth process, the rate of changes taking place on the ridges slows down. The type of food and work performed may slightly increase the size of fingerprints even up to the age of 60 years (e.g. the process of gaining weight results in the increase in body surface and thus, the surface of the palm also increases causing changes in the width of epidermal ridges). It is assumed that the number of skin ridges per 5 mm ranges from 15 to 18 in children up to 12 years of age. Adults from the age of 20 years have 9–10 skin ridges with a length of 5 mm.

Analysis of the average width of the ridges for dactyloscopic identification in archeology was also used by Králík et al. (2002). They examined a fingerprint left on the statue of Venus of Dolní Věstonice I, which was discovered on July 13, 1925 in Dolní Věstonice, South Moravia (Czechoslovakia at that time) carried out by Karel Absolon. It is a figurine made of fired clay, approx. 11.5 cm high, depicting a female figure with rounded shapes. The figurine was found in two parts, in the central fireplace in the upper part of the village of Dolni Věstonice. It is dated

to the period of the Upper Palaeolithic of the Gravesian Culture, which peaked between 27,000 and 24,000. years ago. Only 75 years after the discovery of the figurine, it was examined from the paleodermatoglyphic perspective, i.e. x-rayed in search of the imprints left on it. Analysis revealed the presence of a fingerprint which was located on the left side of the figure's back, directly above the upper oblique fold, where the left fingerprints were almost parallel to the fold in the figure (Fig. 14). The dimensions of the dermatoglyph were estimated at 3x5 mm. Seven lines were recognized (Králík et al. 2002).



Fig. 14. Minutia on the back of the VěstoniceVenus; highlighting of ridges (Králík et al. 2002).

In the next research stage, the average width of the ridges was used in orderto determine the age of the individual to whom the imprint left on the figurine belonged. The analysis showed that the average width of the ridges was 0.37 mm, with a standard deviation of 0.029 mm. Taking into account the condition of the figure before the shrinkage of the ceramic material (clay, which shrinks during drying and firing – by an average of 7.5%), the average width of the ridge amounts were 0.40 mm, with a standard deviation of 0.031 mm. Using the equation, proposed by Kamp et al. (1999), to calculate the average width of the ridges,: y = 614x - 112 (y - age in months, x – lamella width in mm), the age of the individual was estimated at 11.13 years. Experiments have shown that this type of age estimation deviates from the actual age by only 1.9 years, with a standard deviation of 1.36 years. The majority of estimates never differ by more than 4 years from the actual age. Therefore, it can be concluded that the person whose imprint was found on the figurine was between 7 and 15 years old. If the correlation between the width of the ridges and the age was the same in the Upper Paleolithic times as it is now, it becomes obvious that the print could not have belonged to an adult male. With more accuracy and with respect to the shrinkage of the ceramic material, the age estimation can be shifted more towards adulthood. This allows the possibility that the imprint was left by a young adolescent woman, or even a young adult woman. Morover, the study of Kralik et al. (2002) also shows, that the accurate age estimations are not possible for fingerprints. The study also mentions that sex assessment from fragmented fingerprints is impossible.

Conclusion

The above examples, both the author's own research and those cited, show how the analysis of the average width of the skin ridges can be used in archaeological research. As the average width of the ridges varies with the age of an individual, it can be argued that the average width of the ridges correlates with age. Taking into account the research on dermatoglyphs, the average width of the ridges and possibly the estimated age, conclusions can also be drawn about the sex of the individual who left his imprint on the vessel or material. Therefore, as these measurements are used to determine the sex and age of a person, it is useful not only in forensic research, but also in the case of archaeological studies; for example, in determining who was the craftsman of a given vessel or tool. However, it is important to remember that some studies also show serious limitations of such methods. The accurate estimation of age may not always be possible for fingerprints due to many different factors including shrinkage of the ceramic material, limited number of imprints, poor preservation of the imprints. Furthermore, it is also not possible to determine the sex of the owner's imprints from fragmented fingerprints.

Conflict of interests

Authors declare none conflict of interests

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Case study: trepanation or injury? An example of an early medieval skull from Płock (Poland)

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ABSTRACT: The aim of the study was to analyze a skull found in ossuary material in Płock, dated between the 16th and 19th centuries. The skull was subjected to a comprehensive anthropological analysis due to the number of pathological changes occurring to it. These changes testify to both the diseases experienced by the individual and the acquired bone injuries. The study aimed to find out whether the pathological changes on the skull appeared as a result of the injury or intentional trepanation. The examined skull shows three injuries, of which only one may resemble trepanation processes.

Comparative analysis of the skull showed that the observed marks were generated ante-mortem. The presence of compact regenerated bone tissue with a significant thickness attests to this statement. Based on the trauma marks, it was determined that two of them ("A" and "B") had been struck by a sharp-edged instrument. They do not, however, match any known trepanation techniques. They should be categorized as purposeful injuries that are not trepanations, based on the proportions and shape of the incisions, as well as the comparative study. The "C" trace, when examined and compared to the literature, appears to be an oval depression caused by an impact with a blunt-edged object rather than a healed trepanation mark.

KEY WORDS: trepanation, injury, trauma, skull, modern, Płock, palaeopathology



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Introduction

Palaeopathology derives from three fields of knowledge: anthropology, medicine, and archeology (Gładykowska-Rzeczycka 1993). The diseases observed on the skeletal material include developmental changes, injuries, specific and non-specific inflammatory diseases, degenerative changes, disorders of the endocrine system, metabolic disorders and neoplastic changes (Gładykowska-Rzeczycka 1978). The afore mentioned fields allow for a more detailed analysis of historical populations and, together with other anthropological features, reveal an image of the living conditions of the past.

Traumatic bone injuries are one of the most important palaeopathological data (Owens 2007). They can be a consequence of accidental or deliberate actions. The first of the injuries mentioned above may be work related or to lifestyle factors, while deliberate injuries are the result of confrontational combat, robbery, execution or surgery (e.g. trepanation, amputation). Therefore, injuries are aprimary sources of data providing information related to interpersonal violence in past populations. Injuries may be antemortem when the subject has survived the injury or perimortem when the injury was fatal to the victim or occurred just before his death.

Skull injuries can be divided into many types, including active and passive (Teresiński 2002). This division is established on the basis of their location in relation to the so-called the "hat brim line", i.e. the hypothetical plane of the widest circumference, at the height of a hat's brim. All injuries above this line are considered passive injuries, i.e. those resulting from a fall, while those in the vicinity of this line and below it, are considered active. It should be noted that this criterion can be used when falling from the so-called 'vour own height'. The nature of skull fractures depends more on the speed and energy of the injury than on the size of the tool inflicting the blow. Tomasz Kozłowski (1993) states that out of the 578 examined skulls, 5.4% have visible injuries. Moreover, they are more common on the left side of the skull. He noted a higher frequency of injuries on the parietal bones (almost half of the cases) and the frontal bones (over 25%), followed by the nasal, temporal and occipital bones. In male skulls, cuts were dominant, followed by indentations, while in females it was the opposite, albeit, the differences between the frequencies were small. Post-traumatic conditions showing healing features accounted for over 90% of all injuries. One skull showed signs of trepanation.

Trepanation is a procedure that has been carried out since Neolithic times and involves making a hole in the skulls' cranial vault along with the removal of a part of the bone. Trepination has been used within the ritual context, as well as in therapy. The most probable reason for the use of trepanation was to relieve chronic headache, migraines and epilepsy (Negahnaz et al. 2015). Trepanations were characterized by the level of post-surgery survival that varied depending on the country and epoch (approximately 25% in Great Britain during the Iron Age, 78% in Switzerland at the same time, 72% in the Iron Age, and 50% in the early Middle Ages). Trepanations were performed on all continents, of which in Europe most of the skulls with post-trepanation traces were found in France, from where this custom spread to Eastern Europe in the Neolithic. Polish literature provides information on at least 50 trepanated skulls from Poland (Kozłowski 2012).

Majchrzak and Olender (2015), in their work on trepanation of Andean skulls, define four methods of opening skulls (after Lisowski: 1967): a) cutting lines to create a square or rectangular hole in the bone, b) scraping the bone to obtain a hole, which was the slowest, but at the same time the most accurate and safest method, c) drilling a hole or a series of small holes in the skull, which was used to break off a piece of the skull, d) gouging (sawing) cutting out a fragment of the bone in a circular. According to the authors, most trepanations were performed on the occipital bone, and on the left and middle frontal and parietal bones. Drilling at the skull base, as well as in the temporal bones was avoided. Lorkiewicz et al. (2018) mention surgical therapy which was used to treat head injuries as the goal of trepanation procedures.

The aim of the study was to analyze traces of intentional surgeries on a specific skull. Additionally, attempts were made to determine whether they were remnants of injuries or trepanations to the skull.

Materials and methods

The skull was found in an ossuary in Płock material dated between the 16th and 19th centuries. The ossuary was created as a result of moving the cemetery due to the construction of a bridge over the Vistula in the period preceding World War II. It presents a number of pathological changes, which testify to both diseases suffered by the individual and acquired bone injuries.

The research included the assessment of the sex of the subject (Steckel et al. 2005; Malinowski and Bożiłow 1997). Skull measurements were made using the Martin technique (Martin, Saller 1957), using standard measuring equipment twice, and the result was averaged. On their basis, the craniometric indicators were calculated (Malinowski and Bożiłow 1997). The characteristics of pathological changes were determined in accordance with the guidelines of Gładykowska-Rzeczycka (1976) and Ortner's atlas (1998). In addition, measurements of the found traces were made in order to determine their extent.

Results

The skull was preserved in very good condition, without the mandible (Calvarium). Postmortem lesions of the skull base and left zygomatic arch, as well as loss of incisors and canines without signs of obliteration of the alveolar sockets were noted.

Dimorphic features (in particular, clearly marked smoothness, no marked frontal and parietal nodules, the shape of the eye sockets and their rounded upper edge, as well as the massiveness of the mastoid process and strongly pronounced supraorbital arches) indicate that the examined skull belonged to a man. On the basis of obliteration of cranial sutures and traces of tooth wear, the age of the individual was determined as maturus/ senilis.

Twenty-five brain and craniofacial measurements were performed and averaged results are presented (tab. 1). Some of them are burdened with a higher risk of error due to the degree of obliteration of the individual's sutures. This applies to the ba-b measurement, which has a strongly approximate bregma point. It is similar with the measurement of co-co due to signs of overgrowth of the coronary suture. In the case of the n-ns value, the measurement uncertainty results from the chipping of the nasal spine. Part of the dimensions, including the total height of the skull, could not be measured due to the lack of a mandible.

Table	1.	Measurements	of	the	skull

No.	Measurement	mm
1	g-op	187
2	n-b	150
3	i-o	38
4	n-ba	115
5	ba-o	37
6	ba-b	138*
7	eu-eu	139
8	ast-ast	115
9	ft-ft	99
10	со-со	115*
11	au-au	110
12	mst-mst	111
13	The width of the foramen magnum fol-fol	30
14	Horizontal circcumference	506
15	arch n-o	300
16	arch n-b	130
17	n-pr	75
18	n-ns	54*
19	pr-ba	101
20	ol-sta	40
21	ek-ek	97
22	mf-mf	18
23	mf-ek	P-49 L-45
24	The height of the eye socket sbk-spa	P - 35 L - 35
25	apt-apt	28

* - uncertain measurement

The performed measurements were used to calculate the craniometric indicators (Table 2). The subject had a long and quite narrow orthognathic skull, with a wide forehead, a narrow occipital opening, and a wide nose. The eye sockets were characterized by asymmetric size, with the right socket being in the low category and the left socket in the medium height category.

Indicator	Equation	Result	Description
Width and length	eu-eu/g-op*100	74,33	dolichokranius
Height and length	ba-b/g-op*100	73,80	Orthokranius
Height and width	ba-b/eu-eu*100	99,28	Akrokranius
Average height and width of the skull Hrdlicki-Kocki	ba-b/(g-op+eu-eu)/2*100	84,66	Mesosemic by Steward, Kocka and Vallois, megasemic by Hrdlicka
Fronto-parietal	ft-ft/eu-eu*100	71,22	eurymetopus
Foramen magnum	fol-fol/ba-o*100	81,08	narrow
Orbital fissure of the skull	sbk-spa/mf-ek*100	R- 71,43 L - 77,77	R – chamaekonch L – esokonch
Nose, skull	apt-apt/n-ns*100	51,85	chamaerrhinus
Maxillary	pr-ba/n-ba*100	87,83	orthognath

Table 2. Craniometric indicators

The skull depicted 3 trauma marks. In order to determine whether the analysed traces were traumas or trepanations, an additional computed tomography was performed (photos 4, 5 and 6). The examination consisted of scanning the skull and creating several hundred layers, which were then downloaded to a computer and analysed.

The first of these, injury 'A' was U-shaped. It ran along the region of the coronal suture, then turned at a right angle and continued along the sagittal suture where it bent halfway along the left parietal bone (Fig. 1 and 2). It was 87 mm long and 74 mm wide in total. It was inflicted from above at an angle and caused a skull fracture, the remnant of which is a fissure 34 mm long and 6 mm thick. The location of the trace corresponds to the most common injury sites (left side of the skull, parietal bone and frontal bone). However, it was made with great force from the medial side towards the external side. The fact that it was an extensive, fairly wide wound also supports the classification

of the mark as an injury, which may indicate that it was inflicted with a sharpedged object. Its shape also cannot be attributed to any trepanation technique. The CT scan (Fig. 5) shows bone thickening and compaction characteristic of healing marks leading to partial occlusion of the hole.

Trace "B" is located on the left side of the occipital bone of the examined skull and overlaps the cephalic suture that connects the occipital bone with the parietal bones (Fig. 3 and 8). It is 71mm long and 41 mm wide. The object that caused the mark made contact with the bone from above and caused a deep incision 7mm thick with an opening length of 45mm. This is an area where trauma rarely occurs. The incision of the bone was made with great force, penetrating well into the occipital bone, but not causing a skull fracture. This, however, does not exclude the classification of the mark as a post-operation wound. It is possible that the wound was made with a surgical instrument in order to, for example, decompress a hematoma

or equalize intracranial pressure. Nevertheless, it was not a post-trepanation wound, as evidenced by the shape of the opening. The CT scan which was performed (Fig. 4 and 5) shows evidence of wound healing - visible traces of thickening and densification of bone tissue, which caused a significant portion of the hole to be sealed up.



Fig. 1. Trace 'A' on the frontal and parietal bone seen from above (superior).



Fig. 2. Trace 'A' on the left frontal and parietal bone seen from the anterior side.



Fig. 3. Trace "B". – incision on the occipital bone seen from behind (posterior).



Fig. 4. Section of the skull in the transverse plane (transversum). Visible hollow in the skull at the site of trace "B" and signs of wound healing.

The third element is an oval scar trace. It is located approximately in the middle of the right parietal bone, above the almost completely obliterated scale suture (Fig. 6 and 8). It is 45 mm long and 35 mm wide. The scar could have been caused by the use of a blunt-edged tool or a cicatrized post-trrepanation hole. There were no traces of a fracture of the skull bone and no concentric inward healing characteristic of trepanations performed using the drilling technique, which could have been indicated by the shape of the injury. The observed trace was depressed and uniformly healed, which was confirmed by the CT scan (Fig. 7). This phenomenon indicates that for this individual the fracture did not prove to be fatal. It also shows the growth of bone tissue.



Fig. 5. Section of the skull in the sagittal plane from the left side. Visible hollow in the skull at the site of mark "A" and signs of healing of wounds "A" and "B".



Fig. 6. Trace 'C' - indentation on the right parietal bone (dexter) visible from the side.



Fig. 7. Section of the skull in the posterior side. Visible hollow in the right side coresponding to mark "C". Visible signs of bone tissue thickening indicative of wound healing.



Fig. 8. Traces 'B' and 'C' visible from the rear-lateral (dexter) side. Fragment of the "A" mark on the front left side of the skull is also visible.

Discussion

This study attempted to determine what factors may have contributed to the appearance of the observed changes to the skull in question. Three traces were identified. Each was described on the basis of 3D photographs and scans as well as CT images. The observations were compared with examples from the literature.

The study of Brzobohata et al. (2017) where they described a skeleton that belonged to a woman between 20 and 25 years of age, in which there was an oval depression on the right parietal bone was used as comparative material. It was probably formed by a blow from a sharp object, probably an axe. Based on features considered diagnostic for this type of injury, it was determined to have formed around the time of death. It showed no signs of healing or infection, suggesting a direct cause of death for this woman (intracranial bleeding). It is somewhat similar to the shape of the "B" injury on the occiput of the studied individual, but is much shallower and narrower.

A similar case, which was used in the comparative analysis of injuries on the skull of a male from Plock, was described by Nagar et al. (2018). In their work they analyzed 4 injuries located on the skull of a male individual aged 30-40 years, 3 of which were peri-mortem, with the last blow being the immediate cause of death of the individual. The injuries were located near the midline of the frontal bone (near the coronal suture), posterior to the parietal bone, and posterior to the sagittal suture. The first injury was determined that it was inflicted by a sharp instrument falling from above; the second injury was speculated to be a force injury inflicted at an oblique angle also by a sharp instrument - probably the same one. The third injury was caused by a blow to the back of the skull. It was presumed to have been inflicted with a sword, as evidenced by the lack of serrations on the mark. The largest injury ran from the left parietal bone cutting through the sagittal sutures and scapular sutures to the base of the zygomatic process of the right temporal bone. It was an open cut which indicated a blow delivered with a sword from the upper right side, which was assumed as being the immediate cause of this individual's death. These marks differ from the "A" and "B" injuries analyzed in this paper in the depth and width of the cuts. However, they were similar in shape suggesting a high force pressure with a sharp-edged instrument.

Another example are the 5 skulls deposited in the Hungarian Museum of Natural History (Marton et al. 2016). On the first skull, a puncture wound was observed on the frontal bone, which also crosses the upper orbital wall. It may be classified as an arrow-inflicted mortal wound, as no signs of healing were observed at the edges. On the second skull, a defect was found above the orbit and a thickening on the eyebrow arch, but the victim survived the injury as healing marks were visible. This skull also had an injury on the parietal side, which was not fatal neither. The third skull had traces of being struck by a sharp instrument descending from above, passing through the right parietal bone and temporal bone to the frontal bone. This skull also had healing marks in the form of rounded bone edges, which precludes the injury from being considered fatal, although, they had not been fully healed, so death may have occurred some time after the event. The fourth example was a chop wound running in a lower to upper direction with no signs of healing, confirming that this was a fatal blow. The last skull described in this publication bore signs of a chop wound that caused the bony edges to be raised. These were rounded and healed at the edges, indicating that the blows were not fatal. From the examples presented in the publication, traces similar to those observed in the individual from Plock were found on skull number three, where the blow caused the skull fragment to split and shift. Morover, the top edge resembled trace "B" on the occipital (which, however, did not cause the skull fragment to split). A similarity to trace "A" on the skull from Plock was also evident on skull numer four, on which, however, the trace was very thin despite its considerable length.

Steyn et al. (2009) in their work on ante-mortem trauma describe a case of a healed cut wound located on the right frontal bone. It is much wider and shallower than marks "A" and "B", but has a similar shape.

Also, Nerlich et al. in their 2015 paper described an interesting case of a skull mark that most likely came from an injury caused by a sharp-edged instrument. This trace is similar to trace "A" on the skull from Plock. It had a similar course, but led to a complete separation of the vault from the rest of the skull and does not contain any signs of healing.

Examples of trepanation include the skulls described by Lorkiewicz et al. (2018). The skulls belonged to 6 adult males. The first one had a trepanation hole in the upper part of the vault, at the cranial suture, with perpendicular (except for the anterior oblique part) walls obliterated by healing. They indicate sawing as this technique is for making the hole. On the second skull, on the right frontal shell and parietal bone, a large oval hole with traces of healing was found, surrounded by a wide ring of rubbed bone, indicating the use of the scraping method. On the third skull, a pair of nearly oval post-traumatic marks separated by a small bone bridge was visible on the right parietal bone. In both cases, the bone was excised with a sharp instrument positioned perpendicular to the skull surface, probably to drain some pus. Morover, there were two additional defects on the left parietal bone that may also be trepanation marks made by drilling. All of the above-mentioned lesions were fully healed. Another skull had a large, oval-shaped hole in the left parietal bone made probably by a mixed

technique, in which the left and upper left portions were sawn out, while the rest were made by scraping or extracranial drilling. On skull number five, there were two holes on the left half of the cranial vault: one was large and L-shaped, while the other was smaller and oval-shaped. The cranial vault showed two healed lesions made with a sharp instrument. The larger hole was made at the site of an extensive fracture by removing bone fragments and cleaning the edges. The second, smaller hole, most likely represents a typical trepanation made with a sawing technique. The right side of the vault of the last skull showed evidence of extensive blunt force trauma in the form of a circular, depressed area surrounded by a line of concentric fracture and three radiating fractures. All of these lesions were fully healed. There was an oval, funnel-shaped opening adjacent to the frontal bone defect. The most likely interpretation of this lesion was trepanation performed by a combination of drilling and scraping. The alternative diagnosis of this lesion as blunt force trauma was less likely given the CT images confirming that the bone was removed from the site rather than indented inward.

However, all of these injuries differ significantly in the shape and edges of the openings from the studied skull, making it less likely that the analyzed injuries could be classified as trepanation. The blunt trauma trace from the literature was also not similar to the "C" trace.

Conclusion

Comparative analysis of the skull showed that the observed marks were formed ante-mortem. This is evidenced by the presence of compact regenerative bone tissue of considerable thickness.

Based on the analysis of the trauma marks, it was concluded that two of them ("A" and "B") were inflicted with a sharp-edged object. At the same time, they did not match any known trepanation techniques. Considering the dimensions and shape of the incisions as well as the comparative analysis, they should be classified as intentional injuries which are not trepanations. Observation of the "C" trace and its comparison with the literature indicates that it resembles an oval depression resulting from an impact with a blunt-edged instrument rather than a healed trepanation mark. However, the CT scan did not show this.

In conclusion, the analyses which were carried out are an important part of the research on distinguishing trepanation marks from trauma. They made it possible to determine the nature of the observed changes with a fairly high probability.

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Authors' contribution

Daria Gromnicka (50%): prepared a draft, collected and analysed data (including anthropometric measurements, CT scans and photographs), collected references.

Aleksandra Partyńska (50%): designed the study, prepared a draft, collected references.

Conflict of interests

Authors declare none conflict of interests

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Application of the lateral angle method for sex determination of cremated individuals from burials of the Lusatian culture cemetery in Czernikowice, Poland

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ABSTRACT: Research of cremated human remains are limited by severe analytical constraints. Estimation of basic anthropological parameters such as sex of individuals or their age at death is often uncertain. A method for assessing the sex of cremated individuals measures the lateral angle of the petrous part (PP) of the temporal bone, known as the lateral angle (LA) method.

In the cemetery of the Lusatian culture in Czernikowice (51.317389°N, 15.871469°E), 6 well-preserved PP were identified. The analyzed PP belonged to 6 different individuals: 3 adults and 3 children. Based on standard anthropological methods, sex was estimated for adults individuals: 2 males and 1 female. The identified PP served as the basis for application of the LA method. The bones were scanned by computed tomography (CT) and the tomographic imaging allowed measurement of the lateral angle.

The absolute values of intra-observer errors did not exceed 1°. Relative technical errors of measurements (rTEM) fell in the range below 5%, which is indicative of their high precision. Individuals for which the LA value was greater than or equal to 45.0° were qualified as females and those for which it was less than 45.0° – as males. The LA values for female individuals ranged from 48.0 to 49.1°, (average $48.5\pm0.78^\circ$, median 48.4°) and for male individuals were in the range of $24.9-37.5^\circ$ (average $33.4\pm5.80^\circ$, median 35.5°). The absolute difference between the average values for female and male individuals was considerable (15.1°) and statistically significant (p < 0.001).

The LA method provides good reliability of measurements when it comes to this analysis with regard to cremated osteological material, and the use of non-invasive CT enhances its value in the context of archaeological remains. However, its capability for sexing subadult individuals should be approached with caution and requires further research.

Key words: LA method, computed tomography, cremated human remains, Urnfield culture, Late Bronze Age

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Introduction

When pursuing the research on communities connected with the Urnfield tradition on the present-day territory of Poland, we face a large-scale spatio-temporal phenomenon; duration of some cemeteries extended for over one thousand years (Lasak 1996; Stolarczyk et al. 2020). Identification of variability of biological parameters of prehistoric communities is crucial for analysing consequences of their cultural progress based on the evolution of adaptative mechanisms within the population. Accordingly, anthropological data can be of fundamental importance for socio-cultural inferences and changes in funeral rites. The current progress in anthropological methods based on advanced technology provides new information that is especially important in the research on populations of the Urn field culture, as their communities commonly used cremation in their burial practices.

Investigations of cremated bioarchaeological materials are limited by considerable analytical constraints. Estimation of basic anthropological parameters such as sex of individuals or their age at death is very often uncertain. This is particularly true for assessing the sex of individuals on the basis of cremated bone fragments (McKinley 2015). Cremated remains are generally considerably fragmented and deformed, which makes it impossible to use the methods of sex determination that apply to skeletal materials (Strzałko et al. 1973; Dokládal 1999; White et al. 2011). The methods of estimating sex of subadult individuals based on morphology of the mandible and ilium (Schutkowski 1993; Loth and Henneberg 2001), as well as biochemical methods (Rebay-Salisbury et al. 2020; Gowland et al. 2021), cannot be applied

to cremains. Of the methods employed for sexing adult individuals, the use of both the descriptive as well as metric ones can result in errors due to a variety of changes in shape and dimensions of particular parts of a skeleton in the process of cremation (Strzałko et al. 1973).

Petrous parts of temporal bones (PP) are among bones with the best preservability (Masotti et al. 2013; Bonczarowska et al. 2021). Therefore, several methods of sexing were developed which use measurements of different anatomical structures constituting PP (Wahl 1981; Schutkowski and Herrmann 1983; Wahl and Graw 2001; Norén et al. 2005; Ward et al. 2020: Boucherie et al. 2021). One of them is a technique based on the measurement of PP's lateral angle, known simply as the lateral angle (LA) method. As extensively presented in numerous publications on the subject (cf. e.g. Wahl 1981; Graw et al. 2005; Norén et al. 2005; Kozerska et al. 2020), regardless of the adult age categories or origins of individuals studied, there were significant differences between the size of the LA in both sexes. The method has already been applied for estimating the sex of cremated prehistoric individuals, for instance, those discovered at Stonehenge, dated to the Late Neolithic/ Early Bronze Age (Willis et al. 2016). Due to its applicability to individuals from cremation graves, the method gave basis also to the analyses of such prehistoric finds like the ones presented below.

Material and methods

The anthropological analyses were performed on cremated bone remains from the burial ground of the Urnfield culture discovered at site 2 in Czernikowice, Poland (51.317389°N, 15.871469°E) (Stolarczyk et al. 2020). Osteological material was recovered in the 1960s and 1970s and since then most of it has been lost. Out of about 200 burials, cremains from just 24 features, i.e. 20 graves and 4 bone clusters, could have been reanalyzed. However, for 100 burials archival anthropological reports of Brunon Miszkiewicz survived, and these were included in the current analyses and interpretation of the burial ground (Stolarczyk et al. 2020).

The majority of the graves (70.7%) are dated to the Bronze Age (BA) V, 8.8% of them – to BA IV, 8.0% – to BA IV-V, and 3.2% – to the end of the Bronze Age and the beginning of the Early Iron Age. The remaining graves were dated generally to a long period lasting from the Late Bronze Age to the Early Iron Age, viz. around 1300–400 BC.

The weight of the cremated bones was measured for 11.1% burials and ranged from 2 to 1288g. Heavily burnt bone fragments (97.4%) were of white, chalkwhite or cream colour.

The MNI value for 124 analysed graves and bone clusters was estimated to be 146, of which age at death was determined in the case of 140 (95.2%) individuals.

The division into age categories was used according to Kwiatkowska (2005): Infans I (younger children) – from 0 to 7 years old, Infans II (older children) – from 7 to 14 years old, Juvenis (youthful ones) – from 14 to 22 years old, Adultus (young adult) – from 22 to 35 years old, Maturus (middle adult) – from 35 to 55 years old, senilis (old adult) – over 55 years old. The most numerous age group was that of children (69.2%: Infans I – 52.7%, Infans II – 16.4%), less numerous was the set of adults (13.0%), and the least numerous were the oldest individuals (1.5%). Based on the estimated ages at death and mortality table, life expectancy at birth (e0) was estimated to be 6.5 years, whilst individuals in the reproductive age on average did not live past 30 years (e_{20} =8.9). The value of the potential reproduction index was high (R_{pot} =0.883), but so was the proportion of deaths in childhood (79.1%). This situation was mirrored by a low value of the biological state index of the population (I_{bs} =0.179), indicating its poor biological condition.

The sex of the individuals was estimated according to the methods proposed by Strzałko, Piontek, Malinowski (1973), based on morphological dimorphism of the skull and measurements of the zygomatic bone and the bones of the postcranial skeleton. The sex of most individuals was indeterminable (72.1%). This was caused by the predomination of individuals classified within the youngest age categories (69.2%). Based on standard anthropological methods, from among 146 individuals 22 were identified as males and 17 as females.

In the reanalysed osteological material, there were 6 well-preserved PP (4 right ones – PP R, and 2 left ones – PP L) that belonged to 6 different individuals of various ages at death and sex determined for 3 adult individuals using conventional anthropological methods (Strzałko et al. 1973; Dokládal 1999). The sex of these 3 adult individuals was determined on the basis of at least two features of the skull morphology (individuals from graves 69 and 78) and measurements of the bones of the postcranial skeleton (individuals from graves 78 and Cz/18). On this basis, it was determined that an individual from grave 69 had 80% probability of being male, and an individual from grave 78 with a 56% probability of being female (Strzałko et al. 1973; Stolarczyk et al. 2020). For the individual from grave 78, the proximal femur epiphysis (head diameter) was measured, identifying this individual as female. Regarding an individual from the grave Cz/18, measurements of the humeral head and radius were taken and classified as male.

The identified PP offered an opportunity to apply the lateral angle (LA) method in order to assess the sex of individuals and determine the results for individuals whose sex was determined also with the use of standard anthropological methods (Strzałko et al. 1973; Dokládal 1999; White et al. 2011; Stolarczyk et al. 2020).

The LA method relies on measuring the angle between the internal auditory canal and the medial surface of the petrous part of the temporal bone (Wahl 1981). In this study, we applied the variant of the LA method (Akansel et al. 2008) that is adapted to measurements of the lateral angle made on tomographic scans (Figure 1).



Fig. 1. Preserved petrous parts (PP) of temporal bones of individuals from cremation graves (Gr.) from the burial ground at Czernikowice and the technique of measuring lateral angle (LA). Bottom to the right – the most easily recognizable inner ear structures: cochlea, vestibulum and internal acoustic canal (IAC).
The CT examination of preserved PP was performed in the Department of General Interventional Radiology and Neuroradiology of the Wroclaw Medical University with a VCT Lightspeed, 64-row scanner (GE Healthcare, USA). The scanning parameters: tube voltage 120 kVp; beam collimation was 0.6 mm; scan range cover all bones, scan rotation time was 1 s., automatic exposure control (AEC) was activated, the noise index (NI) was 2.8. Scanning was performed in sequential mode. The filtered back projection (FBP) algorithm was used for image reconstruction. Image matrix size of 512×512 was the standard for the CT application. Images were reconstructed in axial and coronal planes with a resolution ranging from 0.3 to 10.0 mm, according to requirements. Image analyses were performed in soft tissue and bone window. Image post-processing techniques included 2D and 3D reconstructions (MPR - multiplanar reconstruction, MIP - maximum intensity projection, VR - volume rendering).

LA measurements on the CT scans of each PP were taken twice at monthly intervals. The intra-observer error was calculated on the basis of the absolute. as well as relative differences in values of measurements and their standard errors (SD; Popović and Thomas 2017). Additionally, technical errors of measurements (TEM) and relative technical errors of measurements (rTEM) were calculated. In this case, the latter is equivalent to the coefficient of variation expressed in percentage. Values of rTEM that are below 5% indicate that the repeated measurements were precise (Ulijaszek and Kerr 1999; Weinberg et al. 2005) and should be considered as reliable. A detailed intra-observer error analysis was made for each of the 6 investigated PP. Differences

in results of measurements in accordance with age categories (between children and adults) and sex classes (between female and male individuals) were also verified. For each of the four categories, precision of measurements was verified by calculating their coefficient of reliability (R). R values range between 0 and 1, where those equal or close to the upper limit indicate that there was no measurement error (Weinberg et al. 2005). Differences in intra-observer errors for measurements on the left and right PP were not calculated, because there was only one preserved PP for each individual. As all the measurements were taken by a single person, inter-observer errors were not taken into consideration.

The sex of each individual was determined on the basis of the arithmetic mean of the LA measurements. he obtained value was then compared to and classified in accordance with the known ranges of its variation for male and female individuals. As a sectioning point 45.0° was applied. Individuals for which the LA value was greater than or equal to 45.0° were qualified as females and those for which it was less than 45.0° - as males ($F \ge 45^\circ < M$), (Wahl 1981; Norén et al. 2005; Akansel et al. 2008; Kozerska et al. 2020). The small sample of the PP was tested. Therefore the difference between the mean values of LA measurements performed for both the sexes was tested with the use of the non-parametric Mann-Whitney U-test.

Results

In the cases of individuals buried in graves 69, 78 and Cz/18, their sex determinations based on standard anthropological methods were confirmed with the LA method (Table 1). Also, the lateral angle measurements made it possible to estimate the sex of children from graves 22, 74 and 82. Individuals from burials 74 and 82 were determined to be males, whereas the one from grave 22 – was female (Table 1).

Table 1. General characteristics of individuals and individual mean values of PP lateral angle with standard deviation (SD); results of LA measurements are provided in degrees (°)

Grave number	Age at death	Standard sex determination	PP under analysis	Mean ± SD	LA method sex determination
22	Infans I	Undetermined	Right	48.0 ± 0.6	Female
69	Maturus	Male	Right	36.5 ± 0.3	Male
74	Infans II	Undetermined	Right	24.9 ± 0.6	Male
78	Adultus	Female	Left	49.1 ± 0.9	Female
82	Infans I	Undetermined	Right	34.7 ± 0.9	Male
Cz/18	Adultus/Maturus	Male	Left	37.5 ± 0.4	Male

The values of the LA angle ranged from 24.9 to 49.1°, with the mean of $38.4\pm9.02^{\circ}$. The LA values for female individuals fell in the range from 48.0 to 49.1° , with the mean value of $48.5\pm0.78^{\circ}$, and median of 48.4° . The LA values for male individuals fell in the range between 24.9 and 37.5° , with the mean value of $33.4\pm5,80^{\circ}$, and median of 35.5° (Figure 2). The absolute difference between the mean values for female and male individuals was high (15.1°) and was proven to be statistically significant (p < 0.001; Mann-Whitney *U*-test). However, the low number of tested PP should be taken into account.



Fig. 2. Comparison between the results of lateral angle (LA) measurements on male and female individuals form Czernikowice with analogical ranges from selected studies (Wahl 1981; Norén et al. 2005; Akansel et al. 2008).

The results obtained for intra-observer errors indicate high repeatability of the measurements. Absolute differences between values of every first and every repeated measurement were lower than 1.0°, and the relative technical error of measurement (rTEM) for particular individuals did not exceed 1.5% (Table 2). There were no differences in the precision of the measurements, either in regard to the sex of individuals or their age at death. For all the categories, values of the coefficient of reliability (R) were close to 1.0 (Table 3), which confirms that the measurements were repeatable.

Table 2. Intra-observer errors calculated on the basis of absolute and relative variability values of measurements of lateral angles with their individual standard deviations (SD), as well as technical errors of measurements (TEM) with relative technical errors of measurements (rTEM)

	Absolute intra-observer variability		Relative i var			
Grave number	Difference	Individual SD	Difference (%)	Individual SD (%)	TEM	rTEM
22	0.33	0.52	1.3	1.1	0.13	0.28
69	-0.40	0.28	1.0	0.8	0.16	0.45
74	0.20	0.52	2.3	2.1	0.08	0.33
78	-0.93	0.80	2.0	1.6	0.52	1.07
82	-0.73	0.80	3.3	2.3	0.30	0.86
Cz/18	0.53	0.38	1.3	1.0	0.22	0.58

Table 3. Mean values of lateral angle (LA) measurements and the parameters of their errors with the coefficient of reliability (R) for individuals categorized according to sex and age-at-death; SD – standard deviation, TEM – technical error of measurement, rTEM – relative technical error of measurement

Category	Mean ± SD	TEM	rTEM	R
Children	35.8 ± 10.6	0.19	0.54	0.9996
Adults	41.0 ± 6.06	0.27	0.66	0.9980
Female	48.5 ± 0.90	0.28	0.59	0.9036
Male	33.4 ± 5.27	0.21	0.62	0.9984

Discussion

The process of cremation negatively affects the potential of anthropological analyses performed on human remains. A vast majority of the Lusatian culture cremains belong to individuals of the subadult age category (Szybowicz 1995; Stolarczyk et al. 2020). Reliable assessment of their sex would be of great importance not only for reconstructions of sex structures within the population but also for approaching socio-cultural issues.

The LA method is among very few procedures of sex estimation that are applicable to cremated human remains from archaeological contexts (Wahl 1981). It must be stressed, that hitherto no research on PP alterations (including changes of LA) caused by the process of cremation has been conducted (Gonçalves et al. 2015). However, the studies based on modern cremains of adult individuals have demonstrated that LA values can correspond with their actual sex (Masotti et al. 2013). Additionally, the research confirmed that there were no statistically significant differences between LA measurements taken on the right versus those taken on the left PP of a given individual (Goncalves et al. 2015; Afacan et al. 2017; Kozerska et al. 2020: Bonczarowska et al. 2021). The LA method has been tested on subadult individuals, but despite statistically significant differences between both sexes (Gonçalves et al. 2011), its use in sex estimation in children and juvenile is questioned (Akansel et al. 2008; Gonçalves et al. 2011; Afacan et al. 2017; Kozerska et al. 2020), and in a recent study its effectiveness is uncertain even for adults (Bonczarowska et al. 2021).

Conducted clinical research also has not confirmed differences between female and male individuals to be statistically significant (Morgan et al. 2013). Moreover, some scholars point out the unreliability of the results regarding subadult individuals and those over the age of 70, showing large discrepancies between the already known sex and the one estimated on the basis of LA measurements (Masotti et al. 2013; Afacan et al. 2017; Masotti et al. 2019). The applicability of the LA method for sexing subadult individuals has been challenged by Afacan et al. (2017). Their research conducted on individuals in the age range from 0-18 years showed no differences between the sexes, but age-related decrease of LA values were observed, especially in subadult male individuals. Similar age-related observations have been reported by Akansel et al. (2008). The method applied by Afacan et al. (2017) was based on the lateral angle measurements taken on images obtained with magnetic resonance (MRI), and the sample group consisted of children with different pathologies of hearing organs, excluding patients with severe pathologies from the study. Therefore, even the authors themselves could not exclude the possibility that their research was conducted on a biased sample and that the measurements were collected disregarding the previous procedure (Wahl 1981; Graw et al. 2005).

Initially, the LA method was based on the measurements of the angle taken on a cast of the internal acoustic canal (IAC) made of dental casting material (Wahl 1981; Graw et al. 2005; Norén et al. 2005; Gonçalves et al. 2011). However simple the technique may appear, either preparation of the casts without damaging cremated bones, or repeatable LA measurements made on them, were never easy to perform. Therefore, the technique was improved by intersecting the silicone replicas with the aim of making their photocopies or scans. The measurements were firstly taken manually on paper copies (Graw et al. 2005), and then on digital copies with the use of computer software (Gonçalves et al. 2011). From a bioarchaeological point of view, the most critical phase of the procedure is the operation performed prior to casting the silicon replicas of the IAC. For the ease of extracting solidified silicone-based material, the outer surface of PP and the inner surface of the IAC must be covered with vaseline or other substances with similar properties. Since PP are commonly subject also to biochemical analyses, including isotopic ones (Harvig et al. 2014; Chmielewski et al. 2021), any contamination with modern

matter must not be accepted. Currently used non-invasive methods based on computed tomography are more appropriate for the frangible and otherwise sensitive archaeological osseous material (Akansel et al. 2008; Panenková et al. 2009; Morgan et al. 2013; Willis et al. 2016; Afacan et al. 2017; Gibelli et al. 2021; Pezo-Lanfranco and Haetinger 2021).

Based on the investigation conducted on the 6 PP from the burial ground at Czernikowice, it can be argued that the difficulties in applying CT scans to LA measurements result first and foremost from poor preservation of the bone structures. Additionally, any contamination of anatomical structures of PP with sediments also limit the possibility of their digital imaging and thereby correct measuring of the lateral angle. Moreover, sincepetrous parts of temporal bones are usually separated from skulls, placing them in the gantry of a scanner in their anatomical position can be difficult. Consequently, in some cases, the measurement of the LA value must be taken on MPR or VR reconstructions instead of axial reconstructions. However, regarding the PP investigated within the present study, this had no effect on the repeatability of the measurements.

Various authors have been concerned about low values of technical errors for both intra- as well as inter-observer measurements (Norén et al. 2005; Pezo-Lanfranco and Haetinger 2021). Discrepancies between the repeated intra-observer measurements taken for the needs of the mentioned studies generally did not exceed 1°, and the highest rTEM was approximately 1%. However, in some research, the value of intra-observer error was above the threshold of acceptance. This indicates that the overall reproducibility of measurements was not precise, while their repeatability (coherence of measurements taken by a single observer) was maintained (Gonçalves et al. 2011; Gonçalves et al. 2015).

The most serious problem though, is the considerable overlap of LA ranges measured for female and male individuals (Pezo-Lanfranco and Haetinger 2021; see Figure 2). In some studies, the differences between mean values of measurements taken for individuals of the two sexes turned out to be statistically insignificant (Morgan et al. 2013; Gonçalves et al. 2015). Nevertheless, the majority of works has already proven the difference to be statistically significant (Graw et al. 2005; Norén et al. 2005; Akansel et al. 2008; Masotti et al. 2019). However, even in these cases, the thresholds of discrimination between LA ranges for both the sexes differ considerably. The most often provided value is that of 45° (Norén et al. 2005; Akansel et al. 2008), but others report 41° (Masotti et al. 2019). Akansel et al. (2008) not only measured the threshold at 45° but also determined the probability of discrimination between both sexes. Scholars calculated the limit value of 60° for female individuals, where such determination of sex has a 97% probability, and the limit value of 35° for male individuals, below which such sex determination hasa 93% probability. Similarly, Graw et al. (2005) provide the value of 65° for females and 40° for males. In our study, the threshold of 45° was accepted, and individuals for which values were lower than 40° were regarded as being males. The difference between the mean values calculated for both the sexes was considerable and statistically significant, though it must be underlined that the analysed sample was very small (just 6 individuals).

Other important questions raised in works addressing the subject concern the

specificity of studied populations and possible differences between the cremation as it is carried out nowadays, and the way it was performed in prehistory. Modern human populations that were investigated in regard to LA variability are characterized by a relatively low phenotypic variability (Pezo-Lanfranco and Haetinger 2021). For that reason outcomes of these studies might be irrelevant to the results of analogical research conducted on prehistoric populations, which have been characterized by a stronger dimorphism of LA. The morphological diversity concerns also the shape of the IAC. There are few main morphotypes of the IAC that can be distinguished within modern populations: the funnel-shaped, the cylinder-shaped and the bud-shaped one (Margues et al. 2012). The variability is of great significance for performing LA measurements. Therefore, when applying the variant of the LA method that uses tomograms, it is proposed to take the LA measurement starting from the apex of the IAC instead of measuring it parallel to the anterior lip of the IAC (Akansel et al. 2008; Gibelli et al. 2021; Pezo-Lanfranco and Haetinger 2021). However, not all researchers using the LA method based on CT imaging take the lateral angle measurement in the exact same way (Panenková et al. 2009; Morgan et al. 2013; Afacan et al. 2017). This causes problems when comparing research results, both in regard to the obtained LA ranges, as well as threshold values discriminating individuals between the sexes. Therefore, the determinations of LA ranges and thresholds for sexing individuals from archaeological contexts should be, at best, based on statistical calculations made for each investigated population (burial ground).

With regards to the burial ground from Czernikowice, the difference between the

sexes was apparent (Table 3), although it must be kept in mind that the number of studied individuals was low. In the case of three adult individuals, sexing with the use of the LA method confirmed the results of earlier analyses based on standard anthropological procedures. Based on the adopted sectioning point 45° (F $\ge 45^{\circ} < M$), the sex of subadult individuals was estimated (Table 1). No other method would allow determining the sex of subadult individuals on the basis of their cremains. However, although the LA method seems to be a promising tool for estimating sex of cremated human bones, there is no certainty that the estimated sex is the "real sex", especially for infant and juvenile individuals (Akansel et al. 2008; Afacan et al. 2017).

Conclusions

The LA method based on CT imaging, despite being widely questioned, can be applied to cremated osteological material from archaeological contexts. The use of computed tomography scanning makes the method non-invasive, non-destructive, thereby enhancing its value. The method is also relatively inexpensive. However, poor preservation of inner anatomical structures of the petrous bone and its possible high density contamination with sediments can limit the effective use of the technique.

The LA method can be applied as an ancillary protocol of sex estimation in regard to adult individuals from archaeological contexts. However, its applicability to the subadult individuals should be approached with caution, calling for further comparative studies, as do the effects of the process of cremation on the morphology of petrous parts of temporal bones, as well as age-related developmental changes.

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Authors' contributions

AH designed the research and secured its financing. MG completed and reported computed tomography. The resulting data were analysed and interpreted by AH. The paper was drafted and revised by AH and approved by MG.

Conflict of interest

The authors declare that there is no conflict of interest.

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Evolution of modern humans is a result of self-amplifying feedbacks beginning in the Miocene and continuing without interruption until now

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ABSTRACT: Humans are a part of the complex system of life. This consists of a multitude of feedbacks among all parts of living systems. In the case of human origins, many feedbacks became positive rather than homeostatic, thus producing self-amplifying effects in basic morphological and behavioural characteristics of emerging humans: erect bipedalism, social structure, tool-making, food procurement and environmental management, symbolic communication, sexuality, extended childhood, and mental capacities. These, plus many other human characteristics, changed gradually, though at varying rates, over the last 6 million years, producing directional variation in extant morphological and behavioural characteristics of what are considered modern humans. The change through time and geographic space of those characteristics is an ongoing dynamic process, thus it is futile to pose essentialist questions about the precise date and place of the modern human origins. Modernity is a process, not an endpoint.

Key words: autocatalytic, evolution, brain, hominins, human



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Introduction

Although the current understanding of human evolution away from the common ancestor with apes prefers the "branching tree model" there was only one way to produce presently living humans: by subjecting some late Miocene hominid to evolutionary forces that ultimately produced the reader of this text and contemporaneous human conspecifics. While all published trees by convention are shown branching upward from our Miocene ancestor to ourselves, they are imaginary (a fact that can be inferred from their very variety) and very few of them are validated by anything but repetitive group-think. An example of such a tree is in Figure 1.



Fig. 1. An example of an "evolutionary tree" of humans. Authors own drawing not based on any specific considerations of links between "fossil species". Just an example.

Instead, it is preferable to reverse perspective and look backward through the telescope that connects the broad observable reality of our current species with – from any reasonable evolutionary perspective – its logically essential and paleontologically documented (Galik et al. 2004) antecedent populations. The history of this subject, as history usually is with its failures and achievements, is what we are trying to explain here. Although any particular explanation may be only hypothetical and uncertain, it is absolutely necessary that the evolutionary process producing modern humans actually happened - look in the mirror if uncertain. With the current branching model of hominin evolution misrepresenting members of various populations such as Neandertals and even Flores "hobbits", as purported failures to become "moderns" (in the sense of morphology and behaviour rather than their time of existence), it is unavoidably essential that there was a direct ancestor-descendant line that. proceeding through the copulations and births in every 250,000 antecedent generations, produced modern humans from some Miocene/Pliocene ancestral population. Differential reproduction in each of those 250,000 generations enabled natural selection to enhance biological processes that underlie complex cognition of modern human individuals. Interaction of members of each generation with their surroundings and with other members of their populations influenced epigenetic processes shaping the course of individual ontogenies, especially their early parts: infancy, childhood and adolescence, and resulted in altered natural environments and social structures with which all our ancestors had to interact.

The fossil record of hominin morphologies and the corresponding archaeological records of results of hominin behaviours are all imperfect due to the vagaries of taphonomy and geological preservation. Nonetheless, where fossil or archaeological sampling has provided reasonable sample sizes of skeletal remains or artefacts, it can be seen that the evidence shows improvements in bodily characteristics or technological abilities of some hominins increasing at a double exponential rate through the last few million years. The characteristic investigated should be measured on an interval (continuous) scale.

The general form of the double exponential equation is

$$y = a^{b^{3}}$$

where y - a dependent variable, x - an independent variable (e.g. time), a, b, constants.

As an example, for the increase in hominin cranial capacity representing the increase in body size the equation is (Fig. 2)

$$CC = 190.0(7.5^{0.9997^{DATE}})$$

Where CC is measured in millilitres (ml) and DATE in thousands of years (ka).

The double exponential rate of increase is the basic formal characteristic of self-amplifying systems.

Double exponential regression of hominin cranial capacities found until the end of Pleistocene (N=233) on their dates has a strong fit to actual data (r=0.92). Variance of actual capacities around the fitted line (stdev=164) does not differ significantly from the variance of brain size among the one species of modern humans (stdev=157, F=1.09; Henneberg 1990).

Cranial capacity is easy to quantify, while technological advances or other traces of behaviours do not lend themselves easily to reliable quantification (Levendis et al. 2019). This evidential constraint, however, does not mean that cultural innovations are not a result of self-amplifying feedback relations. The following basic behavioural characteristics often are mentioned, as being involved in the operation of the human system of positive feedbacks (Bielicki 1969; Tobias 1981; Henneberg 1992; McKee 2017): erect bipedalism, food procurement, technologies, symbolic communication, extended childhood, sexual behaviours, social organisation, cognitive abilities. Erect bipedalism, however acquired, became a very energetically efficient way of locomotion on the ground and in shallow waters, freeing up the upper extremities. These could be then used for transporting food, weapons/ tools, raw materials and children in a position facing the carrier. Similar to trunks of proboscideans, this shift extended the range of individuals' manipulation of the environment, and added to it increased interaction between care-givers and offspring. The erect posture liberated hands that then could manipulate various objects including foodstuffs, tools/weapons, fuels and building materials even during locomotion and without use of teeth and mouths; mouth then could be used to

send voice signals improving interindividual communication. Such effects of erect bipedalism clearly enhanced food acquisition and food sharing, and facilitated development of technologies, rearing of offspring, symbolic communication that in their own way interacted with the enhanced mental abilities and social organisation. Concomitant changes in sexual behaviours improved cooperative parenting and cooperation in food acquisition (to a large extent multi-person foraging, scavenging and hunting). Changed sexual behaviours benefitted from ovulatory crypsis (Lovejoy 2009) allowing more frequent, nearly continuous, female sexual receptivity, promoting monogamous unions with males that might also contribute to cooperative parenting.



Fig. 2. Illustration of the quantitative operation of the self-amplifying feedbacks on human body size as represented by the volume of the cranial cavity. Data taken from De Miguel and Henneberg (2001) and Henneberg and De Miguel (2004) supplemented by data on new finds (Jacob et al. 2006; Berger et al. 2010; De Ruiter et al. 2019).

Collaborative parenting provided an opportunity to prolong childhood, a period of rapid and extensive learning. A prolonged childhood period enabled individuals to acquire knowledge and skills needed to use more sophisticated technologies and to participate in more intricate social structures. All these changes eventually required more learning thus demanding a further extension of the period of childhood. Improved technologies produced more efficient ways of food procurement resulting in greater access to nutrients. This interactive complex resulted in larger number of individuals being able to live together and cooperate in child rearing and food procurement but complicated social relations that had to be modified.

Improved social relations enhanced collaborative parenting and coordinated food procurement that resulted in increased group sizes requiring further development of social organisation. Such improvement was made possible by developed mental capacities and inter-individual communication (language). Improved mental abilities and more efficient (eg. symbolic) communication contributed to better social organisation that enhanced production and use of new technologies and more abundant food acquisition. Greater food abundance allowed larger group size, and so on and so on, iteratively through vast stretches of time... The scheme has many uncertain details but its general heuristic value is undeniable (Figure 3).

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Fig. 3. A schematic representation of autocatalytic feedbacks operating during human evolution.

Evidence for the operation of this set of self-amplifying feedbacks extends far beyond the simple example of cranial capacity increase. Initial low population density of hominins, evidenced by just a few fossil finds from the Miocene and Pliocene, expanded exponentially into Pleistocene leaving fossil evidence everywhere in the Old World, even if one discounts obvious taphonomic limitations and natural vagaries of preservation. The initial scarcity of (mostly stone) artefacts of Lomekwi and of Oldovan technologies in time flourished into Acheulian industries of Lower and Middle Pleistocene and abundance of human made objects, not only technologically useful but also decorative, and into natural surfaces' alterations (e.g. "cave art") of the Upper Palaeolitic and climaxed in the management of plants, animals, soils, watercourses and building materials of the Neolithic. The abundance of human ways to manage the environment that occurred in the Neolithic, altered direction of some feedbacks so that, for example, increasing body and brain size as an effect of better food acquisition, producing collateral effects of greater protection against attacks of predators and imoproved hunting efficiency, later reversed towards reduction in body size (gracilisation, Debec 1960; Schwidetzky 1962) and brain size (Henneberg 1988; Henneberg and Stevn 1993) to lower metabolic costs of body mass in the situation of strong group protection and more reliable, though quantitatively limited, food supply from agricultural activities.

The mechanism described here produced, at any time it operated, a variety of morphologies and behaviours that still were capable of engaging in the set of positive feedbacks, and continued towards characteristics that we now perceive as those of modern humans. The rates of morphological and behavioural changes may have varied at different periods, but continuity of change has not been interrupted in the set of our ancestral populations. Therefore, search for a single moment in time when 'modernity' appeared, or for the individual who could be the earliest holotype of modern *Homo sapiens* is a futile exercise because human variation in time and geographical space is shaped by ongoing processes, not a single act of ... creation... by forces, however natural.

Authors' contributions

Both authors contributed equally to this paper's contents.

Conflict of interest

Authors declare no conflict of interests.

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Association of early menarche with elevated BMI, lower body height and relative leg length among 14- to 16-year-old post-menarcheal girls from a Maya community in Yucatan, Mexico

Sudip Datta Banik

ABSTRACT: Human body segments have different timing and tempo of growth. Early menarche (EM) as an indicator of early reproductive maturity results in a shortened height and leg length. Relatively larger trunk may increase risk for more body fat deposit and higher body mass index (BMI) due to the allometry of total body fat with body proportions. The objective of the study was to assess the association of EM with BMI, absolute body size [height, sitting height (SH), subischial leg length (SLL)] and relative body dimensions [sitting height to subischial leg length ratio (SHSLLR), relative subischial leg length (RSLL) among 14- to 16-yearold post-menarcheal girls from a rural Maya community in Quintana Roo, Yucatan, Mexico. In a cross-sectional study, post-menarcheal girls (n=51) aged 14 to 16 years had EM (n=22) (<12 years of age) and not early menarche (NEM, n=29). Anthropometric measurements of height, weight, and SH were recorded. Derived variables were BMI, height and BMI-for-age z-scores, SLL, SHSLLR, and RSLL. Mean value of age at menarche (AM) was 13 years (EM 11 years, NEM 14 years). Mean values of height (EM 159 cm, NEM 164 cm), BMI (EM 20 kg/m², NEM 19 kg/m²), sitting height (EM 81 cm, NEM 78 cm), SLL (EM 79 cm, NEM 85 cm), SHSLLR (EM 102.93%, NEM 92.03%), and RSLL (EM 49%, NEM 52%) were different (p<0.05) in the two groups. BMI showed significant negative correlation with AM (Pearson's r=-0.29, p<0.04). Linear regression models adjusted for age showed that EM had different interrelationships (p < 0.05) with body dimensions: positive with BMI, SH, SHSLLR, and negative with height, SLL, and RSLL. Earlier AM was associated with higher BMI, SH, SHSLLR and lower SLL, RSLL, explaining lower body height and leg length among the participant EM girls. In the light of life history theory, EM results in a growth trade-off, short stature and larger trunk relative to leg length that might enhance risk for body fat gain.

KEY WORDS: Early menarche, BMI, sitting height, leg length, body proportion



Introduction

Early menarche (EM), defined by at least one menstruation before the twelfth birthday of a girl (Arcoverde et al. 2020; Must et al. 2005) is reported to be associated with lower body height in comparison with peers of not early menarche (NEM) when socioeconomic and other environmental factors remain constant in both groups (Conway et al. 2012; Kang et al. 2019). A previous study also reported the association of EM with early age at peak height velocity and low leg length-to-sitting height ratio (Conway et al. 2012). EM acts as a risk factor for developing overweight and obesity in adolescence and adulthood (Martínez et al. 2010; Rosenfield et al. 2009). Several studies have examined issues regarding menarche and adiposity: Brazil (Arcoverde et al. 2020), Iran (Pejhan et al. 2013), Korea (Oh et al. 2012), Kuwait (Al-Awadhi et al. 2013), Mexico (Datta Banik et al. 2015), Spain (Labayen et al. 2009), and the USA (Adair and Gordon-Larsen 2001). However, it is not clear from the literature review whether obesity and higher body fat are the responsible factors for EM or earlier age at menarche (AM) plays an important role in the development of body fat and other health risks like cardiovascular diseases and even cancer in adulthood (Datta Banik and Dickinson 2014, 2016; Méndez-Domínguez 2011).

Data on menarche and physical growth in girls are reported from the northeastern states of Mexico (Méndez-Estrada et al. 2006) and other regions like Oaxaca (Malina et al. 2004) and Yucatan (Méndez-Domínguez 2011). However, reports on the association of EM with differential growth patterns of height and relative body dimensions (leg length and trunk length relative to height) are not available. AM has been reported to be associated with household socioeconomic factors, mother's and grandmother's childhood living conditions in Yucatan, Mexico (Azcorra et al. 2018; Datta Banik and Dickinson 2016). Poor household socioeconomic status and living conditions of the Maya community in Yucatan are important factors that are responsible for poor nutrition and physical growth of children (Azcorra et al. 2016, 2018).

Life history theory integrated with a biocultural approach indicated that early reproductive maturity results in a growth trade-off and short stature in a sample of adult women in the U.S. (Rivara and Madrigal 2019). Leg length and other body dimensions relative to height among individuals are used as proxy of early life environmental conditions that influence physical growth in children (Bogin 2012; Bogin and Varela-Silva 2010). Shorter relative subischial leg length (RSLL) is a marker that indicates negative impacts of adverse environmental conditions on physical growth in early childhood; infection, disease burden, poor nutrition, and chronic energy deficiency may result in growth trade-off with respect to absolute size and relative body dimensions (Bogin and Varela-Silva 2010). Lower leg length (knee height) showed earlier age at maximum increment compared to standing height and, thereby, indicated earlier maturity among boys and girls from Merida, Mexico (Datta Banik et al. 2017) that was explained by the facts of differential growth and maturity patterns of different body dimensions (Bogin 2021). In such a condition, risk for body fat gain is caused by larger trunk relative to leg length due to earlier onset of puberty. In addition, early puberty is reported to be associated with psychosocial stress developed due to poor household socioeconomic status, absence of father, etc. (Braithwaite et al. 2009; Deardorff et al. 2011, 2014; Henrichs et al. 2014; Steppan et al. 2019; Sun et al. 2017; Webster et al. 2014). Studies also reported that growth failure and delayed maturation may occur due to persistent nutritional stress, particularly in small-scale societies (McIntyre and Kacerosky 2011); however, growth rate increases and menarche occurs earlier if energy supply elevates that might lead to growth trade-off, shortened height, and higher body fatness (McIntyre and Kacerosky 2011; Wells 2018). Studies from Yucatan showed an association between better living conditions and higher body fat with earlier AM (Azcorra et al. 2018; Méndez-Domínguez 2011).

Association between higher BMI and body fat with lower relative leg length has been reported from the USA (Bogin and Varela-Silva 2008) and India (Datta Banik 2022). In the present study, I was interested to test the hypothesis in a new sample from another non-European and low-tomiddle-income country (Mexico), which was higher BMI associated with lower RSLL, especially among girls with EM. The objective of the present study was to find an association of EM with BMI, absolute body size [height, sitting height (SH), subischial leg length (SLL)] and relative body dimensions [sitting height to subischial leg length ratio (SHSLLR), relative subischial leg length (RSLL)] among 14- to 16-year-old post-menarcheal girls from a rural Maya community in Quintana Roo, Yucatan, Mexico.

Participants and methods

A cross-sectional study was carried out in 2019 at Quintana Roo, a rural Maya community in Yucatan, Mexico. The present report is descriptive in nature. Participants were 14- to 16-year-old girls. The study was approved by the bioethics committee of the Centre for Research and Advanced Studies (Cinvestav-IPN); the parents / caregivers and two witnesses from the community signed the consent form as per guideline of the committee and the participants gave their verbal consent before the commencement of the study.

According to the official records, the population size of the community in Quintana Roo (942 individuals in 2010) was the smallest among the 106 municipalities covering rural and urban areas of Yucatan (INEGI 2011). The population may be declining, as a recent household survey carried out in 2018–19 (López-Moreno 2021) recorded 780 individuals (385 males, 395 females) living in the community. However, individuals of the community who lived in the nearest cities and abroad for work were not counted in that survey.

Selection criteria for the participants in the present study were: 14 to 16 years old girls with post-menarcheal status, without any reported physical and mental health burden, residents of Quintana Roo municipality since birth, and both parents had at least one surname of Maya origin. Previous studies in Yucatan reported very few girls above 14 years of age had premenarcheal status (Datta Banik and Dickinson 2014).

A household sociodemographic survey recorded menarcheal status and 65 girls in this age group (14 to 16 years) reported age at menarche that was confirmed by their mothers and grandmothers. Early menarche (EM) was defined as menarche before the 12th birthday (Must et al. 2005); 25 girls of the present study had EM. Other participant girls (n=40) had menarche after 12 years of age (not early menarche or NEM). Only two girls were non-menstruating in the age group, and they were not included in the study. Sampling was probabilistic; sample size (EM=21, NEM=29) was estimated separately from the two sub-populations (EM=25, NEM=40) with 10% margin of error at 95% confidence level, and participants were selected at random from the list of individuals in each group. One EM girl in the community wished to participate in the survey and the final sample (n=51) of the present study included 22 EM and 29 NEM girls.

In Mexico, people use both paternal and maternal surnames. Maya surname was used as a proxy of genetic background of the population and to represent Maya ancestry (Vázquez-Vázquez et al. 2013); surnames were identified by an expert (see Acknowledgements). The parents of the girls had at least one surname of Maya origin and therefore, Maya ancestry of the participants was considered. Data of household characteristics (housing pattern, education, and occupation) were collected.

Anthropometric measurements were recorded following standard protocol (Lohman et al. 1988). Height (cm) and sitting height (SH in cm) were measured to the nearest 0.1 centimeter using a standard stadiometer with platform (Seca, Germany). Stadiometer and a standard anthropometric box [40 cm $(tall) \times 50 \text{ cm} (wide) \times 30 \text{ cm} \text{ deep}$ were used to measure SH. Body weight was measured to the nearest 0.1 kg using a digital scale (Tanita Corp., Japan). Subischial leg length (SLL) was obtained from the difference between stature and sitting height. Other derived variables were body mass index (BMI, kg/m²), relative sitting height to subischial leg length ratio (SHSLLR) (SH/SLL x 100), and relative subischial leg length ratio (RSLL) (SLL/

height×100). Height-for-age and BMIfor-age Z-scores were calculated (de Onis et al. 2007; WHO 2007). All measurements were recorded on household visits by a single researcher (the author) and intra-observer technical error of measurement was within acceptable limits (Ulijaszek and Kerr 1999). An adult woman from the community or caregiver/mother was present when anthropometric measurements were being recorded from the participants. No girls were menstruating at the time of recording anthropometric measurements.

SPSS statistical software (version 15.00) was used for data analysis and descriptive characteristics of the sample for age and anthropometric variables (mean values and standard deviation) were calculated; significant differences of mean values of parameters between EM and NEM girls were estimated using Student's t-test. Linear regression models predicting height, BMI, SH, SLL, SHSLLR, and RSLL were developed to explain the interrelationships between EM and the dependent variables, after adjusting for age. The level of statistical significance was set at p < 0.05 in all analyses.

Results

Household socioeconomic characteristics of the participant girls were similar; land ownership (100%), housing pattern (construction, number of rooms, toilet, kitchen), water connection and electricity (100%), parents' education (majority had school education) and occupation (daily wage laborer and small-scale cultivators in different government programs), civil status (84% of mothers were married and others were either unmarried or divorced), and average monthly per capita income (\$90 US). Mean age (± SD) of the girls was 15.06 years that did not vary significantly (p>0.05) between EM (14.84 years) and NEM girls (15.22 years); t and p-values referred to the differences between EM and NEM groups (Table 1). Mean value of age at menarche (AM) of the participant girls was 12.91 years; significant difference (p<0.05) of AM was found in EM (11.03 years) and NEM (14.33 years) girls.

Table 1. Descriptive statistics of age, age at menarche, and anthropometric characteristics of post-menarcheal girls (n=51)

Variables	All (n=51) Mean (SD)	EM (n=22) Mean (SD)	NEM (n=29) Mean (SD)	t	p-value
Age (years)	15.06 (1.52)	14.84 (1.64)	15.22 (1.42)	-0.90	0.37
AM (years)	12.91 (2.01)	11.03 (0.76)	14.33 (1.39)	-10.02	< 0.0001
Height (cm)	161.87 (7.38)	159.32 (7.29)	163.80 (6.95)	-2.24	0.03
Weight (kg)	50.62 (7.26)	50.95 (8.54)	50.36 (6.25)	0.28	0.78
BMI (kg/m²)	19.28 (2.19)	20.00 (2.61)	18.73 (1.66)	2.10	0.04
Sitting height (cm)	79.42 (3.42)	80.67 (3.45)	78.47 (3.13)	2.38	0.02
SLL (cm)	82.45 (5.78)	78.65 (5.40)	85.33 (4.23)	-4.96	< 0.0001
SHSLLR (%)	96.74 (7.31)	102.93 (6.90)	92.03 (2.56)	7.84	< 0.0001
RSLL (%)	50.90 (1.84)	49.33 (1.68)	52.08 (0.73)	-7.93	< 0.0001
					1

EM: Early menarche; NEM: Not early menarche; SD: Standard deviation; AM: Age at menarche; BMI: Body mass index; SLL: Subischial leg length; SHSLLR: Sitting height to subischial leg length ratio; RSLL: Relative subischial leg length; t and p-values refer to the differences between EM and NEM groups.

Anthropometric parameters had shown significant differences (p < 0.05) of mean values between EM and NEM girls, except body weight. The NEM girls were taller, had longer subischial leg length (SLL) and relative to height (RSLL), lower BMI, sitting height, and sitting height to subischial leg length ratio (SHSLLR), in comparison with the corresponding characteristics among EM peers (Table 1). Only one EM girl had height-for-age Z-score below -2.0 SD; other girls (n=50)were not stunted (low height-for-age). No girl had low or high BMI that could have been attributed as suffering from either undernutrition or having excess weight (overweight and obesity), respectively. Significant correlation (p<0.001) was observed between AM and anthropometric characteristics (BMI r = -0.30, SH

r = -0.67, SHR r = -0.62, SLL r = 0.42, RSLL r = 0.62, and SHSLLR r = -0.61).

These results (not presented in tables) raised my interest to further explore the interrelationships between EM and anthropometric characteristics (absolute body size and relative to height). Linear regression models were used to find the interrelationships between EM (yes=1, no=0) and anthropometric characteristics (SH, SHR, SLL, SHSLLR, and RSLL), after adjustment for age among girls (Table 2). Parameter estimates of the response variable within 95% confidence interval for the coefficient showed significant interrelationships between variables that was estimated by ANO-VA (p < 0.05). The regression models accounted for >60% of total variability explained by adjusted R². EM was found

Dependent	Predictors	В	SEE	t	<i>p</i> -value	95% Confidence Interval for B	
variable						Lower	Upper
Model 1: Height (cm)	Constant	148.36	10.09	14.70	< 0.001	128.07	168.66
	Age (years)	1.01	0.66	1.54	0.13	-0.31	2.34
	EM	-4.09	2.00	-2.05	< 0.05	-8.11	-0.08
	Constant	14.46	3.03	4.77	0.00	8.37	20.55
Model 2: BML (kg/m^2)	Age (years)	0.28	0.20	1.42	0.16	-0.12	0.68
	EM	1.37	0.60	2.29	0.03	0.17	2.58
	Constant	73.85	4.72	15.64	< 0.001	64.36	83.35
Model 3: SH (cm)	Age (years)	0.30	0.31	0.99	0.33	-0.32	0.92
	EM	2.32	0.93	2.48	0.02	0.44	4.19
	Constant	74.51	6.76	11.02	< 0.001	60.92	88.10
Model 4:	Age (years)	0.71	0.44	1.61	0.11	-0.17	1.60
	EM	-6.41	1.34	-4.80	< 0.0001	-9.10	-3.72
	Constant	99.47	7.08	14.05	0.00	85.24	113.71
Model 6:	Age (years)	-0.49	0.46	-1.06	0.29	-1.42	0.44
5115LLI(//0)	EM	10.71	1.40	7.65	< 0.0001	7.90	13.52
	Constant	50.16	1.77	28.42	0.00	46.61	53.71
Model 5: RSLL (%)	Age (years)	0.13	0.11	1.10	0.28	-0.11	0.36
K3LL (70)	EM	-2.70	0.35	-7 75	< 0.0001	-3 40	-2.00

Table 2. Regression models to establish interrelationships between early menarche (EM) and body dimensions among post-menarcheal girls (n=51)

SH: Sitting height; EM: Early menarche (EM=1, Not early menarche=0); B: Regression coefficient; SEE: Standard error of estimate; SLL: Subischial leg length; SHSLLR: Sitting height to subischial leg length ratio (%); RSLL: Relative subischial leg length (%).

to be significantly related to the dependent variables (height, BMI, SH, SLL, SHSLLR, and RSLL) separately, after adjusting for age among girls (p<0.05). Interrelationships between EM and anthropometric parameters were different; regression coefficients were negative with height, SLL, RSLL, and positive with BMI, SH, SHSLLR. It was observed that EM girls had a risk for having 4.09 cm lower height, 1.37 kg/m² higher BMI, 2.32 cm higher trunk size (SH), 6.41 cm lower SLL, 10.71% higher SHSLLR, and 2.7% lower RSLL than NEM peers, holding age as another predictor in the model constant. In the normality tests (Shapiro-Wilk tests), distribution of the residuals was normal (p>0.05) and that showed no patterns. Relatively high tolerance (>0.97) and low variance inflation factor (<1.02) indicated no multicollinearity among predictors.

Discussion

The results of the present study among 14- to 16-year-old post-menarcheal girls from a Maya community in rural Yucatan, Mexico, showed significant interrelationships between EM and growth of body dimensions in terms of their absolute size and relative to height. It was observed that a relatively larger trunk (sitting height), and its relative size (SHSLLR) resulted in shorter leg length (SLL), its relative estimate (RSLL), and higher BMI among EM girls in comparison with NEM peers. The results from a rural community of a non-European and low-to-middleincome country (Mexico) further support the hypothesis that higher BMI is associated with lower RSLL (Bogin and Varela-Silva 2008, 2010).

Age at menarche (AM) as an indicator of sexual maturity was reported to be associated with physical growth, body fatness and health among girls during adolescence and physical and mental health adulthood (Méndez-Domínguez in 2011). A trend of decline in AM in the previous decades has been observed in different countries (Demerath et al. 2004; McDowell et al. 2007; Gomula and Koziel 2018). In Yucatan, Mexico, mean values of AM among girls (12.09 years in Merida City and 12.24 years in the municipality of Progreso) were lower than their mothers' AM (12.66 years and 12.41 years, respectively) (Wolanski et al. 1994). Other studies in Merida, Yucatan reported lower mean AM (11.83 years, Datta Banik et al. 2015, 11.21 years, Datta Banik and Dickinson 2016, 11.57 vears, Datta Banik et al. 2020). Decline of AM over decades in some other Latin American countries was also reported from Colombia (Chavarro et al. 2004; Ireton et al. 2011) and Chile (Codner et al. 2004; Hernández et al. 2007). However, the participant girls (n=51) in the present study had a mean age at menarche of 12.91 years that was higher than that reported earlier from the Merida City,

Yucatan. The result needs verification in future studies with a larger sample size.

Secular trend of height and improvement of leg length in terms of relative increase of knee height (lower leg length) among daughters of women representing the Maya community in Yucatan explained intergenerational influences, rural to urban migration, and substantial improvement in their living conditions (Azcorra et al. 2015). Maternal short stature has been reported to be associated with higher body fat in Maya children in Merida, Yucatan (Azcorra et al. 2016). Higher body fat has been found to be positively correlated with EM in girls from Merida, Yucatan (Datta Banik et al. 2015).

AM as a maturity indicator among girls depends on the environmental factors as well as genetic predisposition (Mukherjee and Datta Banik 2009: Morris et al. 2010, 2011). A study in Yucatan showed better living conditions experienced by mothers and adult daughters during their childhood lowered AM (Azcorra et al. 2018). Therefore, consistent with life history theory and intergenerational influences, maternal biological capital (short stature, body fat) may transmit characteristics to the offspring that influence child growth and development like early maturity in girls, as well as higher levels of body fat and short stature (Wells 2018; Wells et al. 2019). In the present study, EM as a maturity indicator, probably caused by several environmental factors including maternal life history trade-off, was found to be associated with growth trade-off of leg length. However, data on maternal anthropometric characteristics were not available, which was a limitation of the study. On the other hand, NEM girls with relatively slower maturation had more time for growth of leg length, its absolute size and

relative to stature and trunk that might have lowered the risk of body fat gain as indicated by lower BMI in the present study.

Differential growth patterns of body dimensions (absolute size and relative to height) in children and adolescents are responsive to environmental conditions including nutrition and diet, disease, and household psychosocial and economic factors. Leg length and its relative size to trunk (sitting height) and stature (standing height) are the markers of quality of early life living conditions and probable epigenetic effects that determine physical growth and increased risk of body fatness. Developmental plasticity (Bateson et al. 2004) shaped by natural selection is one of the adaptive strategies in response to unfavorable environmental conditions, chronic diseases, and higher energy expenditure for recovery, leads to a consequential growth trade-off and results in smaller absolute size and relative body dimensions. Fetal origins of developmental plasticity, mother to offspring energy pathways and metabolic adjustment explain how early life environmental conditions and maternal capital (biological, social, material) can influence child growth, nutrition, and development (Kuzawa 2005; Wells 2010). The adaptive model of developmental plasticity is explained as developmental adjustment in response to maternal nutritional deficits in its prenatal life. The Developmental Origins of Health and Disease (DOHaD) hypothesis also supports the phenomenon of influences of early life nutrition and growth that shape adult phenotype (Barker 2004, 2007). The adaptive model of developmental plasticity explained the adjustment of phenotype in utero, in anticipation of postnatal life environmental constraints; however, reports stated

that adaptive models did not emphasize maternal phenotype as the initial sources and information received by the offspring (Wells 2010, 2012, 2017, 2018). In light of the thrifty phenotype hypothesis, it can be explained that influences of adverse early life experience include shortened final body size and relative leg length (relative subsichial leg length or RSLL) – a relatively larger trunk may result in central obesity and more body fat deposit and their association with enhanced risk for non-communicable diseases in adulthood and "maternal capital" plays an important role (Hales and Barker 1992; Wells 2011).

Neonates smaller at birth undergo catch-up growth with faster increment in size and enhanced risk for body fat gain in late pre-school age (Ong et al. 2000) that may have negative consequences including early puberty, maturity, and shorter stature, compared to their peers who were larger neonates. A child born with suboptimum energy resources will face risk of growth failure, particularly in lowand middle-income countries (Mertens et al. 2020; Roth et al. 2017). However, higher input of energy, later in childhood and adolescence due to unhealthy food habits of consumption of processed and ultra-processed foods and sugary drinks and low physical activity may cause excess weight and body fat gain (metabolic loads) (Torres-Arroyo 2018) that might be associated with earlier onset of maturity and consequent shortened final body size, and relative leg length and elevated risk for the development of NCDs in adulthood (Méndez-Domínguez 2011). Coexistence of short stature and excess weight (overweight and obesity) in terms of nutritional dual burden is common in Yucatan population (Varela-Silva et al. 2012). Along with, consumption of non-essential energy-dense, industrially processed and ultra-processed foods and drinks are very high in Mexican populations (Bogin et al. 2014; Illescas-Zárate et al. 2021; López-Moreno 2021; Mendoza et al. 2017)

Life history strategies determine physical growth, development, sexual maturity, reproductive fitness, and survival (Rivara and Madrigal 2019). A mismatch between available energy and limited capacity to access may result in trade-offs that can be interpreted by a biocultural approach of evolutionary life history theory (McDade 2003). Sexual maturity in adolescence determines reproductive health and success in the life of women: proximate and ultimate causes and consequences of EM (Gillette and Folinsbee 2012) may have negative impacts on fecundity, quality, and quantity of offspring that can be analyzed in an evolutionary framework (Rivara and Madrigal 2019). EM as an indicator of early sexual maturity and its association with short stature and lower relative subischial leg length (RSLL) can also be explained in terms of evolutionary mechanisms of life history trade-off and intergenerational transmission of maternal capital, both in utero and postnatal. In this regard, Social-Economic-Political-Emotional (SEPE) inequalities and insecurities are important factors for growth failure in children (Bogin and Varea 2020; Scheffler et al. 2019). A life course, intergenerational model of SEPE factors (stressors) are viewed in human ecological perspectives of biology and behaviour, child growth, development, and health. In a literature review, Wells (2018) hypothesized that in an intergenerational perspective on developmental plasticity, life history trade-offs in the maternal generation favor the emergence of similar trade-offs

in the offspring generation. Allocation of relatively higher energy to growth during pregnancy and lactation promotes the development of capacity in offspring for greater fat-free mass and metabolic turnover in life, adult size, and reduced risk for NCDs (Wells et al. 2016).

Intergenerational transmission of maternal life history trade-offs was explained in a study from the United Kingdom that showed mothers with earlier menarche were obese, short in height, and their daughters also had faster infant growth, EM, shorter final size and also gained fat in adolescence (Ong et al. 2007). EM causes earlier maturation of leg length and therefore, results in short stature and RSLL (Conway et al. 2012). Association of EM with higher BMI, obesity, and body fatness was also reported from the USA. (Adair and Gordon-Larsen 2001). Household socioeconomic status and lifestyle habits have been correlated with the development of an obesogenic environment and EM in girls, as reported from Chile (Amigo et al. 2012; Codner et al. 2004; Hernández et al. 2007), Iran (Pejhan et al. 2013; Shalitin and Phillip 2003), Korea (Oh et al. 2012), Poland (Wronka and Pawlinska-Chmara 2005), Portugal (Padez 2003), Spain (Labayen et al. 2009) and other countries (Al-Awadhi et al. 2013; Freedman et al. 2002, 2003; Martínez et al. 2010; Rosenfield et al. 2009). Prenatal and early childhood growth might have a larger effect on sexual maturation (Yermachenko and Dvornyk 2014) and therefore, higher body fat and early sexual maturity in terms of EM may be predicted.

Early sexual maturity among adolescent girls can be viewed as an adaptive strategy to cope with the adverse conditions of SEPE inequalities and insecurities, caused by several biocultural and environmental factors that influence the early start of reproduction, predicting lower probability of survivorship. Therefore, EM is a mismatch between biological and psychosocial maturation that can be interpreted as an alternative reproductive tactic, determined by several environmental factors, and assists a woman to gain phenotypic plasticity, alteration of behaviour. Furthermore, EM results in a life history trade-off that might be transmitted through generations (Gillette and Folinsbee 2012; Wells 2018). A study from Finland reported women were compatible with trade-offs between reproduction and growth; compromised adult height at the cost of early AM. Therefore, women gained fitness benefits by an early start of reproduction but not by taller final size (Helle 2008). Early menarche results in the growth tradeoff of height and leg length in exchange of reaching reproductive age earlier, and thereby increases the risk of body fat deposit in higher relative trunk size. The results in the present study showed that EM girls in their post-menarcheal age had lower height and leg length (SLL) relative to height and sitting height in comparison with NEM peers.

The present study in a cross-sectional design limits the temporal association of EM with elevated BMI, shortened height and leg length. In addition, data of maternal phenotype (height, leg length, BMI, body fatness) were unavailable that could enrich the study. Household socioeconomic status of the participant girls was similar, and data were not used to find association with AM. Significant differences in socioeconomic status might be found in larger samples. The results need further verification in future studies representing other populations in different biocultural environmental situations.

Conclusion

In summary, the present study showed association of EM with lower body height and relative leg length that support earlier studies. Early onset of puberty and sexual maturity among adolescent girls have impacts on demographic shifts in fertility, fecundity, birth rates that are associated with biocultural environmental factors in the prenatal and postnatal stages of growth and development. Maternal capital, environmental factors, SEPE inequalities and insecurities in a woman's early life and during pregnancy, as well as nutritional status, consequently determine the qualities of offspring like birth weight, length at birth, immune response, growth and development trajectory, adult body size, relative body dimensions, and body fat that can be interpreted by the intergenerational transmission of evolutionary trade-offs in growth, maturity, and development.

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Conflict of interest

The author declares no conflict of interest.

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Impact of Infectious Disease on Humans and Our Origins

Petar Gabrić

Abstract: On May 16, 2020, the Center for Academic Research and Training in Anthropogeny organized the symposium "Impact of Infectious Disease on Humans and Our Origins". The symposium aimed to gather experts on infectious diseases in one place and discuss the interrelationship between different pathogens and humans in an evolutionary context. The talks discussed topics including SARS-CoV-2, dengue and Zika, the notion of human-specific diseases, streptococci, microbiome in the human reproductive tract, *Salmonella enterica*, malaria, and human immunological memory.

Key WORDS: human evolution, paleogenetics, immune system, anthropogeny, SARS-CoV-2, dengue, Zika, streptococcus, *Salmonella*, malaria

The symposium, "Impact of Infectious Disease on Humans and Our Origins" was held virtually on May 16, 2020. The symposium was organized by the Center for Academic Research and Training in Anthropogeny (CARTA) of UC San Diego, a virtual organization promoting transdisciplinary research into human origins. The symposium aimed to gather experts on infectious diseases in one place and discuss the interrelationship between different pathogens and humans from an evolutionary context. The gathering was organized as a response to the ongoing COVID-19 pandemic. As of June 1, 2020, the videos from the symposium are available in digital format on CAR-TA's official website (https://carta.anthropogeny.org). A highly informative glossary for non-experts on infectious diseases is available on the event's page. While all speakers managed to be, in a general way, understandable for non-experts on infectious diseases, readers interested in evolutionary anthropology are especially encouraged to check out the talks by Nissi Varki, Amanda Lewis, and Elizabeth Winzeler.

The symposium was opened by Pascal Gagneux, associate director of CARTA,



Original article © by the author, licensee Polish Anthropological Association and University of Lodz, Poland This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution license CC-BY-NC-ND 4.0 [https://creativecommons.org/licenses/by-nc-nd/4.0/] Received: 2021-07-30. Accepted: 2022-02-16 who briefly introduced the concept of anthropogeny and its transdisciplinary character (cf. Gagneux 2021). Gagneux also remarked that eventual paleoepidemics in human evolutionary history would have yielded "surviving populations" and would have, thus, presumably shaped human evolutionary trajectories. In the Q&A section, Susan Kaech similarly noted that such events would have produced founder effects.

Robert Schooley described the new coronavirus SARS-CoV-2 from biological and clinical perspectives, in many regards repeating the information already available to the general public via media. Schooley further gave a historical and epidemiological overview of the previous SARS and MERS outbreaks. Schooley emphasized the role of non-human animals in the transmission of these pathogens to humans, differentiating between (putative) hosts (different bat species) and intermediate species (e.g., the dromedary camel in the case of MERS). The role of bats was particularly highlighted in this context as they can harbor viruses that can infect other mammals and cause diseases in them, and simultaneously remain, at least relatively, unaffected by the virus, partly as they have no bone marrow, and therefore, no B cells. Interestingly, Schooley noted that there exists archaeogenetic evidence that at least three coronavirus species were extant in association with human populations in the last 1,000 years: HCoV-NL63 (~ 500-800 years ago), HCoV-229E (~ 200-300 years ago), and HCoV-OC43 (~ 120 years ago) (Pyrc et al. 2006). Finally, although, Schooley welcomed the search for a COVID-19 vaccine, he found that research on antiviral treatments should receive more attention.

Sujan Shresta described the flaviviruses dengue and Zika from biological and clinical perspectives. These viruses are transmitted to humans by Aedes mosquitos. Infections with the viruses lead to dengue fever and Zika fever, respectively. Possible complications include congenital disorders such as congenital Zika syndrome and microcephaly. Crucially, after some 70 years of research, there are no effective treatments or vaccines for these two diseases. The usual antibody vaccine approach is not applicable for the dengue fever as there are four dengue virus serotypes that can infect the host independently of whether the same host was previously infected by another serotype. Indeed, secondary dengue infections are associated with more severe symptoms. Shresta and her team found in their investigations with mouse models that prior infections by the dengue virus provide cross-protection against Zika infections in the short run (Elong Ngono et al. 2017; cf. Mugabe et al. 2021; Pedroso et al. 2019). However, the Zika virus can 'evolve' to become so virulent that the pre-existing dengue immunity no longer affords protection against Zika. Curiously, Shresta noted in the Q&A section that the African genetic variation might be protective against dengue and Zika. Still, not much is known, and, problematically, the true prevalence and incidence of these diseases remain unknown due to the qualities of the health systems in areas affected by the epidemics.

Nissi Varki introduced the concepts of "definite", "probable", and "possible" candidates for human-specific diseases. Most putatively human-specific diseases are limited to infectious diseases. Varki compared causes of death in humans (historically vs. contemporarily) and chimpanzees and further empha-
sized the evolutionary importance of sialic acids in the differences between humans and other primates (and mammals) in the susceptibility to different pathogens. More precisely, several evolutionary changes in 2-6-linked sialic acid expression have occurred since the split from the last common ancestor of humans and the great apes, suggesting evolutionary conservatism of this phenomenon in non-human great apes and relatively sudden major changes in the human lineage (Gagneux et al. 2003). Furthermore, there is evidence that the human-specific loss of CMAH (cytidine monophospho-N-acetylneuraminic acid hydroxylase) is associated with some human-specific infectious diseases, including cholera (Alisson-Silva et al. 2018) and typhoid fever (Deng et al. 2014) (cf. Okerblom et al. 2017a).

Victor Nizet described the Group A streptococcus and Group B streptococcus from biological and clinical perspectives. Although these bacteria have been detected in other animal species (e.g., GAS: wild chimpanzees, GBS: some fish, cows), Nizet noted that non-human animal GBS infections highly differ from those described in humans. Nizet yet again emphasized the role of sialic acids and "molecular mimicry" in human infections by these bacteria. Namely, these bacteria contain sugar molecules resembling sugars found in humans (the so-called mechanism of "wolf in sheep's clothing disguise"), thus increasing bacterial disease potential (cf. Carlin et al. 2009).

Amanda Lewis discussed the microbiome in the human reproductive tract, which has been understudied. Notably, the vaginal microbiome can influence reproductive success, and thus, has the potential to be involved in selective

mechanisms. Lewis introduced two general macro-types of the human vaginal microbiome: the Lactobacillus-dominant microbiome and the "diverse" microbiome, which often consists of many bacterial types, with Gardnerella vaginalis displaying higher proportions compared to other bacteria (cf. Miller et al. 2016). Approximately 30% of women with the latter type also have bacterial vaginosis associated with limited reproductive success due to various mechanisms, including an overgrowth of anaerobic bacteria and a relatively high vaginal pH. Thus, the presence of G. vaginalis might be considered disadvantageous compared to the dominant presence of Lactobacilli. Interestingly, based on studies of non-human primates (Yildirim et al. 2014) and other empirical arguments, Lewis postulates that the "Gardnerella" microbiome type might be the "ancestral state" (cf. Gilbert et al. 2021; Tortelli et al. 2021). E.g., the lactobacilli dominance is not found in other primates, while women with bacterial vaginosis show similar microbiome compositions to healthy baboons and macaques, but the pH-values still tend to be lower in women with bacterial vaginosis compared to non-human primates (Miller et al. 2016). Furthermore, there is evidence that women with bacterial vaginosis display sialic acid depletion from epithelial glycans. On a side note, Lewis discouraged women from using vaginal "hygiene" products, stressing that "the vagina is a self-cleaning oven".

Manuela Raffatellu described Salmonella enterica from biological and clinical perspectives. While *S. enterica* comprises over 2,000 serovars that can colonize a variety of hosts, a few of these serovars infect only humans and are categorized as typhoidal Salmonella, as they cause the human-specific disease typhoid fever (cf. Behnsen et al. 2015). Intriguingly, Raffatellu pointed out in the Q&A section that *Salmonella* has been isolated in human remains dated to approx. 6,500 years BP and that there are indications that the human-adapted *Salmonella* emerged with neolithization (Key et al. 2020). Furthermore, Raffatellu noted that *Salmonella* has been isolated in pig remains dated to approx. 4,000 years BP, yet it is suggested that it was, in this case, transmitted from humans to pigs.

Elizabeth Winzeler described malaria from biological and clinical perspectives. Malaria is associated with high mortality in pre-reproductive children. Indeed, it is estimated that approx. 20% of children in the affected areas would have died due to malaria had antimalarial treatments not been discovered, thus, highly influencing the gene pools of these populations. Winzeler added that in some parts of Africa, it is frequently the case that the same individuals experience multiple infections in a given year. It is known that specific genetic variations protect against severe forms of malaria (and therefore, from malaria-caused death), suggesting the role of selective mechanisms in the evolutionary past. The most famous example is the sickle-cell allele. However, Winzeler emphasized that the exact mechanisms underlying this protection against malaria were not well understood and that this was not the only protective phenomenon, adding, e.g., the Duffy blood group antigen to the list. Furthermore, Winzeler discussed whether the evolutionary loss of CMAH in humans provides protection against severe malaria (cf. Rabinovich et al. 2017; Winzeler 2008; Okerblom et al. 2017b).

Susan Kaech described the mechanisms of human immunological memory, a phenomenon in which the immune system recognizes an antigen of a pathogen it has already encountered and successfully eliminated in the past. thereby enabling the system a relatively quick and efficient immune response. Kaech explained the process of the development of memory T cells, which, together with memory B cells, generally comprise the immunological memory. During infection, naïve T cells become effector T cells, indicating they are involved in the immune response during primary infection. Approximately 5% to 10% of effector T cells survive in the system as memory T cells after the infection has passed. Research has further shown that not all effector T cells have the same potential to become memory T cells. While research is limited, studies show that this potential depends on both genetic and environmental factors. Specifically, recent research has related memory T cell long-term survival with Interleukin-7 receptor (IL7R) expression (Kaech et al. 2003; Joshi et al. 2007).

The talks presented at the symposium offered a comprehensive overview of specific infectious diseases and/or infection-related phenomena for attendees interested in anthropogeny. This is incredibly valuable, given that these aspects have been at best understudied in the context of human evolution. Nevertheless, most of the talks were strictly focused on the biomedical aspects of these infection-related phenomena, while their relations to specific anthropological and/or archaeological data were only seldom established. This relative one-sidedness is perhaps somewhat unexpected given that topics merging human evolution and history, and infectiology are not unknown in the scientific literature: e.g., the presence of herpes simplex virus 1 in the hominin lineage as early as the split from the last common ancestor of humans and chimpanzees (Wertheim et al. 2014), the evolutionary association between humans and gorillas in the context of specific Pthirus species (sucking lice; Reed et al. 2007), the introduction of syphilis and other pathogen-caused diseases via Columbian migrations (Harper et al. 2011; Majander et al. 2020), etc. Still, a large amount of the presented data might bear implications for human evolution, highlighting the need for further integration of knowledge between infectologists and human evolution experts and future symposia of this type. In this regard, CARTA's general initiative is to be applauded and encouraged without qualification.

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Sex-Based Differences in Age-Related Changes of the Vertebral Column from a Bronze Age Urban Population in Ancient China

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ABSTRACT: The health disparities between males and females in bioarchaeological settings are important indicators of gender-based differences in socioeconomic roles. In this study, sex-based differences of the vertebral column in spine pathology were investigated in human skeletons excavated from a Bronze Age cemetery of the Western Zhou Dynasty at the Dahekou site in Shanxi, China. Results demonstrated that females had a higher prevalence of vertebral compressive fractures, with the majority found in those between twenty-five and thirty years old, suggesting that the fractures were a consequence of osteoporosis and its early onset in females. In contrast, males expressed overall more severe ageing in all vertebral divisions compared to females. Males also had a higher prevalence of vertebral facet joint osteoarthritis in cervical and thoracic divisions than females. Likewise, the incidence of facet joint osteoarthritis was more asymmetric between the left and right joints in males than in females. These findings reflect disparities of vertebral health between the two sexes in an urban setting, in which ageing and injuries of the vertebral column might be driven by different mechanisms. Age-related changes in female vertebral columns may have been more influenced by conditions of hormone deficiency such as menopause, while male vertebral columns might have been more prone to age-related changes due to heavy labor-induced physical stressors. Further studies on the differentiation of ageing mechanisms between the two sexes based on physiology, socioeconomic roles, and living conditions are warranted. The studies are necessary in understanding how multiple sociocultural and physiological factors contribute to health disparities in historic and contemporary environments.

Key words: Paleoepidemiology, degenerative joint diseases, osteoporosis, gendered labor division, health disparities

Original article



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Introduction

Health disparities refer to the differences in health between groups resulting from socioeconomic and environmental disadvantages; these disparities occur across many categories, including ethnic groups, gender, socioeconomic roles, and disability status, which result in apparent health inequalities across such divisions (Braveman 2006). The disparity in health outcomes/care between males and females has been of special interest because it is a natural biological problem in health policy making (Wamala and Lvnch 2002: Ostrowska 2012). In addition to its significance in health policy, sex/gender-based inequality also interests bioarchaeologists. These researchers are interested in examining this disparity in relation to male and female lifestyles and socioeconomic roles played during ancient times, and how this sexgender-based difference has influenced health and social equality in human society (Schepartz et al. 2017). Initial differences between males and females have been explained using the biomedical model, i.e., that postmenopausal women have higher risks because of their higher prevalence of osteoporosis (Cummings et al. 1985, 1989; Cooper et al. 1992; Cummings and Melton 2002). However, this trend has been complicated by social socioeconomic stratification during recent human evolution. For example, it has been shown that the hierarchical nature of an ancient Greek population informed gender-based inequalities which affected dietary differences and dental health (Schepartz et al. 2017). Thus, epidemiological studies on sex-based health inequalities should combine biomedical and socioeconomic models to determine factors for perpetuating these differences (and how they are linked). In this research, we examined the age-related changes of the spinal column. Our work elucidates that sex/ gender-based differences in physiological and physical stresses influence the etiology and pathophysiology of spinal degenerative diseases in a bioarcheological setting. Age-related changes of the vertebral column are generally advanced by daily work activities (i.e. heavy labor) or by chronic and habitual stress (Merbs 1996; Katzman et al. 2010; Pili et al. 2018; Aguirre et al. 2020; Yustos et al. 2021). Weposit that it is beneficial to study male-female differences in the age-related change patterns of the vertebral column in light of the male-female differences in biological and sociocultural elements.

As a result of the natural ageing process, vertebrae of the spine experience several changes, including a decrease in bone mass which may lead to osteopenia and osteoporosis, as well as an increase in degenerative joint diseases (DJD). The latter category involves problems as osteoarthritis, degenerative such disk disease, calcification of associated ligaments, and weakening of attached muscles (Benoist 2003). These changes eventually lead to a number of "painful and debilitating disorders" (Papadakis et al. 2011), including discomfort, pain in individual sections or the whole of the spine, or loss in mobility. Osteophytosis and Vertebral Compressive Fracture (VCF) are two primary outcomes of these age-related changes and pathological processes. While VCF is one of the most common symptoms of osteoporosis (Cummings et al. 1989), osteophytic formation is an adaptive bone remodeling response to daily life stressors and the progressive reduction of bone mineral density and mass, which increases load-bearing surface areas (Fraser et al. 1997). Other less studied consequences of age-related changes in the spine are arthritis of the vertebral body and facet joint osteoarthritis, caused by a combination of multiple factors (i.e. amount of use, profession-related overload, age-related changes, genetics, and injury). Symptoms of spinal osteoarthritis include pain, swelling, and loss of flexibility (Dreyfuss 1994; Manchikanti et al. 2004).

The age-related changes of vertebral bodies have been widely studied in contemporary, historical, and prehistoric populations. Different health outcomes have been linked to nutrition, lifestyles involving poor posture and incorrect ergonomics, levels of sex hormones, trauma, and labor activities (Stewart 1958; Chapman 1972; Burrell et al. 1986; Riggs et al. 1986; Miller et al. 1988; Jurmain 1990; Waldron 1991; Lovell 1994; Knusel et al. 1997; Sofaer-Derevenski 2000; Brickley 2002; Steckel and Rise 2002; Fillingim 2003; Manchikanti et al. 2004; Leveille et al. 2005; Van der Merwe et al. 2006; Rojas-Sepulveda et al. 2008; Bailey 2009; Novak and Slaus 2011; Kim et al. 2012; Shimoda et al. 2012; Zukowski et al. 2012; Missikpode et al. 2015; Hou et al. 2017; Steckel et al. 2019). Specifically, attention has been called for "activity-related osseous change" in historical populations. For example, a comparison study of populations from Wharram Percy (a medieval site) and Ensay (a 16th-19th century site) in the U.K. reveals that sex-based differences in osseous changes of the spine; these findings have been related to dissimilarities in physioanatomy and life history (i.e., females undergoing child bearing and menopause) along with gender-based divisions of labor (i.e., males undertaking heavy-duty labor) (Sofaer-Derevenski 2000). Thus, we propose that differing age-related and spinal osseous changes between sexes could be important indicators in differentiating between gender-based socioeconomic roles in ancient human societies. Knowledge of how physiological and physical sex-based differences affect the etiology and pathophysiology of skeletal degenerative diseases can assist in distinguishing these elements from other sociocultural factors that may have contributed to health disparities in bioarchaeological studies.

In this study, sex-based differences in the ageing of the spine were tested in human skeletons excavated from a Bronze Age cemetery of the Western Zhou Dynasty (1045-771 BCE), at the Dahekou site (Baidu Map: N35.749438°, E111.788635°) located in Yicheng County, Shanxi Province, China (Fig. 1). Between 2007 and 2017, approximately 2,200 tombs were excavated during thirteen phases. Funeral objects were found, including bronze vessels, pottery, jade bone tools, lacquered wood items, tin vessels, shell tools, and sacrificed dog remains (Xie et al. 2011; Guo 2015; Li et al. 2021). From the inscriptions on bronze funeral vessels, scholars realized the existence of a small city-state Ba during the Western Zhou Dynasty (1045–771 BCE), and that this cemetery was used by people of the State Ba (Xie et al. 2011). The State Ba was a small city-state, but had no record in the history scrolls for the Western Zhou Dynasty. The density and numbers of tombs found at the Dahekou cemetery indicates that it was in an urban setting with a relatively large population size, most likely the capital city of the recovered Ba State.



Fig. 1. Location of the Dahekou site.

The socioeconomic structure during the Western Zhou dynasty was a form of feudalism similar to the socio-economic mode of medieval Western Europe (Feng 2014), in which the king owned all of the land and lived in the capital city. Lords of regional states lived in towns and controlled the land, while the peasants lived outside of the towns and worked on the land. Future archaeological study of the Dahekou site would help in reconstructing the daily life of its ancient inhabitants. In total, the remains of 2201 individuals (549 in excavation areas one to six, and 1652 from excavation areas seven to thirteen) were excavated from the Dahekou site (Han 2019). The sex of 1762 individuals was identified among this group. There were 873 males and 889 females, suggesting a balanced male vs. female ratio (0.982), or no significant loss of male individuals. The ratio was a sign of a rather peaceful period for this ancient Chinese region. Our preliminary study also revealed that there were very low cranial fractures (i.e., only two males showed signs of skull fractures: one in the mandible, the other one along a nasal bone). This finding indicated a low incidence of intentional interpersonal violence (Lovell 1994; Zhang et al. 2021), and peaceful existence in Ba State society. (Appendix Table 1). Thus, the Dahekou site provided a revealing window into an ancient urban population with minimal disturbance from interpersonal conflicts for our ageing analysis. In this study, we investigated the influence of sex-based differences and health inequalities on the age-related changes of the vertebral column, including fractures and osteoarthritis. Accordingly, we showed how sex-based physioanatomy and a gender-based division of labor affected the spinal column during a relatively peaceful time of feudalism.

Materials and methods

The skeletons from excavation phases seven to thirteen were used in this study (Table 1). Based on current archeological evidence, the section (excavation areas seven to thirteen) at the Dahekou cemetery was an area used by the commoners (1652 skeletons in total), not the social elites of Ba State; they are proposed to have engaged in an urban lifestyle within the walls of a city, likely the capital of the Ba State. The spines were well preserved in 120 of the skeletons, with nearly 90% of vertebrae from the three segments combined (i.e., cervical, thoracic, and lumbar) (Appendix Table 2). Sex was determined using skeletal features from the skull (including the mastoid process, supraorbital ridge and glabella, the nuchal crest, and the mental eminence). The pelvis was also used (including the morphology of the medial margin of the inferior pubic branch, the subpubic angle, and the greater sciatic notch) following Buikstra and Ubelaker (1994). Age at death was estimated by grading the auricular surface degeneration of the pelvis (Lovejoy et al. 1985). Furthermore, age at death was categorized into three periods based on eight phases: one to two as a young adult (twenty to thirty-five years old), three to six as a middle adult (thirty-six to fifty years old), and seven to eight as an older adult (>50 years). Age was then examined using dental wear stages (Lovejoy 1985), with consideration of the phenomenon that wear is more advanced in pre-contemporary populations due to a more abrasive diet (Molnar 1972; Kaidonis 2008). At Dahekou, there were one hundred and twenty full adult skeletons (forty-six males and seventy-four females) with good preservation from phases seven to thirteen (areas excavated at different times).

Individuals with an age at death estimated at ≥20 years with a reliable determination of sex were selected from two age groups: young adults (Adult Young or AY: 20–35 years) and middle-aged adults (Adult Middle or AM: 36–50 years). The objective was to examine the frequency of degenerative diseases such as VCF and osteophytosis at both the vertebral body and joint facets. Individuals in the Old Adult (>50 years) category were not possible to study due to unfavorable postmortem vertebral preservation conditions.

Table 1. Individuals included in this study from excavation phases seven to thirteen at the Dahekou cemetery. Individuals were grouped in four sex-age groups in this investigation. The first number represented the number of individuals for investigations of vertebral compressive fracture (VCF) and vertebral body osteophytosis (VBO); the second number represented the number of individuals used for joint facet osteoarthritis investigation (JFO)

	Adult Young (20-35 Years)		Adult Middle (3	36–50 Years)	Sum		
	VCF+VBO	JFO	VCF+VBO	JFO	VCF+VBO	<u>JFO</u>	
Male	19	9	27	13	46	22	
Female	42	41	32	30	74	71	
TOTAL	61	50	59	43	120	93	

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A macroscopic visual analysis of each vertebra was carried out, and VCF was recorded with the identification of vertebrae (i.e., T10 or L5) and their type of fracture (i.e., wedge, biconcavity, or crush deformity fracture).

Next, osteophytes on the edge of the vertebral body were investigated and graded in the cervical, thoracic, and lumbar vertebrae. The samples were graded using a five-stage standard following Snodgrass (2004) and Van der Merwe et al. (2006) (Fig. 2). Briefly, Grade 0 has no signs of osteophytosis, and Grade 1 has osteophyte points or slight lipping on the vertebral body margins. Grade 2 has more lipping on

the margins (no more than 2mm projecting horizontally from the vertebral body), while Grade 3 exhibits advanced lipping projecting horizontally from the vertebral body at larger than 2mm or almost fusion of the osteophytes. Grade 4 exhibits the fusion of osteophytes on adjacent vertebrae. In order to compare the severity of osteophytosis in different vertebral divisions, the mean osteophytic value (MOV) was calculated as the mean osteophytic grade value for each vertebra (grades of the upper and lower margins) in different sex-age groups. In total, vertebrae (C1 to L5) from 120 individuals in four sex-age groups were scored (Table 1).



Fig. 2. Five stages of osteophytosis (vertebral body). All specimens were from the Dahekou site. From left to right: M9363 L1-L2 (Grades 0 and 1), M9045 T7-T8 (Both Grade 2), M9045 T9-T10 (Both Grade 3), M7215 C5-C6 (Both Grade 4). Images not scaled.

Lastly, signs of degenerative joint diseases were investigated and graded in the facet joints of cervical and thoracic vertebrae (details given below). The facet joints in the spine (zygapophysial joints) are synovial joints between the articular processes of two adjacent vertebrae. Facet joint osteoarthritis has been investigated intensively inmedicine due to its association with neck and back pain (Manchikanti et al. 2004). Facet joints render the spine flexible; in each spinal motion segment, there are two facet joints formed by neighboring vertebrae. Thus, a vertebra has two superior (left and right) and two inferior (left and right) joint facets. Facet joint osteoarthritis is attained when these joints become inflamed and painful due to the degeneration of intervertebral discs and development of vertebral body osteoarthritis (Manchikanti et al. 2004). Age-related changes in the intervertebral disc lead to narrowing of the intervertebral space (Adams and Dolan 2012), which results in pressure overload of the facet joints and destruction of joint cartilage. This situation gives rise to localized facet joint osteoarthritis (Manchikanti et al. 2004), in addition to vertebral body osteoarthritis. In this study, we established a fivestage standard (Fig. 3) scoring system for this condition which has been explained earlier.



Fig. 3. Five stages of facet joint osteoarthritis. All specimens were from the Dahekou site. From left to right: M12288 C2 superior joint facets (Grades 0), M12026 T1 inferior joint facets (Grade 1), M12125 T11 superior joint facets (Grade 2), M12026 T1 superior joint facets (Grade 3), M12125 T9-T10 fused (Grade 4). Images not scaled.

Our facet osteoarthritis investigation was limited to the cervical and thoracic segments (C1 to T12), and the lumbar segment was not included due to unfavorable postmortem preservation of the joint facets in lumbar vertebrae. The number of individuals suited for this section of the investigation was smaller and more imbalanced. There were twenty-two males and seventy-one females (reasons for the less favorable postmortem preservation of joint facets in males compared to females were unknown). Both the superior and inferior facets of the left and right sides were recorded.

Data was then analyzed using a statistical analysis program named Graph-Pad 6.0 for Windows (GraphPad Software Inc., La Jolla, CA). Mean scores of osteophytosis in a vertebra were based on the means of the upper and lower margins of the same vertebra or four joint facets of the same vertebra. Mean scores of the segments were means of mean scores of all vertebrae preserved in a segment. Group differences in prevalence/frequencies were compared between the different sex-age groups using a non-parametric Chi-square for independence in a contingency table. The significance level was set at $\alpha = 0.05$.

Results

Vertebral Compressive Fracture (VCF)

In total, forty vertebrae (1.6% in terms of prevalence per vertebra) in twenty-one individuals (17.5% in terms of prevalence per individual) carried signs of VCF (Table 2; Appendix Table 3). Two types of compressive deformity factures were observed in the Dahekou population: wedge deformity fractures (anterior height < middle height < posterior height) and crush deformity fractures (in which the whole body was compressed evenly) (Appendix Table 3; Fig. 4). Half of the VCFs (twenty out of forty) were wedge deformity fractures, and the other half were crush deformity fractures. Nevertheless, there were no biconcavity deformity fractures. Nearly half of the individuals (47.6%) had multiple fractures in separated or continuous vertebrae, mostly in the thoracolumbar spine (Appendix Table 3). The prevalence of continuous fractures was 28.6% (two out of seven individuals with VCF) in the female young adult (AY) group, 36.4% (four out of eleven individuals with VCFs) in female middle adult (AM) group, and 33.3% (one out of three individuals with VCF) in male AM group (either wedge or crush deformity varieties). No VCF was found in male AY (0%).

	Cervical	Thoracic	Lumbar	Sum
Female Adult Young	0	0.2%	4.5%	1.1%
Female Adult Middle	1.0%	1.2%	12.8%	3.6%
Male Adult Young	0	0	0	0
Male Adult Middle	0	1.0%	1.4%	0.9%
Total	0.3%	0.5%	3.7%	1.6%

Table 2. Distribution of vertebral compressive fracture in three spine segments from four sex-age groups

Among the group of forty VCFs found, most were in the lumbar segment (82.2%), followed by thoracic (11.1%) and cervical (6.7%) segments (Table 2; Fig. 4). In particular, the majority of the locations were in the lower lumbar vertebrae. Of the seventeen individuals with lumbar VCF, L5 was involved in fifteen

cases and eight of these were isolated to L5 alone (categorized as either the aforementioned wedge or crush deformity). As opposed to the lower lumbar vertebrae, a cervical segment was involved in only one individual (C5-6 from M9054, female, age at death around 40 years) (Appendix Table 3).



Fig. 4. Compressive vertebral fracture. Left: M9066, female, L1 Wedge deformity fracture; Right: M9066, female, L2 Crush deformity fracture. Scale: 10mm per segment.

The VCF prevalence per vertebra was higher in females than in males (Table 2). Throughout this sample, 10 out of 896 vertebrae (1.1%) in the female AY group. 25 out of 687 vertebrae (3.6%) in the female AM group, and 5 out of 588 vertebrae (0.9%) in the male AM group were involved in VCF. Remarkably, no vertebrae from the male AY group showed signs of VCF, indicating the impact of sex-age status (Chi-square test: X^2 = 29.0, P < 0.0001, DF = 3). The lumbar segment had the highest involvement concerning VCF prevalence per vertebral segment. The VCF prevalence was 0.3% in cervical, 0.5% in thoracic, and 3.7% in lumbar segments, demonstrating the effect of position in VCFs (Chi-square test: $X^2 = 66.9$, P<0.0001, DF =2).

Concerning VCF prevalence per individual (Table 2; Appendix Table 2), females had a higher VCF prevalence than males (Female 18 out of 74 or 24.3%) vs. Male 3/46 or 6.5%; Chi-square test: $X^2 = 6.23$, P=0.0013, DF =1). In addition, the prevalence of VCF was higher in the female AM group than in the female AY group. In males, no cases were found

in the Male AY group and the prevalence was 11.1% in the Middle Adult group $(35-45 \pm \text{ years})$; in females, VCF prevalence was at 16.7% in the Adult Young group and 34.4% in the Middle Adult group (Table 2; Appendix Table 2). In the female Adult Young group, six of the seven individuals were clustered in an age range between twenty-five and thirty vears old.

Vertebral Osteophytosis

Cervical, thoracic, and lumbar osteophytosis were observed in all sex-age groups (Tables 5-9). Males tended to have more vertebrae affected in their spine than females in two age groups (Table 3). In terms of mean osteophytic grades, there was a trend that osteophytic formation increased from young to middle adults. However, males expressed higher overall osteophytic stages in all vertebral segments than females (Table 3; Fig. 5a-b). For example, the prevalence of Grade 2 and above was 21.9% vs. 10.5% in the young adult group, while it was 34.7% vs. 19.7% in the middle adult group (Table 3).

Table 3. Summary of the grand mean of osteophyte grades (Mean ± SD) of three vertebral segments (Ce	er-
vical segment including C1) in four sex-age groups, and summary of the prevalence of high severity	of
osteophytosis (Grade 2 and above) in four sex age groups	
	_

		Cer	vical	Thoracic		Lumbar		SUM		
		Male	Female	Male	Female	Male	Female	Male	Female	Com- bined
Osteo-	Adult Young	0.63 ± 1.00	0.38 ± 0.76	0.76 ± 0.80	0.42 ± 0.55	1.13 ± 1.11	1.08 ± 1.03	0.80 ± 0.92	0.55 ± 0.79	0.63 ± 0.84
phytosis grade	Adult Middle	0.87 ± 1.27	0.56 ± 0.91	1.05 ± 1.13	0.51 ± 0.68	1.94 ± 1.08	1.53 ± 1.13	1.20 ± 1.23	0.74 ± 0.95	0.95 ± 1.11
Preva- lence of	Adult Young	26 (22.2%)	25 (10.2%)	35 (16.8%)	14 (3.1%)	28 (34.1%)	55 (27.8%)	89 (21.9%)	94 (10.5%)	183 (14.0%)
Grade 2 and above	Adult Middle	40 (24.2%)	31 (16.2%)	77 (26.6%)	33 (9.51%)	87 (64.9%)	71 (48.0%)	204 (34.7%)	135 (19.7%)	339 (26.6%)



Fig. 5. Osteophyte grades for cervical, thoracic and lumbar segments in male and female adult young groups (a) and male and female adult middle groups (b). There was a trend that the osteophytic formation increased from young to middle adults, and that the males expressed higher overall osteophytic stages in three vertebral segments than females. Abbreviations: MAY – Male Adult Young; FAY – Female Adult Young, MAM – Male Adult Middle; FAM – Female Adult Middle.

In addition, males and females had similarly positioned patterns of vertebral osteophytosis in terms of Grade 2 and above expressions. For instance, we detected thoracic < cervical < lumbar, except in male AM cervical < thoracic, while the percentage of Grade 2 and above was higher in males than in females (Tables 4–5). In different sexage groups, 21.9% (89 out of 407) in the male AY group exhibited Grade 2 and above, whereas 10.5% (94 out of 896) in the female AY group exhibited Grade 2 and above. Furthermore, 34.7% (204 out of 588) of vertebrae in the male AM group exhibited Grade 2 and above, while 19.6% (135 out of 1275) in the female AM group exhibited Grade 2 and above (combined: AY – 14.0% or 183 out of 1303; AM - 26.6% or 339 out of 1275).

Osteophyte Grade	Cervical		The	oracic	Lumbar	
(Addit Ioulig)	Male	Female	Male	Female	Male	Female
0	78 (66.7%)	184 (75.4%)	90 (43.3%)	279 (61.5%)	27 (32.9%)	67 (33.8%)
1	13 (11.1%)	35 (14.3%)	83 (39.9%)	161 (35.5%)	27 (32.9%)	76 (38.4%)
2	17 (14.5%)	17 (7.0%)	29 (13.9%)	14 (3.1%)	18 (22.0%)	29 (14.6%)
3	9 (7.7%)	8 (3.3%)	6 (2.9%)	0 (0.0%)	10 (12.2%)	24 (12.1%)
4	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	0 (0.0%)	2 (1.0%)
Ν	117	244	208	454	82	198
Mean	0.63	0.38	0.76	0.42	1.13	1.08
SD	1.00	0.76	0.80	0.55	1.11	1.03

Table 4. Osteophyte grades of three vertebral segments in Adult Young Groups (Age 20-35 years)

Table 5. Osteophyte grades of three vertebral segments in Adult Middle Groups (Age 36–50 years). In each sex-age group, the number of cases with percentage in parenthesis represented the numbers of vertebrae

Osteophyte Grade	Cervical		Tho	racic	Lumbar		
	Male	Female	Male	Female	Male	Female	
0	98 (59.4%)	128 (66.7%)	115 (39.8%)	205 (59.1%)	14 (10.4%)	33 (22.3%)	
1	27 (16.6%)	33 (17.2%)	97 (33.6%)	109 (31.4%)	33 (24.6%)	44 (29.7%)	
2	12 (7.3%)	19 (9.9%)	38 (13.1%)	31 (8.9%)	41 (30.6%)	33 (22.3%)	
3	19 (11.5%)	12 (6.3%)	27 (9.3%)	2 (0.6%)	39 (29.1%)	36 (24.3%)	
4	9 (5.5%)	0 (0.0%)	12 (4.2%)	0 (0.0%)	7 (5.2%)	2 (1.4%)	
Ν	165	192	289	347	134	148	
Mean	0.87	0.56	1.05	0.51	1.94	1.53	
SD	1.27	0.91	1.13	0.68	1.08	1.13	

Overall, the lumbar spine had the highest prevalence of osteophytes that were at Grade 2 and above in the Adult Middle groups, yielding 64.9% in males and 48.0% in females (Appendix Table 4). Specifically, the male Adult Young group had a high prevalence of osteophytosis (Grade 2 or above) in C5-7 (33.3–44.4%), T6-12 (16.7–31.6%), and L1-5 (17.6–43.8%), with L3 and L5 displaying the highest prevalence (43.8%). The male Adult Middle group had high values from

C3-7 (18.2–41.7%), with 41.7% in C6 to higher values from T10 (44.0%) and L4 (77.8%). In the female Adult Young group, high values were from C5-6 (22.0–25.5%) and L2-5 (22.5 to 42.5%), while they were generally low in the thoracic segment. However, the female Adult Middle group demonstrated high values from C5-6 (40.7%), while they were 33.3–62.1% from L1-5 and 20.0% from T8-9 (Appendix Table 4).

In terms of the mean osteophytic value (MOV), different sex-age groups had different patterns, yet all lumbar elements, lower thoracic elements, C5, and C6 generally exhibited high stages of osteopathic formation (Appendix Table 5; Fig. 6). In the male Adult Young group, the five top values were as followed (in the same decreasing order hereafter): L5, L4, L3, T9, and C5 (range 1.38–1.06). Moreover, the female Adult Young group had top five figures from L4, L5, L3, L2, and C6 (range 1.45–1.06). Additionally, the top five calculations in the male Adult Middle category were from L4, L3, L2, L5, and L1 (range 2.19–1.59), followed by T10, C6, T11, T9, and C5 (range 1.52–1.26). By comparison, the top five values in the female Adult Middle group were in L5, L4, L3, L2, and C5 (range 1.83–1.26), followed by L1, C6, T7, T8, and T10 (1.20-0.73).



Fig. 6. The mean osteophytic value (MOV) (mean osteophyte grades of individual vertebra) in the four sexage groups for the Dahekou skeletons. Abbreviations: MAY – Male Adult Young; FAY – Female Adult Young, MAM – Male Adult Middle; FAM – Female Adult Middle.

Facet Joint Osteoarthritis

Facet joint osteoarthritis was investigated in 6082 joint facets from 1520 vertebrae in twenty-two adult males (nine in the young group and thirteen in the middle group), as well as in seventy-one adult females (forty-one in the young group and thirty in the middle group). Overall, the prevalence of joint facet arthritis was relatively low: 235 of 6082 joint facets (3.9%) exhibited osteoarthritis at Grade 2 or above (Grade 2: 140 or 2.3%, Grade 3: 77 or 1.3%, and Grade 4: 18 or 0.3%). Generally, the percentages of Grade 2 and above were higher in males than in age-matched females (Table 6). Accordingly, the prevalence and severity of facet joint osteoarthritis was low in the female groups. However, signs of joint facet osteoarthritis were more prevalent in the male groups. In fact, the male Adult Young group had

relatively higher thoracic joint facet arthritis, while the male Adult Middle group had higher cervical joint facet arthritis (Appendix Table 6). The side difference in affected bone was unremarkable in females. By comparison, males expressed a general trend in which the left side had higher severity of facet joint osteoarthritis than the right side in both young and middle adult groups; this pattern was especially high in C3 and C7 (left > right) within the male Middle Adult groups (Fig. 7; Appendix Table 6).

Table 6. Grades of joint facet arthritis at left and right sides in four sex-age groups. In each sex-age-side group, the number of cases with percentage in parenthesis represented numbers of joint facets. Cervical and thoracic vertebrae combined

	Male Ad (Individ	ult Young ual N=9)	Female Adult Young (Individual N=41)		Male Adı (Individu	ult Middle al N=13)	Female Adult Middle (Individual N=30)	
	Left	<u>Right</u>	Left	<u>Right</u>	<u>Left</u>	<u>Right</u>	<u>Left</u>	<u>Right</u>
0	230 (79.9%)	225 (78.1%)	1220 (91.7%)	1220 (91.7%)	382 (82.5%)	374 (80.8%)	826 (86.0%)	824 (85.8%)
1	30 (10.4%)	39 (13.5%)	101 (7.6%)	101 (7.6%)	31 (6.7%)	48 (10.4%)	92 (9.6%)	104 (10.8%)
2	17 (5.9%)	14 (4.9%)	5 (0.4%)	4 (0.3%)	27 (5.8%)	24 (5.2%)	25 (2.6%)	24 (2.51%)
3	7 (2.4%)	8 (2.8%)	4 (0.3%)	5 (0.4%)	17 (3.7%)	11 (2.4%)	17 (1.8%)	8 (0.8%)
4	4 (1.4%)	2 (0.7%)	0 (0.0%)	0 (0.0%)	6 (1.3%)	6 (1.3%)	0 (0.0%)	0 (0.0%)
Ν	288	288	1330	1330	463	463	960	960
Mean	0.35	0.34	0.09	0.09	0.35	0.33	0.20	0.18
SD	0.81	0.76	0.33	0.34	0.85	0.79	0.57	0.5



Fig. 7. Side difference of joint facet arthritis in Dahekou skeletons. Abbreviations: MAY – Male Adult Young, FAY – Female Adult Young, MAM – Male Adult Middle, FAM – Female Adult Middle, L – Left, R – Right.

Discussion

There was discordance in the paleoepidemiology of vertebral compressive fractures (VCF) and age-related degenerative joint diseases (DJD) at the Bronze Age Dahekou site. While females had an overall higher VCF prevalence and earlier manifestation of VCF than males, males had a higher severity of DJD than females. These sex-based differences in the patterns of VCF and DJD may indicate different mechanisms for attaining spinal changes in males and females, both naturally and socioeconomically.

Vertebral Fractures in Urban and Agricultural Populations

In contemporary humans, the prevalence of vertebral fractures increases with age for both males and females; postmenopausal women have higher risks because of their higher prevalence of osteoporosis (Cummings et al. 1985, 1989; Cooper et al. 1992; Cummings and Melton 2002). At Dahekou, females had an overall higher VCF prevalence that was not at variance with the modern population, thus suggesting the effects of osteoporosis. Females at Dahekou also demonstrated an earlier manifestation of VCF than males, which may indicate an early onset of age-related changes and osteoporosis within females rather than males. In our opinion, a higher prevalence in females is most likely related to sex-based differences in pathophysiology, a circumstance in which females could have higher hormone deficiency-related osteoporosis than males. Moreover, osteoporotic fractures are uncommon in physically active populations (Jónsson et al. 1992; Sanders et al. 2002; Pisani et al. 2016). In this regard, the low VCF in males at Dahekou may suggest higher physical

stress in males among various biological and physical factors. Vice versa, these trends suggest that women at Dahekou were generally involved in low physical stress during their daily lives, a phenomenon of gender-biased divisions of labor that was corroborated by the severity of DJDs (high in males and low in females).

Physioanatomy and life history, along with gendered divisions in socioeconomic statuses and roles (hence different labor-related stressors) might be responsible for the different patterns found at Dahekou. Females from this location generally had high grades of osteophytosis in the lumbar and cervical segments and very low grades in thoracic segments. On the contrary, although males had high lumbar osteophytotic development to a lesser degree, they did exhibit relatively higher osteophytosis at the thoracic area compared to females. This pattern of sex-based differences at the Dahekou site might have been the consequence of a gender-based division of labor: males and females both engaged in physically demanding jobs, yet the types of jobs were different. Hence, what type of labor division did the Dahekou population engage in as a result of its socioeconomic mode?

The age of the Ba State was in the era of the feudalism system. During the economic mode of the feudalism system, civilian men practiced agriculture in rural areas or specialized in craft and service industries within urban areas, including cities and towns (Barford 2005; Agnew et al. 2015). Under this structure, the overall injury patterns could be interpreted in light of feudal socioeconomic relations and class conflict. As mentioned earlier, limited evidence (including the balanced male vs. female ratio and lack of signs for skull injury (Appendix Table 1) indicated a lack of intense interpersonal conflicts in a relatively peaceful period.

Risks of skeletal injuries include occupational risks (i.e., from farm labor in agriculture), environmental health risks (i.e., rugged terrain), and interpersonal conflicts (i.e., war, battles, structural violence, or fights and assaults) (Agnew et al. 2015). There are remarkable differences in VCF prevalence among different populations throughout the Eurasian continent (Table 7), due to differences in settlement patterns, subsistence methods, social stability, age-related factors of change, mortality patterns, and genetic factors. Differences in the frequency of bone fractures among different populations are related to differences in social status (Geber 2015), environment/terrain (Kilgore et al. 1997), occupation (Grauer and Roberts 1996), or economic setting, i.e., rural vs. urban (Agnew et al. 2015).

Table 7. Vertebral Compressive Fracture in various populations/groups (Methods of investigation may vary among sites)

Den lation/City	Denie 1/A	Economic	VCF Pr	evalence	C	
Population/Site	Period/Age	mode	Male	Female	Source	
Dahekou, Yicheng, Shangxi, China	Western Zhou (1045-771 BCE)	Urban	6.5% (3 of 46) Middle aged man	24.3% (18 of 74) Young to Middle aged women	This study	
Xitun, Yanqin, Beijing, China	Han (202BCE-220CE)	Millet Agriculture	0% (0 of 126)	0.6% (1 of 167)	Zhou, 2014	
Gouwan, Xichuan, Henan, China	Late Neolithic (5000-3000BCE)	Rice and Millet Agriculture	2.6% (1 of 39)	0% (0 of 26)	Wang, 2015	
Matengkong, Xi'an, Shaan Xi, China	Eastern Zhou (770-256BCE)	Agriculture	12.5% (3 of 24)	7.7% (2 of 26)	Wang, 2019	
Xuecun, Henan, China	Han (202BCE-220CE)	Agriculture	2.6% (2 of 76)	1.6% (1 of 61)	Sun, 2013	
Da'an, Jilin, China	Warring States to West Han (475BCE-8CE)	Hunting- Fishing	2.8% (1 of 36)	0% (0 of 67)	Xiao, 2014	
Lamadong, Beipiao, Liaoning, China	Jin (31-420CE)	Millet Agriculture	0.5% (1 of 214)	0% (0 of 190)	Chen, 2009	
Changle, Zhongwei, Ningxia, China	Han (202BCE-220CE)	Agriculture	5.6% (2 of 36)	10.3% (3 of 29)	Zhang, 2018	
Taojiazai,Xining, Qinghai, China	Han-Jin (202BCE-420CE)	Agriculture	0.6% (1 of 175)	0% (0 of 167)	Zhang, 2008	
Jucun, Jiangxian, Shanxi, China	Western Zhou (1046-771BCE)	Agriculture	0.8% (2 of 258)	0.7% (2 of 296)	Zhao, 2018	
Gaohong, Liulin, Shanxi, China	Late Shang (1250-1046BCE)	Agriculture	27.3% (3 of 11)	0% (0 of 5)	Liang, 2017	
Xiaonanzhuang, Taiyuan, Shanxi, China	Warring States (475-221BCE)	Agriculture	0% (0 of 7)	10.0% (1 of 10)	Hou, 2017	
Niedian, Jinzhong, Shanxi, China	Zhou (1046-256BCE)	Agriculture	9.1% – 5 (all male & fema	female) of 55 ale combined	Hou, 2017	

Dopulation/Site	Deriod/Ago	Economic	VCF Pre	evalence	Source	
Population/Site	Penou/Age	mode	Male	Female	Source	
North Liujiazhuang, Anyang, Henan, China	Late Shang (1250-1046BCE)	Agriculture	6.1% – 3 (all male & fem	female) of 49 ale combine	Yuan, 2010	
Chuanzhang, Zungeer, Inner Mongolia, China	Warring States to Han (475BCE-8CE)	Agriculture	0% (0 of 165)	0.8% (1 of 132)	Ana'er, 2018	
Liangwangcheng, Peizhou, Jiangsu, China	Late Neolithic (2800-2500BCE)	Rice Agriculture	1.2% – 1 (s known) of female c	sex-age un- 86 male & ombined	Zhu et al., 2013	
Chongpingyuan, Yichuan, Shaanxi, China	Early Spring-Autumn Period (770-650BCE)	Agriculture	14.3% (1 of 7)	9.0% (1 of 11)	Chen et al., 2018	
Giecz, Poland	950-1250 CE	Rural	50.0%, Adults ≥ 18 years	29.8%, ≥ 18 years	Agnew et al., 2015	
Poznań-Sródka, Poland	950-1250 CE	Urban	0%, Adult ≥ 18 years	3.7%, ≥ 18 years	Agnew et al., 2015	
Modern American Women				5-10% mid- dle aged women	Ensrud and Schousboe, 2011	

Table 7 (cont.)

Agriculture has been one of the most dangerous occupations since its origin due to high physical demands (McCurdy and Carroll 2000), and is still dangerous in modern times with the involvement of machinery (Myers 2001; Chari et al. 2017). Past bioarchaeological studies support the conclusion that rural areas with agricultural practices such as farming and animal husbandry had higher frequencies of trauma than urban areas (Judd and Roberts 1999; Djurić et al. 2006; Agnew et al. 2015). For example, Agnew et al.'s (2015) study of medieval rural and urban settings shows a distinct difference in the prevalence of trunk fractures (including VCF), with VCF prevalence being higher in rural areas than urban areas. Overall, agricultural populations engaged in a laborious lifestyle, reflected in a variety of injuries related to repetitive and highrisk activities. Although urban populations engaged in craft specialization

and participated in repetitive activities, their lifestyle resulted in a lesser risk of fracture (Agnew et al. 2015). The lack of skull fractures at Dahekou also suggests a low risk of injury from intentional interpersonal conflicts, while the relatively high incidence of limb and rib bone injuries in males might indicate a lifestyle with higher physical stress. At the rural Medieval Poland Giecz site, the prevalence of VCF was 50%, whereas the prevalence of VCF at the urban Medieval Poland Poznań-Sródka site was close to 0% (Agnew et al. 2015). The prevalence of VCF was 6.5% in males at Dahekou, corroborating the presumption of an urban lifestyle at Dahekou. The relatively low prevalence of VCF suggests that the economic activities males at Dahekou participated in were less stressful than a typical agricultural lifestyle in terms of the stress on the spine, though nutrition and genetic adaptation (hence better bone quality) might be among the biological and environmental factors in addition to sample bias in these studies. Nonetheless, the low prevalence of vertebral fractures corroborates with the presumption of an urban lifestyle for the economic mode of the Dahekou population.

Spinal Osteophytosis and Gender-Based Labor Divisions

At Dahekou, the high limb and rib bone fractures might also suggest that males from the site had higher environmentally specific physical stressors than females (Appendix Table 1), which might indicate a gender-biased division of labor within in an urban lifestyle. On the other hand, it is proposed that females would be more involved in childcare and household chores, as well as the business and service industries (Agnew et al. 2015). However, females from the Dahekou site would have engaged in some physically demanding tasks, such as water transportation, child-carrying, food processing, and needlework; heavy loading in these scenarios would be on the cervical (head-loading) and lumbar (compressive and bending forces) vertebral segments. It is worth noting that there would have been an efficient water supply system during the Zhou Dynasty, in which joined clay pipes equipped with cleaning pools and valve trenches were installed underground to transfer water into the city (An 1992; Du and Chen 2007). In the city of the Ba State, fresh water from water cisterns might have been transferred to households using pots. Women might have used tumplines to carry water home as in other ancient human societies (Bridges 1994), or for other items such as children, materials and products (Moromizato et al. 2007). In addition, there were certainly more factors complicating this gender-divided labor model, yet it formed a simplified economic model for further studies involving other factors. Researchers would need to consider the life history of females (pregnancy, postpartum, weaning, and menopause) and other environmental and socioeconomic parameters.

Osteophytic grades were generally high in the lumbar area for both males and females, as exhibited by the grades on L4 and L5 (Table 8). Akin to VCF, these vertebrae contribute most to the ergonomic support for upright positions and are more prone to compromise. At Dahekou, the high severity of osteoarthritis in males at both the vertebral body and facet joints suggest an arduous (though less dangerous) urban life than rural life.

Joint facet osteoarthritis in the spine is caused by a combination of multiple factors including usage, overload, age-related changes, injury, and possible genetic predisposition. Manifestations of the condition involves pain, swelling, and loss of flexibility (Butler et al. 1990; Dreyfuss et al. 1994; Dreyer and Dreyfuss 1996; Manchikanti et al. 2004; Cavanaugh et al. 2006; Kalichman and Hunter 2007; Gellhorn et al. 2013; Kim et al. 2015). The overall severity of joint facet osteoarthritis at Dahekou was low, yet males had higher development of the condition than females, indicating multiple factors that included overall body mass, higher physical stress, genetic predisposition, or sample bias. Males also exhibited asymmetry in facet joint osteoarthritis [similar to a finding by Bridges (1994) in prehistoric Native Americans], suggesting relatively more lateral bending activities than anterior or posterior bending involved in urban professions (including

Site	Date	Economic Mode	Vertebrae most severely affected by osteophytosis	References
Dahekou, China	1,045 – 771 BCE	Urban	C5-6, T7-L5 in males	This study
Joseon People, South Korea	1,500 – 1,900 CE	Agriculture	C5, T9-10, L4-5	Kim et al., 2012
Raymond Dart Collection & University of Pretoria Collec- tion, South African	Contemporary skeletal collections derived from cadavers	Mixed con- temporary lifestyles	C5, L4-5	Van der Merwe et al., 2006
Ca-Ala-329, California, U.S.A.	1,200 –1,600 CE		L4-5	Jurmain, 1990
Perry Site Lu 25, Alabama, U.S.A.	4,000 – 1,000 BCE	Hunter- gatherers	L3-4, C5-6, T7-9	Rojas-Sepulveda et al., 2008
Koprivno, Croatia	1,600 – 1,900 CE	Rural Agriculture/ Pastoral	C6, T4, L5	Novak & Slaus, 2011
Sisak, Croatio	1,600 – 1,900 CE	Urban	C5, T7, L5	Novak & Slaus,2011
Spitalfields, England	1,800 – 1,900 CE	Urban	T8-9	Waldron, 1991
El Mirador cave, Spain	4,880 – 4,390 BP	Agriculture and animal husbandry	L>T>C	Yustos et al., 2021

Table 8. Comparison between Dahekou and other groups in vertebrae mostly affected by osteophytosis

metallurgy, pottery making, glass working, stone cutting, and brick making), in addition to right-favored handedness. Female patterns of joint facet osteoarthritis were more symmetrical, suggesting more axially oriented movements or a greater relation to the flexion and extension of urban female activities, such as head-loading (i.e., water transportation), childcare, trade, and work in the service industry. It is worth noting that the status of women during the Bronze Age may have been different between China and Europe. For example, archaeological working in southeastern Spain revealed that women could have been correlated to the ruling class in the Bronze Age El Argar society (about 4,000 years ago) (Lull et al. 2021), which would denote a totally different patterns of labor division.

Inequality of Spinal Health at Dahekou

Ultimately, our research demonstrates that there were VCF-related, female-biased health inequalities with a high prevalence of osteoporotic fractures in females at the Dahekou site (an urban setting with a feudal economy). This socioeconomic context was essential in identifying a sex-based health issue within this population. We did uncover a male-biased health inequality in terms of vertebral osteoarthritis. Males experienced more severe cases of the condition, indicating more male-biased and physically demanding socioeconomic roles. As mentioned above, bone loss-induced fractures were uncommon in physically active populations (Jónsson et al. 1992; Sanders et al. 2002; Pisani et al. 2016). Thus, the high osteoarthritis coupled with the low VCF in males at Dahekou (and high prevalence of rib fractures) suggests higher physical stress in males as well. Vice versa, the low osteoarthritis coupled with high VCF in females would suggest that women at Dahekou were generally involved in low labor-intensive activities.

However, even though stress on the trunk from daily activities was higher in males than females, females not only had more VCF but also early onset of VCF. Although we proposed that the Dahekou population was an urban setting based on the overall low male injuries and other indicators, females at Dahekou had a VCF rate at 24.3%, which was similar to the prevalence rate of 29.8% of females in the rural setting of the Giecz site in Medieval Poland. Modern-time vertebral fractures are typically the result of high-energy trauma, such as a fall from height, autmobile accident, sports accident, or gunshot wounds. Nevertheless, why did Dahekou females have such a high frequency of VCF? Could this high rate of vertebral fractures be the outcome from a high incidence of trauma, such as from accidental falls or violence against women? Would this pattern indicate an unrecognized high risk in a localized urban setting at Dahekou, overall poor bone quality in females at Dahekou, or just a sample bias? There were no signs of intensive interpersonal conflict at Dahekou and no evidence of systematic violence against women, as depicted in the high incidence of head injuries and rib fractures in females from the Heartland of the Wari Empire, Peru (Tung 2012); thus, this alternative explanation is less likely. Previous stable isotopic analysis of human bones from the Zhou Dynasty have demonstrated dietary differences favoring males (i.e., Dong et al. 2017; Barbera et al. 2020; Miller et al. 2020),

while findings indicating egalitarianism between males and females in access to food resourses have been recovered from some archaeological sites of the Zhou Dynasty as well (Lan 2017; Ling 2010; Ling et al. 2017; Pechenkina 2018). These data indicate the possibility of health inequalities that were linked to a male-biased inequality of food access in ancient China. In our opinion, the possible maior causes of VCF in the females of Dahekou were due to hormone deficiency, poor nutrition, and/or lack of physical activities compared to their age-matched male counterparts. The latter conclusion was in accordance with an urban lifestyle, while the former might indicate that females had lower socioeconomic statuses compared to males in ancient societies in China (Holmgren 1981), as has been seen in other ancient complex societies globally (Schepartz et al. 2017) and therefore, had lower diet standards and poorer nutrition than males (i.e., Dong et al. 2017; Barbera et al. 2020; Miller et al. 2020; but see Lan 2017; Ling 2010; Ling et al. 2017; Pechenkina 2018). High prevalence of low back pain is also found in pregnant, postpartum, and osteoporotic females (Katonis et al. 2011; Krishnakumar et al. 2016; Wang et al. 2016). In this regard, females at Dahekou had an overall lower spinal health status.

As earlier mentioned, the Dahekou population provides a useful urban setting: a relatively peaceful and feudalistic society whose production mode fostered various age and work related skeletal pathologies. However, a full body investigation is thus warranted, including study of the skull, trunk, and extremities (along with arthritis of the limb joints).

Nonetheless, the findings of this study demonstrate the disparities of vertebral health between the two sexes. While age-related changes of female vertebral column may be more susceptible to hormone deficiency-related pathophysiologies such as osteoporosis, age-related changes of the male vertebral column may be more prone to heavy, labor induced physical stress. Accordingly, the male-female health inequality found in our study is probably the outcome of combined factors. These factors include sex-based differences in physioanatomy (an image analysis of bone qualities including bone mass, distribution and biomechanical strength is warranted), sex-based stress levels (differences between non-specific stress indicators on the skeletal remains of males and females), gendered divisions of socioeconomic roles in an urban setting, nutrition (an isotopic analysis for diet reconstruction is warranted), and social statuses (a mortuary analysis of grave goods and grave values is warranted). Coupled with imaging, isotropic, and mortuary analyses, knowledge of how sex-based differences in pathophysiology and different levels of physical stress (due to gendered divisions of labor) influence the etiology and pathophysiology of skeletal degenerative diseases can reveal the impact of anatophysiological and sociocultural factors on health disparities in bioarcheological studies.

Limitations of This Study and Future Research Directions

In this study, the model involving sexbased differences in anatophysiology and gender-based divisions of labor between males and females was simplified. Additional factors such as overall skeletal health/pathology status, bone quality analysis (total bone mass, patterns of bone distribution, and bone biomechanical strength) should be considered. A dietary analysis along with grave goods and burial values should be included as well. For example, it is not possible at the current stage to confirm poor nutrition and lack of physical activity in females at the Dahekou site. The age group division was arbitrary and used as a convenient method to generate sex-age groups comparable in males and females. However, the life history of females (i.e., pregnancy, breastfeeding, a weaning, fertility, and menopause) should be taken into consideration in the future.

The excavation at Dahekou continues. An expanded study to have all skeletons studied, especially those from elite classes (with larger grave chambers and richer grave goods), would be helpful in studying the impact of social stratification (Zhou et al. 2021). For example, an investigation of females of different social classes during the Bronze Age using the life course approach following Agarwal (2016) would be suitable. Moreover, radiological analysis of vertebrae would also assist in better characterizing bone changes in aging spines. Furthermore, we suggest that spine pathology would be better studied within the context of the whole skeleton.

There has been limited information on facet joint osteoarthritis occuring after the degeneration of the intervertebral disc (Butler et al. 1990). This suggests that facet joint diseases are secondary to mechanical changes in the overall loading patterns of the spine. However, due to differences in sample sizes, a paired study of osteophytosis in vertebral body and joint facets was not carried out in the current investigation. This process would be possible in the future when more skeletal remains at Dahekou are available.

The term degenerative joint disease (DJD) was used in this study, though there

are arguments that they are age-related adaptations (Anderson and Loeser 2011; Yustos et al. 2021). The scoring grades were still rough; for example, the readings for joint facet osteoarthritis, relied more on the development of osteophytes and mean scores of osteophytosis. For the joint facet investigation, the number of individuals suited for this section of our study was imbalanced (71 females vs. 22 males). However, because our study was the first systematic investigation of facet joint osteoarthritis in skeletons from Northern China, the results were reported to prompt more studies in this direction. For the data analysis, mean values of osteophytosis were averages of two or four readings in a vertebra, which might affect the severity of osteoarthritis. All of these results and analyses need to be refined in the future. The reported results focused on trends, with statistical significance in differences, which was due to the small or biased sample sizes and relatively high Coefficient of Variations (for example, the scores of osteophytosis in vertebral body was 1.22 for all specimens combined), which lowered confidence in the interpretation of our results.

In addition, three vertebral segments in this study (cervical, thoracic, and lumbar) were investigated to assess the development of osteophytosis and prevalence of VCF. From the one-hundred and twenty individuals in the Dahekou population included in this study, there were 2578 total vertebrae (Appendix Table 1). On average, 21.5 vertebrae out of 24 (89.6%) in these three spinal segments combined were in good condition, and therefore, included in this study. Thus, there is a need tobe cautious of sampling bias and exaggeration of difference compared to other sites with less favorable preservation status (Tables 7-8).

Conclusion

Vertebral compressive fractures, osteophytosis of the vertebral body, and joint facet arthritis were investigated in skeletal remains from the Bronze Age Dahekou site in China. Patterns of VCF and DJD in different sex-age groups indicated a sex-based difference in paleoepidemiology, mostly likely due to differences in pathophysiological and socioeconomic roles during a peaceful and feudalistic urban economy. Overall, females had a higher prevalence of VCF, with the majority of cases found in females between twenty-five and thirty years old. This result suggests increased cases of osteoporosis in females, a physiological disease that affected more females. The prevalence of osteophytosis of the vertebral body was higher in the more senior group for both males and females, signifying natural development with skeletal use along with age-related changes. Despite the knowledge that age was a factor, there was also a sex-based difference, as the overall severity of osteophytosis was higher in males than in females. This finding might demonstrate the consequences of gender-based labor divisions or sex-based genetic dispositions. Likewise, males had more severe joint facet arthritis than females, indicating higher physical stress and lower spinal health status in males. These patterns of DJD in different sex-age groups point towards a sex-based difference in paleoepidemiology, suggesting the combined effect of gendered differences in life history and socioeconomic roles. Hence, these findings signify a sex-based health disparity. Comparable to other groups with similar socio-economic structures, the Dahekou population lived in a feudalistic urban area during a peaceful time.

A comprehensive study is warranted to ascertain whether city-state Ba exemplifies ancient Chinese life during the Bronze age. Knowledge of how sex-based differences in pathophysiology, as well as different levels of physical stress (due to gendered divisions of labor) influence the etiology and pathophysiology of skeletal degenerative diseases is beneficial. This view will help distinguish these elements from other sociocultural factors contributing to health disparities in historic and contemporary environments.

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Conflict of interest

None.

Contributions from individual authors

T.H.: Data collection and analyses. W.Z., H.Z.: Data collection. Y.X.: Specimen accumulation and curation. X.Z.: Data analysis and statistics; Q.Z.: Project conception and data analysis. Q.W.: Project conception, data collection, data analysis, writing manuscript.

Data availability statement

Research data and images will be available in the public domain after the completion and publication of the study's findings. Entities include Jilin University and Texas A&M University.

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Individual	Sex	Age	Trauma/Fractures
M9161	F	30-35	Right fibular
M12199	М	25-30	6 left 2 right ribs
M12170	Μ	30-35	Nasal bone
M9045	Μ	35±	1 right rib
M9236	М	35±	Left ulnar distal
M12084	М	35±	2 left ribs
M12115	М	35±	Left ulnar, right clavicle, 2 ribs
M9351	М	36-40	3 (? Left) ribs
M10070	М	36-40	Right scapular, 1 left rib
M11201	М	36-40	1 left rib
M12004	М	36-40	Right humerus, left 3rd metacarpal
M12100	М	36-40	Left tibia two fractures, left radius distal
M12127	Μ	36-40	Right ulnar, 1 left rib
M11203	М	$40\pm$	2 right ribs
M12003	М	$40\pm$	Mandible right condyle, right & left ribs
M12118	М	40±	2 left ribs
M12076	М	40-45	2 right ribs

Appendix Table 1. Signs of trauma or fractures at the Dahekou site (Phases 7-13)

Appendix Table 2. Sample size of vertebrae in spine segments in four sex-age groups. The first number is the total vertebrae from cervical (C1-7), thoracic (T1-12), and lumbar (L1-5) segments well preserved enough to be examined in this study; the second number in the parenthesis is the average number of vertebrae preserved well enough to be included in this study

	Cer	vical	Tho	racic	Lur	nbar	Sum		
	Male	Female	Male	Female	Male	Female	Male	Female	Combined
Adult	117	244	208	454	82	198	407	896	1303
Young	(6.2)	(5.8)	(10.9)	(10.8)	(4.3)	(4.7)	(21.4)	(21.3)	(21.4)
Adult	165	192	289	347	134	148	588	687	1275
Middle	(6.1)	(6.0)	(10.7)	(10.8)	(5.0)	(4.6)	(21.8)	(21.5)	(21.6)
Total	282	436	497	801	216	346	995	1583	2578
	(6.1)	(5.9)	(10.8)	(10.8)	(4.7)	(4.7)	(21.6)	(21.4)	(21.5)
Com- bined	718	(6.0)	1298	(10.8)	562	(4.7)		2578 (21.	5)

	Individual	Sex	Age (Years)	Location	VCF Type	
Female Adult Young	M10081	F	25±	L4-5	Crush	
	M12108	F	25-30	L5	Wedge	
	M7013	F	25-30	T4	Wedge	
	M8010	F	25-30	L3-4	Wedge	
	M8265	F	25-30	L5	Crush	
	M9056	F	25-30	L5	Crush	
	M9161	F	30-35	L1, L5	Crush	
Female Adult Middle	M12128	F	35-40	L5	Crush	
	M9234	F	35-40	L5	Crush	
	M9374	F	35-40	Τ8	Crush	
	M9052	F	$40\pm$	L4-5	Wedge	
	M9054	F	$40\pm$	C5-6	Crush	
	M9057	F	$40\pm$	L5	Wedge	
	M7103	F	40-45	L5	Wedge	
	M12057	F	45-50	L4-5 (W); L2-3 (beginning of CR)	Wedge & Crush	
	M9066	F	45-50	T8, L2-3 (CR); T12, L1, L5 (W)	Crush & Wedge	
	M9163	F	45-50	T7, L1(W); L5 (CR)	Wedge & Crush	
	M9380	F	45-50	L1 (CR); L2, L5 (W)	Crush & Wedge	
Male Adult Middle	M12127	М	35-40	L1	Crush	
	M12143	М	35-40	T10-12	Wedge	
	M12288	М	45±	L5	Crush	

Appendix Table 3. Individuals exhibited Vertebral Compressive Fracture (VCF) in the Dahekou population. Abbreviations: W – Wedge deformity fracture; CR – Crush deformity fracture

	Adult Young							Adult Middle					
		Male			Female			Male			Female		
	Ν	≥G2	%	Ν	≥G2	%	Ν	≥G2	%	Ν	≥G2	%	
C1	14	1	7.1	35	0	0	24	1	4.2	28	0	0	
C2	17	2	11.8	33	0	0	24	3	12.5	31	0	0	
C3	16	1	6.3	31	1	3.2	23	6	26.1	27	1	3.7	
C4	17	2	11.8	34	4	11.8	22	4	18.2	24	5	20.8	
C5	18	8	44.4	36	9	25.0	23	8	34.8	27	11	40.7	
C6	18	6	33.3	36	8	22.2	24	10	41.7	27	11	40.7	
C7	17	6	35.3	39	3	7.7	25	8	32.0	28	3	10.7	
T1	18	1	5.6	39	0	0	25	5	20.0	30	0	0	
T2	18	1	5.6	38	0	0	25	3	12.0	29	0	0	
Т3	17	2	11.8	39	0	0	24	5	20.8	28	0	0	
T4	15	0	0	38	2	5.3	24	6	25.0	27	0	0	
Т5	17	1	5.9	39	1	2.6	23	5	21.7	27	1	3.7	
Т6	17	3	17.6	37	0	0	23	4	17.4	28	4	14.3	
T7	17	4	23.5	36	0	0	22	7	31.8	28	5	17.9	
Т8	17	5	29.4	37	2	5.4	23	6	26.1	30	6	20.0	
Т9	17	4	23.5	38	3	7.9	25	10	40.0	30	6	20.0	
T10	18	5	27.8	36	4	11.1	25	11	44.0	30	5	16.7	
T11	19	6	31.6	39	2	5.1	25	9	36.0	30	3	10.0	
T12	18	3	16.7	38	0	0	25	6	24.0	30	3	10.0	
L1	16	4	25.0	39	1	2.6	27	13	48.1	30	10	33.3	
L2	17	3	17.6	40	9	22.5	26	17	65.4	30	13	43.3	
L3	16	7	43.8	39	13	33.3	27	19	70.4	30	13	43.3	
L4	17	7	41.2	40	17	42.5	27	21	77.8	29	17	58.6	
L5	16	7	43.8	40	15	37.5	27	17	63.0	29	18	62.1	
TOTAL	407	89	21.9	896	94	10.5	588	204	34.7	687	135	19.7	

Appendix Table 4. Prevalence per individual vertebrae of high severity of osteophytosis (Grade 2 and above) in four sex age groups

			Adult	Young					Adult	Middle		
		Male			Female			Male			Female	
	Ν	Mean	SD	Ν	Mean	SD	Ν	Mean	SD	Ν	Mean	SD
C1	14	0.14	0.53	35	0.00	0.00	24	0.25	0.68	28	0.00	0.00
C2	17	0.41	0.87	33	0.03	0.17	24	0.50	1.18	31	0.00	0.00
C3	16	0.31	0.79	31	0.23	0.62	23	0.87	1.49	27	0.41	0.69
C4	17	0.47	0.87	34	0.56	0.86	22	0.82	1.37	24	0.71	1.00
C5	18	1.06	1.26	36	0.69	0.98	23	1.26	1.36	27	1.26	1.20
C6	18	1.00	1.08	36	0.72	0.94	24	1.42	1.21	27	1.11	1.09
C7	17	0.88	1.05	39	0.38	0.71	25	1.00	1.22	28	0.54	0.69
T1	18	0.39	0.61	39	0.10	0.31	25	0.56	0.82	30	0.07	0.25
T2	18	0.33	0.59	38	0.08	0.27	25	0.40	0.71	29	0.03	0.19
Т3	17	0.53	0.72	39	0.21	0.41	24	0.88	1.15	28	0.21	0.42
T4	15	0.60	0.51	38	0.47	0.60	24	1.04	1.12	27	0.37	0.49
Т5	17	0.76	0.56	39	0.59	0.55	23	0.91	0.73	27	0.44	0.58
Т6	17	0.94	0.83	37	0.57	0.50	23	1.00	0.85	28	0.57	0.74
T7	17	0.94	0.90	36	0.56	0.50	22	1.23	0.97	28	0.82	0.72
Т8	17	0.88	0.86	37	0.62	0.59	23	1.17	1.03	30	0.80	0.85
Т9	17	1.18	0.88	38	0.66	0.63	25	1.36	1.22	30	0.73	0.78
T10	18	1.00	1.03	36	0.44	0.69	25	1.52	1.42	30	0.73	0.74
T11	19	0.89	0.88	39	0.41	0.59	25	1.40	1.50	30	0.70	0.75
T12	18	0.72	0.75	38	0.32	0.47	25	1.08	1.32	30	0.60	0.67
L1	16	0.94	0.93	39	0.51	0.56	27	1.59	1.12	30	1.20	1.16
L2	17	0.94	0.97	40	0.93	0.80	26	2.04	1.11	30	1.47	1.17
L3	16	1.19	1.11	39	1.23	1.13	27	2.11	1.09	30	1.47	1.07
L4	17	1.24	1.03	40	1.45	1.13	27	2.19	1.00	29	1.69	1.07
L5	16	1.38	1.09	40	1.28	1.15	27	1.78	1.05	29	1.83	1.14
TOTAL	407	0.80	0.92	896	0.55	0.79	588	1.20	1.23	687	0.74	0.95

Appendix Table 5. The mean osteophytic value (MOV) (mean osteophyte grades of individual vertebra) in four sex-age groups
		Male Adult Young			Female Adult Young			Male Adult Middle			Female Adult Middle (Individual N=30)		
		N	Mean	SD	N	Mean	SD	N	Mean	SD	N	Mean	SD
C1	L	12	0.00	0.00	66	0.05	0.27	24	0.04	0.20	49	0.02	0.14
	R	12	0.00	0.00	66	0.03	0.17	24	0.04	0.20	51	0.04	0.20
C2	L	16	0.13	0.50	64	0.08	0.32	2.5	0.40	1.00	57	0.14	0.40
	R	16	0.06	0.25	64	0.08	0.32	2.5	0.60	1.19	55	0.16	0.42
C3	L	18	0.33	0.84	62	0.10	0.39	26	1.35	1.32	48	0.31	0.78
	R	18	0.11	0.32	62	0.06	0.31	26	0.96	1.18	48	0.17	0.56
C4	L	18	0.44	0.86	64	0.08	0.41	24	0.92	1.14	44	0.34	0.91
	R	18	0.44	0.86	64	0.03	0.18	24	0.88	0.99	44	0.20	0.59
C5	L	18	0.22	0.94	70	0.09	0.41	24	0.71	0.95	48	0.40	0.87
	R	18	0.28	0.46	70	0.07	0.26	24	0.88	0.95	48	0.21	0.41
C6	L	18	0.33	0.97	70	0.06	0.23	22	0.59	0.96	47	0.26	0.61
	R	18	0.22	0.43	70	0.04	0.20	22	0.41	0.67	47	0.19	0.54
C7	L	18	0.39	1.04	74	0.04	0.20	24	0.96	1.16	48	0.29	0.77
	R	18	0.39	1.04	74	0.03	0.16	24	0.71	0.91	48	0.27	0.61
T1	L	16	0.44	1.03	76	0.05	0.22	26	0.42	0.90	54	0.20	0.56
	R	16	0.50	1.10	76	0.07	0.25	26	0.46	0.81	54	0.17	0.47
Т2	L	16	0.13	0.34	72	0.04	0.20	26	0.04	0.20	52	0.10	0.30
	R	16	0.13	0.34	72	0.08	0.40	26	0.08	0.27	52	0.10	0.30
Т3	L	14	0.36	0.63	78	0.04	0.19	26	0.04	0.20	48	0.02	0.14
	R	14	0.50	0.94	78	0.08	0.39	26	0.08	0.39	48	0.02	0.14
T4	L	12	0.50	0.90	72	0.03	0.17	24	0.00	0.00	52	0.06	0.31
	R	12	0.67	1.15	72	0.03	0.17	24	0.00	0.00	52	0.08	0.33
T5	L	14	0.21	0.58	74	0.08	0.27	22	0.00	0.00	48	0.08	0.28
	R	14	0.21	0.58	74	0.08	0.27	22	0.00	0.00	48	0.17	0.48
Т6	L	12	0.33	0.78	70	0.09	0.28	24	0.00	0.00	52	0.12	0.32
	R	12	0.33	0.78	70	0.11	0.36	24	0.00	0.00	52	0.15	0.46
Т7	L	12	0.42	0.79	70	0.06	0.23	22	0.00	0.00	50	0.12	0.33
	R	12	0.42	0.79	70	0.10	0.42	22	0.00	0.00	50	0.16	0.42
Т8	L	14	0.29	0.73	72	0.07	0.26	24	0.04	0.20	54	0.11	0.37
	R	14	0.29	0.61	72	0.06	0.23	24	0.04	0.20	54	0.13	0.44
Т9	L	14	0.57	1.16	68	0.15	0.36	24	0.13	0.34	54	0.31	0.58
	R	14	0.57	1.16	68	0.16	0.37	24	0.17	0.48	54	0.28	0.60
T10	L	16	0.75	1.13	66	0.23	0.42	24	0.17	0.48	54	0.30	0.60
	R	16	0.75	1.13	66	0.21	0.41	24	0.54	1.22	54	0.24	0.47
T11	L	16	0.38	0.62	70	0.26	0.56	26	0.38	1.13	53	0.38	0.77
	R	16	0.38	0.62	70	0.24	0.55	26	0.38	1.13	53	0.38	0.74
T12	L	14	0.43	0.65	72	0.21	0.50	26	0.31	1.09	48	0.29	0.68
	R	14	0.36	0.50	72	0.21	0.50	26	0.00	0.00	48	0.38	0.79
SUM		576	0.35	0.79	2660	0.09	0.33	926	0.34	0.82	1920	0.19	0.53

Appendix Table 6. Mean severity of joint facet osteoarthritis in cervical and thoracic segments

Notes for Authors



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